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Challenges and innovations for grasslands resilience

Edited by

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Foreword

The Sociedade Portuguesa de Pastagens e Forragens welcomes the delegates to the 31st General Meeting of the European Grassland Federation (EGF). It is with enormous pleasure and honour that we organise and once again host this event in Portugal, after 40 years. Over these four decades, Portuguese and European grassland farming have undergone tremendous changes. Several major drivers have increasingly shaped grassland systems: climate change, rural land abandonment, competing land-use demands (agroforestry, energy, rewilding), biodiversity loss, wildfire risk, soil and water conflicts, changing diets, shifts in livestock systems, and geopolitical instability. Scientific and technical innovations, adaptive practices, emerging technological tools, and supportive public policies are essential for strengthening the role of grasslands in resilient European food systems and their capacity to buffer future shocks. These critical issues will be approached within the General Meeting theme, “Challenges and Innovations for Grasslands Resilience”, structured in five sessions:

- **Session 1.** – Climate change and grasslands soil and water management
- **Session 2.** – Agroforestry, pastoralism and society
- **Session 3.** – Breeding for new challenges
- **Session 4.** – Sustainable livestock production systems based on grasslands: Challenges and opportunities
- **Session 5.** – Grasslands and rural land policy tools

To strengthen the EGF network with other researchers, the organizing committee agreed a cooperation with the European Federation for Animal Science (EAAP), which resulted in the organization of session 4.

In the Mid-tours, participants can choose from visits to 11 private commercial farms, the National Plant Breeding Station and one private enterprise of Plant Breeding, organised into 6 excursions. We will observe and discuss a range of farming approaches and technologies, from traditional to innovative, within the Mediterranean agro-forestry environment.

The conference contributions explore how grasslands can be managed, improved, and reimaged as multifunctional systems that simultaneously support climate change mitigation and adaptation, biodiversity, soil health, and sustainable livestock production. They integrate advances in ecology, agronomy, animal science, breeding, agroforestry, digital technologies, and social sciences to address trade-offs, enhance resilience, reduce greenhouse gas emissions, and strengthen the links between grassland management, farmer decision-making, and societal goals.

The post-conference tour will visit the Azores Island of ‘Terceira’. We will observe the local traditional animal production systems based on permanent grasslands under the natural beauty of this island’s landscape.

At the end of the General Meeting, we expect to identify strategic knowledge for grassland stability, adaptive and mitigation solutions. It will also be relevant to highlight knowledge gaps for future research, provide insights to policymakers, industry, educators, and farmers and, finally, but fundamental, to stimulate young researchers’ interest in grassland topics.

The heart of the General Meeting is the participants: thus, we thank all authors for their contributions and the large group of reviewers for their time and essential guidance, which ensures the integrity and success of the event. We would also like to thank the Scientific Committee, the Organising Committee, sponsors, supporters and suppliers who have contributed to the delivery, support and organisation of the General Meeting.

Ultimately, we hope that the 31st General Meeting of the European Grassland Federation will serve as an outstanding platform to provide attendees with significant new knowledge about grassland resilience, along with opportunities to socialise and expand their professional contacts.

Prof. J.P. Fragoso Almeida

Chair of the Organising Committee

Prof. Henrique Trindade

Chair of the Scientific Committee

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Session 1.

**Climate change and grasslands soil and
water management**

Insights from Portuguese grasslands

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Abstract

In Portugal, significant changes in land use have occurred over recent decades, characterised by a reduction in the area occupied by annual crops (particularly autumn–winter cereals) and an increase in the proportion of grasslands within the Utilised Agricultural Area. At present, grasslands cover more than 2 million ha; however, over half of this area consists of natural grasslands. Livestock production accounted for approximately one third of the value generated by agricultural production in Portugal. Approximately 46% of the national livestock population is composed of cattle, followed by poultry (22%), pigs (21%), and sheep and goats (11%). Grassland and livestock production are especially significant in the Alentejo region, where the agro-silvo-pastoral system known as the *Montado* predominates. Grasslands constitute a vital ecosystem, providing essential social, economic, and environmental functions that require preservation. Consequently, appropriate grassland management and adaptation to climate change are essential, with precision agriculture and plant breeding representing key tools to support these objectives.

Keywords: extensive livestock farming, Mediterranean grasslands, Portugal

Introduction

Over the last six decades, the factors of production in Portuguese agriculture (land, water, labour, intermediate goods and services, and capital) have evolved in different ways. The average size of farms almost tripled. Land and labour productivity have increased significantly, reflecting the progress of the national agri-food complex and the strengthening of knowledge and innovation in the sector (Avilez, 2024). The Utilised Agricultural Area (UAA) remained stable between 1962 and 2022; however, arable land (mainly the area occupied by cereals) has declined over the last three decades (2.3 and 1.0 million ha in 1989 and 2019, respectively), while the area occupied by permanent pastures (PP) has increased consistently (0.9 and 2.0 million of ha in 1989 and 2019, respectively) (INE, 1992, 2021), mainly due to the expansion of natural grasslands.

Grassland production in Portugal (mainland and the Açores Archipelago) is constrained by distinct soil and climatic factors, with the main differences relating to rainfall availability and regularity and to temperature range. In mainland Portugal, the majority of the territory is under mediterranean climate, *i.e.* winter rainfall and hot dry summers, whereas in the Açores the humid Atlantic climate, with well-distributed rainfall and mild temperatures all over the year, allows for more continuous and stable grassland production. Thus, in mainland Portugal, particularly in the southern regions, natural grasslands predominate, with high floristic diversity, alongside some improved or sown PP. The natural grasslands are usually composed of native annual species, possibly including some self-reseeding annual

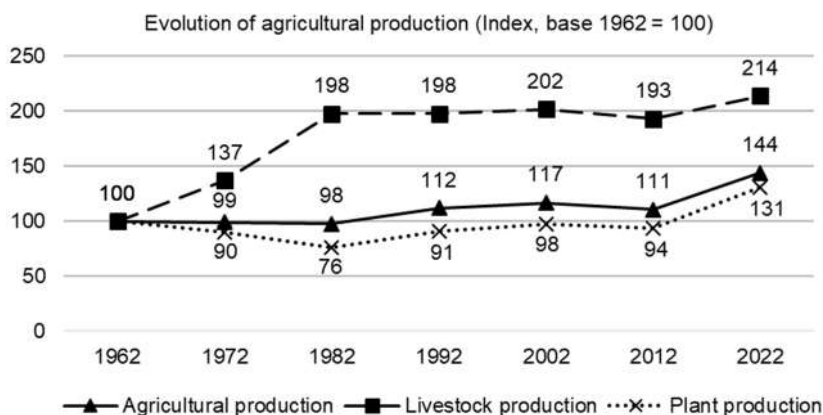


Figure 1. Evolution of agricultural production volume in Portugal ($PV_{\text{const}}^{\text{"1962"}}$ = 100). Source: Avillez (2024).

legumes and some well-adapted annual and perennial grasses but are generally not improved to ensure acceptable yields (Freixial and Barros, 2012). In all cases, production is concentrated in spring, with a marked decline during summer, thereby requiring the use of forage crops supplementation. In central and northern mainland Portugal, annual rainfed pastures are replaced by mountain PP, which includes meadows, mesophile perennial *Nardus* grasslands and other perennial species.

In the Açores archipelago, permanent grasslands, grazed all year-round, are dominated by perennial grasses and legumes and, occupy approximately 75% of the regional UAA. This region is, therefore, the most suitable region for dairy production in Portugal.

The purpose of this paper is to provide an overview on Portuguese grasslands sector. In addition to presenting the evolution of agricultural production in the country, it analyses the development of livestock production and animal production systems and concludes with a discussion of current constraints and future prospects for grassland production in Portugal.

Brief notes on the evolution of agricultural production in Portugal

Portuguese agricultural production accounts for around 2% of the European Union's total (Nunes, 2024). Between 1962 and 2022, agricultural production volume (measured by the value of output at constant prices) grew at an average annual rate of only 0.61% (Figure 1).

Over the last decade, crop production has grown more rapidly ($+3.3\%$ year⁻¹) than livestock production ($+1.0\%$ year⁻¹). According to Pordata (2024), in 2023 livestock production accounted for approximately one third of the value generated by agricultural production in Portugal (€12 157 million).

Evolution of livestock production in Portugal

Portugal holds a total of 2.42 million Livestock Units (LU), corresponding to a stocking rate of 0.6 LU ha⁻¹ of Utilized Agricultural Area (UAA) (INE, 2021). Over the last 40 years, livestock heads have fluctuated between 2.13 and 2.47 million (Trindade and Pereira, 2023). Approximately 46% of the total livestock population (LU basis) consists of cattle, 22% poultry, 21% pigs and 11% sheep and goats.

Over the last 20 years, the average cattle—meat herd size per farm has more than tripled. Of the total herd of 1.5 million head, 84.5% are intended for meat production (INE, 2021).

Regarding sheep production, the 2.2 million head registered in 2019 were distributed across approximately 43 thousand farms, with the majority located in the Alentejo region. The number of dairy sheep has been declining, representing 11.3% of the total sheep population in 2019.

The goat population in the same year amounted to 372.3 thousand heads, with 23.0% of the total population located in the Alentejo region. Dairy goats represent 29.4% of the total goat population.

Considering the meat sector, in 2024 domestic production supplied 75.2% of the quantity required to meet national market demand (INE, 2025). Sheep and goat meat production was sufficient to ensure national self-sufficiency. However, the lower levels of self-sufficiency in pork (72.8%) and beef (50.9%) indicate favourable conditions (particularly in the domestic market) for a continued increase in livestock numbers, especially cattle.

Changes in agricultural production and consequences for grassland areas

According to INE (2024), agricultural holdings occupied 5.1 million ha in 2023, corresponding to 55.5% of the national territory. The UAA, 3.86 million ha, represent 77.4% of the total area of agricultural holdings and has remained relatively stable over the past 60 years. The number of farms declined markedly, from 813 500 in 1968 to 261 500 in 2023 (INE, 2024). Smaller farms (particularly family holdings, located mainly in the north) have largely disappeared. Conversely, the number of larger farms has increased, a trend that is likely to influence the optimization of production processes and, consequently, productivity levels, particularly in livestock farming.

Over recent decades, land-use patterns have also changed substantially, with significant annual reductions in cultivated area, especially cereals. Over the last decade, the cereals area fallen by 40% (–125 000 ha) (Barradas *et al.*, 2025). The low unit yields recorded for autumn-winter cereals, provide a strong explanation for this sharp reduction. In contrast, the proportion of meadows and grasslands within the UAA increased from 22% in 1962 (Avillez, 2024) to 54% in 2023, corresponding to approximately 2.2 million ha (INE, 2024).

The substantial expansion of permanent grassland can be attributed to a combination of social, political, economic and ecological—adaptation to the low fertility soils, factors. These include the abandonment of agricultural land due to rural exodus in search of better living conditions, leading to its transformation into permanent meadows and grasslands through natural regeneration or human intervention; the conversion of arable land previously used for annual rainfed crops (e.g. cereals); and the implementation of the Common Agricultural Policy (CAP). The CAP has promoted the preservation of PP and its expansion by the replacement of low-productivity arable land and, simultaneously, encouraging sustainable agricultural practices. In addition, CAP incentives for soil and biodiversity conservation, together with its role in climate change mitigation, have contributed to this trend. Further drivers include the growth of extensive livestock systems (particularly in the Alentejo and Trás-os-Montes regions) and the increased emphasis on quality livestock production and cost reduction (Trindade *et al.*, 2024).

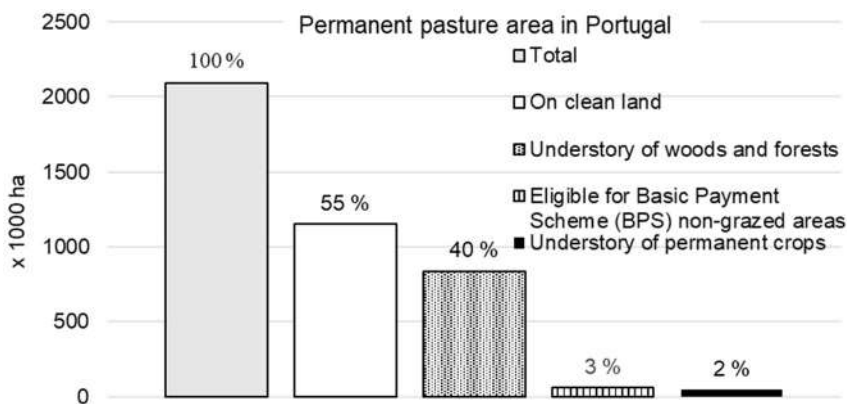


Figure 2. Permanent pasture area in Portugal (INE, 2021).

According to the 2019 Agricultural Census (Figure 2), approximately 70% of the PP area consists of natural pasture. The Alentejo region accounts for around 64% of the total area of PP, of which only 24% corresponds to improved spontaneous and sown pastures. In contrast, in the Açores region, improved and sown pastures represent 91% of the total PP area in the archipelago (Trindade and Pereira, 2023).

According to Vilão *et al.* (2024), PP are the main type in organic farming, representing around 71.3% of the total area (around 760 000 ha in 2022).

Animal production systems with ruminants

Although Portugal is a small country, animal production systems differ substantially across regions, reflecting differences in geography and climate, soil types, landholding size and structure, cultural traditions, ownership patterns and the social characteristics of the population.

According to Fernandes *et al.* (2005), the extensive grazing systems in mainland Portugal are characterized primarily by low external nutrient input, low stocking density, minimal or no use of agrochemicals, preferential use of native breeds, absence of widespread irrigation, limited mechanization, and permanent grazing of plots. Two main types were identified, determined by bioclimatic factors: (1) the “Galaico Duriense” grazing system, which includes the inland mountain systems of Trás-os-Montes, and the (2) Mediterranean systems, characterized by increasing degrees of aridity. The latter include the subsystems of: “Terra Fria” (“Mirandês” and “Beira Alta” plateaus) in the north; the “Serra da Estrela” grazing subsystem and the “Maciço Calcário Estremenho” grazing subsystem in the center; the “Mixed Alentejo” grazing subsystem and the “Serra Algarvia” grazing subsystem in the south.

The (1) “Galaico Duriense” grazing system develops in an area occupied by heathland (*Calluna vulgaris*), gorse (*Ulex* spp.), and other scrub in wet depressions. In the lowlands, these areas are complemented by grasslands dominated by matgrass (*Nardus stricta*). This mosaic includes scrubland dominated by ericaceous (*Ericaceae*) in the higher-altitude zones, which serves as a complementary resource for cattle and is occasionally grazed by predominantly sheep flocks. This grazing system is based on family farms, highly dependent on rented land, based on permanent semi-natural meadows called “Lameiros”, dominated by grasses (e.g. *Lolium perenne* L. and *Agrostis castellana* Boiss. and

Reut.). Typically located in areas of high-water availability and fertile soils, often at altitudes above 700 m and in the mountain regions of northern and central mainland Portugal.

Further east, where the continental and Mediterranean influences overlap with the Atlantic domain, lie the “Terra Fria”, “Mirandês” and “Beira Alta” plateau subsystems. These subsystems include the cattle production, focused on the “Mirandesa” breed or crossbreeds, and the sheep and goat production (on short distance transhumances), complemented with “Lameiros”. In this region the family predominates also. However, all subsystems have experienced a clear decline due to inadequate production structures, limitations in the land market, and a reduction in the social and economic status of livestock producers.

The “Serra da Estrela”, located in central interior region of the country, is located at the highest mountain area in mainland Portugal. The pastures of the Serra da Estrela are characterized as being natural, high-altitude, and semi-natural, composed of spontaneous perennial grasses, legumes such as white clover and subterranean clovers, adapted to acidic soils and extreme climates. The traditional pastoral system of the Serra da Estrela aims sheep’s cheese production. The former transhumance system, which involved long-distance seasonal movements, is now largely confined to much shorter displacements between the surrounding lowlands and the mountain areas.

The “Alentejo Mixed” subsystem is a complex agro-pastoral system characterized by the predominance of large private properties, with livestock production becoming significantly more important than cereal production.

The “Montado”, one of Portugal’s most emblematic ecosystems, particularly in the Alentejo region, is a multifunctional, human-shaped system. It is composed of an arboreal layer dominated by cork oak (*Quercus suber* L.) and holm oak (*Quercus ilex* ssp. *rotundifolia* Lam.), occurring in either pure or mixed stands, together with an PP (herbaceous layer largely consisting of rainfed annual species) extensively grazed by livestock all year around, including sheep, cattle, goats, and pigs (Serrano, 2019). In traditional systems, pigs of the local ‘Alentejano’ breed graze the Montado areas and are finished on acorns at 14–18 months of age. Several causes have led to an increase in cattle replacing the former traditional sheep production. The most emblematic local cattle breeds are the “Mertolenga” and “Alentejana”. Nevertheless, some farms continue to operate under the traditional Mixed Alentejo system, maintaining the cereal–fallow rotation.

In the Algarve Mountain region (south Portugal), grazing systems are located in highly marginal areas with very low productivity. These systems are predominantly family type and operate on small-scale farms. Despite the use of a high-quality native breed with significant market value (the “Algarvia” goat), competition from more attractive income opportunities, combined with the physical effort required by the activity, has led to a general decline in this system.

At the national level, the Açores archipelago (particularly the islands of São Miguel and Terceira) constitutes a major dairy production region, accounting for 30% of total Portuguese dairy output (Almeida *et al.*, 2021). Grazing systems in the Açores are based on extensive dairy, using PP (Ferreira, 2020). These pastures are managed mainly through continuous and rotational grazing, all year round. During periods of some forage scarcity, especially in dry summers and autumns (particularly at lower altitudes) and during winter, animals are supplemented with concentrate feeds and maize or grass

silage. A typical farm comprises several plots of differing sizes that are rarely contiguous, requiring dairy farmers to move livestock and equipment (milking and feeding systems) between plots via public roads and pathways (Almeida *et al.*, 2021). Silva and Barbel (2005) identified three types of grazing systems in Açores dairy farms: (1) extensive systems (fewer than 1.4 cows ha⁻¹); (2) moderately intensive systems (1.4–2.4 cows ha⁻¹); and (3) intensive systems (more than 2.4 cows ha⁻¹).

Current constraints on grassland production in Portugal

The climate of mainland Portugal is generally classified as Mediterranean, characterized by hot, dry summers and mild, rainy winters, with a significant interannual variability. In the absence of irrigation, agricultural activity is predominantly rainfed, accounting for approximately 83% of the UAA of mainland Portugal (Madeira, 2024), dependent on soil moisture, groundwater, and occasional rainfall. Consequently, agricultural production depends largely on soil moisture, groundwater, and sporadic rainfall. These conditions favour extensive farming systems, in which pastures, forage crops, and permanent crops represent the main land-use options, typically distributed within an agroforestry mosaic (Diniz, 2024). Under this context, pasture production is irregular, with periods of high productivity (110 kg DM ha⁻¹ day⁻¹) alternating with phases of very low (<20 kg DM ha⁻¹ day⁻¹) or even null production (Pysillos *et al.*, 2022). Therefore, in rainfed PP based on annual species—mainly self-reseeding annual legumes—and some perennial grasses exhibiting summer dormancy, critical periods for pasture availability occur during autumn (particularly in years with delayed onset of rainfall), winter, and summer. This pronounced seasonal variability renders ruminant-based livestock systems strongly dependent on preserved forage resources to overcome periods of scarcity and/or reduced forage quality.

In addition to climatic constraints, the predominance of soils with low or very low fertility further limits crop productivity. Most Portuguese soils exhibit low OM content (70% of agricultural land <1%), acid pH (83% of agricultural land with pH_{water} <5.5) and low cation-exchange capacity, with only 4.2% exceeding 20 meq (100 g of soil)⁻¹ (Carvalho and Lourenço, 2014). According to Fernandes *et al.* (2005), soils supporting semi-natural mountain pastures generally present pH values ranging from 4.0 to 5.8, except in soils derived from basic crystalline rocks, where pH values are higher (5.9–6.6). In mountainous regions, OM content typically exceeds 2.5%, while phosphorus (P) and potassium (K) levels are highly variable but generally low to medium.

A study carried out in Montado, in Évora region, reported significantly higher concentrations of OM, N, P, K and Mg in soils beneath cork oak canopies compared with soils located outside the canopy. Mean values beneath versus outside the canopy were, respectively: OM (3.1% vs. 1.7%), N (0.2% vs. 0.3%), P (117.7 vs. 68.2 mg kg⁻¹), K (359.3 vs. 180.5 mg kg⁻¹), and Mg (115.0 vs. 76.3 mg kg⁻¹) (Serrano *et al.*, 2017). These findings indicate that pH correction, the establishment of favourable soil chemical conditions for pasture growth (e.g., appropriate Mg/Mn ratios), and rational fertilisation strategies—particularly with respect to phosphorus—can substantially enhance both the quantitative and qualitative potential of pastures.

Under rainfed conditions, average dry matter yields typically range from 0.5–1.0 t DM ha⁻¹ year⁻¹ in semi-natural grasslands established on marginal soils, to 6.0–7.0 t DM ha⁻¹ year⁻¹ in agriculturally improved grasslands (Huyghe *et al.*, 2014). Within the scope of the GEEBovMit LA 3.4 project, annual dry matter productivities of sown pastures and natural pastures in “Alto Alentejo” in 2024, ranged from 5333 to 9571 kg DM ha⁻¹ and from 5611 to 7610 kg DM ha⁻¹, respectively (Simões,

2024). Belo *et al.* (2014), summarising results from several studies, reported productivity values for PP in Montado systems of approximately 800 kg DM ha⁻¹ year⁻¹ (Lourenço *et al.*, 1994), rarely exceeding 1500 kg DM ha⁻¹ year⁻¹ (Crespo, 1997), as well as seasonal yields of 695 kg DM ha⁻¹ during autumn–winter and 2014 kg DM ha⁻¹ in spring. In contrast, mountain pastures generally exhibit higher productivity, ranging from 4 to 6 t DM ha⁻¹ year⁻¹, and exceeding 12 t DM ha⁻¹ year⁻¹ in “Lameiros” (Moreira *et al.*, 2001 cited in Pôças *et al.*, 2006). These production differences are primarily associated with water availability and, consequently, with vegetation type and site-specific environmental conditions.

Prospects for grasslands production

One possible strategy to ensure the economic viability of rainfed farms is to reinforce their extensive nature, focusing on the use of endogenous resources and reducing dependence on external inputs, while maintaining a productive environment that safeguards natural resources (Madeira, 2024).

The Alentejo region presents a land structure suitable for year-round grazing. In addition, its climatic conditions are favourable for the development of leguminous species and forage crops, enabling improvements in pasture productivity and quality at relatively low cost, while contributing to soil fertility restoration. Grasses, in particular, increase pasture productivity and promote greater seasonal regularity, especially during winter. In addition, they contribute to a more balanced energy-to-protein ratio and prevent the invasion of nitrophilic species, such as thistles. Given that 70% of the PP area is classified as natural (INE, 2021), pasture improvement should be actively encouraged. Such improvement may involve appropriate soil fertilisation, correct grazing management, and/or the introduction of improved species through techniques that preserve the existing herbaceous cover, such as direct seeding. This technique, which has significant potential for wider adoption in Portugal, allows pasture renewal through the introduction of improved grass and legume varieties, while taking advantage of the existing substrate and facilitating livestock feeding planning. Furthermore, direct seeding contributes to reducing soil CO₂ emissions, susceptibility to erosion, a particularly relevant form of land degradation in Portugal.

Beyond their role in food security, grasslands provide a wide range of ecosystem services (Bengtsson *et al.*, 2019). These include water regulation, erosion control, the provision of medicinal plants, and habitat for non-plant organisms such as pollinating insects (Richter *et al.*, 2021). Grasslands can also contribute to stabilising farmers' incomes and diversifying agricultural activities (Carlier *et al.*, 2009), i.e. through rural tourism (Parente and Bovolenta, 2012), thereby supporting rural population retention and reducing land abandonment.

The severe depopulation in inland areas, the ageing of the rural population, the lack of effective land management policies, and increasing drought conditions associated with climate have contributed to a rise in both the frequency and intensity of wildfires, particularly in the central and northern interior of Portugal. According to available data (Pordata, 2025; ICNF, 2025), during the period 2010–2024 approximately 1.8 million ha burned nationwide, including forest stands (48%), scrubland (45%), and agricultural land (7%). The establishment of networks of biomass management reduction is one of the main fire prevention strategies (Ferreira, 2024). In this context, grazing, particularly in the mountain regions, can play an important role by mitigating wildfire risks. In addition, grasslands are crucial for climate change mitigation, storing more than 30% of the world's carbon stocks (Chen *et al.*, 2022). According to Almeida *et al.*, (2024), Portuguese pasture may function as net carbon sinks

regardless of regional differences in management intensity. Although higher emissions were observed in northern farms (where grasslands are generally more productive and intensively managed than in the south), these were offset by higher carbon assimilation. Consequently, the estimated soil carbon sequestration potential (emissions—assimilation) did not differ significantly between regions, amounting to -2049 ± 602 and -1327 ± 207 kg C ha⁻¹ year⁻¹ for northern (Trás-os-Montes) and southern (Alentejo) farms, respectively.

According to the Mediterranean Institute for Agriculture, Environment and Development, 82% of the Montado pasture area is classified as natural grassland. Carvalho (2014) highlights the decisive role of pastures in restoring Montado soil fertility, as the prolonged absence of soil mobilisation protects root “network”, prevents soil erosion, and promotes the organic matter accumulation. However, the same author emphasises the need to eliminate limiting factors and to adjust stocking rates to pasture productivity to fully realise the system potential. Therefore, increasing the area of improved grasslands in Montado systems, combined with appropriate management practices, represents the key strategy.

Pasture management involves the selection and management of suitable species and cultivars, soil fertility management, fencing, water distribution, timing, and intensity of grazing, among other factors. Effective decision-making requires access to reliable information. For example, monitoring pasture biomass and quality can directly influence fertilisation strategies and grazing frequency.

Traditional monitoring methods are labour-intensive, costly, and often characterised by low accuracy. Consequently, the potential of precision agriculture—currently adopted in only 4.2% of the utilised agricultural area and 1.6% of livestock units in Portugal (INE, 2021)—should be more extensively exploited in pasture and grassland management. According to Serrano (2025), improved knowledge of pasture productivity obtained through proximal sensors, such as the rising plate meter, enables more precise planning of animal rotation, optimising pasture utilisation and preventing under- or overgrazing. The same author highlights the Normalised Difference Vegetation Index (NDVI) as a robust indicator of pasture quality. Nevertheless, the adoption of precision grassland management and precision grazing technologies (e.g. GPS collars or virtual fencing) remains limited due to their complexity, high costs, and the generally low level of technological expertise among farmers (Ali and Kaul, 2025). These constraints require targeted training and education initiatives, led by researchers and extension services, which can enhance the benefits of digital farming for productivity, feed quality, and environmental and societal outcomes (Ali *et al.*, 2024).

Surface temperatures in the Mediterranean region are currently approximately 1.5°C above pre-industrial levels (Ali *et al.*, 2022). The intensity, frequency, and duration of temperature extremes and heatwaves have increased over recent decades (particularly during summer) and are projected to continue rising. Precipitation is expected to decrease by around 4% per 1°C of global warming across all seasons, especially in the southern Mediterranean basin (Ali *et al.*, 2022). While climate change may not directly alter the chemical composition of grasses and legumes in mixed pastures, it may substantially modify their relative abundance (Dumont *et al.*, 2015).

Mediterranean grasslands are among the most responsive to changes in precipitation regimes (Catorci *et al.*, 2021). In addition to affecting productivity, altered rainfall patterns can drive changes in plant community diversity, composition, and structure (Carmona *et al.*, 2015; Song *et al.*, 2025). Drought conditions may shift competitive balance in favor of native species over introduced ones (Frost *et*

al., 2025). Annual grasses and forbs are more likely to decline than perennials, while certain taxa (particularly legumes) may increase in relative abundance (Tilman and Haddi, 1992). Drought may also favour small-stature species and those with low specific leaf area (Alés *et al.* 1993).

Plant breeding will be fundamental in developing new varieties better adapted to altered seasonal patterns and increased variability in temperature, precipitation and growing periods driven by climate change. For Mediterranean agro-ecosystems, genotype selection should prioritise traits such as high winter vigour and rapid establishment, shorter growth cycles and earlier flowering, efficient use of water and soil nutrients, tolerance to waterlogging, high biomass and seed production, a high proportion of hard seeds or the ability to bury seeds, reserve accumulation and dormancy in perennial species, and high grazing tolerance (Carita *et al.*, 2025).

Considering all these, the future of grasslands in Portugal is likely to involve an expansion of permanent and temporary pastures, particularly under organic production systems, combined with the use of more resilient livestock breeds and increased adoption of technological tools. These developments are driven by the need for agricultural systems that are better adapted to climate change and more resilient to environmental variability. Accordingly, there is a need of research efforts to develop new species, varieties and management practices adapted to increasingly variable climatic conditions. Nevertheless, challenges such as appropriate grazing management remain critical and require specialised knowledge, supported by research, technological development, and effective transfer to farmers.

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Land-use-driven ecological thresholds defining limits to grassland management

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Abstract

Grasslands cover a large portion of the Earth's land surface. They support a unique biodiversity and agricultural productivity. However, they are among the most threatened ecosystems, as land-use intensification reduces plant diversity, homogenizes functional diversity, and destabilizes ecosystem functioning. Identifying ecological thresholds—the points at which small environmental changes trigger abrupt shifts—is critical for sustainable grassland management. Here, we synthesize key results from a multi-site, multi-year study conducted in temperate grasslands that examined the dynamics of trait distributions to characterize community organization and its response to land-use intensification (Le Bagousse-Pinguet *et al.*, 2025). This study revealed two successive thresholds along a country-wide intensification gradient: a first threshold corresponding to the onset of fertilization, and a second threshold occurring beyond 80 kg N·ha⁻¹·year⁻¹ or 500 livestock units·ha⁻¹·year⁻¹. Crossing these thresholds causes grasslands to shift from a stable, biodiversity-friendly state to a low-diversity and unstable state dominated by functionally homogeneous species. These thresholds delineate safe limits of management intensity that help maintain grassland structure, stability, and multifunctionality. Overall, this work highlights the relevance of trait-based approaches as system-level indicators to anticipate ecosystem responses to human pressures and to inform sustainable grassland management under global change.

Keywords: ecological thresholds, functional traits, grasslands, land use intensification, management, plant functional diversity

Introduction: Grasslands are a unique ecosystem

Grasslands, defined as ecosystems mainly dominated by herbaceous species, cover about 40% of the Earth's land surface, and naturally occur across vast regions such as the Eurasian steppe, the African savanna, the American prairie, and Patagonia—areas where tree growth is limited by soil or climatic conditions (Bardgett *et al.*, 2021). Contrary to what one might think, they are also native to Western Europe (Pearce *et al.*, 2023). Before the arrival of humans in the Pleistocene (<12 000 years), the presence of large mammals (elephants, mammoths, aurochs, etc.) and a diverse megafauna maintained open landscapes largely composed of herbaceous plants (dicotyledons; Bråthen *et al.*, 2021). With the arrival of humans and the decline of megafauna (Svenning *et al.*, 2024), grasslands have been maintained through livestock grazing and agriculture up to the present day.

This major shift—from natural to semi-natural ecosystems “managed” by humans—led to significant changes in grassland composition, particularly through a marked increase in graminoids driven by agropastoral activities (Gibson and Newman, 2019). A wide variety of grasslands has since developed under the combined effects of different soil–climate conditions and agricultural practices. In certain climatic and management contexts, grasslands can reach remarkable levels of plant diversity (Wilson

et al., 2012). At the square-meter scale, they are considered among the most diverse ecosystems on the planet, rivaling tropical forests, whose diversity peaks at the square-kilometer scale. Extensively managed grasslands can easily harbor 40 to 60 plant species per m² (world record is 89 species per m² in the White Carpathians; Wilson *et al.*, 2012, Čertoryje, Czech Republic). Today, when human activities threaten biodiversity everywhere (IPBES, 2019), grasslands represent a striking counterexample—showing a pathway in which the combined work of humans, animals, and nature has created and sustained, over millennia, one of the most remarkable ecosystems on Earth.

Grassland diversity plays a key role in ecosystem functioning and in shaping ecosystem responses to climate change. In this context, grasslands represent a model system for research aimed at evaluating the relationship between biodiversity and ecosystem functioning. Grassland biodiversity sustains higher yields and greater temporal stability in response to climate fluctuations (Cardinale *et al.*, 2012). Accordingly, it supports livestock production systems through the supply of forage, which varies in temporal stability and nutritional value (Allart *et al.*, 2024; Maestre *et al.*, 2022), and holds high economic importance. In addition, grassland biodiversity contributes to the regulation of biogeochemical fluxes (Gross *et al.*, 2017) and climate regulation by influencing atmospheric carbon sequestration and storage (Maestre *et al.*, 2022), enhancing water infiltration and nutrient cycling at the watershed scale, improving water purification, reducing greenhouse gas emissions such as N₂O, and protecting soils from erosion (Lavorel, 2019). It also promotes pollination and biological control through the regulation of crop pests, thereby contributing significantly to associated crop yields (Badenhausser *et al.*, 2020; Scheper *et al.*, 2023). Finally, grasslands represent a cultural heritage reflecting the long history of the relationship between humans and nature, and provide essential cultural services linked to hunting activities, tourism, education, and art (Bardgett *et al.*, 2021).

Grasslands as one of the most threatened ecosystems

A direct consequence of the close relationship between humans, animals, and nature in grasslands is the strong dependence of biodiversity on land use and agricultural practices. While traditional agropastoral practices persisted until the last century, the Green Revolution and the decline of mixed crop–livestock systems from the 1960s onwards led to profound changes in grassland ecosystems, with deleterious effects on their biodiversity. At the landscape scale, a sharp decline in grassland area has been observed since the 1970s. For example, between 1970 and 1997, 3.5 million hectares of grasslands in France were converted into annual crops. These land-use changes resulted in a loss of landscape heterogeneity and disrupted connections between habitats in agricultural matrices, leading to significant landscape fragmentation (Le Provost *et al.*, 2020). The simplification of agricultural landscapes is a major driver of biodiversity decline (Tscharntke *et al.*, 2012). This decline is largely due to the destruction of semi-natural habitats such as grasslands, hedgerows, and woodlands, which serve as source habitats and biodiversity reservoirs within agricultural landscapes (Newbold *et al.*, 2016). The loss of these habitats reduces the species pool at the landscape level and limits dispersal between communities, thereby affecting all trophic levels—from plants to birds (Le Provost *et al.*, 2020).

These extensive landscape transformations have been accompanied by the intensification of agricultural practices at the local field scale, particularly in intensively farmed lowland regions. The use of inputs such as fertilizers and pesticides exerts both direct and indirect effects on organisms (Isbell *et al.*, 2013). For example, fertilizer application alters the physicochemical properties of soils (Tilman *et al.*, 2002) and can lead to a decline in local diversity (Suding *et al.*, 2005). Fertilization favors fast-growing, highly competitive species, which exclude less competitive ones and homogenize grassland

floras (Gossner *et al.*, 2016; Grime, 1973). Such shifts in local plant communities have cascading consequences for higher trophic levels by altering the diversity of plant resources available (e.g., Le Provost *et al.*, 2021), as well as by modifying habitat structure and heterogeneity (Gross *et al.*, 2009). Agricultural intensification, whether at the landscape or local scale, is thus a key driver of biodiversity decline in agricultural ecosystems (Newbold *et al.*, 2016). This process is so pervasive and global (Bardgett *et al.*, 2021) that temperate grasslands are now among the most threatened habitats worldwide (Newbold *et al.*, 2016).

Impacts of land-use intensification on grassland ecosystem functioning

Land-use intensification has major consequences for the functioning of grassland ecosystems by decoupling biodiversity from ecosystem processes (Tilman *et al.*, 2002). Intensively managed grasslands are characterized by species-poor plant communities dominated by functionally similar species. Such functional homogenization limits opportunities for complementarity in resource use among species, and thus reduces the potential positive effects of diversity on productivity (Gross *et al.*, 2009; Schaub *et al.*, 2020; Weigelt *et al.*, 2009). Fertilization may also disrupt plant–soil feedbacks and the ability of diverse microbial communities to provide essential nutrients to plants (Fontaine *et al.*, 2024). More broadly, emerging evidence suggests that the loss of species and functional diversity reduces the capacity of grasslands to sustain multiple ecosystem services (Allart *et al.*, 2024; Soliveres *et al.*, 2016), particularly under environmental fluctuations such as drought or increased climate variability (García-Palacios *et al.*, 2018). However, land-use intensification can also provide benefits for farmers. Fertilization practices promote grassland productivity, as nitrogen is one of the most limiting factors for plant growth (Tilman *et al.*, 2002). Fertilization can additionally stabilize yields and promote grassland recovery after drought in some cases (Bloor and Bardgett, 2012; Luna *et al.*, 2023). This has led to an apparent paradox: the most productive grasslands generally exhibit the lowest levels of biodiversity, even though it is widely acknowledged that biodiversity plays a key role in sustaining productivity and ecosystem functioning. Resolving this paradox requires understanding how land-use intensification fundamentally alters the organizational structure and functioning of grassland ecosystems.

Recent evidence suggests that land-use intensification has shifted grassland ecosystems from a high-biodiversity, stable state toward a low-biodiversity, more fragile state (Le Bagousse-Pinguet *et al.*, 2025). This insight stems from the identification of ecological thresholds that mark changes in grassland structure and stability along intensification gradients. These thresholds set the maximum levels of management intensity that can be applied without fundamentally altering the functioning of these ecosystems or compromising their resilience to external disturbances—information that is essential for designing conservation strategies and restoration actions to prevent widespread grassland degradation under climate change.

Identifying ecological thresholds from functional traits

Ecological thresholds are points at which minor changes in environmental conditions can trigger disproportionately large impacts on ecosystems (Hillebrand *et al.*, 2020). Understanding where these thresholds lie is therefore essential for predicting ecosystem responses to environmental change and for establishing ‘safe operating spaces’ to guide sustainable management. While the concept of ecological thresholds is widely acknowledged, their frequency and predictability in natural ecosystems remain poorly understood (Hillebrand *et al.*, 2020). Identifying these thresholds is particularly challenging because it requires selecting appropriate system variables, i.e., response metrics that accurately reflect the intrinsic organization of ecosystems. This challenge stems from the high spatio-temporal

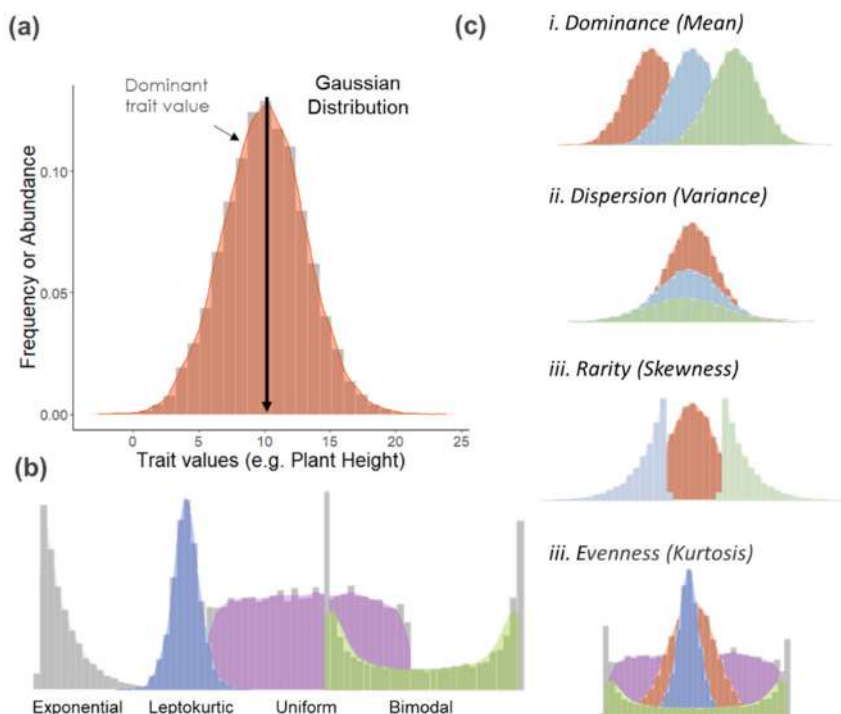


Figure 1. Trait distributions reflect how species assemble within communities and influence ecosystem dynamics and functioning. Trait distributions describe the relationship between different trait values and their abundance within a community (here, plant height in cm). Trait distributions are often represented as Gaussian distributions, where the mean reflects the trait value of the dominant species (a). However, trait distributions are not always Gaussian and can exhibit various shapes (b), including peaked, asymmetric, or bimodal distributions. Uniform distributions represent, for instance, the maximum of trait evenness, where all trait values occur at equal abundance. Importantly, the moments of trait distributions can be used to quantify different facets of trait diversity (c). While the mean and variance characterize functional dominance and dispersion in a normal distribution, skewness and kurtosis provide insights into the shapes of complex trait distributions encountered in naturally assembled communities. Skewness measures asymmetry: high negative or positive values indicate left- or right-tailed distributions, often resulting from rare species with infrequent trait values compared with the bulk of the distribution, defining functional rarity. Kurtosis reflects the relative “peakiness” of the distribution, where low kurtosis values indicate functionally even distributions.

variability of natural systems and the complex interplay of abiotic and biotic factors that drive their dynamics.

The functional traits of plant species—i.e., any morphological or physiological attributes of plants—reflect how plant species share resources, and influence ecosystem functioning (Lavorel and Garnier, 2002). The distributions of traits within communities therefore describe how plant communities are organized (Fig. 1). In this context, trait distributions may serve as suitable system variables to detect ecological thresholds as they reflect how species assemble within communities and how they respond to environmental changes. Trait distributions often deviate from a Gaussian distribution (Fig. 1a) and exhibit complex shapes that convey meaningful biological information (Enquist *et al.*, 2015; Gross *et al.*, 2017). For instance, peaked and narrow (i.e., leptokurtic) distributions (Fig. 1b) or asymmetric ones (e.g., exponential distributions) reflect low trait evenness, i.e., low trait diversity. Such distributions occur when a few species dominate the community and functionally rare species are missing (e.g., in the case of asymmetric light competition or environmental filtering, Grime, 1973). In contrast, even trait distributions (i.e. uniform distributions; Fig. 1b) indicate high trait diversity through

the co-occurrence of species with contrasting resource-use strategies (e.g., niche differentiation, Maire *et al.*, 2012). Investigating the dynamics of trait distributions in response to land-use intensification can illuminate how plant species characterized by contrasting ecological strategies interact and organize in response to human disturbances.

In Le Bagousse-Pinguet *et al.* (2025), we relied on a long-term, multi-site dataset (2008–2020) encompassing 150 agricultural grasslands distributed across farms in Germany (Fischer *et al.*, 2010) to assess whether trait distributions can be used to identify ecological thresholds associated with land-use intensification ($n = 1,950$ observations over 13 years). The selected grasslands spanned a wide and well-documented gradient of management intensity, from extensively managed systems without mineral inputs to highly intensified grasslands receiving more than $400 \text{ kg} \cdot \text{ha}^{-1} \cdot \text{year}^{-1}$ of fertilizer. We specifically examined the distribution of specific leaf area (SLA) within these grasslands, a widely recognized indicator of plant resource-use strategies that strongly influences plant growth and ecosystem productivity under land-use change (Lavorel and Garnier, 2002; Newbold *et al.*, 2016). Trait distributions were analyzed using a recently developed framework (see Le Bagousse-Pinguet *et al.*, 2025 for methodological details; Fig. 2) designed to explicitly account for the pronounced spatial and temporal variability of trait distributions.

Our analyses identified two successive thresholds beyond which grassland structure, diversity, and stability changed abruptly. The first threshold coincided with the introduction of fertilization practices, whereas the second occurred when fertilization rates exceeded $80 \text{ kg N} \cdot \text{ha}^{-1} \cdot \text{year}^{-1}$ or when grazing pressure surpassed $500 \text{ livestock units} \cdot \text{ha}^{-1} \cdot \text{year}^{-1}$. These thresholds were consistently detected both within and across years and were observed across grasslands differing in pedoclimatic conditions and species composition (Fischer *et al.*, 2010). Together, they delineate the level of anthropogenic pressure that grassland ecosystems can withstand before experiencing rapid and pronounced structural and functional reorganization.

Ecological thresholds reveal alternative grasslands states

The identification of ecological thresholds associated with land-use intensification reveals the presence of alternative states in grassland ecosystems that differ markedly in their organization and temporal stability (Fig. 2). Semi-natural grasslands, managed without fertilization and subjected to low grazing and mowing pressure, display remarkably similar functional structures across Germany, with trait distributions consistently approaching a uniform shape (System A; Fig. 2). Because uniform distributions correspond to the theoretical maximum of trait evenness, this pattern indicates that extensively managed grasslands operate close to this upper limit. Such high trait evenness is likely maintained by niche differentiation processes, including complementarity in resource use and positive or higher-order species interactions, which together facilitate the stable coexistence of contrasting ecological strategies and the persistence of functionally rare species. Notably, comparable patterns of maximized trait evenness have been documented in other natural ecosystems, including drylands and forest systems worldwide (Gross *et al.*, 2017; Liu *et al.*, 2020), suggesting that semi-natural grasslands share fundamental organizational features with a broad range of natural ecosystems.

Crossing land-use intensity thresholds pushes grasslands toward a low-diversity and highly unstable state (Fig. 2, system B). When fertilization exceeds $80 \text{ kg} \cdot \text{ha}^{-1} \cdot \text{year}^{-1}$ or cattle density exceeds $500 \text{ livestock units} \cdot \text{ha}^{-1} \cdot \text{year}^{-1}$, trait distributions in managed grasslands become more variable over time

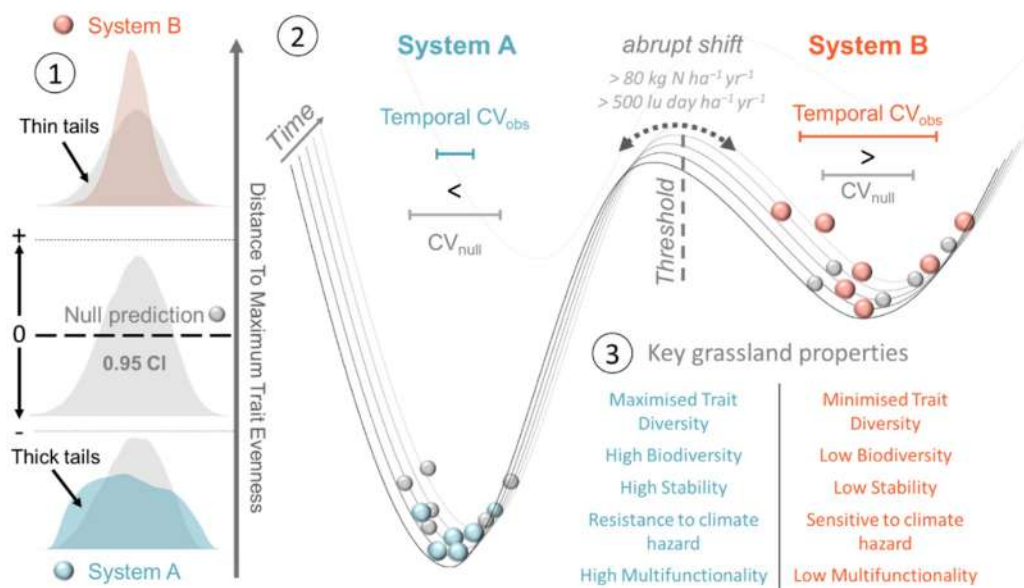


Figure 2. Effect of land-use intensification on community assembly and dynamics. The “ball-in-a-cup” metaphor summarizes results from Le Bagousse-Pinguet (2025). (1) Observed trait distributions in extensively and intensively managed grasslands (Systems A and B, respectively) were compared to null predictions simulating random distributions of trait values across communities. (2) For each system, the distance to maximum evenness was quantified as the deviation of observed distributions from the uniform distribution, which represents the mathematical maximum of trait evenness. Abrupt shifts in community assembly and dynamics were observed when land-use intensification exceeded thresholds: fertilization $> 80 \text{ kg N ha}^{-1} \text{ yr}^{-1}$ or grazing $> 500 \text{ livestock units} \cdot \text{ha}^{-1} \cdot \text{yr}^{-1}$. Extensively managed systems are characterized by stable trait distributions (lower temporal coefficient of variation, CV, than null predictions) and high evenness. In contrast, intensively managed grasslands exhibit unstable, highly peaked distributions. (3) Changes in the dynamics of trait distributions have profound consequences for key grassland properties, including biodiversity, stability, and ecosystem functioning.

and increasingly peaked and narrow. Intensification minimizes trait evenness by promoting the over-dominance of fast-growing species (high SLA values) and excluding functionally rare species. These drastic changes in organizational structure have several implications:

- (i) **Low biodiversity:** Intensively managed grasslands are dominated by a homogeneous plant type (Gossner *et al.*, 2016), and the highly variable, thin-tailed distributions indicate that functionally rare species cannot persist over time within communities. This pattern likely reflects strong competitive asymmetries occurring in productive environments (Enquist *et al.*, 2015; Schamp *et al.*, 2008), threatening the maintenance of high grassland biodiversity.
- (ii) **High sensitivity to climate variability:** During the 2008–2020 time series, intensively managed grasslands showed marked variability during the 2014–2018 period, coinciding with the worst multi-year drought in Central Europe in the last 250 years. Specifically, our time series revealed that trait distributions shifted toward extreme specialization, and rare phenotypes were excluded during the 2018 drought. This result suggests that climatic extremes exacerbate the effects of land-use intensification on grassland diversity and functioning.
- (iii) **Reduced multifunctionality:** Intensively managed grasslands in Germany exhibit lower ecosystem multifunctionality (Felipe-Lucia *et al.*, 2020; Saiz *et al.*, 2022). Some complementary studies (e.g.

Allart *et al.*, 2024) even suggest that fertilization above the 80 kg·ha⁻¹ threshold reduces grassland multifunctionality and resistance to temperature-driven impacts of climate change.

Overall, land-use intensification strongly reduces trait evenness, leading to unstable trait distributions that are highly vulnerable to additional disturbances, including climate extremes. From a functional perspective, maintaining high levels of trait evenness therefore emerges as a key objective for enhancing the long-term sustainability of grassland ecosystems under global change.

Identifying ecological thresholds for optimal ecosystem management

The occurrence of alternative grassland states may shed new light on the biodiversity–ecosystem productivity paradox. The way species interact and assemble within communities strongly influences ecosystem functioning and stability. We found that intensively managed grasslands (System B in Fig. 2) exhibit low functional trait diversity, reflecting limited opportunities for species coexistence. In this context, processes such as niche differentiation and complementarity in resource use may have a weak influence on ecosystem productivity (Maire *et al.*, 2012). In intensively-managed systems, ecosystem functioning may therefore be mostly driven by the traits of dominant species (Grime, 1998; Lavorel and Garnier, 2002) and by high fertilization rates. In contrast, natural and semi-natural grasslands (System A in Fig. 2) are characterized by a high temporal stability associated with a maximized trait diversity. These systems may be primarily driven by endogenous factors such as biotic interactions, where niche differences and complementarity among species could promote the observed stable coexistence of functionally dissimilar species (Maire *et al.*, 2012). In these grassland states, positive relationships between biodiversity and ecosystem productivity are expected to be strong and, in some cases, may match the high productivity observed in highly fertilized systems (Gross *et al.*, 2009; Schaub *et al.*, 2020; Weigelt *et al.*, 2009).

Management practices do not necessarily entail a trade-off with biodiversity. Between the two alternative states, A and B (Fig. 2), Le Bagousse-Pinguet *et al.* (2025) found an additional state, a transition phase between unfertilized and highly fertilized grasslands, in which grasslands can maintain a high stability and trait diversity, although plant species richness and functional diversity remain lower than in unfertilized systems. This transition begins with the introduction of fertilization and ends when the 80 kg·ha⁻¹·year⁻¹ threshold is reached. These abrupt transitions suggest that fertilization acts as a primer, altering ecosystem structure and dynamics regardless of the total quantity applied. This pattern is consistent with recent studies showing that increasing fertilization beyond 80N kg·ha⁻¹·year⁻¹ or 500 livestock units·ha⁻¹·year⁻¹ no longer increases plant biomass but instead leads to higher nutrient leaching and reduced ecosystem multifunctionality (Felipe-Lucia *et al.*, 2020; Saiz *et al.*, 2022). Considering that land-use intensification and eutrophication are major threats to biodiversity and ecosystem functioning (Newbold *et al.*, 2016), identifying ecological thresholds helps define the safe limits of intensification that grassland ecosystems can absorb without undergoing a complete reorganization. Recognizing the intermediate transition phase also provides a framework for targeting sustainable management and fertilization practices that promote high yield while preserving the functional integrity of grassland systems.

Conclusion

Grasslands comprise a broad range of habitats shaped by contrasting floras, pedoclimatic conditions, and management regimes. Yet, the body of evidence synthesized here indicates a consistent distinction between natural and semi-natural grasslands and their intensively managed counterparts. Extensively

managed grasslands exhibit high levels of trait evenness—a structural feature also observed in other natural ecosystems—underscoring their strong degree of naturalness. In contrast, land-use intensification operates as an external forcing that disrupts the biotic self-regulation characteristic of natural systems. By identifying thresholds along gradients of management intensity, this work provides quantitative benchmarks for the levels of disturbance beyond which grasslands undergo marked departures from their natural structural organization. These thresholds define the upper bounds of management and fertilization practices that can be applied while maintaining the intrinsic structure and resilience of grassland ecosystems, thereby offering critical guidance for conservation and restoration strategies aimed at preventing widespread grassland degradation.

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Climate change impacts and adaptation in grasslands: Insights from Western Europe

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Abstract

Climate change poses significant challenges for European grasslands, including frequent droughts and heatwaves, warmer and wetter winters, longer growing seasons, and shifting rainfall patterns. This study reviews the impacts of climate change on grass production in dairy cattle farming systems in The Netherlands, Ireland, and France, and explores potential adaptation strategies. Data were synthesized from modelling, field experiments, and farmer surveys. In The Netherlands, modelling results indicate an approx. 9% decline in grass yield for a representative dairy system on dry sandy soils by 2050, along with more than 20% increase in irrigation demand, while on peat soils excessive rainfall limits grazing opportunities. In Ireland, although yield increases of up to 3.4% are projected, summer droughts and spring and autumn waterlogging threaten grazing stability. In France, grass growth starts earlier in spring and extends later into autumn, but severe summer declines and increased inter-annual variability jeopardize feed security. Adaptation options include higher forage reserves, soil quality improvement, diversification of fodder resources, use of drought- and heat-tolerant species in pastures, improved water management (e.g., irrigation and winter water retention), and adjustments in stocking rates. Findings highlight that adaptation must be tailored to local conditions, combining short- and long-term strategies to sustain dairy production and grassland ecosystem services. However, under more adverse climate conditions, adaptation may ultimately require adjusting stocking rates to the carrying capacity of the system.

Keywords: climate change adaptation, dairy farming systems, France, grasslands, Ireland, The Netherlands

Introduction

Climate change is already impacting Europe, with more heatwaves and altered rainfall patterns (IPCC AR6, 2022). In Western Europe, flooding and droughts are expected to intensify. Towards mid-century, droughts across hydrological, agricultural, and ecological systems will threaten fresh water resources, agricultural productivity, and natural ecosystems. Agriculture in Europe faces severe climate risks, with major production losses projected, especially in Southern regions (IPCC AR6, 2022). Key threats include rising water scarcity, and drought and heat stress impacting plants and animals. While adaptation may sustain agriculture at 1.5–2°C of warming, it is unlikely to suffice at 3°C, elevating risks of crop loss and ecosystem degradation.

Grasslands are vital to Europe's agriculture, sustaining livestock in areas unsuitable for crops and covering about 30% of agricultural area (EUROSTAT, 2023). Their adaptability stems from wide

ecological tolerance, wind pollination, flexible C3/C4 pathways, species richness, resilience to grazing, fire, or defoliation, and reproductive versatility (Tomaškinová *et al.*, 2025). These characteristics provide grasslands with a relatively high adaptive capacity to climate change compared with many other agro-ecosystems, although they remain vulnerable to increasing climatic extremes. Permanent grasslands deliver key ecosystem services, including carbon sequestration, water purification, erosion and flood control, biodiversity support, and cultural values, while ley pastures enhance cropping systems through soil conservation, nutrient cycling, water retention, and pest control when well-integrated (Schils *et al.*, 2022; Martin *et al.*, 2020). Grassland conversion undermines these services; yet EU permanent grasslands declined by approx. 30% between 1967–2007, with regional losses up to 50% (Huyghe *et al.*, 2014; Van Den Pol-Van Dasselaar *et al.*, 2008). The EU Common Agricultural Policy limits declines to 5% and protects sensitive areas, a goal increasingly critical under climate change (EU, 2019; Schils *et al.*, 2022). Climate change also threatens grass yield, quality, grazing regimes, and pasture-based system sustainability. Because soil moisture depends on precipitation (Gauly *et al.*, 2013), prolonged droughts increase green water deficits, intensifying reliance on blue water under rising temperatures. While warming may extend growing seasons and enhance CO₂ uptake or crop opportunities (e.g. forage and maize, *Zea mays*), these benefits are often offset by extreme events such as droughts, floods, and heatwaves (EAA, 2019). The potential exposure to water stress due to limited blue water availability under climate change may lower nutrient uptake efficiency, increases fertilizer leaching/runoff, and intensifies nitrate pollution, eutrophication, and soil degradation (IPCC, 2022). Erratic rainfall both stimulates nitrogen mineralization and increases leaching, while reduced pesticide efficiency allows weeds to expand, further lowering nitrogen uptake and degrading yield quality (Gauly *et al.*, 2013).

Climate change impacts extend beyond grasslands to the entire dairy farming system. For example, declining yields and quality may force greater reliance on purchased roughage and concentrate feed, raising costs for dairy farmers (Van Binsbergen *et al.*, 2024; De Haan *et al.*, 2019). While some opportunities may increase (e.g., forage conservation), grazing and field operations such as manure spreading may be constrained by drought, heat stress, or extreme rainfall (Reidsma *et al.*, 2010).

Given the substantial variability in grassland and dairy system responses across Western Europe's agroecological zones, climate-resilient development requires localized understanding of impacts and tailored adaptation strategies to ensure sustainable production. The objective of this study, therefore, is to show how impacts of climate change on grass production in dairy farming systems differ between The Netherlands, Ireland and France, along with an overview of locally suitable adaptation strategies for the three countries. This study focuses on managed permanent and rotational grasslands used in dairy farming systems in Western Europe, and does not explicitly address extensive rangelands or nutrient-poor grasslands in southern or mountainous regions. We provide an overview of country-specific climate change projections, observations on the effects of currently implemented adaptation measures, and recommendations for future periods based on the modelling outcomes. We discuss the adaptation measures and their relevant risks.

Methods and data

Study area

Dairy production systems: Three Western European countries—The Netherlands, Ireland and France μ were selected as case studies due to contrasting grassland systems, climates, and permanent grassland prevalence. Ireland relies almost entirely on pasture-based grazing under a mild, wet climate;

Table 1. General overview of dairy production systems in the study area in the reference year 2024.

Indicator	The Netherlands	Ireland	France
Dairy farms (x1000)	14.3	18.3	44.1
Milking cows (x1000)	1543	1624	3103
Milk production (billion kg)	14.4	8.7	24.3
Share in EU milk production (%)	9.2	5.6	15.5
Milk yield per cow (kg)	9300	5360	7820
Milk production per farm (x1000 kg)	1030	477	542

The Netherlands combines grazing and zero-grazing across diverse soils and management intensities; and France spans varied dairy systems and climates, from wet western to dry southern regions, using both permanent and rotational grasslands. These contrasts enable a comprehensive assessment of sustainability, climate impacts, and adaptation strategies. Table 1 shows a general overview of the dairy production systems in the study area. France, The Netherlands and Ireland contribute 15.5, 9.2 and 5.6%, respectively to Europe's milk production. Dairy farms in The Netherlands show a higher milk yield per cow and a higher milk production per farm.

Dutch dairy farming is characterized by a high milk yield, year-round calving, and 80% of farms using partial pasture grazing in 2024 (CBS, 2025). Much grass is conserved as silage for winter feeding, supplemented with by-products and concentrates. Grassland covers 65% of farmland (962 000 ha), of which 70% is permanent grassland (CBS, 2025). About 40% lay on drought-sensitive sands and 18% on wet peat soils prone to subsidence and CO₂ emissions. Grasslands are predominantly ryegrass-dominated, but clover-based mixtures are increasingly used to improve functional diversity and resilience compared to ryegrass monocultures.

Grasslands dominate Irish agriculture, covering 82% of farmland (CSO, 2021). The dairy sector uses a spring calving system with extended grazing to align peak milk yield with grass growth. Cows graze from February to November, weather permitting, while silage is the main winter forage, supplemented by approx. 1.6 tonnes of concentrate per cow. Average milk yields are 5,500 to 6,000 kg annually. Pastures are mainly perennial ryegrass (*Lolium perenne*), though white clover (*Trifolium repens*) is increasingly promoted to increase functional diversity and reduce fertiliser dependence on derogation farms.

France hosts diverse dairy systems shaped by heterogeneous pedo-climatic conditions, from plains to mountains. Pastures cover 41% of Utilised Agricultural Land, with 69% being permanent grassland (Godoc and Crestey, 2022). This study focuses on plain systems to align with those in Ireland and The Netherlands. Two modelled farms from the INOSYS Réseaux d'élevage illustrate diversity: a mixed system with higher productivity (9000 vs. 7000 l/cow) linked to greater concentrate use (2000 vs. 1400 kg) and forage crop-based feed; and a specialised system relying mainly on grazed grass. Even specialised farms (wheat, barley, sugar beet, etc.) till on 26% of utilized agricultural area (UAA), offering adaptation potential.

Recent changes in key climate indicators: Table 2 compares climate indicators for 1985–1994 and 2015–2024, indicating a shift in climatic conditions across all study areas. Annual precipitation rose (0.1–10.8%), while mean temperatures increased by 0.7–1.5°C and the number of hot days increased by up to 175%. Consequently, ETP–P increased by 31–88%, suggesting increased climatic pressure

Table 2. Average climate indicators for 1985–1994 and 2015–2024

Indicator	France				Ireland		The Netherlands	
	Poitou-Charentes		Normandie		Cork Airport		De Bilt	
	1985–1994	2015–2024	1985–1994	2015–2024	1985–1994	2015–2024	1985–1994	2015–2024
Average annual temperature (°C)	11.3	12.9	10.7	11.8	9.5	10.2	9.8	11.3
Annual cumulate rain (mm)	626	663	709	710	1214	1295	859	952
ETP-P during growing period (April to September) (mm)	−310	−456	−203	−283	102	88	−149	−195
Number of days per year with $T_{\max} > 25^{\circ}\text{C}$	53	74	19	29	0.4	1.1	23	32

and variability during the growing season, particularly in drier regions. Notably, climatic pressure is not limited to increasing water deficits; in wetter regions, excess rainfall and reduced trafficability can be equally constraining for grass utilization and field operations.

National climate projections: Climate projections in this study are based on national assessments, reflecting country-specific impact and adaptation planning. For cross-country comparison, harmonized indicators are reported relative to a common reference period (1985–1994). By mid-century (approx. 2050), national projections indicate mean annual temperature increases of approx. 0.9–1.6°C in The Netherlands, 1–1.6°C in Ireland, and up to 4°C in France under a 3°C global warming pathway. Annual precipitation changes are modest (e.g. −2% to 3% in The Netherlands), but rising evapotranspiration increases seasonal P–ETP across all countries. Despite similar magnitudes of change, impacts diverge due to contrasting baseline climates, with drought-prone regions experiencing amplified water stress and wetter regions remaining vulnerable to waterlogging and variability.

Methods

This paper provides an overview of results from selected studies on climate change impacts and adaptation strategies for grasslands in The Netherlands, Ireland, and France. The studies were chosen as illustrative case studies rather than to provide a comprehensive or systematic review. The evidence presented is based on modelling approaches, field experiments, farmer surveys, or combinations of these methods.

Results

Impacts of climate change on grasslands and dairy farming

In the following section, we describe how observed impacts differ among The Netherlands, Ireland, and France. We focus on both ends of the hydrological spectrum: more frequent drought/heat stress as well as periods of excessive rainfall and waterlogging.

Observed impacts: Observed impacts reflect both ends of the hydrological spectrum—more frequent drought/heat stress as well as periods of excessive rainfall and waterlogging—depending on local conditions.

Prolonged droughts and heat: In The Netherlands, climate change has increased weather variability, with four dry years (2018–2020, 2022) and two wet years (2023–2024). Drought lowered grass and maize yields on sandy soils by 23% and 14%, respectively (Van Binsbergen *et al.*, 2024). Irrigation mitigated drought impacts, and its use rose from 34% of farms in 2017 to 64% in 2020, mostly

relying on groundwater. Crude protein levels of grass were higher in 2018 (192 g/kg DM), which was mainly due to the timing of the drought, which onset after the first cut and caused a relatively high share of the first cut in the total annual production. Droughts also damaged swards, requiring additional tillage and sometimes herbicides, while complicating the 80/20 grassland–maize ratio under derogation. Droughts also increased soil nitrogen surplus and phosphorus, with N-surplus 56% higher in dry years than in 2021, which raised nitrate in surface and groundwater, and the phosphorus balance changing from negative (depletion) to positive (surplus). Field trials confirmed that irrigation reduces N surplus and potentially nitrate leaching by enhancing uptake (Hoving *et al.*, 2024), and supporting yields.

Irish grassland farms have faced successive extreme weather, testing their resilience (Wingler *et al.*, 2024). Fodder crises occurred in 2012–2013 and 2017–2018, when poor growing seasons, harsh winters, and summer droughts reduced grass growth and increased reliance on imported feed. In April 2018, the government intervened to supply fodder. Since then, repeated dry summers and excessive rainfall, including the 2023 drought and wet 2024, have further limited grass growth (1.2t DM/ha in 2024), delayed grazing, and strained feed supplies. Droughts are common in France, with 14 episodes recorded during 1975–2006 (Lemaire and Pimlin, 2007). Drought frequency and intensity have increased, with at least one drought season each year since 2020 (except 2024).

Excessive rainfall: In The Netherlands, climate change impacts on grasslands strongly depend on soil type, with drought mainly affecting sandy soils, while excess wetness and waterlogging are more prominent on peat soils. A study on rewetted peat meadow showed reduced grass yields and grazing days due to oxygen stress in roots (Pijlman *et al.*, in prep). Climate change may accelerate peat oxidation, making rewetting necessary to prevent degradation, greenhouse gas emissions, and subsidence. However, rewetting combined with changing precipitation patterns may increase wetness risks on peat soils.

In Ireland, despite relatively positive rain–ETP balances, excess rainfall and poor trafficability can pose greater practical challenges for grassland management than drought. From July 2023 to March 2024, precipitation was 30–60% above average, leaving soils saturated through spring and delaying turnout and grazing. Cooler-than-normal conditions in June–July further limited growth, with cumulative herbage production by early July 2024 about 1.2 t DM ha⁻¹ lower than recent years, increasing pressure on feed supplies. In France, increasing drought coincides with excess-water risks. In 2024, wet, warm springs delayed grazing and harvesting, reducing fodder quality, with similar conditions in southern France in 2025 highlighting growing spring climate variability.

Prolonged growth season: A study of Dutch dairy farms showed many farmers experienced prolonged growing seasons in the past years, with various benefits, such as more harvests, earlier spring grazing, longer autumn grazing, higher forage yields, and additional autumn forage for barn feeding (Eckelder and Hilhorst, 2021). These developments offer opportunities for adaptation through more flexible grazing management and increased forage conservation, if supported by appropriate soil and nutrient management.

In Ireland, however, such trends are not yet evident. Slower temperature increases limit the effects of earlier springs or later autumns on grass yields. Additionally, recent nitrogen fertiliser regulations, including delayed spring applications, postpone the growing season. These management constraints

may mask or counteract any climate-driven changes, preventing clear observation of extended growing periods in Irish grasslands.

Predicted impacts: Grass growing period, yield and quality. Several studies have attempted to predict the impact of climate change on grasslands. In The Netherlands, a modelling study for a Dutch dairy farm on sandy soils projected about 9% decline in grass yield and 18% decline in maize yield by 2050 compared to average yields in 2000–2019, mainly due to green water deficit (Hoving *et al.*, data not shown). The larger projected yield decline for maize compared to grass is mainly related to its shorter growing period and stronger dependence on summer conditions, which limits its ability to benefit from wetter spring and autumn periods, rather than to differences in photosynthetic pathway alone. Additionally, grassland and maize irrigation demand is expected to rise by more than 20 and 60%, respectively.

For Irish grasslands, biophysical growth models simulated significant increase in grass production everywhere, with yields ranging from 10 to 16.8 t/ha (Shrestha *et al.*, 2014). More recent application of mechanistic models supports yield increase projections while emphasize on major seasonal shifts and greater inter-annual variability (Streethran *et al.*, 2024; Ruelle, 2022). Grass growth is projected to increase in spring (April) and autumn (October–November) but decline in summer (July–August) due to higher drought risk. While higher spring and autumn rainfall may enhance growth, poor drainage can restrict grazing and silage production (Ruelle, 2022). Increasing year-to-year variability and extreme events—droughts, irregular rainfall, and prolonged wet periods—raise the risk of fodder crises, as seen in 2012–2013 and 2018 (Streethran *et al.*, 2024). Consequently, Irish grasslands face three key challenges: reduced summer growth and fodder insecurity, poor utilization under wetter winter–spring conditions, and greater interannual variability increasing multi-year feed risks.

French grassland simulations indicate earlier spring growth and extended autumn–winter production, but reduced or stalled summer growth with increasing interannual variability. Regions typically productive in summer may require additional fodder in dry years, while already drought-prone areas face heightened risks of species mortality under intensified drought and heat. Earlier and more vigorous spring growth may allow earlier grazing and fodder accumulation (Debaeke and Durand, 2025), but rapid growth and excessive rainfall can complicate management, delaying grazing or harvest and reducing herbage quality. Extended late-season growth may enable additional winter grazing, although poor soil bearing capacity may limit its use in France.

Examples of climate adaptation

The IPCC defines climate change adaptation as adjustments in natural or human systems to reduce harm or exploit opportunities. On dairy farms, adaptation lowers vulnerability, mitigates impacts, and enables new opportunities (De Vries *et al.*, 2023). Adaptation can be reactive, incremental, or transformative. Short-term measures include irrigation, drainage, grazing adjustments, drought-tolerant species, and multispecies grasslands, while long-term strategies may involve shifts from pasture-based systems to mixed feeding or zero-grazing to enhance resilience under climate change. Sudmeyer *et al.* (2016) identify nine adaptation categories for dairy farms under climate change, including: housing and animal health; feed quantity and quality; crop and grazing strategies; precision agriculture; feed conservation; feeding management; water quantity and quality; soil erosion control; and risk management. These measures span improvements in infrastructure, forage and water management,

soil protection, and system diversification. The following sections illustrate these adaptation strategies using case-study examples.

Grassland and forage management: To produce enough forages. In The Netherlands, irrigation is commonly and increasingly used to sustain yields and has been shown to be the most effective strategy to mitigate effects of droughts on grass yields (De Vries and Van Riel, 2024). At experimental farm De Marke in The Netherlands (2050 scenario), a field trial applied 90,628 m³ of surface water over 14.5 ha (14.6 mm/day, 626 mm total) to test groundwater recharge in early 2022. Winter water retention raised grass yields by 7.5% via enhanced spring growth, while maize benefited less due to delayed growth. Spring irrigation of grassland proved more efficient than season-long irrigation (25 vs. 18 kg DM/mm). This likely reflects more favorable temperature and soil moisture conditions in spring, while in summer higher temperatures, evaporative demand, and insufficient irrigation reduced water-use efficiency. About 74% of farmers indicated that irrigation is important for grassland production; 65% supported the option of irrigating silage maize, while 55% reported that, under limited water availability, maize would be prioritized over grass. In Haaksbergen, sub-irrigating maize with wastewater (approx. 4 mm/day) via the KAD system prevented drought stress, reaching 2.5 m into the root zone with minimal pharmaceutical residues (Bartholomeus *et al.*, 2016). Beyond irrigation, Dutch dairy farms use other water management measures (De Vries *et al.*, 2023): drainage on 36% of farms (mainly to remove excess water), farm dams (21%), bales in culverts (13%), and adjustments to beams, sluice gates, or pump-driven ditches to control water levels.

Current and future restrictions, however, will require more efficient use of water, and thus alternatives such as drought-tolerant grasses, grass-herb mixtures, winter rye, or improved water retention. A modelling study showed that deeper-rooted herb-rich grasslands increased yield by 1% but reduced irrigation demand by 28% on rich loamy sand and 5% on poor loamy sand under 2050 climate (Hoving *et al.*, data not shown). A recent field trial confirmed higher dry matter yields from grass-herb mixtures than from pure perennial ryegrass, with better performance of mixtures with cocksfoot (*Dactylis glomerata*) or tall fescue (*Festuca arundinacea*) under no irrigation in a dry year than under drip irrigation (Van Schooten, 2025). Cocksfoot and red clover (*Trifolium pratense*) proved particularly competitive against ryegrass, plantain (*Plantago lanceolata*) and chicory (*Cichorium intybus*).

Forage maize is considered as a viable option to offset production losses in Dutch grasslands. Studies have shown higher yields under warmer temperatures (Wolf *et al.*, 2010), less yield reduction on sandy soils, and greater drought tolerance outside sensitive growth periods. However, maize is more sensitive to waterlogging and ozone exposure, and benefits less from elevated CO₂ (Mills *et al.*, 2018). It also faces rising mycotoxin risks, faster weed growth, and shifting pests and diseases, while some diseases decline in warmer, drier summers. Diversification helps but remains climate-sensitive (De Vries *et al.*, 2023).

In Ireland, reduced summer growth and higher July–September variability threaten forage supply and winter feed security, increasing the need for larger, higher-quality silage reserves, potentially for lactating rather than dry animals. Projected autumn growth (October–November) may partly offset silage demand if weather allows. A key adaptation strategy is sward diversification and genetic improvement; multispecies swards consistently outperform monocultures under stress, with higher resilience, yield, and drought recovery (Finn *et al.*, 2013; Grange *et al.*, 2021). However, gaps remain regarding tolerance to winter waterlogging, recovery under poor grazing, and long-term persistency, highlighting

the need for multi-site, long-term field experiments under typical grazing management (Jezequel *et al.*, 2024).

In France, a common adaptation is increasing conserved forage in livestock diets. Expanding maize silage can buffer summer deficits but increases reliance on irrigation and protein supplements. Alternative strategies include harvesting immature cereal–protein mixtures in late spring, using fast-growing cover crops (e.g. Italian ryegrass, forage rape) for autumn forage, and adopting single-cut sorghum (*Sorghum bicolor*), a drought- and heat-tolerant maize substitute with comparable nutritive value in BMR varieties (Emile *et al.*, 2008; 2016; Blot and Tourneux, 2020). Grass-based systems also require species diversification, favoring tall fescue or *Poa pratensis*, legumes such as red clover and alfalfa (*Medicago sativa*), and forbs like chicory or plantain in multispecies swards (Grange *et al.*, 2021; Lüscher *et al.*, 2022). As growth shifts earlier in spring and later in autumn, flexible grazing—supported by on/off grazing when soil bearing capacity is limited—becomes essential. Overall, forage diversification and adaptive grazing are key to sustaining productivity under increasing climatic variability.

The creation of new water reserves often faces administrative constraints and increasing societal tensions. Irrigation is most commonly used for field crops: in 2020, only 11% of irrigated land was used for fodder production (grasslands and maize silage), according to the French agricultural census (Scotti and Loubier, 2024). During years of drought, existing reserves are frequently depleted early in the year, and withdrawals from surface water or groundwater are generally subject to restrictions by local authorities. For example, at the end of August 2025, three-quarters of the national territory was subject to restrictions on water withdrawals for irrigation, including 17% under total prohibition, 15% under 50% restriction and 24% under 30% restriction.

Soil and nutrient management: In Netherlands, farmers identified key nutrient management adaptations to cope with climate change, including increasing soil organic matter (98%) and preventing soil compaction (97%), with many already applying practices like early manure application or avoiding slurry on dry soils. Adding clay improves soil structure for organic matter buildup, while manure storage enables timely application, preventing soil compaction. Less common measures included adding clay to sandy soils (57%) or expanding manure storage. About 60% highlighted the need for regulatory changes to allow more flexible grassland-arable ratios. Reducing chemical fertilizer use, improving manure quality, and expanding alternatives to synthetic fertilizers were also emphasized as important adaptation strategies.

Herd management: To adapt or reduce the animal demand. In The Netherlands, measures against cow heat stress focus on shade (avoiding direct sunlight), barn adaptations (stimulating heat loss), and management changes (reducing heat production) (De Vries *et al.*, 2023). For shade, farmers prefer housing cows in barns with free-range access; night grazing is another option. For barn modifications, farmers favour roof insulation and fans, while side walls are usually open for ventilation. Fresh drinking water is a given. They accept some milk yield loss in hot periods, noting that lowering concentrate feeding helps recovery. More frequent feeding of smaller fresh rations reduces fermentation and acidosis risks, which can also be prevented with rumen buffers.

In France, herd-management adaptations include allocating lower-quality fodder to young stock to enable compensatory growth in spring or autumn (Pottier *et al.*, 2007), and reducing feed demand

during shortages through early weaning, drying-off, or culling. In grazing systems, dual calving seasons (March/April and September/October) better align feed demand with grass availability, lower stocking rates, spread milk deliveries, and reduce heifer nutritional requirements, enhancing system resilience under variable forage and climate conditions (Delaby and Fiorelli, 2014; Delaby *et al.*, 2020). Ultimately, if climate change increases forage shortages, farms should adapt stocking rates to local agro-climatic conditions by expanding fodder areas or reducing herd size to maintain self-sufficiency.

In Ireland, while larger fodder reserves and diverse swards increase system flexibility, high stocking rates or major shifts in grass growth can still cause problems. Research shows optimal stocking depends on local soil and weather: heavy soils grow more but limit grazing days, while free-draining soils face drought shortages. Smart stocking should focus on grass utilization rather than total growth, with heavy soils requiring lower rates despite higher yield (Ruelle *et al.*, 2022).

Other options: In The Netherlands, a farmer review identified approx. 60 climate adaptation measures across water, soil, pasture, crop, fodder, feeding, and housing management. Most measures (78%) are already implemented on dairy farms, while about one-third—mainly related to soil, pasture, and fodder conservation—are considered important for future climates. Promising but not yet widely adopted options include enhanced water retention (e.g. sub-irrigation, controlled drainage), new forage crops, and heat-stress mitigation through grazing adjustments and barn cooling. The most promising strategies include crop diversification and rotation, increased conserved feed stocks, soil cover and reduced tillage, use of byproducts, nutrient-loss reduction, barn and feeding-time optimization, genetic selection for resilience, and a focus on managing climate variability rather than long-term change.

In Ireland, adaptation measures include increasing fodder and slurry storage, using multispecies swards (limited by persistency), and on/off grazing to manage poor trafficability. Decision-support tools such as PastureBase Ireland (PBI), used by over 6,000 mainly dairy farmers, support grazing and fodder management by tracking farm cover, growth, and paddock performance, and visualizing supply-demand through the grass wedge (Hanrahan *et al.*, 2017). PBI is evolving into a climate-adaptation tool by integrating models (e.g. MoSt Grass Growth) to predict feed shortages or surpluses and support winter fodder budgeting, enhancing resilience to increasing climatic variability.

In France, diverse climates, soils, and livestock systems offer multiple short- and long-term adaptation levers that must be coordinated at farm level to avoid trade-offs and limit environmental impacts. A holistic example is the OasYs system experiment at INRAE Lusignan, launched in 2013 in a drought-prone region (Novak *et al.*, 2025). The system integrates multispecies grasslands, sorghum, forage beet (*Beta vulgaris*), trees, shrubs, and cover crops, combined with dual calving, extended lactations, and crossbreeding to improve forage use and heat tolerance. Together, these strategies demonstrate strong agroecological adaptation potential across environmental, economic, and social dimensions (Novak *et al.*, 2022).

Conclusions

Alongside reviewing literature and projects, we assessed how climate change affects dairy systems in The Netherlands, Ireland, and France, and how these countries adapt. Climate change impacts grasslands differently. In The Netherlands, rising temperatures and recurring droughts reduce grass yields on sandy soils and raise irrigation needs, while excessive rainfall on peatlands lowers forage quality and grazing days. In Ireland, high year-to-year rainfall variability and summer droughts stress pasture

systems, which increases silage reliance. In France, longer spring and autumn growing seasons extend grazing, but summer droughts and heat stress threaten species and grassland sustainability. Overall, benefits from longer growing periods might be offset by extreme events, making outcomes context-dependent. At the same time, prolonged growing seasons may offer adaptation opportunities, such as more flexible grazing calendars and increased forage conservation, provided that soil bearing capacity and nutrient management constraints are adequately addressed. Adaptation strategies reflect local systems. Dutch measures include water management, multi-species swards, adjusted grazing regimes, precision irrigation, and maize silage. In Ireland, precise grazing management, growth prediction tools, and silage reserves are keys. In France, conserved forages, forage diversification, dual-purpose crops, and adapting stocking rates or calving seasons strengthen autonomy. However, adaptation measures might have trade-offs with dairy production. For instance, maintaining permanent grasslands supports carbon storage, soil protection, and feed security, but may conflict with milk productivity. Hence, farmers must balance ecosystem services with production through diverse, flexible adaptation to sustain resilience across Western European grasslands. While management adaptations can mitigate climate impacts, stocking rates must ultimately follow the carrying capacity of local agro-climatic conditions, and herd size reductions may be unavoidable. According to the results, it could be concluded that climate change creates major challenges for dairy farming across The Netherlands, Ireland, and France. Adaptation strategies need to be tailored to local conditions and management systems, balancing milk production with ecosystem services to sustain resilience. Future research would benefit from harmonized climate scenarios, baseline periods and common quantitative indicators across regions, combined with greater integration of socio-economic dimensions such as farmer decision-making and economic feasibility, enabling more robust cross-country comparisons and supporting broader assessments, such as identifying the most relevant grass-based dairy production systems based on future climate conditions.

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The value of long-term experiments in agriculture: grassland resilience on the North Wyke Farm Platform, a case study

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Abstract

Long-term agricultural experiments are essential for understanding system-level interactions among soil, plants and livestock, and their management. This understanding allows to disentangle the factors driving grassland resilience in the context of food production and climate change; two grand challenges in the 21st century. The North Wyke Farm Platform (NWFP), a UK National Bioscience Research Infrastructure (NBRI) established in 2011 in south-west England, provides a unique, fully instrumented facility to study the sustainability of grassland-based livestock and arable production. It comprises three 21-ha farmlets where hydrology, nutrient and sediment fluxes, greenhouse gas emissions, productivity, and biodiversity are continuously monitored. The NWFP has undergone a series of controlled management transitions, from wholly permanent pasture to high-sugar or deep-rooting grasses, grass-legume mixtures, and more latterly to arable cropping, allowing the long-term consequences of land-use and management change to be quantified. The research from the NWFP demonstrates that well-managed grasslands can act as carbon sinks and has informed metrics for environmental and economic sustainability, contributing to national greenhouse-gas inventories and mitigation strategies. Overall, the NWFP illustrates the irreplaceable value of long-term, field to farm-scale research platforms in evaluating whole-system responses to climate and management pressures, and in guiding the transition of livestock systems towards net-zero.

Keywords: crops, livestock, metrics, sustainability

Introduction

Grasslands occupy approximately 25% of the Earth's ice-free land surface (Steinfeld, 2006). They are used predominantly for forage and livestock production and are considered to contribute significantly to carbon (C) sequestration (Staerfl *et al.*, 2012) and increased biodiversity (Jerome *et al.*, 2014) particularly in the presence of grazing livestock.

The Utilised Agricultural Area (UAA), as of 2022, was 41.619 million acres (16.843 million hectares), approximately 69% of the land area of the UK (AUK, 2024). Grasslands can also be a source of N and C losses to water from leaching or runoff and to the atmosphere via pollutants such as ammonia (NH₃) or nitric oxide (NO), which is also involved in ozone generation in rural locations (Mosier, 2001) as well as the greenhouse gases (GHGs) carbon dioxide (CO₂), nitrous oxide (N₂O) and methane (CH₄) (IPCC, 2001). Thus, while grasslands may sequester carbon, they can simultaneously contribute to other emissions and losses, meaning net environmental outcomes depend on local conditions and management.

Grasslands contribute to around 50% of total UK N₂O emissions comprised by emissions from mineral fertilizer and manures application, excreta deposited to soils and manure management. Livestock contribute to 56% of total UK Agriculture emissions as CH₄ due to enteric fermentation and manure management.

However, generally and in the long-term, grasslands can sequester C due to improved sward and livestock management, with occasional reseeded increasing biodiversity and promoting the return of organic matter due to excreta deposition.

Field and farm-scale experiments are useful because they provide a more representative platform for full and detailed systems level studies including biogeochemical nutrient cycling studies, encompassing soils, plants, livestock and associated management (Cardenas *et al.*, 2016). When these are of long-term nature and hence considered more a monitoring platform, the data collected allows disentangling the key factors causing change in system outcomes. Long-term monitoring presents, for example, opportunities to explore the effects of natural (climate) or forced (management) change in agricultural systems that can assist knowledge procurement and tools to help farmers and land managers build resilience. Where data collection is required for different compartments of the agroecosystem, such collections demand large investments but provide robust platforms for investigating process interactions over the longer term. The North Wyke Farm Platform (NWFP), a UK National Bioscience Research Infrastructure (NBRI) is a leading example of such platform. It is located in the southwest of England (50°46'10"N, 3°54'5"W), a region typically devoted to grazing livestock systems. The soil (Harrod and Hogan, 2008) is mainly comprised of two similar soil series: Hallsworth (Dystric Gleysol) and Halstow (Gleyic Cambisol) (Avery, 1980) with about 36% clay in the top-soil. Three 21-ha farmlets were established in 2011 (see Figure 1) to obtain baseline data on hydrology, nutrient cycling and productivity for 2 years. The farmlets had not been reseeded and had been under grassland for at least 20 years; permanent pasture (PP). After 2013, two farmlets were modified by either (i) scheduled ploughing and reseeded with grasses that were: bred for enhanced sugar content or deep-rooting traits (improved pasture, IP), or; (ii) ploughing and sowing of grass and legume mixtures to reduce nitrogen fertiliser inputs (grass/white clover mixed pasture, GWC) (Orr *et al.*, 2016). The third farmlet was left as PP. Later in 2019, the high sugar pasture was ploughed and converted to arable cropping. Beef cattle and sheep graze the pastures (PP, IP, GWC) in the spring and summer, but are housed in the winter since the soils are prone to seasonal waterlogging. Measurements of productivity, losses of sediments and nutrients to water, and of losses of nutrients to the atmosphere are carried out as described in previous studies (McAuliffe *et al.*, 2018a) (Orr *et al.*, 2016), (Cardenas *et al.*, 2022). To date, the NWFP has collected over 100 million data points since 2011: including sensor and field/laboratory records spanning hydrology, sediment and nutrient fluxes, atmospheric emissions, and management/livestock data, allowing system-level outcomes and trade-offs to be quantified over time, including legacy effects and responses to climate and management variability that underpin grassland resilience.

Against the above context, this paper summarises some of the key findings from the NWFP, based on the 14 years of data collection since 2011 in exploring the sustainability of livestock (and more latterly arable) systems and to illustrate the value of these long-term platforms in assessing the co-benefits and trade-offs of modern agriculture. Building resilience for grassland involves an optimal combination of soil properties/climate, plant species and livestock.

System management and assessment of impacts

Whole system (farm) best management is essential to optimize productivity, minimizing losses of nutrients and therefore income, whilst maintaining a high standard of welfare, both for livestock and farm workers themselves. Sustainable intensification aims to increase productivity from a given land area, whilst minimizing the impact on the environment and relies on careful consideration of Best Management Practices (BMP) (Peukert *et al.*, 2014).

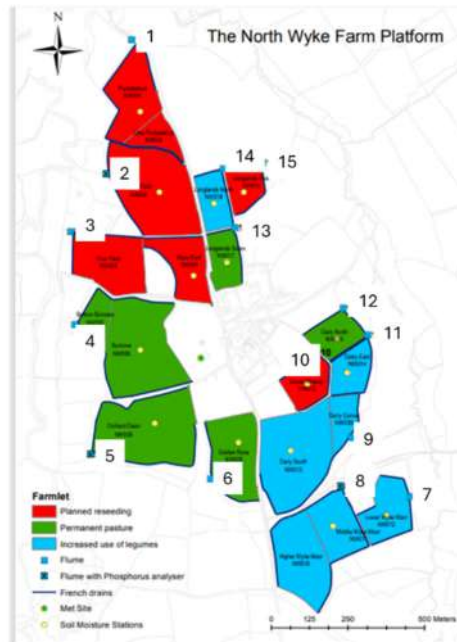


Figure 1. Map of the NWFP. Fields 4, 5, 6, 10 are the permanent pasture (PP) farmlot, fields 1, 2, 3, 15 were originally permanent pasture until 2013 when they were converted to improved pasture (IP) until 2019, and then converted to arable; blue fields 7, 8, 9, 11, 14 originally permanent pasture until 2013 when they were converted to a grass/legume mixed sward (GWC). The permanent pasture had not been ploughed before 2013 for at least 20 years.

We next discuss the risks to the environment due to agricultural practises when BMP are not applied. We include soil, sward and livestock management in the discussion linking directly to the resilience and sustainability of agriculture and particularly grassland production.

Regarding sediment loss from agricultural fields, a recent examination of the wettest winters (December to February, inclusive) in the NWFP dataset compared measured sediment (particulate) exports with background sediment loss rates estimated from paleolimnological evidence for the UK (Collins *et al.*, 2025). Paleolimnological reconstructions use dated lake sediment layers to infer past catchment sediment delivery, providing a baseline against which present-day exports can be evaluated. This work revealed that in circumstances where scheduled ploughing and sward reseedling preceded the winters in questions, measured sediment loss exceeded modern background rates by up to 2.4 times. In comparison, sediment loss from the recently introduced arable treatment on the NWFP exceeded modern background rates by up to 21.7 times.

Regarding atmospheric losses, grasslands can be a sink of CO₂ fluxes, as shown in e.g. a study from 14 European grasslands (Hortnagl *et al.*, 2018), resulting in values between -1783 to -91 g CO₂ m⁻² year⁻¹. The three pastures from the NWFP were monitored using Eddy Covariance towers. The annual values were -540 and 620 g CO₂ m⁻² year⁻¹ in 2017 and 2018, respectively (Cardenas *et al.*, 2022), demonstrating their potential to be a net source. In addition, an event such as land conversion from grassland to arable, can result in major C losses. The latter study on the NWFP demonstrated that overall, the 3 pastures acted as sinks of C, but more consistently on the permanent grassland that had not been ploughed for over 20 years. The other pastures that had been scheduled ploughed and

reseeded 8–10 years previously, showed more variability from year to year. Further, the conversion to arable caused a 280% decrease in total CO₂ uptake compared to the high sugar grass pasture (Guo, personal communication).

Optimizing genetics and nutrient balance both in pasture and livestock will improve nutrient use efficiency (Cardenas *et al.*, 2019; Carswell *et al.*, 2019) and animal performance and the last two studies demonstrated this using data from the NWFP (McAuliffe *et al.*, 2018b). Various metrics have been suggested as a way to define and monitor pathways to achieving productivity, welfare, soil nutrients and emissions to both air and water. Poore and Nemecek (Poore and Nemecek, 2018) used e.g. land use (for crops, yield represents output for a single harvest); freshwater withdrawals weighted by local water scarcity; and GHG, acidifying, and eutrophying emissions as critical metrics for sustainability. They addressed the issue of variability in certain practices, including conservation agriculture, organic farming, and integrated systems of best practice by disaggregating each environmental indicator into its sources or drivers. Their mitigation framework rests on data being validated against known ranges for each system outcome (e.g., maximum yields achievable given specific inputs). Takahashi *et al.* (2018) (Takahashi *et al.*, 2018) highlighted the value of sustainability metrics in agriculture and used data from the NWFP to develop some illustrative examples. These ranged from (i) environmental and ecological indicators including soil organic carbon (SOC) and its heterogeneity, sediment export, greenhouse gas emissions and botanical diversity; (ii) animal health indicators; (iii) nutrient provision; (iv) managerial decisions, to (v) economic performance. The study used a data-based approach to assess economic and environmental trade-offs relevant to pasture-based cattle and sheep production systems. They found that there appears to be a systematic interaction between ‘soil health’, ecological surroundings and livestock grazing, so that a higher level of soil organic carbon (SOC) stock is associated with better animal performance and less nutrient losses to watercourses, and a higher stocking density with greater botanical diversity and elevated SOC.

Sward management

Sward management influences grassland resilience by determining the capacity of the soil–plant system to maintain productivity while regulating carbon storage, nutrient cycling, and losses of sediment and nitrogen under variable climate and grazing pressures. Well managed grazed grasslands function as C sinks (Chang *et al.*, 2021; Bai and Cotrufo, 2022). This stored C is lost if there is cultivation via the conversion to arable use through ploughing (Orr *et al.*, 2016), causing accelerated soil degradation and loss of nutrients and sediment with concomitant loss of fertility. Nutrient losses also result in pollution of watercourses via eutrophication. There are opportunities for managing pastures for benefits to soil, e.g., deep rooting grasses, that can increase C sequestration into the soil by placing it in deeper layers (Gill and Burke, 2002) or high sugar grasses that have the potential to increase soil C. The latter can also be beneficial to grazing animals, by improving digestibility, increasing nutrient uptake and reducing animal-related losses. Mixed swards also provide multiple benefits by providing a variety of nutrients as well as giving the animal choices when grazing. The inclusion of legumes brings atmospheric nitrogen to the soil, decreasing the reliance on mineral fertilisers. However, it has also been shown that in the long-term, legumes might be detrimental to C stocks (Herridge, 2016).

Swards need to match the N requirements and release of energy in the rumen (Lee *et al.*, 2001). In the South-West of England permanent grassland (not ploughed and reseeded for at least 5 years) dominated by *Lolium perenne* is commonly used for grazing livestock. Recent improvements in plant genetics offer opportunities for reducing environmental impacts whilst retaining production (Ellis *et*

al., 2011), via improved properties such as deep rooting, nutritional quality (digestibility, water-soluble carbohydrate content), or by using a grass/clover sward, managing soil drainage, nutrient retention and generally better nutrient cycling (Ellis *et al.*, 2011; Orr *et al.*, 2016).

Pastures species with a high water soluble carbohydrate (sugar) content have been found to be useful in improving N use efficiency (Lee *et al.*, 2002) and decreasing enteric CH₄ emissions (Rivero *et al.*, 2020) whilst deep rooting pastures can potentially also help sequester C. The introduction of legumes provides N saving via avoidance of fertiliser application, and can also increase the potential for C sequestration due to improved root biomass from a beneficial complementarity between plant species (Rutledge *et al.*, 2017). However, the inclusion of legumes can also increase N₂O emissions, due to the decomposition of plant residues and rhizodeposition (Fuchs *et al.*, 2018).

Livestock management

Considerable debate exists regarding the impact of livestock on climate, as well as on air and water pollution. This is not only due to the animal-related direct and indirect emissions of GHGs and ammonia, but also due to the need to use arable land to produce feed for animals. A series of measures have been listed to ensure livestock sustainability, based on decreasing the amount of food suitable for human consumption fed to animals, using breeds appropriate for the location, maintaining healthy animals, using supplements to benefit the animal in addition to increasing productivity, consuming good quality food, not large quantities, keeping livestock management according to cultural practices, and consideration of other benefits such as biodiversity and adopting best practices. The latter could be interpreted as for example: 'growing the right crop in the right place'.

Livestock management influences grassland resilience because decisions on grazing and housing determine how effectively pasture-based systems function under local soil and climate constraints, with consequences for productivity, soil condition, and environmental losses. Animal management is influenced by geography, particularly the soil and the climate. In temperate zones, livestock and particularly cattle, are housed in the winter due to unsuitable ground conditions for grazing on heavy soils prone to seasonal waterlogging and due to limited grass availability. Full year housing (feedlot systems as for example in the US) can rely on feeding human-edible crops (e.g., maize, cereals and soy), which can reduce food-system efficiency. Grazing is an efficient way to convert pasture into protein for human consumption. Livestock are also recycler of nutrients, by depositing the fraction of the feed which is not retained in the form of available nutrients and organic matter within their excreta. This will maintain or increase soil fertility, especially in those cases where repeated scheduled cultivation for sward improvement can make the soil poor in nutrients. However, contaminants such as heavy metals are also present in livestock diets at background concentrations due to their use in diet supplements and presence in drinking water (Petersen *et al.*, 2007). Some of these contaminants are regulated for example, in Europe (Commission, 2003).

Grazing livestock can produce pervasive elevated soil compaction in detriment of yield due to a decrease in root length (Unger and Kaspar, 1994), but mild compaction may increase yield (Voorhees *et al.*, 1986). We found in grazing paddocks at the North Wyke campus, neighbouring the NWFP (Fajardo *et al.*, 2025), that soil recovers during the winter when animals are in housing, suggesting that best grazing management can help unwarranted soil damage, not only via rest periods, but also by controlling the stocking rate and annual grazing strategy. Equally, best grazing management can help avoid unintended consequences related to elevated erosion and sediment loss (Pulley *et al.*, 2022).

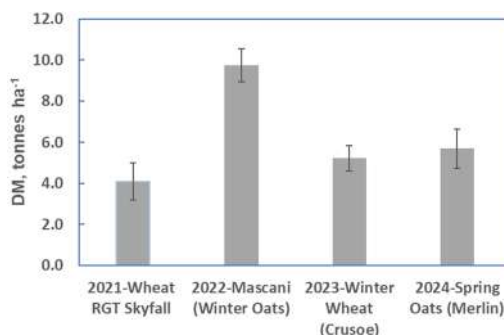


Figure 2. Dry matter yields of cereals cultivated on one of the farmlets of the NWFP, previously a high sugar grass (IP) that was ploughed in 2019 and converted to arable cropping.

Using data from the NWFP, it was suggested that the role of animal variability on emissions heterogeneity is a critical factor to consider, indicating that lower performance animals (high emitters) should be identified (McAuliffe *et al.*, 2018a). This can support targeted management and/or selection decisions, provided rankings are stable through time. We have recently made an assessment based on several years of data from the NWFP. We analysed data from five cohorts of animals monitored at the NWFP between 2019 and 2023. Within these 443 animals, ranked as high, middle, or low CH₄ emitters, 33% changed their position in the ranking in February relative to their cohort in January, although only 7% of those shifts were from high to low or vice versa. By March, around 40% of animals had changed their position in the ranking, but only 5% showed a major shift compared with their January classification. This further supports the concept of long-term monitoring, to be able to capture variability through time, and to disentangle the causes of change, whether management or climate or their interactions.

Heavy soils are not considered suitable for growing arable crops, but they are for growing grass and therefore support livestock. This adds to the argument of the value and impact of diets for humans as mentioned earlier. However, we consider it is not only related to the comparison between a good grassland supporting livestock and a good arable crop cultivated in a lighter optimal soil. It is also about changing the purpose of the land, in this case converting a grassland that thrives in the right soil/climate type into cropland. Takahashi (Takahashi *et al.*, 2018) indicated that for a full comparison of nutritional provision capacity from the same area of land, wheat yield on the same NWFP soil should also be taken into consideration (Takahashi *et al.*, 2018). This had not been measured at the time of the latter publication, but as one of the farmlets on the NWFP was converted to arable cropping in 2019, we are now able to make this comparison. The resulting yields of the cereals cultivated on the NWFP between 2021 and 2024 was suboptimal compared to typical yields in the UK. Figure 2 shows values between 4 and 5 tonnes ha⁻¹ for 3 of the years.

In the case of wheat, values are much lower than the 10 tonnes ha⁻¹ that can be achieved. Winter oats in 2022 produced 9.7 tonnes ha⁻¹ what seems to have been an outlier, possibly helped by weather, but further analysis is needed to understand the reasons for this outcome. The quality of the crops (Figure 3) was also generally lower in our NWFP crops compared to optimal values. For wheat moisture must be above 15% and CP more than 13%.

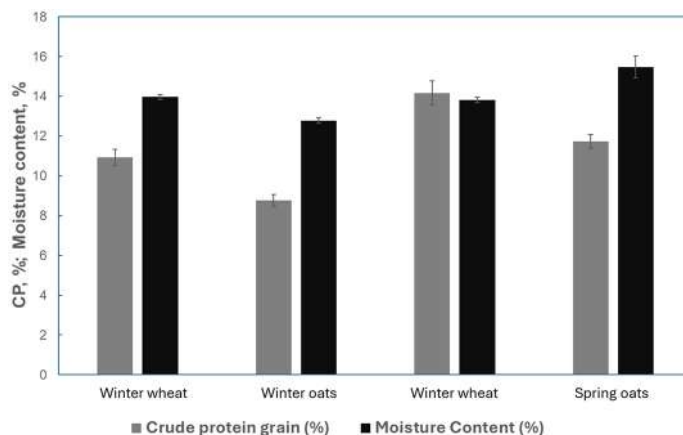


Figure 3. Quality parameters for cereals cultivated on one of the farmlets of the NWFP, previously a high sugar grass (IP) that was ploughed in 2019 and converted to arable cropping.

Soil management and legacy effects

Agricultural management influences soil physics, levels of nutrients and microbial populations. Peukert *et al.* (2016) discussed the role of short- vs long-term management in the interpretation of resulting properties and that a baseline understanding of the management is required to be able to assess the effect of change.

They also discuss that averaging soil properties does not fully describe these impacts and that the resulting spatial variation *between* and *within* agricultural fields needs to be quantified so that any change in either the overall mean or in the spatial distribution of soil properties can be attributed to changes in management. Their study indicated that agricultural fields with different management histories (more than 6 years), but similar recent land use, cannot be assumed to be similar in their mean soil properties. Therefore, long-term management history is helpful when interpreting soil data at field scale. They found that the permanent grassland (these are considered at least 20 years old) did not reduce diffuse pollution and it is possible that long-term effects of management would take decades for water quality to improve. In another study, located near the NWFP, it was found from measurements of greenhouse gas emissions that the effect of a decrease in management intensity on emissions of N_2O from arable rotation and grassland UK soils was not detectable in the first 10 years of the land use change (Horrocks *et al.*, 2015). Intensive management can have a long-lasting legacy on soil properties and function, as shown in a forest ecosystem (Compton and Boone, 2000) and steppe grassland (Mosier *et al.*, 1997) concluding that the soil N cycle was still influenced by former management after 50 and 8 years of agricultural abandonment, respectively.

Ploughing grasslands releases large amounts of carbon, both to watercourses and to the atmosphere. The conversion from grassland to arable on the NWFP released considerable amounts within a few months of the plough event as mentioned earlier (Guo, *pers. comm.*).

Grassland soils also support a variety of insects, from direct pests of grass (e.g., Tipulidae) or nearby arable crops (e.g., wireworms), also providing benefit to natural enemies which can help to suppress pest populations to non-damaging levels, as the study by Benefer *et al* on the NWFP shows (Benefer *et al.*, 2016).

Regenerative practises

Regenerative farming is a movement embracing various BMPs targeting six principles: farming context, minimizing disturbance, maximizing crop diversity, keeping soils covered, maintaining a living root and integrating livestock. Recent estimates of the potential for mitigating global warming using ‘nature climate solutions’ (NCS) needed to avoid warming to $<2^{\circ}\text{C}$, state that up to 25% of mitigation can be achieved from grassland and agricultural measures (Griscom *et al.*, 2017); also called conservation agriculture practises (CA). These include optimising grazing intensity and the inclusion of legumes. In the UK, it is estimated that the maximum mitigation potential for NCS is 1.31 and 8.53 Tg $\text{CO}_2\text{e year}^{-1}$ from optimal grazing intensity and the inclusion of legumes, respectively. This represents about 20% of emissions in 2019 (Brown *et al.*, 2021) from agriculture in CO_2e . Other suggested measures include zero or minimum tillage, the retention of organic residues, or other means to build soil organic carbon (SOC), the diversification of cropping systems through rotations or inter-cropping and the introduction of silvopasture. Of these, the inclusion of legumes (white clover) in the sward has been tested on the NWFP which provides savings in fertilizer applications as shown in the study by Cardenas *et al.* where the PP received 509 kg N-1 in 2017 whilst the HSC received 60 kg N, as well as, the retention of winter stubbles in the arable treatment, returning C and providing some soil cover out of cropping season. In the case of the latter, the recent work by (Brown *et al.*, 2021) illustrated how the inclusion of over-winter stubbles on the arable treatment on the NWFP during winter 2023–2024 ensured that recorded sediment export did not exceed modern background rates in two of the five arable catchments. Here, the fact that over-winter stubbles did not prevent exceedance of modern background rates in three of the arable catchments points to the need for better over-winter cover. The new agri-environment instrument in England (Environmental Land Management—Sustainable Farming Incentive) incentivizes farmers to sow a multi-species cover crop for improved soil cover for regenerative farming and to improve over-winter soil cover compared with stubbles.

Concluding remarks

Grasslands provide multiple benefits, and this potential can be maximized if management is optimized. Using metrics of sustainability can be useful, but these must be robust in their representation of system level outcomes. Here, short term measurements can generate biased data, and although they are useful for exploring mechanistic interactions, longer term datasets are needed to get a robust picture of critical interactions and system outcomes, particularly given changing drivers related to climate and management.

Further strategic investment is needed to support long-term monitoring and to maintain platforms that enable testing new strategies and technologies for agriculture. As more data is collected, new applications such as AI will increase our predictive capacity in the context of change.

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Forage legumes enhance durum wheat yield and weed suppression in Mediterranean cropping systems

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Abstract

Forage legume intercropping represents a sustainable agroecological strategy that enhances soil fertility, reduces reliance on chemical inputs, and improves overall crop system performance. This study evaluated the effects of intercropping two forage legumes—*Sulla coronaria* L. and *Medicago polymorpha* L.—with durum wheat (*Triticum turgidum* subsp. *durum*) under organic farming conditions in a Mediterranean area. Both scarified and unscarified seeds were tested to assess their impact on wheat productivity, weed suppression, and forage quality. Intercropping significantly increased wheat grain yield and reduced weed biomass compared to monoculture, particularly in combinations with *S. coronaria*. A negative linear relationship was found between legume biomass and weed pressure. Additionally, *S. coronaria* produced higher protein content, while *M. polymorpha* had higher fibrous fractions. These findings demonstrate that intercropping with forage legumes, especially *S. coronaria*, can enhance the productivity and ecological resilience of organic wheat systems, even under dry growing conditions.

Keywords: durum wheat, intercropping, *Medicago polymorpha*, *Sulla coronaria*, weed suppression

Introduction

Wheat-legume intercropping is the simultaneous cultivation of two or more species in the same space that offers an advantageous solution compared to monoculture. This agroecological practice enhances soil health by increasing ecosystem biodiversity through synergistic interactions between species and it exploits the legumes' nitrogen-fixing capacity, making it available for cash crops, reducing chemical fertilizer use and increasing soil fertility (von Cossel et al., 2025). At the same time, weed control is achieved and higher grain protein content and yield are produced (Scordia et al., 2024). In this regard, the study evaluated the effects of the introduction of legume forage species in the Mediterranean wheat system to develop a sustainable agri-food chain by improving environmental performance. The aim of this study was to evaluate the potential of two forage legumes, *Sulla coronaria* and *Medicago polymorpha*, intercropped with durum wheat, to improve wheat yield, suppress weeds, and enhance forage quality under Mediterranean conditions.

Materials and methods

The experiment was carried out in Patti (Messina, Italy, 38°11'N, 14°99'E; 196 m a.s.l.) at an organic farm in 2023–2024. The experimental site is characterized by a Mediterranean climate with mild-wet winters and hot-dry summers. Over the twenty years, meteorological data were recorded by the SIAS weather station in Patti (Servizio Informativo Agrometeorologico Siciliano, www.sias.regione.sicilia.it). The 20-year mean values were: 21.4°C, 11.4°C, 16.4°C maximum, minimum, and mean air temperatures; 565.8 mm rainfall; and 676.7 mm evapotranspiration. Specifically, the 2023/24 growing season experienced temperatures very similar to the 20-year average, with average maximum, minimum and mean air temperatures of 21.2°C, 11.3°C and 16.3°C. Rainfall and

evapotranspiration in the season were lower than the 20-year reference (294.2 mm and 481.3 mm, respectively). The growing season was dry. The soil was characterized by the following physical and chemical properties: 58.1% clay, 12.0% silt, and 29.9% sand; pH of 7.8; organic matter content of 2.23 g kg^{-1} (Walkley and Black method); total CaCO_3 of 2.9 g kg^{-1} (Scheibler method); total nitrogen of 0.15 g kg^{-1} (Kjeldahl method); available P of 80.25 mg kg^{-1} (Olsen method); and available K of 209.0 mg kg^{-1} (ammonium acetate test method). The experimental factors investigated were: (i) forage legume species (*Sulla coronaria* L. (local ecotype), *Medicago polymorpha* L. var. Anglona), intercropped (IC) with durum wheat (*Triticum turgidum* subsp. durum (Desf.) Husn, var. Core); (ii) two levels of scarification treatment (S), i.e. mechanical scarified and unscarified seeds, along with an absolute control of a wheat sole crop [control (C)] according to a randomized block design replicated three times. The seedbed was prepared using conventional tillage practices, consisting of a summer ploughing and an autumn disc harrowing at 30 cm and 20 cm depth, respectively. The sowing was carried out manually in December 2023 at the highest seed density for each species, followed by soil rolling. Harvest was carried out at the wheat grain hardening and complete dryness and death of the plant (BBCH 99) on 12 Jun 2024. Wheat, legumes and weeds were collected in 0.25 m^2 quadrats at harvest. Legume and weed samples were dried to constant weight in a forced air oven at 65°C to determine the dry matter content. Durum wheat grain and straw were separated by a mechanical thresher. Grain yield, legume biomass, and weed dry biomass were derived and expressed per unit land area (Mg ha^{-1}). Legume protein and fibrous fractions (NDF, ADF, ADL) were determined using Near-Infrared Spectroscopy (NIRS DS2500, Foss Analytical, Italy) at harvest (June 2024). Data were subjected to analysis of variance (ANOVA) to determine significant differences among the experimental factors (forage legumes and scarified seed treatment) and their possible interactions on wheat yield, weed control, protein content and fibrous fractions of forage legumes. Post-hoc comparisons among different treatments were achieved using Tukey's HSD test ($p \leq 0.05$).

Results and discussion

The ANOVA revealed significant effects of legume IC on wheat grain yield and weed biomass, whereas the effects of scarification treatment were not significant. The first-order interaction was significant for wheat grain yield and weed dry biomass.

Intercropping with forage legume species did not result in significantly increased durum wheat grain yield compared to the wheat monoculture, except the IC with *S. coronaria* unscarified (4.49 Mg ha^{-1}) which showed the highest result (Fig. 1a). These findings are in agreement with those obtained by Scordia et al. (2024).

There were also significant differences in weed dry biomass among IC treatments compared to the control. The IC with *S. coronaria* unscarified (0.69 Mg ha^{-1}) was the most effective in reducing weed pressure compared with the IC with both *M. polymorpha* scarified and unscarified (1.02 and 1.00 Mg ha^{-1} , respectively); the control was the worst (with 1.30 Mg ha^{-1} weed biomass) (Fig. 1b).

The linear relationship between weed dry biomass and legume biomass showed a negative trend (Figure 2a). For each additional dry tonne of legume biomass, weed biomass decreased by 0.66 Mg ha^{-1} . Moreover, when yields of wheat and legume were combined weed biomass decreased by 0.54 Mg ha^{-1} (Figure 2b).

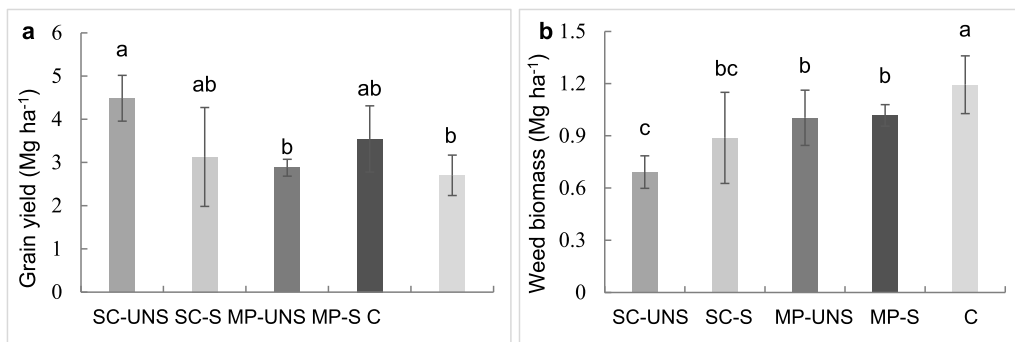


Figure 1. Means $\pm$ standard errors of durum wheat yield (a) and weed biomass (b) for main effects of the interaction forage legumes $\times$ scarification treatment (*S. coronaria* unscarificated (SC-UNS), *S. coronaria* scarificated (SC-S), *M. polymorpha* unscarificated (MP-UNS), *M. polymorpha* scarificated (MP-S) and control (C)). Different letters indicate significantly different means ($p \leq 0.05$).

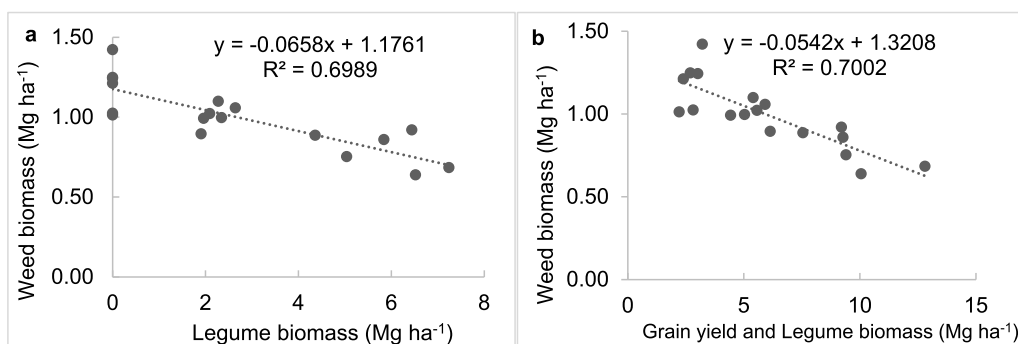


Figure 2. Linear relationship between (a) legume dry matter biomass and weed dry biomass; (b) cumulated wheat grain yield and legume dry matter biomass and weed dry biomass. Significance at $p \leq 0.05$.

Additionally, protein content and fibrous fractions (NDF, ADF, ADL) of forage legumes were significantly different between IC species (except for ADF), but were not affected by scarification treatment. Protein content was significantly higher for *S. coronaria* than *M. polymorpha* (14.82% and 9.11%, respectively). Conversely, NDF and ADF showed higher values for *M. polymorpha* (63.79% and 45.17%, respectively) than *S. coronaria* (45.94% and 40.35%, respectively). The ADL was higher for *S. coronaria* than *M. polymorpha* (14.93% and 9.44%, respectively).

Conclusions

The findings demonstrated the potential of IC to enhance weed control in organic cereal cropping systems. However, IC with *S. coronaria* achieved a good performance in both durum wheat grain yield and weed reduction compared to IC with *M. polymorpha* and wheat sole crop. The benefits of IC were evident even during a dry growing season that compromised wheat development in many areas across Sicily. This effectiveness could be attributed to higher nitrogen supply provided by legumes and improved weed control compared to monoculture. Wheat-legume IC improved forage nutritional quality by increasing protein content and modifying fibre fractions. Specifically, IC with *S. coronaria* showed the most favourable nutritional profile (14.82% protein, lower NDF and ADF). This study demonstrates that agroecological practices such as IC could represent an important strategy towards the resilience and sustainability of cereal cropping systems.

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Nitrogen import trends on commercial pasture-based farms incorporating legumes

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Abstract

The Clover150 program, established in 2020, monitors farm gate nitrogen (N) balances on 30 commercial grassland farms across Ireland. Using an annual farm area target of 10% reseeding and 15% over-sowing, farms incrementally increased area previously containing perennial ryegrass (*Lolium perenne*; PRG) monoculture swards to areas containing PRG/white clover (*Trifolium repens*; WC) swards from 45% in 2021 to 81% by 2024. Within paddocks, yearly average sward WC content increased from 12% in 2021 to 19% in 2023. Mean farm gate N balance (N imports—N exports) varied between year; from 170 kg N ha⁻¹ in 2021 to 128 kg N ha⁻¹ in 2022. Fertiliser N accounted for the largest N imports (75%). As a percentage of total N imports, total feed N imports increased ($P < 0.01$) as fertiliser N imports decreased ($P < 0.01$). Reduced N balance was due to reduced fertiliser N imports. To date, the Clover150 programme has demonstrated successful WC incorporation and reductions in farmgate N balance on commercial farms.

Keywords: concentrates, fertiliser, forage, nutrient balance, white clover

Introduction

Achieving European food sovereignty amid geopolitical uncertainties and reducing nutrient loss to the environment are key objects of the European union's 'a vision for agriculture and food' (EU; European Commission, 2025). Attaining food sovereignty comes with a significant challenge of doing so in an environmentally benign manner. Nutrient loss, particularly nitrogen (N) to surrounding ecosystems, can have detrimental effects due to the eutrophic nature of N (Murphy *et al.*, 2024). Estimating potential N loss on commercial farms can be a challenge but metrics such as farm gate N balance can be a useful indicator (Schröder *et al.*, 2003). Farm gate N balance is highly correlated ($r^2 = 0.82$) with N leaching loss (Murphy *et al.*, 2024), thus, reducing farm N balance can help mitigate the N proportion vulnerable to leaching. Previous studies (Treacy *et al.*, 2008) on commercial farms have identified white clover (*Trifolium repens*; WC) incorporation as a strategy to reduce farm N balance. In mixed swards, WC has been shown to fix up to 124 kg N ha⁻¹ in Irish climatic conditions (Burchill *et al.*, 2014), while also demonstrating the potential to alleviate N inputs to swards containing WC relative to perennial ryegrass (*Lolium perenne*; PRG) monocultures (Egan *et al.*, 2018). To achieve goals outlined by the EU, farms must reduce N balance while maintaining farm forage production. On this basis an on-farm study was commenced with the objectives to: (i) maintain dry matter (DM) production ≥ 14 t DM ha⁻¹; (ii) reduce N balance < 130 kg N ha⁻¹; (iii) reduce N fertiliser to ≤ 150 kg N ha⁻¹ year⁻¹; (iv) achieve average sward WC content in grasslands of 20–25%.

Materials and methods

In 2020, an on-farm study was launched by Teagasc Moorepark, the 'Clover150' programme. Now in its 5th year of a 6-year programme, the group was set up to examine WC incorporation on commercial

farms. A total of 30 farmers are involved in the project across Ireland, from a range of farming systems. In spring of 2021, all farms were provided with a detailed plan for WC establishment and post-sowing grazing and fertiliser management. Farms have an annual farm area target of 10% reseeded and 15% over-sowing. For newly established paddocks (reseeded or over-sowing), a pre-grazing herbage mass of $<1100 \text{ kg DM ha}^{-1}$ was advised for up to 4 months, with reduced N fertiliser levels to reduce competition from PRG, as well as maintaining a lower pre-grazing herbage mass on these paddocks over the autumn and winter period. Sward WC content was assessed visually using the 'Teagasc Clover score card' (Teagasc, 2024) three times per year (spring, summer and autumn) by a trained technician; farmers were then provided with tailored management guidelines for their farm. For paddocks with spring WC content of $\geq 20\%$, N fertiliser was reduced from May onwards. All farm management (grazing events, DM production, N fertiliser etc.) was recorded by each individual farmer and uploaded to PastureBase Ireland (Hanrahan *et al.*, 2017). Farmgate N balance was calculated at the end of each production year using PastureBase Ireland as per Buckley *et al.* (2016); total N imports—total N exports, divided by total utilisable agricultural area. Imports account for N in the form of fertiliser, concentrate and forage feed, livestock and slurry. Outputs account for milk, forage, and livestock sales, as well as slurry exports. Total feed-N accounts for all concentrate and forage feed imports. All data were analysed in SAS 9.4 (SAS Institute, Cary, NC, USA). The model included year as a fixed effect, with farm included as a random effect.

Results and discussion

Reduced farmgate N-balance can help mitigate potential N loss to the environment (Murphy *et al.*, 2024), and ultimately achieve EU nutrient loss reduction targets. Over the last 4 years of this six-year programme, N balance varied ($P < 0.01$; Table 1). Average N balance in 2021 and 2024 was 22% higher than in 2022 and 2023. Total N input followed the same trend as N balance: 15% higher in 2021 and 2024 compared to 2022 and 2023. Similar to Buckley *et al.* (2016), fertiliser N dominated N inputs accounting for, on average, 75% of N input over the 4 years. Dominating fertiliser N imports reflect the importance of low cost forage production, which underpins the economic resilience of pasture-based systems (Dillon *et al.*, 2008). Fertiliser N as a percentage of total N imports reduced by 9% on average from 2021 to all other years; in the same period, total feed N as a percentage of total N imports increased by 8% on average. The recorded shift in N import trends towards a greater dependence on feed N is a cause for concern as it deviates from an emphasis on homegrown feedstuffs production and minimal feed imports (O'Donovan *et al.*, 2021). Increased reliance on feed N coincides with an 11% reduction in herbage production. Despite the marked increase in farm area containing WC, achieving $>20\%$ sward WC content year $^{-1}$ is a continuing challenge. Burchill *et al.* (2014) reported that the sward WC content level is related to sward N fixation ($r^2 = 0.86$). Balancing fertiliser N input with sward WC content is a challenge that farmers must adapt to, in order to achieve herbage production targets. In 2024 there was a return to the N balance recorded in 2021, despite the farm area and sward WC content increasing. Average 2024 herbage production recorded in Ireland was $1200 \text{ kg DM ha}^{-1}$ lower than the previous 10-year average (O'Donovan and Egan, 2025); thus, the dominating influence of climatic conditions may have had an overriding influence on results obtained.

Conclusion

The ongoing clover 150 programme has demonstrated successful WC incorporation and farmgate N balance reductions on commercial farms. Continued work is required to balance fertiliser N input and sward WC content to achieve herbage production targets. Monitoring N import trends in pursuit of reduced N balance is important to maintain resilience of pasture-based systems.

Table 1. Percentage (%) farm area containing white clover (WC) area, sward WC content (%), herbage production (kg DM ha⁻¹) total N imports (kg ha⁻¹), fertiliser and feed imports as % of total N imports, and farm gate N balance (kg ha⁻¹).

Year	WC Area	Sward WC Content	Herbage production	Total N inputs	N inputs (%)		N balance
					Fertiliser N	Total Feed N	
2021	45 ^c	12 ^c	14512 ^a	250 ^a	81.5 ^a	17.5 ^b	170 ^a
2022	64 ^b	15 ^b	12971 ^b	212 ^b	71.8 ^b	26.2 ^a	128 ^b
2023	68 ^b	19 ^a	12812 ^b	210 ^b	73.4 ^b	25.2 ^a	134 ^b
2024	81 ^a	18 ^a	13108 ^b	246 ^a	73.2 ^b	25.9 ^a	167 ^a
SE	2.1	1.7	298	9.1	1.79	1.75	4.9
P value	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01

Superscripts (a, b, c) denote differing means ($P < 0.05$) within variables.

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Peat pasture productivity with raised groundwater tables

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Abstract

Dutch peatlands have long been drained to be used primarily as pastures for dairy production, improving productivity but causing high greenhouse gas (GHG) emissions and soil subsidence due to peat degradation. Raising the groundwater table (WT) from the conventional 50 cm below field level to 20 cm using an active water infiltration system (AWIS) is a possible solution to reduce peat degradation. This paper highlights the effect of raising WT on grass production and soil nitrogen supply (SNS) by using experimental plots with different WT and fertilisation levels from 2020–2024. Regression analyses resulted in an average decrease of 11% in herbage dry matter yield (DMY) when raising WT from 50 to 20 cm below field level, with significant variation between years. The reduced DMY could be explained by the decrease in SNS in wetter conditions, due to a lower mineralisation rate. Increased N fertilisation could mediate DMY losses, but N losses from soil would increase. Although results were obtained at plot level, and exclude losses from harvesting and grazing, they indicate that raising WT is compatible with relatively high grass production.

Keywords: active water infiltration systems (AWIS), grass yield, dairy, nitrogen, peat soil

Introduction

In The Netherlands, peat soils are predominantly used as permanent pasture for dairy production. A long history of drainage of peat soils has resulted in productive pastures, with a high soil N supply (SNS) due to oxidation of the peat layer and historic additions of manure and slurry (Pijlman *et al.*, 2020). Drainage of peat soils, however, also leads to high greenhouse gas (GHG) emissions and soil subsidence (Poppe *et al.*, 2021). In the Climate Agreement, the Dutch government aims to reduce GHG emissions from peat soils by 1 million tons of CO₂ equivalents and reduce soil subsidence by 50% by 2030. The most prominent solution to reach this aim is to rewet previously drained peat soils, by increasing the groundwater table. Raising groundwater tables can be achieved by making use of an active water infiltration system (AWIS), which consist of pressurised submerged drains that can regulate the groundwater table throughout the year (Hoving *et al.*, 2021). Raising groundwater tables affects dairy farm management, as the wetter conditions alter soil conditions. In particular, raising groundwater tables can reduce the mineralisation rate of the peat layer, resulting in less available nitrogen for the grass to take up, leading to reduced grass production and quality (de Vos *et al.*, 2010). Here, we explore how raising groundwater tables affects herbage dry matter yield and SNS.

Materials and methods

Data were collected at the experimental farm KTC Zegveld from 2020–2024. The experimental farm was equipped with two separate farming systems, a high-water system (HW, target groundwater table

of 20 cm below field level) and a low-water system (LW, target groundwater table of 50 cm below field level) (van Dijk *et al.*, 2026). Each system was equipped with experimental plots (HW, $n = 9$, LW $n = 6$), which were divided into three strips (2.5×10 m). Each strip was treated with a fertilisation rate of 0, 125 or 250 kg N ha⁻¹ year⁻¹ in the form of calcium ammonium nitrate (CAN). In the HW system, the target groundwater table of 20 cm below field level was regulated with the AWIS, while in the LW system, no AWIS was present. Groundwater tables were measured in every plot every two weeks, and averaged over the growing season. The average measured groundwater table was significantly different between HW and LW (average of 27 and 45 cm below field level for HW and LW respectively). The three strips per plot were harvested in 4–5 cuts per year, and the herbage dry matter yield (DMY, kg ha⁻¹ year⁻¹) was determined. SNS (kg N ha⁻¹ year⁻¹) was calculated as the N yield without fertilisation. The main effects and interactions of groundwater table (in cm below field level), N fertiliser and year (2020–2024) were estimated with a linear mixed model using R studio.

Results and discussion

Both DMY and SNS were significantly ($P < 0.001$) affected by the main effects of groundwater table and year. For DMY, this resulted in an average decrease of 44 kg DM ha⁻¹ year⁻¹ per cm increase in groundwater table, which is shown in Figure 1. DMY differed per year, with an average increase of 967 kg DM ha⁻¹ year⁻¹ (range 208–2036) compared to 2020. The predicted reduction in DMY when increasing the groundwater table from –50 cm to –20 cm was on average 1.4 Mg DM ha⁻¹, or an 11% decrease. In addition, the significant main effect of fertilisation ($P < 0.001$, not shown in Figure 1) resulted in an increase of 10 kg DM ha⁻¹ year⁻¹ per kg of fertiliser N applied, without being affected by year or groundwater table (average of 10.3, 11.7 and 12.9 Mg DM ha⁻¹ year⁻¹ with 0, 125 and 250 kg N ha⁻¹ year⁻¹ respectively). N fertilisation could therefore be used to compensate the losses in yield, but increased fertilisation results in further N losses to the environment, which could counteract the climate gains from raising groundwater tables (van der Laan *et al.*, 2024).

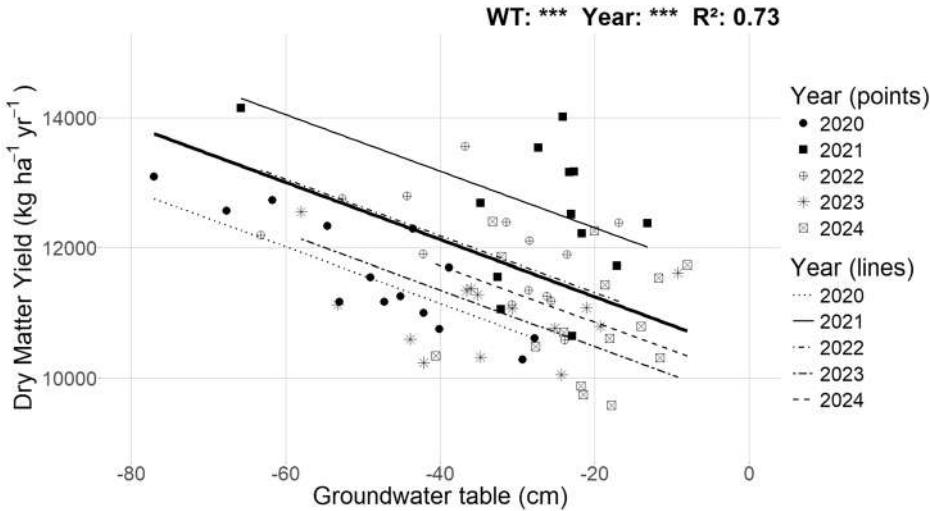


Figure 1. Dry Matter Yield (DMY, in kg ha⁻¹ year⁻¹) fertilised with 125 kg N ha⁻¹ year⁻¹, across the average measured groundwater tables (WT) per plot in 2020–2024. The thick black line indicates the average effect over the 5 years.

SNS decreased on average by $1.2 \text{ kg N ha}^{-1} \text{ year}^{-1}$ per cm increase in groundwater table. SNS differed per year due to varying weather conditions, with an average of an additional $16 \text{ kg N ha}^{-1} \text{ year}^{-1}$ (range -14 to 45) compared to 2020. When moving to the target HW groundwater table, SNS reduced on average by 13% compared to the target LW groundwater table (average of 245 and $282 \text{ kg N ha}^{-1} \text{ year}^{-1}$ for HW and LW, respectively). Decreased SNS with higher groundwater tables can explain the lower DMY, since the wetter conditions can lower the mineralisation rate (Pijlman *et al.*, 2020).

Our findings are at plot level, and show the potential obtainable productivity with higher groundwater tables. This excludes losses that occur during grazing, trampling by cows or trafficking by machinery. Furthermore, the lower load bearing capacity in wetter conditions can result in delayed timing of harvesting and fertilisation (Janssen *et al.*, 2023). These additional losses can impede realizable grass yield and quality, which in turn affects milk production and farm economy. Nonetheless, our findings show that average predicted DMY with higher groundwater tables can still be considered productive ($11 \text{ Mg DM ha}^{-1} \text{ year}^{-1}$ with $125 \text{ kg N ha}^{-1} \text{ year}^{-1}$). Since raising groundwater tables is an important practice to achieve the goals set in the Climate Agreement, farmers should be encouraged to implement the practice on their farms. However, the associated losses should be compensated to incentivise farmers to transition towards farming with higher groundwater tables. Alternatively, new business models could be designed that value farms not only for milk production, but for peat conservation as well.

Conclusions

In conclusion, DMY and SNS were both affected by groundwater table and varied during the study years. The observed reduction in DMY with higher groundwater tables could partly be explained by the reduced SNS due to lower mineralisation rates in wetter conditions. While the predicted reductions with increased groundwater tables were on plot level, which exclude potential losses on farm level, they nonetheless show that pasture productivity can be maintained. To encourage raising groundwater tables on dairy farms, the consequences of higher groundwater tables on farm level should be further explored.

Acknowledgements

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Impact of grassland overseeding on optimization of carbon sequestration in mineral and organic soils

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Abstract

Due to changes in animal feeding systems in many European countries, a significant portion of grasslands are subject to degradation due to abandonment or reduced use for feed production, which has resulted in a change in their floristic composition. At the same time, maintaining environmental functions of grasslands, particularly soil protection, is also crucial. In this context, a study was conducted on such grasslands to assess soil organic carbon (SOC) in mineral soils (2 sites) and soil organic matter (SOM) in organic soils (2 sites) following overseeding using two types of seed mixtures sown in three different dates: spring, late summer, and autumn. The seed mixtures included grasses and legumes with or without addition of extra herbs species. The experiment comprised also a control treatment without overseeding. Higher SOC in mineral soils and higher SOM in organic soils were recorded in overseeded sites compared to the control. The effect of sowing date was observed only in mineral soils, with the highest SOC content found in sites overseeded in spring. Precise overseeding is recommended for degraded grasslands, as it can effectively improve both SOM and SOC content.

Keywords: degraded grasslands, grasses, legumes, soil organic carbon, soil organic matter

Introduction

In recent years, increasing attention has been paid to carbon sequestration in grassland soils, including drained peatlands and degraded or semi-arid grasslands, where vegetation plays a key role (Deru *et al.*, 2023; Tian *et al.*, 2016). Besides their environmental function, grasslands must ensure adequate species composition to provide forage for ruminants. In cases of low vegetation cover and a lack of high-quality forage species, overseeding is commonly used to improve forage quality and quantity, while also increasing soil organic carbon (SOC) (Bondaruk *et al.*, 2020). Some studies show that overseeding can increase biomass yield by 50% and maintain stable organic carbon levels in the topsoil (Kohler *et al.*, 2020). Legumes are a key component of overseeding (Bondaruk *et al.*, 2020), and their combination with perennial grasses and nitrogen fertilization further enhances SOC sequestration (Poepplau, 2021; Conant *et al.*, 2017). Carbon sequestration in soils is positively correlated with annual precipitation, soil moisture, species richness, and grass proportion (Tian *et al.*, 2016). Few studies focus on peat grasslands, where practices like grazing, mowing, and fertilization significantly affect soil organic matter (SOM). The selection of overseeded species also influences SOM dynamics—greater diversity and the inclusion of legumes can enhance nutrient availability and SOM storage (Rumpel *et al.*, 2015). However, the impact of different overseeding mixtures and dates on soil parameters in peatlands remains understudied. This study evaluated how overseeding of degraded grasslands with two seed mixtures in different seasons affects carbon sequestration in mineral and organic soils.

Materials and methods

The research was conducted in eastern Poland in the Lublin and Podlaskie Voivodeships on four farms on meadows with mineral (2 sites) and organic soil (2 sites). In 2021, oversowing was carried out on

each site on three dates: I (spring), II (end of summer) and III (autumn) using an innovative prototype of a seeder equipped with, among others, a coulter, a harrow, or a scarifier. Two seed mixtures were used at each date: M1 (grasses 70%, legumes 30%); M2 (grasses 69%, legumes 29%, herbs 2%), and M0—no-oversowing (i.e., control). The seed mixture included grasses: *Dactylis glomerata*, *Festuca arundinacea*, *Festuca pratensis*, *Lolium perenne*, and *Phleum pratense*; legumes: *Trifolium pratense* and *Trifolium hybridum*; and herbs: *Matricaria chamomilla*, *Taraxacum officinale* and *Plantago lanceolata*. The control treatment consisted of an unsown meadow located in the same habitat conditions. At each site, randomization of treatments and the same experimental design with four replicates was used. The meadows had previously been regularly mowed, but during the experiment, they were mowed three times and fertilized at 120 kg N ha⁻¹, 31 kg P ha⁻¹ and 60 kg K ha⁻¹ per year. Each plot area was 500 m². In the second year after oversowing, in the autumn of 2022, soil samples were collected from a depth of 0–30 cm using a soil auger to assess environmental effects, i.e., organic carbon content in mineral soil (SOC) and organic matter in organic soil (SOM). The results are given as an average of 2 sites. The obtained results were statistically analysed using two-way ANOVA with Tukey's HSD post-hoc test.

Results and discussion

In mineral grassland soils SOC is carbon bound to minerals, while in organic soils SOM is the main soil mass of accumulated plant residues. Studies on mineral soil showed that spring overseeding resulted in significantly ($p < 0.05$) higher SOC content (25.7 g kg⁻¹) compared to summer and autumn (both 23.1 g kg⁻¹). SOC was also significantly higher in plots with M1 (27.5 g kg⁻¹) and M2 (27.0 g kg⁻¹) mixtures compared to the control (Fig. 1), with no difference between the mixtures, indicating no effect of herbs (Bondaruk *et al.*, 2020). The mixtures included grasses and legumes species, which can enhance nitrogen fixation and SOC (Poeplau, 2021; Conant *et al.*, 2017). Overseeding improved species composition in degraded meadows. Other studies also confirm that species richness and grass content promote higher SOC (Tian *et al.*, 2016), as well as favourable precipitation and soil moisture.

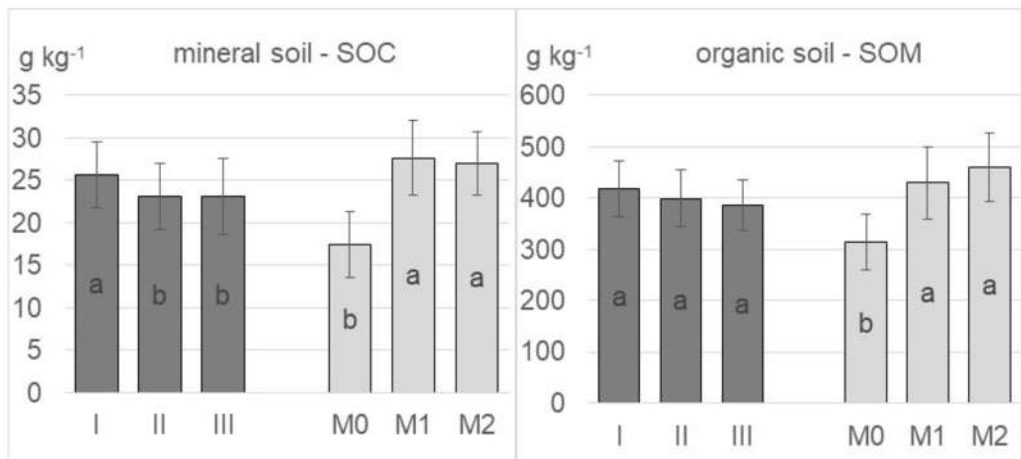


Figure 1. The content of soil organic carbon (SOC) and soil organic matter (SOM) depending on the oversowing date and the mixture used. Oversowing date: I (spring), II (summer), II (autumn); Seed mixtures: M0 (control), M1, M2 (see Methods); error bars represent $\pm$ SD; different letters indicate significant differences ($p < 0.05$)

In organic soils, SOM content did not significantly differ between sowing dates, though the highest value was recorded in spring (417.7 g kg^{-1}). Overseeding with M1 (429.4 g kg^{-1}) and M2 (459.6 g kg^{-1}) significantly increased SOM compared to the control. Other studies confirm that high species diversity and legume use enhance nutrient availability and SOM storage (Rumpel *et al.*, 2015). In summary, the overseeding of degraded grasslands with high-quality forage grasses and legume species improves SOC and SOM, particularly when combined with fertilization (Conant *et al.*, 2017; Bondaruk *et al.*, 2020; Poeplau, 2021), adequate rainfall, and soil moisture (Tian *et al.*, 2016). Spring sowing showed the highest values, though longer growing time may have influenced the results. Because these results presented here are based only on a one-year period, they should be considered as trends rather than definitive conclusions.

Conclusion

Higher SOC in mineral soil and higher SOM in organic soil were observed in overseeded treatments compared with the control. No significant differences were found between treatments overseeded with the two different mixtures. The effect of overseeding date was observed only in mineral soils, with significantly highest SOC content recorded when overseeding occurred in spring. However, it should be noted that in these meadows, the overseeded plants had more time to grow and develop. Precise overseeding is a recommended practice on degraded grasslands, as it can improve SOM and SOC levels. This improvement can be further supported by greater plant cover, as well as other factors not considered in this study.

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Comparison of biostimulants and fertilisation programmes in relation to Bermudagrass performance during autumn and spring season

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Abstract

Bermudagrass (*Cynodon* spp.) is a warm season turfgrass species commonly used in areas subjected to drought and high temperatures. This study investigates the effect of fertilisation and biostimulants on three bermudagrass cultivars (Sultan, MBG002, Arden 15) over a two-year period. Conducted at the University of Padua's experimental farm, three fertilisation programmes were compared: Programme 1 (urea alone, supplying 125–150 kg N ha⁻¹ year⁻¹), Programme 2 (urea+high-dose Hicure), and Programme 3 (urea+low-dose Hicure). Hicure is a peptide- and amino-acid-based biostimulant (non-microbial) applied at 90–180 l ha⁻¹ (high dose) or 45–90 l ha⁻¹ (low dose). Evaluations included turf quality, colour, NDVI, spring green-up, autumn dormancy, and stolon carbohydrate concentrations. Statistical analyses revealed that turf quality and colour were significantly influenced by treatment ($p < 0.001$), with Programme 2 consistently producing the highest scores. Cultivar-specific differences were evident for NDVI and carbohydrate accumulation, with Sultan and MBG002 outperforming Arden 15. These improvements are consistent with reported mechanisms by which amino acids enhance nitrogen assimilation, chlorophyll synthesis, and stress tolerance in warm-season turf species. The findings suggest that integrating peptide-based biostimulants with balanced urea fertilisation can improve bermudagrass performance in transition-zone climates, especially when aligned with cultivar-specific management strategies.

Keywords: bermudagrass, biostimulants, fertilisation, NDVI, turfgrass quality

Introduction

Turfgrass management is evolving under the pressure of climate change, especially in Mediterranean and transition-zone environments. Warm-season grasses such as bermudagrass (*Cynodon* spp.) have gained prominence due to their resilience to drought, high temperatures, and variable rainfall patterns (Guerrero, 2016). However, the challenge is to maintain high turf quality while reducing dependency on synthetic inputs. Despite growing interest in plant biostimulants, few studies have evaluated their effectiveness under practical field conditions across multiple cultivars (Calvo *et al.*, 2014). This study addresses this gap by examining how fertilization programmes, particularly those integrating amino acid biostimulants influence bermudagrass performance across autumn and spring seasons.

Materials and methods

The study was conducted from 2022 to 2024 at the University of Padua experimental farm in Legnaro (45°20'N, 11°57'E) on silty loam soil (pH 8.1). Three bermudagrass cultivars (Sultan, MBG002, Arden 15) were grown in 2 × 2 m plots using a strip-plot layout with three replicates. Plots were randomised within blocks. All programmes received the same annual nitrogen rate (125–150 kg N ha⁻¹), supplied through urea.

The following fertilisation programmes were tested:

Programme 1: Urea only

Programme 2: Urea+high-dose Hicure (90–180 l ha⁻¹)

Programme 3: Urea+low-dose Hicure (45–90 l ha⁻¹)

Hicure is a non-microbial biostimulant composed of amino acids and peptides.

Evaluations included digital image analysis for green cover, NDVI (RapidSCAN CS-45), visual scoring (1–9 scale), and HPLC-based quantification of soluble carbohydrates and starch. Statistical analysis was performed using mixed-effects linear models in R (R Core Team, 2021).

Results and discussion

Treatment significantly influenced turf colour and quality ($p < 0.001$), with Programme 2 outperforming Programmes 1 and 3 across all cultivars. NDVI readings revealed strong cultivar $\times$ treatment interactions ($p < 0.01$), where Sultan and MBG002 consistently exhibited higher NDVI values than Arden 15. Spring green-up was primarily cultivar-dependent, whereas autumn dormancy was delayed in biostimulant-treated plots, indicating improved stress resilience. Sultan displayed the highest total non-structural carbohydrate (TNC) content, supporting previous findings linking carbohydrate storage to enhanced stress tolerance (Zhang *et al.*, 2017). Together, these results demonstrate that both cultivar choice and biostimulant-enhanced fertilisation strategies play synergistic roles in turf performance under climatic stress.

Conclusion

Integrating peptide-based biostimulants with urea fertilisation significantly improved bermudagrass performance under transition-zone conditions. Programme 2 emerged as the most effective, yielding the highest improvements in colour, quality, NDVI, and carbohydrate accumulation. Sultan and MBG002 were the most responsive cultivars. These results highlight the value of combining cultivar-specific management with biostimulant-supported fertilisation to enhance turfgrass sustainability and resilience.

Acknowledgements

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Effect of cricket frass treated through different methods as organic fertilizer for ryegrass biomass production

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Abstract

The present study is a contribution to evaluate the agronomic potential of cricket frass (either in the original form (FO), by physical heat treatment (FH), or by biological stabilization at 35% (F35) and at 60% humidity (F60)) as organic fertilizers, through the measurement of biomass production and the photosynthetic activity of ryegrass (*Lolium multiflorum* Lam.), growing in a calcareous soil, treated with two different doses (1 and 2) of those products compared to a mineral control (recommended rate of mineral fertilizer for ryegrass). Within this aim, a pot experiment was conducted in a semi-controlled greenhouse. Biomass production was significantly affected by the use of frass. An assessment of certain physiological parameters related to primary production revealed that photosynthetic rate and total chlorophyll content values were consistently higher across all treatment modalities than in the control. These findings align with the production increase. The water-use efficiency (WUE) values suggested that the plants were experiencing optimal water status, demonstrating high efficiency and effective stomatal regulation, with no apparent damage to the photosynthetic machinery. Among the frass treatments, the FT and F35 groups, independently of the rate, were particularly noteworthy, exhibiting elevated chlorophyll levels (C_{a+b}) as well as improved WUE, without compromising net photosynthesis rate (P_n).

Keywords: cricket frass, photosynthetic performance, pot trial, ryegrass yield, treated frass

Introduction

One of the by-products resulting from insect production is frass, which has proven to be of interest as an organic fertilizer (Houben *et al.*, 2020; Menino *et al.*, 2021). However, according to EU legislation, it must undergo a temperature treatment for 1 hour at 70°C for microbiological stabilization (Regulation, 2021), for a mature and stable end product. This treatment, when dealing with very large volumes of frass, becomes expensive and time-consuming, which is why more sustainable alternatives have been considered. In the present study, we evaluated the effect of different stabilizing treatments on the agronomic potential of cricket frass used as fertilizer for ryegrass plants, with regard to biomass

production and the assessment of certain physiological parameters related to primary production, such as the photosynthetic rate, water-use efficiency, and total chlorophyll content.

Materials and methods

Soil: Haplic calcisol, according to IUSS-FAO (2006). Surface layer (0–25 cm), analysed as described in Rehan *et al.* (2024), has a pH of 7.9, $14.5 \text{ g (kg organic matter)}^{-1}$, and low levels of nutrients. Cricket frass: either in the original form (FO), heat treated (FH) according to the EU Regulation (2021), biologically stabilized (by the action of microorganisms naturally present in the material) till 35% humidity (F35), and till 60% humidity (F60). The composition of frass (dry matter) revealed values for total N, P, and K contents ranging between 5 and 7%, 1.5 and 2.3%, and 2.4 and 3.7%, respectively. The results for *E. coli* and *Salmonella* were negative. Greenhouse pot experiment: in INIAV (Portugal), using Italian ryegrass (*Lolium multiflorum* Lam.) grown in cylindrical plastic pots (diameter = 24 cm, height = 20 cm), filled with 3 kg of soil, watered daily with deionized water to maintain soil moisture near to 60% of water holding capacity. Temperature varied between 18 and 25°C. Treatments: mineral (M), the recommended rates of mineral fertilizers for ryegrass (25% DAP and 75% calcium ammonium nitrate 27%), based on the N demand of the crop (estimated in 160 kg N ha^{-1}); and two rates (1 and 2) of each of the different types of frass (FO, FH, F35, and F60), replacing, respectively, 25 and 50% of the mineral fertilizers, and considering a mineralization rate of 50%. There were five replicates, totalizing 45 pots. Before sowing, the frass was mixed with soil. Seeds, in a density of 40 kg per hectare, were surface sown. Gas exchanges measurements and photosynthetic pigments: made in adult expanded leaves of 6-week-old plants, prior to cut. Net photosynthesis rate (P_n), stomatal conductance (g_s) and transpiration rate (E) were determined using a portable infra-red gas analyzer (LI6400, Licor, Lincoln, NE, USA), at 20–25°C, with a light supply of $700 \mu\text{mol m}^{-2} \text{ s}^{-1}$, as described in Semedo *et al.* (2021). Water use efficiency (WUE) was calculated as P_n/E . Total chlorophyll levels (C_{a+b}) were determined on the same leaves and conditions used for gas exchange measurements and extracted from approximately 50 mg of fresh weight, using 100% methanol and quantified spectrophotometrically, according to Lichtenthaler (1987). Biomass evaluation: after measurements, plants were cut at 2 cm above the soil surface. Weighted for fresh weight, then washed with deionized water, and dried for dry weight. Data: analysed statistically with ‘Statistix 9.0’, Analytical Software, by using one-way ANOVA with a significance level of $p \leq 0.05$, which was applied to test for differences between treatments in respect to fresh and dry weight, and gas exchange parameters. Tukey HSD test for mean comparison was performed (for a 95% confidence level).

Results and discussion

From the results obtained for ryegrass biomass production (Table 1), it is possible to observe that the highest yield was obtained in the treatment FO-2, by a significant difference compared to treatments M, FO-1 and F60, with both rates. The lowest biomass values, both fresh or dry, were obtained in treatment M. Compared to the M treatment, all frass-amended treatments showed significantly higher P_n values, with increases ranging from 15 to 29%. Concerning WUE, results indicated effective stomatal regulation, which did not compromise the high P_n rates (Lawson and Vialet-Chabrand, 2019). In most treatments, plants maintained relatively low g_s while sustaining efficient primary production.

Regarding C_{a+b} , although some variation was observed between treatments (all exceeding the control), this did not appear to affect P_n performance negatively. However, it is important to highlight the similarity in the pattern observed for fresh weight and C_{a+b} in the treatments FO-2, FH-1, FH-2,

Table 1. Mean values ($n = 5$) for the first cut of ryegrass biomass, expressed as fresh (FW) and dry weight (DW) per pot, net photosynthesis rate (P_n), water use efficiency (WUE) and total chlorophyll content (C_{a+b}), in each treatment

Treatment	Fresh weight (g)	Dry weight (g)	P_n ($\mu\text{mol m}^{-2} \text{s}^{-1}$)	WUE ($\mu\text{mol CO}_2$ ($\text{mol H}_2\text{O}$) $^{-1}$)	C_{a+b} (mg (g DW)^{-1})
M	32 c	5.0 c	12.2 e	5.1 c	8.3 c
F0-1	45 bc	6.3 bc	17.0 ab	4.9 c	10.1 bc
F0-2	62 a	8.2 a	15.1 cd	6.9 a	12.6 abc
FH-1	50 ab	6.5 b	14.6 d	5.6 bc	13.6 ab
FH-2	59 ab	7.6 ab	15.8 abcd	6.8 ab	16.2 a
F35-1	54 ab	7.5 ab	15.7 bcd	6.9 a	12.5 abc
F35-2	56 ab	7.6 ab	16.0 abc	5.5 c	13.9 ab
F60-1	46 bc	6.9 ab	15.6 bcd	4.4 c	10.1 bc
F60-2	44 bc	6.7 b	17.1 a	4.7 c	9.7 bc

Means in the same column with the same letter do not differ significantly ($p \leq 0.05$), as assessed by the Tukey test.

F35-1, and F35-2. There appears to be a trend associating higher biomass production with increased levels of photosynthetic pigments.

Conclusion

Under the conditions of this study, the fertilizing value of cricket frass is confirmed, as well as the possibility of partially replacing mineral fertilization without loss of production. It also corroborates the potential of alternative approaches to the mandatory heat treatment of frass.

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Interests and limits of genetic diversity in multispecies swards

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Abstract

Multispecies swards (MSS) are gaining popularity on French livestock farms for their potential to improve productivity despite climate challenges, and they provide versatile, high-nutrition grass suitable for grazing and mowing. MSS typically combine 2–3 grass species with 2–3 legume species. INRAE studies in Lusignan (URP3F) revealed that incorporating contrasted genetic diversity (e.g., growth start timing, heading date, ploidy), within MSS can stabilize mix composition and reduce yield variability (Litrico *et al.*, 2015). To validate these findings, a multisite trial was launched in 2020 under the CLIMATVEG project in Pays de la Loire. This trial compared monovarietal MSSs with those featuring multivarietal diversity, using species like perennial ryegrass, tall fescue, cocksfoot, white clover, and either red clover or alfalfa. After three years, results differed from earlier studies. Genetic diversity neither boosted productivity nor reduced yield variability. A key factor was the dominance of cocksfoot, which proved aggressive and led to a significant simplification of the mixtures, undermining the introduced genetic and species diversity. The study highlights the need for a registration framework for new varieties specifically adapted for use in MSSs, emphasizing the importance of a “sociability” criterion to ensure compatibility within mixtures.

Keywords: adaptation, diversity, multispecies swards, varieties

Introduction

Grasslands are increasingly exposed to climatic hazards, particularly summer droughts. In this context, farmers are seeking grassland systems that can maintain productivity, preserve a balanced botanical composition, and offer satisfactory forage quality. Multi-species swards (MSS), combining several grasses and legumes, have proven to be more productive (+1.5 t DM/ha) and more stable than simple mixtures (Coutard and Pierre, 2012). Recent research at INRAE Lusignan (Poitou-Charentes) has shown that genetic diversity within MSS enhances their resilience, botanical stability, and forage quality (Litrico *et al.*, 2015; Meilhac *et al.*, 2019). This study aims to evaluate the effect of genetic diversity on the drought tolerance of MSS in the Pays de la Loire region.

Material and methods

A randomized block trial (4 blocks) was conducted from September 2021 to autumn 2024 at three experimental sites in the Pays de la Loire region: Derval, Les Etablères, and Thorigné-d'Anjou. A total of five MSS mixtures were evaluated, each implemented in either a single-variety (SV) or multi-variety form (MV). The main species selected for their drought tolerance were cocksfoot (*Dactylis glomerata*), tall fescue (*Festuca arundinacea*), alfalfa (*Medicago sativa*), and red clover (*Trifolium pratense*), with species choice adapted to local soil and climate conditions (alfalfa on drought-prone soils at Les

Table 1. Details of MSS species and varieties tested and sowing rates for each site (kg ha⁻¹)

Species	Varieties	SV1	MV1	SV2	MV2	SV3	MV3	SV4	MV4	SV5	MV5
Perennial ryegrass	Careva	4	4	4	4	4	4	4	4	4	4
Cocksfoot	RGT Veraly	3	1	5	1.7			8	2.7		
	Archibaldi		1		1.7				2.7		
	Tardi		1		1.7				2.7		
Tall fescue	Lydie	10	2.5			10	2.5			15	3.8
	Volupta		2.5				2.5				3.8
	Aliénor		2.5				2.5				3.8
	Prosper		2.5				2.5				3.8
White clover	Brianna	2	2	2	2	2	2	5	5	5	5
Red clover: Derval	Ganymed	6	2	6	2	6	2				
	Lestris		2		2		2				
	Atlantis		2		2		2				
Lucerne: Les Établières and Thorigné d'Anjou	Sibemol	10	2	12	2.4	10	2				
	Luzelle		2		2.4		2				
	Felicia		2		2.4		2				
	Occitane		2		2.4		2				
	Barvince		2		2.4		2				

Etablières and Thorigné d'Anjou, red clover at Derval on soils with alternating moisture levels). Perennial ryegrass (*Lolium perenne*) and white clover (*Trifolium repens*), known for their good ground cover capacity, were included in all treatments. All mixtures were designed for mowing use. The varieties selected within the dominant species were chosen to maximize behavioural contrasts, in order to test the effect of genetic diversity.

The plots measured 9 m×1.5 m. All blocks were balanced for soil P, K, and pH. A composted manure application of 15 t ha⁻¹ was carried out in November 2023. Measurements were taken at each harvest cycle, corresponding to typical grassland management periods: C1 (end of winter), C2 (peak growth), C3 (summer), and C4 (autumn regrowth). The following variables were assessed: (1) dry matter (DM) yield, (2) botanical composition (grasses, legumes, and other species (C1, C3, C4); proportion of each species (C2)), and (3) chemical composition analysed in laboratory (C1, C2, C4). Botanical compositions were estimated using fixed quadrats installed at sowing. Harvests were triggered when sward height reached 18 cm (platometer) for SV1 during cycles C1 and C2 with a 45-day interval, and at 10 cm for the summer and autumn cycles (C3 and C4). Statistics were carried out using a two-factor ANOVA.

Results and discussion

None of the tested mixtures showed consistently higher yields, regardless of site (Fig. 1). The strong interannual variability—ranging from a very dry year in 2022 to wetter conditions in 2023 and 2024—masked any potential effects of genetic diversity on productivity or its stability.

No statistical differences were detected between SV and MV mixtures; only year effects were observed, reflecting the strong variation in climatic conditions. Perennial ryegrass largely disappeared after the summer of 2022 (0% in 2024), while still in the establishment phase, whereas cocksfoot became dominant in the mixtures (Fig. 2). Its ability to regrow after drought and its deep root system allowed it to prevail. Previous studies showed its dominance in simple multi-species grasslands on shallow

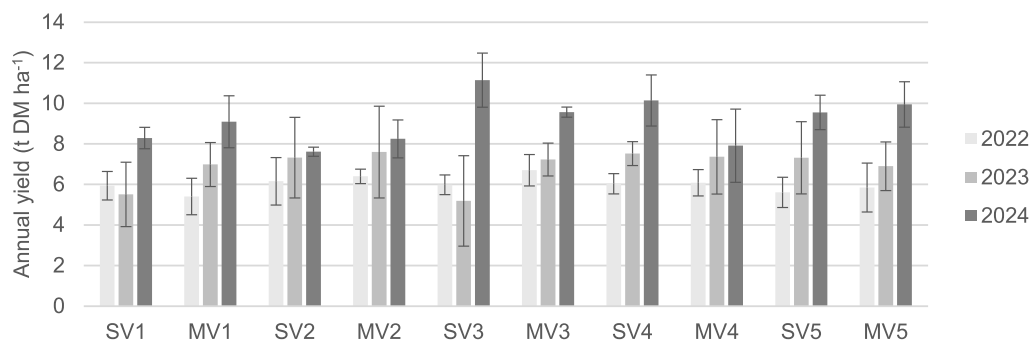


Figure 1. Cumulative yields and standard deviations per year at Les Etablères site.

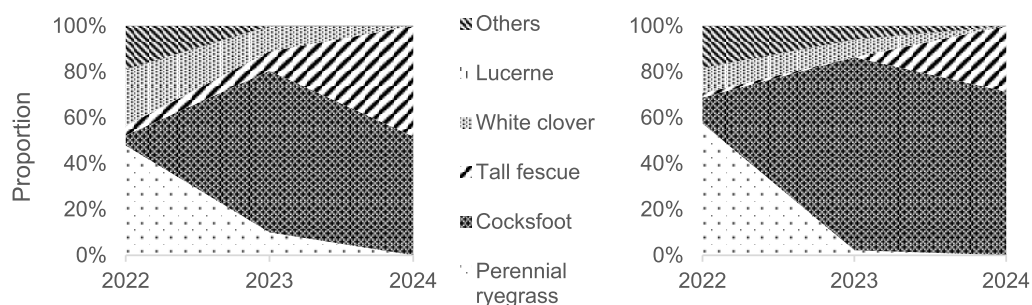


Figure 2. Assessment of the botanical composition of SV1 (left) and MV1 (right) at Les Etablères.

soils (Coutard & Pierre, 2012). Tall fescue also established well and showed good tolerance to contrasting climatic conditions. Legumes were less persistent. White clover declined progressively (0% in 2024), while alfalfa never successfully established (0% since 2022), confirming that autumn is not a favourable period for it to be sown. Initial weed pressure, high in 2022, decreased over time as the sown species progressively colonized the plots. No difference was observed between the SV and MV mixtures regarding the evolution of the botanical composition.

The decline in crude protein (CP) content over the years was linked to the progressive reduction of legumes, which limited nitrogen availability. This shortage particularly penalized cocksfoot, a protein-rich but nitrogen-demanding species, thereby lowering the nutritive value of mixtures where it predominated. The best CP content was observed in autumn (12 % CP (C4) against 9% (C1, C2, C3)), highlighting the value of autumn utilization. In contrast, summer CP (C3) remained highly variable, reflecting year-to-year climatic variability.

Conclusion

After three years of trial, multi-variety MSS did not demonstrate clear superiority over single-variety MSS in terms of productivity, yield stability, or grass-legume balance. The lack of visible effects may be attributed to the time required to establish a durable sward equilibrium, as well as disappearance of certain varieties over time. Variety choice thus emerges as a critical lever that requires further refinement. Revising monoculture-focused selection criteria could foster greater varietal compatibility, as shown in Switzerland (Mosimann and Charles, 1995).

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Effect of foliar application of biostimulants on selected mineral nutrient ratios in grass dry matter

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Abstract

The aim of the study was to evaluate the effect of Tytanit and Stymjod applied separately and together on the ratios of K:(Ca+Mg) and Ca:P in dry matter of *Dactylis glomerata* L. and *Festulolium braunii* (K. Richt.) A. Camus. Stymjod is a liquid nano-fertilizer created on the basis of iodine and Tytanit is a biostimulant created on the basis of titanium. The plant material in the experiment consisted of two grass species. The study was conducted from 2021 to 2023 based on a micro-plot experiment. Factors of the experiment were: plant species: *Dactylis glomerata* 'Amera', *Festulolium braunii*, 'Felopa' and treatment: Control, control with distilled water; Ti, liquid spray with 0.04% Tytanit; Sj, liquid spray with 2.5% Stymjod; Ti+Sj, combined use of Tytanit and Stymjod spray solution. The preparations were applied to each of the three re-growths. Spraying was performed when the plant reached the stem elongation phase and then the spraying was repeated 10 days after the first treatment. The applied preparations differentially affected the concentration of selected macronutrients and ionic ratios in the biomass of the analysed grasses.

Keywords: calcium, *Dactylis glomerata*, *Festulolium braunii*, magnesium, phosphorus, potassium

Introduction

The chemical composition of permanent grassland is highly variable. Good quality feed should contain adequate amounts of macro- and microelements in appropriate proportions. According to Grzegorzczak *et al.* (2013), a synthetic criterion for assessing feed quality is the K:(Ca+Mg) ratio. The literature suggests that for feed to be of good quality, this ratio should be between 0.66 and 0.98 (Gaweł, 2009). Another ionic ratio that provides a lot of information about feed quality is the Ca:P ratio. According to the NRC (1996), the Ca:P ratio in feed should be between 1.0 and 2.0. Studies (Ayan *et al.* 2010) have highlighted that a Ca:P ratio above 2.0 in the diet of animals has a negative impact on their health.

Materials and methods

The research was conducted based on experiments in rings located at the Experimental Station of the University of Siedlce (52°10'3"N; 22°17'24"E) during three years, 2021–2023. In April 2021, polyurethane rings (diameter = 20 cm, height = 20 cm) were dug in at a spacing of 0.8 m x 0.8 m then filled with soil. Next, 10 grass seeds were sown manually in each ring. The three strongest plants were left in each ring. The experiment was conducted in four replicates. The following research factors were used in the experiment:

Factor I—grass species: *Dactylis glomerata* L. 'Amera' and *Festulolium braunii* 'Felopa'.

Factor II—type of biostimulant: Control (distilled water), 0.04% solution of Tytanit (Ti), 2.5% solution of Stymjod (Sj), 0.04% Tytanit + 2.5% solution of Stymjod (Ti+Sj).

Tytanit (Intermag, Olkusz, Poland) and Stymjod (Jeznach, Wojtówka, Poland) were applied to each of the three growth cycles: spring, summer and autumn. Spraying was performed when the plant reached

the stem elongation phase and repeated 10 days after the first treatment (i.e., six sprayings per year). Per application, 50 ml of working fluid was used to thoroughly spray the plants. In the case of Ti+Sj treatment, the first operation was spraying with Tytanit and Stymjod was applied after 30 minutes. During the growing seasons, three grass shoots were collected at the heading stage. The dry weight of individual morphological parts of plants was determined by oven-drying. The plant material prepared for chemical analysis consisted of ground material from leaves, stems and inflorescences collected from a single ring. The total content (g kg^{-1}) of selected macroelements (P, K, Ca, Mg) was determined in the plant dry matter using the ICP-AES method, and the quantitative ratios $\text{K}:(\text{Ca}+\text{Mg})$ and $\text{Ca}:\text{P}$ were calculated. Laboratory tests showed that the soil in the rings was slightly acidic (pH_{KCl} 5.8), with organic matter content of 2.08%, and average content of available forms of nitrogen and minerals was (mg kg^{-1}): 30.3 $\text{NO}_3\text{-N}$; 49.0 $\text{NH}_4\text{-N}$; 81.6 P; 182.4 K; 417.2 Ca; 66.1 Mg. The soil content of P, K, Ca and Mg (assimilable forms) was determined using the ICP-OES method, using a Perkin-Elmer Optima 8300 emission spectrometer, after extraction of components from the soil using the Mehlich 3 method (Mehlich, 1984).

Statistical analysis was performed using Statistica 13 software (TIBCO Software, Palo Alto, CA, USA). Data were analysed at a significance level of $p < 0.05$. Differences between means were assessed using ANOVA, followed by Tukey's HSD test. Statistically significant differences among means are indicated by different letters in the tables.

Results and discussion

Depending on the application of the biostimulant, the obtained results of the $\text{K}:(\text{Ca}+\text{Mg})$ ion ratio ranged from 0.67 to 0.81 (Table 1). The lowest value was obtained from the biomass of the control treatment, and the highest after the application of Tytanit. No statistically significant difference in the ion ratio of the above mentioned macroelements was observed between plant species. The results for both species ranged from 0.73 to 0.77. Similar values were reported in the literature for *L. multiflorum*, where after the application of different doses of Tytanit, the values ranged from 0.80 to 0.87 (Sosnowski et al., 2023). Higher $\text{K}:(\text{Ca}+\text{Mg})$ values were observed in plants in 2021 and 2023. This was probably due to hydrothermal conditions. Dry conditions prevailed in the first and third years of the study, which probably affected the balance between K and Ca and Mg. In the case of interaction between preparation $\times$ year, statistically significant differences in the $\text{K}:(\text{Ca}+\text{Mg})$ ion ratio were observed. The effect of the Tytanit preparation significantly increased the ion ratio in question in all years of the study, which may suggest that it is a biostimulant that affects the plant regardless of the prevailing hydrothermal conditions. The effect of Stymjod was different according to the year of the study. In the second year, a year in which dry periods alternated with wet periods every month, plants treated with Stymjod showed similar results to the control.

The use of biostimulants increased the $\text{Ca}:\text{P}$ ratio in relation to the control object by 8.2% (Tytanit) to 19.4% (Stymjod, Table 1). The very wide ion ratio in the tested plant biomass was caused by too low P levels in the soil. Both *D. glomerata* and *F. braunii* were characterised by an excessively wide ion ratio of the above elements, which would make these plants too poor in P as feed. According to Grzegorzczak et al. (2017), Ca and P levels in the diet of animals should be balanced in order to increase their intake. Ciepiela and Godlewska (2017) also studied the effect of a biostimulant on the $\text{Ca}:\text{P}$ ion ratio in grass feed. The authors found that the average ratio was 2.64 for *D. glomerata* and 2.78 for *F. braunii*. Grzegorzczak et al. (2017) found that the $\text{Ca}:\text{P}$ ratio in herbs and legumes, ranging from 3.9 to 7.8, significantly exceeded the recommended standards. The narrowest $\text{Ca}:\text{P}$ ion ratio was recorded

Table 1. Mineral nutrient ratios in the dry matter of *D. glomerata* L. and *F. braunii* depending on the preparation used

Nutrient ratio	Factor	Year			Mean
		2021	2022	2023	
K:(Ca+Mg)	Control	0.68 Ab	0.65 Ab	0.69 Ac	0.67 b
	Ti	0.80 Aa	0.79 Ba	0.83 Aa	0.81 a
	Sj	0.78 Aa	0.69 Bb	0.76 Ab	0.75 ab
	Ti+Sj	0.78 Ab	0.70 Bb	0.78 Ab	0.76 ab
	Mean	0.76 A	0.71 B	0.77 A	
	<i>Dactylis glomerata</i>	0.75 Aa	0.68 Bb	0.74 Ab	0.73 a
	<i>Festulolium braunii</i>	0.77 ABa	0.74 Ba	0.80 Aa	0.77 a
Ca:P	Control	5.60 Ab	5.65Ab	5.80 Ab	5.68 b
	Ti	5.89 Aab	6.74 Bab	5.83 Aab	6.15 ab
	Sj	5.67Ab	7.98 Ba	6.70 Aa	6.78 a
	Ti+Sj	6.76 Aa	6.52 Bab	6.35 Aa	6.54 a
	Mean	5.98 B	6.66 A	6.17 AB	
	<i>Dactylis glomerata</i>	6.10 ABa	6.67 Aa	5.99 Bb	6.25 a
	<i>Festulolium braunii</i>	5.74 Ba	6.80 Aa	6.48 ABa	6.34 a

Values in columns within the five treatments and within the two species marked with the same lowercase letters do not differ significantly; Values in rows marked with the same uppercase letters do not differ significantly.

in plants in the first year of our study, which was probably due to the young age of the plants and the higher P content in the soil. Statistically significant interactions between plant x year for the Ca:P ion ratio were also observed. In the case of Stymjod, plants in the second and third years of the study showed an increase in the Ca:P ion ratio compared to plants from the Control.

Conclusion

Tytanit had a significant effect on the macroelement content in the dry matter of *D. glomerata* and *F. braunii* plants. The use of Tytanit optimised the potassium, calcium and magnesium content in plant biomass, thereby improving the K:(Ca+Mg) ion ratio.

The use of biostimulants exacerbated the unfavourable Ca:P ratio, probably by improving the availability of Ca to plants in P-deficient soil.

The results obtained can be used to develop recommendations for the use of biostimulants to improve feed quality under unfavourable hydrothermal conditions.

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Investigating biological nitrogen fixation differences between white clover varieties under varying nitrogen rates

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Abstract

White clover (*Trifolium repens* L.) is an important component of grassland systems due to its capacity for biological nitrogen (N) fixation (BNF). The evaluation of white clover varieties for recommended lists does not quantify the BNF capacity of varieties under varying N management systems. This would better inform farmers on varieties most beneficial to their system from a chemical N reduction perspective. The objective of this experiment was to evaluate the effect of variety and N application rate on BNF. The experiment consisted of five white clover varieties sown with AberChoice (perennial ryegrass, *Lolium perenne* L., PRG) managed at three N fertilisation rates (0, 75 and 150 kg N ha⁻¹ year⁻¹). The natural abundance method compares the $\delta^{15}\text{N}$ of a legume paired with a non-legume reference plant to estimate %Ndfa (N derived from the atmosphere). Prior to defoliation, a selective herbage sample, obtaining fractions of grass and clover from each plot was dried, milled and analysed for atom% ¹⁵N content and %N content. Nitrogen fertilisation rate had a significant ($P < 0.001$) impact on the BNF. Nitrogen fixation was reduced from an average 102 kg N ha⁻¹ year⁻¹ to 48 kg N ha⁻¹ year⁻¹ when nitrogen application was increased from 0 to 150 kg N ha⁻¹ year⁻¹.

Keywords: clover content, clover dry matter yield, natural abundance, *Trifolium repens* L.

Introduction

Nitrogen is a major limiting nutrient in herbage production, applied in large quantities to sustain yields (Sainju *et al.*, 2019). Both the EU and Ireland have established reduction targets in relation to fertiliser application to alleviate the pollution associated with the use of chemical N. EU policy aims to reduce chemical N fertiliser usage by at least 20% by 2030 (Farm to Fork Strategy; (European Commission, 2020)). Clover incorporation into swards can enable a reduction in chemical N usage and associated greenhouse gas emissions while sustaining overall grass production (Herron & Shalloo, 2022), through its capacity for BNF. The objective of the current study was to determine the N-fixation response when assessed under varying N rates across different white clover varieties. This will facilitate the identification of superior white clover varieties in relation to BNF under different N management regimes to facilitate the transition to low N grassland systems on farm.

Materials and methods

The experiment was established in 2022 at the Teagasc, Animal and Grassland Research and Innovation Centre, Moorepark, Fermoy, Co. Cork, Ireland (50°7'N, 8°16'W; 40 m asl). Five white clover varieties classified by both leaf category and size (Table 1) were sown with AberChoice PRG in 7 m×1.5 m plots. A three-replicate randomised block design was fertilised at 0, 75 and 150 kg N ha⁻¹ year⁻¹ (0 N, 75 N and 150 N, respectively). Fertiliser N (46% Protected Urea) was applied after each defoliation from February to September. Data recording began in 2024. The plots were mechanically defoliated when an average herbage mass of 1400 kg dry matter (DM) ha⁻¹ was reached. Pre-grazing herbage

Table 1. Relationship between varieties, leaf size, nitrogen rate and clover DM production

Variety	Leaf size	Leaf category	Clover DM production (kg DM ha ⁻¹)		
			0 N	75 N	150 N
PRG + AberAce	0.24	Small	3128	2459	1537
PRG + AberHerald	0.54	Medium	5004	3226	2582
PRG + Chieftain	0.67	Medium	3861	2615	1881
PRG + Clodagh	0.76	Large	4239	3075	3433
PRG + Brianna	1.12	Large	3866	1769	2375
SE	—	—	169.4	169.4	169.4

Leaf size (relative to Aran at 1229 mm²) and category as determined by the DAFM Recommended List of clover varieties.

mass was measured by cutting each plot with an Etesia mower harvester (Etesia UK, Warwick, UK), with a 100 g subsample of the mown herbage dried at 60°C for 72 hours for DM determination.

Samples were subsequently milled through a 0.2 mm sieve prior to crude protein (CP) analysis. Sward clover content was measured seasonally (spring, summer, autumn). A random 70 g sample was taken along a transect of each plot, cut by Gardena shears at 4 cm (Gardena, Ulm, Germany) prior to defoliation. The herbage was separated into clover, grass and weed fractions and dried at 90°C for 16 hours to determine clover DM proportion. The natural abundance (NA) method compares the $\delta^{15}\text{N}$ of a legume paired with a non-legume reference plant to estimate %Ndfa (N derived from the atmosphere) (Cox *et al.*, 2022). Prior to each defoliation event, a representative herbage sample was obtained, comprising a 50 g fraction each of grass and clover. This was dried, milled and analysed for atom%¹⁵N content and %N content. The %Ndfa was calculated as follows: Ndfa (%) = ($\delta^{15}\text{N}$ reference plant– $\delta^{15}\text{N}$ clover)/($\delta^{15}\text{N}$ reference plant–B_S) (Unkovich *et al.*, 2008). The above-ground N² fixation (kg ha⁻¹) was derived from the %Ndfa multiplied by the clover N yield ((herbage yield×clover %×CP %)/6.25). Data were analysed using SAS 9.4 statistical programme (SAS Institute, Cary, NC). The PROC MIXED function was used to determine the effect of variety, season, nitrogen rate and associated interactions on nitrogen fixation.

Results and discussion

The annual N fixed by grass-clover mixtures differed significantly ($P < 0.001$) between N application rates. There was a strong negative correlation between N rate and BNF ($r = -0.69$) as the amount of N fixed reduced as N rate increased (102, 62 and 48 kg N fixed ha⁻¹ year⁻¹ at 0 N, 75 N and 150 N, respectively). This confirms (Laidlaw, 1988) who showed that N fertiliser reduces clover N contribution to total yield due to a reduction in clover DM production but not the efficiency of the individual clover plants to fix N. There were numerical differences between the amount of N fixed by the varieties (Figure 1), but not significantly so in this short Feb-Sept test period.

This variation did not align with variety leaf size but can be attributed to their differences in clover DM production, given the strong positive correlation with BNF ($r = 0.82$). Similar to Burchill *et al.* (2014), differences in clover DM yield were the largest contributor to annual variation in BNF.

Biological N fixation was significantly ($P < 0.001$) impacted by season. Nitrogen fixation increased from spring into summer and autumn (Figure 2). As previously outlined by Kessler *et al.* (1990), N fixation exhibits a positive response to higher shoot temperature, governing this season response. Clover

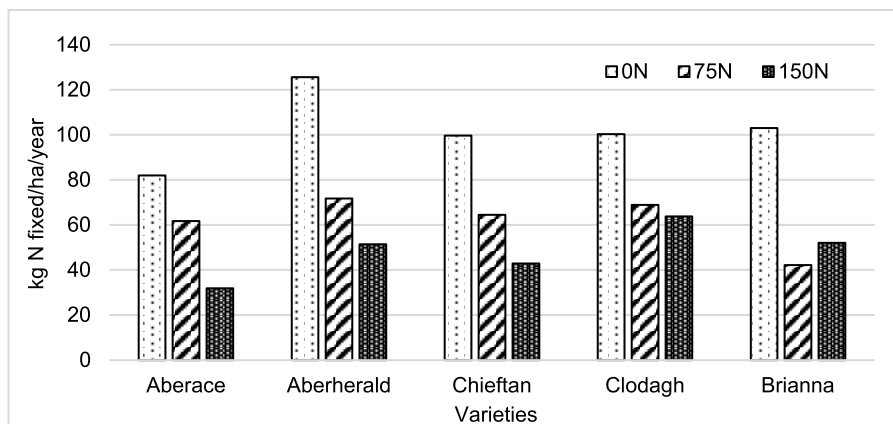


Figure 1. Relationship between varieties, nitrogen application rate and biological nitrogen fixation.

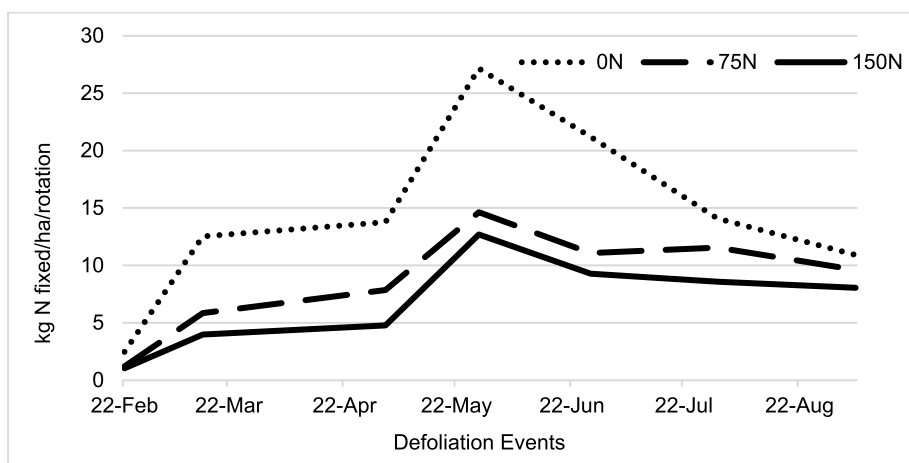


Figure 2. Level of biological nitrogen fixation between nitrogen application rates across rotations.

proportion increased as the year progressed. This increase in clover DM production and temperature increased BNF from spring to summer and autumn.

Conclusion

In the first year of this study, the expected trends of decreasing BNF with increasing N application rates were observed. The amount of N fixed by white clover was closely related to the clover DM production which was affected by both variety and N application rate. This study will be continued to seek significant differences in BNF capacity between clover varieties. As part of this, more efficient, cost-effective methods of quantifying BNF will be tested, with the aim to provide a simple evaluation protocol for quantifying varietal differences in BNF capacity.

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Effect of seeding rate and timing on the establishment of plantain in existing grazed pastures in Ireland

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Abstract

The inclusion of ribwort plantain (*Plantago lanceolata* L.; PL) in grazing swards is of interest to researchers as it has been shown to impact on nitrogen (N) cycling in pasture based grazing systems. There is substantial interest in the establishment of PL in existing mixed swards of perennial ryegrass (*Lolium perenne* L.; PRG) and white clover (*Trifolium repens* L.; WC) in Ireland. This study examined the effect of PL sowing rate and timing of sowing on the establishment of PL in existing mixed swards of PRG & WC. In spring and autumn of 2023, 12 plots (36 m²) were grazed to 3.5 cm by dairy cows and subsequently over-sown with PL at 2.5, 5.0 or 7.5 kg seed ha⁻¹. Ribwort plantain germinated and established in all spring sown plots but only sporadically in autumn sown plots; average sward PL content in the spring and autumn plots was 35% and 0%, respectively, in March 2024. Sowing rate was associated with PL content in spring sown plots ($P < 0.05$) where PL content was 20, 39 and 44% of March 2024 sward dry matter in the 2.5, 5 and 7.5 kg ha⁻¹ sowing treatments, respectively; no differences were observed between sowing rates in autumn sown plots.

Introduction

Ribwort plantain (*Plantago lanceolata* L. (PL)) can play a role in reducing N losses from grazing systems as it has been shown to impact N cycling at animal and soil interfaces (Mangwe *et al.*, 2019, Welten *et al.*, 2019). Researchers have shown that PL needs to account for at least 30% of sward dry matter (DM) (Minnée *et al.*, 2020) in order to impact the N cycling in grazing systems. Recent studies have reported various contents of PL in grazing swards (Baker *et al.*, 2023, Hearn *et al.*, 2023). Where PL content is reduced to less than 30% of sward DM, remedial action is required to maintain sward ecosystem service functionality. The current study is a follow up to Hearn *et al.* (2024) where differences in sward PL content due to different sowing rates were reported in the first 5 months post a spring over-sowing event. There is evidence of over-sowing date influencing establishment of WC in PRG-dominant grazing swards on commercial farms in Ireland (Egan *et al.*, 2022); for WC establishment, spring over-sowing events tend to be more successful than autumn over-sowing events. The objective of the current trial was to determine whether the sowing rate and timing of sowing could impact on the level of PL establishment in existing grazing swards of PRG and WC in Ireland in the first full growing season post over-sowing.

Materials and methods

The experiment took place in 2023 and 2024 at Teagasc Moorepark (52°16'N; 8°26'W) where the soil type is a free draining acid brown earth soil of sandy loam texture; soil nutrient status of the plot area in January 2023 was 11.9 and 187 mg l⁻¹ of P and K, respectively, and soil pH was 6.4. Plots from a grazing experiment completed in 2022 were used for the current study—the existing plots consisted

of mixed swards of PRG (cv. AberGain and AberChoice) and WC (cv. Buddy)—the pre-experimental botanical composition of these plots were 73, 19, 0 and 8% PRG, WC, PL and weeds, respectively.

As per Hearn *et al.* (2024), the protocol for this trial was developed to incorporate best practices from previous over-sowing studies (Egan *et al.*, 2022, Raedts and Langworthy, 2020). Briefly, all plots (36 m²) were grazed to 3.5 cm compressed sward height (CSH) by 40 lactating dairy cows. Ribwort plantain (cv. Tonic) seed was applied on to the soil surface immediately post grazing using an Einböck Pneumaticstar PRO Seeder, for each of the sowing rate treatments (2.5, 5 and 7.5 kg ha⁻¹). Compound chemical fertiliser was applied following the sowing, at a rate of 12 kg N ha⁻¹, 12 kg P ha⁻¹ and 24 kg K ha⁻¹. The above process was carried out on separate but adjacent plot areas in spring (April) and autumn (September) 2023. Plots were arranged in a block design with 4 replicates of each sowing treatment within each block.

Following over-sowing, all plots were grazed when the average plot area pre-grazing CSH was 8 cm; all plots were grazed together over a period of 24–36 h to a target residual CSH of 4 cm. Botanical composition of each plot was measured on two occasions in 2024 (March and September), using the methodology of Hearn *et al.* (2024). Briefly, samples were collected in each plot using a mechanical hand shears immediately prior to grazing. Samples were separated by hand into grass, clover and plantain and dried for 48 h at 90°C to determine sward content on a DM basis.

Compound chemical fertiliser was applied at a rate of 12 kg N ha⁻¹, 12 kg P ha⁻¹ and 24 kg K ha⁻¹ following each grazing to ensure appropriate N was provided to a grazing sward including WC (Murray *et al.*, 2023) while not limiting the availability of soil P or K.

Data analysis was conducted using SAS 9.4; a mixed model was used to estimate species sward DM content differences where sowing rate and sowing month, and associated interactions, were included as fixed effects, and replicate was included as a random effect.

Results and discussion

The level of PL was associated with over-sowing date ($P < 0.05$) and over-sowing rate ($P < 0.05$). All sowing rates used for over-sowing in spring resulted in the establishment of PL whereas little establishment was recorded in plots which were over-sown in autumn (Figure 1). Similar to Raedts and Langworthy (2020), the results of the current trial indicate that spring over-sowing is an effective method of establishing PL in existing grazing swards; these results indicate that autumn over-sowing of PL is not effective. The current results reflect the findings of Egan *et al.* (2022) where it now appears that spring over-sowing is more effective for both WC and PL establishment in existing grazing swards in Ireland.

Sowing rate influenced the level of PL establishment in spring over-sown plots where higher PL sowing rate correlated to higher levels of PL in the grazing sward (Figure 1). There was some variation in the difference of sward PL content between sowing rate treatments at different measurement points. In the spring of 2024, the sowing rates of 5 and 7.5 kg ha⁻¹ provided sufficient sward PL DM content to influence N cycling in grazing systems (Figure 1). However, due to sward dynamics, in particular the growth habit of WC in Irish grazing swards, sward PL content was below the required level of 30% in autumn of 2024. This result is similar to that of Hearn *et al.* (2023) where it was reported that sward species content can vary widely within a growing season.

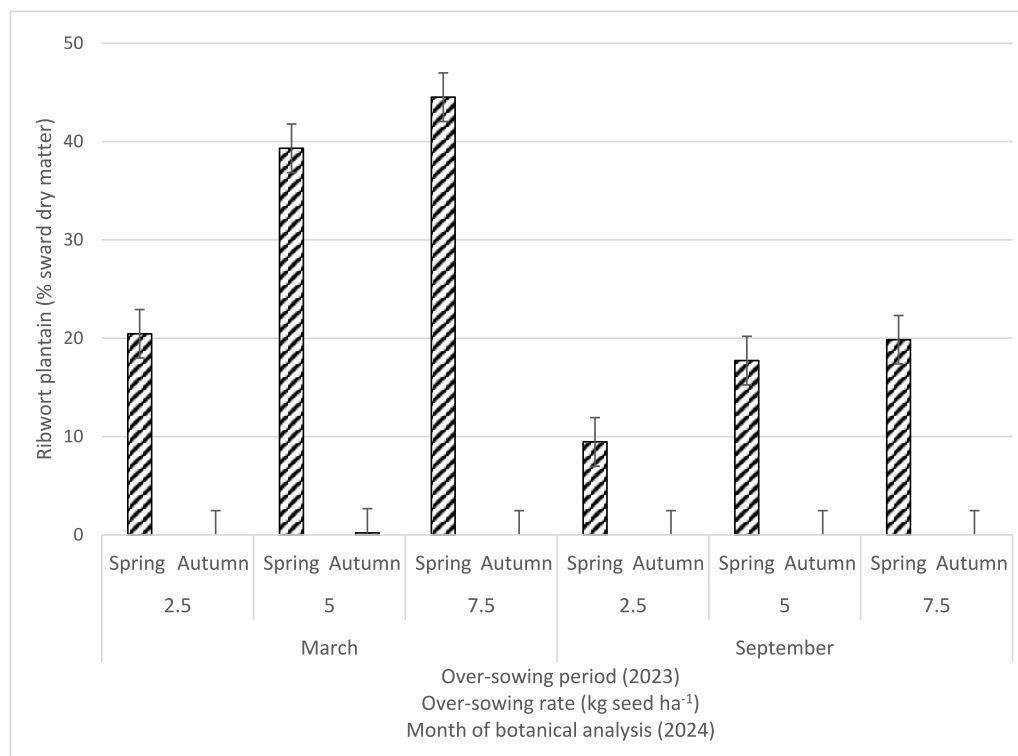


Figure 1. Ribwort plantain content (% sward dry matter) of 2023 spring and autumn over-sown plots in March and September 2024; error bars represent standard error.

Conclusion

The current work indicates that spring over-sowing, using a minimum rate of 5 kg seed ha⁻¹, can be a viable method of maintaining sward PL content at levels required to impact N cycling on farm. While the current trial needs to be replicated using different sowing years, researchers need to be conscious of the possible effects of PL variety and site on PL establishment after over-sowing.

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Impacts of drought-resistant species on forage yield and nutritive value during the sowing year

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Abstract

Summer droughts induce strong declines in forage yield in mountain grasslands. The aim of the DryMount project is to determine the most suitable mixtures that can provide satisfactory forage yield under drought. Three species-rich mixtures co-dominated by *Alopecurus pratensis* and *Dactylis glomerata* and differing in their expected proportion of drought-resistant species (DRsp, 0, 15, 30%, including *Plantago lanceolata*, *Lotus corniculatus*, *Agrostis capillaris* and *Festuca rubra*) were sown in 2023 with four replicates at four mountain sites (around 1100 m asl) with different pedoclimatic conditions across Switzerland. In the two harvests of the sowing year, drought-resistant species abundance significantly impacted forage yield. At the drier sites, the mixtures with DRsp had higher yields than the mixtures without DRsp, whereas at the wetter sites they showed similar yields than the mixtures without DRsp. Overall, at the first harvest, all forage mixtures showed good nutritive value in terms of crude protein, fibre and sugar content, with differences mainly observed among sites. However, mixtures with 30% DRsp had higher sugar and lignin contents than the other mixtures. Altogether, these preliminary results highlight the benefits of drought-resistant species for forage yield under drought without any major reduction in nutritive value during their establishment in the year of sowing.

Keywords: drought, forage quality, multi-species mixture, soil moisture, yield

Introduction

Mountain grasslands are particularly sensitive to climate change. More frequent and intense summer droughts reduce forage yield (Vitra *et al.* 2019) and have variable effects on forage nutritive value (Deléglise *et al.*, 2015). However, increasing plant diversity enhances grassland yield stability, as well as resistance to drought (Lüscher *et al.*, 2022) as many species provide greater guarantees that some will maintain functioning even if others fail (i.e., insurance hypothesis, Yachi & Loreau, 1999). The aim of this study was to assess how the inclusion of drought-resistant species in commonly used productive grassland mixtures affect forage yield and nutritive value across contrasting climatic conditions in mountain areas.

Material and methods

Grassland mixtures were established at four mountain sites with contrasting pedoclimatic conditions at around 1100 m a.s.l. in the Swiss cantons of Valais (VS), Uri (UR), Bern (BE) and Ticino (TI). In spring 2023, three species-rich mixtures differing in their expected proportion of drought-resistant species (DRsp, 0, 15, 30%), including *Plantago lanceolata*, *Lotus corniculatus*, *Agrostis capillaris* and *Festuca rubra*, were sown. All mixtures were co-dominated by *Alopecurus pratensis* and *Dactylis glomerata* and also included five commonly-used forage species: *Lolium perenne*, *Phleum pratense*, *Poa pratensis*,

Trifolium pratense and *Trifolium repens*. Each site comprised four blocks, with one replicate of each mixture per block. Forage yield was harvested twice and summed to determine annual yield. Nutritive value was assessed at the first harvest by measuring crude protein (CP), neutral detergent fibre (NDF), acid detergent lignin (ADL) and water-soluble carbohydrates (WSC) content. Soil moisture was continuously monitored using EC-5 moisture sensors plugged to ZL6 dataloggers (MeterGroup, München, Germany) and average soil moisture content during both growth periods were calculated at each site. Statistical analyses were conducted using R version 4.3.3 (R Core Team, 2024). The effects of DRsp abundance on forage yield and nutritive values were evaluated for each site using linear mixed-effects models, with ‘block’ included as a random factor. The influence of soil moisture on the response of DRsp mixtures (15 and 30% DRsp) compared to control mixtures (0% DRsp) was analysed using a second-degree polynomial regression model.

Results and discussion

Forage yield increased with increasing abundance of drought-resistant species at the first and second harvests in the UR and BE site, which was then also reflected on the annual yield (Table 1). At the VS site, annual forage yield tended to increase with the increasing abundance of drought-resistant species whereas it tended to decrease at the TI site (although only significant for the first harvest). Interestingly, these contrasting impacts of the abundance of drought-resistant species were explained by the average soil moisture during the two growth periods (Figure 1). Mixtures with drought-resistant species (DRsp 15 and 30%) outperformed control mixtures (DRsp 0%) under drier conditions, while yield differences diminished under wetter conditions. In drier conditions, 30% DRsp mixtures showed greater yield gains than 15% DRsp mixtures, corresponding to up to 60% and 40% yield increase compared to control mixtures, respectively. However, this advantage decreased with increasing soil moisture. Both *L. corniculatus* and *P. lanceolata*, germinated quickly after sowing, and formed a dense vegetation cover, particularly in dry conditions (based on visual observations). This may be due to their higher seed weight (1.2 g and 2 g per thousand seeds, respectively, based on our seeds) compared to some grasses (lower weights), which makes them more robust in the early life stage as they have greater reserves. These two drought-resistant species are also known to achieve higher yields with lower soil moisture (Küchenmeister *et al.*, 2014), which can explain the greater yield gain of DRsp mixtures.

Table 1. Forage yield (two harvests and annual) and nutritive value at first harvest of the sowing year across sites and DRsp abundance levels (mean±SE).

Site	DRsp (%)	Yield (t ha ⁻¹)			Nutritive value (g kg ⁻¹)			
		1st harvest	2nd harvest	Annual yield	CP	NDF	ADL	WSC
VS	0	3.0 ± 0.2	1.6 ± 0.1	4.6 ± 0.2	211 ± 4	307 ± 10	35 ± 2	52 ± 2
	15	3.1 ± 0.1	1.6 ± 0.1	4.6 ± 0.1	194 ± 7	319 ± 11	38 ± 1	57 ± 4
	30	3.3 ± 0.2	1.7 ± 0.1	4.9 ± 0.3	204 ± 12	296 ± 6	42 ± 1	56 ± 5
UR	0	1.6 ± 0.5a	0.8 ± 0.1a	2.4 ± 0.6a	178 ± 3	392 ± 11	59 ± 9	67 ± 3a
	15	2.3 ± 0.3ab	1.0 ± 0.2ab	3.3 ± 0.4ab	171 ± 8	382 ± 11	47 ± 5	70 ± 2a
	30	3.0 ± 0.1b	1.3 ± 0.1b	4.2 ± 0.2b	168 ± 4	367 ± 10	52 ± 3	77 ± 1b
BE	0	2.8 ± 0.4a	2.0 ± 0.1a	4.8 ± 0.4a	169 ± 13	395 ± 15	25 ± 7a	60 ± 5
	15	3.3 ± 0.2ab	2.9 ± 0.1b	6.2 ± 0.2b	181 ± 10	355 ± 5	30 ± 2a	59 ± 3
	30	4.0 ± 0.3b	2.9 ± 0.1b	6.9 ± 0.4b	162 ± 3	343 ± 13	54 ± 7b	69 ± 6
TI	0	3.4 ± 0.08a	2.1 ± 0.1	5.4 ± 0.1	155 ± 3	452 ± 8a	40 ± 1a	60 ± 2
	15	3.0 ± 0.17ab	2.1 ± 0.2	5.1 ± 0.3	145 ± 6	437 ± 6a	53 ± 2b	67 ± 3
	30	2.9 ± 0.19b	2.0 ± 0.2	4.9 ± 0.6	142 ± 4	414 ± 4b	63 ± 4c	67 ± 2

Different letters within site indicate significant differences among DRsp mixtures ($P < 0.05$).

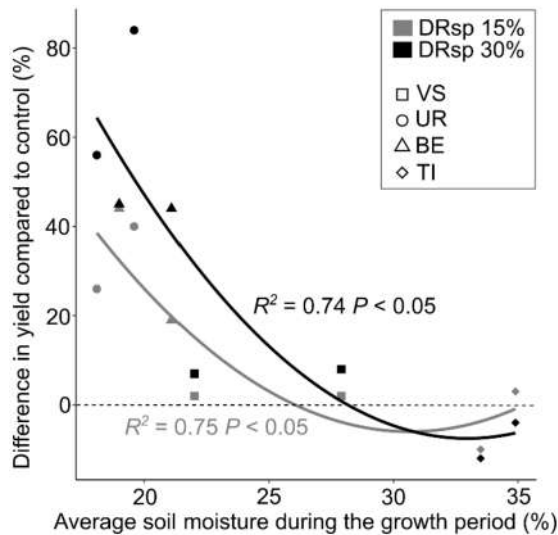


Figure 1. Relative yield differences of drought-resistant species (15% and 30% DRsp) mixtures compared to the control (0% DRsp), as a function of the average soil moisture during the two growth periods of the sowing year per site.

Forage from all mixtures had good crude protein content at each site (Table 1) and this was not impacted by the abundance of drought-resistant species, likely because both *P. lanceolata* and *L. corniculatus* showed satisfactory crude protein content (Küchenmeister *et al.*, 2014, Marley *et al.*, 2006). Forage NDF content tended to decrease whereas sugar content (WSC) tended to increase with increasing abundance of drought-resistant species, but this was only significant for the 30% DRsp mixture at the TI site for NDF and UR site for WSC. Forage ADL content increased with increasing abundance of drought-resistant species at the BE and TI site, which again could be related to the abundance of *P. lanceolata*, which is known to have higher ADL content than other forage species (Elgersma *et al.* 2014).

Conclusions

The inclusion of drought-resistant species increased forage yield under lower soil moisture without compromising yield under wetter conditions, confirming their suitability across a range of climatic conditions. Overall, the presence and increasing abundance of drought-resistant species did not reduce forage nutritive value, with only an increase in ADL content at some sites. These preliminary findings highlight the potential of such mixtures for use in mountain areas exposed to dry conditions. However, a precise assessment of the botanical composition of the mixtures and their persistence over time has to be evaluated in the coming years.

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Protective effect of *Byssochlamys spectabilis* against drought in subterranean clover plants

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Abstract

Fungal endophytes give their host plants biological advantages for surviving. Endophytic fungus *Byssochlamys spectabilis* was inoculated into clover plants which were subjected to drought. Seeds of *Trifolium subterraneum* were sown in a greenhouse and regularly watered up to inoculation. Half of the pots were subjected to deficit irrigation (70% of field capacity) till the end of the trial. Drought influence was estimated by using a scale 1 to 5 (from 1—Healthy plant to 5—Plant completely dead). Biomass was analysed obtaining colorimetric parameters, stoma size of the leaves, fibre, protein, phenolic and antioxidant activity. There was a positive effect of the endophyte for many parameters: the presence of the endophyte (E+) protected the plants against drought, showing E+ greener colour than E- plants. Antioxidant capacity and protein were also higher in E+ plants (more than 35 mg Trolox/g fresh weight and about 13% respectively) with deficit irrigation when compared with E- ones (26 mg Trolox/g fresh weight and 11%), while fibre showed lower values in E+ plants (about 27%). Stomata size seems to decrease in E+ plants (3.6 vs. 5.1 μm). Inoculation of *B. spectabilis* in pasture plants could help to overcome drought in a climate change scenario.

Keywords: biomass quality, drought resistance, endophytes, *Trifolium*

Introduction

Endophytic fungi are plant microbes that live asymptotically within plant tissues, being ubiquitous and highly diverse. Each plant analysed for this purpose has been found to harbour at least one species of endophytic fungi (Saikkonen *et al.*, 2000), and more than 100 different species of endophytic fungi can be found in a single plant specimen. Research on endophytic fungi has increased in recent years, focusing on the benefits that these microbes confer on their host plants: resistance against phytopathogens, improved nutritional status of the host plant (Rodrigo *et al.* 2017), or improved photosynthetic efficiency (Spiering *et al.* 2006), are some of them. In this research, the potential of the endophytic fungi *Byssochlamys spectabilis*, were tested as a protector of the plant against drought in subterranean clover plants.

Materials and methods

Seeds of *Trifolium subterraneum* ssp. *Brachycalycinum* cv Dom Dimis were surface disinfected with 2.5% NaClO, and sown in 4-l plastic pots containing soil substrate:sand (5:1). In the meantime, *Byssochlamys spectabilis* (Genbank accession number: KP899436) was grown at 23°C in dark conditions in PDB (Potato Dextrose Broth) to obtain the inoculum. Once the plants were correctly

established and fungus mycelium separated from spent broth, half of the plants were sprayed with the homogenized mycelium in sterile water and the rest were sprayed with sterile water to act as control treatment. The complete experiment was replicated 15 days later giving the name Trial 1 (older plants sown first) and Trial 2 (younger plants sown 15 days after Trial 1). All pots were placed on greenhouse benches and half of the pots of each treatment were subjected to deficit irrigation (approx. 70% of field capacity measured by difference of weight). The rest of the pots were regularly irrigated to maintain field capacity. Drought (deficit irrigation-DI) influence was estimated through the percentage of the plant affected by symptoms of decay visually observed in each pot (symptoms considered were yellowing and drying). The levels of influence were established as follows: 1, Healthy plant with bright green leaves; 2, Slight symptoms of drought (affecting less than 50% of the plant); 3, Half of the plant with yellow leaves and/or stems and beginning of dryness; 4, Almost dry with severe signs of decay; 5, Plant completely dead. After the observations, the herbage of the plants was harvested and taken to the lab for processing. Part of the samples were conserved to determine stoma size by means of a Scanning Electron Microscope (SEM) SU3800, and an aliquot was dried and ground to analyse acid detergent fibre (ADF) and crude protein (CP) by means of official methods and expressed on a dry matter basis. Milled freeze-dried leaves were subjected to colorimetric analysis to determine greenness and brightness of the leaves by using a Minolta Colorimeter CR-300. Total phenolic content (TPC) and antioxidant activity measured as DPPH free radical scavenging capacity (RSC) were obtained following the methods described by Yadav *et al.* (2014) and Liu *et al.* (2007) respectively. The effect of the variables was evaluated by analysis of variance (ANOVA) test or Kruskal-Wallis test when non-parametric data. Tukey test for multiple comparison was used when significant differences ($P < 0.05$) were found in the ANOVA. All these analyses were performed with the XLStat (Addinsoft, 2023) ‘add-on’ for Microsoft Excel.

Results and discussion

According to Kruskal–Wallis analysis, there was a great significant effect when analysing the data of physiological state of the plant; however, in the triple interaction trial×irrigation regime×fungus, the significantly lower data of physiological state of the plants subjected to field capacity irrigation (FCI) masked significant effects of the fungus. Thus, completing analysis only with deficit irrigation (DI) data, results showed that the protection of the fungus is present (E408) compared to the control treatment (E0) (Table 1).

Regarding stomatal size, ANOVA indicated that leaves of plants with FCI had significantly ($p < 0.001$) higher values, 7.2 μm on average, with reference to the 4.33 μm of the stomata in the leaves with DI. Results of analysis of the DI plants indicated that the fungal inoculation led smaller stomata size ($p < 0.001$), resulting in 3.6 vs. the 5.0 μm as an average of the E0 plants. Studying CP, only

Table 1. Kruskal–Wallis and comparison analysis for the interaction trial×fungus for plant physiological state

Source	Means of ranks in physiological state*
Trial 1	
E0	21.00a
E408	10.67b
Trial 2	
E0	7.83b
E408	10.50b

* $p < 0.0001$.

the main parameter fungus, resulted in a significant difference: E408 plants showed an average of 13.3% while E0 plants hardly reached 10.0%. Conversely, ADF in E408 plants were significantly lower (27.4%) than that of E0 plants (31.5%). However, the interaction irrigation×fungus indicated that no significant differences in ADF values were found between FCI plants and DI plants when they were inoculated with E408 (values ranged between 26.7% and 28.2%). In the case of E0-DI plants, the ADF values were significantly ($p < 0.0001$) higher (34.9%). With reference to TPC, younger plants (Trial 2) showed higher values of mg of gallic acid/g of fresh weight than older plants, the first ranging between 40 and 48, and the latest not reaching 32. When studying the interaction irrigation×trial, higher values of TPC were found in younger plants subjected to DI, showing more than 52 mg gallic acid/g fresh weight vs. less than 32 mg gallic acid/g fresh weight found in the rest of the treatments. Table 2 shows results for the triple interaction irrigation×trial×fungus, for the RSC values and colour parameter *a*, because L parameter was not significant. Higher RSC values were found in inoculated (E408) younger plants subjected to DI. Regarding the color, lower values of *a* parameter indicate greener colour, and FCI plants showed lower values than DI. The analysis also indicates that in both trials, when plants were subjected to DI, the inoculation with E408 resulted in greener plants.

All the results given above are in consonance with previous studies where endophytes can exert plant protection against drought, as simulated here by DI. Previously, Hubbart *et al.* (2014) found that inoculating root isolated endophytes in wheat plants, when well-watered, E+ and E—plants did not differ in some attributes such as height; the same as can be seen for colour here (in Trial 2, Table 2). Conversely, the effects of the endophytes appear to protect the plant when plants are under stress. In our case, lower fibre content was found for plants that were subjected to DI, they were inoculated with E408. Regarding the antioxidants, in Trial 2, inoculated plants subjected to DI, showed higher RSC, which is in accordance with Nie *et al.* (2024), who found higher antioxidant components when plants were inoculated with fungal endophytes and subjected to water stress. But not only physiological modifications, such as this difference in antioxidants, are generated by the endophytes to protect the plants against drought. Morphology of the stomata was previously studied by Aimone *et al.* (2023), who stated that one of the strategies of the endophytes to protect plants against drought was by reducing water expenditure, reducing the size of the stomata, as we also observed here.

Table 2. ANOVA results and Tukey analysis for RSC and *a* parameter in the triple interaction

Source	Trial 1				Trial			
	FCI		DI		FCI		DI	
	E0	E408	E0	E408	E0	E408	E0	E408
RSC (mg Trolox/g fresh weight)*	25.83b	25.83b	25.83b	25.83b	25.83b	25.83b	26.68b	35.10a
<i>a</i> *	−5.36e	−6.18f	2.08a	−3.38d	−6.46f	−6.22f	−1.76b	−2.14c

RSC, DPPH free radical scavenging capacity; FCI, field capacity irrigation; DI, deficit irrigation.
 * $p < 0.0001$.

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Winter grazing for lactating dairy cows: effects on pasture and organic milk production

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Abstract

With climate change, the grass growth in Western France will be higher in late autumn and late winter, when the pastures' load-bearing capacity is not always sufficient for grazing. This can create a high grass cover that leads to the deterioration of the pasture's quality if not utilized before spring. This study aims to assess whether winter grazing by dairy cows is a satisfactory way to exploit this grass and whether it can contribute to higher protein self-sufficiency for organic dairy farms. During 3 winters, 23 lactating cows of the Trévarez experimental farm herd grazed 3 hours per day to supplement a grass-silage-based diet. They produced 0–2.22 kg milk cow⁻¹ day⁻¹ more than a control group that remained indoors. To assess the impact of winter grazing, ungrazed zones were set in the middle of five paddocks each year. Both zones were sampled to estimate the forage mass, density (median = 250 kg DM cm⁻¹) and nutritive values (crude protein of 19.8%), showing that the winter grass is a valuable roughage for lactating cows. The botanical composition and cover degradation were assessed visually, showing that winter grazing had no deleterious effect on the pastures the following year.

Keywords: dairy cows, organic, winter grazing

Introduction

Climate change is modifying the grass growth in Western France (Moreau *et al.*, 2020). There will be more growth at the end of autumn, at a time when the bearing capacity of soils is often not good enough to enable utilization of this grass. Winter grazing seems to be a suitable way to valorize this grass, thus preventing the presence of a large amount of biomass at the beginning of spring that might result in reduced herbage nutritive value (Holmes *et al.*, 1992). On the other hand, winter rations in organic dairy farms are often limited in terms of protein intake (Martin *et al.*, 2024). Few solutions are viable to enrich the cows' feed in winter. Protein-rich concentrates are expensive. Farm-produced concentrate can be used, but its nutritive value is too variable (Pelzer *et al.*, 2014). Grass, with its richness in protein (18–20% of crude protein), could be grazed to ensure a complementary supply of protein (INRAE, 2018). This study aims to assess if winter grazing is an appropriate practice in terms of its effects on animal performance and impact on the pasture.

Materials and methods

This study was repeated during 3 winters (2022–2025; winter being recognized as the period between the end of November and the beginning of March) during 59, 69 and 69 days, on the Trévarez organic experimental farm (Saint-Goazec, France). Each year two groups of 20 to 23 crossbred dairy cows on average (Prim'Holstein×Normande×Jersey) were studied. The control group (C) had a grass-silage-based ration (Table 1). The test group (T) was fed the same ration and was maintained on a pasture for 3 hours per day on paddocks of 0.47 ha on average. The grazing was organized in two rotations:

the first of two days per paddock, the second of only one day per paddock. The milk production of each cow was monitored daily and analysed through one mixed linear model (MLM) per year. Fixed effects were the group (C/T), the parity and the milk production during the 3 weeks prior to the study. The random effect was the individual animals' identification. The amount of dry matter (DM) of forages fed to the cows was weighed daily. The amount (as DM) of feed refusal was determined 3 times each week. The grass intake was estimated by the Herb'Valo method (Delagarde *et al.*, 2017). Forages and concentrates were sampled every 15 days to determine nutritive values. Each pasture was sampled before grazing to measure the grass nutritive values.

Ungrazed zones (UZ) were delimited in the middle of 5 pastures per year. Grazed zones (GZ) and control zones were sampled four times (before, in the middle, at the end of the study, and before the spring grazing rotation) to measure biomass, density and nutritive values of grass. Grass samples were all cut at 5 cm, by hand. Botanical composition (in 3 categories: grass/legumes/others) was assessed visually. Cover degradation was assessed visually at 5 fixed points (by GPS) in the GZ and on the UG of 3 pastures during the last 2 years, at the same measurement periods as described above and before and after each grazing. Herbage height was measured every two weeks with a platometer (Jenquip Manual Platometer).

Results and discussion

The mean pre- and post-grazing heights were in the ranges 5.9 to 7.1 and 4.4 to 5.6 cm., respectively. The pasture swards were >80% grasses. The mean dry matter content was $15.8 \pm 2.3\%$. The mean crude protein (CP) and organic matter digestibility were, respectively, 19.8 ± 3.5 and $67.2 \pm 5\%$. The median densities of grass in the 3 winters were, respectively, 249, 255 and 219 kg DM cm⁻¹ ha⁻¹. The lesser density observed during the last year might be due to the greater percentage of younger pastures and was ignored. The density measured is in the same range as the French references (Defrance *et al.*, 2004). Using a density value of 250 kg DM cm⁻¹ ha⁻¹, the mean grass growth ranged between 2 and 7 kg DM day⁻¹ ha⁻¹.

The MLM showed significant ($P < 0.05$) effects on milk production for T compared to C cows in the two first winters: +2.25 (estimates of 11.39 and 13.64, both SE of 0.86 kg cow⁻¹ day⁻¹) and +1.49 kg cow⁻¹ d⁻¹, (estimates of 14.66 and 16.15, SE of 1.11 and 1.19 kg cow⁻¹ day⁻¹) but no significant difference for the third winter (mean milk productions: 14.3 kg cow⁻¹ day⁻¹ and 15.7 kg cow⁻¹ day⁻¹). The milk response during the first winter can be explained by the lowest intake of grass silage (Table 1). This forage had a high mineral concentration of 18.7% of DM, which is probably a sign of a severe contamination with soil. As a result, the grass intake seems to have compensated for its ingestion. The difference in milk response between the last two winters is not explained by differences in ingestions or nutritional values of stocked feed, which are similar.

Before the first spring rotation in March, the average biomass was lower for GZ than UZ by 280 kg DM ha⁻¹. The estimated cows' intakes ranged from 300 to 514 kg DM ha⁻¹. Once grass growth resumed in spring, the growth rate in UZ was superior to the GZ, but only during the first 15 days of March and they were equivalent after that. These elements suggest that winter grazing has no or very little impact on the herbage yield of pasture swards. Winter grazing also had no impact on the botanical composition, with 77% and 81% grasses in GZ and UZ in the spring. As regards the cover degradation, some pastures (the most recently sown) reached up to 50% of bare soil at the end of the

Table 1. Intake in control and test groups (expressed as kg DM cow⁻¹ day⁻¹)

	Year 1	Year 2	Year 3
Estimated intake of the control group			
Grass silage	7.0	8.1	8.6
Maize silage	4.8	5.1	4.8
Alfalfa haylage	1.1	1.7	2.2
Triticale-oat-pea mixture	2.3	2.6	2.2
Minerals	0.2	0.1	0.2
Total ingestion of the control group	14.5	16.7	16.8
Differences in estimated intake for the test group (test-control): positive meaning higher in the T group			
Of stocked feed	0	-0.8	-1.1
Of grass	+3.3	+2.5	+2.0
Difference in total ingestion	+3.2	+1.7	+0.9

two rotations. Yet, there was no difference in cover degradation in spring between UZ and GZ zones on all studied pastures.

Conclusion

The general increase in milk production and the absence of deleterious effects on the pastures indicates that winter grazing may be beneficial to organic dairy farms.

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Timing of spring fertilization affects grassland yield and herbage feed quality

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Abstract

Climate change, marked by rising temperature, has led to earlier grass growth and an extended growing season. This study investigated how timing of spring fertilization and the first cut affects yield and forage quality in grassland on Norway's west coast. Early spring fertilization was applied at the estimated start of growth, while normal timing occurred 3–4 weeks later. The early first cut was taken at 450 growing degree days (GDD), and the standard cut followed 75 GDD later. Second cuts were made 500 GDD after the first, and third cuts occurred in late August. Early spring fertilization increased first-cut yield by 39% and annual yield by 15% compared to normal timing. It also improved N-use efficiency by 15%, measured as dry matter yield per kg total N applied. Although early cutting slightly reduced annual yield, it enhanced forage quality, both protein and fibre content. The yield benefit from early spring fertilization was concentrated in the first cut, with no gain in later cuts. While early fertilization can boost productivity and better utilize the growing season, it may come at the cost of reduced forage quality.

Keywords: harvesting time, herbage quality, ley, nitrogen fertilization

Introduction

Climate change has become increasingly evident since the previous standard measurement period (1961–1990). In the new normal (1991–2020), the most notable difference is the rise in average temperatures during autumn and winter. For example, at Fureneset, a representative agricultural research station for coastal areas in Western Norway, the annual average temperature has increased by 0.7°C, with the most pronounced seasonal rise occurring from January to March. During this period, the average temperature has risen from 1.8°C to 2.5°C. April to June remains the driest part of the year, coinciding with peak solar radiation. While a longer growing season extends favourable temperatures into autumn, plant growth is limited by low light levels. Additionally, heavy autumn rainfall complicates harvesting operations. Western Norway's dry and cold spring conditions allow more time for the slow-acting manure to take hold. Therefore, early application of manure and fertilizer may enhance biomass production. Previous studies conducted in Ireland, UK, and Norway have shown a small but significant increase in dry matter yield with early application of mineral N fertilizer (Håland, 1978; Stevens *et al.*, 1989; Long *et al.*, 1991). Since the Norwegian study was carried out, harvesting frequency has increased and different grassland species are now in use. A combination of early fertilization and an early first cut may improve both annual herbage feed quality and the distribution of yield across cuts. However, the anticipated improvements in herbage yield and forage quality from early fertilizer application and harvesting remain largely unconfirmed. Therefore, the objective of the present study was to address this knowledge gap by testing the effects of spring fertilizer timing and harvesting strategy on herbage yield and feed quality.

Materials and methods

A field trial was conducted using a first production year of a ley composed of perennial ryegrass (*Lolium perenne*), festulolium (*Lolium* spp. $\times$ *Festuca* spp.), timothy (*Phleum pratense*), and cocksfoot (*Dactylis glomerata*) at the Fureneset research station (EUREF89N 61°17.6'E 5°2.9', 10 m asl), on a sandy loam soil. The experiment followed a 2 $\times$ 2 factorial design, testing two spring fertilization timings (Early or Normal) and two first-cut harvest timings (Early or Standard) across three seasonal cuts. Early Spring fertilization was applied at the estimated onset of growth, while normal fertilization occurred 3–4 weeks later. Growth onset was defined as the first date following three consecutive five-day periods with mean daily soil temperature at 5 cm depth above 5°C. Early first cut was defined as harvesting at 450 growing degree days (GDD, base temperature 0°C) after onset of growth, while the standard first cut occurred 75 GDD later. The second cut was taken 500 GDD after each respective first cut, and the third cut was performed at the end of August across all treatments. Fertilised plots measured 2.8 m $\times$ 8.5 m, with a harvest plot area of 1.5 m $\times$ 7 m. The experiment ran for three years (2022–2024). Mineral fertiliser (YaraBela™ Opti-NS 27-0-0 (4S)) was applied at rates 70, 50 and 50 kg N ha⁻¹ in spring, after first, and after second cut, respectively. Cattle manure was also applied, equivalent to 170 kg total-N ha⁻¹ in spring and 87 kg total-N ha⁻¹ after the first cut. Plots were harvested using a Haldrup grass harvester (J. Haldrup, Løgstør, Denmark) to a stubble height of 7 cm. Herbage samples were dried at 60°C for 48 h, milled to 1 mm, and analysed for dry matter (DM), ash, CP (N; AOAC method 2001.11; AOAC (2023)) and ash-corrected NDF_{om} (Ankom200 Fiber Analyzer). Data were analysed using the GLIMMIX procedure in SAS, with fertilization timing (F), harvest timing (H), cut number (C), and their interactions as fixed effects, and year and replicate as random effects. Repeated measures were accounted for using the RANDOM statement. Significance of effects were declared at $P \leq 0.05$ and trends at $0.05 < P \leq 0.10$.

Results and discussion

On average across all years, early fertilization increased the DM yield by 1.56 Mg ha⁻¹ in the first cut and by 1.32 Mg ha⁻¹ annually (Table 1). Although grassland soils can contain substantial amounts of N, the availability of mineral N in spring is typically low. This is because organically bound N must first be mineralized, a temperature-dependent process, before it becomes accessible to plant roots (Macduff and White, 1985). The positive effect of early fertilization is therefore attributed to an increased supply of readily available N, which reinforces itself as leaf mass expands with greater light interception, enhanced growth and improved nutrient uptake from the soil. However, early spring fertilization resulted in lower yields in the second cut than normal timing. Similar findings were reported by Stevens *et al.* (1989), who observed that late spring fertilization produced a slightly larger second cut compared to early fertilization. The yield benefit of early spring fertilization was thus confined to the first cut, with no significant effect on the third cut. An early first cut led to an average yield reduction of 0.90 Mg DM ha⁻¹ in the first cut and 0.83 Mg DM ha⁻¹ in total annual yield compared to the standard timing. Harvesting first cut at an early developmental stage contributed 48% to the annual yield, while standard first cut contributed 53%. Notably, the timing of spring fertilization had stronger influence: with early spring fertilization, the first cut accounted for 55% of the annual yield, compared to 45% with normal timing. Leaf proportion of total biomass tended to be lower following early fertilization compared to normal timing. Early first cut increased the leaf proportion in the first cut but reduced it in the third relative to the standard cutting schedule. On average, Early Spring fertilization resulted in 15% higher annual N-use efficiency than Normal timing (24.2 *vs.* 21.0 kg DM kg⁻¹ N applied, $P < 0.001$). Harvesting at the Standard time for first cut led to a 17% higher N use

efficiency than harvesting Early (21.9 *vs.* 18.1 kg DM kg⁻¹ N applied, *P* < 0.001). Early fertilization resulted in a 30 g kg⁻¹ DM lower herbage CP content in the first cut compared to normal timing, with only minor effects observed in the subsequent cuts (Table 1). The reduced CP content in the first cut following early spring fertilization is likely due to a dilution effect caused by increased yield (Reyes *et al.*, 2015). Fibre content is primarily influenced by the phenological development of the plants, which is largely governed by day length and temperature. Therefore, grassland harvested on the same day but subjected to different N fertilization regimes will generally exhibit similar fibre content (Termonen *et al.*, 2020). This explains the absence of any significant effect of spring fertilization time on NDF_{om} content (Table 1). Herbage feed quality after early spring fertilization combined with an early first cut would likely have benefitted from a shorter regrowth period between cuts and potentially a fourth cut. However, a more frequent mowing pattern may decrease DM yield.

Table 1. Herbage DM yield, yield distribution, leaf proportion of total biomass, and chemical content in grassland fertilized (F) at growth start in spring (Early F) or at normal time (Normal F), harvested (H) early (EH) or at standard (SH) time in the first cut.

Item	Cut no. (C)	Treatments				Significance ^a (<i>P</i> <)						
		Early F		Normal F		SE	F	H	F × H	C	C × F	C × H
		EH	SH	EH	SH							
DM yield, (Mg ha ⁻¹)	1	5.08	6.04	3.58	4.42	0.177	0.001	0.01	ns	0.001	0.001	0.001
	2	2.66	2.98	3.09	3.21							
	3	2.22	1.82	2.01	1.83							
Leaf biomass of DM yield (g g ⁻¹)	1	0.43	0.34	0.42	0.37	0.074	0.10	ns	ns	0.001	ns	0.05
	2	0.49	0.46	0.49	0.45							
	3	0.64	0.71	0.73	0.75							
CP (g (kg DM) ⁻¹)	1	119	99	148	129	6.4	0.001	ns	ns	0.001	0.001	0.001
	2	117	106	111	106							
	3	96	109	98	116							
NDF _{om} (g (kg DM) ⁻¹)	1	497	530	481	533	11.2	ns	ns	ns	0.001	ns	0.01
	2	496	498	504	506							
	3	554	526	530	537							

Average across three production years, *n* = 9.
^aThree-way interaction (F × H×C) was not significant for any of the variables.

Conclusion

Early spring fertilization gave higher annual DM yield and N utilization, but lower herbage crude protein content. In farm situations where area is limited, early spring fertilization combined with standard harvesting timing for high dry matter yield can be recommended. Otherwise, normal fertilization timing an early harvesting regime aimed at high herbage quality is likely the best strategy.

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Investigating the use of drone imagery to detect disease in perennial ryegrass swards

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Abstract

Disease incidence in perennial ryegrass is a growing threat to Irish grassland systems, especially as the anticipated changes in climate are expected to increase weather conditions conducive to disease development. A study investigating the incidence of disease was conducted on perennial ryegrass plots that were experiencing high disease pressure from Crown Rust (*Puccinia coronata*) and *Drechslera*. Eighty-five candidate perennial ryegrass varieties were assessed for disease prevalence, herbage yield and nutritive quality. Disease scoring was visually assessed as the percentage of the plot infected by disease, multiplied by the percentage of leaf infected. Drone images were obtained and used to calculate the Green Normalised Difference Vegetation Index (GNDVI). Regression models highlighted negative relationships between disease scores and herbage yield and nutritive quality variables. Higher disease scores corresponded strongly to lower GNDVI ($r = -0.80$). Varietal differences were observed for disease resistance with corresponding increases in herbage yield, quality and greenness for varieties with greater resistance. On commercial farms, an indication of varietal disease resistance will be valuable for future agricultural production. Practical implications suggest that drones may provide a tool for detection and monitoring of disease.

Keywords: crown rust, disease, drone, GNDVI, multispectral imaging, perennial ryegrass

Introduction

In Ireland, warmer and wetter conditions are expected to become more common due to climate change (Nolan & Flanagan, 2020), leading to increases in diseases like Crown Rust, which negatively impacts perennial ryegrass herbage yield and quality. Moisture retention and reduced nitrogen are also known to promote fungal spread (Critchett, 1991), and reduced permissible nitrogen application rates may further increase disease development. The ability to identify disease incidence could help future-proof grassland farming. Drone technology may potentially be a useful monitoring tool, allowing for early detection and giving farmers opportunities to respond before herbage yield and quality are significantly reduced. Traditionally, Crown Rust presence in pastures is only detected once visible symptoms appear and quality has declined. Currently, disease monitoring relies heavily on visual scoring; however, this is labour-intensive. There is an increasing need for early, technology-driven methods of disease detection, such as drone-based monitoring. The objective of the study was to use multispectral drone imagery to investigate whether disease incidence can be detected through Green Normalized Difference Vegetation Index (GNDVI) and investigate the effect of disease on perennial ryegrass dry matter yield (DMY) and dry matter digestibility (DMD).

Table 1. Summary statistics for disease severity, for dry matter yield (DMY), dry matter digestibility (DMD) and Green Normalized Difference Vegetation Index (GNDVI).

Variable	Min	Max	Mean	SD
Disease Score	3.6%	90%	27.9%	18.4%
% plot infected	26.7%	100%	71.3%	17.3%
% infection/leaf	10%	90%	40.2%	16.1%
DMY (kg DM/ha)	864	3570	1989	453.52
DMD (g/kg)	591.3	866.55	799.6	39.7
GNDVI	−0.045	0.142	0.053	0.029

Materials and methods

The area analysed consisted of 200 plots containing 88 different perennial ryegrass (PRG, *Lolium perenne*) variety crosses, established in 2023 in a randomized complete block design with 2–3 replicates, with the aim of selecting new varieties. Sampling took place in October 2024 in Teagasc Oakpark, Carlow, Ireland. Herbage yield was measured for each plot by cutting a 0.25 m² area within a quadrat using Gardenia hand shears to approx. 4 cm. The fresh weight was recorded and a 100 g sub-sample was oven dried at 90°C and reweighed to calculate DMY. Dry matter yield (kg DM/ha) was calculated as: (Herbage fresh weight (kg)×DM%)×40 000. The herbage sub-sample was milled through a 1 mm screen using a Cyclotech 1093 Sample Mill (Foss, Hillerød, Denmark) and analysed using near infrared reflectance spectrometry (NIRS) for estimation of DMD and water-soluble carbohydrates (WSC). On the day of sample collection, disease scoring was visually assessed, based on discoloration and leaf damage, by 3 trained individuals estimating the % of the plot infected and % leaf area affected (disease score = % of the plot infected×% leaf area affected). Disease scoring was carried out once when the disease burden was at peak, similar to Arojju *et al.* 2018, who reported this is sufficient to evaluate disease severity and susceptibility. Imagery was captured at flight heights of 12 m and 50 m using a DJI Mavic 3 Enterprise drone. Images were processed using DroneDeploy to generate GNDVI values. Statistical analysis was undertaken using linear regression and ANOVA in RStudio and R version 4.4.1.

Results and discussion

Disease severity was widespread and highly variable, as summarised in Table 1. Varieties differed significantly in disease severity, highlighting a genetic basis for resistance (Kimbeng, 1999), indicating that breeding resistant varieties remains a sustainable management strategy.

Considerable variability was seen in DMY and DMD between varieties. Regression analysis indicated that both DMY and DMD were negatively associated with disease score (Figure 1). The relationship between DMD and disease score was strong ($r = 0.72$), while the DMY and disease relationship was weaker, but still significant, with yield decreasing by approximately 6.2 kg/ha for each percentage increase in disease score ($r = 0.25$). The results are in agreement with Potter, (1987) who found higher disease scores directly reduced yield and quality. This reduction of DMY with increasing incidence of disease ultimately limits overall herbage production and reduces the potential for producing animal feed, particularly during autumn when grass growth rate can be limiting (O'Donovan *et al.*, 2016).

The GNDVI calculated from drone imagery ranged from −0.045 in diseased plots to 0.14 in healthier ones. It was positively correlated with both DMD ($r = 0.66$, $p < 0.001$) and DMY ($r = 0.32$,

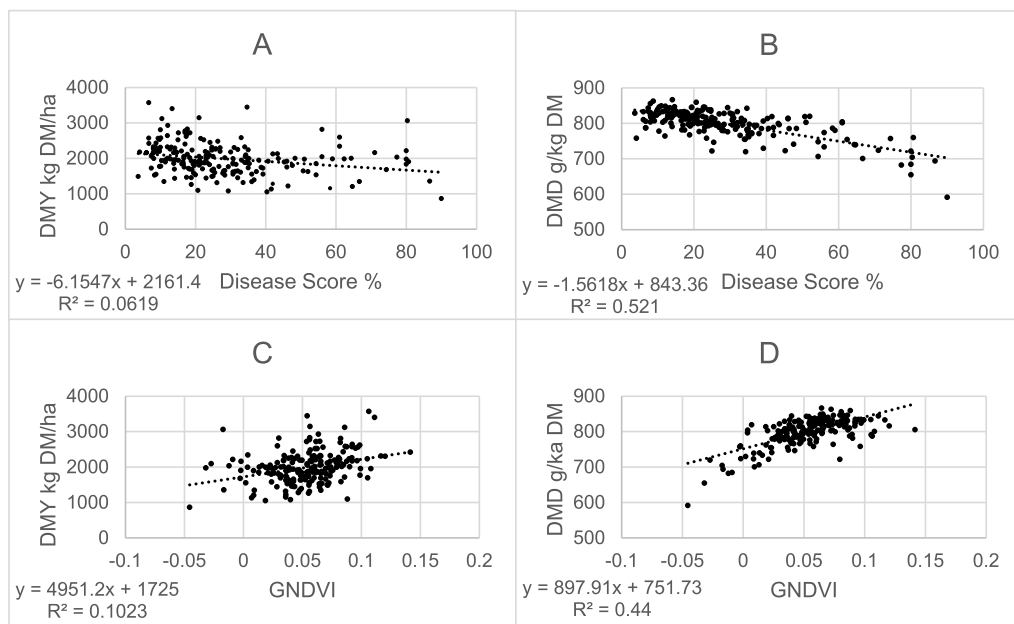


Figure 1. Relationship between (A) Dry Matter Yield (DMY; kg DM/ha) and disease score, (B) Dry Matter Digestibility (DMD, g/kg DM) and disease score. (C) DMY and Green Non-Differentiated Vegetative Index (GNDVI) and (D) DMD and GNDVI.

$p < 0.001$), suggesting that plant greenness is related to herbage quality and productivity. Varietal differences were statistically significant ($p < 0.001$) for disease score, WSC and GNDVI. Drone imaging is affected by flight height, light, and weather, and while GNDVI captured stress well, more research is required to investigate whether drone imagery can reflect specific disease symptoms.

Conclusion

Increased disease scores reduced perennial ryegrass herbage yield and quality. Drones present themselves as a rapid, non-invasive tool for monitoring disease incidence. Under increasing climate pressure, monitoring and developing resistant varieties will be key technology to managing future disease threats. Drone imaging can play a large role in the breeding and evaluation of resistant varieties.

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Predicting the impact of climate change on grass growth in the Republic of Ireland

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Abstract

The objective of this study was to simulate grass growth across the Republic of Ireland under potential climate change scenarios. Future grass growth was simulated using the Moorepark St Giles grass growth model and weather scenarios provided by Met Éireann's TRANSLATE project. Daily grass growth was simulated with a resolution of 1 km² for a typical year under five different climate change scenarios ranging from 1.5–4.0°C of global warming above a historical baseline. Annual national average yields are projected to increase by 0.4–1 t DM ha⁻¹ (+ 3.4–9.2%), varying by scenario. Seasonal gains are concentrated in the spring (+ 3.4–13.7%) and autumn (+ 8.4–18.0%) with summer yields relatively static (+ 0.0–1.6%). However, the impacts of climate change on yield differ markedly between regions. Summer yield in the NUTS 2 Eastern & Midland region reduced by 0.7–4.2%, while it increased by 1.3–4.0% in the NUTS 2 Northern & Western region. Region-specific adaptation strategies will be essential to maintaining the resilience of Irish grassland systems.

Keywords: adaptation, climate change, grassland resilience, simulation

Introduction

In the Republic of Ireland (ROI), 92% of the utilised agricultural area is dedicated to either grassland or rough grazing. The temperate maritime climate allows an extended grass-growing season and non-irrigated yields which are among the highest in Europe, with peak grass growth typically occurring between May and August. Climate change is projected to alter rainfall patterns and increase temperatures in the ROI, with warmer, drier summers and warmer, wetter winters projected over the coming century (O'Brien & Nolan, 2023). In order to assess the future consequences of climate change on Irish grasslands, this study simulated daily grass growth under different potential future climate scenarios across the ROI.

Materials and methods

The Moorepark St Giles grass growth (MoSt GG) model was used to predict the impact of future climate change on grass growth in Ireland using weather scenarios from Met Éireann's TRANSLATE project. MoSt GG is a dynamic, mechanistic model which simulates the growth of perennial ryegrass (*Lolium perenne*) swards with a daily time step (Ruelle *et al.*, 2018). It has been widely evaluated in the Irish context. Daily precipitation, as well as daily maximum and minimum temperature from Met Éireann's TRANSLATE project were used to simulate grass growth under different climate scenarios at the 1 km² resolution (O'Brien & Nolan, 2023). The six scenarios selected consisted of a historical baseline scenario (1976–2005) and five global warming levels of 1.5, 2.0, 2.5, 3.0, and 4.0°C above this baseline. It should be noted that the ROI is projected to warm slower than other regions, with 4.0°C of global warming associated with a warming of 2.2°C in the ROI. The weather data represent

a typical annual profile for each scenario, without accounting for intra-annual variability. Simulations were carried out at the paddock level, with one paddock for each of the 77 083 cells in the TRANSLATE grid. Each paddock was grazed eight times from mid-February to mid-October, with 150 kg of nitrogen applied in six separate applications on fixed days, representative of a typical Irish grazing system. All simulations were carried out on a free draining soil.

Results and discussion

Grass yield increased from a national average of 11.6 t DM ha⁻¹ under the historical baseline scenario to 12.7 t DM ha⁻¹ under the 4.0°C scenario (Figure 1). The predicted impacts varied across the country, with yield increases ranging from 0.6 t DM ha⁻¹ at the 10th percentile to 1.6 t DM ha⁻¹ at the 90th percentile, under the 4.0°C scenario. A pronounced increasing east-west yield gradient emerged beyond 3.0°C of global warming, areas with annual yields exceeding 12.7 t DM ha⁻¹ were almost exclusively in the West and along the South coast.

The monthly distribution of grass growth also shifted under climate change scenarios (Figure 2). Under the 2.5°C warming scenario, the median impact across the country in August was a decrease of 61 kg DM ha⁻¹ (−3.4%) per month relative to the baseline scenario. However, the monthly impact of climate change on yield was also highly dependent on location, as the 10th and 90th percentile impacts in July under the 2.5°C warming scenario were a decrease of 145 kg DM ha⁻¹ (−8.2%) and an increase of 203 kg DM ha⁻¹ (+12.0%), respectively.

In the Eastern and Midland region, summer yields declined under all climate change scenarios, whereas the Northern and Western region showed increases across all scenarios, relative to the baseline (Table 1).

Conclusions

Although annual yields are projected to increase, changes in the seasonal distribution of grass growth will require adaptation of management practices. Increased spring and autumn growth highlights the importance of grazing infrastructure to maximise grass utilisation during these periods. Further research examining the subsequent impacts of the changing seasonal distribution of grass growth on farm level strategies, such as optimal stocking rate, is required to improve the resilience of the Irish

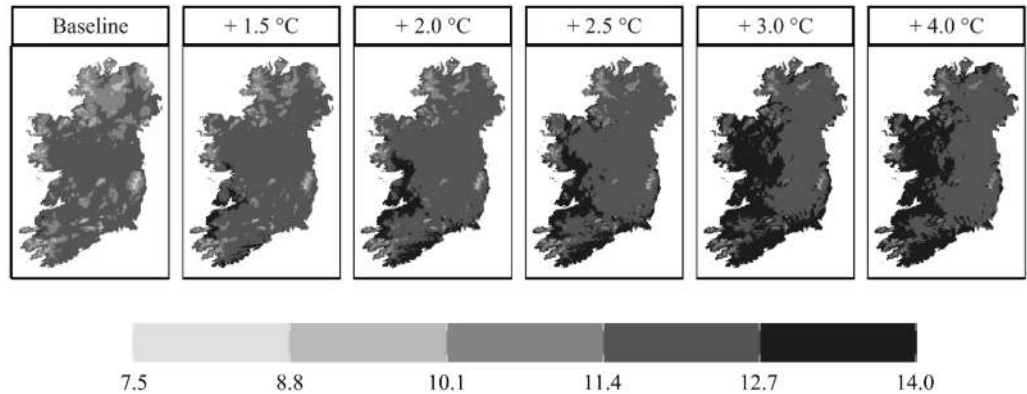


Figure 1. Annual yield (t DM ha⁻¹) across the ROI under different Global Warming Levels.

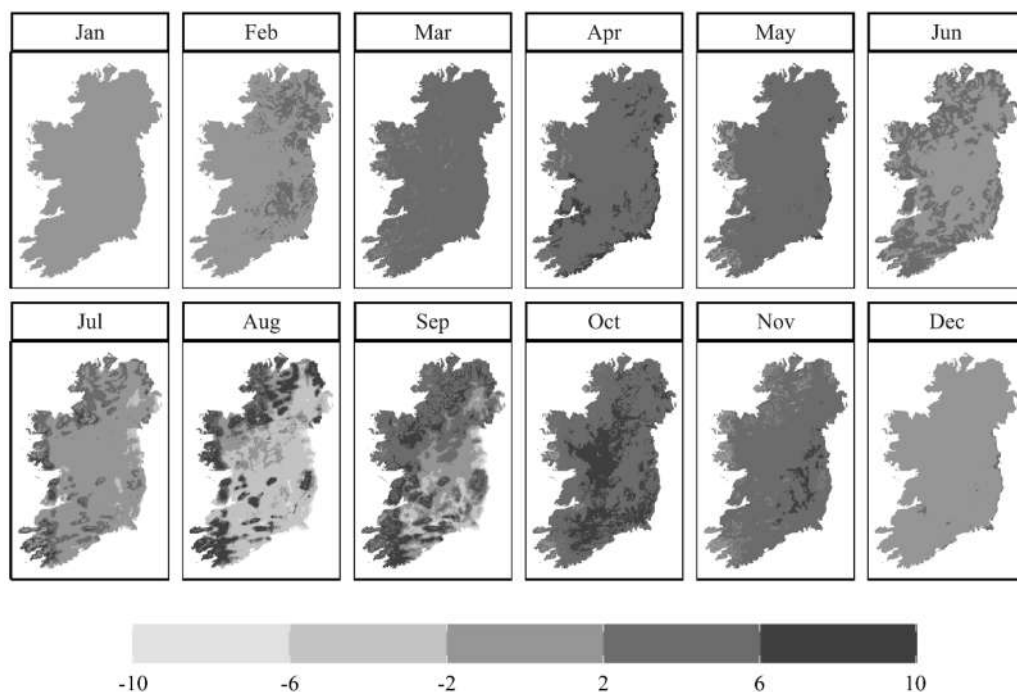


Figure 2. Change in average daily yield (kg DM ha⁻¹) between the baseline scenario and 2.5°C Global Warming Level by month.

Table 1. Mean yield (t DM ha⁻¹) and percentage change relative to the baseline for each Global Warming Level by season and NUTS 2 region

Region	Season ¹	Baseline	+1.5°C	+2.0°C	+2.5°C	+3.0°C	+4.0°C
Northern and Western	Spring	3.3 (-)	3.4 (3%)	3.5 (6.1%)	3.6 (8.5%)	3.7 (12.3%)	3.8 (12.9%)
	Summer	5.3 (-)	5.4 (1.3%)	5.5 (2.7%)	5.5 (2.9%)	5.6 (4%)	5.5 (3.5%)
	Autumn	2.2 (-)	2.4 (8.9%)	2.5 (13%)	2.6 (15.8%)	2.7 (19.4%)	2.7 (20.8%)
	Winter	0.5 (-)	0.5 (10.3%)	0.5 (18%)	0.6 (23.8%)	0.6 (29.3%)	0.6 (32.2%)
Eastern and Midland	Spring	3.4 (-)	3.6 (4.2%)	3.7 (7.4%)	3.8 (9.9%)	3.9 (14.1%)	3.9 (14.6%)
	Summer	5.3 (-)	5.2 (-1.2%)	5.3 (-0.7%)	5.2 (-1.6%)	5.1 (-2.8%)	5.1 (-4.2%)
	Autumn	2.4 (-)	2.6 (7.7%)	2.7 (11.8%)	2.7 (13.6%)	2.8 (15.3%)	2.8 (15.6%)
	Winter	0.5 (-)	0.5 (13.2%)	0.6 (21.6%)	0.6 (28.4%)	0.6 (35.8%)	0.7 (39.8%)
Southern	Spring	3.4 (-)	3.6 (3.4%)	3.7 (7.2%)	3.8 (9.5%)	3.9 (13.6%)	3.9 (13.9%)
	Summer	5.3 (-)	5.4 (0.7%)	5.4 (1.7%)	5.4 (1.4%)	5.4 (1.1%)	5.3 (-0.9%)
	Autumn	2.4 (-)	2.6 (8.3%)	2.7 (11.6%)	2.7 (13.7%)	2.8 (16.4%)	2.8 (17%)
	Winter	0.5 (-)	0.6 (10.9%)	0.6 (18.1%)	0.7 (23.8%)	0.7 (29.8%)	0.7 (32.9%)

¹ Spring: Mar–May, Summer: Jun–Aug, Autumn: Sep–Nov, and Winter: Dec–Feb.

pasture based system. In addition, further investigation is needed to assess potential increases in intra-annual variability in grass growth.

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Soil bacteria diversity responses to long-term fertilization and liming in acid grasslands

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Abstract

Long-term nutrient enrichment and liming influence soil microbial communities in temperate grasslands. We studied the effects of six decades of fertilization and liming at the Zachenschöberl Grassland Experiment in Austria, using a factorial design with phosphorus (P), potassium (K), nitrogen (N), and lime treatments. Soil bacterial communities (via 16S rRNA gene sequencing) were assessed across seven treatments and in an unfertilized control. Bacterial alpha diversity (Shannon, observed richness) was lowest in control and in unlimed NPK treatments and highest in PK and limed plots. Beta diversity analyses revealed clear treatment-driven clustering of microbial communities. Acidophilic taxa (e.g. *Acidobacteriales*) dominated control soils, while fertilized plots were enriched in *Gaiella*, *Bradyrhizobium*, and *Chloroflexi* KD4-96. Liming and P+K enhanced microbial diversity and altered community structure. Our results highlight the divergent trajectories of grassland biodiversity under prolonged fertilization and suggest that moderate nutrient input together with liming can balance productivity with ecological sustainability.

Keywords: gene, genera, legumes, liming, microbial communities, nutrient enrichment

Introduction

Soil acidification from long-term fertilization alters both plant and microbial communities, yet their combined responses in permanent grasslands remain underexplored. Nutrient enrichment, particularly nitrogen, often promotes dominance of competitive grasses and reduces diversity by excluding forbs and legumes (Ward *et al.*, 2017). Historic experiments, including Park Grass (UK), have shown that even liming could not fully reverse these species losses (Ward *et al.*, 2017). Nevertheless, liming can alleviate acidity, improve nutrient availability, and favour colonization by pH-sensitive species like clover (Ward *et al.*, 2017). Microbial communities are also pH-sensitive. *Acidobacteria* dominate under low pH, while higher pH supports greater bacterial richness and copiotrophic taxa (Wenyika *et al.*, 2025). In a liming trial, bacterial diversity nearly doubled as pH increased from approx. 4 to 8 (Rousk *et al.*, 2010). Fertilization may increase nutrients and organic inputs but often leads to acidification and microbial diversity decline (Yang *et al.*, 2021). The extent to which liming can offset these microbial shifts remains uncertain. Moreover, feedbacks are expected between plant and microbial communities: changes in plant functional groups (grasses, forbs, legumes) may alter microbial substrates and interactions. To address this, we used selected treatments at the long-term Kalkdünungsversuch Zachenschöberl grassland experiment in Austria (est. 1964; pH approx 4.5; 1297 m asl) to assess fertilization and liming impacts on microbial diversity. We hypothesized that (1) long-term

fertilization favours acid-tolerant microbial taxa, and (2) liming increases diversity by improving soil pH and supporting pH-sensitive groups.

Materials and methods

The long-term fertilization experiment was set up in 1964 in mountain meadow, province of Styria (Austria), (47°27'48"N;14°4'19"E; altitude 1297 m asl). All treatments were cut regularly two times a year. For the study seven treatments were selected (Table 1).

Table 1. Overview of selected treatments at the Zachenschöberl Grassland Experiment, including fertilizer (N, P, K annually) and mixed agricultural lime (every two years).

Treatment code	Treatment name	Description
Control	Control	No fertilizer or lime
11	PK	Phosphorus (21 kg P ha ⁻¹) and potassium (100 kg K ha ⁻¹) fertilizer
12	PKCa ₅₀₀	PK+500 kg ha ⁻¹ mixed agricultural lime
14	PKCa ₁₅₀₀	PK+1500 kg ha ⁻¹ mixed agricultural lime
21	NPK	Nitrogen (80 kg N ha ⁻¹)+P and K fertilizer
22	NPKCa ₅₀₀	NPK+500 kg ha ⁻¹ mixed agricultural lime
24	NPKCa ₁₅₀₀	NPK+1500 kg ha ⁻¹ mixed agricultural lime

DNA was extracted (Qiagen PowerSoil) from 0.25 g subsamples. The V4 region of the 16S rRNA gene was amplified using primers 530F/802R and sequenced on the Ion Torrent PGM platform. PCR products were purified (Ampure XP), quantified (Qubit), and barcoded. PCoA based on Bray-Curtis dissimilarity visualized microbial community structure. The alpha diversity metrics (Observed, Shannon) were computed on rarefied data. Principal Coordinates Analysis (PCoA) was used to compare communities across various samples. The Bray-Curtis dissimilarity metric was employed to measure the differences between the communities based on their relative abundances without any rarefaction. QIIME 2 outputs were processed using the phyloseq R package.

Results and discussion

Soil bacterial diversity followed a pH-driven pattern. Alpha diversity (Shannon H') was lowest in the unfertilized, unlimed control (approx. 5.2) and highest in limed plots (approx. 6.0). NPK without lime slightly reduced microbial diversity, likely due to acidification (pH approx. 4.3) and dominance by a few nutrient-exploitative taxa (Figure 1). Fertilization plus liming maintained high microbial diversity (H' approx. 5.8), indicating that pH correction buffered the effects of long-term fertilization. Because sampling was conducted in September, shortly after the main growing season and following biomass removal, soil conditions likely reflected both peak nutrient uptake and seasonal shifts in microbial activity. Microbial diversity patterns may therefore partly capture post-harvest recovery processes and the accumulation of organic residues, which could accentuate treatment effects. Liming shifted communities toward more diverse, neutrophilic assemblages.

Conclusion

Decades of fertilization and liming significantly altered both microbial communities in this mountain grassland. Soil bacterial diversity was closely linked to pH; unlimed plots had low diversity dominated by acidophilic taxa, whereas limed plots supported richer, more balanced communities. Liming improved microbial diversity and shifted its composition but did not fully reverse fertilization impacts.

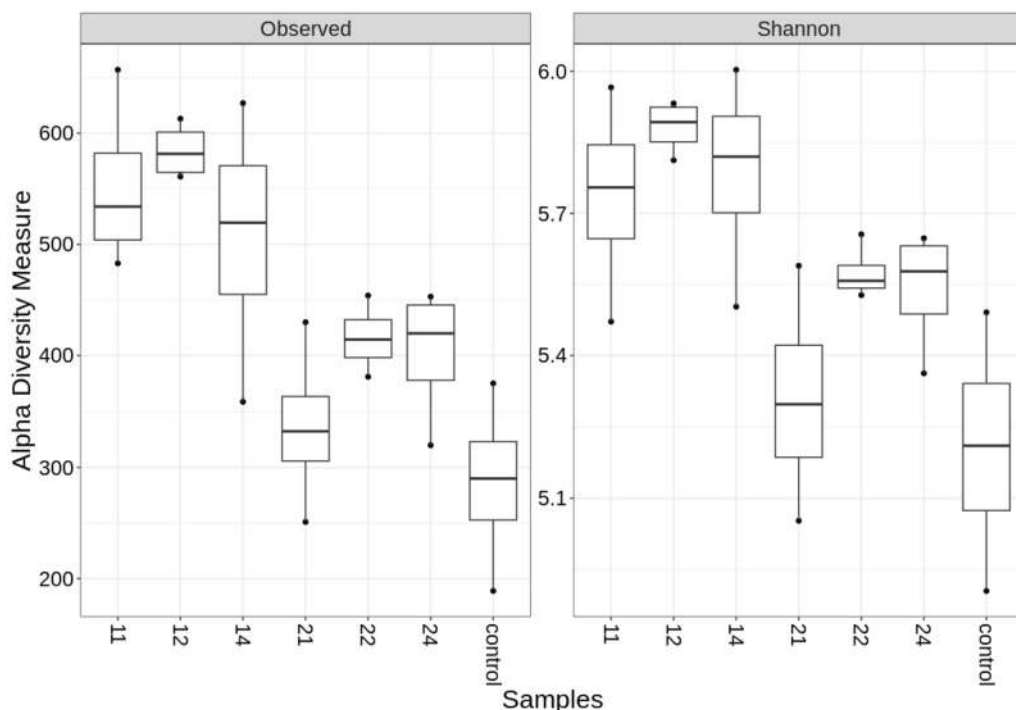


Figure 1. Alpha diversity of soil bacterial communities under fertilization and liming treatments. Boxplots show observed ASV richness (left) and Shannon index (right) from 16S rRNA data. Treatments: Control (no inputs), PK (code 11), PKCa₅₀₀ (12), PKCa₁₅₀₀ (14), NPK (21), NPKCa₅₀₀ (22), NPKCa₁₅₀₀ (24).

Overall, soil pH emerged as a key driver of microbial patterns. Sustainable management should aim to balance nutrient inputs with periodic liming to maintain biodiversity. Although liming enhances soil health and supports diverse communities, it cannot fully offset the ecological costs of long-term nutrient enrichment.

Acknowledgements

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Water productivity and growth parameters of two grass species under deficit irrigation in Saudi Arabia

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Abstract

This study aimed to evaluate the drought stress response of Fawn-Tall fescue and Tekapo-orchard grass. The trials were conducted at the Qassim University Agricultural Research Station (26°18'28"N, 43°46'E). The grass varieties were grown under four irrigation treatments: I1 equivalent to 0.3 of standard crop evapotranspiration (ET_c), I2 equivalent to 0.65 ET_c, I3 equivalent to 0.75 ET_c, and I4 equivalent to 1.2 ET_c. Plant height, fresh weight, and dry weight were measured, and water productivity (WP) was calculated. The results showed a reduction in growth for both grass genotypes under increasing drought stress, as indicated by shorter plants and reduced fresh and dry weights. However, WP results showed that Fawn-Tall fescue tolerated drought stress more effectively than Tekapo-orchard grass, as indicated by the relatively constant WP values across all irrigation treatments.

Keywords: arid, grass species, stress, water deficit

Introduction

Tall fescue (*Festuca arundinacea* Schreb.) is one of the most important forage plants grown extensively in Europe and the United States. Water deficit is by far the most important abiotic environmental factor contributing to yield losses in crops. Transpiration is the dominant component of evapotranspiration. Previous studies have suggested that Fawn-Tall fescue is a forage crop capable of enduring drought (Al-Ghumaiz and Motawei, 2011; Cougnon *et al.*, 2014; Cross *et al.*, 2013) and would be a suitable forage crop to establish in arid zones, where water supply may be limited during certain periods of the year. This study aimed to investigate the effect of deficit irrigation on the yield and water productivity of Fawn-Tall fescue plant and Tekapo-orchard grass (*Dactylis glomerata*).

Materials and methods

The field experiment was conducted at the Qassim University Agricultural Research Station. (26°18'28"N, 43°46'E). Fawn-Tall fescue and Tekapo-orchard grass were seeded in 3-m² plots (ten rows per plot) as a randomised complete block design (RCBD) with three replications. A one-horsepower hydraulic pump was used to deliver water to the experimental units. The pump supplied water through a 50-mm polyethylene main line connected to four sub-main lines, each 32 mm in diameter. Irrigation treatments for the experiment were as follows:

- I1 is equivalent to 0.3 of the standard crop water requirements in the study area ET_c
- I2 is equivalent to 0.65 of the standard crop water requirements in the study area ET_c
- I3 is equivalent to 0.75 of the standard crop water requirements in the study area ET_c
- I4 is equivalent to 1.2 of the standard crop water requirements in the study area "ET_c"

Crop evapotranspiration (ET_c) was calculated using the FAO Penman-Monteith equation via the "CropWat" software developed by the FAO, along with available meteorological data records for the

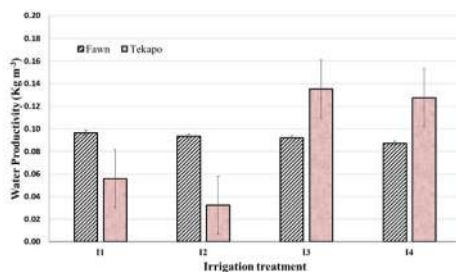


Figure 1. Water productivity (kg m^{-3}) of Fawn-Tall fescue and Tekapo-orchard grasses under different irrigation treatments.

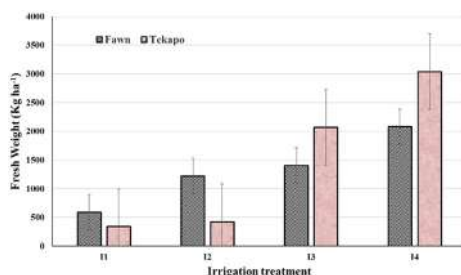


Figure 2. Fresh weight of Fawn-Tall fescue and Tekapo-orchard grasses under different irrigation treatments.

study area. Measured growth parameters included plant height (cm), fresh weight (kg ha^{-1}), dry matter yield (kg ha^{-1}), and total irrigation water ($\text{m}^3 \text{ ha}^{-1}$). Water productivity (WP) was calculated according to Geerts and Raes (2009). Analysis of variance (ANOVA) was performed using the least significant difference at $p < 0.05$. All statistical computations were performed using JMP Ver. 11.

Results and discussion

The effect of irrigation treatments on the water productivity (WP) of the grass genotypes is shown in Fig. 1. The WP trends of the two grasses Fawn and Tekapo were different. The WP of Tekapo decreased as the level of irrigation decreased from I4 to I2. For the Fawn genotype, the WP remained nearly constant across all irrigation treatments, and the differences between treatments were statistically insignificant (Fig. 1).

Regarding fresh weight, our results showed that the fresh weight of Fawn decreased significantly when irrigation levels decreased from I4 to I3 (Fig. 2). However, the decrease of fresh weight from I2 to I1 was not statistically significant. The reductions in fresh weight, calculated as percentage from the fresh weight for irrigation treatment I4, were 32, 41 and 71% for irrigation treatments I3, I2, and I1, respectively. For the Tekapo genotype, fresh weight decreased significantly as irrigation decreased from I4 to I2, followed by an insignificant decrease at I1 (Fig. 2). The reductions in the fresh weight were 32, 86 and 88% for irrigation treatments I3, I2 and I1, respectively.

The dry weight of Fawn also decreased as irrigation was reduced from I4 to I1. The corresponding percentage reductions in the dry weight, calculated as percentage from the dry weight for irrigation treatment I4, were 26, 58 and 71% for treatments I3, I2, and I1, respectively. Tekapo dry weight significantly declined with reduced irrigation (Fig. 3). The reductions in the dry weight were 23, 80 and 84% for irrigation treatments I3, I2 and I1, respectively.

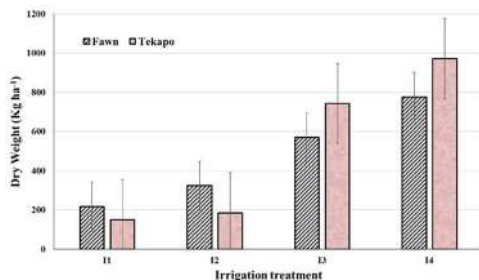


Figure 3. Dry weight of Fawn-Tall fescue and Tekapo-orchard grasses under different irrigation treatments.

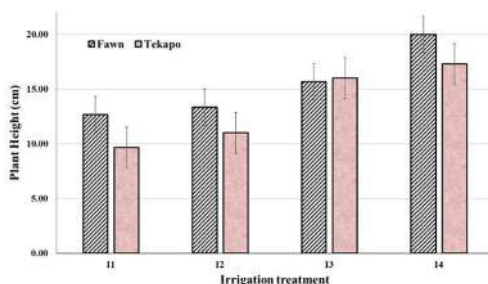


Figure 4. Plant height (cm) of Fawn-Tall fescue and Tekapo-orchard grasses under different irrigation treatments.

Plant height followed a trend similar to that of dry weight. The plant height of Fawn decreased as irrigation was reduced from treatment I4 to I1. The reductions in the plant height, calculated as percentage from the plant height for irrigation treatment I4, were 21, 33 and 36% for irrigation treatments I3, I2 and I1, respectively. Tekapo also showed decreased plant height under reduced irrigation (Fig. 4). The reductions in the plant height were 7.5, 36 and 44% for irrigation treatments I3, I2 and I1, respectively.

Conclusion

This study demonstrated variation between the two grass genotypes (Fawn-Tall fescue and Tekapo-orchard grass) in their adaptation to water stress. Fawn produced higher dry weight yield than Tekapo under deficit irrigation. Furthermore, the water productivity of Tekapo decreased sharply with reduced irrigation, whereas the water productivity of Fawn-Tall fescue remained nearly constant across all irrigation treatments.

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Effects of raised groundwater tables and grassland use on load-bearing capacity of agricultural peatlands

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Abstract

Dutch agricultural peatlands are typically drained to promote production. However, this drainage leads to peat oxidation and CO₂ emissions. Rewetting by raising groundwater tables can reduce these impacts but may compromise load-bearing capacity (LBC). This study examined the relationships between raised groundwater tables (to –20 cm, using an active water infiltration system), seasonal variation, grassland use, and LBC—measured as penetration resistance—in an on-farm experiment. A linear mixed model was developed to predict LBC. Raising the groundwater table from –50 to –20 cm reduced LBC by 0.25–0.34 MPa. LBC was on average 0.38 MPa higher in autumn than in spring, and 0.07–0.12 MPa higher in grazed fields than in mown fields. Understanding the interplay between seasonal variation, grassland use, and groundwater tables in determining LBC and accessibility of agricultural peatlands can refine models that assess the financial and practical implications of rewetting for farmers.

Keywords: active water infiltration system (AWIS), dairy farming, penetration resistance, rewetting

Introduction

Approximately 8% of the land area of The Netherlands consists of peatland, of which 77% is used for agriculture, predominantly as grassland-based dairy farming. Agricultural peatlands are typically drained to around 50 cm below field level to promote production and increase load-bearing capacity (LBC). LBC, measured as penetration resistance, usually ranges between 0 and 2 MPa, and values below 0.7 MPa are considered insufficient to prevent sward damage from livestock and machinery. However, deeper drainage leads to oxidation and decomposition of peat, resulting in CO₂ emissions and soil subsidence. Rewetting peatland to 20 cm below field level is seen as a key strategy to mitigate this (Querner *et al.*, 2012). To predict the practical and financial implication of raised groundwater tables for farmers, it is important to improve our insight into the impact of raised groundwater tables on LBC in agricultural peatlands. The aim of this study was to quantify the effect of raised groundwater tables on LBC and to assess how seasonal variation and grassland use influence this relationship.

Materials and methods

An on-farm experiment (2021–2024) was conducted at the Knowledge Transfer Centre (KTC) Zegveld experimental farm (52°8′25.8″N, 4°50′19.7″E) located on drained peat soil in the western peat region of the Netherlands. Sixteen 10 × 10 m monitoring plots were established in fields with either raised groundwater tables (–20 cm; high-water (HW) fields) or reference groundwater tables (–50 cm; low-water (LW) fields), under two grassland use strategies: mowing only (silage fields)

and combined grazing of dairy cows and mowing (pasture fields). The raised groundwater table was achieved using a pressurized submerged drainage system with active water infiltration (AWIS). LW fields had no active drainage in place. All fields received both artificial fertilizer (70–85 kg total N ha⁻¹ year⁻¹) and cattle slurry (173 kg total N ha⁻¹ year⁻¹). LBC was measured as penetration resistance (MPa) using a penetrometer (Eijkelkamp, The Netherlands) with a cone diameter of 5 cm² and a 60° apex angle, taking 15 measurements per plot. Groundwater tables were measured in monitoring wells which were located adjacent to each plot. Gravimetric soil moisture content (SMC, %) at 0–10 cm was determined in a bulk sample of 20 soil cores per plot. Basal vegetation cover (used as a proxy for tiller density) was measured once in spring and autumn using a point intercept method, taking 10 measurements per plot. All other measurements were conducted biweekly during spring and autumn. Pearson correlation coefficients between all variables were retrieved. A linear mixed-effects model was fitted with LBC as the response variable and groundwater table, season, basal cover and all two-way interactions among these variables as fixed effects, using the ‘lme4’ package in R (Bates *et al.*, 2015). Non-significant interaction terms ($p > 0.05$) were removed.

Results and discussion

On HW fields, the median groundwater table was -19 cm (range: -51 to 0) and the median LBC was 0.63 MPa (range: 0.24 to 1.64). On LW fields, the median groundwater table was -35 cm (range: -90 to 0) and the median LBC was 0.67 MPa (range: 0.24 to 1.84). There was a negative correlation between LBC and groundwater table ($r = -0.45$, $p < 0.001$), an even stronger correlation between LBC and SMC ($r = -0.63$, $p < 0.001$), and a positive correlation between groundwater table and SMC ($r = 0.66$, $p < 0.001$). These relationships indicate that variation in LBC is associated with both groundwater table and SMC. Further research should aim to disentangle the individual contributions of SMC and groundwater tables to LBC.

The fitted model (conditional $R^2 = 0.60$; Figure 1) showed that in spring, each 1 cm increase in groundwater level corresponded to a 0.0085 MPa decrease in LBC ($p < 0.001$). Thus, raising the groundwater table from -50 to -20 cm would reduce LBC on average by approximately 0.25 MPa. A significant interaction between groundwater table and season indicated that the negative effect of higher groundwater tables on LBC was stronger in autumn than in spring (estimate = -0.0028, $p = 0.014$). In autumn, raising the groundwater table from -50 to 20 cm would reduce LBC by 0.34 MPa. However, under similar groundwater tables, LBC was on average 0.38 MPa higher in autumn than in spring, partly offsetting this seasonal effect. This difference may be explained by seasonal patterns in the relationship between soil moisture and LBC, known as hysteresis (Schlotzhauer & Price, 1999).

The basal cover (used as a proxy for tiller density) was on average 65% for pasture fields and 44% for silage fields, confirming the positive effect of grazing on tiller density. This 21% increase in basal cover translated to a 0.13 MPa higher LBC on pasture fields compared to silage fields in spring (estimate = 0.621, $p < 0.001$), which is in line with Janssen *et al.* (2023). A significant interaction between basal cover and season (estimate = -0.30, $p = 0.017$) showed that the effect of basal cover on LBC was smaller in autumn with a 0.07 MPa difference between pasture and silage fields. This effect may also be explained by different soil dynamics between seasons (Schlotzhauer & Price, 1999), possibly resulting in different impacts of sward structure on soil strength.

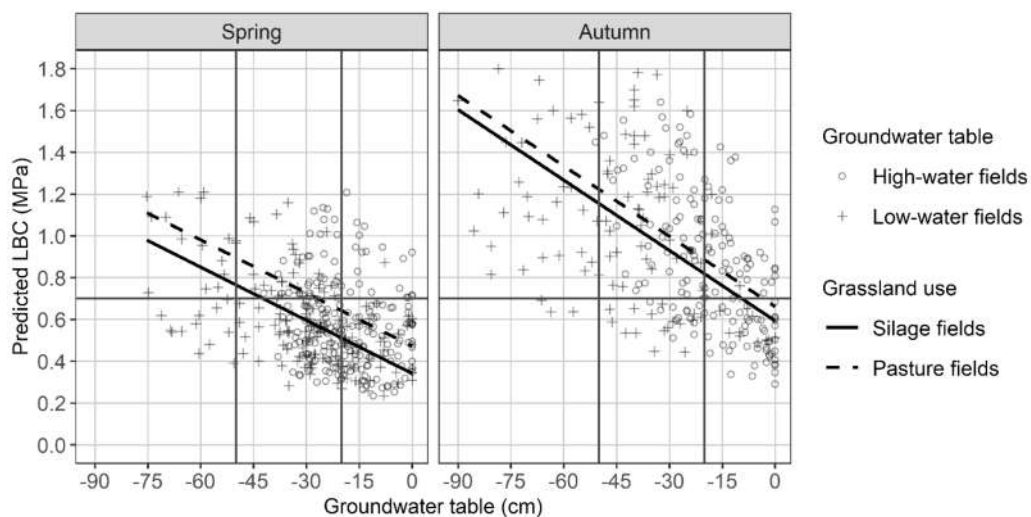


Figure 1. Predicted LBC (MPa) in spring and autumn as a function of groundwater table (cm below field level) for pasture (basal cover = 0.65) and silage (basal cover = 0.44) fields, based on the linear mixed model. Vertical lines mark groundwater tables of -50 cm and -20 cm, the target levels for LW and HW fields, respectively. The horizontal line indicates the 0.7 MPa threshold below which LBC is considered insufficient to prevent sward damage from livestock and machinery. Points represent observed values for LW and HW fields. The final model was: $LBC = \beta_0 + \beta_1 \times GWT + \beta_2 \times Season + \beta_3 \times BC + \beta_4 \times (GWT \times Season) + \beta_5 \times (BC \times Season) + u(Field) + u(Year) + \epsilon$, where LBC is load-bearing capacity (MPa), GWT is groundwater table (cm below field level), BC is basal cover (proportion), $\beta_0 \dots \beta_5$ are fixed-effect coefficients ($\beta_0 = 0.068$, $\beta_1 = -0.0085$, $\beta_2 = 0.38$, $\beta_3 = 0.62$, $\beta_4 = -0.0028$, $\beta_5 = -0.30$), $u(Field)$ and $u(Year)$ are random intercepts for field and year, and ϵ is the residual error.

Conclusion

This study demonstrated that elevating groundwater tables from -50 to -20 cm increased the likelihood of insufficient LBC (<0.7 MPa penetration resistance). However, the relation between groundwater tables and LBC was strongly influenced by season and grassland use. In spring, raising the groundwater table from -50 to -20 cm reduced LBC by 0.25 MPa, whereas in autumn the reduction was greater (-0.34 MPa) but was offset by a generally higher LBC in autumn ($+0.38$ MPa). Grazing, which increases basal vegetation cover, also enhanced LBC, particularly in spring. Further exploring the interplay between seasonal variation, grassland management, and groundwater tables in determining LBC can help refine models that predict field accessibility and assessing the financial impacts of rewetting agricultural peatlands for farmers (De Vos *et al.*, 2006).

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Access to grazing reduces enteric methane emission of high-producing dairy cows

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Abstract

Methane emissions from cattle significantly contribute to agricultural greenhouse gases. This study evaluated the impact of autumn grazing on enteric methane emission of dairy cows. Twenty-seven cows were subjected to a two-period incomplete Latin square design with three dietary treatments: full indoor feeding (A), grazing (6 h) on perennial ryegrass+indoor feeding (B), and grazing (6 h) on a multispecies sward of perennial ryegrass, white clover and ribwort plantain + indoor feeding (C). Methane and CO₂ emissions were measured via GreenFeeds. According to the VEM-method, treatment B and C had a comparable fresh grass dry matter intake, respectively 3.42 kg day⁻¹ and 4.18 kg day⁻¹. Fat and protein corrected milk (FPCM) yield was not significantly affected but was numerically higher for B and C (36.9 kg day⁻¹) than for A (35.8 kg day⁻¹) ($P = 0.273$). Treatment B and C showed lower methane production, methane intensity and methane yield compared to treatment A ($P < 0.001$). These results suggest that integrating grazing on conventional and multispecies pastures can reduce methane emission, without compromising productivity. This management innovation aligns with climate change mitigation goals and highlights the potential of diversified grazing strategies to enhance the resilience and sustainability of dairy farms.

Keywords: clover, dairy cows, enteric methane, grazing, plantain, ryegrass

Introduction

Enteric methane (CH₄) from dairy cattle is a significant contributor to agricultural greenhouse gas emissions and is a key target for climate mitigation (Beauchemin *et al.*, 2022). Nutritional mitigation strategies, including grazing intensity and altering pasture composition, are gaining attention for their potential to reduce emissions (Costigan *et al.*, 2024, Lardy *et al.*, 2024). Multispecies swards have been suggested to influence rumen fermentation dynamics, potentially leading to lower methane yields compared to monoculture swards (Soder and Brito, 2023). Empirical evidence remains scarce, especially under commercial grazing regimes common in Western Europe, where dairy processors often require a minimum of 6 hours of grazing per day for 120 days annually to qualify for the pasture milk label (Krizsan *et al.*, 2021). In this context, diversified grazing systems that incorporate species such as white clover (*Trifolium repens*) and ribwort plantain (*Plantago lanceolata*) may offer an opportunity to reduce methane. This study aimed to evaluate the effect of restricted autumn grazing on methane emissions and productivity in dairy cows, focusing on two pasture compositions: a conventional ryegrass sward and a multispecies sward including white clover and ribwort plantain. We hypothesized that integrating restricted grazing for 6 hours on conventional and multispecies pastures, supplemented by indoor feeding, would reduce methane emission without negatively affecting milk yield.

Materials and methods

The experiment was conducted in 2024 at the research facility of ILVO (Belgium) with Holstein-Friesian dairy cows ($n = 27$) in a two-period incomplete Latin square design with three dietary

treatments: Full indoor feeding (A), restricted (6 h) grazing on perennial ryegrass (*Lolium perenne*) with indoor feeding (B), restricted (6 h) grazing on perennial ryegrass, white clover and ribwort plantain, with indoor feeding (C). Each cow received two out of three treatments over two 4-week periods (22 August 2024–16 October 2024). Four pastures ranging from 0.92 to 1.29 hectares were used: two monocultures for treatment B and two multispecies swards for treatment C. Each pasture was divided into four strips, and cows were rotated over the strips every 3–4 days. A central paddock within each pasture provided access to the GreenFeed system (C-lock, Rapid City, SD, USA) and a water bin. All cows were milked twice daily, and cows from treatment B and C grazed after morning milking from 07:30 h to 13:30 h. Two partial mixed rations (PMR) were formulated for the indoor (treatment A) and grazing (treatment B and C) groups, which differed slightly to ensure the same nutritional value between all treatment groups. Treatment A comprised: 40% maize silage, 43% grass silage (perennial ryegrass), 8% pressed beet pulp supplemented with 7% soybean meal and 2% crushed barley. Treatments B and C comprised: 45% maize silage, 35% grass silage (perennial ryegrass), 9% pressed beet pulp supplemented with 8% soybean meal and 3% barley supplementation. Additional concentrates were tailored based on milk yield and body weight, but ranged from 2.5 to 6.5 kg cow⁻¹ day⁻¹. Intake was measured using roughage intake control (RIC) feed bins; fresh grass intake during grazing was estimated via VEM (Netto Energy of Lactation, CVB, 2016) and the n-alkane technique using synthetic C32 dosed via concentrate (Mayes *et al.*, 1986). GreenFeed systems measured individual gaseous emissions, both indoors and outdoors. Milk was recorded at each milking, and samples, at the last week of each period, were analysed by FTIR (Fourier-transform infrared) spectroscopy for composition.

Results and discussion

Total dry matter intake (DMI) according to the VEM-method and the concentrate intake did not differ between the treatment groups ($P = 0.140$ and $P = 0.671$, respectively; Table 1). According to the n-alkane method the feed intake was lower compared to the VEM-method, and total DMI was significantly highest for treatment A (26.6 kg DM day⁻¹), followed by treatment C (25.1 kg DM day⁻¹) and lowest for treatment B (24.1 kg DM day⁻¹; $P < 0.001$). While the fresh grass supply for both treatments was similar, the difference in total DMI between treatment B and C was caused by a difference in fresh grass intake (0.9 kg day⁻¹ versus 2.0 kg day⁻¹). Although smaller, a similar difference in fresh grass intake was observed according to the VEM-method, one possible explanation could be the palatability of the grass. The fresh grass intake according to the n-alkane method was lower than expected with the VEM-method. This underestimation has also been observed in previous studies (e.g., Klootwijk *et al.*, 2018). One possible explanation is that the faecal recovery rates for C32 and C33 applied may not have been accurate in this study due to the supplementation with maize silage (Van den Pol-Van Dasselaar *et al.*, 2006).

No statistically significant differences were observed among treatments for any of the milk yield or compositional parameters. Across all metrics for CH₄ emission significant treatment effects were observed ($P < 0.001$; Table 1). The CH₄ production, CH₄ intensity and CH₄ yield was significantly highest for cows in treatment A compared to both treatment B and C. The CH₄ yield based on the DMI according to the VEM-method and n-alkane method showed the same results. The effect of treatment C was not statistically different from treatment B, suggesting that the impact of sward diversity in this context may be limited. No significant difference was found for CO₂ emission.

Table 1. Least square means (in kg cow⁻¹ day⁻¹) of the dry matter intake (DMI) of the fresh grass (based on the VEM-method or n-alkane method), total DMI ((based on the VEM-method or n-alkane method)), CO₂ emission, methane emission (CH₄), methane intensity (CH₄ per kg fat and protein corrected milk (FPCM)) and methane yield (CH₄ per kg dry matter intake (DMI)) based on VEM-method and n-alkane method. per treatment group.

	Treatment			<i>P</i>
	A	B	C	
DMI intake				
Fresh grass (kg day ⁻¹ , VEM-method)	—	3.4	4.2	—
Fresh grass (kg day ⁻¹ , n-alkane method)	—	0.9	2.0	—
Total (kg day ⁻¹ , VEM-method)	26.5 ^a	26.6 ^a	27.5 ^a	0.140
Total (kg day ⁻¹ , n-alkane)	26.5 ^a	24.1 ^c	25.1 ^b	<0.001
Emission				
CO ₂ production (g day ⁻¹)	15372	15422	15764	0.100
CH ₄ production (g day ⁻¹)	561 ^a	466 ^b	493 ^b	<0.001
CH ₄ intensity (g (kg FPCM) ⁻¹)	15.3 ^a	13.1 ^b	13.5 ^b	<0.001
CH ₄ yield (g (kg DMI) ⁻¹ , VEM-method)	21.3 ^a	17.6 ^b	18.0 ^b	<0.001
CH ₄ yield (g (kg DMI) ⁻¹ , n-alkane method)	21.2 ^a	19.4 ^b	19.7 ^b	<0.001

Results with a different superscript are significantly different from each other.

Conclusion

This study showed that restricted grazing, in combination with a corresponding indoor diet, could result in a reduction, compared to full indoor feeding, in CH₄ emission without compromising production provided that the overall nutritional quality of the total diet is maintained. However, no additional CH₄ mitigation benefit was observed from including white clover and ribwort plantain compared to a monoculture of perennial ryegrass. These findings support the viability of partial grazing strategies as a practical and effective approach to reduce the environmental footprint of dairy production. Further research is warranted to explore the long-term effects of sward diversity.

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Functional trait-based design of cover crops for maize yield, resource use, and nitrogen dynamics

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Abstract

Cover crops are usually grown to conserve nutrients and make them available to the succeeding crop. The type and extent of this conservation depends, among other things, on the functional traits of the cover crops and is influenced in particular by rooting depth, biological N₂-fixation and winter hardiness. If cover crops are not terminated before the following main crop, but are maintained as living mulch (LM) systems, there are competitive effects that are reflected in biomass yield and nutrient uptake. However, little is known about effects of the functional composition and much less about niche occupancy in the root zone when different LM combinations are grown with maize. We evaluated eight cover crop treatments, as LM or non-living mulch (NLM), on maize yield and nitrogen (N) uptake. NLM treatments did not significantly affect maize yield (control: 23.7 Mg DM ha⁻¹; NLM: 22.4 Mg DM ha⁻¹), whereas LM significantly reduced yield (10.1 Mg DM ha⁻¹), regardless of functional composition. At harvest, mineral N (N_{min}) was lowest in LM, intermediate in NLM, and highest in the control, reflecting differences in root competition and N uptake. After this first year of evaluation, functional traits of cover crops seem of much lesser importance than the cover crop management.

Keywords: cover crops, functional traits, living mulch, maize yield, nutrient uptake

Introduction

Cover crops are a sustainable agronomic practice that improve nutrient cycling, soil structure, and weed suppression, thereby enhancing resource use efficiency, subsequent crop productivity, and long term sustainability of cropping systems (Quintarelli *et al.*, 2022). The realization of these benefits depends largely on cover crop functional traits, such as rooting depth, biological N₂-fixation, and winter hardiness, as well as management optimization, among other factors (Kühling *et al.*, 2023). In maize-based systems, terminated cover crops primarily act as nutrient catch crops, releasing resources to the following maize crop, whereas LM persist throughout the season, providing continuous ground cover, supporting biodiversity, and reducing nitrate leaching (Huss *et al.*, 2022). However, LM may also compete with maize for light, water, and nutrients, raising questions about whether functional trait diversity can mitigate these trade-offs. Despite their potential, little is known about how functional trait combinations in cover crops influence maize performance, particularly in terms of root-zone niche occupancy, and belowground resource capture under LM systems. Therefore, in this study, we assessed silage maize above and belowground biomass and soil N_{min} to address the research question: How do

cover crop functional traits and management strategies influence maize yield, root distribution, and N dynamics?

Materials and methods

The experiment was conducted at the Reinshof experimental site, Göttingen, Germany, as part of a multi-year, multi-site field study. The soil is a silty loam Phaeozem (pH 7.1). Eight cover crop treatments and a control (fallow) were arranged in a randomized complete block design with four replicates. Treatments included single and multi-species mixtures differing in key functional traits and managed as either LM or NLM. Cover crops were sown in August 2023 following barley harvest, at 15 kg ha⁻¹. LM treatments included white clover (*Trifolium repens*), sorghum (*Sorghum sudanense*) with white clover, and a four-species mixture of red fescue (*Festuca rubra*), sorghum, white clover, and Persian clover (*Trifolium resupinatum*). NLM treatments included sorghum, white clover, sorghum with white clover, and two four-species mixtures: one with red fescue, sorghum, white clover, Persian clover, and the other with red fescue, sorghum, oilseed radish (*Raphanus sativus*), and winter rapeseed (*Brassica napus*). Silage maize (*Zea mays*) was sown in mid-May 2024 at 100 000 seeds ha⁻¹ with 75 cm row spacing in 10 × 10 m plots. Fertilization included 120 kg N, 46 kg P, and 120 kg K ha⁻¹. Aboveground biomass was sampled at maize BBCH 33, 65 and 85 by harvesting ten plants per plot from two central rows; subsamples were oven dried at 60°C to constant weight. Root biomass was collected at maize BBCH 33 and 65 using a soil corer of 9 cm diameter at 0–30 and 30–60 cm depths and three positions relative to the maize row: maize row (MR), near maize row (NMR), and between maize rows (BMR). Soil N_{min} (NO₃⁻ and NH₄⁺) was collected in triplicate per plot at 0–30, 30–60 and 60–90 cm at five time points (before and after winter and maize BBCH33, 65 and 85). Statistical analyses were performed in R (v4.2.2) using linear mixed effects models ('nlme' package) with block as a random factor. Post hoc comparisons of estimated marginal means were performed using the 'emmeans' package with Tukey adjustment.

Results and discussion

Maize aboveground biomass at BBCH 85 was not significantly affected by the functional composition of cover crop mixtures but was strongly influenced by management (Table 1). LM significantly reduced maize biomass (10.1 ± 0.76 Mg DM ha⁻¹) compared to control (23.7 ± 1.31 Mg DM ha⁻¹) and NLM (22.4 ± 0.59 Mg DM ha⁻¹), which did not differ from each other, reflecting competition with living cover crops possibly for light, nutrients, and water. Root biomass at 0–30 cm differed among treatments and positions (Table 2). In the MR, LM significantly reduced root biomass (2.08 ± 0.20 Mg DM ha⁻¹) relative to control (3.47 ± 0.44 Mg DM ha⁻¹) and NLM (4.52 ± 0.24 Mg DM ha⁻¹), indicating strong belowground competition, whereas LM significantly increased root biomass in NMR (0.60 ± 0.11 Mg DM ha⁻¹) and BMR (0.50 ± 0.10 Mg DM ha⁻¹), suggesting lateral root development to avoid direct competition. No significant differences were observed at 30–60 cm depth, showing that competition was concentrated in the upper soil layer where maize and LM roots

Table 1. Mean aboveground maize biomass at BBCH85 under control, living mulch, and non-living mulch treatments.

Treatment	Aboveground biomass (Mg ha ⁻¹)
Control	23.7 ± 1.31 b
Living mulch (LM)	10.1 ± 0.76 a
Non-living mulch (NLM)	22.4 ± 0.59 b

Values are mean ± SE; different letters indicate significant differences (Tukey-adjusted, *P* < 0.05).

Table 2. Mean root biomass of maize at 0–30 cm depth across three positions under control, living mulch, and non-living mulch treatments.

Treatment	MR (Mg ha ⁻¹)	NMR (Mg ha ⁻¹)	BMR (Mg ha ⁻¹)
Control	3.47 ± 0.44 b	0.17 ± 0.10 a	0.17 ± 0.10 ab
Living mulch (LM)	2.08 ± 0.20 a	0.60 ± 0.11 b	0.50 ± 0.10 b
Non-living mulch (NLM)	4.52 ± 0.24 b	0.16 ± 0.05 a	0.12 ± 0.04 a

Values are mean±SE; different letters indicate significant differences (Tukey-adjusted, $P < 0.05$). No significant differences were observed at 30–60 cm depth. MR, maize row; NMR, near maize row; BMR, between maize rows.

Table 3. Mean soil mineral N (0–90 cm) after winter (March 2024) and at final harvest (September 2024) under control, living mulch, and non-living mulch treatments.

Treatment	After winter (kg N ha ⁻¹)	Final harvest (kg N ha ⁻¹)
Control	89.7 ± 16.66 b	68.4 ± 12.70 b
Living mulch (LM)	45.6 ± 04.89 a	31.0 ± 03.33 a
Non-living mulch (NLM)	45.2 ± 03.75 a	38.1 ± 03.16 ab

Values are mean±SE; different letters indicate significant differences among treatments within each sampling date (Tukey-adjusted, $P < 0.05$).

co-occur. Soil $N_{\min}$ (0–90 cm) also varied among treatments and seasons (Table 3). After winter, control plots contained 89.7 ± 16.7 kg N ha⁻¹, nearly double that of LM (45.6 ± 4.9 kg N ha⁻¹) and NLM (45.2 ± 3.8 kg N ha⁻¹), which did not differ significantly from each other, indicating that both LM and NLM effectively captured N over winter, and likely reduced leaching. At harvest, $N_{\min}$ was lowest in LM (31.0 ± 3.3 kg N ha⁻¹), intermediate in NLM (38.1 ± 3.2 kg N ha⁻¹), and highest in control (68.4 ± 12.7 kg N ha⁻¹). In LM, strong belowground competition limited N availability, whereas in NLM, maize accessed N efficiently and residue mineralization contributed additional N, resulting in intermediate $N_{\min}$ level. In the Control, no cover crop competed for N, so maize uptake alone possibly left the highest soil $N_{\min}$.

Conclusion

Cover crop management, more than functional trait composition, determined maize yield and soil N dynamics. LM maximised N retention but reduced yield, whereas NLM sustained yields comparable to control while lowering residual $N_{\min}$, offering a more balanced trade-off between productivity and N conservation.

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N₂O emissions from timothy grassland under different fertilization regimes in boreal climate

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Abstract

Cultivated grasslands can be significant sources of nitrous oxide (N₂O), due to application of fertilizers. Using cattle slurry as a fertilizer is a common grassland management practice; however, it has been shown to increase N₂O emissions. Current knowledge on the effect of different fertilizer types and their rates on N₂O emissions are limited for boreal region. Here, we present N₂O flux results from first year (out of two) of the study conducted using cattle slurry and mineral nitrogen (N) fertilizer on a mineral soil cultivated with timothy (*Phleum pratense*) in central Finland. We measured N₂O fluxes using closed static chamber for snow-free period and snow-gradient method for snow-cover period. We found increased N₂O emissions after slurry and slurry with mineral N applications compared to only mineral N application, especially during dry period of the growing season. However, mineral N fertilizer also increased N₂O emission similar to slurry application when soil received enough moisture after precipitation events. Our result highlights that soil properties in combination with fertilizer types regulate the soil N₂O emissions from boreal grasslands. More multiyear studies are needed to assess the impacts of cattle slurry application on N₂O emissions and grassland productivity in boreal grasslands.

Keywords: boreal grassland, cattle slurry, chamber measurements, nitrous oxide

Introduction

Nitrous oxide (N₂O) is a strong greenhouse gas, with approximately 60% of the global emissions originating from agriculture (Shukla *et al.*, 2022). Mineral nitrogen (N) fertilization, soil moisture and temperature, and vegetation are some of the key factors regulating N₂O production in agricultural soils. Additionally, slurry application has been found to increase the N₂O emissions from grasslands (Jones *et al.*, 2007; Schils *et al.*, 2008; Xia *et al.*, 2020), although it can also improve soil organic carbon (SOC) stock and fertility (Xia *et al.*, 2020). Both mineral N and organic N fertilization are common management practices for boreal grasslands for ensuring soil fertility and biomass productivity. However, our current knowledge on the effect of different fertilization types and rates on N₂O emissions in boreal grassland is still scarce. The aim of this study was to quantify N₂O emissions and yields from different rates of cattle slurry and mineral N fertilization in boreal grassland.

Materials and methods

The study was conducted *in situ* on mineral loamy soil with 3.4% SOC in Maaninka, eastern Finland, with timothy (*Phleum pratense*) as grass species (established one year prior). The experimental design was a split plot with two main treatments: mineral N fertilization and cattle slurry (30 t ha⁻¹; 102 kg total N and 53 kg soluble N (sol-N) ha⁻¹) applied for the first and second harvests. Within each main plot, three subplots were established (0 or slurry N only, 250 and 350 kg soluble N ha⁻¹ year⁻¹) and the sol-N was divided between three harvests using ratios 0.44:0.36:0.20. In the slurry plots, the

sol-N from slurry was supplemented with mineral N to reach the target level and in the no slurry plots, all N was from mineral fertilizer. Slurry was spread to a depth of 5–7 cm using a plot-sized disc slurry injector. We measured N₂O fluxes from May 2024 to April 2025 with closed chamber method in snow-free period and with the snow gradient method during snow-cover. The N₂O concentration was measured in real time with a portable N₂O gas analyser (LI-7820) from chamber headspace during chamber measurement whereas gas chromatography was used for snow gradient method. The N₂O flux measurements were made weekly during growing season (GS, May to September) and biweekly during the non-growing season (NGS, October to April). To capture immediate effect of fertilization, N₂O flux measurements were conducted daily for one week after fertilization and every other day during the following week. N₂O fluxes were calculated based on linear increase in headspace N₂O concentration over time for the chamber method whereas it was based on the N₂O concentration gradient and diffusion rate of N₂O through the snowpack for snow gradient method. Biomass was harvested manually from N₂O flux measurement spots to estimate yields.

Results and discussion

Our preliminary findings suggest that N₂O emissions from timothy monoculture on mineral boreal soil is affected by organic N (here cattle slurry) addition. We found that slurry addition increased N₂O emissions by 3.5 folds compared to plots that received only mineral fertilizer (mean annual flux for slurry: 1.27–1.54 mg m² day⁻¹ and for mineral fertilizer: 0.24–0.59 mg m² day⁻¹). An increasing trend in N₂O emissions following slurry addition compared to mineral N addition was found in both first and second fertilization events (Fig. 1a).

These emissions could likely be a combined result of moisture and carbon-rich slurry, which have been shown to promote N₂O production in soil (Xia *et al.*, 2020; Jones *et al.*, 2007). No increase in N₂O emissions from the mineral-N fertilized plots during the first and second fertilization events could be associated with limited soil moisture, since the early part of the GS was dry and precipitation free. This could be supported by a similar N₂O emission trend with no distinct variations in emission rates across all treatments following third fertilization, when only mineral N was applied, and consistent precipitation was recorded. Mineral fertilized plots tended to produce higher yields compared to slurry fertilized plots, while the nonfertilized treatment (“no slurry 0 N”) produced the lowest yields.

The following NGS started with smaller emission rates in all plots. Interestingly, slurry-treated plots showed occasionally higher emission rates compared to the mineral-N fertilized plots (Fig. 1b) and those increased emissions coincided with fluctuating freezing temperatures. The fluctuating freezing temperatures are known to trigger freeze-thaw cycles during NGS, which are known to stimulate N₂O emissions (Lind *et al.*, 2019). Similarly, all treatments exhibited a marked increase in N₂O emission rates in April, coinciding with the onset of warmer temperatures, suggesting that temperature induced changes are also crucial for N₂O emissions, regardless of fertilizer types and their application rates.

Conclusion

This study suggests that using cattle slurry as a fertilizer led to higher N₂O emissions compared to mineral N fertilizer from timothy grassland cultivated on mineral soil, especially during precipitation-free and drought conditions. Nevertheless, our result demonstrated that mineral N fertilization could also increase N₂O emissions similar to that from organic N fertilization under conditions favourable

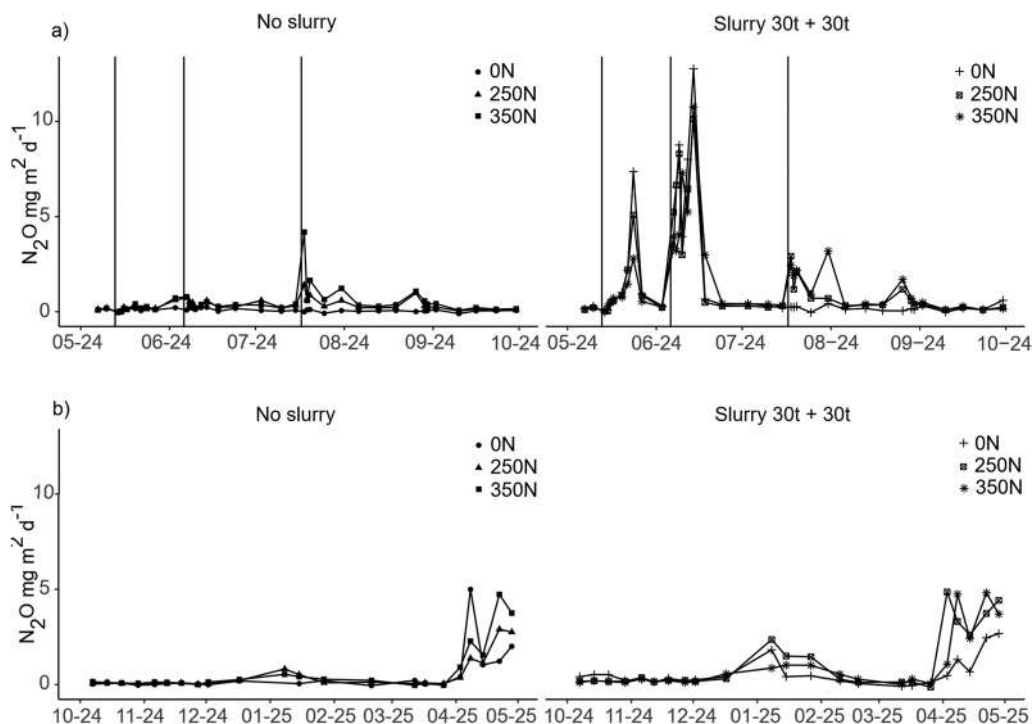


Figure 1. Measured N_2O fluxes ($\text{mg m}^{-2} \text{ day}^{-1}$) during the growing season (a) and non-growing season (b). The left panel shows mineral fertilizer plots, and the right panel shows cattle slurry + mineral N plots. Within each main treatment, three different supplemented mineral N levels are indicated by different symbols. The values represent treatment means ($n = 4$), and error bars are omitted for clarity. The vertical lines indicate timing of fertilization.

for N_2O production, e.g., with enough moisture. Assessments of climatic impact of agricultural management strategies should incorporate also economic viability and agronomic performance. Emission reductions that compromise productivity may not be beneficial for farmers in the long term. Therefore, we suggest that studies with multiyear N_2O emission assessment and biomass yield from the application of fertilizer types and rates are needed. An appropriate conclusion from this study will be derived by including and analysing data from second measurement year.

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High carbon content of a permanent grassland in western Norway: Too much of a good thing?

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Abstract

The decline of soil organic carbon (SOC) content is a major concern in agricultural soils, and reduction of tillage frequency is proposed as a measure to counteract this tendency. Here, we assessed the effect of tillage and renewal frequency on grassland SOC content based on a long-term experiment at Fureneset, Western Norway. The objective was to compare permanent and unrenewed grassland treatments with treatments ploughed and renewed 6 to 15 times from 1974 to 2016. Mean SOC content of the permanent grassland was $64 \pm 9 \text{ g kg}^{-1}$ (one standard deviation) at 0 to 40 cm depth and soil contained 210 Mg C ha^{-1} , compared to $60 \pm 6 \text{ g kg}^{-1}$ and 190 Mg C ha^{-1} for grassland renewed with ploughing. Higher SOC contents were associated with reduced forage dry matter yields (DMY) of the permanent grassland, but not in frequently renewed grasslands. High SOC contents correlate with high porosity and water content, as soil properties approach those of organic soils. This may cause a wetter soil and reduced plant growth and increase soil compaction. In areas with generally high SOC contents in agricultural soils, increased carbon content due to no tillage may thus make the soils more agronomically challenging to manage.

Keywords: grassland management, no tillage, ploughing, renewal, wet soil

Introduction

It is a common goal to increase soil organic carbon (SOC) content, as it alleviates atmospheric CO_2 loading and may improve soil physical properties and biomass production (Johnston et al., 2009). Reducing the frequency of soil tillage is believed to increase the amount of soil carbon (C), but there is no consensus yet (Fornara & Higgins, 2022).

To assess whether there is a potential for more C capture and storage by increasing grassland age, we analysed soil samples from a long-term grassland experiment established in 1974 at NIBIO Fureneset (61.3°N, 5.0°E), Western Norway.

Materials and methods

Fureneset has a long-term annual mean precipitation of 2300 mm and mean temperature of 7.8°C (1991–2020). The experiment is located on a medium peaty topsoil with a poorly drained subsoil with marked gleyic features. The history of the grassland experiment treatments is shown in Table 1. In spring 2016, all renewed treatments were chemically fallowed before either soil tillage by ploughing and harrowing, or direct sod-seeding. Extremely high rainfall in 2016 limited sward establishment and therefore a second direct seeding was carried out in May 2017.

The experiment includes different fertilizer strategies (see Sturite & Øpstad, 2022), but in this paper all fertilizer treatments are pooled together for simplicity. Six unfertilized plots are excluded from

Table 1. Treatment history of the experiment.

	PG1	PG2-S	PG2-R	R6	R12
Renewal rate 1974–1991	Permanent	Every 3rd year	Every 3rd year	Every 5th year + 1 year green fodder crops	Every 6th year
Renewal rate 1992–2016	Permanent	Permanent	Permanent	Every 3rd year	Every 6th year
Renewal rate 2017—	Permanent	Renewed in 2017 by sod-seeding	Renewed in 2016 by full tillage	Every 6th year	Every 12th year
Soil core 2023	6 plots	6 plots	6 plots	6 plots	0

Table 2. Soil organic carbon (SOC) concentration and SOC stocks by various soil depths.

Treatment	SOC concentration (g kg ⁻¹)			SOC stock (Mg ha ⁻¹)			
	0–5	5–20	20–40	0–5	5–20	20–40	Sum
PG1	108 ± 5.9 ^a	87 ± 4.5 ^a	35 ± 1.9 ^{ab}	74 ± 3.2 ^a	68 ± 2.6 ^a	34 ± 1.7	208 ± 4.9 ^{ab}
PG2-S	98 ± 6.5 ^a	84 ± 4.9 ^{ab}	41 ± 2.6 ^{ab}	70 ± 3.6 ^a	67 ± 3.0 ^{ab}	40 ± 2.3	215 ± 5.5 ^a
PG2-R + PG12	81 ± 5.7 ^b	69 ± 4.3 ^c	35 ± 1.5 ^b	60 ± 3.0 ^b	56 ± 2.4 ^c	35 ± 1.4	183 ± 4.7 ^c
R6	77 ± 5.9 ^b	76 ± 4.5 ^{bc}	42 ± 1.9 ^a	58 ± 3.2 ^b	60 ± 2.6 ^{bc}	40 ± 1.7	199 ± 4.9 ^b

Values are mean ± SE. Treatments sharing the same letter are not significantly different ($P < 0.05$).

analysis in this paper. The plant biomass was normally harvested 3 times during the growing season and subsamples were dried at 60°C for 48 h to determine dry matter yields (DMY). In very wet summers like in 2022, there were only 2 cuts. DMY data from 2018 to 2022 are used, when the sward age of all ploughed and reseeded treatments in 2017 coincide.

Before R6 renewal in spring 2023, duplicate cores of 100 cm³ were taken at 5–9 cm and 15–19 cm soil depths as presented in Table 1. Collected samples were analysed for water retention. In autumn 2024, soil samples were taken from all 72 plots at three depths (0–5, 5–20 and 20–40 cm), sieved (<2 mm) and analysed for soil organic carbon (SOC). Soil bulk density was determined by mixing the soil and then weighing a scoop with a known volume. Data were grouped based on calendar date of the last ploughing and reseeded event. R was used for statistical analysis (R core team 2023): SOC concentration and SOC stock was regarded as a function of sward renewal rate with replicate as a random effect, while DMY was regarded as a function of sward renewal rate and SOC concentration 0–20 cm, with year and replicate as random effects.

Results and discussion

Both types of permanent grassland (PG1 and PG2-S) had higher SOC stocks than seldomly renewed grasslands (PG2-R and R12), but PG1 was not significantly different from R6 (Table 2). This was mainly due to a somewhat higher C accumulation below ploughing depth in R6.

DMY of all renewed grassland treatments were higher than that of PG1. We found no correlation between DMY of renewed grasslands and SOC. High SOC content in PG1 surprisingly reduced DMY (Fig. 1a). This may be due to very high SOC concentration, combined with unfavourable conditions for drainage. Campos-Cáliz *et al* (2024) found agronomic benefits in cereal production from increasing SOC concentration to level off at around 14 g kg⁻¹, and this may be valid for grasslands as well.

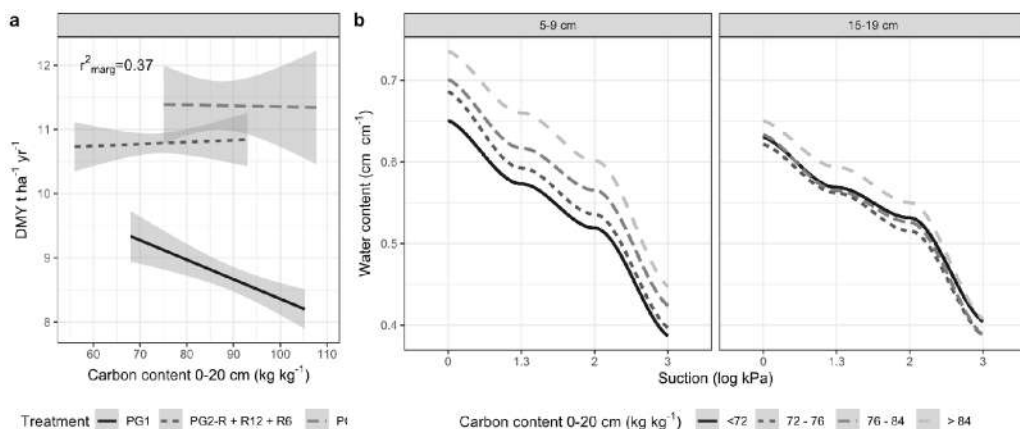


Figure 1. (a) Yearly DMY 2018–2022 as a function of carbon content (0–20 cm) measured in 2024. (b) Volumetric water content at different suction and depth; at 5–9 cm and 15–19 cm. Each line represents a quartile of observations.

High SOC concentrations have led to increased water content and moisture retention in the soil (Fig. 1b), which may hamper plant growth and shorten the periods of trafficability, especially in a climate and a soil as consistently wet as it is at Fureneset. There are likely other effects as well, linking SOC and DMY, which have to be investigated in future studies.

Conclusions

This study shows that grassland SOC stocks can be high after omitting tillage for 30–50 years. However, high SOC stocks were associated to decreased DMY in the oldest grassland, which is likely linked to excessive water holding capacities. Renewal, either by tillage or sod seeding, mitigated the negative effect of SOC on DMY. Further research is needed to investigate whether there exists an upper threshold for optimal SOC.

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Optimising boreal grassland production through species selection, nitrogen management, and harvest strategy

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Abstract

Achieving a high-yielding and stable grass sward for silage production requires careful coordination of the nitrogen (N) fertiliser applications, harvesting strategies, and the selection of grass species. To find the best combinations, a plot experiment was conducted in east-central Finland in 2023–2024 on sandy loam with three management strategies: reduced N fertiliser application with three harvests (3hR), two harvests (2 h) and three harvests (3 h). All strategies included the application of cattle slurry for the second harvest and were implemented for three grass mixtures (G; timothy, fescues, and perennial ryegrass) and two grass-legume mixtures (L; clover added). On average, in both years, the DM yield of G3h was higher than G2h and G3hR, which were equal. In contrast, L3hR produced a higher DM yield than L2h in both years and in 2024 was equal to L3h. Perennial ryegrass was abundant in the first year but suffered winter damage later. The second year was favourable for red clover. Maximising the benefits of optimal cultivation strategies requires paying attention to the winter hardiness of species and varieties, and the implementation of management practices that account for the legume component in the mixture.

Keywords: forage, grass, grass-legume, mixture, nitrogen

Introduction

Multi-species grass mixtures improve resilience to climate extremes (Isbell *et al.*, 2015) and enhance yield and nutritional quality (Sturludóttir *et al.*, 2013). Nitrogen-fixing grass legumes offer the potential to reduce mineral nitrogen (N) fertiliser use, but further research on their use in mixtures, especially in boreal conditions, is still needed (Ergon & Bakken, 2022). This study tested five commercial seed mixtures under various harvest and fertiliser application strategies, with the hypothesis that different management strategies influence species composition, forage yield, and nutritional quality.

Materials and methods

The experiment was conducted in 2023–2024 as a split-plot design with three replicates in east-central Finland (63°9'N, 27°20'E) on sandy loam on plots established in 2022. The main plots included three cultivation strategies: reduced mineral N fertiliser application with three harvests (3hR; 70+93+0 kg soluble-N (sol-N) ha⁻¹), two harvests (2 h; 100+93 kg sol-N ha⁻¹) and three harvests (3 h; 100+83+60 kg sol-N ha⁻¹). All strategies included cattle slurry injection for the second harvest (on average, 30 t ha⁻¹, 53 kg sol-N and 102 kg tot-N ha⁻¹). Mineral P and K were not used for 3hR while recommended rates were given for 2 h and 3 h. The five subplots consisted of three grass mixtures (G) and two grass-legume mixtures (L; Table 1). The first harvest was implemented at the same time for all plots, and the second harvest of 2 h was delayed compared to the others. In 2024, herbicide (amidosulfuron at 15 g ha⁻¹) was applied after first harvest because of the high amount of dandelion (*Taraxacum*). Dry matter yield (DMY, kg DM ha⁻¹) was measured. Botanical composition (fresh weights, sample size 300–500 g) was determined using manual sorting in the first harvest

Table 1. Grass mixtures (G1, G2 and G3) and grass-legume mixtures (L1 and L2) and their composition (species and cultivars), % of sowing mixture (25 kg ha⁻¹, except G2 30 kg ha⁻¹).

Mixture	Species	Timothy		Meadow fescue		Tall fescue	Perennial ryegrass	Red clover	White clover
	Variety	'Tuure'	'Rhonia'	'Klaara'	'Eevert'	'Karolina'	'Riikka'	'Selma'	'Silvester'
G1		75		10		15			
G2		35	40	25					
L1		36	24	10			10	15	5
G3		35	25	20		10	10		
L2		20	40		10	10	10	10	

Timothy, *Phleum pratense* L.; meadow fescue, *Lolium pratense* Huds.; tall fescue, *Lolium arundinaceum* Schreb.; perennial ryegrass, *Lolium perenne* L.; red clover, *Trifolium pratense* L.; white clover, *Trifolium repens* L.

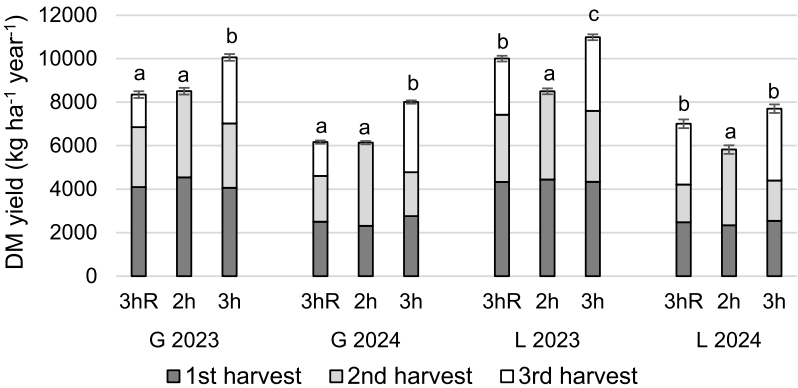


Figure 1. Dry matter yields (DMY) of grass mixtures (G) and grass-legume mixtures (L) in 2023 and 2024. 3hR, three harvests with reduced nitrogen (N) fertiliser application; 2 h, two harvests; 3 h, three harvests. Means marked with different letters indicate statistically significant differences ($P < 0.05$) within year and mixture type (G or L). Error bar is the standard error of the mean (SEM).

from two replicates. Statistical analysis was conducted using ANOVA (the MIXED procedure of SAS 9.4; the split plot design). Years were analysed separately, and mixture types (G and L) were analysed both separately (Figure 1) and together (Figure 2). The Kenward-Roger approximation for degrees of freedom and Tukey-Kramer test for pairwise comparisons were used.

Results and discussion

On average, in both years, the DMY of G3h was higher than G2h and G3hR, which were equal (Figure 1). In contrast, with the nitrogen-fixing clover involved, L3hR produced a higher DMY than L2h in both years and in 2024 was equal to L3h. However, this logical result appeared only in the third harvest, where 3hR was unfertilised. The strategy of G3hR maintained as high DMY as G3h in the first and second harvest of both years. The results indicate that the N rates used in the 3hR were sufficient to meet the plants' needs during the relatively dry growing periods of the first and second yields.

Perennial ryegrass was abundant in the first year, producing high DMY, but it suffered winter damage, causing a sharp decline in DMY in the first harvest of 2024 (Figure 2). The growing season of 2024 was warm and dry, resulting in an earlier harvest time and reduced DMY, and a higher red clover content. On average, 3 h produced higher DMY in the first harvest compared to 3hR in 2024, but not in 2023. The amount of red clover in the second harvest in 2024 was low (4%) likely because of

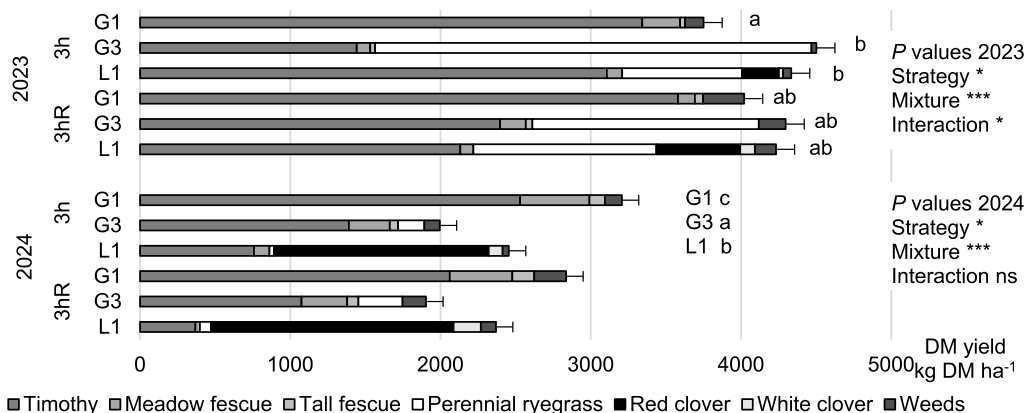


Figure 2. Dry matter yields (DMY) of grass mixtures (G) and grass-legume mixtures (L) in 2023 and 2024. 3hR, three harvests with reduced nitrogen (N) fertiliser application; 2 h, two harvests; 3 h, three harvests. Means marked with different letters indicate statistically significant differences ($P < 0.05$) within year and mixture type (G or L). Error bar is the standard error of the mean (SEM).

herbicide use, which reduced the total DMY of 3hR. However, the relative abundance of red clover recovered to 31% in the third harvest of L3hR.

Conclusion

The strategy of three harvests instead of two, and N fertiliser application also for the third harvest, is recommended for attaining the highest DMY. In this experiment, the benefits of biological N fixation by legume forages might have been more pronounced under lower N fertiliser application and without herbicide. Finding optimal cultivation strategies requires the selection of species with adequate winter hardiness, as well as management practices that account for the legume component in the mixture, such as appropriately reducing N fertiliser application.

Acknowledgement

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DNDC simulation of N₂O fluxes under mineral and organic nitrogen management in North Savo, Finland

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Abstract

Nitrous oxide (N₂O) is a potent greenhouse gas with 273 times the global warming potential of carbon dioxide (CO₂). In managed grasslands, N₂O fluxes are strongly influenced by soil properties, management practices, and climatic conditions. We evaluated the HE-DNDC (heat exchange-denitrification-decomposition) model performance in simulating N₂O fluxes under mineral (N_{min}) and organic (N_{org}) nitrogen management in a boreal grassland at Maaninka, Finland. Simulations over a three-year rotation (May 2017–May 2020), including re-establishment, were compared with eddy covariance measured N₂O fluxes. Cumulative measured N₂O fluxes were 7.5 kg N ha⁻¹ for N_{min} and 10.9 kg N ha⁻¹ for N_{org} treatment. The model simulations agreed well with the measured fluxes in N_{min} treatment (6.9 kg N ha⁻¹), while they underestimated the fluxes under N_{org} (7.3 kg N ha⁻¹). Annual simulated N₂O fluxes varied, with both over- and underestimations across years for both treatments. Notably, underestimations were prominent during the re-establishment phase for both the treatments, likely due to the model's limited ability to accurately represent enhanced microbial activity and organic matter decomposition owing to grassland renovation practices. Further refinement and long-term validation of HE-DNDC using diverse field data in boreal grasslands are warranted to improve the simulation accuracy.

Keywords: boreal grassland, eddy covariance, HE-DNDC model, nitrogen management, nitrous oxide

Introduction

The North Savo region of Finland, with its short growing seasons, supports intensive dairy and beef production, with milk contributing 70% of farm income (OSF, 2023). Livestock production accounts for 14.5% of national GHG emissions (Gerber *et al.*, 2013), underscoring the importance of grassland soils in mitigation. Nitrogen management is a key driver of N₂O emissions, but field-based assessments are resource-intensive and temporally constrained. Process-based models, such as the Heat-Exchange DNDC (HE-DNDC), simulate underlying biogeochemical processes and offer a cost-effective means to evaluate mitigation strategies across diverse conditions. In this study, we assessed the HE-DNDC performance in simulating N₂O emissions under mineral and organic nitrogen management in boreal grasslands using eddy covariance measurements.

Materials and methods

The study was conducted from 2017 to 2020 in Maaninka, eastern Finland (63°9′49″N, 27°14′3″E; 89 m asl), where the mean annual temperature is 3.8°C and precipitation is 619 mm. The soil is silty loam and classified as Haplic Cambisol (USDA). The ley was established as a mixture of timothy (*Phleum pratense*), red clover (*Trifolium pratense*) and barley (*Hordeum vulgare*) sown as cover crop with seed rates of 15, 5 and 160 kg/ha respectively. The 6.3 ha site was divided into two treatments: (i) N_{min}, receiving mineral fertiliser of 106:28:50 kg NPK ha⁻¹ year⁻¹, with half the N and all P

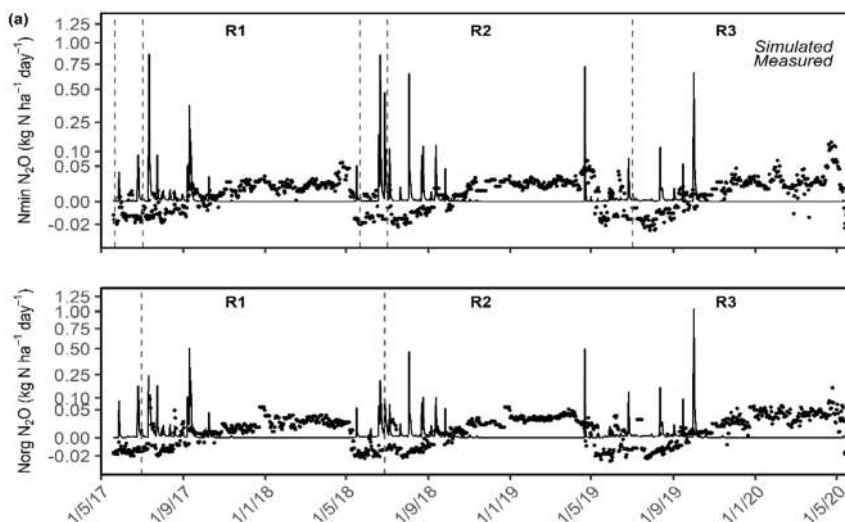


Figure 1. Measured and simulated N_2O fluxes: (a) $\text{N}_{\min}$ treatment, (b) N_{org} treatment. Dashed vertical lines indicate mineral and organic fertilizer application events, respectively.

and K applied as a basal dose, and the remaining N after the first cut; and (ii) N_{org} , receiving organic fertiliser at $98:13:83 \text{ kg NPK ha}^{-1} \text{ year}^{-1}$ (98 kg total N, of which 53 kg was soluble) from biogas digestate slurry applied by injection after the first cut. Rotational cycles R1, R2, and R3 spanned 20 May 2017–19 May 2018, 20 May 2018–19 May 2019, and 20 May 2019–19 May 2020, respectively.

N_2O fluxes were measured using a centrally located eddy covariance tower with a pulse quantum cascade laser spectrometer (APR-QC-TILDAS-76 CS; Aerodyne Research Inc., USA) (Li *et al.*, 2023). Simulations were performed with the HE-DNDC model, a modified version of the DeNitrification–DeComposition model incorporating surface heat exchange, soil carbon and nitrogen fluxes, and freeze–thaw dynamics for cold climates (Deng *et al.*, 2020).

Results and discussion

Measured daily N_2O fluxes showed sustained winter emissions (Dec–Apr) and occasional growing-season uptake (Fig. 1), with peaks emissions linked to fertilizer/manure applications and freeze–thaw events. HE-DNDC captured the timing of major peaks but underestimated persistent winter emissions and missed uptake events, indicating limited representation of overwinter microbial activity, frost depth variability and plant-mediated uptake (Baral *et al.*, 2022). Annual measured emissions were stable in R1 and R2 but increased sharply in R3 (re-establishment year) by 55% for $\text{N}_{\min}$ and 81% for N_{org} relative to R2. HE-DNDC simulated R1 and R2 totals with reasonable accuracy, but substantially underestimated R3 emissions in both treatments. This underestimation coincided with atypical winter conditions during R3, including variable snow depth, repeated freeze–thaw cycles, and crop termination in autumn R2, factors known to enhance microbial activity, organic matter mineralisation, and denitrification (Liu *et al.*, 2023), which were only partly accounted for in the model.

Over the three-year period, HE-DNDC simulated $\text{N}_{\min}$ totals reasonably (6.9 vs. 7.5 kg N ha^{-1} in total; 8.3% underestimation) but underestimated N_{org} by 33% (7.3 vs. $10.9 \text{ kg N ha}^{-1}$), with the largest gaps in R3 ($\text{N}_{\min}$: 1.1 vs. 3.4 ; N_{org} : 1.7 vs. 5.4 kg N ha^{-1}). Underestimation in N_{org} reflects

Table 1. Measured and simulated N₂O emissions (kg N ha⁻¹) during R1 (2017–2018), R2 (2018–2019), and R3 (2019–2020) for mineral (N_{min}) and organic (N_{org}) treatments.

Study period	N _{min} measured	N _{min} simulated	N _{org} measured	N _{org} simulated
R1	1.9	2.1	2.6	3.5
R2	2.2	3.7	3.0	2.1
R3	3.4	1.1	5.4	1.7
Total	7.5	6.9	10.9	7.3

partial representation of residual organic inputs, manure-related biogeochemistry and biological N fixation, as the model treated the sward as perennial grassland rather than a legume-grass ley (Thentu *et al.*, 2025).

Conclusion

HE-DNDC effectively reproduced multi-year cumulative N₂O emissions in cold-region grasslands but underestimated winter and organic amendment-related fluxes due to simplified crop parameterisation and limited representation of winter dynamics and seasonal uptake. Targeted refinements will enhance its reliability for nutrient management assessments in boreal dairy and beef systems.

Acknowledgement

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Temperature and water determine pasture productivity along an environmental gradient, with implications under climate change

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Abstract

Productivity patterns in rain-fed pastures are closely linked to climate variability. Shifts in annual plant growth cycles as a result of climate change have been documented widely in the past decades. These changes have major implications for pasture productivity, forage availability and long-term ecosystem resilience. Understanding these seasonal growth patterns and what drives them is essential for identifying at-risk areas and adapting management practices accordingly. We analysed pasture growth across a large climatic and topographic gradient using high-resolution remote sensing data. Using cluster analysis, we identified three distinct pasture types based on the timing and intensity of growth phases. These patterns were spatially coherent and linked to key environmental drivers for three different years, 2017, 2018 and 2022. We found that summer temperatures, along with aridity and other water availability proxies such as soil properties and topography, play a major role in shaping growth timings and productivity regionally. These relationships were often non-linear, indicating potential tipping points in ecosystem function under future climate conditions. We further describe how these regions may behave under climate change scenarios with implications for management. Our results suggest that region-specific strategies should safeguard pasture productivity and resilience in the face of increasing climate variability.

Keywords: aridity, phenology, remote sensing, space-for-time, tipping point

Introduction

Rain-fed grasslands managed for livestock cover 40% of land surface, mostly in drylands (UNCCD, 2017). Their productivity and healthy ecological functioning sustain millions worldwide and provide essential ecological services such as carbon sequestration and storage (Bengtsson *et al.*, 2019). Annual growth dynamics in pastures and their phenology patterns are determined by historical and present climate conditions, which renders these ecosystems particularly vulnerable to climate change's predicted increases in temperature and precipitation variability (Berdugo *et al.*, 2020). Studying phenology patterns along large-scale spatial climate gradients provides insights into future trajectories of pasture phenology through a space-for-time substitution approach. However, phenology is shaped not only by climate but also by soil, topography, biodiversity and management, creating complex spatial and temporal patterns that vary with scale (Jenerette *et al.*, 2010). While local methods provide mechanistic insights, remote sensing (RS) enables large-scale monitoring, albeit with challenges of resolution and accuracy. New high-resolution datasets can overcome these limitations. This study uses high-resolution RS and machine learning to describe pasture phenology along a spatial climate

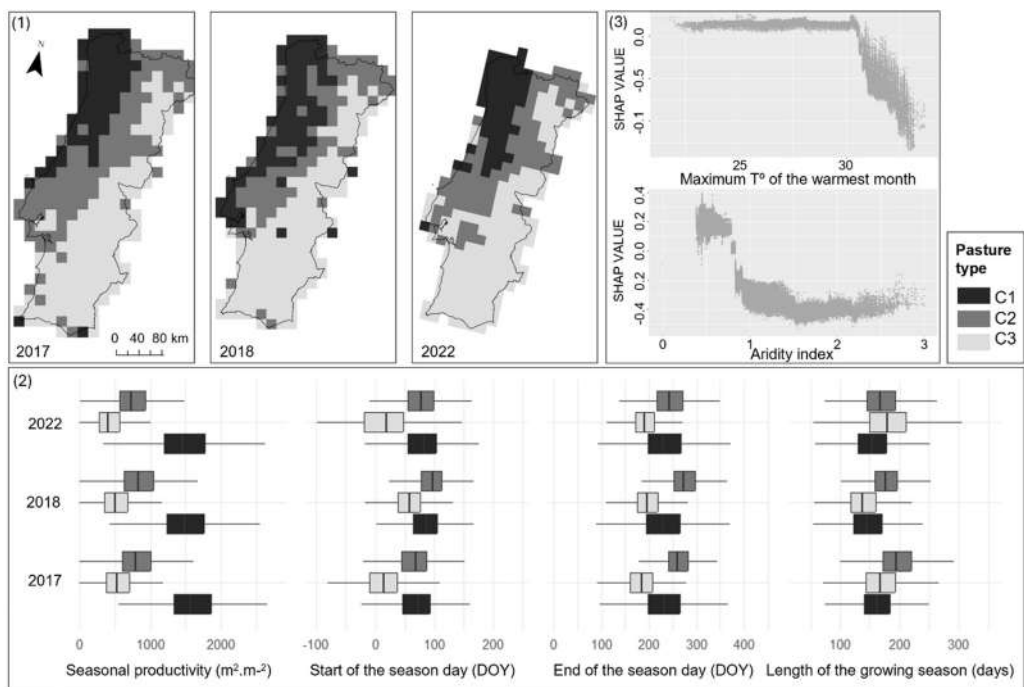


Figure 1. (1) Maps of dominant clusters, in each year; (2) Boxplots of seasonal productivity, green-up day, green-down day and length of the season in each cluster, by year. (3) Dependence plots of cluster classification to summer temperatures (classification of C2) and aridity (classification of C3), in 2018.

gradient and identify its main environmental drivers. We hypothesize that abiotic factors, particularly precipitation and temperature, shape distinct phenological typologies.

Materials and methods

We analysed annual phenological patterns of pastures in mainland Portugal (Mediterranean climate, strong latitudinal, longitudinal and elevational gradients) in 2017, 2018 and 2022, using High Resolution Vegetation Phenology Parameters from Copernicus. These consist of 13 yearly variables with 10 m resolution, such as seasonal productivity and start of season day (CLMS, 2023). Environmental predictors included topography (EU-DEM), topographic wetness index, climate (CHELSA, WorldClim, ERA5-Land, IPMA), soil (ESDAC chemical and physical properties), and socioeconomic indicators (BGRI population density, building vacancy). All variables were aggregated as mean values at the farm level using IFAP land-use polygons. K-means clustering (R, stats package) was used to identify phenology-based groups from the 13 phenology metrics. The optimal number of clusters ($k = 3$) was chosen using multiple indices and stability checks, aiming at balancing spatial continuity with interpretability. Relationships between clusters and environmental drivers were modeled using XGBoost, achieving between 68 and 69% accuracy. We used SHAP values to then quantify variable importance for each year.

Results and discussion

Clustering revealed three distinct pasture types aligned with north–south and east–west climate gradients (Figure 1(1)). Cluster 3 (C3, south/east) consistently had the earliest start, shortest season,

and lowest productivity (Figure 1(2)). XGBoost identified summer temperature and water availability (aridity index) as primary drivers (Figure 1(3)), with thresholds around 27–30°C and an AI of 0.79 (for 2018), similar in all three years. C3 pastures were linked to high aridity whereas C1 (north/littoral) was associated with higher wetness and C2 (north/centre) with cooler summer temperatures. These patterns indicate water limitation in the south/east and temperature limitation in temperate northern areas, modulated locally by soil and topography, are determining pasture phenology. Water availability strongly influenced productivity and phenological timing, with semi-arid areas showing earlier senescence and shorter seasons possibly due to water stress intensified by high radiation, high summer temperatures and evapotranspiration. Northern pastures lagged in green-up due to cooler pre-season temperatures but maintained growth longer, possibly due to cooler summer temperatures. Predicted climate change, which will cause the northward expansion of semi-arid zones, higher summer temperatures, and reduced rainfall, may shift northern phenology towards patterns closer to southern ones, particularly if identified tipping points ($AI < 0.8$) are crossed as suggested by our data. In the future, this could shorten seasons, lower forage quality and yield, increase costs, disrupt grazing schedules, and reduce biodiversity and ecosystem resilience. Local edaphic and topographic conditions may buffer or intensify climate impacts.

Conclusions

Current one-size-fits-all policies in mainland Portugal risk overlooking the differences identified in the current study. Management strategies must integrate climatic thresholds, seasonal growth trajectories, and site-specific conditions to safeguard productivity and ecosystem services under future warming and drying trends.

Acknowledgements

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Drought periods in summer increase nitrate leaching in cut grassland

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Abstract

In cut grassland the offtake of nitrogen (N) with harvested biomass is a major sink for N and often exceeds the N fertiliser input, resulting in comparatively low leaching losses of nitrate over winter. Climate change leads to generally drier summers with more frequent drought periods and increased precipitation during winter in regions with Atlantic climate. Prolonged drought can lead to reduced N efficiency during the vegetation period and, subsequently, to higher nitrate losses over winter even in cut grassland systems. We hypothesize that the combination of effects controlling the N-use efficiency during summer and the amount of leaching water and temperature during winter differ strongly among years.

In a four-year field experiment in North-West Germany with different systems of N return, including mineral and organic fertiliser, we found that N drought periods in summer subsequently resulted in higher nitrate-N leaching over winter (34–139 kg ha⁻¹ for synthetic fertiliser and 27–48 kg ha⁻¹ for organic fertiliser; N input: 0, 170, 240, and 360 (170 organic) kg ha⁻¹) compared to a year with average rainfall patterns (12–15 kg ha⁻¹). To avoid nitrate losses under changing climatic conditions a more dynamic adaptation of grassland management is required.

Keywords: climate change, grassland, N efficiency, nitrate leaching

Introduction

Usually, nitrate leaching is lower in grassland than in arable land. In cut grassland, especially, the offtake of nitrogen (N) with harvested biomass is a major sink for N (Kayser *et al.*, 2015; Nyfeler *et al.*, 2024). On more coarse-textured soils, prolonged drought periods can lead to reduced N efficiency in the vegetation period resulting in high residual soil N in autumn which poses a risk of higher NO₃-N leaching over winter (Kayser *et al.*, 2020). Against the background of increasing climatic change with drier summers and more frequent and increased precipitation in the winter months, this study investigates how the effects that control the N use efficiency during summer and the amount of leaching water and temperature during winter differ among years and how they affect NO₃-N leaching.

Materials and methods

We used data from a four-year experiment with organic and synthetic fertilisers on a cut grassland site in North-West Germany. The experiment was located in a maritime climate on a coarse-textured soil (sand to loamy sand). The long-term average temperature is 8.7°C and the average annual rainfall 778 mm (see Table 1). The sward is based on a ryegrass-dominated mixture (*Lolium perenne* L.) with

Table 1. Precipitation during the main vegetation period and leaching period for the years 2021–2024.

	2021/2022 (mm)	2022/2023 (mm)	2023/2024 (mm)	Long-term average (mm)
Precipitation vegetation period Apr–Sep	482	299	538	398
Precipitation leaching period Oct–Mar	404	398	658	380
Water leached over winter Oct–Apr	298	258	657	265

Regional long-term average (1991–2020) for precipitation; water leached from modelled data for the experimental site.

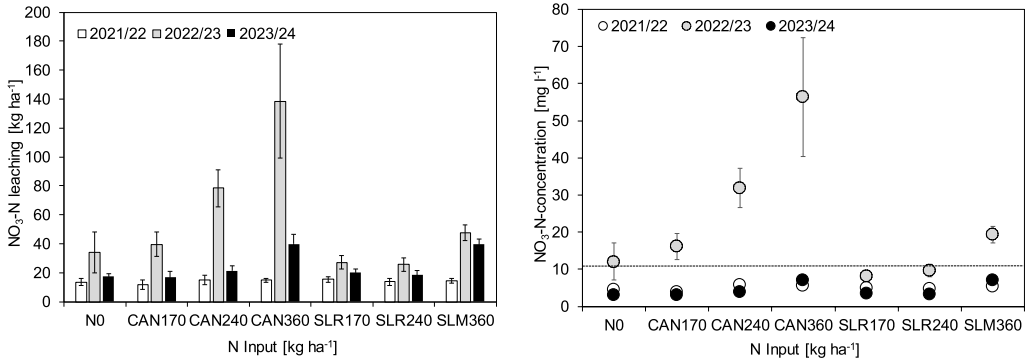


Figure 1. (A) NO₃-N leaching losses (kg ha⁻¹) over winter for 2021/22, 2022/23, and 2023/24, annual means with standard error (4 replications); (B) NO₃-N concentrations (mg l⁻¹) in leached water for 2021/22, 2022/23, and 2023/24 annual weighted means with standard error (4 replications); dotted line is drinking water threshold of 11.3 NO₃-N (mg l⁻¹).

no clover. From a wider experimental setting, we extracted treatments whose plots had suction cups installed. N input levels were 0, 170, 240 and 360 kg N ha⁻¹ in either synthetic (CAN, calcium-ammonium-nitrate) or organic (SLR, cattle slurry, dairy; applied by sliding-shoe technique; SLM, 360 kg N as 170 kg N ha⁻¹ with slurry and 190 kg ha⁻¹ CAN) form. The plots were mown four times and grab samples were taken from each plot. Herbage was oven-dried (60°C, 48 h), ground to <1 mm, and analysed for N. In each plot, six suction cups at 75 cm depth allowed sampling of leached water every 2–3 weeks. NO₃-N was analysed photometrically with a flow-injection analyser. Amount of water leached was determined by a water-balance model (Table 1; DVWK, 1996). For data analyses, mixed models and polynomial contrast analyses based on log-transformed values were conducted in Genstat (VSN International, 2024). The vegetation period 2022 had less rainfall than average and periods of drought (12 hot days and 1,146 hours of sunshine compared to 1 hot day and 795 hours of sunshine in 2021). In contrast, in 2023/24 rainfall was high in summer and winter with exceptionally high amounts of water leached (Table 1).

Results and discussion

Nitrate leaching losses and concentrations differed strongly among years with a significant interaction of year and fertiliser form (cattle slurry or CAN) (Figure 1). Polynomial contrast analysis for 2021/22 confirmed no increase in NO₃-N losses and concentrations with increasing fertiliser input, but a linear increase in 2022/23 for CAN ($P = 0.002$). This was not the case for input with cattle slurry in that year, but in 2023/24 NO₃-N leaching losses and concentrations showed a linear increase for fertilisation with slurry ($P < 0.001$) and for CAN ($P = 0.006$); with higher values mainly for N input >240 kg ha⁻¹ (N360).

Nitrate-N leaching losses were in a range of 12–15 kg ha⁻¹ for 2021/22 and 17–40 kg ha⁻¹ for 2023/24, but were exceptionally high in 2022/23, especially for N input with synthetic fertiliser (CAN) ranging from 34 kg ha⁻¹ for the Control to 78 kg ha⁻¹ for input of 240 kg N ha⁻¹ and 139 kg ha⁻¹ for N360. N leaching in that year was considerably less for fertilisation with cattle slurry and amounted to 27 for N170 and N240, and 48 kg ha⁻¹ for input of 360 kg N ha⁻¹ with slurry and CAN (170/190). This might be explained by NH₃ losses at application and incorporation of N in soil organic matter. Average annual NO₃-N concentrations were well below the threshold level for drinking water (11.3 mg l⁻¹) for 2021/22 and 2023/24, but exceed the threshold in several cases in 2022/23. Less rainfall and a drought period during summer that year led to reduced DM yields and less N offtake resulting in high residual N in autumn. N yields as a major N sink in grassland only amounted to, on average, 50%–70% of total N input.

Main drivers for high NO₃-N leaching in 2022/23 were the subsequently high NO₃-N concentrations for CAN combined with average water leached, and for 2023/24 the comparatively low NO₃-N concentrations combined with exceptionally high amounts of water leached (Table 1). In that respect, the year 2021/22 can be seen as a reference: low NO₃-N concentrations with a weak response to N-input level and average water leaching over winter.

At the same site, the drought period in 2018 also resulted in subsequent high NO₃-N leaching losses of 22 kg N ha⁻¹ at N0, and 146 and 221 kg N ha⁻¹, for N240 and N360 (combination of cattle slurry and CAN, 50/50 and 67/33), respectively; NO₃-N leaching was 87 kg N ha⁻¹ when N240 was applied solely as slurry (Kayser *et al.*, 2020). These findings are in line with findings by Klaus *et al.* (2020) from a pot experiment where the combined treatment of drought and fertilisation led to a three-fold higher nitrate leaching compared to the unfertilised control. Droughts reduce biomass yield and N offtake, which normally constitutes the main N sink, damage the sward and subsequently lead to increased mineralisation after rewetting of the soil (Klaus *et al.*, 2020; Kayser *et al.*, 2020). The reaction in grassland management to more frequent droughts is limited. Recommendations include: dividing fertilisation into several small applications; focusing on a first cut, and generally, to have realistic yield expectations, using variable cutting frequencies, and by the choice of more suitable species and varieties.

Conclusion

A key finding of the study is that longer periods of drought significantly increase the risk of nitrate leaching, even in cut grassland and more so from synthetic N fertiliser. This requires a more dynamic adaptation of grassland management to changing weather conditions. With on-going climate change it is likely that NO₃-N concentrations above the EU threshold will occur more often unless fertiliser recommendations are modified to better incorporate drought events. This is relevant for drinking water protection areas that are often located in regions with coarse-textured soils.

Acknowledgements

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Biomass yield potential of cut grassland on raised bogs in North-West Germany

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Abstract

Drained raised bogs in Northwest Germany are commonly used as grasslands. Along with current demands for re-wetting of peatland there is an ongoing debate on the appropriate nitrogen (N) fertilization of these grasslands: questions remain open on the actual biomass yield potential and the efficiency of fertiliser-N use, and how these are affected by differences in site conditions and by weather effects in different years. Drained raised bogs have a pronounced hydrological sensitivity. Drought conditions during the vegetation period, which under conditions of climate change are likely to occur more frequently, will lead to smaller potential biomass yields and a reduced N-use efficiency. We use data from field experiments with linearly increasing N input (0–600 kg N ha⁻¹ in 120 kg N ha⁻¹ increments; synthetic N) on cut grassland over five years and from five sites on drained raised bogs in Northwest Germany to model the yield potential and determine N response curves. There is a strong variability in yield among sites and years which is not reflected in official and uniform N fertiliser recommendations. In drier years, especially, yield curves flattened out much earlier reflecting a lower N need than assumed.

Keywords: climate change, fertiliser, N-use efficiency, nitrogen losses

Introduction

Drained raised bog soils (DA Bogs) in Northwest Germany are widely used for grassland production. This is associated with mineralization and losses of carbon and nitrogen (N), and measures like re-wetting are discussed. These soils have a low nutrient retention capacity and a pronounced hydrological sensitivity which further promotes N losses (Szajdak *et al.*, 2020). Current regulations in Germany stipulate a basic reduction of N fertilization by 50 kg N ha⁻¹ per year (BMEL, 2017) to reduce the risk of N losses (EU Nitrate Directive, 2008). However, this does not account for heterogeneous site conditions and variability of weather over the years (Deru *et al.*, 2019). This study analyses data from a fertilizer field trial on five sites on DA bogs over five years to assess N response patterns. The aim is to identify the main influencing factors on biomass yield on DA bogs as a prerequisite for the development of site-adapted N fertiliser strategies that consider both, yield performance and environmental protection.

Materials and methods

Data basis for the study were field experiments conducted by the Lower Saxony Chamber of Agriculture on five drained peatland (DA bog) sites and one marsh soil site (as a reference) in Northwest Germany over five consecutive growing seasons (2019–2023). The trials examined the yield response of grassland to six nitrogen fertilisation levels (0–600 kg N ha⁻¹, in 120 kg increments; synthetic fertiliser, calcium-ammonium-nitrate; plot size 7 × 1.5 m) with four replications. The use of synthetic

N and the range of N applied is far beyond that used in practice (EU Nitrate Directive, 2008) and was chosen to be able to assess and model a potential N response. To minimize carry-over effects, plots rotated between two field areas annually. Grassland swards were managed with up to five cuts per year depending on growth conditions. Swards were harvested and sampled by a forage harvester and dry matter content determined by oven-drying (60°C, 48 h). Samples from two replicates per treatment were analysed by NIRS (Near-Infrared Spectroscopy). Nitrogen content in biomass was calculated by dividing crude protein content by 6.25, and subsequently corrected by treatment-wise factors derived from analysis of a subsample by elemental analysis. Annual precipitation and growing condition varied among years (2019: 854 mm, drought period; 2000: 830 mm, dry spell in spring; 2021: 967 mm, evenly in summer; 2022: 744 mm, dry spell summer; 2023: 1,220 mm, dry spring, wet summer and winter).

Statistical analysis focused on DA bog sites and was conducted using linear mixed models to test effects of nitrogen level, year, and site (fixed or random), including interactions. Threshold models (Linear Plateau, Quadratic Plateau, Mitscherlich-Baule) were applied to determine optimal N input levels regarding DM yields. All analyses were conducted in R.

Results and discussion

Across the experimental years (2019–2023), the marsh site consistently produced higher DM yields than DA Bogs, especially at N input levels ≥ 360 kg N ha⁻¹. In contrast, yields on DA Bogs began to plateau around a N fertiliser input of 270–360 kg N ha⁻¹ and comparison of means did not show any differences between fertiliser level >360 kg N ha⁻¹. DM yields on DA bogs showed considerably inter-annual variability of on average over five years of 7.6 t ha⁻¹ to 11.6 t ha⁻¹. The highest yields were observed in the year 2021, and the lowest in 2019 (Figure 1A). Nitrogen fertilisation, site, and year had a significant effect on dry matter yield on DA Bogs (fertilisation: $p < 0.0001$; year: $p = 0.006$), with a significant interaction between fertilisation and year ($p = 0.0002$). Variance partitioning showed that location/site explained 70% of the total variance associated to random effects, exceeding the year-specific variance component (14%).

When modelled with location/site as a fixed factor, site-specific yield responses to nitrogen fertilisation were evident. High-yielding sites responded strongly to increasing N input reaching on average 17.6 t ha⁻¹ DM yield at 600 kg N ha⁻¹, whereas less responsive sites rarely exceeded 10.0 t ha⁻¹ DM yield. Nitrogen concentrations in harvested biomass were higher on DA Bogs than on marsh soil across all treatments. This partly compensated for lower DM yields on DA bogs and resulted in comparable or even higher N removal at similar input levels. The poorer model fit and higher response variability observed for DA Bogs reflect the combined effects of site heterogeneity, climatic extremes, and poorer retention capacity of nutrients and water. Compared to marsh soils, DA Bogs showed stronger yield reductions under hydrological stress like drought in 2019 and high rainfall in 2023. Overall, to achieve the same DM yield on DA Bogs as on marsh soils at an N input level of 360 kg N ha⁻¹ required approximately 85 kg N ha⁻¹ more N fertiliser input.

For DA Bogs, the Linear Plateau model estimated a N fertiliser threshold of 271 kg N ha⁻¹ (95% CI: 231–312 kg N ha⁻¹) with a plateau yield of 11.5 t ha⁻¹ (Figure 1B). This is in line with only small yield gains beyond the N input threshold. The Quadratic Plateau model (416 kg N ha⁻¹) and Mitscherlich-Baule model (95% threshold 312 kg N ha⁻¹) gave higher estimates. This can, at least partly, be explained by a tendency to overestimate optima with wider treatment intervals for the

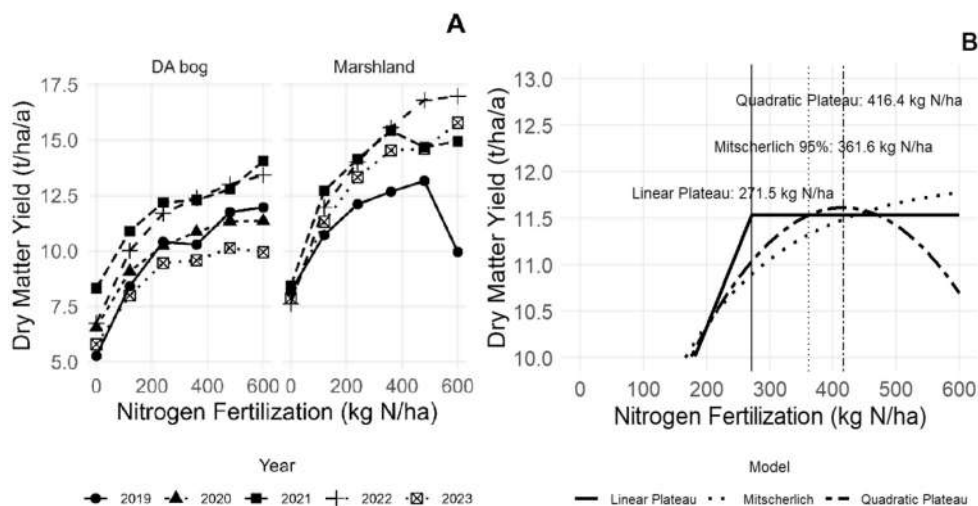


Figure 1. (A) Dry matter yield (t ha^{-1}) accumulated per year as a function of nitrogen fertilisation (kg ha^{-1}) from 2019 to 2023 on DA bog (average of 5 sites; and marshland (1 site)). (B) Yield response curves on DA Bogs based on three model types: Linear Plateau, Quadratic Plateau, and Mitscherlich-Baule.

Quadratic Plateau function (Taube, 2021), and also with smaller sample size and wider spaced data for the Mitscherlich-Baule model (Dhakal *et al.*, 2020). In our study, the Linear Plateau approach provided the most robust estimate of the nitrogen threshold on DA Bogs, identifying a range of 231–312 kg N ha^{-1} with narrow confidence intervals and good agreement with nitrogen balance data.

Conclusion

This study highlights the limited N-use efficiency and strong yield variability of DA Bogs. A modelled yield response curve following the Linear Plateau approach indicate a N fertiliser threshold of 271 kg N ha^{-1} . Nitrogen inputs beyond that level on DA Bogs offer little yield benefit and increase the risk of N surplus and N losses to the environment. Tailored strategies, such as cut-based or moisture-adapted fertilisation, could help to balance agronomic performance with environmental protection.

Acknowledgement

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Warm-climate forage performance and soil properties affected by soil, biochar, manure, and tillage

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Abstract

Biochar (BC) amendment to soils can negatively affect crops by binding essential nutrients; however, little data exist on its effects on warm-climate forage systems. We compared the effects of BC, dairy manure, soil type, and tillage on three forage species performance (*Cynodon dactylon* (perennial); *Zea mays* and *Sorghum drummondii*×*bicolor* (annuals)) and soil properties over three years. The experimental design included three split-block whole plots ($n = 3$). ANOVA and LSD tests were performed with R. BC did not affect any studied species production or nutritive value to cattle. Manure and tillage increased perennial and annual species production, respectively. Biochar increased soil C only for the perennial species while manure for all studied species, indicating potential benefits for soil health and carbon sequestration. BC did not affect soil P or N for any species, while manure increased both nutrients for the perennial species and only N for the annuals. Tillage increased soil P for all species while decreased N only for sorghum-Sudan (*Sorghum drummondii*×*bicolor*). Consider the crop type and nutrient makeup is key before choosing the proper tillage and amendments. Longer study periods may produce different results since, over time, BC can act as a slow-release source of nutrients.

Keywords: biochar, dairy manure, nutrient uptake, soil nutrients, tillage, warm-climate forages

Introduction

Biochar (BC) amendment to soils can affect grain crop and soil parameters; however, few data exist on its effects on warm-season forage performance and pasture soil properties. Several studies, mainly in annual row crops under different soil types, documented positive BC effects (10 to 42% average annual yield increases; Joseph *et al.*, 2021; Yi *et al.*, 2023) while others reported neutral or even negatives effects (decreases of 16%; Purakayastha *et al.*, 2019). However, biochar amendment efficacy for ameliorating pasture/crop soil characteristics and increasing forage yields and nutritive values in perennial and annual warm-climate forage systems has not been widely tested and remains controversial. We studied wood BC (BC (0, 5, 10 Mg DM ha⁻¹) and wood-Calcium modified-BC (5 Mg DM ha⁻¹)), dairy manure (0 and 10 Mg DM ha⁻¹), soil type (loamy sand (LS), sandy loam (SL), clay loam (CL)), and amendment application type (soil surface; topsoil tillage) effects on *Cynodon dactylon* (perennial); *Zea mays* (silage) and *Sorghum drummondii*×*bicolor* (annuals)) as well as soil properties after the third continuous crop season in southcentral North America.

Materials and methods

Our experiment was conducted over two growing seasons (March 2022 to November 2023). Results from the implantation year (growing season 2021) can be found in Hays *et al.* (2023) and Cooper *et al.* (2024). The trial was a four-factorial experiment ($r = 3$) since each species was analysed as an independent experiment: (1) three soil types; (2) two dairy manure application rates; (3) four BC application treatments; and (4) two application types. Amendment incorporation treatments (soil

surface; topsoil tillage) were applied in strip plots. Manure (no manure (0 Mg ha⁻¹); manure (10 Mg ha⁻¹)) and BC treatments (wood-BC: no biochar (0 Mg ha⁻¹); BC rates: low biochar (5 Mg ha⁻¹); high biochar (10 Mg ha⁻¹) and wood-Ca modified-BC (5 Mg ha⁻¹) were applied in a completely randomized design within each block. Wood-BC was applied in March 2021 and incorporated in tilled microplots, while wood-Ca modified-BC was applied in March 2022. The experimental units were 3 × 3 m microplots with 1.5-m alleys between. Samples were taken with a 0.25 m² metal frame from the inner 2 × 2-m microplot and harvesting 1 m of both central rows at 10-cm stubble height for the perennial and the annuals, respectively. Samples were dried and weighed to determine DM percentage, and ground for subsequent nutrient analyses. Total herbage N and C and minerals content were analysed. Soil sampling (0 to 10 cm) was conducted at the end of each growing season. Total C, N and P (Mehlich 1978, Mehlich 1984) were analysed. Statistical analysis was performed using a linear model implementation from the R Base package, and the Least Significant Difference (LSD) and p-values were calculated for multiple comparisons using the Agricolae R package. We considered significance at $P \leq 0.05$ and did not report individual probabilities in the text unless they were $P > 0.05$ and relevant to the discussion.

Results and discussion

Biochar did not affect ($P > 0.05$) any studied species production or nutritive value to cattle. Manure increased Bermudagrass production after the third year (2023) across all soil types, as well as application type did for sorghum-Sudan (except when grown in clay loam soil). Application type also increased maize production across all soil type but only after the second year (2022). Wood-BC (5 and 10 Mg ha⁻¹) both increased soil total C for the perennial species only after the second year (2022), while manure increased it for the perennial and the annual sorghum-Sudan also after the second year (2022). Biochar did not affect ($P > 0.05$) soil P or N for any species, while manure increased both nutrients for the perennial species (P increased in clay loam but decreased in loamy sand soil) and only N for the annual species. Tillage increased soil P for Bermudagrass and maize while decreased N for sorghum-Sudan only in 2022.

Within each year, interaction or main effect, and parameter, values within each column (upper case) and each line (lower case) followed by the same letter do not differ ($p \leq 0.05$) according to a least significant multiple mean separation.

Conclusions

Biochar showed a neutral effect on annual or perennial species production and nutritive value across all soils, 2 and 3 years after application. However, manure application increased Bermudagrass dry matter yield in clay loam in 2022 and across all soil texture types in 2023. In 2022 and 2023, most plant parameters for both, annual and perennial species, reached the minimum concentration required for adequate growing and finishing cattle (NASEM, 2016).

Furthermore, BC and manure application improved soil total C content after the second year, for Bermudagrass (14%) and for Bermudagrass and sorghum-Sudan (13.5%), respectively, indicating potential benefits for soil health and carbon sequestration. The fact that biochar did not increase soil total C for the annuals and that it only increased it during the second year for the perennial species, probably indicates the adverse priming effect of biochar.

Table 1. Dry matter yield (DMY) and soil parameters as affected by soil type, biochar, manure, and application type with Tukey's test displaying the mean±SE after the 2nd and 3rd continuous growing season of Bermudagrass, maize, and sorghum-Sudan.

2022				
maize				
DMY (kg/ha/year)				
Application type (p = 0.0001)	Soil surface	Topsoil tillage		
	7734.5±403.5 b	9623.1±381.0 a		
sorghum-Sudan				
DMY (kg/ha/year)				
soil×application type (p <0.0000)				
	Clay loam	Sandy loam	Loamy sand	
Soil surface	2884.7±241.5 b A	6973.8±494.7 a B	6195.1±1079.7 a B	
Topsoil tillage	2573.9±158.4 b A	9828.9±808.1 a A	9039.7±255.6 a A	
Total C (g / kg)				
Manure (p = 0.0002)	No manure	Manure		
	43.63±2.60 b	49.97±2.91 a		
Total N (g / kg)				
Application type (p = 0.0007)	Soil surface	Topsoil tillage		
	3.75±0.13 a	2.94±0.15 b		
Bermudagrass				
Total C (g / kg)				
Biochar (p = 0.0012)	No biochar	Low biochar	High biochar	Calcium biochar
	42.23±3.53 c	47.41±3.72 ab	48.93±3.99 a	45.21±3.43 bc
Manure (p = <0.0000)	No manure	Manure		
	43.1±2.4 b	48.8±2.7 a		
2023				
maize				
P (mg / kg)				
Application type (p = 0.0092)	Soil surface	Topsoil tillage		
	38.87±5.18 b	51.55±5.88 a		
Total N (g / kg)				
Manure (p = 0.0462)	No manure	Manure		
	2.30±0.09 b	2.53±0.09 a		
sorghum-Sudan				
Total N (g / kg)				
Manure (p = 0.0046)	No manure	Manure		
	2.41±0.08 b	2.73±0.09 a		
Bermudagrass				
DMY (kg/ha/year)				
Manure (p = 0.0002)	No manure	Manure		
	7706 7706.9±248.4 b	9239.9±248.3 a		
P (mg / kg)				
soil×manure (p = 0.0009)				
	Clay loam	Sandy loam	Loamy sand	
No manure	8.46±1.17 b B	76.36±4.28 a A	56.79±4.10 a A	
Manure	16.24±1.17 c A	61.91±4.29 a A	36.01±4.09 b B	
Application type (p = 0.0390)				
Total N (g / kg)	Soil surface	Topsoil tillage		
Manure (p = 0.0462)	29.24±2.46 b	37.60±2.45 a		
	No manure	Manure		
	2.38±0.09 b	2.67±0.37 a		

Biochar did not affect soil P or N for any species, while manure increased both nutrients for the perennial species and only N for the annuals, indicating that fertilizer amendment should be based on manure N. Tillage increased soil P for Bermudagrass and maize while decreased N for sorghum-Sudan in 2022. It is essential to consider crop type and nutrient makeup before choosing the proper tillage and amendments. Longer study periods may produce different results since, over time, BC can act as a slow-release soil nutrient source.

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Effect of soil receiving different long-term fertilizer treatments on *Plantago lanceolata* characteristics

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Abstract

This study examined the effects of soil receiving different long-term fertilizer treatments on the growth and biomass of *Plantago lanceolata* cultivated in pots. Soil was collected from a grassland trial in Černíkovec (established in 1966), which included 11 treatments in 1991: five actively fertilized treatments with PK alone, N+PK (N₅₀PK, N₁₀₀PK, N₁₅₀PK and N₂₀₀PK) at N: 50 to 200, P: 40, K: 100 kg ha⁻¹ year⁻¹; five treatments where fertilization ceased (T) in 1991 but received the same levels as the actively fertilized treatments before 1991; and one unfertilized control. The highest *P. lanceolata* sward height (>16.0 cm), number of flowers (>6), above-ground dry weight biomass (>2.1 g/pot), and total dry weight biomass (>4.4 g/pot) were recorded in the actively fertilized N+PK (N_(50 to 200)+PK)- and PK-alone fertilized treatments, along with some fertilizer-terminated treatment (e.g., N₂₀₀PKT). However, no differences were found between other terminated treatments (e.g., PKT, N_(50–150) PKT) and the control. Similarly, there were no differences in the number of leaves and longest leaf width between any of the treatments, except for the lower leaf width values in the control. Some ceased treatment maintained stable soil P and K, supporting sustained productivity. Soil phosphorus and potassium were positively correlated with key *P. lanceolata* growth and biomass traits. These findings highlight the lasting legacy of fertilization on soil fertility and the potential for natural recovery in grasslands, although with limitations in productivity in the absence of nutrient inputs.

Keywords: fertilization cessation, nutrient legacy effects, plant traits, ribwort plantain, soil nutrient

Introduction

Fertilization is vital for boosting plant biomass and nutrient management in grasslands, especially amid rising demand for ruminant products. However, long-term use impacts soil properties and sustainability. In Czechia, long-term fertilization trials have assessed productivity, species richness, and soil quality (Honsová *et al.*, 2007; Hrevušová *et al.*, 2009; Hrevušová *et al.*, 2015). Despite existing knowledge, linking past fertilizer use to current soil function and plant growth remains inconsistent. Long-term fertilization effects can last over a decade, slowing recovery in alluvial grasslands. This highlights the limits of single-season studies and the need for long-term monitoring, as legacy effects are influenced by climate and other external factors. This study examined how soils from such long-term trials affect *P. lanceolata* growth, hypothesizing that continued fertilization enhances biomass compared to unfertilized soils, with terminated treatments showing intermediate responses.

Materials and methods

The *P. lanceolata* pot experiment was conducted in a greenhouse in 2019 in Liberec (50°46'N, 15°2'E), located 90 km north of Prague. Soil samples for the greenhouse experiment were collected from the

0–30 cm depth of a long-term fertilization trial (11 treatments) on a flat alluvial grassland near the village of Černíkovice, 35 km south of Prague (49°47'N, 14°45'E; elevation: 363 m a.s.l.), conducted between 1991 and autumn 2018. The long-term experiment began in 1966 with four treatments (Control, PK, N₁₀₀PK, N₂₀₀PK). The experiment was arranged into four randomized blocks, each with six treatments (Control, PK, N₁₀₀PK, N₂₀₀PK, N₃₀₀PK, N₄₀₀PK) on 5 × 6 m plots, with N₃₀₀PK and N₄₀₀PK treatments added in 1975. In 1991, nitrogen rates were halved, and each plot was divided into two 5 × 3 m subplots. This resulted in five actively fertilized treatments (PK, N₅₀PK, N₁₀₀PK, N₁₅₀PK, N₂₀₀PK), where PK alone received P: 40 and K: 100 kg ha⁻¹ year⁻¹, and the N_(50–200)+PK treatments received the same PK dosage with nitrogen levels ranging from 50 to 200 kg ha⁻¹ year⁻¹. Additionally, five treatments with terminated fertilization (PKT, N₅₀PKT, N₁₀₀PKT, N₁₅₀PKT, N₂₀₀PKT) had received the same fertilization rates as their actively fertilized counterparts until 1991, after which fertilization was discontinued. One plot remained as the unfertilized control. The *P. lanceolata* seeds were sown in May 2019 in pots (1 litre) containing soil from long-term experimental plots. The pot experiment was conducted on 11 soil treatments replicated 4 times. The pots were arranged in a completely randomized design. *Plantago lanceolata* characteristics were measured before the final harvest, and the experiment was completed in July 2019. Soil properties (pH, P, K, Ca, Mg, C, N) and *P. lanceolata* growth traits, such as number of leaf and flower, sward height, longest leaf width, and biomass traits (leaves, stems, flower blooms, and roots dry weight), were measured. Data were analysed using R (V.4.4.3) with one-way ANOVA and post-hoc Tukey test ($P < 0.05$), along with regression analysis to explore relationships between soil and plant traits.

Results and discussion

Plantago lanceolata growth and reproductive traits such as sward height, and number of flowers varied significantly among treatments (Table 1). Actively fertilized treatments (N_(50–200) PK and PK alone) generally produced the tallest plants (>16.0 cm), most flowers (>6), and moderately wider leaves (>2.2 cm), though some terminated treatment (e.g., N₂₀₀PKT) showed similar results. The control showed the lowest values overall, though some terminated treatments performed similarly (e.g., PKT, N_(50–150) PKT) (Table 1). Similarly, biomass attributes such as total aboveground biomass, and total above- and belowground dry weight biomass, were significantly affected by the treatments (Table 1). However, there were no significant differences in the number of leaves among the treatments. The highest values for total aboveground biomass (>2.1 g/pot), and total above- and belowground biomass (>4.4 g/pot) were recorded in actively fertilized treatments (N_(50–200) PK and PK alone) and some fertilization-terminated treatments, particularly N₂₀₀PKT. In contrast, the control and certain fertilization-terminated treatments (e.g., PKT, N_(50–150) PKT) exhibited lower values for these *P. lanceolata* characteristics. This is in line with Hejčman *et al.* (2007), who reported that legacy soil nutrient levels can sustain plant growth and biomass even after fertilization has ceased.

Soil phosphorus and potassium were positively correlated with key *P. lanceolata* growth and biomass traits. For example, soil phosphorus significantly affected the number of flowers ($R^2 = 0.36$, $p < 0.03$), leaf biomass ($R^2 = 0.31$, $p < 0.001$), aboveground biomass ($R^2 = 0.32$, $p < 0.001$), and total above and below-ground biomass ($R^2 = 0.30$, $p < 0.001$) confirming their potential as predictors of *P. lanceolata* growth and biomass performance. Similarly, Moeneclaey *et al.* (2022) reported increases in the growth and biomass of grasses and forbs with rising soil phosphorus levels.

Table 1. Analysis of variance (ANOVA) of *P. lanceolata* growth and biomass characteristics.

Treatment	SH (cm)	L _L W (cm)	LN (number)	FN (number)	ADW (g/pot)	TDW (g/pot)
Control	12.1 ± 1.6 b	2.0 ± 0.3 b	17.2 ± 3.9 a	4.1 ± 1.1 c	1.5 ± 0.2 b	2.6 ± 0.7 b
PKT	12.3 ± 1.8 b	2.1 ± 0.3 ab	17.6 ± 3.3 a	4.9 ± 1.7 c	1.5 ± 0.3 b	3.0 ± 0.8 b
PK	16.2 ± 1.5 a	2.3 ± 0.2 a	21.0 ± 4.5 a	7.1 ± 1.8 a	2.1 ± 0.3 a	4.1 ± 1.0 a
N ₅₀ PKT	12.5 ± 1.7 b	2.0 ± 0.3 ab	17.4 ± 3.1 a	4.5 ± 1.7 c	1.6 ± 0.3 b	3.1 ± 0.8 b
N ₅₀ PK	16.4 ± 1.6 a	2.2 ± 0.2 ab	18.2 ± 3.7 a	6.6 ± 1.6 ab	2.1 ± 0.4 a	4.0 ± 1.1 a
N ₁₀₀ PKT	12.5 ± 1.2 b	2.2 ± 0.3 ab	17.6 ± 3.9 a	4.9 ± 1.3 c	1.6 ± 0.3 b	2.7 ± 0.7 b
N ₁₀₀ PK	16.3 ± 2.0 a	2.3 ± 0.3 a	18.2 ± 3.5 a	6.7 ± 1.8 a	2.1 ± 0.3 a	4.2 ± 1.0 a
N ₁₅₀ PKT	13.5 ± 1.3 b	2.1 ± 0.2 ab	18.8 ± 4.2 a	5.0 ± 1.4 bc	1.6 ± 0.4 b	3.1 ± 0.7 b
N ₁₅₀ PK	16.8 ± 1.7 a	2.3 ± 0.3 a	19.9 ± 3.3 a	6.9 ± 1.8 a	2.2 ± 0.4 a	4.3 ± 1.1 a
N ₂₀₀ PKT	16.6 ± 2.0 a	2.3 ± 0.3 a	20.5 ± 3.5 a	6.7 ± 1.2 a	2.2 ± 0.4 a	4.3 ± 1.0 a
N ₂₀₀ PK	16.8 ± 2.0 a	2.3 ± 0.3 a	20.9 ± 4.8 a	7.2 ± 1.7 a	2.4 ± 0.4 a	4.4 ± 1.2 a
p value	<0.001	<0.01	NS	<0.001	<0.001	<0.001

SH, sward height; L_LW, longest leaf width; LN, number of leaves; FN, number of flower blooms; ADW, total above-ground dry weight biomass; TDW, total above- and below-ground dry weight biomass. Different letters in the same column indicate significant differences (post hoc Tukey test; same letters indicate no difference). Values are mean ± SD.

Conclusion

Some treatments where fertilization had ceased maintained stable levels of soil phosphorus and potassium, contributing to sustained *P. lanceolata* productivity over time. Terminated treatments, particularly N₂₀₀PK, played a proportional role in supporting plant growth and biomass comparable to that of actively fertilized treatments, indicating effective support without continuous fertilization even after 27 years. These results emphasize the enduring impact of past fertilization on soil health and the potential for partial natural recovery in grassland ecosystems, although productivity remains limited without continued nutrient input.

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The effect of regenerative agriculture practices on soil health: case studies from the Tech4RegenAg project

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Abstract

In Portugal, as in most mediterranean regions, regenerative agriculture principles are key to improve pasture soil health and mitigate the effects of climate change. The Tech4RegenAg project aims to demonstrate the feasibility of implementing such systems in multiple environments across Portugal. Three pilot studies were implemented in different agricultural contexts: in the São Miguel Island of the Azores in a dairy cow farm (A), and in Beira Baixa region in two distinct farms of dairy goats (B) and beef cattle (C). In farm (A), the selected field was one dedicated to the production of maize and grass for silage. Farm (B) was monitored in respect to an annual pasture. In farm (C), plots with permanent grasslands and with annual pastures, with different degrees of soil mobilization, were selected for time-dependent comparison of the effects of different practices. In each case, the soil microbiome (16S, ITS) was studied in combination with standard soil physical-chemical parameters. Preliminary results reflect different management practices, with heavily fertilized and mobilized fields having excessive nutrients and low base exchange capacity, whereas soils under low-till and use of cover crops reduce chemical fertilizer inputs and maximize soil biodiversity.

Keywords: climate change, regenerative agriculture, soil health, soil microbiome

Introduction

The Mediterranean region is experiencing soil erosion at a faster rate than the rest of Europe, primarily due to its higher susceptibility to climate change events (Ferreira *et al.*, 2022). By implementing regenerative agriculture practices that reduce tillage and increase soil cover, one can enhance soil health, which in turn boosts plant health and productivity. As a result, there is a decreased reliance on synthetic fertilizers and pesticides, which can improve farmers' profit margins while promoting environmental sustainability (Musto *et al.*, 2023). Additionally, technologies like next generation sequencing (NGS) can provide valuable insights into soil health and function (Garg *et al.*, 2024). This study aims to describe the measurable benefits of regenerative agriculture practices in relation to soil health.

Materials and methods

This study was conducted in Portugal, specifically in the Azores and Beira Baixa regions, between 2024 and 2025. In the Azores, a dairy cow farm (Farm A) was monitored for approximately 2 hectares of land, farmed conventionally, to produce two monocrops for silage: maize and ryegrass. In the Beira Baixa region, a dairy goat farm (Farm B) with a 4-hectare plot with a biodiverse, annual pasture; and lastly, an organic beef cattle farmer (Farm C) employing a traditional grazing system with beef cows. In farm C, soil samples were collected from three different plots: (i) a plot where cows had grazed (ii)

Table 1. Summary of the physical-chemical analysis of soils samples across three different farms in the Azores (São Miguel Island, Farm A) and Beira Baixa (Farms B and C).

Variable	Farm A	Farm B	Farm C		
			Grazed	No till	No graze, no till
pH (H ₂ O)	6.2	6.8	6.2	5.9	6.4
Organic matter (% of soil)	7.20	2.2	1.4	1.2	1.3
Total Nitrogen (% of soil)	0.41	0.14	0.10	0.07	0.07
Carbon:Nitrogen	10.2	9.3	7.5	10.1	10.9
Phosphorus (mg P ₂ O ₅ /kg soil)	<LQ	228	79.2	119	<LQ
Potassium (mg K ₂ O /kg soil)	299.3	348	106.9	176	130
Calcium (mg CaO /kg soil)	2841	1100	752.8	529	1500
Sulphur (mg S/kg soil)	<LQ	14,1	4,8	3.8	5.6
Molybdenum (mg Mo/kg soil)	<LQ	<LQ	< LQ	< LQ	<LQ
Base exchange capacity (cmol ⁺ /kg soil)	11,5	4,45	4.0	4.15	11.8
Base saturation (% of cation exchange capacity)	100	100	92.0	90.3	98.4

LQ, limit of quantification.

a plot that had not undergone any soil disturbance for at least a decade, and (iii) a plot without any disturbance. The soil samples were analysed for physical and chemical parameters, including pH, total nitrogen, organic matter (OM), phosphorus, potassium, calcium (Ca), sulphur, molybdenum (Mo), cation exchange capacity, and base saturation. These samples were analysed with NGS 16S (bacteria) and ITS (fungi) amplicon sequencing.

Results and discussion

The physical-chemical analysis of the sampled soils is presented in Table 1.

Overall, the levels of OM were lower in Beira Baixa, particularly on Farm C. The pH was close to neutral on most farms, likely due to liming, as evidenced by the high presence of Ca and base saturation. Farm B exhibited optimal soil conditions, such as low presence of pathogens and high metabolic activity of key nutrient pathways. All samples showed high abundance of *Pythium sp.* fungi, which can cause root rot and adversely affect crop yields (Harvey *et al.*, 2008). This issue was more pronounced in plots with frequent soil mobilization, such as Farm A, which produces two annual crops each year with conventional tillage. Biodiverse cropping systems and subsoiling are potential means to facilitate the early-stage implementation of cover crops or pastures (Raper, 2005). Additionally, the use of subsoiling could help incorporate keylines in the dryland plots of Beira Baixa, improving water retention (Sun *et al.*, 2017). For instance, in the Azores, transitioning from monocrop ryegrass-maize rotations to a mixture that includes *Lolium multiflorum* (40%), *Bromus catharticus* (26%); and clovers (34%, including *Trifolium resupinatum*, *Trifolium michelianum*, *Trifolium squarrosum* and *Trifolium incarnatum*) can contribute to a more diverse diet for grazing ruminants, as well as create the conditions for a more resilient soil, when accompanied with minimum tillage. Additionally, soil inoculation with *Trichoderma sp.* could incrementally reduce the pressure of pathogenic fungi, reducing productivity losses and nutrient loss (Naseby *et al.*, 2000). Direct seeding would be recommended for all plots in order to prevent further soil compaction and losses of organic matter. It is important to note that micronutrients, such as Mo, are often below the limit of quantification. This suggests that legumes may not reach their potential in producing crude protein, given the critical role this nutrient plays in the nitrogenase activity of *Rhizobium* (Li *et al.*, 2023; Alam *et al.*, 2015).

Conclusions

These results confirm the potential for improving both conventional and organic agricultural systems by adopting regenerative and context-based approaches. Employing conventional practices, such as tillage, leads to soil compaction and loss of soil microbiome diversity. This work, albeit limited in number of farms and replicates, lays a foundation for future studies, providing a science-based approach to measure the impact of conventional practices to determine the outcomes to improve following regenerative agriculture principles. This transition is crucial for farmers, given the current political and ecological landscapes. It is essential to demonstrate to farmers that increasing the environmental sustainability of their farms does not necessarily lead to a loss in productivity.

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Effects of nitrogen rate and white clover inclusion on soil microbial communities in grassland swards

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Abstract

Soil microbial communities play a critical role in regulating nutrient cycling and supporting grassland resilience. Nitrogen application rates and plant species composition can strongly influence soil microbial communities and their function. This study examines the effects of varying inorganic N rates and sward types, perennial ryegrass (*Lolium perenne* L.; PRG) and white clover (*Trifolium repens* L.; WC) on soil microbial communities in a long-term farm systems trial at Teagasc Moorepark, Fermoy, Cork, Ireland. Soil cores were obtained in May 2024 from grassland swards under three treatments: PRG-only (200 kg N ha⁻¹ year⁻¹), PRG-WC (150 kg N ha⁻¹ year⁻¹), and PRG-WC (100 kg N ha⁻¹ year⁻¹). These soil cores underwent shotgun metagenomic sequencing to quantify the microbial communities present. Multivariate analyses revealed no significant effects of the combined treatments, which varied in nitrogen application rates and sward types, on microbial community composition. Organic matter and soil pH were identified as key drivers significantly correlated with various bacterial genera ($p < 0.01$ and $p < 0.05$ respectively). These findings suggest that soil physicochemical properties may play a greater role on microbial community composition in grasslands, compared to sward type and N application rates.

Keywords: clover, grass, organic matter, soil microbiome, soil pH

Introduction

Grassland soils are the foundation for pasture-based livestock systems, supporting nutrient cycling, water retention, and forage production. In intensively managed grazing systems dominated by monoculture perennial ryegrass (*Lolium perenne* L.; PRG) swards, high levels of inorganic nitrogen (N) fertiliser applications (>200 kg N ha⁻¹ year⁻¹) are applied to achieve high levels of herbage production (Enriquez-Hidalgo *et al.*, 2016). While effective, excessive use of high inorganic N inputs can lead to shifts in the diversity and functional traits of soil microbial communities, which are critical for overall soil health and fertility (Leff *et al.*, 2015). One method to reduce reliance on inorganic N inputs is incorporating legumes such as white clover (*Trifolium repens* L.; WC) into PRG swards (Lüscher *et al.*, 2014), as WC can fix atmospheric N through biological nitrogen fixation (BNF), making it available for plant growth (Enriquez-Hidalgo *et al.*, 2016). White clover has been shown to fix up to 200 kg N ha⁻¹ year⁻¹ through BNF, but this process can be significantly reduced by up to 90% under high inorganic N fertiliser application rates (>150 kg N ha⁻¹ year⁻¹) compared to unfertilised swards (Enriquez-Hidalgo *et al.*, 2016). While the advantages of PRG-WC swards aboveground in terms of improved herbage production, forage quality, and animal performance (Lüscher *et al.*, 2014) are well known, their impact belowground on the soil microbiome is less understood. Soil microorganisms especially nitrogen-fixing bacteria, including rhizobia, play a vital role in nutrient cycling, regulating soil biochemical processes, and sustaining the ecological resilience of grassland ecosystems (Köhl *et*

al., 2014). The objective of this study is to assess how WC inclusion in PRG swards receiving varied inorganic N application rates influence soil microbial communities in a long-term grazing system.

Materials and methods

The experiment was conducted at Teagasc Moorepark, Fermoy, Co. Cork, Ireland, as part of a long-term farm systems trial. The soil type is characterised as a free draining acid brown earth with a sandy loam-to-loam texture, with phosphorus and potassium levels classified with an average index of 3 and 4 respectively (Wall and Plunkett, 2021), and an average pH of 6.4. Soil samples ($n = 33$) were collected in May 2024 from paddocks assigned to three treatments: Grass200 (PRG-only, 200 kg N ha⁻¹ year⁻¹), Clover150 (PRG-WC, 150 kg N ha⁻¹ year⁻¹), and Clover100 (PRG-WC, 100 kg N ha⁻¹ year⁻¹), with eleven paddocks per treatment ($n = 11$). Within each paddock, samples were taken in a W-shaped pattern using a sterilised soil core sampler (10 cm depth), collecting a minimum of 20 soil cores per paddock. Cores were homogenised, and an approx. 1 g subsample of composite soil was transferred into sterile centrifuge tube, and snap frozen in liquid N on site, and transported to a -80°C freezer. Soil DNA was extracted using the DNeasy PowerSoil Pro Kit (Qiagen) following the manufacturer's guidelines and quantified with a Qubit dsDNA Broad Range Assay (Life Technologies). Sequencing libraries were prepared using a miniaturised Nextera XT protocol on a Labcyte Echo 525 and shotgun metagenomic sequenced on an Illumina NovaSeq 6000 (2 × 150 bp) at the Teagasc Sequencing Centre. Raw paired end reads were quality filtered using Fastp following removal of the PhiX spike in control with Fasnica and Bowtie2. Each soil sample yielded ~20.3 million (M) paired end reads (± 4.0 M SD). Taxonomic classification was performed using Kraken2 with a custom GTDB-tk database, and Bracken was applied to refine abundance estimates at genus level. Data were analysed in R (v4.4.2; R Core Team, 2024) using packages from the tidyverse ecosystem, along with vegan. Beta diversity (Bray-Curtis dissimilarity) with treatment, soil pH, organic matter, and their interactions included as fixed factors were evaluated using PERMANOVA (adonis2, permutations = 999). All p -values were adjusted for multiple comparisons using the Benjamini-Hochberg procedure to control the false discovery rate (FDR), and statistical significance was set at adjusted $p \leq 0.05$.

Results and discussion

Soil microbial community composition at the genus level had no statistically significant differences between treatment types. The treatment explained 5.7% of the variation in microbial community composition at the genus level ($R^2 = 0.057$, $F = 0.88$, $p = 0.615$), which is consistent with Wang *et al.* (2022), who also found soil microbial community composition did not respond to N treatments. Both organic matter and soil pH significantly influenced microbial community composition across all paddocks, soil pH averaged 6.45 ± 0.25 SD (range 6.1–6.9), while organic matter averaged $7.97 \pm 1.11\%$ SD (range: 6.2–10.4%) (Table 1). These findings align with Zhelnina *et al.* (2015), who found that soil pH was the primary factor influencing microbial composition, having a stronger effect than fertiliser applications or plant species diversity due to its effects on microbial activity and nutrient availability.

Organic matter content was significantly correlated with 33 bacteria genera (adjusted $p \leq 0.05$), with seven of them meeting the strongest Spearman rank correlation associations of ($|\rho| \geq 0.70$). The strongest positive and negative correlations were observed for *Deftuviicoccus* and *Acidibrevibacterium* (positive) and *Saccharomonospora* and *Thermopolyspora* (negative) (Table 2). These patterns suggest distinct ecological preferences amongst taxa with soil organic matter, Pérez-Guzmán *et al.* (2021) found

Table 1. Effects of organic matter, soil pH, on all three treatments on soil microbial composition

Variable	<i>R</i> ²	<i>F</i>	<i>p</i> -value
Organic matter	0.0848	2.75	0.008
Organic matter×Treatment	0.0458	0.74	ns
Soil pH	0.0707	2.27	0.017
Soil pH × Treatment	0.0644	1.04	ns

*R*² shows sequential variance explained; *F*, pseudo-*F* statistic; *p*-values are based on 999 permutations using adonis2 with by = "terms" (vegan package); ns, not significant (*p* > 0.05).

Table 2. Soil microbial genera most strongly correlated with organic matter across all three treatments

Genus	Spearman ρ	Adjusted <i>p</i> -value	Mean abundance	SE	IQR
<i>Deffluviococcus</i>	+0.7474	0.000172	0.0575	0.0041	0.0500
<i>Acidibrevibacterium</i>	+0.7080	0.000966	0.0075	0.0008	0.0025
<i>Saccharomonospora</i>	−0.8067	0.000029	0.0581	0.0020	0.0100
<i>Thermopolyspora</i>	−0.7797	0.000084	0.0266	0.0019	0.0100

ρ , Spearman rank correlation coefficient; adjusted *p*-value, FDR-corrected *p*-value (Benjamini–Hochberg); mean abundance, average relative abundance across all samples and treatments; IQR, interquartile range.

that functional microbial groups were positively associated with soil organic matter in perennial grass systems.

Conclusion

Organic matter and soil pH had a significant effect on soil microbial community composition, while treatment effects, including sward type and chemical N fertiliser, were not significant. However, as this study was conducted from a single time point, future research for multiple time points is needed to capture the potential seasonal or yearly variations in the soil microbiome.

Acknowledgements

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Restoration potential of two native grasses (*Stipa tenacissima* and *Lygeum spartum*) in degraded arid and semi-arid rangelands

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Abstract

In eastern Morocco, rangelands mainly situated in arid and semi-arid zones, are undergoing increasing degradation, threatening pastoral production systems and livelihoods of rural communities. This study investigates the restoration potential of two native perennial grasses, *Stipa tenacissima* and *Lygeum spartum*, known for their remarkable ability to survive in harsh and arid climatic conditions. Germination experiments conducted under different pretreatments revealed high germination rates for both species. Observations in field further demonstrated the ecological adaptability of *S. tenacissima*, while forage analyses indicated that both grasses maintain acceptable nutritional value. These results show that integrating these native grasses into restoration programmes could contribute to the slowing down of pasture degradation and support the recovery of these ecosystems.

Keywords: drought resilience, ecological restoration, forage quality, *Lygeum spartum*, rangeland degradation, *Stipa tenacissima*

Introduction

In the drylands of arid and semi-arid regions, rangelands serve as essential ecological and socio-economic systems. They not only support livestock and pastoral livelihoods but also contribute to the conservation of plant and animal biodiversity (FAO, 2020). Unfortunately, these ecosystems are increasingly threatened by a combination of anthropogenic pressures and climate-related challenges, further compromising plants cover and forage availability (IPCC, 2022). In the Mediterranean rangelands of eastern Morocco, the decline of native perennial grasses has significantly worsened the advancing degradation, compromising ecological stability and sustainability (Le Houérou, 2001). The ability of perennial grasses like *Stipa tenacissima* and *Lygeum spartum* to adapt to stressful conditions makes them particularly promising for rehabilitation and long-term sustainability of pastoral systems.

Materials and methods

The study examined two native perennial grasses, *Stipa tenacissima* L. and *Lygeum spartum* L., typical of Mediterranean steppe ecosystems in North Africa. Seeds were collected in the summer of 2023 from degraded rangelands, particularly in Jerada Province (Oriental Morocco). Germination trials were conducted under controlled laboratory conditions (20°C, 12 h light/12 h dark), with seeds sterilized and soaked for 24 or 48 h before placement in moist Petri dishes (4 replicates of 25 seeds per treatment). Germination was recorded daily over 21 days.

Field monitoring of *S. tenacissima* was carried out in the arid zone of Ain Beni Mathar over one year, with monthly evaluation of the species survival to assess drought adaptation under natural conditions. For forage quality, aerial biomass of both species was dried, ground, and analysed for crude protein

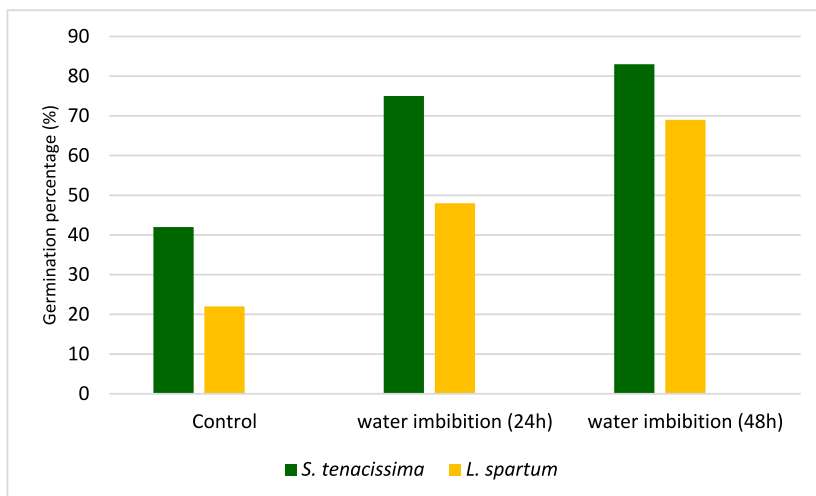


Figure 1. Germination percentage of *S. tenacissima* and *L. spartum* seeds following water imbibition pre-treatments.

(Kjeldahl method), fibre fractions (NDF and ADF via Van Soest), and ash content, following standard protocols (Van Soest *et al.*, 1991; AOAC, 2019)

Results and discussion

Stipa tenacissima and *L. spartum* both showed marked increase in germination rate after water imbibition treatments. Germination rates increased from 42 to 83% for *S. tenacissima* and from 22 to 69% for *L. spartum* following 48 hours of soaking (Figure 1). This pre-sowing treatment reduces physiological dormancy and improves seedling establishment, reinforcing the potential of these species for rangeland restoration, in line with findings reported by Aidi *et al.* (2012).

Field monitoring of *Stipa tenacissima* in Aïn Beni Mathar showed a sharp decline in survival during the first three months post-plantation (from 100 to 49%) (Figure 2), likely due to transplant shock and slow root development under moisture stress. Since the fourth month, survival stabilized between 45% and 51%, suggesting successful adaptation to arid conditions (Figure 2). These results highlight the species drought tolerance after establishment, confirming its suitability for long-term dryland rehabilitation (Le Houérou, 2001).

The chemical analysis of *L. spartum* and *S. tenacissima* showed notable differences in nutritional quality (Table 1). *Stipa tenacissima* showed higher dry matter and fibre contents (crude cellulose, NDF, ADF), indicating lower digestibility, while *L. spartum* contained higher contents of crude protein and mineral matter, suggesting better potential to meet ruminant nutritional needs. Lignin levels were comparable for both species. These findings are in agreement with previous studies, showing that *L. spartum* offers a more balanced protein-to-fibre ratio compared to *S. tenacissima* (Aidi *et al.*, 2012). Despite their high fibre and lignin contents limiting digestibility, both species are valuable for biomass production and persistence in arid degraded rangelands, making them important forage options for grazing systems and restoration efforts under climate variability.

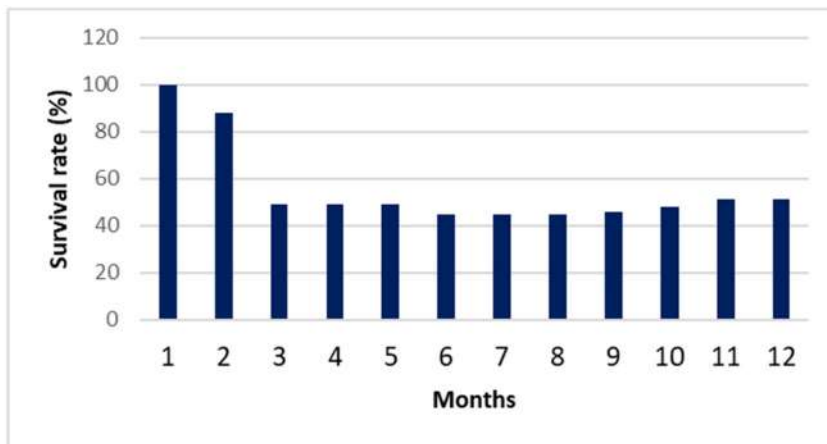


Figure 2. Survival rate of *Stipa tenacissima* 12 months after plantation at the Ain Beni Mathar station.

Table 1. Chemical composition of *S. tenacissima* and *L. spartum* (% of dry matter)

Species	Dry matter	Protein	Crude cellulose	NDF	ADF	ADL	Mineral matter
<i>Lygeum spartum</i>	74.49	5.01	42.16	76.44	46.75	10.1	6.1
<i>Stipa tenacissima</i>	76.33	3.49	45.08	77.81	49.61	9.47	5.09

Conclusion

The results of this study confirm the relevance of *S. tenacissima* and *L. spartum* as valuable native grasses for restoring degraded Mediterranean rangelands. The significant improvement in germination following simple water-soaking treatments highlights their potential for use in cost-effective revegetation programmes. In field conditions, the relatively high drought tolerance of *S. tenacissima* demonstrates its suitability for arid zones. In parallel, forage quality assessments showed that both species can contribute significantly to sustainable pastoral systems, combining ecological restoration with economic utility. These insights reinforce the role of climate-resilient native species in improving vegetation cover, forage supply, and overall rangeland functioning under ongoing climate pressures. Further studies focused on long-term survival success and involving local communities will be essential to successfully broaden the use of these species in arid rangelands restoration efforts.

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The influence of meteorological variability on the peak of herbaceous biomass production in a Sahelian semiarid savanna ecosystem

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Abstract

Savanna ecosystems in the Sahel play a crucial role in ensuring food security. Like other ecosystems, they are heavily impacted by climate change. This study was conducted in Dahra, in the Senegalese Sahel, located in the northern half of the country (15°20'N, 15°29'W). It aimed to assess the influence of certain meteorological variables on the peak of herbaceous biomass production. The biomass was collected on 28 plots of 1 m² each at a 10-day frequency. Meteorological data were recorded by sensors installed on a flux tower. The effects of climatic variables on biomass production were calculated using the XGBoost regression model. Results show that the soil temperature (5 cm depth) is the most impactful variable, followed by the amount of rain and the number of times there is no rain (the number of periods without rain).

Keywords: herbaceous biomass, meteorological variability, Sahel pastures

Introduction

Savanna ecosystems of the Sahel are already heavily affected by climate change. Many subtropical dry regions are expected to become drier, leading to reduced vegetation, which may in turn amplify the initial drying (Zeng & Yoon, 2009). These ecosystems play a fundamental role in food security of the region, and an improved understanding of how they are affected by weather events is thereby critical. Several studies have shown that biomass dynamic follows the amount of rainfall (Diatta, 2021). Many factors other than rain can also play a role in the herbaceous biomass production. Among factors that influence the herbaceous biomass, some authors specify that rain is the main factor of biomass fluctuation in the Northern Sahel (Bakhoum, 2013). To conclude this, the authors did not compare climatic parameters between them, but compared rainfall to the mineral elements contained in the soil. Unlike these studies, this study aims to compare the influence of certain meteorological parameters on the herbaceous biomass production.

Material and methods

This study was conducted in Dahra, northern Senegal. The climate of this area induces 372 mm rainfall, 1.45 m.s⁻¹ wind speed, 8 hours/day insolation, and 30°C annual average temperature (Ndiaye, 2015).

The herbaceous biomass was monitored during the rainy season (July to October) with a 1 m² square quadrat throughout two 1,000 m long perpendicular transects with 14 measurement points each. The

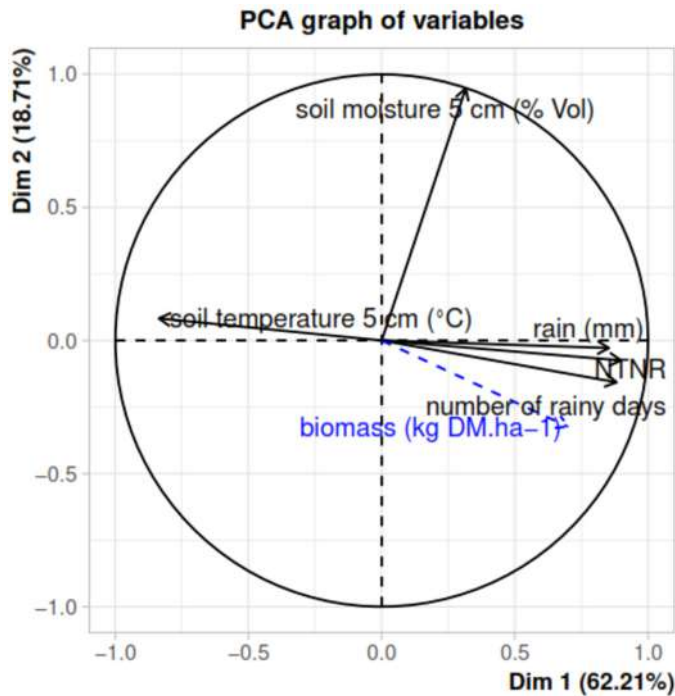


Figure 1. Relationships between variables. The PCA used meteorological variables as explanatory variables and added biomass as a supplementary variable.

biomass was collected at the soil level, using the integral harvesting direct method, starting ten days after the first rain, enough to allow plant germination, with a 10-day interval. Every measurement day, the collection points were moved one metre away from the last collection point to the North. The biomass of the 28 plots was averaged for each measurement date, and the maximum average was used as the biomass amount of the year. From 2008 to 2018, the measured variables were soil moisture, soil temperature at 5 cm depth, rainfall, number of times there was no rain (number of periods without rain) (NTNR), and number of rainy days, using sensors installed on a tower.

A Principal Component Analysis enabled us to identify the relationships between biomass and meteorological variables, and the XGBoost regression model assessed the contribution of each meteorological variable to the peak of herbaceous biomass. The XGBoost model was chosen due to its ability to handle complex nonlinear relationships and interactions between variables without strong assumptions about data distribution. This makes it particularly well-suited for analysing agro-meteorological variables, whose effects can be non-additive and seasonal.

Results and discussion

Figure 1 shows a positive correlation of the rainfall variables (rain, number of rainy days, and NTNR) with the biomass. At the same time, the soil temperature appears negatively correlated with the biomass and rainfall variables. Soil moisture does not show any correlation with biomass and soil temperature. Our results did not confirm the findings of Deng *et al.* (2016) regarding the positive effect of soil moisture on biomass production, but are similar to those of the same authors for the positive effect of rainfall on biomass production.

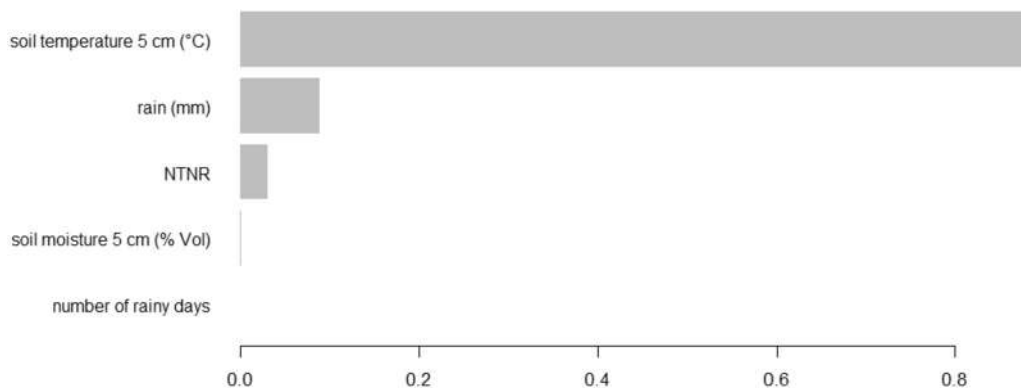


Figure 2. The relative contribution of meteorological variables to herbaceous biomass as determined by the XGBoost model. The horizontal axis represents the relative importance of the variables.

The soil temperature (5 cm) was the most impactful variable, followed by the amount of rain and the NTNR (Figure 2). The soil moisture (5 cm) and the number of rainy days had a very low impact on biomass production. We hypothesize that soil temperature can have both positive and negative effects. It is positive in that it is responsible for litter decomposition, which frees nutrients to the soil, making them available for the plants. However, it is responsible for the soil surface layer drying, making water unavailable for the plants. Since the soil is poor in the Sahelian ecosystems, the main effect of soil temperature may be drying by evaporation.

Conclusions

The temperature of the soil surface layer is a key parameter for understanding the dynamics of Sahelian savanna ecosystems.

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Mountain ranges as refugia for grassland communities under climate change in the Central Apennines

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Abstract

Grassland ecosystems support pastoral systems, deliver key ecosystem services, and are protected within networks such as Natura 2000, yet their biodiversity is increasingly threatened by climate change and human pressures. This study projects shifts in grassland habitats across three Central Apennine ranges using the predictive model MaxEnt, trained on 37 environmental variables and 600 occurrence points. Projections were based on the MPI-ESM1-2-HR climate model under three Shared Socio-economic Pathway scenarios (126, 370, 585) for the years 1995, 2055, and 2085, across four elevation belts: 0–500, 500–1000, 1000–1900, and 1900–2500 m asl. Model performance was high, with an average Area Under Curve of 0.90. By 2085, projected changes relative to 1995 ranged from –5% to +4%, depending on habitat, elevation, and climate scenario. *Sesleria apennina*/*S. juncifolia* and *Nardus stricta* grasslands remained largely stable, whereas *Bromopsis erecta* exhibited contrasting distribution changes along the altitudinal gradient, with expansions mainly between 500 and 1900 m under optimistic scenarios. For this grassland medium and pessimistic scenarios indicated clear elevation-dependent changes in distribution. The results are consistent with previous findings and highlight that the most pronounced distribution changes are expected below 1900 m, where management practices play a critical role in maintain priority conservation habitats.

Keywords: CHELSA, IPBES, predictive vegetation models, protected areas, rangelands

Introduction

Grassland ecosystems provide vital forage resources for pastoral systems and deliver a broad array of other ecosystem services that are essential to human well-being. However, climate change poses one of the greatest challenges, threatening both pastoral systems and the rangeland habitats that have co-evolved with them over millennia (Ickowicz & Moulin, 2023). Since the 1980s, inspired by the Theory of Island Biogeography, large protected area networks have been established worldwide—most notably in Europe through the Natura 2000 network—to safeguard habitats and plant and animal species at risk of extinction. Mountain ranges serve as important refugia for biodiversity (Buschke *et al.*, 2020). Nevertheless, studies have shown that their protective effectiveness is insufficient to halt habitat fragmentation over time, largely driven by anthropogenic pressures (Lawrence *et al.*, 2021). Climate change may further exacerbate this process. Given the marked altitudinal gradient of the Central Apennine chain in the Marche region, the impacts of climate change on grasslands are expected to vary across altitudinal zones. This study uses predictive modelling to project future changes in the distribution of the three most relevant Natura 2000 grassland habitats in mountain ranges under climate change scenarios.

Materials and methods

The study area includes the Natura 2000 sites of three mountain groups in the Marche region (central Italy), namely, from north to south: Monti Catria–Nerone (19 273 ha; 1703 m asl), Monte San Vicino

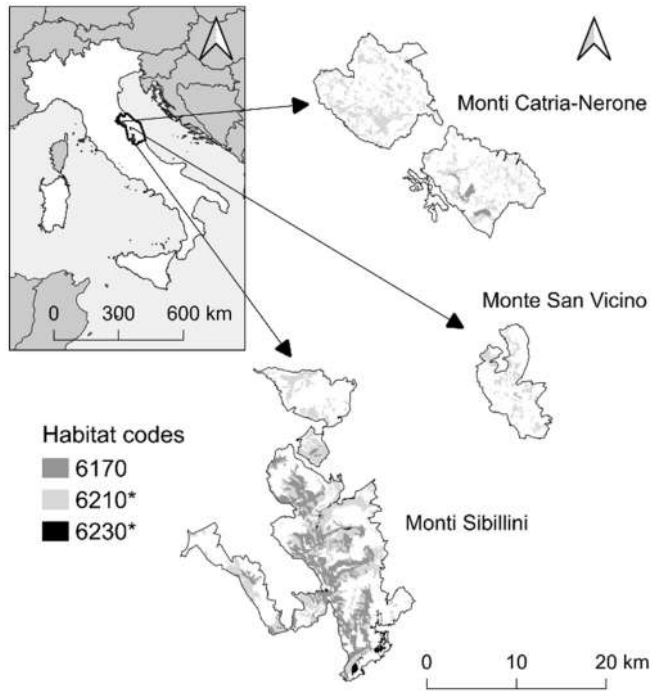


Figure 1. Overview of the three mountain ranges and the Natura 2000 grassland habitats.

(5394 ha; 1483 m asl), and the Monti Sibillini (27 980 ha; 2476 m asl) (Figure 1). The climate along the Apennine chain is classified as “temperate, fully humid, warm summer” (Cfb) according to the Köppen–Geiger climate classification. The geology consists of limestones (Maiolica, massive limestone, and Scaglia Rossa) and Pleistocene-Holocene slope deposits. The MaxEnt predictive model (Phillips *et al.*, 2006) was applied to project changes in Natura 2000 grassland habitats, hereafter referred to as: *Sesleria apennina*/*S. juncifolia* grasslands (alpine and subalpine calcareous grasslands, habitat code 6170), *Bromopsis erecta* grasslands (semi-natural dry grasslands and scrubland *facies* on calcareous substrates, *Festuco-Brometalia*, habitat code 6210*), and *Nardus stricta* grasslands (species-rich *Nardus* grasslands on siliceous substrates in mountain and submountain areas in Continental Europe, habitat code 6230*, where * denotes priority conservation habitats (Biondi *et al.*, 2009). These grasslands are the most widespread and constitute the primary forage resource for local grazing systems (Figure 1).

The MaxEnt model was trained with 37 environmental variables (19 bioclimatic, 3 topographic, geology, and 14 soil parameters). Raster layers were pre-processed in R v.4.1.1 (resampled to 10 m resolution, cropped, aligned) and saved in .asc format for MaxEnt v.3.4.4. A total of 600 randomly displaced occurrence points was used to sample habitats (60 per habitat code) from the Regione Marche, (2013) web repository. The analysis employed the MPI-ESM1-2-HR global circulation model (CHELSA database) under three greenhouse gasses emissions scenarios, i.e. Shared Socio-economic Pathways (SSP): ‘optimistic’ SSP 126, ‘medium’ SSP 370, and ‘pessimistic’ SSP 585. MaxEnt was run with 10-replicated bootstrap, a 25% test split, and 500 iterations, for three time slices (baseline 1995, 2055, 2085). To assess elevational patterns, digital elevation models of the three study areas were processed in QGIS v.3.40.6, divided into four belts (0–500, 500–1000, 1000–1900, 1900–2500 m asl),

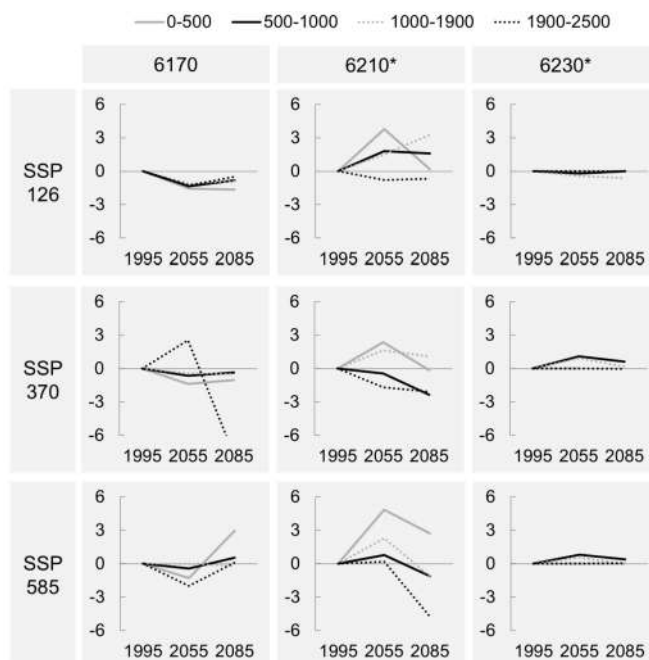


Figure 2. Projected percentage difference in habitat area from the baseline 1995 to 2085, under the three SSP scenarios.

vectorised, and intersected with all model outputs. The resulting areas were calculated and exported to Excel, and model performance was evaluated using the Area Under the Curve (AUC).

Results and discussion

Model performance was consistently high across all analyses, with an average test AUC of 0.91 (± 0.03 SD) for SSP 126, and 0.90 (± 0.03 SD) for both SSP 370 and SSP 585. Among geomorphological variables, slope, elevation and geology were, on average, the most contributing variables, while isothermality and precipitation seasonality were the most contributing bioclimatic predictors. Overall, projected changes by 2085 relative to the 1995 baseline ranged from -5% to $+4\%$ (Figure 2). Under the optimistic scenario, *S. apennina*/*S. juncifolia* grasslands showed slight declines across all elevation belts, whereas *B. erecta* grasslands increased mainly between 500 and 1900 m. In the medium scenario, *S. apennina*/*S. juncifolia* grasslands decreased markedly above 1900 m, while *B. erecta* grasslands increased at 500 and 1900 m but declined at highest elevation. In the pessimistic climate change scenario, *B. erecta* grasslands increased at the lowest elevation but dropped sharply at the highest. *N. stricta* grasslands remained stable across all scenarios. The observed variation range is consistent with Bianchini *et al.* (2025) for Monti Sibillini rangelands. These findings also align with Dibari *et al.* (2015) on the Alps, who reported that *Sesleria* and *Nardus* grasslands remained largely unchanged, whereas the most xeric grasslands—here partially including some *Bromopsis erecta* grasslands—showed the greatest variation. Similarly, Zhang *et al.* (2025) found that the most significant global grassland changes occur below 1200 m, which was also observed here for *B. erecta*, a priority habitat closely linked to management. At low elevations, these grasslands may be constrained by agricultural intensification or land abandonment, while at higher elevations they may face competition from other grassland types.

Conclusion

The changes predicted by MaxEnt were modest but varied by habitat, elevation, and climate scenario. For some vegetation types, mountain areas serve as refugia, highlighting the importance of careful grassland conservation, requiring active management, particularly below the 1900 m elevation belt.

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Soil CO₂ emissions in response to mowing and fertilization in *Bromopsis erecta*-dominated grasslands

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Abstract

Reducing soil greenhouse gas (GHG) emissions can help mitigate rising atmospheric CO₂ levels by limiting the release of carbon from soils. Permanent grasslands, which cover approx. 13% of global agricultural land, can contribute to climate change mitigation through targeted management practices, including mowing regimes and nitrogen (N) application. However, the effects of such management practices on soil respiration (i.e., CO₂ emissions) are not fully understood. This study evaluated the effects of two mowing frequencies (twice per year vs. monthly) and nitrogen fertilisation (0 vs. 90 kg ha⁻¹ year⁻¹) on soil respiration, as well as on soil temperature and moisture—the primary drivers of soil respiration—in a *Bromopsis erecta*-dominated grassland in the Central Apennines. Measurements were conducted biweekly during the growing seasons of 2016, 2018, and 2024 with a portable infrared gas analyser. Mowing frequency had no effect on soil respiration, whereas fertilisation significantly influenced soil respiration depending on the year. This highlights the role of interannual climatic variability. This study suggests that increasing the traditional mowing frequency from twice per year to monthly does not influence soil respiration. However, the effects of mowing and fertilisation on soil carbon storage warrant further investigation.

Keywords: climate change, GHG emissions, grassland management, mowing frequency, nitrogen fertilization, soil respiration

Introduction

Soil respiration is one of the main carbon fluxes between terrestrial ecosystems and the atmosphere, and it is strongly influenced by agricultural practices (Balogh, 2018). Permanent grasslands, particularly those dominated by *Bromopsis erecta*, are high-value ecological habitats and cover extensive areas across Europe (Gigante *et al.*, 2024). Despite their potential role in climate change mitigation, the effects of management practices on soil respiration remain poorly understood and underexplored in these ecosystems. The present study monitored soil respiration, temperature, and moisture over three years (1, 3, and 9 years after the first treatment) to assess the responses to different mowing regimes (twice per year *vs.* monthly) and N fertilisation in a *B. erecta*-dominated grassland in the Central Apennines. We hypothesised that increasing mowing frequency, especially in combination with N fertilisation, would significantly influence the primary drivers of soil respiration, including soil temperature and moisture.

Materials and methods

The study was carried out in a semi-natural permanent grassland dominated by *B. erecta* near Monte San Vicino (900 m asl, Central Apennines, Italy), within the Natura 2000 site SPA IT5330025. The area represents a priority calcareous grassland habitat (6210*, EU Habitats Directive 92/43/EEC). The

site has a northeast-facing slope (approx. 10%) and a temperate-oceanic climate (mean annual temperature: 13°C; precipitation: 865 mm). At the beginning of the experiment, the soil had a clay loam texture, with a pH of 6.79, total C content of 5.92%, and total N content of 0.53%. A randomised complete block design with three replicates per treatment was used (plots 2 × 2 m). Treatments included: (i) customary mowing, twice annually (June and October); and (ii) intensive mowing, monthly from April to October, resulting in 4 to 6 cuts per year when biomass exceeded 0.5 t ha⁻¹. Mowing was performed at 5 cm height, with biomass removed immediately. Fertilisation consisted of an annual application of 90 kg N ha⁻¹ as ammonium nitrate in April. Plots were managed according to these treatments continuously, even when measurements were not being conducted, and were fenced to prevent disturbance by animals. Soil respiration was measured in situ with an infrared gas analyser (EGM-4) using 10 cm PVC collars inserted about 9 cm into the soil. Soil temperature (10 cm depth) was measured using a thermometer probe, and soil moisture (0–10 cm) was determined by collecting samples and drying them at 105°C until constant weight. Measurements were taken every 2–3 weeks. Vegetation regrowth in collars was manually removed. Mixed-effects models assessed treatment effects; model assumptions were verified using Q-Q plots and Levene's test ($p > 0.05$).

Results and discussion

A significant interaction between monitoring year and fertilisation was observed (Table 1). Soil CO₂ efflux was highest in 2016 (0.57 g CO₂ m⁻² h⁻¹), lowest in 2018 (0.35 g CO₂ m⁻² h⁻¹), and intermediate in 2024 (0.48 g CO₂ m⁻² h⁻¹) (Figure 1). Mean soil temperature differed across years, being higher in 2024 (18.3°C) than in 2018 and 2016 (16.8°C), whereas soil water content showed an opposite trend, with higher values in 2016 (26%) and lower in 2018 (22%) (Figure 1).

Mowing intensity, as well as its interactions with fertilization and year, had no significant effects on soil CO₂ efflux, soil temperature, or soil water content (Table 1). In line with previous studies, mowing intensity had no detectable effect on soil CO₂ emissions (Francioni *et al.*, 2020; D'Ottavio *et al.*, 2023). In contrast, the effect of fertilization on soil CO₂ efflux varied across years, indicating a significant interaction with year and the influence of interannual climatic variability (Zhao *et al.*, 2025). Despite the observed rise in soil temperature over time—aligned with climate change projections—this was not accompanied by increased CO₂ emissions, indicating that temperature alone does not limit soil CO₂ emissions (Francioni *et al.*, 2020; D'Ottavio *et al.*, 2023). Variations in soil moisture, likely influenced by differences in annual rainfall, appeared to affect both the autotrophic and heterotrophic soil respiration (Balogh *et al.*, 2016).

Table 1. *F*-values and associated *p*-values from the analysis of main effects and interactions of monitoring year, fertilization (Fert), and management (Man)

Factor	Soil CO ₂		Soil temperature		Soil water content	
	<i>F</i>	<i>p</i>	<i>F</i>	<i>p</i>	<i>F</i>	<i>p</i>
Year	39.234	< .001	36.871	< .001	65.907	< .001
Year × Fert	6.813	0.007	1.791	0.214	0.489	0.522
Year × Man	1.342	0.289	0.843	0.408	0.895	0.381
Year × Fert × Man	0.699	0.512	0.32	0.637	0.175	0.711
Fert	0.183	0.68	0.004	0.952	1.133	0.318
Man	0.011	0.92	0.323	0.586	1.833	0.213
Fert × Man	0.003	0.96	0.014	0.909	0.002	0.963

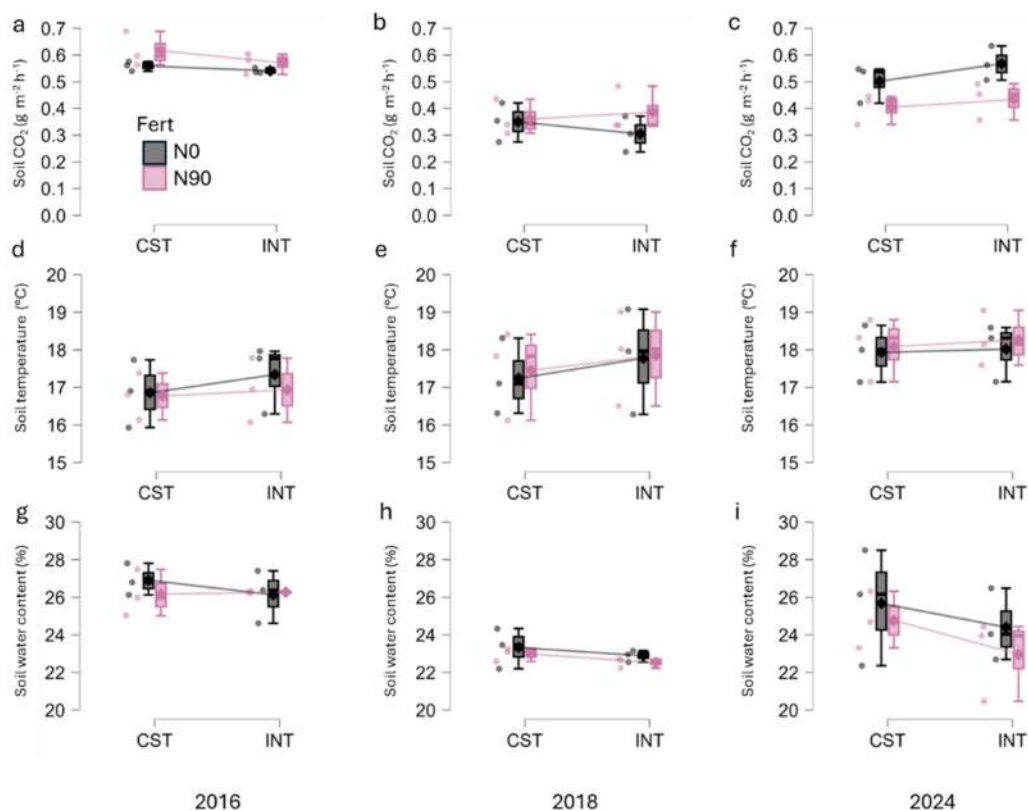


Figure 1. Mean soil CO₂ emissions, temperature, and water content during the monitoring years (from March to October). CST, customary mowing; INT, monthly mowing.

Conclusion

N fertilization emerged as a key driver of soil respiration, although its effect varied between years. In contrast, increasing mowing frequency had negligible effects on soil CO₂, temperature, and moisture. These results highlight the importance of management practices that conserve biodiversity-rich grasslands while supporting climate change mitigation. In this context, moderately increasing the customary mowing frequency (e.g., twice a year) could be a viable strategy for maintaining secondary grasslands where farmers require more intensive use, achieving a win-win situation between conservation and production objectives.

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Drought and soil type affect the emergence of meadow species and their initial emergence timing

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Abstract

Rising global temperatures are increasing the risk of drought, which could potentially delay seedling emergence and hinder vegetation renewal in lowland hay meadows. Also, soil properties influence the drought resistance of plants. This study investigated the effect of reduced soil moisture on the initial emergence time and seedling numbers of 23 meadow species across various soil types. A 30-day drought simulation experiment was conducted in pots using a split-plot design: the main plot included two moisture levels (control: 60–80% and drought: 20–40% water capacity), and the subplot included three soil types: universal (pH 6.0), universal + sand (pH 6.3), and acidic peat soil (pH 4.0). Mixed-model ANOVA analysis revealed that seedling emergence was significantly reduced and delayed under drought conditions and in acidic or sandy soils compared to the universal soil, which was at the optimal moisture level. Based on the predicted values at the species level, *Centaurea jacea*, *Arrhenatherum elatius*, *Dianthus deltoides*, *Succisa pratensis* and *Allium angulosum* showed stable and high emergence at low soil moisture levels, making them suitable candidates for direct sowing. These traits are important for successful meadow restoration under climate change conditions, in environments where natural regeneration is limited.

Keywords: drought, lowland hay meadow, plant emergence, soil type

Introduction

Semi-natural grasslands in Europe are protected within the Natura 2000 network, which supports ecosystem functions such as regulating groundwater supply (Ritzema *et al.*, 2016). Lowland hay meadows (habitat code: 6510) are classified as a semi-natural grassland with high floristic diversity and various soil types (Roleček *et al.*, 2014). However, loss of biodiversity and decline in productivity under drought conditions have threatened plant communities in this habitat, causing changes in vegetation through delayed emergence time and colonization by invasive species (Gigante *et al.*, 2024). To understand how drought may adversely affect plant emergence, this study aimed to evaluate the impact of reduced soil moisture on seedling emergence and initial emergence time across different soil types. Finally, based on these results, this study can recommend species that are less sensitive to drought conditions at the emergence stage.

Materials and methods

The study covered 23 meadow species (Table 1) in a split-plot design and three replications, with water capacity level as the main plot (60–80% of water capacity as control, and 20–40% of water capacity as drought condition) and soil type as subplot (universal soil, pH 6.0; universal+sand = 70% universal soil+30% sand, pH 6.3; acidic peat soil, pH 4.0). The universal soil consists primarily of deacidified peat with the addition of drainage sand, ground and composted pine bark, and fertilizer. The volume of the pot was 700 cm³, and the total number of pots was 138. Control pots were watered once every two days, while simulated drought pots were watered once every four days. During the 30-day

Table 1. The twenty-three meadow species included in this study

<i>Achillea millefolium</i>	<i>Centaurea jacea</i>	<i>Kadenia dubia</i>	<i>Sanguisorba officinalis</i>
<i>Allium angulosum</i>	<i>Dianthus deltoides</i>	<i>Knautia arvensis</i>	<i>Silene flos-cuculi</i>
<i>Alopecurus pratensis</i>	<i>Eryngium planum</i>	<i>Lathyrus pratensis</i>	<i>Succisa pratensis</i>
<i>Arrhenatherum elatius</i>	<i>Galium boreale</i>	<i>Leontodon autumnalis</i>	<i>Tragopogon pratensis</i>
<i>Briza media</i>	<i>Geranium pratense</i>	<i>Leucanthemum vulgare</i>	<i>Vicia angustifolia</i>
<i>Campanula patula</i>	<i>Hypericum perforatum</i>	<i>Plantago lanceolata</i>	

Table 2. Two-way mixed-model analysis of variance (ANOVA) on the number of emerging seedlings and the initial time of emergence (t_0) of 23 meadow species.

Source of variation	Number of seedling emergence			t_0 (days)		
	df	F-value	p-value	df	F-value	p-value
Water capacity level (A)	1	55.62	***	1	43.89	***
Soil type (B)	2	8.76	***	2	15.99	***
A $\times$ B	2	1.1	ns	2	3.61	*

t_0 , the initial time of emergence (days); ns, non-significant; * significant at $p < 0.05$; *** significant at $p < 0.001$.

experimental period, the initial emergence time (t_0 ; i.e., the first time of emergence) and the number of emerging seedlings were determined. Analysis of variance (ANOVA) was then performed using a mixed model with species and replication as random effects; means of significant treatment effects were compared by the Tukey post-hoc test. To obtain the recommended species for sowing, we extracted the BLUP (Best Linear Unbiased Predictor) value from the ANOVA model. All statistical analyses were conducted by R Studio 4.4.1.

Results and discussion

The interaction between water capacity level and soil type factors occurred in t_0 (Table 2). On the contrary, all factors independently affected the numbers of seedlings emerging.

Figure 1a shows that, regardless of the soil type, seedling emergence rates were lower in drought conditions than under control conditions, ranging on average from 21.4% (acidic soil) to 26.7% (universal soil). Soil acidic conditions resulted in the lowest number of seedlings emerging (21.4%). Souza *et al.* (2022) explained that drought conditions lead to an increase in osmotic stress, which forces seeds to utilize nutrients in the face of an insufficient water supply, potentially reducing their viability. The interaction between drought and acidic and universal + sand, soils significantly delayed t_0 (by 2.6 and 4.8 days, respectively), compared to drought and universal soil (1.3 days) (Figure 1b). From an ecological perspective, delaying the initial emergence moment may gradually alter the dominance of native species, particularly in the case of perennial plants in grasslands, resulting in reduced regeneration capacity of vegetation (Orbán *et al.*, 2023). Figures 2a and 2b illustrate the analysis of BLUP values for seedling emergence percentage and t_0 , showing that species such as *C. jacea* (63.00%, 8 days), *A. elatius* (59.82%, 6 days), *D. deltoides* (53.50%, 6 days), *S. pratensis* (53.24%, 6 days) and *A. angulosum* (47.18%, 10 days) exhibited greater stability across three different soil types.

Conclusions

Drought conditions have a serious impact on the rate and timing of seedling emergence. Growing the tested species in universal soil with a full water supply results in higher average seedling emergence and accelerates their emergence compared to development in other soil types, at both levels of water

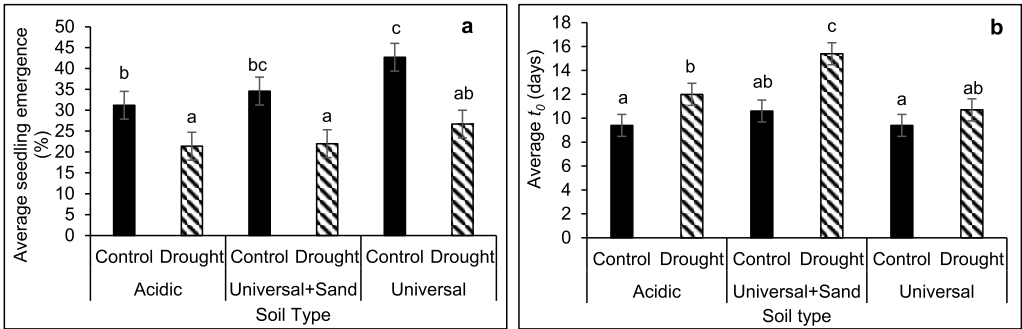


Figure 1. Effect of drought and soil types on the emergence of 23 meadow species. (a) Average seedling emergence (%), and (b) average initial time (t_0). The same letter indicates a non-significant difference at $p < 0.05$.

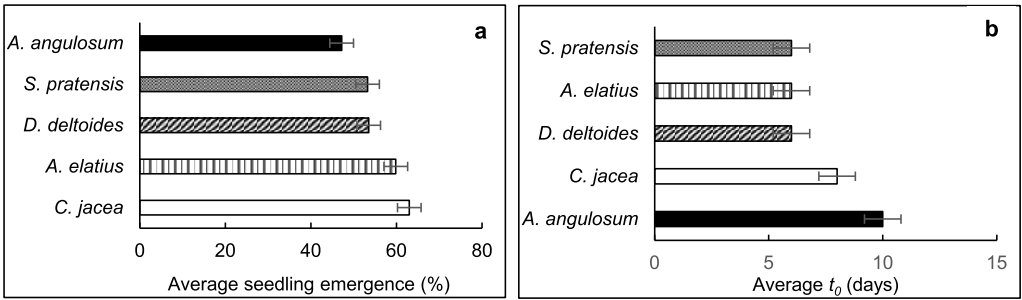


Figure 2. (a) Average seedling emergence (%), and (b) t_0 , average initial time of seedling emergence (days) of the best five species based on BLUP analysis.

capacity. Moreover, *C. jacea*, *A. elatius*, *D. deltoides*, *S. pratensis*, and *A. angulosum* can be recommended as less sensitive to drought conditions during the emergence stage due to their higher proportion of emerging seedlings and the fastest emergence rates.

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Biochar application and pasture GHG emissions: links to soil temperature and moisture at peak growth

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Abstract

Biochar has the potential to mitigate climate change by reducing greenhouse gas (GHG) emissions. This study examines the effects of biochar on GHG emissions, carbon dioxide (CO₂), methane (CH₄), and nitrous oxide (N₂O) in hard-setting pasture soil under current and reduced rainfall conditions. A woodchip biochar was incorporated in the topsoil (0–20 cm) in a sown biodiverse pasture in Portugal's Centre region, at 30 g kg⁻¹. The two-factor experiment (biochar, rainfall) had four treatments: (i) C100: no biochar, 100% rain; (ii) C50: no biochar, 50% rain; (iii) B100: biochar, 100% rain; (iv) B50: biochar, 50% rain. During the peak growth period in April 2024, GHG emissions were monitored weekly, along with soil temperature, moisture, and inorganic nitrogen (NH₄⁺ and NO₃⁻). Results showed that biochar, particularly under 50% rainfall conditions, significantly reduced CO₂ emissions. Changes in soil moisture and temperature were linked to these effects, suggesting that biochar alters microclimate conditions and affects nitrogen dynamics, namely N₂O emissions. These findings support the use of biochar as a promising strategy to reduce GHG emissions and enhance resilience in rainfed Mediterranean pasture systems facing future climate challenges.

Keywords: climate change mitigation, hard-setting soil, rainfall

Introduction

GHG emissions, including CO₂, CH₄, and N₂O, have increased significantly in recent decades due to anthropogenic activities, particularly in agriculture (Decock *et al.*, 2015). Biochar was investigated as a potential solution for reducing GHG emissions and moderating soil temperature fluctuations (Xiong *et al.*, 2020). This study aimed to assess the effects of biochar and reduced rainfall on cumulative GHG emissions and soil inorganic nitrogen (NH₄⁺ and NO₃⁻), as well as to explore how these responses are related to soil temperature and moisture conditions during the growth peak in a sown biodiverse pasture with hard-setting soil. Soils that dry into a dense, cement-like mass, restricting water, air, roots and seedlings, are considered hard-setting (Daniells, 2012).

Materials and methods

The study was conducted at Quinta da França, near Covilhã, Portugal, on a sandy loam Dystric Cambisol. The area has a Mediterranean climate, with 858 mm of annual rainfall, 90% of which falls between October and May. Woodchip biochar (produced at 620°C from pine woodchips) was incorporated to 20 cm depth at a concentration of 30 g kg⁻¹. Twenty plots (3 m×3 m), half with rainout

Table 1. Average ($n = 5$ replicates) of medians for daily maximum temperatures ($^{\circ}\text{C}$) in April.

Treatment	Soil temp	Surface temp	Air temp
C100	16.9 a,b	23.1 a,b	27.5 a
C50	17.4 b	22.5 b,d	26.2 a,b
B100	15.5 a,c	20.0 c,d	24.4 b,c
B50	15.7 a,c	19.4 c	24.1 b,c

In each column, treatments that do not share a letter are significantly different ($p < 0.05$) based on Tukey's HSD test.

shelters, were arranged in a random block design with five replicates for each of the four treatments: (i) C100: no biochar, 100% rainfall; (ii) C50: no biochar, 50% rainfall; (iii) B100: biochar, 100% rainfall; (iv) B50: biochar, 50% rainfall. All treatments were on a newly re-sown (September 2022) biodiverse pasture, and the sward age was the same across control and biochar plots. The microclimate temperature (T) was recorded at +12 cm, the soil surface T at +2 cm, and the soil T at -6 cm, and the soil moisture content (SMC) at -6 cm via a TOMST Extreme sensor in each plot, logging every 15 min. GHG emissions were monitored weekly in April 2024, following the method described by Fangueiro *et al.* (2017). One-way ANOVA was performed using Python 3.10.

Results and discussion

Correlation analysis showed that CO_2 emissions were positively related to maximum surface T ($r = 0.60$), air T ($r = 0.49$), and soil T ($r = 0.50$). CH_4 emissions correlated slightly with minimum T and SMC. N_2O emissions also showed a negative relationship with SMC and a strong inverse correlation with minimum T . Only CO_2 emissions correlated with soil T at 6 cm depth; CH_4 and N_2O were associated with surface and air T . Considering the hard-setting soil of the field site, with roots present only in the first 5 cm, this suggests that GHG emissions may occur from just a few cm below the soil surface.

Biochar significantly (ANOVA, $p < 0.05$) reduced the maximum soil T , soil surface T , and 12 cm T in April 2024 (Table 1), with no effect from rainout shelters or interaction of biochar and rainout shelters. Since CO_2 emissions showed a strong positive correlation with soil T , this suggests that biochar may help reduce CO_2 losses by keeping the soil cooler. Biochar may cool soil by increasing evaporative cooling and by lowering bulk density and thermal conductivity, which slows heat transfer and stabilises temperature (Xiong *et al.*, 2020).

CO_2 emissions were highest in control plots (C100), while biochar significantly (ANOVA, $p < 0.05$) reduced (34%) emissions, especially under reduced rainfall conditions (B50) (Figure 1). CH_4 emissions trended to be reduced by all treatments compared to control plots (C100), but not significantly. N_2O emissions were strongly reduced under 50% rainfall (Figure 1) and moderately reduced by biochar under 100% rainfall. These findings were consistent with the study by Abagandura *et al.* (2019), which found that applying 1% biochar to sandy loam soils reduced cumulative N_2O and CO_2 , but had a limited effect on CH_4 emissions. Soil ammonium (NH_4^+) and nitrate (NO_3^-) levels were not significantly different among treatments. NO_3^- levels have a slight positive correlation with N_2O emissions, and biochar reduced N_2O losses even under elevated NO_3^- conditions. Overall, the results highlight biochar's potential to mitigate GHG emissions, particularly under future climate scenarios with reduced precipitation.

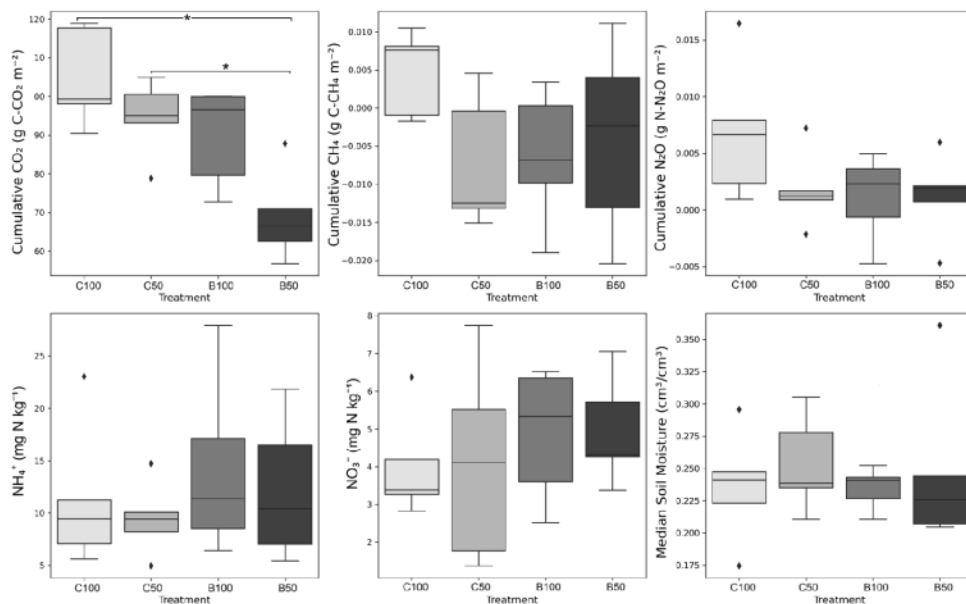


Figure 1. Cumulative greenhouse gas (GHG) emissions, ammonium (NH_4^+) and nitrate (NO_3^-), and median soil moisture levels in April 2024. * Significant differences (ANOVA, $p < 0.05$). Black dots are outliers.

Conclusion

This study shows that biochar can effectively reduce GHG emissions in pasture soils even under reduced rainfall conditions. These results support the use of biochar as a climate-smart amendment in rainfed Mediterranean pasture systems, particularly in light of expected climate change scenarios.

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Carbon farming in grassland-based system: how to get the most from farm diagnosis?

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Abstract

For over 20 years, Idele has supported farmers in low-carbon initiatives by developing CAP'2ER[®], a multi-stakeholder tool for assessing the environmental impacts of livestock farming. To date, over 45,000 farm diagnoses have been carried out in response to climate-related challenges. This is particularly relevant in France, where 38.5% of farms involve significant herbivore production. This article shows grassland-based system (GS) CAP'2ER[®] results while highlighting two key insights: i) How experienced GS farmers use it on-farm, ii) what was missing from the farmers' point of view, and their requests for refinements. These insights stem from contrasted territories: Pathways EU project (GS₁) implemented in western France, main dairy region, a lowland area with grassland- maize silage leys, and Decarbon'Alpes project (GS₂) carried out in alpine mountain region, a pastoral grassland area.

Keywords: carbon farming, C footprint, C sequestration, GHG emissions, low input farming

Introduction

With limited earth resources and increasing world population, agriculture is challenged to provide healthy food while preserving other ecosystem services. In France, 38.5% of farms involve significant herbivore production, and the fodder areas (FA) is 47% of UAA. Approximately 55% of agricultural greenhouse gases (GHG) are emitted from ruminant farms but those animals also help to manage 11.5 million hectares of grassland with high soil organic carbon stock. Under business-as-usual management, the organic carbon stocks (SOC) in French permanent grassland should still increase over the next decades (Pellerin *et al.*, 2019). CAP'2ER[®], an Iso certified LCA environmental assessment tool developed by Idele, is widely used in France and in several EU countries (over 45 000 applications). Nevertheless, grassland-based farms (GB), usually encouraged to protect watershed basin, could be challenged by this new approach on climate mitigation strategies when focussing on impact per product (C footprint). That's why two GS farmer hubs wanted to trial it: what is the best farming system development option for their region, how can they apply the results of the farm assessment and what should be further developed?

Materials and methods

Farms were assessed by CAP'2ER[®] level 2 in two region: 10 dairy cattle farms GS₁ located west in Normandie/Bretagne/Pays de la Loire, a lowland oceanic zone (<500 m altitude) where maize silage with grassland leys are dominant in the fodder area (Pathways Eu study); 41 ruminant farms located from north to south French Alps (500–2500 m high), where permanent grassland is dominant fodder use (GS₂- Decarbon'Alpes study). Both regions are rainy with gradient from 600 to 1400 mm/year (1-E/W, 2-N/S).

Table 1. Environmental assessment (CAP'2ER[®] 2023) of grassland systems versus Baseline.

Assessment over 1 year	unit	GS ₁	West ¹ w-Base	GS ₂	Fr. GL ² lowland
Data collection	year	2022–2023	2013–2021	2022–2024	2013–2021
Grassland in UAA	%	94	39	96	54
Farm Mineral Fertilizer	kg. ha ⁻¹	0	53	12	42
Direct energy use by fuel	l ha ⁻¹	49	200	112	121
Farm nitrogen balance on UAA	kg N. ha ⁻¹	29	117	57	85
Nourishing human on UAA	Nb person. ha ⁻¹	13	26	16	15
Farm Emissions GHG	Mg eq CO ₂ ha ⁻¹	3.8	8.8	3.7	5.81
Farm Carbon storage	Mg eq CO ₂ ha ⁻¹	2.17	0.7	1.9	1.06
Dairy Milk GHG emitted	kg eq CO ₂ . l ⁻¹	0.96	0.98	1.32	1.02
Net C milk footprint (GHG-storage)	kg eq CO ₂ . l ⁻¹	0.47	0.88	0.64	0.78
Biodiversity	ha eq. ha ⁻¹	2.68	1.28	2.5	1.4

¹ Dairy western baseline where maize silage is over 30% of fodder area.

² 145 dairy cattle grassland system located in a blend of French lowland region.

Results

The results (Table 1) focus on dairy cattle as they exist in both regions (all GS₁, $n = 13$ for GS₂). Farmers were interested in comparing their hub average result to local dominant business model for GS₁, and to national French average lowland grassland milk for GS₂. At a territory scale, both GS₁ and ₂ have lower nitrogen inputs per hectare than the baseline, the nitrogen balance excess as well as the GHG emissions are lower. The Biodiversity on UAA is higher than baseline.

In the lowland region, GS₁ (4481 l milk/cow, 23% replacement, 1.1 LSU ha⁻¹ FA) compared with regional trend (7923 l milk/cow, 32% replacement, 1.8 LSU ha⁻¹ FA), it was expected that C footprint would be higher as methane is the first source of GHG emission in French milk. But GS₁ farmers implement various techniques to improve grass management as a measure of standing grass stock, ball grazing, winter grazing; and several farms are spring calving, with mixed breed. It increases the grazing days from a baseline in the W-Base of 144 days to 283. Rotational grazing on mixed swards requires no mineral fertilizer while still meeting the needs for good quality grass in the spring, at the peak of the cow's requirement. This coupled with moderate milk yield means that requirements for concentrates is very low for the cow (18 g/l milk GS₁, 171 g/l W-base) and the heifer 82 kg LSU⁻¹ (versus 677 kg W-base). The longer grazing period, which reduces the period of housing and the quantity of crops to be harvested, reduces the direct fuel consumption by 28% per litre of milk sold. Thanks to all this, the total GS₁ GHG emitted per product is similar to W-base. Moreover, GS₁ keep more carbon in soil with 70% permanent grassland in UAA (i.e., W-base 12%) and maintains around 120 metres linear/ha (like W-base) of hedges for grazing on the land used by the dairy cows, which is 40% higher hedge ratio in the landscape for GS₁ at the level of the whole Farm W base. Indeed, the Soil data national survey indicates a 49 % higher soil C stock for permanent grassland than arable land (0–30 cm depth, west Fr. measurements). Farmers promote that changing from W-Base to GS₁ farming system would improve overall carbon footprint by 46% according to CAP'2ER[®] (Net milk C footprint). Unfortunately, the statistical trend is in the opposite direction, as the grassland area has decreased by between 2 and 6% in those regions between 2015 and 2023. At last, if GS₁ increases the biodiversity and helps to preserve the ecosystem with no pesticide use on the one hand, the number of human feeds per hectare of farm agriculture area is much lower than W-base on the other hand. GS₁

farmers pointed out that they rely on fewer external areas, which also means less risk of disturbance for ecosystems elsewhere in the world.

In Alpine region, GS₂ includes 25% pastoral area in total land use compared with lowland grassland milk production with negligible pastoralism. The milk production is moderate (5 563 litres of corrected milk/Cow, 6 570 Fr-GL Base), and the fields are smaller and steeper, with typically more dispersed plot structure than Fr-GL Base. Thus, the energy consumption is higher per unit of milk produced despite a significant proportion of grassland in UAA. The first source of GHG (CH₄) is also more important due to lower herd productivity; so even if GHG emission due to nitrogen fertilisation (spreading manure and mineral fertilisers processing) are lower than baseline, the GS₂ overall GHG emissions are 29% higher than the mean lowland French GL milk (per l sold). However, after deducting carbon storage, the net C-footprint of mountain milk is 18% lower.

Discussion and conclusion

Both farmer groups were very interested in the results as CAP'2ER[®] delivers farm and product impacts, not only on GHG but also on water, soil, air quality as well as services provided (food, energy, fertility, biodiversity) so that they can answer the questions asked by visitors to the farm. GS₁ decided to organise special farm open-day tours to promote GS techniques to the younger generation from areas where maize silage predominates. Two hundred and eighty 15–22-year-old students participated. CAP'2ER[®] results were reinforced by information presented on the economic performance and the labour requirements. Students evaluate GS as a credible farm option. 72% found GS to be reliable for sustainable agriculture under climate change, with farmers being more autonomous from firm services (80%) and having less work (89%).

GB farmers underlined that a fast communication limited to global C footprint is poor. It is important to raise awareness on the sources of emissions: CH₄ emitted and atmospheric C captured in soil are natural short-term processes whereas CO₂ mainly comes from fuel, long-term C released. They also suggest to i) emphasise the use of outside surfaces by farms, ii) include feed-food competition calculation, iii) better integrate pastoral area in results. As C sequestration, especially in pastoral systems offsets a large proportion of the emissions in the final GS C footprint, the farmers have requested that there is better integration of soil/climate/grassland management in the tool. GS₂ measured soil current C stocks to implement CarSolEl tool (Lafleur *et al.*, 2022) on a set of typical FA land use. It shows that C sequestration varies widely: two pastoral zones with actual C stock 103 and 70 Mg C ha⁻¹, both managed with low grazing pressure (10–50 LSU grazing days/ha, no mineral N input, nor manure spreading, sequester –610 (soil losing organic C) to +510 kg C ha⁻¹ (soil C increase) per year. Difference in altitude and soil texture are important drivers, as well as management. All those considerations are valuable to further development of GS assessment.

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Characterisation of Welsh Mountain sheep wool as a tool to manage water run-off from grasslands

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Abstract

Sheep wool is a versatile, renewable fibre that has potential for use in agricultural settings as a fertiliser, mulch or as a geotextile. Such applications support the development of added-value products for coarse wool, such as Welsh Mountain wool, that can support sustainable farming practices and may address the effects of climate change. Understanding the appropriate form of wool for development of products for use in an agricultural setting is a key part of this process. Here, a model to study the properties of raw, scoured and felted Welsh Mountain wool as a tool to manage surface water run-off in the event of heavy rainfall is described. Imported coconut coir is used to manage water run-off from construction sites, therefore, was used as a comparison material. Coir or wool was placed in a Tullgren funnel and wetted with distilled water or 10% w/w soil in water, and the filtrate collected. Results showed sheep wool has potential to be used to reduce water run-off from grasslands as indicated by high water holding capacity (WHC) and slower outflow rates. However, wool needed to be processed to reduce the risk of nutrient loading in run-off, as may occur when using raw wool.

Keywords: climate change, sheep wool, water run-off

Introduction

Sheep wool is a versatile, natural, renewable porous fibre that displays key functional qualities including hygroscopicity and sorbency. For coarse sheep wool, i.e., wool with a fibre diameter $>30\ \mu\text{m}$ that is not suited to the apparel industry, such as is produced by Welsh Mountain sheep, developing novel applications will valorise these low-grade wools. This will support the sheep farming industry by addressing economic challenges facing farmers alongside mitigating the effects of climate change. Climate change has increased the occurrence of extreme weather events such as heavy rainfall (Clarke *et al.*, 2022). Coconut coir is used as a barrier material to manage surface water run-off from construction sites; however, it is an imported fibre. There is growing evidence that sheep wool can also act as a filter material, removing organic contaminants from water (Periolatto and Gozzelino, 2015; Pang *et al.*, 2022), and may offer an alternative to coir barriers to manage water run-off. The present study used a bench-top model to study WHC and filter properties of various forms of Welsh Mountain wool, compared to coconut coir, in the context of its potential utilisation as a tool for water run-off from grasslands. The work sought to identify the form of wool suited for development of a tool for water run-off management.

Materials and methods

The study assessed three forms of Welsh Mountain wool: raw, scoured or felted wool (50 mm thickness); and coconut coir. The oven dry matter (DM) and WHC of each fibre was assessed. The WHC was calculated from the difference between the dry weight of a known mass of fibre, compared to the saturated weight of fibre after immersion in water for 2 hours. For the filtration experiment, trial

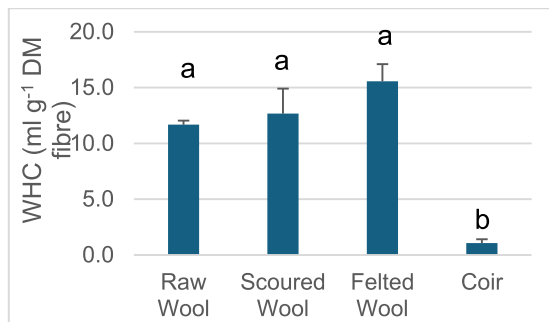


Figure 1. The water holding capacity (WHC) of the different fibre types after immersion in water for two hours. Different superscripts indicate a significant difference ($P < 0.001$).

fibres (18.5 ± 1 g DM/funnel, $n = 3$ replicates per treatment) were placed in Tullgren funnels, with a collection sample placed below each funnel. Separate funnels were set up to test either deionised water or a 10% soil-water solution each ($n = 24$, 4 fibre treatments $\times$ 2 water treatments $\times$ 3 reps). Flow rate was assessed by applying water using a pump system at a rate of 697.0 ± 40 ml min⁻¹, to 11.5 g DM $\pm$ 1 g filter material, and measuring the outflow in the collection vessel.

To assess the chemical content of filtrate, samples were wetted from above with 500 ml of either deionised water or 10% (w/w) soil in deionised water. The pH, sulphur (S), phosphorus (P) and potassium (K) content of the filtrate were assessed. Data were analysed using ANOVA with Bonferroni post hoc in Genstat (Payne *et al.*, 2011). Comparison of filtrate nutrient concentration to initial 10% soil-water test solution was performed in SPSS (v 29, IBM SPSS Statistics) using ANOVA and Dunnett's post hoc. Significant differences were attributed where $P < 0.05$.

Results and discussion

The DM of each fibre was 849, 902, 888 and 896 g kg⁻¹ for raw wool, scoured wool, felted wool and coconut coir, respectively. The WHC of coir was significantly lower than any of the wool samples (Figure 1, $P < 0.001$). Wool processing did not alter WHC (Figure 1).

The outflow rate for each fibre is shown in Figure 2. Felted wool had a slower outflow rate than coir ($P < 0.05$), with all other treatments intermediate. The pH and chemical composition of filtrate from the wool and coir samples are shown in Table 1.

Effect of fibres on the filtrate where deionised water was the test solution (Table 1)

The filtrate from raw wool was more alkaline than that the filtrate from other fibre treatments when deionised water was the test solution ($P < 0.05$). Water filtrate from coir was more acidic than the filtrate from felted wool ($P < 0.05$). Raw wool filtrate was associated with highest concentration of sulphur, compared to the filtrate from the other fibres ($P < 0.05$). Concentrations of P and K were higher in both coir and raw wool filtrates, compared to the filtrates from scoured or felted wool ($P < 0.05$). Raw wool filtrate had highest K concentration of all filtrates ($P < 0.05$). These results indicate that raw wool leaches S, P and particularly K into the test solution. Suint in raw wool is rich in K salts (Rippon, 2013), and a likely source of the elevated K concentration observed in this study.

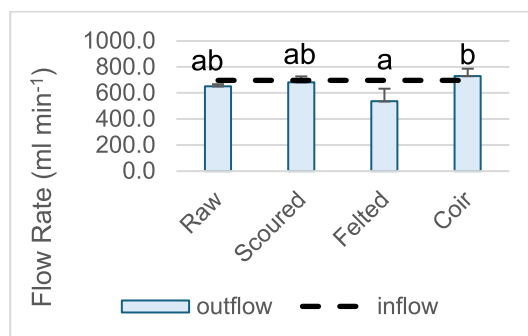


Figure 2. Outflow rates of water for different fibres. Superscript indicates differences between fibre types ($P < 0.05$). Inflow rate is indicated by a dashed line.

Table 1. Effect of filter material on the physical and chemical composition of filtrate. The pH and chemical composition of the solution applied to the filter set up is also shown [#].

	Test solution	Raw wool	Scoured wool	Felted wool	Coir	Filtrate effect	
						SED	<i>P</i>
Test solution: water							
pH	6.31	8.58 ^c	6.62 ^{ab}	6.88 ^b	6.01 ^a	0.203	< 0.001
Sulphur (mg l ⁻¹)	0.04	1.12 ^a	0.11 ^b	0.12 ^b	0.26 ^b	0.150	< 0.001
Phosphorus (mg l ⁻¹)	0.02	0.46 ^a	0.02 ^b	0.02 ^b	0.52 ^a	0.075	< 0.001
Potassium (mg l ⁻¹)*		124.83 ^c	0.60 ^a	0.81 ^a	8.94 ^b	0.15	< 0.001
Test solution: 10% (w/w) soil in water							
pH	5.82	6.36 ^{a,**}	5.98 ^{ab}	5.91 ^b	5.72 ^b	0.113	0.003
Sulphur (mg l ⁻¹)	0.81	1.93 ^{a,**}	0.61 ^b	0.48 ^b	0.67 ^b	0.217	<0.001
Phosphorus (mg l ⁻¹)	0.47	1.26 ^{a,**}	0.69 ^b	0.84 ^{ab}	0.82 ^{ab}	0.145	0.020
Potassium (mg l ⁻¹)*	9.15	162.98 ^{a,**}	9.73 ^b	9.09 ^b	10.84 ^b	0.141	<0.001

Different superscripts between columns indicate differences between the filtrate from different fibres, $P < 0.05$. *Geometric mean is shown as log10 transformation was used for statistical analysis.

** For soil-water test solution only, filtrate pH or chemical composition was significantly different from the initial solution.

The deionised water filtrate from scoured and felted wool had the lowest concentrations of S, P and K, compared to the filtrates from other fibres. Findings indicate the use of coir as buffer material may also contribute significant quantities of P and K to the environment. Nutrient leaching, in particular P, poses a significant risk for water quality due to increased eutrophication (Misselbrook *et al.*, 1995).

Effect of fibres on the filtrate of a 10% soil-water solution (Table 1)

Similar to the results for deionised water, the 10% soil-water solution filtrate was more alkaline when raw wool was used as the filter material, and this differed significantly from the felted wool or coir filtrate pH (Table 1, $P < 0.05$). S and K were increased in the filtrate from raw wool, compared to other wool samples and coir ($P < 0.05$). P was significantly higher in the raw wool filtrate than scoured wool ($P < 0.05$), with other samples being intermediate. Raw wool filtrate had significantly higher pH, S, P and K compared to the initial soil-solution ($P < 0.05$). Whilst S concentration was numerically lower in the filtrate from scoured wool, felted wool and coir compared to the initial test solution, this was not to a significant extent ($P < 0.05$).

Conclusion

Sheep wool has potential to be used to water run-off from grasslands as indicated by high WHC and slower outflow rates. The natural impurities of raw wool, i.e. grease, suint, dirt and vegetable matter (Rippon, 2013), appeared to readily leach nutrients into aqueous solutions in a filtration set-up, indicating that raw wool is not a good candidate fibre for managing water run-off. Wool needs to be processed to reduce the risk of nutrient loading in run-off, for example by using scoured or felted wool. Further testing is needed to develop a mode of application of processed wool that can be applied at field-scale to protect waterways.

Acknowledgements

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Using LAI and NIR as rapid indicators for assessing weed impact in permanent meadows

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Abstract

Incorrect management practices in permanent meadows in north-eastern Italy may promote the spread of invasive species leading to a reduction in both productivity and forage quality. This study aimed to investigate variations in the leaf area index (LAI) and NIR-spectrum trend of permanent meadows' canopies invaded by *Erigeron annuus* and *Sorghum halepense* and to link these variations to forage quality. LAI, forage quality, and NIR-spectrum were measured in four permanent meadows located in two farms characterized by different levels of invasive species (15 vs 0% in cover abundance for *Erigeron annuus*; 30 vs 0% in cover abundance for *Sorghum halepense*). Forage quality (dry matter content, crude protein, NDF, ADL, ash, and ether extract content) and LAI were conducted using the Polisphec NIR and Li-Cor LAI-2200 Plant Canopy Analyzer, respectively. LAI values were equal and lower (for meadows invaded by *E. annuus* and *S. halepense* respectively) in areas dominated by invasive species. Forage quality was not affected by the presence of weed species; however, the NIR revealed distinct spectrum trends separating invaded and non-invaded meadows. Our results indicated that LAI and NIR-spectrum trend can be used as effective indicators for rapidly assessing the presence of such species on permanent meadows.

Keywords: bioindicator, *Erigeron annuus*, forage quality, *Sorghum halepense*

Introduction

Permanent meadows are important for maintaining animal and plant biodiversity, especially in lowlands where intensive agriculture has reduced and simplified seminatural areas. In northern Italy, meadows located in lowlands are subject to invasion by weeds, particularly *Erigeron annuus* and *Sorghum halepense*. Incorrect management (Song *et al.*, 2018) and climate change promote the spread and establishment of these species, with possible repercussions on the quality.

The aim of the study was to investigate green forage quality through NIR technology and LAI in meadows with high and nil weed coverage, in order to assess the use of these tools for quantifying the invasion stage of weed species for the improvement of management practices.

Materials and methods

The experiment was conducted in two meadows at the experimental agricultural farm of Padova University in Legnaro, (Padova, 45°21'3.5"N 11°57'12"E) and two meadows in Spilimbergo (Udine, 46°7'47.7"N 12°49'47.7"E), with different levels of *S. halepense* and *E. annuus* invasion, respectively. Legnaro has an average annual temperature of 13.9°C and an annual rainfall of 828 mm (ARPAV, 2025; 30-year average). Spilimbergo has an average annual temperature of 13.5°C and an annual rainfall of 1564 mm (ARPAFVG, 2025; 34-year average). The meadows in Legnaro were fertilized with slurry (200 kg N ha⁻¹ year⁻¹) in spring at the beginning of the spring regrowth and cut four times per year. The first mowing was performed on 29 April and the second on 12 June. The meadows of

Table 1. Results of the analysis of variance testing the effect of invasive species (*Erigeron annuus* and *Sorghum halepense*) in Leaf area index (LAI), dry matter (DM), crude protein (CP), neutral detergent fibre (NDF), acid detergent lignin (ADL), ash (Ash), fibre (F), ether extract (EE) content, and plant cover (PC)

	LAI	DM	CP	NDF	ADL	Ash	F	EE	PC
<i>Erigeron annuus</i>	NS	NS	NS	NS	NS	NS	NS	NS	**
<i>Sorghum halepense</i>	*	**	NS	NS	NS	NS	NS	**	***

p* < 0.05; **p* < 0.01; **p* < 0.001; NS, not significant.

Spilimbergo were fertilized with chemical fertilizer (200 kg ha⁻¹ year⁻¹; NPK: 20, 10, 10) in March; they are cut twice per year, and the first mowing was performed on 27 June. Two days before the June mowing, a floristic survey was conducted using the Braun-Blanquet method, recording all species in a 100 m² area and estimating percentage cover of each of them. For each survey, five LAI measurements using the Li-Cor LAI-2200 Plant Canopy Analyzer and five measurements with the Polisphec NIR were performed on a 1 m² sub-area to assess forage quality and canopy spectrum. Forage quality included dry matter (DM), crude protein (CP), neutral detergent fibre (NDF), acid detergent lignin (ADL), ash, fibre (F), and ether extract (EE) content in fresh fodder. Immediately after the mowing, six images of the ground surface were taken with a camera (Pentax K30) at a fixed height of 130 cm, and analysed with Turf Analyzer for ground plant cover (PC). The data collected were analysed with R (R Core Team, 2025), performing analysis of variance for each parameter to test the effect of weed invasion. The spectrum of the 206 samples (5 for each of the 4 meadows) was visualized by non-metric multidimensional scaling (NMDS) of Bray–Curtis distances of leaves. To further test the effect of weed invasion, permutational multivariate analyses of variance (PERMANOVA) were carried out. The data collected in meadows invaded by *E. annuus* and *S. halepense* were analysed separately

Results and discussion

The two meadows located in Legnaro belong to Molinio-Arrhenatheretea, one characterized by high coverage of *Lolium multiflorum* (35%), *Trifolium repens* (15%), and *Trifolium pratense* (10%), while in the other by *L. multiflorum* (30%), *S. halepense* (30%) and *Convolvulus arvensis* (10%), with *T. repens* (3%) and *T. pratense* (5%) having a low coverage. The two meadows located in Spilimbergo belong to Arrhenathalia elatioris, one characterized by high coverage of *Trisetaria flavescens* (35%), *Arrhenatherum elatius* (10%), *Avenula pubescens* (10%), and *Poa pratensis* (15%), while the other had a high coverage of *A. pubescens* (40%), *A. elatius* (15%), *T. flavescens* (8%), and *E. annuus* (15%).

Weed presence did not affected LAI, DM, CP, EE, NDF, and F while it affected the PC (Table 1), with meadow not invaded having a lower percentage than the lawn with *E. annuus* (18.28% vs. 26.95%). In meadows with and without *S. halepense*, no significant results were found for CP, ADL, F, NDF and ash (Table 1). Significant differences were found for LAI, DM, EE, and PC. LAI and PC were higher in the non-invaded meadow (4.43 vs 3.42 and 35.45% vs 8.90%), while DM and EE were higher in the meadow with a high presence of *S. halepense* (data not shown).

The dense canopy of high-productive meadows favours species with an erect growth habit, and this limits the growth of plants with a creeping habit or small size, leaving uncovered spaces at ground level (Gusmeroli, 2012). We found that *S. halepense* seemed to be a strong competitor with highly productive plant species, resulting in lower ground cover where present, whereas *E. annuus* occurs in canopies with greater ground cover due to the presence of creeping-habit species under *E. annuus*. However, at advanced stages of herbage maturity, LAI may progressively decrease with forage yield because of the increasing proportion of non-leafy plant material contributing more to the yield than to LAI (Peratoner *et al.*, 2021).

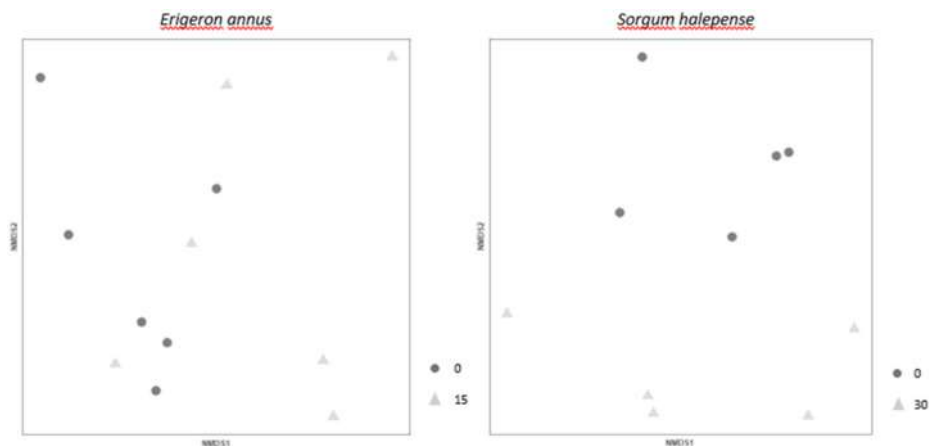


Figure 1. Spatial ordination resulted from NMDS on NIR-spectrum of meadows invaded by *Erigeron annuus* (0 vs 15% in cover abundance; on the left) and *Sorghum halepense* (0 vs 30% in cover abundance; on the right).

The graphical visualization of NMDS (stress value = 0.02) displayed a good separation of weed invasion for *S. halepense* but not for *E. annuus* (Fig. 1). However, the separation was supported by PERMANOVA, which revealed a high significance of weed invasion for both species ($p < 0.001$ and $p < 0.01$, respectively).

Conclusion

These preliminary results of a wider study, covering twenty-two meadows in north-eastern Italy, are promising for a broader study of the spread of these weeds in forage meadows. Estimating the presence of weeds can be a way to monitor their spread, implement the best containment techniques, and favour native grass species resulting in better forage quality. The leaf area index and NIR-spectrum trends appeared promising in their estimation of degree of invasion; however, further insights are needed to gain a deeper knowledge of the effect of weeds on LAI and forage quality at different stages of weed growth and on which spectrum bands can be used in remote sensing to identify invaded meadows.

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Rewetting peat soils in Dutch dairy systems: A case study on *Galba truncatula* presence

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Abstract

Extensive drainage has made Dutch peatlands suitable for intensive dairy farming, but also causes peat oxidation, greenhouse gas emissions, and land subsidence. Raising groundwater levels is a key mitigation measure, though it may reduce grassland productivity, grazing opportunities, and increase parasite risk. This case study explored how elevated groundwater levels relate to grassland functioning and *Galba truncatula* presence within two contrasting peatland dairy farm contexts in the North of the Netherlands. Over 382 days, 25 fixed monitoring points per pasture were measured biweekly for volumetric water content, soil temperature and pore water conductivity. The presence of *G. truncatula* was observed on five points. Soil samples were analysed for pH and organic matter content. Binary logistic regression assessed the relationship between soil properties and snail presence. Soil moisture, temperature, and pH were positively associated with snail presence (Nagelkerke $R^2 = 0.303$). The conventional farm's thicker clay layer, higher organic matter, and lower pH likely suppressed *G. truncatula*, highlighting that elevated water levels alone do not determine parasite risk. These case-study observations illustrate how local soil and water conditions influence both grassland function and parasite risk under rewetting and improve our understanding of the impact on the Dutch peatland dairy systems.

Keywords: Dutch dairy, *Galba truncatula*, peatland, rewetting

Introduction

Dairy farming is central to the Dutch peat meadow regions. Historic drainage enabled intensive agriculture but caused peat oxidation, subsidence, and greenhouse gas emissions (Van den Akker *et al.*, 2018). Mitigation via (e.g.) raised ditch levels and infiltration slows degradation (Van den Akker *et al.*, 2007) but reduce bearing capacity, grazing days, and yield, while increasing parasite risk (Holshof *et al.*, 2011). Liver fluke (*Fasciola hepatica*) is a parasite thriving under wet conditions due to higher presence of its intermediate host, *Galba truncatula* (Hoving *et al.*, 2021). Infected livestock show non-specific symptoms, including reduced productivity and weight loss (Radostitis *et al.*, 2007). This case study was conducted on two peatland dairy farms in the North of the Netherlands. It aims to explore soil conditions and management strategies related to *G. truncatula* occurrence under elevated groundwater levels and examine the multifaceted impact of rewetting on Dutch peatland dairy systems.

Materials and methods

This case study was conducted on two peatland dairy farms in the northern part of the Netherlands: a biodynamic (Demeter) farm and a conventional farm. Farms were selected based on liver fluke antibodies in bulk milk samples, grazing of dairy and young stock, and on-farm rearing of young stock. On each farm a pasture was selected and monitored for 382-days (April 2024–May 2025). Each pasture had 25 fixed monitoring locations. Points one to 20 were semi-randomly assigned across wet/dry zones, vegetation patterns, and proximity to drainage channels. Volumetric water content (VWC) (%), soil temperature (ST) (°C), and pore water conductivity (ECp) (mS m⁻¹) were measured biweekly

Table 1. Mean (M) and Standard Deviation (SD) of the 6 observed variables on both farms over the complete 382-days research period.

Variable	Unit	Conventional	Biodynamic	Interpretation / relevance
pH (KCl)	–	4.3 ± 0.4	5.0 ± 0.3	Lower pH in Lemmer below optimal range (4.5–6.0) for <i>G. truncatula</i>
Organic matter (OMC)	%	34.4 ± 14.3	18.6 ± 4.2	Higher OMC in Lemmer indicates more peat influence
Clay layer thickness	cm	30.6 ± 13.0	40.5 ± 9.2	Thicker clay in Lemmer, affecting drainage and soil chemistry
Volumetric water content (VWC)	%	40.0 ± 15.0	48.5 ± 14.0	Differ between locations, but not consistent with <i>G. truncatula</i> presence
Soil Temperature (ST)	°C	15.0 ± 5.7	13.9 ± 5.6	Differ between locations, but not consistent with <i>G. truncatula</i> presence
<i>G. truncatula</i> presence	–	Absent	Present (17 ind.)	Found only under higher-pH conditions

Values are mean ± SD.

using a WET2-sensor (Delta-T Devices), four times within a 1 m radius. Both soils were classified as ‘Waardveengrond’ (Histosol) with reed- and sedge-based peat under a clay cover (BOFEK). Ditch water levels were obtained from Wetterskip Fryslân. Soil sampling was conducted once per pasture at 10 representative locations. Organic matter content (OMC) was determined using Kopecky rings and loss-on-ignition (105°C drying, 500°C ashing, duplicate). Soil pH (KCl method) was measured in duplicate (approx. 40 auger cores collected within 1 m radius). The presence of *G. truncatula* was assessed biweekly at five GPS-marked ditch locations (points 21 to 25) per site. Additionally, semi-structured interviews were conducted with both farmers, focusing on water management, grazing, forage quality, cattle health, and farm performance. Data were presented in summary tables to characterize soil and hydrological conditions for each farm and compare locations with and without *G. truncatula* within a descriptive case-study framework. A binary logistic regression was performed to assess whether and which soil conditions contributed to the presence of *G. truncatula*. The dataset used for this analysis was restricted to the observations from the biodynamic farm.

Results and discussion

The farms represent contrasting systems. One is a high-input conventional farm (9300 kg cow⁻¹ year⁻¹), using liquid manure, strip grazing, and mowing every 4–5 weeks. The other a low-input biodynamic farm (5600 kg cow⁻¹ year⁻¹), using solid manure, Nieuw Nederlands Weiden, and mowing twice per year. Summer water levels were similarly high: 0.05–0.35 m-se and 0.01–0.30 m-se respectively. In total, 17 individual *G. truncatula* were found during the monitoring period, all on the biodynamic farm. Based on Smith *et al.*, (2024) higher ST and VWC are positively correlated with the snail’s presence. Although VWC and ST differed (not significantly) between both farms (Table 1), no consistent pattern with snail presence was observed, suggesting other soil properties may have played a more defined role.

Organic matter content on the conventional farm (M = 34.4, SD = 14.3) was higher than on the biodynamic (M = 18.6, SD = 4.2) likely due to its thinner clay layer (30.6 ± 13.0 cm vs. 40.5 ± 9.2 cm), suggesting partial sampling of the underlying peat. This aligns with values typical of peat-rich soils (>30% OMC; Bakker & Schelling, 1996). *G. truncatula* prefers clay over peat (Smith *et al.*, 2024) and tends to avoid high-organic soils, partly due to associated low pH. The pH on the conventional farm was below the optimal range for *G. truncatula* of 4.5–6.0 set by Bishop in 1981; this may explain its absence alongside lower VWS and ST. Solid manure use on the biodynamic farm likely contributed to the higher pH and improved soil conditions. A robust binary logistic regression was performed to assess whether ditch location (on the biodynamic farm) could predict *G. truncatula* presence. The model was significant ($\chi^2(4) = 14.06, p = 0.007$) and explained around 18% of the variation (Nagelkerke $R^2 = 0.184$). But no clear location was selected by the model as the primary location

for *G. truncatula* presence. Therefore, a further binary logistic regression was performed assessing the influence of soil moisture, temperature, ECp and pH (KCl) on the presence of the liver fluke snail. The model was significant ($\chi^2(4) = 18.549$, $p < 0.001$) and explained around 30% of the variation (Nagelkerke $R^2 = 0.303$). Indicating a significant positive influence of soil moisture ($B = 0.108$, $SE = 0.053$, $p = 0.042$), soil temperature ($B = 0.37$, $SE = 0.0176$, $p = 0.036$) and soil pH ($B = 19.265$, $SE = 6.696$, $p = 0.004$) on the presence of *G. truncatula*.

Conclusion

The two farms represented contrasting peatland contexts with clear differences in soil conditions and parasite risk. Groundwater levels were similar, but *G. truncatula* was not observed on the conventional farm, indicating that a thinner clay layer, higher organic matter and lower pH rather than elevated water tables explained the observed differences, even though moisture and temperature conditions appeared suitable. The binary logistic regression (executed on the biodynamic farm data) showed a positive relation between soil moisture, temperature and pH and the presence of *G. truncatula*. The observed differences in pH and organic matter between both farms not only affected *G. truncatula* occurrence but also likely influenced grass productivity, linking soil conditions under rewetting to both animal health and grassland performance. This case study suggests that rewetting peat without a clay layer does not necessarily increase liver fluke risk, while clay-covered peat soils may pose a greater risk under high water tables. As it influences soil moisture, temperature and mainly pH levels, the farmers' management strongly influences parasite risk and grassland performance, highlighting delicate balance between productivity and resilience in rewetted peat systems.

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Novel machine learning tools to identify factors regulating CO₂ fluxes in a boreal legume grassland

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Abstract

Grasslands are crucial for the global carbon (C) cycle and the livestock sector. Rapidly changing climate in the boreal region is a considerable threat to the sustainability of Finnish dairy and livestock industries. To project grassland responses to future climate, it is essential to study their current behaviour of C sequestration and greenhouse gas (GHG) fluxes. We measured GHG fluxes using the eddy covariance (EC) technique and supporting micrometeorological variables (2017 to 2020) from a boreal legume grassland in eastern Finland. The grassland was intensively managed with 2–3 cuts per year, along with synthetic or organic fertilizer application (106 kg N ha⁻¹ year⁻¹). In addition, the grassland sward was renewed every 3–4 years. We applied tree-based machine learning model and Shapley Additive exPlanations (SHAP) analyses for explaining the variations in sub-daily (30-min average) CO₂ fluxes with meteorological and management data. We present here the results of these analyses. We aim to investigate intra- and inter-annual changes in CO₂ fluxes and regrowth after grass cuts and examine grassland responses to sward renewals. We hypothesize that ML tools can effectively elucidate the seasonal variations in grassland CO₂ fluxes, with insights into climate-smart management options and securing sustainability of the Finnish livestock sector.

Keywords: boreal environment, climate-smart agriculture, eddy covariance, management practices, milk and beef production

Introduction

CO₂ flux tower measurements have become a cornerstone in understanding carbon dynamics in grassland ecosystems. These towers, using the eddy covariance method, provide continuous, high-resolution data on net ecosystem exchange (NEE), capturing the balance between photosynthesis and respiration (Kheradmand *et al.*, 2023). Environmental variables such as soil temperature, air temperature, latent heat, and sensible heat are critical inputs for modelling NEE. Studies have shown that these factors significantly influence CO₂ fluxes, with soil and air temperature playing dominant roles in ecosystem respiration. Management practices, including fertilization and cutting, also affect CO₂ exchange. For instance, intensively managed grasslands exhibit higher gross primary production and ecosystem respiration compared to unmanaged sites, sometimes rivalling forest ecosystems in carbon uptake (Gilmanov *et al.*, 2010). Recent advances in machine learning have enhanced the predictive power of flux tower data. A novel model combining GA and Fourier series (IOGAFS) achieved determination coefficients of 0.81 for grasslands, demonstrating its utility in areas lacking direct measurements. Long-term flux tower datasets have enabled global assessments of grassland CO₂ dynamics. Analysis of 316 site-years revealed that managed grasslands can act as significant carbon sinks, with net CO₂ uptake exceeding 700 g CO₂ m⁻² year⁻¹ in some cases. However, the fate of this carbon—whether retained in biomass or exported off-site—remains a key consideration in evaluating true sequestration

potential. Sub-daily (30-minute average) CO₂ fluxes in grassland ecosystems can be effectively modelled using tree-based machine learning algorithms. These models, such as Random Forest, XGBoost, and LightGBM, can capture complex nonlinear relationships between CO₂ fluxes and input variables. Meteorological data including air temperature, soil moisture, solar radiation, and wind speed, along with management data such as fertilizer application and grass cutting schedules, are used as predictors in the model.

Materials and methods

The study was conducted at an agricultural field (6.3 ha; 280 m×220 m) located in Maaninka, eastern Finland (63°9′N, 27°14′E, 89 m a.s.l.). The region experiences a continental boreal climate with 30-year (1981–2010) mean annual temperature of 3.2°C and precipitation of 612 mm. The soil is classified as a Haplic Cambisol/Regosol (Hypereutric, Siltic), with silt loam texture (25 ± 6% clay, 53±9% silt, 22 ± 8% sand). Soil properties include pH 5.8 ± 0.2, organic carbon 3.0 ± 0.5%, total nitrogen 0.2 ± 0.03%, C:N ratio 15 ± 0.4, and bulk density 1.1 ± 0.1 g cm⁻³. The experimental grassland was established in 2015 with timothy (*Phleum pratense* L. cv. Nuutti; 15 kg ha⁻¹) and red clover (*Trifolium pratense* L. cv. Ilte; 5 kg ha⁻¹), reseeded in May 2017, and it underwent complete renewal in spring 2019. The study spanned three years covering an entire grassland rotation cycle. The site received an average annual fertilization of 106 kg soluble N, 28 kg P, and 50 kg K ha⁻¹, applied in two splits (May and mid to late June). Grass harvesting occurred twice annually in the first and second years (late June and mid-August) and once in the last year (early August). Grassland renewal in autumn 2018 involved glyphosate application, ploughing, and winter bare fallow. Re-establishment in June 2019 included seeding with timothy (15 kg ha⁻¹), red clover (3 kg ha⁻¹), and barley (*Hordeum vulgare* L.; 160 kg ha⁻¹) as a cover crop.

An eddy covariance (EC) tower was centrally positioned at the study site. The system comprised: (1) a sonic anemometer (R3-50, Gill Instruments, UK) at 2.5 m height measuring three-dimensional wind velocity and sonic temperature; (2) an infrared gas analyser (LI-7000, Li-Cor, USA) for measuring CO₂ and water vapour concentrations. For further details on flux data processing, data quality control, CO₂ flux partitioning into gross primary productivity (GPP) and total ecosystem respiration (TER), please refer to Li *et al.* (2023). We used tree-based machine learning models, such as Random Forest, XGBoost for predicting sub-daily (30-min average) CO₂ fluxes from the grassland study site, as these models are particularly effective for handling nonlinear relationships and interactions among meteorological and management variables (Lee and Lee, 2024). The input data included CO₂ flux measurements from the eddy covariance tower, along with environmental variables such as soil and air temperature, solar radiation, and vapour pressure deficit. Management data, including fertilization and mowing events, were also incorporated to capture anthropogenic influences on carbon exchange. The modelling process began with data preprocessing, including handling missing values, normalizing features, and encoding categorical variables. The dataset was then split into training and testing subsets. To interpret the model and understand the contribution of each feature to predictions, Shapley Additive exPlanations (SHAP) analysis was employed. SHAP values provide a unified measure of feature importance by quantifying the impact of each variable on individual predictions. SHAP summary plots and dependence plots were then used to visualize feature importance and interactions.

Results and discussion

Net ecosystem CO₂ exchange (NEE) of a vegetated ecosystem such as the grassland study site under investigation in this work is a net function of the amount of atmospheric CO₂ fixed as GPP by the

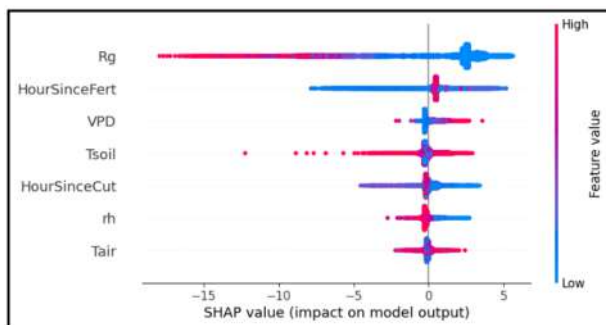


Figure 1. SHAP value vs Feature value plot describing the impact of features or explaining variables on the predicted net ecosystem CO₂ exchange (NEE) at the mineral soil grassland in eastern Finland.

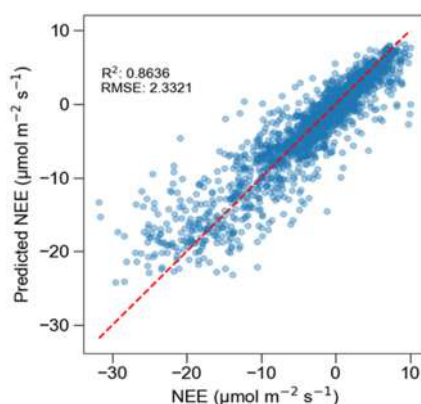


Figure 2. A 1:1 scatter plot of measured vs predicted net ecosystem CO₂ exchange (NEE) at the mineral soil grassland in eastern Finland.

vegetation and that lost to the atmosphere as TER. NEE depends on several meteorological variables such as solar radiation (R_g), vapour pressure deficit (VPD), soil (T_{soil}) and air temperature (T_{air}) and relative humidity (RH). Among these meteorological variables solar radiation plays a major control on the photosynthetic CO₂ uptake by the vegetation. This is clearly seen in Figure 1 below, which highlights in terms of SHAP values how each feature included as an explaining variable in the machine learning model employed in this study fares compared to all other features. This is a powerful way of visualizing how sensitive is the grassland NEE to different climatic variables. We can also see those other variables such as VPD, air and soil temperature present a moderate control over NEE in this ecosystem. This is consistent with numerous studies (e.g., Shurpali *et al.*, 2025) where individual non-linear regressions between NEE and climatic variables have yielded similar observations. The advantage with machine learning model application is that one can clearly visualize the relative importance of each meteorological variable in regulating NEE. Moreover, while the impact management activities (e.g., hours since fertilization (HoursSinceFert) and grass cuts (HoursSinceCut)) is difficult to quantify, the machine learning models make it possible to understand their role in grassland ecosystem CO₂ exchange (Figure 1).

Conclusion

The results of the XGBoost model for predicting NEE based on NEE measurements and governing climatic and management variables agree well with observations (Figure 2) with an R^2 value of 0.86 at 95% confidence interval. While ML models are often criticised for overfitting, owing to autocorrelation among explaining variables, simulations done in this study agree with the accepted, fundamental plant physiological concepts of variation in GPP and TER with their governing variables such as Rg, VPD, Tsoil and management practices.

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Estimating soil carbon sequestration potential in Portuguese grassland applying the IPCC's Tier 1 method

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Abstract

Soil carbon sequestration (SCS) is a nature-based, low-cost climate mitigation strategy that also contributes to the climate adaptation of agricultural systems. Grasslands can have their soil organic carbon (SOC) stocks enhanced through practices such as improving legume species varieties and reducing grazing. However, uncertainties remain both in estimating and measuring the impact of the application of land use and management changes, as it varies with soil, climate, and historic land use. IPCC (Intergovernmental Panel on Climate Change) guidelines are commonly used to estimate SOC and SOC sequestration potentials at different tiers. Here, the IPCC's Tier 1 methodology was applied to estimate the sequestration potential of the conversion of rainfed and irrigated crops to improved grassland and the improvement of degraded and non-degraded grasslands for $n = 5062$ unique agricultural cropland and grassland sites in mainland Portugal. The improvement of poor degraded grasslands resulted in the highest national sequestration potential, equal to $0.6 \text{ Mt C year}^{-1}$ ($2.2 \text{ Mt CO}_2\text{eq year}^{-1}$), about 4% of Portugal's emissions in 2021, if applied in all managed grassland areas.

Keywords: grassland management, IPCC guidelines, mitigation, soil organic carbon

Introduction

Greenhouse gas (GHG) emissions from agriculture, forestry and other land uses (AFOLU) represent 22% of total worldwide emissions (Calvin *et al.*, 2023). This considerable impact can be reduced with adequate changes in agricultural practices and land-management which also offer other co-benefits (Lal, 2008; Paustian *et al.*, 2016; Smith *et al.*, 2008; Verchot *et al.*, 2007). Grasslands represent a significant component of Portugal's agricultural land and play a crucial role in both forage production and soil carbon dynamics. Rotational grazing, irrigation and improved grass species have been popularized as effective measures towards SCS in grasslands. A project in Portugal involving the adoption of sown biodiverse permanent pastures rich in legumes (SBPPRL) sequestered an estimated cumulative 1.54 Mt CO_2 between 2009–2014 (Ravaioli *et al.*, 2023; Teixeira *et al.*, 2015). However, accurately estimating sequestration potentials remains challenging due to regional heterogeneity in soils and climate (Scharlemann *et al.*, 2014; Schrumpf *et al.*, 2011), data scarcity, limited national soil monitoring (Ramos *et al.*, 2017), and methodological constraints, such as the use generalist default emission factors, or resource-intensive sampling experiments (Vetter *et al.*, 2022). This study applies the IPCC Tier 1 methodology to quantify SOC gains from grassland management improvement and cropland-to-grassland conversion scenarios across mainland Portugal.

Table 1. Average SOC and total national sequestration potential estimates

Identifier	Transition	Average SOC ($\text{t C ha}^{-1} \text{ year}^{-1}$)	Total national potential (Mt C year^{-1})
M3A	Rainfed cropland to improved grassland	0.82	0.45
M3B	Irrigated cropland to improved grassland	1.05	0.18
M5A	Degraded grassland to improved grassland	0.84	0.60
M5B	Nominal grassland to improved grassland	0.39	0.28

Materials and methods

A georeferenced database was developed for 5062 managed cropland and grassland sites across mainland Portugal. Data to characterize these sites in terms of land-use, climate, soil type and SOC measurements, was compiled from COS, IPMA, the Soil Atlas of Europe and LUCAS. Four land-use transition scenarios were assessed: Annual Cropland to Improved Grassland (M3A, M3B)—conversion of rainfed crops and irrigated annual crops into improved grassland, respectively; Grassland Improvement (M5A, M5B)—conversion of degraded grassland and nominal grassland to improved grassland.

SOC changes were computed for each transition. This was based on the IPCC Tier 1 method, which estimates SOC changes by computing the difference between steady state SOC levels in two different LU or Management regimes, over a 20-year period. SOC stocks for each LU/management are described by:

$$\text{SOC} = \text{SOC}_{\text{REFc,s}} \times F_{\text{LU}_{\text{c,i}}} \times F_{\text{MG}_{\text{c,i}}} \times F_{\text{I}_{\text{c,i}}}, \quad (1)$$

where $\text{SOC}_{\text{REFc,s}}$ is the organic carbon in soils in undisturbed conditions in tC ha^{-1} , $F_{\text{LU}_{\text{c,i}}}$, $F_{\text{MG}_{\text{c,i}}}$ and $F_{\text{I}_{\text{c,i}}}$ are dimensionless stock change factors (SCFs) which represent respectively, the impact of different land-use types (LU), management regimes (MG) and input levels (I) on soil carbon stocks; “c”, “s” and “i” represent climate, soil and the set of defining land-use/management conditions, respectively. National potentials were obtained by scaling per-hectare changes to managed areas.

Results and discussion

The conversion of irrigated crops to improved grasslands (M3B) leads to the highest yearly average SOC sequestration per unit area ($1.05 \text{ t C ha}^{-1} \text{ year}^{-1}$). Improvement of degraded grasslands results in the highest maximum national SOC sequestration potential ($0.6 \text{ Mt C year}^{-1}$), if applied in all managed grasslands. Table 1 shows the estimates obtained for each transition. Higher sequestration potentials were obtained for sites on podzol soils and in warm temperate moist conditions, sites on sandy soils and in warm temperate dry conditions had lower estimates. The SOC stocks obtained following IPCC's Tier 1 were lower than the in situ LUCAS values.

Our estimates exceed rates reported in long-term cropland-to-grassland studies (approx. $0.54 \text{ t C ha}^{-1} \text{ year}^{-1}$) but fall below modelled rates for degraded pasture improvement (approx. $1.78 \text{ t C ha}^{-1} \text{ year}^{-1}$). These discrepancies reflect climatic and methodological differences. Key limitations include: (i) generalized management representation due to limited site-specific data and use of highly generalized SCFs, and (ii) assumed linear SOC accumulation, which may oversimplify temporal dynamics and hinder comparability.

Conclusion

Improving degraded pastures offers the largest national SOC sequestration potential in Portugal, offsetting approx. 4% of annual emissions. For robust policy design, future work should adopt Tier 2/3 methods with localized factors and expand soil monitoring.

Acknowledgements

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Structuring and production of *Opuntia* cv. Miúda forage under different irrigation depths

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Abstract

This study evaluated the yield and vegetative structure of the forage cactus *Opuntia cochenillifera* cv. Small, under different irrigation depths in relation to the field capacity of the soil. The experiment was carried out at the Federal Institute of Espírito Santo (IFES), Santa Teresa campus, Brazil, using three rows of 45 m of cactus plants submitted to five irrigation depths (1 mm, 2 mm, 3 mm, 4 mm and 5 mm), corresponding to approximately 0.25, 0.5, 0.75, 1 and 1.25 of the soil capacity of the field, respectively. Morphometric analyses were performed following a randomized block design (RBD) in a 4 × 6 factorial scheme, with biweekly measurements of height, width, number of cladodes and plant biomass. The data were analysed using analysis of variance (ANOVA) after checking the assumptions of normality (Shapiro–Wilk test) and homoscedasticity (Levene test). The results showed that irrigation at 4 mm, equivalent to field capacity, was the most efficient to maximize dry biomass production, while excess irrigation above field capacity (5 mm) reduced productivity due to possible oxygen limitation in the root zone. The growth of cladodes (height, width and thickness) increased with irrigation up to 4 mm and then stabilized. Therefore, keeping irrigation close to field capacity optimizes biomass yield and ensures robust cladode development in forage cactus cv. Miúda.

Keywords: biomass, cladodes, forage cactus irrigation depth, *Opuntia cochenillifera*

Introduction

The cactus, originally from Mexico, was introduced in Brazil as a host of the cochineal insect (*Dactylopius coccus*), used in the production of the red carmine dye (Frota *et al.*, 2015). The variety known as small palm or sweet cactus (*Opuntia cochenillifera*) stands out for its greater nutritional value and ease of management, as it has no spines (Lopes, 2009). Its crassulaceous acid (CAM) metabolism allows it to capture solar energy during the day and fix CO₂ at night, significantly reducing water loss through evapotranspiration (Ramos *et al.*, 2011; Sampaio, 2005). This adaptation makes it one of the most suitable forage options for regions with low rainfall, with dry matter yields reaching up to 80 t ha⁻¹ (Menezes *et al.*, 2005).

Although not commonly practised in cactus cultivation, irrigation can influence growth, productivity, and bromatological composition, with conflicting findings reported in the literature (Yosef *et al.*, 2009; Rostamza *et al.*, 2011; Islam *et al.*, 2012; Simsek *et al.*, 2011; Ul-Allah *et al.*, 2014). Its low water demand is attributed to CAM metabolism and dry tillage (Consoli *et al.*, 2013), although it produces less dry matter compared to other forage crops in the semi-arid region (Oliveira *et al.*, 2010). This study aimed to evaluate the yield, growth and vegetative structure of forage cacti under different irrigation depths, evaluating biomass production, as well as morphogenesis and vegetative development.

Materials and methods

The experiment was carried out in the Large Animal Sector of the Federal Institute of Education, Science and Technology of Espírito Santo, Santa Teresa Campus (19°48'S, 40°40'W; 150 m above sea level).

To establish the treatments, the area was deforested and the forage cactus plants were standardized to a height of 45 cm. Next, a localized irrigation system (drip irrigation) was installed using different emitters, flow measurement and calculation of the Christiansen Uniformity Coefficient (CUC) to evaluate the efficiency of the system.

Five in-depth irrigation treatments were established (0.25, 0.5, 0.75, 1 and 1.25 of field capacity), based on a crop coefficient (K_c) of 4.5 mm, with daily irrigation applied for 53 minutes. The experiment was carried out in an area composed of three rows of 45 m of forage cacti, where, fortnightly, the height and width of the plant canopy, number and area of cladodes, and biomass production per linear meter were evaluated.

A completely randomized design (CRD) was used, in a 5×3 factorial scheme, with three replications, totalling 48 experimental units. The variables were submitted to analysis of variance (ANOVA), after verification of normality (Shapiro–Wilk test) and homoscedasticity (Levene test).

Vegetative structuring was determined by evaluating the number, length, width, thickness, perimeter and area of cladodes (CA), cladode area index (CAI), green matter production (GM) and dry matter (DM). The cladode area was estimated using the equation: $CA = -211.5104 + 8.8649P$ (Santos *et al.*, 2010), and the CAI was calculated as the ratio of the cladode area to the soil area occupied by the plant. Green and dry biomass was obtained by cutting and weighing a 1-m linear section of the plants. For dry matter determination, ~1 kg samples were dried at 80°C in a forced air furnace, following the methodology of Silva and Queiroz (2002). Linear measurements were made using a tape measure, and the thickness of the cladode was measured with a digital caliper.

Results and discussion

The study evaluated the yield of forage cacti under different irrigation depths, using 0.48 m² (80 × 60 cm) lots, arranged in double rows. It was estimated that one ha contained 2174 m of cactus rows, covering approx. 1305 m². A correction factor of 2.719 was applied to convert dry matter (DM) to kg ha⁻¹. The median yield of the boxplot analysis was low (Figure 1), with little variability, indicating water limitation. As irrigation increased from 0.25 to 0.75 of the required depth, median yield and variability also increased, reflecting a progressive improvement in biomass accumulation. However, the maximum yield was observed below 4 mm of irrigation, equivalent to the field capacity, while the excess of irrigation (1.25) did not increase the yield and tended to reduce it, probably due to reduced soil aeration and possible physiological stress.

Conclusion

The ideal irrigation depth for forage cacti, corresponding to 100% of the soil capacity in the field, proved to be the most effective to maximize the production of dry biomass. However, the recommendation of this level of irrigation should be supported by a cost-benefit analysis, considering the economic feasibility and efficiency of water use under local climatic and soil conditions. In addition, an irrigation management strategy that prioritizes optimal water application during the first 30 days

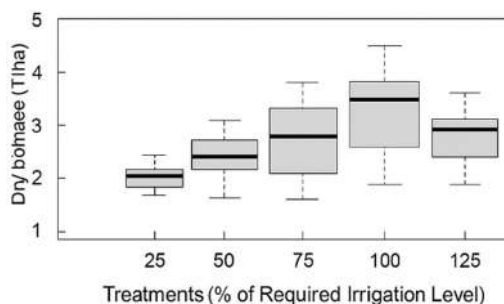


Figure 1. Dry biomass yield.

of cultivation is effective in promoting significant increases in cladode thickness, length, and width, thus contributing to more robust and efficient plant development.

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Char-induced changes in GHG fluxes, organic carbon, and yield in boreal grassland systems

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Abstract

Char amendments can benefit agricultural soils, but effects vary by type. In a three-month mesocosm study using a red clover–timothy mix, we tested biochar and hydrochar with and without nitrogen (N) on GHG emissions, soil carbon fractions (MAOC and POC), and biomass yield. Only N₂O emissions were affected: biochar with N increased emissions and nitrification, while hydrochar with N reduced both. Hydrochar without N showed the lowest N₂O emissions, with some uptake during active plant growth. While MAOC remained unchanged, both chars increased POC. Biomass yield was unaffected overall, though biochar with N reduced timothy yield. These findings highlight complex interactions between char type, soil, vegetation, and N management.

Keywords: biochar and hydrochar, boreal legume-forage, GHG flux, gross nitrogen transformation, nitrogen fertilization, POC and MAOC

Introduction

Due to inefficient nitrogen use, agriculture often loses fertilizer N to water bodies, causing eutrophication and acidification, and to the atmosphere as N₂O, contributing to climate change and ozone depletion (Ravishankara *et al.*, 2009). N fertilization can also increase CO₂ emissions through enhanced microbial activity (Sun *et al.*, 2022). To mitigate these impacts, one promising solution is the use of char, such as biochar, in agricultural soils. We conducted a mesocosm study on a typical boreal forage-legume mix (Timothy and red clover) with three objectives: (1) GHG Emissions: Assess biochar and hydrochar effects on CO₂, N₂O, and CH₄ emissions. We hypothesize reductions due to improved soil microbial biomass and nitrogen dynamics. (2) Biomass and NUE: Evaluate impacts on yield and NUE, expecting improvements from both char types. and 3) SOC Fractions: Investigate changes in particulate and mineral-associated organic carbon (POC and MAOC, respectively), hypothesizing increases via enhanced microbial carbon assimilation.

Material and methods

Mesocosms were set up in a greenhouse with 3.7 kg of sieved (2 mm) sandy soil packed into PVC rings (Ø 18 cm×12 cm) at field bulk density (1.01 g/cm³). Biochar (B, pyrolysis at 600°C) and hydrochar (H, hydrothermal carbonization at 220°C) were mixed into the top 6 cm. Timothy and red clover were sown and fertilized (75 kg N ha⁻¹) following standard farming practice. Soil moisture was maintained at 45% WFPS and adjusted every other day. We took two approaches to assess char's effect: (1) when

char was applied at a constant rate (hereafter, RAR—relative to char application rate) with fertilizer N, and (2) when char was applied considering char's C amount (hereafter, RCC—relative to carbon content) without fertilizer N. The latter approach assumes that char C is one of the key intrinsic properties that influences soil properties and GHG fluxes. The RAR included three treatments: N, NB, and NH with 20 t ha⁻¹ char applied. The RCC included two treatments: B and H without N, however with same amount of char C. We assessed char's effect in the presence and absence of fertilizer N by comparing results between RCC and RAR. Before comparison, we normalized the results from NB and NH in RAR by dividing with added C (kg) via char. We included a treatment without char and N (NBH0) to assess background effects. We measured GHG fluxes over 99 days using a closed static chamber method and analysed their concentrations via gas chromatography. Flux rates were calculated using gas concentration changes and the ideal gas law. Fluxes were converted to standard units (g m⁻² day⁻¹ for CH₄ and CO₂; mg m⁻² day⁻¹ for N₂O), and gaps between measurement days were filled using linear interpolation. Daily fluxes were summed per replicate to calculate cumulative emissions, then averaged by treatment. Soil MAOC and POC were analysed using size and density fractionation according to Skipp & Brownfield, (1993). Fresh biomass was measured at two harvests, then oven-dried at 60°C for 48 h to determine dry biomass, which was used to calculate yield (g dw m⁻²) and nitrogen use efficiency (NUE). NUE was assessed as partial factor productivity (NUE_{PPF}, g dw g⁻¹ N) and agronomic efficiency (NUE_{Ag}, Δg dw g⁻¹ N). We compared mean values of variables in Fig 1 using one-way ANOVA. However, we used two-way ANOVA for comparing timothy and red clover yields in each treatment in Fig. 1b and Fig. 1c.

Results and discussion

Only N₂O emissions were affected, with significantly ($p < 0.05$) higher levels in NB (1.56×) and NH (1.4×) than control (N) in RAR, and lower in H than B, NH, and NB in RCC (Fig. 1a). NB also emitted more N₂O than NH ($p = 0.003$). This increase aligns with some studies (Prommer *et al.*, 2014), but contrasts with most (e.g., Joseph *et al.*, 2021), possibly due to enhanced nitrification. Timothy yield differed significantly only in NB (lower than N in RAR) and NH (higher than H in RCC) (Fig. 1b–c). Timothy outperformed red clover in N (RAR) and NH (RAR, RCC), while red clover yielded more only in H. Overall, timothy did better in fertilized soils (N, NB, NH), and red clover in unfertilized ones (NBH0, B, H) where the former showed a significant positive correlation with the applied N amount. NUE_{PPF} and NUE_{Ag} showed no significant differences, though NH had 34% higher mean NUE_{Ag} than NB. Red clover yield increase with biochar aligns with earlier studies, likely due to improved root traits and nitrogenase activity (Xiang *et al.*, 2017). Soil MAOC was unaffected by NB and NH, though slightly higher than control in RCC (Fig. 1d). In contrast, POC significantly ($P < 0.05$) increased in NB and NH, indicating char as the main source. Normalized POC remained similar across char treatments, regardless of fertilization (Fig. 1e), consistent with Garbuz *et al.* (2020).

Conclusion

Contrary to our first hypothesis, we found that only N₂O emission was affected by biochar and hydrochar amendments. Our study recommends that biochar with fertilizer N should be avoided in legume-forage system because it has potential to increase N₂O by promoting nitrification. In contrast, hydrochar can offer N₂O mitigation option by limiting nitrification and also potentially by enhancing soil N₂O reduction in the absence of fertilizer N. Although our second and third hypotheses remained neutral, we found that hydrochar maintained MAOC levels and increased microbial biomass and, in contrast, biochar showed the opposite effects on MAOC and microbial biomass. This suggests that hydrochar has better potential than biochar for SOC stabilization. Our results highlighted the effect

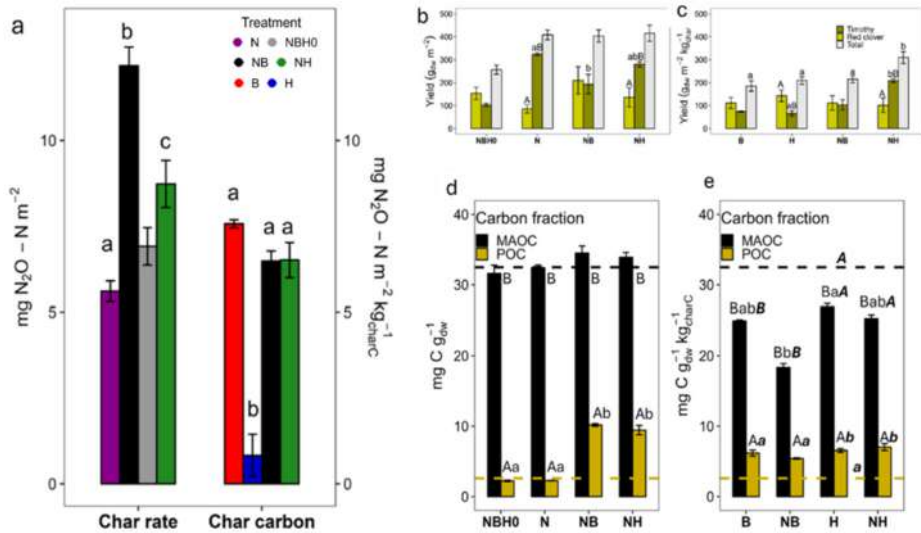


Figure 1. (a) Cumulative N_2O emissions, biomass yield in RAR (b) and RCC (c) respectively and POC and MAOC in RAR (d) and RCC (e). Legend colour in each plot represents experimental treatments or studied variables. In plots b, c, d, and e lowercase denote significant differences between the treatments whereas uppercase letters denote significant differences between the variables in each treatment. In plot e, italic bold lowercase and uppercase letter denote significant differences between initial (dashed lines) and end POC and MAOC respectively. Absence of letters denote no statistical difference.

of chars on GHG flux, biomass yield and SOC at mesocosm scale and at shorter time scale. We recommend future studies to explore char's effect for longer period in field conditions for providing realistic scenarios and for recommending large-scale use of char in agricultural soils.

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As it became warmer from 1982 to 2024, yield decrease of a hay meadow depended on the frequency of defoliation

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Abstract

Predicting the effect of climate change on grassland yields necessitates considering the potential interactions with grassland management. Our 42 years of data from a permanent grassland managed at two fertilizer regimes (PK or NPK) and two defoliation frequencies (two or three cuts a year) give insight into such interactions. The site is located in the Swiss Jura Mountains at 930 m a.s.l. At this site, mean air temperature has risen by 2°C over the last 42 years, while annual rainfalls did not change significantly. Considering the water deficit during the vegetation period (March–October) allowed us to account for one third of the large interannual yield variability (coefficient of variation 18%). Over the 42-year period, yields decreased by an average of 24% under the 3-cut but held steady under the 2-cut treatments. The yield decrease under more frequent defoliation cannot be explained by nutrient depletion, nor by a degradation of the botanical composition compared to the 2-cut treatments. A similar yield evolution over the decades was observed for all harvests of the year, without difference between the spring and the summer harvests. In conclusion, 42 years of data show that the frequency of defoliation may influence long-term grassland resilience to rise in temperature.

Keywords: biomass production, climate change, long-term experiment, management intensity

Introduction

As the average annual temperature increases, the length of the vegetation period increases (Vuffray *et al.*, 2016), which could benefit forage production in mountain regions. On the other hand, more frequent drought and heat periods are detrimental to forage production (De Boeck *et al.*, 2016), and grasslands on shallow soils over limestones, like those found in the Jura Mountains, could be severely affected by such effects. Moreover, Meisser *et al.* (2013) showed that the effect of drought stress on grassland yields might be aggravated by frequent defoliations. The combined impact of these factors on forage production in lower mountains remains to be quantified. In this study, we analyse a series of yield and weather data from a long-term experiment spanning 42 years, to assess the long-term effect of climate change on the forage productivity of a hay meadow under two different mowing frequencies.

Materials and methods

The long-term experiment is located at 930 m a.s.l. in the Swiss Jura Mountain (47°19′49.7″N 7°39′54.5″E), on a gleyic cambisol over limestone. The experiment was first established in 1972. We here analyse the yield data from 1982 to 2024. Over the last 42 years, average temperature was 7.8°C and average precipitations 1200 mm year⁻¹. The initial plant community was a typical hay meadow of the lower mountain (*Arrhenatherion elatioris*). Plot size is 15.9 m², from which the 7.8 m²-large middle stripe was used for measurements. The plots are harvested at 5–6 cm above ground level with a motor mower either twice or thrice a year. In the 2-cut treatment, the first harvest was taken at the

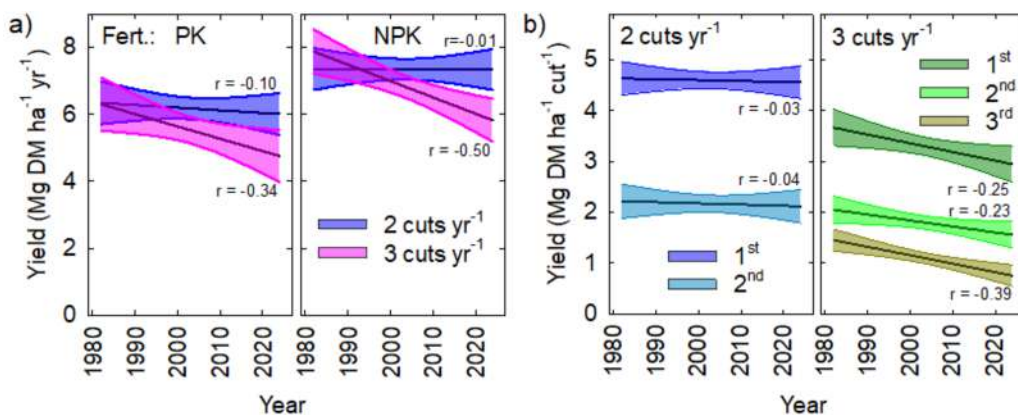


Figure 1. (a) Evolution from 1982 to 2024 of the annual yield of a hay meadow receiving either only PK or NPK fertilizer, and harvested either twice or thrice a year (abbreviated yr), and (b) evolution during the same timespan of the yield of the different yearly harvests (1st, 2nd, 3rd: first, second and third harvest of the year). Shown are the linear regression lines and their 95% confidence intervals. In (b) the two fertilizer treatments are pooled.

beginning of July and toward the end of September. In the 3-cut treatment, the harvests were taken at the beginning of June, beginning of August and toward the end of September. Since 1982, the period between the first and the last harvest of the year has, on average, increased by 6.5 and 7.5 days every 10 years in the 2- and 3-cut treatments, respectively. The second factor tested in the experiment is the rate of N application in a factorial design with three replicates: the NPK treatments receive yearly 75 kg N ha⁻¹ (ammonium nitrate, divided into equal applications before each regrowth), 26 kg P ha⁻¹ and 113 kg K ha⁻¹. The PK treatments receive the same amount of P and K fertilizer as the NPK treatments, but no N. Yields were assessed every year, while the botanical composition of the meadows was assessed every four years by visual estimation of the relative biomass abundance of the species. The daily temperature, rainfalls and relative sunshine duration data, specifically for the experimental site, were provided by MeteoSwiss. These local time series were generated using spatial interpolation from station data on to a regular grid, followed by bilinear interpolation and local linear regression with altitude (MeteoSwiss, Grid-Data Products: Local Time Series). The potential evapotranspiration was evaluated according to Turc (1961) and the water deficit was calculated as the difference between rainfalls and potential evapotranspiration. The data were analysed by ANCOVA with the cutting regimes and N fertilizer rates as categorical variables and the years as continuous predictor using StatisticaTM 13, as well as by simple linear regressions of the biomass yield as a function of the years 1982 to 2024 for the single treatments.

Results and discussion

The number of cuts impacted the yield evolution during the last four decades (number of cuts × year interaction $p < 0.01$), while the rate of N application did not. With two cuts a year, yield was maintained over the last forty years but decreased significantly with three cuts a year (Fig. 1a). Indeed, with three cuts a year, yield decreased on average by 42.9 kg DM ha⁻¹ per year (SE 11.8; $p < 0.001$) across both N application rates. This corresponds to a 24% yield decrease over the 40-year period. At this site, annual mean air temperature has risen by 2°C over the last 40 years ($p < 0.0001$). The slight trend toward decreasing rainfalls was not significant, both for the whole year and for the vegetation period (March–October; data not shown). But because of the increasing evapotranspiration, the water deficit of the March–October period increased on average from about –210 mm to about –240 mm

($p < 0.0001$) during the considered timeframe. The interannual yield variability was very large, with a mean coefficient of variation of 18%. The March-October water deficit was the weather parameter best explaining this variability, but considering it allowed to account for only one third of the interannual yield variability. In the 3-cut NPK treatment, P and K fertilizer applications corresponded on average to 3.8 kg P and 16.5 kg K applied per Mg of DM yield and were therefore sufficient to compensate for the export of P and K with the harvested biomass (Huguenin-Elie *et al.*, 2017). The yield decrease observed in the 3-cuts treatments was therefore not due to nutrient depletion, which was confirmed by soil analyses data (not shown). In 2019 and 2023, the legume proportion was about 15% larger in the PK than in the NPK treatments. On the other hand, the botanical composition was similar between the two defoliation treatments at the same N application level, this not only for the proportions of grasses, legumes and forbs, but also with respect to the proportions of the main grass species (*Arrhenatherum elatius*, *Trisetum flavescens*, *Dactylis glomerata*). For instance, the grass proportion was 48 and 50%, and 60 and 61%, respectively, for the two fertilizer treatments at two and respectively three cuts per year. The difference in yield decrease in the 3-cut treatments can therefore not be explained by a loss of productive species in these treatments. Because the cause of the loss of productivity in the 3-cut treatments is neither nutrient depletion nor a degradation of the botanical composition of the meadow, we hypothesize that it is related to interaction effects between climate change and defoliation frequency (Meisser *et al.*, 2013; Batbaatar *et al.*, 2023). In the 2-cut treatments, the average yield of both the July and the September harvests remained similar over the years, while in the 3-cut treatments the yield decrease was observed for all three harvests (Fig. 1b). The yield decrease in the 3-cut treatments therefore seems to be related to a general decrease in vigour, rather than to a short-term effect of the summer cuts. The results of Batbaatar *et al.* (2023) indicate that the negative impact of the combination of defoliation and drought stress might be larger on root than on aboveground biomass production, which could help explain the decrease at all harvests observed in our experiment. Further investigations will be necessary to explain the processes underlying the difference in yield evolution between the two defoliation regimes.

Conclusion

Forty-two years of data from an experiment on a hay meadow indicate that more frequently mown meadows may be less resilient to the current rise in temperature than meadows mown only twice a year, even without clear degradation of their botanical composition.

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Effects of grassland diversity, fertilization, and proportion in the crop rotation on soil health indicators

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Abstract

Integrating grasslands into crop rotations has gained popularity due to positive agronomic impacts. Grasslands are also believed to improve soil properties, such as soil organic carbon (SOC) content and structural stability. However, the impacts of diversified grasslands on SOC and structural stability remain unexplored. Hence, we investigated the effects of grassland mixtures, fertilization, and proportion in the crop rotation on wet stability of aggregates (WSA) and SOC in the plough layer (0–20 cm). Samples were collected from two crop rotations (2 and 4 years of grassland in a six-course rotation), which had two mixtures, i.e. 3-sp (*Lolium perenne* L., *Trifolium repens* L., *Trifolium pratense* L.), and 12-sp (3-sp+5 forbs+4 additional legumes), either unfertilized or receiving 300 kg total-N ha⁻¹ in cattle slurry. Fertilization and increasing grassland proportion significantly increased both WSA and SOC. The more diverse grassland increased WSA, but increase in stability was not related to SOC. Thus, the increase may be related to belowground species complementary, with 12-spp mixture having a larger variety of roots acting as stabilizing agents. Hence, WSA appears to be a sensitive indicator of soil health for assessing management effects, as it changes more rapidly than SOC.

Keywords: grassland leys, organic dairy/crop rotation systems, soil carbon, soil health, soil structural stability

Introduction

In dairy/crop rotation systems, grassland leys play a major role as feed for livestock. Further, the positive impacts of these grassland leys on soil health have also gained popularity in recent years. SOC sequestration is a substantial component of the approaches to climate change mitigation, and studies have shown increased SOC contents after integrating grassland leys (Jensen *et al.*, 2022; Knudsen *et al.*, 2019; Stumpf *et al.*, 2018). SOC, or soil organic matter, is directly related to soil structural stability (i.e., aggregate stability), and several studies (Martin *et al.*, 2020; Puerta *et al.*, 2018) report an increased aggregation when ley pastures are integrated into arable crop rotations. Soil structural stability is directly related to the resistance of soil to the erosion process; therefore, knowledge of soil structural stability is highly important in cropping systems. The species composition and duration of the grassland phase in the rotation vary between farms and, especially in northwestern Europe, farmers apply cattle slurry as an organic N-fertilizer in the grassland phase. These factors may directly influence the SOC and aggregate stability in temperate croplands, and their effects on soil properties are yet to be explored. Therefore, this study aimed to evaluate the impacts of grassland mixtures, organic fertilizer (cattle slurry), and grassland proportion in the crop rotation on SOC concentration and aggregate stability in the plough layer. We hypothesized that increasing cattle slurry application, more diverse grassland mixtures, and increasing the grassland proportion in the crop rotation can increase SOC levels and aggregate stability.

Materials and methods

For this study, soil samples were collected from a long-term field study in Foulumgaard Experimental Farm, Denmark (56°29'N, 9°34'E), representing two crop rotations (2 and 4 years of grassland in a

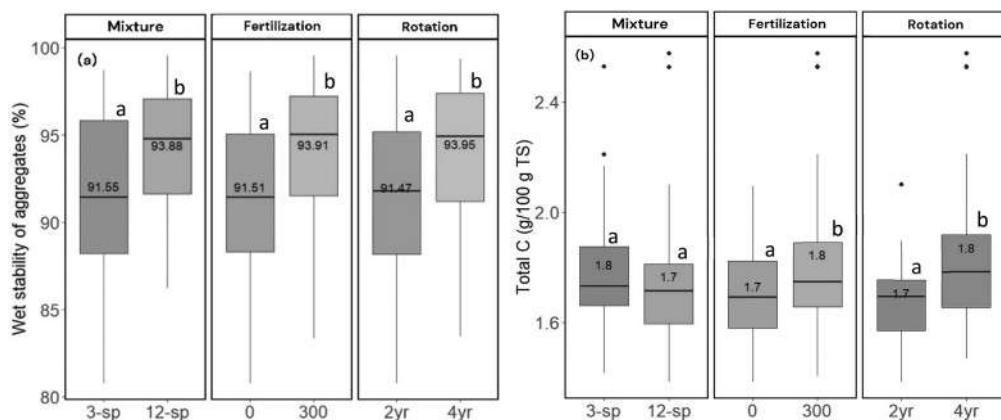


Figure 1. The wet stability of aggregates (WSA) (a) and soil organic carbon content (b) in the plough layer (20 cm) in two crop rotations (2 or 4 years of grassland in a six-course rotation) which in the ley-phase was divided in two grassland mixtures (3- and 12-species) and two fertilization levels (0 and 300 kg total-N ha⁻¹ in cattle slurry). $n = 48$. Lines within the boxes represent median values, box boundaries include the 25th and 75th percentiles, and the whiskers extend from the box boundary to the 10th and 90th percentiles. The mean values are indicated. Letters denote statistical significance at $p < 0.05$.

six-course rotation), two grassland mixtures (3-sp and 12-sp), and unfertilized and fertilized conditions (300 kg total-N ha⁻¹ in cattle slurry). The experiment consists of 12 fields with 2 blocks, each containing four plots. More information about the experimental design is provided in Jensen *et al.* (2025). The 3-sp was a grass-clover mixture consisting of perennial ryegrass, red clover, and white clover, while the 12-sp consisted of the 3-sp mixture with 4 additional legume species (lucerne (*Medicago sativa* L.), birdsfoot trefoil (*Lotus corniculatus* L.), melilot (*Melilotus officinalis* L.) and sainfoin (*Onobrychis viciifolia* Scop.)) and 5 forbs (plantain (*Plantago lanceolata* L.), chicory (*Cichorium intybus* L.), caraway (*Carum carvi* L.), yarrow (*Achillea millefolium* L.) and salad burnet (*Sanguisorba minor* Scop.)). The soil type of the site is Hapludult, and the soil texture is loamy sand (8% clay, 10% silt, 82% sand). The pH of the soil is 5.9. Soil blocks were collected from each plot in the 5–15 cm soil layer, representing the ploughing layer. The first 5 cm was removed to obtain a homogeneous basis for samples across the different crops, and samples were stored fresh in sealed plastic boxes at 2°C until analysis (Qi *et al.*, 2022). A sub-sample (approx. 100 g) from each soil block was air-dried, crumbled, and sieved using a 2 mm sieve, and SOC was quantified using the Vario Max Cube. Another sub-sample (approx. 100 g) was gently broken down by hand through an 8 mm sieve to get a homogeneous basis for the test across the samples. The test for wet stability of aggregates was conducted using Eijkkelkamp's wet sieving apparatus (Rocco *et al.*, 2024). Data analysis was conducted using linear mixed-effect models in the R project software package version 4.3.3 (R Foundation for Statistical Computing). For the analysis, mixture, rotation, and N application rate are considered fixed effects, and field and block are considered random effects, with block nested within field. Two-way and Three-way interactions of fixed effects were assessed, and there were no significant differences ($p < 0.05$); hence, only the main effects were considered in the analysis.

Results and discussion

Figure 1 shows the WSA (Fig. 1a) and SOC (Fig. 1b) in the plough layer (20 cm). Fertilization and increasing the grassland proportion in the crop rotation significantly increased both WSA and SOC. These findings follow the previous studies that reported increments in soil aggregation (Martin *et al.*, 2020; Puerta *et al.*, 2018) and SOC (Jensen *et al.*, 2022) when grassland is integrated into arable crop

rotations. We assume that cattle slurry can influence the binding of organic matter-mineral particle associations, while increasing the grassland proportion in the rotation relates to microaggregate bonding through plant roots and fungal hyphae. Increasing the grassland proportion promotes higher SOC and WSA due to greater input of above- and belowground plant residues, an increased abundance of mesofauna, fungal hyphae, and a denser root network, which bind and stabilize aggregates, making them more resistant to mechanical damage (Jensen *et al.*, 2022; Puerta *et al.*, 2018). Further, roots and hyphae might be persistent since they are replaced annually and remain undisturbed due to the absence of tillage when grassland is grown in the crop rotations. Although more diverse grasslands increased WSA, SOC was not significantly affected. Thus, the increase may be related to belowground species complementary, with 12-sp. mixture having a larger variety of roots acting as stabilizing agents (Puerta *et al.*, 2018), which were not captured in the SOC measurements. Furthermore, the WSA values across all treatments in this study were considered as being very stable (Pulido Moncada *et al.*, 2015). However, no significant interactions were observed between the mixture effects, fertilization, and grassland proportion in the crop rotation.

Conclusion

This study showed that increasing grassland proportion and fertilization increased SOC and aggregate stability, while a more diverse mixture increased the aggregate stability without affecting SOC. Thus, aggregate stability appears to be a sensitive indicator of soil health for assessing management effects, as it changes more rapidly than SOC.

Acknowledgements

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Interpretability of a machine learning model for predicting crude protein in leaves of Tamani grass (*Panicum maximum* cv. BRS Tamani)

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Abstract

Machine learning models have been used for prediction tasks. However, understanding how input variables influence model predictions is equally important. This study aimed (i) to interpret the impact of pre-grazing height (H_{pre}) and post-grazing height (H_{post}), interval between grazing, nitrogen (N) doses, light interception, and weather conditions on the XGBoost used to predict crude protein (CP) in Tamani grass leaves; (ii) compare variable influences under low and high N supply scenarios. The Shapley Additive Explanations (SHAP) approach was applied to assess the magnitude and direction of each variable's contribution to CP prediction. The model revealed that $240 \text{ kg ha}^{-1} \text{ year}^{-1}$ of N positively influenced CP prediction increasing the predicted value, while $80 \text{ kg ha}^{-1} \text{ year}^{-1}$ had a negative effect. Higher H_{pre} , H_{post} , and solar radiation values tended to reduce predicted CP, whereas lower values increased it. Rainfall between 100 and 180 mm enhanced CP content, while levels outside this range decreased it. Under high N input, the influence of climatic factors diminished, whereas under low N, precipitation became the main driver of CP variation. Nitrogen availability modulated the relative importance of management and climatic factors, highlighting adequate N supply as a key strategy to improve crude protein in Tamani grass.

Keywords: algorithms, *Panicum maximum*, pasture management, precision livestock farming, shapley additive explanations

Introduction

Knowledge of forage chemical composition is essential for adjusting management strategies and, consequently, improving animal performance (Rouquette, 2016). However, traditional methods for evaluating forage composition are generally time-consuming and costly (r'Mannetje *et al.*, 2000). New technologies have emerged as promising alternatives to provide rapid and accurate information to support decision-making in pasture management. Among these technologies, predictive modeling based on machine learning (ML) algorithms stands out. Despite their strong predictive performance, machine learning models often exhibit high complexity, which hinders the interpretation of the factors driving specific predictions. In this context, the SHapley Additive exPlanations (SHAP) technique stands out for enabling the understanding of both the strength and direction of the effect of each variable, thus promoting a global interpretation of the model (Lundberg *et al.*, 2018).

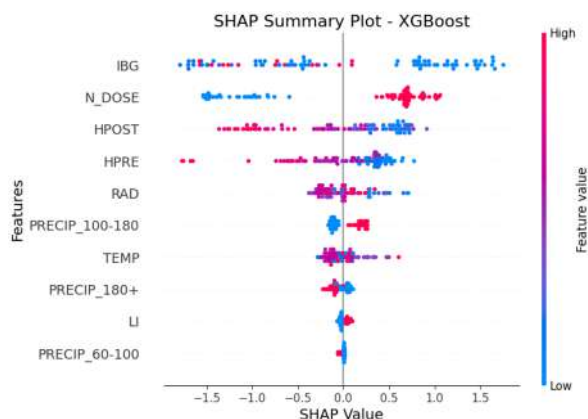


Figure 1. Global interpretation of the XGBoost model using SHAP values. IBG, interval between grazing; HPRE, pre-grazing height; HPOST, post-grazing height; TEMP, temperature; RAD, solar radiation; PRECIP_100–180, precipitation between 100 and 180 mm; PRECIP_180+, precipitation above 180 mm; PRECIP_60–90, precipitation between 60 and 90 mm; LI, light interception. SHAP values on the x-axis indicate the direction and magnitude of their contribution, and colour denotes feature value (pink = high; blue = low).

The pasture ecosystem is a dynamic system influenced by management practices and abiotic factors, including water availability, light, nutrient supply, and temperature. We hypothesized that management factors exert a stronger influence on crude protein content than climatic variables. Therefore, this study aimed to apply the SHAP technique to interpret the impact of climatic and management variables on the XGBoost model in the task of predicting the crude protein (CP) content of leaves of *Panicum maximum* cv. BRS Tamani.

Materials and methods

Data were collected from a pasture area at Embrapa Beef Cattle, comprising 0.96 ha of Tamani grass (*Panicum maximum* cv. BRS Tamani) between December 2020 and April 2022. The dataset was split into 80% for training and validation and 20% for testing, using 5-fold cross-validation with 100 iterations. The SHAP technique was applied to interpret the prediction results (Lundberg *et al.*, 2018). For the SHAP analysis, the following variables were used as inputs in the XGBoost model to predict the crude protein (CP) content in Tamani grass leaves: temperature, solar radiation, precipitation, nitrogen rates (80 and 240 kg ha⁻¹ year⁻¹), light interception (LI) levels (90% and 95%), interval between grazing (IBG), pre-grazing height (Hpre), and post-grazing height (Hpost). To assess the influence of variables under contrasting management conditions, the dataset was further divided into two subsets corresponding to low and high nitrogen application rates (80 and 240 kg ha⁻¹ year⁻¹, respectively).

Results and discussion

The variables are ranked in descending order of importance in the model's predictions (Figure 1). Each point represents an observation from the dataset, with its position along the horizontal axis indicating both the magnitude and direction of that variable's contribution. The colour denotes the feature's magnitude: pink corresponds to higher values (1 for binary variables or the upper range for continuous ones), whereas blue corresponds to lower values. Positive SHAP values indicate a positive contribution, increasing the predicted value, while negative SHAP values reflect a negative contribution, decreasing the prediction. The IBG variable demonstrates that low values can either increase or decrease predictions, depending on their interaction with other features. Positive SHAP values were observed for the

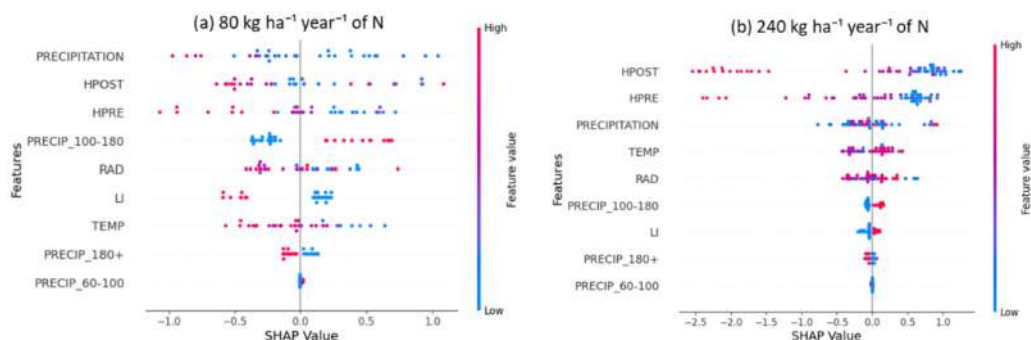


Figure 2. SHAP summary plot for crude protein (CP) prediction split into two datasets: (a) 80 kg N ha⁻¹ year⁻¹ and (b) 240 kg N ha⁻¹ year⁻¹.

highest N dose, increasing the model's prediction, while the lowest dose showed negative SHAP values. High values of H_{post} , H_{pre} , and Rad tend to reduce the predicted CP content, whereas lower values of these variables increase it. Additionally, precipitation in the 100–180 mm range (Precip 100–180 = 1) had a positive effect on the model's prediction (Figure 1).

In pastures receiving 80 kg ha⁻¹ year⁻¹ of N (Figure 2a), precipitation was the primary factor influencing CP content, followed by canopy heights H_{post} and H_{pre} . Among the precipitation classes, only rainfall between 100 and 180 mm and below 180 mm showed distinct positive effects on the model, suggesting that this range favours the accumulation of nitrogen compounds. LI at 90% had a positive impact, whereas LI at 95% negatively affected the model. In contrast, at the rate of 240 kg N ha⁻¹ year⁻¹ (Figure 2b), management heights variables showed greater overall importance, especially H_{post} , followed by H_{pre} . Under this scenario, climatic variables had a lower magnitude of impact on the model's prediction (Figure 2b).

Conclusion

Positive SHAP values were associated with the highest nitrogen dose (240 kg ha⁻¹ year⁻¹), indicating an increase in the model's predictions, while negative SHAP values were observed for the lowest dose (80 kg ha⁻¹ year⁻¹). The SHAP analysis showed that the factors influencing crude protein prediction in Tamani grass vary with nitrogen availability. Under low nitrogen levels, precipitation was the main driver of crude protein variation, suggesting that adequate water availability (100–180 mm) promotes the accumulation of nitrogen compounds in the forage. In contrast, with higher nitrogen supply, management heights, particularly pre- and post-grazing, became the primary determinants of crude protein content, whereas the influence of climatic variables was less pronounced.

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Establishing climate-resilient pasture mixtures by sod-seeding

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Abstract

An on-farm experiment was carried out in Sardinia in the framework of the Operational Group 'Paschinres' to promote the use of permanent pastures and conservation tillage on four sheep farms. Two innovative pasture mixtures (Mix A, Mix B), sown after minimum tillage (MT) and by sod-seeding (SS) on bare soil, were compared to temporary grasslands (TG) typically sown on the farms. Each mixture featured some core species combined with farm-specific species from three functional groups (grasses, G; legumes, L; and herbs, H). Mixtures were sown in autumn 2023 in large fields. One-way ANOVA and LSD test were carried out to assess differences for seedling establishment, forage yields and its botanical composition and forage quality among farms, soil management before sowing and grassland type. G and L established better in Mix A and on farm 1, but no differences were assessed for soil management type and H establishment. On average, Mix B yielded higher than Mix A but similarly to TG. The proportion of functional groups in mixtures was similar for both sowing after MT and SS. SS succeeded in the establishment of biodiverse pasture mixtures featuring seeds of different sizes on soils with variable traits.

Keywords: forage quality, herbs, legume-based mixtures, pasture production, sod-seeding

Introduction

The use of permanent pastures, especially if combined with sustainable and environment-friendly agronomic techniques, would be a turning point for increasing the profitability of Sardinian sheep farms in the medium and long-term, compared to the conventional production system based on annual forage crops and synthetic fertilizer applications. Among the environment-friendly agronomic techniques, sod-seeding and the use of mixtures based on assorted functional groups stand out. Sod-seeding can be carried out with special equipment, on soils with herbage residues or no-tilled bare soil after chemical weeding with desiccants (glyphosate), typically on weeds allowed to germinate after a rainfall of 20–30 mm at the end of summer. Pastures featuring a variety of species, functional groups and plant reproductive strategies are expected to exhibit higher performances than monocultures, because of the complementarity of adaptation traits, symbiotic relationships, and mutual benefits (Blaix *et al.*, 2023). However, many farmers lack the knowledge to successfully adopt these techniques, especially with regards of weed control and timing of management interventions. The use of innovative mixtures and sod-seeding was implemented in an innovation programme aimed at reducing variable costs of milk production and improving the quality of products in four dairy-sheep farms, which were partners of the Operational Group 'Paschinres'. This paper describes the agronomic performances of two mixtures, based on three plant functional types, both sown after minimum tillage and by sod-seeding, compared to the temporary grasslands.

Materials and methods

Two innovative mixtures (Mix A, Mix B) were sown in farm fields (n. 4 farms×2 ha) both with sod-seeding (SS, n. 4 farms×1 ha) and minimum tillage (MT, n. 4 farms×1 ha) in October 2023 to be compared to farm temporary grasslands (TG), i.e. barley in pure stand sown after soil MT in the same

month. Chemical weeding (using glyphosate, 0.5 l ha^{-1}) preceded sod-seeding of mixtures, carried out with a sod-seeder which cut furrows (1 cm depth), where seeds were settled, by means of discs. MT consisted in a soil scarification with chisels followed by affination with hoes (8–15 cm) which was followed by seeding carried out with a seed drill. Each mixture featured some core species from three functional groups (grasses, G; legumes, L; and herbs, H), namely *Lolium rigidum* (15% weight/total weight), *Medicago polymorpha* (20%), *Plantago lanceolata* (15%) and *Cichorium intybus* (15%) in Mix A and *Hordeum vulgare* (10%), *T. spumosum* (15%), *T. michelianum* (15%), *P. lanceolata* (15%) and *C. intybus* (15%) in Mix B. Farm-specific legume species were added (i.e. *T. subterraneum*, *T. resupinatum*, *Sulla coronaria*) so that the total legume seed ratio was 55% and 60% for Mix A and Mix B, respectively. Seeds rates were 35, 52 and 120 kg ha^{-1} for Mix A, Mix B, and TG. Fertiliser DAP (150 kg ha^{-1}) was spread by broadcast application in all treatments. Seedling establishment and re-establishment was recorded on twenty quadrats ha^{-1} ($25 \times 25 \text{ cm}$). Forage DMV values were estimated from sampling under six exclusion cages ($100 \times 100 \text{ cm}$) per ha in spring 2024. Subsamples were dried at 80°C for 72 h and weighted. Forage Crude Protein (CP), Neutral Detergent Fibre (NDF), Acid Detergent Fibre (ADF) and Lignin (ADL) were determined according to Van Soest *et al.* (1991) procedure.

One-way ANOVA and LSD test were carried out to assess differences for DMVs, botanical composition, and establishment rates among farms, pastures and tillage type. SS mixtures did not establish in farm 3 and they were excluded from the statistical analysis for this treatment.

Results and discussion

Rains fell late after summer 2023, which delayed sowing to late autumn. Winter was mild and dry and favoured seedling establishment, but at the same time it caused the shortening of the vegetative phase. G and L seedlings established better in Mix A and on farm 1, while no differences were assessed for H establishment at the beginning of winter and between tillage/sowing type. In fact, herbs completed their establishment late in spring, with chicory performing better than plantain in all farms.

Forage yields varied greatly among farms, mixtures and, to a lesser extent, between soil management techniques (Figure 1). Farm DMV values ranged from 2.3 to 4.2 t ha^{-1} . On average, Mix B yielded higher than Mix A but not differently from TG. The soil management before seeding did not affect significantly the mixture DMV values, despite the SS-sown pastures showing slightly higher production. The incidence of weeds was nearly the same in all types of pastures.

Forage quality analyses highlighted the good protein content, especially in Mix A, due to its higher presence of legumes (Table 1). In Mix B, protein content was similar to that of TG. Medium-high values were observed for NDF and ADF, with no substantial qualitative differences between the two mixtures. ADL content was higher in Mix A, probably due to the greater proportion of spontaneous species and legumes that lignified early in the season. Soil management before seeding did not significantly influence any of the quality parameters.

Conclusion

The establishment year is critical for perennial mixtures to achieve a long-lasting and productive pasture in the following years. In this trial, the use of two mixtures allowed us to assess that Mix B showed better agronomic performance compared to Mix A. Nonetheless, Mix A showed a more balanced botanical composition and a better forage quality than Mix B, that could make it preferable for farms seeking pasture quality improvement. In fact, the vigorous growth of the cereal component in

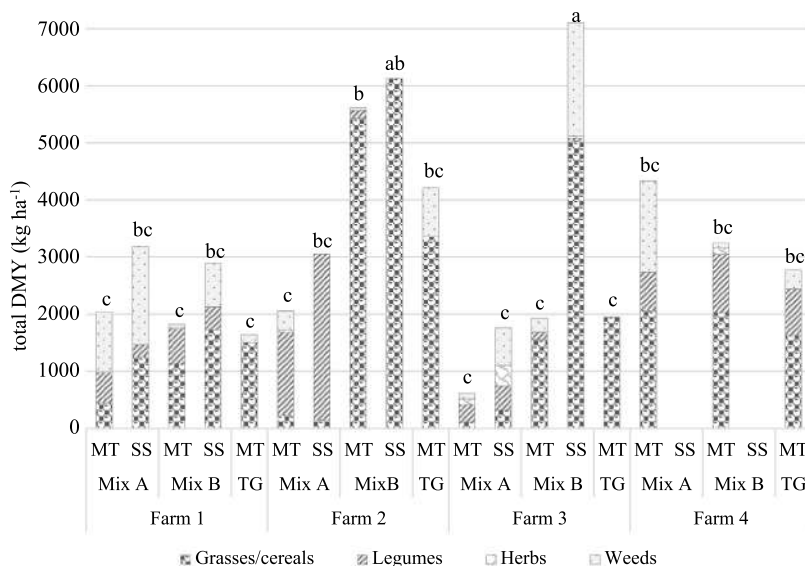


Figure 1. Forage production (DMY) in spring 2024 of different forage resource (Mix A, Mix B and temporary grasslands, TG), sowing technique (MT, minimum tillage; SS, sod-seeding) and farms, and contribution of functional groups (grasses, legumes and herbs) and unsown species to mixtures and temporary grasslands. Different letters indicate significant statistical differences among values ($p < 0.05$).

Table 1. Average forage quality of mixtures (mix A, mix B) and temporary grassland (TG) in spring 2024.

	Crude protein (% DM)	NDF (% DM)	ADF (% DM)	ADL (% DM)
Mix A MT	20.0 ^(a)	50.3	33.8	12.7 ^(a)
Mix B MT	13.8 ^(b)	51.6	31.1	3.4 ^(b)
Mix A SS	18.1 ^(a)	47.2	33.7	11.6 ^(a)
Mix B SS	10.8 ^(b)	54.2	34.9	3.9 ^(b)
TG MT	12.4 ^(b)	49.7	32.9	3.3 ^(b)

Temporary grasslands were sown after minimum tillage (MT), while mixtures also by sod-seeding (SS). Different letters indicate significant statistical differences among values ($p < 0.05$).

the early stages in Mix B affected negatively the development of the legume component, increasing the similarities of the mixture with temporary grassland. Sod-seeding proved to be effective in three out of four farms, where the operational aspects that might have hindered the satisfactory establishment were successfully faced (e.g., timing of intervention, soil conditions, etc.) thanks to the knowledge and experience of the farmers about their farm soil characteristics.

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Precision agriculture to assess winter-damaged grasslands in northern Sweden

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Abstract

Forage production in Sweden is primarily based on grasslands, specifically ley systems (harvested forage as part of a crop rotation). Winter survival is a critical aspect of these systems, and climate fluctuations and species differences further affect their resilience. Remote sensing tools such as low-cost unmanned aerial Vehicles (UAVs) with RGB cameras can be used to monitor crop growth processes relevant to assessing winter damage, such as mapping stand density. In this context, this study aimed to use a UAV-RGB to develop a practical framework to assess grassland swards damaged during winter. We flew a UAV over a grassland field during the spring season of 2025 at an altitude of 20 m (Ground Sampling Distance (GSD) of 0.93 cm/pixel). A U-Net Deep Learning (DL) model was used to classify the damaged areas. The input dataset consisted of 100 subset images (128 × 128 pixels; 80% for training and 20% for validation), generated by a four-fold data augmentation. Preliminary results show the potential of the DL model to precisely map winter-damaged patches in ley fields (overall accuracy = 0.90, recall = 0.90, and F1 score = 0.95 in the validation dataset). Further analyses using an extended and multi-temporal dataset are under development.

Keywords: deep learning, ley system, remote sensing, swards, winter kill

Introduction

Forages are essential in Swedish agriculture, covering a high proportion of the arable land, especially in northern Sweden, where perennial forage crops occupy around 80% of the agricultural landscape. Livestock production is based on grasslands, specifically ley systems (“vall”; harvested forage as part of a crop rotation). Leys are vulnerable to winter kill—damage generally caused by fluctuating temperatures, variable snow cover, ice encasement, and pathogens. This damage typically occurs in irregular patches, making visual assessment and management decisions, such as resowing, renovation, or weed control, challenging. Integrating precision agriculture tools for field monitoring and re-sowing these patches with fast-growing forage mix species offers potential for rapid assessment and recovery.

Recent advances in remote sensing, particularly UAV imaging, enable high-resolution field-level data with rapid revisit times and high-resolution assessment of vegetation parameters.

Low-cost UAV-RGB can be attractive for farmers due to their affordability and flexibility. When combined with artificial intelligence, RGB imagery could support agricultural tasks relevant to winter kill monitoring, including stand density estimation (Wang *et al.*, 2022) and weed detection (Anderegg *et al.*, 2023). In this context, this study aimed to use a low-cost UAV-RGB and a DL model to map winter-damaged grasslands in northern Sweden.

Table 1. Overall accuracy, recall and F1 score metrics of the DL model for the calibration and validation dataset

Dataset	Overall accuracy	Recall	F1 score
Training	0.94	0.99	0.97
Validation	0.90	0.90	0.95

Materials and methods

UAV flights were performed over ley fields of a farm in Umeå, Sweden, during early spring (after snow melt and early re-growth) in 2025. In this study we show only the images over one field taken on 25 May 2025. This field was partially sprayed with herbicides in autumn 2024 to mimic winter-damaged areas, as part of a field experiment in the same research project (more details available at <https://www.slu.se/en/research/research-catalogue/projekt/v/vall-reparera/>).

High-resolution RGB imagery was captured using a DJI Mavic 3 UAV at 20 m above-ground altitude (GSD of 0.93 cm/pixel), with 70 and 80% of side and frontal overlapping, respectively. The acquired images were processed using Pix4Dmapper software to generate an orthomosaic image. The orthomosaic (1227×1215 pixels) was split into small subset images (128×128 pixels), and a selection of these subsets (25 images; input dataset) was used to create label masks, where value = 1 represents green vegetation ($\approx$ no damage) and value = 0 represents no green vegetation ($\approx$ damage). The labels were created and visually checked using semi-automated annotations with the Segment Anything Model (SAM-2) assistance in the DigitalSreeni Image Annotator tool (Bhattiprolu, 2024). The input dataset was divided into training (80%) and validation (20%). Data augmentation techniques, such as geometric (image flipping and rotating) and spectral manipulation (brightness, saturation and contrast) were applied to the training dataset to create new synthetic images and increase the input data (by a factor of 4, totalling 100 images).

We implemented a U-Net DL architecture (Ronneberger *et al.*, 2015) incorporating a pre-trained VGG16 Convolutional Neural Network (CNN) (Simonyan and Zisserman, 2014) as the contracting path for feature extraction. The VGG16, which was originally trained on the ImageNet dataset, consists of sequential convolutional and max-pooling layers. The U-Net design used two convolutional and two up-sampling blocks, as suggested in Knoth (2020). The classification quality was assessed by the overall accuracy, the recall, and the F1 score metrics for both the training and validation datasets.

Results and discussion

Table 1 shows the overall accuracy, recall and F1 score metrics for the validation and training datasets. All the metrics indicated a good performance of the DL model (values close to 1), indicating that it could be an approach to classify winter damage in grassland swards. The metrics were higher for the calibration dataset, but the model showed a relatively consistent performance when applied to the validation dataset, and no clear overfitting pattern was identified, suggesting that the model should be stable when generalising the results. However, these are still preliminary results, and the small input dataset used to calibrate the model might not be enough to drive final conclusions about the modelling performance. A more comprehensive and extended evaluation using a large input dataset is under development. Moreover, the generalisation of the models would be better analysed once we test them on an independent UAV image (testing dataset).

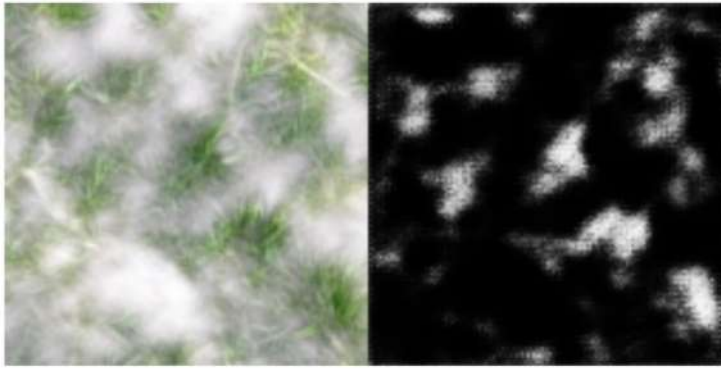


Figure 1. Example of an RGB image subset from the validation dataset (left) and the classified image when applying the DL model (right). In the left image, the green pixels represent the green vegetation, and the others represent no green vegetation ($\approx$ damage). Subsequently, the white pixels in the right image represent the areas classified as no damage, and the black ones classified as damage.

Figure 1 shows an example of one RGB image subset from the validation dataset and the no-damage/damage classified image performed using the model on the same image. It visually illustrates the model performance in identifying pixels that were more prone not to have green vegetation (black pixels, right image), which were then considered damaged. This classification approach is a proxy of the damage, since it does not consider areas that might not have green vegetation due to other reasons (such as machinery paths). A post-processing approach will be performed to better account for other spatial factors. Moreover, we also did not consider damaged areas covered with weeds. One of the reasons was the time of the UAV flight, during which no weeds were detected. A next step in this study will be to consider flights taken at several fields and multiple times over the spring and early summer, to account for the weeds and, consequently, identify when to get UAV imagery to assess damage before weeds are visible.

Conclusion

The preliminary results demonstrate that a low-cost UAV-RGB combined with a U-Net DL model can accurately map non-green patches in ley fields (overall accuracy = 0.90, validation data). Although the approach shows potential for practical winter-damage monitoring in grasslands, the small initial dataset and the lack of an independent testing dataset limit robust conclusions. Moreover, it currently does not differentiate for other damage sources (e.g., machinery paths) or weed-covered areas, reinforcing the need for post-processing steps. Further work with larger and multi-temporal datasets is under development to better refine and generalise this framework.

Acknowledgement

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Mapping small-scale variations in grassland productivity by remote sensing to support pasture management in mountain areas

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Abstract

Mountain pastures are characterised by strong heterogeneity, marked topography, and large expanses, which make them difficult to monitor from the ground. Hence, there is great potential for remote sensing to facilitate the integration of information on growth conditions and phenology over time. In a case study, we analysed a time series of high-resolution (10-m) Sentinel-2 images from 2016 to 2024 for an alpine valley in eastern Switzerland. Based on all available cloud-free images, we calculated the area under the curve of the Enhanced Vegetation Index (EVI) as an annual growth indicator and related it to weather data. The analysis showed a good agreement of the indicator with summer weather conditions (bias of -6.8 to $+1\%$ on the average biomass indicator) but a considerable small-scale variability due to local site conditions, up to more than $\pm 50\%$ of biomass indicator values with respect to the considered 8-year average. The results can be used to identify vulnerability levels along the elevation profile or to map zones where growth varies the most from year to year as a function of local weather variations. In these zones, pasture management will need extra flexibility measures and real-time monitoring to adapt to annual fluctuations of a future climate change.

Keywords: annual variation, EVI, growth pasture, remote sensing, Sentinel-2

Introduction

Pastures are a major land use in mountain areas world-wide. In Switzerland, they make up one quarter of the country's surface and are characterized by strong heterogeneity, marked gradients and undulating topography (Lüscher *et al.*, 2019). They are grazed by ruminant livestock and provide important ecosystem services to society (Pauler *et al.*, 2024).

Another feature of mountain areas are highly fluctuating weather conditions, both between and within years. Weather conditions affect pasture yields and herbage quality as well as other ecosystem services. These variations conflict with fixed carrying capacities attributed to each mountain pasture farm by authorities, which need to be met to obtain full support payments for grazing (Meyer *et al.*, 2025). In productive years, more biomass is available than can be grazed by the allowed number of animals, but in poor years, it is hard to fulfil the asked grazing days and exceptions must be applied for. Since 2016, the European Space Agency (ESA) program Sentinel-2 has been allowing a high (10-m) resolution observation of the Earth by means of satellite images. The reflected light in the frequency range of the near-infrared, detected from satellite, is indicative of living vegetation. We use this signal to evaluate the annual dynamics of pasture growth in an alpine region of Switzerland (Maighels valley, Grisons), in particular the seasonal biomass presence variations. This provides a scientific basis for the development of a more flexible pasture management system that adapt the carrying capacity of mountain pasture farms to annual weather fluctuations.

Materials and methods

Satellite images (ESA Sentinel-2) at 10-m space resolution were acquired on a 5-day basis, filtered for cloud-coverage less than 30%, and further masked for cloudy and contaminated pixels using the Sentinel-2 Scene Classification Layer. Based on the difference of the reflected-light intensity in the near-infrared and red frequency bands, the Enhanced Vegetation Index (EVI) was computed. Light in these frequency bands is reflected or absorbed by photosynthetic vegetation, respectively, determining EVI values from 0 to 1 for increasing standing biomass density. As shown by previous studies in alpine regions (e.g. Bayle *et al.* 2019), it is possible to track vegetation growth by computing vegetation indices on regularly acquired images over the year. EVI was chosen here because it saturated less quickly than the closely related Normalized Difference Vegetation Index (NDVI) and detected a more differentiated growth evolution in spring and autumn. In Oriani *et al.* (2025) the area under the NDVI curve (AUC) was used as an indicator of biomass presence during the entire season for different pasture habitats. Similarly, we computed here the AUC on EVI, but for single pixels, to obtain high-resolution maps of biomass presence. Annual anomalies were computed pixel-wise as deviations from the corresponding 2016–2023 averages and displayed in maps. For comparison, monthly weather data was derived from monthly climate grids of MeteoSchweiz (TabsM and RhiresM products). The Maighels study area is located in the central Swiss Alps (46.6303, 8.6883), oriented towards north, covered by 8.9 km² of mountain pastures at elevations of 1700–2400 m asl., and grazed by 260 heifers during summer.

Results and discussion

The aggregated EVI AUC for the entire test area varied considerably between years, with average values of –6.8%, –2.3% and +1.0% in 2016, 2018 and 2020, respectively, relative to the average for 2016–2023. Variation among years for individual pixels within the area was also large. Across all three test years, values in 95% of all pixels varied between –33.4% and +37.5% relative to the 8-year average.

The spring of 2016 was characterised by above-average precipitation and temperatures slightly below average (Figure 1). This resulted in values of the biomass productivity indicator below average for a substantial part of the test area (average deviation of –6.8). Nevertheless, local positive deviations are shown (green areas), suggesting the need of targeted adaptation of the grazing activity. The summer of 2018 was hotter and drier than average, with pixel values weakly deviating from the 8-year average (–2.3% on average). These local deviations were tightly interspersed, likely due to diverse water storage capacities of the terrain. Conversely, in 2020, spring was warmer and drier than average and a large area in the lower part of the valley (top of the map) had biomass productivity indicator values above the average (up to +50%). In contrast, values were below average in the upper of the valley (down to –50%, bottom of the map).

The data show that values of the EVI AUC deviations are fine-grained. Zones with values above and below average are located close to each other. This means that forage availability varies considerably over short distances. In cases where there is forage shortage in one zone, animals can potentially find forage nearby. To make use of this forage, it is essential that pasture management can be adjusted in a flexible way, allowing faster or slower movement of livestock across the landscape and/or employing mobile fencing systems.

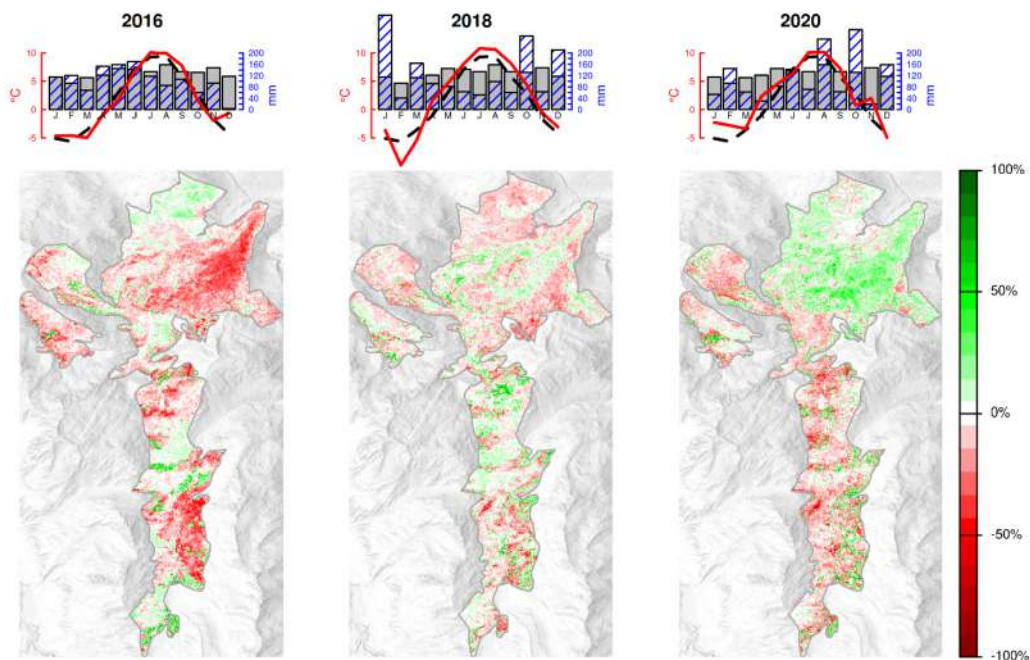


Figure 1. Top row: Monthly precipitation and temperature in the test area in the years 2016, 2018 and 2020 (blue bars and red line) compared with the 1991–2020 long-term average (grey bars and black line). Bottom row: Annual percentage deviation from the average biomass productivity indicator for the years 2016–2023 based on Sentinel-2 images. Green areas indicate positive deviations, while red areas indicate negative deviations.

Conclusion

This study shows that in mountain areas, biomass productivity can vary substantially across space and between years. Areas with below-average forage availability are neighboured by areas with above-average forage availability over relatively short distances. The use of high-resolution remote-sensing imagery provides means to detect these variations. The information can support herdsmen in their efforts to balance forage shortage within a grazing area by a flexible allocation of pastureland to livestock. Future research developments could benefit from high-resolution climate data, if available, to better analyse the statistical relations with local biomass anomalies.

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Effects of biochar on productivity and community structure in a legume-rich pasture in Portugal

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Abstract

Biochar has gained attention for its potential to improve soil properties and enhance plant productivity, yet its effects in grasslands remain underexplored. This study reports on a large-scale field experiment in a legume-rich pasture in Portugal, assessing the impacts of biochar on plant productivity and community structure under ambient (100%) and reduced (50%) precipitation, over the course of two growing seasons. Biochar resulted in a boost in productivity, increasing aboveground biomass by approx. 50%. Legume species benefitted more from biochar than the other functional groups. As a result, biochar altered the community structure. In the first season biochar significantly increased legume abundance at the expense of the forbs, while in the second season it was at the expense of the grasses. The productivity boost and changes in community structure with biochar amendment resulted from modifications to soil biological and physicochemical properties, enhancing nutrient availability. Precipitation manipulation had little to no impact on productivity, community structure and soil parameters, as water availability was not a limiting factor for plant growth. Results indicate that biochar amendment can represent a win-win scenario for pastures, contributing towards securing productivity and livestock nutrition as well as overall ecosystem health and resilience to global climate change.

Keywords: community composition, Mediterranean grasslands, rainout shelter, *Trifolium*

Introduction

Biochar amendment can enhance soil properties such as pH, cation exchange capacity, water retention and microbial activity, which in turn have the potential to enhance plant productivity (Jeffery *et al.*, 2024). However, its effects vary with soil type, biochar characteristics and environmental conditions (Dai *et al.*, 2020). While extensively studied in croplands, the effects of biochar amendment in grasslands remains underexplored. In Portugal, Sown Biodiverse Pastures (SBP)—legume-rich permanent pastures—are highly productive but vulnerable to climate change, particularly reduced precipitation, emphasizing the need for adaptation strategies. This study presents a large-scale manipulation experiment, investigating the impacts of biochar amendment on plant productivity and community structure in SBP under ambient (100%) and reduced (50%) precipitation, over the course of two growing seasons. We hypothesize that biochar increases plant productivity and that the sponge function of biochar will compensate for a reduction in precipitation, upholding systems productivity under scenarios of climate change.

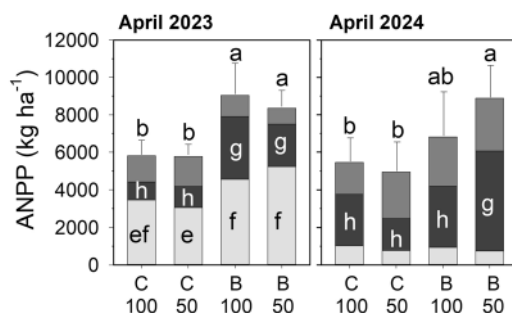


Figure 1. ANPP in the four treatments at the peak of the growing season in 2023 and 2024 (■ grasses, ■ legumes, ■ forbs). Data represents mean $\pm$ SD, $n = 5$. Different letters indicate significantly different means (Tukey's HSD post-hoc, $p < 0.05$) between treatments at each harvest.

Materials and methods

The study was conducted at Quinta da França farm near Covilhã, Portugal, on a sandy loam dystic Cambisol. The region has a Mediterranean climate, with wet winters and hot, dry summers. Long-term average annual precipitation is 858 mm, with 90% of the precipitation being confined to the October–May period. The experiment consisted of a balanced factorial design with two factors: biochar amendment and precipitation, resulting in four treatments: (1) C100: control, 100% precipitation, (2) B100: biochar addition, 100% precipitation, (3) C50: control, 50% precipitation, and (4) B50: biochar addition, 50% precipitation. Each treatment was replicated five times, and each experimental plot was 3 m $\times$ 3 m. In October 2022, biochar (4%, w/w), produced from pyrolysis (700°C) of untreated woodchips (Novocarbo, Hamburg, Germany) was mixed into the topsoil, and the pasture was sown with a legume-rich seed mixture. In November 2022, stationary rainout shelters were constructed for partial interception of rainfall. Aboveground net primary productivity (ANPP) was assessed in January, March, April and May of 2023 and 2024, by harvesting all plant material in a quadrat in each experimental plot, subsequently sorted into three functional groups (grasses, legumes, forbs), then oven-dried and weighed. Soil and plant nutrient analyses were performed using standard laboratory techniques, and meteorological conditions and soil moisture were monitored continuously. Statistical analyses included ANOVAs to evaluate treatment effects on aboveground biomass, relative abundance of each functional group, soil parameters, and tissue nutrients.

Results and discussion

In both growing seasons, precipitation manipulation did not have a significant effect on ANPP (Fig. 1), as water availability was not the limiting factor for plant growth. However, biochar amendment led to a significant increase ($p < 0.001$) in ANPP in April of approx. 50% in both growing seasons, in agreement with our first hypothesis. These biomass increases are much higher than the values quoted in meta-analyses on plant productivity with biochar amendment, i.e., approx. 10–15% (Jeffery *et al.*, 2024).

The productivity boost with biochar may be due to an improved water holding capacity of the soil. Our data indeed indicates that biochar did increase soil moisture (by 24–29%), thereby compensating for the reduction in precipitation, in agreement with our second hypothesis. However, the soil moisture increase with biochar was not responsible for the observed increase in productivity, as comparison of the biochar plots receiving reduced precipitation (B50) with the control plots receiving ambient precipitation (C100) revealed a 44–63% increase in ANPP in the B50 plots, although soil moisture

in B50 and C100 was not significantly different. The productivity boost with biochar in our grassland was most likely the result of a combination of the liming and fertilizer-like effect of biochar. Our results indicate that the addition of biochar led to a significantly higher pH ($p < 0.001$), which in turn may have increased nutrient availability in the soil. Indeed, biochar amendment significantly ($p < 0.001$) increased herbage nutrient accumulation per-square metre (68, 60 and 90% for nitrogen, phosphorus and potassium, respectively). These large increases are evidence that biochar led to enhanced nutrient availability, and supports the idea that biochar had a fertilizer-like effect.

High variability in species responses to biochar has been reported in many studies (e.g. Jeffery *et al.*, 2024). In our grassland, the biomass of all the three functional groups was significantly affected by biochar (Fig. 1), with the largest growth responses to biochar being recorded in legumes. In April 2023, biochar increased grass and legume biomass by 51 and 167%, respectively. In contrast, forb biomass decreased by 33% in the biochar plots. In April 2024, biochar increased legume and forb biomass by 116 and 30%, respectively, while grass biomass tended to decrease with biochar addition.

To date, little is known about how biochar affects plant community composition, especially in grasslands, probably due to the difficulties associated with integrating biochar incorporation into the soil with normal agricultural practices. In our grassland, biochar altered the community structure, the latter expressed as the relative abundance of each of the three functional groups. In both growing seasons, biochar led to a significant increase in legume abundance. In the first growing season, this was at the expense of the forb species, while continuing legume supremacy decreased the abundance of grasses in the second growing season. Thus, our results indicate that legumes benefitted disproportionately from the biochar-induced changes in soil parameters, driving shifts in community composition. The competitive advantage of legumes with biochar in our grassland is most likely the result of the increase in soil pH and potassium availability. These changes may particularly have been beneficial to the legumes, as several studies have reported enhanced biological nitrogen fixation in legumes following biochar application due to a higher pH (Farhangi-Abriz *et al.*, 2024) and/or increasing potassium availability (Oram *et al.*, 2014), subsequently increasing legume growth. This biochar-induced higher productivity of legumes may have increased nitrogen-input, providing another reason for the observed large positive effects of biochar on system productivity.

Conclusion

Our results indicate that amending soils with biochar increases growth of pasture herbage through improved soil characteristics. With increasing pressures from intensive land management, and climate change threatening the capacity of Mediterranean pastoral systems to maintain productivity for livestock, biochar amendment can represent a win-win scenario for pastures, contributing towards securing herbage productivity and, simultaneously, to livestock nutrition, as well as overall ecosystem health and resilience to global climate change. Long-term, field-scale, in-situ biochar experiments are needed to further investigate the long-term behaviour of biochar in soil and the possibility of a moisture-related mechanism of biochar, especially when vegetation experiences severe drought conditions.

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Nitrogen fixation and weed suppression in legume–grass–herb mixtures

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Abstract

Biological nitrogen fixation (BNF) by legumes offers a sustainable alternative to synthetic nitrogen in grasslands. This study examined how legume inclusion and functional group diversity affects yield, BNF, and weed suppression in multispecies grasslands. A field experiment in Denmark tested red (*Trifolium pratense* L.) and white (*Trifolium repens* L.) clovers in monocultures and in combinations with two grasses and two herbs with increasing diversity. Across five harvests, BNF was assessed using the ^{15}N isotope dilution method. Mixtures showed the highest proportion of nitrogen derived from atmospheric fixation (86–96 %Nd_{fa}), significantly exceeding monocultures (<75% Nd_{fa}). Legume contribution to total dry matter declined with increasing diversity (41–76% in monocultures vs. 9–23% in mixtures), limiting nitrogen inputs despite enhanced fixation efficiency. Accordingly, total nitrogen fixed was greatest in red clover monocultures due to higher legume biomass, despite lower %Nd_{fa}. Weeds significantly reduced net yields by 24–59% in legume monocultures, compared to 0.3–6% in mixtures. These results indicate that monocultures offer high nitrogen fixation potential but are vulnerable to weed pressure, while diverse mixtures provide stable yields and effective weed suppression, offering practical guidance for designing grasslands that balance productivity, nitrogen input, and long-term resilience.

Keywords: biological nitrogen fixation, monocultures vs. mixtures trade-offs, plant functional diversity, resilience, weed suppression

Introduction

Grassland systems are recognized for their potential to deliver resilient and sustainable forage production under variable environmental conditions. A key driver of this resilience is the diversity of plant functional groups, which can enhance resource use efficiency, productivity, and ecological stability (Jensen *et al.*, 2025). Legumes play a pivotal role through their capacity for biological nitrogen fixation (BNF), contributing to soil fertility and supporting biomass production in mixed swards. However, the performance of legumes and their nitrogen-fixing function can vary depending on species composition and mixture complexity (Rasmussen *et al.*, 2012). This study investigates the effects of increasing functional group diversity, ranging from monocultures to six-species mixtures on aboveground biomass, and the percentage of nitrogen derived from the atmosphere (%Nd_{fa}) and total nitrogen fixed by legume species. Additionally, we assess weed presence across treatments, as weed suppression is a critical ecosystem service often enhanced by diverse plant communities. Our findings aim to inform the design of multifunctional grassland systems that optimize productivity, nitrogen inputs, and ecological resilience.

Materials and methods

This study was conducted in 2023–2024 at Aarhus University's experimental field in Foulum, Denmark (9°34'E, 56°29'N) as part of the LegumeLegacy project, which identified the benefits of multi-species grassland leys across three functional groups: grasses (timothy (*Phleum pratense*) and ryegrass

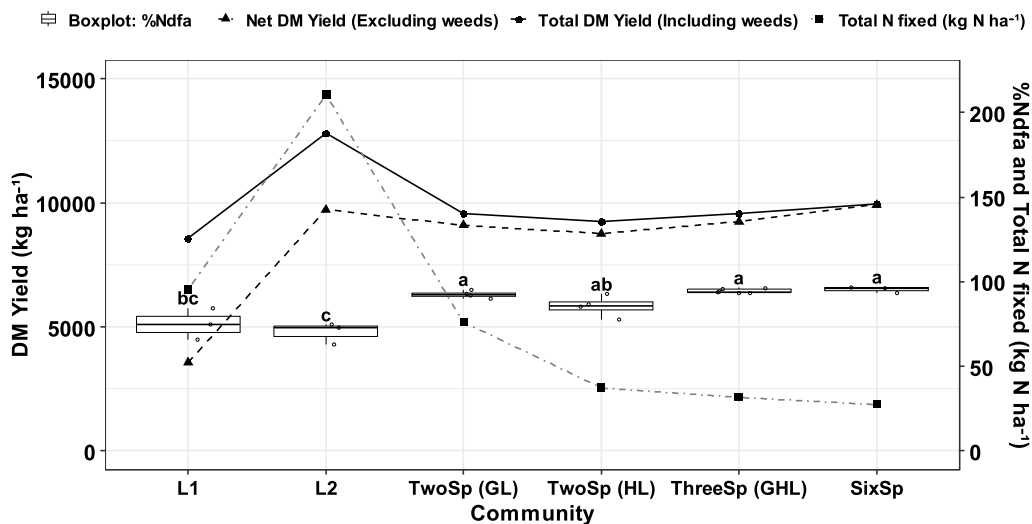


Figure 1. The yield, %Ndfa and total nitrogen fixed in legume monocultures vs. grass-herb-legume mixtures. Community: L1-white clover; L2-red clover; TwoSp (GL)/(HL)-2-species mixture with grass/herb and legume; ThreeSp (GHL)-3-species mixture with grass-herb-legume; SixSp-6-species mixture with two grasses, two herbs and two legumes. Different letters indicate significant differences in %Ndfa between communities at $p \leq 0.05$.

(*Lolium perenne*)), legumes (red and white clover), and herbs (chicory (*Cichorium intybus*) and plantain (*Plantago lanceolata*)) with varying species proportions based on a simplex design (Kirwan *et al.*, 2009). To analyse the impact of species composition on nitrogen fixation, this study only used a subset of all plots containing at least one legume. All plots of 3×9 m were sown in April 2023 at locally recommended densities and fertilized with 150 kg N ha^{-1} in 2023 and 174 kg N ha^{-1} in 2024 (as NH_4NO_3), along with 30 kg P ha^{-1} , and 158 kg K ha^{-1} each year. Given the poor legume establishment, legumes were resown in all plots in August 2023. BNF was assessed using the ^{15}N isotope dilution method (Rasmussen *et al.*, 2012). In February 2024, ^{15}N -labeled ammonium sulfate, enriched to 98% atom was applied to 1×1 m subplots. Subsequently, biomass was harvested five times from 0.5×0.5 m quadrats within each ^{15}N -labeled subplot, sorted by species (including weeds), dried at 60°C for 48 h, and weighed. Subsequently, the ^{15}N enrichment was analysed at UC Davis Stable Isotope Facility using continuous flow isotope rate mass spectrometry after combustion to N_2 gas at 1000°C in an on-line elemental analyser. The %Ndfa was calculated based on excess ^{15}N in legumes relative to non-fixing reference species (McNeill *et al.*, 1994) using the following equation:

$$\% \text{Ndfa} = 1 - \left(\frac{\text{excess atom\%}^{15}\text{N legume (labeled)} - \text{excess atom\%}^{15}\text{N legume (unlabeled)}}{\text{excess atom\%}^{15}\text{N reference (labeled)} - \text{excess atom\%}^{15}\text{N reference (unlabeled)}} \right) \times 100$$

Biomass, total N fixed and %Ndfa data were analyzed in R using ANOVA followed by pairwise comparisons with Tukey's HSD test at $p \leq 0.05$.

Results and discussion

The %Ndfa, total nitrogen fixed, and dry matter (DM) yield varied significantly across plant community compositions (Figure 1).

The highest %Ndfa was observed in the diverse mixtures: Sixsp (95.5%), ThreeSp (GHL, 94.4%) and TwoSp (GL, 92.4%). TwoSp (HL) mixture had a slightly lower %Ndfa (85.5%) though not statistically different from the other mixtures at $p \leq 0.05$. By contrast, legume monocultures: white clover (74.9%) and red clover (70.1%) had significantly lower %Ndfa ($p \leq 0.05$). These results suggest that legumes fix nitrogen more efficiently when grown with other functional groups, likely due to competitive stimulation and reduced soil nitrogen availability. Although all communities were fertilized to control for management effects, which can suppress BNF (Cabeza *et al.*, 2024), the presence of grasses and herbs in mixtures may have mitigated this effect by creating localized nitrogen-limited conditions that promote fixation. Thus, fertilization may have inadvertently reduced the nitrogen contribution from legumes in monocultures, where fertilization might not have been necessary. Despite this, red clover still achieved the highest legume DM yield (9,731 kg ha⁻¹) and total nitrogen fixed (211 kg N ha⁻¹), followed by white clover (3555 kg ha⁻¹ and 95.2 kg N ha⁻¹ fixed). In contrast, mixtures such as GHL and the six-species mixture had much lower legume biomass (1,108 and 892 kg ha⁻¹, respectively), resulting in 31.7 and 27 kg N ha⁻¹ fixed, respectively, despite their higher %Ndfa values. The %Ndfa reflects the proportion of nitrogen derived from atmospheric fixation, but not the total amount fixed. The total nitrogen fixed is a function of both %Ndfa and legume biomass, which was substantially reduced in the more diverse mixtures. This suggests that while facilitative interactions in mixtures (e.g., reduced soil N availability due to grasses and herbs) may enhance fixation efficiency, the overall contribution of legumes to system-level nitrogen inputs is constrained by their lower biomass in these communities. Moreover, the narrowing gap between total and net yields in diverse mixtures (Figure 1) further supports the role of diversity in reducing weed pressure, consistent with reports by Jensen *et al.* (2025), which demonstrate that species-rich mixtures enhance productivity through complementary resource use and improved weed suppression. In this study, weeds were predominantly grass species, likely thriving under the high nitrogen availability provided by legume fixation and fertilizer inputs in monocultures. This highlights the vulnerability of monocultures to nutrient-responsive weeds, whereas mixtures can buffer against weed dominance through competitive interactions. Although conducted over one production year, the study coincides with the typical persistence window of short-lived species such as red clover, reflecting peak performance and a useful reference for understanding species interactions and productivity in the early years of grassland management.

Conclusion

Increasing species and functional group diversity enhanced %Ndfa, indicating more efficient nitrogen fixation in mixtures compared to legume monocultures. Despite their high DM yield, legume monocultures suffered from significantly greater weed infestation, which reduced their net productivity. Nevertheless, they maintained the highest proportion of legume biomass, correlating with the highest total nitrogen fixed. In mixtures, legume biomass and total nitrogen fixation declined with increasing diversity, despite higher %Ndfa values. These results suggest that carefully designed mixtures can balance the benefits of nitrogen fixation with productivity and resilience where including legumes ensures substantial nitrogen input, while increasing diversity can limit weed pressure and stabilize yields, providing practical guidance for managed grasslands.

Acknowledgement

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Potential of grassland species as carbon sinks in the Mediterranean dehesa

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Abstract

Grasslands play a key role in climate change mitigation by acting as carbon sinks through photosynthetic carbon fixation. This study compared the carbon sequestration potential of the main pasture species in Mediterranean dehesa. A randomised complete block trial was conducted with ten species (four grasses and six legumes) and two replicates, evaluating carbon concentration (g kg^{-1}) and the amount of carbon sequestered (Mg ha^{-1}) in above-ground biomass over three consecutive seasons (2021–2024). The mean carbon concentration was 370.2 g kg^{-1} , showing a highly significant correlation with spring precipitation ($R^2 = 0.998$). Grasses averaged 384.2 g kg^{-1} , while legumes reached 362.2 g kg^{-1} . *Ornithopus compressus* had the highest carbon concentration (394.0 g kg^{-1}) in 2023/2024. *Trifolium subterraneum* ssp. *subterraneum* had the highest carbon sequestration capacity (14.78 Mg ha^{-1}), followed by *Festuca arundinacea* (11.85 Mg ha^{-1}), *Lolium rigidum* (10.07 Mg ha^{-1}) and *Ornithopus compressus* (9.15 Mg ha^{-1}). These results highlight the importance of selecting specific pasture species to maximise the role of Mediterranean grasslands as carbon sinks and enhance the sustainability of livestock systems.

Keywords: biodiversity, carbon sequestration, climate change, ecosystem services, grasses, legumes

Introduction

The dehesa, a traditional agrosilvopastoral system in south-western Iberia, is characterised by high biodiversity and the capacity of its grasslands to act as carbon sinks, contributing to climate change mitigation through CO_2 fixation and storage in biomass and soil. Globally, grasslands can store up to 30% of the world's soil carbon (Schuman *et al.*, 2002). Grasses, with deep root systems, promote carbon sequestration in deeper horizons, while legumes contribute by fixing nitrogen and stimulating microbial activity, enhancing soil carbon accumulation (Fornara & Tilman, 2008). Functional diversity, together with extensive management practices such as grazing and rotation, supports productivity, stability, and persistence of pastures (Llera & Galea-Gragera, 2025a), improves soil health (Llera & Galea-Gragera, 2025b), and helps maintain the carbon sink function even under extreme climatic conditions (Chen *et al.*, 2018; Sun *et al.*, 2024). The objective of this study is to analyse the carbon sequestration potential of above-ground biomass in each pasture species, as their individual contribution has been little explored to date.

Materials and methods

The trial was conducted at the Valdesequera experimental farm (Badajoz, south-western Iberia) on clay-loam soil (25.5% clay, 0.87% organic matter, CEC 17.8 meq/100 g , pH 6.0). Plant material consisted of annual forage legumes from the official active collection of the CICYTEX Germplasm Bank (Galea-Gragera & Llera, 2025). Two traditional species (*Trifolium subterraneum* ssp. *subterraneum*

Table 1. Carbon concentration (g kg^{-1}) and sequestered carbon (Mg ha^{-1}) in above-ground biomass across three seasons and the eleven evaluated pasture species.

	C Content (g kg^{-1})		C Amount (Mg ha^{-1})	
Season				
2021/2022	369.9	B	11.05	A
2022/2023	353.3	C	4.67	B
2023/2024	387.5	A	9.61	A
Species				
<i>T. subterraneum</i>	364.2	C	14.78	A
<i>T. glomeratum</i>	361.5	C	6.21	DE
<i>T. cherleri</i>	341.1	D	6.38	DE
<i>O. compresus S</i>	358.6	C	9.25	BC
<i>O. compresus C</i>	365.3	BC	9.05	BCD
<i>B. pelecinus</i>	368.7	BC	5.96	E
<i>M. polymorpha</i>	376.6	AB	5.45	E
<i>D. glomerata</i>	385.4	A	7.84	CDE
<i>P. tuberosa</i>	385.2	A	6.05	E
<i>F. arundinacea</i>	383.6	A	11.85	B
<i>L. rigidum</i>	382.4	A	10.07	BC
Significance				
Season	*		*	
Species	*		*	
Season×Species	*		NS	
Overall mean (g kg^{-1})	370.2		8.44	
CV (%)	2.85		28.76	

Different letters in a column indicate significant differences (LSD, $p < 0.05$). * $p < 0.0001$. NS, not significant; CV, coefficient of variation.

(TS), *Medicago polymorpha* (MP)) and four second-generation species (*Ornithopus compressus*—with and without testa—(OCC, OCS), *Biserrula pelecinus* (BP), *Trifolium glomeratum* (TG), *Trifolium cherleri* (TCh)) were evaluated, together with four perennial grasses (*Dactylis glomerata* (DG), *Phalaris tuberosa* (PT), *Festuca arundinacea* (FA), *Lolium rigidum* (LR)). A randomised complete block design was used with 11 species (including both *O. compressus* variants) and two replicates. Although limited replication could affect statistical precision, no relevant effects on data consistency were detected. Over three seasons (2021–2024), carbon concentration in above-ground biomass was determined using the Dumas method with a LECO analyser (LECO, 1998), and dry matter yield (Mg ha^{-1}) was recorded. Carbon sequestration (Mg ha^{-1}) was calculated from these two variables. Statistical analysis (ANOVA, $p < 0.05$) considered season and species as factors, and significant differences among means were identified using the LSD test in R (v. 4.4.3).

Results and discussion

The mean carbon concentration in the above-ground biomass of the evaluated species was 370.2 g kg^{-1} . In all seasons (Table 1), grasses exhibited higher carbon concentrations than legumes. The difference between the mean values for grasses (384.2 g kg^{-1}) and legumes (362.2 g kg^{-1}) was highly significant. TS recorded the highest amount of sequestered carbon among all evaluated species (14.78 Mg ha^{-1}), followed by FA (11.85 Mg ha^{-1}), LR (10.07 Mg ha^{-1}), and both OCS and OCC (9.25 and 9.05 Mg ha^{-1} , respectively), with no significant differences observed between them.

Interannual variability and differential species responses reinforce the importance of maintaining high functional diversity in grasslands, as such diversity is associated with greater carbon accumulation and stability in the soil, even under adverse climatic conditions (Chen *et al.*, 2018; Sun *et al.*, 2024). These

findings are consistent with previous reports from the Tibetan Plateau, where nature-based solutions enhance carbon sequestration through restoration and adaptive management (Sun *et al.*, 2024). This highlights the relevance of adaptive management strategies and the selection of resilient species to maximise the long-term carbon sink function.

Although the absolute values obtained in this study correspond to specific experimental conditions, similar results have been observed in other production systems. For instance, Zhang *et al.* (2024) demonstrated that legume-based covers significantly increase soil organic carbon compared to non-legume covers (mainly grasses), due to greater inputs of plant residues and stimulation of microbial activity.

Conclusion

The results confirm that grasses have a higher carbon concentration than legumes and that pasture species in the Mediterranean dehesa exhibit a remarkable potential as carbon sinks (mean: 8.44 Mg ha⁻¹). *Trifolium subterraneum* ssp. *subterraneum* showed the highest sequestration capacity (14.78 Mg ha⁻¹). These findings emphasise the importance of diversification and the selection of suitable species for Mediterranean grazing systems, as well as sustainable pasture management to enhance climate change mitigation through their carbon sequestration capacity. Optimising such management has relevant practical implications in productive, environmental, and economic terms.

Acknowledgement

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Variability of grassland vegetation quantity and quality as induced by photovoltaic (PV) modules

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Abstract

A growing demand for renewable energy is leading to the expansion of free-field photovoltaic systems (FFPV). PV modules modify microclimatic conditions on site, which can impact grassland vegetation quantity and quality. To determine these effects, we examined the grassland vegetation at different positions from the PV modules at a dummy PV system in northern Germany. At harvests, compressed sward height (CSH) was determined, soil plant analysis development (SPAD) values were used to non-destructively measure leaf chlorophyll content of *Plantago lanceolata*, and crude protein and lignin content of biomass was assessed by near infrared spectroscopy (NIRS). Compared to plants in full sun, plants in shade showed high SPAD values indicating increased chlorophyll production to compensate for low light conditions. Additionally, high crude protein content in shade was observed which could have led to instability of the plants and in turn to a low sward height due to vegetation lodging. The results contribute to our understanding of grassland characteristics in combination with PV with implications for feed quantity and quality.

Keywords: biomass, CSH, NIRS, photovoltaic, SPAD

Introduction

In Europe, free-field photovoltaic (PV) systems on grasslands are expanding in order to mitigate climate change as set out in the Renewable Energy Sources Act 2023 (EEG 2023). In grasslands, PV modules induce microclimatic changes which were shown to affect plant development, biomass of functional plant groups, and chlorophyll content of a newly sown multispecies plant mixture (Baumann *et al.* 2025). In the present study, we assessed the impact of dummy PV modules on plant quantity and quality of extensively managed grassland in northwest Germany. Specifically, we examined the sward height, chlorophyll content, plant biomass, biomass crude protein content and lignin content. We further elucidated the relationships of these parameters with particular consideration of the observed vegetation lodging effect.

Materials and methods

In April 2024, dummy PV constructions made from heavy-duty shelves were installed (5 m×1 m; tilt angle 35°, lower edge 80 cm above ground) on three plots (4 × 10 m) on extensively managed grassland on a podzol soil in northwest Germany. Three plots without construction served as control in a randomized block design.

Sampling took place in June, August and October 2024, within frames of 50 × 50 cm in shade and half-shade areas of PV plots and within two frames for each control plot (full sun). Within the frames, we determined compressed sward height (CSH), soil plant analysis development (SPAD) values from

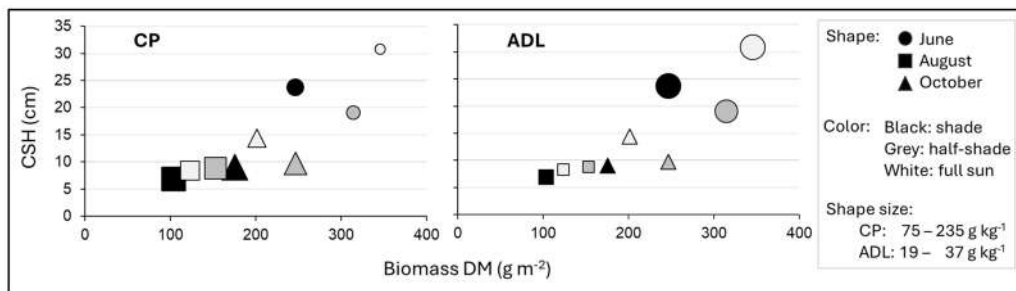


Figure 1. Mean amounts of crude protein (CP) and lignin compounds (ADL) in relation to compressed sward height (CSH) and biomass dry matter (biomass DM) over time for the different PV areas.

Table 1. Correlation coefficients ρ of all harvests for investigated parameters depending on light conditions.

Light condition	Parameter	CSH	Biomass (DM)	SPAD	CP	ADL
Shade/half-shade	CSH	–	0.53	0.19	–0.69**	0.72**
	Biomass (DM)	0.88**	–	–0.23	–0.93***	0.75**
	SPAD	–0.18	–0.33	–	0.13	–0.05
	CP	–0.90***	–0.93***	0.18	–	–0.90***
	ADL	0.69**	0.71**	0.07	–0.85**	–
Full sun/all	CSH	–	0.79***	–0.40**	–0.87***	0.67***
	Biomass (DM)	0.95***	–	–0.37*	–0.85***	0.70***
	SPAD	–0.92***	–0.88**	–	0.52**	–0.08
	CP	–0.90***	–0.93***	0.85**	–	–0.72***
	ADL	0.95***	0.88**	–0.87**	–0.92***	–

CSH, compressed sward height; Biomass (DM), biomass dry matter; SPAD, soil plant analysis development; CP, crude protein; ADL, lignin compound; shading of cells: shade, black; half-shade, dark grey; full sun, light grey; all, white; area $n = 9$, all $n = 18$; *** $P < 0.001$, ** $P < 0.05$, * $P < 0.1$.

Plantago lanceolata L. as a proxy for chlorophyll content, and biomass of which crude protein (CP), and lignin compounds (ADL) were analysed by near infrared spectroscopy (NIRS) after drying and milling.

Mixed ANOVA with repeated measurements followed by Tukey post-hoc test, as well as Spearman correlation coefficients ρ were applied using the statistical software R (v. 4.4.3; R Core Team 2023). Significant differences refer to $P \leq 0.05$.

Results and discussion

Biomass dry matter (DM) was lowest in the shade area and decreased over time (Fig. 1) indicating the negative effect of reduced light on biomass production. In general, biomass was positively related with CSH confirming the plate meter as a powerful tool to assess vegetation dry matter (Murphy *et al.*, 2021; Table 1).

However, looking at different light conditions, the strength of this relationship decreased in the order full sun > shade > half-shade, without being significant in half-shade anymore. In the present study, this might be explained by vegetation lodging which was observed particularly in half-shade indicated by a non-linear CSH-biomass relationship; Fig. 1).

In the shade area, the lower correlation strength might additionally be due to a comparably high surface roughness caused by animal activity (moles, mice) underneath the PV modules.

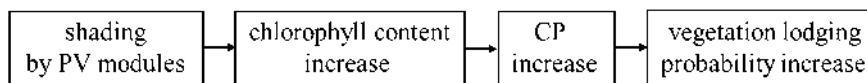


Figure 2. Visible effect of a changed plant physiology caused by PV shading.

Vegetation lodging can occur if the plant tissue strength is low, which can be attributed to factors such as a low photosynthetic active radiation (PAR), a high N content and /or low lignin content in the plant (Xue *et al.* 2017; Li *et al.* 2022). In June, the lignin content was independent of the light conditions, but a significantly higher CP content was observed in plants growing in shade and half-shade than in full sun (Fig. 1). SPAD values decreased with increasing light conditions indicating highest chlorophyll content in shade (data not shown). The increased production of nitrogen-containing chlorophyll molecules (high SPAD values) compensating for low light conditions in shaded areas (Sieve *et al.* 2024) most likely contributed to the higher CP content (Fig. 1). Figure 2 illustrates the reaction chain in which PV shading leads to an increase in plant chlorophyll content, which increases biomass CP content, which in turn increases the probability of vegetation lodging.

In full sun without any shading effects, SPAD values were also strongly positively correlated with CP and negatively correlated with lignin content (Table 1), but that was caused by changes in plant physiology during the vegetation growth.

Conclusion

In our study, differences in light conditions created by dummy PV on grassland strongly affected plant physiology which in turn influenced biomass production and crude protein content. This has to be taken into account when using grassland under PV for animal feed.

The reduction in biomass DM due to low light conditions caused by the PV modules could not be compensated for by a higher plant chlorophyll content. The results further display the dependence of CSH measurements on factors such as the physiology of the plants.

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Can diverse pastures improve the climate resilience of Australia's grazing-based dairy industry?

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Abstract

Conventional rainfed grazing-based dairy farming in southern Australia, based on perennial ryegrass (*Lolium perenne*), is often unproductive under dry conditions. We assessed whether diverse (multi-species) pastures are better at withstanding dry conditions to build drought resilience in Australia's dairy industry. We used commercial rainfed farms (eleven, 2022–2024, with three extended to 2025) in Victoria, Australia. This paper reports findings from one farm. The farm has a temperate climate (cold wet winters, hot dry summers). Average rainfall: 530 mm, predominantly May to October (growing season). Mean minimum and maximum temperatures: 9.0 and 18.4°C, respectively. Pasture biomass and composition, and soil moisture (0–80 cm depth) measurements were taken from April 2023–April 2025. Multispecies pastures increased annual biomass by 19% (15.35 to 18.22 Mg ha⁻¹), primarily due to summer and 'edge of growing season' growth, where the multispecies could access deeper water. The multispecies pasture composition changed seasonally. Perennial ryegrass was dominant in winter and spring, chicory (*Cichorium intybus*), tall fescue (*Festuca arundinacea*) and cocksfoot (*Dactylis glomerata*) increased in autumn, and red (*Trifolium pratense*) and white clover (*Trifolium repens*) increased in summer. Increasing pasture diversity can facilitate adaptation to climate change in the dairy industry.

Keywords: herbs, legumes, multispecies pastures, pasture biomass, perennial ryegrass

Introduction

The Australian dairy industry relies heavily on fertilised ryegrass-dominant pastures to maximize production. Under optimal growing conditions these pastures are highly productive, but they exhibit poor persistence under more constrained conditions, such as drought. This leads to increased costs from purchase of feed and pasture re-establishment, and environmental damage from bare soils (Cullen *et al.*, 2009). Agricultural production in Australia's southern regions is projected to be significantly affected by changes in climate and water availability in the future, creating unfavourable conditions for ryegrass. Multispecies pastures, comprised of plants from the three functional groups: grasses, legumes and forbs, could address this problem. Their diversity supports functional redundancy, enabling systems resilience (Biggs *et al.*, 2020), and retention of productivity during or following a climate event (Lin, 2011). There are many reported benefits of diversity in grassland ecosystems (Tilman *et al.*, 2001) but a lack of empirical evidence regarding their impact in Australian dairy systems. Our hypothesis is that use of multispecies pastures will improve pasture production relative to ryegrass, in more challenging conditions experienced in Australia's southern dairy system, by provided greater 'edge of season' and summer growth.

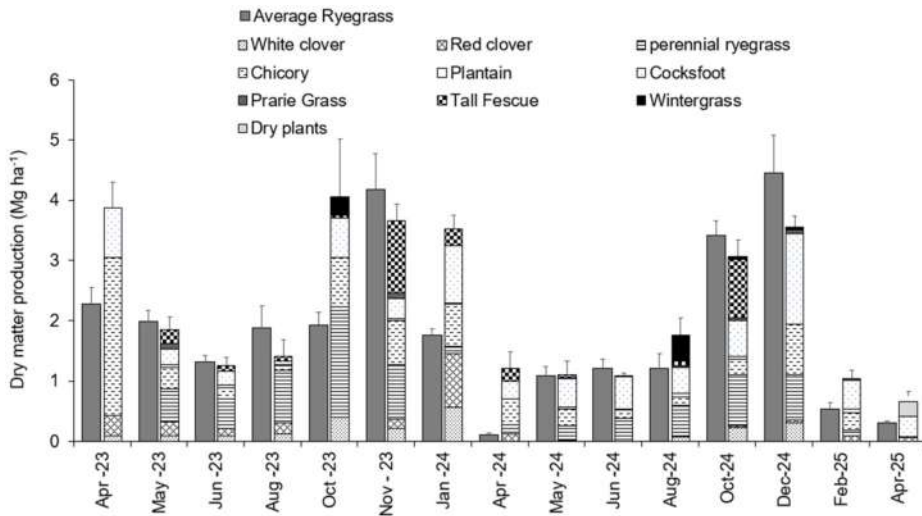


Figure 1. Pasture dry matter production (April 2023 to April 2025) for ryegrass and multispecies pastures. Error bars are total biomass standard errors ($n = 4$).

Materials and methods

Comparison was made between a ryegrass and a multispecies pasture situated on the same soil type at a farm located in the Western District of Victoria (38.42°S, 143.55°E), covering the period from April 2023 to April 2025. Pastures at this site had been established for six years.

Each pasture was split into 4 quadrats of equal size. Pasture exclusion cages (4 per quadrat per pasture type) were used to enable grazing to occur within each paddock. Samples were collected from under the exclusion cages following the grazing rotation. Composition of the multispecies was assessed visually prior to harvest. Biomass was collected from a quadrat (50×50 cm) under each exclusion cage. To assess climate resilience, data on pasture dry matter production, nutritional quality (seasonally) and species composition, and soil moisture (capacitance probe, 80 cm depth) were collected. Meteorological data were sourced from the Australian Bureau of Meteorology's Mount Gellibrand (38.23°S, 143.79°E) (temperature) and Colac (Shire Office) (38.34°S, 143.58°E) (rainfall) stations. The long-term annual rainfall is 530 mm, with mean minimum and maximum temperatures of 9.0°C (5.4–12.8°C) and 18.4°C (11.7–26.1°C) respectively. Differences were assessed using ANOVA (0.05 significance).

Results and discussion

Pasture dry matter production from both pastures was seasonally driven, with low production over the autumn (March–May) and winter (June–August) months (Figure 1).

Using a multispecies pasture sward in place of ryegrass led to a 19% increase in annual pasture herbage production (from 15.35 to 18.22 Mg ha⁻¹), due primarily to greater production in drier months and at the edges of the growing season (growing season May (autumn) to late October (spring)). However, there were differences in the findings between years due to rainfall variation. The benefits of the multispecies were observed where the sampling month followed a dry month. For example, a low September 2023 rainfall led to increased biomass with multispecies in October 2023, whereas the higher September 2024 rainfall meant the October benefit was not seen. Consistent time of better performance with the multispecies were April (autumn) and summer (January or February) due to low levels of stored moisture. The findings reflect the sampling period's climate conditions (Figure 2).

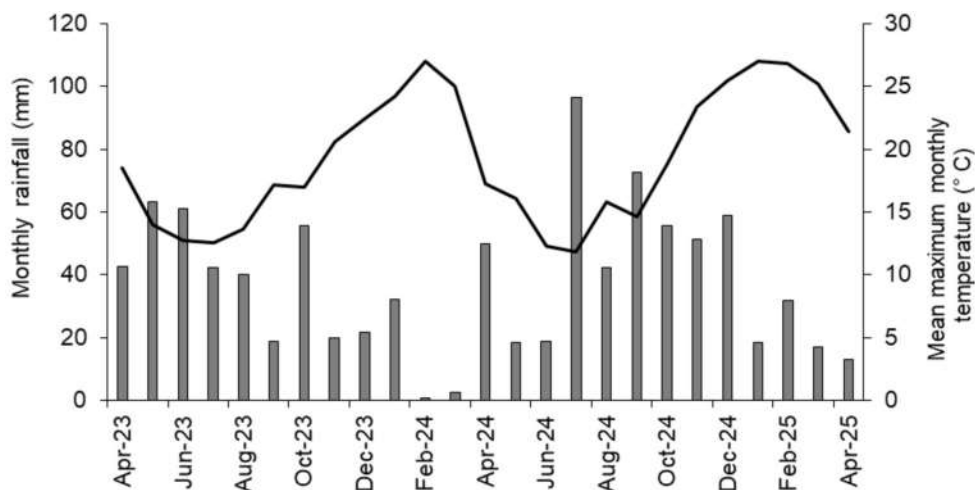


Figure 2. Monthly rainfall and mean maximum temperature from April 2023 to April 2025.

The soil moisture profile to 80 cm depth showed that in the multispecies pastures, greater moisture was drawn from depth (40 cm+) than in the ryegrass pasture over late spring and summer (data not shown), but this was not evident in the top 40 cm of soil, suggesting that the multispecies pasture was better able to access additional water from depth to grow in the drier months. During the drier months, the contribution of ryegrass to the pasture mix was reduced relative to winter and spring, and chicory became more dominant along with more drought tolerant grasses (tall fescue, cocksfoot) (Figure 1) supporting the theory that there are deeper roots present in the multispecies pastures relative to ryegrass that can access water from depth.

Conclusion

The study found that there are benefits from using multispecies swards in pastures for dairy production in southern Australia as a strategy to increase drought resilience. This benefit arises from the deeper-rooted species within the mix being able to access deep soil moisture.

Acknowledgements

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CO₂ and drought effects on biomass production and quality in tall fescue and perennial ryegrass

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Abstract

Despite ongoing climate change, there is a lack of quantitative studies detailing how the co-variation CO₂ and water availability will affect biomass production and quality in grasslands. To this aim, plants of tall fescue and perennial ryegrass were grown in controlled growth conditions at either 200 or 800 ppm and subjected to severe drought. Individual transpiration rates and Soil Relative Water Content were calculated for a subset of plants and Leaf Elongation Rate (LER) was measured daily. Plant dry mass, as well as Water Soluble Content (WSC), Crude Protein (CP) and digestibility were measured at the end of the drought treatment. Our results showed that elevated CO₂ markedly mitigated drought impacts on LER and delayed the onset of growth cessation. Elevated CO₂ dramatically increased plant dry mass in both water supply regimes and species. WSC increased with CO₂, except for ryegrass plants exposed to drought, whereas CO₂ decreased CP in all treatments. Overall, these findings provide valuable insights into the genetic variability of grass response to CO₂ and drought combinations, which may help us to anticipate how plants will respond to climate change.

Keywords: climate change, CO₂, drought, forage quality, grass, leaf elongation

Introduction

Grasslands form the basis of many agrosystems, but current species, cultivars and practices are at risk due to climate change (Durand *et al.*, 2010). Anticipating how grasslands will respond to future conditions is challenging, because climate change variables—mainly CO₂, water availability and temperature—interact in complex ways with plant functioning, which also depends on the genotype (Acker *et al.*, 2024). Leaf growth is one of the most important determinants of biomass production (Nelson, 2000) and is particularly sensitive to climate variables. Although individual effects of CO₂ and water availability on plant growth are relatively well understood, their concomitant variations under climate change will lead to unprecedented growth conditions (Acker, 2021). Therefore, the main objective of the present study was to quantify the effect of CO₂ on the response of leaf growth to drought and its ultimate effects on biomass production and quality. For that purpose, an experiment was conducted under controlled growth conditions using plants of perennial ryegrass (*Lolium perenne*) and tall fescue (*Festuca arundinacea*).

Materials and methods

The experimental protocol is fully described in Acker *et al.* (2024). Briefly, the experiment was conducted in two growth chambers set either at 200 or 800 ppm of CO₂ in order to have the maximal range of plant response. A total of 96 plants per species were arranged in individual plastic pots in each chamber. Plants were irrigated daily with a standard nutritive solution until the emergence of the seventh leaf. Thereafter, water was withheld from half of the plants while the others remained watered. Leaf length was measured daily on the main stem of 10 plants per treatment. LER per tiller was calculated by summing the elongation of all visible leaves on the main stem. Plant transpiration and Soil Relative Water Content (SRWC) were computed gravimetrically using a system of load cells.

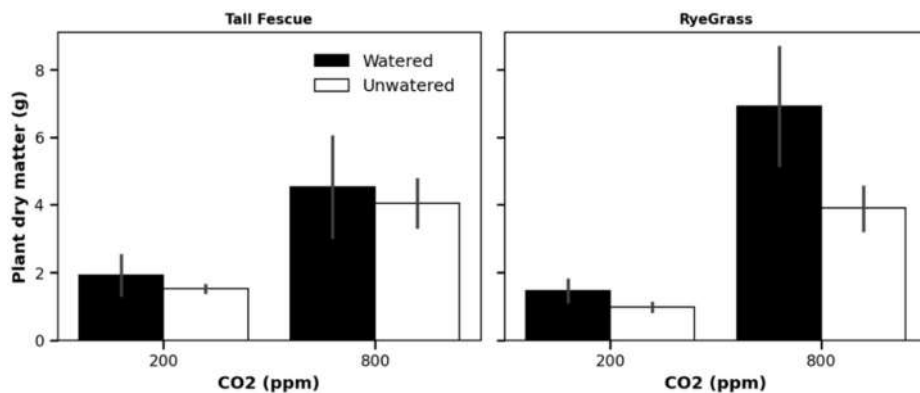


Figure 1. Individual plant dry matter (g per plant) measured at the end of the drought experiment for tall fescue (left panel) and ryegrass (right panel). Data are shown for two levels of CO₂ (200 and 800 ppm) applied to watered (black bars) and unwatered (white bars) plants. Error bars stand for SD.

When leaf elongation ceased in the unwatered treatment, individual plant dry mass was measured, as well as WSC, CP, ADF, NDF and the enzymatic digestibility were predicted from NIRS analyses.

Results and discussion

Our results showed that elevated CO₂ markedly mitigated drought impacts on LER and delayed the onset of growth cessation by three days in both species (Acker *et al.*, 2024). A positive correlation was observed between LER and SRWC in both species, while elevated CO₂ positively affected the offset of this relation, probably due to enhanced stomatal regulation (Acker *et al.*, 2024). The negative effect of drought on the total plant dry matter harvested at the end of the experiment was more severe in ryegrass than in tall fescue (Figure 1). Under watered conditions, elevated CO₂ increased plant dry matter compared to low CO₂, from 1.93 to 4.53 g (+235%) and from 1.47 to 6.92 g (+471%) for tall fescue and ryegrass, respectively. Under unwatered conditions, plant dry matter was lower than in watered conditions and was positively affected by CO₂ (+265 and +397% for tall fescue and ryegrass, respectively). The mitigation of drought by CO₂ was mainly explained by the reduced rate of water consumption observed at elevated CO₂, allowing plants to grow for a longer period (Acker *et al.*, 2024).

CO₂ and water supply treatments also affected biomass composition, as illustrated in Figure 2. WSC content was very low at 200 ppm CO₂ (10–11% for both species) and strongly increased at elevated CO₂ by 286% and 204% for tall fescue and ryegrass, respectively. At elevated CO₂, drought reduced WSC in ryegrass (–26%) but had a positive effect in tall fescue (+21%), while the effect of drought on WSC was not significant at low CO₂. CP content strongly decreased at elevated CO₂ (down to 6.5% for tall fescue plants in the unwatered treatment), which can be attributed to the enhancement of biomass production with CO₂ as reported in Figure 1. Despite the dilution of CP per gram of biomass at elevated CO₂, the total amount of crude protein remained higher than that measured at low CO₂. Drought also decreased CP at 200 ppm CO₂ by 15% and 13% in tall fescue and ryegrass, respectively. At elevated CO₂, drought also reduced CP in tall fescue (–52%) but slightly increased CP in ryegrass. Analyses of ADF and NDF showed that fibre content was hardly affected by CO₂, but increased under drought for both species (data not shown). Enzymatic digestibility was similar between CO₂ treatments for watered plants (approx. 83% and 86% for tall fescue and ryegrass, respectively). By contrast, drought decreased digestibility in all treatments, but the reduction was mitigated at elevated CO₂, particularly for tall fescue.

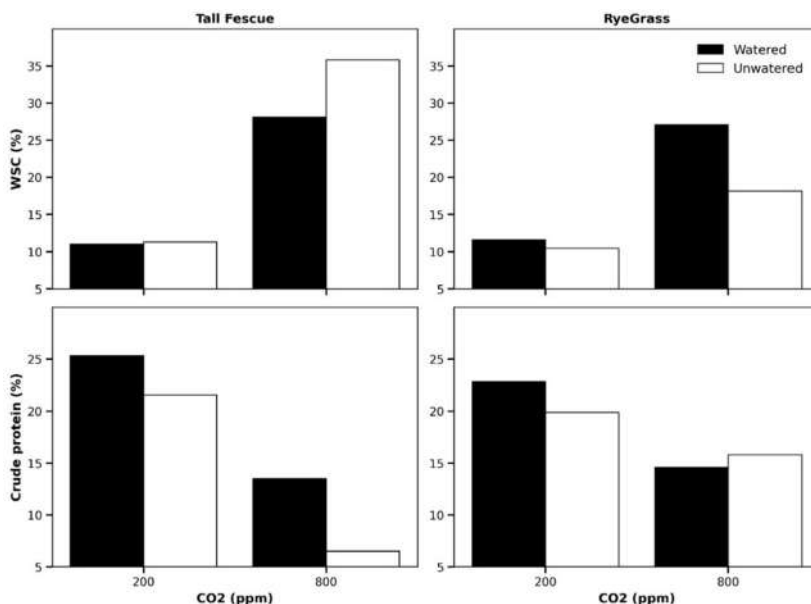


Figure 2. Water soluble carbohydrate (% upper panels) and crude protein content (% lower panels) in tall fescue (left) and ryegrass (right) at the end of the drought treatment. Data are shown for two levels of CO₂ (200 and 800 ppm) applied to watered (black bars) and unwatered (white bars) plants.

Conclusion

The results of this study highlight that CO₂ can strongly affect the effects of drought on grasslands production and forage quality, thus affecting livestock management. Elevated CO₂ enhanced biomass production and mitigated the effect of drought in both species. Biomass composition showed contrasted response to CO₂ and drought between species. In order to further anticipate the effects of climate change on grasslands, new field experiments would be required with different CO₂ levels based on the main IPCC scenarios, using Free-Air Carbon dioxide Enrichment systems for instance. Also, such data would be very useful for the parameterisation of plant models dedicated to assist breeders and agronomists for the adaptation of grasslands to future climates.

Acknowledgement

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Temporal changes in N, P, and K concentrations in soil and herbage following heifer faeces application

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Abstract

Cattle faeces are a key source of nutrient inputs influencing nutrient cycling and plant–soil interactions in grassland ecosystems. This study investigated the effects of heifer faeces deposition on N, P, K concentrations in the soil and herbage using a randomised block design experiment established within the area of a long-term grazing experiment in Betlém, Oldřichov v Hájích, Jizerské Mountains, Czechia. Thirty samples of fresh heifer faeces (each weighing 1 kg) were applied to the pasture surface, and soil, residual of faeces and herbage were sampled in these places. P and K concentrations in the faeces residues decreased over the duration of the study. Faeces application induced significant temporal changes in K concentrations only in the soil, but a consequent reduction in herbage N and P concentrations was detected. There were no significant correlations between soil and herbage nutrient concentrations. These findings provide new insights into nutrient redistribution processes following faeces deposition in an area under long-term pasture management.

Keywords: cattle, grassland management, grazing, nutrients, pasture

Introduction

Semi-natural grasslands cover approximately 34% of the European Union's agricultural area, yet their extent has markedly declined due to land use intensification and abandonment in recent decades (Bandak *et al.*, 2024; Kassahun *et al.*, 2020). These ecosystems are an integral part of agricultural landscapes, supporting high plant diversity and providing essential forage for livestock. Livestock grazing is a broadly used management practice across European grasslands, influencing soil and plant systems through trampling, selective defoliation, and nutrient deposition (Zhang *et al.*, 2022). Among these, faeces inputs under grazing generate nutrient-rich microsites that alter soil nutrient characteristics and microbial activity, thereby influencing biomass production and herbage quality (Brummerloh *et al.*, 2023). Cattle faeces represent a significant source of nutrients for biomass production and play a key role in regulating carbon and nitrogen cycling within grassland ecosystems. Despite their functional importance, the temporal dynamics of faeces inputs on soil nutrient characteristics and herbage quality remain insufficiently understood. To address this knowledge gap, this study investigates the effects of heifer faeces deposition on nutrient, P, K concentrations in the soil and herbage in Central European grassland over a two-year period.

Materials and methods

The heifer faeces experiment was established in 2019 within a long-term grazing experiment (established in 1998) located in Betlém, Oldřichov v Hájích, Jizerské Mountains, Czechia (50°50.34'N, 15°05.36'E; 420 m asl). According to data from the Liberec Meteorological Station, the site has a

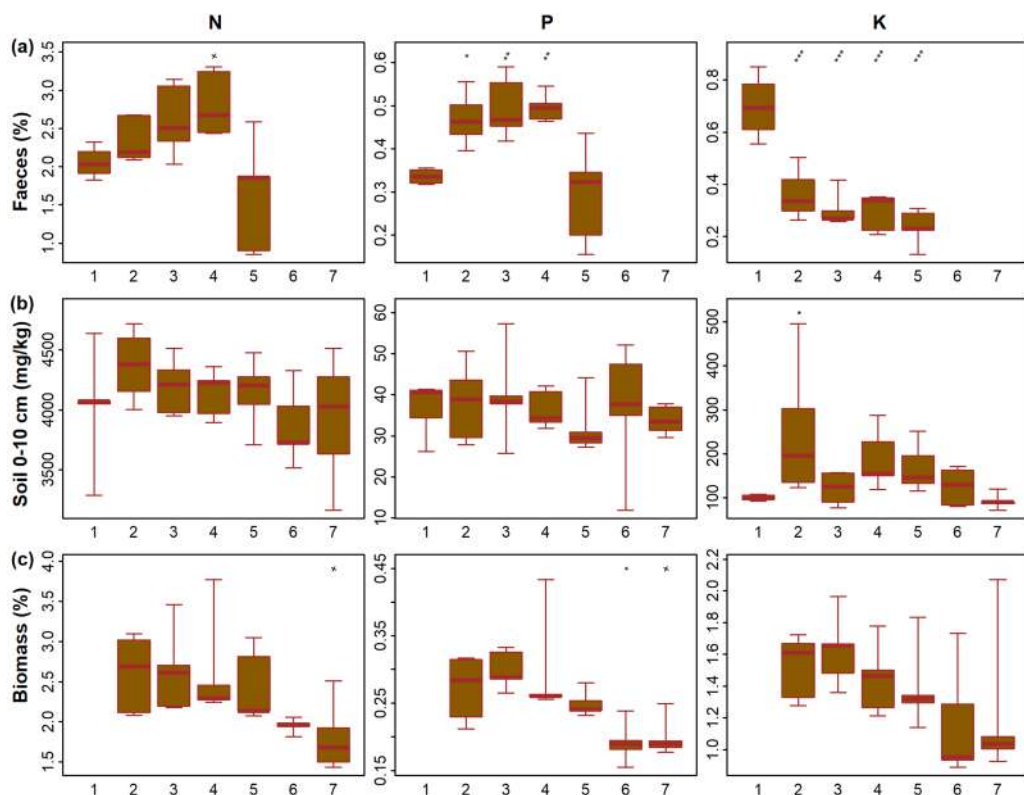


Figure 1. Temporal changes of N, P, K nutrient concentrations in faeces, soil (plant available P, K), and herbage biomass across seven sampling terms. Boxplots show median, quartiles and range of all variables used in this study. Results of Dunnett test compared to the first available date are shown above each box with asterisk coding; $^{+}0.1 > p > 0.05$; $^{*}0.05 > p > 0.01$; $^{**}0.01 > p > 0.001$; $^{***}0.001 > p$.

mean annual temperature of 7.2°C and mean annual precipitation of 803 mm. The heifer faeces experiment was arranged in a randomised block design comprising five blocks of six deposits of fresh heifer faeces (in total 30 circular plots of 30 cm in diameter). Herbage sampling was done in six terms (in the first term one month after faeces application, sampling was not done as insufficient herbage was available and the sward was still covered by faeces). Soil sampling was done in seven terms: (i) before faeces application (28 June 2019); (ii) one month after faeces application (31 July 2019), (iii) two months after faeces application (30 August 2019), (iv) three months after faeces application (1 October 2019), (v) four months after faeces application (31 October 2019), (vi) one year after faeces application (11 June 2020), and (vii) two years after faeces application (3 August 2021). Herbage was sampled from the same plots as soil samples. All chemical analyses were performed in an accredited laboratory of the Czech Agrifood Research Centre (Chomutov, Czechia). For this, only the concentrations of N, P and K in faeces, soil and herbage were selected. Dunnett test and correlation between nutrients in the soil and herbage was used for the statistical analyses.

Results and discussion

There was found to be consequent increase of N and P concentrations in the faeces until the fourth sampling period (Figure 1a), but K concentrations decreased over the study time. However, an increase in K concentration was found in the soil (0–10 cm depth) after faeces application (Figure 1b).

Despite nutrient inputs through faeces application, plant nutrient uptake appeared to be constrained; this was likely due to nutrient losses via leaching, nutrient imbalances, or immobilization processes in the soil, which would likely have reduced the availability of essential nutrients for plant growth. Concentrations of N and P in the herbage biomass decreased through the study period (Figure 1c). Similarly, there was a tendency for decreasing concentrations of K in the herbage.

The correlation analysis revealed no statistically significant relationships between nutrient concentrations in soil and herbage. This outcome aligns with findings by Kassahun *et al.* (2020), who emphasise that the presence of nutrients in the soil does not ensure their effective transfer to the plants. Similarly, Pavlů *et al.* (2019) also found no positive effect of faeces presence on the soil nutrient concentration in tall sward height patches. While some studies have reported positive effects of faeces-based amendments on soil fertility and herbage nutrient content (Rajpoot *et al.*, 2024), others highlight the overriding influence of environmental variability and site-specific conditions.

Conclusions

Heifer faeces application led to elevated of K in the soil at depth of 0–10 cm, only immediately during the period after application. However, there were no correlations between soil and herbage nutrient concentrations; this finding suggested an inefficient nutrient transfer to the plants. These findings emphasize the importance of depth-resolved nutrient monitoring and better alignment of nutrient availability with plant demand to improve nutrient use efficiency and minimize environmental threats through leached nitrates or eutrophication caused by excess of nutrients in grazed grassland systems.

Acknowledgement

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Temporal dynamics of soil carbon and nitrogen in southern Australian dairy pastures

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Abstract

Australia's dairy industry in southern Australia is predominantly reliant on perennial ryegrass pastures. Recent studies suggest that increasing pasture diversity, by incorporating a range of plant forms, offers significant benefits compared to ryegrass pastures, including in soil health, carbon (C) and nitrogen (N). This study compared soil C and N in multispecies and conventional ryegrass pastures across four commercial dairy farms in Victoria's Gippsland region, using a paired paddock approach. Analyses of soil samples at depths of 0–10 cm, 10–20 cm, 20–30 cm and 30–60 cm were done in spring 2023 and autumn 2024. While no significant differences in overall nutrient content in soil were found between multispecies and ryegrass pastures (except for active C), both systems exhibited higher ammonium N ($\text{NH}_4^+\text{-N}$) in spring and higher nitrate N ($\text{NO}_3\text{-N}$) in autumn. Stable C and N forms (Total N, Total Organic C, Total C) demonstrated consistency across seasons and pasture types, indicating resilience to short-term environmental changes, whereas labile forms ($\text{NH}_4^+\text{-N}$, $\text{NO}_3\text{-N}$, Active C) showed greater variability and sensitivity to seasonal factors, suggesting their potential as indicators of short-term soil condition changes. Significant differences in soil C and N content were observed among the four sites ($P < 0.001$), attributable to variations in climatic conditions.

Keywords: dairy systems, multispecies, ryegrass, soil carbon, soil nitrogen

Introduction

In southern Australian dairy farms, particularly in Victoria and Tasmania, perennial ryegrass (*Lolium perenne* L.) has long been the dominant pasture species (Garcia *et al.*, 2014). However, the sustainability of these intensive monoculture systems is increasingly questioned due to their heavy reliance on mineral fertilizers (Lüscher *et al.*, 2014) and poor drought tolerance (Wang *et al.* 2024). Multispecies pastures, combining grasses, legumes, and herbs, may provide benefits including high productivity with reduced nitrogen fertiliser input, improved drought resilience, and increased carbon sequestration potential (Thomson & Albornoz, 2023). Despite these advantages, there is a notable lack of studies examining soil C and N dynamics between multispecies and conventional ryegrass pastures, particularly in Australian climates. In addition, the long-term outcomes on soil C and N when relying purely on biologically fixed N in multispecies pastures is unknown, particular as many of these pastures are established on sites that have a history of heavy fertilizer application. To address this, a two-year farm-scale study was conducted across four commercial farms in Gippsland in Victoria, Australia, which measured soil C and N under multispecies pastures and perennial ryegrass monocultures.

Materials and methods

Four field sites were established on commercial dryland dairy farms across Gippsland (Table 1). Paired paddocks were used at each site, with grazing rotations varying from 20 to 40 days seasonally. Fertiliser types and soil amendments varied between farms but were consistent across pasture types within each farm (Table 1). Soil samples were collected at two time points: Spring 2023 and Autumn 2024. The

Table 1. Details of experimental sites

Location	Coordinates (Lat. S, Long. E)	Long term avg RF (mm)	Soil texture pH (1:5 CaCl ₂) (0–10 cm)	Multispecies composition	Fertilizer and soil management
Newmerella	37.739974, 148.46269	769	Silty loam R: 5.7; M: 6.0	Perennial ryegrass, white clover, plantain, chicory, forage turnip	Compost (10 t ha ⁻¹ year ⁻¹) Silica, gypsum and lime (1 t ha ⁻¹ year ⁻¹)
Goon Nure	37.967227, 147.54513	644	Silty loam R: 6.6; M: 6.6	Perennial ryegrass, white clover, red clover, plantain, chicory, oats	Urea (100 kg ha ⁻¹ year ⁻¹), Pellet chicken manure (400 kg ha ⁻¹ year ⁻¹)
Nilma North	38.10613, 145.96523	923	Loamy sand R: 5.3; M: 5.5	Perennial ryegrass, lucerne, white and red clovers, plantain, chicory	Effluent+Urea (120 kg ha ⁻¹ year ⁻¹), Pellet chicken manure (200 kg ha ⁻¹ year ⁻¹) lime, gypsum 1 t ha ⁻¹ year ⁻¹),
Waratah Bay	38.79954, 146.07317	881	Loamy sand R: 5.3; M: 6.0	Perennial ryegrass, white clover, plantain, chicory	K humate 26% (8 kg ha ⁻¹ year ⁻¹), Urea (30 kg ha ⁻¹ year ⁻¹)

RF, rainfall; R, ryegrass; M, multispecies.

soil sampling methodology involved extracting four deep soil cores (0–60 cm) from a transect (approx. 71 m) at 4 points (approx. 14 m intervals) along the transect in four pre-defined quadrats (50 m×50 m) within each treatment paddock (area approx. 1 ha). Samples from each transect were composited to yield four composite samples per paddock (1 per quadrat). The deep cores were stratified into five depth intervals: 0–10 cm, 10–20 cm, 20–30 cm, 30–40 cm and 40–60 cm. Collected soils were analysed for total C (TC) and N (TN), total organic C (TOC), labile/active C, mineral N (ammonium N (NH₄⁺-N), nitrate N (NO₃⁻-N), and potentially mineralizable nitrogen (PMN). The data were analysed using a linear mixed model (LMM) approach implemented in GenStat software.

Results and discussion

Soil carbon: Topsoil C was high in both multispecies and ryegrass pastures (5.04 ± 0.49 and 5.09 ± 0.55%, respectively). Carbon content decreased markedly with soil depth in both systems (Table 2). Pasture type (multispecies vs. ryegrass) had no significant effect on TC ($P > 0.991$), TOC ($P = 0.606$), or active carbon ($P = 0.856$) (Table 3). Season significantly ($P < 0.05$) affected TC and TOC (Table 3), with higher values in spring compared to autumn, but not active C ($P = 0.789$). However, active C analysis revealed an influence of the interaction between season, pasture type, and soil depth that was not apparent with TC and TOC (Table 2). The consistency in the four-way interaction (site×pasture type×soil depth×season) across TC, TOC ($P > 0.001$ for both TC and TOC) and active C ($P = 0.001$) underscores the intricate nature of soil C dynamics. These results collectively suggest that while overall C content may not be significantly affected by pasture type in the short term, the more labile active C fraction shows significance relationships with pasture, soil layer and season (Table 3). Rutledge *et al.* (2017) also found no evidence of increased C retention in more diverse swards compared to ryegrass-clover swards over 4-year study period.

Table 2. Soil C variation in pasture systems

	Carbon type			
	Multispecies		Ryegrass	
Soil layer	0–10 cm	30–60 cm	0–10 cm	30–60 cm
Total C (%)	5.04 ± 0.12	1.10 ± 0.12	5.09 ± 0.55	1.40 ± 0.19
Organic C (%)	2.61 ± 0.46	0.57 ± 0.10	2.89 ± 0.51	1.05 ± 0.19
Active C (mg/kg)	1221 ± 216	248 ± 44	1103 ± 195	341 ± 60

Table 3. *P*-values from a linear mixed model (LMM) analysis of soil C and N parameters across sites, soil depths, seasons, and pasture types, and the interaction between them.

	TC	TOC	Active C	TN	PMN	NH ₄ ⁺	NO ₃ ⁻
Farm	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	0.133
Soil depth	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
Season	0.006	0.022	0.789	0.539	0.205	<0.001	<0.001
Pasture type	0.991	0.606	0.856	0.358	0.970	0.513	0.696
Pasture type×season	0.318	0.485	0.002	0.762	0.457	0.731	0.372
Pasture×soil depth×season	0.201	0.984	<0.001	0.860	0.980	0.047	0.291
Farm×pasture type×depth×season	<0.001	0.001	0.001	<0.001	0.197	0.002	0.859

Soil nitrogen: There were significant differences for TN, PMN, and NH₄⁺-N (all $P < 0.001$) among farms; however, not for NO₃⁻-N ($P = 0.133$). The lack of significant sites effects for NO₃⁻-N could be attributed to its high mobility and rapid transformation in soil depending on climate conditions, (such as RF), leading to more variable levels across sites. Pasture type did not significantly affect any N measure. However, seasonal effects varied among N forms. NH₄⁺-N levels were higher in spring (7.11 kg ha⁻¹) compared to autumn (5.15 kg ha⁻¹), while NO₃⁻-N showed the opposite trend with higher levels in autumn (16.92 kg ha⁻¹) than in spring (9.97 kg ha⁻¹) (both $P < 0.001$). These findings align with Ridley *et al.* (2001) who reported that soils under pastures in southeastern Australia contained more mineral N in autumn than in spring with NO₃⁻ comprising 70–77% of the mineral N. These results highlight the importance of climate (as evidenced by the variable RF across the four sites; Table 1), in determining N dynamics. The contrasting seasonal effects on NH₄⁺-N and NO₃⁻-N suggest that readily available N pools are more sensitive to seasonal fluctuations than TN pools.

The diverse management practices, combined with variations in soil texture, rainfall, and multispecies composition, likely contributed to the complexity of our results.

No significant changes in stable soil C and N were observed within the one-year study period, consistent with the understanding that such changes typically require longer timeframes which is a key limitation of this study.

Conclusion

While total soil C and N content was comparable between pasture systems, distribution varied across farms and soil layers. Stable forms of C and N remained consistent across seasons and pasture types, confirming their reliability as long-term soil quality indicators. In contrast, labile forms showed variability, suggesting their utility as short-term soil condition markers. Future long-term studies are necessary to track changes in stable C and N forms, which can take decades to be seen, to gain a proper understanding of the long-term outcomes of pasture species selection and management.

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Biodiversity and ecosystem functioning after 60 years of fertilization: insights from the Ossekampen grassland experiment

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Abstract

The biodiversity-productivity relationship of four different fertilization treatments (unfertilized control, Ca, PK, and NPK) has been assessed at the Ossekampen Grassland Experiment in 2022. The long-term fertilization experiment was established in 1958, on a species-rich old pasture on heavy clay soil near Wageningen, the Netherlands. The long-term fertilization resulted in grassland ecosystems with a distinct botanical composition, confirming a significant relationship between aboveground biomass production (productivity) and plant species richness. However, the response to the fertilization on a selection of productivity and biodiversity indicators, above and belowground, were different between treatments. The NPK treatment reached highest productivity (yield, nitrogen yield and root density) as well as highest belowground biodiversity for the indicators bacterial and fungal biomass. In the liming (Ca) treatment highest above ground biodiversity (richness in plant species, and flowering) as well as belowground biodiversity (earthworm abundance) were measured. Our findings highlight that depending on the type of fertilization, the direction and strength of the biodiversity productivity relationship is affected.

Keywords: biodiversity, long-term fertilisation experiment, productivity

Introduction

Species-rich grasslands are of high importance, supporting biodiversity while providing multiple ecosystem services. Although it is well established that moderate management intensity enhances biodiversity and multifunctionality, the long-term effects of individual management practices on above- and belowground biodiversity and how these are linked to ecosystem service provision remain poorly quantified due to a lack of long-term experiments. The Ossekampen experiment, a long-term fertilization experiment, established in 1958 to improve hay quality and biomass production, offers a unique opportunity to address this knowledge gap. After more than 64 years of continuous management, the treatment plots have developed into distinct grassland ecosystems, with clear differences in botanical composition and yield characteristics between treatments (Bufe *et al.*, 2025). A trade-off between plant species richness and productivity was confirmed in previous studies (e.g., Korevaar & Geerts, 2015; Berendse *et al.*, 2021). In this study, we sampled four fertilization treatments during the 2022 growing season, aiming to address the following research question: What are the effects of long-term fertilization with Ca, PK and NPK on indicators of productivity and above- and belowground biodiversity?

Table 1. Mean values per treatment (control, Ca, PK and NPK) and corresponding ANOVA results for selected indicators of productivity, biodiversity, and soil quality assessed in 2022.

Indicator	Treatment (mean (SD))				ANOVA
	Control	Ca	PK	NPK	<i>F</i> (df 3,4)*
Dry matter yield (kg DM ha ⁻¹)	3391 (401)	5564 (53)	5480 (690)	9492 (748)	43.3*
N yield (kg N ha ⁻¹)	51 (0.7)	92 (0.7)	84 (17)	141 (18.4)	17.8*
Root density 0–20 cm (1–10; low to high)	7.5 (0)	8.0 (1)	8.6 (0.2)	9.3 (0.4)	ns
Plant species richness (n)	28 (1.8)	31 (3.2)	26 (3.5)	15 (0.4)	14.7*
Richness flowering plants (n)	22 (3.5)	24 (0.7)	22 (4.2)	13 (0.7)	6.3*
Richness insect species (n)	22 (0)	27 (2.8)	25 (5.6)	23 (2.1)	ns
Earthworm abundance (n per 0.04 m ⁻² ; 0–20 cm)	0.5 (0.7)	1.5 (1.4)	0.3 (3.5)	0.3 (3.5)	s
Earthworm burrows (n per 0.04 m ⁻² 0–10 cm)	2.5 (1.4)	2.8 (1.0)	3.0 (2.1)	2.0 (2.1)	ns
Bacterial biomass (mg kg ⁻¹ soil; 0–20 cm)	866 (79.2)	665 (19.1)	813 (9.2)	911 (16.3)	13.1*
Fungal biomass (mg kg ⁻¹ soil; 0–20 cm)	546 (22.6)	558 (19.1)	560 (29.7)	592 (61.5)	ns

* Significant at $P < 0.05$; ns, not significant.

Materials and methods

In 2022, we assessed a wide range of indicators of selected treatments at the Ossekampen experiment (Wageningen, The Netherlands). It was a very dry year, with a precipitation deficit of 318 mm from April to September. The selected treatments received an annual fertilization of Ca (357 kg ha⁻¹), PK (22 kg P ha⁻¹, 166 kg K ha⁻¹) and NPK (160 kg N ha⁻¹, 33 kg P ha⁻¹, 311 kg K ha⁻¹) and were replicated twice (plot size 40 m²). Indicators of productivity were dry matter yield, N yield and root density. Indicators of above ground biodiversity were plant species richness, flowering plants and insect richness. Indicators of belowground biodiversity were earthworm abundance, earthworm burrow abundance, and bacterial and fungal biomass. Indicators of soil quality were organic matter and pH. For detailed information on the experimental design and on sampling protocols for dry matter yield and N concentration see Berendse *et al.* (2021). Plant species richness was assessed by the Braun Blanquette method in June 2022. A transect method combined insect sampling and flowering plant observations (7 times during April - September). From each replicate two sods 0.04 m⁻² were assessed for counting earthworm abundance (0–20 cm), earthworm burrow abundance; and root density (0–10 cm) was scored on a scale from 1 (low) to 10 (high). Bulk samples (0–20 cm) were analysed for bacterial and fungal biomass by phospholipid acid analysis (PLFA), organic matter content (%) and soil pH. Statistical analysis (ANOVA and linear regression) was carried out with Jamovi software. The outcomes of the measured indicators in the fertilized treatments (Ca, PK and NPK) were expressed relative to the unfertilized control treatment.

Results and discussion

Productivity indicators (dry matter yield, N yield and the root density) were lowest in the control treatment and highest in the NPK treatment (Table 1). Highest plant species richness, flowering plants and insects were observed in the Ca treatment.

Lowest plant richness and flowering plants were measured in the NPK treatment. Insect richness was not significantly different per treatment. The main insect groups observed were bees, hoverflies and butterflies. Results of above ground biodiversity did not match belowground biodiversity. The highest number of earthworms was found in the Ca treatment and the highest number of earthworm burrows in the PK treatment (Table 1). Earthworm numbers were extremely low, probably due to the

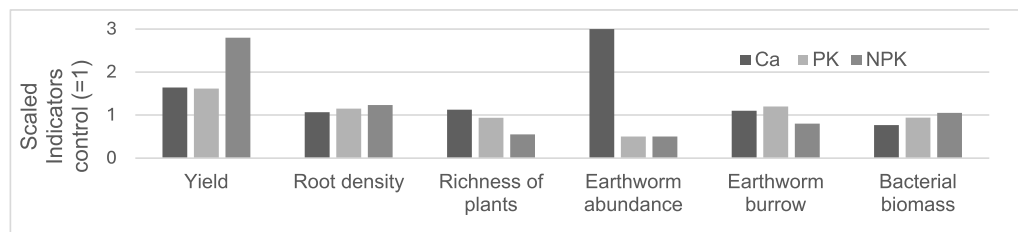


Figure 1. Scaled effects of fertilization (Ca, PK, NPK) relative to the control treatment (control = 1) on a selection of indicators on productivity (dry matter yield and root density) and biodiversity (plant richness, earthworm abundance and burrows, bacterial biomass).

drought. In the NPK treatment, bacterial biomass was significantly higher. The soil quality indicators of organic matter and pH were not significantly different between treatments, OM: Control 12.6%, Ca 11.8%, PK 12.0%, NPK 12.9%; and pH: Control 4.5, Ca 5.9, PK 4.4 and NPK 4.5. In this study we confirmed a significant biodiversity-productivity relationship between yield and plant richness ($R = -0.82$, $P < 0.05$). An overview on the effect size and direction of different fertilization treatments (Ca, PK and NPK) relative to the unfertilized control on the indicators of productivity and above and belowground biodiversity is presented in Figure 1. The productivity indicators (yield and root density) increased in all three treatments with fertilizer application. Plant richness increased in the liming treatment. Figure 1 highlights that the direction and strength of the productivity biodiversity relationship depends on the type of fertilization.

Conclusions

This study confirmed that fertilization management for increased biomass production leads to a significant reduction of aboveground biodiversity. Our findings highlight that depending on the type of fertilization, the direction and strength of the biodiversity productivity relationship is affected. The NPK fertilization reached highest productivity levels and highest bacterial and fungal biomass while above ground biodiversity was lowest. Above ground biodiversity and earthworm numbers were highest in the liming treatment. Long term experiments allow for integrated assessments of biodiversity and multifunctionality.

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Enhancing forage maize resilience with digestate irrigation: A circular approach to climate-induced drought

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Abstract

In the Atlantic Arc region, rainfall is frequent and evenly distributed, and temperatures are moderate. However, there has recently been a decrease in total rainfall, coupled with prolonged summer droughts. Consequently, water stress in maize crops has been observed during the summer months when greater quantities of water are required to support plant and cob development. Field trials in northern Spain have explored the potential of using the liquid fraction of digestate from livestock slurry to strategically irrigate maize crops. The experiment was replicated over two consecutive summers. Drip irrigation was applied in July or July and August, depending on the treatment. Using the liquid fraction of digestate for summer drip irrigation can statistically increase plant weight by up to 61 g per plant. This suggests that the yields of protein, starch and energy increase by up to 23, 12 and 18%, respectively, in irrigated plants. However, these higher yields were not statistically significant. Despite interannual weather variability must be considered, the findings suggest that liquid digestate irrigation may improve maize crop resilience under water-limited conditions. This circular economy-based strategy may help reduce agriculture's water footprint, a growing concern amid increasingly frequent summer droughts under the current climate change scenario.

Keywords: circular economy, maize, climate change adaptation, slurry digestate, strategic irrigation, water stress

Introduction

The Atlantic Arc area in the north of Spain, once known for its mild climate and regular rainfall, is now facing reduced precipitation and prolonged summer droughts. These changes threaten forage maize (*Zea mays* L.) production, particularly during the summer when water demand peaks, compromising vegetative growth and cob development. As a result, yield and feed quality are at risk, undermining livestock production in the region. In response to these challenges, there is a growing interest in circular economy strategies that enhance resource efficiency and increase the resilience to climate change. One such approach involves the use of liquid digestate, a nutrient-rich by-product of the anaerobic digestion of livestock slurry, as an alternative irrigation source. While digestate has been widely studied for its fertilising properties (Möller & Müller, 2012; Tambone *et al.*, 2010), its potential role in mitigating drought stress through strategic irrigation remains underexplored. This study evaluates the use of summer drip irrigation with liquid digestate to improve the yield and nutritional performance of forage maize under water-limited conditions in northern Spain. It explores whether this practice could enhance drought resilience, promote nutrient recycling, and reduce reliance on conventional water sources. By addressing this knowledge gap, the study contributes to the development of integrated, climate-adaptive practices for the sustainable forage production.

Table 1. Agronomic performance and nutritional composition of forage maize under different irrigation treatments.

	C	R1	R2	SD	P
Plant height (m)	2.87	2.96	2.91	0.259	0.566
Cob insertion height (m)	1.27	1.22	1.21	0.126	0.271
Plant weight (g plant ⁻¹)	200 ^b	249	261 ^a	46.3	0.043
Yield (Mg DM ha ⁻¹)	17.97	21.54	20.79	3,959	0.272
Cob (%)	54.40	57.76	53.74	5.582	0.434
Foliage (%)	45.60	42.24	46.26	5.582	0.434
Dry matter at harvest (g kg ⁻¹)	365.2	352.3	330.1	34.81	0.218
Greenery index	45	41	41	6.6	0.481
Dry matter digestibility (%)	75.56	76.21	77.33	2.036	0.336
Crude protein (Mg ha ⁻¹)	1.31	1.49	1.61	0.262	0.128
Starch (Mg ha ⁻¹)	6.50	7.95	7.28	1,931	0.459
Metabolizable energy (GJ ha ⁻¹)	210	253	247	48.3	0.256

^{a,b} Different letters in the same row indicate significant differences among treatments.

Materials and methods

The field experiment was conducted over two consecutive summers (2023 and 2024) in a forage maize cropping system located in northern Spain, within the Atlantic Arc region. The site is characterised by a temperate oceanic climate, with historically regular rainfall patterns that have become increasingly erratic due to climate change (IPCC, 2023). A randomised complete block design with three replicates per treatment was used, maintaining consistent plot and treatment assignment across years. Each 16 m² plot received either conventional management (non-irrigated control) or strategic drip irrigation with liquid digestate from anaerobically digested livestock slurry. Three treatments were compared: a non-irrigated control (C), moderate irrigation in July (R1), and full irrigation in July and August (R2), meeting the crop's total water needs. The irrigation was applied during peak summer months, coinciding with the critical stages of maize development. Agronomic parameters measured included fresh and dry biomass yield, plant height, and cob development. The nutritional quality of the harvested plants was assessed through laboratory analysis of protein, starch, and organic matter digestibility, and energy content was estimated following ARC (1980) guidelines. Statistical analyses were conducted using RStudio (v2024.12.1). Data normality and variance homogeneity were checked prior to applying ANOVA or Kruskal-Wallis tests. Post hoc comparisons were performed using Tukey's HSD or Dunn's test at a 95% confidence level ($p < 0.05$).

Results and discussion

The application of liquid digestate through drip irrigation during the summer months showed a positive influence on forage maize performance (Table 1), although the effects were not consistent across all measured parameters. Notably, plant height remained unaffected by the irrigation treatment, suggesting that vertical growth was not limited by water availability under the conditions of this study. By contrast, individual plant weight increased significantly in the irrigated plots, indicating that water supply during the critical growth period played a key role in biomass accumulation.

Although irrigated treatments led to higher dry matter yields due to increased plant weight, no statistical differences were found between treatments. The two targeted irrigation events in July and August enabled the plants to withstand seasonal drought stress more effectively during the crop's peak water demand. Our findings are consistent with previous research on maize's sensitivity to water deficit during reproductive and grain-filling stages (Çakir, 2004), and reinforce the importance of a timely water

supply to maintain productivity under climate-induced stress conditions. Improvements in forage quality were also observed beyond dry matter yield. Plants irrigated with liquid digestate revealed higher concentrations of protein and starch, along with increased metabolizable energy, although these improvements were not statistically significant. These enhancements are likely linked to the dual function of digestate as both a water source and a carrier of available nutrients, particularly nitrogen and potassium. This aligns with earlier studies that demonstrated the fertilising potential of digestate (Möller & Müller, 2012; Tambone *et al.*, 2010), underscoring its relevance under drought-prone field conditions. The results support the idea that digestate irrigation enhances maize resilience and productivity under water scarcity, while contributing to circular economy goals by valorising livestock waste and reducing freshwater use. While long-term effects (e.g. nutrient accumulation or soil salinization), were not assessed, consistent results across years suggest digestate irrigation as a promising adaptation strategy to summer droughts.

Conclusion

This study demonstrates that strategic summer irrigation with liquid digestate contributes to forage maize resilience under drought conditions. While enhancing individual plant biomass and overall dry matter yield per hectare, no significant effect on plant height was found. Besides, improvements in protein, starch, and energy content highlight the dual role of digestate as both a water and a nutrient source. Despite the need for further research on long-term soil impacts and optimisation of application strategies, these results support the use of liquid digestate as a sustainable and circular approach to climate adaptation in agriculture.

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Evidence on nitrogen fertilisation in tropical pastures: aspects related to experimental conditions and research gaps

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Abstract

Nitrogen (N) use in tropical pastures is important for intensifying animal production. This study aimed to verify through a systematic literature review the effects of N fertilization in tropical pastures on forage production, animal performance, and greenhouse gas (GHG) emissions. The research question formulated followed the PICOS strategy. A total of 84 629 documents were retrieved from six databases, 34 of which met the inclusion criteria. The retrieved studies dated from 2004 to 2023, with 61.7% published in the past five years and 91.2% conducted in Brazil. Overall, 69.7% were conducted under grazing management and 30.3% in plots. Among the grazing studies, 58.3% used continuous stocking and 37.5% used rotational stocking. The most studied forage species was *Brachiaria brizantha*, reported in 52.9% of the studies, while the most common cultivar was Marandu, representing 50% of the results. Among nitrogen fertilizers, urea was the most common source (53.2%). Of all studies, 47.1% evaluated animal performance measures, while 55.8% evaluated GHG emissions, N loss as ammonia volatilization, and/or soil carbon and N levels. Only 2.94% of the studies evaluated both. These results allow us to identify gaps for future research on N use in tropical pastures.

Keywords: research gaps, systematic review, warm climate forage

Introduction

The rational use of N in tropical pastures is an indispensable practice for food production, maximising land use and reducing the need to open up new areas for livestock farming (Bastidas *et al.*, 2024). This study aimed to assess, through a systematic review of the literature, the effects of nitrogen fertilisation on tropical pastures regarding forage production, animal performance, and greenhouse gas (GHG) emissions.

Materials and methods

This is a Systematic Literature Review (SR), conducted and reported in accordance with the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) guidelines (Page *et al.*, 2021). In order to ensure the correct procedure for searching and extracting articles, a review protocol was developed, registered and is available on the Open Science Framework platform at <https://osf.io/ujt7q/>.

A high-sensitivity search strategy was used to meet the research objective. The guiding question was defined based on the PICOS strategy: the population (P) was tropical forage grasses, the intervention (I) was nitrogen fertilization (types of N), the comparison (C) was the use of different doses of nitrogen,

and the outcome (O) was forage production, animal performance, and greenhouse gas emissions. The study type (S) included only experimental studies. This strategy led to the following question to be answered: “What are the effects of nitrogen fertilization on forage production, animal performance, and greenhouse gas emissions in tropical pastures?”

Regarding eligibility criteria, only complete research articles were included in the SR, available in full text from in five databases and one electronic library, namely: SCOPUS (Elsevier), Web of Science (Main Collection), Springer Link, Science Direct (Elsevier), SciELO, and the Cab Direct electronic library. Only articles reporting well-designed experimental studies, in any language and with no publication date restrictions, were accepted. Thus, other retrieved documents were excluded, such as editorial letters, letters to the editor, expert opinions, reviews, correspondence, book chapters, theses and dissertations, abstracts, lectures, and books. Works in which nitrogen fertilisation came from organic sources or exclusively from nitrogen-fixing microorganisms, works with native forage crops, works with grass and legume consortia without nitrogen fertilisation treatment (evaluating only the effect of legumes as a source of N) were excluded.

A highly sensitive electronic search was conducted using descriptors and Boolean operators in the six information sources. For this purpose, the CAPES Journal Portal was used, through the proxy of the Federal University of Mato Grosso do Sul (UFMS, Brazil). The selection of studies was performed by two previously trained reviewers. The studies were evaluated for methodological quality using the Critical Appraisals Skills Programme (CASP, 2018) instrument for experimental studies (CASP Randomised Controlled Trial Standard Checklist). The risk of bias was assessed using the Joanna Briggs Institute (JBI) critical appraisal tool for controlled trials, described by Barker *et al.* (2023), and was considered low due to the objectivity of the results.

Results and discussion

The study identified 84 629 documents, of which 66 976 were scientific articles. Among these, 34 335 articles were duplicates and were excluded, leaving 32 641 articles, which were evaluated for title, abstract, and keywords. After this stage, 32 587 articles were excluded because they did not meet the inclusion criteria for this SR. This left 54 articles selected for full-text reading and classified as potential sources of relevant data. Of these, 34 articles were considered eligible and included in the SR due to their methodological robustness for data extraction.

The studies retrieved date from 2004 to 2023, with 61.7% published in the last five years and 91.2% conducted in Brazil. Of the total, 69.7% were conducted under pasture management and 30.3% in plots experiments. Among the pasture management studies, 58.3% involved continuous stocking, while 37.5% involved rotational stocking. The most studied forage species was *Brachiaria brizantha*, featured in 52.9% of the studies, with the Marandu cultivar being the most common, accounting for 50% of the results. Among nitrogen fertilisers, urea was the most common source (53.2%). This fact is related to the representativeness of the use of this genus in tropical climate regions. In Brazil, it is estimated that 99 million hectares are planted with *Brachiaria* spp. forage crops (Jank *et al.*, 2014). The frequent use of urea in research is also related to its lower cost per kilogram of nitrogen compared to other sources (Longhini *et al.*, 2024).

Among the 34 studies analysed, 16 presented some variable of animal performance, namely: stocking rate (SR), evaluated in 16 studies (47.1%), average daily gain (ADG), evaluated in 12 studies (35.2%)

and gain per area (GPA), evaluated in 10 studies (29.4%). Of the total sample of studies, 18 (55.8%) presented variables linked to GHG emissions, such as nitrous oxide (N₂O), methane (CH₄ enteric and soil), carbon dioxide (CO₂), N loss as volatilised ammonia (NH₃), soil carbon (C) and N contents. The variable with the highest number of assessments was N loss through NH₃, found in 26.4% of the studies, followed by N₂O emissions found in 23.5% of the studies, CO₂ found in 8.8%, and soil N and C contents found in 5.8 and 2.9% of the studies, respectively. However, only one study evaluated the effect of N on forage production, animal performance, and GHG emissions in tropical pastures jointly.

The analysis revealed a fragmented research focus, with most studies addressing either animal performance or GHG emissions in isolation. Few studies integrated these components, which limits understanding of the trade-offs between productivity and environmental outcomes in tropical grazing systems (Bastidas *et al.*, 2024). Despite frequent assessment of stocking rate and average daily gain, there is a clear lack of studies simultaneously evaluating forage production, animal performance, and GHG emissions. Moreover, research has largely concentrated on N-related gaseous losses (NH₃ and N₂O), while soil C and N dynamics remain underexplored. These gaps highlight the need for integrated experimental approaches that capture both productive and environmental responses to N fertilization in tropical pastures, enabling a more holistic evaluation of system sustainability.

Conclusion

The use of N is an important tool for maintaining the productivity of tropical pastures. It is a recurring theme in research in tropical climate regions, which focuses on proposing strategies for the rational use of fertiliser in order to mitigate environmental effects. These results also allow us to identify gaps for future research on the use of N in tropical pastures.

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Effect of ley botanical composition on root biomass in temporary grasslands

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Abstract

Combining species with contrasting traits has been shown to increase resource use efficiency by plant communities. However, knowledge about mixture effects on root growth is still very limited. We assessed the effects of mixing two grasses (perennial ryegrass, timothy), two legumes (white and red clover), and two herbs (common chicory, ribwort plantain) on aboveground biomass till three harvests, and belowground (root) biomass at the third harvest (0–30 cm depth). Red clover achieved the highest aboveground biomass among the monocultures. The six-species equi-proportional mixture revealed significant positive mixing effects resulting in an aboveground biomass similar to that of red clover. Perennial ryegrass had the largest root biomass among the monocultures, and there were no beneficial mixing effects on root biomass of the six-species equi-proportional mixture. Correspondingly, the root biomass of the six-species mixture was significantly lower than that of perennial ryegrass. As a consequence, community root biomass did not correlate with aboveground biomass. We conclude that in mixtures combining grasses, legumes, and herbs in temporary grasslands (≤ 1 year after sowing), species interactions resulting in positive mixture effects on aboveground biomass do not relay on mixture effects on root biomass.

Keywords: belowground biomass, LegumeLegacy project, mixture effects, root-to-shoot ratio, species interactions

Introduction

There is ample evidence that combining grasses and legumes in multi-species mixtures enhances grassland performance relative to mono-specific swards across a range of indicators related to the N economy of the system (Suter *et al.*, 2015, 2021; Fox *et al.*, 2020). Herb species could also contribute to sward performance with specific traits, like deep rooting systems and spatial and/or temporal complementarities in nutrient uptake (Husse *et al.*, 2017). Importantly, interactions between species also affect community root biomass (Yang *et al.*, 2019; Abalos *et al.*, 2021) and seem to depend on the species being combined (Yang *et al.*, 2019; Skersiene *et al.*, 2024). Cong *et al.* (2017) observed a positive effect of the addition of *Plantago lanceolata* L. and *Carum carvi* L. on the root biomass of grass-clover mixtures. Less is known, however, about the effects of grass-legume-herb multispecies mixtures on root biomass in intensively managed grasslands. The objective of this study was to assess such effects in temporary grasslands using two grass, two legume and two herb species.

Materials and methods

A field experiment was conducted on a deep, arable, silty clay loam on the Swiss Plateau, with two grasses (*Lolium perenne* L., *Phleum pratense* L.), two legumes (*Trifolium repens* L., *Trifolium pratense* L.), and two herbs (*Cichorium intybus* L., *Plantago lanceolata* L.). Communities systematically varied in species richness and composition following a simplex design, and species richness ranged from 1

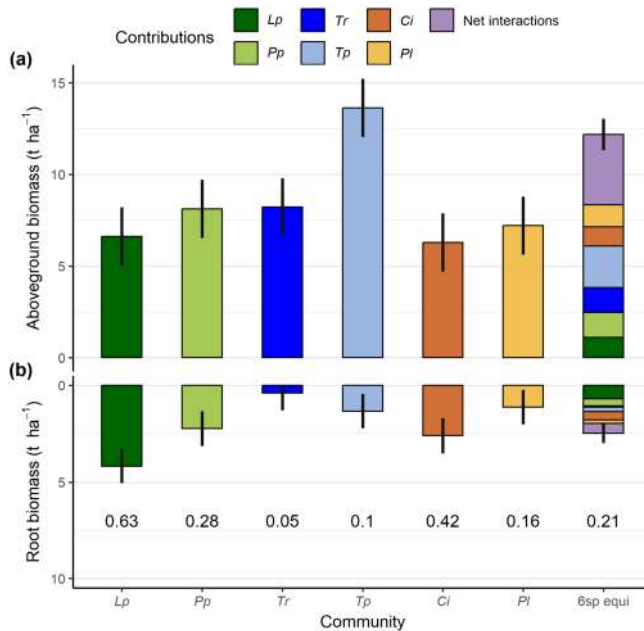


Figure 1. Aboveground biomass (a) and root biomass (b) of *L. perenne* (Lp), *P. pratense* (Pp), *T. repens* (Tr), *T. pratense* (Tp), *C. intybus* (Ci) and *P. lanceolata* (PI) grown as monocultures and in a six-species equi-proportional mixture (6sp equi). For the 6sp equi, the contribution of each species expected from their biomass in monocultures (identity effect) is represented by a distinct colour, and the purple section represents the additional biomass resulting from species interactions. Values below the bars (in b) are root-to-shoot ratios. All values are derived from the regression model, with error bars showing the 95% confidence interval.

to 6. A selection of 33 plots was used for measurements on root biomass. Root samples were taken to 30 cm soil depth using one monolith (25 cm×25 cm) for the 0–20 cm depth and 6 soil cores (4.8 cm diameter) for the 20–30 cm depth. Root sampling was performed in July 2024 after the third harvest, 11 months after sowing the swards. Here, we present results for monocultures and the six-species equi-proportional mixture, reflecting species identities and interaction effects on aboveground and root biomass (0–30 cm depth). The cumulative yield from the start of the vegetation period till root sampling was used for aboveground biomass. All plots received mineral fertiliser applications of 120 kg N, 50 kg P and 300 kg K ha⁻¹ year⁻¹. Data were analysed with linear regression using a Diversity-Interaction (DI) model that estimated species identity effects and an average interaction among species (Moral *et al.*, 2023).

Results and discussion

Aboveground biomass of *T. pratense* was about twice as large as the mean biomass of the other monocultures (Fig. 1a). In the six-species equi-proportional mixture, a positive mixing effect from species interactions was evident ($P < 0.001$), resulting in a 46% higher aboveground biomass than expected from monocultures. This mixture performance was not significantly different from the best-performing monoculture *T. pratense* ($P = 0.129$ for the difference) (Fig. 1a).

In contrast, *L. perenne* had the highest root biomass among all communities (Fig. 1b), consistent with Hofer *et al.* (2017) and likely reflecting the greater below-ground investment strategy of grasses

for effective nitrogen (N) capture compared to N₂-fixing clovers. Importantly, no significant mixture effect was observed with respect to root biomass ($P = 0.157$). Correspondingly, the root biomass of the six-species mixture was significantly lower than that of the best-performing monoculture (*L. perenne*) ($P = 0.004$ for the difference). The contrasting pattern in the above- and belowground biomass resulted in root-to-shoot ratios ranging from ≤ 0.1 in the legumes to ≥ 0.28 in the non-fixing species, except for *P. lanceolata* that had an intermediate root-to-shoot ratio. This pattern is in agreement with Philadelphi *et al.* (2025), where the root-to-shoot ratios of *L. perenne* and *C. intybus* were much higher than that of *T. repens*. Similarly, in a grassland experiment, Marley *et al.* (2024) found that *L. perenne* and *C. intybus* had a larger root biomass than *T. repens* and *T. pratense* in the 0–30 cm soil layer (with *P. lanceolata* root mass in-between). Taken together, our results reveal that in these grass-legume-herb mixtures, root biomass (of swards less than one year old) does not correlate with aboveground biomass and that positive mixture effects evident aboveground cannot be used as indicators for mixture effects on root biomass. Yet, we also show that the positive mixing effects on aboveground biomass can develop with a root biomass expected from the constituent species grown in monocultures.

Conclusion

We conclude that species acquiring their N solely from the soil allocate more resources to root development than species relaying on symbiotic N₂ fixation. In mixtures combining N₂-fixing and non-fixing species, species interactions resulting in positive mixture effects on aboveground biomass do not relay on mixture effects on root biomass. Further analyses in the framework of this project will show if this holds true for N yield, which would point at an increased N uptake per unit of root mass by the root system of grass-legume-herb mixtures.

Acknowledgements

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Increasing sward diversity decreased weed invasion

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Abstract

Weed invasion in productive grasslands can reduce both forage quantity and quality. Increasing the species richness in the sward is one of the ecological strategies for suppressing weeds. The species used in the experiment were two grasses (G1: *Lolium perenne*, G2: *Phleum pratense*), two legumes (L1: *Trifolium repens*, L2: *Trifolium pratense*) and two herbs (H1: *Cichorium intybus*, H2: *Plantago lanceolata*). These species were selected based on their complementary functional traits which may minimise interspecific competition when combined in a mixture. We investigated weed invasion across 34 grassland communities: six monocultures (one for each species), 27 mixtures (two-, three-, four- and six-species equi-proportional mixtures), and a high nitrogen G1 grass monoculture (as a reference for conventional ley grassland management). The communities were established across 53 plots and measured over two growing seasons. Weed percentages were determined by manual biomass separation at each harvest. The findings indicate that mixtures, on average, led to 34% lower weed invasion than the high nitrogen monoculture. Despite the disparity in the high nitrogen monoculture treatment compared to the unfertilized mixtures, there was no difference in the weed invasion. We concluded that increasing species richness reduced weed invasion overall which can be attributed to the strong contribution of the interspecific interaction effect within the mixtures.

Keywords: beta regression, grassland, multi-species mixtures, weed invasion

Introduction

Weed invasion in productive grasslands reduces forage quantity and quality (Naylor, 2002). However, increasing the number of species sown in a productive grassland can decrease weed establishment (Sanderson *et al.*, 2012; Connolly *et al.*, 2018). Low to intermediate species richness with differences in growth habit and phenology is sufficient for optimal weed suppression (Storkey *et al.*, 2015; Suter *et al.*, 2017). Suter *et al.* (2017) proposed that three main mechanisms drive the suppression of weeds: symbiotic nitrogen fixation in leguminous plants; effective light capture due to aboveground biomass production; and competition from non-leguminous plants for nitrogen in the soil. This study investigates how manipulating the sown species diversity of up to six species affects weed invasion.

Materials and methods

A field experiment was established at Brody, Poland, in April 2023, on a loamy sand soil (topsoil composition: 78% sand, 19% silt, 3% clay, pH 6.02). This site is part of the LegumeLegacy project, which follows a standard experimental design across multiple locations in Europe and Canada. The project optimises the numerous benefits of grass, legume, and herb mixtures in crop rotations by modelling underlying mechanisms and legacy effects. The species used in the experiment were two grasses (G1: *Lolium perenne*, G2: *Phleum pratense*), two legumes (L1: *Trifolium repens*, L2: *Trifolium pratense*) and two herbs (H1: *Cichorium intybus*, H2: *Plantago lanceolata*). The experimental design included 34 grassland communities: six monocultures (one for each species), 27 mixtures (two-, three-, four- and six-species equi-proportional mixtures), and a high nitrogen (N) G1 grass monoculture

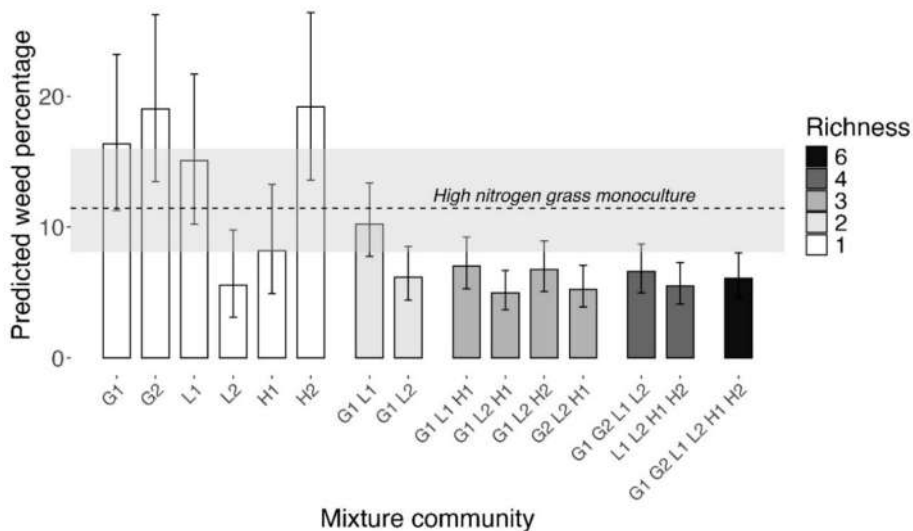


Figure 1. Predicted weed invasion (mean $\pm$ 95% confidence interval) of all monocultures and a selection of mixture communities: two 2-species mixtures, four 3-species mixtures, two 4-species mixtures, and the 6-species mixture. The high nitrogen reference grass monoculture (HighN) is indicated by a horizontal dashed line, with 95% confidence interval (grey band).

(as a reference for conventional ley grassland management—referred to as HighN hereafter). The communities were established across 53 plots measuring 21 m² (7×3 m) four times a year over two growing seasons. The HighN treatment received 100 kg ha⁻¹ of N per regrowth, while all other plots received no nitrogen fertiliser. Each year in early spring the soil was fertilized with P and K in the dose of 20 and 40 kg ha⁻¹ while the soil content of these nutrients was at the level of 0.294 and 0.179 mg g⁻¹, respectively. Weed percentages were determined by manual biomass separation at each harvest (two per year), and a plot-level weed invasion response was calculated by averaging over all four harvests. The effects of increasing species diversity (up to six-species mixtures) on weed invasion were analysed using Diversity-Interactions models (Kirwan *et al.*, 2009), employed within the beta regression modelling framework (Cribari-Neto & Zeileis, 2010) to handle the proportional response appropriately. DI models can jointly assess the effects of species identity, richness, composition and interspecific interactions on ecosystem functions. Inference was conducted using Wald χ^2 tests. The analysis was implemented in R with the *betareg* package (Cribari-Neto & Zeileis, 2010).

Results and discussion

Compared to other monocultures, the lowest weed invasion was recorded in the L2 monoculture (5.6%), followed by H1 (8.2%) and the HighN community (11.5%) (Figure 1).

The L2 monoculture had significantly lower ($P < 0.05$) weed invasion than all monocultures except H1. No monoculture showed significantly lower weed invasion than the HighN community. Weed invasion decreased significantly ($P < 0.001$) with increasing species richness, with the six-species mixtures exhibiting the lowest weed invasion compared to the average of mixtures containing two-, three-, or four-species. A similar effect of increasing species richness on reduced weed establishment has been documented before, but it was argued that the key factor is not richness per se, but rather the species identity of the dominant species (Sanderson *et al.*, 2012; Suter *et al.*, 2017; van Ruijven *et al.*, 2003). While we observed similar identity effects, we also found significant interaction effects

between species, highlighting that interspecific interactions strongly contribute to weed suppression in mixtures. Overall, mixtures on average (7.6%) had lower weed invasion than all monocultures except L2. Furthermore, mixtures on average had lower weed invasion than both the HighN community (a 34% reduction in weed invasion, $P < 0.05$) and the G1+L1 mixture (a 25% reduction in weed invasion, $P < 0.05$). Despite the substantially higher nitrogen input in the HighN monoculture treatment (100 kg N ha^{-1} per regrowth) compared to the unfertilized mixtures (0 kg N ha^{-1}), none exhibited significantly greater weed invasion, which is remarkable. Furthermore, six mixtures had significantly lower weed invasion than the HighN treatment, underscoring their agronomic potential under low-input conditions. From the composition of the mixtures that perform better than our HighN, we can deduce that a strong species identity effect is driving suppression of weeds. On average, mixtures containing L2 performed significantly better than those without it ($P < 0.001$), and a similar positive effect was observed for the inclusion of H1 ($P < 0.05$). Also, the average of mixtures that include both L2 and H1 is significantly better than the average of mixtures that only contain one of them ($P < 0.001$). This implies that weed invasion can be reduced by including the L2 and H1 species in a mixture.

Conclusion

Mixtures on average exhibited significantly lower weed invasion than both the HighN and G1 monocultures. Increasing species richness reduced weed invasion overall which can be attributed to the strong contribution of the interspecific interaction effect within the mixtures. Additionally, our results indicate that species with specific functional traits (shown by strong identity effects), such as tall growth and fast emergence, contributed to this effect. These traits play a key role in shaping canopy structure, improving light interception and resource acquisition, which are crucial in suppressing weeds. The enhanced performance of mixtures containing L2 and H1 supports the theory that plant height and emergence were key in suppressing weeds under nitrogen-limited systems.

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Root biomass distribution in grassland monocultures and multispecies mixtures

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Abstract

Multispecies grasslands can provide high yields and forage quality while enabling sustainable forage production. Although niche complementarity between species in diverse grasslands is known to increase resource use efficiency, the underlying belowground growth patterns remain poorly understood. Enhancing belowground complementarity may support designing resilient and resource-efficient grasslands. We investigated how species diversity affects root biomass and its vertical distribution in monocultures and multispecies grassland mixtures using grasses, legumes, and herbs. Grassland plots were established in Denmark in spring 2023 as part of the LegumeLegacy project. Aboveground overyielding was observed in mixtures. Root mass was more evenly distributed over depth layers in mixtures than in monocultures (0–50 cm), suggesting a better spatial niche utilization. There was no overyielding of total root mass, likely due to young sward age. However, significant root overyielding occurred at a depth of 25–50 cm depth (51%). This may reflect plastic root growth of particular species, better niche utilization, or complementarity effects. It underlines the importance of deep-rooting herbs in mixtures for sward resilience and ecosystem services.

Keywords: multispecies grassland, root biomass distribution, transgressive root overyielding

Introduction

Diverse grassland mixtures often yield more than monocultures, while maintaining good forage quality and requiring less fertilizer inputs. This is possible due to higher resource use efficiency when combining species from different functional groups with complementary resource use strategies (Kirwan *et al.*, 2009). While aboveground benefits and interactions are widely investigated, belowground growth in temporary grasslands is poorly understood. Previous studies have reported root overyielding and deeper root distributions in diverse swards (Mommer *et al.*, 2010; Mueller *et al.*, 2013), often attributed to complementarity effects and plasticity in root allocation. This study aims to investigate the root biomass and its vertical distribution in monocultures and grassland mixtures of grasses, legumes, and herbs. This may help design resilient pastures that not only deliver high aboveground yields but also grow deeper and more evenly distributed root systems. Such rooting patterns could reduce competition, enhance resource use efficiency and drought tolerance by improving resource uptake from deep soil layers. These grasslands may even provide ecosystem services such as a higher organic matter input and improved soil structure via deep and diverse rooting.

Materials and methods

Grassland monocultures and six-species mixtures of grasses (*Lolium perenne* (LP), *Phleum pratense* (PP)), legumes (*Trifolium repens* (TR), *Trifolium pratense* (TP)), and herbs (*Cichorium intybus* (CI), *Plantago lanceolata* (PL)) were established in spring 2023 in Tjele, Denmark. Due to poor clover establishment, the clovers were reintroduced in August 2023. All plots received 150 kg N ha⁻¹ in 2023 and 173.7 kg N ha⁻¹ in 2024, each split into two applications in spring, and 30 kg P ha⁻¹ and

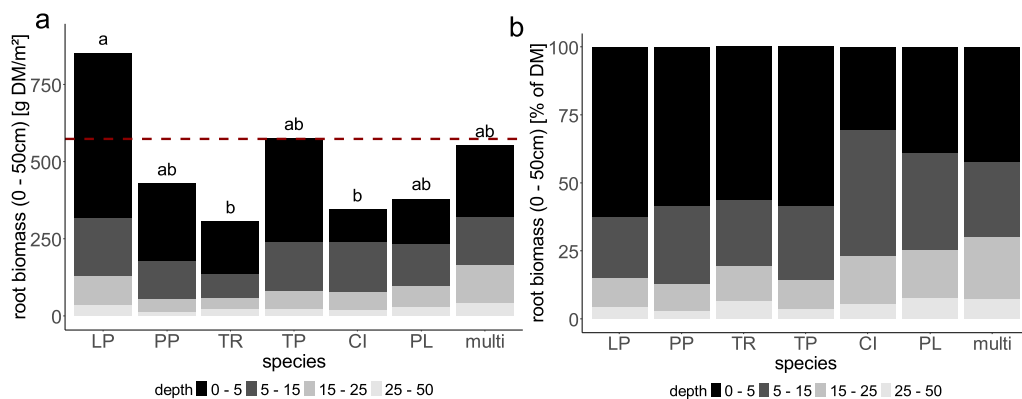


Figure 1. Total root mass (0–50 cm) by depth layer (cm) (a) and the relative proportions (b) in the seven different treatments (LP, *Lolium perenne*; PP, *Phleum pratense*; TR, *Trifolium repens*; TP, *Trifolium pratense*; CI, *Cichorium intybus*; PL, *Plantago lanceolata*; multi = six species mixture), different letters indicate significant differences of total root mass. Dotted line indicates expected root biomass in multispecies plots.

159 kg K ha⁻¹ annually. The plots were harvested five times using a Haldrup forage plot harvester in 2024. The second harvest was done in June 2024, where the species composition was determined by separating subsamples into sown species and weeds. Root samples were taken two weeks prior with a self-constructed soil core installation, mounted on a tractor in monocultures and six-species mixtures (3 replications each). The samples were taken at a 45° angle to a vertical depth of 50 cm and in vertical depth intervals of 0–5 cm, 5–15 cm, 15–25 cm and 25–50 cm. In each plot, three cores with an inner diameter of 5.2 cm were taken and pooled within each depth layer. The samples were stored at 2°C, the roots were washed and then dried at 60°C for 48 h to determine the root dry mass. Linear models and paired t tests were done with base R. Mixed models were defined with the 'nlme'-package at $P \leq 0.05$ significance. Post-hoc multiple comparisons were done with the 'emmeans'-package. Expected root mass was calculated based on observed root mass in monocultures and the recorded aboveground biomass composition.

Results and discussion

The total root mass (0–50 cm) varied significantly between treatments ($P < 0.05$), ranging from 306 ± 47 g DM m⁻² in TR plots to 851 ± 225 g DM m⁻² in LP plots (Figure 1a), with mixtures having 553 ± 228 g DM m⁻². The grass and clover monocultures had 55% (TR) to 62% (LP) of their biomass in the first layer (0–5 cm) and 80% (TR) to 88% (TP) in 0–15 cm depth. The herbs and the multi-species plots showed a more even distribution of their biomass across depths, having only 33% (CI) to 40% (mixtures) of the root mass in the first layer (0–5 cm) and 70% (mixtures) to 73% in 0–15 cm depth (Figure 1b).

Shallower-rooting ryegrass and white clover allocated most of their root mass in upper layers, while deeper-rooting herbs had a more even root distribution, which is in line with the findings of Mommer *et al.* (2010) and Bakker *et al.* (2019). However, no total root mass overyielding occurred in mixtures. This contradicts the findings from long term grasslands, where root biomass increased with species diversity both in upper and lower soil layers (Mueller *et al.*, 2013). Young sward age and limited species richness in our mixtures could explain these results, as overyielding of root biomass increases over time and with diversity (Ma & Chen, 2016). Separation confirmed negligible clover proportions in mixtures (data not shown), likely due to poor establishment. While shallower rooting grasses and

clovers might have been well estimated, total root mass in herb monocultures and mixtures, hence potential root mass overyielding, was probably underestimated in this study. As chicory and plantain can root much deeper than 50 cm already in the first growing season (Czaban *et al.*, 2023; Pol *et al.*, 2024), true total root mass in herb monocultures and mixtures was probably higher than shown here.

Nonetheless, transgressive root overyielding in the deepest layer (25–50 cm, 51%, $P < 0.05$) and deeper root mass distributions in mixtures was found. Mixture yields can increase above- and below-ground when combining species from different functional groups, by reducing competition and enhancing resource use efficiency, leading to complementarity effects and a better niche utilization (Kirwan *et al.*, 2009; Ma & Chen, 2016). The functional group diversity in mixtures might have caused deep root overyielding, possibly through a more plastic root mass allocation (Mueller *et al.*, 2013) or complementarity effects between species. Bakker *et al.* (2019) attributed positive species interactions in their grasslands mostly to the selection effects of dominating species. As we calculated the expected root mass with actual aboveground proportions, selection effects are unlikely to have caused deep root overyielding. To verify assumptions about complementarity effects and spatial niche differentiation, information about the actual belowground composition is therefore needed.

Contrary to our results, Mommer *et al.* (2010) found overyielding mostly in upper layers, where root mass clumped instead of being more evenly distributed, and it was mostly one deep-rooting species occupying deeper layers. Reduced intra-species competition within soil layers and efficient resource uptake across soil layers might have caused the low root mass in upper layers in our case. The small clover proportions and the efficient nutrient uptake in diverse mixtures probably limited the nitrogen availability, possibly causing the deeper or more flexible root distribution (Mommer *et al.*, 2010): Herbs might have been outcompeted by fibrous rooting grasses in upper layers and instead grown more into deeper layers.

Conclusion

Temporal diverse grasslands are not only high yielding but also grow deeper and have more evenly distributed root systems. This underlines their importance to increase resilience and the provision of ecosystems services such as an increased carbon input and better soil structure. However, mixtures did not exhibit root overyielding overall, thus depth-specific analyses are important. Complementarity effects and improved niche use have likely caused deep root overyielding, but differentiated interpretations require the actual root species composition.

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Assessing water management in grassland-based sheep and beef systems in Wales

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Abstract

Water is a key component of sheep and cattle production in Wales. High rainfall and natural water features make a significant contribution to the sustainability of grassland-based ruminant systems. Potable and mains water can represent significant financial and carbon costs when used in farming. To determine the sources of water used in sheep and cattle systems in Wales, Hybu Cig Cymru levy payers were surveyed using an online questionnaire. There are a number of management techniques that can be deployed to minimise the run-off of water and soil nutrients. These are becoming increasingly important as the prevalence of extreme weather events increases across the globe. Respondents were asked which methods, if any, they used to minimise run-off on their farms.

Introduction

The aim of the pilot survey was to quantify the source and volume of water utilised for red meat production, and to explore soil and water management methods in order to set a baseline and identify any areas for improvement. In Wales, beef cattle and sheep benefit from water that is naturally plentiful across many farms including their own private water supply, such as boreholes and springs. There can also be a need to use mains water. There are positive implications for both the cost of production and carbon audits where mains water is not the primary source of water.

Materials and methods

The English and Welsh bilingual survey was accessed via the online platform ‘Survey Monkey’ and was open from 3–17 March 2025. There were 10 questions relating to farm information, livestock numbers and water usage and management. Red meat producers in Wales were targeted through social media platforms, email newsletter and in-person attendance at livestock auction markets and events, along with targeted emails through the Hybu Cig Cymru (HCC) levy board producer database.

Results and discussion

A total of 221 respondents participated in the survey. The majority were upland farms (46%), followed by hill (29%), and then lowland (25%). Across the 221 farms, the respondents recorded a total of 142 613 head of animals: 11 974 head of beef animals, 1294 dairy animals, and 129 345 head of sheep and lambs. Most of the beef animals (47%) were located on upland farms, with just over a third (31%) on lowland farms. For sheep and lambs the majority (46%) were found on upland farms whilst a further 38% of sheep were situated on hill farms.

The survey asked respondents to identify their primary sources of water for red meat production on their farm, with the option to select more than one source. A total of 181 responded to this question, with the majority (73%) noting they only used natural sources of water (i.e., surface, ground or rain water) and did not use any mains water at all as their primary source. A further 22% noted they used both mains water and natural sources for their red meat production, whilst just 4% of respondents

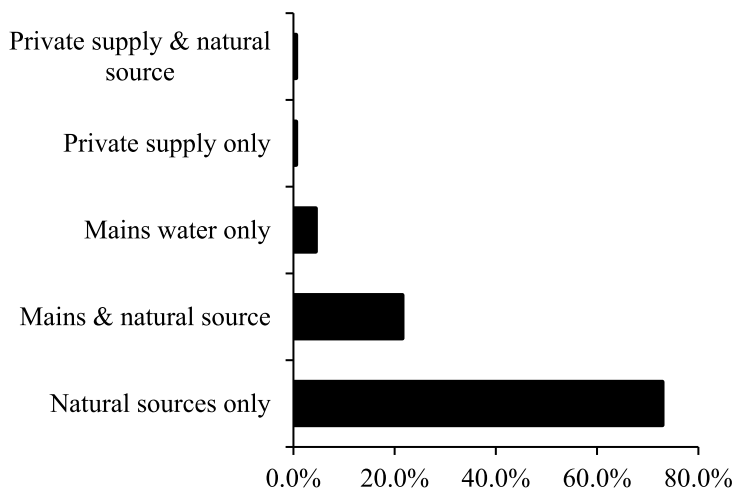


Figure 1. Survey responses to the question “What are your primary sources of water for red meat production? Please select all that apply”. The percentage of respondents is shown.

selected mains water as their sole supply of water on the farm. Around 1% noted the use of a private supply.

Of the respondents that used a natural water source in their red meat production system (whilst they were able to select more than one source as an option) the majority (74%) utilised surface water (rivers, streams, ponds, springs, etc); a further 55% utilised groundwater (such as boreholes and wells etc) and 29% utilised rain water (collected or direct use) as a source. This suggests that mains water remains supplementary, highlighting the opportunity for lower production costs and carbon emissions associated with potable water treatment.

Of the respondents that had a beef-only farm; the majority (63%) utilised only natural sources for water, with just 11% relying solely on mains water, and 26% on a combination of both natural and mains. Sheep-only farms also mainly utilised natural sources (65%), with 30% utilising both natural and mains, and just 5% solely on mains. Mixed farms (sheep and beef) utilised more natural water sources in comparison to both beef- and sheep-only farms, with fewer relying on mains-only water (3%). Farms situated in lowland areas appear to utilise the most natural water sources, followed by hill farms, whilst those situated in upland areas tend to utilise more mains water.

As water and soil management are closely linked, the survey asked the respondents if they used any methods to reduce runoff from their fields. Among the 162 respondents for this question, there was clear variation in the uptake of runoff reducing methods, as shown in Table 1 below. Seventy percent of respondents stated that they used one or more of the soil and water management techniques to minimise runoff and retain nutrients. No-till/reduced-till farming (19%) and buffer zones/drainage systems (14%) were most commonly implemented methods within the survey. Grassed waterways, wetlands, and cover crops were adopted less.

Results from upland farms were variable and reported the highest use of mitigation but variation in farm size, exposure, or agri-environment incentives may have influenced this. Hill farms have a modest but more balanced uptake, particularly in engineered solutions like leaky dams and wetlands.

Table 1. Results from the survey question: “Water and soil management are closely linked. Which of these methods do you use to reduce runoff from fields?”

	Buffer zone	Nothing	No-till/reduced-till farming	Retention ponds/wetlands	Scrapes	Leaky dams	Grassed waterways	Cover crops	Coir logs	Controlled/subsurface drainage
Farm type										
Hill	2	9	5	3	1	2	2	1	0	4
Lowland	4	4	4	2	0	0	1	2	1	4
Upland	7	17	10	2	1	1	4	2	0	6
Livestock species										
Beef	1	4	2	1	0	0	1	1	1	1
Sheep	6	8	7	2	0	1	1	1	0	6
Beef and sheep	7	17	10	4	2	2	5	4	0	7
Grand total	14	29	19	7	2	2	7	6	1	14

Data show the percentage of respondents that selected a specific management method, within either farm type or livestock species.

Lowland farms adopted techniques such as buffer zones and no-till/reduced-till farming, which reflect the farming systems; this is perhaps due to less acute runoff risk.

Sheep-only farms lean more toward natural mitigation (e.g. cover crops and reduced tillage), possibly reflecting grazing patterns or soil health priorities. Mixed farms (beef+sheep) show the broadest engagement with mitigation strategies, likely owing to more diverse operations, economies of scale, or increased environmental opportunity. For example, grassed waterways were mostly utilised by mixed farms, likely benefitting from larger scale or varied terrain.

This survey confirms that Welsh sheep and beef farms predominantly rely on natural water sources, with 73% of respondents using natural water sources exclusively, and only 26% supplementing with mains water. Within the 26% who noted use of mains water, just 4% recorded that mains water was their exclusive water source. The sector’s deep connection to Table 1.

regional hydrology underscores its resilience but also its vulnerability to climatic shifts. Based on the survey results, it is evident that water runoff management practices across Welsh red meat farms are widespread but vary significantly. While some producers are actively engaging in soil and water management techniques, adoption rates are variable—particularly among beef-only and lowland farms. It would be useful to further explore the context of the of these methods, as not all methods may be inappropriate on some farms.

Conclusions

Enhancing runoff control will not only improve production efficiency, and soil and ecosystem health, but also ensure compliance with regulation. It could be an opportunity to enhance the reputation of Welsh red meat as a responsible, high-quality product in increasingly sustainability-conscious markets. This baseline demonstrated that as use of natural water sources is significantly higher than mains water, and the implications for costs and carbon emissions will be positive compared with standard global figures. Strengthening awareness and delivering tailored guidance on runoff management will bolster environmental stewardship and enhance the sustainability credentials of Protected Geographical Indication (PGI)Welsh Lamb and PGI Welsh Beef.

Acknowledgement

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The resilience of forage mixtures: superior performance over monocultures under climate change

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Abstract

Climate change threatens to reduce the yield in grassland ecosystems. Sown diversity could be a crucial adaptation strategy for maintaining grassland productivity and increasing resilience to extreme climatic events compared to monocultures. Grasslands with higher sown diversity tend to be more resilient to drought, due to a more efficient resource use across the niche space and mutualism among different species. Multi-species mixtures in intensively managed grasslands show strong evidence for yield advantages that may be enough to out-yield the best-performing monoculture. This paper investigates how climate change, local climate, soil conditions, and management practices impact the benefits of average mixtures compared to the best-performing monoculture in terms of productivity, weed suppression, and forage quality. Based on the COST Action 852 Experiment (2002–2011), our analysis included data from 31 locations across Europe, comprising 30 grassland communities, consisting of 4 monocultures and 11 mixtures of 4 species, at high and low sowing densities. We employed a hierarchical mixed-effects linear model approach to investigate the relationship with environmental variables. Our results show that the benefits of sown diversity were maintained for yield at positive temperature anomalies and moderate precipitation shifts, weed suppression at low P fertilization and moderate precipitation anomalies, and forage protein under negative precipitation anomalies without fertilization.

Keywords: climate change, grassland, mixtures, monoculture, sown diversity, transgressive overyielding

Introduction

The decline of biodiversity has fuelled many experiments quantifying the relationship between different biodiversity levels and ecosystem functioning (Hector *et al.*, 1999; Kirwan *et al.*, 2007). Multiple studies addressed the transgressive effect, where the average mixture performs better than the best-performing monoculture, explicitly focusing on transgressive overyielding and weed suppression (Connolly *et al.*, 2018; Kirwan *et al.*, 2007). Under the current climate change scenario and with agricultural systems facing numerous challenges (IPCC, 2022) It is essential to understand how the transgressive effects vary under different climate and soil conditions (Lüscher *et al.*, 2022). Our paper examines the environmental conditions under which the average mixture surpasses the best monoculture, and the effects of climate anomalies on sown diversity benefits over diverse forage yield, weed suppression, and forage protein content.

Material and methods

We used the COST Action 852 Agrobiodiversity Experiment data (2002–2011) (Kirwan *et al.*, 2014) to investigate the overperformance indices of sown forage mixtures over monocultures, the average performance of mixtures compared against the best performing monoculture for yield (transgressive over

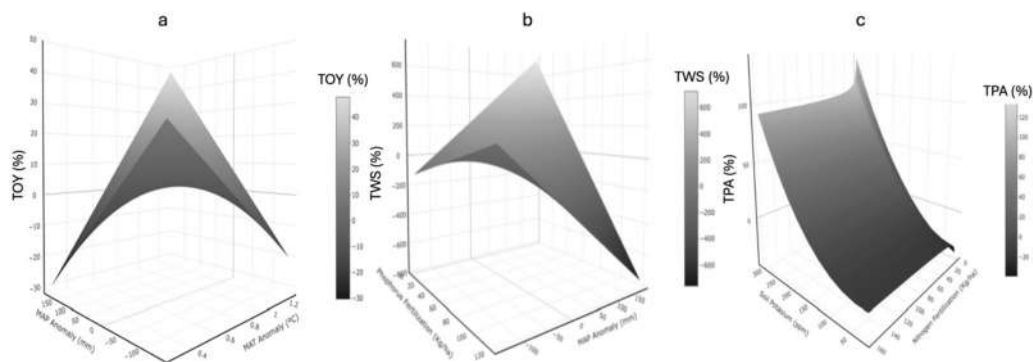


Figure 1. Relationship between (a) TOY and the interaction of MAT anomaly with MAP anomaly. (b) TWS and the interaction of MAP anomaly with phosphorus fertilization. (c) TPA and the interaction of soil potassium with nitrogen fertilization.

yielding, TOY), weed suppression (transgressive weed suppression, TWS) and protein content (transgressive protein availability, TPA). A simplex design was implemented at 31 locations (30 in Europe and 1 in Canada, including a total of 30 grassland communities per site, consisting of 4 monocultures and 11 mixtures of 4 species (2 legumes and two grasses), at high and low sowing densities (low being 60% of high) (Connolly *et al.*, 2001). Climate data were acquired from WorldClim 2.1 (Fick & Hijmans, 2017), and their corresponding anomaly variables (centred around 0) were calculated by subtracting the averaged data between 1970 and 2000 from the period averages of the experiment per site.

We used mixed effects linear models to assess how climate, management, and soil variables shaped the overperformance indices. We checked for variance inflation factor and curved relationships (squared or log-transformed). We added drivers according to a fixed sequential order: local climate, anomaly variables, management, soil texture, and soil nutrients (Rodríguez *et al.*, 2020). Model terms were selected automatically by a genetic algorithm included in the R package *glmulti*, using the corrected Akaike information criterion (AICc). Explanatory power of the final models was evaluated by the marginal variance explained by the fixed effects, using the R package *MuMIn*.

Results and discussion

TOY was maintained under warming (MAT anomaly) of +1.3°C and a MAP anomaly of −40 mm and increased with increasing MAP anomaly. The diversity effect on yield (TOY) increased with warming at moderate anomalous increases in precipitation but decreased again when both the precipitation deficits and the warming anomalies increased (Figure 1a), as reported by Sebastià *et al.* (2024). TOY was found to be negative in very dry and hot or very wet and cold conditions (Smith & Dukes, 2013). Our results are aligned with other studies that demonstrate precipitation as the primary climatic factor influencing grassland productivity, as well as temperature (Ghani *et al.*, 2022).

Phosphorus fertilization regulated the relationship between the positive effect of sown diversity on weed suppression and precipitation anomalies: TWS decreased with phosphorus fertilization above 72 kg ha^{−1} per year when the precipitation anomalously increased above 106 mm (Figure 1b). This suggests that the application of fertilizer weakened the nutrient competitiveness of forage mixtures against weeds (Nadeem *et al.*, 2018). The benefits of sown diversity over the forage protein content (TPA) were related to soil nutrients and decreased with decreasing soil K concentrations (Figure 1c);

however, this effect was slightly amplified when no N fertilization was applied. TPA was more related to soil potassium than nitrogen fertilization concentrations (Dietrich et al., 2023; Obasoro et al., 2025).

Conclusion

The positive effect of mixtures on TOY was held under moderate climate changes, but it was still very sensitive to strong climatic anomalies. At the same time, fertilization negatively affected TWS and TPA. It could interact with climate and soil nutrient status, highlighting the strong influence of climate change and management practices on the benefits of sown diversity. Our models suggest that sown diversity offers a promising strategy to enhance resistance and resilience in the face of an uncertain climatic future, under some thresholds. It can also boost forage production and quality, reduce farmers' reliance on agrochemicals, and improve agricultural systems' sustainability, while supporting plant and animal biodiversity. This paves the way for future studies involving drought-tolerant species and trait combinations that strengthen diversity effects.

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Developing productive buffers on livestock farms to reduce water run-off from grasslands

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Abstract

The aim was to develop farmer-driven solutions to reduce risks to water quality through optimal farm management practices, with a focus on soil resilience and biodiversity. Water infiltration into soils is one factor that can influence the likelihood of surface water run-off from agricultural land. In this study, the potential role of different productive buffer strips was monitored for changes in soil water infiltration and soil biodiversity, to help capture diffuse pollution. A key distinction of the work was that the buffers were utilised for agricultural production. Four commercial farm sites on the project, each with buffer containing two forage treatments in triplicate plots (9 m long×6 m wide) established in summer 2023 (Year One), with red clover (*Trifolium pratense*) as Treatment 1 and perennial ryegrass (*Lolium perenne*) as Treatment 2. Water infiltration (WIR) measurements taken each autumn to spring in 2023–2024 (Period 1) and 2024–2025 (Period 2) differed between red clover and ryegrass plots in Period 2 in Oct to Feb ($P < 0.05$), with a tendency in March 2025 ($P = 0.08$). Earthworm biomass and numbers determined spring and autumn in 2024 were higher under red clover compared to ryegrass in autumn 2024 ($P < 0.05$).

Keywords: flooding, forages, red clover, soil and water management, water infiltration

Introduction

Grasslands have an important role both locally and globally in many ecosystem functions, including water resource management (Bengtsson *et al.*, 2019). There is an urgent need to develop and manage grasslands to help alleviate flooding risk. Given that grasslands cover 30% of the land area globally, with 70% of the global agricultural area as grasslands, this equates to a total of 3.5 billion hectares (Lemaire *et al.*, 2011); providing a substantial platform to help alleviate the impacts of climate change. The use of contrasting forage species, such as clovers, plantain and chicory, in grassland-based ruminant systems can help boost livestock productivity whilst reducing the environmental impact of these systems. Water infiltration into soils is one factor that can influence the highlighted how a shift from perennial ryegrass (PRG)-dominated pastures to swards with more contrasting forage species may provide an ecohydrological approach to mitigating flood risk. Replicated studies on monospecific swards of red clover (*Trifolium pratense*), white clover (*Trifolium repens*) and chicory (*Cichorium intybus*) species, compared to perennial ryegrass (*Lolium perenne*) swards, significantly increased WIR (Marley *et al.*, 2024). Here, we present the findings from the application of this knowledge to develop productive buffer zones on commercial livestock farms. A key distinction of the work was that the buffers were utilised for agricultural production, not set aside from the farm, with areas being either grazed or cut for silage as part of the farm system. The objective was to determine whether the use of contrasting forages in these buffer zones would still provide benefits for reducing surface water run-off once under a commercial farm setting.

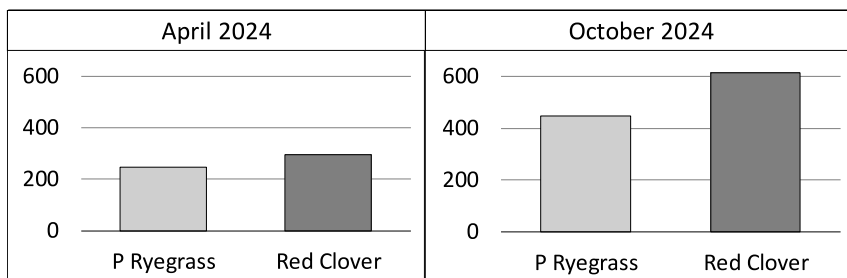


Figure 1. Earthworm biomass (g m^{-2}) and abundance (numbers m^{-2}) determined spring and autumn in 2024.

Materials and methods

The farm sites included a range of conventional and organic beef, sheep and dairy farms located in the Bannau Brycheiniog (Brecon Beacons) National Park in Wales, UK. Prior to buffer establishment, soil samples were taken to a depth of 15 cm and soil nutrient indices for P and K standardised across sites through fertiliser application and lime applied to achieve pH 6.2. Forage buffers at each site included two forage treatments sown in triplicate plots (9 m long $\times$ 6 m wide) within each buffer in June 2023 (Year One). All farms had Treatment 1 as red clover (cv. AberClaret) sown at a rate of 16 kg ha^{-1} and Treatment 2 as perennial ryegrass (cv. Aberwolf) sown at a rate of 35 kg ha^{-1} in each buffer. A third forage option (for example, lucerne (*Medicago sativa*) or plantain (*Plantago lanceolata*)) was included, as chosen by the farmer, but this third treatment was not included in the analysis and findings presented here as data were not comparable. Forage measurements were taken to validate that the forage treatments were as described and included plant populations after establishment in 2023 and the forage yields and botanical composition during the first harvest year in 2024 (data not shown). Measurements of WIR were taken on all replicate plots ($n = 24$) on four dates each autumn to spring in 2023–2024 (Period 1) and 2024–2025 (Period 2). WIR measurements were taken on the third dry day after heavy rain (when soils were at field capacity) using a double ring infiltrometer (30 cm $\varnothing$) method described by Gregory *et al.* (2005). Earthworm biomass and numbers were determined in spring and autumn in 2024. Earthworm biomass (g m^{-2}) and abundance (number m^{-2}) were quantified by hand-sorting from a cube of soil 30 cm $\times$ 30 cm $\times$ up to a depth of 30 cm from each plot. For the statistical analysis, Treatment 1 and 2 forages established on four farm sites in spring of Year One were compared. For these forage treatments, WIR and earthworm data was analysed using REML. WIR and earthworm data was either Log^{10} or square root transformed to normalise the data prior to statistical analysis. All data analysis was performed using Genstat[®] for Windows[™] (22nd edn.) and $P < 0.05$ taken as significant.

Results and discussion

There were no differences between treatments in spring 2024, but both the earthworm numbers and biomass were higher under red clover plots compared to ryegrass plots in autumn 2024 (Period 2) ($P < 0.05$) (Figure 1). WIR differed between red clover and ryegrass plots in Period 2 in October, December, February 2025 ($P < 0.05$), with a tendency for higher WIR on red clover plots in March 2025 ($P = 0.08$). Numerical differences were also observed in Period 1, but the differences were not significant ($P > 0.05$) (Figure 2). REML analysis comparing square root adjusted data at each time point showed a similar pattern in earthworm numbers and abundance as was found for WIR. Although this suggested that earthworms may be a key driver of increased WIR under red clover on the farms,

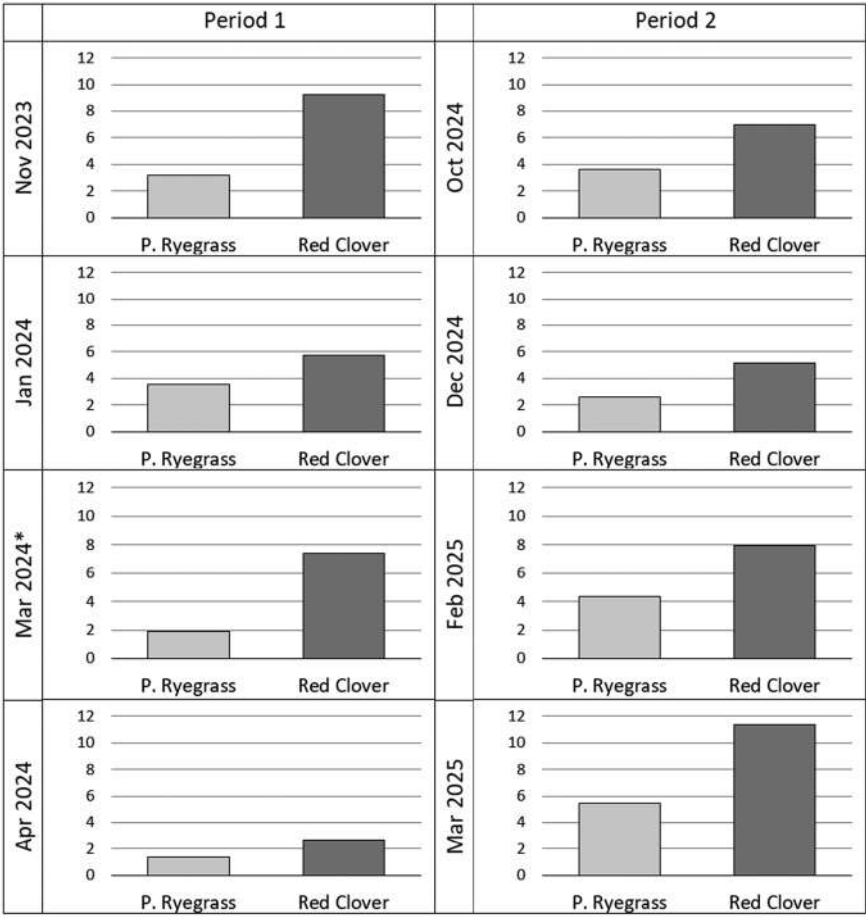


Figure 2. Mean WIR (cm h⁻¹) from four sites 2023–2024 (Period 1) and 2024–2025 (Period 2). *3 sites on this date as wet weather prevented further measurements.

in other work (Marley *et al.*, 2024), it was the root diameter of the red clover that was the main underlying mechanism by which this forage was altering WIR.

Conclusion

Overall, the findings showed that the use of productive buffers on farms had huge potential to reduce surface water run-off from agricultural land. Further work is needed to understand how these forages alter soil resilience to grazing and machinery compaction to ensure these buffers continue to protect water under different management when utilised on commercial farms.

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Assessing soil organic carbon in grasslands with hybrid dynamic modelling

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Abstract

Soil carbon sequestration (SCS) is increasingly recognized as an effective means of mitigating atmospheric carbon. To enhance the accuracy of SCS models, this study introduces a hybrid modelling framework that integrates a process-based model (PBM) with a machine learning (ML) approach and applies it to estimate soil organic carbon (SOC) in Portuguese grasslands. For the PBM we used a simplified zero-dimensional SOC model, and the ML algorithms were artificial neural networks (ANNs). These ANNs were calibrated to estimate the two PBM parameters: carbon input (K) and the mineralization rate (α). Through random variable selection aimed at dimensionality reduction and a versatile network architecture, the ANNs effectively leveraged diverse satellite-based radiometric, soil, and climatic datasets. The resulting hybrid model yielded R^2 values of approximately 0.71 for the training dataset, and around 0.43 for validation and testing datasets (root mean squared errors approximately 0.49 for training and 0.64 for validation/testing sets). Feature importance analysis further underscored significant contributions from variables such as soil moisture, temperature, and vegetation indices. By combining mechanistic knowledge with machine learning techniques, this proposed framework provides a reliable, interpretable, and scalable solution for improved SOC modelling, thus bolstering soil carbon sequestration programs and enhancing global carbon offsetting initiatives.

Keywords: artificial neural networks, carbon sequestration, machine learning, process-based modelling, soil organic matter, sown biodiverse pastures

Introduction

The increase in atmospheric CO₂ is a major driver of climate change. Addressing this challenge requires not only emission reductions but also strategies to remove carbon from the atmosphere through technological and land-based negative emissions solutions. Among these, boosting soil organic carbon (SOC) through improved land management has garnered attention for its dual benefits: enhancing carbon sequestration while supporting climate adaptation, reversing land degradation, and improving food security (Amelung *et al.*, 2020; Bradford *et al.*, 2019).

However, direct measurements of SOC changes are logistically demanding and resource-intensive, limiting their feasibility for large-scale monitoring. To overcome these limitations, researchers have increasingly turned to modelling approaches. Two main strategies dominate: process-based models (PBMs), which rely on ecological principles to simulate soil carbon dynamics, and machine learning (ML) models, which detect complex patterns in data without assuming predefined relationships. While PBMs are grounded in theory, their accuracy often suffers when applied outside the conditions for which they were calibrated. ML models, though flexible and data-efficient, lack the mechanistic grounding to predict long-term responses to disturbance or climate shifts. A recently proposed alternative is the use of hybrid modelling to blend the strengths of both (Zhang *et al.*, 2024). This study

develops a framework that uses an ML approach, artificial neural networks (ANNs), to estimate the parameters of a simplified PBM for SOC modelling. Applied to biodiverse pasture systems in Portugal (Teixeira *et al.*, 2015), the model aims to produce spatially explicit, physically consistent predictions of SOC change that support scalable assessments of land-based carbon sequestration potential.

Materials and methods

This study presents a hybrid modelling approach that couples the simplest 0D dynamic process-based model of SOC dynamics taken from Teixeira *et al.* (2011), with only two parameters: carbon input (K) and mineralization rate (α), according to

$$\text{SOM}(t + \Delta t) = \frac{K}{\alpha} \cdot \left(1 - e^{-\alpha \cdot \Delta t}\right) + e^{-\alpha \cdot \Delta t} \cdot \text{SOM}(t). \quad (1)$$

Unlike conventional modelling workflows where parameter estimation precedes simulation (Morais *et al.*, 2018), two ANNs in this framework represent the two parameters and are integrated into the model's training loop. During training, predicted values of K and α are directly used within the SOC equation, and the loss function is computed as the deviation between modelled and observed SOC values. This method ensures that machine learning predictions are continuously informed by the physical model. The two ANNs use flexible architectures with randomized input selection to reduce overfitting and prevent bias. A ReLU-activated hidden layer is followed by dropout for regularization, and final outputs are passed through constrained activation functions (Softplus for $K > 0$ and a bounded sigmoid for α ensuring that mineralization is lower than 20%, as expected from the literature), ensuring that predicted parameters remain within physically meaningful ranges.

The model was trained and validated using a multi-year dataset (four years of production from 2017–2018 to 2020–2021) of paired SOC measurements from eight grassland farms in Portugal and one in Spain, including radiometric, climatic, and soil variables. By combining satellite imagery, soil data, and topographic metrics, the model generates location-specific parameter estimates without requiring site-by-site recalibration. The total number of SOC observations was 1074. To improve generalization, the dataset was partitioned into training, validation, and testing sets (60/20/20 split), with hyperparameters optimized via Ray Tune. The final architecture leveraged SHAP (Shapley Additive Explanations) values to assess variable importance, offering insight into which predictors most influenced K and α . Weights were initialized using Xavier uniform distribution, and a learning-rate scheduler was implemented to refine convergence.

Results and discussion

The optimized hybrid model showed robust performance across all data partitions. The ANN predicting K used a larger hidden layer, while the one estimating α benefitted from moderate regularization. Hyperparameter tuning revealed that intermediate values for dropout and random input selection struck the best balance between model complexity and generalization. On the training set, the model achieved a high coefficient of determination ($R^2 \approx 0.71$) and a root mean squared error (RMSE) below 0.49. While R^2 values dropped to approx. 0.43 on the validation and test sets, predictive accuracy remained stable, suggesting only minor overfitting. Average K estimates were around 0.82 kg SOC per 100 kg soil annually and α averaged near 0.18. Importantly, the distributions of predicted K and α values were consistent across splits, confirming the model's capacity to generate physically plausible outputs even when faced with unseen data.

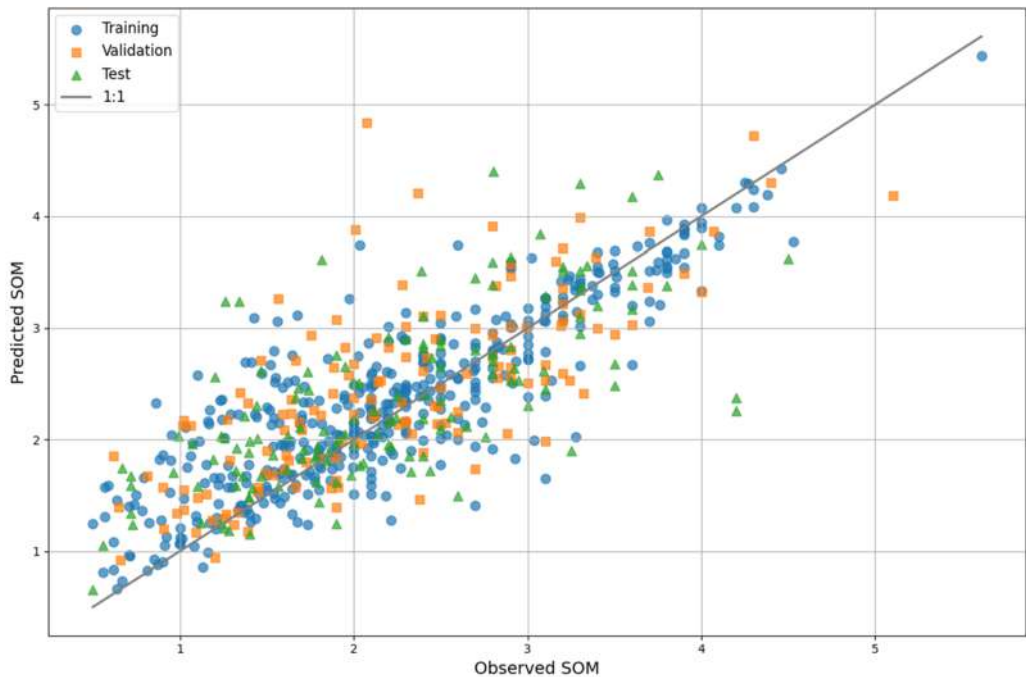


Figure 1. Scatter plot of the predicted versus observed soil organic carbon (SOC) values for the training (blue circles), validation (orange squares), and test (green triangles) datasets, with the 1:1 reference line included.

An analysis of feature influence using SHAP values provided additional interpretability. For K , predictors related to soil moisture, temperature, and pH dominated the model's output, underscoring the role of immediate edaphic and climatic conditions in controlling carbon inputs. In contrast, α was more responsive to topographic metrics and vegetation indices such as NDVI and SAVI, highlighting the importance of microclimatic variation and plant activity in shaping decomposition rates. The resulting model accurately replicated observed SOC changes, with most predicted values aligning closely with the 1:1 line in scatter plots across training, validation, and test sets (Figure 1).

Conclusion

This study developed a hybrid modelling approach that integrates process-based equations with ANN to estimate SOC dynamics in grasslands using farm-sourced and remotely sensed data. By combining physical interpretability with the flexibility of data-driven learning, the model produced accurate, spatially explicit predictions of carbon input and mineralization. Its consistent performance across datasets and ability to generalize across sites highlight its potential as a scalable tool for soil carbon monitoring and climate mitigation planning. Overall, this hybrid approach not only advances predictive accuracy but also retains scientific interpretability, making it suitable for both operational forecasting and policy-relevant applications in soil carbon monitoring.

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Impact of climate change on the organic extensive livestock farming sector of the Catalan Pyrenees

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Abstract

Mediterranean grasslands face low productivity due to increased warming above the global mean and to decreased precipitations. Therefore, the Catalan Pyrenees, on the west side of the Mediterranean, constitute a vulnerable region within the climate change framework. Extensive livestock is the traditional grassland management in this region. In order to identify methodologies for practice adaptation of the organic extensive livestock sector to climate change, we conducted fifteen interviews and four focus groups with key stakeholders in four locations in the Catalan Pyrenees. We developed a cognitive mapping analysis with the interview data, detecting the main problems, causes, consequences and measures for climate change adaptation, which were presented in each focus group to ground the information in the studied areas. We determined a lack of forage and water scarcity as the main problems, with effects on both livestock and ecosystems. Some measures proposed were a re-assessment of the number of heads of livestock per exploitation, the building of water troughs, and the promotion of ovine-caprine livestock whose numbers have been reduced in the last decades. This information serves to propose good practices for climate change adaptation in the Catalan Pyrenees.

Keywords: extensive farming, livestock, organic, Pyrenees

Introduction

The emergence of organic production schemes has brought about new opportunities for grazing livestock farming to counteract the current trend of abandonment. This is particularly the case for pastoralism in the Mediterranean basin, as in Spain, with ruminant pasture-based farming systems and the production systems based on ruminant grazing, where conventional and organic management are similar (López, 2020). Extensive organic livestock farming is a fundamental pillar for the sustainability of mountain areas. In Catalonia, this production model combines environmental, cultural and economic values. However, climate change is testing its viability, and the literature indicates the limited information available on the impacts, vulnerability and adaptation to climate change of extensive livestock production systems at the Spanish level (Rubio 2017). Our objective was the identification of organic management systems that are being applied in extensive livestock farms located in specific areas of the Catalan Pyrenees around several natural parks, through a qualitative diagnosis with stakeholders, including farmers and the administration.

Materials and methods

We conducted fifteen semi-structured interviews with livestock farmers, technicians and managers of natural parks in the Catalan Pyrenees, and three participatory territorial workshops held in Ripoll, Berga and Pont de Suert, municipalities east-west across the region, who were representative of each

Table 1. Ecological livestock farms and pasture surface in Catalonia

	2015	2016	2017	2018	2019	2020	2021	2022	2023	2024
Livestock farms (n)	791	834	913	927	971	1017	1092	1096	1135	1122
Meadows, pastures and fodder (km ²)	902.8	1178.2	1395.9	1457.6	1601.1	1792.3	1830.7	1810.6	1880.6	1860.6

territory. The questions covered the following topics and were answered individually and extensively: (i) management of the agricultural and livestock operation for adaptation to climate change; and (ii) socioeconomic perspectives: aims to collect the current challenges while establishing organic production; the steps to make production more sustainable in the face of climate change; support and training; and the situation of women in the sector. The information obtained allowed the development of an analysis of problems regarding the adaptation of livestock farming to climate change, with special emphasis on the ecological model, to identify possible solutions.

Results and discussion

As of 2024, Catalonia had 1122 organic livestock farms registered in the CCPAE (Table 1). The sector has experienced a cumulative growth of 45.53% in the last decade, with a peak of 1135 farms registered in 2023. The predominant subsector continues to be beef cattle.

The mountain regions located around the Natural Parks included in this study concentrate a significant number of farms benefiting from subsidies from the PDR.cat programme. In 2022, those organic farms covered 95 228 ha, benefiting 644 exploitations in 28 municipalities. In comparison, in 2016 they covered 60 289 ha. This increase reflects a progressive consolidation of the sector in the mountain regions.

Organic farms in the Pyrenees provide clear benefits: they maintain the agroforestry mosaic, promote biodiversity and contribute to food sovereignty (Vargas et al., 2020). However, the results from our qualitative analysis have allowed the identification of several structural tensions that especially affect organic farms in the Pyrenees (Table 2).

First, the lack of fodder and the limitation of water resources stand out as direct effects of the recurrence of intense droughts. This situation reduces the availability of natural food for livestock, raising production costs and causing problems in the viability of farms. Secondly, a high dependence on public aid was detected, especially those linked to the number of livestock. This allocation criterion can induce a livestock overload above the real capacity of the pastures, generating pressure on the environment.

In addition, interviews identified that many farms do not close the production cycle, which limits their capacity for self-sufficiency and contribution of organic matter to the soil. That is, most farms concentrate their activity on the cows that raise the calves, but do not fatten those, neither do farmers take advantage of the produced organic matter to fertilize the meadows. This fragmentation reduces circularity and increases dependence on external resources.

Finally, there has been a change in the composition of livestock compared to recent decades, with a reduction in the number of sheep and goats, species historically linked to the territory, and which allowed for better management of the mountains, cattle being currently favoured.

This phenomenon is also associated with the loss of active shepherds in the high pastures, often linked to the ageing of the sector and the lack of generational change, plus the economic diversification of the livestock farmers themselves.

Table 2. Simplified framework of structural problems for the management of operations

Structural causes	Central problem	Direct consequences	Proposed measures
Dependency on subsidies per head. Change of model (more heads, less land). Non-closure of the ecological cycle.	Lack of forage and water	Pressure on wet ecosystems. Reduced fertility of meadows. Increased costs. Dependence on public subsidies	Installation of water troughs. Local slaughterhouses. Direct sales and cooperatives. Close the cycle with fattening. External consulting.

Conclusion

The results of this work point to the need to strengthen the structural elements that make a more resilient ecological livestock model possible. The closure of the cycle, understood as the capacity to complete all phases of the production process within the farm or in a network with other actors in the territory, emerges as a central strategy to improve autonomy, reduce external dependence and provide added value to products. In parallel, the importance of a system of public subsidies more adapted to the territorial reality of the mountain, which recognizes the environmental and social benefits of this type of livestock farming, becomes evident.

At the environmental level, the focus must be placed on the recovery of pastures, the improvement of irrigation systems and resilience to droughts, while favouring the conservation of natural resources. This approach should be accompanied by a regulatory reform that makes administrative requirements more flexible and allows better adaptation to the particularities of mountain farms.

Finally, the future of the sector will also depend on its ability to attract and retain new professional profiles. For this reason, it is essential to continue promoting the incorporation of young people and women, facilitating their training and ensuring their access to land, resources and support networks.

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Effects of water availability on forage quality of an intensively managed mountain pasture

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Abstract

On intensively used pastures in mountain areas, high forage quality is essential to ensure a high milk production performance, with a reduced use of concentrates. However, in addition to natural seasonal variation, forage quality is influenced by extreme weather events, which are expected to become more frequent due to climate change. When forage quality changes, grazing systems, as well as supplementary feeding, must be adjusted to maintain animal health and production. Therefore, knowledge about quality of pasture herbage and its seasonal dynamics under varying meteorological conditions is crucial. Using yield and forage quality data from a simplified Corral-Fenlon on a short sward pasture in Dietenheim/Bruneck (South Tyrol, NE Italy), we investigated the effects of water availability expressed by means of the aridity index (ratio between precipitation and potential evapotranspiration sum in the 4 weeks preceding sampling) on crude protein (CP) and neutral detergent fibre (NDF) estimated by NIRS over a 5-year period. We found that the effect of the aridity index shifted over the course of the year. In spring, lower aridity index values—indicating drier conditions—were associated with higher CP and lower NDF, whereas in autumn they were linked to lower CP and higher NDF.

Keywords: aridity index, climate change, crude protein, NDF, short sward grazing

Introduction

In mountainous areas, intensively used pastures are a cheap and convenient way to feed high-yielding dairy cows. Their forage quality can be very high but is subject to constant change. This poses challenges for farmers, as unstable forage quality can have negative effects on production and animal health. Therefore, grazing systems and supplementary feeding must constantly be adjusted accordingly. Fibre content (typically estimated by NDF (neutral detergent fibre)), as well as crude protein (CP), are key forage quality parameters. Their annual variation due to plant phenological development is well documented, but predictions for very early or very late growth stages—often observed in grazing—are less certain. Additionally, varying meteorological conditions can have an impact (Grant *et al.*, 2014) which is still poorly understood. The aridity index, which considers rainfall as well as the potential evapotranspiration, is a well-suited tool for assessing weather conditions over a certain time-period. Since both extreme drought and heavy rainfall are expected to become more frequent because of climate change, reliable information about forage quality of pastures under unpredictable meteorological conditions is needed to adapt feeding and grazing strategies. The objective of this study is therefore to evaluate how the aridity index affects herbage quality to allow informed decision about grazing management and animal feeding.

Materials and methods

We used yield and forage quality measurements (224 for CP and 221 for NDF) on a short sward pasture with three paddocks in Dietenheim/Bruneck (South Tyrol, NE Italy; 46°48'10.3"N 11°57'28.3"E, 903 m asl), in which the growth rate was monitored over a 5-year period from 2019 to 2023. In each paddock, two areas of 10 × 1.35 m were mown alternately every two weeks from May to August (simplified Corral Fenlon according to Mosimann, 2001), collecting forage samples which were dried at 60°C until constant weight. CP and NDF content were analysed by NIRS (NIRSystem 5000, Foss, Hillerød, Denmark). For each sampling event, the value of the respective aridity index (AI) was calculated using weather data over the last four weeks. For the estimation of the potential evapotranspiration (PET), the method of Penman Monteith was used (Allen *et al.*, 1998). For the calculation of growing degree days (GDD), the base temperature was set at 5°C and the cumulation began at the estimated start of the vegetation period (Schaumberger, 2011). Twelve observations with AI values beyond 4 (5.3 and 5.4% of the CP and of the NDF dataset, respectively) were dropped from the dataset as they only occurred in the last phase of the grazing season with very high GDD. The data were analysed by means of linear mixed models, starting from a baseline model accounting for the random effects of paddock and year, as well as the effect of GDD. The models were developed stepwise-forward using AIC as an indicator of model fit and optimising also the degree of polynomials. All fixed predictors were mean-centred before analysis. Normal distribution of residuals and variance homogeneity was assessed by means of diagnostic plots. Data were analysed using the function *lmer* within the package *lme4* in R (4.4.1).

Results and discussion

A high forage quality was observed throughout the investigation period under all conditions of water availability, with a mean CP of 214.9 ± 38.5 SD g (kg DM)⁻¹ and a mean NDF of 426.0 ± 63.1 SD g (kg DM)⁻¹. Both CP and NDF were significantly explained by polynomials of GDD (a second-degree one for CP and a third-degree for NDF) and AI (a second-degree one for CP and a first-degree for NDF) ($P < 0.05$ for the highest degree term of each polynomial), as well as by an interaction effect of GDD and AI ($P < 0.001$ for both CP and NDF). The conditional R^2 was 0.55 for both models, but the marginal R^2 was higher for the CP model (0.47) than for the NDF model (0.22). The random effect of paddock accounted for 3.5 and 3.1% of the variance in CP and NDF, respectively, while 9.9% of the variance in CP and 39.4% of the variance in NDF were explained by the random effect of year. This indicates that a substantial amount of the variance in NDF can be attributed to differences between years.

The interaction between GDD and AI was highly significant ($P < 0.001$) in both models, suggesting that the effect of moisture availability on forage quality depends on the level of temperature accumulation. This confirms what had already been observed for CP in similar climatic contexts in permanent meadows (Peratoner *et al.*, 2020). At low GDD (early season), CP was highest under dry conditions (low AI), and lowest under wet conditions. With increasing GDD, these differences decreased and near the end of the season CP was slightly higher under wet conditions. This reversal was more pronounced for NDF, which was lowest under dry conditions in spring, and under wet conditions in autumn (Figure 1). Forage quality was lower in wet springs, which can be tentatively related to a nitrogen dilution effect, where increased biomass due to better growing conditions reduces the nitrogen content. In the second half of the growing season however, the influence of these two effects appeared to diminish as higher aridity indices resulted in overall better forage quality. Under high moisture contents in autumn, a higher leaf-to-stem ratio, together with better nitrogen uptake and growing conditions,

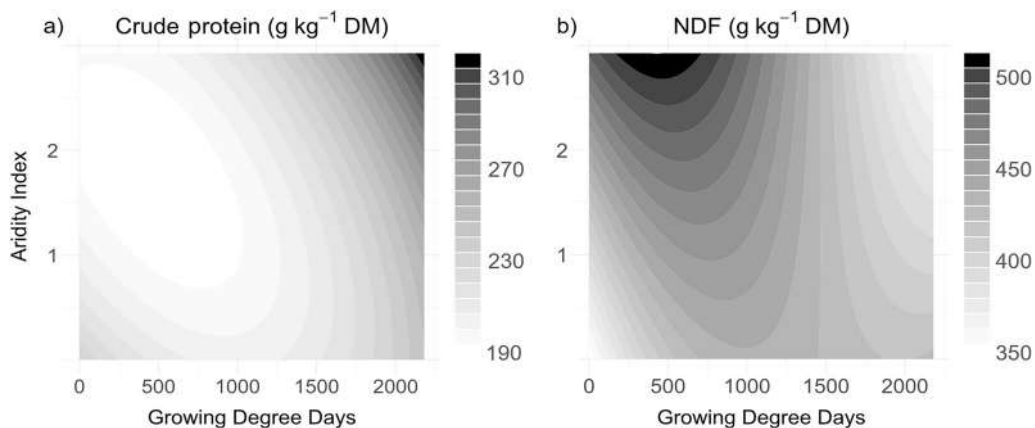


Figure 1. Effect of growing degree days and aridity index on (a) crude protein and (b) NDF. Predicted values obtained from the fitted model are shown.

presumably increased CP and lowered NDF contents. These patterns were particularly evident due to the short and consistent sampling intervals.

Our results suggest that grass phenological growth patterns in addition to a nitrogen dilution effect cause dry conditions to improve forage quality only in spring, but no longer in autumn. Understanding this seasonal divergence is important, as it implies that grazing strategies and supplementary feeding need to be adjusted differently throughout the year.

Conclusion

Our results offer an approach for the prediction of seasonal forage quality dynamics of pastures under temperate climate managed with high defoliation frequency, depending on water availability and the progress of the growing season. During dry springs, higher protein contents and lower NDF should be anticipated, which can result in high passage rates and unbalanced nitrogen intake, whereas in autumn the same problems are more likely to occur under wet conditions. To mitigate potential animal health issues arising from these forage characteristics, stocking rates should be reduced and/or supplemental feed should supply adequate fibre and energy to maintain rumen function.

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Microclimate and biomass dynamics of peat grassland under photovoltaic modules

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Abstract

To mitigate climate change effects, measures that reduce the emission of greenhouse gases from the use of fossil energy or from carbon mineralisation in drained peatland are discussed. The use of photovoltaic (PV) systems on peat with grassland combines CO₂ reduction and provides income for farmers. Little is known about how microclimate affects biomass and quality. We conducted an experiment at a peatland site with existing grassland in North Germany, where a photovoltaic system was installed in 2021. Measurements were taken over two growing seasons in 2023 and 2024 across three distinct sections: (i) adjacent grassland not affected by PV modules as a control, (ii) directly under the PV modules, and (iii) in the aisle between PV module blocks. Eight TOMST sensors per section continuously recorded soil moisture and surface temperature. Grass samples were collected in a three-cut system to assess herbage biomass production and nutritive value. Our results indicate slightly reduced biomass under PV modules, with higher crude protein and lower water-soluble carbohydrates. The study contributes to the understanding of the combined use of PV and grassland.

Keywords: herbage, microclimate, peat, photovoltaic, sensors

Introduction

There is growing interest in the use of photovoltaic (PV) systems on agricultural land as a means of combining renewable energy production with continued forage cultivation. (Weselek *et al.*, 2019). The PV modules and the technical design of the PV system can influence microclimatic conditions, which in turn may affect the quantity and quality of biomass (Zinken *et al.*, 2024). This study examines a PV system installed on peatland to analyse surface temperature, biomass yield, crude protein, and water-soluble carbohydrates across three treatments: free grassland, under modules, and in the aisle between modules.

Materials and methods

This investigation took place at the ‘Solarpark Lottorf’ in Schleswig-Holstein, Germany. The PV modules used in the park feature a single-axis tracking system, measure 2 m in length, and have an edge height ranging from 640 mm up to 2170 mm above ground level. Rows are spaced 4 metres apart. Starting in April 2023, a total of 24 TMS-4 sensors (TOMST, Prague, Czech Republic) were used to continuously monitor soil moisture and temperature. A single sensor measured values across three vertical zones relative to the soil surface: 6 cm below, 2 cm above, and 15 cm above ground. The treatment areas were studied: (i) a free grassland area without PV coverage, (ii) an area directly under the modules, and (iii) the aisle between the module blocks. Each treatment was represented by a transect measuring 4 m×10 m, along which sensors were placed approximately one metre apart. In the ‘under’ transect, sensors were placed in a strategic arrangement beneath two adjacent rows of modules, located at their

Table 1. Annual mean surface temperature (°C) for the five positions in the two areas across the two sampling years

	Surface temperature (°C)				
	left_edge	under	right_edge	between	free
2023					
Aisle	15.57	15.49	15.58	15.42	15.9
Module	14.53	14.56	14.71	14.81	15.9
2024					
Aisle	14.6	14.6	14.84	14.77	14.86
Module	14.06	14.01	14.11	14.24	14.86

lateral edges, directly under the modules and between the modules. Data logging was performed at 15-minute intervals, covering two entire growing seasons (2023 and 2024). Biomass sampling followed a three-cut regime, with harvests conducted on 27 May (both years), 26 July (2023)/6 August (2024), and 28 September (2023)/7 October (2024). Samples were taken from a 30 cm×60 cm area located east of each sensor within every transect, resulting in 24 grass samples per sampling event. After drying, the grass samples were analysed by Near-Infrared Spectroscopy (NIRS) to determine, among others, crude protein (CP) and water-soluble carbohydrates (WSC). Groups of four sampling points from the free area were averaged and used as the corresponding fifth position for the other two areas. Annual yields were summed for each year, and CP and WSC are given as weighted means. EMMs for DM, CP, and WSC were obtained from linear models with year, area, and position as fixed effects; mean surface temperature is shown descriptively.

Results and discussion

Across both the aisle and module areas, dry matter (DM) yields in 2023 were consistently higher than in 2024 (Fig. 1). The year effect was significant in the mixed model ($p=0.003$) and varied by sampling position, with some positions (e.g., right edge in the aisle area) showing larger reductions in 2024 than others. In the aisle area, the presence of rushes (*Juncus* spp.) may have influenced DM values. Mean surface temperatures (Table 1) were slightly higher in 2023 than in 2024. In both years, temperatures were marginally lower in the aisle area and lower under the modules compared to the free position, with left_edge and under showing the coolest values, followed by right_edge and between. This gradient corresponds to the changing shading pattern created by the module tracking system. Even in the aisle area, which is not directly beneath modules, reduced temperatures suggest partial shading from adjacent module rows. Crude protein concentrations were generally higher under the modules than in the aisle. Within the aisle area, the five positions yielded similar CP values, whereas under the modules, CP was markedly higher, especially at the left edge and directly beneath the modules. Reduced light availability could slow down plant development, consistent with Portner *et al.* (2023), who reported reduced biomass but higher CP under PV shading. The lower CP values in 2024 may partly reflect the slightly later sampling dates in that year. Differences in WSC concentrations were smaller and varied more among positions. In the aisle area, weighted mean WSC values of the four module positions were similar to values in the free position, whereas under the modules, in both years, WSC values were consistently lower than in the free (unshaded) positions. The lowest values occurred at the left edge and directly under the modules, with slightly higher values at the right edge and between positions. This pattern may be related to the PV module tracking system, which tilts westward during the day, reducing shading in the right-edge positions. WSC appeared less affected by the factor year than CP or DM. The mixed-model analysis (Year×Position×Area) confirmed significant main effects of year for DM and CP ($p < 0.05$) and a noticeable Year×Position interaction pattern for DM, suggesting that the

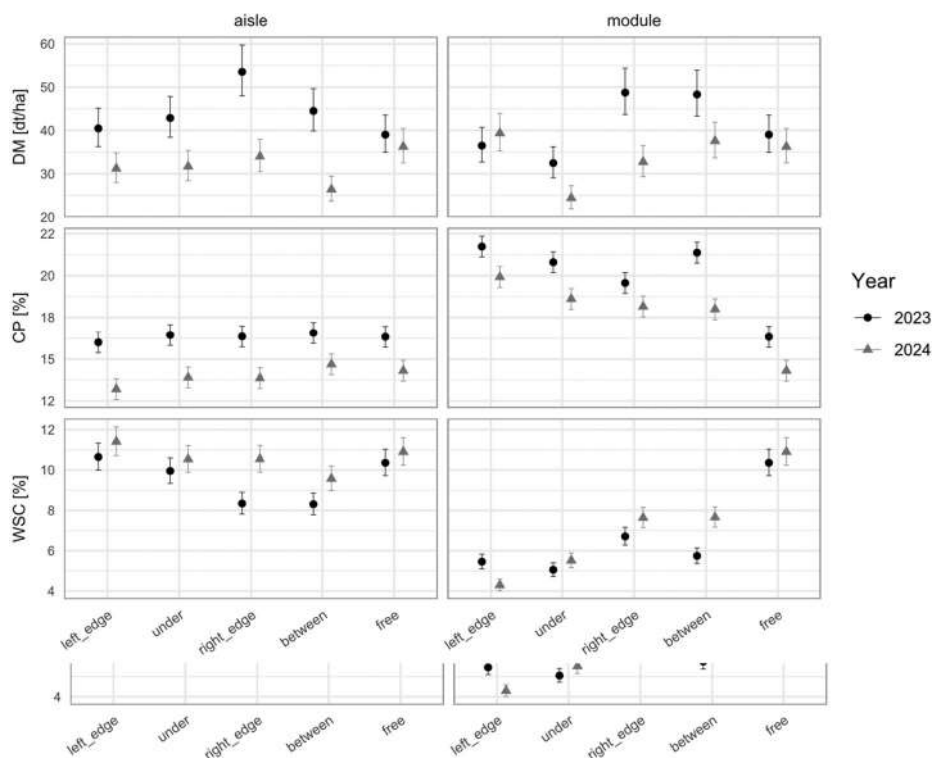


Figure 1. Estimated means of dry matter (DM), crude protein (CP) and water-soluble carbohydrates (WSC) by area (aisle and module) and position relative to PV modules across two years, based on linear models. Positions: left edge, under, right edge, between, free. Error bars indicate ± 1 standard error for each group.

magnitude of interannual differences depended on sampling position. Overall, 2024 was characterised by reduced biomass and protein content, whereas carbohydrate concentrations were more stable.

Conclusion

Shading from PV modules altered both forage yield and quality, with biomass generally reduced under the modules but crude protein concentrations often higher, particularly in more shaded positions. Water-soluble carbohydrates were less affected and showed mainly position-specific variation. These patterns, consistent across two growing seasons, highlight the importance of microclimatic conditions created by PV arrays in shaping productivity and nutrient composition.

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Soil nitrogen variability under different grassland management practices

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Abstract

As part of a research project at the Faculty of Agriculture and Life Sciences, soil samples were taken from 24 permanent grassland areas in north-eastern Slovenia in autumn 2020. The aim was to evaluate the effects of different grassland management practices—three annual cuts (3C), four annual cuts (4C) and a combination of cutting and grazing (CG)—on mineral nitrogen ($N_{\min}$) and soil pH. Samples were taken from three depths (0–30, 30–60 and 60–90 cm) and analysed for nitrate (NO_3^-), ammonium (NH_4^+), total mineral nitrogen ($N_{\min}$) and pH. $N_{\min}$ and NO_3^- were significantly higher in CG than in 3C, while NH_4^+ values were similar in all practices. All nitrogen forms decreased with depth, reflecting microbial activity at the surface. Soil pH differed between practices, but not significantly. The results suggest that grassland management influences nitrogen dynamics in the soil, especially in the upper layers, and that a combination of cutting and grazing improves nitrogen availability without increasing the risk of leaching, as higher mineral N was confined to the topsoil and did not accumulate in deeper layers.

Keywords: grassland management, soil depth, soil mineral nitrogen, soil pH

Introduction

Understanding nitrogen (N) dynamics in grassland soils is crucial for optimising fertilisation and mitigating environmental impacts. While fertiliser inputs are relatively easy to quantify, N uptake by plants, biological fixation and losses through leaching, denitrification or volatilisation are more difficult to assess and depend on soil type, pH, land use intensity and season (Yang *et al.*, 2023; Yao *et al.*, 2025). The management of grassland, such as the frequency of cutting, grazing and fertilisation, strongly influences nitrogen mineralisation and thus the availability of plant-available nitrate (NO_3^-) and ammonium (NH_4^+) (Yang *et al.*, 2023; Zhao *et al.*, 2022). The concentration of mineral N generally decreases with soil depth, which is due to lower microbial activity and reduced organic inputs (Watroš *et al.*, 2018). In pasture systems, the uneven deposition of urine and faeces causes spatial fluctuations in soil mineral N content and availability (Boagert *et al.*, 2000; Bison *et al.*, 2024). Soil pH regulates nitrogen conversion: a low pH value favours NH_4^+ accumulation, while a near-neutral pH value promotes nitrification (Skowron, 2004; Sapek, 2010; Watroš *et al.*, 2018). In this study, the effects of management intensity, sampling depth and soil pH on the content of mineral nitrogen in permanent grassland are investigated in order to better understand the N cycle under different utilisation practices.

Materials and methods

The study was conducted in autumn 2020 on 24 permanent grassland sites in the Podravska (46.55°N, 15.65°E) and Savinjska (46.24°N, 15.27°E) regions in Slovenia. The soils included pseudogleys, hypogleys and brown earths with pH values between 4.0 and 7.0 and textures ranging from loam to

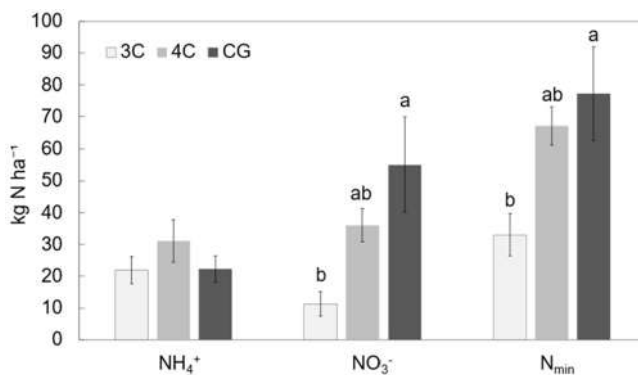


Figure 1. Concentration of NH_4^+ , NO_3^- and $\text{N}_{\min}$ (0–90 cm) under three different grassland management systems.

silty clay (Vidic *et al.*, 2015). Grassland utilisation and management systems were evenly distributed across the different soil types. All sites were located on livestock farms within the Natura 2000 network, characterised by medium- to low-intensity use. In 2020, the average annual air temperatures (11.2 and 10.8°C) were slightly above the long-term averages (10.04 and 10.02°C) and the precipitation totals (916 mm and 1174.6 mm) were close to the average (927.2 mm and 1119.6 mm) in both regions (ARSO, 2023). Soil samples were collected from grassland areas with different regimes: three cuts (3C), four cuts (4C) and three cuts with autumn grazing (CG). At each site, 15 subsamples were randomly collected at 0–90 cm depth and divided into 30 cm thick layers (0–30, 30–60 and 60–90 cm). Mineral nitrogen (NO_3^- and NH_4^+) was determined according to modified ISO protocols (SIST ISO 13395:1999, SIST ISO 11732:2005, SIST ISO 14255:1999) after extraction with 0.01 M CaCl_2 and measured by molecular absorption spectrometry. The pH of the soil (0–30 cm) was determined according to SIST ISO 10390:2006. Data were analysed using linear mixed models (IBM SPSS v20) with management and depth as fixed factors and region as a random factor. Results are presented as estimated means $\pm$ standard error (SE). Total $\text{N}_{\min}$ (0–90 cm) was compared using a one-way ANOVA. Significant differences were evaluated with a Bonferroni test at the 0.05 level.

Results and discussion

The results confirmed that grassland utilisation, sampling depth and soil pH significantly influence the $\text{N}_{\min}$ content of permanent grassland soils (Figure 1). The CG system showed the highest $\text{N}_{\min}$ values across all layers (total 0–90 cm: 77.3 kg ha⁻¹), followed by the 4C (67.2 kg ha⁻¹) and 3C (33.0 kg ha⁻¹) systems. These differences were statistically significant ($P = 0.005$) and indicate that grazing integration improves soil N availability, probably due to additional organic nitrogen input via animal excreta and increased microbial turnover, as also observed by Boagert *et al.* (2000) and Zhao *et al.* (2022). A similar trend was observed for NO_3^- content, which was also highest under CG and lowest under 3C management. In contrast, NH_4^+ concentrations were comparable in all management systems, suggesting that the variation in total $\text{N}_{\min}$ content was largely due to differences in NO_3^- accumulation.

Within each practice (Table 1), the $\text{N}_{\min}$ content decreased with depth, with the highest concentrations measured in the 0–30 cm layer. This gradient reflects the typical vertical distribution of $\text{N}_{\min}$ in grassland soils, where microbial activity and organic matter input are concentrated in the surface layer

Table 1. Concentration of NH_4^+ , NO_3^- and N_{min} under different soil depths and grassland management systems

Management systems	N form (kg ha^{-1})	Soil depth (cm)			Sig. ($P <$)
		0–30	30–60	60–90	
3C	NH_4^+	13.79 ± 3.01 B a	5.07 ± 3.00 b	3.74 ± 3.15 b	0.011
4C		21.28 ± 3.01 A a	7.00 ± 3.01 b	3.45 ± 3.20 c	0.001
CG		14.27 ± 3.00 B a	4.93 ± 3.01 b	4.10 ± 3.32 b	0.009
Sig. ($P <$)		0.059	0.798	0.986	
3C	NO_3^-	7.79 ± 4.79 C	1.96 ± 4.80	2.02 ± 5.04	0.516
4C		22.78 ± 4.80 B a	8.75 ± 4.79 b	5.45 ± 5.03 b	0.010
CG		35.95 ± 4.79 A a	15.90 ± 4.80 b	4.23 ± 5.33 c	0.001
Sig. ($P <$)		0.001	0.060	0.852	
3C	N_{min}	21.41 ± 6.31 C a	7.02 ± 6.32 b	5.83 ± 6.52 b	0.024
4C		44.06 ± 6.32 B a	15.79 ± 6.31 b	9.00 ± 6.53 c	0.001
CG		50.20 ± 6.31 A a	20.83 ± 6.32 b	8.32 ± 6.78 c	0.001
Sig. ($P <$)		0.001	0.080	0.879	

(a, b, c): Different letters indicate significant differences among soil depths within the same management system (Bonferroni test, $P < 0.05$). (A, B, C): Different letters indicate significant differences among management systems within the same soil depth (Bonferroni test, $P < 0.05$). Values are presented as estimated fixed-effect means $\pm$ SE.

(Watroš *et al.*, 2018; Yao *et al.*, 2025). The NH_4^+ content followed a similar trend and was significantly higher in the topsoil, especially in the 4C system (21.3 kg ha^{-1}), which may reflect increased mineralisation with more intensive biomass decomposition (Yang *et al.*, 2023). NO_3^- concentrations were highest in the CG system, especially in the top layer (35.9 kg ha^{-1}), indicating more favourable conditions for nitrification under the CG management. Soil pH, determined only in the 0–30 cm layer, ranged from 6.36 in 3C and 6.65 in 4C to 6.79 in the CG system. Although the differences were not statistically significant, this near-neutral pH range could support microbial processes such as nitrification (Skowron, 2004; Watroš *et al.*, 2018), which explains the higher NO_3^- values in the CG system.

Conclusion

This study shows that the combination of cutting and grazing increases the availability of mineral nitrogen in the soil more effectively than cutting alone, especially in the topsoil. The lack of nitrogen accumulation in deeper layers indicates a low risk of leaching under current management and soil conditions. These results support mixed grassland management as a promising approach to improve nitrogen cycle while maintaining ecological sustainability in permanent grassland systems.

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Effect of nitrogen application rate on herbage production in perennial ryegrass and perennial ryegrass-white clover swards

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Abstract

The inclusion of white clover (*Trifolium repens* L.) in perennial ryegrass (*Lolium perenne* L.) swards can play a role in reducing artificial nitrogen (N) fertiliser input to intensive grazing systems. White clover is an important forage legume in temperate parts of world, but benefits can be limited due to low clover content in the sward, often associated with artificial N fertiliser application. This experiment investigated the effect of N fertiliser application rate (0, 100, 150, 200, 250 kg N ha⁻¹ year) to two sward types: perennial ryegrass only (grass-only) and perennial ryegrass-white clover (grass-clover) swards. White clover inclusion increased herbage production on the 0, 100 and 150 kg N ha⁻¹ grass-clover treatments (+1282, 1006 and 626 kg DM ha⁻¹, respectively) compared to grass-only swards at the same N application rates. Herbage production was significantly lower on grass-only and grass-clover swards receiving no nitrogen fertiliser application compared to those receiving N fertiliser. Herbage removed increased by 69% for grass-only and 35% for grass-clover when 250 kg N ha⁻¹ was applied compared to zero N application. Sward clover content was significantly greater ($P < 0.05$) when 0 and 100 kg N ha were applied (26.3 and 18.1%, respectively) compared greater N application rates of 150, 200 and 250 kg N ha⁻¹.

Keywords: crude protein, herbage production, nitrogen yield, perennial ryegrass, white clover

Introduction

In some temperate regions, such as Ireland, sown grass monocultures have traditionally been dominant but grass-white clover mixtures are increasingly now sown, largely due to their ability to supply biologically fixed nitrogen and reduce the reliance on synthetic fertilizer (Delaby *et al.*, 2022), as well as animal production benefits. Grass-based milk production systems, such as those common in Ireland, rely on a long growing season to ensure high proportions of grass in the diet (>80%) as it is a low-cost feed (O'Brien *et al.*, 2018; Dillon *et al.*, 2005). The sustainability of these systems is increasingly challenged by the volatility of fertiliser prices and environmental concerns resulting in restrictions on artificial N fertiliser use. The supply of N within pasture-based systems is crucial for plant growth, and in a scenario of reduced artificial N fertiliser allowances, better use of alternative N sources, such as biological N fixation (BNF) by legumes, is important to maintain herbage production. The inclusion of white clover (*Trifolium repens* L.) in perennial ryegrass (PRG) swards coupled with reduced artificial N fertiliser use can maintain or increase herbage production, herbage quality and animal performance compared to grass-only swards receiving higher N fertiliser input (Dineen *et al.*, 2018; Enriquez-Hidalgo *et al.*, 2018). The application of high levels (>200 kg N) of artificial N fertiliser has been reported to negatively affect sward legume content (Harris *et al.*, 1995). The objective of this study was to identify the optimum artificial N fertiliser application rate for grass-only and grass-clover swards grazed 8–9 times per year.

Table 1. Effect of nitrogen fertiliser application rate on herbage production, sward clover content, sward crude protein content and N yield.

		N application rate (kg N ha ⁻¹)					SE	N rate	Sward type	Sward type × N rate
		0	100	150	200	250				
Herbage removed (t DM ha ⁻¹)	GO	6.7	8.5	9.7	10.5	11.3	0.26	<0.001	<0.001	<0.01
	GC	8.0	9.5	10.3	10.6	10.8				
Clover content (%)	GC	26.3	18.1	16.0	12.9	11.3	1.06	<0.001	—	—
Crude protein (g (kg DM) ⁻¹)	GO	183	182	187	198	204	2.7	<0.001	<0.001	NS
	GC	198	198	201	206	209				
N yield (kg N ha ⁻¹)	GO	202	234	280	315	349	6.7	<0.001	<0.001	<0.001
	GC	250	286	312	327	346				

NS, not significant; GO, grass-only; GC, grass-white clover.

Material and methods

This experiment was undertaken at Teagasc, Animal and Grassland Research and Innovation Centre, Moorepark, Fermoy, Co. Cork, Ireland (52°9'N; 8°15'W) in 2019–2021. The experiment was a 2×5 factorial design with four replicates. Plots were 9 m×1.5 m and were established in August 2017. Treatments consisted of two sward types: grass-only and grass-clover, and five artificial N fertiliser application rates (0, 100, 150, 200, 250 kg N ha⁻¹; hereafter referred to as 0 N, 100 N, 150 N, 200 N, 250 N). Plots were grazed eight to nine times per year. Immediately prior to each grazing, herbage mass was estimated using an Etesia mower (Etesia UK, Warwick, UK) by cutting one strip in each plot to a height of 4 cm. Harvested herbage was weighed, and a 100 g sub-sample was taken for dry matter (DM) determination at 90°C for 15 h. A further subsample was dried at 60°C for 48 hours and milled through a 1 mm sieve and stored for analysis. Pre- and post-grazing sward heights (cm) were taken with a rising plate meter (Jenquip, Fielding, New Zealand) immediately before and after grazing. Herbage removed was determined at each grazing by adjusting the yield at 4 cm to the actual height grazed. Sward clover content was measured in the grass-white clover plots at the same time as herbage estimation as described by Egan *et al.* (2018). Herbage crude protein content and herbage N content were estimated using NIRS (Burns *et al.*, 2010). Herbage N yield was calculated by multiplying the herbage DM yield by the herbage N content. Data were analysed in SAS using Proc Mixed. Nitrogen rate, sward type and associated interactions were included as fixed effects and replicate was used as random effect.

Results and discussion

There was an interaction between sward type and N rate on herbage removed ($P < 0.01$); herbage removed was greater on grass-clover than grass-only at 0 N and 100 N and similar at other N application rates (Table 1). Nitrogen rate had an effect ($P < 0.001$) on sward clover content; increasing N rate reduced clover content (Table 1). Sward white clover increased as the year progressed and was greater in the June to September period on 0 N, 100 N and 150 N than 200 N and 250 N (Figure 1).

Low N rates resulted in lower crude protein content particularly in grass-only swards (Table 1). Grass-clover swards (202.4 g (kg DM)⁻¹) had greater CP content ($P < 0.001$) than grass-only swards (190.7 g (kg DM)⁻¹) across all N rates. A significant N rate×sward type interaction was observed for N yield (Table 1). At lower N rates, GC swards had higher N yields than the GO swards; however, this effect reduced as N rate increased (Table 1). At the highest N rate (250 N), there was no difference in N yield between the GC and GO swards (Table 1).

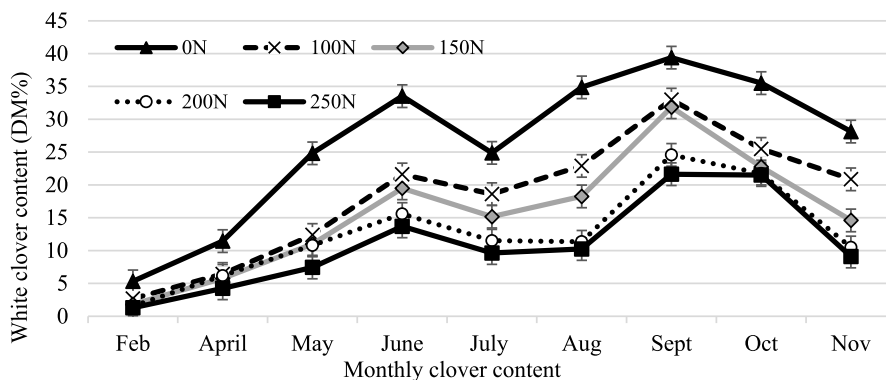


Figure 1. Effect of nitrogen (N) application rate (0: ▲, 100: ×, 150: ◆, 200: ○ and 250 kg N/year: ■) on sward white clover content in 2019, 2020 and 2021. Data presented as LSM. Vertical bars represent SEM (1.7) N rate $P < 0.001$.

Conclusion

Incorporating white clover into perennial ryegrass swards receiving 150 kg N ha⁻¹ resulted in similar amounts of herbage removed to grass-only swards receiving 200 and 250 kg N ha⁻¹ indicating potential for reducing N fertilizer input to enable positive impacts on economic and environmental sustainability. Grass-clover swards had similar crude protein content to grass-only swards receiving 200 or 250 kg N ha⁻¹.

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Examining the influence of nitrogen and weather on grass growth over time

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Abstract

Perennial ryegrass underpins pasture-based systems in Ireland. Grass growth is influenced by weather conditions and nutrient availability, in particular nitrogen (N). A plot study was conducted at Teagasc, Fermoy, Co. Cork, Ireland from 2003 to 2023 examining the impact of N fertiliser (four rates: 0, 150, 300 and 650 kg N ha⁻¹) and weather on growth. Meteorological data were collected at the experimental site. Plots were harvested weekly in rotation from February to November. Annual and seasonal dry matter (DM) production and N response were analysed using PROC MIXED in SAS. A TreeExplainer-based Shapley Additive exPlanations (SHAP) framework was used to identify drivers of growth. Average annual DM production was 5.3 (0 N), 8.95 (150 N), 11.4 (300 N) and 13.9 (650 N) t DM ha⁻¹. Spring N fertiliser application significantly ($P < 0.001$) influenced early spring growth. On average, 99, 76, 71 and 66 days were required from January 1st for growth rates to reach 10 kg DM ha⁻¹ day⁻¹ for 0 N, 150 N, 300 N and 650 N, respectively. Day length, soil and temperature, soil moisture deficit, rainfall and solar radiation significantly influenced ($P < 0.05$) grass growth.

Keywords: climate, fertiliser, grass growth, herbage production, N response, nitrogen

Introduction

Perennial ryegrass (*Lolium perenne* L.) underpins pasture-based milk and meat production systems in temperate regions. Meteorological conditions and nutrient availability influence growth. Nitrogen (N) fertiliser plays an important role in providing sufficient N to meet the production requirements of grass and is the most limiting nutrient for grass growth in Ireland and other temperate regions (Parsons & Chapman, 2000). It is the most effective management input for manipulating DM production within the limitations imposed by the environmental factors like soil type, temperature and moisture (Morrison *et al.*, 1974). Insufficient N supply lowers the overall capacity of the sward to capture light and hence photosynthesise, restricting of the growth of individual leaves and the number of tillers that develop (Parsons & Chapman, 2000). The objective of this study was to examine the effect of N fertiliser application rate (0, 150, 300 and 650 kg N ha⁻¹) and meteorological conditions and their interactions on herbage production.

Materials and methods

A long-term plot experiment was conducted at Teagasc, Animal and Grassland Research and Innovation Centre, Moorepark, Co. Cork, Ireland (52.16°N, 8.26°W) from 2003 to 2023. Plots (1.5 m × 5.0 m) were established with late-heading diploid perennial ryegrass and rotated every 3–5 years. Four annual artificial N fertiliser treatments (0, 150, 300 and 650 kg N ha⁻¹ as CAN) were applied, each replicated four times. Daily meteorological data, including rainfall, air and soil temperatures, solar radiation, were recorded at the site, and soil moisture deficit (SMD) was calculated using equations described by Schulte *et al.* (2005). Plots were rotationally defoliated once every 28 days, as described

Table 1. Mean seasonal herbage production (kg DM ha⁻¹) and N response (kg DM (kg N applied)⁻¹) for perennial ryegrass swards receiving 0, 150, 300 and 650 kg N ha⁻¹ year⁻¹ across a 21-year period (2003–2023).

	Herbage production						N response				
	0	150	300	650	SE	P value	150	300	650	SE	P value
Spring	1,045	2,034	2,443	2,970	59.9	<0.001	17.9	12.3	8.6	0.6	<0.001
Early summer	2,130	3,532	4,455	5,350		<0.001	27.7	22.4	15.8		<0.001
Late summer	1,457	2,268	3,087	3,868		<0.001	23.8*	22.4*	16.4		<0.001
Autumn	846	1,241	1,559	1,804		<0.001					

Spring, January to April; early summer, May and June; late summer, July and August; autumn, September to December.

*Average N response for late summer and autumn calculated together to account for residual N effects after fertiliser application ceased.

by Corrall & Fenlon (1978), by harvesting one strip (1.2 m×5.0 m) from each plot to using an Etesia mower (Etesia UK, Warwick, UK). Mown herbage was weighed and a 100 g sub-sample dried at 60°C to determine DM content. Grass growth was calculated using the methodology described by Corrall & Fenlon (1978). The start of the grass growing season was defined as when daily growth rate reached 10 kg DM ha⁻¹ day⁻¹ in spring (Hurtado-Uria *et al.*, 2013). Herbage production response to the N fertiliser applied (N response; kg DM kg⁻¹ N applied; O'Donovan *et al.*, 2004) was calculated for each year and each season (spring (January–April), early summer (May–June), late summer combined with autumn (July–December)). Annual and seasonal DM production, and N response were analysed using PROC MIXED in SAS (version 9.4). A Tree Explainer-based Shapley Additive exPlanations (SHAP) framework was used to estimate the marginal contribution of each predictor variable (i.e. meteorological factors) to the response variable (grass growth).

Results and discussion

There was a significant ($P < 0.001$) effect of N fertiliser rate on seasonal (Table 1) and annual herbage production (5.3 (0 N), 8.95 (150 N), 11.4 (300 N) and 13.9 (650 N) t DM ha⁻¹). As N fertiliser application increased, the response to additional N fertiliser reduced (Table 1). The response to N fertiliser application was lowest in spring (Table 1), similar to O'Donovan *et al.* (2004). On average, 99, 76, 71 and 66 days were required from January 1st for growth rates to reach 10 kg DM ha⁻¹ day⁻¹ for 0 N, 150 N, 300 N and 650 N, respectively.

Daylength was the most consistent and influential driver of grass growth (Figure 1) and had a larger effect at higher N fertiliser rates. Soil temperature at 10 cm had a strong positive effect on grass growth (Figure 1), while SMD had a strong negative effect. The analysis identified a critical SMD threshold of 40 mm, beyond which a sharp decline in growth occurred at all N fertiliser rates. The extent of the impact of SMD on growth varied by N fertiliser rate, with more prolonged reductions in growth at low compared to higher N rates (data not shown), similar to Akmal & Janssens (2004). Solar radiation and air temperature had secondary but synergistic effects on growth, particularly in spring and early summer.

Conclusion

Grass growth is strongly influenced by both N fertilization and meteorological conditions. The variability in grass growth increased at lower N fertiliser application rates, suggesting that N availability amplifies grass growth sensitivity to climatic fluctuations.

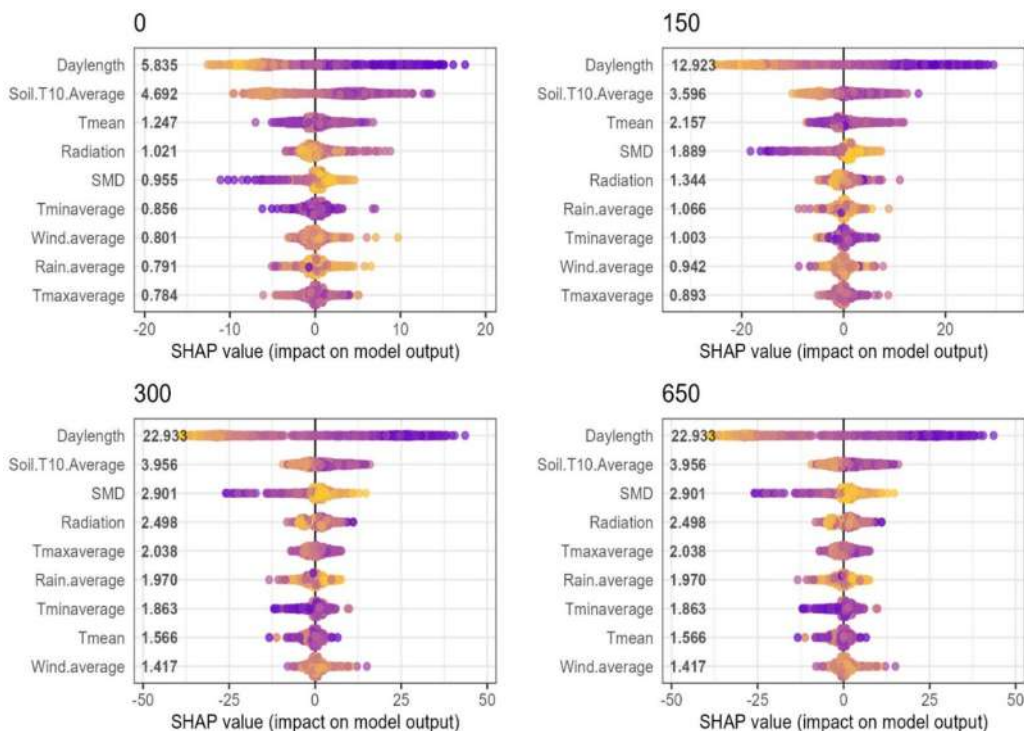


Figure 1. Ranking and range of SHAP scores for each meteorological variable on the growth of perennial ryegrass swards receiving 0, 150, 300, 650 kg N ha⁻¹. Positive SHAP scores indicate increased grass growth; negative SHAP scores indicate a limitation to grass growth.

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Farmer action plans to reduce greenhouse gas emissions or increase carbon sequestration

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Abstract

To achieve climate targets, farmers are increasingly focussed on reducing greenhouse gas (GHG) emissions from and increasing carbon (C) sequestration within their production systems. The Horizon2020 Innovation Action Project, ClieNFarms, aims to co-develop locally relevant solutions to empower farmers to achieve climate neutrality and climate resilience. There are 20 farmer networks within the ClieNFarms project each with 3 to 10 commercial farmers. Through a co-creation process (Creative Arena), interaction with the ClieNFarms Catalogue of Solutions, and exchanges with experts, each project farmer has developed an Action Plan to implement solutions on their farm. Each farmer selected two or more solutions which were appropriate to their system and location. The number and type of solutions selected within each network varied. The dairy and beef networks selected broadly similar solutions including genetics and animal breeding, slurry and fertiliser management, soil fertility, and feed management. Crop networks selected solutions including cover crops and fertiliser management. In general, solutions selected were broadly similar across enterprise categories, but specifics around methods used and implementation were locally appropriate.

Keywords: C sequestration, co-creation, GHG emissions

Introduction

Agriculture, like all sectors, has a range of climate targets to achieve. A key target is of the European Green Deal Farm to Fork strategy is climate neutrality by 2050. Farmers are increasingly focussed on reducing greenhouse gas (GHG) emissions from, and increasing carbon (C) sequestration within, their production systems. The Horizon2020 Innovation Action Project, ClieNFarms, aims to co-develop locally relevant solutions to empower farmers to achieve climate neutrality and climate resilience. This paper will describe the process whereby farmers identified solutions appropriate to their farms, and summarise the main solutions selected by animal and crop farmers.

Materials and methods

The central operational focus of the ClieNFarms project is the case-study structure that allows a strong empowerment of farmers and the supply chain accompanied by dissemination and replication of the tested innovations. Called I3S (Innovative Systemic Solutions Space), the demonstration structure is based on demonstration farms (DFs), lead commercial farms (LCFs), outreach farms all working together with the supply chain in a living-lab like approach. The I3S structure typically enables one demonstration farm to reach 10 lead commercial farms, which in turn influence 100 outreach farms. There are 20 I3S in ClieNFarm each with 3 to 10 commercial farmers (dairy×7; beef×2; sheep×1; tillage crops×5; specialist crops (tomato and olive)×2). Each of the LCFs developed an Action Plan in which they listed the solutions that they implemented during the project, an explanation of how the

Table 1. List of solutions included in action plans in dairy, beef, sheep, crop and specialist crop I3Ss (Innovative Systemic Solutions Space).

Theme	Solution	No. farms in each network type choosing this solution (total no. farms in network type)				
		Dairy (72)	Beef (20)	Small ruminants (6)	Crop (37)	Specialist crop (14)
Animal nutrition	Reduce crude protein content in diet	14		3		
	Optimise type and/or quantity of concentrate	36	4	4		
	Low emissions feed ingredients	23				
Animal management	Improve forage quality	13	8	2		
	Improve daily liveweight gain		9			
	Manage number of youngstock and unproductive stock	8	5			
Animal health	Improve animal health	16	8	2		
Animal breeding	Genetic improvement of livestock	15	3	6		
	Improved breeding management	2				
	Reduce age of calving	11	1			
	Improve reproductive performance	12	7	4		
Grassland management	Improve grazing/grazing management	16	10			1
Crop or grassland fertilisation	Fertiliser type and quantity applied	20	6		21	4
	Better use of slurry (timing and/or method of application)	23	7			
	Soil fertility	12	4		1	2
	Improve slurry/manure management	14				
	Improve slurry storage time	1				
	Use organic fertilisers	7	3		5	9
Crop/grassland management	Reduce soil tillage	17	3		17	7
	Incorporate legumes (grassland or crops)	21	8	3	9	1
	Diversify rotation	4			20	1
	Intersown crops	9			10	10
	Introduce cover crops	15			31	13
	Crop residue incorporation	3			14	13
Energy	Use renewable energy	13			2	2
	Produce renewable energy	24	8		1	3
	Reduce energy use	10			2	
Other	Plant and manage hedgerows	17			10	
	Plant and manage trees/agroforestry	6			9	

solution will be implemented, the indicator and unit used to monitor the solution implementation, the indicator value before implementation, target indicator value, year of implementation and a list of people helping to choose and implement each solution (ClieNFarms Consortium, 2024). Selection of solutions to be included in the action plans was facilitated by the creation of the ‘ClieNFarms Catalogue of Solutions’ (<https://cliefarms.eu/solutions/>), hosting of Creative Arenas (Bergez & Lanigan, 2022; Browne et al., 2024) which were multi-actor workshops in each I3S, and exchanges with experts including, but not limited to, advisers, nutritionist, veterinarian, the I3S demonstration farm and other farmers, as well as their I3S Coordinator.

Results and discussion

Each farmer selected two or more solutions which were appropriate to their system and location. The number and type of solutions selected by farmers within each network varied. The dairy and beef networks selected broadly similar solutions including ones related to genetics and animal breeding, slurry and fertiliser management, soil fertility, and feed management (Table 1). The crop networks selected solutions including cover crops and fertiliser management (Table 1).

These solutions, although adapted to a particular production system, can be applied on different farms. As part of the ClieNFarms project, the LCFs demonstrate the implementation of these solutions to inspire other farmers at, for example, open days and farm walks. The identification of categories of levers depending on the type of farm may also enable stakeholders in the sectors to promote levers adapted to archetypes.

Conclusions

The number and type of solutions selected within each network varied. In general, solutions selected were broadly similar across enterprise categories, but specifics around methods used and implementation were locally appropriate.

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Soil compaction and legacy forages effects on yield and mineral offtake

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Abstract

Different forages vary in their mineral composition and alter soil biology and physical parameters, possibly due to differences in rooting, and sward micro-climates. There has been relatively little research to study either the legacy effects of these forages on the mineral composition and offtake of the following forage crop, or how soil compaction affects yield and mineral composition. Monoculture of perennial ryegrass (PRG), red clover, white clover, chicory and plantain were established and harvested between August 2017 and April 2021. Following this, the same plot areas were resown with PRG. In the autumn of 2021 and 2022, one half of each plot was compacted with machinery. The experiment investigated the legacy effects of the previously sown forage as well as the effect of compaction on the forage yield and mineral composition of PRG. Machinery compaction reduced forage DM yield ($P < 0.001$) resulting in a lower offtake of macro and micro-minerals. Areas previously sown to plantain, red clover and white clover had higher forage yields than those sown to ryegrass or chicory ($P < 0.05$) resulting in changes in macro and micro-minerals offtake.

Keywords: compaction, forage, legacy effects, legumes, minerals

Introduction

Livestock rely on a supply of essential macro- and micro-nutrients to maintain health and optimal productivity. Swards containing different forage species have been shown to contain higher concentrations of these essential nutrients when compared to perennial ryegrass (PRG) swards (Marley *et al.*, 2021). Soil degradation may be exacerbated by land management practices and agricultural intensification, with machinery compaction typically reducing grassland productivity, although recent research has sought to find ways to improve grassland resilience to compaction through use of different forages (Muhandiram *et al.*, 2020). Use of legumes to increase soil nitrogen within rotations has been the topic of many studies (Kakraliya *et al.*, 2018). However, there has been little research into either the legacy effects of different forages on the mineral composition and offtake of the following grass crop or how soil compaction affects yield and macro- and micro-nutrient offtakes from grasslands. Here, we report on the legacy effects of the previously sown forage as well as the effect of compaction on the forage yield and mineral composition of PRG harvested for silage.

Materials and methods

The experimental area was set up on a stony, well drained loam of the Rheidol series at Aberystwyth University Wales (52.4296°N, 4.0226°W). Plots were uniformly ploughed (175 mm depth) and soil P and K indices were standardised. Lime was applied at 5 Mg ha⁻¹. The experiment was a randomized block design of four replicates (12 × 5 m) comprising PRG (*Lolium perenne*), chicory (*Cichorium intybus* (CH)), white clover (*Trifolium repens* (WC)), red clover (*Trifolium pratense* (RC)) and plantain (*Plantago lanceolata* (PL)) sown as pure swards in August 2017 at rates of 35, 7, 6, 16 and 9 kg ha⁻¹,

respectively. During 2018–2020, plots were cut on five or six occasions during May to November. PRG, CH and PL received 230 kg N ha⁻¹ year⁻¹. No N was applied to RC and WC. On the same plots, and after applying glyphosate herbicide, PRG was established by direct drilling on 26 April 2021 at a rate of 35 kg ha⁻¹. Plots were maintained by harvesting on four occasions during 2021 and five occasions in 2022–2024. All plots received 300 kg N, 52 kg P, 149 kg K and 8 kg S ha⁻¹ year⁻¹. On 15 October 2021 and 14 October 2022, half of each plot was compacted using a weighted tractor and trailer (combined weight 7.66 Mg, travelling at 0.73 km h⁻¹, equivalent to 443.8 kPa). Soil bulk density was determined in soil cores (8 cm Ø, 0–15 cm) in November of 2021 and 2022. Forage yield, harvested at the height of 6 cm from Cut 1 of 2023–2024 (11 May and 8 May, respectively) was determined by weighing the material cut from 8 m×1.5 m within each plot. Sub-samples of forage, as harvested, were taken to determine dry matter (DM) and chemical composition by ICP-MS and ICP-OES. Data were analysed for effects of previous forage (PF), compaction (C) and year according to the split plot design.

Results and discussion

Machinery compaction increased soil density from 1.00 to 1.07 g cm⁻³ after the first compaction and from 0.97 to 1.08 g cm⁻³ after the second compaction ($P < 0.05$; SEM 0.009 (38.8 df)) but previous forage had no legacy effect ($P = 0.577$). There was no effect of previous forage or soil compaction ($P > 0.05$) on ryegrass DM yield in the first harvest year following a single compaction treatment in the preceding autumn. However, following further compaction in the autumn of the first harvest year, effects of previous forage and compaction on yield, chemical composition and hence, mineral offtakes became apparent in the second and third harvest years. This was despite differences in soil density between years within compaction level not differing significantly ($P > 0.05$; SEM 0.0088 (30 df), suggesting that the compaction effect may have been a direct effect on the forage *per se* in addition to soil changes. There was no evidence ($P > 0.05$) of PF × C or C × year interactions (except for Co). Main effects of previous forage and compaction on forage DM yield and mineral offtakes in the ryegrass crop over the second and third harvest years are presented. Offtakes increased ($P < 0.001$) between the second and third harvest years with the exception of K and Se which decreased ($P < 0.001$) and Mo which was unchanged ($P = 0.194$) (data not presented). Forage yield was influenced by PF ($P < 0.001$) and reduced by more than 300 kg ha⁻¹ in plots previously under PRG or CH compared those under PL or clover ($P < 0.05$) (Table 1). Compared to PRG, WC increased ($P < 0.05$) P offtake in a subsequent PRG crop with other legacy forages intermediate. Similarly, PL and WC increased potassium offtake, PL increased magnesium, PL, WC and RC increased sulphur and copper and PL and WC increased zinc offtake ($P < 0.05$). Calcium offtakes were lower following PRG than any other forage. Compaction reduced ($P < 0.001$) DM yield by 483 kg ha⁻¹ (11%) and reduced offtake of all minerals ($P < 0.05$) except for molybdenum ($P = 0.143$) (Table 2).

Conclusion

Findings showed the legacy effects of previous forages altered the mineral offtake of a subsequent ryegrass crop with, for example, higher P and Mg offtake following white clover and plantain, respectively, highlighting benefits beyond the initial higher mineral found in the previous forage. This work also demonstrated that machinery compaction reduced forage productivity and nutritive value with regards to all mineral offtakes, except molybdenum.

Table 1. Main effect of previous forage on mean yield and mineral offtakes of subsequent ryegrass crop over second and third harvest years (2024–2025).

	Previous forage					SEM	Prob.
	Perennial ryegrass	Chicory	Plantain	Red clover	White clover		
Yield (kg DM ha ⁻¹)	3881.9 ^a	3906.5 ^a	4260.7 ^b	4222.8 ^b	4218.5 ^b	68.55	0.003
P (kg ha ⁻¹)	13.33 ^a	13.67 ^{ab}	14.91 ^{bc}	14.74 ^c	14.90 ^c	0.373	0.026
K (kg ha ⁻¹)	74.2 ^{ab}	65.4 ^a	83.8 ^{bc}	71.9 ^{ab}	91.0 ^c	3.20	<0.001
Mg (kg ha ⁻¹)	3.66 ^a	3.97 ^{ab}	4.27 ^b	4.05 ^{ab}	3.95 ^{ab}	0.099	0.014
Ca (kg ha ⁻¹)	17.7 ^a	19.3 ^b	20.0 ^b	20.9 ^b	19.4 ^b	0.40	0.002
S (kg ha ⁻¹)	7.90 ^a	8.02 ^{ab}	8.79 ^c	8.77 ^c	8.80 ^{bc}	0.228	0.027
Cu (g ha ⁻¹)	12.8 ^a	13.4 ^a	15.6 ^b	15.2 ^b	15.4 ^b	0.52	0.007
Fe (g ha ⁻¹)	217.6	212.0	233.5	245.2	240.9	9.15	0.098
Zn (g ha ⁻¹)	54.2 ^{a'}	54.4 ^{a'}	61.7 ^{b'}	59.4 ^{ab'}	61.0 ^{b'}	1.86	0.034
Mo (g ha ⁻¹)	0.64	0.78	0.92	0.83	1.07	0.133	0.272
B (g ha ⁻¹)	12.1	12.2	13.0	13.1	12.5	0.30	0.135
Se (g ha ⁻¹)	0.21	0.19	0.24	0.25	0.27	0.048 [§]	0.222
Co (g ha ⁻¹)	0.23	0.18	0.18	0.22	0.20	0.029 [§]	0.068

a, b, c Means differ ($P < 0.05$) based on Student–Newman–Keuls test.

a', b' means differ at $P < 0.10$.

§ applies to means on log₁₀ scale.

Table 2. Main effect of compaction on yield and mineral offtakes of subsequent ryegrass crop.

	Year	Uncompacted	Compacted	SEM	Prob.
Yield (kg DM ha ⁻¹)		4339.7	3856.4	49.26	<0.001
P (kg ha ⁻¹)		15.29	13.33	0.208	<0.001
K (kg ha ⁻¹)		82.0	72.6	1.44	<0.001
Mg (kg ha ⁻¹)		4.35	3.61	0.067	<0.001
Ca (kg ha ⁻¹)		20.6	18.3	0.37	<0.001
S (kg ha ⁻¹)		9.16	7.75	0.161	<0.001
Cu (g ha ⁻¹)		15.8	13.2	0.34	<0.001
Fe (g ha ⁻¹)		243.1	216.5	5.25	0.003
Zn (g ha ⁻¹)		63.6	52.7	1.16	<0.001
Mo (g ha ⁻¹)		0.91	0.79	0.052	0.143
B (g ha ⁻¹)		13.3	11.9	0.24	0.001
Se (g ha ⁻¹)		0.25	0.21	0.016 [§]	0.010
Co (g ha ⁻¹)	2023	^A 0.12 ^a	^A 0.17 ^b		C × Year
	2024	^B 0.29	^B 0.26		0.031

^{A, B}, means within columns differ ($P < 0.05$) based on LSD with Bonferroni adjustment for multiple comparisons.

^{a, b}, means within row differ ($P < 0.05$) based on LSD with Bonferroni adjustment for multiple comparisons.

§; applies to means on log₁₀ scale.

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Permanent grasslands in Switzerland: CO₂ fluxes and C budgets in response to climate change

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Abstract

Permanent grasslands (PG) cover about 70% of the Swiss agricultural area and provide many ecosystem services, including climate regulation. Thus, carbon (C) budgets of PG are of great interest to mitigate anthropogenic climate change. But PG are also affected by extreme events, demanding a better understanding of their resilience to climate change. Ecosystem greenhouse gas (GHG) fluxes, alongside information on climate, soil and management, are available for three long-term PG sites along an elevation gradient in Switzerland. Responses of PG to climate change, including weather extremes have been studied using machine learning. C budgets have been validated with changes in soil C stocks. Overall, the PG were a small C sink, unless organic fertilization was reduced or the sward was renewed. Lowland PG were strongly impacted by summer heatwaves and droughts, while alpine PG benefitted from a large winter snowpack. Thus, GHG fluxes and C budgets were affected by both climate and management practices, with unexpected trade-offs between securing C sinks and reducing nitrous oxide (N₂O) emissions. Moreover, farmers of intensive PG could keep regrowth rates after mowing stable over about 20 years, despite high climatic variations. Our results provide crucial insights into the development of climate-smart management practices.

Keywords: carbon sequestration, greenhouse gas fluxes, permanent grasslands, regrowth, sward renewal, weather extremes

Introduction

Permanent grasslands (PG) cover about 34% of the European and about 70% of the Swiss agricultural area, being located from the lowlands up to alpine elevations (BFS, 2024). Moreover, PG provide many ecosystem services, in particular feed production for livestock, but also climate regulation, soil protection and biodiversity (Richter *et al.*, 2024). PG soils are also rich in carbon (C), containing about 20% of global soil C stocks (Bai & Cotrufo, 2022). Therefore, one goal in sustainable management of grasslands is to maintain or increase these soil C stocks to mitigate anthropogenic climate change (IPCC, 2021). However, PG are also affected by extreme events, demanding a better understanding of their resilience to climate change (Gharun *et al.*, 2020). Thus, the objectives of this study were to: (i) quantify long-term trends in GHG fluxes, (ii) identify drivers of GHG fluxes, and (iii) assess impacts of management practices on GHG fluxes.

Materials and methods

Ecosystem greenhouse gas (GHG) fluxes, i.e., carbon dioxide (CO₂), and water vapour (H₂O), have been measured over recent decades with the eddy covariance technique, alongside climate variables (Eugster & Merbold, 2015). Methane (CH₄) and nitrous oxide (N₂O) fluxes are also available for selected years, together with soil and management information. Here, we used data from three long-term PG sites, located along an elevation gradient in Switzerland: Chamau (CH-Cha, at 393 m asl, since 2005; Feigenwinter *et al.*, 2023a,b), Frübüel (CH-Fru, at 982 m asl, since 2005; Rogger *et al.*, 2022) and Alp Weissenstein (CH-Aws, at 1978 m asl, summer fluxes since 2006, year-round since

2015). Responses of PG to climate change, including weather extremes, have been studied using tree-based machine learning models (e.g., random forest (RF)) as well as Shapley Additive exPlanations (SHAP) analyses to identify flux drivers. Negative fluxes denote GHG uptake by the PG, positive fluxes denote GHG release to the atmosphere. C budgets have been validated with changes in soil C stocks.

Results and discussion

Net ecosystem CO₂ exchange (NEE) of all PG sites with the atmosphere showed pronounced intra- and inter-annual variations, driven by both management activities (i.e., cutting, fertilization, renewal) and meteorological conditions. While NEE of the lowland PG Chamau was $-260 \pm 172 \text{ g C m}^{-2} \text{ year}^{-1}$ (2005 to 2024), NEE of the subalpine PG Frübüel was $-357 \pm 76 \text{ g C m}^{-2} \text{ year}^{-1}$ (2006–2019), and NEE of the Alpine PG Alp Weissenstein averaged $-152 \pm 82 \text{ g C m}^{-2} \text{ year}^{-1}$ (2015–2024). Despite significant increases in air and soil temperatures and decreasing soil water contents at Chamau and Alp Weissenstein, NEE did not show any significant trend over time, likely due to adjustments of cutting and subsequent fertilization dates. Gross primary production (GPP) and ecosystem respiration (Reco) rates between cutting dates at the lowland Chamau remained stable. Similar results were found for the Alpine site Alp Weissenstein. Overall, the PG were small C sinks, also validated with soil C stock measurements (Chamau only).

The most important drivers of long-term NEE and GPP fluxes at the lowland site Chamau were light, management, and temperature, while Reco fluxes were mainly driven by temperature, GPP, and management. For the subalpine site Frübüel, light and temperature were most relevant. Similarly, temperatures and growing-degree-days were the main drivers of NEE for the Alpine site Alp Weissenstein, clearly indicating that all three PGs were energy-driven. On the other hand, observed climate change and extreme events impacted the three Swiss grasslands very differently in the last 20+ years. While the Alpine grassland could benefit from a large winter snowpack during a following summer heatwave and drought, the lowland grassland was strongly impacted by the lack of soil water and high vapour pressure deficit. Thus, with future climate change, net ecosystem CO₂ uptake and grassland productivity will most likely decline, making PG management more challenging.

Considering C imports via organic fertilization and C exports via harvests, the lowland grassland was typically a small C sink, unless organic fertilization was drastically reduced or the PG was completely renewed. Particularly during these renewal events, large N₂O emissions occurred, leading to up to $10.7 \text{ kg N}_2\text{O-N ha}^{-1}$ of N losses within a 3-month period, compared to about $7 \text{ kg N}_2\text{O-N ha}^{-1}$ in normal years. However, N₂O emissions were lower when ploughing was avoided, and only shallow tilling was used. Although reduced fertilization successfully reduced N₂O fluxes and nitrate leaching, it also reduced the C sink, even triggering C source behaviour. These unexpected trade-offs between securing the soil C sink and reducing N₂O emissions demand the development of climate-smart management options.

Conclusion

Overall, we found that long-term GHG fluxes and C budgets were affected by both climate and management practices. Our results provided crucial insights into the development of climate-smart management practices, with PG renewals via ploughing clearly to be avoided.

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Nitrogen leaching from grazed grass only and grass-white clover swards at varying nitrogen inputs

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Abstract

To reduce cost inputs and environmental impacts of inorganic nitrogen (N) use, there is renewed interest in the incorporation of legumes, white clover in particular, in grazing systems. The objective of this study was to quantify N losses from perennial ryegrass and perennial ryegrass-white clover swards with reduced nitrogen inputs. Three grazing treatments were used for this study: grass-only swards receiving 200 kg N ha⁻¹ (Gr200), and grass-white clover swards receiving 100 (Cl100) or 150 kg N ha⁻¹ (Cl150) annually. Twenty cows were assigned to each treatment and swards were rotationally grazed at a stocking rate of 2.4 cows ha⁻¹. In the second year of the study, N leaching concentrations were measured from start of October to end of April at 2-week intervals. Nitrogen leaching concentrations were numerically highest for Cl150 treatment. Replacing chemical N inputs with clover in the sward did not reduce N leaching concentrations.

Keywords: dairy cows, grazing, herbage production, nitrate

Introduction

In grass-based systems, to reduce input costs and environmental impacts of inorganic nitrogen (N) use (Carran and Clough, 1995) and to increase farm gate N-use efficiency (NUE; N inputs vs. outputs that come through the farm gate such as feed, fertiliser and milk sales), there is a renewed interest in the incorporation of legumes, and white clover (*Trifolium repens* L.; WC) in particular, in perennial ryegrass (*Lolium perenne* L.; PRG)-based production systems (Lüscher *et al.*, 2014). Grass-only based grazing systems are highly efficient and low-cost but are ultimately dependent on high N fertilizer levels (>200 kg N ha⁻¹) to achieve high levels of herbage production (Enriquez-Hidalgo *et al.*, 2016). White clover can offset some of these N requirements through its ability to biologically fix atmospheric N to facilitate grass production (Crush, 1987).

Nitrogen losses from swards and grazed swards can be a source of pollution to water bodies, especially in nitrate (NO₃) leaching vulnerable zones. The application of N to soil from chemical fertiliser, animal excretions and N fixation are drivers of N losses to waterways. Nitrate leaching from grazing systems occurs primarily during the drainage period during autumn, winter and spring, when rainfall exceeds evapotranspiration. The maximum admissible concentration (MAC) limit of NO₃ in rivers, set out in the Water Framework Directive (WFD), is 50 mg NO₃ l⁻¹ (11.3 mg NO₃-N l⁻¹). A number of methods have been developed to measure NO₃ leaching from grazed swards. Depending on site and soil type, these methods include the use of ceramic cups to measure NO₃-N concentrations in soil water at the root zone level (≤ 1 m below the soil surface). Ceramic cups were the method employed

Table 1. Nitrogen fertiliser application strategy (kg N ha⁻¹)

Rotation (rot) and date	Gr200	CI150	CI100
February	28	28	28
Mid-March	28	28	19
15th April (2nd rot)	20	20	19
6th May (3rd rot)	20	13	9
27th May (4th rot)	17	9	0
17th June (5th rot)	17	9	0
8th July (6th rot)	17	9	0
29th July (7th rot)	17	9	0
19th August (8th rot)	17	9	9
Mid-September	19	16	16
Total	200 kg	150 kg	100 kg

by this study. The objective of this study was to determine the effect of WC inclusion in PRG swards and N fertiliser rate on NO₃-N concentrations from ceramic cup data from grazing systems.

Materials and methods

The experiment was carried out at Moorepark Teagasc, Fermoy, Co. Cork, Ireland from February 2023 to November 2024. The experiment contrasted two sward types (PRG-only and PRG-white clover) receiving varying levels of fertiliser. This resulted in three separate grazing treatments: a PRG-only sward receiving 200 kg N ha⁻¹ (Gr200), a PRG-white clover sward receiving 150 kg N ha⁻¹ (CI150), and PRG-white clover sward receiving 100 kg N ha⁻¹ (CI100). There were 20 cows per treatment assigned per treatment through a randomised complete block design based on calving date, parity, milk yield, body weight, body condition score and genetic merit. Each treatment group was then equal and was stocked at 2.4 cows ha⁻¹. Treatments were rotationally grazed from early-February to mid-November and target post-grazing sward height was 4 cm. Nitrogen fertiliser applications were applied according to Table 1. Each farmllet was walked weekly to monitor herbage availability (Hanrahan *et al.*, 2017). All treatments were supplemented with concentrate; on average, 716 kg concentrate was fed across all treatments each year. Sward WC proportion was measured before grazing in each paddock in each rotation by cutting 15 random grab samples to 4 cm with Gardena hand shears, separating the sample into PRG and WC fractions and drying at 90°C for 16 hours. Milk yield was recorded daily and milk composition weekly by taking milk samples from a consecutive evening and morning milking.

Porous ceramic cups are an established method for determining NO₃ concentration of soil water draining from grassland swards to 1 m depth in the soil (Murphy *et al.*, 2024). The porous (1 µm) cups are capable of absorbing soil water, by means of suction applied via flexible tubing, and can be buried below the cultivation depth of the soil. The soil water can then be sampled and analysed to determine NO₃-N concentration and when combined with soil water drainage can be used to estimate total NO₃-N leached. Porous ceramic cups have been installed on a 20% of the grazing area during the trial. The cups were sampled every two weeks during the effective drainage period (October 2024 to March 2025). Herbage characteristics (Table 2) were analysed using Proc MIXED in SAS (SAS 9.4). Terms included in the model were Paddock ID, rotation, sward type, fertiliser rate treatment and NO₃-N concentrations. Individual paddock was the experimental unit, with paddock included as a random factor and rotation as a repeated measure. Milk data were also analysed using Proc MIXED. Terms included in the model were sward type treatment, fertiliser rate treatment and parity.

Table 2. Effect of sward type (ST) and nitrogen fertiliser rate (FR) on grazing characteristics and nitrate-N ($\text{NO}_3\text{-N}$, mg l^{-1}) concentrations of soil water

	Treatment			P-value		
	Gr200 ¹	Cl150	Cl100	SE	ST	FR
Pre-grazing yield (kg DM ha^{-1})	1599	1422	1498	76.1	0.077	0.217
Pre-grazing height (cm)	11.78	11.31	11.36	0.325	0.266	0.546
Post-grazing height (cm)	4.42	4.39	4.43	0.040	0.782	0.786
Clover content (%)	—	19.8	20.1	1.03	—	0.886
Leachate nitrate-N ($\text{NO}_3\text{-N}$, mg l^{-1})	8.37	11.02	10.57	0.981	0.003	0.586

Gr200, PRG-only sward receiving 200 kg N ha^{-1} ; Cl150, PRG-white clover sward receiving 150 kg N ha^{-1} ; Cl100, PRG-white clover sward receiving 100 kg N ha^{-1} .

Results and discussion

There was no effect of sward type or N fertiliser application rate on any grazing characteristics (Table 2). There was also no reduction in production for the two WC treatments receiving less chemical N compared to the Gr200 treatment, similar to Murray *et al.* (2024). Sward type did have an effect on $\text{NO}_3\text{-N}$ concentration with the Cl150 treatment having the highest $\text{NO}_3\text{-N}$ concentration ($P < 0.01$). White clover inclusion had a significant ($P < 0.001$) positive effect on milk solids production, whereas N fertiliser rate had no effect. The Gr200 treatment produced 5601 kg milk and 504 kg milk solids (kg fat+protein) cow^{-1} . In comparison, the PRG-WC swards produced on average 5858 kg milk and 535 kg milk solids cow^{-1} .

Conclusion

The inclusion of WC in PRG swards had a significant effect on $\text{NO}_3\text{-N}$ concentration with the Cl150 treatment having the highest concentration. These results are preliminary from one year’s collection, requiring further over-years analysis. Additional ceramic cups will be installed in more of the grazing area to capture a greater understanding of the variation depending on WC content.

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Transfer of soil mineral N in various organic mixed grass-legume swards to subsequent winter wheat

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Abstract

The benefits on the agronomic properties of winter wheat (WW) succeeding mixed grass-legume (MGL) leys grown for three harvest years were studied in a field experiment farmed organically. Crop residues of red clover (RC), white clover (WC), birdsfoot trefoil (BT), and lucerne (LU) grown in mixed swards with timothy were incorporated prior to seeding of WW, cv. Informer. N-uptake, estimated with N-sensor at growth stage DC 39, was significantly higher for WW grown after LU compared to RC. Consequently, grain yield (GY) and grain protein content were significantly higher after LU than after RC, as GY increased by 43% when grown after LU compared to after RC. The benefits of WC and BT were intermediate and not significantly different from LU or RC. This study shows the impact of the individual legume species in MGL leys, providing several positive benefits for the successive crops, thus reducing the need for fertilisers.

Keywords: *Lotus corniculatus*, *Medicago sativa*, *Trifolium pratense*, *Trifolium repens*

Introduction

Mixed leys containing forage legumes and grass species are widely used for livestock feed in the Nordic countries. Legume crops are an essential component of sustainable agricultural systems as legume plants provide biologically fixed nitrogen (BFN) in symbiosis with soil rhizobia in root nodules (Granstedt & L-Baekstrom, 2000). The incorporated mixed grass-legume (MGL) swards (below-ground biomass and crop residues) constitute a valuable source of soil mineral N (SMN) to the succeeding crop. Previous studies on short-term rotational leys showed benefits for the subsequent winter wheat (WW, *Triticum aestivum*) crop after one, two, or three years of MGL leys with red clover (RC, *Trifolium pratense*) (Høgh-Jensen & Schjoerring, 1997; Granstedt & L-Baekstrom, 2000), birdsfoot trefoil (BT, *Lotus corniculatus*) (Nuttall *et al.*, 2008), or lucerne (LU, *Medicago sativa*) (Salehi *et al.*, 2025). RC is considered a short-lived legume due to its poor persistence, originating in root rot caused by infection of soil-borne pathogens (Wallenhammar *et al.*, 2020; Nilsdotter-Linde *et al.*, 2025). In a previous study, strategies were identified and developed for sustainable forage legume cropping as persistence and production capacity were studied for MGL leys with RC, BT, LU and white clover (WC, *Trifolium repens*) intercropped with timothy (TI, *Phleum pratense*) across a three-year period (Wallenhammar *et al.*, 2020). The objective of this study was to compare the benefits of four preceding binary grass-legume long-term swards on the subsequent WW, in terms of WW yield, nitrogen uptake, and grain quality.

Materials and methods

The field experiment was organically managed and located at the Kvinnersta field experimental farm, Örebro municipality (59°21'N, 15°14'E) in south-central Sweden. The topsoil was clay loam (39% clay, 51% silt, and 8% sand; 2.8% organic matter; pH 6.3, K (AL) = 20.9 mg 100 (g soil)⁻¹; P (AL) =

Table 1. Legume proportion of preceding crop to winter wheat at last forage cut in 2019, nitrogen uptake of winter wheat at DC39, grain yield, and quality measurements of grain and straw grown after 3 years of mixed grass-legume swards of timothy (TI) intercropped with red clover (RC), white clover (WC), birdsfoot trefoil (BFT), or lucerne (LU) in a field experiment near Örebro, south-central Sweden.

Preceding crop	Legume proportion (%) ¹	N-uptake at DC 39 (kg N ha ⁻¹)	Grain yield (15%, w/w) ²	Protein content (% of DW)	Volume weight (g l ⁻¹)	No of spikes (m ⁻²)	Straw yield (kg DW ha ⁻¹)	N-content in straw (%)
RC+TI	72 b ³	11 b	3119 b	8.6 b	806 ab	360	9986	0.27 b
WC+TI	60 c	15 ab	3695 ab	8.7 ab	804 b	370	11040	0.26 b
BT+TI	68 bc	12 b	3314 ab	8.9 ab	814 a	339	9477	0.27 b
LU+TI	83 a	18 a	4426 a	9.0 a	812 ab	374	13468	0.33 a
P-value	<0.001	0.008	0.022	0.033	0.011	ns	ns	<0.001
CV	6.4	17.9	14.7	1.9	0.5	9.5	22.3	5.8

¹ Legume proportion at last forage cut in 2019; 5th September for RC, WC, and LU; 5th August for BT.

² Calculated using dry weight (DW) content to 15% water weight by weight.

³ Similar characters indicate no significant difference according to Tukey's HSD test ($P < 0.05$).

4.6 mg 100 (g soil)⁻¹). Annual average precipitation is 697 mm, and annual average temperature is 7°C. The experimental set up was arranged in a randomised complete block design, plot size 12 × 3 m, with four replicates following a three-year MGL ley established in 2016, where the following legume species were grown: RC, WC, BT, and LU intercropped with TI (Wallenhammar *et al.*, 2020). All plots were cut three times each harvest year, 2017–2019, except for BT plots which were cut twice each harvest year (Wallenhammar *et al.* 2020). The last cut was performed on 5 August for BT and 5 September 2019 for RC, WC and LU. The experimental area was turned with a mouldboard plough on 10 September and WW (cv Informer) was seeded on 20 September 2019. No fertilisation or plant protection measures were carried out until harvest on 6 August 2020. The amount of SMN (the sum of NH₄-N and NO₃-N) was determined on 1 April 2020 as samples were randomly taken plot-wise from the following depths: 0–0.3 (12 cores), 0.3–0.6, and 0.6–0.9 m (6 cores each). The samples were pooled into one composite sample per layer for each treatment and stored frozen at –18°C until processing at Agrilab, Uppsala. The nitrogen uptake in the WW crop was estimated with a Yara N-sensor[®] at growth stage DC 39 on 2 June. At harvest, grain yield (GY) was measured and analysed for grain protein content (GPC), volume weight, and thousand kernel weight (TKW) at Agrilab, Uppsala. To determine straw biomass and N-content in straw, plants in two randomly located frames (0.5 m×0.5 m) in each plot were cut at the soil surface and the material was pooled and analysed plot-wise at the Soil and Plant Laboratory, Swedish University of Agricultural Sciences, Uppsala. Statistical analyses were performed using ANOVA (JMP[®] 17.2 Statistical Discovery, Cary, NC, USA). Pairwise comparisons were performed using Tukey's HSD test ($P < 0.05$).

Results and discussion

Legumes, notably deep-rooted species, enhance benefits for the following wheat crop due to BFN of the legumes and the soil structure stabilisation by the roots. In this study, SMN at the onset of growth in April was 55, 50, 45 and 57 kg ha⁻¹ (0–0.9 m) for RC, WC, BT and LU, respectively, but was not significantly different across treatments. However, the estimated N-uptake at stem elongation was significantly higher for WW grown after LU than after RC (Table 1).

This difference demonstrated an impact on WW agronomic properties as both GY and GPC were significantly higher after LU than after RC (Table 1). GY increased by 43% after LU compared to GY after RC. The benefits of WC and BT were intermediate and not significantly different from LU or RC. TKW ranged from 54 to 58 g, and the volume weight from 42.6 to 42.7 g, but neither were

significantly different across preceding legume crops. The straw yield (SY) varied across the preceding crops and although mean value was highest for LU, no significant differences were found. The proportion of straw yield was ca 75% of the plant weight (GY+SY) for all preceding crops. Nitrogen content in straw was significantly higher after LU and shows the valuable benefits of LU. The benefits for subsequent cereal crops depend on the composition of MGL swards. In our study, the legume proportion (LP) for the last cut was highest for LU (83%) followed by RC (72%) (Table 1), which is higher than reports from comparative studies where two grass species were included (Nilsson-Linde *et al.*, 2025) as TI is less competitive. The grassland legacy effect of mixtures was demonstrated by Grange *et al.* (2022) and a LP of at least 33% was required to achieve high crop performance for the succeeding *Lolium multiflorum*. Most previous studies are reported from cropping systems with RC as a legume crop and, to our knowledge, this study is the first to investigate long-term swards in Sweden whilst also revealing the benefits from WC, BT and LU, where LU showed outstanding properties. In addition to BFN, legume-based management strategies improve soil N mineralisation and microbial diversity growth (Jalal, 2022). Thus, long-term MGL leys also improve the soil physical characteristics for both the agronomic and economic outcomes in a sustainable agricultural production system.

Conclusions

This study demonstrated the importance of legumes in crop production. The forage legumes RC, BT, WC and LU, grown in binary long-term leys with TI without fertilisers, resulting in a high legume proportion, offers valuable N benefits to subsequent cereal crops.

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Resilience of permanent grasslands and pastures in Portugal over seven years: implications under climate change

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Abstract

Permanent grasslands and pastures are an important part of agricultural and extensive livestock production systems. Climate change can cause significant declines in grassland productivity leading to forage shortages and financial difficulties for farmers. Therefore, farmers and policymakers need to understand the resilience of permanent grasslands and pastures to climate change and how management may enhance this resilience. Here we examine the resilience of permanent grasslands and pastures in Portugal between 2017 and 2023 based on growing season vegetation productivity obtained from high resolution satellite data. We analysed temporal variability in grassland productivity and the response to the extreme 2022 drought across mainland Portugal comparing herbaceous, shrub, and oakwood permanent grasslands. Overall, permanent grasslands in Portugal had peak productivity in 2020 and the lowest in 2022. The herbaceous permanent grasslands exhibited higher productivity during the study period. However, oak permanent grasslands show a lower temporal coefficient of variation and higher resilience to the 2022 drought. Our findings suggest that oak grasslands are more resilient to ongoing climate change. We discuss the implications of our findings for grassland management under climate change.

Keywords: drought, oak pastures, permanent pastures, productivity

Introduction

Permanent grasslands and pastures correspond to 13% of the EU's area and comprise the biggest agricultural land cover in Portugal (Eurostat 2022, INE 2021). Permanent grassland and pastures are present across a broad range of environmental conditions and provide most of the animal feed in extensive livestock production. Permanent grasslands promote biodiversity and provide ecosystem services including water regulation, erosion control and carbon sequestration (Bardgett *et al.*, 2021). Climate change threatens the socio-ecological systems of permanent grasslands as vegetation productivity is tightly coupled with climate. Thus, it is critical to understand the resilience of permanent grasslands to climate change in order to adapt policies and farming systems. Here we aim to investigate the resilience of permanent grasslands and pastures in mainland Portugal between 2017 and 2023 comparing herbaceous, shrub, and oak permanent grasslands.

Materials and methods

The study focuses on the three most frequent types of permanent grasslands and pastures in mainland Portugal: herbaceous, shrub and oak permanent grasslands. Permanent grasslands include land used permanently to grow herbaceous fodder and forage crops, through cultivation or naturally for 5 years or more, which is not included in the crop rotation. The shrub and oak permanent grasslands are sub-types where the herbaceous crops are interspersed with shrubs or have an overstory of *Quercus* tree species (e.g., cork oak, *Q. suber*). The study sites are plots that were consistently occupied

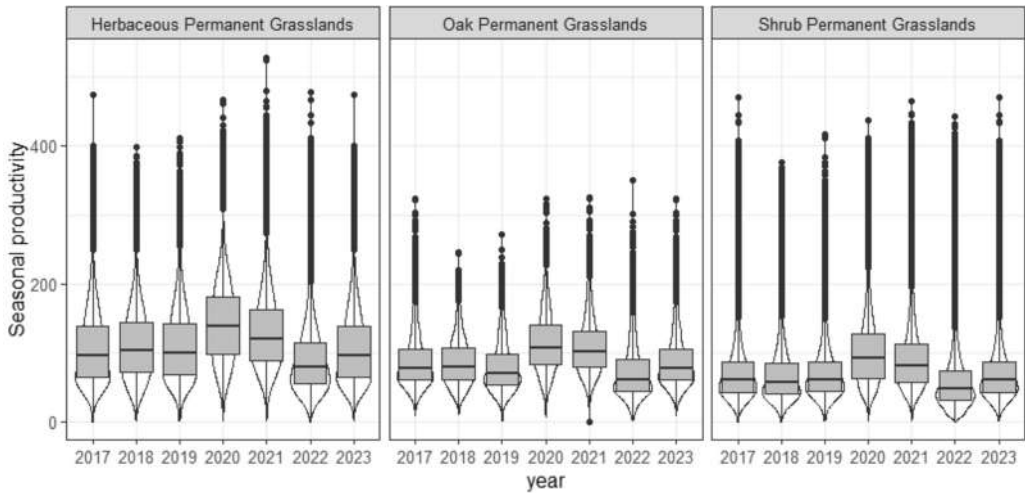


Figure 1. Boxplots of seasonal productivity per year and permanent grassland type.

by the same grassland type between 2017 and 2023 based on the national agricultural land use data. We used growing season vegetation productivity as a descriptor of the system since it represents the amount of biomass available and thus how many animals it can support. Growing season vegetation productivity was obtained from the High-Resolution Vegetation Phenology and Productivity product for the period 2017–2023 at 10 m spatial resolution. We evaluated the resilience of permanent grasslands by calculating the temporal variability of vegetation productivity in each pixel across the years 2017–2023 using the coefficient of variation, standard deviation, minimum, maximum and mean. Then, we aggregated the values of the indicators at plot level using the mean. We also calculated indicators of resilience to drought taking 2022 as a focal event. The resistance, recovery and resilience to drought were calculated as a ratio of the seasonal productivity before (2020), during (2022) and after the drought (2023) (Lloret *et al.* 2011). The grassland types were compared using Kruskal-Wallis and Wilcoxon Mann-Whitney tests. The spatial data were processed in ArcGIS Pro 3.5.1 and data analysis carried out in R.

Results and discussion

The seasonal productivity of permanent grasslands shows the highest median values for the year 2020 (114.50 PPI day⁻¹) and the lowest for the year 2022 (64.04 PPI day⁻¹) across all grassland types (Fig. 1). The decline in productivity in 2022 coincides with the third driest hydrological year since 1931, recording just 33% of the average precipitation (488.3 mm).

The drought affected the rainy season and by summertime most of the country was under severe (60%) to extreme drought (40%). Farmers required direct financial assistance for livestock feed and other mitigation measures (e.g., fallow use). Permanent herbaceous grasslands retained higher mean, minimum and maximum productivity values during this period (Fig. 1, Table 1). Shrub grasslands showed the lowest productivity values ($P < 0.01$). The results suggest that permanent grasslands can maintain higher productivity since the herbaceous crops do not compete with shrubs and trees for light, resources, and space. However, the standard deviation and coefficient of variation are significantly lower in permanent grasslands and the resilience to drought is significantly higher in oak grasslands ($P < 0.01$, Table 1). The results suggest oak grasslands are more stable and resilient to droughts, even

Table 1. Average and median values of temporal productivity and resilience in different types of permanent grasslands.

Temporal variability indicator	Mean per grassland type			Median per grassland type		
	P-Herb (<i>n</i> = 267175)	P-Oak (<i>n</i> = 6010)	P-Shrub (<i>n</i> = 218564)	P-Herb (<i>n</i> = 267175)	P-Oak (<i>n</i> = 6010)	P-Shrub (<i>n</i> = 218564)
Temporal minimum	66.1	56.1	42.2	58.7	48.1	34.8
Temporal maximum	169	135	119	166	125	108
Temporal mean	112	90.7	74.8	104	81.2	65.6
Temporal variance	17479	9950	10288	13764	7683	6828
Temporal standard deviation	34.3	26	25.4	32.6	24.3	22.8
Temporal coefficient of variation	3.65	3.42	4.04	3.42	3.26	3.81
Resistance	0.0737	0.0743	0.0685	0.0629	0.065	0.0566
Recovery	0.162	0.157	0.175	0.124	0.128	0.127
Resilience	0.0907	0.0945	0.0883	0.075	0.0812	0.0699

P-Herb, herbaceous permanent grasslands; P-Oak, oak permanent grasslands; P-Shrub, shrub permanent grasslands.

though the magnitude of the effect may be small. Trees can mobilize water from deeper soil layers and regulate the microclimate decreasing air temperature, thus improving the productivity of herbaceous crops. However, this facilitation effect may be limited to the tree's vicinity in these open systems, thus showing a weaker signal at broader scales (Moreno Marcos *et al.*, 2007). Shrub grasslands show lower resilience to drought and higher coefficient of variation ($P < 0.01$, Table 1). This suggests shrub grasslands may be less stable and contribute to higher landscape fire risk due to drier vegetation and higher fuel loads (Lecomte *et al.*, 2019).

Conclusion

Permanent grasslands can retain higher productivity levels while oak permanent grasslands are more stable and more resilient to drought. The differences among permanent grassland types are significant, nevertheless there is still a large amount of variability likely related to other environmental and management factors that will be explored in further studies. The results suggest permanent grasslands may be more vulnerable to climate change and particularly extreme droughts despite higher productivity. Oak overstory may help mitigate the impacts of droughts on permanent grassland productivity.

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Developing and validating a model for estimating effects of rewetting peat meadow on yield and feed value of grass

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Abstract

Rewetting peat soils is considered to be a key measure in Dutch peatland policy to reduce land subsidence and greenhouse gas emissions due to peat decomposition. The question is how this affects the yield and feed value of grass, grassland use, and finally farm income losses. A new model, SG-CMPM, was developed to quantify the effects of rewetting in a practical and reliable way at field level. This study evaluates whether SG-CMPM can be used, instead of the more extensive integrated farm model Waterpas, in a to-be-developed future compensation methodology for farmers in peat areas. Calculations were conducted for six peat meadow farms in the province of Friesland, comparing a reference ditch water level on those farms, varying between –112 and –55 cm, with a –35 cm (future) scenario. SG-CMPM integrates the empiric grass growth model GRASS2007 into the hydrological model SWAP, enabling simulation of direct effects (e.g. drought or oxygen stress) and indirect effects (e.g. delayed harvest due to reduced trafficability) on grass yield and feed value. Results for the reference situation were highly consistent between SG-CMPM and Waterpas. Small differences in the –35 cm scenario were attributed to simplified grassland use representation in SG-CMPM. Validation against farm data (ANCA model) showed generally good agreement, although feed value reduction may be overestimated. SG-CMPM offers an efficient, reproducible tool for estimating effects of rewetting, though further research on feed value and trafficability impacts is recommended.

Keywords: ditch water level, feed value, grass yield, peat meadow, SWAP model

Introduction

Rewetting peat soils is essential for achieving Dutch peatland goals, including reduced land subsidence and greenhouse gas emissions due to peat decomposition. According to Dutch law, arrangements exist so that damage associated with legitimate government intervention must be compensated. Compensation levels are determined with a model-based calculation method (Compensation Methodology Peat Meadow). To assess its adequacy, effects of higher ditch water levels were first calculated for six dairy farms using the integrated farm model Waterpas (De Vos *et al.*, 2006), a combined model of Dairy-Wise (Schils *et al.*, 2007) and the Soil Water Atmosphere Plant model (SWAP, Van Dam *et al.*, 1997, Kroes *et al.*, 2017). Results matched observed reality, but the method is complex and time-consuming. Therefore, the grass growth module from DairyWise (GRASS2007) was directly integrated into SWAP, creating SG-CMPM (SWAP-GRASS2007 Compensation Methodology Peat Meadow). Simulations with SG-CMPM are performed at field level instead of farm level (Waterpas). The complexity of dynamics in grassland management is therefore simplified compared to Waterpas. Suitability of SG-CMPM was tested by calculating effects of elevated ditch water levels for the six farms with both Waterpas and SG-CMPM, and by comparing reference results (no water level change) with farm data

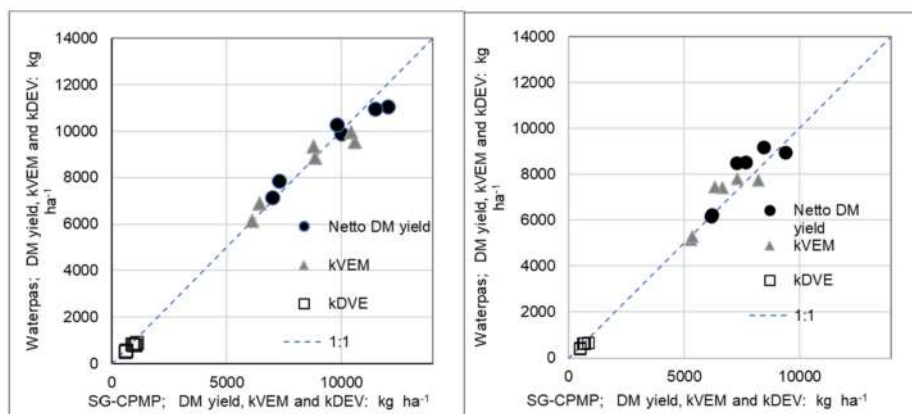


Figure 1. Calculated net DM yield, kVEM yield and kDVE yield ($\text{kg ha}^{-1} \text{ year}^{-1}$) with SG-CMPM (x-axis) and Waterpas (y-axis) for reference water table (left) and adjusted water table to -35 cm (right).

used for the ANCA model (De Vries *et al.*, 2020). ANCA also estimates net DM and kVEM yields from farm input–output data.

Materials and methods

Calculations were based on actual conditions of six peat meadow farms in the province of Friesland. The reference ditch water levels varying between -112 and -55 cm for the six farms were compared with an elevated level of -35 cm . Two models were applied: Waterpas and SG-CMPM. This integrated model instrumentation translates changes in hydrology (water table depth) and nitrogen availability (both affected by water level rise and adapted farm management) into changes in grass yield and feed value. SG-CMPM provides estimates of indirect and direct effects on grass production. Indirect effects include delayed harvests due to reduced trafficability under wet conditions; direct effects involve reduced transpiration from drought stress or oxygen stress under waterlogging. The SG-CMPM module calculates current and future grass yield and feed value on field level, expressed as net dry matter (DM), Feed Unit Milk (VEM), and Intestinal Digestible Protein (DVE). For the reference situation those data were compared with data from Waterpas. Both SG-CMPM and Waterpas data were compared with ANCA-data. For the elevated ditch water level -35 cm a comparison was made only with the Waterpas data.

Results and discussion

SG-CMPM results were validated against Waterpas for the six farms. For the Reference situation (Figure 1, left), both models produced very similar outputs. For the -35 cm water table scenario (Figure 1, right), SG-CMPM gave slightly lower net DM and kVEM than Waterpas, likely due to its simplified representation of grassland use.

Reference results from both models were compared with ANCA data (Table 1). For these model results, no statistical tests are possible. kVEM represents grass energy yield, calculated as net DM yield per cut multiplied by feed energy content (VEM). DVE is not reported by ANCA. Average net DM and kVEM yields from ANCA, SG-CMPM, and Waterpas for the Reference are shown in Table 1.

For conventional farms, net DM matched ANCA closely, while kVEM was 3–5 percentage points lower. For organic farms, net DM was 5–9 points lower and kVEM 8–12 points lower. Given the

Table 1. Net DM and kVEM yield ($\text{kg ha}^{-1} \text{ year}^{-1}$) calculated with Waterpas and SG-CPMP and compared with ANCA data for the reference water table and percentual differences with ANCA data

Model	Conventional farm		Organic farm	
	Net DM yield ($\text{kg ha}^{-1} \text{ year}^{-1}$)	kVEM (energy) ($\text{kg ha}^{-1} \text{ year}^{-1}$)	Net DM yield ($\text{kg ha}^{-1} \text{ year}^{-1}$)	kVEM (energy) ($\text{kg ha}^{-1} \text{ year}^{-1}$)
ANCA ¹	10.571	9.937	7.895	7.121
SG-CPMP	10.817 (102%) ²	9.655 (97%)	7.148 (91%)	6.271 (88%)
Waterpas	10.557 (100%)	9.429 (95%)	7.517 (95%)	6.517 (92%)

¹ Including conservation losses.

² Percentage compared to ANCA data.

small sample size, differences in weather over years and possible variation in nitrogen supply, these comparisons are indicative rather than conclusive. Larger deviations for kVEM than for DM suggest that feed value reduction may be overestimated in the model calculations. Waterpas functionalities for modelling trafficability affects grassland losses were applied in SG-CPMP. Changes in nitrogen supply potential and feed value shifts due to altered botanical composition were successfully implemented. SG-CPMP and Waterpas were aligned through integration of the GRASS2007 module from DairyWise, using identical trafficability functions and updated concepts for nitrogen supply, botanical composition, losses, and water uptake. Model results from six case farms showed close alignment, confirming SG-CPMP's suitability for estimating re-wetting effects on grass yield, feed value, and income loss. Consistency across farms with varying soil, water table and management conditions support robustness. However, evidence for feed value reduction due to higher water tables remains limited. Further research is needed on botanical composition effects under wet conditions and on trafficability thresholds under waterlogging.

Conclusions

SG-CPMP has been successfully aligned with Waterpas and performs reliably across diverse peat meadow farm conditions. It is suitable for estimating the impact of rewetting on yield, feed value, and economic outcomes. To improve accuracy, further insights into botanical shifts, feed intake, and trafficability under wetter conditions are required.

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Sun, shade and soil: A case study on how free-field photovoltaic systems alter microclimate and soil

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Abstract

In the context of climate change mitigation efforts, the growing demand for renewable energy is driving the expansion of free-field photovoltaic systems (FFPV), causing significant land-use changes with poorly understood impacts on grassland ecosystems. To investigate the impact of modules on microclimate and soil, TMS-4 sensors were installed in a FFPV in Northern Germany. We defined three areas (= treatment): area directly affected by modules (module), area where modules in a row have been removed (window), and area unaffected by modules (open) with eight sampling positions each. Soil moisture and temperatures were measured at each position to assess the influence of different shading positions within the FFPV. In addition, we analysed soil samples taken with an auger soil sampler at the same positions. We found an increased variability of all parameters in the areas ‘module’ and ‘window’ compared to the area ‘open’ in the rank order: open < window < module. Nitrogen and carbon content as well as temperatures underneath the module and window were reduced; soil moisture was highest underneath the modules. Our results enhance understanding of system-design-driven, position-dependent variations in soil and microclimate within FFPVs.

Keywords: carbon, dual land-use, marginal grassland, nitrogen, photovoltaic, TMS-4 sensors

Introduction

The growing demand for renewable energy sources and the imperative to alleviate climate change impacts, particularly heat stress on flora and fauna, have substantially intensified research in free-field photovoltaic systems (FFPV). In a way the modules also represent anthropogenic structural elements within landscapes, exerting measurable influences on the local microclimate (Armstrong *et al.*, 2016). The present study investigates the influence of varying shading conditions on soil parameters and microclimate within an existing FFPV. Specifically, we compared soil moisture, temperature at three heights, and soil nutrient distribution of two PV-affected areas and one unaffected area.

Materials and methods

The present study was carried out from June to November 2024 in the FFPV ‘Solarpark Klein Rheide’ in Schleswig-Holstein, Germany. The FFPV was built in 2018. A technical feature of this FFPV is that modules are omitted at regular intervals in each module row to allow for more sunny areas (Fig. 1; window). The grassland vegetation was not sown but grew naturally after the FFPV was built and is mowed once a year. For data analyses, the study period was divided into seasons: summer (June–August) and autumn (September–November). Average air temperature and summed precipitation during summer and autumn, recorded by a weather station at a distance of approximately 7 km from the trial site, were 16.21°C and 231.58 mm and 8.96°C and 213.19 mm, respectively. We defined

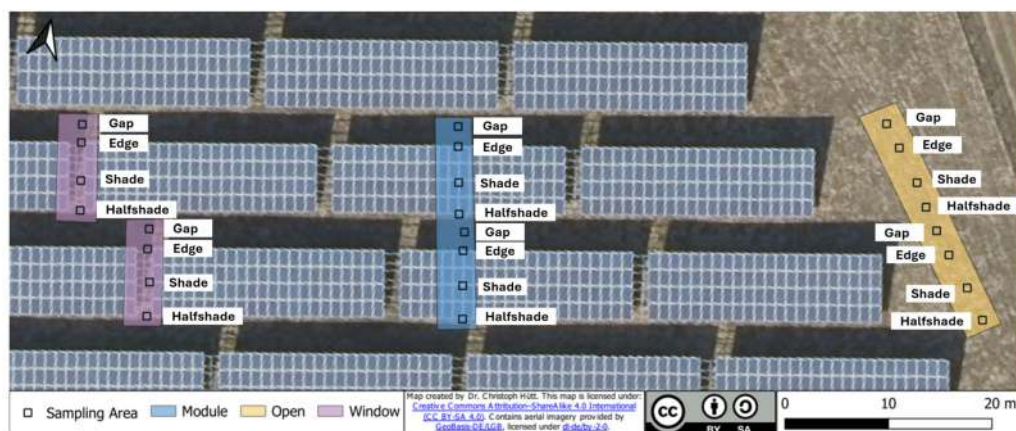


Figure 1. GPS positions of each position per area (= treatment) in the Solarpark Klein Rheide.

three treatments: area directly affected by modules (module), area where modules in a row have been removed (window) and area unaffected by modules (open) (Fig. 1). Samples were taken from different positions within each treatment referred to as gap, halfshade, shade and edge—which is following the spatial sequence in the area ‘module’. In May 2024 (after mowing), at each position one TMS-4 sensor (TOMST, Prague, Czech Republic) was installed at a depth of 9 cm measuring soil moisture and temperatures at 15 min intervals at -6 cm, $+2$ cm, and $+15$ cm from the surface. In November 2024, soil was sampled from a depth of 0–10 cm at each position ($n = 10$) using an auger soil sampler. Carbon (C) and nitrogen (N) content were analysed to investigate the effect of shading on soil nutrient distribution. Generalised linear mixed effect models (R package ‘glmmTMB’) with the fixed effects area ($n = 3$), position ($n = 4$) and the interaction of both were calculated for the target variables N, C, soil moisture and temperatures. The sample ID served as random effect. Multiple contrast tests according to Tukey’s HSD test (R package ‘emmeans’) were conducted to analyse the main influencing factors.

Results and discussion

Within the ‘open’ area, measured parameters were similar, whereas in the ‘module’ area significant variability across all measured parameters was detected (Tables 1 and 2). However, the interaction between area and position was significant for all parameters, except for temperature at -6 cm in autumn; in that case only the effect of area was significant. C and N content were lowest in shade of the areas ‘module’ and ‘window’ (Table 1). N content in all areas ranged from $1.03\text{--}1.49$ g (kg DM) $^{-1}$, which is in the range of N contents reported by Blume *et al.* (2016) for Ap-horizons of mineral soils in humid climate. Although the ‘window’ area was not consistently shaded, the lowest C content was found at position ‘shade’ whereas it was highest at its position ‘edge’, which may be explained by a reduced vegetation cover and varying microclimate conditions affecting soil fauna and microbial activity underneath PV modules compared to open areas (Krasner *et al.*, 2025). Grazing sheep may be used to redistribute nutrients in the position ‘shade’ in FFPVs, as modules provide preferred resting sites that increase trampling and defecation. Li *et al.* (2025) found an enhanced diversity of temperature through niche differentiation in a single axis mover FFPV with lowest air temperature at halfshade/edge (combined position in this type of FFPV). In the present study, both air temperatures in the area ‘module’ revealed the lowest values in halfshade, whereas edge revealed the highest values respectively (see Table 2), highlighting the effect of different technical module installations on the microclimate.

Table 1. Estimated means±SE (standard error) of generalised linear mixed effect models for soil moisture, nitrogen (N) and carbon (C).

Area	Position	N (g (kg DM) ⁻¹)	C (g (kg DM) ⁻¹)	Soil moisture (m ³ m ⁻³)	
				Summer	Autumn
Open	Gap	1.46 ± 0.03 ^a	22.2 ± 0.5 ^a	0.24 ± 0.01 ^a	0.27 ± 0.008 ^a
	Halfshade	1.47 ± 0.03 ^a	22.4 ± 0.5 ^a	0.24 ± 0.01 ^a	0.26 ± 0.008 ^a
	Shade	1.45 ± 0.03 ^a	22.1 ± 0.5 ^a	0.22 ± 0.01 ^a	0.26 ± 0.008 ^a
	Edge	1.49 ± 0.03 ^a	22.4 ± 0.5 ^a	0.23 ± 0.01 ^a	0.26 ± 0.008 ^a
Module	Gap	1.29 ± 0.03 ^b	20.1 ± 0.5 ^b	0.23 ± 0.01 ^{ab}	0.27 ± 0.008 ^b
	Halfshade	1.23 ± 0.03 ^b	18.7 ± 0.5 ^b	0.19 ± 0.01 ^a	0.23 ± 0.008 ^a
	Shade	1.03 ± 0.03 ^a	15.4 ± 0.5 ^a	0.26 ± 0.01 ^b	0.29 ± 0.008 ^b
	Edge	1.30 ± 0.03 ^b	19.9 ± 0.5 ^b	0.22 ± 0.01 ^{ab}	0.26 ± 0.008 ^b
Window	Gap	1.26 ± 0.03 ^a	20.2 ± 0.5 ^a	0.22 ± 0.01 ^a	0.28 ± 0.008 ^b
	Halfshade	1.27 ± 0.03 ^a	20.1 ± 0.5 ^a	0.23 ± 0.01 ^a	0.26 ± 0.008 ^{ab}
	Shade	1.26 ± 0.03 ^a	19.3 ± 0.5 ^a	0.19 ± 0.01 ^a	0.24 ± 0.008 ^a
	Edge	1.40 ± 0.03 ^a	22.5 ± 0.5 ^b	0.12 ± 0.02 ^a	0.25 ± 0.001 ^{ab}

Lower case letters indicate significant differences within areas, light grey indicates lower values, while dark grey indicate higher values compared to the area 'open' at the respective positions.

Table 2. Estimated means±SE (standard error) of generalised linear mixed effect models for temperature (Temp) at -6 cm, 2 cm and 15 cm

Area	Position	Temp. (°C) at -6 cm		Temp. (°C) at 2 cm		Temp. (°C) at 15 cm	
		Summer	Autumn	Summer	Autumn	Summer	Autumn
Open	Gap	18.3 ± 0.1 ^a	11.6 ± 0.07 ^c	18.1 ± 0.09 ^a	10.7 ± 0.07 ^a	17.2 ± 0.06 ^a	10.15 ± 0.06 ^a
	Halfshade	18.7 ± 0.1 ^a		18.2 ± 0.09 ^a	10.6 ± 0.07 ^a	17.2 ± 0.06 ^a	10.25 ± 0.06 ^a
	Shade	18.4 ± 0.1 ^a		18.2 ± 0.09 ^a	10.6 ± 0.07 ^a	17.1 ± 0.06 ^a	10.04 ± 0.06 ^a
	Edge	18.6 ± 0.1 ^a		18.2 ± 0.09 ^a	10.5 ± 0.07 ^a	17.2 ± 0.06 ^a	10.12 ± 0.06 ^a
Module	Gap	17.1 ± 0.1 ^b	10.8 ± 0.07 ^a	17.4 ± 0.09 ^c	10.1 ± 0.07 ^a	16.8 ± 0.06 ^c	9.22 ± 0.06 ^a
	Halfshade	15.5 ± 0.1 ^a		15.9 ± 0.09 ^a	10.1 ± 0.07 ^a	16.1 ± 0.06 ^a	9.69 ± 0.06 ^b
	Shade	15.4 ± 0.1 ^a		16.3 ± 0.09 ^b	10.4 ± 0.07 ^b	16.5 ± 0.06 ^b	10.25 ± 0.06 ^c
	Edge	16.8 ± 0.1 ^b		17.7 ± 0.09 ^c	10.8 ± 0.07 ^c	17.2 ± 0.06 ^d	10.29 ± 0.06 ^c
Window	Gap	18.0 ± 0.1 ^b	11.1 ± 0.08 ^b	17.7 ± 0.09 ^b	10.0 ± 0.07 ^a	17.0 ± 0.06 ^c	9.64 ± 0.06 ^a
	Halfshade	16.4 ± 0.1 ^a		16.6 ± 0.09 ^a	10.1 ± 0.07 ^{ab}	16.3 ± 0.06 ^a	9.54 ± 0.06 ^a
	Shade	16.4 ± 0.1 ^a		16.7 ± 0.09 ^a	10.4 ± 0.07 ^c	16.5 ± 0.06 ^b	10.14 ± 0.06 ^b
	Edge	17.5 ± 0.2 ^b		17.6 ± 0.13 ^b	10.3 ± 0.10 ^{bc}	17.0 ± 0.09 ^c	10.14 ± 0.06 ^b

Lower case letters indicate significant differences between areas for temperature at -6 cm in autumn and within areas for all other parameters. Italics indicates lower values, while underlined indicates higher values compared to the area 'open' at the respective positions.

Conclusions

Our findings indicate that PV installations induce position-dependent variations driven by system design parameters such as module size, inter-module gaps, and row spacing. This underlines the need for further scientific investigations into differently equipped FFPVs.

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Impact of acidified cattle slurry on reactive nitrogen dynamics in temperate grassland microcosms

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Abstract

The behaviour of reactive nitrogen in agricultural soils is influenced by multiple interacting factors, leading to spatial variation in nitrogen (N) fertilizer efficiency. This study assessed the impact of different N sources on key N pathways in a typical grassland of northern Germany. Twenty-eight PVC microcosms were sown with Italian ryegrass and fertilised with cattle slurry (acidified (CS+A) or untreated (CS)) at 85 or 170 kg N ha⁻¹, or with ammonium sulphate (AS) at 170 kg N ha⁻¹, all labelled with ¹⁵N. Ammonia (NH₃) and nitrous oxide (N₂O) emissions were measured using static chambers. CS+A significantly reduced total NH₃ volatilisation by 63–78% ($p < 0.05$) compared to CS. Acidification enhanced N uptake (by 139–165%) and reduced nitrification/denitrification losses at the lower N rate. Fertilizer N uptake ranged from 31–85%, with AS twice that of CS. Higher N application rates increased biomass uptake, but also increased N loss. Volatilization and nitrification/denitrification accounted for 7–61% and <0.2% of the applied N, respectively, while 7.6–15.6% remained in the soil. Results highlight the potential benefits of slurry acidification for reducing N losses and improving efficiency in intensive grassland systems.

Keywords: acidification, ammonia volatilisation, ¹⁵N tracing, nitrification, slurry grassland

Introduction

Nitrogen (N) is vital for plant growth, but organic fertilizers like cattle slurry often have lower N-use efficiency (NUE) than mineral fertilizers and contribute to reactive N losses such as ammonia (NH₃) volatilisation and nitrous oxide (N₂O) emissions, which harm air and water quality, climate, and biodiversity (Guo *et al.*, 2020). EU regulations (e.g., Directive 2008/50/EC) increasingly limit N fertilizer use to reduce such impacts. Slurry supplies mineral and organic N that can stimulate nitrification and denitrification under favourable soil conditions, while acidification reduces NH₃ losses but its impacts on N₂O emissions and plant uptake remain uncertain (Butterbach-Bahl *et al.*, 2013; Nyameasem *et al.*, 2022). This study uses a ¹⁵N labelling approach to trace N dynamics in temperate grassland soil from acidified and untreated cattle slurry and ammonium sulphate. We hypothesised that acidification would reduce gaseous N losses and boost plant N uptake, though it may increase denitrification at high application rates.

Materials and methods

A greenhouse experiment was conducted at Christian-Albrechts University of Kiel using 28 PVC microcosms (33 cm diameter, 20 cm height) filled with loamy Eutric Luvisol soil collected from an organic grassland (0–20 cm depth) in Schleswig-Holstein, Germany. Soil moisture was maintained

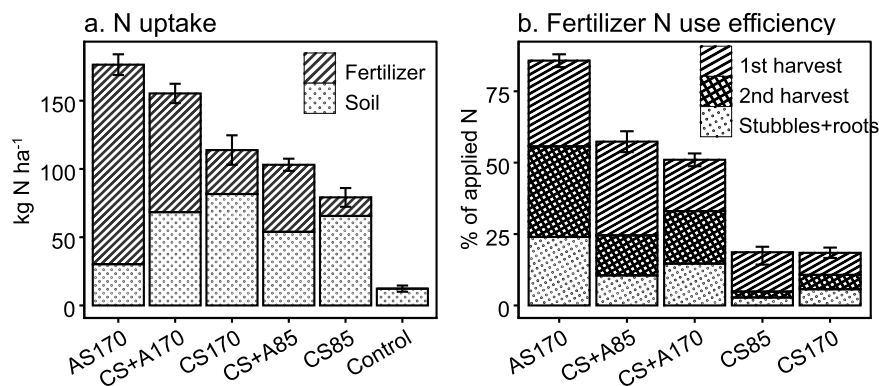


Figure 1. N source and rate effects on (a) N uptake and (b) Fertilizer N use efficiency (% of total N applied); Error bars represent standard error of the mean. ^{abc} Means with different letters/symbols are significantly different ($p < 0.05$).

at 60% water-filled pore space, and phosphorus, potassium and sulphur were pre-applied to prevent nutrient deficiencies. Italian ryegrass (*Lolium multiflorum*) was sown at a rate equivalent to 30 kg ha⁻¹ and later thinned to 100 plants per microcosm. A randomised complete block design with four replicates tested six fertilizer treatments: untreated and acidified cattle slurry (CS, CS+A) at two N rates (85 and 170 kg N ha⁻¹), and ammonium sulphate (AS) at 170 kg N ha⁻¹, all enriched with ¹⁵N. CS was band-applied (simulating a trailing shoe application), while AS was broadcast. Above- and below-ground biomass were harvested and analysed for total N and ¹⁵N enrichment using isotope ratio mass spectrometry. NH₃ volatilization was measured using the dynamic tube method (Pacholski *et al.*, 2006), and N₂O emissions were sampled from static chambers and analysed by gas chromatography, with fluxes calculated based on concentration changes over a 60-minute period. The experiment lasted 78 days and included two biomass harvests. The harvested material was oven-dried at 55°C to a constant weight to determine dry matter yield, and its nitrogen content was measured using near-infrared reflectance spectroscopy. Nitrogen recovery and balance calculations incorporated fertilizer-derived N uptake, gaseous N losses (NH₃ and N₂O), and changes in soil N pools. Data were analysed using linear mixed-effects models in R (v3.5.0), with significant differences ($p < 0.05$) assessed using Tukey's HSD test.

Results and discussion

Dry matter (DM) yields did not differ significantly among treatments ($p > 0.05$), yet slurry acidification increased N yields by 24–42 kg N ha⁻¹ and improved N use efficiency. N uptake from acidified slurry (CS+A₁₇₀) equalled those from mineral fertilizer (AS₁₇₀) (Fig. 1a). Fertilizer N recovery was highest for ammonium sulfate (AS; 86%), moderate for acidified slurry (51–57%), and lowest for untreated slurry (17–18%). Doubling the N rate from 85 to 170 kg N ha⁻¹ increased total N uptake by 44–51% and fertilizer-derived N uptake by 50–61%, with no gain in DM yield. Acidification enhanced fertilizer N uptake three- to fourfold and slightly reduced soil N uptake (approx. 17%). Fertilizer N recovery peaked at the first harvest (11–36%) but remained higher at the second harvest (16–32%) and in stubbles and roots (11–24%) under acidified and mineral fertilizers than under untreated slurry (3–5% and 4–7%, respectively; Fig. 1b). These findings support slurry acidification as a viable substitute for mineral fertilizers (Nyameasem *et al.*, 2022).

Table 1. Fertilizer N pathways recovery/partitioning in grassland as affected by N source and application rate.

	N pathway (kg N ha ⁻¹ (%))				
	CS_85	CS+A_85	CS_170	CS+A_170	AS_170
Biomass	26.3 ± 1.99 ^a (30.9)	46.0 ± 2.39 ^b (54.1)	53.4 ± 2.72 ^b (31.4)	97.0 ± 3.94 ^c (57.1)	145.0 ± 5.39 ^d (85.3)
Soil	13.2 ± 0.28 (15.5)	13.3 ± 0.15 (15.6)	13 ± 0.36 (7.6)	13.2 ± 0.92 (7.8)	13.4 ± 0.39 (7.9)
N ₂ O	0.11 ± 0.02 ^d (0.13)	0.02 ± 0.01 ^a (0.02)	0.04 ± 0.09 ^b (0.03)	0.11 ± 0.019 ^c (0.06)	0.12 ± 0.05 ^c (0.07)
NH ₃	19.6 ± 1.13 ^b (23.1)	0.3 ± 0.13 ^d (0.4)	70.6 ± 1.19 ^c (41.5)	0.8 ± 0.23 ^d (0.5)	2.96 ± 0.52 ^a (1.7)
N diff ¹	25.8 ± 2.73 ^b (30.4)	25.3 ± 2.50 ^b (29.8)	33 ± 3.52 ^b (19.4)	58.9 ± 4.56 ^c (34.6)	8.79 ± 5.81 ^a (5.2)

Means with different letters in a row are significantly different ($p < 0.05$); $n = 4$.

¹ Unaccounted N.

Nitrogen partitioning differed markedly with acidification and N rate (Table 1). Biomass ¹⁵N recovery increased by 75–82% following acidification, while AS showed the highest recovery due to greater N availability. Soil ¹⁵N recovery remained low (7.6–15.6%) and unaffected by treatment, likely reflecting limited microbial turnover under controlled greenhouse conditions. NH₃ losses were highest from untreated slurry at 170 kg N ha⁻¹ but were reduced 88-fold by acidification (Table 1). Losses showed a strong inverse relationship with ¹⁵N recovery. Doubling N input raised NH₃ emissions 3.6-fold for untreated slurry but only 2.7-fold for acidified slurry. Cumulative N₂O emissions were minimal (0.02–0.12 kg N ha⁻¹; 0.02–0.13% of applied N) but increased with rate and fertilizer type ($P < 0.05$). Acidification did not cause pollution swapping, consistent with field observations (Nyameasem *et al.*, 2023). Despite 19–35% of slurry N remaining unaccounted for, likely lost as N₂ or N oxides, these microcosm results indicate that acidification effectively reduces gaseous N losses and improves N efficiency.

Conclusion

Slurry acidification markedly reduced NH₃ emissions without increasing N₂O losses, thereby increasing plant N uptake and improving N-use efficiency. While higher N rates may enhance other nitrification or denitrification losses, the findings suggest that acidification boosts fertilizer value, reduces gaseous N losses, and supports climate-smart, sustainable grassland nutrient management.

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Litter decomposition of Marandu grass under nitrogen doses in the Cerrado biome

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Abstract

Brazil is the world's top beef exporter, mainly relying on pasture-based systems that shape production quality and sustainability. Nitrogen (N) fertilization is a strategy used to increase forage productivity. Fertilization may affect litter mass decomposition, thereby improving nutrient cycling in this system. This study aimed to evaluate litter decomposition of *Brachiaria brizantha* 'Marandu' under rates of N fertilisation. The experimental design was a randomized block design, with two blocks and four nitrogen rates (45, 90, 135, and 180 kg ha⁻¹ year⁻¹ of N, split into applications of 45 kg N). Marandu grass litter was placed in nylon bags and incubated for 0, 4, 8, 16, 32, 64, 128, and 226 days. There was a significant effect of N rate on the remaining litter mass ($p = 0.00011$). The percentage of litter incubated between days 0 and 226 decreased at 45, 90, 135 and 180 kg N ha⁻¹ year⁻¹, resulting in a decrease of approximately 32, 51, 45, and 46% in litter mass disappearance, respectively. The highest decomposition rates occurred at 90, 135, and 180 kg N ha⁻¹, with -0.0029 , -0.0039 , and -0.0031 g g⁻¹ day⁻¹ OM, respectively. Increasing N fertilisation applications in Marandu grass increases litter decomposition rates.

Keywords: *Brachiaria brizantha*, Cerrado biome, litter decomposition, nitrogen fertilisation, tropical grasslands

Introduction

Soil N bioavailability increases forage production by generating new tillers and leaves, which can enhance the productivity of grassland ecosystems (Cardoso *et al.*, 2020). The presence of N can impact nutrient cycling dynamics (Ren *et al.*, 2024) through increased biomass, the input of plant residues, such as litter deposited on the soil, and the provision of substrate for microbial N decomposers. Warm-climate grassland pastures in Brazil are typically sown on acidic and low-fertility soils, leading to their degradation due to the continuous extraction of nutrients through forage or animal production (Sanchez, 2002).

Furthermore, pasture fertilisation is not a common practice in Brazil, unlike other types of agriculture. When nutrient replenishment is non-existent, organic matter (OM) mineralization becomes the main source of nutrients, especially N and K (Dubeux *et al.*, 2007; Freitas *et al.*, 2011). Litter of warm-climate grass (C4) has low nutritional quality, delaying decomposition and favouring nutrient immobilization. In this sense, nitrogen fertilisation acts as an inducer that directly benefits decomposing microorganisms, thereby accelerating the mineralization process and increasing nutrient availability for the soil-plant-animal system.

Studies with *Panicum maximum* 'Mombaça' reported that increasing N doses improves the plant's morphophysiological and biological nutritional responses, allowing the identification of the optimal cost-benefit ratio between fertilisation levels (Euclides *et al.*, 2022). In this sense, the use of nitrogen fertilisers, in addition to increasing forage production, also provides an additional strategy to accelerate the rate of nutrient decomposition and mineralization. However, there are few studies on the effects of N fertilisation on warm-climate grass litter. Therefore, the objective of this study was to evaluate the effect of N doses on litter decomposition in *Brachiaria brizantha* 'Marandu' pastures.

Materials and methods

The experiment was conducted at Embrapa Gado de Corte, in Campo Grande, MS (20°27'S, 54°37'W, 530 m above sea level), from November 2024 to March 2025. The soil of the experimental area is classified as an Oxisol. A randomized complete block design was used, with two blocks and four rates of N fertilisation (45, 90, 135, and 180 kg N ha⁻¹ year⁻¹, split into applications of 45 kg N). The pasture sward was *Brachiaria brizantha* 'Marandu' under intermittent stocking, managed at a grazing height of 30 cm and a stubble height of 15 cm. The F1 Angus×Nelore crossbred steers, approximately 20 months of age and with an average initial weight of 327 kg, were used. The initial stocking rates were 5.35, 5.80, 4.74 and 4.78 AU ha⁻¹, and the final stocking rates were 3.75, 3.78, 4.02, and 3.96 AU ha⁻¹ at the 45, 90, 135, and 180 kg N rates, respectively. The average grazing days were 10, 12, 9, and 9, and the average of rest days was 44, 32, 32, and 26 for the 45, 90, 135, and 180 kg N rates, respectively.

Five nylon bags (10 × 20 cm) were used per incubation period by treatment, consisting of two empty bags and three bags with litter, each with a porosity of 75 µm. The nylon bags were oven-dried at 55°C before being placed with the litter samples. Senescent Marandu grass litter was collected while still attached to the plant before the N application. The senescent material was oven-dried at 55°C for 72 hours, and each nylon bag received 12 g of litter sample.

For each treatment, bags were incubated in situ in the field in contact with the soil, without overlapping, and covered with a thin layer of aboveground plant material. The existing litter mass was collected by sampling two 0.25 m² quadrats per plot. Samples were oven-dried at 55°C for 72 hours and weighed to estimate total litter dry mass. Nitrogen fertiliser was applied on the first day of the incubation period, and the remaining doses were subdivided and used each time the animals left the paddock. The litter bags were removed at 0, 4, 8, 16, 32, 64, 128 and 226 days of incubation. Two empty bags were collected per period to determine the bag weight. At each incubation period, the litter bags were collected, manually cleaned, and oven-dried at 55°C for 72 hours, and then weighed. All data were expressed as OM to eliminate the effects of mineral particles on nutrient concentration.

ANOVA of remaining litter mass was analysed using Python statistical models (version 0.14.0), with N rates a fixed effect and block and days as random effects, adjusted to a 5% significance level. Litter decomposition was obtained by fitting the double exponential nonlinear model proposed by Weider & Lang (1982). $Y = Ae^{-k_1t} + (1-A)e^{-k_2t}$, where Y = mass remaining at time t , A = fraction of rapid decomposition, $(1-A)$ = fraction of slow decomposition, k_1 and k_2 = decay constants and e = associated error. Estimated as the difference between the initial and final litter mass stocks.

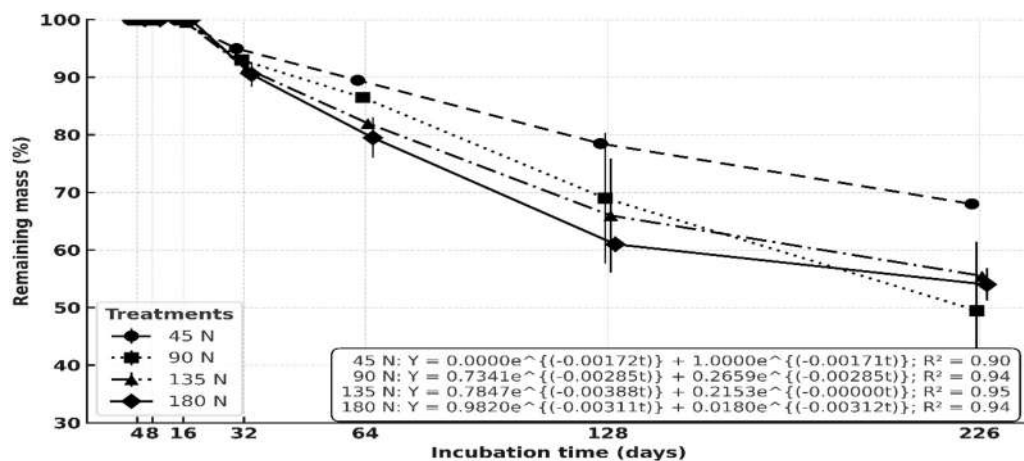


Figure 1. Litter mass remaining in Marandu grass after application of different nitrogen fertilisation.

Results and discussion

There was a significant effect of N rate on the remaining litter mass ($p = 0.00011$; Figure 1). The percentage of remaining incubated litter between days 0 and 226 decreased at 45, 90, 135, and 180 kg N ha⁻¹ year⁻¹, resulting in a decrease of approximately 32, 51, 45, and 46% in litter mass disappearance, respectively. The double exponential model separated litter decomposition into two fractions: easily decomposed (fast) and recalcitrant (slow). It was observed that 73, 78 and 98% of the decomposition at 90, 135 and 180 kg N ha⁻¹, respectively, occurred in the fast fraction, and for the rate of 45 kg N ha⁻¹, practically 100% occurred in the slow fraction. The highest decomposition rates occurred at 90, 135, and 180 kg N ha⁻¹, with -0.0029 , -0.0039 and -0.0031 g g⁻¹ day⁻¹ OM, respectively. The results for decomposition rates are similar to those observed by Ramalho et al. (2019) when using 150 kg N ha⁻¹, which may indicate a greater availability of labile material (Xiang *et al.*, 2024) in the incubated litter subjected to these N fertilisation rates.

Conclusion

Models suggest that increasing the rates of nitrogen fertilisation applied to Marandu grass litter accelerates its decomposition. Further studies on lignin, carbon, nitrogen, and their ratios in litter are needed to better understand the dynamics of nutrient mineralization.

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Effect of fertilisation on species composition and yield of lowland hay meadows under dry conditions

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Abstract

This study aimed to assess the effect of NPK fertilisation on the yield and species composition of meadow sward under drought conditions. The following were determined: soil cover with plants, yield, share of various plant groups and individual species in the yield, and biomass in the topsoil. The study was conducted during a period of drought. In the treatments without fertilisation, the results indicated an increase in the share of *Festuca rubra*, a species tolerant to a wide range of site conditions and lack of fertilisation (from 17.7% in the first year to 25.9% in the third year of the study), but the yields remained at a similar level of about 3 Mg DM ha⁻¹. NPK fertilisation resulted in vigorous growth and increased the competitive ability of tall grasses, especially *Dactylis glomerata* (up to 26.7% in the third year). On the other hand, a noticeable decrease in the share of legumes and other dicotyledonous plants was observed. In the third year, yields increased 2.7 times compared to yields from unfertilised treatments. The results also showed that the quantitative effect of NPK fertilisation on sward cover and biomass development in the topsoil layer was significantly smaller (statistically insignificant on average) than its effect on aboveground biomass.

Keywords: below-ground biomass, fertilisation, permanent meadow, species composition, sward cover, yielding

Introduction

Extreme weather phenomena, including long-term droughts, which have been intensifying in recent years (Łabędzki, 2007), increasingly determine the productivity of grasslands (Smith *et al.*, 2016). These phenomena are of particular concern in permanent meadows located on mineral soils, which are poorly supplied with groundwater, characterised by large fluctuations in soil moisture during the year, and low soil trophicity (Golińska *et al.*, 2016). Maintaining the proper species composition of the sward of these meadows is becoming increasingly complex. Additionally, their degradation intensifies due to the limited or complete omission of fertilisation. The study aimed to assess the effect of mineral fertilisation (NPK) on the yield of lowland hay meadows after several years of non-fertilisation. Furthermore, the study aimed to determine the interactions between species under drought conditions and depending on fertilisation.

Materials and methods

The study was carried out in the Pisia Tuczna river valley (central Poland), on a natural, moderately dry meadow site, on a degraded permanent meadow that was extensively managed (one or two cuts per year) and had not been fertilised for several years. The meadow was located on mineral soil (black earth), which was poor in potassium and had high phosphorus and magnesium concentrations. The meadow contained a low share of valuable tall grass species (less than 20%) and was infested with dicotyledonous herbs and weeds (approximately 40%). Sward cover was less than 50%. The experiment was designed as a complete randomised block with four replications. Mineral fertilisation was

Table 1. Average air temperature, precipitation totals, and climatic rainfall index in growing seasons (IV–IX) of subsequent years of study and in the multi-year period

	First year	Second year	Third year	Multi-year
Average air temperature (°C)	13.9	15.0	16.8	15.3
Sum of precipitation (mm)	304.2	271.2	346.7	366.1
Climatic rainfall coefficient ($\sum \text{mm} \cdot \sum ^\circ \text{C}^{-1}$)	0.119, very dry	0.098, extremely dry	0.113, very dry	0.131, dry

applied in the following annual doses in kg ha^{-1} : 180N (in three equal doses), 30 P (in one dose, in spring), and 100 K (in two equal doses—in spring and after the first cut). In subsequent years, three cuts were made per year. Every year, soil plant cover (visual assessment) using a 50×50 cm frame), yield (mower width 1.65 m $\times$ plot length 7.5 m), botanical composition, and biomass in the topsoil (0–10 cm, 10–20 cm, using a steel cylinder with a diameter of 110 mm and a height of 100 mm) were determined. The data were statistically analysed using analysis of variance (ANOVA). The significance of differences was verified using the Tukey test ($p \geq 0.05$). During the three-year study, weather conditions were unfavourable for plant development with high temperatures (in the second and third year) and long periods without rainfall. These were very dry periods, with the second growing season extremely dry (Table 1). The values of the Vinczeffy (1984) climatic hydrothermal index ranged from 0.098 (second year) to 0.119 (first year), with optimal conditions for the growth and development of meadow vegetation occurring when this index ranged from 0.2 to $0.25 \text{ mm } ^\circ \text{C}^{-1}$. The water table usually drops below 150 cm at the end of July, but in the second year of the study, it dropped already in the first days of June.

Results and discussion

Grasses dominated the yields from unfertilised plots (-NPK), with a high share of dicotyledon plants and the lowest share of legumes (Table 2). Among the grasses, *Arrhenatherum elatius* was the most abundant. In the following years, the share of species tolerant to various habitat conditions (drought, high and low air temperatures) and lack of fertilisation increased; primarily *Festuca rubra*. Legumes disappeared, especially *Trifolium dubium*, a 1–2-year-old species that regrows poorly after subsequent mowings. The share of other dicotyledonous plants decreased, especially low-growing species: *Achillea millefolium*, *Taraxacum officinale*, and *Leontodon autumnalis*. The applied fertilisation (+NPK) resulted in strong development of grasses and increased their competitive ability (Table 2). It particularly stimulated the growth and development of *Dactylis glomerata*. The high competitiveness of tall grasses has decreased the share of short grasses, especially the slower-growing *Festuca rubra*, which has lower soil nutrient requirements but is also less competitive. At the same time, in the first year after reintroduction of fertilisation, there was a significant decrease in the share of dicotyledonous plants, which accounted for (on average) half of the yields from unfertilised plots. Short species such as *Plantago lanceolata*, *Leontodon autumnalis*, and *Taraxacum officinale* declined. This demonstrates the significant potential for regenerating meadow sward productivity through fertilisation. The height of the meadow sward in the fertilised soil was significantly higher than in the unfertilised plots due to the strong growth and development of tall grasses. However, the sward cover was like that in the unfertilised plot. NPK fertilisation had no significant effect on sward cover due to the tussock-like habit of the dominant species (*A. elatius* and *D. glomerata*). The below-ground biomass in the fertilised treatment did not differ significantly from that in the unfertilised variant. This indicates a much smaller quantitative effect of NPK fertilisation on the development of biomass in the topsoil layer (0–10 cm), compared to the response to fertilisation of above-ground biomass. The plants produced the most

Table 2. The effect of fertilisation (+NPK) on the specified features of the meadow sward compared to unfertilised treatment (–NPK) in the years after the start of fertilisation

	First year		Second year		Third year		Mean for years	
	–NPK	+NPK	–NPK	+NPK	–NPK	+NPK	–NPK	+NPK
Group of plants and more important grass species (g kg ^{–1}):								
Grasses total	71.12	84.5	76.34	85.18	79.15	89.75	75.53	86.48
<i>Arrhenatherum elatius</i>	30.68	31.25	38.56	31.75	29.23	43.13	32.82	35.38
<i>Bromus inermis</i>	4.94	6.68	4.10	6.53	5.53	7.98	4.86	7.06
<i>Dactylis glomerata</i>	8.56	13.82	6.99	16.80	8.40	26.70	7.98	19.11
<i>Festuca rubra</i>	17.69	15.66	15.54	12.42	25.91	3.75	19.71	10.61
<i>Lolium perenne</i>	1.16	2.63	1.42	1.08	0.78	0.00	1.12	1.24
<i>Fabaceae</i>	4.44	2.90	1.19	2.43	0.01	0.00	1.88	1.78
Other dicotyledonous	24.44	12.60	22.47	12.39	20.84	10.25	22.59	11.75
Sward height (cm)	16.8 a	28.3 b	19.0 a	36.7 b	23.5 a	43.9 b	19.8 a	36.3 b
Sward cover (%)	50.7	52.7	44.4	45.0	51.6	51.4	48.9	49.7
Below-ground biomass (kg DM·m ^{–2})								
0–10 cm	0.882	1.293	1.596	1.672	1.252	1.171	1.243	1.379
Yield (kg DM ha ^{–1})	2898 a	4608 b	3292 a	6741 b	2667 a	7363 b	2952 a	6237 b
Average for years	3753.0 a		5016.5 b		5015.0 b			

Means in rows with different superscript letters indicate a significant difference at $P < 0.05$.

significant root mass in the 0–10 cm layer in the driest, second year. The increase in meadow sward yields under NPK fertilisation, compared to yields from unfertilised plots, amounted to approximately 111% on average. This confirms the very high yield potential of fertilisation. Yields increased in subsequent years of the study; in the second year, they were twice as high, and in the third year, they were as much as 2.7 times higher, compared to the yields obtained from the unfertilised plot.

Conclusion

The production potential of unfertilised lowland hay meadows in the Pisia Tuczna river valley is approximately 3 Mg DM ha^{–1}. The yield is closely linked to weather conditions, fertilisation practices, and the presence of forage grasses, which respond positively to fertilisation. The study indicated that yields can double in the second year and increase 2.7 times in the third year, compared to yields from unfertilised plots. The regeneration of the sward primarily resulted from the growth of tall grasses such as *A. elatius*, *D. glomerata*, and *B. inermis*, which are characterised by a high ability to utilize nutrients from fertilisers and tolerance to drought and high temperatures. Over the years, the share of these species in the yield increased, and in the last year, it was approximately 35% higher than in the yield from the unfertilised plot.

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Changes in floral biodiversity in Bavarian grasslands

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Abstract

The Bavarian State Research Center for Agriculture maintains an archive of more than 43,000 grassland vegetation surveys conducted across Bavaria (southern Germany) over a period of 70 years. These surveys document the occurrence of all vascular plant species and estimate the yield proportion of each species. The vegetation surveys were digitized, which now allows for an in-depth investigation of changes in plant species richness and plant community composition in and between the major natural regions of Bavaria. Preliminary analyses indicate that the average plant species richness on Bavarian agricultural grasslands has decreased throughout the decades, and that this decrease in species richness is accompanied by a shift in plant community composition. Three of the highly valuable forage species for intensive defoliation, *Lolium perenne*, *Poa pratensis* and *Trifolium repens*, increased significantly in yield proportion. In contrast, frequency and yield proportion of *Trifolium pratense*, which is less tolerant to defoliation, decreased. This suggests that due to the intensification in grassland management, competitive plant species outcompete less competitive species. Further investigations into the functional characteristics of both increasing and declining species are currently underway to shed light on the underlying processes.

Keywords: feeding value, frequency of *Trifolium pratense*, yield proportion of competitive grassland species

Introduction

Over the last few centuries there has been a massive loss of species worldwide. To investigate whether this global trend is also reflected in a decline in biodiversity at the regional level, biodiversity surveys over a longer period are necessary. Here, the current and historical vegetation surveys on agricultural permanent grasslands all over Bavaria conducted between 1950 and 2020 offer great potential for investigating changes in the diversity of Bavarian grasslands and possible causes over a period of 70 years. This will allow an in-depth investigation of changes in plant species richness and plant community composition in and between the major natural regions of Bavaria. Here we show first preliminary results.

Materials and methods

Over 43 000 vegetation surveys were conducted by the Bavarian State Research Center for Agriculture between 1950 and 2020 on agricultural permanent grassland throughout Bavaria, including 32 000 surveys as part of evidence preservation procedures and 11 000 surveys as part of the ‘Bavarian grassland monitoring’ project (2002–2020; Heinz *et al.*, 2023). In 2024/2025, we digitized the data using automatic text recognition (OCR) and georeferenced the records by digitizing paper maps. The vegetation surveys include yield proportions (visual estimation of biomass proportion) for each plant species (method Klapp/Stählin; Voigtländer and Voss, 1979). Using the feed value of each plant species according to Briemle *et al.* (2002, scale 1 to 9) and the yield proportion of each species, the community weighted mean of feed value was calculated for each survey. The vegetation surveys are

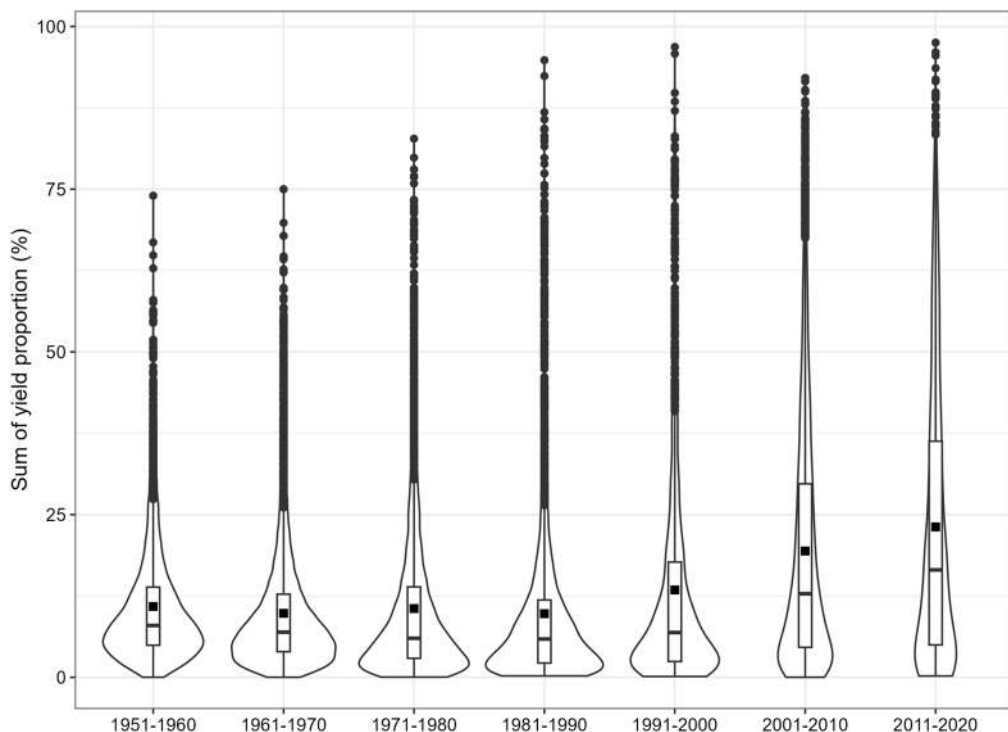


Figure 1. Changes of the sum of yield proportion (%) of *Lolium perenne*, *Poa pratensis* and *Trifolium repens* between 1951 and 2020.

evaluated both for Bavaria as a whole and separately for natural regions, and the temporal changes in the frequency and proportion of individual grassland species is calculated.

Results and discussion

Preliminary analyses indicate that the average plant species richness in Bavarian agricultural grasslands has declined significantly over the decades accompanied by a change in the plant community composition. This pattern applies both to individual natural areas and to Bavaria as a whole (Sweet *et al.*, 2025; Meyer *et al.*, not shown). These changes in species composition also affect the agronomic parameters of grassland communities.

But which plant species are the 'winners' and which are the 'losers'? The summed-up yield proportion of three grassland species (*Lolium perenne*, *Poa pratensis*, *Trifolium repens*) that are of high value for forage production in intensively used grasslands has increased over the decades (Figure 1). This increase can be attributed to an increased defoliation frequency by grazing or mowing and higher fertilisation rates, as well as reseeding.

However, this increase in the number and amount of competitive, use-tolerant grassland species has been accompanied by a decline in the frequency and yield proportion of less use-tolerant, yet valuable forage species. For example, the frequency of *Trifolium pratense* in one of the largest natural regions ('Unterbayrisches Hügelland and Isar-Inn-Schotterplatten') in which a relatively high proportion of all vegetation surveys ($n = 11574$) were carried out, declined over time. Between 1951 and 1960, *T. pratense* was present in almost every permanent grassland field (in over 95% of vegetation surveys).

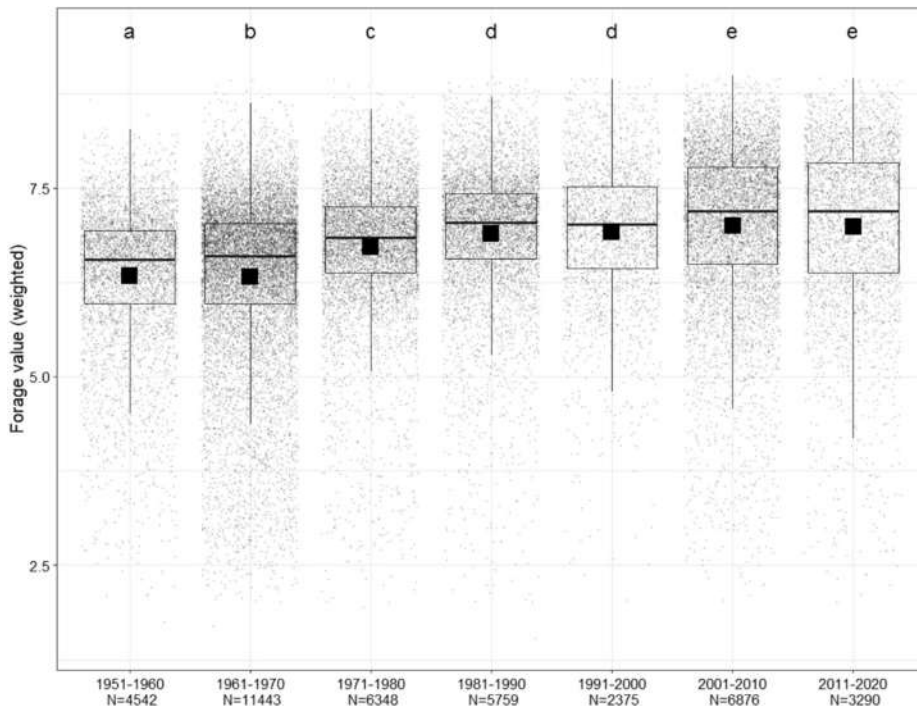


Figure 2. Changes of community weighted mean feeding value between 1951 and 2020.

Its occurrence has declined significantly over the decades, with *T. pratense* only being found in one of three grassland fields in the last three decades (1991–2020).

The community weighted mean feeding value averaged 6.2 for the 1951–1960 surveys and increased significantly to 7.4 for the 2011–2020 surveys (Figure 2). This indicates species shifts over time, with an increase in plant species valuable for forage production at the expense of species less competitive and less productive. Such a shift in composition likely contributed to the decline of plant diversity observed in Bavarian grasslands. Further investigations will analyse how species diversity in agricultural grasslands have been affected by local environmental and agronomic changes in the last 70 years.

Conclusions

Preliminary analyses show an increase of highly valuable and defoliation-tolerant grassland species, which is accompanied by a decrease in total species richness, frequency and yield proportion of less competitive grassland species.

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How earth observations and AI can support grassland understanding and management—examples from Switzerland

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Abstract

More than two-thirds of Switzerland's agricultural land consists of grassland used as meadows or pastures. These areas host a wide variety of plant species and communities, spanning diverse environmental conditions—from highly fertile and productive lowland sites to more marginal locations in the Alps. Consequently, there are a large range of uses and intensities of grasslands. Moreover, weather and other environmental factors influence grass growth, leading to considerable variation in productivity. Due to climate change and the need for optimized management, grasslands are under significant pressure. As a result, practices to adapt to climate change and mitigate its effects on grassland must be developed and tested.

In the last years we have developed the Agricultural Landscape Intelligence System that allows us to monitor grassland growth of the entire landscape. In this contribution, we outline how we use our platform to monitor grassland growth, identify risk areas, and train data-driven models to predict grassland growth based on environmental variables.

Keywords: agricultural management, artificial intelligence, earth observation, grassland monitoring

Introduction

More than two-thirds of Switzerland's agricultural land consists of grassland used as meadows or pastures. These areas host a wide variety of plant species and communities, spanning diverse environmental conditions—from highly fertile and productive lowland sites to more marginal locations in the Alps. Consequently, there are a large range of uses and intensities of grasslands. Moreover, weather and other environmental factors influence grass growth, leading to considerable variation in productivity.

Due to climate change and the need for optimized management, grasslands are under significant pressure. As a result, practices to adapt to climate change and mitigate its effects on grassland must be developed and tested. Additionally, grassland provides multiple ecosystem services. While grazing may help sustain these services (for instance, by controlling shrub encroachment), overgrazing combined with climate extremes can cause the opposite. This situation is likely to become more acute with increasingly variable weather conditions including more frequent periods of drought. Meanwhile, digital technologies are still widely underutilized and could make significant contributions to the development of more sustainable and efficient land and food systems by offering valuable insights to support the sustainable management of pastures by reducing labour demands and addressing emerging challenges.

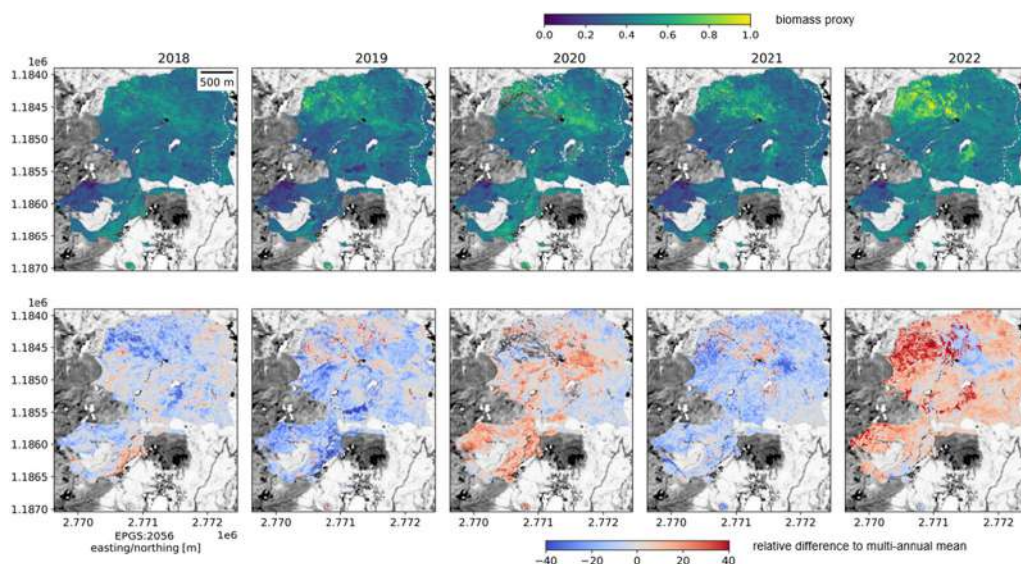


Figure 1. Biomass proxy maps (top) and relative difference of a year to the average of all years (bottom).

Materials and methods

Agricultural Landscape Intelligence System (ALIS). In the last years we have developed the ALIS which allows us to monitor grassland growth of the entire landscape. ALIS integrates multi-source data—including remotely sensed imagery, environmental data (e.g., weather, topography, soil), and in situ observations—to provide a comprehensive view of agricultural ecosystems. We interpret these data streams using state-of-the-art machine learning, mechanistic models grounded in ecophysiological processes, and expert knowledge from Earth Observation (EO), ecology, and agriculture. ALIS has demonstrated its capabilities also in the domain of grassland applications. Figure 1 shows an example of estimations of biomass per year and biomass fluctuations of a grassland area. In this contribution we show how this platform can be used to assess different growth response of mountain rangeland habitats to inter-annual weather fluctuations.

Annual Growth Curve Analysis. To extract pasture growth patterns, we selected pasture plots and calculated plant growth information, e.g. by the means of NDVI time series from all available images of these plots. From these data, a pixel-level database was created. For each pixel, the database stores NDVI value, habitat type, spatial grid coordinates, shadow mask, elevation, aspect, time stamp. Aspect and shadow mask values were derived from a DEM. This database enabled later filtering based on terrain orientation and illumination, ensuring more reliable growth curve generation. Annual NDVI growth curves per grassland plot were extracted for each year from 2016 to 2023. To reduce noise caused by complex mountain topography, the analysis considered only south-facing pixels (aspect 90–270°) and pixels outside mountain shadows. The growth curves were generated for four elevation bands: 2000–2200 m, 2200–2400 m, 2400–2600 m, 2600–2800 m.

Growth Season Indicators. From the growth curves, key statistical indicators were derived: (i) Start of Greening (SOG): First DOY where the median NDVI exceeds 0.05 for more than five consecutive days; (ii) End of Season (EOS): First DOY after which NDVI remains below 0.05 for more than five days. This fixed-threshold approach was chosen over dynamic SOG/EOS detection because it was

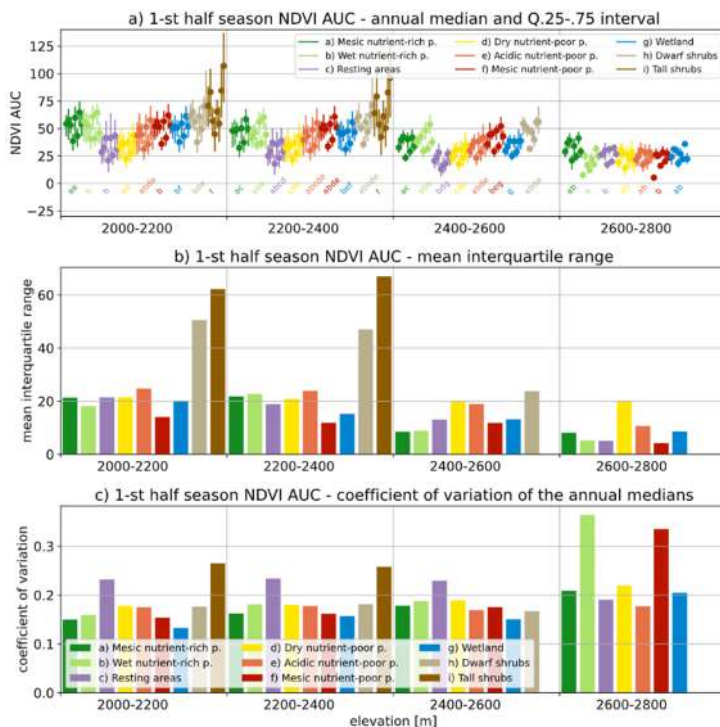


Figure 2. Plot of the NDVI AUC1 (see section 3.3) for different habitat units (different colors), years (same colour bars), and elevation classes (separated by vertical grid lines): (a) annual median (dots) and 25th–75th quantile envelope (error bar) for different years (2016–2023). Letter combinations in the same colour of the habitats show statistical similarity according to Tukey's HSD test. (b) Mean interquartile range (equivalent to the mean error bar length in a). (c) Coefficient of variation of the annual medians (dots in a) for every habitat unit.

robust to data scarcity and provided consistent indicator computation across habitats; (iii) Area Under the Curve (AUC): The AUC was calculated as a proxy for cumulative vegetation growth, computed in three ways: (iii.a) AUC1: From SOG to mid-season (fixed at August 1); (iii.b) AUC2: From mid-season to EOS; (iii.c) AUC: Entire season from SOG to EOS August 1 was chosen as the mid-season marker because it roughly corresponds to the transition between growth plateau and senescence in most habitats, aligns with pastoral practices, and is independent of the exact peak NDVI date, which is often noisy.

AUC was also calculated for the 0.25 and 0.75 quantile curves.

Results and discussion

From the analysis, two main types of growth patterns were identified—those belonging to dry pastures and those to wet pastures. This distinction is linked to vegetation responses to snow persistence and changes in elevation. The AUC (Figure 2) statistics show that mesic and wet pasture habitats have a larger cumulative growth and less spatial variability compared to dry pastures and resting areas, particularly during the whole growth season at elevations from 2000 to 2400 m a.s.l. At higher elevations, these differences diminish. As suggested by plot studies along elevation transects, this homogenization of pasture growth among habitats can be explained by changes in species composition caused by water scarcity and lower temperatures typically found at mountain tops. Under such conditions, more

productive and wet habitats may shift toward a species composition similar to that of dry pastures, resulting in reduced productivity.

The polygon-wise analysis of AUC in relation to elevation reveals a significant correlation between growth and elevation in dry and less productive pasture habitats. Growth during the first half of the season appears to be consistently more dependent on elevation than growth in the second half.

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Interaction of fire and freeze-thaw cycles alters soil nutrients and microbial communities

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Abstract

Pyric herbivory, the interaction of fire and grazing, is commonly used to maintain open habitats in mountain grasslands, which often experience freeze-thaw cycles in winter. Prescribed burns are typically applied at winter's end, but the interaction between fire and prior freeze-thaw stress on soil processes remains unclear. This study examined how fire affects soil nitrate availability and the composition of nitrifier populations under contrasting thermal conditions. We collected 30 intact soil blocks from a temperate mountain grassland in the western Pyrenees and subjected them to five treatments: (1) control (constant 5°C, no burning), (2) constant 5°C+burning, (3) freeze-thaw cycles (FTC)+no burning, (4) FTC+burning at 5°C, and (5) FTC+burning below 0°C. We measured nitrate availability, along with gene copy numbers of nitrifiers. FTC, alone and in combination with fire, increased soil nitrate availability. Fire significantly altered the microbial community structure by increasing the abundance of ammonia-oxidizing bacteria (AOB), while other nitrifiers remained unaffected. These results highlight that the combined effects of fire and freeze-thaw cycles can influence nutrient cycling and microbial dynamics, which should be considered when applying fire as a management tool in cold mountain ecosystems.

Keywords: burning, freeze, thaw cycles, N cycle, nitrifiers, nutrient availability

Introduction

Pyric herbivory—the interaction between fire and grazing—is a management tool used to maintain open habitats in mountain grasslands (Fuhlendorf *et al.*, 2009; San Emeterio *et al.*, 2023). Soils in these ecosystems often undergo freeze-thaw cycles (FTCs) during winter, while prescribed burning is typically conducted at the end of the cold season. FTCs can increase nutrient availability and nitrogen (N) losses due to microbial cell lysis, aggregate breakdown, enhanced mineralisation, and shifts in microbial communities (Gao *et al.*, 2018; Matzner & Borken, 2008). Prescribed burning can also lead to a temporary increase in nutrient availability (Múgica *et al.*, 2018). However, the interaction between fire and prior freeze-thaw stress on soil processes remains poorly understood. This study examines how simulated burning affects soil nitrate availability and the microbial communities of nitrifiers involved in nitrification enzyme activity under contrasting thermal conditions.

Materials and methods

We collected 30 intact soil blocks (20 × 20×20 cm) from a temperate mountain grassland located in the Special Area of Conservation Roncesvalles-Selva de Irati in the western Pyrenees at 1100 m asl. The soil blocks were incubated in the dark for two months under two different thermal regimes: a control at constant 5°C and a freeze-thaw cycle (FTC) consisting of 24 hours at –15°C followed by 72 hours at 5°C. During the freezing periods, soil temperatures remained mostly around –5°C,

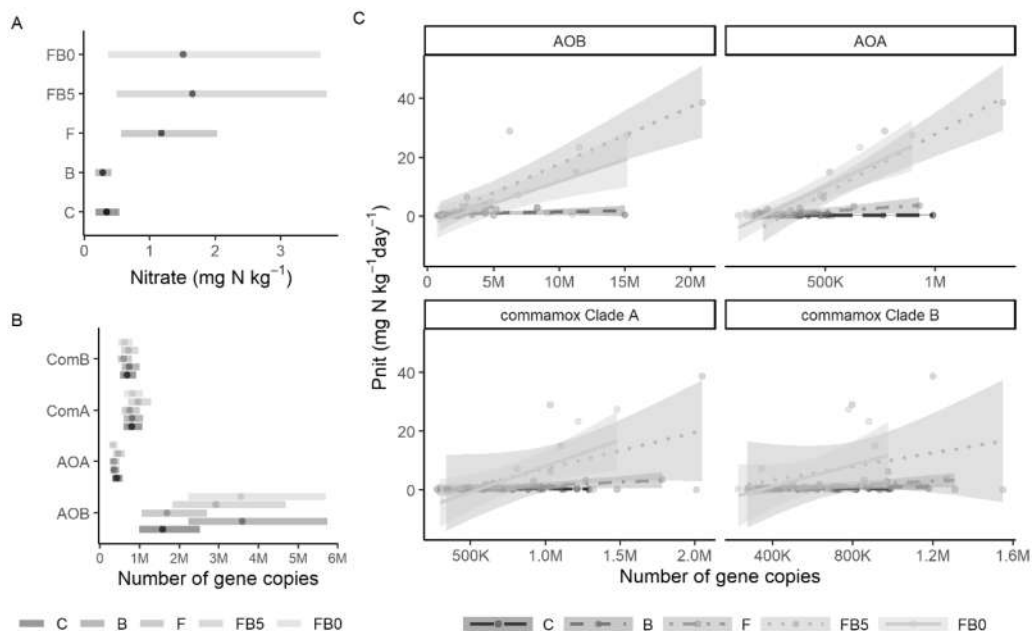


Figure 1. Nitrate availability (A), number of gene copies of nitrifiers (B) and potential nitrification rate (Pnit) as a function of the gene copies of nitrifiers (C) in soil under different combinations of freeze-thaw cycles (FTC) and simulated burning. C, control; B, burning; F, FTC; FB5, FTC+burning at 5°C; FB0, FTC+burning below 0°C.

occasionally dropping below this threshold. After this period, simulated burning was performed by placing 200 g of dry *Ulex gallii* Planch. (a dominant shrub in the area) on top of each block and igniting it with a cooking torch. The flame temperature reached approximately 750°C, and the residence time (duration above 50°C at the soil surface) was around 4.5 minutes. The combination of incubation and burning treatments yielded five experimental groups: (1) control (constant 5°C, no burning, C), (2) constant 5°C+burning (B), (3) FTC+no burning (F), (4) FTC+burning with soil at 5°C (FB5), and (5) FTC+burning with soil below 0°C (FB0). We measured soil nitrate and ammonium availability in 2M KCl extracts using an Autoanalyzer 3 (Bran Luebbe, Germany), immediately after burning and after 1 week. Potential nitrification rate and the gene copy numbers of nitrifiers were quantified following Miambi *et al.* (2022). Data were analysed using mixed linear models with soil blocks as a random effect.

Results and discussion

FTC treatments, either alone or combined with burning, significantly increased both the availability and variability of soil nitrate (likelihood ratio test (LRT) = 21.3, $P < 0.001$; Figure 1A). Ammonia-oxidizing bacteria (AOB) were the dominant nitrifying group (Figure 1B). Although ammonia-oxidizing archaea (AOA) are typically more prevalent in acidic soils, the AOB/AOA ratio observed here was consistent with that reported in other acidic environments (Petersen *et al.*, 2012). AOB abundance increased in response to burning (alone or in combination with FTC), while other nitrifier groups remained relatively stable (LRT for the interaction between nitrifier type and treatment = 64.2, $P < 0.001$; Figure 1B). Potential nitrification was low in undisturbed control soils (0.0–0.5 mg N kg⁻¹ day⁻¹), but increased substantially—up to 40 mg N kg⁻¹ day⁻¹—when both stressors were combined, correlating with increases in AOB and AOA (LRT for interaction between gene copy numbers and

treatment: AOB = 16.3, $P = 0.003$; AOA = 26.7, $P < 0.001$; Figure 1C). In contrast, potential nitrification was not significantly affected by the abundance of complete ammonia oxidizers (comammox) Clade A (LRT = 3.7, $P = 0.450$) or Clade B (LRT = 3.7, $P = 0.441$).

The higher nitrate levels after FTCs alone are likely due to nitrate release from microbial cell lysis rather than increased nitrification or shifts in nitrifier community composition. Conversely, burning—especially when combined with FTCs—stimulated AOB populations, the dominant nitrifiers in these soils, although nitrification potential and nitrate accumulation only rose when FTCs were also present. This indicates that post-fire nitrate enrichment likely results from the nitrification of mineralised organic compounds released during combustion and microbial cell lysis, rather than from direct nitrate release from cell lysis caused by thermal shock. FTCs appear to be more detrimental to microbial communities than low-intensity fires, where soil temperatures only briefly reach lethal levels in the uppermost millimetres.

Conclusion

This study demonstrates that FTCs significantly increase soil nitrate availability, primarily through microbial cell lysis rather than enhanced nitrification. In contrast, prescribed burning promotes the abundance of AOB, the dominant nitrifiers in these acidic mountain grassland soils, but only leads to increased nitrification potential when combined with prior FTC exposure. These findings suggest that the timing and sequence of thermal disturbances play a critical role in regulating nitrogen dynamics. While FTCs act as a direct stressor disrupting microbial cells, burning appears to stimulate nitrification indirectly by promoting the microbial processing of fire-released organic substrates. Understanding these interactions is essential for predicting nutrient cycling under combined disturbance regimes and for optimizing fire-grazing management strategies in temperate mountain grasslands for minimizing nitrate losses.

Acknowledgements

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Establishment of ryegrass pastures in mixture with clovers to reduce nitrogen fertilization and enhance sustainability in southern Chile

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Abstract

The sustainability of pasture-based livestock production systems is becoming increasingly important. One effective strategy to enhance sustainability is to include clovers in mixtures of ryegrasses while simultaneously reducing synthetic nitrogen (N) fertilization. To evaluate this approach, two independent trials were established: one with combinations of perennial ryegrass (PR) and white clover (WC) and the other with combinations of hybrid ryegrass (HR) and red clover (RC). Results from the establishment year indicated that including WC in mixture with PR, and RC in mixture with HR, enabled a reduction or complete elimination of nitrogen fertilization without compromising forage production and increasing forage quality. Further evaluations are currently underway to assess second-season performance, as well as the potential benefits of clover inclusion on N balance and overall system sustainability.

Keywords: hybrid ryegrass, nitrogen fertilization, red clover, sustainability

Introduction

Ryegrasses are key forage species for animal production in southern Chile. In recent decades, they have commonly been sown as monoculture swards and managed with high rates of synthetic nitrogen fertilizer. However, the pursuit of more sustainable pasture-based production systems remains a priority. Incorporating clovers in mixture with ryegrasses in pastures represents a promising strategy to enhance sustainability. The aim of this study was to assess the impact of including white clover in perennial ryegrass pasture swards, and red clover in hybrid ryegrass pasture, alongside reduced nitrogen fertilization, on the agronomic performance of ryegrass pastures.

Materials and methods

Two independent field trials under irrigation were established in September 2024 at the INIA Carillanca Research Station in Chile (38°41'S, 72°25'W; 188 m asl). In each trial, four treatments were arranged in a randomized complete block design with four replicates, using plots of 10.5 m² (3 × 3.5 m). All treatments received the same basal fertilization, based on results of soil analysis from a 0–20 cm soil sample taken before sowing. The pre-sowing soil analysis indicated an organic matter content of 17.5%, water pH 6.0, aluminium saturation of 1.0%, available nitrogen of 7.3 mg kg⁻¹, Olsen P of 14.6 mg kg⁻¹, exchangeable K of 247.5 mg kg⁻¹, and a base sum of 7.1 cmol(+) kg⁻¹. Treatments were as follows: T1, ryegrass (R) monoculture fertilized with 190 kg N ha⁻¹ during the first season; T2, R–clover (C) mixture fertilized with 80 kg N ha⁻¹; T3, R–C mixture without N fertilization; and T4, C monoculture without N fertilization. Nitrogen (N) was applied as urea in five split applications in T1: 30 kg N ha⁻¹ pre-sowing and four applications of 40 kg N ha⁻¹ during the season. In T2, 20 kg N ha⁻¹ were applied four times during the season. Ryegrass was sown in rows 18 cm apart

and clover broadcasted. In the perennial ryegrass (*Lolium perenne* L., PR) and white clover experiment (*Trifolium repens* L., WC), PR cv. Hustle AR1 (15 kg ha⁻¹) and WC cv. Legacy (4 kg ha⁻¹) were used; in the hybrid ryegrass (*Lolium x boucheanum* Kunth, HR) and red clover experiment (*Trifolium pratense* L., RC), HR cv. Shogun (20 kg of seed ha⁻¹) and RC cv. Perduro INIA[®] (10 kg of seed ha⁻¹) were sown. Forage yield was evaluated five times in the PR-WC trial and four times in the HR-RC trial, sampling 6.48 m²/plot with a bar mower at a cutting height of 4–5 cm. We estimated dry matter content (DM), forage yield by species (kg DM ha⁻¹) and forage quality (crude protein, CP; neutral detergent fibre, NDF; metabolizable energy, ME) in a composed sample per treatment of the March harvest. We also evaluated chlorophyll content previous to each harvest using a SPAD-502 meter (Minolta Camera Co. Ltd., Japan), measuring five leaves per species in each plot and time. Data were analysed using a repeated measures mixed model, including fixed effects of treatment (T), time and their interaction, and random effects of block and plot nested within treatment. When the treatment effect was significant ($P \leq 0.05$), treatment means were compared using Tukey's HSD test. If interactions were significant ($P \leq 0.05$), treatment means were compared within each time using Tukey's HSD.

Results and discussion

The soil analysis (0–10 cm) at the end of the season, averaged across both trials, showed an organic matter content of 17.5%, a pH (water) of 6.3, aluminium saturation of 0.3%, available nitrogen of 13.9 mg kg⁻¹, Olsen P of 16.9 mg kg⁻¹, exchangeable K of 193.3 mg kg⁻¹, and a base sum of 7.8 cmol(+) kg⁻¹.

In the PR–WC trial, significant effects of T, time, and their interaction ($T \times \text{time}$) were observed on the forage yield of the sown species (Table 1). In four of five harvests, there were no significant differences in forage yield among treatments. However, in the third harvest, T2, T3, and T4 outyielded T1. Regarding PR forage yield, in two harvests, T1 produced higher PR yields than T2 and T3. In contrast, WC yield was only affected by sampling time ($P < 0.001$), being highest in March and lowest in December and June. Crude protein (CP) and neutral detergent fibre (NDF) values in March were 14.0–47.7%, 17.7–39.6%, 18.6–37.7%, and 21.5–30.4% for T1, T2, T3, and T4, respectively. The higher CP of T2, T3 and T4 is consistent with the greater SPAD values observed in WC compared with PR (43.2 vs. 31.8).

In the HR–RC trial, significant effects of T, time, and their interaction were observed on the forage yield of the sown species (Table 1). In the first, third, and fourth harvests, no significant differences were detected among T1, T2, and T3, whereas in the second harvest, T2 and T3 outyielded T1. Whereas HR yield of T1 was higher than the mixtures (T2 and T3), with no significant differences ($P > 0.95$) between the two mixture treatments. On the other hand, RC monoculture (T4) outperformed T2 and T3 in the second harvest, and was also superior to T2 in the third harvest. Crude protein (CP) and neutral detergent fibre (NDF) values in March were 13.3–40.3%, 15.4–41.3%, 18.4–38.7%, and 20.3–37.6% for T1, T2, T3, and T4, respectively. The higher CP of T2, T3 and T4 is consistent with the greater SPAD values observed in RC compared with HR (46.8 vs. 34.2).

We obtained a high contribution of clovers, averaging 69% for WC and 55% for RC. Nyfeler *et al.* (2011) reported that PR–WC mixtures were more productive and efficient than monocultures, and defined an optimal WC proportion of 40–60%. By contrast, Chapman *et al.* (2018) reported an average WC proportion of 20% in PR mixtures under grazing conditions.

For HR–RC, Harrison *et al.* (2024) observed a RC contribution exceeding 50% in the first year, decreasing to approximately 40% by the third year. In our PR–WC experiment, mixtures outyielded

Table 1. Forage yield (kg DM ha⁻¹) during the establishment year in two independent trials: perennial ryegrass-white clover and hybrid ryegrass-red clover.

	Treatments (T)				SEM	p value		
	T1	T2	T3	T4		T	Time	T × Time
Perennial ryegrass-white clover trial (five times of evaluation)								
Perennial ryegrass	7738	3611	2670	—	542.8	<0.0001	<0.0001	0.0002
White clover	—	6899	6949	8911	753.6	0.0851	<0.0001	0.1662
Sown species	7738	10510	9619	8911	456.8	0.0106	<0.0001	0.0020
Hybrid ryegrass-red clover trial (four times of evaluation)								
Hybrid ryegrass	11140	6409	4965	—	598.4	0.0002	0.0501	0.3993
Red clover	—	6647	7156	10637	618.1	0.0055	<0.0001	0.0134
Sown species	11140	13055	12121	10637	585.6	0.0142	<0.0001	<0.0001

Treatments: T1, ryegrass (R) monoculture with 190 kg N ha⁻¹; T2, R+clover (C) with 80 kg N ha⁻¹; T3, R+C without N fertilization; T4, C monoculture without N. SEM: standard error of means (n = 4 blocks); —, treatment does not include the respective species.

the fertilized PR monoculture by 30%, whereas in the HR–RC experiment this difference averaged 13%. We also observed an improvement in forage quality when clovers were included in the mixtures. The advantages of grass–clover mixtures observed in this study are consistent with previous research, which has highlighted not only their higher productivity but also their potential to reduce nitrogen fertilizer requirements and enhance forage quality (Lüscher *et al.*, 2014; Chapman *et al.*, 2018; Harrison *et al.*, 2024; Nyfeler *et al.*, 2024).

Conclusions

It is concluded that, during the establishment season, the inclusion of WC in mixtures with PR and RC in mixtures with HR allowed a reduction or even complete elimination of nitrogen fertilization, without compromising forage production and while improving forage quality. The performance during the second season, as well as additional potential benefits of clover inclusion—such as enhanced system sustainability—is currently being evaluated.

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Session 2.
Agroforestry, pastoralism and society

Agroforestry Innovations in Europe

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Abstract

Agroforestry (AF) is a sustainable land use practice and system able to contribute to all EU CAP goals. AF also contributes to the delivery of ecosystem services as it is based and promotes biodiversity increases biomass production and resilience against climate change when positively affecting water quality and contribute to the climate change mitigation. However, only 10% of the grasslands and 0.01% of the arable lands are currently using agroforestry in Europe attending to the LUCAS data. A set of Regional Agroforestry Innovation Networks were established in Europe thanks to the EU projects AFINET and AF4EU to understand the reason behind the reduced implementation of agroforestry in Europe. Main reasons where the lack of knowledge, the lack of a tool to understand the future agroforestry scenarios optimizing different sustainable parameters that are adapted at local level. AF4EU developed a tool based on three representative farms in 10 countries and 11 regions of Europe that were parametrized based on economic, environmental and social indicators to help farmers to incorporate agroforestry in their farms aided by an adequate establishment of an advisory formation tool.

Keywords: agrosilviculture, agrosilvopasture, decision support tool, silvopasture

Introduction

Agroforestry is one of the agroecological or land management techniques linked to the temporary and spatial use of biodiversity able to fulfil most of sustainable goals provided by United Nations (UN, 2026) linked to the economic, environmental and social progress in the world. From an agroecosystem perspective, supporting agroforestry as land management to foster worldwide sustainability is a form to protect waters, enhance nutrient cycling, combat climate change and preserve and use biodiversity while optimizing the use of the resources (Mosquera-Losada *et al.*, 2018b). However, there is a small share of agroforestry in Europe initially caused by the green revolution that promoted productivity without considering environmental and social aspects in rural areas. The EU CAP did contribute to the destruction of large number of woody perennials including trees across the EU landscape due to the land consolidation and the reduction of payments linked to the woody perennial component presence (Santiago-Freijanes *et al.*, 2018).

The environmental and societal challenges associated with the green revolution and the subsequent promotion of the land management intensification caused strong challenges still persisting nowadays such as biodiversity losses in Europe (Walder *et al.*, 2006). An assessment of the state of nature shows that 81% of habitats, 39% of birds and 62% of non-bird species of **community interest** are in a poor or bad state. The reduction of the environment quality was caused mainly by high fertilization rates affecting the water quality, the landscape fragmentation due to the land consolidation, no-renewable energy use that heavily impacted environment by enhanced extreme weather events (EEA, 2025).

The Sustainable Development Goals (SDGs) adopted by United Nations in 2015 integrates different economic, environmental and social challenges to foster sustainability across the world, contributing agroforestry to most of them as highlights the International Institute for Sustainable Development (IISD). AF can contribute to the SDG1, 2, 3, 4, 5, 6, 12, 13 and 15 (Anderston, 2018). Moreover, agroforestry is also able to foster the global and European policies to foster agricultural sustainability as highlighted Santiago-Freijanes *et al.* (2021). However, and in spite of the global benefits that agroforestry may play, the extent of agroforestry in Europe is reduced (Mosquera-Losada *et al.*, 2018) in spite of the specific policy support provided by the CAP in the implementation of the past as the measures 222 and 8.2 for the CAP 2007–2013 and 2014–2020 period (Mosquera-Losada *et al.*, 2018a).

Fostering adoption of agroforestry should be based in different pillars linked to the development of research, technology and innovation at both agroecosystem and agrifood levels to understand the holistic approach of the agroforestry concept based on the agricultural and forestry spatial and temporary use of biodiversity and the impact it has on rural revitalization and development. Agroforestry research is needed to understand the adequate combination of agroforestry components and species to a specific environment, agroforestry knowledge is needed to understand agroforestry practice and business models implementation and agroforestry innovation is needed to foster adoption based on adequate advisory services assessing to EU foresters and farmers to reach an adequate share of agroforestry at EU level.

This keynote aims at understanding the agroforestry concept linked to the CAP policy promotion in Europe considering the whole agrifood system from the agroecosystem to the business models development.

Agroforestry, CAP and sustainability

Agroforestry, as an agroecological land management technique, but also a sustainable system linked to multiple products delivery, is able to fulfil most of the CAP objectives associated with the main sustainability goals described by the FAO: economic, environmental and social. Figure 1 shows the main CAP 2023–2027 objectives linked to the different EU strategies and the main FAO components of agroecology where agroforestry takes place and can provide benefit to all systems.

The economic 2023–2027 CAP goals are linked to the promotion of the fair income, competitiveness and the food value chain and are based in the EU circular economy, bioeconomy and the farm-to-fork strategies, and linked to the agroecology concepts of efficiency, recycling and circular and solidarity economy. Agroforestry as a multiproduct system is able to increase the amount of products delivered from a unit of land therefore fostering bioeconomy linked to the territory. As the main product is related to the biomass, this can be processed and used from multiple purposes such as energy, furniture, compost, among others. The agricultural component of agroforestry is directly linked to the farm to



Figure 1. CAP 2023–2027 aims (black), FAO agroecology elements (green) and EU linked strategies (red).

fork strategy as usually agroforestry products are related to extensive practices based on biodiversity use that has a high appreciated value (i.e., honey, meat, milk, among others) and are usually associated with short value chains.

The environmental 2023–2027 CAP goals are linked to climate change, environmental care and landscapes and are associated with the climate change, forest and biodiversity EU strategies and related to the FAO agroecological concepts of Resilience and Diversity. The capacity of the woody component of agroforestry is linked to a high capacity of sequestering carbon in both aboveground and in the soil. Environmental care is ensured thanks to the largest and deepest root network that is able to prevent nutrient leaching such as nitrates towards the water bodies and reintegrate it in the agroforestry system through tree residue depositions. Landscape is promoted as the combination of trees/shrubs and grasslands/arable crops is very well perceived by consumers.

The social 2023–2027 CAP goals are based on one of the most relevant EU social challenges such as ageing, poor development and health related to the CAP goals of generational renewal, rural areas and food-health. These CAP goals are linked to the agroecology FAO component of culture and food traditions and human and social values. The fact that agroforestry is able to deliver a high number of products per unit of land enhances productivity and therefore the farmers income if adequate value chains are developed that therefore ensures rural areas vitality.

The tenth EU objective is the knowledge and innovation linked to the FAO agroecology component of knowledge share and cocreation. In Europe, this objective is linked to the development of the EIP-Agri in 2014 developed until 2020 and further integrated in the CAP-Network after 2020, therefore mostly encompassing the CAP periods. The EIP-AGRI/CAP-Network are core to develop operational groups and H2020 and HEuropa multi-actor projects that involves relevant actors to co-create and favour sustainable practices uptake. Agroforestry have been involved in different EU multi-actor Research and Innovation Projects such as AGFORWARD and AGROMIX linked to agroecosystems, DIGITAF

and DIGIRANGELAND linked to AF digitalization and the Coordination and Support Actions (CSA) AFINET and AF4EU that gathers information to support AF agroecosystem, digitalization, advisory, value chains and business models.

Agroforestry definition

Policy makers need clear and easy definitions to promote sustainable land management practices such as agroforestry to understand the baseline of the practice at territory scale and to establish a monitoring system to further evaluate the agroforestry policy impact. Agroforestry definition has been largely discussed since Nair (1985, 2022). Agroforestry—the practice of growing trees and crops in interacting combinations—is recognized the world over as an integrated approach to sustainable land-use” that opened the discussion about the definition of a practice combining different productive species always including woody perennials. Many papers have been written about the definition of agroforestry (McAdam *et al.*, 2009; Mosquera-Losada *et al.*, 2009; Nair, 1985; Nair *et al.*, 2022). They include simple definitions such as from “growing trees on farms” (Young, 1997) to more technical definitions. Within the AGFORWARD project, building on definitions by MacDicken and Vergara (1990), Nair (1994) and Mosquera-Losada *et al.* (2009) defined agroforestry as “the practice of deliberately integrating woody vegetation (trees or shrubs) with crop and/or animal systems to benefit from the resulting ecological and economic interactions”. This definition is also similar to the definition used by the European Agroforestry Federation (EURAF, 2017) which is that agroforestry is “the integration of woody vegetation, crops and/or livestock in the same area of land. Woody vegetation can be inside parcels or on the boundaries (hedgess)”.

The above definitions are useful in an academic and promotional context as they envisage agroforestry as a range of components, there is an emphasis on interactions, also implying that there are benefits from the interactions. By contrast, in a policy context the definitions of agroforestry tend to relate more specifically to agroforestry practices and land cover and land use. Our definition is in line with most of the policy definitions across the world (i.e., ICRAF as cited by FAO (1993), FAO (2015), or AFTA (2015) in North America) and stands as “A collective term for land management systems where woody perennials (trees, shrubs, etc.) are deliberately integrated with agricultural crops and/or animals, in some form of spatial arrangement or temporal sequence” An argument for a focus on “trees and shrubs” as woody perennial rather than “trees” is that shrubs can provide many of the same productive, environmental or social benefits of trees. Moreover, tree definitions vary across countries and trees can also be cultivated in a shrub shape, while providing the same environment and social benefits (Mosquera-Losada *et al.*, 2016b). For this report, agroforestry is defined within a specific edaphoclimatic context as “the integration of woody vegetation (first component) in at least two vertical layers on land, with the bottom layer providing an agricultural product such crops or forage/pasture (second component) which may be consumed by animals (third component). The distribution of the woody vegetation can be uneven or evenly distributed and the woody component can deliver an agronomic product (fruit, forage) or some other ecosystem service”.

Agroforestry practices links the presence of woody vegetation (trees or shrubs) with an agricultural product obtained from the understory and managed by the farmer (FAO, 2015). The ‘forestry’ component can include shrubs, or fruit/nut trees, as well as timber and biomass trees. Agroforestry can be implemented in a spatial or temporary scale depending on the use of the woody perennials. Temporary agroforestry practices consist of using woody vegetation (mainly legumes) that is used to subsequently improve soil fertility (Mosquera-Losada *et al.*, 2009) for subsequent cropping or animal rearing. This

Agroforestry practice	Description	
Silvopasture		Combining woody with forage and animal production. It comprises forest or woodland grazed and pastoral land with hedgerow isolated/scattered trees or trees in lines or belts
Agrosilviculture		Widely spaced woody vegetation inter-cropped with annual or perennial crops. Trees/shrubs can be distributed following an alley cropping, isolated/scattered trees, hedges and line belts design
Agrosilvopasture		Combination of woody perennials with crops that are grazed. Such as the grazing of triticale by pigs in the Spanish dehesa (Santamaria et al 2017)

Figure 2. Spatial agroforestry practices in Europe.

practice is still practised globally and was traditionally used in Europe. The most common spatial agroforestry practices in Europe include silvopasture, agrosilviculture and agrosilvopasture (Figure 2). Agroforestry can be applied at plot as a practice, at farm as a system both delineating the landscape scale.

Agroforestry practices are typically implemented at a plot level and can have the woody component spatially (i.e., multipurpose trees in silvopasture or hedgerows) and/or temporally (alternation of silvopasture and silvoarable or including woody vegetation in fallow lands) combined in different farming systems. Agroforestry requires the expertise of woody vegetation specialists/farmers like foresters (to understand and manage long term cropping) and arable and livestock farmers/experts (to understand and manage the short-term crops and/or livestock production). From a policy point of view, agroforestry policies should support and promote the adequate management of woody vegetation at plot level to provide productive and ecological benefits while ensuring more sustainable farming systems (Mosquera-Losada *et al.*, 2016) also considering farm and landscape scales (Figure 2).

Agroforestry practices implemented at plot scale can be combined at farm (agriculture or forest) level creating different types of agroforestry systems. Thousands of types of agroforestry systems comprising different combinations of woody vegetation, animals and herbaceous components adapted to local conditions (Figure 3).

Silvopasture practice could be carried out in both open (e.g., wood pasture and parkland) or more closed areas (e.g., grazed or meadow orchards), and with different woody vegetation distribution (e.g., hedgerows and windbreaks). Because arable crop production is generally mechanised, trees have been destroyed and crop varieties adapted to open sites have been selected. By contrast, pasture understories (which are often biodiverse) can usually tolerate greater levels of shade than arable crops and thereby can remain relatively productive in such situations (Pardini *et al.*, 2010). These are some of the reasons why agrosilviculture practices are not usually associated with “forestland”. However, there is the potential to identify some better shade- adapted crop species and crop varieties. There are also crops

Land use and agroforestry practice		Common name	Brief description
AGRICULTURE	Silvopasture	Wood pasture and parkland.	Typically areas of widely-spaced trees that are also used for forage and animal production.
		Meadow orchards	This practice includes fruit orchards, shrubs which are grazed or sown with pastures, but also olive groves and vineyards
		Hedgerows and windbreak system	Here the woody components are planted to provide shelter, shade, or parcel demarcation to a crop and livestock production system
	Agrisilviculture	Alley-cropping systems	Widely spaced woody perennials inter-cropped with annual or perennial crops. It comprises alley cropping with scattered trees and orchards and line belts within the plots. These practices are sometimes found only during the first few years of the plantation
		Riparian buffer strips	Areas of tree and shrubs allowed to establish along croplands/pastures and water sources such as streams, lakes, wetlands, and ponds to protect water quality, can be identified as silvoarable or silvopasture.
	Agrosilvopasture	Dehesa	Areas where trees and/or shrubs are combined with crops and grazed by animals.
FOREST	Silvopasture	Forest grazing	Although the land cover is described as forest, the understory is grazed
	Agrisilviculture	Forest farming	Forested areas used for production or harvest of naturally standing specialty crops for medicinal, ornamental or culinary uses
URBAN AND PERIURBAN	Agrisilviculture	Homegardens	Combining trees/shrubs with vegetable production usually associated with peri-urban or urban areas
	Agrosilvopasture	Grazed homegardens	Combining trees/shrubs with vegetable production usually associated with peri-urban or urban areas that has animals such as chicken

Figure 3. Agroforestry practices linked to main farm types and land use (agriculture, forest or peri-urban).

that prefer to have shade conditions instead of being grown in open sites, particularly in southern EU countries where light intensities and temperatures are high.

Agroforestry extent

Agroforestry is poorly present in Europe, silvopasture is only implemented in around 10% of the potential grassland area, while only the 0.01 % of the arable lands (Mosquera-Losada *et al.*, 2018b) are managed under the agrosilviculture or agrosilvopasture agroforestry practices (Figure 4). This fact is against the main CAP goals linked to the sustainability. The main reason behind this low AF adoption is linked to the lack of a clear definition of agroforestry to be supported at local level, the lack of knowledge, the lack of supportive business environment.

The AFINET project searched the reasons behind the low degree of acceptance of agroforestry and prioritized them as can be seen in Table 1.

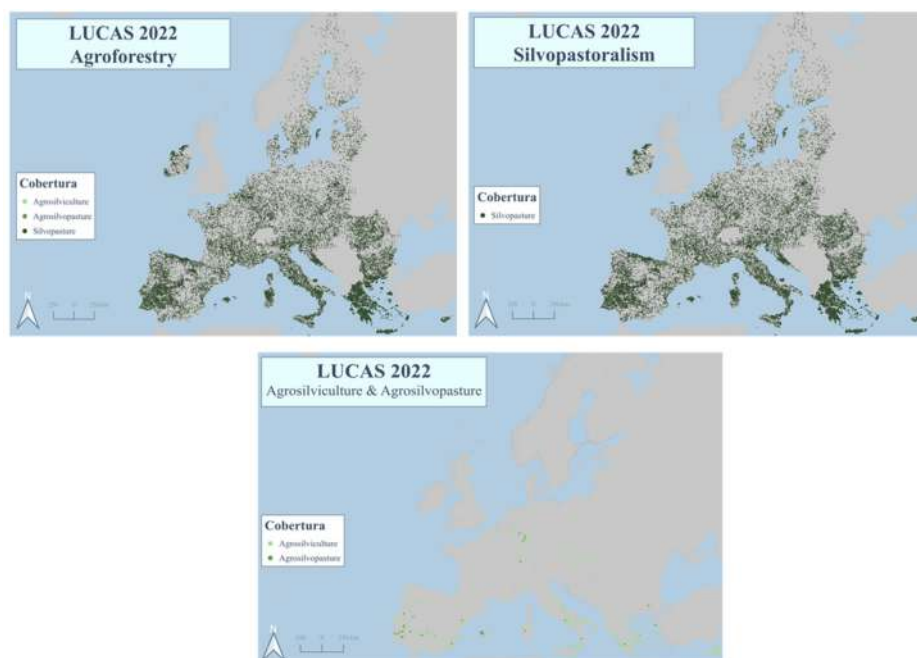


Figure 4. Agroforestry, silvopasture, agrosilviculture and agrosilvopasture extent in Europe.

Table 1. Main economic, technical, educational and policy challenges to foster agroforestry in Europe

Economic	Technical
Farming systems economic value DST // alternative use of trees and shrubs through bioeconomy	Planting the right tree in the right place // enhancing agroforestry// agroforestry as a climate change tool
Educational	Policy
Lesson modules at all education levels//AF online map// local challenges inventory	Inventory of problems of each region and how was it solved//Agroforestry, CAP, aims and sustainable development goals

Agroforestry, biodiversity and ecosystem services

Agroforestry is clearly fostering the delivery of several ecosystems services such as those linked to productivity, water protection and climate change based on the promotion and use of biodiversity with different biodiversity traits including herbaceous and woody perennials.

Biodiversity and agroforestry

Agroforestry promotes biodiversity as it is linked to the presence of at least two different ecological traits (woody and herbaceous) plant components. But this fact generates heterogeneity in the territory enhancing both beta and alpha biodiversity. Generally, grasses are dominant below the tree while dicots are dominant far away from the tree due to the higher nutrient inputs (from the tree) below the tree and the shade that modifies the temperature and humidity favouring different plant species. These patches of plant biodiversity are encompassed of different microbiota, arthropods and polychaetas families. Moreover, when we speak about silvopasture, biodiversity is enhanced by the selection of different species by the livestock promoting ones over another species, trampling and dung patches that generates habitat heterogeneity.

Productivity and agroforestry

The amount of biomass generated in an agroforestry system is higher than in agricultural lands without trees, this is due to the largest amount of the vertical aboveground biomass photosynthetically active (woody perennials + herbaceous). The eointensification or the optimization of the use of the resources generates an increase in biomass production and therefore causes a sustainable way of land intensification. The land equivalent ratio (LER) indicator reveals that 1 ha of AF is equivalent to 1.4 ha of crop+forestry biomass production when they are grown separately (Seserman *et al.*, 2018). At the end, productivity is enhanced due to the increased biodiversity allowing the multipurpose use of land and optimizing the resources use (radiation, temperature, water, and nutrients), if adequate components (tree/shrubs/crops/animals) are integrated and adequately managed by considering the positive interactions among them (Mosquera-Losada *et al.*, 2016; Santos *et al.*, 2022).

Climate change

The woody vegetation growth is essential for both climate change mitigation and adaptation (Mosquera-Losada *et al.*, 2017). Concerning mitigation, AF has an enormous capacity of increasing carbon (C) sequestration in agronomic and forestland, while avoiding or counteracting emissions and the effects of catastrophic events mostly due to the increase of resilience. Agroforestry is one of the most cost-effective technologies to sequester carbon and therefore mitigate climate change as provided by the IPCC in the Global Warming of 1.5°C report. Moreover, AF has been demonstrated to be useful in reducing the risk of GHG emissions as a fire prevention technique when the “understory fuel” is consumed by animals (Damianidis *et al.*, 2021). In terms of adaptation, there is fragmented information where AF practices have demonstrated to be able to reduce temperature variation by (a) the shade the trees cause, (b) the reduction of wind speed and its desiccation effect that tree barriers cause to surrounding croplands, and (c) the reduction of the effects of catastrophic events such as flooding (due to the reduction of the impact of heavy rainfall on soil and the better soil structure that the root of the trees provide which enhances water infiltration and reduces runoff) on crop production. Moreover, several researchers have estimated the capacity of AF to mitigate climate change, by counteracting the emissions at the farm scale, due to carbon (C) sequestration in the subsoil (Howlett *et al.*, 2011b). In addition, trees are able to maintain temperature during the hot waves, which reduces the exchange of greenhouse gases (GHGs) with the atmosphere (Rigueiro-Rodríguez *et al.*, 2009).

Agroforestry agroecosystem and agrifood innovations

The best way to promote agroforestry is by promoting the commercial and sustainable use of the enhanced biomass produced in AF systems. AF4EU described for the first time the AF value chains in 10 countries of Europe based on Regional Agroforestry Innovation Network (RAIN). The RAINs highlighted the linkages that AF systems have to extensive rural areas production by using local resources for surrounding urban areas reinforced by the interactions among the value chains. The main EU value chains were linked to agrotourism, beef, nuts, honey, spasture, goat, timber, berries, fungi, wine, aromatic plants and olives (Figure 5). Agrotourism as a service that connects rural and urban areas were described by 90% of the countries (Spain (two regions: Galicia and Andalusia), Greece, Germany, Belgium, Italy, Slovakia, Finland, Portugal and France) following by silvopasture beef production. It can also be highlighted the large share of value chains across Europe with the exception of those linked to crops (i.e., Olives or Aromatic) that are specific of some regions of Europe (Mediterranean).

Figure 6 shows the value chain of beef silvopasture that highlights that most of the products are produced based on the own farm resources with some inputs such as mineral fertilizers, water, seed and

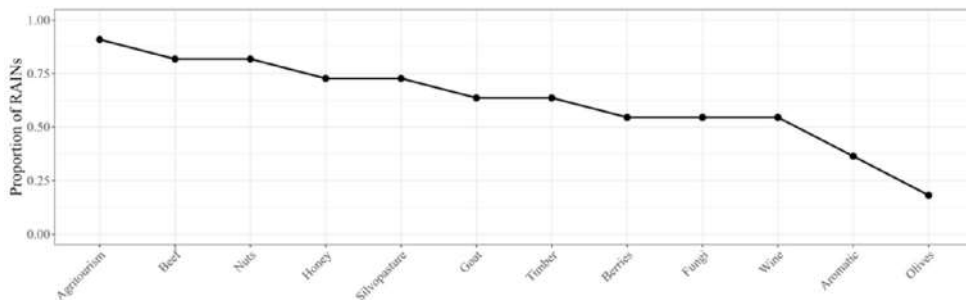


Figure 5. AF value chain description across Europe by the Regional Agroforestry Innovation Networks (RAINs).

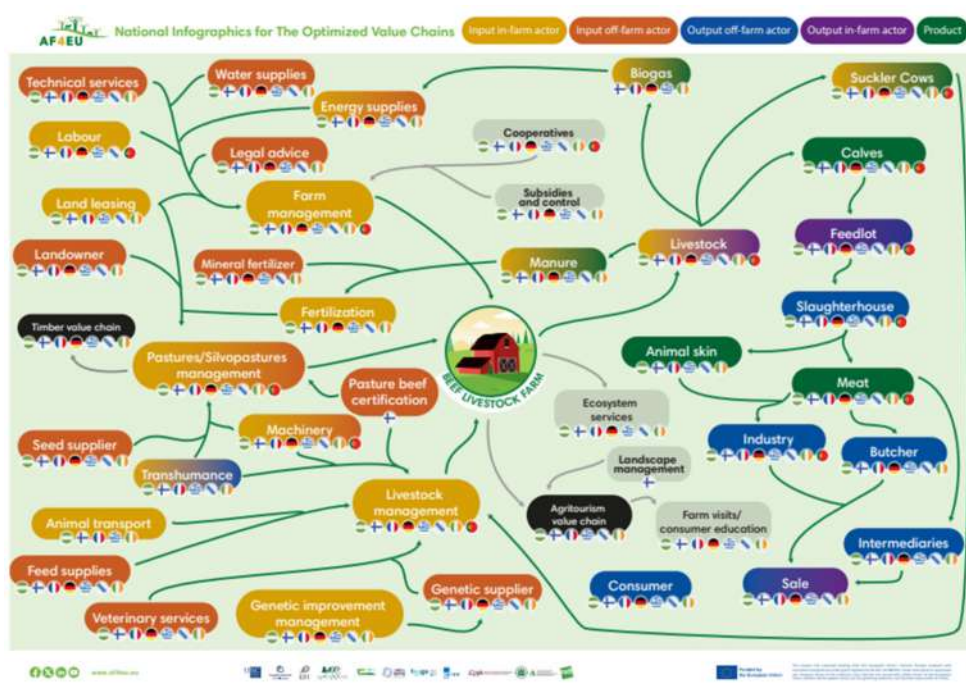


Figure 6. The value chain of beef silvopasture.

feed supplies coming from external resources, being most of the services from outside of the farm (manure, legal advice) technical services, legal advice, technical services, landowner, certification or veterinary services). The beef farm value chain linked to silvopasture is also related to circularity and bioeconomy concepts based on the reuse of manure, livestock, biogas or suckler cows. The value chain has as main products calves, skin or meat that can be sold directly or through intermediaries to the consumers. The value chain is connected with the timber and the agritourism and recognize the role of cooperatives, subsidies, the provision of ecosystem services, landscape management and consumer education.

Agroforestry has a huge potential to increase EU sustainability. Fostering AF should be linked to the understanding of both the business environment and the business models to be adopted by farmers considering the value chains development and connectivity at long and short term as developed in

the AF4EU DST fostered by the AF4EU advisory knowledge tools including the AF4EU-Knowledge Cloud, AF4EU MOOC tools.

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Agro-silvo-pastoral land uses for wildfire risk mitigation

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Abstract

Wildfires are increasingly reshaping landscapes in Europe, driven by climate change, land abandonment, and fire-suppression-based policies. In southern Europe, spontaneous forest expansion on former agricultural lands and grasslands has increased fuel continuity, reducing landscape resilience and raising wildfire hazards. Increasing research evidence shows that traditional agro-silvo-pastoral practices, often encompassed within High Nature Value (HNV) farming systems, can contribute to wildfire risk mitigation by maintaining heterogeneous, multifunctional landscape mosaics. Incentive-based mechanisms such as Payments for Environmental Services demonstrate how public funding can effectively link biomass management with supporting rural livelihoods. However, the scalability of these approaches remains limited by fragmented governance, discontinuous funding and insufficient monitoring frameworks. Building fire-resilient landscapes requires integrated strategies that align agricultural, forestry, civil protection, and climate policies under long-term, participatory governance. Strengthening the economic viability and social recognition of extensive farming and agroforestry systems is essential to prevent further land abandonment and achieve sustainable wildfire mitigation in Mediterranean socio-ecological systems.

Keywords: fire-resilient landscapes, High Nature Value (HNV) farming, landscape governance, payments for environmental services (PES)

Introduction

Wildfires have emerged as a growing concern across Europe, with recent years witnessing unprecedented fire events even in regions historically less affected (Elmqvist *et al.*, 2025). Extreme droughts and heatwaves, exacerbated by climate change and land-use transformations, have led to large, high-intensity fires that threaten both ecosystems and livelihoods (UNEP, 2022; Wunder *et al.*, 2021). In southern Europe, rural exodus and land abandonment since the mid-20th century triggered spontaneous forest expansion in marginal agricultural and pasture lands. While enhancing carbon sequestration and certain biodiversity values (Varela *et al.*, 2020), this process also increased biomass accumulation and fuel continuity, elevating wildfire risks (Moreira *et al.*, 2020).

Traditional fire-suppression strategies have reduced small fires but contributed to build-up of fuel and to extreme fire events (Keeley *et al.*, 1999; Xanthopoulos *et al.*, 2020). This has prompted a paradigm shift toward proactive wildfire risk mitigation, emphasizing fuel management and landscape-level interventions (Canadas *et al.*, 2023; UNEP, 2022).

This paper examines how agroforestry, grazing, and other extensive land uses can foster fire-resilient Mediterranean landscapes.

Wildfires as a socio-ecological phenomenon

In recent years, unprecedented fire events have occurred in Europe. While regions historically with fewer fires, experienced severe fires driven by extreme droughts and heatwaves, fire-prone regions in

southern Europe are projected to experience even more extreme conditions in terms of longer summer seasons with more days of exposure to heatwaves and high temperatures (Elmqvist *et al.*, 2025).

Wildfires are processes driven by complex interactions in social-ecological systems. The increase in large wildfires and their severe impact across Europe over recent decades is intrinsically linked to land-use changes, in addition to the effects of climate change (Oliveira *et al.*, 2017; Cervera *et al.*, 2019), resulting in wildfires more often encountering the fuel and weather conditions conducive to becoming destructive (i.e. very dry vegetation, high temperatures and warm winds) (UNEP, 2022) and becoming a major environmental and socioeconomic threat across many regions worldwide (Wunder *et al.*, 2021).

Land abandonment and rural exodus that took place in southern European countries since the middle of the 20th century, resulted in overall reduction of forest extraction leading a spontaneous forest expansion in former marginal agricultural and pasture lands (Cervera *et al.*, 2019). The resulting forest expansion and densification of existing forests is occurring rather as an unplanned encroachment, i.e. a side effect of industrialisation, than as a viable solution to its environmental impacts in terms of reducing CO² emissions or compensating for biodiversity losses (Infante-Amate *et al.*, 2022).

Ecosystem services arising from this forest expansion include increased carbon sequestration, water infiltration, provision of forest products, soil retention, and forest coalescence (Varela *et al.*, 2020), representing an opportunity to reduce fragmentation and enhance the conservation of forest specialist species such as birds or large carnivorous species. This process is however subject to controversial opinions among natural scientists, since forest-centred conservation policy can hamper the maintenance of the multifunctional mosaic human-shaped landscapes that largely depend on traditional agro-silvo-pastoral practices to support biodiversity and secure the provision of ecosystem services, being wildfire regulation a paramount one. Furthermore, the evolution of these new forests towards more diverse successional stages can be hindered by increasing forest disturbances, being wildfire one of the paramount risks faced by these forests. The traditional rural mosaic that would make a sufficient fuel fragmentation is becoming scarce in European Mediterranean countries (Moreira *et al.*, 2020), leading to high biomass content and continuity in early successional stages and hence making these landscapes more vulnerable to severe wildfires. Landscapes changes towards homogenization create favourable conditions for the spread of wildfires by connecting fuel loads available to burn (; Moreira *et al.*, 2011; Sil *et al.*, 2019; Kelly *et al.*, 2020; Spadoni *et al.*, 2023) resulting in a reduced capacity of the landscape to regulate fire and provide fire protection.

Fire suppression and the shift toward proactive mitigation

Over the last four decades, emergency response and fire exclusion have dominated wildfire management in EU Mediterranean countries (Aquilué *et al.*, 2019; Elmqvist *et al.*, 2025). Efficient suppression has protected both non-fire-adapted ecosystems and expanding wildland–urban interfaces but has also reduced small fires while promoting fuel accumulation and extreme wildfire events (Keeley *et al.*, 1999; Xanthopoulos *et al.*, 2020). These large fires, intensified by land-use and climate change, now account for most burned areas (Canadas *et al.*, 2023). Even in technologically advanced countries, suppression fails under extreme weather, as fire behaviour exceeds control capacity (Tedim *et al.*, 2016). Without complementary fuel management, these policies cannot prevent large-scale disasters (Moreira *et al.*, 2011), creating the so-called “firefighting trap” where immediate success leads to higher long-term risks (Xanthopoulos *et al.*, 2020).

A broad consensus now supports rebalancing investments from reactive suppression to proactive wildfire mitigation (UNEP, 2022; Canadas *et al.*, 2023). Direct measures, such as fuel breaks, prescribed burns, and thinning, reduce fuel continuity, protect wildland-urban interfaces, and improve suppression safety (Castellnou *et al.*, 2019; Spadoni *et al.*, 2023). Yet, they remain costly, temporary, and dependent on public funding (ECA, 2025; Ascoli *et al.*, 2023), rarely achieving the scale needed to alter landscape flammability (Sil *et al.*, 2019). This underscores the need to integrate fuel management within broader land-use planning and adaptive, cross-sectoral governance (Moreira *et al.*, 2020; Wunder *et al.*, 2021; Stoof *et al.*, 2024).

Landscape mosaics and extensive land uses

Viewing wildfires as a landscape problem highlights the importance of non-forest uses and traditional Mediterranean mosaics, where agricultural plots, pasture lands, silvopastoral uses and forests are intermingled, in reducing fire risk (Lecina-Díaz *et al.*, 2023; Pulido *et al.*, 2023). Indirect mitigation measures, such as sustainable forest management, agroforestry, and extensive grazing, modify vegetation structure and fuel patterns, improving landscape fire regulation (Lasanta *et al.*, 2022). Evidence from Spain and Italy shows that areas with active agro-silvo-pastoral management experience fewer and less severe fires, enhancing suppression efficiency (Aquilué *et al.*, 2019; Spadoni *et al.*, 2023; Pérez-Izquierdo *et al.*, 2025).

Research further indicates that heterogeneous landscapes and High Nature Value (HNV) farming systems are particularly effective in mitigating fire impacts by reducing fuel connectivity and promoting recovery (Lecina-Díaz *et al.*, 2023; Pulido *et al.*, 2023) and can be considered as indirect measures for wildfire risk mitigation. In contrast, direct measures based on forestry interventions alone often remove biomass where burn probability is low, with limited benefits for fire reduction (Pérez-Izquierdo *et al.*, 2025). Integrating direct and indirect measures through “fire-smart territories” (Tedim *et al.*, 2016; Pulido *et al.*, 2023) offers a cost-effective pathway to reduce hazard while revitalising rural economies. Combining prescribed burns with grazing (San Emeterio *et al.*, 2023) or promoting non-timber forest products can enhance both ecological resilience and the financial viability of these interventions (Varela *et al.*, 2020).

The social dimension of indirect wildfire risk mitigation

While most large-scale fuel management is carried out by public agencies without engaging territorial stakeholders, local managers implementing indirect measures such as cultivation of rainfed tree crops, resin-tapping or extensive livestock grazing, can better integrate communities and generate economic returns that encourage wildfire risk reduction at the landscape scale (Spadoni *et al.*, 2023; Pérez-Izquierdo *et al.*, 2025). Viewing fire-resilient landscapes through this integrated lens helps bridge science and practice, diversifies stakeholder participation, and strengthens the design and maintenance of “fire-smart territories.” Combining complementary perspectives also supports tailored policies for specific landscape patterns, minimising trade-offs and enhancing synergies for ecosystem service provision.

Fire management beyond suppression must address the coupled social and environmental dimensions of wildfire (Fischer *et al.*, 2016) to foster fire-adapted ecosystems and resilient societies capable of coping with increasingly severe events (Moritz *et al.*, 2014; Tedim *et al.*, 2016; Aquilué *et al.*, 2019). Social dimensions include land-use practices, governance arrangements, economic incentives, and stakeholder interactions, while subjective dimensions encompass risk perceptions, values, norms, trust, and attitudes toward fire, prevention, and management interventions (Higuera *et al.*, 2019; Wunder

et al., 2021). Integrating these dimensions into wildfire science is essential especially since the detachment of local populations has accompanied top-down suppression approaches that leave communities unprepared for extreme fires (Steelman, 2016; Bacciu *et al.*, 2022). Strengthening wildfire governance systems, this is the social architecture that supports collective action and innovation, can enhance local responsiveness to changing fire risk (Essen *et al.*, 2023). A shift toward adaptive governance focused on reducing damage rather than area burned (Moreira *et al.*, 2020) requires transforming underlying institutional structures and assumptions (Darnhofer *et al.*, 2017).

High Nature Value farming systems and wildfire risk regulation

Firewise measures advocate for the recovery and maintenance of well-managed grasslands, annual crops, and agroforestry mosaics that limit the continuity of flammable vegetation. These landscapes are closely linked to the concept of High Nature Value (HNV) farmland, which includes permanent grasslands, arable and permanent crops, and agroforestry systems traditionally associated with high species and habitat diversity, or the presence of species of European conservation concern. Beyond their biodiversity value, these systems provide important wildfire regulation services by maintaining low-flammability vegetation structures at the landscape scale.

HNV farming is characterized by low input intensity, extensive livestock use, and the presence of large areas of semi-natural vegetation forming diverse mosaics. These features enhance biodiversity and the supply of multiple ecosystem services (Plieninger and Bieling, 2013). Regulating services are particularly tied to maintaining permanent grasslands and low-intensity livestock management (Tóth *et al.*, 2016; Pulido *et al.*, 2017), which help sustain low flammability at the landscape scale.

However, the current extent and condition of HNV farming in fire-prone regions is insufficient to fully deliver these regulating functions. HNV systems are predominantly located in marginal and mountainous areas, making them prone to abandonment and hence vulnerable to recurrent wildfires (Den Herder *et al.*, 2017). Ongoing abandonment, declining livestock numbers, and farm succession constraints are leading to the contraction and fragmentation of these landscapes, reducing their capacity to regulate wildfire risk.

While HNV farming systems are well aligned with wildfire-resilient landscape objectives, their fire-mitigation potential is increasingly threatened and cannot be taken for granted. Strengthening and maintaining HNV farming in fire-prone regions requires targeted policy support and incentive schemes that explicitly recognise wildfire regulation as a co-benefit of extensive farming. To date, research and policy attention have focused primarily on biodiversity and provisioning services, while regulating and cultural services remain under-represented (Varela *et al.*, 2025). Greater recognition of these functions would enhance the societal relevance of HNV farming and support its long-term conservation in wildfire-susceptible landscapes.

Incentivising active landscape management

Since firefighting alone will not be enough to counteract the abandonment path in rural economies, broader models of rural development strategies that include climate, biodiversity cultural and economic targets and civil protection would be required (Wunder *et al.*, 2021). Conversely, the amounting evidence of the effects of landscape transformations at large scale in enhancing fire resilience can be powerful triggers towards improved rural economies and better managed forestlands with the bio-economy as umbrella concept (Verkerk *et al.*, 2018; Varela *et al.*, 2020).

From an economic point of view, wildfires can be considered a public bad or disservice from landscapes that accumulate biomass since fire impacts can affect not only landowners' own property, but also society at large, creating a spatial externality problem (Wunder *et al.*, 2023). In this situation landowners may lack not only knowledge and capacity, but also sufficient motivation to engage in socially optimal fire-mitigating activities (Fraccaroli *et al.*, 2025). Therefore, beyond improving organization and awareness among landowners encouraging proactive land use practices in fire-prone rural areas, which simultaneously reduce wildfire risk and enhance ecosystem services, may require public intervention through economic incentives that can avoid societally sub-optimal underinvestment (Wunder *et al.*, 2023; Fraccaroli *et al.*, 2025).

Payments for environmental services (PES)

Payments for environmental services (PES) are voluntary transactions between service users (e.g. municipalities concerned over escalating fire risk) and service providers (e.g. landowners with little resources to engage in risk-reducing actions) conditional on agreed-upon rules of natural resource management for generating offsite services (Wunder, 2015). The offsite services term that is included in this definition emphasizes links to the concept of externality. PES generally are characterized by voluntariness, where ecosystem service (ES) providers can choose to either respond or not to the monetary incentives offered by the potential user. Participation involves a contract between the ES beneficiary (or its representative) and the service provider. This way, resources flow from beneficiaries to provider, possibly via an intermediary, and are directly tied to the environmental goals.

Conditionality and additionality are also important features in differentiating PES from other types of incentives. Conditionality refers to the requirement that payments are only made if the service provider (e.g., landowner or community) delivers specific environmental outcomes and this is linked to having monitoring systems in place. Additionality in PES takes place when outcomes are measurable and it is verified that these have gone beyond what would have happened in the absence of the scheme, thereby producing a change in the behavior of the service provider compared to a non-PES scenario.

A PES scheme has been used to pay shepherds for reducing biomass accumulation through targeted livestock grazing with notable examples in Spain (Varela *et al.*, 2018; Wunder *et al.*, 2023). Through this PES, public administrations reward shepherds for the biomass control services that they provide through targeted grazing. Payments are generally modulated according to the level of accomplishment in biomass reduction and this is verified through monitoring by technicians of public administration. Targeted grazing involves managing livestock to achieve specific vegetation outcomes. It offers a cost-effective means to reduce fine fuels, particularly in rangelands or areas where mechanical treatments are infeasible (Bruegger *et al.*, 2016). Compared to mechanical treatments, targeted grazing often receives greater public support (Varela *et al.*, 2014) and can be more affordable compared with other methods, for example mechanical means (Bruegger *et al.*, 2016).

The Network of Grazed Firebreaks of Andalusia (RAPCA), launched in 2007 in southern Spain, is the most established program of this kind. It contracts over 200 shepherds to graze more than 6,000 hectares of fuelbreaks on around 43 000 ha public forestlands, offering result-based payments linked to biomass consumption levels (Varela *et al.*, 2018). Monitoring is conducted by technical staff, and similar programs have since been replicated in other Spanish regions (Wunder *et al.*, 2023). Local shepherds are remunerated by the regional administration on behalf of the Andalusian society according

to the biomass reduction accomplished in fuelbreaks located in public forests. Compliance is estimated based on levels of vegetation consumed, and experience in herd management by the shepherds is required as an eligibility criterion.

Following the example of the RAPCA, the region of Catalonia developed a similar program since 2016 funded through the II Pillar of the Common Agricultural Policy. Currently it covers 5800 hectares of fuel breaks in strategic areas (these are areas of recurrent wildfires and from where rapid suppression interventions can be very effective in stopping wildfire spread) and involves 97 shepherds. In the Canary Islands, the Gran Canarias Pastorea program started in 2022 where contracts were signed with 35 shepherds for grazing about 2000 ha. The scheme features some design innovations, such as sophisticated spatial analyses identifying wildfire high-risk areas in and activity monitoring through herd-installed GPS, enabling a clearer mode of establishing payment conditionality (Wunder *et al.*, 2023).

These programs can be seen as a middle ground between direct measures, since most of them pay for fuel break maintenance, and indirect support for extensive grazing in surrounding reinforcement areas. In these areas, the browsing and trampling of animals can have additional positive ecological effects. Also, these schemes may create opportunities for new actors to participate in wildfire risk mitigation.

The success of active landscape management in wildfire-prone regions will ultimately depend on the landholders who manage them. Understanding their perceptions as the reasons underpinning their willingness to undertake certain practices and/or uptake innovations will be crucial for the maintenance of mosaic landscapes where extensive agriculture intermingles with agro-forestry and extensive grazing use. Involving local landowners in the design of PES schemes can therefore improve the efficiency and uptake of these programs (Varela *et al.*, 2022; Varela *et al.*, 2025).

Market-based incentives for fire-resilient landscapes

Differently from the previous examples on PES schemes, value chain labels work through commodity markets, by certifying agricultural or forest products that contribute to wildfire risk mitigation and mosaic landscape maintenance. The underlying assumption is that provision of wildfire prevention services may constitute a sustainability attribute appreciated by consumers and hence these products would fetch their willingness to pay a price premium for these products. The generated extra income would in turn reward certified producers, and thus also allow them to continuously finance their future risk mitigation efforts. Effectively communicating the benefits of products from these systems involves significant challenges given that most sustainability attributes are credence attributes (i.e. consumers cannot verify these characteristics either prior to consumption or even after consumption).

Initiatives include the LIFE Granatha project in Italy that reduced biomass and shrub cover while creating marketable organic brooms (Ascoli *et al.*, 2023). In Catalonia (north-eastern Spain) the Fire Flock label identifies meat and milk from herds grazing in high wildfire risk areas (Nuss-Girona *et al.*, 2022). These initiatives demonstrate how indirect wildfire risk reduction can align with rural economic development and, if appreciated by consumers, can stimulate new business opportunities through labelling (Lecegui *et al.*, 2023). However, these labels are frequently linked to research projects, at a local scale and in an incipient status (Nuss-Girona *et al.*, 2022; Pulido *et al.*, 2023).

Overall, most value-chain labels have so far remained at a proof-of-concept stage, with limited impact on local markets (Wunder *et al.*, 2023). Their demand has not generated enough traction to confirm

their future potential as standalone strategies towards wildfire resilience. Furthermore, very little is known about the perception of consumers with respect to these wildfire-labelled products. To fill the gap, a study was conducted investigating consumer preferences and willingness-to-pay for beef and lamb meat from silvopastoral systems associated to the provision of wildfire prevention service and explored two alternative ways of labelling this service (Lecegui *et al.*, 2023). The results provide key insights for further improvement of these initiatives, highlighting that the proximity of production determines most of the consumer preferences and willingness to pay for lamb meat and beef. Consumers also placed higher value on targeted grazing than on forest grazing. Therefore, complementing information on distance with the type of pasture could increase the meat quality perceived by consumers, where both labelling of forest grazing and targeted grazing can influence the choice for beef and lamb meat positively.

Sustainable funding and scaling of fire-resilient landscape initiatives

Achieving wildfire-resilient landscapes requires long-term, coordinated investment strategies that go beyond fragmented and short-term project funding. However, reliance on EU funds has often been perceived as complex and difficult to navigate, particularly due to the segmentation of competencies in wildfire management (Ascoli *et al.*, 2023). This fragmentation can hinder the development of shared governance structures and reduce the effectiveness of public policies aimed at wildfire risk mitigation.

To overcome the current lack of investment capacity in fire-prone territories, it is essential to ensure the efficient and coherent use of multiple funding sources, avoiding redundancies, gaps in funding, and conflicting objectives. Strategic alignment between international, European, national, and regional funding is necessary, with investments directed toward pre-planned actions that reflect local and regional fire risk mitigation needs (Ascoli *et al.*, 2023; Spadoni *et al.*, 2023). European funding programs such as the Rural Development Programme and LIFE+ can play a transformative role in mitigating wildfire impacts under climate change scenarios, while also fostering continuity and stability that can attract private investment (Moreira and Pe'er, 2018; Verkerk *et al.*, 2018; Ascoli *et al.*, 2023; Spadoni *et al.*, 2023). A territorial approach that integrates the EU Bioeconomy Strategy, European Forest Strategy, and CAP could enhance the effectiveness of these measures (Varela *et al.*, 2020).

Despite the ecological importance of HNV farming systems and their potential role for wildfire risk mitigation, particularly in extensively managed semi-natural areas, current CAP funding schemes have often been poorly adapted to the needs of low-input and small-scale farming (Martino and Muenzel, 2018; Pe'er *et al.*, 2020; Sumrada *et al.*, 2021). A shift toward supporting multifunctional farming systems, such as silvopastoral systems, and implementing PES schemes for biomass reduction and wildfire prevention could yield broader societal benefits (Muñoz-Ulecia *et al.*, 2021; Varela *et al.*, 2020).

While EU funding supports research and pilot initiatives, many projects have not been replicated or integrated into broader funding mechanisms, limiting their impact (ECA, 2025). Thereby, the scaling-up of successful demonstration projects remains a challenge. Authorities in several member states have failed to incorporate lessons from successful LIFE projects into the design of Structural and Investment Fund measures, missing opportunities to enhance the effectiveness of EU funding (ECA, 2025).

Moreover, the lack of robust monitoring and evaluation frameworks impedes the assessment of program performance. Existing indicators are often output-focused and do not capture results or impacts, reducing the Commission's ability to evaluate the effectiveness of wildfire-related investments across

the EU (ECA, 2025). Strengthening these frameworks with performance indicators that track inputs, outputs, results, and impacts is essential for improving transparency and guiding future funding decisions.

Finally, the long-term viability of fire-resilient landscapes, such as HNV farming systems, depends not only on financial support but also on social and institutional measures, including advisory services and knowledge transfer. These are critical to ensure that marginalised farmers are not excluded from funding opportunities and can actively participate in landscape-level wildfire prevention strategies (Martino and Muenzel, 2018; Sumrada *et al.*, 2021).

Concluding remarks

Addressing increasing wildfire risk requires a paradigm shift from reactive suppression to proactive, landscape-level mitigation (UNEP, 2022; Elmqvist *et al.*, 2025). Integrating forestry, agriculture, agroforestry and extensive livestock systems under coherent management principles can contribute to a transition from wildfire-prone to wildfire-resilient landscapes.

High Nature Value (HNV) farming systems and other low-intensity agro-silvo-pastoral practices provide both rural livelihoods and wildfire-regulating services by maintaining landscape heterogeneity (Varela *et al.*, 2020; Lecina-Diaz *et al.*, 2023; Pulido *et al.*, 2023). Their recovery, maintenance and adoption depend on viable economic and institutional mechanisms. Payments for Environmental Services schemes, such as Andalusia's RAPCA and Catalonia's result-based grazing programs, effectively link public investment and local stewardship (Wunder, 2015; Varela *et al.*, 2022, 2018). Yet, as Fracaroli *et al.* (2025) show, rigorous impact evaluation and spatial targeting in high-risk areas remain limited, and scaling-out locally adapted initiatives appears more feasible than centralised approaches.

Landscape interventions must also balance trade-offs with carbon storage and other ecosystem services (Pérez-Izquierdo *et al.*, 2025). Building resilient territories therefore calls for integrated governance aligning forestry, agriculture, civil protection and climate policies (Elmqvist *et al.*, 2025).

Finally, strengthening public-private partnerships among public administrations, private landholders, and local communities will be key to fostering adaptive, socially inclusive wildfire governance and achieving fire-resilient socio-ecosystems (Essen *et al.*, 2023). This collective effort must be underpinned by long-term investment strategies that integrate EU and national funding instruments with local initiatives, ensuring both continuity and social engagement.

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Changes in grass biomass and quality in silvopastoral grasslands under the influence of poplar rows

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Abstract

In many temperate silvopastoral systems, grassland is managed extensively, with timber production often the main focus. This raises the question of how the proximity of trees affects the biomass and nutritional value of grassland when it is harvested once a year. In a one-year field experiment conducted at two sites in the temperate climate of northern Germany, we investigated how the distance to tree lines affects herbage biomass accumulation and the nutritive value in relation to the harvest date. Herbage biomass accumulation was determined in the primary growth at four successive harvest dates and at three distances from the tree line within the agroforestry system and in an open-field control ($n = 4$ replicates). Herbage biomass accumulation was significantly affected by the interaction harvest date $\times$ distance position from the tree line ($P < 0.01$). At closer distance to the tree line the increase in herbage accumulation in spring was lower and the decrease during summer slower compared with further distance but showed a pronounced decrease in autumn. The crude protein content showed an interaction between site $\times$ harvest date $\times$ distance position ($P < 0.001$), driven by the timing of harvest. The CP was lowest in the early summer.

Keywords: agroforestry, grass biomass, grass quality, poplar rows, silvopastoral system

Introduction

Modern agriculture is under increasing pressure to become more sustainable and biodiversity-friendly. Grassland management is crucial for preserving biodiversity and storing carbon. Integrating trees into grassland in the form of silvopastoral systems serves the dual purpose of timber production and increasing the economic value of less productive grassland which is otherwise at risk of abandonment. Since grassland is less important in such cases, producers aim to achieve the highest possible agronomic return with the least possible input. Consequently, one cut systems are often practised, prompting the question of the extent to which tree lines influence the accumulation of grassland biomass and the nutritional value when the harvest date shifts from spring to late summer.

The aim of this study is to answer the question of how the timing of the first harvest date in poplar agroforestry systems affects the herbage biomass accumulation and nutritive value of grasslands and what effects trees have in this respect.

Materials and methods

The study was conducted in 2024 at two sites in Germany: Site A (Lüneburg, 53°12'22.8"N, 10°21'58.5"E) and site B (Niederer Fläming, 51°56'14.8"N, 13°12'27.1"E) as part of a silvopastoral experiment where poplar trees are planted in order to produce construction wood with an anticipated tree age of 10–15 years. The soil texture at both sites was sandy loam. Site A had a long-term average annual precipitation of 695 mm, while site B was considerably drier, with a long-term average annual

precipitation of 539 mm (DWD Climate Data Center (CDC), 2025). The poplars on grassland site A (Lüneburg) were planted in 2016, and those on site B (Niederer Fläming) in 2021. The experimental design followed a split-plot design in which the factor distance to the tree line was varied. Three fixed distances were placed 1 m, 4 m and in the centre between two tree lines within the agroforestry systems. One open field reference plot was established in areas of the same field without effect of trees. Each distance position had four replications. The second experimental factor consisted of the timing of the first harvest during the primary growth, which had four stages: May, June/July, August and September. At each harvest date, the standing herbage biomass was determined by means of manually cutting the above-ground biomass at a stubble height of 3 cm using electric hand shears (ComfortCut, Gardena) in a 0.18 m² steel frame. In the laboratory, the fresh biomass was manually separated into living green and senescent brown herbage biomass and the dry matter (48 h at 60°C in a forced air-oven) determined. Thereafter, the samples were rebulked and ground to 1 mm using a centrifugal mill (ZM 300, Retsch). The herbage nutritive value was determined using near-infrared reflectance spectroscopy (NIRS; Phoenix 5000, BlueSun Scientific) by scanning each sample twice. In the present study the crude protein concentration (CP) and the acid-detergent fibre in organic matter (ADF) are considered. The files were processed and a calibration data set was used that was based on a large data set for grass and forage (VDLUFA Qualitätssicherung NIRS, Kassel, Germany). The statistical analysis was performed in RStudio (R Core Team, version 4.0.2) using linear mixed-effects models in the package 'nlme' (Pinheiro *et al.*, 2025) The fixed effects included site, distance position to the tree line, harvest date and their interactions. The replication was used as random effect. Comparisons of means were performed posthoc using Tukey's HSD method ($P < 0.05$) in the package 'emmeans' (Lenth, 2022).

Results and discussion

The herbage biomass accumulation was significantly affected by the interaction harvest date x distance position ($P < 0.01$). The herbage mass increase in spring was lower at a close distance to the tree line while the decrease in summer was slower compared with a further distance. The lowest values were measured in September in close proximity to the tree line, and the highest value in June/July in the open field, suggesting a time-depending growth-suppressing effect of trees (Figure 1A). This is in line with the observation that a stronger tree effect was found at the older site (tree age: 8 years, estimated height 17.20 m) compared to the younger site (tree age 3 years, estimated height 4.45 m).

In May the proportion of biomass of senescent tissue was low, increasing with vegetation dynamics and reached highest values in June/July. An exception was the first distance position (1 m), which showed higher values in May, but then lower values compared to other distances positions, obviously related to the lower biomass growth (Figure 1B). The CP concentration was affected by a site x distance x harvest date interaction ($P < 0.01$); however, the dominating effect was the time of harvest with a decline from May to June/July, and recovery in late summer due to regrowth. Comparing the sites, site A had generally higher CP concentrations than site B (Figure 1C–D). Similar results had been found by Opitz von Boberfeld (1996) with decreasing CP concentrations below 12 % after May. There was a significant interaction of site x harvest date ($P < 0.05$) on the ADF concentration. Related to the dynamics of the CP concentration, at site A the ADF values increased from 27.5% in May to 40.8% in August and 42.7% in September. At site B, the ADF concentration increased from 26.7% in May to 43.2% in August and 42.9% in September. The dynamics of the CP- and ADF-concentrations indicate the considerable decrease of the nutritive value, with harvest date being more influential than tree distance. The increasing proportion of senescent biomass during the growing season reduced the overall nutritive value of the herbage, further reinforced by the concurrently rising fibre content. The

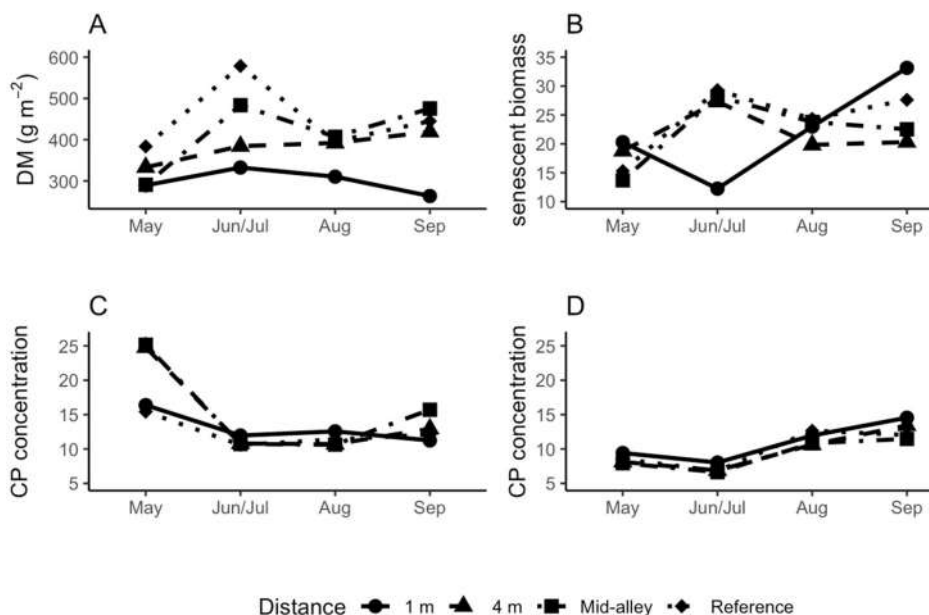


Figure 1. Herbage biomass accumulation (g DM m⁻²) (A). Results of ANOVA: S (site) × H (harvest date), $P < 0.01$; D (distance to the tree line) × H, $P < 0.01$), proportion of senescent herbage biomass in the dried sample (%) (B: S × H, $P < 0.01$; D × H, $P < 0.05$; S × D, $P < 0.001$) and CP concentration (%) (S × H × D, $P < 0.001$) for Site A (C) and Site B (D) of the four different distances to the tree line (1 m distance, 4 m distance, mid-alley (centre between the rows of trees) and reference) over the four harvest dates (May, June/July, August, September).

herbage nutritive value in the present study is to be considered too low for dairy cows, while sufficiently high CP concentration was achieved for beef cattle (DLG, 2025).

Conclusion

Trees in silvopastoral systems affect the dynamics of herbage mass accumulation and nutritive value of the herbage. There is indication that the effect of trees on the herbage accumulation and tissue senescence is stronger than on the nutritive value of the herbage. However, the response is affected by complex interactions with site and the age and height of the trees. This has to be studied in more detail with more years and sites in future research.

Acknowledgement

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New insights into the role of agroforestry for controlling nitrate leaching from farrowing-sow paddocks

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Abstract

Agroforestry is beneficial to outdoor animal husbandry but knowledge on its role in reducing nitrogen (N) surplus remains elusive. A field experiment was conducted on a commercial farm in Denmark with lactating sows and their piglets housed in 4 paddocks with grass-clover ley and poplars in the centre either cut (TM; biomass as wood chips used as surface mulch in the tree area) or uncut (TU), each with feed and hut on same (RS) or opposite side (RO) of the trees. Soil nitrate across each paddock was depicted by 51 suction cups at 1 m depth. Nitrate leaching estimated with support of a model varied seasonally and spatially from 7 to 860 kg N ha⁻¹ year⁻¹. Although RO changed soil N distribution, no clear leaching effect occurred. Higher leaching in the tree zone of TM indicated that cutting poplar reduces N uptake. On average, nitrate leaching in the study year was in the order TM-RO>TM-RS>TU-RO>TM-RS, i.e., 231^a > 214^{ab} > 170^b > 163^b kg N ha⁻¹ across zones. These results highlight the role of trees and resource position for distributing nitrate leaching in outdoor pig systems.

Keywords: outdoor pig, pasture, poplar, silvopastoral, spatial analysis

Introduction

Despite benefits for animal welfare, outdoor pig production on pasture is characterised by an uneven manure distribution and destruction of vegetation cover, leading to hotspots of nitrate leaching (Kongsted *et al.*, 2020; Salomon *et al.*, 2007). High nitrogen (N) surplus and leaching pollute the ground- and the surface waters. Integrating trees may enhance N uptake and influence the spatial distribution of soil N (Jakobsen *et al.*, 2019; Manevski *et al.*, 2019; Ullfors *et al.*, 2025). However, such systems remain elusively explored. This study explores the impact of poplar tree management and position of resources for the animals on the spatial and seasonal variation of NO₃⁻ leaching in pig farrowing paddocks.

Materials and methods

A field experiment was conducted from April 2022 to April 2024 on a commercial farm in Denmark with four 63 × 15 m paddocks, each with a central zone (B) with poplars (*Populus spp.*, planted in 2011 at 2–3 m spacing, 1900 trees ha⁻¹) flanked by grass-clover ley zones (*Lolium perenne*-*Trifolium repens*) sown in 2022 (A and C). Each paddock had combined two treatment factors: tree management (T) and resource position (R). Trees were either unmanaged (TU) or harvested at 2 m height and woodchips surface applied as mulch in April 2023 (TM). Resources (feeding trough and hut) were placed either on the same (RS) or on opposite sides (RO) with tree zone in between. Each paddock housed a lactating sow with piglets for 11 weeks per batch, with three succeeding batches introduced

Table 1. Model estimated annual nitrate leaching ($\text{kg N ha}^{-1} \text{ year}^{-1}$) for year 1 (before the experiment) and year 2 (during the experiment) and each zone (grass, A and C; tree, B) under different resource positions and tree management as least-square means (LS-mean) with standard error (SE) and the significance level (*p*-value) of post hoc comparison of treatment levels.

Year	Zone	Resource position					Tree management				
		RO		RS		RO/RS	TM		TU		TM/TU
		LS-mean	SE	LS-mean	SE	<i>p</i> -value	LS-mean	SE	LS-mean	SE	<i>p</i> -value
1	A	216	57	151	40	0.001	191	51	170	45	0.301
	B	133	38	143	42	0.593	143	41	132	39	0.576
	C	88	23	76	21	0.243	67	18	100	27	0.001
2	A	211	56	256	68	0.087	240	64	225	60	0.563
	B	163	48	144	43	0.417	239	70	98	29	< 0.001
	C	227	60	177	48	0.054	192	51	209	57	0.514

RO, on opposite sides of trees; RS, on the same side; TM, tree harvest with wood chip mulched; TU, unmanaged trees.

in April, July and November 2023. Soil water was sampled biweekly from 51 suction cups per paddock installed in April 2022 at 1 m soil depth. Nitrate concentrations in soil water were determined colorimetrically and values interpolated to daily with the percolation-weighted concentration method of Shepard *et al.* (1996), with percolation estimated by the Daisy model (Manevski *et al.*, 2019). Leaf Area Index was estimated from DJI Mini 3 Pro UAV with RGB camera at ca. 40 m altitude in season to feed the model for parameterising poplar trees canopy dynamics. For the grass ley, weekly visual assessments of the cover were used. Annual leaching for year 1 (April 2022 to March 2023) and year 2 (April 2023 to March 2024) were statistically analysed by a generalised mixed model (GLM) and seasonal leaching by generalised additive model (GAM).

Results and discussion

Annual nitrate leaching varied considerably between years and locations within the four paddocks (Table 1). Year 1 prior the experiment had lower leaching than the study year 2, but still considerable, suggesting residual soil mineral N from past stocking. Spatial analysis is a relatively unique future of the study, and for grass zones revealed the highest leaching in areas around feed and hut, e.g. specific subzones in the zone with feed and, in RS, hut. Opposite to expectation, RO did not reduce paddock leaching but tended to even out leaching across zones (Table 1). Clear effects were lacking.

The tree zone exhibited 59% lower leaching for TU compared to TM with 239 kg N ha^{-1} in year 2 (Table 1). In this year with sows present, leaching of TU tended to be lower in the tree zone B compared to the grass zones (ratio B/A: 0.4 (SE = 0.9); B/C: 0.5 (SE = 0.2)). Also, TM increased the overall paddock annual leaching compared to TU, e.g. in year 2 from 167 (SE = 27) to 223 (SE = 35) kg N ha^{-1} ($p < 0.001$). The investigation of seasonal variation revealed that leaching differences between TM and TU treatments emerged prominently during the main leaching season in autumn and winter (Figure 1). Thus, the hypothesis that tree management with wood chip mulching would reduce leaching by N immobilisation could not be confirmed. This may reflect reduced tree uptake after biomass removal and limited microbial access to the organic carbon (C) of wood chips due to their large size (predominantly 1–5 cm) and lack of incorporation. Surface N input from feed and other sources in the experiment year totalled 338 $\text{kg N ha}^{-1} \text{ year}^{-1}$ averaged across the two years, with an output of 234 kg N ha^{-1} primarily via piglets. Thus, estimated paddock leaching was 143–170% of the N surplus, emphasising the magnitude of losses under current management.

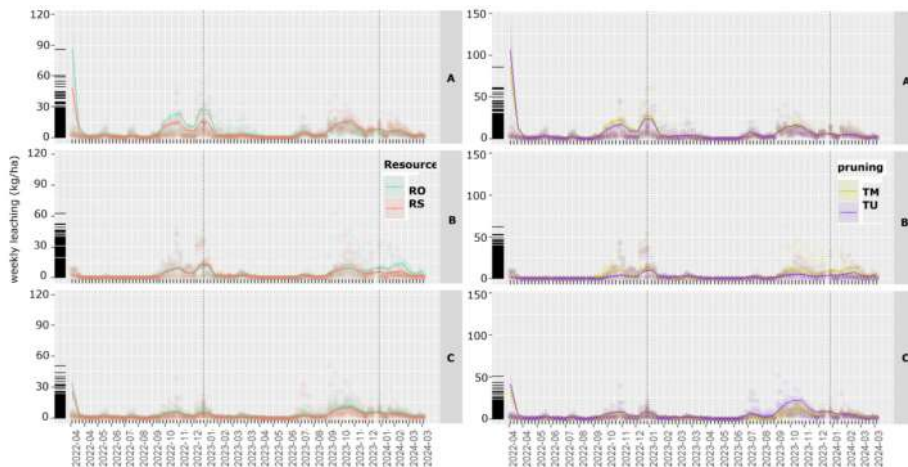


Figure 1. Weekly nitrate leaching comparing resource position (opposite (RO) or same (RS) sides) and trees either cut (TM) or uncut (TU) derived from a generalized additive model.

Conclusion

This study showed that tree management influences spatial and temporal patterns of nitrate leaching in outdoor pig systems. Unmanaged poplar trees showed lower leaching compared to trees harvested and mulched with wood chips, especially during the leaching season in autumn and winter. Tree management treatment appeared to reduce N uptake with no effect of C potentially mineralised from wood chips. Placing resources on opposite sides should redistribute soil mineral N but not necessarily reducing nitrate leaching. Hence, agroforestry designs should carefully consider the uptake potential of tree vegetation and the use of wood chip mulch in addition to pig behaviour when aiming to mitigate nutrient losses.

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Productivity and profitability of integrated crop-livestock systems with mature trees

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Abstract

Analysing the economic aspects of integrated systems is essential for their widespread adoption, as these systems support sustainable farming intensification. This study evaluated two integrated systems, crop-livestock (CL) and crop-livestock with mature trees (CLT), under two nitrogen (N) levels (90 and 180 kg N ha⁻¹). Livestock performance, crop productivity, and economic returns were assessed over two years following drastic tree thinning in the CLT system. Cool-season pastures grazed by beef cattle preceded corn cultivation (2017–2018) and soybean (2018–2019). In the first year, revenue increased by 41%, costs rose by 30%, and profit grew by 59% compared to the second year. Significant differences in revenue were observed between systems (CL = US\$ 1814 ± 136.3 ha⁻¹, CLT = US\$ 1445 ± 140.8 ha⁻¹), regardless of N level. Reducing tree density did not equalize systems performance. Total costs ranged from US\$ 940 ± 84.0 ha⁻¹ (CLT N90) to US\$ 1241 ± 90.0 ha⁻¹ (CL N180). No significant differences in profit were found, averaging US\$ 552 ± 63.2 ha⁻¹. However, profitability in CLT treatments was only achieved through tree components. While both systems are profitable, they require further adaptation to cope with increased tree competition, especially in shallow soils, and climatic challenges.

Keywords: agroforestry, beef cattle, *Eucalyptus dunnii*, nitrogen levels, thinning

Introduction

Integrated crop-livestock systems (ICLS) have emerged as promising strategies to enhance agricultural sustainability and resilience by improving resource use efficiency and soil quality through diversified land use (Lemaire *et al.*, 2023). Despite their greater management complexity compared to specialized systems, ICLS can mitigate climate and market risks (Delandmeter *et al.*, 2024) while maintaining productivity and profitability. Integrating trees into these systems introduces new dynamics, offering environmental benefits such as carbon sequestration and improved animal welfare, but also posing challenges due to competition for light, water, and nutrients. Effective tree management, combined with optimized nutrient inputs, particularly nitrogen (N), is essential to balance ecological and economic outcomes. This study investigates the economic performance of ICLS with and without mature trees under varying N levels in cool-season pasture, following a strategic reduction in tree density, to evaluate the impact of this intervention on system productivity and profitability.

Materials and methods

This field experiment, covering 13.1 ha, was conducted at IDR-Paraná in Ponta Grossa-PR, Brazil (25°7'22"S, 50°3'1"W), located in a subtropical region. The site features sandy-textured soils, classified as Typic Distrudept and Rhodic Hapludox. The experiment followed a randomized block design

with three replicates and four treatments, combining two N fertilization levels, 90 kg N ha⁻¹ (N90) and 180 kg N ha⁻¹ (N180), applied as urea approximately 40 days after pasture sowing, and two ICLS: without trees (CL) and with trees (CLT).

In 2006, three tree species (eucalyptus, pink pepper, and silver oak) were planted in 6 of the 12 experimental units, spaced 3 × 14 m apart (238 trees ha⁻¹). From 2010 onward, cattle grazing was introduced during cool-season pastures following warm-season crops. Beginning in 2013, progressive thinning reduced tree density to 40 trees ha⁻¹, with a final spacing of 9 × 28 m. This new arrangement was assessed from 2017 to 2019 to evaluate its effect on the profitability recovery of CLT, just before the final tree harvest in April 2019. During the winters of 2017 (April) and 2018 (May), a mixture of black oat and ryegrass was sown at seeding rates of 45 and 15 kg ha⁻¹, respectively, with 400 kg ha⁻¹ of NPK 4-30-10 fertilizer. Maize (Balu 280 Pro) was sown in October 2017 (62 500 seeds ha⁻¹), receiving the same fertilizer rate at sowing and a topdressing of 250 kg ha⁻¹ urea. Soybean (cv. Apollo) was planted in November 2018 at 10 seeds per metre, with 230 kg ha⁻¹ of 02-20-18 fertilizer. Soybean and corn rotation is a common agricultural practice in Brazil. Three beef heifers were maintained in each paddock to assess animal performance, with stocking rates adjusted to maintain a 20 cm sward height. Grazing took place from mid-June 2017 and early July 2018, lasting approximately 105 days each year. Grain yields were measured and adjusted to 13% moisture. Tree volume was estimated by sampling 10% of the population, measuring diameter and height, and calculating volume using the Smalian method. The economic analysis considered revenue, cost, and profit, following the methodology of Volsi *et al.* (2020). Revenue was calculated based on livestock, grain, and forestry outputs, using official market prices (SEAB, 2020). Costs included all agricultural inputs, services, labor, and associated fees, with technical coefficients applied for field operations. Tree-related costs were excluded, as they were offset by prior thinning. All values were corrected to June 2023 using Brazil's official inflation index (IPCA) and converted to USD at the rate of 1 BRL = 0.2075. Variance analyses were performed using a split-plot model in Statgraphics Centurion XV. Treatments (system × N level) were the main plots, and years were the subplots. Treatments and years were treated as fixed effects, with blocks as a random effect.

Results and discussion

Each tree yielded an average of 5.28 m³ of wood, totalling 190 m³ ha⁻¹ and generating US\$ 556 ha⁻¹ year⁻¹ in revenue over 13 years. CL systems significantly outperformed CLT, regardless of N level (Table 1). Livestock production in 2017 (407 ± 36.7 kg LW ha⁻¹ year⁻¹) was significantly higher than in 2018 (175 ± 24.6 kg LW ha⁻¹ year⁻¹) due to more favourable rainfall. In 2018, 35% below-average precipitation and tree shading in CLT systems reduced output by up to 42%, resulting in lower weight gains and economic losses of US\$ 261 ± 29.0 ha⁻¹. Conversely, higher livestock performance in 2017 and in CL systems contributed up to 32% of total profit, enhancing winter revenues. Total revenue, cost, and profit were higher in the first year (US\$ 2048 ± 88.3, 1264 ± 43.0, and 783 ± 67.0 ha⁻¹, respectively), compared to the second year (US\$ 1211 ± 70.5, 891 ± 33.6, and 320 ± 49.6 ha⁻¹), when soybean was used. Despite soybeans' tolerance to moderate shading, their productivity in CLT systems dropped sharply, by 69% compared to corn (38%), even after tree thinning. This decline was linked to reduced pasture residue, lower crop area (-14%), low soybean prices in 2018–2019, and early drought conditions that intensified tree-crop competition. In contrast, corn cultivation in 2017 and CL treatments yielded better results, contributing to higher summer revenue and more stable profits. The total cost ranged between US\$ 940 ± 84.0 ha⁻¹ and US\$ 1241 ± 90.0 ha⁻¹ (Table 1). Fertilizers accounted for 39% of total input costs. Higher N levels did not increase profit, suggesting

Table 1. Grain and animal production (live weight gain per area) and economic parameters under different integration (CL, crop-livestock only and CLT, crop-livestock-trees systems) and nitrogen treatments (N90, 90 kg N ha⁻¹; N180, 180 kg N ha⁻¹).

	CL N90	CL N180	CLT N90	CLT N180	P
Animal (kg LW ha ⁻¹ year ⁻¹)	350 ± 69.6a	398 ± 64.9a	201 ± 52.2b	236 ± 45.9b	0.0004
Corn (kg ha ⁻¹) 2017–2018	8993 ± 741.3a	10452 ± 590.8a	6222 ± 333.7b	5904 ± 717.9b	<0.001
Soybean (kg ha ⁻¹) 2018–2019	3707 ± 95.7a	2929 ± 312.1a	1033 ± 125.4 b	1045 ± 142.3b	0.0001
Revenue (US\$ ha ⁻¹)	1787 ± 159.6a	1841 ± 236.5a	1483 ± 205.8b	1407 ± 210.4b	0.0234
Cost (US\$ ha ⁻¹)	1151 ± 92.3a	1241 ± 90.0a	940 ± 84.0c	978 ± 71.8b	<0.001
Profit (US\$ ha ⁻¹)	635 ± 91.6a	600 ± 151.4a	543 ± 122.2a	429 ± 149.4a	0.3841

LW, live weight. Within rows, means with the same letter do not differ significantly (LSD test, $P > 0.05$). In CLT systems, grain production was based on 86% of the area dedicated to intercropping, while 14% was occupied by trees.

that lower doses, aligned with system nutrient cycling, may help reduce costs. Net profit remained consistent across treatments, averaging US\$ 552 ± 63.2 ha⁻¹, supporting wider adoption of these sustainable systems.

Conclusion

Integrated crop-livestock systems demonstrated profitability (US\$ 552 ha⁻¹), with CLT profits largely driven by forestry revenue. However, droughts reduced livestock gains in CLT due to increased water competition. To ensure stable farmer income and minimize losses, early tree thinning and the use of shade-tolerant species are recommended. Adjusting inputs as trees mature and recognizing the value of ecosystem services can further enhance returns from carbon-neutral, tree-based products.

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Integrating grazing behaviour and local knowledge to classify Mediterranean rangeland resources and characterise their forage quality

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Abstract

Efficient use of spontaneous vegetation in Mediterranean rangelands is challenged by strong spatiotemporal variability in forage quantity and quality. This heterogeneity, exacerbated by climate change, complicates real-time assessment and the availability of shared, operational knowledge on forage value. This study aimed to develop a functional classification of rangeland resources to support grazing management. In spring and autumn 2024, we monitored the feeding behaviour of sheep and goats on three farms in southern France. We analysed 165 samples of plant parts consumed by animals using near-infrared spectroscopy (NIRS). Unsupervised classification primarily grouped the samples by plant part (leaf, stem, etc.) rather than botanical species, and captured seasonal shifts in resource quality and availability. The resulting functional classes provided a simplified but informative view of the grazing diet, revealing species-specific feeding behaviours. In parallel, interviews with 18 pastoral farmers explored their knowledge and expectations regarding rangeland use. They assessed forage value based on plant species, animal behaviour, and seasonal changes. By combining grazing behaviour, NIRS analysis, and local knowledge, we developed a typology validated through farmers' feedback and field observations, that will serve a decision-support tool to optimise the use of spontaneous vegetation in Mediterranean pastoral systems.

Keywords: functional classification, knowledge, local grazing management, Near-Infrared Spectroscopy (NIRS), pastures

Introduction

Mediterranean livestock systems rely on spontaneous forage resources, which feed small ruminant flocks from 2 to 12 months per year, depending on animal production strategies (Deschamps and Jouven, 2025). Mediterranean rangelands display high botanical diversity, multi-layered vegetation and strong spatiotemporal variability, which call for an accurate, adaptive grazing management (Jouven *et al.*, 2010). These features are not compatible with classical indicators of grazing resource such as grass height, and limit the transferability of scientific results acquired in a given context. As a result, few technical references are available to guide grazing management on Mediterranean rangelands: farmers and advisers rely mainly on empirical knowledge built from their own practical experience involving direct observation of plant x animal interactions (Fernandez-Gimenez and Fillat, 2012). Such knowledge is now challenged by the frequent disruptions in seasonal dynamics brought in by climate change. Consequently, farmers ask for scientific knowledge about the impact of climate change on pastoral vegetation, and for tools providing real-time assessment of the availability and quality of pastoral resources. To address this need, we developed a functional classification of pastoral resources

based on grazing behaviour and NIRS analysis of plant parts consumed, and examined its relevance for grazing management by confronting it with farmers' knowledge.

Materials and methods

The study was conducted in two departments of southern France. Hérault displays a typical Mediterranean climate, with garrigue shrublands, wooded areas, and woody shrublands. Ardèche is more mountainous but still under Mediterranean influence, with grasslands and broom shrublands. We implemented two complementary approaches based on surveys and direct observation of animal behaviour: (1) Eighteen small ruminant pastoral farms were selected to reflect a gradient of range-land utilization. We carried out semi-structured interviews in order to analyse how farmers perceived rangelands and what observations they relied on to manage grazing (e.g. key species, palatability, seasonal changes); (2) Behavioural observations were conducted on three of the investigated farms in spring and autumn 2024. The observers identified the plant parts (PP) ingested by sheep and goats, then recorded grazing sequences of 20 bites every 2.5 min (4 animals/farm, 4 days, 6–8 hours/day) using a coding system inspired by Mikicic *et al.* (2023). The plant parts consumed were then sampled in a way that mimicked the animals' foraging behaviour. Samples were dried, ground, and analysed using near-infrared spectroscopy (NIRS). Spectra were pre-processed (SNV, detrending, Savitzky–Golay smoothing), and an unsupervised classification (hierarchical clustering using Ward's method) was performed based on spectral data. In parallel, chemical composition was predicted using calibration models. The classes were confronted with farmers' observations during feedback sessions, revealing both convergences and discrepancies and allowing us to refine the interpretation of the classes as an operational tool for grazing management.

Results and discussion

Farmers assessed and managed grazing areas using multiple indicators, particularly vegetation characteristics such as the presence of key plant species, species palatability (often interpreted through animal behaviour), and resilience to grazing. These indicators, which would change seasonally, informed the pastoral value of a site and its role within grazing circuits. Physical attributes of the rangeland (e.g. soil type, slope and aspect, accessibility) and animal feeding behaviour played a key role in the choice of the area to be grazed at a given season or depending on the weather.

The NIRS-based classification (see Table 1) grouped 165 samples of plant parts into five functional classes: (1) green and mixed herbaceous leaves; (2) stems and leafy stems; (3) young leaves and flowers; (4) green and senescent woody leaves; and (5) reproductive organs. Based on intra- and inter-class inertia, this classification organises grazed feeds by plant organs rather than taxonomy. It complements botanical typologies (Hanisch *et al.*, 2020) and morphological typologies (Agreil & Meuret, 2004) by integrating seasonality and nutritional features. The groups display distinct chemical compositions across sites and seasons, suggesting applicability in varied contexts. A comparison with farmers' knowledge revealed both convergences and divergences. For instance, *Aphyllantes monspeliensis*, considered highly palatable and used for deferred grazing, shifted from Cluster 1 in spring to Cluster 2 in autumn, indicating seasonal decline in quality. Conversely, *Fraxinus excelsior* and *Bituminaria bituminosa* remained in protein-rich Cluster 3, while *Cytisus spp.* showed functional plasticity depending on plant part. *Brachypodium retusum*, grouped in Cluster 1, was described as unpalatable by farmers, a nuance confirmed by behavioural observations but not by the classification. These cases show how functional classification can clarify perceptions, reveal seasonal dynamics, and capture differences between plant parts. Farmers considered the classification clear and relevant. While species remain

Table 1. Chemical characteristics of the five functional classes derived from clustering

Cluster	Plant parts	Species	CP (% DM)	NDF (% DM)	IVOMD (% OM)	Farmers' perceptions
1	Green and mixed herbaceous leaves	<i>Gramineae</i> + <i>Aphyllantes monspeliensis</i>	10.6 ^a ±0.8	64.9 ^b ±3.2	42.3 ^b ±0.9	High spring palatability; early grazing but decline rapidly+used for deferred grazing
2	Stems and leafy stems	<i>Phillyrea</i> , <i>Olea</i> , <i>Cytisus oromediterraneus</i> , <i>Smilax</i> , <i>Rhamnus</i> . . .	11.1 ^a ±1.8	47.3 ^a ±1.3	56.0 ^a ±2.3	Valued ligneous resources; highly appreciated by goats (<i>Rhamnus</i> sp) during spring and autumn
3	Young leaves and flowers	<i>Fraxinus</i> , <i>Cytisus oromediterraneus</i> , <i>Cytisus scoparius</i> , <i>Bituminaria bituminosa</i> . . .	14.8 ^b ±1.2	39.9 ^c ±0.3	67.8 ^c ±0.3	High-quality resources; <i>Bituminaria</i> and <i>Cytisus</i> seen as excellent protein-rich forage
4	Green and senescent ligneous leaves	<i>Quercus alba</i> , <i>Pistacia</i> , <i>Rubus</i> . . .	11.6 ^a ±2.4	46.5 ^a ±2.3	54.4 ^a ±2.3	Species important to ensure animal intake (<i>Rubus</i> sp, <i>Quercus</i> sp. . .)
5	Reproductive organs	<i>Cytisus oromediterraneus</i> , <i>Cytisus scoparius</i> , <i>Rubus</i> . . .	9.6 ^a ±2.5	46.7 ^a ±3.7	56.7 ^a ±1.3	Seasonally used parts; contribute to diet diversification at specific time (pods, fruits. . .)

CP, Crude protein; NDF, neutral detergent fibre; OMD, organic matter digestibility. Letters indicate clusters that are significantly different according to Tukey's test ($p < 0.05$)

their primary reference, the classes provided added insight at the organ scale, refining forage quality assessment. Unexpected groupings prompted discussion on dietary roles, offering a practical tool for describing diets and managing spontaneous vegetation. This classification does not estimate resource biomass but provides a functional grid to qualify resources available at a given time for a given animal species. By linking farmers' observations to nutritionally meaningful groups, it offers a framework to optimise use of pastoral resources, guide decisions on flock movements, and adjust feed supplementation.

Conclusions and perspectives

The proposed classification of feeds grazed on Mediterranean rangelands offers a robust foundation for shared references between science and pastoral practice. It complements farmers' empirical knowledge with objective information and provides a transferable classification of rangeland resources in contexts shaped by ecological heterogeneity and seasonal variability. To ensure its practical relevance, the classification should be further validated—with supplementary data and with groups of farmers—to assess its usability and integration into local management strategies. Its utility could be enhanced by integrating farmers' ecological knowledge and remote sensing for spatiotemporal monitoring.

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Forage availability of a grass and legume mix in a silvopastoral system with native trees

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Abstract

The silvopastoral system (SPS) is a form of livestock production associated with strategies for adapting livestock to climate change. This study evaluated the biomass of palisadegrass and pigeon pea mix at different distances between tree rows in a SPS in São Carlos-SP, Brazil. The SPS consisted of Piatã palisadegrass (*Urochloa brizantha* cv. BRS Piatã) with rows of native trees, spaced 17 m apart, established in 2008, directly sown pigeon pea (*Cajanus cajan* cv. BRS Mandarin; sown in 2022). Monthly assessments were taken from March 2022 to October 2023 at five sample points along a transect in the strip between two rows of trees, with four replicates. Data for palisadegrass, pigeon pea, and total biomass were subjected to analysis of variance, and means were compared using Fisher's test ($P < 0.05$). The distance between tree rows significantly influenced forage availability in the grass-legume mixture, with greater biomass observed in areas receiving higher levels of photosynthetically active radiation. Throughout the experimental period, SPS demonstrated substantial monthly forage availability, with average dry matter (DM) yields of 2466 and 8253 kg DM ha⁻¹ for palisadegrass and pigeon pea, respectively, during the rainy season, and 1568 and 5181 kg DM ha⁻¹ during the dry season.

Keywords: agroforestry, *Cajanus cajan*, consortium, solar radiation

Introduction

One of the significant challenges in tropical pasture-based livestock production systems is ensuring year-round forage availability to meet animals' nutritional demands. Generally, this environment exhibits a seasonal forage growth pattern, being vigorous during spring and summer and significantly reduced during autumn and winter. In this context, the intercropping of grasses and legumes, mainly Pigeon pea (*Cajanus cajan*) in Brazil, has emerged as a promising strategy (Pezzopane *et al.*, 2024), due to the grazing pattern that allows forage biomass to accumulate during productive months, to be consumed during periods of forage scarcity (Furtado *et al.*, 2023). Additionally, the adoption of silvopastoral systems (SPS) is also increasing as a strategy for pasture-based livestock production in the country due to several environmental benefits (Brunetti *et al.*, 2025). However, the understanding of legume forage production under grazing in SPS remains limited, particularly in systems with native trees. The main objective of this paper is to evaluate the forage biomass production of pigeon pea intercropped with *Urochloa brizantha* cv. BRS Piatã in a SPS composed of native trees.

Materials and methods

This study was performed at Embrapa Southeast Livestock, located in São Carlos-SP, Brazil (21°57'42"S, 47°50'28"W, 854 m asl) from March 2022 to October 2023 and divided into three periods based on grazing stages: Dry 2022 (June to October 2022); Rainy 2022/23 (November 2022 to June 2023); Dry 2023 (June to October 2023). The local soil is classified as a typical Haplustox with a sandy loam texture (Soil Survey Staff, 2014), characterized by flat terrain to gently undulating terrain. The climate is classified as subtropical highland (Köppen: Cwa), with two distinct seasons: a dry season (from April to September) and a wet season (from October to March).

The SPS pasture (2.0 ha) was established in 2008 with *Urochloa brizantha* cv. BRS Piatã palisadegrass and triple-row strips of native tree species of the Atlantic Forest Biome. The tree rows were spaced 17 m apart, oriented northeast-southwest, perpendicular to the terrain's slope, with 2.5 m between tree lines and 2.5 m between trees within each row. In the central lines of each tree strip, native tree species were planted, comprising *Anadenanthera colubrina* (angico-branco), *Peltophorum dubium* (canafistula), *Zeyheria tuberculosa* (ipê-felpudo), *Cariniana estrellensis* (jequitibá-branco), and *Piptadenia gonoacantha* (pau-jacarê). For structural support (tutoring), the lateral lines were alternately planted with *Guazuma ulmifolia* (mutambo) and *Croton floribundus* (capixingui) (population density 545 trees ha⁻¹). These lateral tree rows were used as reference points for determining forage sampling distances. In April 2022, the trees were measured, indicating an average height of 9.6 and 7.7 m, and an average diameter at breast height (1.3 m above ground) of 19.0 and 20.9 cm, respectively, for the central row and lateral row species.

In January 2022, a grass-legume intercrop was established, combining the pasture with pigeon pea (*Cajanus cajan* cv. BRS Mandarin). The pasture was mowed to enable sowing of the legume in the strips between the tree rows. The legume was sown at 0.45 m distance between rows, with approximately 10 seeds per linear metre, aiming for a stand density of about 200,000 plants ha⁻¹. Initial fertilization consisted of 400 kg ha⁻¹ of potassium chloride (KCl) and 150 kg ha⁻¹ of the 03-17-00 (N-P-K) fertilizer formulation. The intercrop developed without grazing until June 2022, when grazing was allowed in the experimental area. In October 2022, the pigeon pea was cut to approximately 25 cm above ground, following recommended management practices for the species (Pezzopane *et al.*, 2024). Following mowing, fertilization was applied with 250 kg ha⁻¹ of single superphosphate and 150 kg ha⁻¹ of KCl. The area remained ungrazed until January 2023, when grazing resumed and continued until October 2023, the end of the experimental period.

During grazing periods, Canchim beef cattle (3/8 *Bos indicus* (Nellore)×5/8 *Bos taurus* (Charolais)) were used under continuous stocking, with variable stocking rates adjusted according to forage availability.

Forage biomass (pigeon pea+palisadegrass) was assessed using a 1.0×0.5 m frame. All aboveground biomass was harvested to soil level and weighed using a precision balance. Monthly assessments were taken at five sample points along a transect in the strip between two rows of trees, with four replicates per point. Samples of each species were collected: for palisadegrass, 250 g was dried at 60°C for 72 hours to determine its dry matter (DM). For pigeon pea, three plants were dried at 60°C until constant weight (≈148 h) to determine DM.

Table 1. Biomass of the consortium of Piatã palisadegrass and pigeon pea in a SPS, during three seasons of the year

Season	Biomass (kg DM ha ⁻¹)		
	Palisadegrass	Pigeon pea	Palisadegrass + Pigeon pea
1st Season (Dry 2022)	1306.1 ± 50.1 c*	2755.7 ± 167.6 b	4059.6 ± 157.9 b
2nd Season (Rainy 2022/23)	2466.6 ± 58.6 a	8253.5 ± 668.6 a	10424.2 ± 630.9 a
3rd Season (Dry 2023)	1830.2 ± 57.9 b	7608.9 ± 771.3 a	9361.8 ± 771.9 a
Significance (<i>p</i> <)	0.0001	0.0001	0.0001

* Means followed by the same letter in the column do not differ from each other (*p* ≤ 0.05) by Fisher's test.

Data of palisadegrass biomass, pigeon pea biomass, and their summed biomass were submitted to analysis of variance considering species and seasons as fixed effects. Means were compared using the Fisher test at 5% significance level, as implemented in the PROC MIXED procedure of SAS (SAS Institute, Cary, NC, USA).

Results and discussion

Differences were observed in palisadegrass biomass, pigeon pea biomass, and their summed biomass across three post-grazing seasons (*P* < 0.0001). Table 1 presents biomass (kg DM ha⁻¹) for palisadegrass, pigeon pea, and the total biomass (palisadegrass+pigeon pea) at three post-grazing seasons: Dry 2022, Rainy 2022/23, and Dry 2023.

In our study, the effect of light between tree rows altered the microenvironment throughout the experimental period. Under such conditions, shaded grasses and legumes undergo morphological and physiological adaptations to optimize light-use efficiency (Pang *et al.*, 2017), which influences forage availability, resulting in greater biomass observed in areas receiving higher levels of photosynthetically active radiation. Additionally, following the introduction of animals and the first cutting after the 1st season, the biomass of all components peaked during the 2nd season, demonstrating substantial monthly forage availability. However, due to competition for resources as plants grow, grazing pressure, effects of mowing, and the absence of vegetative regrowth in pigeon pea the stand density decreased to 140 000 plants ha⁻¹ in the 2nd season, and by the end of the experiment, following the 3rd season, the pigeon pea population naturally declined to 80 000 plants ha⁻¹. Although numbers of pigeon pea plants ha⁻¹ decreased, higher productivity was observed, thanks to greater individual plant growth. This underscores the importance of intercropping for forage supplementation during dry periods (Furtado *et al.*, 2023; Pezzopane *et al.*, 2024).

Conclusion

Throughout the experimental period, SPS demonstrated substantial monthly forage availability, yielding 2466 and 8253 kg DM ha⁻¹ for palisadegrass and pigeon pea, respectively, during the rainy season, and 1,568 and 5,181 kg DM ha⁻¹ during the dry season. The forage availability, mainly during dry periods, supported a stocking rate of 2.2 Animal Unit (AU) ha⁻¹, confirming that the pigeon pea–palisade grass intercrop in a SPS is a promising strategy to reduce seasonal forage variability while simultaneously increasing system biodiversity.

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Impact of tree and nitrogen fertilization on yield in integrated crop-livestock systems

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Abstract

This study assessed the yield advantages of integrated crop-livestock systems with and without trees (CLT and CL, respectively) over 10 years in southern Brazil. Two nitrogen (N) levels (90 and 180 kg N ha⁻¹, N90 and N180, respectively) were tested. Trees (eucalyptus, pink pepper, silver oak) were planted in 2006 and selectively thinned between 2013 and 2015 to reduce shading. Grazing by beef cattle occurred on pasture following soybean or maize crops. Relative yield (RY = CLT/CL) from 2009–2019 showed that CLT systems generally had lower yields, especially as trees aged, with RY ranging from 0.36 to 1.89. Initial tree benefits (RY > 1.0) declined mainly after year four (RY < 1.0), and N90 (0.76) had better RY than N180 (0.59), indicating limited response to N under shade. The study highlights the need to adjust N rates over time and to prioritize shade-tolerant crops or intensive tree management to maintain productivity in CLT systems.

Keywords: agroforestry, beef cattle, corn, shading, soybean

Introduction

Globally, there is a growing interest among land managers in integrated crop-livestock systems (ICLS) as a pathway toward more sustainable agriculture and diversified land use that boosts income without degrading the environment (Lemaire *et al.*, 2023). When properly managed, the synergistic interactions among ICLS components can enhance overall productivity. Integrating trees into production systems increases complexity, making it essential to enhance beneficial interactions while reducing competitive effects among components (Jose *et al.*, 2017). As tree canopies close, light becomes a limiting factor, potentially reducing crops and pasture yields. Therefore, effective tree management, such as regular thinning, is essential for maintaining light availability and sustaining productivity of both understorey crops and grazing cattle over time. This study aimed to evaluate the relative yield dynamics of crops and beef cattle within an ICLS with trees, compared to those in a tree-free (crop-livestock only), over a ten-year period that included successive thinning interventions. We also evaluated the effects of two nitrogen (N) application rates during the livestock phase on relative yield. Our hypothesis was that, in a long-term ICLS with trees, progressive management of tree canopy cover would enhance solar radiation penetration to alley crops, thereby supporting the stability of intercropping, especially under higher N levels.

Materials and methods

This field experiment, covering 13.1 ha, was conducted at IDR-Paraná in Ponta Grossa-PR, Brazil (25°7'22"S, 50°3'1"W), situated in a subtropical region. The site has sandy-textured soils, classified as Typic Distrudept and Rhodic Hapludox. The experiment followed a randomized block design with three replicates and four treatments, integrating two N fertilization rates (N90, 90 kg N ha⁻¹ and

N180, 180 kg N ha⁻¹), applied as urea approximately 40 days after pasture sowing, and two ICLS: without trees (CL) and with trees (CLT).

In 2006, three tree species (eucalyptus, pink pepper and silver oak) were planted in 6 of 12 experimental units, spaced 3 × 14 m apart (238 trees ha⁻¹). The species were interspersed within the same rows, arranged crosswise to the slope. Eucalyptus and silver oak were included for timber wood production, while pink pepper was introduced to generate environmental services and non-wood products such as fruits. Eucalyptus trees were pruned in October 2008, August 2009 and July 2011. Pink pepper trees were pruned in August 2009 and silver oak in August 2009 and July 2011. During the summer of 2013, the area was thinned to 159 trees ha⁻¹ by removing pink pepper trees, many of which had been damaged by cattle activity. Furthermore, the experimental area was thinned to 79 trees ha⁻¹ during the spring of 2014 by thinning silver oak, and to 40 trees ha⁻¹ (9 × 28 m spacing) during spring 2015 by thinning eucalyptus (thinning entire rows to decrease the shading level provided by trees). The final tree harvest was performed in April 2019. From 2010 onward, cattle grazing was introduced during cool-season pastures (black oat and ryegrass) following warm-season crops (corn (*Zea mays*) or soybean). Fertilizers (NPK) were applied annually at sowing and at variable rates for each culture, and cover fertilization with N was applied to corn (150–270 kg ha⁻¹ of urea) and pastures according to the treatments. Three beef heifers were maintained in each experimental unit to assess animal performance, with stocking rates adjusted to maintain a 20 cm sward height. Grazing took place from mid-June/early July, lasting approximately 100 days each year. Grain yields were measured and adjusted for 13% moisture. The study compared relative yields (RY = CLT/CL) using data from 2009 to 2019, where RY > 1 signifies higher CLT productivity. Variance analyses for RY were conducted using a GLM model in Statgraphics Centurion XV, with intercrops (corn, soybean and beef cattle), N levels, and years as fixed effects, and blocks as random.

Results and discussion

The results showed no significant differences in RY between intercrops ($P = 0.804$). However, significant effects were observed across years ($P < 0.001$) and N rates ($P = 0.045$). Higher RY values were recorded at N90 (0.76 ± 0.11) compared to N180 (0.59 ± 0.04), indicating a limited response to N under shaded conditions. In other words, system differences were amplified at the higher N rate, raising concerns about the utility of increased N inputs in CLT systems over time, as they also fail to offset tree-induced competition. Thus, in CLT systems, N rates should be dynamically adjusted in accordance with changing shading conditions, as noted by Komainda *et al.* (2024).

Relative yield (RY) ranged from 1.89, observed three years after tree planting, to a minimum of 0.36 eight to nine years after system initiation (Figure 1). During the early stages, up to three years of age, the trees appeared to have little to no negative impact on intercrop productivity. However, once the trees surpassed this age threshold, their influence became increasingly detrimental, with yields in the CLT system declining below those observed in the CL system. The most pronounced reductions were recorded approximately 8 to 12 years after tree planting, indicating a strong competitive effect as the trees matured (Figure 1). Following a substantial thinning intervention in 2015, which widened the spacing from 14 × 7 m to 28 × 9 m, a modest recovery in yield was observed. Nonetheless, productivity declined again in the final year of the study, suggesting persistent challenges in long-term system balance. Given the spatial configuration of the trees in this system, thinning operations must be frequent and carried out early, involving trees of varying ages and sizes. This approach enables the

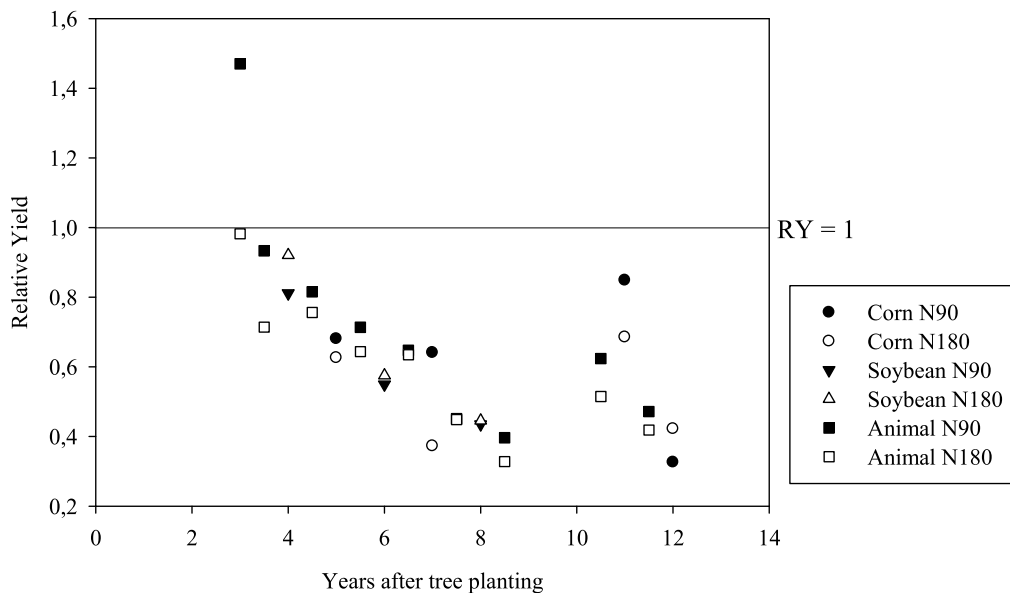


Figure 1. Relative yield (RY) of different crops and beef cattle in integrated crop-livestock systems. RY = 1 indicates that the relative yield of crops or beef cattle in integrated systems with trees is equal to that in systems without trees. RY > or < 1 indicates that the relative yield of crops or animals in integrated systems is greater or lower than that in systems without trees.

harvesting of diverse wood products while reducing competition between trees and crops, thereby enhancing light penetration to the alley crops over time.

Conclusion

The study revealed that while the type of annual intercrop had no significant impact on relative yield (RY), both N levels and system aging influenced performance. Lower RY at higher N rates suggests limited nutrient responsiveness under tree shading. As the trees matured, competition intensified, although thinning provided a temporary boost in productivity, maintaining long-term balance remained a persistent challenge. These results highlight the need for adaptive N management and thoughtful system design to maintain productivity in CLT over time.

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Floristic diversity of a Mediterranean grazed forest–grassland mosaic in northern Morocco

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Abstract

Forest–grassland mosaics are transitional ecosystems occurring at the interface of tree- and grass-dominated landscapes. In Northern Morocco, particularly in the Chefchaouen region, plant composition and diversity within these mosaics remain understudied. This study aimed to characterize the floristic composition and diversity of a forest–grassland mosaic in this southern Mediterranean area. A comprehensive plant species inventory was conducted, and a digital herbarium was developed. The flora is primarily dominated by Asteraceae, Fabaceae, Poaceae, and Lamiaceae families. Mediterranean species represent about 74% of the recorded flora, with a total of 264 species. Three major vegetation groups were identified: shrubs, trees, and herbaceous species, mainly grasses. Floristic composition varied significantly across seasons and under different grazing intensities. Commonly observed species include *Aira caryophylla*, *Anthoxanthum odoratum*, *Brachypodium distachyon*, *Cynodon dactylon*, *Hordeum murinum*, *Hyparrhenia hirta*, and *Lolium rigidum*. The findings underline the high plant diversity of this forest–grassland mosaic compared to other regions in the southern Mediterranean. These ecosystems are ecologically significant and vulnerable, particularly oak-dominated areas, which require targeted conservation efforts to preserve their biodiversity. This work provides a baseline for future monitoring and sustainable management of Mediterranean forest–grassland systems under increasing environmental and anthropogenic pressures.

Keywords: floristic composition, forest–grassland mosaic, grazing impact, Mediterranean ecosystem, pastoral landscapes, plant diversity

Introduction

Forest–grassland mosaics form ecological zones that support extensive small ruminant systems throughout the Mediterranean. In northern Morocco, forests cover about 544 291 ha and sustain large goat populations, with forest pastures providing roughly 43–100% of energy and 38–100% of protein in goat diets (Chebli *et al.*, 2022). These silvopastoral areas also carry significant socio-economic weight for local households (Chentouf *et al.*, 2011). Yet these rangelands are under increasing pressure; overgrazing reduces highly palatable species, leads to less palatable species dominance, and even to toxic plants, while broader regional trends show that Mediterranean forests harbour low endemism and face persistent pressures, especially on the southern shore (Chebli *et al.*, 2018, 2020). In northern Morocco specifically, decades of drought, fire, soil erosion, and human activity have driven substantial forest losses, underscoring the need for up-to-date floristic baselines to guide management. Despite their importance, the composition and diversity of grazed forest–grassland mosaics in the Chefchaouen region remain understudied. This study aimed to characterize the floristic composition and diversity of a forest–grassland mosaic in northern Morocco.

Materials and methods

The study was conducted in the forest rangelands at Chefchaouen region in northern Morocco, located on the southern Mediterranean coast. The area has a Mediterranean climate, with annual rainfall

ranging from 250 to 700 mm. Vegetation ranges from degraded shrublands dominated by *Cistus* spp. to mature oak (*Quercus* spp.) stands, interspersed with *Arbutus unedo*. Goat flocks average fewer than 80 head per farm, grazing mainly in spring, summer, and autumn, with reduced access in winter.

Botanical surveys were conducted for one year, from September to August, covering both blooming and fruiting stages of pastoral plants. Floristic composition was assessed using the quadrat method, with quadrat size and number determined after preliminary tests to minimize heterogeneity effects. In total, 80 quadrats of 40 m² were used for shrubs, and 80 quadrats of 1 m² were used for herbaceous vegetation, covering representative forest rangeland formations in the study area. Three-line transects were established based on visible changes in vegetation cover. Species identification followed standard botanical keys, and life forms and chorotypes were classified using established systems (Emberger and Maire, 1941; Maire, 1952 and 1987; Fennane and Tattou, 1999). The perturbation index was calculated following the formula of Hebrard *et al.* (1995). This index reflects the degree of anthropogenic disturbance, as chamaephytes and therophytes are indicative of rangeland degradation, while the denominator represents overall floristic richness. Palatability was evaluated using standard indices, combining direct field observations with literature references (Le Houérou, 1979; Sarson and Salmon, 1976).

Results and discussion

The floristic survey recorded 358 plant taxa across 228 genera and 66 families in the northern Moroccan forest rangelands. Herbaceous species dominated (86.1%), followed by shrubs (11.7%) and trees (2.2%). Asteraceae, Fabaceae, Poaceae, and Lamiaceae were the most represented families, together accounting for 42.7% of total species. The richest genera included *Trifolium*, *Cistus*, *Medicago*, *Ranunculus*, and *Vicia*. Perennials comprised 66% of recorded species, reflecting adaptation to local climatic constraints, while annuals represented 34%. Life-form analysis showed a predominance of therophytes (48%), followed by hemicryptophytes (24%) and phanerophytes (14%). This high proportion of annuals and therophytes indicates a trend toward “therophytisation”, a common feature in degraded Mediterranean ecosystems under grazing and human pressure (Benabadji, *et al.*, 2009). The calculated perturbation index was 54%, lower than in some nearby regions but still signalling significant anthropogenic impact, mainly due to fire, fuelwood collection, and agricultural expansion (Chebli *et al.*, 2018). Chorological analysis revealed that Mediterranean species dominated the flora (73.7%), followed by European (9.2%), widely distributed taxa (8.4%), and a small proportion of endemics (2%). This strong Mediterranean imprint reflects both the biogeographic setting and climatic conditions of the study area (Mele *et al.*, 2006). Palatability assessments, based on herder knowledge and direct observation, covered 95 key taxa. Among these, 93% were considered potential forage resources for goats, with species such as *Quercus coccifera*, *Q. suber*, *Arbutus unedo*, *Cistus* spp., *Erica arborea*, and *Pistacia lentiscus* ranking among the most preferred. Highly palatable and very palatable species together represented 42% of the assessed taxa, while unpalatable species accounted for just 7%. Overgrazing, however, risks reducing the abundance of highly palatable species and promoting the spread of less desirable plants, as documented in other southern Mediterranean rangelands (Chebli *et al.*, 2018).

Overall, the results confirm the high botanical richness of these silvopastoral systems compared to other southern Mediterranean sites. Yet, the trends toward therophytisation, moderate degradation, and pressure on palatable forage species underscore the need for adaptive management measures, such

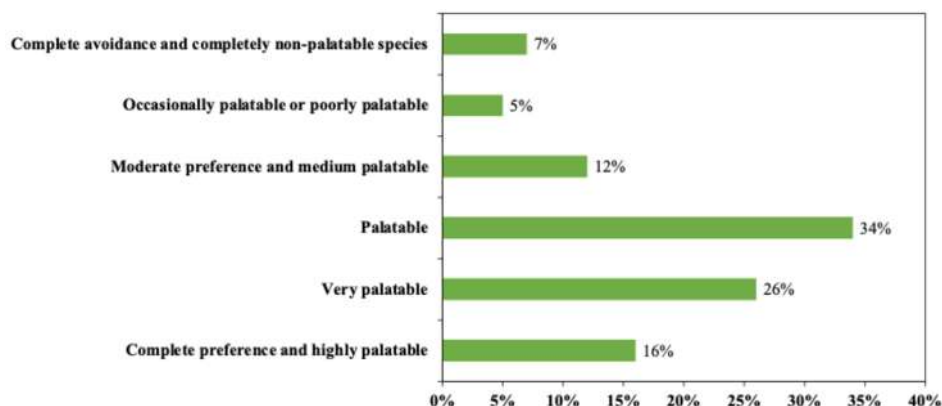


Figure 1. The importance of palatability classes of plant species in the forest rangelands of northern Morocco, based on assessments by local herders.

as controlled stocking rates and seasonal grazing rotations, to maintain both biodiversity and pastoral productivity (Figure 1).

Conclusion

The forest-grassland mosaic of northern Morocco supports remarkable floristic diversity, with a strong Mediterranean imprint and a high proportion of species valued for their pastoral, aromatic, and medicinal uses. However, signs of degradation, such as the prevalence of therophytes, moderate disturbance levels, and the decline of highly palatable species, reflect ongoing pressures from overgrazing, recurrent fires, and agricultural encroachment. Safeguarding these ecosystems requires targeted protection of oak-dominated stands, adaptive grazing strategies, and long-term monitoring of species composition and palatability trends. Such measures are vital to maintaining both biodiversity and the socio-economic functions these silvopastoral landscapes provide.

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Smartphone-based photogrammetry for biomass assessment in North African highland rangelands

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Abstract

Moroccan pastoralism, sustaining millions across 70% of agricultural lands, faces desertification, overgrazing, and climate variability in semi-arid steppes, threatening grassland resilience. This study develops a cost-effective protocol to estimate pasture biomass, enhancing sustainable grazing management. In a field mission in Ifrane-Timahdite, 60 vegetation circles (66 cm diameter, 0.33 m²) across three pastoral farms, selected for species diversity (33–63 species), were filmed using iPhone 11 Pro smartphones with hemispherical scanning for high-density 3D reconstruction (4.81 points/m²). Photogrammetry via Pix4Dmapper generated digital surface models to derive vegetation volumes, and an RGB index was calculated to characterize vegetation cover. Dry biomass, measured through destructive sampling (oven-dried at 70°C for 48 h), was correlated with volumes using a linear regression (80% training, 20% test), achieving a robust correlation. This non-destructive, scalable approach, leveraging accessible smartphones, enables precise pasture monitoring despite spatial heterogeneity. By supporting adaptive grazing, it strengthens resilience in pastoral systems, balancing pastoral livelihoods and biodiversity conservation. Future integration with drone and satellite data will enhance national-scale predictions, offering a replicable framework for sustainable pastoralism in vulnerable semi-arid regions.

Keywords: biomass estimation, linear regression, photogrammetry, rangelands, semi-arid ecosystems, smartphone

Introduction

Pastoralism in Morocco, supporting millions across 70% of agricultural lands, relies on natural pastures in semi-arid regions like Timahdite (Mahyou, 2022). It faces challenges from land degradation, with 31% of lands affected by erosion, and desertification driven by overgrazing and climate-driven droughts, which have increased by 50% in the Mediterranean (Boutallaka *et al.*, 2025). Socio-economic shifts, including a 63% decline in nomadic populations from 2004 to 2014, exacerbate resource access issues (Brisebarre & Mahdi, 2021). Accurate biomass estimation is critical but hindered by heterogeneous rangelands and unscalable traditional methods (Atzberger, 2013). Smartphone-based photogrammetry, using Structure-from-Motion (SfM), offers a cost-effective solution, producing high-resolution digital surface models (DSMs) for biomass prediction (Taugourdeau *et al.*, 2022; Lussem *et al.*, 2019). This study adapts the Structure-from-Motion photogrammetry approach of Taugourdeau *et al.* (2022) and Lussem *et al.* (2019) by replacing UAV imagery and ground camera measurements with smartphone-only video acquisition, and tailoring the processing workflow to

the specific conditions of Moroccan highland rangelands in Timahdite aiming to provide a practical, low-cost, and replicable approach for non-destructive biomass monitoring and improved sustainable grazing management.

Materials and methods

The study was conducted in Timahdite, Morocco, across three pastoral farms selected for species diversity (33–63 species per farm). Each farm contained four parallel transects spaced 40–200 m apart, and each transect included five circles (66 cm diameter, 0.33 m²) spaced 20 m apart, yielding 60 circles. A commercial GPS tracker box (14 × 9 × 7 cm) was placed beside each circle as a scaling reference. For each circle, two 25–30 s videos were captured with an iPhone 11 Pro using a handheld hemispherical scan from ground level to approx. 1 m, generating dense 3D reconstructions (4.81 points/m²). Vegetation was then clipped at ground level, weighed fresh with a spring scale, oven-dried at 70°C for 48 h, and reweighed for dry biomass.

Videos were processed in Pix4Dmapper (v4.8) using the “3D Maps” template (point cloud, DSM, orthomosaic; DSM resolution of 8.6 cm), suitable for hemispherical scans. In QGIS (v3.28), the Normalized Difference Green-Red Index (NGRDI), calculated as (green–red)/(green+red), isolated vegetation pixels by leveraging green reflectance. A threshold of –0.07 adjusted between –0.04 and 0 for varying lighting conditions (Chen, 2025), determined by histogram analysis for each image was applied to create a binary map (1 = vegetation, 0 = non-vegetation). The binary raster was polygonized using the Polygonize tool. The DSM was clipped with the circle’s interior vector, then with vegetation pixels using Clip Raster by Mask Layer. Vegetation volume was calculated using Raster Surface Volume (base level: lowest elevation, method: count above base). Volumes were scaled by matching GPS tracker box dimensions in QGIS to their known size.

A table of 60 data points (circle number, dry biomass, volume) was compiled. Seven entries were removed because of clearly aberrant volume-to-biomass ratios that were very likely caused by field errors during biomass cutting and collecting. The remaining 53 data points were analysed in Google Colab (Python 3.8) and data was split 80% training, 20% testing.

Results and discussion

The study area is dominated by *Carex divisa*, *Bromus mollis*, and *Stipa parviflora*, with low vegetation cover under extensive grazing typical of the Timahdite highland rangelands.

Linear regression yielded an R^2 of 0.64, explaining approx. 64% of dry biomass variance from volume. The Pearson correlation coefficient was 0.76, reflecting a strong linear relationship and accounting for 57% of shared variance (0.76²). Model errors remained moderate: the mean absolute error (MAE) was 12.20 g per 0.33 m² plot, which is acceptable relative to the biomass range of 0.2–111 g, while the mean squared error (MSE) reached 465.45 g² and the root mean squared error (RMSE) was 18.37 g, indicating moderate accuracy but larger errors for high-biomass samples.

The results align with SfM-based studies in semi-arid rangelands, where volume-based models achieve moderate to high accuracy (Taugourdeau *et al.*, 2022; Lussem *et al.*, 2019). The R^2 and correlation confirm smartphone photogrammetry’s potential for low-cost high-resolution biomass estimation, capturing fine-scale variability in heterogeneous rangelands more than coarser-resolution satellite

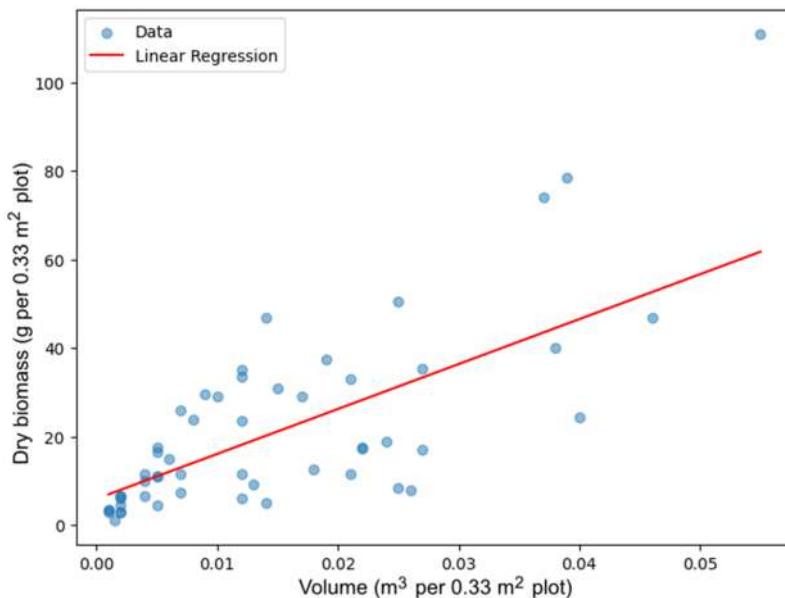


Figure 1. Plot of volume vs. dry biomass with regression line.

methods like Sentinel-2 (Mahyou, 2022). The MAE indicates practical utility for herders adjusting grazing pressure, while the RMSE suggests challenges in predicting high-biomass patches, likely due to the small dataset (53 circles). Lighting variability affected NGRDI thresholding, potentially introducing errors in sparse vegetation. Compared to drone-based studies (Fraser *et al.*, 2024), smartphones reduce costs and regulatory barriers, making the method accessible to Moroccan herders and institutions like INRA. The small dataset limits model variability, necessitating larger samples for improved generalization. Future work will integrate smartphone calibrations with drone acquisitions and Sentinel-2 imagery to enable biomass monitoring from the plot to the national scale, following the multi-scale frameworks successfully tested in Senegal and West Africa (Nungui-Pambu *et al.*, 2023; Taugourdeau *et al.*, 2022).

Conclusions

Smartphone-based photogrammetry offers a scalable, cost-effective solution for biomass estimation in Moroccan rangelands, with linear regression achieving a reasonably reliable R^2 of 0.64. Despite limitations from a small dataset, the method supports adaptive grazing to mitigate degradation. Expanding sample sizes and integrating multi-scale remote sensing will enhance the method's robustness and render it directly applicable for sustainable pastoralism and rangeland management in Morocco.

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Nutritional value of green alder leaves and their selection by livestock on encroached mountain pastures

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Abstract

Green alder is an encroaching species on mountain pastures, which causes several agro-environmental issues. Targeted grazing could be a useful tool to limit its spread. We evaluated the nutritional value of green alder leaves and their selection by different livestock categories, such as Capra Grigia and Chamois Coloured goats, Highland cattle, and heifers of productive breeds. We collected green alder leaves in summer and analysed their chemical composition and *in vitro* gas production and digestibility. Mean crude protein content was 201 g (kg DM)⁻¹, and condensed tannins ranged from 3.4 to 40.9 g (kg DM)⁻¹. *In vitro*, a diet containing 20% green alder leaves showed the highest digestibility and lowest CH₄ emissions at the beginning of summer, with lower CH₄ emissions compared to a 100% hay diet. Feeding observations showed that green alder accounted for 45% of goats' diet, 12% of Highland cattle diet, and 1.4% of heifers' diet. Green alder was actively selected by goats, while Highland cattle were indifferent, and heifers avoided it. Our results suggest that green alder can be a valuable forage, especially at the beginning of the summer. Moreover, robust livestock can exploit green alder and its incorporation into animal diets could reduce greenhouse gas emissions.

Keywords: *Alnus viridis*, digestibility, forage, feeding preferences, goats, Highland cattle

Introduction

Over the past century, agricultural land abandonment in mountainous areas has led to the expansion of green alder (*Alnus viridis* (Chaix) DC) across Central Europe, causing negative effects, such as reduced biodiversity and herbaceous forage availability, nitrogen leaching, and soil acidification (Zehnder *et al.*, 2020). Although the use of Highland cattle grazing has been proposed as a biological control method for green alder encroachment (Pauler *et al.*, 2022), a comprehensive analysis of the forage potential of green alder along the growing season and its selection by different livestock categories is still lacking. In this context, our objective was to evaluate the forage value of green alder and its selection by different livestock species. To achieve this, we synthesised data from three studies (Nota *et al.*, 2024; Svensk *et al.*, 2024; Mochi *et al.*, 2025). Specifically, we assessed the seasonal variation in green alder leaf chemical composition, *in vitro* digestibility, and gas production over two years, and we determined, through direct observations, the feeding preferences of Highland cattle, Capra Grigia and Chamois Coloured goats, and heifers of production-oriented cattle breeds.

Materials and methods

Green alder nutritional value

Green alder leaves were harvested during June, July, and August of 2020 and 2021 across four summer pastures: one in the Italian Alps (Val Vogna) and three in the Swiss Alps (Bovonne, Champlong, and Alp Weissenstein). Five green alder shrubs were sampled per site, ensuring canopy-level access up to 1.8 m in height to simulate livestock browsing. Air temperature was measured at each site, and cumulative growing degree days (GDD) were calculated as a proxy for seasonal temperature progression (Grigorieva *et al.*, 2010).

Leaf samples were split into two subsamples: one was dried and ground for laboratory analysis, and the other was freeze-dried for tannin quantification and *in vitro* assays. Nitrogen content (N) was analysed using the Dumas method (ISO 16634–1:2008), with crude protein calculated as $N \times 6.25$. Phosphorus (P) was determined via ICP-OES after microwave digestion. Fibre content—including neutral detergent fibre (aNDFOM), acid detergent fibre (ADFOM), and lignin (ADL)—was measured gravimetrically following Van Soest protocols. Total phenols and tannins (including condensed tannins) were quantified using modified Folin-Ciocalteu and butanol-HCl assays according to Makkar (2003). *In vitro* organic matter digestibility (IVOMD) and methane (CH₄) and carbon dioxide (CO₂) production were assessed via 24-hour incubation using the Hohenheim Gas Test, simulating cattle diets with a) 20% green alder leaves and 80% hay, and b) a 100% hay diet as a control. We tested the effects of GDD on leaf chemical composition, phenolic content, IVOMD and fermentation gas production, using Linear Mixed-Effects Models (LMM). GDD was treated as a fixed effect, with summer pasture and year as random factors; and the cow used in the digestibility experiment was included as an additional random factor. We also performed t-tests for comparing the IVOMD and fermentation gas production between a diet including green alder leaves and a control diet (100% hay).

Animal foraging behaviour

To assess feeding preferences, we carried out direct observations on three livestock categories: (1) Highland cattle; (2) Capra Grigia and Chamois Coloured goats; and (3) heifers from production-oriented breeds (Holstein, Simmental, and Fleckvieh) on green alder-encroached pastures. Observations on Highland cattle were conducted in the summer of 2021 in the summer pasture of Bovonne. Observations on goats and heifers were conducted in the summer of 2024 across three summer pastures in the Swiss Alps (Bovonne, Grand Clé, and Conche). Observations were carried out in 15-second intervals, during which each observer visually estimated the relative abundance and consumption of plant species within an area of 1.8 m in height and 0.5 m in radius around one animal's head. All woody species and tall herbs were identified to the species level; other plants were categorized as either 'herbaceous' or 'ferns.' The proportion of green alder in the diet and Jacobs' selection index (Jacobs, 1974) were then estimated. Jacobs' index ranges from –1 (total avoidance) to +1 (total preference), to evaluate feeding selection by livestock.

Results and discussion

Throughout the growing season, green alder leaf N content decreased, but remained above global averages for woody species (Table 1; Tian *et al.*, 2018). This resulted in consistently elevated crude protein levels (mean = 201 g (kg DM)^{–1}). Green alder P content also declined with GDD, but its values remained comparable to levels in *Alnus glutinosa* and other deciduous woody plants (Luske and van Eekeren, 2017; Tian *et al.*, 2018). The fibre fractions of green alder leaves increased significantly during the season, which aligns with previous findings on other species showing lower forage quality in

Table 1. Summary of linear models relating green alder (*Alnus viridis*) leaf composition to cumulative growing degree days (GDD).

Variable	Range (g (kg DM) ⁻¹)	P-value	R ² (cond.)	R ² (marg.)	Estimated equation
Nitrogen (N)	22.6–47.0	<0.001	0.58	0.30	N = 37.27–0.012 GDD
Phosphorus (P)	0.9–5.8	<0.001	0.72	0.51	P = 1.33–0.001 GDD
aNDFOM	202.8–454.6	<0.001	0.76	0.62	aNDFOM = 219.27+0.232 GDD
ADFOM	116.1–353.5	<0.001	0.51	0.24	ADFOM = 151.36+0.105 GDD
ADL	41.6–257.3	<0.001	0.73	0.48	ADL = 30.30+0.167 GDD
Condensed tannins	3.4–40.9	<0.001	0.58	0.05	CT = 2.24+0.001 GDD

late summer (Gonzalez-Hernandez *et al.*, 2000). In a diet with 20% green alder, IVOMD decreased over the season (from 668 to 601 g (kg DM)⁻¹), likely due to increasing fibre and lignin content (Table 1). Condensed tannin concentrations increased over the season. In the diet containing 20% green alder leaves, CH₄ production was reduced by 17% and CO₂ emissions by 5%, compared to the 100% hay diet. However, CH₄ production increased over the season, ranging from 34.54 to 49.86 ml g⁻¹.

Feeding observations showed that green alder accounted for 45% of the goats' diet, 12% of the Highland cattle diet, and only 1.4% of the heifers' diet. Jacobs' index values were 0.41 for goats, indicating an active selection for this species; approximately zero for Highland cattle, showing that green alder was consumed proportionally to its abundance in the environment; and –0.56 for heifers, which avoided it.

Conclusion

Considering its forage quality and the feeding preferences observed among different livestock categories, we conclude that robust animals such as Highland cattle and goats can effectively forage on green alder-encroached pastures, helping to control its spread in mountain pastures. Moreover, incorporating green alder into cattle diets during early summer may contribute to reduce enteric methane emissions.

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Shared innovations for enhancing agroforestry in the mediterranean: the SHARE Project approach

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Abstract

Agroforestry systems, combining woody perennials with crops and/or livestock, are key Mediterranean land uses rooted in local traditions and valued for their biodiversity and ecosystem service (ES) provision. However, they remain threatened by land abandonment, market marginalisation, low profitability, and socio-institutional constraints. The SHARE project tackles these issues through a transdisciplinary, multi-actor approach spanning six Mediterranean countries—Italy, France, Portugal, Cyprus, Tunisia, and Morocco. Using the ES framework, SHARE co-designs innovations to enhance agroforestry efficiency, productivity, and sustainability. Living Labs engage stakeholders in co-developing business models, policies, and practices to build capacity and foster inclusive value chains. By addressing ecological and institutional barriers and testing innovations, they will generate policy briefs, training materials, and strategies for products like Halloumi cheese and Argan oil, supporting rural resilience and sustainable Mediterranean landscapes.

Keywords: agroforestry systems, ecosystem services, Living Labs, Mediterranean, sustainability

Introduction

Agroforestry systems are inherently more complex than specialised systems, enhancing structural diversity and functional diversity throughout resource efficiency, especially in nutrient, light, and water use (Guillot *et al.*, 2019). Due to this feature, their management requires constant adaptation to local conditions, supported by a blend of traditional knowledge and innovation (Kmoch *et al.*, 2018). In the Mediterranean, agroforestry has long shaped rural, often arid and marginal, landscapes and is increasingly recognised in policies like the European Green Deal, the Common Agricultural Policy,

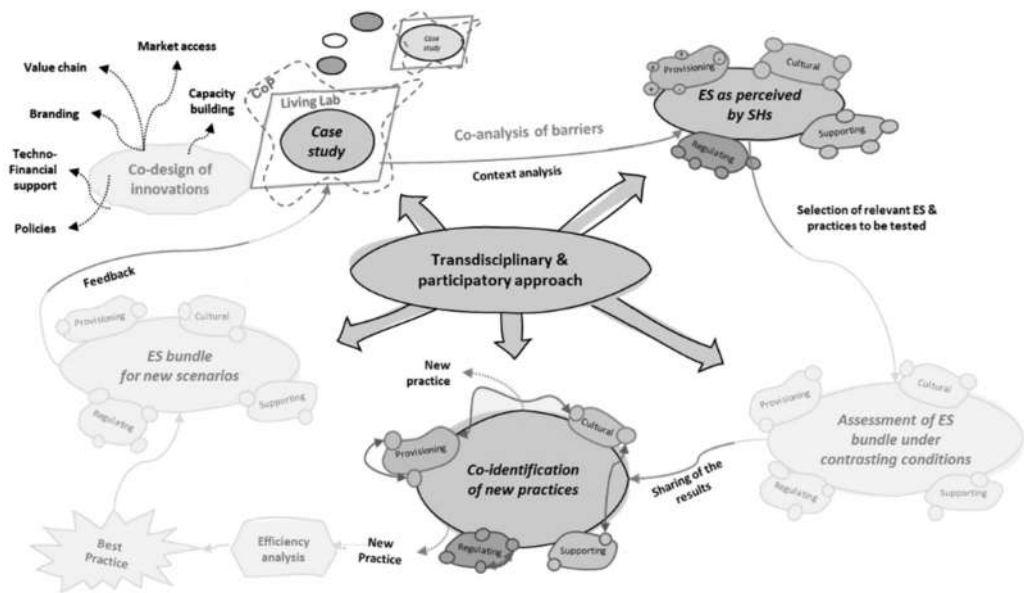


Figure 1. SHARE process of co-design for innovative business models, practices, and strategies.

and the FAO's GIAHS programme. Despite this support, challenges remain that limit sustainability and benefits for rural communities (Sollen-Norrlin *et al.*, 2020). The EU-funded SHARE project addresses these issues through a transdisciplinary, multi-actor approach by establishing Living Labs (LLs) in six Mediterranean countries. These LLs engage farmers, researchers, policymakers, and civil society to identify barriers, co-design innovations, and assess practices in real world farms and plots within those farms. The SHARE project pursues the following objectives: (i) to evaluate trade-offs and synergies among ES within Mediterranean agroforestry systems through the application of the Millennium Ecosystem Assessment framework, including their economic valuation; (ii) to develop context-specific scenarios that integrate biophysical, socio-economic, and policy dimensions to explore alternative management pathways; (iii) to deliver evidence-based strategies that enhance the resilience, productivity, and socio-ecological value of Mediterranean agroforestry systems.

Materials and methods

Six case studies across the Mediterranean, each representing a distinct agroforestry system, serve as demonstration sites to implement and test the SHARE methodology. Throughout the project, LLs will operate continuously to collaboratively identify barriers and co-develop innovative business models, practices, and strategies that integrate technical, socio-economic, and policy-market perspectives (Figure 1). Besides the internal exchange in each LL, the ongoing exchange between LLs will facilitate mutual learning, enable tailored solutions, strengthen capacity building, and foster the creation of Communities of Practice (CoP). The methodology adopted in the SHARE project is grounded in a transdisciplinary and participatory research framework, designed to be both scientifically rigorous and highly replicable across similar agroecological contexts. Central to this approach is the integration of hybrid knowledge, achieved through iterative co-learning processes that combine local expertise (e.g., farmers and practitioners), policy and institutional insights (e.g., decision-makers, extension officers), and scientific evidence (e.g., researchers in biodiversity, socioeconomics, and governance). This integration is operationalised through a combination of participatory workshops, stakeholder

mapping, and co-design sessions within each LL. Semi-structured interviews and focus group discussions are conducted to identify locally perceived challenges and opportunities, while Delphi surveys and multi-criteria analysis are used to prioritise management options and assess trade-offs among ES. Quantitative data from field measurements (e.g., soil fertility, vegetation structure, biomass, and carbon stock) and remote sensing are coupled with farm-level socio-economic surveys to model system performance using ES assessment tools (e.g., InVEST). The integration of qualitative and quantitative evidence ensures a robust analytical framework to support context-sensitive, evidence-based innovation pathways in Mediterranean agroforestry systems.

Results and discussion

The expected impacts of the SHARE project will closely depend on the co-identification, co-assessment, and co-design of new models, practices, and strategies for Mediterranean agroforestry systems. Due to SHARE's bottom-up approach (Toderi *et al.*, 2017), the most relevant issues for stakeholders will be identified during the process and are expected to vary—from creating firebreaks to buffer wildfires in arid regions (e.g., Morocco, Tunisia) to increasing soil cover by vegetation in temperate areas (e.g., Italy, France). The co-identification of relevant challenges will be supported by a continuous flow of hybrid knowledge, regularly updated by LL members through tools such as predictive models to address the most critical ES (e.g., crop and livestock production, soil health, fertility, biodiversity, carbon sequestration, water regulation). Key ES provided by agroforestry systems will be assessed using both the Millennium Ecosystem Assessment and Nature's Contributions to People frameworks (Dean *et al.*, 2024). These ES bundles will be evaluated across current, innovative, and trial practices, considering specific site conditions. To complement this, farm-level data on inputs, outputs, and socio-economic factors will be analysed to benchmark technical and economic efficiency, helping to identify best practices that can be adopted more widely.

Conclusion

The SHARE project is expected to generate measurable ecological, economic, and social benefits for Mediterranean agroforestry systems. Preliminary outcomes indicate increased soil organic carbon (+15–25% in pilot sites), enhanced vegetation cover stability, and higher crop-livestock integration efficiency. Economic analyses suggest improved farm profitability through diversified income streams and reduced input dependency. At the social level, LL have strengthened stakeholder cooperation and local innovation capacity. By aligning with the EU Biodiversity Strategy for 2030, SHARE contributes to the restoration of degraded ecosystems and the co-design of climate adaptation strategies with local communities. Its actions support key UN Sustainable Development Goals (SDGs)—notably biodiversity conservation (SDG 15), climate action (SDG 13), food security (SDG 2), and poverty reduction (SDG 1). Through inclusive business models and the creation of Communities of Practice, SHARE fosters long-term adoption of resilient agroforestry practices, positioning Mediterranean agroforestry as a model for multifunctional and climate-smart land use.

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Rewilding with semi-wild horses: effects on vegetation structure and composition in the Côa Valley, Portugal

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Abstract

Mediterranean landscapes feature diverse vegetation types and are highly vulnerable to wildfires, with grazing playing a key role in regulating vegetation structure and composition. This study explores the early effects of semi-wild horse grazing in two rewilding areas in Portugal. Using grazing exclusion areas as control, we tested whether this extensive regime could manage vegetation structure and composition, to mitigate fire hazard and promote biodiversity. We assessed grass height, shrub height, cover and aboveground biomass, plant species richness, species turnover, forbs-to-grasses ratio (F:G) and functional group importance value (IV). Results showed a greater increase in grass height in ungrazed plots, suggesting horse grazing helps limit grass height. Shrub metrics showed no significant differences between treatments, indicating horses had limited control of woody vegetation. The F:G ratio and forb IV were higher under horse grazing, suggesting potential benefits for anthophilous insects. Species richness remained stable, but temporal turnover was higher in ungrazed plots. Overall, horse grazing contributed to controlling grass dominance, reducing local fire hazard while potentially enhancing resources for insects. However, its limited impact on woody vegetation highlights the need for diverse grazing systems to achieve broader vegetation management goals in rewilding approaches.

Keywords: ecosystem management, grazing, plant community, Rewilding, semi-wild horses, vegetation structure

Introduction

Large herbivores play a key role in ecosystem functioning by regulating vegetation structure and composition, promoting nutrient cycling, and facilitating seed dispersal (Pringle *et al.*, 2023). In Mediterranean landscapes, grazing is being valued as a management tool for maintaining open habitats and preventing shrub encroachment, ultimately promoting landscape's fire resilience (Kirkland *et al.*, 2024). Rewilding has emerged as a restoration strategy to enhance ecological processes, including herbivory, in landscapes where wild herbivore populations are absent or where traditional livestock farming is infeasible (Shackelford & McDougall, 2023). This study assesses the early effects of a rewilding approach on vegetation structure and composition in two rewilding areas in the Côa Valley, Portugal. To assess vegetation responses, we compared areas grazed by horses with ungrazed control plots. We hypothesized that: (a) horse grazing would regulate vegetation structure compared to grazing exclusion; (b) species richness would decline in ungrazed areas, due to absence of grazing disturbance; (c) plant community composition would diverge between treatments driven by differences in grazing pressure; and (d) the relative representation of grasses and forbs would shift, with horse grazing favoring forb representation, which is relevant for anthophilous insects (Bonavent *et al.*, 2023). This study contributes to understanding how semi-wild herbivores can restore herbivory processes, promote habitat heterogeneity, and support fire hazard mitigation in Mediterranean rewilding contexts.

Table 1. Summary of key findings by vegetation metric and site between grazed and ungrazed plots.

Metric	Site 1	Site 2	Interpretation
Grass height	Taller in ungrazed plots ($p < 0.001$)	Taller in ungrazed plots ($p = 0.002$)	Supports horse grazing role in controlling fine fuels
Shrub cover, height, biomass	No significant differences	No significant differences	Horse grazing insufficient to regulate woody vegetation
Species richness	No significant differences	No significant differences	Short timeframe may limit richness response
Species turnover	No significant differences	Higher in ungrazed plots ($p = 0.045$)	Ungrazed plots may be undergoing faster compositional change
F:G ratio	Increased under grazing ($p = 0.009$)	No grazing effects but increased over time	Grazing maintains forb presence, beneficial for pollinators
<i>Stipa gigantea</i> frequency	Increased in ungrazed plots	Minimal occurrence	Grazing helps contain spread of fire-prone grasses
Importance value (IV)	Forb IV increased in grazed plots	Forb IV increased in grazed plots	Functional shift toward more flowering plants, beneficial for anthophilous insects

Materials and methods

The study was conducted between 2021 and 2024 in two rewilding sites in the Greater Côa Valley, Portugal. Both sites feature Mediterranean woodland-shrubland mosaics and had a history of livestock grazing. In 2021 and 2022, Rewilding Portugal introduced Sorraia horses under low-intensity continuous grazing regimes. Both sites are managed to restore herbivory-related ecological processes and enhance habitat heterogeneity.

At each site, six paired survey sites (40 m×40 m) were established, each containing two plots grazed by horses and two fenced ungrazed plots for control (10 m×10 m each). Surveys were carried out every spring between 2021 and 2024. Structural metrics included mean grass height, mean shrub height, fractional shrub cover, and estimated aboveground shrub biomass. Composition metrics included species richness, the forb to grass ratio (F:G) and the cumulative importance values (IV) of forbs and grasses (Kent & Coker, 1994). In addition, species turnover and the frequency of *Stipa gigantea*, a tall fire-prone grass, were analysed as indicators of community change and fuel structure. Linear mixed models were used to analyse vegetation structure, with grazing treatment, year, and their interaction as fixed effects, and survey site as random effect. Community composition was assessed using ANOVA, paired t-tests, and temporal turnover analyses. All analyses were conducted in *R* software (R Core Team, 2022)

Results and discussion

Results indicate that across both study sites, horse grazing significantly reduced grass height over time, particularly in 2024, while ungrazed plots showed much taller grasses (Table 1), supporting the role of horses in controlling the accumulation of fine fuels, which are critical contributors to fire hazard in Mediterranean ecosystems (Cardoso *et al.*, 2022).

In contrast, grazing effects were not observed for any of the shrub-related metrics at either site, suggesting that horse grazing alone may not exert enough pressure to control shrub encroachment (Ribeiro *et al.*, 2024). Vegetation species richness did not differ between grazing treatments at either site. However, temporal turnover was significantly higher in ungrazed plots at Site 2, indicating greater shifts in community structure with a dominance of grasses in the absence of grazing. The F:G ratio increased

in grazed plots at Site 1, while remaining stable in ungrazed plots, suggesting that grazing disturbances may favour forbs by reducing competition with grasses (Tuomi *et al.*, 2019), which suggests benefits for anthophilous insects (Bonavent *et al.*, 2023). Supporting this pattern, the IV of forbs increased under horse grazing at both sites, while those of grasses declined. Notably, *Stipa gigantea* became more frequent and dominant in ungrazed plots, which reinforces concerns about fine fuel accumulation in the absence of grazing (Davies *et al.*, 2022). To fully meet restoration and management goals, promoting a diverse herbivore assemblage and using complementary strategies, such as shrub clearing or prescribed burning, may be needed (Castro *et al.*, 2022).

Conclusions

This study provides evidence that semi-wild horse grazing can help mitigate local fire hazard by reducing grass dominance in Mediterranean landscapes, where ungrazed areas risk accumulating fine fuels. By enhancing forb representation, horse grazing is also promoting food and shelter resources for insects. However, limited impact on shrub structure suggests that horse grazing alone may be insufficient to control woody vegetation. A multi-herbivore approach and integrating complementary measures may be necessary to achieve broader vegetation management goals. These findings support the role of grazing as a nature-based solution for landscape resilience in Mediterranean ecosystems.

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Effects of incorporating willow silage in ewes' diets on performance and parasite egg excretion

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Abstract

Faced with increasingly unpredictable weather conditions, the use of woody fodder may offer a sustainable solution to ensure sufficient forage stocks for the winter period. This study aimed to evaluate the effects of incorporating willow silage into the diets of ewes on animal performance and egg excretion. One-year-old willow twigs were harvested in August, shredded, and ensiled under plastic sheets. Sixteen ewes aged 10 to 12 months were assigned to two groups: a control group and a willow group. Both groups received hay, grass silage, and a cereal mixture. In the willow group, part of the grass silage was replaced by willow silage. Feed intake, refusals, and body weight were monitored over a two-month period. Internal gastrointestinal parasite egg excretion was assessed before, during, and after the willow silage distribution. The average intake of willow silage was 118 g per ewe per day, representing 14% of the diet's dry matter. Refusals contained small willow twigs (approximately 0.2 cm in diameter). Incorporating 14% willow silage on a dry matter basis did not affect ewe growth. However, a reduction in faecal egg counts of gastrointestinal strongyles was observed following willow silage distribution.

Keywords: ewes, parasite egg excretion, silage, willow (*Salix viminalis* L.)

Introduction

Warm and dry summers, increasingly frequent in Belgium, have significant impacts on feed management. Limited grass growth forces livestock farmers to use winter forage stocks prematurely, while the lack of regrowth in forage crops prevents the replenishment of reserves. This situation threatens the forage autonomy of farms. Trees, thanks to their deep root systems, can access water sources unavailable to conventional forage crops, making them more resilient to drought. Among these, willow (*Salix viminalis* L.) presents promising potential as a woody fodder. Due to their tannin content, willow leaves may help reduce gastrointestinal nematode burdens and decrease female worm fecundity in young sheep (Mupeyo *et al.*, 2011). This study aimed to evaluate the effects of incorporating willow silage into the diet of ewes on zootechnical parameters such as feed intake and live weight gain, in order to assess the feasibility of including this forage in sheep diets. Additionally, internal parasite egg excretion was monitored to investigate the potential anthelmintic properties of willow when administered as silage.

Materials and methods

One-year-old twigs of basket willow (*Salix viminalis* L.) were harvested in August 2023 using a forage harvester. The twigs were shredded and ensiled in two 1 m³ silos covered with plastic sheets. To enhance preservation, salt was added at a concentration of 27 g·kg⁻¹ in fresh matter (FM). Willow silage used

Table 1. Chemical composition of diets with or without silage given to the ewe

	Control	Willow
Dry matter (%)	59.3	65.9
Ash (g (kg DM) ⁻¹)	35.4	64.1
Crude fibre (g (kg DM) ⁻¹)	231	240
Crude protein (g (kg DM) ⁻¹)	130	120
Calcium (g (kg DM) ⁻¹)	5.41	5.59
Phosphorus (g (kg DM) ⁻¹)	3.77	3.29
Sodium (g (kg DM) ⁻¹)	0.78	2.64
Magnesium (g (kg DM) ⁻¹)	1.75	1.66

in this study had the following composition: dry matter (DM) content was 51% FM, crude fibre was 232 g (kg DM)⁻¹, and crude protein was 64.4 g (kg DM)⁻¹. Sixteen Ardennes Red ewes, aged between 10 and 12 months, were included in the trial, which began in March 2024 while the animals were housed indoors. Prior to the experiment, all ewes were weighed and assigned to two homogeneous groups along liveweight. The Control group received a conventional diet composed of grass silage, hay, a grain mixture, and mineral supplements and the Willow group received a similar diet, but with a substitution of grass silage by willow silage. In the Willow group, the inclusion rate of willow silage in the dietary dry matter was progressively increased over the course of the trial: 6% from day 0 to 17, 9% from day 18 to 25, 12% from day 26 to 32, and 14% from day 33 to 56. The chemical composition of the diets is presented in Table 1. Feed intake and refusals were recorded daily for a period of 56 days. Ewes were weighed biweekly to monitor growth. At the end of the indoor feeding period, all animals were moved to a pasture that had not been grazed by ruminants in previous years, to minimize parasite contamination.

Faecal samples were collected at the beginning of the trial, 33 days and 56 days after (T0, T1 and T2) to count internal gastrointestinal parasite eggs. A supplementary counting was realised 42 days after the end of willow distribution (T3). The internal parasites targeted were: *Capillaria* sp., non-pathogenic coccidia, pathogenic coccidia, *Dicrocoelium lanceatum*, *Fasciola hepatica*, *Moniezia* sp., *Nematodirus* sp., *Paramphistomum* sp., *Skrjabinema* sp., other gastrointestinal strongyles, *Strongyloides papillosus*, and *Trichuris* sp. A linear mixed-effects model was fitted to assess the effects of group, parasite species, and sampling date, as well as their interactions, on parasite egg excretion. The model included individual animal ID as a random intercept to account for repeated measures and inter-individual variability. Interaction terms were retained to capture potential differential parasite responses across groups and time points. We applied a log(OPG+1) transformation to stabilize the variance of count data and fitted a linear mixed-effects model (log(OPG+1)~Group*Parasite*Date+(1|ID)) using the lme4 package in R. Post-hoc analyses were conducted using the emmeans package to estimate marginal means for each Group–Date combination within each Parasite group. These contrasts enabled the identification of statistically significant differences in OPG values between experimental conditions, while accounting for model structure and covariates. Model assumptions were verified through residual diagnostic.

Results and discussion

The crude protein content of the willow silage was relatively low compared to values reported by Muklada *et al.* (2021), which ranged between 10 and 14% DM. The elevated sodium content in the willow silage was attributed to the salt added during the silo preparation to improve preservation. Total feed intake per ewe was 910 g day⁻¹ in the willow group and 890 g day⁻¹ in the control

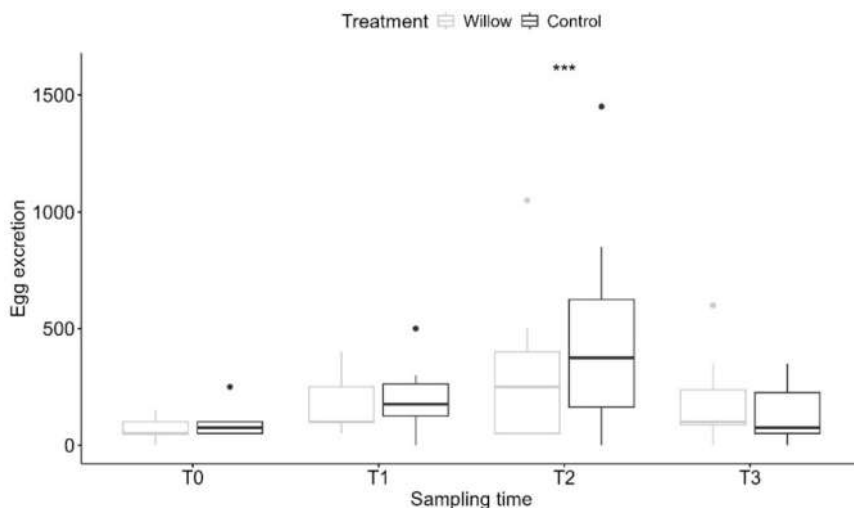


Figure 1. Gastrointestinal strongyles eggs per g (EPG) in faeces of ewes receiving a control diet or a diet including willow silage.

group. Willow silage intake averaged 118 g day^{-1} , representing 14% of the diet's dry matter. Refusals contained willow twigs of approximately 0.2 cm in diameter, indicating partial rejection of coarse material. Both groups gained weight steadily over the trial, with slightly more variation in the willow group. Average daily liveweight gain was $126 \pm 31 \text{ g day}^{-1}$ in the Control group and $142 \pm 47 \text{ g day}^{-1}$ in the Willow group ($p = 0.44$). Faecal egg counts showed a dynamic evolution throughout the trial, with a peak at T2 followed by a decline in both groups. No statistically significant differences were observed between the two groups for most parasites, except for gastrointestinal strongyles. At T2, the control group exhibited significantly higher excretion of gastrointestinal strongyles ($p = 0.001$; Figure 1). These findings are consistent with previous studies: Mupeyo *et al.* (2011) observed reduced parasite egg excretion in lambs fed chopped willow and Lira *et al.* (2008) reported that willow fodder blocks could reduce anthelmintic drench use in New Zealand. Importantly, the present study shows that ensiled willow—rather than fresh or extracted forms—can also contribute to reducing parasite egg excretion in ewes.

Conclusion

In this study, willow silage was incorporated at 14% of the diet's dry matter, a level likely limited by the high salt content added during the silo preparation for preservation. This inclusion did not affect liveweight gains but had a positive impact on gastrointestinal strongyles egg excretion, suggesting its potential as a natural strategy to support parasite control in sheep. Notably, these effects were observed with ensiled willow, highlighting its relevance even after conservation.

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Winter-spring rainfall amount drives Mediterranean grasslands' productivity and diversity

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Abstract

Mediterranean grasslands and woodlands are highly diverse ecosystems and an important food resource for pastoralism. However, these systems are threatened by climate change, which is predicted to decrease water availability. Understanding how climate impacts these systems, in particular which variables drive plant diversity and pasture productivity, is vital for predicting climate change effects. We assessed pasture biomass and species diversity of holm-oak woodlands in a semi-arid region in Portugal, across 57 sites and monitoring a subset of ca. 13 sites over 11 years (2011–2022). Our results show that pasture biomass and diversity increased with precipitation, and that both correlated more strongly with winter-spring rainfall than with other seasons. Pasture biomass was also significantly and negatively influenced by slope. Our work suggests pasture productivity is particularly dependent on winter-spring water availability, predicting an average biomass change of 1.01 g m^{-2} per year per 1 mm of change of precipitation. As climate change may decrease precipitation in southern Mediterranean areas throughout the 21st century, these pastures will likely become less productive and diverse.

Keywords: dehesa, montado, pastures, plant diversity, *Quercus ilex*, rangelands

Introduction

Grassland ecosystems characterized by seasonal drought, as, for example, in the Mediterranean region, are highly diverse while supporting cattle grazing and other human activities (White *et al.*, 2000). However, climate change may further decrease water availability (Ferreiro-Lera *et al.*, 2025), calling for adapted management and conservation measures that rely on accurate predictions of climate change effects on ecosystems. Here, we aimed to identify the key drivers of pasture productivity and plant diversity in holm-oak woodlands. Specifically, we asked: (1) which season's precipitation is most important for annual pasture biomass accumulation and plant diversity? and (2) what is the influence of seasonal precipitation, temperature and local topography on pasture biomass? To answer these questions, we sampled 57 sites in a semi-arid region in southern Portugal, along a climatic gradient, and followed a subset of sites during 11 years. We then identified which seasonal precipitation period was related with pasture biomass and species diversity, and the role of climatic and topographic variables on the former.

Materials and methods

This study was conducted on Portuguese holm-oak (*Quercus ilex* L.) woodlands called *montado*, a savannah-like system mainly used for extensive cattle grazing. It is considered a High Nature Value Farmland, due to its high plant diversity and low-intensity management (Ferraz-de-Oliveira *et al.*, 2016). The study area has a semi-arid, mediterranean climate, with mean annual precipitation of 573 mm and temperature of 16.4°C. Here, we selected 57 homogeneous sites with similar land-use

(i.e., occupied with holm-oak woodlands with extensive grazing) but with different levels of aridity, spanning from 0.31 to 0.40 (Zomer and Trabucco, 2022). Additional details are given in Nunes *et al.* (2017).

Fieldwork was conducted once per year at the peak of standing biomass, during spring (April–June), between 2011 and 2022. Plant composition was surveyed across all 57 sites in 2012, with a subset of approx. 13 sites distributed along the aridity gradient monitored annually, resulting in 146 total observations (in 2019 and 2020 no field work was conducted). From 2014, pasture annual biomass was also sampled, yielding 117 observations from 41 sites. Species cover was estimated with the point-intercept method, with six 20 m transects. Every 50 cm, all species touching a rod placed vertically on the ground were registered. Plant diversity was calculated as Diversity of order 1 (Hill numbers; Hill, 1973). Above-ground herbaceous biomass was collected in three 30 cm quadrats placed randomly in grassland patches, oven-dried at 60°C during 48 hours and weighed.

To characterize the climate of each observation, we calculated annual and seasonal precipitation and temperature per hydrological year (September to August) between 2010/2011 and 2021/2022 from Copernicus C3S ERA5-Land monthly data at 9 km resolution (Muñoz Sabater, 2019). For topography, we retrieved slope, elevation and Potential Solar Radiation (PSR) from a digital terrain model with 25 m resolution.

We used Pearson correlation to identify which seasonal precipitation period most influenced diversity and biomass accumulation. To further explore factors driving pasture biomass accumulation, we used automatic model selection with GLMMs, using site as a random factor and precipitation, temperature, slope, elevation, PSR and soil type as predictors (numerical predictors were scaled). The final model retained the predictors from the conditional averaged model of the best fitting models, based on $\Delta\text{BIC} = 2$. Analyses were performed in *R* software (R Core Team, 2025).

Results and discussion

Pasture biomass and species diversity showed the strongest correlation with winter-spring precipitation (Table 1). Furthermore, pasture biomass accumulation at peak growth increased significantly with winter-spring precipitation and decreased with slope (Table 2).

While total annual rainfall was significant for productivity, our results show that specific seasons are more critical for these plant communities (Harrison *et al.*, 2015; Muñoz-Gómez *et al.*, 2024). This implies the rainfall regime, not just the annual total, is crucial when predicting climate change effects.

Our findings highlight winter-spring rainfall as the most critical period, which agrees with previous studies in this ecosystem (Muñoz-Gómez *et al.*, 2024) and other mediterranean-climate grasslands (Alexander *et al.*, 2023). More specifically, our model suggests that 1 mm of winter-spring precipitation corresponds to a biomass change of approx. 1.01 g m^{-2} (exponent of 0.003).

For example, in a site with an average slope of 4°, a 20% reduction in the average winter-spring precipitation, 229 mm to 183 mm, would yield a biomass reduction from 4.66 to 4.52 g m^{-2} (a 3.1 percentage points decrease in the annual production).

Table 1. Pearson correlations and associated *p*-value between pasture biomass and species diversity with precipitation accumulated over different periods.

	Pasture biomass (g m ⁻²)		Species diversity	
	<i>ρ</i>	<i>p</i> -value	<i>ρ</i>	<i>p</i> -value
Precipitation hydrological year (Sep.–Aug.)	0.316	0.001	0.160	0.053
Autumn precipitation (Oct.–Dec.)	−0.106	0.255	−0.164	0.047
Winter precipitation (Jan.–Mar.)	0.377	0.000	0.242	0.003
Spring precipitation (Apr.–Jun.)	0.342	0.000	0.304	0.000
Winter–spring precipitation (Jan.–Jun.)	0.438	0.000	0.321	0.000

Table 2. Model output for pasture biomass (unscaled predictors for a more direct interpretation), fit with Gamma error distribution with log. link.

Parameter	Est. coefficient	<i>z</i> -value	<i>p</i> -value
Intercept	4.558 ± 0.203	22.460	<2 × 10 ^{−16}
Winter–spring prec. (mm)	0.003 ± 0.001	3.913	9.13 × 10 ^{−5}
Slope (degrees)	−0.149 ± 0.065	−2.296	0.021

Trigamma $R_m^2 = 0.31$, $R_C^2 = 0.47$

Conclusion

This work sought to identify the key climatic and topographic drivers of pasture productivity and diversity in these semi-arid mediterranean woodlands. The most important factors were winter-spring precipitation and slope, highlighting the critical role of water availability during the seasons prior the peak standing biomass. Future climate change, particularly reduced rainfall in winter and spring will, very likely, impact pasture productivity and diversity in these systems.

Acknowledgements

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***Medicago intertexta* under different management strategies and intercropping systems**

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Abstract

In Mediterranean forage systems, rising climate variability calls for resilient, low-input options. *Medicago intertexta* (L.) Mill. (calvary medick), a self-reseeding annual legume, was evaluated in 2023–2024 in a semi-arid Mediterranean environment typical of inland Sicily, as a pure stand and in substitutive intercropping with *Lolium multiflorum* Lam. (Italian ryegrass; hereafter annual ryegrass) and *Trifolium alexandrinum* (berseem clover). Two utilization strategies were compared: simulated grazing and haymaking. Across treatments, haymaking increased mean dry-matter (DM) supply by 20.5% vs simulated grazing (3005 vs 2493 kg ha⁻¹). Under grazing, the legume–legume mixture *M. intertexta*+berseem achieved the highest total (3982 kg ha⁻¹), about +6.5% over the best monocrop (berseem, 3739 kg ha⁻¹). Under haymaking, the *M. intertexta*+ryegrass mixture led (3966 kg ha⁻¹). A significant resource × utilization interaction supported interpreting mixtures within management ($p = 0.022$). These results indicate that haymaking enhances overall system yield, while simulated grazing favours the legume–legume mixture, positioning *M. intertexta*-based intercrops as practical levers to buffer climate risk in Mediterranean forage systems.

Keywords: forage production, intercropping, Mediterranean forage systems, pasture regeneration, self-seeding legumes

Introduction

In Mediterranean forage systems, increasing climate variability—characterized by prolonged droughts, irregular rainfall patterns, and heatwaves—necessitates the development of more resilient, adaptable, and low-input production strategies. In this context, the use of self-reseeding annual legumes is emerging as a key approach to support pasture regeneration and improve soil fertility without relying heavily on external inputs (Bartholomew, 2014). Calvary medic (*Medicago intertexta* L.) stands out among these species due to its ability to fix atmospheric nitrogen, persist through hard seed dormancy, and thrive in the challenging conditions typical of Mediterranean environments (El-Shafey and Al-Sherif, 2020). These traits make it particularly suitable for integration into ley farming systems, where temporary pastures alternate with arable crops to enhance soil fertility and system sustainability. Despite its promising traits, the agronomic performance of calvary medic under different forage management regimes and in combination with other forage species remains underexplored. Intercropping *M. intertexta* with complementary species (e.g., annual ryegrass or berseem clover) can improve resource-use efficiency and stabilize yields. At the same time, flexible forage management—adjusting cutting frequency, stubble height, and the haymaking–grazing balance—further boosts productivity and the persistence of the legume component under different conditions (Li *et al.*, 2023).

Materials and methods

A field experiment was carried out during the 2023–2024 growing season at the “Sparacia” experimental farm of the University of Palermo (Cammarata, Sicily, Italy). The study aimed to evaluate the

Table 1. Dry matter yield (kg ha^{-1}) of *Medicago intertexta* (MI), ryegrass (R), and berseem (B) grown alone or in mixtures: ANOVA for main effects and interaction

Factor	Df1	F-value	p-value	Significance
Resource (R)	5	3.94	0.0177	*
Utilization (U)	1	3.27	0.0875	**
R $\times$ U	5	3.5	0.022	*

* $p < 0.05$; ** $0.05 \leq p < 0.10$.

Table 2. Dry matter yield (kg ha^{-1}) of *Medicago intertexta* (MI), ryegrass (R), and berseem (B) grown alone or in mixtures: Treatment means (Tukey $\alpha = 0.05$)

	<i>M. intertexta</i>	Ryegrass	Berseem	Total
<i>M. intertexta</i> (M.I.) (G)	2399 ab			2399 a
<i>M. intertexta</i> (M.I.) (H)	3653 b			3653 a
Ryegrass (R) (G)		867 a		867 b
Ryegrass (R) (H)		1699 a		1699 a
Berseem (B) (G)			3739 b	3739 a
Berseem (B) (H)			2480 ab	2480 a
MI+R (G)	939 a	763 a		1702 a
MI+R (H)	2157 ab	1809 ab		3966 a
MI+B (G)	1616 a		2366 ab	3982 a
MI+B (H)	1864 a		1681 ab	3545 a
B+R (G)		1367 a	905 a	2272 a
B+R (H)		462 a	2227 a	2689 a

Different letters indicate Tukey-adjusted differences ($\alpha = 0.05$).

forage yield, regrowth capacity, and persistence of calvary medic when cultivated both as a pure stand and in a substitutive intercropping system (alternating rows) with two widely used forage species in the Mediterranean region: annual ryegrass and berseem clover. A split-plot design with four replications was adopted. Two forage utilization strategies were evaluated: (i) simulated grazing, implemented as three cuts every 28 days to a 3-cm stubble height (G), and (ii) hay production, achieved with a single cut at flowering. This two-pronged management framework spans contrasting production scenarios—from intensive defoliation to haymaking—and enables a comprehensive assessment of species interactions, competitive dynamics, regrowth capacity, and tolerance to disturbance. Statistical analyses were performed in R (version 4.5.1; R Core Team) using linear mixed-effects models (*lme4*, *lmerTest*) and estimated marginal means/contrasts with Tukey adjustment (*emmeans*, *multcomp*).

Results and discussion

Results are presented by crop treatment and utilization strategy. Table 1 reports the ANOVA summary and Table 2 the treatment means for total dry matter.

At the plot scale, the Resource effect was significant ($F = 3.94$, $p = 0.0177$), Utilization showed a trend ($F = 3.27$, $p = 0.0875$), and the Resource $\times$ Utilization interaction was significant ($F = 3.50$, $p = 0.0220$); therefore, responses are interpreted within each utilization. Under simulated grazing (G), the legume–legume mixture MI+B achieved the highest total (3982 kg ha^{-1}), about +6.5% above the best monocrop (berseem, 3739 kg ha^{-1}), and—together with berseem—formed the upper CLD group, in line with intercrop efficiency gains reported for Mediterranean forages (Saia *et al.*, 2016). Under haymaking (H), MI+R led (3966 kg ha^{-1}), while differences among mixtures were not

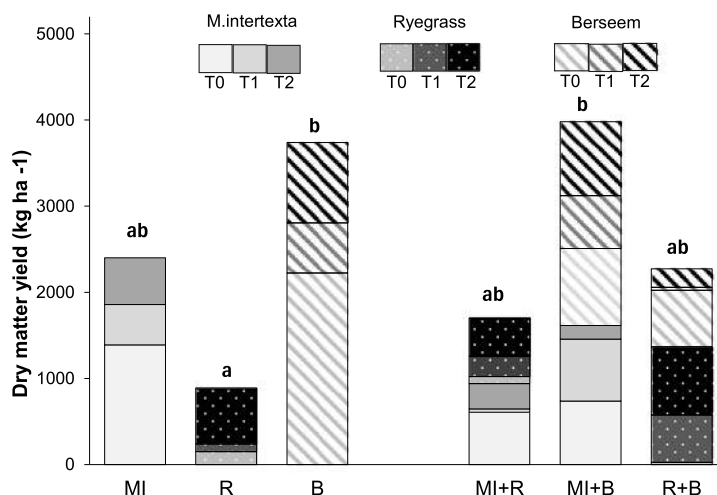


Figure 1. Dry-matter yield (kg ha^{-1}) under simulated grazing (G); stacked bars show contributions at T0, T1, and T2 for monocultures and mixtures. Patterns/colours denote species, while colour intensity within each species indicates harvest timing (T0 light, T1 medium, T2 dark).

significant, consistent with complementarity and N-related benefits in legume–ryegrass associations and their sensitivity to arrangement and seeding ratio under cut-and-carry systems (Bacchi *et al.*, 2021). Within mixtures, the only clear utilization shift was MI+R ($H > G$), whereas other pairs were not significant at $\alpha = 0.05$. For context only, unadjusted marginal means suggested higher DM under haymaking (+20.5%; 3005 vs 2493 kg ha^{-1}), but these are not interpreted inferentially due to the significant interaction (Bacchi *et al.*, 2021).

Overall, the pattern indicates species complementarity and differential sensitivity to defoliation method, with haymaking enhancing system-level yield and simulated grazing favouring the legume–legume mixture (Bacchi *et al.*, 2021; Saia *et al.*, 2016).

Berseem and the MI+B mixture stand out with the highest totals, while annual ryegrass is clearly the lowest; *M. intertexta*, MI+R, and R+B are intermediate. The stacks illustrate different harvest dynamics: T0 contributes strongly for *M. intertexta* and MI+B, berseem maintains substantial contributions across all three cuts, annual ryegrass adds only modest amounts at each cut, and the mixtures vary in how they build yield over T0–T2 (MI+R modest in every cut, R+B increasing more in the later cuts). Overall, the figure highlights contrasting patterns of yield build-up across the three harvests.

Conclusion

Yield is clearly management-dependent: under grazing, a legume–legume mixture (e.g., MI+B) is preferable, whereas under haymaking a legume–ryegrass association (e.g., MI+R) is better suited. In this context, intercropping is a practical lever to raise productivity and stability while buffering climatic variability. *Medicago intertexta*, a self-reseeding legume, emerges as a keystone species for low-input systems: integrating it into mixed pastures, ley rotations, and rotational hayfields provides a concrete route to more productive, resilient, and resource-efficient Mediterranean forage systems—provided mixtures are tailored to the intended use.

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Yield and chemical composition of six fodder tree species planted in hedgerows in temperate permanent grasslands

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Abstract

Agroforestry for forage production is a promising solution to provide additional tree-based forage during periods of drought-induced forage losses in grasslands. However, the yield and chemical composition of fodder tree leaves remain poorly studied in temperate climates. To address this, we planted fodder tree hedgerows at six sites with different pedo-climatic conditions in western Switzerland in 2021. The hedgerows included six species: *Morus alba*, *Fraxinus ornus*, *Tilia cordata*, *Tilia platyphyllos*, *Salix caprea* and *Alnus cordata*. In August 2024, we calculated basal area (BA) and estimated leaf biomass production of 20 individuals per species per site. The leaves of each species were also collected at each site, and their chemical composition was analysed. On average, *A. cordata* had the greatest leaf biomass (334 g DM for a BA of 15 cm²), followed by *F. ornus*, *S. caprea*, *M. alba*, *T. cordata* and *T. platyphyllos* (94 g DM for a BA of 15 cm²). *M. alba* had the leaves with the highest CP and Ca content and lowest fibre content whereas *A. cordata* had the highest fibre and lowest P contents. Altogether, our findings highlight the potential of tree leaves to provide additional forage sources in late summer across a range of climatic conditions.

Keywords: agroforestry, fodder hedgerows, forage production, nutritive value

Introduction

Climate change has led to increases in the frequency, intensity, and duration of droughts globally, with serious consequences for ecosystem functions such as primary production (Moravec *et al.*, 2021). In Switzerland, permanent grasslands have registered decreases in yield of up to 40% during the vegetation season under extreme drought conditions (Calanca *et al.*, 2022). Agroforestry systems, which integrate trees into permanent grasslands, represent a promising solution to provide additional tree-based forage during summer droughts as trees have better drought resistance. However, more precise and species-specific data on fodder tree productivity and nutritive values are needed before agroforestry systems can be implemented in temperate climates.

Materials and methods

Six tree species were selected to be planted in hedgerows on six farms in western Switzerland along an altitudinal gradient from 450 to 850 m asl. The species included *Morus alba* L., *Tilia cordata* Mill., *Tilia platyphyllos* Scop., *Fraxinus ornus* L., *Alnus cordata* Loisel. and *Salix caprea* L. The hedgerows were planted in the autumn of 2021. At each site, four to ten replicated multispecies hedgerows of 45 m were planted, with three lines of trees and all trees spaced 1 m apart. Multispecies hedgerows comprised five randomly arranged monospecific hedgerows of 9 m each, corresponding to five of the six selected fodder tree species (three sites with *T. platyphyllos* and three with *T. cordata*). In August

Table 1. Power regressions representing the relationship between forage yield and tree basal area for each fodder tree species in August 2024.

Species	Equation	R_m^2	R_c^2	Predicted forage yield (g DM) per basal area		
				5 cm ²	10 cm ²	15 cm ²
<i>Alnus cordata</i>	$y = 41.48x^{0.77}$	0.64	0.82	143.2	244.3	333.8
<i>Fraxinus ornus</i>	$y = 48.34x^{0.67}$	0.46	0.74	142.1	226.1	296.7
<i>Morus alba</i>	$y = 28.72x^{0.75}$	0.50	0.68	96.03	161.5	218.9
<i>Salix caprea</i>	$y = 17.43x^{0.97}$	0.68	0.92	83.0	162.7	241.1
<i>Tilia cordata</i>	$y = 22.77x^{0.59}$	0.36	0.79	58.9	88.6	112.6
<i>Tilia platyphyllos</i>	$y = 10.48x^{0.81}$	0.77	0.83	38.6	67.7	94.0

R_m^2 , marginal R -squared; R_c^2 , conditional R -squared.

2024, the leaves of five trees per species per hedgerow, on four hedgerows per site were counted (i.e., 20 individuals per species per site). Additionally, the dry matter (DM) of 50 leaves per species per site was determined (after drying at 60°C for 72 hours and weighing). The average leaf biomass of each species was then multiplied by the total number of leaves to estimate total leaf biomass production (i.e., forage DM yield). Trees were 2–3 years old at the time of forage yield estimation. Forage yield for *M. alba* was only analysed for the three sites below 600 m asl, as above this altitude this species did not produce leaves due to recurrent late frosts in spring. The basal diameter of all sampled trees (including all trunks in case of multi-trunk individuals) was also measured in August 2024 and basal area was calculated for each tree. Leaves of each species were also collected at each site in August, dried and ground to analyse their chemical composition. Nitrogen (N) content was analysed using the Dumas method (ISO 16634–1:2008), and crude protein (CP) calculated as $N \times 6.25$. Phosphorus (P) and calcium (Ca) were determined via ICP-OES after microwave digestion. Fibre fractions—including neutral detergent fibres (aNDFom), acid detergent fibres (ADFom) and acid detergent lignin (ADL)—were analysed gravimetrically according to Van Soest *et al.* (1991) with a heat-stable amylase, and sodium sulphite. The fibre values were expressed without residual ash. We ran power regressions between forage DM yield and tree basal area and their significance was tested using linear mixed-effect models with ‘hedgerow’ nested within ‘site’ as a random factor. The effect of tree species on leaf chemical composition was analysed using linear mixed-effect models with ‘site’ as a random factor.

Results and discussion

Tree basal area was a good predictor of forage yield for all species across sites (R_c^2 ranging from 0.68 to 0.92 for the six species, Table 1). Forage yield differed among species. *Alnus cordata* had the highest forage yield per 15 cm² basal area (334 g DM), followed by *F. ornus* (297 g DM), *S. caprea* (241 g DM), *M. alba* (219 g DM), *T. cordata* (113 g DM) and *T. platyphyllos* (94 g DM).

The forage chemical composition significantly differed among tree species, likely due to different plant growth strategies (Table 2). CP concentrations were the lowest in *F. ornus* (125 g (kg DM)^{−1}) and the highest in *M. alba* (236 g (kg DM)^{−1}). All species except *F. ornus* had CP values above 140 g (kg DM)^{−1}, which is the target value in most Swiss gramineous-dominated grasslands (>70% grasses; Agroscope, 2017). Lignin concentration, which influences digestibility, was the highest in *A. cordata* (235 g (kg DM)^{−1}) and the lowest in *M. alba* (24 g (kg DM)^{−1}). Accordingly, the highest aNDFom and ADFom concentrations were found in *A. cordata* and the lowest in *M. alba*. Both aNDFom and ADFom concentrations were all lower than those generally found in gramineous-dominated grassland (469 and 260 g (kg DM)^{−1}, respectively; Agroscope, 2017), except for the ADFom value of *A. cordata*

Table 2. Average chemical composition ($\pm$ SE) (in g (kg DM)⁻¹) of the leaves of the six fodder tree species in August 2024.

Species	CP	aNDFom	ADFom	ADL	P	Ca
<i>Alnus cordata</i>	162 $\pm$ 11 ^B	443 $\pm$ 9 ^A	459.4 $\pm$ 6.6 ^A	235 $\pm$ 7 ^A	1.6 $\pm$ 0.1 ^F	10.8 $\pm$ 0.9 ^C
<i>Fraxinus ornus</i>	125 $\pm$ 4 ^C	315 $\pm$ 8 ^B	238.2 $\pm$ 6.1 ^B	56 $\pm$ 4 ^C	1.9 $\pm$ 0.1 ^E	15.6 $\pm$ 1.4 ^B
<i>Morus alba</i>	236 $\pm$ 17 ^A	185 $\pm$ 4 ^D	121.3 $\pm$ 3.2 ^E	24 $\pm$ 2 ^D	4.8 $\pm$ 0.2 ^A	23.6 $\pm$ 1.6 ^A
<i>Salix caprea</i>	166 $\pm$ 8 ^B	284 $\pm$ 12 ^C	197.5 $\pm$ 10.3 ^C	102 $\pm$ 7 ^B	3.0 $\pm$ 0.2 ^C	8.9 $\pm$ 0.58 ^C
<i>Tilia cordata</i>	154 $\pm$ 7 ^B	289 $\pm$ 9 ^C	172.3 $\pm$ 11.9 ^D	77 $\pm$ 13 ^C	2.3 $\pm$ 0.1 ^D	21.2 $\pm$ 1.6 ^A
<i>Tilia platyphyllos</i>	161 $\pm$ 1 ^B	290 $\pm$ 5 ^{BC}	196.7 $\pm$ 10.8 ^{CD}	62 $\pm$ 4 ^C	3.7 $\pm$ 0.1 ^B	22.2 $\pm$ 1.9 ^A

CP, crude protein; aNDFom, neutral detergent fibre; ADFom, acid detergent fibre; ADL, acid detergent lignin; P, phosphorus; Ca, calcium. Different letters indicate significant ($P < 0.05$) differences among species.

(459 g (kg DM)⁻¹). The highest P and Ca concentrations were found in *M. alba* (4.8 g P (kg DM)⁻¹, and 23.6 g Ca (kg DM)⁻¹), whereas the lowest P content was found in *A. cordata* (1.6 g P (kg DM)⁻¹). All species had Ca concentrations above the average values for gramineous-dominated grasslands (6.6 g (kg DM)⁻¹; Agroscope, 2017). Previous studies that analysed leaf chemical composition for the same species found a wide range of values, likely because leaves were collected at varying times of year, sites and stages of maturity (Papachristou & Nastis, 1996; Emile *et al.*, 2016; Novak *et al.*, 2020; Galland *et al.*, 2025). Nonetheless the concentrations of chemical compounds recorded in our study fall within the range of values previously recorded for the six species, except for aNDFom, which was relatively lower than those of other studies. Overall, *M. alba* had the highest CP, P and Ca content and the lowest fibre contents.

Conclusion

All six tree species show potential to provide supplementary forage in late summer when herbage yields might be negatively impacted by drought. *Morus alba* had the leaves with the highest CP and lowest fibre content but did not produce leaves above an altitude of 600 m, whereas *A. cordata* had the highest fibre and lowest CP contents. Trade-offs existed between nutritive value and productivity, as the species with the lowest nutritive value, *A. cordata*, was also the most productive tree species. Overall, *S. caprea* was the species with the best compromise between forage yield and nutrient content. Our study is one of the first to characterize the forage yield and chemical composition for these six fodder tree species across varying temperate pedoclimatic conditions. Continued measurements are planned in the coming years to understand how fodder tree production will evolve.

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Restoring ecosystem services through grazing management in Portuguese agro-silvo-pastoral systems

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Abstract

Montados, multifunctional agro-silvo-pastoral systems predominant in drylands of Portugal, provide essential ecosystem services (ES). While traditional extensive grazing supports their savannah-like structure and biodiversity, overgrazing causes degradation, reducing tree regeneration, soil compaction, and simplifying vegetation structure. This study assessed the effectiveness of grazing exclusion as a passive restoration strategy to enhance ES, considering the influence of grazing intensity (GI), tree cover density (TCD), and potential solar radiation (PSR). An experiment in a semi-arid region in Portugal assessed three ES across 40 plots—20 excluded from grazing (>5 years) and 20 under continued grazing—distributed along gradients of GI, PSR, and TCD. Grazing exclusion promoted forage availability, oak regeneration, shrub cover, and reduced bare soil, enhancing multiple ES. Higher TCD was associated with higher acorn production and oak regeneration. Lower PSR areas supported greater herbaceous diversity and oak regeneration, promoting habitat and biodiversity maintenance. High-GI under high-PSR intensified negative impacts, highlighting the importance of local environmental conditions. Our findings confirm that grazing exclusion (>5 years) is an effective passive restoration measure, and its strategic placement in low-PSR and high-TCD areas can accelerate ES recovery, while targeting harsher zones can prevent long-term degradation but may require more time and/or active restoration to accelerate recovery.

Keywords: agroforestry, ecosystem services, grazing management, land degradation, passive restoration

Introduction

Pastures are the dominant land use in drylands (Maestre *et al.*, 2022) and a key component of Montado systems, where traditional extensive grazing supports ecosystem structure and diversity. However, overgrazing can lead to degradation and negatively affect key ecosystem services (ES) related to tree regeneration, soil structure, and vegetation diversity (Maestre *et al.*, 2022). Therefore, implementing effective grazing management is essential to support the resilience of Montado systems. Implementing grazing exclusion areas in the landscape is a recognized passive restoration measure to enhance oak regeneration and ES delivery (Köbel *et al.*, 2021). In addition to grazing intensity (GI), local environmental factors such as Potential Solar Radiation (PSR) and Tree Cover Density (TCD) also influence ecosystem dynamics and ES provision (Príncipe *et al.*, 2014; Maestre *et al.*, 2022). Therefore, in this study, we aim to identify optimal locations for grazing exclusion areas as a passive restoration measure to maximize the supply of three ES—forage production, habitat and biodiversity maintenance, and ultimately, soil erosion—in the Montado system, considering the influence of GI, TCD, and PSR.

Materials and methods

A restoration test was implemented at Herdade da Coitadinha, Portugal, to assess ecosystem service (ES) delivery, considering grazing (GI), TCD, and PSR. GI was estimated using cattle heads $\text{ha}^{-1} \text{yr}^{-1}$ from 2022, and dung counts in the field (Maestre *et al.*, 2022). PSR, mapped via Area Solar Radiation (Spatial Analyst) in ArcGIS Pro, served as a proxy for microclimatic conditions (Principe *et al.* 2014), with higher PSR associated with south-facing and more exposed slopes, with higher temperatures and lower soil humidity. TCD was derived from High Resolution Layers—Tree Cover Density product (Copernicus Land Monitoring Service, 2023).

Plots were selected based on PSR (low <40th, high >60th percentile), TCD (low < 40th, high > 60th percentile), and GI (excluded for ≥ 5 years vs. grazed). This design resulted in 8 treatments with 5 replicates each (40 plots). In the spring of 2023, ES indicators were measured in all plots (45 × 45 m), excepting acorn sampling, that was measured in November 2023, when acorns were maturing. Sampling followed commonly used methods: (i) bare soil cover, herbaceous and shrub abundance and diversity were estimated using the “point and line-intercept” methods, respectively, with data collection along three 20-metre transects per plot; (ii) herbaceous biomass was collected using three 0.3 × 0.3 m quadrats placed near each vegetation transect within the plots; (iii) natural regeneration of Holm oak was quantified by counting regenerating individuals within 20×2 m belts; (iv) acorn biomass was estimated by counting acorns on two opposite sides of the canopy of up to five Holm oak trees per plot, as well as on the ground beneath those sides using 1 m² quadrats. In total, indicators were classified across 3 ES: forage production, biodiversity and habitat maintenance, and erosion control (CICES; Haines-Young and Potschin, 2018). Generalized Linear Models (GLMs) were built using R software (v4.4.1, R. Core Team, 2025) to test the effects of PSR, GI, and TCD (predictors) on each indicator (response variables).

Results and discussion

Our results revealed distinct effects of environmental gradients and GI on key ES. Areas with higher TCD showed increased acorn biomass and Holm oak regeneration (Table 1), suggesting that denser tree cover enhance conditions for propagule production—a key source of regeneration and forage for livestock, such as the Iberian pig—and for oak seedling establishment. Conversely, in areas with lower TCD, higher herbaceous biomass was observed likely due to lower shading effect.

PSR negatively influenced regulation and maintenance services. Areas with lower PSR supported greater herbaceous diversity and higher oak regeneration (Table 1), suggesting that reduced solar exposition may diminish the impacts of drought stress by creating more favourable microclimatic conditions (Principe *et al.*, 2014)—such as lower soil evaporation and higher moisture retention.

Grazing consistently reduced both structural and taxonomic diversity, negatively affecting all evaluated ES, in line with findings by Maestre *et al.* (2022). Areas with higher GI showed lower herbaceous and shrub abundance, reduced shrub diversity, and increased bare soil cover, reflecting degradation associated with overgrazing (Table 1). These findings reinforce that implementing long-term (≥ 5 years) grazing exclusion areas, scattered within grazed landscapes, is an effective passive restoration strategy to enhance oak natural regeneration and the delivery of ecosystem services in Montado systems (Köbel *et al.*, 2021). Incorporating these areas within rotational grazing systems may prevent resource depletion while maintaining sustainable productive uses.

Table 1. Results of Generalized Linear Models (GLM) for the indicators and related Ecosystem Services.

	ES	Response variable	Mean; max; min value	TCD	PSR	GI	PSR*GI	<i>R</i> ² adj
Provisioning services	Forage production	Acorn biomass (No. of individuals × biomass of acorns)	1739.4; 5009.4; 0.00 (g m ^{−2})	755.108 (211.56)*	n.s.	n.s.	−797.044 (0.0242) *	0.40
		Herbaceous biomass	78.66; 344.69; 14.11 (g m ^{−2})	−0.294 (0.885) *	n.s.	−0.203 (0.085) *	−0.324 (0.111)*	0.43
Regulation and maintenance services	Habitat and biodiversity maintenance	Natural regeneration	12; 67; 0 (individuals)	0.961 (0.199)*	−0.906 (0.184)*	−0.506 (0.219)*	n.s.	0.57
		Diversity of herbaceous (Shannon index)	1.98; 2.92; 0.44	n.s.	−0.393 (0.078) *	n.s.	n.s.	0.50
		Abundance of shrubs (%)	21.34; 97.83; 0	n.s.	n.s.	−1.027 (0.135) *	n.s.	0.52
		Diversity of shrubs (Shannon index)	0.66; 1.40; 0	n.s.	n.s.	−0.171 (0.043) *	n.s.	0.34
	Erosion control	100-Bare soil cover	79.65; 99.19; 36.56 (%)	n.s.	n.s.	−0.1612 (0.026) *	n.s.	0.47

Significant values ($p < 0.05$) are represented with *, non-significant values are represented by n.s., and tabulated alongside respective model estimates and standard error (in parentheses).

A significant interaction between PSR and GI was observed for provisioning services (Table 1). Acorn and herbaceous biomass decreased more strongly under high GI in high PSR areas, suggesting that grazing in areas with high solar radiation magnifies the reduction of provisioning services, which will be more difficult to recover. These findings align with Maestre et al. (2022), who suggested that grazing pressure may interact with climate-related stressors to reduce ES delivery in warmer drylands. Overall, ES were enhanced at grazing exclusion areas, lower PSR, and higher TCD.

Conclusion

Grazing exclusion (>5 years) is an effective passive restoration strategy that enhances multiple ES, including forage provision, biodiversity and habitat maintenance, and soil protection. Its effectiveness also depends on local environmental factors: areas with higher TCD and lower PSR show greater oak regeneration and herbaceous diversity and should be prioritized in ecological restoration efforts aiming at faster ES recovery. In contrast, exclusion areas in harsher environmental conditions (high PSR, low TCD) help prevent further degradation and secure future agricultural use but may require more time and/or active restoration measures to accelerate recovery. Moreover, high GI under higher solar exposure intensifies negative impacts, highlighting the need for careful planning. Therefore, selecting exclusion areas strategically—whether to maximize short-term ES recovery or to prevent long-term degradation—is key to successful restore Montado systems.

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Integrated approach for use and conservation of Apenninic dry grasslands: a model from LIFE “IMAGINE”

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Abstract

Semi-natural dry grasslands are crucial reservoirs of plant biodiversity, recognized under the EU Directive 92/43/EEC. However, they are increasingly threatened by the abandonment of traditional agro-pastoral practices, particularly in the mountainous regions of the central Apennines (Italy). As part of the LIFE Integrated Project “IMAGINE” (LIFE19 IPE/IT/000015), we collected and analysed grassland vegetation relevés from central Italy, focusing on Annex I habitats. Uni- and multivariate analyses identified major vegetation types, indicator species, and key ecological drivers. Pastoral value was calculated to determine their sustainable carrying capacity, guiding the design of rotational Grazing Plans. A Web-GIS tool, integrating Sentinel-2 imagery and EVI-based harmonic modelling, enabled near real-time monitoring of biomass dynamics to support farmer-led, adaptive management. Quality assessments of dairy and meat products were also conducted. This study identified distinct Apenninic dry grassland types with specific management needs, guiding the implementation of site-specific grazing regimes and concrete conservation actions.

Keywords: conservation, Habitats Directive, management, semi-natural grasslands

Introduction

European semi-natural dry grasslands (SNDG) are among the most species-rich plant communities worldwide, and they are fully dependent on regular, low-intensity land-use practices (Biurrun *et al.*, 2021). Many of these grasslands correspond to habitats of community interest protected under Annex I of the Habitats Directive (92/43/EEC), the primary legislative framework for biodiversity conservation in Europe. However, SNDG are among the most threatened habitat types in Europe, with a deteriorating trend (EEA, 2020).

In the central Apennines (Italy), the principal threat to SNDG conservation is the gradual abandonment of traditional agro-pastoral activities, such as extensive grazing and mowing, which has resulted in a decline in SNDG extent, alterations in species composition, and a decrease in overall species richness (Giarrizzo *et al.*, 2017; Malatesta *et al.*, 2019). In this context, as part of the LIFE Integrated Project “IMAGINE” (LIFE19 IPE/IT/000015) in the Umbria region, we developed an integrated approach to manage and preserve the SNDG. This strategy combines the study of plant biodiversity, including floristic-vegetational and ecological diversity of various SNDG types; the assessment of animal husbandry with a focus on the qualitative and quantitative knowledge of forage resources; and geo-informatics, specifically the creation of a Web-GIS app as management support for monitoring the vegetation growth.

Materials and methods

The study area includes the SNDG of Umbria region at an altitudinal range between 380 and 1880 m asl, covering about 450 km². Their botanical composition was investigated through 623 4 × 4 m² phytosociological relevés (Braun-Blanquet method), both collected from previous bibliographic sources and by an in-field campaign of vegetation surveys in 2021–2024. A hierarchical divisive cluster analysis was performed using the modified TWINSpan method (Roleček *et al.*, 2009). Diagnostic, constant and dominant plant species were identified for each grassland type. To define the ecological characteristics of the surveyed vegetation, Ellenberg Indicator Values (EIVs), adapted to the Italian flora, and environmental characteristics (elevation, aspect, slope, bedrock type and bioclimatic indices) were analysed by means of descriptive statistics. In order to define the optimal stocking rate of grazing animals for each grassland type, we calculated the Pastoral Value (PV), applying the method based on the visual estimation of the degree of coverage (Bagella, 2001), an adaptation from Daget & Poissonet (1972). At the same time, herbaceous biomass samples were collected and dried in a 40° ventilated oven for 24 hours and then analysed to determine dry matter (DM), ether extract (EE), crude protein (CP) according to AOAC (1995). Neutral detergent fibre (NDF), acid detergent fibre (ADF) and acid detergent lignin (ADL) were determined according to Van Soest *et al.* (1991), hemicellulose and cellulose were calculated by difference between NDF and ADL and between ADF and lignin, respectively. Non-fibre carbohydrates were calculated by difference between DM content and the sum of EE, CP, Ash and NDF. For the development of the Web-GIS app, we used Sentinel-2 satellite imagery of pixels classified as grassland under category 30 in the ESAWorld Cover 2021, filtered by cloud coverage (<20%) and by Scene Classification Map band. The Enhanced Vegetation Index (EVI) modelled with two harmonics in accordance with vegetative cycles (spring-summer and autumn) was calculated with a 10 m spatial resolution.

Results and discussion

The vegetation analysis allowed us to identify 17 types, and 8 distinct macrotypes (A–H), of SNDG, each with its own peculiar floristic-vegetational and ecological characteristics, and corresponding to habitats 6210*, 6230* and 6170 of Annex I to the 92/43/EEC Directive.

The SNDG macrotypes correspond to the following: *Nardus stricta*-dominated acidophilous montane grasslands (A), *Bromopsis erecta*/*Brachypodium genuense*-dominated mesophilous montane grasslands (B), *B. erecta*-dominated mesophilous hilly grasslands on marly and sandy substrata (C), *Sesleria junceifolia*-dominated, edapho-xerophilous, montane stripped grasslands (D), *Stipa dasyvaginata* subsp. *apennincola*/*B. erecta*-dominated thermo-xerophilous hilly grasslands (E), *B. erecta*/*Festuca inops*-dominated thermophilous hilly grasslands (F), *B. erecta*-dominated xerophilous submediterranean submontane grasslands (G), and *B. erecta*/*Festuca trachyphilla*-dominated montane grasslands (H). Species richness shows a great variability between groups, spanning 8 to 73 species per relevé, with the highest mean value (44) for macrotype B. Results showed the spread of *Brachypodium* species, known to cause a reduction of biodiversity and habitat quality in abandoned grasslands (Bonanomi and Allegranza 2004); in addition, the occurrence of shrubs and edge species (e.g. *Asphodelus macrocarpus*), highlights the ongoing successional processes affecting the pastures. In contrast, in localized areas, the spread of typical species from overgrazed pastures, together with few nitrophilous taxa, underlines phenomena of intense ongoing grazing pressure due to poor or incorrect pasture management.

The PV varies from 0.7 to 39.0, with the macrotype C showing the highest mean value (27.3) while the *Nardus*-dominated stands the lowest (9.3). Based on the site-specific characterization of SNDG,

Grazing and Action Plans aimed at counteracting the detected degradation phenomena were drafted. Grazing Plans included the evaluation of the maximum maintainable and maximum recommended stocking rates, the identification of areas to be excluded from grazing, and the design of rotational grazing systems for a conservation-oriented use of SNDG.

Finally, we developed a Web-GIS app called “Praterie”, accessible from <https://lifeimagine-webgis.users.earthengine.app/view/desktopwebgis>. The map layers of the app show the days remaining until the expected start of the next vegetative maturity period (VMP) and the maximum average greenness (MAG), both calculated by a harmonic model of the previous three-year EVI (Enhanced Vegetation Index) data. In this context the VMP is intended as the temporal window when grassland vegetation is expected to reach its maximum greenness, which is considered a proxy for peak biomass and optimal forage quality. The MAG serves as a predictive proxy map of grassland productivity and potential forage availability. Another layer shows the observed average EVI of the previous 10 days. Users can also build a vegetation growth EVI-based chart for a location of interest showing the observed vegetation growth (red line) and the estimated vegetation growth (green line) obtained through the application of the harmonic model. Operationally, decision-making could be enhanced by using the forecasted vegetative maturity period to better plan grazing activities to align with the period of highest expected forage availability and quality, the maximum greenness to plan the appropriate grazing load based on expected productivity, and the observed greenness to make timely, tactical adjustments based on the actual, current state of the vegetation canopy. While the map layers provide a strategic overview of what should be happening, the growth graph offers a diagnostic tool to see what is actually happening, potentially enabling more precise and timely decision-making.

Conclusion

A comprehensive approach that includes the study of the biodiversity of SNDG, an understanding of their forage resources, and the development of appropriate technological tools is essential for implementing effective grassland management practices and promoting biodiversity conservation. Our study proposes an integrated vegetational, pastoral, and geospatial approach aimed at strengthening the resilience and conservation of these semi-natural grasslands. The web-GIS tool contributes to optimizing livestock management and biodiversity conservation by providing timely and accurate data about grassland status and growth curves. Based on the first achieved results, in collaboration with the Regional Forest Agency, concrete actions of shrubs and edge species removal, both manual and mechanical, have been already implemented to contrast the secondary succession phenomena.

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Beyond grazing pressure: stocking method and grazing intensity jointly affect grassland plant diversity

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Abstract

Although typically managed intensively, grazed permanent grasslands in dairy systems can support high plant diversity, depending on the management context. While numerous studies have examined the effects of grazing intensity (GI) and stocking method (SM) separately, their interactions are often overlooked. This study tests whether the effects of GI and SM on different diversity measures interact, depending on the regional context. We surveyed 68 grazed grassland sites on dairy farms across Germany. Plant diversity was assessed using species richness (SR) and species evenness (Simpson Equitability, EQ). We analysed the effects of GI (livestock unit grazing days, LUGD), SM (rotational vs. continuous), and region (RG, North vs. South), as well as their interactions. The SR declined with increasing GI, independent of RG and SM. In contrast, species evenness responded more complexly: the effect of GI on EQ was significantly affected by SM and RG. These findings highlight that plant diversity is shaped by complex mechanisms and that considering just one of the factors might not be sufficient.

Keywords: CSR, grazing intensity, phytodiversity, stocking method

Introduction

Grazed permanent grasslands hold considerable potential for supporting plant diversity. In dairy production, these pastures are typically managed intensively. Yet, their contribution to phytodiversity depends on various factors such as climate, soil conditions, topography or grazing management (Gaujour *et al.*, 2012). Among the most important and adjustable management factors are grazing intensity (GI) and stocking method (SM). While the effects of these two factors have often been studied in isolation (e.g. Herrero-Jáuregui & Osterheld, 2018; Pavlů *et al.*, 2003), their combined effects on plant diversity are usually overlooked, especially under real-farm conditions. Moreover, the regional context may further modify these relationships due to underlying differences in climate and vegetation. We therefore hypothesize that the effect of GI on the two diversity components plant species richness and evenness on dairy farm pastures is dependent on the SM and the regional context.

Materials and methods

We selected 68 permanent grassland sites from 34 grass-based dairy farms across different regions of Germany. The sites, exclusively used for grazing with occasional mulching, represented each farm's specific grazing practices: 58% were grazed by lactating dairy cows and 42% by other cattle (e.g. heifers, calves) typical for a dairy system. We applied hierarchical clustering based on the long-term annual means of temperature, precipitation and global horizontal irradiation to classify the sites into the two regional clusters 'North' ($n = 35$) and 'South' ($n = 33$) that reflect natural differences in the vegetation along a climatic gradient. Management data for 2021–2023 were compiled from interviews

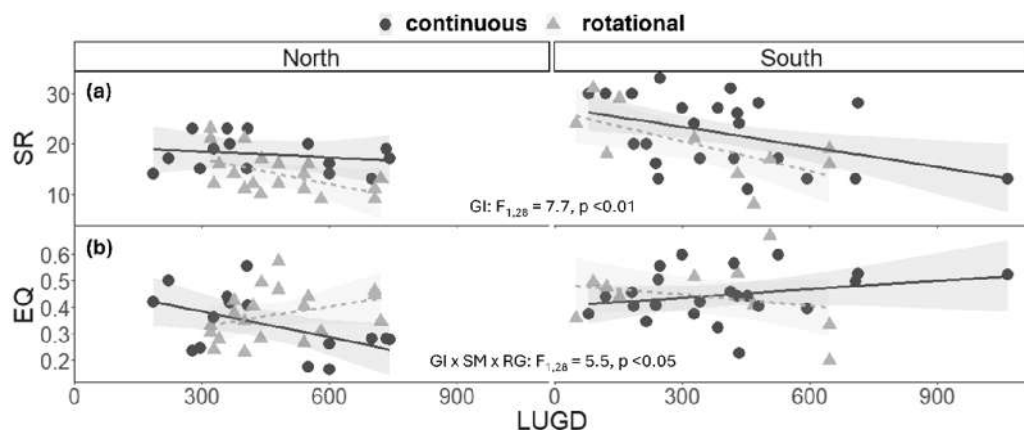


Figure 1. Relationships between GI (grazing intensity) and (a) SR (species richness), and (b) EQ (Simpson equitability), corresponding to the interaction with RG (region) and SM (stocking method). Different point and line shapes represent SM. *F*- and *p*-values are shown for the main effect of GI on SR and the interaction effect of GI $\times$ SM $\times$ RG on EQ.

with the farmers. The SM was classified as either rotational or continuous grazing for each site (Allen *et al.*, 2011). The GI was measured as livestock unit grazing days (LUGD) to standardise comparisons across different livestock groups and grazing durations (1 LU = 500 kg live weight), calculated as follows:

$$\text{LUGD} = \frac{\text{livestock units} * (\text{grazing hours perday}/24) * \text{grazing days per year}}{\text{hectares}}$$

On-site botanical surveys were conducted during 2023 and 2024 within a representative 3 m $\times$ 3 m plot per site, where all vascular plant species were recorded and their dry matter (DM) yield proportions visually estimated (Voigtländer & Voss, 1979). Species richness (SR) quantifies the total species number. Simpson Equitability (EQ), as a measure of species evenness, was calculated by dividing the Gini-Simpson-Index by the SR (Magurran, 2003). For a basic understanding of the ecological context, we classified all species into plant functional types C (competitor), S (stress-tolerant) and R (ruderal), and derived a functional signature for each site based on yield proportions (Grime *et al.*, 1988). For analyses of covariance, linear mixed-effects models were computed in R studio to test main and interaction effects of RG, SM and GI on SR and EQ, including the individual farms as random effect. Pearson's product-moment correlation tests were used to test relationships between functional signature components CSR vs. SI, SR and EQ.

Results and discussion

We found a significant negative main effect of GI on SR, without interactions of SM or RG (Figure 1a). The EQ was affected by the interaction of GI $\times$ SM $\times$ RG (Figure 1b): in the North, EQ decreased significantly under rising GI on continuously stocked and increased on rotationally stocked pastures. Moreover, with increasing GI under continuous stocking, EQ increased significantly in the South and decreased in the North.

An increase in GI was correlated with all three indices of the sites' functional signatures (C: $r = 0.3$, $p < 0.05$; S: $r = -0.5$, $p < 0.0001$; R: $r = 0.3$, $p < 0.05$). Moreover, a higher SR was correlated with

significantly higher S- ($r = 0.6, p < 0.0001$) and lower C-components ($r = -0.5, p < 0.0001$), whereas we found no significant correlations between EQ and any component of the CSR signature.

Despite an output-oriented grassland management, mean plant diversity was relatively high, likely supported by certain characteristics of the studied farms, such as low to medium soil phosphorus contents (mean: 0.06 g kg^{-1}) and nitrogen fertilization (mean: $50 \text{ kg ha}^{-1} \text{ year}^{-1}$), reduced concentrate feeding (mean: 83 g kg^{-1} energy-corrected milk), and diversified grassland use as adapted to the needs of different livestock groups. The observed shift from stress-tolerant towards competitive and ruderal species under high GI reflects a functional adaptation of plant communities to nutrient-enrichment and disturbance and corresponds to the observed linear decline in SR (Grime *et al.*, 1988; Marini *et al.*, 2007). In contrast to the species number, species evenness responded in a more complex way, confirming our hypothesis for EQ but not for SR. The EQ was influenced by the interaction of GI and SM, which was further dependent on the regional context, but showed no significant response to the GI-induced change in functional composition. Other research confirms that the mechanisms controlling species composition are more difficult to understand than the mere species number (Klimek *et al.*, 2007). While EQ was already relatively high in the South, GI and SM seemed to have a larger impact in the North, where topographic and pedoclimatic conditions were more uniform.

Conclusion

While high grazing intensity generally reduced the plant species numbers of dairy pastures, the species evenness was shaped by complex interactions between grazing intensity, stocking method and regional context. Both species number and evenness are essential components of plant diversity. Our findings emphasize the importance of considering both grazing intensity and stocking method when evaluating management systems for an effective conservation of biodiverse grassland.

Acknowledgement

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Grazing effects on soil compaction in Mediterranean oak woodlands

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Abstract

This study evaluates the impact of grazing on soil compaction in two Mediterranean oak woodlands (*Quercus rotundifolia* (Qr) and *Quercus pyrenaica* (Qp)) in the Montesinho Natural Park in north-eastern Portugal. In each habitat, paired fenced plots (grazed versus ungrazed) were monitored. Soil samples were taken at depths of 0 to 10 cm, 10 to 20 cm and 20 to 30 cm to evaluate penetration resistance and moisture content. The Qr plot was grazed by goats at a moderate intensity for 15 consecutive days. Meanwhile, the Qp plot was grazed by sheep at a higher intensity for 55 days, intermittently over an extended period. Soil resistance was determined using an impact penetrometer and, along with soil moisture, analysed using two-way ANOVA (SS Type III). The results showed that grazing significantly increased soil compaction in the Qr plots, but not in the Qp plots. Penetration resistance was negatively correlated with moisture content. Soil compaction varies with moisture, with drier soils harder to penetrate. These findings demonstrate that pre-existing soil properties modulate the effects of grazing on soil compaction, underscoring the importance of site-specific conditions in the sustainable management of Mediterranean silvopastoral systems.

Keywords: penetration resistance, silvopastoral systems, soil physical properties

Introduction

Soil compaction affects root growth, water infiltration, and soil quality, with high values indicating limited aeration and restricted root development. Recognised as a priority in soil protection in Europe, its magnitude is typically quantified by penetration resistance (Virto *et al.*, 2015). Among the aggravating factors, grazing management plays a central role: intensive trampling can increase compaction and degrade the physical structure of the soil (Virto *et al.* 2015). Penetration resistance, measured with penetrometers, is a practical and reliable method for diagnosing compaction and comparing management practices, as demonstrated by several studies (Serrano *et al.*, 2023). This study assesses the impact of grazing on soil compaction and moisture in two Mediterranean oak forests (*Quercus rotundifolia* Lam. and *Quercus pyrenaica* Willd.) in the Montesinho Natural Park, in north-eastern Portugal.

Materials and methods

The study was conducted in Montesinho Natural Park, north-eastern Portugal (41°53.59'N, 6°52.8'W), in natural stands of *Q. pyrenaica* and *Q. rotundifolia*, without silvicultural management and subject to occasional extensive grazing. The area, located at an altitude of 985 m, is part of the supramediterranean bioclimatic zone, with an average annual temperature of 8–12°C and average precipitation of 772.7 mm (1981–2010; IPMA). The study was conducted at two experimental sites, each with two fenced plots of 1.3 ha. The first site was dominated by *Q. rotundifolia* (965 trees ha⁻¹) and with a dense understorey of *Genista falcata* Brot., *Pterospartum tridentatum* (L.) Willk., *Cistus*

salviifolius L., *Halimium lasianthum* subsp. *alyssoides*, *Cytisus multiflorus* (L'Hér.), and *Erica australis* L., as well as a sparse herbaceous layer of annual grasses. The soil, a Leptosol derived from basic rocks, poorly developed and shallow (horizon A: 10–30 cm), has a sandy loam texture (11.15% clay), pH 5.3, high stoniness and horizon C on rock at <50 cm. In July 2024, one of the areas, 25 Preta de Montesinho goats (45–50 kg), in the last third of gestation, grazed for 15 consecutive days (approx. 15 hours per day), were collected at night and supplemented with 25 kg/day of hay and 5 kg/day of concentrate (CP 18.4%). The second site, dominated by *Q. pyrenaica* Willd. (1545 trees ha⁻¹) and undergrowth of *Cytisus striatus* (Hill) Rothm. and *Rubus ulmifolius* Schott, had Leptossolo Úmbrico with a sandy loam humic horizon (8.91% clay), pH 5.1, frequent stoniness and a stony horizon at 10–50 cm. Between May and October 2024, one of the areas, 220 Churra Galega Bragançana Branca sheep (50–60 kg) grazed for 55 days intermittently (approximately 17 hours per day), characterising high-intensity, non-consecutive grazing. Penetration resistance was determined in the field with an impact penetrometer, under conditions close to field capacity and after grazing, according to Stolf *et al.* (1983). Measurements were taken to a depth of 60 cm, with 25 repetitions per treatment (species and management). The procedure consisted of repeatedly dropping a 4 kg weight from a height of 40 cm until it reached the maximum depth or until the rod could no longer penetrate the soil, recording the number of impacts (blows) required to drive the rod through each centimetre of soil, serving as an indicator of soil resistance (Equation (1)).

Soil resistance was calculated using Equation (1):

$$F = 5.6 + 6.89 \times N \quad (1)$$

where F is the soil resistance (kgf cm⁻² × 0.098 = MPa), N is the number of impacts ÷ deepening (cm).

Moisture content was measured at depths of 0–10, 10–20, and 20–30 cm, simultaneously with penetration resistance, using an SM150T capacitive sensor. The samples, collected randomly in both areas, allowed the soil conditions to be characterised, with data up to 30 cm being used to compose the resistance and moisture profiles. Statistical analysis was conducted using two-way ANOVA (SS Type III), considering habitat and grazing as factors, followed by Tukey's test ($p < 0.05$) for paired comparisons. Analyses were performed using SYSTAT® 13.2 software.

Results and discussion

Penetration resistance varied significantly between habitats at all depths, with significant interaction between habitat and grazing. *Quercus rotundifolia* showed higher values under grazing, especially at 0–10 cm (3.03 ± 0.23 MPa), while *Q. pyrenaica* under grazing recorded the lowest values at 20–30 cm (5.14 ± 0.39 MPa) (Table 1).

The edaphic and functional contrasts between *Q. rotundifolia* and *Q. pyrenaica* generate differences in infiltration, water storage, and resistance to trampling, resulting in distinct compaction patterns, even under similar grazing conditions (Rapp *et al.*, 1999). *Q. rotundifolia* grows in shallow, stony Leptosols derived from basic rocks. These soils are characterised by rapid drainage, low to moderate organic matter content and neutral to alkaline pH (Alonso-Forn *et al.*, 2020), which translates into greater susceptibility to increased bulk density and mechanical resistance under the effect of grazing. In contrast, *Q. pyrenaica*, typically found in more favourable soil conditions, particularly in Umbric Leptosols, is characterised by a dark, humic A horizon, sandy loam texture and moderately acidic pH (Calzolari

Table 1. Soil penetration resistance and moisture content by depth in grazed and ungrazed plots of *Q. rotundifolia* and *Q. pyrenaica*.

Habitat		Penetration resistance (MPa)		
		0–10 cm	10–20 cm (MPa)	20–30 cm
<i>Q. rotundifolia</i>	Grazed	3.03 (±0.23) A	6.99 (±0.71) A	8.68 (±0.28) A
	Ungrazed	1.87 (±0.11) B	4.57 (±0.37) B	6.01 (±0.61) C
<i>Q. pyrenaica</i>	Grazed	2.03 (±0.24) B	3.80 (±0.27) B	5.14 (±0.39) C
	Ungrazed	1.70 (±0.18) B	5.32 (±0.49) AB	7.23 (±0.31) B
Significance (<i>P</i> <)		0.004	0.001	0.001

Averages followed by different letters in the column indicate significant differences between habitats for each variable according to Tukey's test at 5% probability, while the values in parentheses represent the standard error.

et al., 2021). The dense network of surface roots and the accumulation of litter under the canopy intensify bioturbation, nutrient cycling and microbial activity, favouring soil structure and porosity, which mitigates compaction even under intense and continuous grazing. (Rakkar *et al.*, 2018).

Conclusion

In Mediterranean mountain systems evaluated, soil compaction is determined mainly by edaphic properties (depth, texture, and organic matter content) and not only by the presence of livestock. *Quercus pyrenaica* in umbric Leptosols maintained stable surface resistance (0–10 cm) despite grazing, while *Q. rotundifolia* in shallow, stony Leptosols showed increased compaction under grazing. Thus, effective silvopastoral management should start with a prior soil diagnosis and adjust stocking rates and practices to the edaphic potential of each habitat.

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Training for pastoralism: issues, systems, tools and networks (case study in France)

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Abstract

Agro-silvo-pastoral (ASP) systems rely on the efficient use of a diversity of grazed resources with limited inputs, to provide a variety of goods and services over the long-term. Such strategy requires systemic, multi-scale approaches and adaptive management based on local knowledge and a close monitoring of the ecosystem. To secure the economic and social sustainability of ASP systems, the various stakeholders involved, from farmers and shepherds to technical advisers and policy makers, need to understand the functioning of ASP systems and the associated performance criteria. Education is thus a major issue. In France, pastoralism is addressed at various levels of agricultural education: shepherd schools, agricultural schools, higher education institutions. Recent projects have fostered exchange and coordination among existing training programmes, in order to share concepts and methods and ultimately facilitate cooperation between future stakeholders operating with or within the livestock sector, who will have opportunities to work together. This communication presents the common features of pastoral education in France, together with the tools and methods co-developed in the framework of recent projects, and the interest of local to international networks.

Keywords: agroecology, education, herbivores, management, rangelands, systemic approach

Introduction

Agro-silvo-pastoralism (ASP) is a type of livestock farming relying on extensive grazing of natural pastures, rangelands or woodlands to feed the flocks/herds, sometimes complemented with cultivated land grazed after harvest (dictionary of Agroecology: <https://dicoagroecologie.fr/en>). In Europe, ASP systems are mainly found in mountain and Mediterranean areas; in the former, they are sometimes the only agricultural activity, which implies a major socio-economic and ecological role; in the latter, they represent an efficient solution to take advantage of the typical mosaic of rangeland, woodland and cultivated areas and provide services ranging from cultural landscapes to reduction of agricultural inputs and lowering of fire risk. In the last decade, the growing interest in agroecology and in the socio-ecological services provided by low-input agricultural systems relying on biological diversity have paved the way to reconsidering the interest of ASP systems, beyond the cultural heritage they represent (Plininger & Huntsinger, 2018). In order to support such systems and encourage their development, they should be understood by a variety of stakeholders: farmers and advisory services, but also land owners, policy makers, operators of product chains, etc. To achieve this, ASP should be introduced into agricultural education.

The ASP systems differ from conventional livestock systems, especially for three distinctive features (Jouven, 2016): (1) they rely on diversity (biological diversity, diversity of products and services, diversity of production strategies), and conserve it; (2) the production strategy includes anticipation

and adaptation to changes (seasonality, climatic hazards, erratic access to land or markets), in both the short- and long-term; (3) the operation of the system relies on specific skills (of both animals and humans) and local knowledge. While conventional livestock systems are well documented and included in the curricula of agricultural studies, ASP systems are rarely mentioned. In fact, their distinctive features challenge education: how do you teach something that is by essence multifaceted, varied and variable? What general understanding should be put forward when local knowledge is concerned? Nevertheless, six shepherd schools exist in France and similar schools have been developed in other European countries (Spain, Italy, Portugal and Switzerland). The ASP is also included in other agricultural education programmes in France, from technical studies to Master's courses. Based on the example of France, this paper aims to analyse how to teach ASP in a variety of educational contexts, and give insight into the possible role of local to international networks.

Material and methods

This work focuses on four educational contexts including ASP that exist at Institut Agro Montpellier (with their main targeted profession): shepherd school (shepherd); technical education and continuous training of the associated teachers (farmer, technical adviser); Bachelor degree in agricultural or environmental science (project manager, at a local scale); Master's degree in agronomy (manager position in government institution, agribusiness, technical institute, association; agricultural research and education). The staff involved in teaching meet regularly, especially in the framework of the mixed technological unit "Unité Mixte Technologique (UMT) Pasto", which has encouraged, since 2015, exchanges between research, education and extension on the topic of ASP. The results reported here are based on various projects carried out in France in the last five years involving the authors, and especially on the PASSTEC project, which aimed at producing shared educational material in order to tackle the agro-ecological and climatic transitions through the study of ASP. In the framework of a series of focus groups, we cross-analysed the training programmes about ASP in the aforementioned four educational contexts, focusing on: (1) the main features of education in ASP; (2) the implications in terms of education tools and methods; (3) the added value of educational networks.

Results and discussion

We identified four common features in the training programmes aimed at grasping the functioning of the ASP social-ecological system, reported in Table 1: (1) multi-scale interdisciplinary systemic analysis; (2) agro-ecological knowledge; (3) observation and monitoring of ASP systems and (4) discussions and debate. These four features lead to the understanding of complex systems, through complementary methods involving direct interactions with a variety of components of the socio-ecosystem: interviews with stakeholders, direct observation of the state and behaviour of pastures and of grazing flocks/herds. Such direct interactions mobilise specific investigation methods, usually adapted for education on the basis of existing research or extension tools. In order to understand and interpret the discourse or observed behaviour of ASP components, the students need a certain amount of knowledge about ecology, livestock and pasture science, social sciences, etc., which can be provided by a few lectures or introduced within the documents used in the field. The combination of a diversity of points of views and case studies supports debates and discussions about ASP strategies and their interests/limits, which enables to draw principles and models of action. Recorded interviews, serious games and simulation models can be used in order to diversify the examples (other regions, other countries), and fuel the debate among students. A variety of tools, with their associated educational setting, are now available on the PASSTEC platform (in French). All tools are applicable to a range of educational contexts, which may seem strange given the diversity of targeted professions. In practice, what changes

Table 1. Common features of education in ASP, with associated tools and methods.

Features of education in ASP	Objectives	Associated tools and methods	Examples
Multi-scale systemic and interdisciplinary analysis	Explore the connections between agricultural, ecological and socio-economic aspects, at various levels of scale	Meeting a variety of local stakeholders; case studies exploring various aspects of ASP; serious games	Interviews (carried out by students or recorded ¹); serious game AEOLE ²
Agro-ecological knowledge	Understand the functioning of the pastoral social-ecological system	Models and serious games combined with discussions with farmers and ecologists	Rami Pasto serious game ¹ ; P@stor-all platform ¹ ; videos of shepherds at work ¹
Observation and monitoring of ecosystems, in various contexts	Use methods and indicators that inform on the state and dynamics of the ecosystem	Observation sheets combining questions and knowledge; methods for eco-pastoral diagnosis	Mil'Ouv method ³ ; observation sheets for pastures and herds/flocks ¹
Discussions, debate	Share and evaluate a variety of strategies, considering the local context and possible evolutions	Cross-presentations of individual or group analysis	Q-sort about ASP ¹ ; methods to trigger and moderate debates

¹ Available on the PASSTEC platform: <https://fermewikisagro.fr/passtec/>.

² <https://www.sidam-massifcentral.fr/developpement/aeole/>.

³ https://idele.fr/fileadmin/medias/Documents/Livret_pedagogique_milouv.pdf

between technical and scientific education is how far the analysis is pushed, and how much theory and models are produced through discussion and debates. Using similar tools to educate a variety of stakeholders is a powerful lever of change, since it provides common ground for future interactions and collaborations.

These advances in understanding, documenting and empowering ASP education were made possible by projects that gathered teaching staff from a variety of training programs. Certain projects produced teaching material, which is available on open internet platforms, usually in the language of the country. European projects such as the Erasmus PastorEA, Shep4Bio, EU4Shepherds, TransFarm or Eco-transH fostered exchanges and advances in ASP education, especially for shepherds. The Pastinova PRIMA project organized international summer schools for scientist and extension services. Based on our results, it would make sense to connect ASP education, at all levels, within and between EU countries, in order to diversify case studies and share educational methods.

Conclusion and perspectives

In order to support ASP, a variety of stakeholders should be able to understand its distinctive features and how these contribute to the provision of a variety of goods and services, in a changing ecological, climatic and socio-economic context. To achieve this, ASP should be introduced into agricultural and ecological education, favouring systemic, interdisciplinary approaches, direct observations, and cross-analysis of a variety of case studies, with discussions and debate. Since similar tools may serve a variety of training programmes, transnational educational networks are powerful tools to share educational material and encourage the teaching of ASP.

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Technical establishment of agrosilvopastoral systems for grazing dairy cows in northern Europe

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Abstract

More frequent periods of drought and heat pose big challenges for pasture based dairy production even in northern Europe. Agrosilvopastoral systems, the integration of trees on pastures, can provide shade for grazing animals and alleviate drought stress in grassland swards. However, integration into modern grazing systems is challenging due to frequent moves of animals, strip wires and mechanical maintenance requirements. Therefore, such systems are rare on research farms and in practice in northern Europe. This paper presents the technical establishment of a silvopastoral system at the research farm of the Thünen-Institute for Organic Farming in northern Germany. The objective is to test methods and designs for establishing trees on pastures that allow systematic research of individual effects in the system. In autumn of 2024, four different high value timber trees with different shading intensities were planted in rows 48 m apart. Two different tree protection methods were tested. The first year showed differences in establishment costs of tree protection. Future evaluations of tree growth, grassland productivity, botanical composition, shade use by animals, SOC changes and establishment and running costs will help farmers with decision making.

Keywords: grazing, pasture based dairy production, shade, silvopastoral systems, trees

Introduction

Agroforestry systems in general and silvopastoral systems in particular, i.e., the combination of trees and grazing animals on grassland, are discussed as a strategy to offset the greenhouse gas emissions associated with production by acting as a sink for carbon in agricultural systems. At the same time, they can serve as a climate change adaptation strategy by mitigating the effects of drought and heat on animals and swards. Through shading and changing wind conditions, the evapotranspiration of grass swards can be reduced (Sutterlütti *et al.*, 2023), thereby possibly counteracting yield losses and feed shortages during drought periods. Additionally, improved environmental conditions (shade, microclimate) are created for keeping different farm animal species. Trees also provide many other ecosystem services such as serving as a supplementary feed source or C storage and provide habitats for biodiversity and create additional market value for farms (Amorim *et al.*, 2023; England *et al.*, 2020).

Planting trees on grazed areas offers extensive opportunities to address numerous questions about sustainable land use systems and should serve as a basis and infrastructure to enable long-term implementation of research approaches in this area. However, investigations into interactions between tree-sward-animal systems have so far been limited to two-dimensional considerations. The positive effect of provision and amount of shade on the productivity and behaviour of grazing dairy cows has been analysed (Nielsen and Wredle, 2023; Palacio *et al.*, 2015; Schütz *et al.*, 2014). Studies looking at shade on grass swards have not shown a negative effect on sward productivity (Rivest *et al.*, 2013). There is a significant research need on interactions in silvopastoral systems, as only a few systems exist and no long-term studies are available. Practical integration in modern grazing systems is, however,

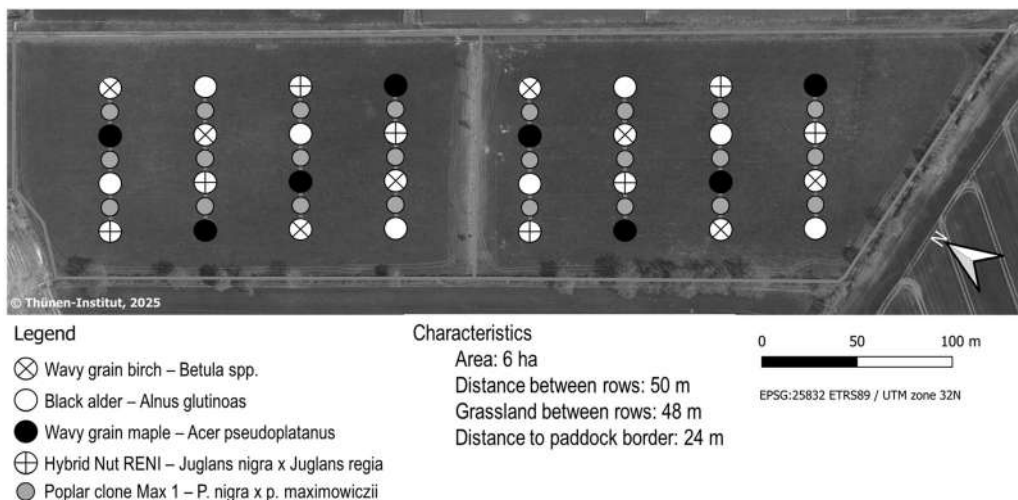


Figure 1. Planting of tree strip system on dairy cow pastures.

challenging due to frequent moves of animals, strip wires and mechanical maintenance requirements. It is also unclear how shade intensity from different trees could support systems with the appropriate amount of shading for animals and swards.

The aim of the project is to establish tree plantings on pastures for dairy cows to address many design and utilisation questions and allow systematic research of individual effects in the system. The focus is on the effect of different degrees of tree shading on grazing animals and grassland swards.

Materials and methods

In November of 2024 a silvopastoral system was established on two dairy cow pastures of 3 ha each at the Thünen-Institute for Organic Farming in northern Germany. The aim was to integrate trees for shading in a wide area of the paddock to allow shade for the herd for the whole day and in all grazing strips. The design was chosen to allow for mechanical harvesting and pasture maintenance work with a working width of 6 m of the agricultural machinery. The design was also required to seamlessly integrate trees in the grazing system with grazing allocations of 24 to 48 hours between tree rows (0.6 ha for 45 cows).

Trees were planted in rows with a distance of 50 m between tree rows (1 m buffer strips to each side; 48 m workable strip) and 12.5 m between individual trees in one row (Fig. 1), resulting in 9.3 trees per ha. Trees were planted in a randomized design in 4 replicates with four different high value timber trees that offer different shading intensities: hybrid nut (*Juglans nigra* x *Juglans regia*; intense shading), wavy grain maple (*Acer pseudoplatanus*; moderate shading), black alder *Alnus glutinosa*; moderate shading) and wavy grain birch (*Betula* spp.; light shading). The wavy grain trees (maple and birch) are expected to produce high-quality wood for special applications and bring added economic value. All selected trees do not drop any fruits etc. that could be harmful for grazing cows. In between the high value timber trees fast growing poplars (*Populus nigra* x *Populus maximowiczii*) were planted for a quick shading effect. Poplars can be removed once the slower growing high value timber trees offer enough shading. Additional gates and flexible water troughs were installed so that cows can enter and leave grazing strips between tree rows individually when tree row separations are used for strip grazing. At

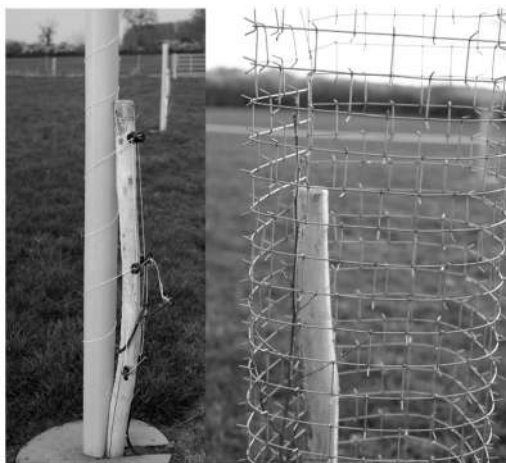


Figure 2. Different tree protection methods used in the silvopastoral systems for grazing dairy cows: plastic covers wrapped with electrified wire (left) and Cactus protector[®] (right).

the same time grazing two or more strips is possible as the tree protection is installed on each tree individually.

Two different tree protection methods against grazing and wild animals were tested: on one paddock trees are protected with the cactus protector[®] (<https://www.cactusprotector.be/en/>, 1.80 m height) and on the other paddock trees are protected with plastic covers (Tubex ventex, 1.80 m height) wrapped with electrified wire (Fig 2). Both methods are also equipped with a Robinia wood stake, a self-made vole basket and a wood fibre mulch board.

Results and discussion

In the grazing season of 2025, the silvopastoral system was grazed for the first time. Trees were not yet large enough to produce considerable shade; hence, the effect on grass swards was not measured but is expected to be minimal. The productive sward area lost by planting (area of mulching plate) was less than 0.1%. Both tree protection methods offered suitable protection of trees against cows. Fewer mulching plates were torn apart by animals where trees were protected with plastic covers and electrified wire. Protection methods resulted in different material costs: the cactus protector[®] including all other required material was at €48 per tree (€448/ha) and the plastic cover was at €19 per tree (€177/ha) when the electrified wire was led from tree to tree to connect to the main power line. With underground wiring the plastic cover method was at approx. 40 € per tree (373 €/ha) but offered the option of free movement between trees (Fig. 2).

Conclusion

Silvopastoral systems can be designed to seamlessly fit into modern grazing management. The loss of productive sward area was minimal. There are options for maintaining establishment and running costs at low levels. A wider network of research farms with silvopastoral systems in northern Europe could offer wider recommendations from different environments.

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Enhancing grassland simulations by evaluating grazing representations in ModVege

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Abstract

Effective grassland management is crucial for carbon sequestration and livestock productivity. Our study improved the ModVege model by adding spatially explicit grazing simulations to see how different grazing methods impact vegetation heterogeneity. We looked at four grazing approaches: uniform defoliation, random grazing, palatability-based selection and Optimal Foraging Theory. We tested these across various stocking rates (approximately 0.7–2 livestock units per hectare per year) and management regimes (continuous vs. rotational). Our results show that heterogeneous grazing patterns, such as random defoliation, increase spatial variance and help preserve reproductive biomass, unlike homogeneous methods. We found that rotational grazing at moderate intensity was the most productive and resilient strategy, while continuous, high-intensity grazing made the ecosystem less resilient. Calibrated for French grasslands, our modular design makes the model a valuable tool for implementing spatially explicit grazing strategies in both temperate and arid ecosystems (e.g. African rangelands that deal with unpredictable rainfall and unique pastoral systems).

Keywords: grassland modelling, grazing management, ModVege, spatial heterogeneity, sustainable agriculture

Introduction

Grasslands cover approx. 40% of the Earth's ice-free land and are essential for carbon storage, biodiversity and livestock support (Dixon *et al.*, 2014). Grazing management strongly influences grassland vegetation, soil health and productivity, yet most models often oversimplify the spatial interactions between plants and herbivores (Ren *et al.*, 2024). While process-based models like ModVege and its CoSMo extension (Piseddu *et al.*, 2022) improve biomass simulation by including plant physiology and grazing, they often neglect spatial heterogeneity - a key factor shaping vegetation patterns and ecosystem resilience (Grandin, 2022). To address these gaps, we developed a spatialised ModVege model to quantify how different grazing methods influence vegetation dynamics. Specifically, we: (i) compare grazing methods for their effects on biomass partitioning and spatial heterogeneity, (ii) assess interactions between grazing intensity and management regimes, and (iii) identify trade-offs between productivity and resilience.

Materials and methods

We used a spatialised Java implementation of ModVege to simulate a 4.2-ha pasture, discretised into 650×650 hexagonal cells of 0.1 m^2 each. The model tracked four biomass compartments: vegetative green (BGV), reproductive green (BGR), vegetative dry (BDV) and reproductive dry (BDR). A two-by-two factorial design investigated stocking rates (low: approx. $0.69 \text{ LSU ha}^{-1} \text{ year}^{-1}$; high: approx. $2.08 \text{ LSU ha}^{-1} \text{ year}^{-1}$) and management regimes (continuous grazing over 150 days vs. rotational grazing in five 10-day cycles). Daily biomass outputs (NetCDF4) were analysed in R (ncdf4, ggplot2),

Table 1. Simulation parameters and experimental design.

Parameter	Description
Pasture size	650 × 650 hexagonal cells (0.1 m ² each) Total: 4.2 ha (42 250 m ²)
Time frame	1 growing season (365 days)
Grazing period	150 days (continuous) or 5 × 10-day cycles (rotational)
Livestock units (LSU)	1 LSU = 500 kg dairy cow
Stocking rates	Low intensity: approx. 0.69 LSU ha ⁻¹ (7 LSU/4.2 ha×150 days/365 days) High intensity: approx. 2.08 LSU ha ⁻¹ 21 LSU/4.2 ha×150 days/365 days) (continuous) 63 LSU/4.2 ha×50 days/365 days (rotational)

Characteristic	Homogeneous Defoliation	Random Grazing	Palatability-Based Selection	Optimal Foraging Theory
Grazing Pattern	Uniform removal	Unstructured movement	Selective grazing	Energy maximization
Management Input	High	Minimal	Moderate	Complex
Animal Performance	Potentially reduced	Potentially reduced	Improved	Improved
Pasture Impact	Improved health	Overgrazing risk	Degradation risk	Overgrazing risk
Best Use Case	Pastures needing even regrowth	Situations needing minimal intervention	Optimizing animal nutrition	Efficient resource utilization

Figure 1. Comparison of four grazing methods. Homogeneous (uniform defoliation: 12.3 kg DM LSU⁻¹ day⁻¹); random (stochastic cell selection); palatability-based (Delagarde *et al.*, 2001); Optimal Foraging Theory (Pyke, 1984).

Table 2. Productivity across grazing regimes (mean±SD; biomass in kg dry matter ha⁻¹).

Management regime	Total biomass	Green vegetative biomass (BGV)	Green reproductive biomass (BGR)	Key implications
Continuous (0.69 LSU ha ⁻¹)	6584 ± 210	4120 ± 185	0 ± 0*	Eliminated reproduction
Rotational (0.69 LSU ha ⁻¹)	6712 ± 185	3980 ± 162	1208 ± 92**	Balanced productivity
Continuous (2.08 LSU ha ⁻¹)	3102 ± 157*	2845 ± 134*	0 ± 0*	Severe biomass loss
Rotational (2.08 LSU ha ⁻¹)	4387 ± 231*	3210 ± 198*	526 ± 74**	Partial recovery

using grassland data from the Massif Central, France (CNIEL and Institut de l'Élevage, 2007). Table 1 details the full simulation parameters, including pasture size, time frame and livestock definitions.

The key characteristics, management requirements and recommended use cases for grazing methods are summarised in Figure 1.

Results and discussion

Grazing implementation method strongly affected pasture dynamics, especially reproductive biomass and spatial heterogeneity (Table 2).

Homogeneous defoliation (as in simplified models) eliminated reproductive biomass, mirroring intensive grazing that prevents plant regeneration (Wentao *et al.*, 2023). Random grazing preserved 15–20% of reproductive biomass, better reflecting natural heterogeneity (Odadi *et al.*, 2017). Spatial variance was lowest under homogeneous grazing (approx. 50 kg² ha⁻²), highest with random grazing (210 kg² ha⁻²) and intermediate with OFT-based grazing (max. 150 kg² ha⁻²), supporting evidence that non-spatial models underestimate pasture heterogeneity. High-intensity continuous grazing reduced total biomass by 53% and delayed phenological peaks, eliminating reproductive output. Rotational grazing mitigated these effects, preserving reproductive biomass and increasing productivity by 41% at high intensity. Overall, low-intensity rotational grazing provided the best balance of productivity and resilience, supporting with global recommendations for grassland management (Ren *et al.*, 2024).

Conclusion

This spatialised modelling framework improves the prediction of pasture patch dynamics and supports adaptive, sustainable grazing across diverse grasslands. It helps identify key thresholds for making it a valuable tool for optimising management. Future work should integrate biotic interactions, validate results with field data and extend modelling to multi-year climate scenarios—especially for regions with high rainfall variability. Overall, spatial models are essential for sustainable grazing management in a changing environment.

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Disturbance and mutualism: how grazing shapes pastoral ecosystems

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Abstract

Grazing herbivores profoundly shape pastoral ecosystems through their presence and behaviour, playing a crucial role in structuring vegetation, influencing biodiversity and regulating nutrient cycles. This paper explores grazing's dual nature—as both a disturbance and a mutualistic force—and its multi-scale interactions with vegetation. We examine how diverse pastoral practices influence plant communities and contribute to ecosystem resilience. Given the inherent variability of pastoral resources, especially with shifting environmental conditions, adaptive strategies and collaborative management are vital for sustainable use. Drawing on theoretical frameworks and empirical evidence, we advocate for balancing ecological risks (e.g. degradation, biodiversity loss, climate change impacts) with benefits like biodiversity conservation and maintaining resilient landscapes. Ultimately, we recommend robust institutional arrangements to support long-term ecological and social well-being.

Keywords: biodiversity conservation, climate change adaptation, ecosystem services, extensive grasslands, grazing ecology, vegetation dynamics

Introduction

Pastoral ecosystems—naturally vegetated grazing lands—are shaped by plant-herbivore co-evolution and grazing dynamics, with animals enhancing vegetation, soil conditions and nutrient cycling through defoliation, trampling, seed dispersal, and fertilisation (Probo *et al.*, 2025). Climate change threatens the sustainability of extensive pastures and rangelands by reducing forage, altering plant communities, and weakening drought resilience (Deléglise *et al.*, 2022). As a keystone process with both homogenising and diversifying effects (Adler *et al.*, 2001), grazing's outcomes depend on pastoral practices. This paper examines regional adaptive strategies to sustain pastoralism under shifting environmental conditions.

Materials and methods

This synthesis investigates alpine and Mediterranean pastoral systems across three key dimensions: vegetation response, grazing impacts and climate interactions. First, we assessed species richness, functional traits and productivity using long-term ecological monitoring data from the Alpes Sentinelles network (<https://www.alpages-sentinelles.fr>). Second, we examined defoliation, trampling and nutrient redistribution through empirical data (e.g. faecal sampling, soil assays from field trials) and relevant literature on grazing intensity and ecosystem feedbacks. Third, we explored phenological shifts and resource variability using station data, field observations of vegetation phenology and scenario-based projections of grazing pressures and ecological responses from the EU LIFE PASTORALP project (<https://www.pastoralp.eu>). This approach combines direct observations with robust secondary sources, enabling a nuanced and context-sensitive interpretation of grazing effects.

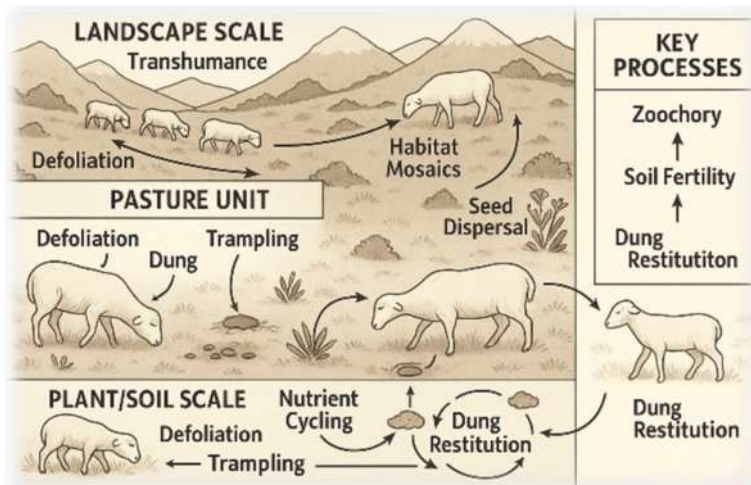


Figure 1. Multi-scale ecological processes in transhumant grazing. Livestock shapes ecosystems across (1) Landscape: transhumance maintains habitat mosaics, aids seed dispersal (zoochory) and affects vegetation structure; (2) Pasture: grazing modifies vegetation (defoliation/trampling); dung/urine affect nutrient cycling/soil fertility; (3) Local/Plant: defoliation, trampling and root turnover impact plant structure/below-ground dynamics. Core processes zoochory, soil fertility and dung restitution drive feedback loops. Figure created with AI assistance (Perplexity for layout and ChatGPT for final rendering) based on original conceptual design and authors' instructions. The illustration does not mimic any published copyrighted figures.

Results and discussion

Grazing acts as both an ecosystem engineer and a nature-based solution, shaping vegetation, species composition, and ecosystem processes across contexts and scales (Figure 1). While continuous grazing tends to homogenise vegetation at landscape scales, individual animal behaviours—like selective foraging and movement patterns—create fine-scale heterogeneity. Grazing pressure affects plant communities in context-specific ways, favouring stress-tolerant traits (e.g. low stature, sclerophylly) in Mediterranean regions, and producing variable impacts on diversity in alpine areas. While studies in the Marais Poitevin (in the Atlantic façade) support the intermediate disturbance hypothesis, where moderate grazing limits dominance and enhances diversity, findings from Alpages Sentinelles show that grazing effects (positive or negative) depend on abiotic constraints like soil depth and nutrient availability. In addition, transhumance corridors promote colonisation via zoochory and mosaic maintenance, boosting species spread by up to 20%.

Grazing shapes ecosystems from plant to landscape scales through direct (defoliation, trampling) and indirect (nutrient cycling) effects (O'Grady *et al.*, 2024). Its impact depends on management—timing, location, intensity, herd structure and behaviour—with long-term monitoring and experiments showing it maintains open habitats via scrub clearance and controlled burns. While moderate grazing boosts soil fertility, nutrient build-up near night camps can cause eutrophication and diversity loss. Adaptive practices like rotational grazing offer context-specific solutions, though their full ecological outcomes remain under study. Climate warming adds complexity, reducing subalpine pasture protein content by up to 15% (Fanlo and Taull, 2018), with interannual variability like hot springs further worsening forage quality declines up to 20% based on alpine monitoring. Grazing may buffer climate-driven plant shifts (Speed *et al.*, 2012), but the shift from mobile to stationary grazing systems in former transhumant areas increases overuse risks. While grazing mitigates wildfire risk in Mediterranean shrublands, overuse may degrade fragile alpine zones. Addressing these trade-offs need flexible, evidence-based

Table 1. Trade-offs and interactions between pastoralism, biodiversity and climate change, with management recommendations (e.g. PASTORALP reports).

Factor	Positive impact (pastoralism)	Climate-related risk/interaction	Management recommendation
Mosaic habitats	Maintains open grasslands and supports species diversity	Overgrazing in sensitive alpine zones can reduce structural diversity	Rotational grazing; mixed-species herds
Nutrient cycling	Dung and urine improve soil fertility (N, P)	Nutrient hotspots (e.g. night camps) may cause eutrophication, favouring nitrophilic species	Use of mobile corrals to redistribute input
Climate resilience	Transhumance and adaptive timing exploit seasonal growth windows	Drought and warming reduce forage quality and increase stress on vegetation	Strategic water-point planning; rotational grazing; flexible transhumance

strategies—mobile/rotational systems, mixed-species herds and even pressure distribution—to support ecosystem resilience (Table 1).

Conclusion

Pastoral ecosystems face environmental strain and heavy reliance on grazing animals, needing evolving, sustainable practices to build resilience. Blending traditional knowledge with scientific expertise is key to creating adaptive grazing strategies that support flexible transhumance and diversified grazing, vital for long-term ecosystem health. Tools like the PASTORALP platform (<http://www.pastoralp.eu/tools>) offers practical solutions to foster resilient, multifunctional landscapes. Protecting these landscapes, essential for biodiversity and rural livelihoods, demands coordinated efforts from policymakers, local stakeholders and the wider community. To safeguard ecological integrity and ensure long-term sustainability, future research should prioritise continuous monitoring (e.g. via Alpages Sentinelles) and robust evaluation of adaptive strategies under shifting environmental conditions.

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Converting *Cistus ladanifer* areas to productive grasslands in southern Portugal

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Abstract

Mediterranean shrublands dominated by *Cistus ladanifer* represent extensive areas with limited agricultural productivity. This experimental study investigated converting these landscapes into productive grasslands in the Algarve region, southern Portugal. Two sites were established in Benafim where *C. ladanifer* was the predominant vegetation. Following mechanical removal of shrub vegetation, areas were seeded with seeds naturally present in animal feed. The experiment monitored vegetation dynamics, biomass production, and floristic composition over multiple growing seasons, comparing intervened areas with non-intervened control sites. Results demonstrate significant transformation potential, with converted areas producing approximately 5 t ha⁻¹ of dry biomass during 2024–2025. There was no loss of floristic composition diversity observed in the converted areas. This land management approach enhances grassland resilience and agricultural productivity in Mediterranean regions: converting low-productivity *C. ladanifer* areas into functional grasslands offers benefits for carbon sequestration, soil stability, and sustainable grazing systems. Using seeds naturally present in animal feed is a practical, economically viable strategy scalable for broader landscape interventions. These findings contribute to grassland development technologies and restoration practices supporting climate adaptation in Mediterranean environments, addressing key challenges in grassland resilience under changing climatic conditions.

Keywords: biomass production, *Cistus ladanifer*, grassland establishment, land conversion, Mediterranean grasslands

Introduction

Natural grasslands in Portugal's rural landscape emerge mainly in abandoned areas, often invaded by shrubs. In southern Portugal, *Cistus ladanifer* L. usually dominates these areas due to its pioneer characteristics and resilience to Mediterranean conditions, where its vigorous growth and leaf litter allelopathic compounds inhibit the development of other species, creating monospecific formations (Mendes *et al.*, 2015). Although *C. ladanifer* has potential economic value, rural landowners face difficulties exploiting these resources due to limited infrastructure and market networks. Climate change can favour invasive shrubs (Caldeira *et al.*, 2015), although shrub removal followed by grazing can be effective in reversing land degradation (Mendes *et al.*, 2015).

Well-managed grasslands provide carbon sequestration, soil stability (Lorenz & Lal, 2018), and sustainable grazing through maintained productivity and biodiversity (Teague & Kreuter, 2020). One of the methods to improve grasslands is mechanical shrub removal followed by seeding (Török *et al.*, 2011), which also improves germination of species present in the seedbank (Mendes *et al.*, 2015, Bohlman *et al.*, 2016).

This study aims to evaluate the effectiveness of mechanical shrub removal, followed by seeding with seeds present in the supplementary forage given to grazing sheep, to convert *C. ladanifer* shrublands into productive grasslands. It is expected that this method will successfully establish productive grasslands without significant biodiversity loss.

Materials and methods

This experiment was conducted in the Algarve, southern Portugal, with a Mediterranean climate (Classification: Csa) characterised by warm and arid summers and mild, rainy winters, with moderate precipitation patterns, averaging 500 mm yearly. Two grassland sites were established at Quinta do Freixo, in Benafim: Pasture A (PA), with 3.4 ha, in 2022, and Pasture B (PB) with 2.9 ha, in 2023. Both sites are on slopes, with soils developed over schist and greywacke, with an acidic pH (approximately 5.7), and low organic matter content. Shrub removal was done with a brush cutter, and hay with seeds was spread, with a mixture of oats and vetch for PA and a biodiverse mixture for PB.

In 2025, biomass production and its floristic composition were measured in both pastures. Biomass samples were collected in a zigzag pattern in both sites and sorted in three main botanical groups: grasses, legumes and others. After weighing the fresh matter, samples were dried in a drying oven at 65°C for 72 hours and weighed again to determine the dry matter (DM). The height of the swards was also measured with a rising platometer, to obtain a relationship between the height and weight of the biomass of each experimental area. In May of the same year, we also conducted a survey of the floristic composition, which involved randomly delimiting 1 m² plots across the pasture and the respective uncleared shrublands, identifying all plant species within, and counting individuals per species. A total of 18 plots were sampled (7 plots in PA and 2 plots in its neighbouring shrubs, 7 plots in PB and 2 plots in its neighbouring shrubs); some specimens were collected for laboratory confirmation and preserved for reference.

Results and discussion

While the starting estimated DM production for PA was lower than for PB, PA quickly outperformed PB along the growing season, as can be seen in Figure 1. As for the floristic composition, both pastures expressed significant variabilities as the season progressed, expressing the dynamic interactions of a Mediterranean pasture system.

Overall diversity was high, with 21 legume species (mainly clovers) and 38 other species across 19 families, though many are of low pasture value. Grasses, while abundant in biomass, were limited to seven genera, with oat showing poor persistence in PA, likely due to lodging and inter-species competition. In PB, the other botanical species percentage was low but highly diverse, which can be a result of hay seeding with a biodiverse mix. A smaller diversity and number of species were observed in the rockrose shrubs, but the persistence of the seedbank was notable, with several species identified, many from the *Compositae* family. Overall, the flora expressed typical Mediterranean herbaceous dynamics, with rapid winter growth, spring peak, and preparation for summer drought dormancy, driven by species-specific phenology and competitive interactions.

The successful conversion of *C. ladanifer*-dominated shrublands into productive grasslands achieved high biomass production levels in 2025. These results may be partially attributed to exceptional precipitation conditions during the study period. The maintenance of high floristic diversity (21 legume species and 38 other species across 19 families) demonstrates that mechanical shrub removal followed

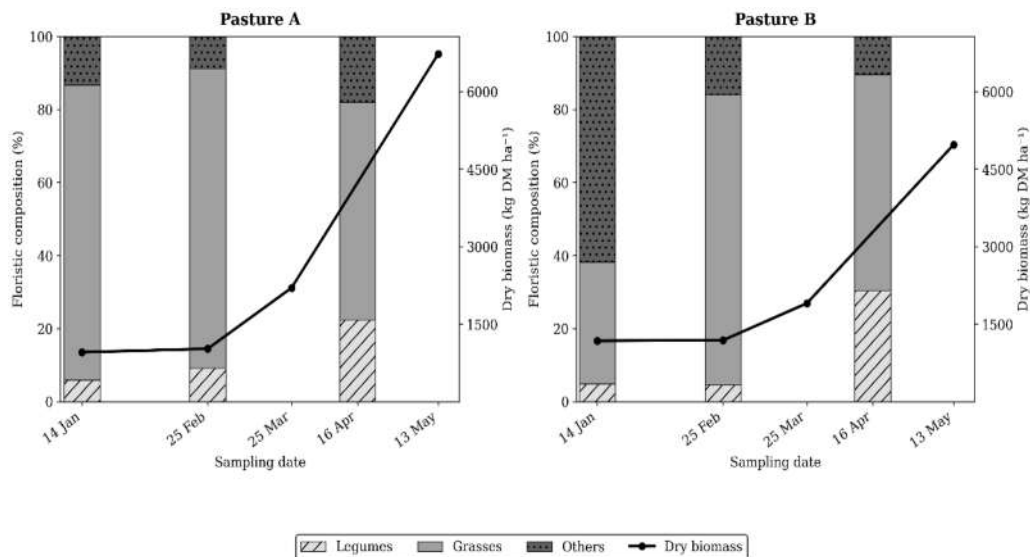


Figure 1. Biomass production and floristic composition of experimental pastures A and B, during the growing season of 2025.

by targeted seeding preserves ecosystem complexity while enhancing productivity. The differential performance between sites, with PA showing superior biomass but PB maintaining greater diversity through biodiverse seed mixtures, highlights the importance of seeding strategy selection. The persistence of native species from the soil seedbank indicates successful preservation of local genetic resources. These findings validate this land management approach as viable for enhancing Mediterranean agricultural productivity while maintaining ecological integrity, though the exceptional precipitation conditions suggest that results may vary under typical climatic scenarios.

Acknowledgements

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Grassland response to drought in dryland Mediterranean oak woodlands

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Abstract

The *Montado* agroforestry system in southern Portugal is a valuable dryland ecosystem facing high desertification risk from climate change and intensified land-use pressure. Its understorey grasslands are crucial biodiversity reservoirs, but are highly threatened by increasing aridity and drought. We studied grassland response to simulated drought (rain exclusion) across three Long-Term Socio-Ecological Research (LTsER) sites distributed along an aridity gradient. Response was assessed via biodiversity, plant productivity, and soil parameters. After two years, we find that plant diversity is strongly linked to interannual climatic variability, with drought response and resilience differing across sites. This underscores the complex site-specific nature of drought impacts and the need for long-term monitoring to inform effective adaptation strategies.

Keywords: biodiversity, ecosystem resilience, montado, rain exclusion

Introduction

Grasslands are vital in dryland ecosystems, supporting soil protection, nutrient cycling, and climate regulation (Safriel *et al.*, 2005). In Southern Portugal, the montado (a High Nature Value agro-silvo-pastoral system highly vulnerable to desertification) hosts understorey grasslands, mainly composed of annual species, critical for biodiversity and livestock. These annual species possess high turnover rates, conferring resilience to interannual climatic variability (Nunes *et al.*, 2017). However, escalating anthropogenic pressures and climatic stress are driving montado decline and biodiversity loss (Plieninger *et al.*, 2021). Projected increases in aridity and drought intensity may accelerate desertification and affect ecosystem functions (Maestre *et al.* 2022). Here, we assessed induced drought effects on plant diversity after two years across three semi-arid montado sites. We hypothesize that (i) drought will reduce plant diversity and (ii) this response will be modulated by the original site aridity, reflecting diverse plant drought-coping strategies (e.g. seed bank dynamics, climate legacy effects, etc).

Materials and methods

Drought simulation experiments (rainout shelters) were established at three Long-Term Socio-Ecological Research (LTsER) montado sites (Table 1), following the Drought-NET protocol (Knapp *et al.*, 2017). Five replicated plots of three treatments: (i) control (unsheltered), (ii) moderate (50% rainfall reduction), and (iii) extreme (1st percentile of 100-year rainfall records per site). Each 2 × 2 m plot was lined (50 cm deep) and covered (80 cm height) to minimize microclimate disruption. Baseline plant composition (2022) and annual sampling (2023, 2024) was recorded at peak productivity (April–May).

Table 1. Site characterization

Site name (abbrev.) (coordinates)	Soil texture	MAP short-term			MAP long-term	AI	MAT (°C)
		2022	2023	2024			
Companhia das Lezírias (CL) (38.83N, 8.82 W)	Sandy	554	359	505	691	0.63	16.7
Herdade da Ribeira Abaixo (HRA) (38.10N, 8.57 W)	Sandy clay loam	456	274	477	636	0.55	15.9
Herdade da Coitadinha (PNN) (38.17N, 7.04 W)	Sandy loam to Clay loam	378	321	453	545	0.43	16.4

Soil texture, short-term mean annual precipitation (MAP) based on annual weather station observations (IPMA, IP, 2025) and long-term (50 years; Muñoz Sabater, 2019); Aridity index (AI); and mean annual temperature (MAT; Fick & Hijmans, 2017).

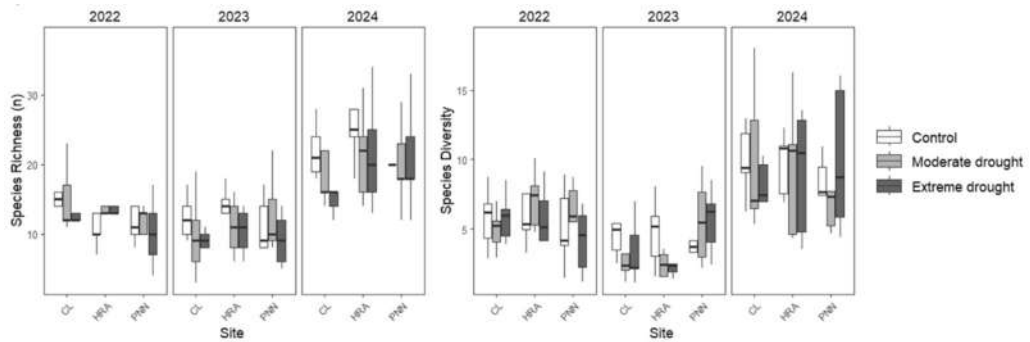


Figure 1. Species richness (left) and diversity (right) on study sites (CL, HRA and PNN), years (2022, 2023 and 2024) and treatments (control, moderate and extreme drought).

Table 2. Effects of Year and Treatment on the LnRR of species richness and diversity (Hill's number), in 2023 and 2024 compared to the initial year (2022) and respective relationship.

	CL			HRA			PNN		
	df	P	Effect	df	P	Effect	df	P	Effect
Richness									
Year	1	0.000	+	1	0.000	+	1	0.000	+
Treatment	2	0.065	=	2	0.006	—	2	0.354	=
Species diversity									
Year	1	0.000	+	1	0.000	+	1	0.000	+
Treatment	2	0.053	=	2	0.063	=	2	0.034	+ (extreme drought)

Diversity was quantified as species richness and as Hill diversity of order 1 (Hill, 1973), corresponding to the effective number of species (Tuomisto, 2010). Treatment effects used the Log Response Ratio ($\text{LnRR} = \ln(\text{Treatment}_{\text{year}}) - \ln(\text{Treatment}_{2022})$). Linear Mixed- Models (LMMs) were fitted per site. Year, Treatment and their interaction were fixed terms, with blocks nested within sites a random effect. Model selection used Akaike Information Criterion ($\Delta \text{AIC} > 2$) and Variation Inflation Factor (VIF) verified multicollinearity. All analyses utilised R software (R Core Team, 2025).

Results and discussion

Evaluation of LnRR of plant richness and diversity via LMMs showed a significant *Year* effect, found for all sites, indicating that both indices increased from 2023 to 2024 (Figure 1 and Table 2).

This response reflects interannual variability in plant community, probably linked to the higher precipitation in 2024 (Table 1). Significant Treatment effects were site-specific: species richness decreased

in HRA (lower in both drought treatments), but diversity unexpectedly increased in PNN under extreme drought, compared to other studies (e.g., Korell *et al.*, 2021). However, this diversity increase, combined with the lack of differences in richness, suggests a local increase of species evenness probably associated with the disappearance of drought-sensitive species. No statistical significance for Year×Treatment interaction was found.

Conclusions

This study assessed the grassland response to two years of simulated drought in Mediterranean drylands. Our findings suggest that while interannual climatic variability strongly influences plant diversity across sites, the highly site-specific responses to drought underscore the complexity of these impacts. To better predict ecosystem responses, future assessments need to consider additional biodiversity metrics (e.g. functional diversity), and integrate climatic factors through long-term monitoring.

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Local perceptions of conservation and management of mountain grasslands under climate change in Iberian National Parks

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Abstract

Alpine communities have a high ecological value and provide relevant ecosystem services, but are vulnerable to climate and land use changes, causing notable variations in plant distribution and composition. Furthermore, alpine communities have relevant socioecological functions, such as extensive livestock farming. We studied the perception of vulnerability to climate change of ecosystem services in alpine grasslands in five National Parks in the Iberian Peninsula. We gathered the perceptions of key stakeholders about the status, management and conservation strategies of mountain grasslands within the climate change framework in these five locations. We distributed 27 online surveys to all five National Parks, which have described the conservation status and management strategies for each studied park. Furthermore, we conducted two focus groups in the National Park *Aigüestortes i Estany de Sant Maurici*, and applied a thematic analysis, where we identified climate change impacts in pasture use and ecosystem services, including the extension of the summer grazing season, and a decrease in diversity, water and grass availability. We highlight key management strategies which include rotating foraging livestock and path cleaning.

Keywords: climate change, ecosystem services, National Parks

Introduction

Mountain regions provide crucial ecosystem services to their inhabitants and the surrounding lowland populations. Greater attention to the ecological and social implications of climate factors is particularly important for assessing future management strategies and enabling adaptation (Clark, 2019), as management options appear to be increasingly limited with the accelerated climate change (Schirpke, 2021). The acceleration of climate change will become the main driver of changes in ecosystem services in mountain grasslands, especially at high altitudes. However, understanding the dynamics of ecosystem service resilience in mountain grasslands under future scenarios is still limited (Kohler, 2017). For this reason, the preservation of ecosystem services provided by mountain grasslands requires effective conservation strategies to ensure the protection of biodiversity and ecological functionality and to contribute to maintaining traditional mountain activities, such as pastoralism (Straffelini, 2024).

Table 1. Perceived factors related to climate change

Observed increase in temperatures.
Occurrence of heatwaves.
Changes in precipitation distribution.
Climate uncertainty.

The objective of this research is to carry out a study of the perceptions of stakeholders of the vulnerability to climate change of ecosystem services in high altitude grasslands in five national parks across the Iberian Peninsula, considering the particularities of each of these spaces through the contributions of representatives from different sectors.

Material and methods

A survey of the stakeholders’ perception on grassland vulnerability to climate change was conducted in five mountain National Parks across the Iberian Peninsula: Sierra Nevada, Sierra de Guadarrama, Ordesa and Monte Perdido, Aigüestortes i Estany de Sant Maurici, and Picos de Europa. Qualitative information was collected from stakeholders across sectors: public administration, private sector, livestock farmers, and local associations. A total of 27 surveys were conducted in the five NPs, structured into sections using the Likert scale. A descriptive qualitative analysis was applied using the Excel software. In addition, two focus groups were conducted in Aigüestortes and Estany de Sant Maurici NP, using the snowball method to identify key stakeholders. The MAXQDA software was used for the qualitative analysis of focus groups transcriptions, and a MAXMap was created to facilitate visualization of the results. We used inductive thematic analysis to identify emerging themes, codes and subcodes. The inductive approach allowed the data to be analysed without a prior analytical framework, avoiding the influence of the researcher’s preconceptions.

Results and discussion

Stakeholders were aware of the main threats of climate change (Figure 1, Table 1). Our results suggest an impact perceived on pasture use due to rising temperatures, which has broadened the grazing period, extending grass availability during the year, modified the grazing hours to avoid the heat, and dried the grass during summer. Changes in water availability are identified due to Atlantic or Mediterranean climatic influence. Also, water scarcity drives livestock gathering at water points during summer, which might influence wetland ecosystems conservation. Furthermore, drought periods in valley-bottom meadows are especially harsh, as forage production is low and farmers need to buy forage. Finally, uncertainty in water projections plays a key role in pasture use.

Also, changes perceived in ecosystem services with climate change were a lower availability of grass with increased drought periods, especially in valley bottoms, as well as decreased water levels, less biodiversity, reduced food security, and decreased sociocultural value of the grasslands. Therefore, there is a need to protect the sector, including managers of the territory and food producers, and, according to stakeholders, prioritize this over the conservation of wildlife, such as bears, which affect livestock management and, indirectly, grassland conservation. The proposals for good management practices of mountain grasslands include pasture rotation to take advantage of less-used pastures and promote plant growth in overgrazed areas; periodic path cleaning; generating a management plan; coordinating and distributing pastures among ranchers; hiring shepherds to manage livestock; applying prescribed

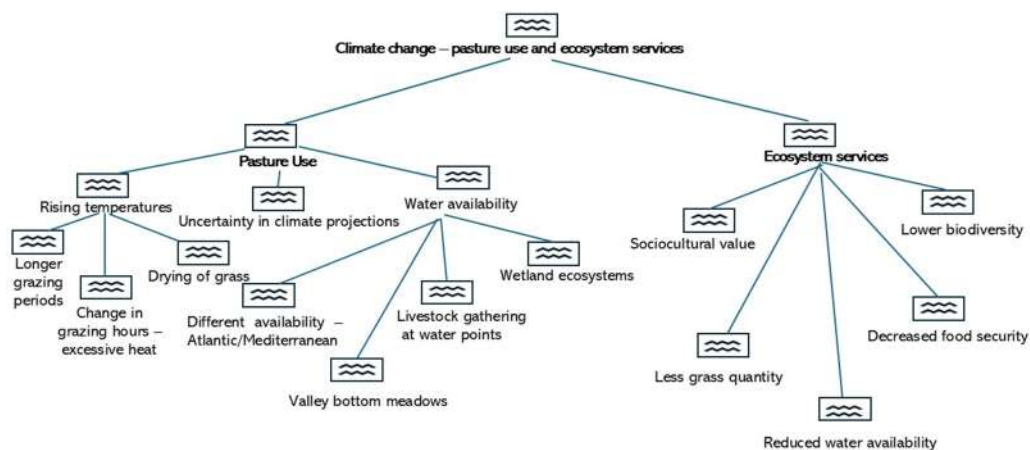


Figure 1. Climate change influencing pasture use and ecosystem services in the NPs. Theme, codes and subcodes represented by boxes.

burns in conjunction with livestock management; installing more watering points for livestock; and directing herds to higher pastures.

Conclusion

Stakeholders' perceptions include awareness of climate change challenges on grassland utilization in high mountains. According to participants, ecosystem services will be influenced by a decrease in biodiversity, grass quantity and water availability. They also mention the importance of providing sociocultural value to the work carried out by shepherds in high mountain areas. Stakeholders propose good practices including a management plan based on grassland rotation and patch cleaning.

Acknowledgement

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Identifying solutions to mitigate GHG emissions and increase carbon sequestration using a multi-actor approach

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Abstract

Reducing greenhouse gas (GHG) emissions and increasing carbon (C) sequestration are important for agriculture to reach our climate targets. The Horizon2020 Innovation Action Project, ClieNFarms, aims to co-develop locally relevant solutions to reach climate neutral and climate resilient farms across Europe. In Ireland, one of the ClieNFarms networks is focused on dairy-beef production systems. A structured workshop using a multi-actor, living lab approach brought together 25 stakeholders from the sector to assess knowledge and discuss solutions to enhance C sequestration and reduce GHG emissions. Workshop participants were provided with 40 solutions and asked to discuss those in terms of likelihood of implementation and impact. Solutions related to slurry spreading and fertiliser application were identified as those with the highest likelihood of implementation. Further discussions provided information on pre-requisites and systemic barriers to four high impact, low likelihood of implementation solutions identified by the stakeholders (reduce herd size, anaerobic digestion, conversion to forestry/nature, renewable energy). Stakeholders identified that cooperation between actors is required to implement high impact, low likelihood of implementation solutions. Overall, farmers were receptive to implementing solutions, however, they require clear guidelines and evidence that solutions will be effective.

Keywords: beef, GHG emissions, grassland, living lab, sustainability

Introduction

European agriculture must reduce greenhouse (GHG) emissions and increase carbon (C) sequestration if the target of reaching climate neutrality by 2050 is to be achieved. A multi-actor approach (Feo *et al.*, 2022) is required to support farmers in innovating, changing and adapting practices to achieve this target. A Living Lab approach places the farmer at the centre, encouraging and supporting actions to be implemented on farm (Bergez *et al.*, 2022; McPhee *et al.*, 2021). Within the Horizon 2020 ClieNFarms Project (clienfarms.eu/), Creative Arenas, based on a living lab type approach, were used for co-creation and user involvement to exchange knowledge and support the development and adoption of innovative solutions to reduce GHG emissions on farms. The aim of the Creative Arena was to initiate a discussion between farmers and other actors in the industry on solutions and practices that can be implemented to reduce GHG emissions and increase C sequestration.

Materials and methods

A structured one-day workshop focused on implementation of a range of solutions to support the reduction of GHG emissions and increase C sequestration on dairy-beef farms was held in Ireland in May 2023, using a similar format to that described by Browne *et al.* (2024). Twenty-five stakeholders attended, including farmers, researchers, advisers, policy makers and industry (meat processors, bankers, agricultural input suppliers). Participants were randomly assigned to one of four groups and guided through a discussion by facilitators. Forty solutions to reduce GHG emissions and increase

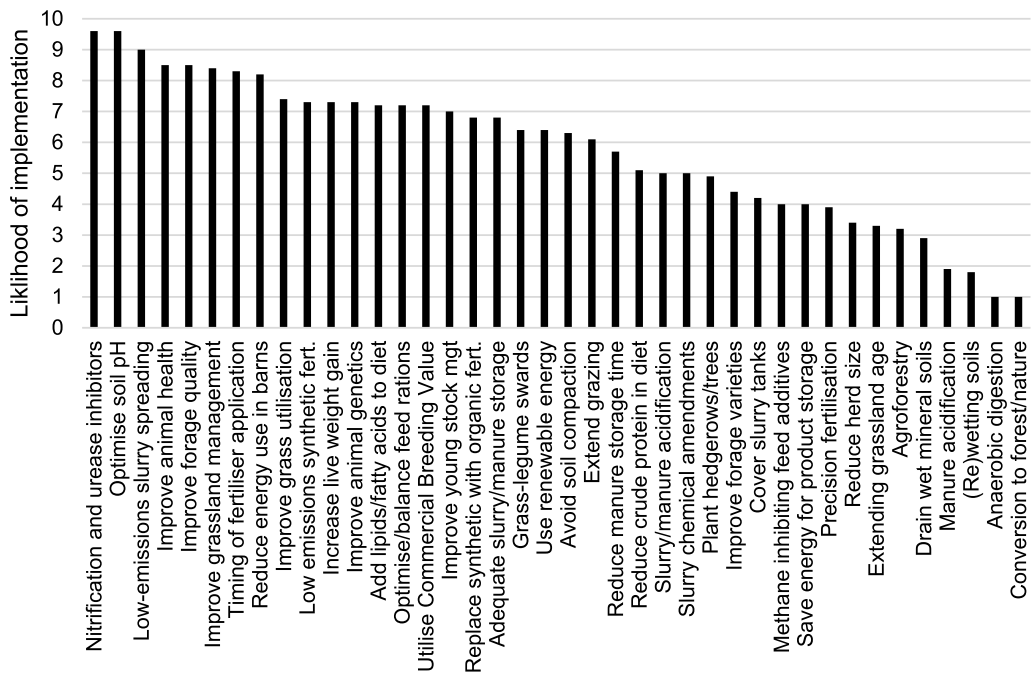


Figure 1. Average likelihood of implementation of each solution from participants (1 = low, 10 = high).

C sequestration were selected from the ClieNFarms list of solutions. Each group was assigned 10 solutions, and each participant in that group was asked to rank and discuss the solutions based on likelihood of implementation using a scale from 1 (low) to 10 high (high). After this, participants ranked solutions on an impact scale from 1 (least) to 5 (most). Prior to the event, four experts involved in grassland systems, GHG emissions and C sequestration research also rated each solution on impact using the same scale, to allow comparisons to be made.

Results and discussion

The solutions with the lowest average likelihood of implementation were anaerobic digestion and conversion to forest/nature (Figure 1). Solutions with the highest likelihood of implementation were all related to optimizing grass production (nitrification and urease inhibitors, improving soil pH and low-emissions slurry spreading (LESS)). Solutions related to fertiliser were rated in the top five for impact by participants (Figure 2), including precision fertilisation, LESS, and timing of fertiliser application. Other top-five solutions were improving young stock management and animal health. The bottom five were mostly grass and feed-related (add lipids/fatty acids, improve grass utilisation, forage varieties, and feed ration), plus reducing energy use in livestock barns. Figure 2 also shows differences in assumed impact between the expert and participant averages. The greatest differences (≥ 2.5) were for precision fertilisation, young stock management, timing of fertiliser to match plant demands, covering liquid manure tanks, and using Commercial Breeding Value; participants rated these higher. No solutions were rated ≥ 2.5 higher by the expert than by participants. Some high impact solutions had low likelihood of implementation, e.g. rewetting of organic soils; an understanding of the challenges and barriers, as well as necessary supports, is required to increase their implementation.

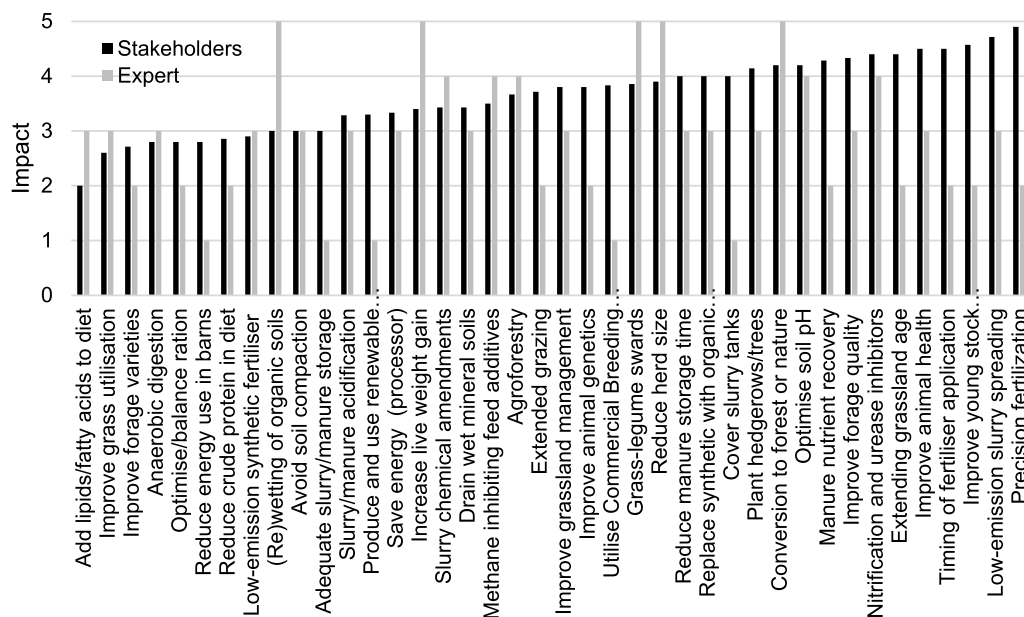


Figure 2. Average assumed impact for each solution (1 = low, 5 = high) for participants compared to the experts assumed impact rating.

Conclusion

The Creative Arena workshop promoted discussion of the impact and likelihood of implementation of solutions to reduce GHG emissions and increase C sequestration on dairy-beef farms by farmers and industry stakeholders. Utilising a multi-actor approach ensured that challenges were looked at from many perspectives.

Acknowledgements

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Endophytes to inhibit *Bothriochloa ischaemum* var. *songarica* germination: a novel tactic for rangeland restoration

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Abstract

King Ranch bluestem (*Bothriochloa ischaemum* var. *songarica*; KRB) is a bunchgrass in the Poaceae family. Native to Europe and Asia, it was introduced to Texas as a drought-resistant forage and used to reduce soil erosion. However, its uncontrolled spread has become an ecological threat, leading to its classification as an invasive species. KRB's rapid growth often outcompetes native grasses, threatening the biodiversity of prairies and rangelands.

This project aims to identify endophytic microbes that influence competitive interactions between native and invasive grasses during germination. The goal is to develop a microbial inoculant that effectively suppresses KRB germination and persistence while promoting native grasses or having no impact on them during restoration of native rangelands. This approach aims to offer a sustainable alternative to herbicides. Native endophytes isolated in 2021 from little bluestem (*Schizachyrium scoparium*; LB) stored in glycerol stock were screened for viability, then tested as an inoculum. Inoculant containing selected endophytes was applied to KRB and LB seeds in petri dishes and incubated in a germination chamber. Germination percentage was compared to assess the effectiveness of the endophytes in altering germination. Whole-genome sequencing was conducted on several impactful endophytes that impact competition between KRB and LB seedlings. Although analysis of variance did not detect statistically significant treatment effects ($p > 0.05$), clear trends emerged. *Pseudomonas canadensis* and *Serratia nevei* reduced KRB germination by 24–25%, while a microbial consortium containing *Delftia tsuruhatensis* and *Pseudomonas spp.* produced the greatest suppression at 29%. In contrast, *Bacillus velezensis* enhanced LB germination by 13%, and the consortium increased it by 8%.

Keywords: endophytic microbes, germination inhibition, invasive species management, King Ranch bluestem, microbial inoculants

Introduction

King Ranch bluestem (*Bothriochloa ischaemum* var. *songarica*; KRB) is a bunchgrass in the Poaceae family, one of the Old-World bluestems which include grasses like *Bothriochloa*, *Dichanthium*, and *Capillipedium* (Wied *et al.*, 2020), with this study focusing specifically on *B. ischaemum* var. *songarica*. This warm-season perennial grass grows 46–122 cm tall and forms dense stands under optimal conditions (Texas Invasive Species Institute, 2014; USDA Forest Service, 2018). Native to Europe and Asia, it was introduced to the United States in the early 20th century for its drought tolerance and forage potential (Clayton *et al.*, 2017; Bull Creek Foundation, 2022) and later became widely promoted as a grazing crop (Clayton *et al.*, 2017). While KRB provides livestock feed, its nutritional value decreases throughout the season (Wied *et al.*, 2020). Its extensive root system also aids in erosion control, and it has been planted along many highway roadsides for that purpose (Hafenrichter, 1959).

Little bluestem (*Schizachyrium scoparium*; LB), a drought-tolerant native grass, is used in restoration projects to improve soil health and combat invasive species (Tober & Jensen, 2013; Sedivec *et al.*, 2009). It also provides fair forage for cattle and supports native wildlife (Tober & Jensen, 2013).

Endophytes, microorganisms that live within plant tissues, can enhance growth and resistance to pathogens (Rosenblueth & Martínez-Romero, 2006). Bacterial endophytes, such as *Arthrobacter*, *Serratia* and *Bacillus*, are known for their beneficial interactions with plants (Khan *et al.*, 2023). The commercial inoculant Exceed, for example, contains *Rhizobium leguminosarum*, which aids in nitrogen fixation (Visjonbiologics, 2024). The objective of this study is to investigate how native endophytes influence the competitive interactions between native and invasive grasses, with a particular focus on potentially inhibiting the germination and establishment of KRB while monitoring native plants for any potential off-target negative impacts. The long-term goal is to reduce reliance on chemical herbicides by developing a sustainable, microbiological alternative.

Materials and methods

This study followed a two-factor factorial design to evaluate the effects of native endophytes on the germination of KRB and LB. Factors included grass species (KRB vs. LB) and inoculation treatment (endophyte vs. uninoculated control), with four replicates per treatment. Each experimental unit was a 100-mm petri dish containing 25 seeds and 45 g of soil, yielding 100 seeds per treatment group. A total of 50 endophytic isolates were initially screened for viability and potential effects on germination.

Endophytic isolates were obtained from LB roots collected in 2021 and preserved at -80°C in glycerol stocks. Prior to use, isolates were revived on Luria–Bertani agar plates and grown in LB broth at 25°C to confirm viability. Microbial suspensions were prepared by culturing isolates to a standard optical density, centrifuging, and resuspending in $0.1\times$ Luria–Bertani broth.

Soil for the germination trials was a fine sandy loam collected from organic field plots at the Texas A&M AgriLife Research and Extension Center (Stephenville, TX, USA), sieved, and stored at room temperature. KRB seeds were collected from Langley Farm (Stephenville, TX, USA), air-dried, cleaned, and examined for maturity under a dissection microscope. LB seeds were purchased from Native American Seed due to limited access to and availability for collection. In each petri dish, 25 seeds were placed on the soil surface, and inoculated treatments received 16 ml of microbial suspension while controls received sterile $0.1\times$ LB broth. Dishes were incubated for 20 days in a Percival growth chamber and watered with reverse-osmosis (RO) water every four days.

To confirm the species of the most effective isolates (over 10% inhibition), DNA was extracted using a Zymo Quick-DNA HMW MagBead kit and sequenced at Eurofins Genomics. Taxonomic assignment was performed by comparing sequences against public databases. Germination percentage was calculated per dish, and treatment effects were analyzed in RStudio using analysis of variance (ANOVA). Where significant differences occurred ($p < 0.05$), Tukey's HSD post-hoc tests were applied.

Results and discussion

The endophytes tested showed varied effects on the germination rates of KRB and LB seeds, but the ANOVA results indicated that the differences were not statistically significant. The most desirable outcomes were observed with endophytes that suppressed KRB while promoting or having no effect on LB germination. *Serratia nevei* (167) reduced KRB germination by about 25% with almost no effect on LB. *Bacillus velezensis* (338–1) slightly reduced KRB but increased LB germination by 13%. The microbial consortium 414 B-1, composed of *Delftia tsuruhatensis*, *Pseudomonas atacamensis*, and *Pseudomonas mercuritolerans*, produced the strongest combined effect by reducing KRB germination by 29% while enhancing LB germination by 8%.

In contrast, some isolates reduced germination in both species, which is less desirable for restoration purposes. *Pseudomonas canadensis* (42BB1) suppressed KRB by 24% but also negatively affected LB germination, making it a weaker candidate for use in promoting native establishment.

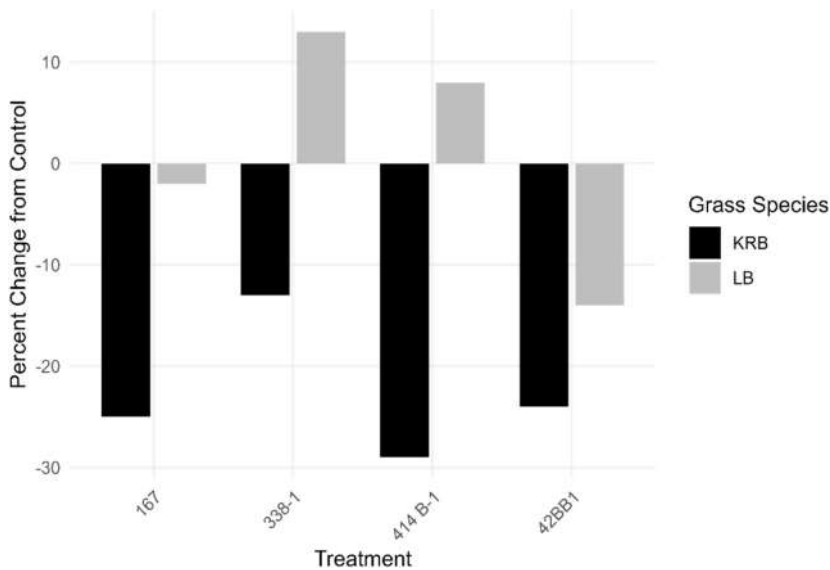


Figure 1. Percent change in germination of KRB and LB seeds inoculated with native endophytes compared to the control. Negative values indicate reduced germination, while positive values indicate increased germination.

Conclusion

These results suggest that certain endophytes may be useful in promoting native grass growth while suppressing invasive species like KRB, offering potential for sustainable land management. While the results were not statistically significant, observed trends indicate that some endophytes may reduce KRB germination without negatively impacting LB. With further testing, increased replication, and future field trials, microbe-based treatments could become a viable alternative to chemical herbicides in native grassland restoration efforts, where the dense seedbed from KRB is a significant challenge for long-term success.

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Long-term trends of both tree and grassland productivity in silvopastoral systems

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Abstract

Dryland silvopastoral systems are primarily composed of grassland and trees. Vegetation productivity within these systems is crucial for sustainable livestock management. While the long-term influence of climate on overall agroforestry productivity has been examined, a clear understanding of the different responses of grassland and trees to climatic conditions at the regional level is lacking. This study aimed to differentiate the productivity baseline between grassland and trees and identify the productivity response to climatic variables in Portugal.

Plant Phenology Index, a remote sensing product with 500 m resolution, was used to distinguish and monitor grassland and trees in Holm oak and Cork oak silvopastoral systems over 20 years. Productivity trends over time for both grassland and tree components were assessed using trend and correlation analysis. Results show no significant or strong trends in the productivity of grassland and trees over 20 years. However, dry years appear to cause a decrease in tree productivity with a delayed response, while grassland showed an immediate productivity decrease. This study uncovers long-term productivity patterns of the two most important components of silvopastoral systems. The observed complementarity in productivity responses of grassland and trees to climatic conditions offers valuable insights for enhancing precision livestock management.

Keywords: dryland, montado, *Quercus rotundifolia*, *Quercus suber*, remote sensing

Introduction

Woodland and grassland in silvopastoral dryland systems determine the forage availability for cattle. The variation of vegetative productivity of trees and of grassland species over time is determinant to better manage livestock. Remote sensing vegetation indices can estimate the differences in productivity at the regional scale and over time (e.g. Ramos *et al.*, 2015). In silvopastoral dryland systems, the strong phenological differences between trees and grassland allow their differentiation through the seasonal cycles. While trees, as evergreen vegetation, remain with a vegetative signal all year round, grasslands show a seasonal pattern. The long-term variability of trees and grassland productivity, and their relationship with climate over time and at the regional scale, needs to be better understood. This study aimed to evaluate the productivity baseline and trend of the tree and grassland components of silvopastoral dryland systems over 20 years, and assess their response to climatic variables in Portugal.

Materials and methods

The study area included all sites classified as Holm oak and Cork oak silvopastoral systems (i.e., 10–60% is covered by Cork oak or Holm oak and understorey with temporary crops and/or pastures), based on the Land Cover Map of Continental Portugal. These sites were selected based on the available data from 1995 to 2018, as having a stable land use, to ensure the same land use over

time (1493 Holm oak data points, 662 Cork oak data points). Annual data on seasonal and total productivity was collected from MR-VPP remote sensing product (Copernicus Land Monitoring Service, 2022). MR-VPP is derived from MODIS products with a 5-day revisit time, from 2000 to 2021, with a 500 m resolution. It uses the Plant Phenological Index (PPI), a dimensionless vegetation index that relates to plant productivity. While PPI seasonal data was used to characterise grassland productivity, the difference between total and seasonal PPI was used to characterise tree productivity (Ramos *et al.*, 2015).

The trend analysis of productivity in each silvopastoral system and vegetation component was determined over 20 years using the Mann-Kendall test (Kendall's Tau). Precipitation and temperature, considered as key climatic variables, were collected from ERA5LAND (Copernicus Climate Change Service, 2022). The sum of precipitation and average temperature of the spring months (April, May and June) for each year was used, as this is the determinant period for vegetative growth in these ecosystems (e.g., Bicho *et al.*, 2024). A Spearman correlation analysis was then performed for tree and grassland productivity among the spring months of the current year and the previous year.

Results and discussion

Long-term productivity trends at the country level showed a weak but significant positive correlation with tree productivity for Holm and Cork oak land uses (Mann–Kendall test; $\text{Tau}_{\text{Holm oak}} = 0.0802$, $p < 0.05$; $\text{Tau}_{\text{Cork oak}} = 0.150$, $p < 0.05$) and no significant trend in grassland productivity (Mann–Kendall test, $\text{Tau}_{\text{Holm oak}} = 0.09$, $p > 0.05$, $\text{Tau}_{\text{Cork oak}} = 0.221$, $p > 0.05$) over 20 years. Other studies report the existence of no trend or a decreasing trend in these ecosystems' productivity over time (e.g., Santos *et al.*, 2016, Aubard *et al.*, 2019), showing a stable to declining productivity pattern. In contrast, our results suggest a weak significant positive trend of tree productivity, representing an increase in tree canopy density.

Cork oak systems showed a higher tree productivity than Holm oak (Figure 1). Holm oak silvopastoral systems dominate in drier regions where overall vegetation productivity and tree cover tend to be lower than in more mesic areas (Santos *et al.*, 2016). However, the differences in grassland productivity between the two silvopastoral systems were much smaller.

Spring (April, May and June) temperature and precipitation were significantly correlated with tree and grassland productivity in both Holm oak and Cork oak systems, but with a higher magnitude in grassland productivity (Table 1). Overall, spring temperature showed stronger correlations with grassland productivity than with tree productivity, while precipitation had stronger correlations with tree and grass productivity. The previous year's spring precipitation showed stronger correlations with tree productivity than the spring precipitation of the current year. Conversely, grassland productivity was more strongly correlated with the precipitation of the current year than of the previous year. Most grassland species have an annual cycle, which is adapted to precipitation seasonality, being more responsive to annual spring precipitation (Liu *et al.*, 2021). Our results suggest that the tree productivity of slow-growth, evergreen trees, such as Holm and Cork oak, have a lagged response to precipitation and temperature. This lagged response was already reported in tree-ring analysis for other oak species from southeast Europe (Stojanović *et al.*, 2018).

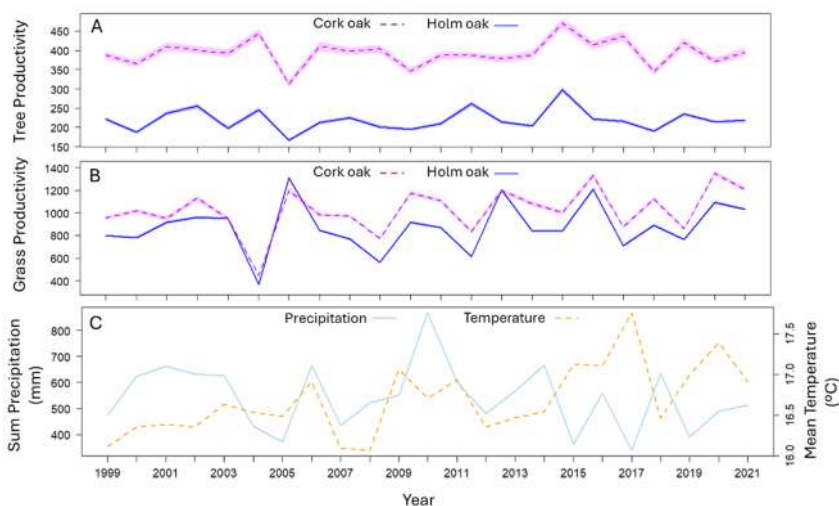


Figure 1. (A) Average tree productivity (PPI) over time. (B) Average grassland productivity (PPI) over time. (C) Annual mean temperature and sum of precipitation for the studied sites from 1999 to 2021. The shaded polygon represents the 95% confidence intervals of each variable for the period 1999 to 2021.

Table 1. Spearman correlation of precipitation and temperature of spring months (April, May and June) for the current and previous year with tree and grassland productivity on Holm oak ($N = 1493$) and Cork oak ($N = 662$) silvopastoral systems.

	Year	Tree productivity		Grass productivity	
		Holm oak	Cork oak	Holm oak	Cork oak
Mean temperature	Current	−0.02	0.04	−0.15	−0.16
	Previous	−0.07	−0.08	0.12	0.04
Sum precipitation	Current	0.07	−0.02	0.21	0.29
	Previous	0.14	0.11	−0.2	−0.09

All Spearman correlation coefficients presented are significant ($p < 0.05$).

Conclusion

Dryland silvopastoral systems are under great pressure due to climate change coupled with land use intensification, making it essential to understand their short and long-term response. This work shows that spring precipitation plays an important role in the productivity of both trees and grassland vegetation. While dry years tend to cause a decrease in tree productivity with a lagged effect the following year, grassland vegetation showed a more immediate decline in response to drought in the same year. The differentiated productivity responses of these essential components of silvopastoral systems to climate conditions, both in the short and long term, provide valuable insights for enhancing precision livestock management in these dryland Mediterranean areas.

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Closer-to-nature forest management: An integrated approach combining grazing, prescribed burning and silvocultural treatments to enhance forest wildfire resilience and ecosystem sustainability.

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Abstract

Mediterranean oak ecosystems are increasingly vulnerable to wildfires due to climate change, land abandonment and historical fire suppression practices. Traditional firefighting strategies have proven inadequate in the face of escalating fire severity. This study presents a holistic closer-to-nature management approach in a native Pyrenean oak (*Quercus pyrenaica*) forest, at Quinta da França, in Portugal. The focus is to promote natural ecological processes with agro-silvo-pastoral treatments that lead to increased forest ecosystem resilience against wildfires. Different treatments were applied as nature-based solutions, namely, prescribed burning, targeted livestock grazing areas and selective silvocultural practices, for understorey fuel load control, not impacting mature tree maintenance and regrowth. Remote sensing monitoring was applied and data collection from forest inventories and field vegetation surveys was used. The results have been evaluated both qualitatively and quantitatively and the case study demonstrates how this strategy can promote a multifunctional forest structure that is more resilient to fire. The study stands as a replicable model of integrated, sustainable land management for other mediterranean forest regions, aligned with the European Union's forest and biodiversity strategies for 2030.

Keywords: closer-to-nature forest management, integrated fire management, nature-based solutions, wildfire risk reduction

Introduction

The Mediterranean region faces increasing wildfire threats exacerbated by rising temperatures, changing precipitation patterns, land abandonment and expansion of the wildland–urban interface. Native Pyrenean oak forests (*Quercus pyrenaica*), characteristic of sub-Mediterranean mountainous regions, are shaped by long-term silvo-pastoral use. However, fire regimes have shifted, becoming more frequent and intense due to the accumulation of fine biomass, mostly due to the abandonment of traditional agro-silvo-pastoralism, alongside with inadequate management strategies, rooted in suppression rather than prevention. Within this context, the present case study, developed within an EU-funded project—SILVANUS—, explores a case study for synergistic application of integrated fire management (IFM) and close to nature forest management (CTNFM) combined treatments, to develop a landscape-level approach for forest fire prevention and ecosystem management. IFM offers a holistic framework that encompasses prevention, preparedness, response, recovery, and adaptation,

integrating ecological, social, and operational dimensions to manage fire as both a hazard and a natural ecological process (Corona *et al.*, 2015). CTNFM complements this by promoting ecological complexity, native biodiversity, and long-term forest resilience (Kalapodis *et al.*, 2025). These strategies are implemented through targeted silvicultural interventions, including prescribed fire and livestock grazing—both recognized as key nature-based solutions (NbS) for fuel management (Keesstra *et al.*, 2024). The present study focuses on evaluating the effectiveness of these measures in creating a resilient forest structure and reducing wildfire susceptibility (Larsen *et al.*, 2022).

Materials and methods

A 220-ha of native Pyrenean oak forest at Quinta da França (QF) site is managed under an agro-silvo-pastoral framework. Multiple fire mitigation strategies have been implemented over the past 20 years for its sustainability. In 2018 half of the forest was fenced for animal grazing. Part (100 ha) is grazed for part of the year by a herd of 60 beef cattle, while part is ungrazed (120 ha). Vegetation survey plots (40 m×40 m) were installed in both areas. Trees, shrubs, grasses, forbs and bare ground were monitored during annual cycles in 2019–2021. Also, three forest inventories were conducted in 2007, 2013 and 2021, with a point grid survey (250 m×250 m). Remote sensing surveying, with satellite and drones (UAV) was used as complement for high resolution land cover mapping and for vegetation vigour index assessment (NDVI). Different silvocultural treatments, such as prescribed fire, selective grazing and selective tree thinning and pruning, were applied. The intervention areas were applied based on CTNFM principles and combined as a whole to enhance forest structural complexity and fuel discontinuity for wildfire resilience. Two main NbS results were analysed: prescribed burning and livestock grazing. Prescribed burning, conducted in January 2024, under expert supervision, was applied in the non-grazed forest area, with a prescribed burning plot (1.4 ha) that was compared with an identical adjacent unburned control plot (1.6 ha), both having similar topology and land cover, and layers assessed for trees, grasses and shrubs. The vegetation structure, biomass, and regrowth were monitored through forest inventories and field plot surveys. The prescribed burning effect was monitored through monthly Sentinel2-derived NDVI time series (2023–2024). All data provided the basis for assessing the interventions' effectiveness in reducing fire risk.

Results and discussion

Prescribed burning treatment seems to result in a significant reduction in shrub and grass biomass during the subsequent dry season. The analysis of the NDVI ratio (prescribed/control), in the summers of 2023 and 2024, revealed a reduction in shrub (4.1%) and grass (3.7%) vigour, and stability for trees (0.4%), suggesting that prescribed fire does not affect the mature trees. NDVI analysis showed a post-burn increase in winter and spring vegetation vigour in the burned plot *versus* the control, indicating stronger herbaceous regeneration and reduced shrub regrowth.

Forest inventories show that tree biomass has increased over the years for the total forest area, particularly in the grazed area (2.6 tons per hectare per year), compared with the non-grazed area (1.7 tons per hectare per year). Mature *Quercus pyrenaica* forests with higher biomass are more stable and resilient (Nunes *et al.*, 2010). Conversely, the average density of trees is higher in the non-grazed area (420 trees per hectare) than in the grazed area (275 trees per hectare). However, the average annual growth rate is faster in the grazed area (7.0%) than in the non-grazed area (0.3%), particularly since the installation of fencing in 2018. These results suggest that cattle do not hinder tree regeneration, or negatively influence the mature trees, but rather contribute to an increase in overall structural complexity and biomass.

Field surveys indicate that cattle could have a marked impact on vegetation structure. Grazing reduced the vertical continuity of vegetation by suppressing tall grasses and intermediate shrubs, thereby limiting the development of ladder fuels (Rouet-Leduc *et al.*, 2021). While shrub cover was not entirely eliminated by cattle, shrub biomass in grazed areas accumulated at a slower rate. Also, shrub cover mapping points to lower shrub cover percentage class (>0–50% of shrub) in the grazed area, comparing to non-grazed area (>50–100% of shrub) (Cherif *et al.*, 2024). Additionally, grazing improved horizontal fuel discontinuity by maintaining open spaces and clearings, which are essential for fire-breaks and firefighting access (Ribeiro *et al.*, 2023; Ribeiro *et al.*, 2024).

Conclusion

The combination of IFM and NbS at Quinta da França seems to be a robust and replicable model for wildfire prevention and forest resilience. Prescribed burning reduces shrub and grass biomass while preserving tree health and soil integrity. Planned grazing complements this by maintaining fuel discontinuities, enhancing forest structure, and supporting biodiversity without impairing regeneration or mature tree growth. Silvicultural interventions, such as targeted pruning and thinning reduce crown connectivity and tree density. All these combined strategies promoted a diverse forest structure, reducing fuel loads and fire risk. NbS interventions with modern monitoring enable proactive fuel management and restoration, with QF's remote sensing and field survey framework ensuring data-driven decisions (Balata *et al.*, 2022). The study suggests that shifting from reactive suppression to proactive, multifunctional forest management is not only feasible but essential in fire-prone Mediterranean landscapes. By aligning with the EU's Forest and Biodiversity Strategies for 2030, the QF model provides a tangible roadmap for sustainable land use, climate adaptation, and community resilience. It underscores the value of combining traditional knowledge with modern technology to guide effective, nature-based wildfire mitigation.

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Peak-season forage performance under a mature agroforestry system in Northern Ireland

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Abstract

The UK government's stated ambition to expand woodlands has facilitated a renewed interest in agroforestry as a sustainable land management strategy, yet there remain concerns amongst livestock farmers regarding its long-term impact on forage production. This study investigated the effect of a mature agroforestry system on peak-season herbage yield and quality, two primary drivers of such scepticism, using new data from a 26-year-old agroforestry trial at AFBI Loughgall, Northern Ireland. Grass performance on three agroforestry fields, consisting of common ash (*Fraxinus excelsior* L.) and permanent pasture, were compared against three adjacent fields with no trees. Grass growth was measured using enclosure cages, with snip samples analysed for nutritive quality using near-infrared reflectance spectroscopy. Botanical composition and soil moisture dynamics were also assessed to elucidate long-term impacts on inter-species resource competition. The results showed that peak-season yield was lower under agroforestry, although the scale of reduction (approx. 28%) was more muted than values previously reported. Neither botanical composition nor key nutritive variables were significantly affected. Peak-season soil moisture was lower under agroforestry, suggesting that a longer grazing season may partially offset peak-season yield reduction through greater whole-season utilisation. The study provides essential local data to evaluate the roles of trees in climate-resilient farming.

Keywords: climate mitigation, grazing, herbage quality, long-term experiment, pasture growth, shading effect

Introduction

As with many European countries, the UK government aspires to expand woodlands to sequester carbon while simultaneously restoring biodiversity. Its seventh carbon budget recommends that the national landmass covered by trees increase from 13% today to 19% by 2040, in order to underpin the legislated transition to a net-zero economy by 2050 (Climate Change Committee, 2025). While this policy landscape has created a renewed interest in agroforestry as a sustainable land management strategy for grassland regions, a large proportion of the farming community remains reluctant to adopt the practice, often due to concerns regarding its long-term impact on forage production and hence the profitability of livestock enterprises (Irwin *et al.*, 2023). The objective of this study, therefore, was to investigate the effect of a mature agroforestry system on peak-season forage yield and forage quality, two primary drivers of such scepticism, thereby providing useful information to support both on-farm decisions and policy design.

Materials and methods

The study was conducted at the long-term agroforestry (LTAF) trial at AFBI Loughgall, Northern Ireland (54°23'53"N, 6°36'41"W; 30 m asl). The site was established in 1989, comprising three fields with common ash (*Fraxinus excelsior* L.) on permanent pasture, and further three fields with permanent pasture alone. The initial density of common ash was 400 stems ha⁻¹, subsequently reduced by thinning to 265, 170 and 128 stems ha⁻¹ in 2004, 2009 and 2023. All six fields are managed

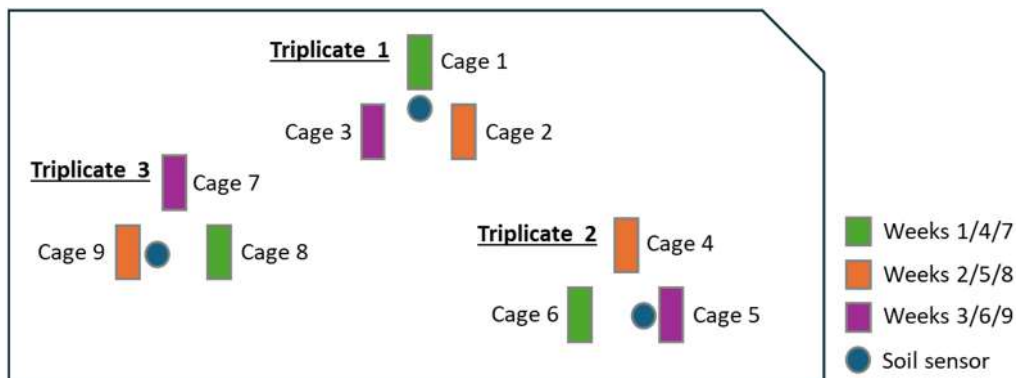


Figure 1. Schematic diagram of the experimental design for each field.

through controlled sheep grazing and receive regular nutrient inputs. The size of individual fields ranged between 0.56–0.80 ha (Fornara et al., 2018).

Each field was instrumented with nine wire-mesh enclosure cages (1.8 m×0.9 m), arranged into three replicates to create a statistically robust replication structure. Triplicates within each field were placed under a geometric layout, with the centroids of triplicates forming the vertices of an equilateral triangle anchored at the centroid of the field (Figure 1). The location of the first vertex was determined by a random compass bearing. Three cages were installed around each vertex, again forming a (smaller) equilateral triangle. All cages were oriented along a north-south axis to minimise the directional bias, and a 1.0 m-wide discard area was established along the perimeter of each cage to create an enlarged plot and hence reduce the border effect. One plot per triplicate was equipped with an SM150T soil sensor (Delta-T, Cambridge, UK) to continuously measure volumetric soil moisture (%).

Grass was cut on a 21-day rotation to simulate a typical grazing practice in the study region. A pre-season trim was conducted on 10th/17th/24th April 2025, respectively, for each of the three plots per triplicate. Data from nine weekly grass cuts thereafter (1st May–26th June inclusive), or three simulated grazing events per plot, were used for subsequent analysis. The 9-week window here was selected to capture the peak grass growing season in the study region, based on 25-year data from the GrassCheck pasture monitoring programme (Patterson *et al.*, 2025). For herbage yield, two parallel strips (8 cm×1 m each) were hand-sheared across the centre of the cage and weighed using a precision balance. Foreign materials identified inside the cage (including leaves and branches from common ash trees) were removed prior to shearing and thus did not contribute to the yield. A subsample was oven-dried at 85°C for 20–24 h and weighed again to obtain dry matter content. For forage quality, weekly fresh samples from each field (across three plots cut) were pooled to create a single representative sample. These samples were analysed for metabolisable energy (ME), crude protein (CP), water-soluble carbohydrates (WSC) and acid detergent fibre (ADF) using near-infrared reflectance spectroscopy. Residual biomass in the plot was mowed and discarded after each sampling.

Botanical composition of the sward was assessed 3–6 days before the pre-season trim. An approx. 50 g herbage sample from each plot was collected and manually sorted into five groups: perennial ryegrass (*Lolium perenne*), meadow grass (*Poa* spp.), bromes (*Bromus* spp.), bentgrass (*Agrostis* spp.) and others (including dead materials; collectively contributing <5%). Again, a subsample was oven-dried and

Table 1. Mean herbage growth ($\text{kg DM ha}^{-1} \text{ day}^{-1}$) and 95% confidence interval under different treatments and cuts as predicted by the mixed-effects model.

Treatment/cut	Predicted mean	Lower limit	Upper limit
Trees/1	48.1	38.2	57.9
Trees/2	46.1	36.3	56.0
Trees/3	28.1	18.2	38.0
No trees/1	52.0	42.1	61.8
No trees/2	57.0	47.1	66.8
No trees/3	62.0	52.2	71.9

reweighed to calculate dry matter content. Species identification followed the Field Studies Council guidelines and used ligule morphology, leaf folding, surface texture, and colouration.

Three-week yield (converted to $\text{kg DM ha}^{-1} \text{ day}^{-1}$) were analysed using a linear mixed-effects model regression, with a fixed model structure of treatment (trees/no trees) $\times$ cut (1st/2nd/3rd) and a random model structure of field/triplicate. Forage quality variables were analysed in a similar manner but with only field as a random effect. Treatment effect on botanical composition was evaluated by Dirichlet regression to account for the error distribution structure associated with proportional data.

Results and discussion

The average yields across the whole peak season with and without trees were 40.8 and $57.0 \text{ kg DM ha}^{-1} \text{ day}^{-1}$, respectively. The majority of this difference stemmed from the third cut (Weeks 7–9) when the tree canopy became more mature and thus was likely to have intercepted more light (Table 1). As a result of this seasonality, the first-order treatment effect was not statistically significant ($p = 0.555$). In contrast, the first-order effect of third cut and the interaction between treatment and third cut were both significant (both $p < 0.001$).

None of the nutritive variables showed a significant first-order treatment effect (all $p > 0.1$). Across both treatment and control, the sample averages and standard deviations were 11.5 ± 0.5 (ME), 17.9 ± 2.4 (CP), 13.0 ± 3.1 (WSC) and 28.1 ± 2.8 (ADF) % DM, respectively. Botanical composition was also unaffected ($p > 0.2$ for all species groups). Across both treatment and control, the sample averages and standard deviations were 26.7 ± 22.1 (perennial ryegrass), 8.2 ± 14.8 (meadow grass), 26.5 ± 27.9 (bromes) and 33.8 ± 25.3 (bentgrass) %, respectively.

The present finding of a $\sim 28\%$ yield reduction is relatively more muted compared to those commonly reported (e.g. Ehret *et al.*, 2015), including a recent modelling study for the same study site that predicted an approx. 80% reduction after 25–30 years of agroforestry (Giannitsopoulos *et al.*, 2025). This is an encouraging result in light of the current policy landscape, particularly as a tendency was observed for peak-season soil moisture to be lower under agroforestry (36.6 vs 38.3%). Should the pattern continue into the non-peak season, this suggests that a longer grazing season owing to improved trafficability may partially offset peak-season yield reduction through greater whole-season herbage utilisation. Care should be taken, however, at inter-study comparison, as tree species and density often vary and, furthermore, few studies provide a seasonal breakdown of the annual yield to allow estimation of peak-season production.

Conclusion

This study has provided essential local data to evaluate roles of trees in sustainable and climate-resilient farming, underpinning informed decision-making by farmers and policymakers alike.

Acknowledgements

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Agroforestry systems adapted to the pastoral context of the Great Taznakht Southeastern Morocco

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Abstract

Agriculture and livestock farming are impacted by climate constraints and water scarcity in many regions of Morocco. The Great Taznakht area, located in Southeastern Morocco, faces years of drought. Agricultural practices are often inadequate, water resources are scarce and arable land is limited, resulting in modest farm incomes. The objective of this study is to assess the feasibility of implementing agroforestry systems (AFS) adapted to local conditions, using a structured, three-phase approach. First, we conducted an exploration and analysis of the edaphic and climatic characteristics of the study area, as well as monitoring vegetation cover and cultivated land, with ArcGIS and Google Earth. Second, we examined stakeholders' perceptions through interviews and focus groups with farmers, herders, and technicians. These interviews provided insights into motivations for adopting systems and highlighted the advantages and disadvantages of each system. Third, we synthesized biophysical and social findings in a SWOT analysis to identify strengths, weaknesses, opportunities, and threats. Based on these findings, two AFS models adapted to the conditions of each zone were proposed, integrating feedback from local stakeholders. However, the success of these AFS depends on the commitment and collaboration of stakeholders, with a long-term vision geared towards environmental sustainability.

Keywords: climate change, arid zone, GIS, SWOT, water scarcity

Introduction

The Great Taznakht region is heavily impacted by climate change, with recurrent droughts and progressive soil degradation. This area is at high risk of desertification, further exacerbated by intensive cultivation, overgrazing, and the uncontrolled exploitation of medicinal and aromatic plants (MAPs). The Great Taznakht region is predominantly pastoral: rangelands cover 326 878 ha. The Utilized Agricultural Area (UAA) totals 38 991 ha, of which 34 822 ha are rainfed and 4169 ha irrigated. Analysis of cumulative rainfall variability from 2016 to 2024 shows a peak of 183.5 mm in 2018–2019, followed by a sharp decline, reaching a minimum of 38 mm in 2019–2020 and zero rainfall in 2023–2024 (ORMVAO, 2024). This alarming trend toward increased drought underlines the urgent need to adopt resilient agricultural practices, such as agroforestry, to ensure the sustainability of agricultural systems in the context of climate change and water scarcity.

In this context, sustainable and resilient agroecological practices are essential to preserve natural resources and sustain subsistence farming. The overall objective of the study is to identify and conceive agroforestry systems that are most appropriate and adapted for the ecological and social context of the Great Taznakht.

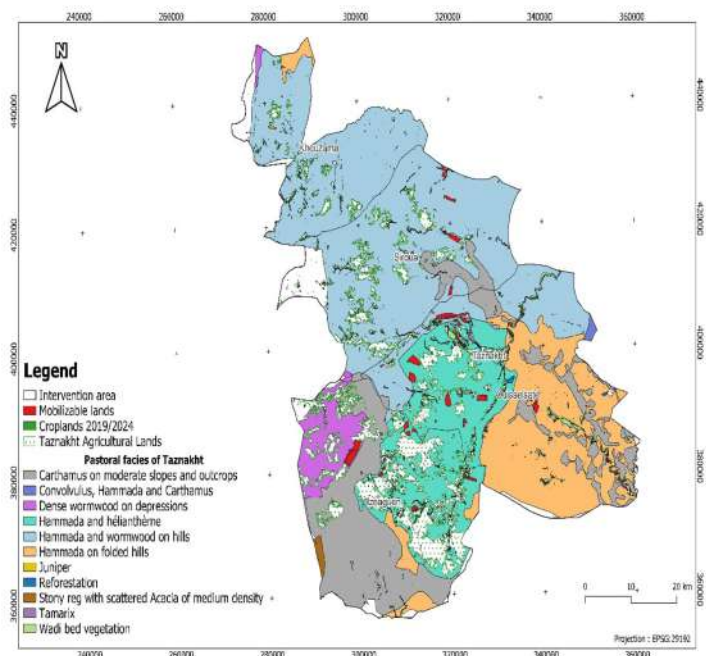


Figure 1. Identification of areas suitable for agroforestry within the study area.

Materials and methods

The study followed a three-phase mixed-methods design comprising exploration, participatory prospective assessment, and synthesis/conceptualization to assess the feasibility of agroforestry systems for Great Taznakht.

- (1) Exploration phase: A structured literature review was conducted (Scopus, Web of Science, and Google Scholar; 2000–2025; English and French) using Boolean queries combining agroforestry, silvopastoral systems, and dryland farming with Morocco, Maghreb, or Drâa-Tafilalet, and thematic focuses on soil, water, rangeland, and desertification. In order to assess land suitability for agroforestry, we analysed the rangeland-units map (Figure 1) to characterize each unit's structural and ecological attributes (e.g., vegetation composition and cover, soil and slope conditions, water availability) and used these descriptors to delineate areas most compatible with agroforestry options (Yessef, 2021).
- (2) Prospective phase: Participatory diagnosis to assess the acceptability of agroforestry systems among local populations. The discussions helped identify the most suitable plant species and practices for the region, while considering both social and ecological challenges; data were collected through semi-structured interviews and two focus groups with 10 and 20 persons among farmers, herders, and commune representatives, then transcribed and thematically analysed.
- (3) Synthesis and conceptualization phase: results of the prospective phase, combined with insights from the bibliographic review, served as basis for formulating concrete recommendations. Sketches illustrating the configurations of the proposed systems, integrating interactions between species and terrain specificities, were developed.

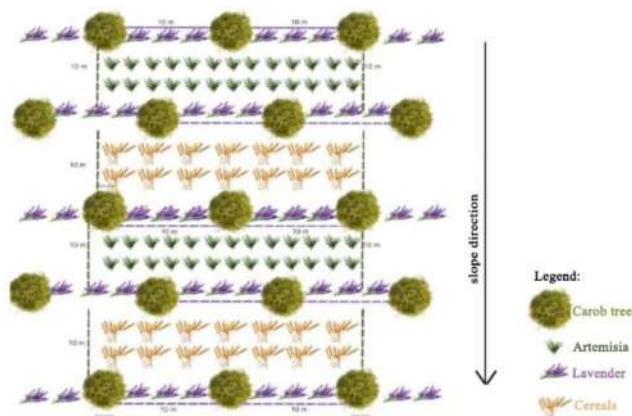


Figure 2. Recommended system for *Hammada scoparia* associated with *Artemisia herba alba* on hills, and *Hammada scoparia* associated with *Helianthemum lippii* on hillslopes.

Results and discussion

In-depth analysis enabled the identification of areas most suitable for the establishment of agroforestry systems, taking into account the environmental conditions specific to each unit.

Three main pastoral units suitable for agroforestry systems were identified:

- (1) *Artemisia herba-alba* in lowlands: shallow soils covered by *Artemisia herba-alba* vegetation, with high agroforestry potential thanks to good water retention;
- (2) *Hammada scoparia* associated with *Artemisia herba-alba* on hills: shallow and saline soils;
- (3) *Hammada scoparia* associated with *Helianthemum lippii* on flat land: Flat topography and deep soils.

The choice of plant material is based on rigorous criteria, combining recommendations from local residents—who possess extensive experience with native plants—and in-depth bibliographic research (Rejeb, 1995; Nguyen *et al.*, 2013; Nair, 2011).

Subsequently, the SWOT analysis screened and prioritized these options by triangulating the bibliographic review, mapping outputs, and stakeholder inputs. The highest-scoring criteria drought tolerance, soil/erosion protection, fodder security, and added value.

This approach aims to identify species best adapted to the region's arid conditions. Based on a mapping analysis of soils and local bioclimatic conditions, two agroforestry systems (AFS) specifically suited to the identified land typologies were proposed with different species combinations and orientations.

The first system consists of associating *Atriplex halimus*, *Helianthemum lippii*, *Artemisia herba alba* and barley. It is a forage-oriented system. The second system is composed of *Ceratonia siliqua*, *Lavandula dentata*, and *Artemisia herba alba*. It is an agroforestry/aromatic and medicinal-oriented system. This system focuses more on perennial trees and aromatic plants, with higher value-added products (carob pods, essential oils, medicinal plants, truffles).

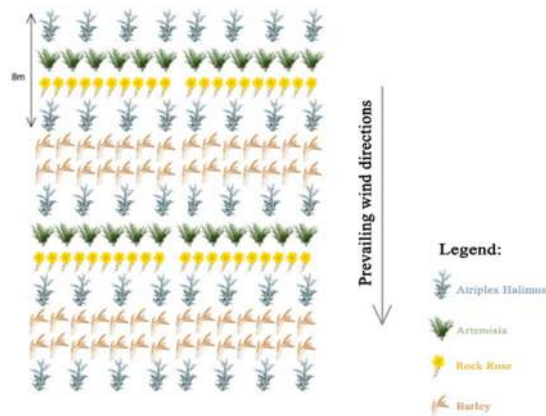


Figure 3. Recommended system for *Artemisa herba alba* in lowlands site.

AFS 1: Hillslopes (carob+lavender+*Artemisia*) We established *Ceratonia siliqua* with *Lavandula dentata* and *Artemisia herba-alba* in $60 \times 60 \times 60$ cm pits, each fitted with a basin and an upstream V-rill to harvest runoff. During establishment, we irrigate $25\text{--}30$ l plant⁻¹, five applications at 15-day intervals. Carob is spaced 10–20 m ($\approx 25\text{--}45$ trees ha⁻¹), while lavender and *Artemisia* are set at 40–60 cm within the inter-rows.

AFS 2: lowlands (*Atriplex* belts+*Helianthemum*+cereals/MAPs) We deployed *Atriplex halimus* as perimeter and internal windbreak belts (row spacing 4 m and 8 m) to frame strips of cereals and MAPs; during establishment, we applied $10\text{--}15$ l plant⁻¹ every two weeks.

Helianthemum lippii is arranged in lines at ≥ 2 m; where applicable, we inoculated with *Terfezia clavervii* and provided moderate irrigation in winter–early spring. Belt orientation minimises wind fetch, and micro-catchments were added where feasible to enhance soil moisture.

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Diet acceptance of typical wetland plants (helophytes) by grazing water buffaloes

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Abstract

Water buffaloes (*Bubalus bubalis*) are increasingly used for the restoration of wet grassland sites in Germany. Their landscape conservation performance depends largely on the degree of grazing competitive helophytes like reeds, sedges and rushes that tend to dominate and reduce biodiversity. To clarify disputed information about water buffaloes' grazing behaviour and landscape conservation services, we investigated the diet acceptance of typical helophytes by a combination of biting scores and GPS-tracking methods at two coastal wet grassland sites during a summer grazing period. We found neither avoidance nor a preference for grazing activities in areas with high helophyte coverage. Biting scores show that reed was preferred over rushes ($p < 0.01$). However, rush was not excluded from the grazing diet. We found a significant advantage of water buffalo over cattle for soft rush acceptance ($p < 0.01$) and thus, rush suppression. There were no indications of a general preference of water buffalo for helophytes. Their partial inclusion in the pasture ration of water buffaloes is apparently due to their general physiological strategy of energy intake (compensation of energy shortages by higher intake) and herd behaviour (more gregarious than scattered). This study demonstrates the suitability of water buffalo for a targeted landscape management in coastal wetlands.

Keywords: *Bubalus bubalis*, grazing preference, phytodiversity, reed, soft rush, wetland grazing

Introduction

Low stocking densities lead to a high proportion of spurned plants in productive grazing areas that require re-mowing to maintain the sward in a good condition. This problem is particularly significant on wet grasslands where, on the one hand, competitive helophytes like sedges, reed belts, and rushes occur and, on the other hand, it is not always possible to use common mowing equipment when water tables are high. Dominant stands of reed plants suppress less competitive and light-demanding plant species, thereby reducing biodiversity. The nature conservation administrations advise grazing with small-framed cattle breeds (such as Highlands or Galloways) and limited stocking densities (0.8 to 1.4 livestock units per hectare) to minimise trampling damage. However, the rigid grazing selectivity of these breeds lead to an undesirable increase in rushes, tall sedges and reeds over time. In this context, the introduction of water buffaloes (*Bubalus bubalis*) is increasingly being discussed as an alternative option for wet grassland restoration in Germany (Sweers *et al.*, 2013a). It is assumed that they bite more intensively on helophytes, thereby achieving the desired landscape conservation effect (Wiegleb & Krawczynski, 2010). So far, the knowledge about water buffaloes feeding behaviour is mostly anecdotal and suffers from a lack of evidence. Therefore, this study aimed to survey the acceptance of typical problematic wetland plants by grazing water buffaloes under the conditions of South-Baltic coastal wetlands.

Materials and methods

All surveys were conducted at two coastal wet grassland sites in Western Pomerania at the South Baltic Sea shore (Wieck, 54°24'9.6"N, 12°33'57.2"E; and Mönchow, 53°50'32.1"N 13°53'23.3"E).

Table 1. Biting score scheme applied to erectophile wetland plants

Score	Meaning	Description
0	No biting	Single plant/tussock completely avoided
1	Light biting	Only plant tops removed
2	Medium biting	Tops and remarkable parts of leaves/stems removed
3	Heavy biting	Large parts of the plant/tussock removed

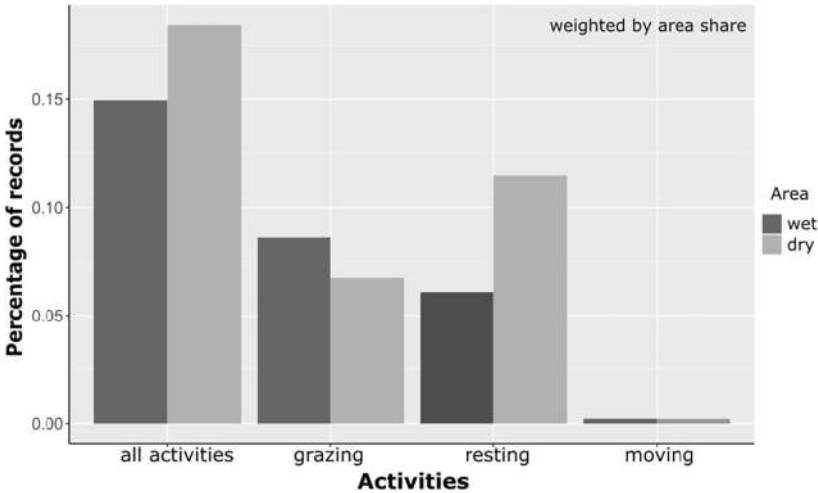


Figure 1. Main activities and space use of a GPS-tracked water buffalo cow depending on the degree of soil wetness at the heterogeneous wet grassland site Mönchow.

River-type water buffaloes of the Murrah breed were used for summer grazing after the first cut was conserved for winter feeding. An adapted continuous free-ranging pasture with a stocking density of 1.1–1.3 LU/ha was practised. At the Wieck site, a neighbouring pasture stocked with beef cattle cross-breed heifers (1.3 LU/ha) served as a comparison treatment. Biting intensity was used as a measure of grazing preference. An equidistant, four-stage scoring scale was applied for assessment (see Table 1). Randomly selected individual plants along transects were assessed according to this scale during five field campaigns (20 July 2023, 9 August 2023, 23 August 2023, 7 September 2023, 24 September 2023).

At the Mönchow herd, a leading buffalo cow was equipped with a GPS collar (Vectronic-Aerospace, Berlin, Germany) to analyse their space use with grazing activity under contrasting soil wetness as derived from a digital elevation model and verified by vegetation analysis (wet sites: mean annual water level <45 cm below ground, dry sites: ≥ 45 cm).

Cumulative link models (CLM) were applied to explore the effects of kind of animal on the biting intensity and to examine the degree of wetness on the behaviour patterns. Resulting models were compared and tested for significance of the factorial effects by sequential deviance analysis.

Results and discussion

The dry areas of the pasture were significantly preferred for resting, while the wet areas were even slightly preferred for feeding (see Fig. 1).

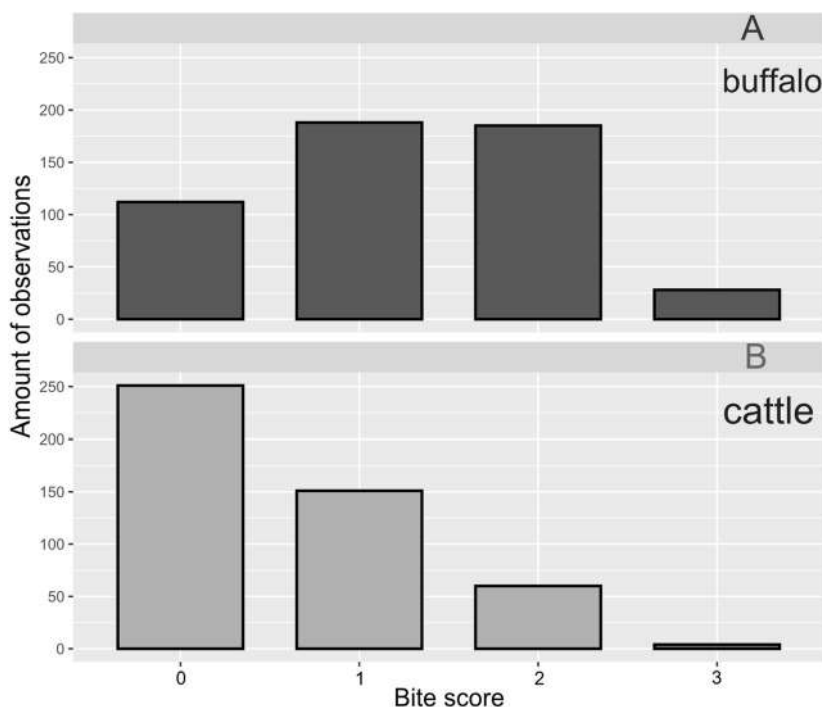


Figure 2. Biting intensity of rushes (*Juncus effusus*) by water buffalo (A) compared to cattle (B) under the same conditions at the Wieck site ($n = 924$, $p < 0.01$).

At the Wieck site, we found significant differences in the biting intensity patterns between water buffaloes and cattle for the helophyte soft rush (see Fig. 2). We were able to confirm the high preference of water buffaloes for reeds (*Phragmites australis*) already observed by Sweers *et al.* (2010b). At the Mönchow site, sedges were also bitten, but not grazed down to the base of their shoots. In contrast, *Ranunculus sceleratus* was completely avoided.

Conclusion

This study demonstrates the suitability of water buffalo for a targeted landscape management in coastal wetlands aimed at dominance breaking of competitive helophytes to foster biodiversity. For animal welfare reasons, it is recommended to include higher, dry pasture areas in wetland grazing systems.

Acknowledgement

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Moderate grazing intensity enhances regrowth without altering spatial heterogeneity in a perennial ryegrass pasture

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Abstract

The key to sustainable pasture management is balancing the needs of grazing herbivores with preserving vegetation condition and growth potential over time. One factor that influences this balance is the spatial heterogeneity of grazed vegetation, a consequence of grazing behaviour and a driver of forage harvesting efficiency in animals. This study assessed how grazing intensity (GI) affects spatial heterogeneity of sward structure and post-grazing regrowth. The experiment was conducted in spring-summer 2022 on a *Lolium perenne* pasture. It compared low GI (25% sward height reduction) to moderate GI (50%) on 180 m² paddocks grazed by sheep in a rotational system with a 3-day occupation time. GI treatments were applied by adjusting herd size, three sheep for low GI and six for moderate GI. Normalized Difference Vegetation Index (NDVI) data, acquired using unmanned aerial systems, were combined with ground-based sward height measurements. Over two grazing cycles (C1 and C2), Moran's global ($P = 0.16$) and local ($P = 0.84$) indices, plus semivariance analyses, revealed increased spatial heterogeneity across paddocks, with no significant differences between GI treatments. However, moderate GI tended to favour regrowth during C2 (0.16 vs 0.08 cm day⁻¹, $P = 0.02$), suggesting more efficient forage utilization under moderate grazing.

Keywords: grazing intensity, grazing management, pasture regrowth, sward height

Introduction

Grazing animals do not harvest herbage uniformly: selective biting, movement between feeding stations, and asynchronous regrowth create vegetation patches at different stages, which shape both pasture productivity and intake efficiency (Rook *et al.*, 2004). Conventional post-grazing targets aiming to maximize harvest rates in rotational settings help maintain a homogeneous sward, but lead to low post-grazing heights, slow plant recovery and reduced intake rates. The “rotatinuous” stocking method proposes to reconcile efficient forage collection with rapid plant regrowth by setting higher post-grazing sward height targets to maintain a high animal intake rate (Savian *et al.*, 2020). Nevertheless, such a strategy leaves more ungrazed vegetation after paddock occupation, with potential short-term effects on sward heterogeneity during subsequent grazing-regrowth cycles. To test this hypothesis, we compared the effects of two post-grazing targets (25 vs. 50% sward height reduction) on sward spatial heterogeneity and plant regrowth in a ryegrass pasture rotationally grazed by sheep.

Materials and methods

The experiment was conducted on a 1.1 ha perennial ryegrass pasture at Gembloux Agro-Bio Tech (Belgium) in spring-summer 2022. Twenty 18 × 10 m paddocks were rotationally grazed by 6-month-old Charolais ewe lambs with a 3-day occupation time. Two treatments were applied: starting from the same pre-grazing sward height (SH) of 12 cm, the moderate grazing intensity (GI) treatment (T2,

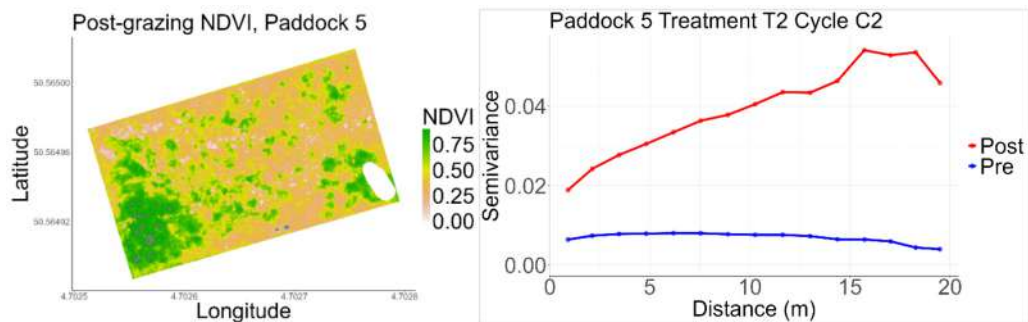


Figure 1. Post-grazing NDVI map of paddock 5 (T2) and corresponding semivariogram, cycle 2.

50% GI) had a post-grazing SH of 6 cm while the low GI treatment (T1, 25% GI) had a post-grazing SH of 9 cm. Lambs were weighed at the beginning and end of each grazing cycle, which corresponds to the time it takes for animals to return to the same paddock. They had an average weight of 41 kg at the start of cycle 1 and 43 kg at the start of cycle 2, with no difference between treatments.

Each treatment was monitored over two grazing cycles: 18 and 30 days (low GI) vs. 24 and 36 days (moderate GI). SH was monitored at 40 georeferenced points per paddock using a sward stick before and after grazing. Multispectral cameras embarked in an unmanned aerial system (DJI P4, 35 m flight height) captured NDVI rasters (1.4 cm resolution; 5.4×10^5 pixels per paddock). Heterogeneity was assessed using global and local Moran's I , along with semivariograms derived from NDVI data. The main parameters calculated were the nugget (C_0), reflecting fine-scale variability; the sill ($C_0 + C$), representing total variance; and the range (α), indicating the spatial extent of autocorrelation. Generalized linear mixed models (gamma distribution, log link) tested the effects of treatment, cycle, and moment (pre-/post-grazing) on sward height, NDVI, and spatial metrics, with paddock included as a random factor. The paddock was the experimental unit ($n = 10$ per treatment).

Results and discussion

Pre-grazing NDVI did not differ between treatments (cycle 1: 0.74 ± 0.08 in T1 vs. 0.75 ± 0.08 in T2; cycle 2: 0.73 ± 0.07 vs. 0.75 ± 0.08 ; Table 1; $P = 0.79$ and $P = 0.66$, respectively). After three days of grazing, NDVI declined to 0.66 ± 0.11 in T1 but to 0.57 ± 0.12 in T2 during cycle 1 ($P < 0.01$) and to 0.65 ± 0.10 vs. 0.52 ± 0.16 in cycle 2 ($P < 0.01$), confirming greater biomass removal under moderate grazing (Table 1). Figure 1 (paddock 5, T2) illustrates the sharper colour shift towards yellow/pink and the steeper post-grazing semivariogram that accompanies this stronger biomass removal.

Global Moran's I increased after grazing in every cycle (cycle 1: 0.75 to 0.82 in T1 and 0.74 to 0.83 in T2; cycle 2: 0.77 to 0.87 and 0.79 to 0.91; Table 1). The treatment effect was non-significant for both global ($P = 0.16$) and local ($P = 0.84$) Moran's I , whereas the moment (pre- and post-grazing) effect was highly significant ($P < 0.01$). Local Moran's I and the semivariogram sill followed the same pattern, confirming greater patchiness once animals had grazed. Post-grazing sills, for example, doubled from 7.2×10^{-3} to 15×10^{-3} in T2 (cycle 1) and from 5.6×10^{-3} to 26×10^{-3} in T2 (cycle 2; Table 1). These results show that a 50% height reduction does not homogenize the sward any more than a 25% reduction because lambs create and maintain patches through selective foraging, avoidance of stemmy or dung-contaminated zones and asynchronous regrowth (Brambilla et al., 2013).

Table 1. NDVI, global and local Moran's I, and semivariogram parameters (nugget C_0 , sill $C_0 + C$, and range α) by treatment (T1, T2), cycle (C1, C2), and grazing moment (pre- and post-grazing)

			NDVI	Global Moran's I	Local Moran's I	C_0	$C_0 + C$	α
T1	C1	Pre	0.74 ± 0.08	0.75 ± 0.03	0.57 ± 1.03	0.0038	0.0051	3.19 ± 0.67
		Post	0.66 ± 0.11	0.82 ± 0.03	0.86 ± 2.44	0.0068	0.011	3.89 ± 0.64
	C2	Pre	0.73 ± 0.07	0.77 ± 0.03	0.34 ± 0.96	0.0032	0.0042	3.26 ± 0.93
		Post	0.65 ± 0.10	0.87 ± 0.01	0.59 ± 1.82	0.0062	0.012	6.23 ± 3.42
T2	C1	Pre	0.75 ± 0.08	0.74 ± 0.03	0.49 ± 0.79	0.0038	0.0072	3.07 ± 0.65
		Post	0.57 ± 0.12	0.83 ± 0.01	0.75 ± 1.88	0.010	0.015	3.64 ± 1.46
	C2	Pre	0.75 ± 0.08	0.79 ± 0.02	0.13 ± 0.70	0.0041	0.0056	4.96 ± 2.18
		Post	0.52 ± 0.16	0.91 ± 0.01	0.35 ± 0.96	0.012	0.026	7.20 ± 3.87

Table 2. Average regrowth rate in cm day^{-1} per treatment and cycle

Cycle	Treatment 1	Treatment 2	P value
C1	0.23 ± 0.16	0.10 ± 0.08	0.069
C2	0.08 ± 0.08	0.16 ± 0.07	0.02

Regrowth rates (Table 2) revealed a treatment×cycle interaction. In cycle 1, lightly grazed paddocks recovered faster ($0.23 \pm 0.16 \text{ cm day}^{-1}$ in T1 vs. $0.10 \pm 0.08 \text{ cm day}^{-1}$ in T2, $P = 0.069$). In cycle 2 the ranking reversed, with T2 regrowing twice as fast ($0.16 \pm 0.07 \text{ cm day}^{-1}$ vs. $0.08 \pm 0.08 \text{ cm day}^{-1}$, $P = 0.02$). This crossover suggests compensatory photosynthesis and enhanced nutrient return triggered by the heavier, yet conservative, defoliation (Ye *et al.*, 2020). Twice as many lambs in T2 also redistributed faecal nutrients more evenly, further stimulating second-cycle growth.

Conclusion

Moderate grazing intensity removed more biomass than low intensity. Still, it maintained similar spatial heterogeneity in the short term due to selective foraging and dung avoidance. It also led to faster regrowth in the second cycle. This suggests moderate grazing as an efficient strategy to optimize forage use and animal performance under favorable weather and soil conditions. Integrating spatial metrics into pasture management could help improve the timing of grazing rotations and avoid overgrazing. Further work should assess seasonal dynamics and forage quality to confirm these results in broader contexts and over longer periods of time.

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Session 3.
Breeding for new challenges

Grass and legume breeding for European grassland farming with emphasis on climate change

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Abstract

Forage breeding must respond to region-specific challenges posed by climate change and, to a lesser extent, increasing interest for certain cropping systems (intercropping; organic; grazing) and key quality traits (stem wall digestibility; protein content; water-soluble carbohydrates). In Europe, an increasing importance of legumes is justified by their protein content and nitrogen (N) fixing ability. We expect alfalfa to expand its cultivation northward, while remaining pivotal in southern Europe by emphasizing drought tolerance selection. Tall fescue and cocksfoot will increase their relative importance among grasses due to their stress tolerance. Changes with respect to priority species and breeding traits will occur across a latitudinal gradient and, for frost tolerance, also a longitudinal gradient. Strategic axes of successful breeding programs will encompass: (a) the regional exploitation of the wide adaptive variation from exotic genetic resources such as landraces and wild populations, facilitated by marker-assisted selection and genome-enabled prediction of germplasm characteristics; (b) large reliance on genomic selection, to face the complexity and reduce the cost of combining different stress tolerance traits and key quality traits; (c) phenomics and envirotyping aimed to reduce the selection costs and improve the characterization of target environments. Agroecological studies could unveil selection strategies, adaptive traits and genetic resources needed to breed for specific environments and cropping systems.

Keywords: adaptation to climate change, drought tolerance, genetic resources, genomic selection, intercropping

Introduction

The adaptation to the changing climate, and crop-livestock system modifications aimed to mitigate climate change, could affect substantially grassland farming in Europe. The changing climate implies: (a) an increasing climate unpredictability; (b) an increasing likelihood and severity of drought (also due to decreasing irrigation water), and greater risk of salinization, land degradation, and desertification; (c) a progressive reduction of cold-prone, boreal climates; and (d) an increase of flooding and ice-encasement (in northern regions) (IPCC, 2019; Hari *et al.*, 2020; Caretta *et al.*, 2022). In this context, the vulnerability of pastoral systems to climate change may be very high in Southern Europe and other regions (IPCC, 2019), especially when based on natural grasslands. For example, the changing climate is decreasing both the productivity and the forage quality of natural grasslands in climatically contrasting regions of Italy mainly because of marked changes in species composition (Dibari *et al.*, 2021). Conversely, a predictively longer growing season may offer advantages in Nordic countries, although it may reduce cold acclimation and affect negatively winter survival due to higher temperatures during the longer autumns (Dalmannsdottir *et al.*, 2016; Ergon *et al.*, 2018). The temperature

and CO₂ increase could increase the productivity of grasslands in regions where drought is not an issue (Durand *et al.*, 2010).

Options for livestock systems with large potential for mitigation of climate change include the production of higher-quality feed, lower mineral N fertilization, and better management of grazed land (IPCC, 2019; FAO, 2010). A likely reduction of animal-based food in human diets could concur to mitigation (IPCC, 2019). However, a sharp reduction of crop-livestock systems is not conceivable, when considering (a) the large number of ecosystem services that these systems provide with respect to nutrient cycling, landscape and biodiversity preservation, carbon sequestration, control of erosion, reduction of wildfires, etc. (Neely *et al.* 2009; Peeters, 2009; Barbieri *et al.*, 2021), (b) the huge economic importance of these systems to produce food products with high added value, and (c) especially in subsistence agriculture, the importance of animal rearing as a buffer against economic shocks or crop failures and for reutilization of farm resources (FAO, 2010).

Greater inter-specific diversity of forage crops could be essential to cope with climate hazards and reduce the risk of substantial crop failure. In particular, legume-grass intercropping could be important for both adaptation and mitigation of climate change, to improve the yield, yield stability and forage quality and to reduce the use of inputs and greenhouse gas (GHG) emissions (Lüscher *et al.*, 2014; Martin-Guay *et al.*, 2018). A moderate species diversity (e.g. four or five species) should be sufficient to provide agroecological and production advantages (Volaire *et al.*, 2014; Lüscher *et al.*, 2022; Bybee-Finley *et al.*, 2023), especially in stress-prone and climatically unpredictable environments (Isbell *et al.*, 2015).

Plant breeding can be pivotal for adaptation to and mitigation of climate change with respect to grassland farming (IPCC, 2019; Rognli *et al.*, 2021). Selecting stress tolerant, resilient cultivars, especially those with greater drought tolerance, is of paramount importance in several European regions to safeguard the production of temporary grasslands and to provide an alternative to natural grasslands whose production has been disrupted by climate change and its associated effects (e.g., overgrazing caused by decreasing forage resources). Specific breeding for intercropping may be required to obtain balanced and productive legume-grass mixtures (Annicchiarico *et al.*, 2019).

Major target species

A greater breeding effort on grain and forage legumes is expected in the next decades (Rubiales *et al.*, 2021). For forage legumes, this is justified by many benefits arising from their cultivation, such as improved forage quality, greater feed protein self-sufficiency, and reduction in the use of N fertilization and GHG emissions (Lüscher *et al.*, 2014; Cellier *et al.*, 2015; Julier *et al.*, 2017; Dineen *et al.* 2018). Alfalfa (*Medicago sativa* subsp. *sativa*) is the main perennial forage legume in temperate regions of Europe and elsewhere, especially where farming systems rely substantially on forage conservation (Annicchiarico *et al.*, 2015a). A recent comparison of alfalfa (in pure stand or in mixture with grasses) *versus* annual legumes such as vetches or pea (in pure stand or in mixture with cereals) in rain-fed Mediterranean environments highlighted the distinct forage yield advantage of drought-tolerant alfalfa material not only under moderate drought but even under severe drought (as determined by about 250 mm of annual rainfall), despite the potential advantage of annual crops under these conditions (Melis *et al.*, unpublished data). These results suggest that alfalfa will remain pivotal in Southern Europe, while expanding its relative importance in non-acidic soils of Northern Europe at the cost of

red clover (*Trifolium pratense*) and white clover (*Trifolium repens*) because of drier and warmer growing conditions. While white clover is particularly suited to grazing systems, red clover, which is the most-grown forage legume in Nordic countries (Annicchiarico *et al.*, 2015a), produces forage containing a polyphenol oxidase that may reduce ruminal proteolysis (Lüscher *et al.*, 2014). Minor breeding efforts could be justified on red and white clover and other legume species in regions to which they are particularly adapted and/or to exploit the advantages of species diversity. Birdsfoot trefoil (*Lotus corniculatus*), sainfoin (*Onobrychis viciifolia*) and sulla (*Hedysarum coronarium*) may have an interest for multi-species mixtures because of their content of condensed tannins, which can reduce the rate of protein degradation in the rumen, decrease the risk of bloat, and provide bioactive compounds to control parasites of animals (Lüscher *et al.*, 2014).

Among grass species, tall fescue (*Festuca arundinacea*) because of its broad adaptability to various stresses, and cocksfoot (*Dactylis glomerata*) because of its outstanding drought tolerance even in shallow soils (Boller *et al.*, 2010), have special perspective importance in the changing climate, albeit with a definite need for forage quality improvement (particularly for stem quality). The importance of perennial ryegrass (*Lolium perenne*), currently the most-grown forage grass in temperate grasslands, will largely depend on the ability to increase its tolerance to drought and high temperatures. New genetic diversity must be exploited for this purpose, since current varieties originate mainly from regions with an oceanic climate (Luis Blanco-Pastor *et al.*, 2019). The cultivation of perennial ryegrass is expected to move further northward as consequence of warmer and longer growth seasons, albeit with a challenge represented by sufficient winter survival (hindered by lack of adaptation to snow cover and susceptibility to pink snow mould, *Microdochium nivale*). A long-term public-private pre-breeding program has been undertaken in the Nordic and Baltic countries to improve the regional adaptation of this species (Rognli *et al.* 2018). Breeding of Italian ryegrass (*L. multiflorum*) is expected to focus on two contrasting variety types, one for very short (6 month) meadows grown in winter for forage stock production, and the other for 3- to 4-year rotational meadows. *Festulolium*, i.e., hybrids between *Lolium* and *Festuca* species (mostly *L. multiflorum* and *F. arundinacea*), combines the good tolerance to abiotic stresses (drought, frost) of fescues with the superior yield and forage quality of *Lolium* (Humphreys and Zwierzykowski, 2020). *Festulolium* cultivars are increasingly popular in some countries of eastern and northern Europe, contributing to more resilient and sustainable forage production in dry and warmer growing seasons. Timothy (*Phleum pratense* L.), which is currently the main grass for conservation cuts in Nordic countries, may remain regionally important if it was improved for greater persistence under intensive management (which is needed to increase its nutritive value).

Breeding for adaptation to increasingly drier regions of Southern Europe

Landraces and wild populations have largely been used as genetic resources by forage crop breeders. These materials evolved under highly diversified environmental conditions and, as such, contain a wealth of genetic variation for adaptive traits and adaptation patterns (Annicchiarico *et al.*, 2015a; Blanco-Pastor *et al.*, 2020). For example, alfalfa farm landraces originated in a rather small region of the Po Valley exhibited remarkable genotype $\times$ environment interaction (GEI) of cross-over type for biomass yield across irrigated and rainfed environments and contrasting soil types, with specific adaptation to the conditions under which they evolved (Annicchiarico and Piano, 2005). Likewise, wide adaptive variation emerged among natural populations of perennial ryegrass across Central and Western Europe (Keep *et al.*, 2020), as well as among cultivars of alfalfa (Annicchiarico *et al.*, 2011a) or cocksfoot (Annicchiarico *et al.*, 2011b) across locations of the Mediterranean basin. All of these studies revealed a trade-off between yield under relatively moisture-favorable conditions (largely reflecting the

cultivar yield potential) and yield and persistence under drought, which increased when passing from moderate to very severe drought. This response pattern agrees with trade-offs between relevant adaptive traits, which concern dehydration avoidance (involving fast water use) under moderate drought, and dehydration tolerance (involving slow water use and possibly summer dormancy) under severe drought (Lüscher *et al.*, 2022; Volaire *et al.*, 2023). For example, the specific adaptation to severely drought-prone Mediterranean environments of the alfalfa landrace Mamuntanas (Annicchiarico *et al.*, 2011a) was associated with a water saving strategy and a combination of drought tolerance traits, such as lower leaf stomata density, greater leaf tolerance to desiccation, delayed leaf senescence, and high water-soluble carbohydrate (WSC) content in storage organs under stress (Annicchiarico *et al.*, 2013; Kang *et al.*, 2021). Likewise, drought tolerant perennial ryegrass populations appeared more resource conservative (Keep *et al.*, 2020); and the specific adaptation to severely drought-prone Mediterranean environments of the cocksfoot cultivar Kasbah (Annicchiarico *et al.*, 2011b; Shaimi *et al.*, 2009; Zhou *et al.*, 2019) relied substantially on its full summer dormancy, which implies long-term persistence along with intrinsically low yield potential (Voltaire and Norton, 2006). However, variety types adapted to moderate drought and with relatively broad adaptability and yield stability across climatically variable conditions could be obtained. In cocksfoot, for example, this was a feature of material with incomplete or absent summer dormancy combined with dehydration tolerance (Piano *et al.*, 2004; Annicchiarico *et al.*, 2011b) and, occasionally, fully summer dormant material featuring high yielding ability in favourable seasons (Kallida *et al.*, 2016). For alfalfa, other relevant adaptive traits include: (a) autumn dormancy, of which the optimal value is expected to decrease across a latitudinal gradient in Europe to profit of milder winter temperatures and the wet season for forage production; (b) the root size, which is primarily important for greater storage of starch and proteins that favours the adaptation to moisture-favourable, relatively frequent mowing conditions (Annicchiarico *et al.*, 2013).

The mentioned, physiology-based trade-off between growth potential and drought tolerance justifies the modest genetic correlation across drought-prone and moisture-favourable environments that has frequently been reported for biomass yield of forage crops cultivars (Annicchiarico *et al.*, 2015a). Breeding for drought-prone areas may be further complicated by low genetic correlations between regions featuring different drought stress pattern, which emerged in alfalfa for biomass yield of half-sib progenies (Annicchiarico *et al.*, 2022). This result is particularly important because it concerns the material used by breeders for a reliable estimation of parent breeding values, also in view of the higher cost-efficiency of half-sib progeny-based selection compared with other selection schemes for alfalfa synthetic variety breeding (Annicchiarico and Pecetti, 2021). Such a remarkable GEI would require the selection of specific cultivars for different drought-prone regions (ideally associated with evaluation work aimed to verify the cultivar stress tolerance and recommendation domain).

The increasing fluctuations for annual rainfall and other considerations may lead some breeding programs to emphasize the selection of varieties with higher yield stability and broader adaptability. Particularly for alfalfa, a promising avenue is selecting semi-hybrid varieties with chance hybridization around 50% for two genetically distant, heterotic populations planted at a seed ratio of 1:1 (Brummer, 1999). In a multi-environment study, two-way and three-way semi-hybrids outperformed a synthetic variety bred from local germplasm (also used as a component of the semi-hybrids) for biomass yield in specific cropping environments (Annicchiarico and Pecetti, 2022). However, identifying highly heterotic exotic germplasm and selecting it for local adaptation may be challenging.

In vitro selection for stress tolerance may be valuable in some cases. This method out-performed other phenotypic selection methods for alfalfa tolerance to salinity of soil and/or irrigation water (Pecetti *et al.*, 2024), a target of increasing importance under climate change.

Breeding for adaptation to regions of Northern and Central Europe

Climate change predictions for Northern and Central Europe prompt selection programs to re-evaluate the bred species (with greater emphasis on alfalfa, tall fescue and cocksfoot) and the target traits. Opportunities (longer growing seasons) and possible challenges (drought; heat; thawing and freezing during winter leading to ice encasement; fungal diseases; etc.) are more diversified in these regions compared with Southern Europe. Improving the adaptation of major species to the new climate and its associated stresses by increasing the genetic diversity is probably the most important strategy, particularly for Northern Europe (Rognli *et al.* 2018; 2021). Field testing of Nordic and Central European cultivars of perennial ryegrass across locations spanning from continental Estonia to maritime Iceland highlighted the superior adaptation of Nordic cultivars albeit in association with large GEI (Helgadóttir *et al.*, 2018). However, the current cultivars will probably not be adapted to the future climate, as the longer growth seasons with warmer autumns may give inferior hardening and freezing tolerance in less adapted species, such as perennial ryegrass (Dalmannsdóttir *et al.*, 2016). Introgressing more exotic genetic resources to change the timing of growth cessation without compromising winter survival will be a major goal in this case. A study on local timothy cultivars found decreasing freezing and increasing ice-encasement tolerance over time (20–30 years) along with changes in genetic compositions, possibly due to seed multiplication outside (south) of the area of adaptation (Pashapu *et al.*, 2025).

Increased seed production in tetraploid material, and greater resistance to root rot, already are crucial breeding targets for red clover (Vleugels *et al.*, 2019; Nilsdotter-Linde *et al.*, 2025). Milder winters are expected to emphasize the importance of root rot resistance, because cold acclimation induces resistance also against this biotic stress. Experiments in controlled conditions found 37% lower survival rate when red clover was subjected to a combination of freezing stress and clover rot infection compared to the effect of freezing and infection alone (Ergon and Amdahl, 2024). The concurrent effect of this stress and other factors lead to low correlation between plant mortality under growth chamber conditions (only due to freezing temperature) and field-based plant mortality, as reported by Zanotto *et al.* (2021). This multi-location study also highlighted the better frost tolerance of several landraces and wild populations compared with commercial cultivars, the large variation in this material for several morphophysiological traits, and the association of accession characteristics with climate at their collection sites that could help identify promising genetic resources.

Breeding for intercropping

The selection for adaptation to legume-grass intercropping is challenged by the presence of possibly different companion species and, within each species, a potentially huge number of possible companion cultivars. This situation is further complicated by the possible impact on competition dynamics of different crop managements with respect to N fertilization amount, sowing rates of each component, sowing mode (bulked seed on the row or alternate rows), and type and frequency of crop exploitation (Harris, 1987). A better understanding of plant-plant interactions and its modelling could allow to decrease experimental efforts by restricting the number of mixtures to be evaluated (Louarn *et al.*, 2020). The early indication by Harper (1977) that the yield efficiency of a mixture depends mainly on the performance of its weaker partner was confirmed by results for legume-grass mixtures reviewed

in Annicchiarico *et al.* (2019). This work suggested to concentrate on the selection for greater competitive ability of the weaker partner species in the target growing conditions, owing to the trend of the genetic correlation for genotype yield responses across pure stand and mixed stand to decrease with increasing competitive stress exerted on the focus species. Such a response occurred regardless of the companion species in a large study assessing a set of white clover genotypes in pure stand and intercropping with eight grass varieties belonging to four species (Annicchiarico and Piano, 1994). Selecting for general adaptation to intercropping is justified by commercial reasons and is supported by the small size of specific mixing ability effects relative to general mixing ability ones in Holland and Brummer (1999).

Alfalfa frequently is a strong competitor against grasses and, accordingly, tends to display fairly high correlation for genotype yield responses across pure stand and mixed stand environments—albeit with possible room for specific selection (Maamouri *et al.*, 2017; Grieder *et al.*, 2021). However, most legume species tend to be out-competed by grasses. A minimum content of about 30% average legume proportion on total biomass is necessary to achieve the expected legume advantages in terms of total biomass (via N transfer) and forage quality (in terms of protein content) (Thomas, 1992; Dineen *et al.*, 2018). Selecting a poor competitor such as white clover for greater competitive ability allowed for both a drastic increase of clover proportion (approaching 30% average content) and a higher total yield of clover-grass mixtures (Annicchiarico and Proietti, 2010).

Selecting for intercropping could be performed under severe competition, by mixing a few vigorous cultivars of different companion species as a tester; or in pure stand according to an index of selection whose traits contribute to high competitive ability (Annicchiarico, 2003). Taller plant stature (which is associated with higher relative growth rate) emerged consistently as a key trait contributing to greater competitive ability in relatively productive environments (Annicchiarico *et al.*, 2019). Several traits contributing to complementarity of resource use have been envisaged (Litrico and Violle, 2015; Lüscher *et al.*, 2022), but their adoption in breeding program has been limited so far (Annicchiarico *et al.*, 2019).

Regardless of the level of species competition, breeding for intercropping may depend on the direct effect of a component for the target trait (e.g., yield) and its indirect effect on the other component(s) (Sampoux *et al.*, 2020). The correlation between these effects is often negative, and a compromise must be sought.

Genomic and marker-assisted selection

Forage yield improvement is hindered by long selection cycles, outbreeding mating systems associated with severe inbreeding depression, low narrow-sense heritability partly due to high non-additive genetic variance (especially in autotetraploid species), fairly low consistency of breeding values across spaced plant and sward evaluation, and large GEI across different cropping conditions and crop utilizations (Annicchiarico *et al.*, 2015a). Genomic and marker-assisted selections may alleviate the challenges of field-based selection. About 15 years after the development of NGS technologies, large marker numbers can be available rapidly and at a low cost, mainly via genotyping-by-sequencing (GBS; Elshire *et al.*, 2011). Novel marker tools, such as the DArTag 3K panel for alfalfa, may further reduce the selection costs (Sipowicz *et al.*, 2024). These developments have allowed to envisage the genomic selection for complex, polygenic traits, as well as to identify major QTLs for less complex traits through GWAS or classic QTL analysis on mapping populations, as in the case of anthracnose

resistance in alfalfa (Pégard *et al.*, 2025). Markers flanking QTL could be used by breeders to introgress the resistant alleles in their elite material. This could be particularly efficient when the donor parent comes from a natural population.

Genomic selection, which combines phenotyping and genotyping data of a set of genotypes representing a reference population to define a statistical model for prediction of breeding values in future plant selections (Meuwissen *et al.*, 2001), may provide remarkable advantages to forage crop breeders. It may allow to: (a) increase the selection intensity and/or facilitate the selection for multiple traits (given the lower cost per evaluated genotype of genotyping relative to phenotyping); (b) shorten the selection cycle. The latter advantage, which is particularly relevant for half-sib or full-sib progeny-based selection, can be realized through a model constructed from genotyping data and dense-sward phenotyping data of plant pools (synthetics and half-or full-sib families), which is then applied on genotyping data of young individual plants (Annicchiarico *et al.*, 2015a; Barre *et al.*, 2022). Particularly for alfalfa, phenotyping half-sib progenies of genotyped parent plants is recommended for construction of the genomic selection model, because of the convenience to focus only on the additive genetic variance (i.e., the relevant variance for synthetic variety breeding) in the presence of high non-additive genetic variance (Bingham *et al.*, 1994; Annicchiarico, 2015). The application of genomic selection is mostly envisaged on plants issued by a preliminary stage of mass selection, but it may also concern within-family selection (Byrne *et al.*, 2024).

Important research topics for genomic selection optimization includes the cost efficiency of the marker tool (Cericola *et al.*, 2018; Sipowicz *et al.*, 2024), the predictive ability of statistical models and the extent of their applicability for other reference populations (Annicchiarico *et al.*, 2015b; Medina *et al.*, 2021; Barre *et al.*, 2022), the incorporation of GEI effects (Sköt *et al.*, 2024), the integration of genomic predictions with phenotypic (Simeão Resende *et al.*, 2014) or phenomics data (Jahufer *et al.*, 2021) and, for autotetraploid species, the cost-efficient estimation of allele frequencies (Annicchiarico *et al.*, 2022).

Predictive ability values (as correlations between predicted and observed values based on cross validations) for biomass yield were mostly in the range of: (a) 0.20–0.35 for grass species in the review by Barre *et al.* (2022) and subsequent work (e.g., Byrne *et al.*, 2024); (b) 0.10–0.35 for alfalfa (e.g., Annicchiarico *et al.*, 2015b, 2022; Medina *et al.*, 2021; Andrade *et al.*, 2022; Pégard *et al.*, 2023) and other forage legumes (e.g. Ehoche *et al.*, 2025), with occasionally higher values (e.g., Sköt *et al.*, 2024). Albeit fairly low in absolute terms, these values may indicate greater predicted yield gain per time and unit cost for genomic selection relative to phenotypic selection, owing to the low narrow-sense heritability of the latter (Annicchiarico *et al.*, 2021; Ehoche *et al.*, 2025). Genomic selection can conveniently be envisaged also for key forage quality traits, owing to increasing evidence for a polygenic control of these traits (Biazzi *et al.*, 2017) and the consequent need to account for small-effect genes. Moderate predictive ability was found for neutral detergent fiber digestibility and protein content of alfalfa (Annicchiarico *et al.*, 2021), and organic matter digestibility and WSC content of perennial ryegrass (Grinberg *et al.*, 2016).

The superiority of genomic selection over phenotypic selection is awaiting experimental confirmation in terms of actual yield gains. Two unpublished studies performed on alfalfa separately for Po Valley germplasm in Northern Italy and Mediterranean germplasm in Algeria indicated significantly greater genetic gains for genomic over phenotypic selection (Annicchiarico *et al.*, unpublished data), despite

predictive ability values of 0.32 for the Italian reference population (Annicchiarico *et al.*, 2015b) and 0.22 for the Mediterranean population in Algeria (Annicchiarico *et al.*, 2022) of the selection models. Importantly, these results concerned a species with particularly low values of genetic yield gains (Annicchiarico *et al.*, 2015a) and narrow-sense heritability of biomass yield (Annicchiarico, 2015). Another example concerns a divergent selection for herbage accumulation and heading date on perennial ryegrass, which revealed the potential of genomic selection but also its limit when a predictive model was used on genetically unrelated material (Faville *et al.*, 2020).

High-throughput phenotyping and envirotyping

New imaging technologies (visible, multispectral, thermal) on different supports (drones, automated systems) may allow for a quick and low-cost estimation of different traits. For example, visible images taken by drones allowed for estimating plant height and canopy cover (Surault *et al.*, 2018; 2021). Drone-based data of RGB imaging and canopy temperature difference provided an effective means to identify drought-resilient alfalfa material (Noushahi *et al.*, 2025). Likewise, remote sensing data from equipment mounted on a phenomobile showed high potential for repeated estimates of biomass yield in alfalfa selection experiments subjected to abiotic stresses (Cazenave *et al.*, 2019). In such experiments, vegetation indices obtained from UAV images could conveniently be combined with spatial analysis (Biswas *et al.*, 2021). Ongoing research efforts include the UAV image-based estimation of a time-consuming trait such as legume content in legume-grass mixtures (Kawamura *et al.*, 2024).

The increasing year-to-year climate variability emphasizes the importance to characterize the environment of each experiment to better exploit its results. Predicting the variety response in new environments could depend crucially by explicit GEI effects (Xu, 2016). Johansen *et al.* (2025) modelled GEI effects for yield, winter mortality and spring cover of 264 perennial ryegrass accessions from multi-environment trials in Nordic and Baltic countries using reaction norm or envirotyping. Genomic prediction improved up to 22 and 9% for spring cover and winter kill, respectively, for unobserved germplasm and environments when GEI was included, confirming the potential importance of envirotyping. A GWAS detected genetic variants with large effects across all environments, along a few environment-specific QTLs.

Yield data from Norwegian VCU trials of timothy cultivars (8 sites in the years 1988–2017) were used to develop a prediction model for biomass yield based on agro-climatic variables (Hellton *et al.*, 2023). Growing degree days in July and the number of rainy days in June–July had the highest predictive ability, explaining 43% of the yield variation. The prediction model was combined with a range of climate ensembles (RCP4.5) for the decades 2050–2059 and 2090–2099, showing that yields may decrease substantially in South-Eastern Norway but increase in Northern Norway due to higher temperatures and changing precipitation patterns.

Envirotyping could also support breeding programs with respect to plant ideotypes to prioritize for specific regions. For example, a factorial regression model for biomass yield of contrasting cocksfoot cultivars as a function spring-summer drought stress of sites of the Western Mediterranean basin was exploited to predict the cultivar responses for 2050 according to expected site-specific climatic values (Annicchiarico, 2020). The results revealed the future importance of the fully summer dormant *hispanica* germplasm, predicted to be higher yielding than non-dormant or partly dormant germplasm in nearly all areas of North-West Africa and Southern Europe (while being currently superior only in inland Morocco).

Conclusions

Grassland plant breeding is going to be severely challenged by climate change because of the increasing demand for cultivars with substantially greater tolerance to drought and other stresses, which will be highly needed for species whose modest economic importance limits the possibility of stress alleviation by management (e.g., irrigation). Compromises between high yield of good nutritive value and stress tolerance shall be sought. Further difficulties would arise from: (a) limited breeding budgets due to the possibly large number of target species and the modest market importance of several of them; (b) the traditionally high reliance of breeders on regional genetic resources (landraces, old cultivars, natural populations) whose local adaptation, which derived from decades or even centuries of evolution under consistent climate conditions, is now disrupted by the changed climate; (c) additional complexities associated with the variation of growing environments and system managements (Caradus and Chapman, 2025) and their interactions with the changing climate. Other breeding targets can be relevant besides those considered here. For example, animal grazing can reduce production costs and the environmental impact of forage crops (Benson, 2008), and specific selection for grazing tolerance can be a key trait in systems based on sheep grazing and/or continuous grazing (Smith *et al.*, 2000; Annicchiarico *et al.*, 2015). Advances in breeding knowledge and supporting technologies, and the careful selection of exotic parent germplasm, will help breeding programs face the remarkable challenges posed by the changing climate. Concurring policy measures supporting public or private breeding efforts could help maintain grassland productivity in various regions and safeguard thereby the environmental and economic importance of European forage-animal systems.

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Response to selection for grain and forage yield in intermediate wheatgrass

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Abstract

Integrating perennial grain crops into agricultural systems can become a major milestone to increase provision of ecosystem services of food production. Intermediate wheatgrass (IWG) is a forage species that is undergoing domestication to improve its grain production as dual-purpose crop. However, methods to optimize response to selection for both grain and forage under are not well understood. Our goal was to understand the genetic relationship among grain and forage yield components, quantify potential trade-offs between vegetative and reproductive allocation, and offer relevant information to optimizing responses to selection in IWG. In two field experiments, thirty half-sib families were space-planted in a randomized complete block design with three replications. Phenological, grain, and forage traits were measured for each plant over three years. Narrow sense heritability estimates were moderate to high for most traits. Secondary traits such as grain yield per spike and spikes per plant could be used for indirect selection for grain yield per plant. No trade-offs were detected between grain and forage yield traits, indicating that simultaneous improvement of both traits is feasible. Dual-use IWG germplasm could therefore efficiently be developed by shortening the time to first evaluation, using secondary traits, and performing selection under dual-use.

Keywords: dual-use, heritability, indirect selection, perennial grain, yield components

Introduction

Intermediate wheatgrass (IWG, *Thinopyrum intermedium* (Host) Barkworth and D.R. Dewey), is a promising dual-use (grain and forage) perennial crop under domestication (DeHaan *et al.*, 2025). It is a perennial cool-season grass species with vernalization requirements (Locatelli *et al.*, 2022), and it provides multiple ecosystem services. The forage production and quality are high (Favre *et al.*, 2019). Improving grain yield is key to the success of the crop. Indirect selection on a secondary trait such as spike fertility, size, and grain yield per spike have been proposed to increase the response to selection for seed yield in IWG (Bajgain *et al.*, 2022). We hypothesized that the response to indirect selection on yield components could be higher than direct selection for yield per plant under dual-purpose system. Forage harvest can have a strong effect on grain yield in dual-purpose crops; therefore, there may be trade-offs between vegetative and reproductive allocation of resources in dual-use crops. The aim was to understand the relationship among grain and forage yield components in IWG under dual-use. Specifically, our goals were to: (1) quantify potential trade-offs between vegetative and reproductive allocation under dual-use management and (2) identify secondary characters to grain yield per plant to be used in indirect selection.

Materials and methods

Seeds were from The Land Institute (USA) breeding program Cycle 4 (De Haan *et al.*, 2025), selected from accessions originally from the former Soviet Union evaluated at the Rodale Institute (USA).

Seedlings were grown in the greenhouse and transplanted to the field at Paysandú, Uruguay (32°55'S, 58°03'W) following Locatelli *et al.* (2023). Soils were fertile loamy clay Typic Argiudoll. From 1,050 plants evaluated, 30 plants were selected as parents of the half-sib families, based on a simple index Z -score of the sum of standardized traits from data for number of spikes per plant and early maturity. The half-sib progeny (seeds) from each selected plant were grown in the greenhouse and then transplanted to the field (Exp. 1) in a randomized complete block design with three replications. Each plot consisted of 12 plants arranged in two rows of 3.6×0.6 m each. Based on the evaluation of Exp. 1 and using the same selection criteria, the best 30 plants were selected to establish a second trial of half-sib families in the same location (Exp. 2). Mature spikes were harvested each summer, and three spikes per plant were used to count the number of spikelets per spike. Forage yield was evaluated by hand-harvesting aboveground biomass of the plants at 2.5 cm above ground at grain maturity (summer forage). Biomass was dried at 65°C for 48 h and then weighed. The harvest index was estimated as the ratio of grain yield per plant and the summer dry matter forage yield. Linear mixed models were used to study differences among half-sib families and to estimate variance components using the lmer function of the lme4 package of the R statistical software. Narrow-sense heritability (h^2) on a trait mean basis was estimated for each year and experiment following Holland *et al.* (2003). The relative selection efficiency (RSE), i.e., the expected rate of improvement in a trait when selecting on x relative to that when selecting on y was estimated following Falconer & Mackay (1996) and Searle (1965) as the correlation among traits times the square root of the ratio of the heritabilities of both traits.

Results

There was no genotype by stand-age interaction for most traits in Exp. 1. Genotypic and stand-age (year) effects were large for most traits, with significant differences among half-sib families and years for most traits. Similar genotypic effects were found in Exp. 2 (data not shown). High and positive genetic correlations between most grain yield components were found in both experiments (0.73 to 0.97). Phenological traits (days to heading and days to maturity) had high and negative correlations with most traits in both experiments. Summer forage yield for both experiments was positively correlated to most grain yield components.

Most traits had moderate to high heritability values in both experiments and for different years (Table 1). Harvest index and traits directly associated with grain yield had the highest genetic gain per year in both experiments. On the other hand, phenological traits had the lowest values of genetic gain. Harvest index for the first year and yield per spike for the second year were the traits that had a relative selection efficiency (RSE) higher than 1 for indirect selection for grain yield per plant in Exp. 1 (Table 1). Similarly, harvest index, number of spikes per plant and yield per spike were the traits that had a RSE higher than 1 for indirect selection for grain yield per plant in Exp. 2. In Exp. 1, the RSE was higher in the first year than in the second year for summer forage yield, harvest index, and grain filling. Nevertheless, for most of the traits, the greatest genetic gains per year are obtained when using the first stand age for selection.

Discussion

Indirect selection for some of the grain yield components such as the number of spikes per plant and the grain yield per spike would result in a higher response to selection than direct selection for grain yield per plant. Grain yield per plant increases from first to second year are commonly observed in spaced-plant studies. In geographic areas where vernalization requirements may not be fully met (as in this study), plants that are able to have more reproductive tillers may be plants with lower

Table 1. Genotypic mean and standard errors (SE), narrow sense heritabilities (h^2), relative selection efficiency for early selection (RSE_E, using the first year to select for second year), and for selecting for grain yield on correlated secondary traits (RSE_C), for agronomic and phenological traits evaluated over two years in Exp. 1 and one year in Exp. 2 in 30 half-sib families of intermediate wheatgrass in Paysandú, Uruguay.

Trait	Mean (SE)		h^2			RSE_E	RSE_C			
	Exp1	Exp2	Exp1	Exp2		Exp1	Exp2	Exp2		
	Yr 1	Yr 2	Yr 1	Yr 1	Yr 2	Yr 1	Yr 1	Yr 2	Yr 1	
Plants headed (%)	71.3 (8.1)	85.0 (6.1)	91.7 (5.2)	0.84	0.93	0.49	0.47	0.75	0.64	0.50
Spikes/plant (No.)	13.0 (3.2)	33.5 (8.8)	13.3 (2.1)	0.45	0.27	0.41	0.88	1.00	0.70	1.11
Spikelets/spike (No.)	21.3 (1.1)	17.2 (0.8)	—	0.46	0.65	—	0.10	0.64	1.00	—
Spikelets/plant (#)	359 (91)	665 (194)	—	0.27	0.10	—	0.87	0.68	0.44	—
Grain/spike (mg)	138 (28)	80 (16)	274 (31)	0.51	0.69	0.44	0.62	0.99	1.06	1.02
Grain/spikelet (mg)	6.9 (1.3)	4.7 (0.8)	—	0.61	0.61	—	0.70	0.90	0.96	—
Grain yield (g plant ^{−1})	2.2 (0.9)	3.3 (1.2)	4.2 (0.9)	0.31	0.35	0.23	0.69	—	—	—
Harvest index (%)	2.1 (0.6)	3.4 (1.2)	7.6 (1.1)	0.62	0.30	0.62	1.04	1.14	0.62	1.18
GDD to heading	3865 (76)	2608 (81)	2931 (64)	0.55	0.49	0.64	0.70	−0.39	−0.68	−0.81
GDD to grain filling	1777 (55)	1640 (63)	1849 (45)	0.47	0.01	0.37	1.99	0.55	0.03	−0.01
GDD to maturity	5640 (82)	4154 (90)	4701 (53)	0.32	0.55	0.67	0.20	0.01	−0.52	−0.84
Forage DM (g plant ^{−1})	99.0 (16.9)	91.6 (21.0)	47.4 (5.7)	0.40	0.13	0.0	1.30	0.57	0.39	0.0

vernalization requirements that should be selected further. Therefore, this trait should have been used in breeding efforts, especially where selection for adaptation to mild temperatures is desired. Therefore, we suggest using the number of spikes per plant as a strong selection criterion in the first cycles and using a lower weight in the following years. Grain yield per spike could be a more universal secondary trait to select for grain yield per plant. Grain yield per spike had also a high relative selection efficiency for grain yield per plant. We did not find any trade-offs between vegetative and reproductive allocation of resources under a dual-crop management. We detected positive and significant genetic correlations between forage yield and several grain yield components, suggesting the possibility of simultaneous improvement for both grain and forage yield. Selection in the first year of production might be used to increase genetic progress. These are encouraging findings for the development of dual-purpose perennial grains in temperate areas.

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Yield and quality of grass-legume swards over the growing season using different lucerne ratios

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Abstract

Lucerne (*Medicago sativa* L.) is one of the most important legumes for forage production because of its high nutritive value and productivity. Field trials were carried out with the aim of investigating dry matter (DM) yield and quality of grass-legume mixtures with different proportions of lucerne: 30, 50 and 70%. The following quality parameters and the mineral contents were determined: crude protein (CP), neutral detergent fibre (NDF), acid detergent fibre (ADF), net energy for lactation (NEL), calcium (Ca), and phosphorus (P). The increased proportion of lucerne in the mixture did not increase DM yield over the year, but contributed to a significant increase in CP content in DM yield of the first cut. Significant differences were found among mixtures for content of ADF and Ca. Significant differences were found among cuts for the DM yield and all stated herbage quality parameters.

Keywords: cut, grass, legume mixture, herbage quality, lucerne ratio, productivity

Introduction

Lucerne is a promising forage crop in the Baltic countries because of its great yield, excellent quality, and high adaptability in different conditions (Lillak *et al.*, 2005). The relationship between the proportion of legumes in the seed mixture and the herbage yield is not well understood. Some research findings show a positive relationship between increased legume proportion in the mixture and herbage yield (Elgersma & Søgaard, 2016); other studies did not observe a general relationship between the proportion of legumes and forage biomass (Sanderson *et al.*, 2013). Mowing management affects the herbage quality of lucerne-based mixtures. Studies show that the maturity of lucerne at harvest could significantly affect the DM yield (Katanski *et al.*, 2018). There is a lack of information on the impact of lucerne proportion on the productivity and quality of lucerne-grass mixtures, as well as on changes in forage quality between harvests in the Baltic region.

Materials and methods

The field trial was conducted at the Research Institute of Agronomy of LBTU in Skriveri (56°37'N, 25°7'E) on an Eutric Retisol with a pH (KCl) of 6.1–6.4; contents of P and K were 50 and 77 mg kg⁻¹, and organic matter of 29–34 g (kg soil)⁻¹. The trial design was a randomised complete block with three replications and a plot size of 8 m². Legume-grass mixtures with different rates of lucerne (30, 50 and 70%) and the following grass species: ×*Festulolium loliaceum*, *Festuca pratensis* L., *Lolium perenne* L., *Phleum pratense* L. in equal parts in mixture, were seeded in 2023 without a cover crop. The results of the first year of sward use (2024) were analysed. The four cuts during the vegetation season with the first cut at the bud stage, and next cuts after 35–40 days were made. Fertilisation comprised 30 kg N ha⁻¹, 34 kg P ha⁻¹ and 112 kg K ha⁻¹ applied in early spring after the resumption of vegetation. Moderate doses of N fertilizer (30 kg N ha⁻¹ after the 1st and 2nd cuts) were applied as a “starter” fertilizer in the first half of the vegetation season, considering the low organic matter

Table 1. Herbage quality depending on the cut and species composition

Lucerne content, % (L)		Content in DM (g kg ⁻¹)					NEL (MJ (kg DM) ⁻¹)
		Crude protein	NDF	ADF	Ca	P	
1st cut	30%	135.4	425.8	320.6	13.4	4.20	6.05
	50%	154.8	441.5	328.4	14.1	3.93	5.99
	70%	181.7	400.7	330.5	17.3	4.27	5.97
	Mean 1st cut	157.3	422.6	326.5	14.9	4.13	6.00
2nd cut	30%	191.3	432.5	275.8	12.5	3.37	6.41
	50%	199.7	432.4	300.6	15.3	3.27	6.21
	70%	188.5	456.8	310.2	14.1	3.23	6.13
	Mean 2nd cut	193.1	440.6	295.5	13.9	3.29	6.25
3rd cut	30%	188.7	458.9	278.3	11.9	3.30	6.39
	50%	194.9	454.1	292.0	13.8	3.23	6.28
	70%	193.9	453.6	281.5	13.3	3.33	6.36
	Mean 3rd cut	192.5	455.5	283.9	13.0	3.29	6.34
4th cut	30%	188.0	446.7	285.8	13.6	3.17	6.33
	50%	196.2	426.2	282.6	15.3	3.23	6.35
	70%	181.0	452.5	305.3	14.3	3.13	6.17
	Mean 4th cut	188.4	441.8	291.2	14.4	3.18	6.28
LSD _{0.05} C		9.62	17.62	14.03	1.20	0.16	0.11
LSD _{0.05} L		8.33	ns	12.15	1.04	ns	0.10
LSD _{0.05} C/L		16.67	ns	ns	ns	ns	ns
P value C		<0.001	0.008	<0.001	0.020	<0.001	<0.001
P value L		0.022	0.932	0.028	0.001	0.362	0.029
P value C/L		0.002	0.052	0.325	0.075	0.354	0.338
F value C		27.074	4.995	15.225	3.946	65.801	14.963
F value L		4.497	0.070	4.156	8.715	1.062	4.103
F value C/L		5.064	2.478	1.233	2.235	1.172	1.205

content and pH level of the soil. The CP, NDF, ADF, P, and Ca content of DM yield were determined with near-infrared reflectance spectroscopy (NIRS). NEL was calculated according formula $NEL = (0.0245 \times (88.9 - (ADF\% \times 0.779)) - 0.12) \times 4.184$ using digestibility coefficients and full-value coefficients. The experimental data were statistically analysed using two-way analysis of variance with “Lucerne Content” and “Cut” as factors. The differences among means were detected by using an LSD at the 0.05 probability level.

Results and discussion

Crude protein, NDF and NEL were lower, whereas ADF and P were significantly higher at the first cut compared with later cuts (Table 1). There were no significant differences in stated quality parameters between 2nd, 3rd, and 4th cuts. The increased proportion of lucerne in the mixtures contributed to successive increase of CP content in the grass of the 1st cut. At the following 2nd, 3rd and 4th cuts the highest CP content was found for mixtures with 50% of lucerne. Legumes accumulate more Ca than grasses; therefore, the content of Ca in DM was closely connected with legume content. In all cuts, a significantly lower Ca content in DM yield was found for the mixture with lucerne at 30%.

The cutting regime considered in the three cuts is the average number of cuts for cultivated grassland swards in Latvia. Four cuts per vegetation season are recommended for mixtures containing grasses and lucerne with good regrowth potential (Gaile, 2005). Differences in yield distribution between cuts were stated for all mixtures. The average dry matter yield of the 1st cut accounted for 45% of the annual yield (Fig. 1). This is in line with the findings of other Baltic researchers (Tamm *et al.*, 2020).

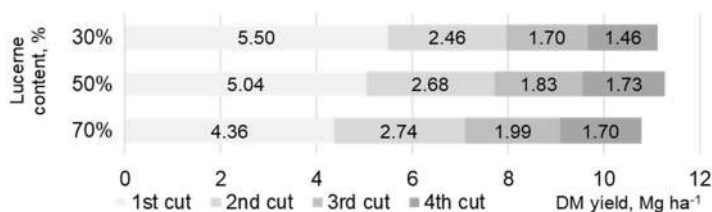


Figure 1. Distribution of annual DM yield by cuts.

The highest 1st cut DM yield for mixtures with lucerne 30% could be explained by cool weather conditions in the spring, which delayed lucerne development in early spring and allowed grasses to dominate in the 1st mowing. No significant ($P > 0.05$) effect of higher lucerne proportion in the mixture on the annual DM yield was found.

Conclusion

Preliminary results show that the cutting had a greater impact on DM yield and quality than lucerne content in the mixture. A higher proportion of lucerne in the seed mixture significantly increased the CP content in the DM yield. Although the 1st cut was taken at the bud stage, the forage quality was lower in the later cuts. This is an important aspect, because the DM yield of the 1st cut accounts for the highest proportion of the total annual yield.

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Improving forage production in Norway: Participatory evaluation of climate-adapted seed mixtures

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Abstract

A stable supply of high-quality forage is essential for the economic sustainability of milk and meat production, the most important agricultural sectors in Norway. Successful forage production relies on selecting species, cultivars, and seed mixtures suited to the diverse agroecological conditions, which vary widely in latitude and altitude. Climate change modifies production boundaries unpredictably. Without on-farm performance data, there is a risk of recommending new varieties or seed mixtures that underperform in farmers' fields. To address this, we propose the *tricot* method: a participatory, large-scale, and cost-effective testing approach where farmers test new seed mixtures directly in their fields. This approach involves many farmers in conducting small, simple experiments. Each trial consists of an incomplete block with three options out of the total ten options in the trial. Farmers evaluate these options based on predetermined criteria using digital tools. These mini trials, combined with data on management, climate, and soil, are collected into the ClimMob software, which performs statistical analyses and provides feedback. When all data are aggregated, this approach can provide robust statistical results on optimal seed mixtures for various local conditions. This method also strengthens collaboration between researchers, advisers, and farmers, contributing to more sustainable and resilient agricultural practices.

Keywords: climate adaptation, collaboration, digital, field trials, local conditions, participatory

Introduction

Traditionally, Randomised Complete Block Design (RCBD) field trials have been used to assess new forage species, cultivars, and mixtures across diverse agroecological zones. However, these trials are labour-intensive and costly, limiting the number of sites and often excluding farmers from the evaluation process, which can reduce practical relevance and adoption. Participatory research addresses this by involving farmers in setting goals, conducting trials, and evaluating outcomes, ensuring that research is tailored to local conditions, and allows for innovation, learning, and ownership (van de Gevel et al. 2020). On-farm experimentation also helps integrate scientific and farmer knowledge (Lacoste et al. 2022). The *tricot* (triadic comparison of technologies) approach is a promising participatory method. Each farmer receives an incomplete block of three options out of a larger pool, ranks them using the best-worst scaling approach, and submits results to the free online software platform ClimMob (<https://climmob.net/>). This incomplete block design enables large-scale, cost-effective data collection, and rapid feedback across varied environments. Although *tricot* has been applied in several studies in the Global South, it is new to Norway. The INNOFORAGE project (2025–2028) aims to establish a cost-effective, farmer-led testing framework to identify optimal forage mixtures for local conditions and promote adoption. We will evaluate if *tricot* can deliver robust results across Norway's diverse climates and integrate it with RCBD trials at research station and selected farms. While *tricot* enables broad participation, metric traits like feed quality and biomass yield still require controlled

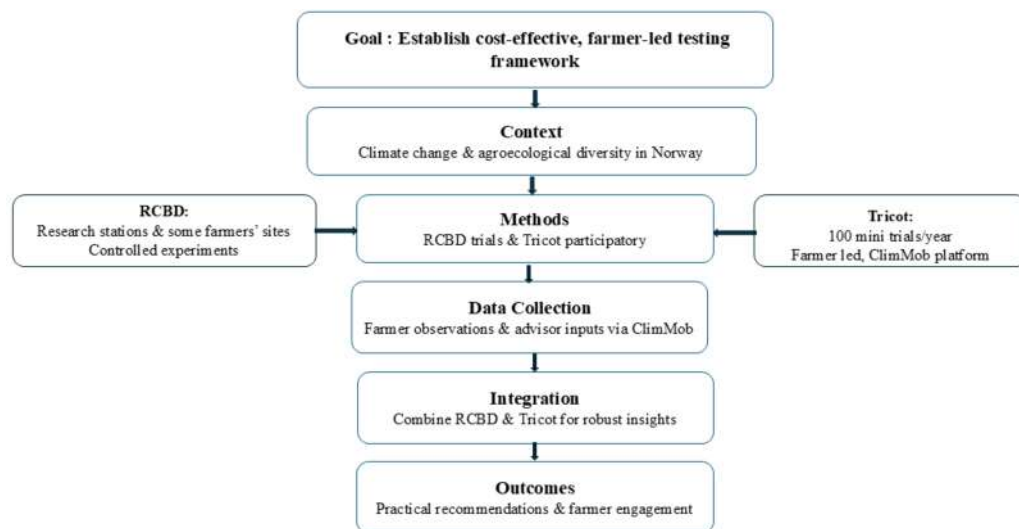


Figure 1. Flow chart of the different activities and work within the project.

experiments. By combining both approaches, we aim to provide more targeted recommendations and assess how participatory research supports farmer engagement and knowledge sharing.

Materials and methods

A flow chart of the work in the project is presented in Figure 1.

Twelve forage seed mixtures were developed in collaboration with seed companies, advisers and researchers (Table 1). Using the ClimMob platform, three seed mixtures were randomly assigned to each mini-trial via a balanced incomplete block design. In summer 2025, 100 mini-trials were established on farms across Norway, each with three $3 \times 7 \text{ m}^2$ plots managed as part of the field by farmers. Management practices, soil characteristics, and agro-climate indices are recorded for each trial and included as covariates in the confirmatory Plackett-Luce model to reduce confounding. The primary outcomes are overall performance rank, winter survival, and yield, with key comparisons across mixture $\times$ region $\times$ year. Secondary outcomes (e.g., weed invasion, plant height, legume cover) are analysed exploratorily. Farmers have provided informed consent and agreed to take part in focus group interviews and surveys regarding their experience with the experimentation. In the establishment year, they assess the three options in autumn based on: cover of sown species and of legumes (which is best/which is worst), weed invasion (least/most), plant height (tallest/shortest). In spring 2026, they will also report on winter conditions, particularly ice cover on fields. At harvest, they will assess yield (most/least), proportion of legumes (most/least), and proportion of weeds (least/most). In addition, they will be asked on overall performance compared to surrounding field (best/worst). They also will provide information on fertilisers (type and quantity). Trials will run for at least two full production years. Another 100 participants will be recruited in 2026, and seed mixtures may be adjusted based on feed-back from farmers and advisors participating in round one.

ClimMob manages all participant and data processes, supports rapid feedback, and includes R packages for statistical analyses and agro-climatic indices (Quiros et al. 2024). Farmer engagement and

Table 1. Overview of seed mixtures

Mixture	<i>Phleum pratense</i>	<i>Lolium pratense</i>	<i>Poa pratensis</i>	<i>Dactylis glomer- ata</i>	<i>Bromus inermis</i>	<i>L. arundi- naceum</i>	<i>Festuca rubra</i>	<i>Agrostis capillaris</i>	<i>L. perenne</i>	<i>Trifolium pratense</i>	<i>T. repens</i>	<i>Medicago sativa</i>	Region
1	70	20	0	0	0	0	0	0	0	10	0	0	Common
2	50	20	20	0	0	0	0	0	0	0	10	0	Common
3	25	10	15	0	0	25	5	0	10	5	5	0	Common
4	10	15	10	0	15	20	5	2	8	10	5	0	Common
5	0	30	0	40	0	0	0	0	10	10	10	0	Common
6	10	0	0	20	0	50	0	0	10	10	0	0	Common
7	10	0	0	0	0	70	0	0	10	10	0	0	Common
8	40	50	0	0	0	0	0	0	0	10	0	0	South/Central
9	40	50	0	0	0	0	0	0	0	10	0	0	North/Mountain
10	25	0	10	0	0	45	0	0	0	10	0	10	Common
11	0	0	0	0	0	0	0	0	90	0	10	0	South/Central
12	45	15	15	0	0	0	5	5	0	10	5	0	North/Mountain

learning will be studied using mixed methods, including interviews, surveys and stakeholder meetings throughout the project period.

Results and discussion

The mini-trials are currently being sown by advisers and farmers. Of the 100 mini-trials planned for 2025, 30 with seed mixtures targeted for northern and mountainous regions have been distributed to farmers located c. 61–70°N. The remaining 70 trials, designed for central and southern sites, have been distributed to farmers between about 59 and 67°N. After all participating farmers are registered, they will receive an initial set of questions regarding the establishment of their seed-mixture plots. We expect the first results to be available in early autumn 2025. Farmers have expressed great interest in taking part in the experimentation. Interest in participating has exceeded expectations, with more than the 100 planned farmers expressing they would like to join. These additional participants will be included in the 2026 round of trials. The advisers have also shown enthusiasm, particularly in using the mini-trials in field days—events where farmers meet, share experiences and discuss results. This project will both support the adaptation of seed mixtures to local agroecological conditions, and improve adoption, ownership, and active engagement among all actors—especially the farmers.

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Intraspecific variability at early establishment and winter survival of some minor forage species

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Abstract

Efforts continue to diversify grasslands with minor forage species to meet the current and future challenges. From a cultivation and breeding perspective, the key question is how much intraspecific variation exists in desired traits, as this determines the potential for future breeding. This study compared six dicot forage species (*Achillea millefolium* (Achmil), *Carum carvi* (Carcar), *Cichorium intybus* (Cicint), *Lotus corniculatus* (Lotcor), *Medicago lupulina* (Medlup), and *Plantago lanceolata* (Plalan)) to investigate intraspecific variation in early establishment (dry matter yield and weed mass) and winter survival. Four to seven cultivars and wildtypes (hereafter accessions) of each species were grown in a single-site field experiment arranged in a randomized block design with three replicates. Data were collected from the first harvest after sowing and the following winter. All plant species showed potential for successful early establishment and winter survival with significant differences among species. Although there was considerable intraspecific variation within species, this variation was statistically significant only for some species. The findings reveal the potential of minor species to be considered in future grassland farming and point out the possibility of improving germplasm through plant breeding.

Keywords: accessions, early establishment, minor forage species, winter survival

Introduction

Grasslands face multiple stressors, including drought, reduced species diversity, and particularly harsh winter conditions, which negatively impact crop physiology and productivity (Andrews, 1987). Enhancing both intraspecific and interspecific diversity has been proposed as a strategy to improve resilience and performance under these stresses (Isselstein & Komainda, 2021). The integration of underutilized species into cultivated grasslands represents a promising approach, yet these species have received limited breeding attention. Legumes and forbs, in particular, offer potential benefits for climate adaptation, forage quality, ruminant health, and nitrogen use efficiency due to their functional traits and secondary metabolites (Hamacher *et al.*, 2021). Since successful establishment and early growth are critical for integrating new germplasms, this study aims to evaluate early establishment and winter survival in selected underutilized species to identify intraspecific variability to better assess suitability for inclusion in grasslands mixtures.

Materials and methods

A field trial testing the effects of cultivars and wildtypes (hereafter accessions) of minor dicot forage species, (i.e., *Achillea millefolium* (Achmil), *Carum carvi* (Carcar), *Cichorium intybus* (Cicint), *Lotus corniculatus* (Lotcor), *Medicago lupulina* (Medlup) and *Plantago lanceolata* (Plalan)) was established in May 2023 in Göttingen (Central Germany) on a Luvisol soil with a silty loam texture in a randomized complete block design with three replications and a plot size of 4.8 m². The number of accessions

Table 1. Estimated forage dry matter yield, weed mass, and median winter survival in the early establishment of *Achillea millefolium* (*Achmil*), *Carum carvi* (*Carcar*), *Cichorium intybus* (*Cicint*), *Lotus corniculatus* (*Lotcor*), *Medicago lupulina* (*Medlup*) and *Plantago lanceolata* (*Plalan*).

Species	n ¹	Dry matter yield (g m ⁻²)			Weed mass (g m ⁻²)			Winter survival		
		Mean	Range ²	Pop	Mean	Range	Pop	Median	Range	Pop
<i>Achmil</i>	4	47.8 b	131.6	0.06	77.4 a	102.4	0.41	8.0 a	5	0.13
<i>Cicint</i>	4	130.7 a	201.5	0.56	49.6 a	113.9	0.4	7.0 ab	5	0.12
<i>Carcar</i>	4	50.3 b	177.6	0.43	79.0 a	166.9	0.22	6.5 ab	8	0.13
<i>Lotcor</i>	7	36.3 b	77	0.48	97.8 a	210.5	0.05	4.0 c	5	0.14
<i>Medlup</i>	4	77.1 b	164.7	0.002	69.3 a	105.2	0.13	7.0 ab	8	0.07
<i>Plalan</i>	7	56.6 b	139	0.009	88.5 a	173.3	0.05	5.0 b	7	0.02

The Pop columns provide *P*-values for the main effect of accessions per species as obtained from analyses of intraspecific variability. Different lowercase letters indicate significant differences among the species (*P* ≤ 0.05).

¹ Number of accessions per species.

² Distance between minimum and maximum values per target variable (range).

differed among species because of variable access in the market (*Lotcor* and *Plalan*: 7 and all other species 4 accessions). After seeding, two cleaning cuts against annual weeds were conducted. The early establishment of plants was assessed using dry matter yield and weed mass in autumn to define their competitive strength against weeds. For this, plots were harvested once in September during the establishment year 2023 using a plot harvester (Model HD 1500 Wintersteiger). Subsamples were dried (60°C, 48 h) to determine the dry forage yield. Winter hardiness was visually assessed in the spring of 2024 (first winter after establishment) using a rating scale from 1 (no plants survived) to 9 (all plants survived). Calcium ammonium nitrate (30 kg N ha⁻¹) was applied to each non-leguminous plot after seeding. Statistical analyses were performed with R studio package ‘nlme’ (Pinheiro *et al.*, 2025) using linear mixed effects modelling for the accumulated forage yield and weed mass. The main effect of species was treated as a fixed and the block was used as a random effect. Tukey’s HSD test was used post hoc to compare species means for forage yield and weed mass. The intraspecific variability was calculated as distance between minimum and maximum values per target variable (Range). A Kruskal–Wallis comparison of medians was applied for winter survival in the package ‘agricolae’ (De Mendiburu, 2023). The main effect of accessions was tested for each species separately with the block as random effect to determine the intraspecific variability per species for forage yield and weed mass. Kruskal–Wallis test was performed to evaluate accessions effects on winter survival.

Results and discussion

The average temperature and rainfall during the growing season (April–October) were 14.8°C and 438 mm, respectively, which was 3% warmer and 12% wetter than the long-term values (average temperature and precipitation were 14.4°C and 385 mm, respectively from 2013 to 2022). During winter 2023/2024 (December–February), the average temperature was 4.6°C which was warmer than the long-term value (3.0°C for 2013–2022). Temperatures remained below 0°C for 19 days in the considered winter period (average –2.8°C). The main effect of species was significant for all target variables (*P* ≤ 0.001), except weed mass (Table 1).

All investigated species were successfully established and survived the first winter. However, clear differences among species were observed, with *Cicint* showing the highest forage yield (*P* ≤ 0.001), whereas differences in weed mass (an indicator of competitive strength) were not statistically significant among species. Notably, with 28% *Cicint* had the lowest proportion of weeds, which was considerably lower compared to the other species. All species showed a huge variation among the accessions within species

but significant effects were found only for *Medlup* (forage yield), *Lotcor* (weed mass), and *Plalan* (forage yield and weed mass) (Table 1). Regarding winter survival, *Lotcor* showed significantly lower survival rates than other species. The values of winter survival were generally above five, indicating moderate survival. *Lolium perenne*, the most important forage grass in Germany, has a winter survival scale of, on average, 4–5 according to the official species variety list (Bundessortenamt, 2024). Interestingly, although there was a wide range of variation among accessions within the species, these effects were not statistically significant for all species. This indicates that the observed variation was not fully explained by the investigated factors, suggesting that the emergence and establishment were also influenced by additional variables that were not identified in this experiment. Such patterns are typical of wildflower species, which often exhibit delayed and variable germination rates.

Conclusion

All species except *Lotus corniculatus* showed moderate to high winter survival rates under the given conditions. However, it is important to note that the results of this study were based on a single site and the first year only. Therefore, there is a need to take a closer look at the variation among accessions within species and consider a broader range of traits, including long-term performance and nutritive value, ideally through studies extending over additional seasons and at multiple locations.

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Valorisation of the endemic legume shrub *Retama monosperma* as a resource for feeding small ruminants

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Abstract

In the framework of the European Protein Strategy, the leguminous shrub *Retama monosperma*, endemic to coastal dunes and arid lands of SW Iberian Peninsula and NW Morocco, was evaluated. Through a climate and environment harshness gradient within its natural range, 38 shrubs were selected; mature seeds harvested and nutritional quality analysed. The species maintained high feed nutritional profile towards higher temperature and drought conditions, with mean values (% dry matter basis) for dry matter, ash, crude protein, ether extract, neutral detergent fibre, acid detergent fibre and lignin of 93.93, 2.56, 25.15, 3.71, 43.38, 29.81 and 2.53, respectively. Seeds showed higher fermentation potential than soybean meal. Methane emissions during fermentation were lower than those of referenced protein feeds, with a methane reduction of 31.8% compared to soybean meal. *Retama monosperma* is of interest as an indigenous high-quality protein source as feed for small ruminants. Further research regarding antinutritional factors and product transfer to final users is needed.

Keywords: degradability, indigenous resource, methane, nutritional value, protein

Introduction

Significant environmental impacts and European reliance on imported protein pose a challenge to livestock farming sustainability (Albaladejo Román, 2023). Issues associated with soybean cultivation, including emissions and habitat loss, have prompted interest in local plant alternatives (Halmemies-Beauchet-Filleau *et al.*, 2018). *Retama monosperma* (L.) Boiss. is a leguminous shrub endemic to the SW Iberian Peninsula and Morocco, well-adapted to harsh environments, that produces abundant fruit and is grazed by sheep and goats (Muñoz-Vallés *et al.*, 2013). Furthermore, it shows strong adaptability in ecological restoration contexts. This study aims to assess the potential of *R. monosperma* as a sustainable protein source for small-ruminant feed.

Materials and methods

Samples of *R. monosperma* fruits were collected from 38 shrubs across four populations along a climatic gradient in the SW Iberian Peninsula: Valdevaqueros (666 mm rainfall, 17.2°C), Tres Piedras (522 mm, 18.5°C), Flecha del Rompido (395 mm, 18.4°C) and Manta Rota (422 mm, 18.4°C). Two zones (inner/semistabilised vs. sea-exposed) were sampled per population. Mature seeds were peeled, dried (40°C, 48 h), and subjected to a thermal-wet debittering: 24 h hydration, cooking (60 min, 100°C), 5-day soaking, and drying (50°C, 48 h). Crude seeds (CS) and treated seeds (TS) were ground

Table 1. Chemical composition of mature seeds (% DM) in the studied areas.

	V		T		F		M		Total
	Ex	In	Ex	In	Ex	In	Ex	In	
DM	91.6 ± 1.1	93.3 ± 0.9	93.1 ± 1.8	95.2 ± 0.3	94.3 ± 0.2	94.7 ± 0.0	94.5 ± 0.1	94.0 ± 0.0	93.9 ± 0.2
Ash	2.7 ± 0.0	2.4 ± 0.1	2.7 ± 0.0	2.5 ± 0.0	2.5 ± 0.0	2.4 ± 0.0	2.5 ± 0.0	2.5 ± 0.0	2.5 ± 0.0
CP	25.1 ± 1.7	24.6 ± 1.6	23.1 ± 1.3	25.3 ± 0.9	27.6 ± 0.6	24.3 ± 0.8	24.7 ± 1.3	25.4 ± 0.3	25.1 ± 0.4
EE	4.1 ± 0.2	4.0 ± 0.1	3.6 ± 0.2	3.6 ± 0.0	3.5 ± 0.5	3.8 ± 0.2	3.3 ± 0.2	3.4 ± 0.0	3.7 ± 0.0
NDF	43.8 ± 1.1	44.5 ± 1.5	43.5 ± 0.7	40.4 ± 0.7	41.7 ± 1.8	42.3 ± 0.1	45.2 ± 1.1	45.4 ± 1.1	43.3 ± 0.5
ADF	30.6 ± 0.9	29.6 ± 0.9	30.4 ± 0.4	27.9 ± 0.5	29.4 ± 2.2	29.4 ± 0.6	31.0 ± 0.3	30.0 ± 0.6	29.8 ± 0.4
Lignin	2.6 ± 0.1	2.0 ± 0.2	2.0 ± 0.3	3.1 ± 0.2	2.8 ± 0.3	2.3 ± 0.2	2.6 ± 0.2	2.4 ± 0.4	2.5 ± 0.1
NFC	24.1 ± 1.3	24.3 ± 0.3	26.9 ± 0.9	28.0 ± 0.7	24.5 ± 0.9	26.9 ± 0.8	23.1 ± 0.9	23.9 ± 0.9	25.1 ± 0.4

Data represent the mean ± SE of 5 independent replicates. V, Valdevaqueros; T, Tres Piedras; F, Flecha del Rompido; M, Manta Rota; Ex, exposed; In, inner; DM, dry matter; CP, crude protein; EE, ether extract; NDF, neutral detergent fibre; ADF, acid detergent fibre; NFC, non-fibrous carbohydrates.

(1 mm). Soybean (SBM) and pea (P) meals served as references. All samples were analysed for dry matter (DM), crude protein (CP), ether extract (EE), ash, neutral detergent fibre (NDF), acid detergent fibre (ADF) and lignin (Guzmán *et al.*, 2020), and non-fibrous carbohydrates (Detmann & Valadares Filho, 2010). Feeds were subjected to an *in vitro* assay: 7 samples (0.5 g) were incubated in 120 ml Wheaton bottles with culture media and rumen fluid from 7 goats; fermentation kinetics were assessed via gas production curves at 2, 4, 8, 12, 24, 48 and 72 h (Romero-Huelva *et al.*, 2013), with methane measured at 24 h. All the bottles were incubated in the same period. Nutritional values were compared by ANOVA across climatic gradients and among areas (inner, exposed). For the *in vitro* assay, Substrate (CS, TS, SBM, P) was a fixed factor, with populations as subgroups within CS and TS. Post-hoc Tukey tests were used to prevent the family-wise error rate using R software (v.4.3.1).

Results and discussion

Mature crude seeds showed medium to high CP content (19.7–30.5% DM). Seeds of *R. monosperma* exhibited an interesting nutritional profile, with CP, EE, and fibre levels (Table 1) comparable to common ruminant feeds like peas and beans (McDonald *et al.* 2010; De Blas *et al.*, 2019). The nutritional quality remained consistent across the climatic gradient, showing resilience to harsh conditions such as drought, soil nutrient scarcity and salinity, with no significant differences among areas. This suggest that *R. monosperma* could be considered as a novel nutritional feed source appropriate for future climate change scenarios, able to grow in degraded soils while maintaining high fruit production and seed nutritional quality. Furthermore, the species provides vital ecosystem services, including soil nutrient enrichment, amelioration of temperature extremes, wildlife refuge or support for bee populations (Muñoz-Vallés *et al.*, 2013).

Retama monosperma seeds exhibited higher fermentation potential ($A = 172$ and 180 ml for CS and TS, respectively) than soybean meal (SBM) (142 ml), but slightly lower than pea (187 ml). As shown in Table 2, treated seeds and peas produced more gas than crude seeds, with SBM showing the lowest value. This is consistent with the results of Bizzuti *et al.* (2023), who found a proportional relationship between non-fibrous carbohydrates and gas production. Finally, *R. monosperma* seeds generated significantly lower methane emissions than SBM (31.8 and 32.2% in crude and treated seeds, respectively). The presence of bioactive compounds could be related to these reduced emissions, due to interference with rumen processes (Christodoulou *et al.*, 2025).

Table 2. Parameters of *in vitro* ruminal fermentation kinetics.

	CS			TS			SBM	Pea	SEM	<i>p</i> -value
	V	T	F	V	T	F				
GP 24 h (ml (g DM) ⁻¹)	138 ^b	140 ^b	138 ^b	145 ^a	146 ^a	145 ^a	115 ^c	144 ^a	1.81	0.001
N-NH ₃ (%)	7.3 ^{bcd}	8.5 ^{bc}	8.9 ^{abc}	5.2 ^{cd}	4.5 ^d	5.9 ^{cd}	13.6 ^a	9.8 ^{ab}	1.32	0.015
CH ₄ (ml (l GP) ⁻¹)	87.1 ^b	89.7 ^b	86.9 ^b	87.3 ^b	88.9 ^b	86.3 ^b	111 ^a	102 ^{ab}	1.76	0.002

CS, crude seeds; TS, treated seeds; V, Valdevaqueros; T, Tres Piedras; F, Flecha del Rompido; SBM, soybean meal; GP 24 h, accumulated gas production after 24 h of incubation. Data from 7 independent replicates. ^{a,b,c,d} Means with different letters within each row differ significantly (*p* < 0.05).

Conclusion

Retama monosperma seeds exhibit a valuable nutritional profile, medium-to-high protein content, and resilience to harsh climates. They demonstrate favourable fermentation kinetics and reduced methane emissions compared to SBM. These preliminary results highlight this species as a promising, novel and endemic feed source. Future research will investigate bioactive compounds, the reduction of antinutrients via debittering, and *in vivo* digestibility to validate its potential.

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Drought tolerance and productivity in *Plantago lanceolata* cultivars under controlled conditions

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Abstract

In a mesocosm pot experiment, seven cultivars of *Plantago lanceolata* (ribwort plantain) were subjected to a drought stress with six replicates per cultivar. The experiment included two drought stress phases, each followed by a recovery phase. Four harvests were conducted: after the first drought phase, after the first recovery phase, after the second drought phase, and after the final recovery phase. A significant cultivar $\times$ treatment $\times$ harvest interaction was found for the dry matter yield ($F_{18,275} = 7.80$, $P < 0.0001$), indicating that cultivar responses to drought differed across harvests. On average, drought stress reduced total biomass yield by 26.7% across cultivars, with losses ranging from 3% in ‘Ecotain’ to over 45% in ‘Captain’. Stomatal conductance (g_s) decreased significantly under drought stress across cultivars ($P < 0.001$), indicating a clear physiological response to water limitation. These results reveal short-term resilience in *P. lanceolata* following moderate drought events.

Introduction

Climate change increasingly challenges forage production due to the rising frequency of drought events (Trnka *et al.*, 2014). To secure productivity in grassland systems, drought-tolerant species such as *P. lanceolata* (ribwort plantain) have gained attention as functional components in forage mixtures (Stewart, 1996; Lee *et al.*, 2015). Stomatal conductance (g_s) is widely recognized as a key physiological indicator of plant response to drought stress (Flexas *et al.*, 2004). Previous studies in crops such as maize and barley have demonstrated cultivar-specific physiological adaptations to drought, including differences in g_s (Li *et al.*, 2021; Lv *et al.*, 2023). The present study aimed to evaluate seven *P. lanceolata* cultivars, including two wild accessions, under drought and control conditions, focusing on productivity and g_s . We hypothesized that (1) cultivars differ in drought tolerance, and (2) stress responses are reflected in physiological adaptation in terms of g_s .

Materials and methods

A two-factorial mesocosm experiment considering the *P. lanceolata* cultivar and drought stress was conducted from September 2023 to December 2024 at the mesocosm station of the Hessen Department of Agricultural Affairs in Central Germany. A total of 84 mesocosms (25.2 cm diameter, 28 cm height, 14–18 kg soil per mesocosm) were used in a randomized complete block design with six replicates. The used soil consisted of a silty sand (63.8% sand, 28.0% silt, 8.2% clay) with a pH of 7.3. Seven accessions of *P. lanceolata* were tested, including five commercial cultivars (‘Captain’, ‘Ceres Tonic’, ‘Diversity’, ‘Hercules’, ‘Ecotain’) and two wild accessions. Sowing took place on 12 September 2023, with 50 seeds per pot (equivalent to 1000 seeds m⁻²) sown without pretreatment. No thinning was conducted, and approximately 20 plants per pot remained on average after winter. In early April 2024, each pot received 0.5 g N, 1.5 g P₂O₅ and 3.0 g K₂O (equivalent to approximately 100, 300 and 600 kg ha⁻¹, respectively). After the first three harvests, additional nitrogen was applied as urea

solution on 19 June, 23 August and 2 October 2024 (0.5 g, 1.0 g and 0.5 g per pot, respectively; = 400 kg N ha⁻¹ total seasonal input).

After successful overwintering, plants were harvested once on 8 May 2024 to 6 cm stubble height. Immediately after this harvest, drought stress was induced in the drought stress treatment by completely withholding irrigation, while control pots remained irrigated and were re-filled to their initial mesocosm weight. The first drought phase lasted for 36 days (9 May–14 June 2024), during which soil moisture declined by approximately 60%, and visible wilting occurred after about 14 days. After the drought phase, all pots were fertilized and fully irrigated to allow recovery. In total, four harvests were conducted: after the first drought, first recovery, second drought, and final recovery phases (December 2024). Plants were cut at 6 cm, separated into leaves and stems, and fresh and dry weights were recorded after drying at 60°C for 48 h to record the dry matter yield (DMY). Stomatal conductance (g_s) was measured weekly on six dates during the first drought stress period using a LI-600 (LI-Cor) with three readings per mesocosm. The mean g_s per mesocosm across dates was used for analysis. The statistical analyses were performed in R studio using linear mixed effects models with cultivar, treatment, and their interaction as fixed and block as random effects. Model formulae were: $DMY \sim \text{cultivar} \times \text{treatment} \times \text{harvest} + (1|\text{block})$ and $g_s \sim \text{cultivar} \times \text{treatment} + (1|\text{block})$. Comparisons of means were performed post hoc using Tukey's HSD method. To complement significance testing, compact effect sizes are reported as percent DMY change between drought and control per cultivar across harvests

Results and discussion

We detected a significant three-way interaction (cultivar $\times$ treatment $\times$ harvest; $F_{18,275} = 7.80$, $P < 0.0001$) for the DMY. This indicates that cultivar responses to drought depended on the harvest (i.e., response patterns varied across the four harvest dates). Percent DMY change between drought and control per cultivar across harvests ranged between -45.7% for 'Captain' and -3.0% for 'Ecotain' (Table 1). These percentage changes summarise the practical magnitude of cultivar responses. The drought treatment had a significant main effect on g_s ($F_{1,50} = 20.68$, $P < 0.001$), with mean g_s decreasing by approximately 18% under drought conditions compared to the control. No significant cultivar $\times$ treatment interaction was observed for g_s , indicating that cultivars responded similarly to drought stress in terms of stomatal regulation although considerable variation was found when comparing means for this interaction (Table 1).

On average, cultivars with significant reductions showed a 19% decline in g_s , while stable cultivars decreased by only 4% (Table 1). This finding indicates some variation in stomatal behaviour among cultivars, although the overall response to drought was consistent. The g_s aligns with the Ball–Berry model, where reduced g_s under drought reflects decreased photosynthesis (Ball *et al.*, 1987). Cultivars with more flexible g_s control may better balance CO₂ uptake and water conservation (Flexas *et al.*, 2004). Although absolute g_s values varied slightly among cultivars, their relative response to drought was consistent. However, in some cultivars g_s did not decline under drought which points to genotypic variability in physiological drought-stress adaptation.

These findings support our hypothesis of cultivar-specific reactions to drought stress and highlight better-performing cultivars such as 'Ecotain' and 'Hercules'. However, differences among cultivars were not linked to g_s and may be related to other physiological or morphological adaptations, such as root allocation, which requires further studies. The wild accessions exhibited lower DMY, likely due to genetic background and lack of breeding selection.

Table 1. Total dry matter yield (DMY, g mesocosm⁻¹ ±SE) and mean stomatal conductance (g_s , mol m⁻² s⁻¹ mean ±SE) of seven *Plantago lanceolata* cultivars under control and drought stress conditions.

Cultivar	Treatment	Total DMY	g_s
Captain	Control	9.4 ± 0.3 Aab	0.29 ± 0.02 aB
	Drought stress	5.1 ± 0.2 Bbc	0.22 ± 0.02 aA
Ceres Tonic	Control	8.7 ± 0.2 Aab	0.33 ± 0.02 aB
	Drought stress	5.0 ± 0.2 Bbc	0.27 ± 0.02 aA
Diversity	Control	9.1 ± 0.3 Aab	0.29 ± 0.02 aB
	Drought stress	5.2 ± 0.3 Bbc	0.23 ± 0.02 aA
Ecotain	Control	10.0 ± 0.2 Aa	0.29 ± 0.02 aA
	Drought stress	9.7 ± 0.3 Aa	0.26 ± 0.02 aA
Hercules	Control	6.5 ± 0.3 Abc	0.29 ± 0.02 aA
	Drought stress	6.1 ± 0.2 Ab	0.28 ± 0.02 aA
Wildtype 1	Control	5.7 ± 0.2 Ac	0.31 ± 0.02 aB
	Drought stress	4.8 ± 0.2 Bbc	0.26 ± 0.02 aA
Wildtype 2	Control	5.9 ± 0.2 Ac	0.27 ± 0.02 aA
	Drought stress	4.2 ± 0.2 Bbc	0.26 ± 0.02 aA

Different lowercase letters indicate cultivar differences within a treatment; uppercase letters indicate treatment differences within a cultivar (Tukey's HSD, $P < 0.05$).

Conclusion

This study confirms significant cultivar-specific variation in drought tolerance among *P. lanceolata* cultivars, with cultivars like 'Ecotain' maintaining physiological integrity and productivity under water stress. The results indicate potential to use drought-tolerant *P. lanceolata* cultivars in climate-resilient forage systems.

Acknowledgements

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Validation of a hand-held NIRS scanner for use in field-based grass breeding trials

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Abstract

Quality attributes are important factors in forage grass breeding, impacting production and animal performance. While forage breeding programmes have moved in recent years towards the use of field-based Near Infrared Reflectance Spectroscopy (NIRS) methods to evaluate dry matter content of forage, laboratory-based NIRS analysis continues to be used to rank candidates for other important quality parameters. The availability of instant results derived from field-based NIRS systems could improve efficiency and increase annual sample volume. The aim of this study was to validate the accuracy of a hand-held NIRS instrument compared with conventional lab-based NIRS methods using a plot-based field trial of early-stage breeding material. Calibrations were imported from the Master NIRS Instrument used for the Forage Analysis Assurance (FAA) Group maintained at AFBI Hillsborough for fresh forage and validated via regression analysis ($R^2 = 0.9084, 0.3938, 0.5194$ and 0.6174 for DM, CP, ADF and WSC, respectively). Testing was then conducted over a two-year period under a conservation cutting regime to compare candidate rankings based on the hand-held and laboratory-based NIRS analysis. As for other field-based NIRS systems, varying accuracy rates were obtained, suggesting that though hand-held systems may be useful for some quality parameters in fresh forage, accuracy remains problematic.

Keywords: field trials, hand-held NIRS instrument, Near Infrared Reflectance Spectroscopy (NIRS), nutritive value, perennial ryegrass

Introduction

The importance of quality parameters in breeding improved perennial ryegrass varieties is well documented. Breeding programmes have used quality parameters in addition to yield data to improve performance for many years using laboratory-based NIRS Near Infrared Reflectance Spectroscopy methods to determine dry matter, digestible organic dry matter, water soluble carbohydrate, fibre (ADF and NDF) and crude protein. As technology has advanced, field-based NIRS has become available and is widely used to evaluate dry matter content of forage using harvester mounted systems but reporting of the accuracy of portable hand-held systems on multiple quality parameters remains unclear. The aim of this study was to assess the accuracy of using a laboratory calibrated hand-held NIRS instrument (Aurora) to determine real-time forage quality parameters in perennial ryegrass breeding trials.

Materials and methods

In order to align the hand-held instrument with the laboratory-based NIRS Tecator 6500 (Hillsborough Master Instrument) the Aurora handheld NIRS instrument was calibrated with four equations (dry matter (DM), crude protein (CP), acid digestible fibre (ADF) and water-soluble carbohydrate (WSC)) from the Hillsborough NIRS Fresh Grass database spectral data and reference data. A batch of fresh samples was scanned with both instruments in order to validate the efficacy of the hand-held

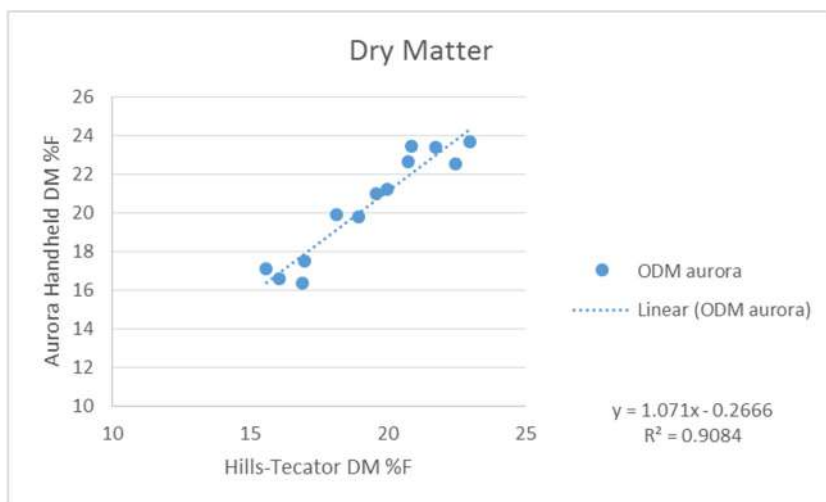


Figure 1. DM comparison of Aurora and Hillsborough NIRS calibrated using the Hillsborough NIRS Fresh Grass equations.

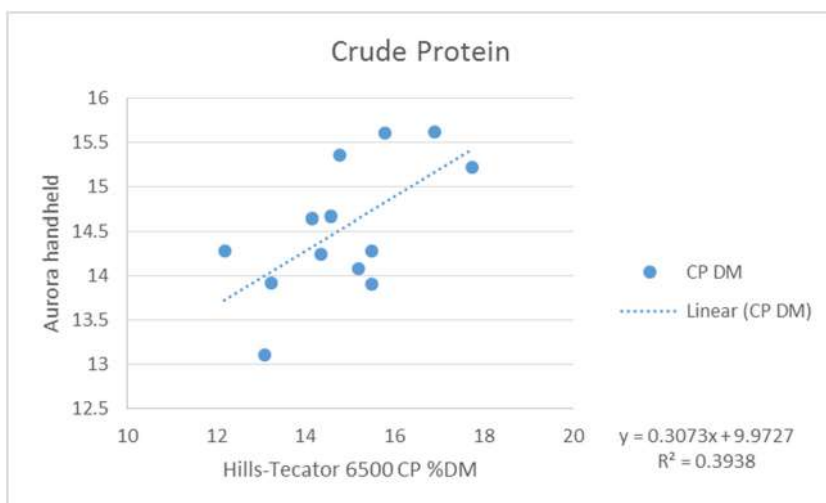


Figure 2. Hillsborough CP calibration—comparison of Aurora calibrated using the Hillsborough NIRS Fresh Grass equations.

instrument. The Aurora was then used to analyse fresh perennial ryegrass samples from a plot-based field trial at AFBI Loughgall before harvesting under conservation management. Obtained quality results were compared with the equivalent standard laboratory NIRS analysis of oven-dried, milled samples taken at harvest.

Results and discussion

Figures 1 and 2 show the DM and CP comparisons from the sample batch scanned by the Aurora and Hillsborough NIRS calibrated using the Hillsborough NIRS Fresh Grass equations. R^2 values of 0.9084 and 0.3938, indicate comparability of results in laboratory tested fresh grass samples. R^2 values 0.5194 and 0.6174 were obtained for ADF and WSC respectively. Following this the Aurora was used to scan a field trial of late diploid perennial ryegrass in AFBI Loughgall and the results

obtained compared with the standard laboratory quality analysis used within the breeding programme. Results (not shown) did not show any significant correlation. Discrepancies between laboratory results and field-based sampling highlight problems with accuracy that are similar to published findings and there are a number of possible indications for this including scanning methodology, discrepancies between homogeneity of samples, and whether the calibrations are suited to a range of environmental conditions (Feng, 2024; Yamada, 2024; Hart, 2020; Cherney, 2021 & Gorla, 2022).

Conclusions

In conclusion, although laboratory testing indicates the use of Aurora on fresh grass samples is possible for DM, CP, ADF and WSC, the efficacy of the instrument when taken out into field plots remains problematic. Further investigation is required to identify and correct these irregularities.

Acknowledgements

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***Lolium perenne* root architecture analysis using 2D and 3D imaging**

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Abstract

Manual root phenotyping techniques can be time consuming and laborious, requiring the extraction of whole root structures to facilitate manual measurements, resulting in the loss of *in situ* root architecture and orientation. Using rhizoboxes can preserve physical structures and allow for semi to fully automated analysis by combining photographs with image processing tools, with no manual root disturbance. Using this approach, we have developed a reliable method for growing perennial ryegrass (*Lolium perenne* L.) to examine a variety of root phenotypic traits. This method has included the use of 2D and 3D X-ray Computed Tomography (CT) scanning techniques to provide a comprehensive examination of root phenotypes.

Introduction

The ability to accurately and rapidly phenotype root systems in perennial ryegrass breeding programmes remains difficult largely because they grow below the soil surface. Methods that allow visualisation such as hydroponics and agar may affect root development, thereby changing the root architectural shape and underlying morphological traits (Kerstens *et al.*, 2021). Using root coring can effectively analyse rooting systems, but it is a destructive method and damages fine roots, while also not allowing for the tracking of continuous root growth (Wharton *et al.*, 2025). Digital annotation methods such as WinRHIZO scanning can reduce manual workload but generally still requires excavation and so are destructive (Han *et al.*, 2020).

Rhizobox based methods can alleviate some of these issues, enabling non-destructive and continuous 2D analysis. They utilise photography and automated digital root segmentation software to obtain root trait data, as seen in Malinowska *et al.* (2025). This omits the need for excavation, serving as a high-throughput root phenotyping method that reduces workloads. Automated photography can also be incorporated to effectively analyse large datasets. However, whilst this method possesses such advantages, the analysis is limited to the two-dimensional plane. This captures the planar projection of a 3D structure, potentially misrepresenting the three-dimensional structure and missing large proportions of the rooting system concealed in the soil.

This study will determine whether integrating 2D rhizobox imaging, WinRHIZO scans, and 3D X-ray CT digital twin reconstructions can provide complementary data, where each method can compensate for the others' limitations. The overall objective was to devise a technique for accurate and comprehensive measurement of varietal variations in root architecture traits.

Materials and methods

Eight individual perennial ryegrass (PRG) genotypes originating from modern PRG cultivars were clonally propagated and transplanted into custom built rhizoboxes, with the clonal propagation ensuring genetic uniformity within each genotype. Five replicates of each genotype were used with two

plants in each box. Rhizoboxes were filled with a homogenized mixture of peat, sand and topsoil, and angled at 60° to the horizontal, to encourage root growth along the transparent side. Plants were grown at 20°C under 16-h daylength. Root depth, total root length, lateral root development, and root angle were captured at specified time points using photography and digital analysis. Semi-automated root segmentation was performed using FIJI's SmartRoot plugin to obtain initial root depth, seminal root length and seminal root angle. Digital markers were used to trace roots and obtain root length, and a digital protractor was used for root angle.

A digital 3D reconstruction was achieved by 3D X-ray CT scanning. Round 3D printed PLA pots of 150 mm depth and 57 mm diameter and a wall thickness of 1 mm were used. X-ray CT scans were performed at 160kV and 250 μ A, with a scan time of 10 minutes. A resolution of 53.0 μ m was obtained. Finally, samples were excavated, washed, and scanned using WinRHIZO (version 2015a, Regent Instruments Inc.) to produce high resolution 2D images showing fine roots and lateral branching patterns. Each WinRHIZO scan provided a full planar projection of the excavated root system in a 2D image.

Results and discussion

Initial 2D analysis revealed that four out of eight PRG genotypes consistently exhibited two statistically significant root depth phenotypes ("shallow" and "deep"), (Fig. 1A). The use of semi-automated root segmentation (Fiji's SmartRoot) was used to significantly reduce manual annotation time, improving throughput without compromising accuracy (Fig. 1B). 3D CT scanning provided a three-dimensional view of the root system capturing structural features not visible in 2D analysis alone. However, due to resolution limitations, fine root features such as lateral roots and root hairs were less discernible (Fig. 1D). WinRHIZO scanning post-excavation addressed this limitation by providing high resolution images of fine roots, root hairs and overall rooting structure. These images can serve two functions; firstly, WinRHIZO scanning can be combined with CT scans to provide location specific morphological data obtaining root specific measurements, such as local root diameters, (Fig. 1E–F). This can theoretically be mapped to the corresponding locations on the digital twin, generated by X-ray CT scanning (Fig. 1D) to provide both spatial data and root point specific data in the 3D system. Secondly, WinRhizo scans can be inputted into Image J to be used in a direct comparison with rhizobox-derived images and WinRHIZO scans (Fig. 1C).

When comparing PRG lines excavated from round pots (scanned using WinRHIZO) to rhizobox derived images through SmartRoot, the 2D representations of the root systems (root length, branching and lateral root development) were generally comparable. However, whilst excavation and flattening of 3D structures can accurately mimic rhizobox images in WinRHIZO scans, the root systems may be distorted during preparation with respect to lateral distribution. Therefore, length to width ratio and root angle measurements may need to be adjusted or carried out solely in rhizoboxes for direct genotype-to-genotype comparisons. Rhizoboxes may prove advantageous here as they omit the need for excavation, likely capturing root angles and length-to-width ratios in their truer state without introducing preparation-induced distortions into the root system.

Conclusions

This study has demonstrated that integrating WinRHIZO scanning with 3D X-ray CT scanning provides detailed insights into fine root morphology that is not captured by either method alone. It can also serve as a valuable tool for comparing root growth differences between rhizoboxes and

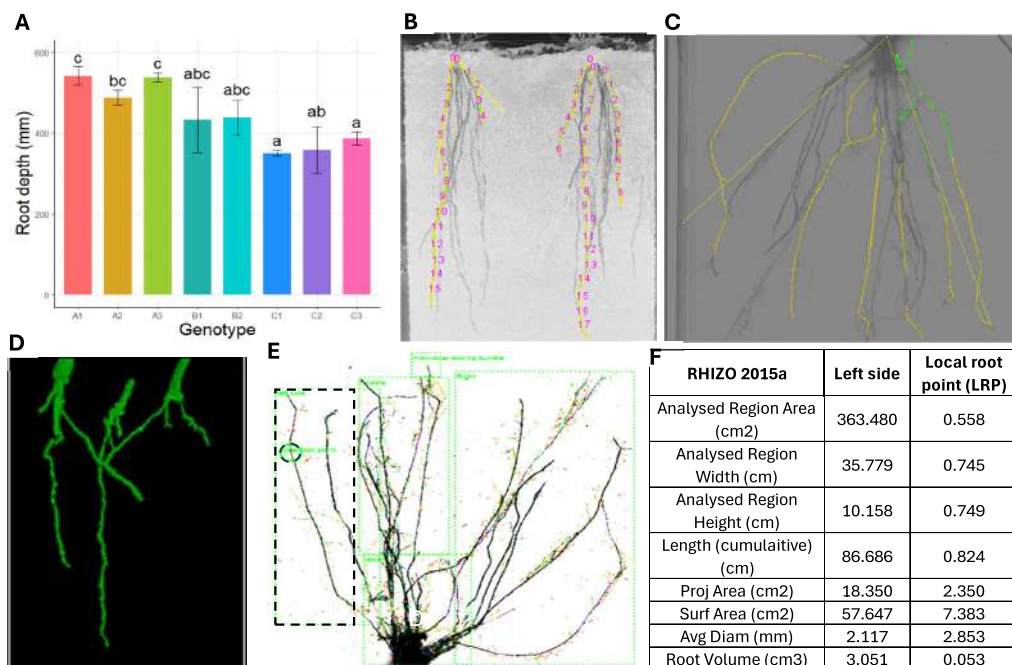


Figure 1. (A) Comparison of PRG genotypes root lengths obtained through using rhizobox images. (B) Rhizobox image of PRG root structures traced in SmartRoot. (C) A WinRHIZO scan of an excavated PRG line cultivated in round tube for CT imaging, partially annotated on SmartRoot. (D) 3D reconstruction of a PRG root structure scanned using CT X-ray scanning. (E) An analysed WinRHIZO scan using its native software showing two highlighted regions of PRG root system. (F) WinRHIZO location specific morphological data (encircled in WinRHIZO scan).

round pots as well as supplementing 3D structural analysis. The combination of 2D rhizobox imaging, CT scanning and WinRHIZO analysis offers a comprehensive root phenotyping approach with each method compensating for the limitations of the other. Combining these enabled the assessment of root angle and length, root biomass, volume and surface area, root hair characteristics and 3D root architecture. This approach may facilitate a better understanding of how complex 3D root structures can be represented as 2D planar projections, through the analysis of overall root architecture and fine scale morphological trait differences between varieties.

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Breeding Mediterranean annual clovers for new challenges and opportunities

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Abstract

Sown annual pasture and fodder legumes are used in areas with Mediterranean climates for their high nutritive value to livestock and their ability to fix atmospheric nitrogen. Annual Legume Breeding Australia (ALBA) is a joint venture between the University of Western Australia and the pasture seeds company, DLF Seeds, to breed more productive and resilient annual pasture and fodder legumes for the farming systems of southern Australia and other global regions with Mediterranean climates. Major species include subterranean clover (*Trifolium subterraneum* L.), Persian clover (*T. resupinatum* L.), balansa clover (*T. michelianum* Savi), and arrowleaf clover (*T. vesiculosum* Savi). Key breeding objectives include matching flowering times to growing season lengths of intended environments, increased winter and spring biomass, increased seed yields and stand persistence, resistance to pests and diseases, and low phyto-oestrogen contents. High hardseededness is a key trait in cultivars for crop rotations and low rainfall areas. Selection is also practiced for waterlogging tolerance in wetter target environments. Future traits include low bloating and methanogenic potential. Climate change is likely to see annual clovers used in marginal areas currently sown to perennial clovers.

Keywords: annual legumes, clovers, forage production, pastures, plant breeding, *Trifolium*

Introduction

Pasture and fodder legumes are highly valued by farmers for their high nutritive value for livestock and their ability to fix atmospheric nitrogen (N) to increase soil fertility. Grasslands of the Mediterranean basin have immense legume diversity (Davis, 1970) and many of these have become naturalised in other global regions (Porceddu *et al.*, 2016). Southern Australia also has a Mediterranean-type climate, but very few Australian native legumes are herbaceous and palatable to ruminant livestock, and many are toxic (Nichols *et al.*, 2012). This has led to selected native Mediterranean annual legumes becoming domesticated, whereby seeds are harvested and traded, and the development of a pasture seed industry. Active cultivar development programmes have operated since the 1950s with the evaluation of germplasm collected from the Mediterranean basin and, more recently, from breeding programmes (Nichols *et al.*, 2012). To date, Australia has domesticated 35 Mediterranean annual legume species and developed improved cultivars of them through breeding and selection (Nichols *et al.*, 2012). Many of these cultivars have subsequently been sown in the Mediterranean basin and other areas with similar climates to improve grassland productivity (Porceddu *et al.*, 2016). Of critical importance to these programs is a large genetic resource, held in the Australian Pastures Genebank, of almost 60,000 accessions, resulting from more than 90 collection expeditions since the early 1950s, particularly to grasslands in the Mediterranean basin and surrounding regions (Smith *et al.*, 2021).

Until recently, most of the breeding and selection of annual pasture legumes in Australia was conducted by public agencies. In 2017, the Western Australian State Government's annual pasture legume breeding program was purchased by the international pasture seed company, DLF Seeds, and a joint

venture, known as Annual Legume Breeding Australia (ALBA), was formed in 2018 with the University of Western Australia (UWA). The aim of ALBA is to breed more productive and resilient annual pasture and fodder legumes for southern Australia and other global regions with Mediterranean-type climates. This paper reports on annual pasture and forage legumes under development by ALBA and their roles in farming systems in Mediterranean-type climates.

Key species

The key species currently in the ALBA programme are subterranean clover (*Trifolium subterraneum* L.), Persian clover (*T. resupinatum* L.), balansa clover (*T. michelianum* Savi) and arrowleaf clover (*T. vesiculosum* Savi). These species total almost 90% of all certified annual pasture legume seed produced in Australia (Australian Seeds Authority, 2023). Much of this seed is sown by Australian farmers, although a high proportion is also exported to farmers in other countries with similar climates, including the Mediterranean basin. Other species will be included as market opportunities are identified.

Selection criteria for improved cultivars

Cultivars of the key species are being developed that match flowering and seed maturation times to growing season lengths of intended environments, with early-flowering cultivars for short-growing season areas and late-flowering cultivars for long-growing season areas.

Subterranean clover, with 53.7% of all annual pasture legume certified seed production, is the most important annual legume in Australia (Australian Seeds Authority, 2023). It is an inbreeding, self-regenerating species used in grazed pastures. Its main agronomic feature is its high grazing tolerance, due to its prostrate habit, flowers located close to the ground, and burial of its seed-bearing burrs, particularly for subspecies *subterraneum* and *yanninicum*. New cultivars of ssp. *subterraneum* and ssp. *yanninicum* are being developed for moderately acidic (pH 4.5–6.5) well-drained and poorly drained soils, respectively, while new ssp. *brachycalycinum* cultivars are being developed for neutral-alkaline (pH 6.0–8.0) soils. Key selection traits include increased winter and spring biomass, increased seed yields and stand persistence, resistance to important pests and diseases, and low phyto-oestrogen content. High hardseededness is also a key trait in cultivars for crop rotations and low rainfall areas.

Breeding and selection are being conducted on both types of Persian clover used in commerce (var. *majus* and var. *resupinatum*). Both types are outcrossing, have waterlogging tolerance (Rogers *et al.*, 2005), and are best suited to soils with pH 5.0–8.0 (Nichols *et al.*, 2012). While var. *resupinatum* is a self-regenerating, dual-purpose grazing and fodder type, var. *majus* is grown as a one-year fodder crop for high-quality hay or silage (although it can also be grazed during winter and early spring). The main breeding focus for var. *majus* is late flowering and high spring biomass with large leaves to maximise legume quantity and quality, while the focus for var. *resupinatum* types is high winter and spring biomass, high seed yield, and increased seedling regeneration in early flowering and midseason types.

Balansa clover is also an outcrossing, self-regenerating, dual-purpose grazing and fodder species. It has high waterlogging tolerance and mild salinity tolerance (Rogers *et al.*, 2005) and is best suited to moderately acidic soils. New early flowering, midseason and late flowering cultivars are being developed with greater seedling vigour, higher winter and spring biomass, high seed yields and greater stand persistence.

Arrowleaf clover is another out-crossing, self-regenerating grazing and fodder species. It is deep-rooted and can remain green after most other annual legume species have senesced. Breeding is mainly focussed on increasing winter and early spring biomass production and seed yields in early and mid-season maturity types.

Cultivars from ALBA

Three new subterranean clover cultivars have been commercialised by the ALBA programme (Nichols *et al.*, 2024). ‘Edison’ and ‘Carver’ are late-flowering ssp. *subterraneum* cultivars, while ‘Franklin’ is a midseason ssp. *yanninicum* cultivar. Early and midseason flowering ssp. *subterraneum* cultivars are under consideration for commercialisation and a ssp. *brachycalycinum* cultivar is under development. Cultivars of Persian clover var. *majus*, balansa clover, and arrowleaf clover are also under consideration for commercialisation.

Conclusions

Farmers in Australia and other regions with Mediterranean-type climates will benefit from more productive and persistent annual legumes from the ALBA programme, with resilience to climate change. Grazing industries will benefit from the production of high-quality feed and the atmospherically fixed N will increase the productivity of companion pasture grasses and herbs and will be available to crops grown in rotation. With climate change, the use of annual clovers is likely to increase into marginal areas currently sown to perennial clovers. Future annual legumes are also being sought with less bloat-risk to stock and a lower carbon footprint on the environment. The partnership with DLF Seeds, an international seed company, leads to the reliable provision of seeds for global adoption.

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Assessing agronomic performance and bioactive compounds in *Plantago lanceolata* ecotypes

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Abstract

Ribwort plantain (*Plantago lanceolata* L.) is a deep-rooted, fast-growing forage species, notable for its high drought tolerance and capacity to enhance nitrogen use efficiency. It synthesizes iridoid glycosides such as aucubin, acetoside and catalpol, contributing to natural parasite control and improved livestock health, supporting its role in sustainable grazing systems. In a two-year field trial conducted at the Georg-August University of Göttingen, 25 ecotypes collected primarily in Germany and two registered cultivars were evaluated for dry matter yield, morphological traits, and concentrations of aucubin, acetoside, and catalpol in leaf and stem tissues. Significant variation in yield and leaf proportion was observed across years and accessions, reflecting genotype-by-environment interactions. The two cultivars showed lower dry matter yield compared to many ecotypes, suggesting limited adaptability under the given site conditions. Bioactive compound concentrations were consistently higher in leaves than stems, supporting the hypothesis that leaves serve as primary sites for secondary metabolite synthesis and accumulation. These findings highlight the agronomic potential of locally adapted *P. lanceolata* ecotypes and their biochemical traits relevant to functional livestock nutrition.

Keywords: acetoside, agronomic traits, aucubin, catalpol, plantain

Introduction

Pastures are the main feed source for grazing ruminants in temperate regions, typically composed of perennial ryegrass (*Lolium perenne* L.) and white clover (*Trifolium repens* L.) (Chapman *et al.*, 2018). However, their growth and nutritional value often decline during summer due to soil moisture deficits and plant maturation, reducing herbage dry matter (DM) intake and animal performance (Thomas, 1984).

Given increasing weather fluctuations, identifying ecotypes suited to specific and changing environments is essential to guide breeding for stable growth and feed value. The present study aimed to assess agronomic performance and secondary metabolite profiles in ribwort plantain to identify accessions with potential for forage use.

Materials and methods

Twenty-five ecotypes, mainly collected in Germany, and two registered cultivars of ribwort plantain (DSV, Herkules), both derived from wild temperate-climate populations, were evaluated in a field experiment using a randomized complete block design with ten replicates over two consecutive years. Each ecotype and cultivar was planted as single plant in the field in April/May 2021, following germination and juvenile growth in the greenhouse. For each individual plant, dry matter yield, and leaf

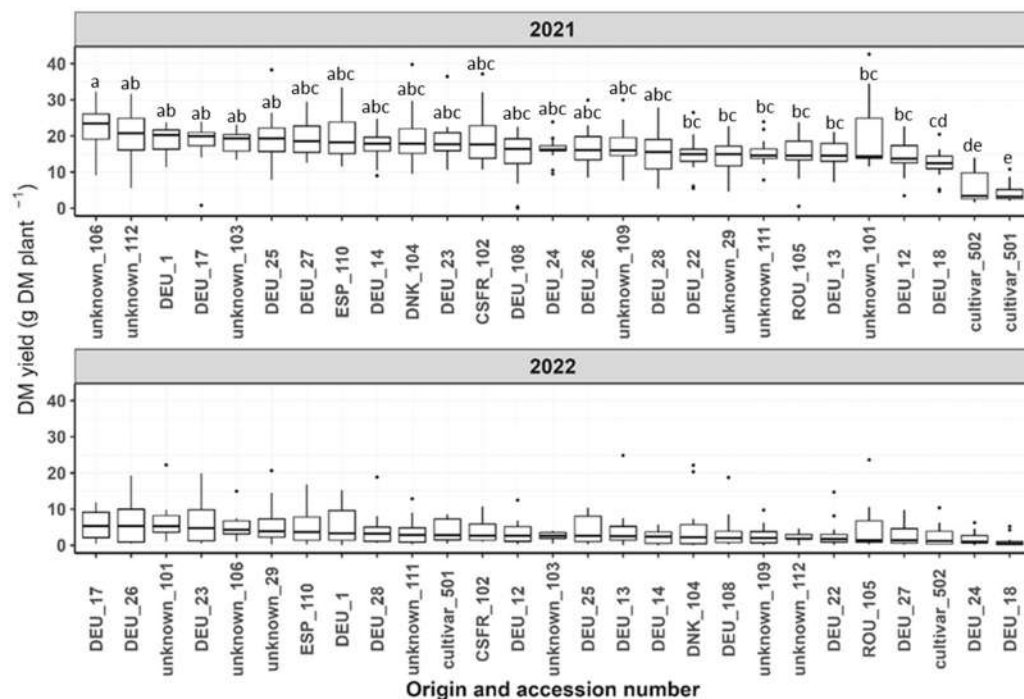


Figure 1. Dry matter yield (g plant⁻¹) of all accessions in 2021 ($n = 9$ per accession) and 2022 ($n = 9$ –16 per accession), ordered by descending median yield. Significant differences among accessions were detected only in 2021. Accessions not sharing a common letter differ significantly (Tukey's HSD test; $\alpha = 0.05$).

and stem biomass, were recorded in 2021 and 2022. Concentrations of the secondary metabolites aucubin, acetoside, and catalpol were quantified in leaf and stem tissues of a subset of plants harvested in 2021 ($n = 58$), which was selected to enable a preliminary assessment of phytochemical variability while accounting for budgetary constraints. This subset was subsequently analysed using HPLC following ethanol extraction of 0.1 g freeze-dried material, according to Navarrete *et al.* (2016). Statistical analyses were performed in R v4.3.0 (R Core Team, 2023) using RStudio v2023.06.1 (RStudio Team, 2023). Mixed-effects models were fitted using the lme function from the 'nlme' package (Pinheiro *et al.*, 2025), and estimated marginal means were calculated with the 'emmeans' (Lenth, 2023) package.

Results and discussion

The mean DM yield per plant across all accessions was 16.5 g in 2021, whereas in 2022 the mean yield decreased to 4.1 g per plant. This yield reduction is most probably due to the hot and dry summer in 2022. Yield declines in the second and subsequent years have been documented previously (Lee *et al.*, 2015; Chesney *et al.*, 2025) albeit to a lesser extent than those observed in the present study. The DM yield per plant of the two cultivars was generally lower than that of the ecotypes (Fig. 1). In 2021, all ecotypes outperformed the cultivars, with an average yield difference of approximately 14 g per plant.

However, under drier conditions in 2022, this trend was less pronounced; not all ecotypes exceeded the performance of the cultivars, and the average yield difference between ecotypes and cultivars was reduced to 0.8 g per plant (Fig. 1).

Concentrations of all three bioactive compounds were significantly higher in leaf tissue compared to stem tissue, supporting the hypothesis that leaves function as the principal sites of secondary metabolite biosynthesis and accumulation (Dalar *et al.*, 2012; Dixon & Dickinson, 2024). Aucubin ranged from 0.026 to 25.64 mg (g DM)⁻¹, catalpol from 0.10 to 19.60 mg (g DM)⁻¹, and acetoside from 0.20 to 123.86 mg (g DM)⁻¹, with mean values of 3.08, 4.29, and 32.75 mg (g DM)⁻¹, respectively. Although variation among accessions was observed for all three compounds, the differences were not statistically significant. This aligns with findings in the literature, where genotype-specific effects on the accumulation of bioactive compounds have been reported, but environmental conditions and plant developmental stage are generally considered to have a more pronounced influence. In particular, mature leaves tend to exhibit higher concentrations of these compounds than younger ones (Bowers & Stamp, 1992; Darrow & Bowers, 1997).

Conclusion

The observed variability in key traits among *P. lanceolata* ecotypes constitutes a valuable genetic resource for the enhancement of forage production and livestock nutrition.

Further evaluation of these accessions is necessary, as they provide a critical genetic resource for targeted breeding programmes.

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Assessment of standing crop biomass using remote sensing tools in rangelands of Morocco

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Abstract

Grassland degradation and climate change threaten livestock productivity and ecosystem services in the Middle Atlas. This study aimed to estimate standing crop biomass (SCB) in the Timahdite rangelands, particularly in the Ait Arfa du Guigou tribe area, using in situ biomass sampling and vegetation indices (VIs) derived from Landsat 8 OLI imagery. Biomass data were collected from 60 sites, and five VIs (NDVI, RVI, DVI, OSAVI, and MSAVI) were computed. Regression models (linear, exponential, power, and logarithmic) were used to assess the relationship between VIs and SCB. The OSAVI showed the strongest correlation with field biomass ($R^2 = 0.77$; MAPE = 11.03%), outperforming the other indices. Site-specific analyses also highlighted the strong model performance in Hebri and Boutassaf using MSAVI and OSAVI. This study demonstrates the potential of remote sensing for reliable SCB estimation, supporting sustainable rangeland management, and improved livestock feeding strategies in mountainous ecosystems.

Keywords: biomass, Landsat 8, rangeland degradation, Timahdite, vegetation indices

Introduction

Livestock productivity faces several constraints due to climate change and degradation of rangelands. Attempts to alleviate these constraints have been implemented through the use of local alternative feed crops (Boukrouh *et al.*, 2023c,d; 2024a,b). However, grassland ecosystems play a crucial role in the livelihoods of local communities and the conservation of biodiversity. They host interesting fodder species that increase livestock productivity (Boukrouh *et al.*, 2023a,b). In the Middle Atlas, particularly in the Timahdite area, they were estimated to cover 55–60% of the nutritional requirements of 22% of Moroccan sheep herds. For sustainable biodiversity conservation and soil erosion prevention, the estimation of standing crop biomass is a key component. Advancements in remote sensing technologies create opportunities to make this estimation (Chawicha and Tussie, 2023). The aim of this study was to develop reliable models with sufficient precision to estimate the standing crop biomass of the Ait Arfa du Guigou tribe, a mountainous grassland in the Middle Atlas, using in-situ biomass collection and vegetation derived from Landsat 8 OLI satellite images.

Materials and methods

Standing crop biomass data were collected during the peak of the photosynthetic capacity of the vegetation (from the end of April to the end of June 2016) and consequently, the maximum remote sensing reflectance values. Standing crop biomass was sampled according to stratified random sampling from 60 different homogeneous (90 m×90 m) sites across all strata (30 in Hebri, 14 in Boutassaf,

Table 1. Regression models for the relationship between standing crop biomass (*Y*) as a dependent variable and vegetation indices (*X*) as predictor variables for grazing areas in the study.

VIs	Equation	<i>R</i> ²	MAE (kg ha ⁻¹)	RMSE (kg ha ⁻¹)	MAPE (%)
Whole Timahdite area					
NDVI	$Y = 1881.7 \times X - 62.42$	0.74	41.88	52.78	12.67
MSAVI	$Y = 3953 \times X - 173.43$	0.67	44.60	60.12	12.83
OSAVI	$Y = -141.59 + 2682.1 \times X$	0.77	37.86	49.47	11.03
DVI	$Y = 4276.3 \times X - 35.607$	0.35	67.72	84.14	19.98
RVI	$Y = 519.2 \times X - 463.33$	0.71	44.82	56.06	13.61
Grazing areas					
Hebri					
MSAVI	$Y = 3595.4 \times X - 95.69$	0.71	37.17	44.71	8.73
OSAVI	$Y = -2449.12 \times X + 529.04$	0.71	37.09	44.97	8.77
Boutassaft					
MSAVI	$Y = 10386X^{1.67}$	0.67	41.86	64.02	13.38
OSAVI	$Y = 5540.7X^{1.66}$	0.63	44.92	67.40	14.32
Bouangar					
NDVI	$Y = 2038.77 \times X - 70.51$	0.47	26.69	34.02	8.39
OSAVI	$Y = 2448.0 \times X - 101.14$	0.43	26.06	31.88	8.14

MAE, mean absolute error; MAPE, mean absolute percentage error; RMSE, root mean square error; NDVI, normalised difference vegetation index; OSAVI, optimised soil adjusted vegetation index.

and 16 in Bouangar) using 300 representative (1 m×1 m) quadrats. The study sites were selected to represent the region’s ecological diversity, encompassing various vegetation types, landscape features, and soil compositions. For the remote sensing data, Landsat 8 OLI satellite images closest to the dates of field vegetation sampling were downloaded. Then, they were pretreated using ENVI v5.1 software. Using ArcGIS software, five vegetation indices, including the normalized difference vegetation index (NDVI), ratio vegetation index (RVI), difference vegetation index (DVI), optimized soil-adjusted vegetation index (OSAVI), and modified soil-adjusted vegetation index (MSAVI), were calculated for each grazing area and sampling period. Regression models (linear, logarithmic, exponential, and power) were then established to relate these indices to the standing crop biomass.

Results and discussion

In the Timahdite area, the overall standing crop biomass averaged 368±107 kg ha⁻¹, which is lower than the 859 kg ha⁻¹ recorded by El Aich *et al.* (2021) in 1979. The total standing crop biomass found in the three grazing areas differed, with values of 441 kg ha⁻¹, 292 kg ha⁻¹ and 311 kg ha⁻¹ for Hebri, Boutassaft, and Bouangar, respectively. These differences in standing crop biomass among grazing areas can be explained by the decrease in precipitation (553 mm recorded at Hebri and 443 mm at Bouangar) and the higher grazing pressure observed in Boutassaft and Bouangar (El Aich *et al.* 2021).

This study demonstrated a strong linear relationship between OSAVI and field observations, leading to the adoption of a linear model for predicting the available forage for sheep in these rangelands. Similarly, Fern *et al.* (2018) suggested that the OSAVI is the most appropriate vegetation index-based estimator of green biomass in the semi-arid regions of southern Texas. Meshesha *et al.* (2020) concluded that although NDVI is the spectral vegetation index most widely used by ecologists, it may not always be the best vegetation index in all regions and for all vegetation types. Analyses performed on the three grazing areas separately showed significant correlation coefficients in the Hebri grazing area (*r* = 0.85) for a linear model based on MSAVI and OSAVI, in the Boutassaft grazing area (*r* = 0.81) for a power model based on MSAVI, and a correlation of 0.69 in the Bouangar grazing area for a

linear model based on NDVI. The findings of Gholami and Oldeland (2019) in the arid rangelands of Iran were different and reported that soil-adjusted indices performed worse than a simple square root transformation of the NDVI.

Conclusions

The OSAVI was the best VI for estimating standing crop biomass, with a high R^2 and low error (MAPE = 11.03%). However, the selection of an appropriate vegetation index for predicting standing crop biomass depends on the specific characteristics of the study area and the type of vegetation.

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Breeding pasture and forage species for Mediterranean climate systems: The INIAV-Elvas approach

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Abstract

Permanent pastures play a crucial role in Mediterranean systems, particularly in Portugal, where they occupy 52% of Portugal's agricultural land use. These pastures are the basis of extensive livestock systems, offering year-round forage in regions limited by seasonal rainfall and drought. The plant breeding of forage species, especially legumes and grasses, aims to enhance pasture and forage productivity, resilience, persistence, and nutritional quality. Conventional plant breeding methods remain central to genetic improvement. At INIAV-Elvas, plant breeding programmes are being carried out on several species of legumes and grasses, based on the collections and genetic germplasm preserved in the Portuguese Plant Germplasm Bank. Once the most promising genotypes/populations have been identified in these collections, autogamous species are selected in the breeding programme; intraspecific diversity is promoted in allogamous species, followed by consecutive years of selection. After this first selection level, agronomic tests are carried out. The aim is to obtain populations/ genotypes that are distinct from those currently available on the market, that demonstrate homogeneity and stability, and are better adapted to the Mediterranean climate and expected climate changes. INIAV is currently the breeder of 40 registered forage varieties.

Keywords: forage grasses, forage legumes, Mediterranean agriculture, permanent pastures, plant breeding

Introduction

In Mediterranean climate regions, livestock agroforestry systems occupy vast areas. Rainfed permanent pastures contribute significantly to these agricultural systems, as they form the basis of ruminant feeding and, when properly managed, provide multiple ecosystem services (biomass production, climate regulation, soil erosion control, biodiversity conservation, water retention, pollination, and recreation) and social benefits.

These ecosystems are particularly vital in extensive livestock systems, supplying forage year-round in regions characterized by relatively mild winters, very hot summers, and high intra and interannual variability in precipitation (Seager *et al.*, 2019).

According to Noleppa and Carlsburg (2021), plant breeding in the EU over the past two decades has contributed significantly to increased yields and production in arable farming and, consequently, to improved market and trade conditions, increased food availability, greater economic prosperity, and additional agricultural income, while avoiding additional land use, greenhouse gas (GHG) emissions, and biodiversity loss.

In the semi-arid regions of the Mediterranean, self-reseeding annual legumes and perennial grasses with summer dormancy can play an important role in feeding livestock, particularly ruminants. The plant

breeding objectives of these species including climate change adaptation, is fundamental to enhance pasture and forage productivity, resilience, persistence and nutritional quality.

For agro-silvo-pastoral systems typical of Mediterranean regions, improved genotypes must consider current and future environmental conditions in order to make permanent rainfed pastures more adapted to changes in seasonal precipitation patterns and rising temperatures caused by climate change, which have consequences during growth periods.

Materials and methods

INIAV–Elvas Innovation Hub, located in south-eastern Portugal, has for many years conducted conventional breeding programmes structured according to the particular objectives defined to each species. At present, they are at different stages of implementation. Moreover, the various phases of such programmes are conducted annually and in parallel.

The working collections and germplasm resources conserved at the Portuguese Plant Germplasm Bank, currently holds 3328 accessions of pastures and forage species, from the wild flora of mainland Portugal and similar ecoregions. These collections comprise *Fabaceae*—particularly the genera *Astragalus*, *Lathyrus*, *Lupinus*, *Ornithopus*, *Medicago*, *Scorpiurus*, *Trifolium* and *Vicia*—as well as *Poaceae*, specifically *Dactylis* and *Festuca*.

The genetic diversity has been utilized in the different breeding programmes, applying specific methodologies to self-pollinated and cross-pollinated species. From these collections, the pre-breeding and breeding programmes select individuals using the maximum number of traits; afterwards, these are incorporated into elite plant material, i.e., new varieties. This is a gradual, phased process, and therefore plant material exists at different stages of evaluation.

In defining the selection criteria for pasture and forage species adapted to Mediterranean regions, the INIAV-Elvas team takes into account ongoing climate change and prioritizes the identification of the most suitable species for each agroecosystem, and within these, the selection of genotypes with the following characteristics: (i) high winter vigour/rapid establishment; (ii) shorter cycle/earlier flowering; (iii) high efficiency in the use of water and soil nutrients; (iv) tolerance to waterlogging and severe stress; (v) high biomass and seed production; (vi) high proportion of hard seeds or the ability to bury seeds; (vii) in the case of perennial species, the ability to accumulate reserves and present some dormancy to cope with increasingly prolonged summer periods; and (viii) high grazing tolerance.

Furthermore, achieving optimal nutritional values (protein content, dry matter digestibility, and fibre components) has always been an integral consideration.

Once the most promising genotypes/populations have been identified in these collections, the most interesting autogamous species are selected to continue in the breeding programme to reach the best solution, and intraspecific diversity is promoted in allogamous species, followed by consecutive years of selection. After the identification of superior genotypes/ populations, agronomic tests are carried out.

Table 1. Improved varieties developed by INIAV and registered in the 2025 National Catalogue of Varieties.

Species	Variety(ies)	Year	Aptitude
<i>Astragalus pelecinus</i> L.	Arga	2016	Pasture
<i>Avena sativa</i> L.	Boa-Fé, S. Aleixo, S. Eulália, Santa Rita	1982/1986/1995/2002	Forage
<i>Dactylis glomerata</i> L.	Delta 1	2011	Pastures
<i>Festuca arundinacea</i> Schreber	Ariel		Pastures
<i>Lathyrus cicera</i> L.	Grão da Comenda, Grão da Gramicha	2001/2002	Forage
<i>Lupinus albus</i> L.	Estoril	1990	Forage
<i>Lupinus angustifolius</i> L.	Giribita	2010	Forage
<i>Lupinus luteus</i> L.	Acos, Cardiga	1996/1990	Forage
<i>Medicago doliaata</i> Carmign.	Atalaia	2002	Pasture
<i>Medicago polymorpha</i> L.	Lentisca	2002	Pasture
<i>Medicago truncatula</i> Gaertn.	Revilheira	2002	Pasture
<i>Ornithopus compressus</i> L.	Cetus	2016	Pasture
<i>Ornithopus sativus</i> Brot.	Ara	2016	Pasture
<i>Trifolium alexandrinum</i> L.	Belém, Sodrê	1990/2025	Pastures
<i>Trifolium hirtum</i> All.	Enigma	2019	Pasture
<i>Trifolium resupinatum</i> L.	Mamede	2024	Pasture
<i>Trifolium suaveolens</i> L.	Maral, Resal	1993	Forage
<i>Trifolium s. ssp. brachycalycinum</i> Katzn. & F.H.W.Morley	Davel, Dom Dinis, Dom Pedro, Dom Sancho, Romel	1996/2016 /2016 /2016/1996	Pasture
<i>Trifolium vesiculosum</i> Savi	Lança	2020	Pastures
<i>Vicia benghalensis</i> L.	Fontainhas, Lage	1984	Forage
<i>Vicia sativa</i> L.	Barril, Gil Vaz, Piedade	1984	Forage
<i>Vicia villosa</i> Roth.	Amoreiras, Casal	1984	Forage
×Triticosecale Wittm. Ex A. Camus	Fronteira, Gavião, Monsaraz	1999/2019/2019	Forage

The aim is to obtain populations/genotypes that are distinct from those currently available on the market, that demonstrate homogeneity and stability, and that are better adapted to the Mediterranean conditions and expected climate changes.

Breeding programmes are carried out on a wide diversity of pasture species and aim the adaptation to different ecological conditions and grazing systems, to ensure optimal establishment, productivity, quality, and persistence.

Results and discussion

The breeding programmes conducted by INIAV-Elvas have consistently obtained pasture and forage varieties that outperform those currently available on the market. These varieties exhibit genetic uniformity and phenotypic stability, and are better adapted to the Mediterranean climate as well as to the scenarios of future climate change. This is in-line with the FAO's suggestions (EEA, 2019) for risk reduction, soil and water conservation and efficient water management.

INIAV is the registered breeder of 40 pasture and forage cultivars from the 2025 National Catalogue of Varieties: 1 astragalus (*Astragalus pelicinus*.); 4 forage oats (*Avena* sp.); 1 cocksfoot (*Dactylis glomerata*); 1 tall fescue (*Festuca arundinacea*); 2 grass peas (*Lathyrus* sp.); 2 lupins (*Lupinus* sp.); 2 narrow-leaf lupins (*Lupinus angustifolius*); 3 annual medics (*Medicago* sp.); 2 birds'-foot (*Ornithopus* sp.); 12 annual clovers (*Trifolium* sp.); 7 vetches (*Vicia* sp.); and 3 forage triticales (×Triticosecale Wittm.) (Table 1).

Conclusion

The high genetic diversity from native species demonstrates the importance of continuing to search and collected this type of material. These programmes are long-term, dynamic and costly processes,

but crucial to obtain varieties that are extremely well adapted. The number of varieties obtained show the success of the breeding programmes.

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A new breeding line for fodder galega (*Galega orientalis* Lam.)

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Abstract

Fodder galega (*Galega orientalis* Lam.) is the most persistent leguminous species cultivated on semi-natural grasslands in Estonia, with a lifespan exceeding ten years. Its ability to fix atmospheric nitrogen symbiotically contributes to lower nitrogen fertilizer requirements. Introduced to Estonia over 50 years ago, one domestically developed variety 'Gale' has been selectively bred for local conditions. In breeding the new line, attention has been given to its potential future use in the production of dehydrated forage and protein concentrates, as well as for use as an energy crop. Sustainable sources of protein are urgently needed for both human food and animal feed. A key objective is to breed a variety which has increased the leaf proportion to enhance self-sufficiency in plant-based protein production under the pressures of climate change. The new breeding line consistently yielded more dry matter and exhibited higher leaf and crude protein yields than the cultivar 'Gale' in all cuts across both trial years. In addition, the nutritive value of the new breeding line LL2008 was slightly higher than that of 'Gale'.

Keywords: DM yield, Galega, leaf proportion, nutritive value

Introduction

With changing climatic conditions, there is an increasing need to identify forage crops that are both economically viable and rich in nutritional value. The growing global demand for protein has intensified the search for sustainable alternative sources for both monogastric feed and human food. Perennial species enhance long-term productivity, resource efficiency, and ecological stability. Fodder galega (*Galega orientalis* Lam.) is a winter-hardy, tap-rooted perennial legume cultivated in Estonia for over fifty years and well adapted to temperate climates (Raig *et al.*, 2001). It can reach heights of up to 150 cm under moist conditions and is valued for its high yields, persistence, and nutritive quality. Its efficient leaf canopy ensures uniform light penetration and nearly complete radiation absorption, supporting high biomass productivity (Raig *et al.*, 2001). Pure stands can persist for over 25 years, yielding 9.56–11.0 Mg ha⁻¹ of dry matter annually without stand thinning (Slepetys & Adamovics, 2011; Adamovics *et al.*, 2011; Biliget *et al.*, 2024). The only Estonian cultivar 'Gale' was developed in 1987. It has been widely distributed across the Baltic region and Scandinavia owing to its high yield, winter hardiness, drought tolerance, and rapid regrowth after cutting. The aim of breeding the new line was to increase the leaf proportion and to explore its potential future use for producing dehydrated forage and protein concentrates. The study compared the dry matter yield and the nutritive value of the forage, leaves and stems of the cultivar 'Gale' with those of the new breeding line LL2008.

Materials and methods

The experimental field was established in 2022 at the Centre of Estonian Rural Research and Knowledge at Jõgeva (58°45'N, 26°24'E; average annual temperature 5.3°C and precipitation 670 mm). The study includes data from two years (2023–2024). The soil was a typical Calcaric Cambic Phaeozem (Loamic) with medium sandy loam texture (IUSS, 2015). Agrochemical indicators were as follows: pH_{KCl} 5.8 (ISO 10390); soil organic carbon content 1.6% (ISO 10694), phosphorus and potassium 191 and 220 mg kg⁻¹ (Mehlich III method), respectively; and Ca 1501 mg kg⁻¹. The trial was

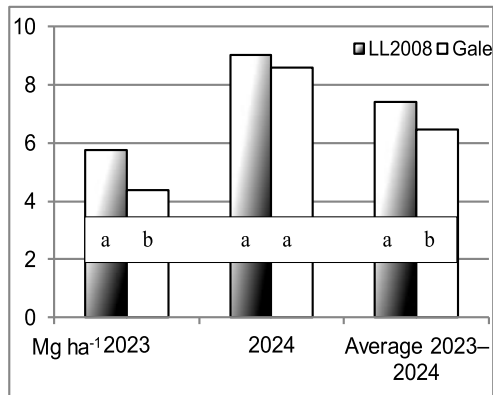


Figure 1. DM yield (Mg ha^{-1}). Different lowercase letters in the columns are statistically different ($p < 0.05$, Fisher LSD test).

arranged in a randomized complete block design with five replicates and a plot size of 6 m^2 . Seeding rate was 25 kg ha^{-1} . Prior to sowing, seeds were scarified and inoculated with species-specific *Rhizobium galegae* strain 740. No fertilizer was used during the experiment. Forage was harvested three times at early flowering in both trial years. No harvesting was conducted during the establishment year. Dry matter yield and quality were assessed; stems and leaves were separated to determine the stem-to-leaf ratio, and nutritive value was analysed.

During the study, the average air temperature from April to end of September was 13.9°C in 2023 and 14.6°C in 2024, which is $1.3\text{--}2.0^\circ\text{C}$ higher than the local long-term average of 12.6°C (1990–2024). Accumulated precipitation differed between years: 273 in 2023 and 364 mm in 2024, compared with the long-term average of 364 mm. Statistical analyses were performed using Agrobase Generation IITM. Data were analysed using one- and two-way ANOVA, and Fisher's LSD post-hoc test ($p = 0.05$) was applied.

Results and discussion

Dry matter (DM) yield of *Galega orientalis* cultivars was significantly dependent on the year (Year $\times$ cultivar $p < 0.001$). The breeding line LL2008 tended to yield more DM than cultivar 'Gale' in all cuts across both trial years (Figure 1). The annual yield in 2024 was greatly reduced by an early summer drought; however, abundant rainfall doubled the yield of the third cut for both cultivars.

A high leaf-to-stem ratio in the DM yield is a desirable trait in forage legumes. Previous studies have shown that the proportion of leaves in galega was on average 35% greater than in alfalfa, with leaf DM values ranging from 497 to 622 g kg^{-1} (Ignaczak *et al.*, 2022). In our study, the percentage of leaves in DM for LL2008 tended to be higher than in 'Gale' in 2023 (64–70% and 54–66%, respectively) and in 2024 (54–67% and 51–66%, respectively) (Figure 2). Under a three-harvest system, leaf protein yield reached up to 1180 kg ha^{-1} in LL2008 and up to 1136 kg ha^{-1} in 'Gale'. LL2008 has greater yield of leaves and crude protein than the cultivar 'Gale' in all cuts and both trial years (Table 1).

Previous studies have shown that the yielding ability can reach $8.5\text{--}10.5 \text{ Mg ha}^{-1}$ of DM and $1.7\text{--}1.8 \text{ Mg ha}^{-1}$ of crude protein (CP), with CP concentrations of $200\text{--}220 \text{ g kg}^{-1}$ DM (Raig *et al.*, 2001; Rinne *et al.*, 2022). Our results showed that the CP concentration of LL2008 was slightly higher than in 'Gale' across all cuts and both years. In 2024, LL2008 had a CP concentration of 208 g kg^{-1} DM in the first cut ('Gale': 205 g kg^{-1} ; range $145\text{--}236 \text{ g kg}^{-1}$), 187 g kg^{-1} in the second cut ('Gale':

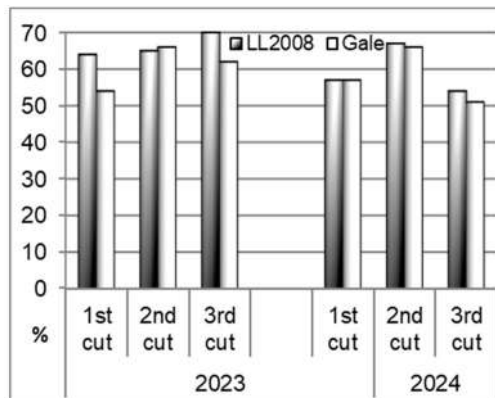


Figure 2. Proportion of leaves in DM.

Table 1. Yield of leaves and crude protein in dry matter 2023–2024

Year	Cultivar	Cut 1		Cut 2		Cut 3		Total	
		Leaf yield (Mg ha ⁻¹)	Leaf protein yield (kg ha ⁻¹)	Leaf yield (Mg ha ⁻¹)	Leaf protein yield (kg ha ⁻¹)	Leaf yield (Mg ha ⁻¹)	Leaf protein yield (kg ha ⁻¹)	Leaf yield (Mg ha ⁻¹)	Leaf protein yield (kg ha ⁻¹)
2023	LL2008	1.18	173	1.51	353	1.11	307	3.80	833
	Gale	0.65	88	1.13	260	0.91	263	2.69	611
2024	LL2008	1.47	362	1.43	298	2.33	520	5.24	1180
	Gale	1.36	346	1.31	282	2.14	508	4.81	1136

183 g kg⁻¹), and 191 g kg⁻¹ in the third cut ('Gale': 190 g kg⁻¹). In 2023, LL2008 had 145 g kg⁻¹ DM in the first cut ('Gale': 139 g kg⁻¹), 199 g kg⁻¹ in the second cut ('Gale': 192 g kg⁻¹), and 236 g kg⁻¹ in the third cut ('Gale': 228 g kg⁻¹). In both years and all cuts, nutritive values were above good levels where LL2008 was slightly better.

Conclusions

The breeding line LL2008 consistently yielded more DM than the cultivar 'Gale' in all cuts across both trial years. LL2008 also exhibited higher leaf and crude protein yields than 'Gale' in all cuts. Additionally, the nutritive value of LL2008 was slightly higher than that of 'Gale'.

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Enteric methane emissions from cows of a rustic-local breed (Salers) and a widespread breed (Charolais) grazing on permanent grassland

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Abstract

Grasslands are increasingly recognised for their role in mitigating enteric methane (CH₄) emissions from cattle, but climate change-induced stressors threaten both the cattle and pasture systems. To explore an adaptive grazing strategy by mixing breeds in the herd, this study measured CH₄ emissions from two cattle breeds on permanent semi-mountain grassland (Auvergne, France). In this trial, 18 Salers and 21 Charolais cows grazed a permanent grassland. The cows were weighed every 35 days. Methane emissions were recorded daily using two GreenFeed[®] systems. Data of CH₄ were averaged on 35 days according to the repeatability calculation results and were adjusted from diurnal variation. No significant difference in CH₄ emission was observed between the two breeds. However, since Charolais had higher metabolic body weight (MBW) than Salers, CH₄ emission per unit of MBW was significantly lower in Charolais. Methane production decreased significantly over time, regardless of breed, possibly because of decreased forage quality during the grazing season. Breed diversification within a herd may enhance the resilience of cattle farming to climate change without affecting the herd's CH₄ emissions.

Keywords: cattle breed, enteric methane emission, grazing, GreenFeed[®], permanent grassland, suckler cows

Introduction

The role of grasslands in mitigating methane (CH₄) emissions from cattle farming is gaining increasing attention, especially with the introduction of a new metric, the GWP*. However, climate change has intensified droughts and heatwaves, threatening both cattle and pastures. Thus, adaptive management strategies, such as identifying robust and adaptable cattle breeds in herd, are essential for sustainable grazing (Dumont *et al.*, 2020). Despite this, data on CH₄ emissions from different cattle breeds under similar grazing conditions remain limited. To address this gap, this study measured enteric CH₄ emissions from two French breeds while grazing on a permanent semi-mountain grassland. A local Salers breed from the Auvergne region was represented by cows from the beef branch of the breed. The Charolais breed, originally from Bourgogne and now widely distributed worldwide, was included for its resilience to heat stress (Brown-Brandl *et al.*, 2006). Based on the study by Dall-Orsoletta *et al.* (2018), which found no differences in daily CH₄ emissions between Holstein and Normande dairy breeds, no significant differences in daily CH₄ emissions between the two suckler breeds were expected.

Materials and methods

The experiment was conducted from June to September 2024 at the Herbipôle experimental farm in Laqueuille (Auvergne, France). Methane emissions were measured using two GreenFeed[®] (GF) sys-

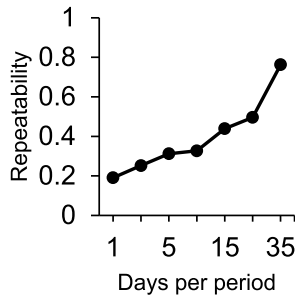


Figure 1. Repeatability of enteric CH₄ emissions in function of the number of days per period.

tems. Prior to the grazing trial, 28 Salers and 28 Charolais cows underwent a training period indoors to habituate them to the GF. Ten Salers and seven Charolais cows failed to visit the GF sufficiently and were excluded. The grazing trial was therefore carried out with 18 Salers (average live weight: 704 kg; average parity of four) and 21 Charolais (average live weight: 769 kg; average parity of four). Salers and Charolais cows grazed together on permanent semi-mountain grassland, rotating every two weeks across five plots (10 ha each, unrestricted size). Two solar-powered GF followed the herd and were placed near the water points to encourage frequent use. The cows grazed all day without forage supplementation, except bait feed offered by the GF (up to 240 g visit⁻¹, 8 visits day⁻¹). The cows were weighed every 35 days. Data recorded from GF at visit duration <2 min and airflow < 26 l s⁻¹ were excluded (Gunter *et al.*, 2017). Additionally, gas data (CH₄ and CO₂) exceeding 3 standard deviations from the mean were removed as outliers, resulting in the use of 6311 visit data (i.e., 92.6% of total). After data cleaning, diurnal variation in CH₄ emissions was corrected using a modified equation from Fresco *et al.* (2023). The total experimental period was divided into three sub-periods of 35 days each to reach a maximal repeatability of 0.76 for CH₄ data (Figure 1) using a modified equation from Arbre *et al.* (2016). A mixed model with repeated measurements was applied to data using SAS, considering breed (n = 2) and subperiod (n = 3) as fixed effects. Cows were included as a random effect, and subperiod was also treated as a repeated measure. Differences were considered significant at $P < 0.05$.

Results and discussion

In our study, grazing animals visited the GF units 2.3 ± 1.52 times day⁻¹, which was higher than the 0.7 ± 0.24 visits day⁻¹ observed on dairy cows under similar environmental conditions (Bouchon *et al.*, 2024), and comparable to the 2.5 ± 0.22 visits day⁻¹ reported on dairy cows in high latitudes (Lardy *et al.*, 2023). Daily CH₄ emissions were similar ($P = 0.91$, Table 1) between the two breeds (284 ± 6.4 vs 285 ± 6.9 g day⁻¹ for Charolais and Salers, respectively). However, CH₄ emissions per metabolic body weight (MBW) were lower ($P = 0.02$) in Charolais than in Salers (1.91 ± 0.04 vs 2.05 ± 0.04 g kg MBW⁻¹, respectively), reflecting greater ($P < 0.01$) MBW of Charolais compared to Salers (148 ± 2.0 vs 140 ± 2.1 kg, respectively). Regardless of breed, CH₄ emissions (g day⁻¹ and g kg MBW⁻¹) declined over time ($P < 0.01$), while MBW remained stable ($P = 0.06$). This decrease in CH₄ emissions may be linked to reduced forage digestibility throughout the grazing season (Boadi *et al.*, 2002).

Conclusion

Breed diversification within a herd is considered as an adaptation strategy to enhance the resilience of cattle farming to climate change. Nevertheless, there is still limited data on the environmental impacts of grazing multiple cattle breeds together in local grassland systems. Our study is meaningful

Table 1. Metabolic body weight and enteric CH₄ emissions during the grazing season in Salers and Charolais suckler cows

	Salers			Charolais			SEM	P		
	SP ² 1	SP 2	SP 3	SP 1	SP 2	SP 3		Breed	SP	Breed×SP
MBW (kg)	139	140	139	148	149	148	2.1	< 0.01	0.06	0.77
CH ₄ (g day ⁻¹)	299	284	273	298	279	276	7.1	0.91	<0.01*	0.38
CH ₄ (g kg MBW ⁻¹)	2.15	2.03	1.96	2.01	1.87	1.86	0.043	0.02	<0.01*	0.43

MBW, metabolic body weight (BW^{0.75}); SP, subperiod.

*Significant difference ($P < 0.05$).

by providing baseline data on enteric CH₄ emissions from both suckler cattle breeds (rustic-local Salers and widespread Charolais) on permanent semi-mountain grasslands. Our data suggest that breed diversity does not have an adverse effect on CH₄ emissions compared to a homogeneous herd. This implies that grazing together different breeds may offer a viable strategy to mitigate the challenges posed by climate change in grazing systems. However, a limitation of our study is the lack of data on forage intake under pasture conditions. Additionally, our dataset includes only two breeds. Future research will aim to collect more diverse data by including a wider range of breeds across broader systems and geographic regions.

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Forage: bioenergy dry matter herbage yield ratios of native Texas bunchgrasses

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Abstract

Interest in both forage production and second-generation biofuels and from grasslands in southern North America has increased in recent years. We measured biomass yield of 42 native bunchgrasses in five Texas locations over 2 years for dual-use forage and biofuel feedstock systems adaptable to market and climate fluctuations. Two types of harvests were evaluated: forage (F) repeated boot stage and end of growing season; biofuel (B) end of growing season. A F/B dry matter yield ratio calculated dual-use flexibility. Promising entries included *Tripsacum dactyloides* (F/B = 0.943), *Andropogon gerardii* (F/B = 0.942), *Trichloris crinita* (F/B = 0.852), *Sorghastrum nutans* (F/B = 0.783), *Schizachyrium scoparium* (F/B = 0.646) and *Trichachne californica* (F/B = 0.600). Native Texas perennial bunchgrasses show potential for dual use, flexible forage and biofuel systems.

Keywords: multipurpose, native bunchgrass, ratio

Introduction

Industrialization and modernization have increased energy demand, which still depends on fossil fuels that contribute to greenhouse gas emissions (Osman *et al.*, 2021). As an alternative, biomass—organic matter of plant or animal origin—shows potential as a renewable energy source (Naviza *et al.*, 2023). This study, part of the Southcentral SUN Grant (USDA, DOT, DOE), evaluated native Texas perennial grass microplots as forage and second-generation biofuel feedstock. These species require low inputs, tolerate drought, and improve soil health (Abraham *et al.*, 2004). Forage harvest systems were compared across four ecoregions, measuring yield and forage-to-biomass ratio.

Materials and methods

A multi-factorial, two-year experiment was conducted across five Texas locations Stephenville, Nacogdoches, Lubbock, Knox City, and Kingsville, using pre-existing established microplots with species varying by site. Forage biomass yields were calculated by combining summer harvest and regrowth from the same plant in the fall. Bioenergy biomass was determined from whole plant, once-per-season harvests each fall. Field plots followed randomized complete block designs, with two plants per replication (except in seed increase plots, where only one was collected). Each site had distinct soil and climate characteristics, ranging from fine loamy soil in Stephenville to sandy soils in Kingsville. From each location, the following species were collected by availability: Little bluestem (*Schizachyrium scoparium*), big bluestem (*Andropogon gerardi*), side-oats grama (*Bouteloua curtipendula*), Indiangrass (*Sorghastrum nutans*), eastern gammagrass (*Tripsacum dactyloides*), switchgrass (*Panicum virgatum*), silver bluestem (*Bothriochloa laguroides*), four-flower trichloris (*Chloris pluriflora*), two-flower trichloris (*Chloris crinita*), and seep Muhly (*Muhlenbergia reverchonii*). However, hooded windmill (*Chloris cucullata*) and Arizona cottontop (*Trichachne californica*) were collected from Lubbock rather than

Table 1. Average forage dry-matter yield: bioenergy (F:B) ratios for each location and year and for the most flexible species/accession per location and year.

	Average	Species/accession	DMY (kg ha ⁻¹)		F/B
	F:B		F	B	
2021					
Kingsville	1.21	Two-flower Trichloris	34964.29	41060.85	0.85
Knox City	1.70	Dropseed- Duck creek TX	7380.49	9423.80	1.10
Lubbock	0.25	Arizona Cottontop- La Salle	807.49	1346.26	0.60
Nacogdoches	1.34	Indiangrass-STN	22934.12	36618.20	0.63
Stephenville	0.37	Little bluestem-9110978	4012.92	8481.42	0.47
2022					
Kingsville	1.23	Big bluestem- Kenedy	3126.43	3319.82	0.94
Knox City	1.19	Indiangrass-41091012	4617.16	2700.00	1.71
Lubbock	—	—	—	—	—
Nacogdoches	—	—	—	—	—
Stephenville	1.09	Eastern gammagrass- local	6578.95	6976.47	0.94

switchgrass because the latter is poorly adapted to the semi-arid climate there. Forage biomass yields were calculated by combining summer harvest and regrowth from the same plant in the fall. Bioenergy biomass was determined from whole plant, once-per-season harvests each fall. Samples were assayed by year, season, species/accession, and location. ANOVA were performed by year and location to assess growth and yield differences among species and harvest type (forage vs. bioenergy), using the GLIMMIX procedure in SAS 9.4 with AR(1) covariance structure. Least squares means were calculated with LSMEANS, and differences were evaluated using PDIF and LINES (with PDMIX800 macro). Kenward–Roger adjustment corrected degrees of freedom. Significance was set at $\alpha = 0.05$.

Results and discussion

Dry matter yield per area indicated the potential for biomass production in each environment. Native bunchgrasses generally produce more biomass when harvested once, late in the growing season, compared to multiple forage harvests (Zhang *et al.*, 2020). However, in several cases in our data, the F:B ratio exceeded 1.0, meaning that the combination of summer forage and autumn regrowth surpassed the total biomass collected once a season in the fall, suggesting strong potential for flexible grazing systems. Average ratios were calculated per species, accession, location, and year. Ratios nearer 0.0 indicated higher biomass relative to forage, while those close to 1.0 suggested comparable forage and biomass yields.

In Kingsville during year (Y) 1, the average ratio was 1.21. The Big bluestem (Kennedy) had the highest DMY for B value (30963.92 kg ha⁻¹) compared to Silver bluestem (9093250), which had the highest DMY for F (10648.15 kg ha⁻¹). The Two-flower Trichloris recorded DMY values of 34964.29 kg ha⁻¹ and for F and 41060.85 kg ha⁻¹ B, resulting in the most balanced ratio (0.85) (Table 1). In Y2, the average ratio was 1.23. The Little bluestem (9089176) was the top DMY for B producer (7177.03 kg ha⁻¹). Switchgrass (PMC) led in DMY F (12091.22 kg ha⁻¹). The species Big bluestem (Kennedy) showed DMY levels of 3126.43 kg ha⁻¹ for F, and 3319.80 kg ha⁻¹ for B, yielding the most flexible use potential (0.94). At Knox City during Y1, the average ratio was 1.70. Arizona cottontop (La Salle) had the highest DMY in B (5048.47 kg ha⁻¹), Side oats grama (El Reno) had the highest DMY F (7590.44 kg ha⁻¹). Indiangrass (4190102) obtained DMY for F 7380.40 kg ha⁻¹, and for B was 9423.80 kg ha⁻¹, which indicates that it had the best dual-purpose

potential (0.78). Under Y2, the average ratio is 1.19. Switchgrass (Alamo) produced the most DMY for B (6892.23 kg ha⁻¹) < compared to Indiangrass (41091012) had the highest F (4617.16 kg ha⁻¹) and for B (2700.00 kg ha⁻¹), demonstrating multipurpose (1.71). During Y1 Lubbock during Year 1, the average ratio was 0.25. Indiangrass (4190102) had the highest DMY for F (3633.72 kg ha⁻¹), while Side oats grama (9112300) had the best DMY for B (8346.80 kg ha⁻¹). Arizona Cottontop (La Salle), where DMY was 807.49 kg ha⁻¹ for F and 1346.26 kg ha⁻¹ for B, derived a ratio of 0.06, the closest to 1 in this location, suggesting flexible use. At Y2, the ratios were not calculated due to plot loss. However, Indiangrass (91123211) had the biggest DMY (4928.75 kg ha⁻¹) for F. During Nacogdoches Y1, the average ratio was 1.34. Switchgrass (Alamo) had the best DMY B production (93564.89 kg ha⁻¹), and Little bluestem (Coastal Plains) had the highest forage (7080.69 kg ha⁻¹). Indiangrass (STN), where DMY was 22934.12 kg ha⁻¹ for F and 36618.20 kg ha⁻¹ for B, showed the best dual use (0.63). In Y2, the data is incomplete to calculate the ratio. The species had the highest DMY for F was Switch grass (9960.94 kg ha⁻¹). During Y1 in Stephenville, the average ratio was 0.371. Local Switchgrass had the highest DMY B (16962.84 kg ha⁻¹), Little bluestem (9110978) produced the most DMY for F (4012.92 kg ha⁻¹), also showing potential for dual-purpose use (0.47). During Y2, the average ratio was 1.09. Side oats grama (El Reno) led in DMY for B (4025.94 kg ha⁻¹), compared to Indiangrass (4190102), which obtained the highest value in F (3186.02 kg ha⁻¹). Eastern gamma grass (local) showed to DMY for F 6578.95 kg ha⁻¹ and for B 6976.47 kg ha⁻¹, resulting in this species the best flexible use (0.94).

Environmental factors influenced results. Stephenville had a higher average annual precipitation (802 mm), but cold snaps limited subtropical species like Seacoast bluestem, which thrives in Kingsville (average temperature 28.4°C; US Climate Data, 2022). Arizona cottontop performed better in dry, cold-prone areas like Lubbock, Knox City, and Stephenville. Biomass yield depended on water availability during the growing season, not just annual rainfall (Zhang *et al.*, 2020). Nacogdoches and Kingsville produced more biomass per plant due to consistent precipitation, while Lubbock produced less due to water scarcity. Species such as the hooded windmill and Arizona cottontop are adapted to arid west Texas. Switchgrass is currently produced for biofuel in southcentral North America (Parrish *et al.*, 2005). It is highly palatable for livestock when immature and shows strong regrowth once established. Eastern gammagrass and Big bluestem also demonstrated potential as dual-purpose grasses for forage and bioenergy systems.

Conclusion

This research highlights the potential of native Texas perennial bunchgrasses as flexible dual-purpose crops for both forage and bioenergy. Rainfall timing was more critical than annual precipitation for biomass production, and certain accessions within species were better suited to specific regions. Little bluestem, Switchgrass, Big bluestem, Indiangrass, Arizona cottontop, and Eastern gammagrass showed promising yields for either forage, biomass, or both. Their deep roots support drought resilience, soil conservation, and wildlife habitat. Future studies should focus on forage nutritive value, and ash content to better assess market-flexible biofuel potential vis-a-vis livestock use.

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Session 4.

Sustainable livestock production systems based on grasslands: Challenges and opportunities

Sustainability and trade-offs in livestock production systems: the role of grassland and local resources

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Abstract

Switzerland is predominantly a grassland country and, therefore, provides a particularly suitable model for examining sustainability measures at the grassland–livestock nexus. The present work synthesizes scientific evidence to critically assess the sustainability of livestock production systems that integrate grasslands and locally available resources in both ruminant and monogastric species. Site-adapted livestock systems are a long-term strategy for producing high-quality food while delivering other key ecosystem services such as biodiversity conservation, carbon sequestration, and landscape preservation. These systems also maximize resource use efficiency, reduce food–feed competition, improve animal health and welfare, and enhance positive externalities. Ensuring multifunctionality requires balancing productivity with environmental performance amid the pressures of climate change, labor shortages, and structural transformation in agriculture. Adaptive pastoral management, the integration of woody forage species, and targeted agricultural policies are essential for reducing emissions and nutrient losses, enhancing circularity within agroecosystems, and achieving the Farm to Fork Strategy objectives under the European Green Deal. Permanent but also temporary grasslands play significant roles, improving soil health, nutrient cycling, and carbon storage, increasing resilience, reducing chemical inputs, and benefiting subsequent crops. Collaboration among researchers, farmers, policymakers, and society is crucial to design adaptive, socially accepted livestock systems that align with the European Green Deal target of carbon neutrality by 2050, ensuring food security, quality, affordability, and sustainable livelihoods.

Keywords: agroecology, ecosystem services, grasslands, monogastrics, ruminants, sustainability

Agroecological framing and system context

We frame our analysis within the FAO's 10 Elements of Agroecology, which articulate ecological functions (recycling, resilience, etc.) together with social and institutional dimensions (responsible governance, circular and solidarity economy, etc.). This framework guides our assessment of livestock sustainability in Switzerland (FAO, 2018; Barrios *et al.*, 2020).

Animal production systems are key contributors to both local and global food security. Yet, their long-term sustainability is contested. Current debates highlight their detrimental environmental impacts, economic performance, and social legitimacy. In Switzerland, livestock farming is a cornerstone of food provision, particularly regarding farm viability and generational renewal. The number of farms has been steadily declining for decades, with around 800 farms disappearing each year (Swissinfo, 2023; BFS, 2024). Between 2000 and 2024, Switzerland lost nearly a quarter of its farms, a trend particularly visible in the dairy and mountain farming sectors (Hediger *et al.*, 2023). Structural change is further accelerated by demographic factors: within the next five years, about 7000 farmers will reach retirement age (Flury *et al.*, 2025). Studies also show that highly specialized dairy farms face

a higher risk of leaving the sector compared to diversified or organic farms (Zimmermann *et al.*, 2022), and that farms without animals tend to achieve higher returns on assets than livestock farms (Zimmermann *et al.*, 2025). Despite subsidies, farming, especially animal husbandry, is perceived as hard work, bureaucratically demanding, and less attractive for the younger generation, which questions its long-term socio-economic viability. According to Pinsard and Accatino (2023), the integration of grasslands and locally available resources emerge as a key strategy to strengthen the resilience of livestock systems, maximize resource use efficiency, improve nutrient cycling, and support biodiversity.

On arable land, the inclusion of temporary grass–legume leys within crop rotations offers multiple benefits. According to Malisch *et al.* (2024), in livestock and/or mixed systems, these leys provide high-quality forage, increase protein self-sufficiency and enhance resilience through diverse botanical compositions adapted to site conditions. Legume-grass leys improve the yields of subsequent crops (Fox *et al.*, 2020), support habitats for pollinators (Table 1), but could also supply biomass for renewable energy production such as biogas. Across all cropping systems, multi-species mixtures contribute to soil carbon sequestration, improve soil structure, substitute for mineral nitrogen inputs (Table 1) (Ten Berge *et al.*, 2016), and suppress weeds (Sebastià *et al.*, 2025), thereby reducing the need for chemical inputs (Lechenet *et al.*, 2016).

Permanent grasslands on non-arable lands represent a striking example of multifunctionality, providing forage for livestock, while simultaneously contributing to biodiversity conservation, and the maintenance of cultural landscapes. Practices such as adaptive pastoral management, including the adjustment of grazing intensity and livestock breed selection, directly influence plant diversity, forage quality, yield stability, and the conservation of rare habitats (Table 1) (Pauler *et al.*, 2025; Probo *et al.*, 2025). According to Mahieu *et al.* (2021), the integration of woody forage species offers additional opportunities to diversify animal diets, enhance animal welfare, and promote the circularity of agroecosystems, supporting more circular, resource-efficient farming systems and reducing dependence on external inputs by enhancing on-farm nutrient recycling. A certain level of functional plant diversity promotes provisioning services and high plant diversity underpins the regulatory and cultural services of grassland ecosystems, while also contributing to improved soil health (Bengtsson *et al.*, 2019).

The systems described above are well suited to ruminants; however, the context for the monogastrics production system is more complex.

Swine and poultry production systems, which typically rely on imported feed supplements, can reduce pressure on arable land, limit deforestation, and lower the overall environmental footprint, by increasing the use of local resources, agricultural by-products, and former food products, combined with agroecological practices (Mottet *et al.*, 2017).

For monogastric and ruminant, future strategies must integrate productive efficiency, recycling, animal health and welfare, adaptation to climate change, and the multifunctionality of grassland systems. Such integrative approaches might facilitate the development of animal production systems with improved sustainability, being able to remain competitive and to meet food security goals. The objective of this synthesis paper is to highlight key findings from recent research on the integration of grasslands and local resources in livestock systems, and to identify the main forthcoming challenges and opportunities.

Table 1. Some examples of quantitative evidences for key ecosystem services in a few selected studies as influenced by specific interventions

Service/outcome	Indicator	System comparison/intervention	Reported quantitative range	Key conditions/trade-offs	References
Forage yield	DM yield ($\text{t ha}^{-1} \text{ year}^{-1}$)	Perennial ryegrass/white clover vs multispecies mixtures	+18% (from +2 to +64%)	Grass-legume-herbs mixtures, modelled across 26 sites (range: min, max among the 26 sites)	O'Malley <i>et al.</i> (2026)
Forage quality	Metabolizable energy content (MJ (kg DM)^{-1})	Grass-legume swards vs grass-only	is maintained despite larger yields	Similar across a large range of legume proportions	Suter <i>et al.</i> (2021)
N efficiency	kg N yield per applied kg fertilizer N	Grass-legume swards vs grass only	2.2 to 3.4 times higher	Depending on N fert applied (here 57 or $150 \text{ kg N ha}^{-1} \text{ year}^{-1}$)	Nyfelner <i>et al.</i> (2024)
N input into the system	N from symbiotic fixation ($\text{kg N ha}^{-1} \text{ year}^{-1}$)	Grass-legume swards vs grass only	+185 to +300 kg	Depends on yield, legume proportion and soil N supply	Oberson <i>et al.</i> (2024); Nyfelner <i>et al.</i> (2024)
Nitrogen leaching	NO_3 concentration in soil solution	Grass-legume at 50 kg Nfert vs Grass at 150 kg Nfert	$\leq \text{NO}_3$ conc. in soil solution	During winters and during the grassland ley phase	Nyfelner <i>et al.</i> (2024)
N_2O emissions	N_2O fluxes ($\text{g ha}^{-1} \text{ day}^{-1}$)	Multispecies mixtures with no N fertilizer vs grass only with N fertilizer	2.9 to 4.6 fold reduction	No trade-off with biomass yield	Hargreaves and Rees (2025)
Soil organic carbon	SOC gain (t C ha^{-1} in 0–50 cm soil depth)	Ley introduction/proportion in the crop rotation	+40 kg C ha^{-1} per 10% more leys	Crop rotations established for 3 decades	Guillaume <i>et al.</i> (2022)
Greenhouse gases	Enteric CH_4 intensity	Higher forage quality through decreased grass maturity	–7 to –18%	Mechanism: higher digestibility; depends on diet composition	Arndt <i>et al.</i> (2022)
Biodiversity	Plant species richness	Reduced fertilizer application/extensification	+20 to +170%	Reduces yield; requires site-specific targets	Probo <i>et al.</i> (2025)
Resistance to weeds	Weed biomass ($\text{t ha}^{-1} \text{ year}^{-1}$)	grass-legume mixtures vs monocultures average	–52% (from –30% to –75%)	Weeds in grass-legume mixtures, modelled across 31 sites and 3 years (range: min, max among the 31 sites)	Connolly <i>et al.</i> (2018)
Resistance to drought	DM yield ($\text{t ha}^{-1} \text{ year}^{-1}$)	4-species grass-legume mixtures vs monocultures average	Ø Monocultures rainfed: $10.5 \text{ t ha}^{-1} \text{ year}^{-1}$ (from 7 to 16 t) Ø Mixtures drought: $12.5 \text{ t ha}^{-1} \text{ year}^{-1}$ (from 10 to 15 t)	Key message: mixtures under drought performed at least as good as the average of monocultures under rainfed conditions	Finn <i>et al.</i> (2018)
Fodder tree biomass	Leaf forage yield ($\text{t DM ha}^{-1} \text{ year}^{-1}$)	Willow/maple hedgerows	0.5 to 13.0	Depends on tree density and cutting regime; seasonal availability	Galland <i>et al.</i> (2025)

Sustainability in ruminant production systems in non-arable lands: ecosystem services and management challenge

Permanent grasslands account for about 34% of the European Union's utilized agricultural area, and 70% of Switzerland's total agricultural land (Bengtsson *et al.*, 2019). In addition to providing forage, they deliver essential regulatory services, such as erosion control, and water regulation, as well as

cultural services, such as aesthetic value and heritage preservation (Richter *et al.*, 2024). Plant functional diversity supports high grassland yields (O'Malley *et al.*, 2026): The inclusion of legumes allows symbiotic nitrogen fixation, enhances productivity, and supports soil health by reducing reliance on chemical fertilizers. High plant species diversity, on the other hand, is a key driver of grassland non-production multifunctionality (Klaus *et al.*, 2024). Management strongly shapes these outcomes: intensive practices increase forage yields but reduce biodiversity and associated regulatory services, like carbon sequestration, nutrient retention and water regulation and cultural services (Table 1) (Neyeaert *et al.*, 2021; Richter *et al.*, 2024), whereas extensive management favours biodiversity and cultural services but limits productivity (Richter *et al.*, 2024). Such trade-offs call for differentiated grassland management at farm and landscape scales that balance ecological requirements with societal expectation and economic resilience. Multifunctionality assessment tools, such as indicators of species richness, provide valuable guidance for decision-making (Klaus *et al.*, 2024, 2025; Tindale *et al.*, 2023). Grazing management strongly influences pasture composition: moderate grazing pressure increases small-scale heterogeneity and promotes plant diversity, while both under and over grazing reduce it by favouring dominance of a few woody or spiny/toxic species (Probo *et al.*, 2025). Moreover, the increased use of high-productive modern cattle breeds has amplified the negative effects of uneven pasture use, particularly in steep mountain areas (Pauler *et al.*, 2020). Therefore, livestock systems based on non-arable land, such as mountain grasslands, support sustainable animal production by making productive use of these areas for dairy and meat products (Meyer *et al.*, 2025). Since December 2023, the Swiss Alpine season ("Schweizer Alpsaison") has been recognized by UNESCO as Intangible Cultural Heritage (Meyer *et al.*, 2025). However, these systems face growing challenges from climate change, including more frequent droughts and variable forage production, as well as labour shortages and the rising presence of large predators, which complicate the balance between productivity and ecosystem services (Meyer *et al.*, 2025). Adaptation strategies often involve replacing dairy cows with suckler cows or, on dairy farms, increasing cheese production (Meyer *et al.*, 2025). The integration of local woody species, such as goat willow (*Salix caprea* L.) or sycamore maple (*Acer pseudoplatanus* L.), into silvopastoral systems or forage hedgerows provides supplementary resources, especially during droughts (Table 1) (Enri *et al.*, 2020; Galland *et al.*, 2025). Fodder trees offer high-quality feed, enhance dietary diversity, reduce emissions, and support circular use of on-farm resources (Mahieu *et al.*, 2021). Yet they present trade-offs: while many species supply protein, digestibility, and minerals, excessive lignin or tannins can reduce intake, requiring careful dietary management (Enri *et al.*, 2020). The most suitable species, such as goat willow and sycamore maple, combine moderate tannin content, good digestibility, and balanced mineral profiles. Strengthening these practices through targeted policies and adaptive management is key to ensuring long-term resilience and multifunctionality of grassland-based systems under changing climatic, economic, and social conditions. These systems also enable production on land unsuitable for human food crops, thus reducing competition with human nutrition (Muscat *et al.*, 2020; Van Zanten *et al.*, 2018). Finally, the spatial mosaic resulting from different management intensities allows multiple functions to coexist, even though optimizing all of them simultaneously remains impossible (Dumont *et al.*, 2022).

Livestock systems in arable lands: integration of temporary grasslands in crop rotations

Based on Cooledge *et al.* (2022), modern agricultural systems, often characterized by simplified rotations and continuous cropping, have led to progressive soil degradation, biodiversity loss, and heavy reliance on mineral inputs. Reintroducing leys into rotations mitigates these effects by improving soil

structure, increasing soil microbial biomass, and enhancing carbon sequestration (Guillaume *et al.*, 2022; Hoeffner *et al.*, 2021). These benefits create legacy effects for subsequent crops, with higher nitrogen availability, improved yields, and longer-term enrichment of soil organic matter. Grassland leys also suppress weeds (Connolly *et al.*, 2018; Suter *et al.*, 2017). By replacing part of mineral N with biological N fixation and improving overall N use efficiency, leys with legumes reduce greenhouse gas emissions linked to fertilizer production and fertilizer-driven N₂O emissions in the field, and enhance the sustainability and productivity of crop rotations (Cummins *et al.*, 2021; Fox *et al.*, 2020; Grange *et al.*, 2022; Zhou *et al.*, 2019). However, these benefits come with a land-use trade-off, as allocating part of the arable area to grassland leys reduces the area available for cash crops and can therefore decrease total crop output at the system level. The discussion should therefore focus on what is the optimal amount of leys in the rotation required to maintain the services they provide, with the least possible loss of total crop output. Knowledge on this optimum is still very patchy.

Integrating crops and livestock further optimizes nutrient cycling, lowers emissions compared to livestock-only systems, and fosters circularity (Loges *et al.*, 2020; Malisch *et al.*, 2024). However, effective management is needed to minimize nitrogen leaching, particularly by adjusting intervention timing and selecting appropriate species and regimes (Cooledge *et al.*, 2022; Nyfeler *et al.*, 2024).

Within these strategies, multi-species mixtures have emerged as a key tool to improve both productivity and environmental performance (Malisch *et al.*, 2024). By combining grasses, legumes, and non-leguminous herbs, they promote overyielding and even transgressive overyielding of biomass through complementarity and synergy (Finn *et al.*, 2013; Nyfeler *et al.*, 2009; O'Malley *et al.*, 2026). They also increase yield stability under climatic variability, particularly during droughts, thanks to diverse root systems and growth strategies (Husse *et al.*, 2017; Haughey *et al.*, 2018; Schaub *et al.*, 2020; reviewed in Lüscher *et al.*, 2022). Used as leys, these mixtures enhance soil aggregation and stability (de Haas *et al.*, 2019; Guest *et al.*, 2022), reduce compaction, and improve porosity in long-term rotations (Jarvis *et al.*, 2017), with positive effects extending to subsequent crops and contributing to erosion and flood control.

The Swiss Standard Mixtures developed by Agroscope exemplify this approach: refined over 70 years, they provide regionally adapted combinations designed to meet climatic challenges and optimize resource use (Lüscher *et al.*, 2025).

Nevertheless, their management requires substantial expertise, particularly to maintain legume proportions and persistence, because legumes are highly responsive to management-driven competition with grasses. Maintaining a stable legume fraction depends on controlling fertilizer N inputs (which favour grasses), applying balanced P and K fertilization and ensuring adequate soil pH, and matching cutting or grazing frequency to the growth dynamics of legumes. In addition, legumes often show reduced persistence under waterlogging, or during periods of high disease and pest pressure, which can lead to rapid declines in their contribution. As a result, farmers need to occasionally adjust management, to prevent legume loss and maintain mixture functionality over time. Nevertheless, the 70 years of practical experience with these mixtures has led to mixture compositions with which grave declines of legume proportion are rare during the anticipated duration of utilization of the leys (Lüscher *et al.*, 2025).

Grassland-based diets and alternatives to feed supplements in ruminant systems

Ruminant livestock systems based on grasslands make efficient use of local resources while minimizing competition with the human food supply (Muscat *et al.*, 2020; Van Zanten *et al.*, 2018). In Switzerland, the Grassland-based Milk and Meat (GMM) program, launched in 2014, promotes production adapted to local forage by limiting supplements to 10% of the ration and requiring 75–85% grassland-based forage depending on altitude. Covering around 80% of national grasslands, it provides significant subsidies but has had limited impact on nutrient cycle closure. Its updated version, GMM+, specifically targets nitrogen surplus reduction by capping crude protein in purchased feeds or per unit of milk, thereby improving resource efficiency and strengthening the link between livestock and local feed production.

Optimizing forage diets is central to sustainability. Tannin-rich legumes such as sainfoin (*Onobrychis viciifolia* Scop.) and bird's-foot trefoil (*Lotus corniculatus* L.) can reduce urinary nitrogen losses, typically on the order of approx. 10–30%, and methane emissions, often approx. 5–20%, depending on inclusion level and tannin concentration (Grosse Brinkhaus *et al.*, 2016; Arndt *et al.* 2022, Lazzari *et al.*, 2023 a,b). However, potential negative effects on digestibility may occur when tannin levels are high, requiring careful dietary management (Mueller-Harvey *et al.*, 2019). Beyond forage choice, dietary adjustments such as lowering crude protein content, reducing the dietary proportion of cereal-based concentrates, or partly replacing cereals with former food products can further mitigate the environmental footprint of milk and meat production, for example by decreasing nitrogen emissions (Schrade *et al.*, 2023) and supporting overall sustainability (Reiche *et al.*, 2025).

However, maximizing grassland-based forage proportions must account for animal welfare and performance (Falk *et al.*, 2018). Not all dairy cow types or genetics are suited to forage-based milk production, underlining the need for locally adapted systems (Heublein *et al.*, 2017; Schori and Munger, 2021). Breeds and cow types with strong functional traits (e.g. health, fertility) and moderate production potential, such as dual-purpose Fleckvieh/Simmental, Original Brown Swiss, Montbelarde, Jersey, and well-designed crossbreds, are generally better suited to forage-based systems than high-yielding Holstein dairy cows. Grazing-based milk production systems are also strongly shaped by climate: increasing heat stress, now evident even in temperate regions (Pontiggia *et al.*, 2023, 2024; Holinger *et al.*, 2024), requires mitigation. Measures such as providing indoor housing during the hottest hours (Pontiggia *et al.*, 2025) or using silvopastoral systems for natural shade have proven effective, simultaneously supporting animal welfare and enhancing biodiversity.

Integration of local resources and implementing agroecology in monogastric systems

While these scenarios align well for ruminants, the question remains: what about monogastrics? Monogastric livestock systems have long relied on soybean-based protein feeds, which impose significant environmental burdens through deforestation and disruption of global nutrient cycles (Kasper, 2024; Mottet *et al.*, 2017). In response, recent advances in animal nutrition emphasize the use of local resources, agro-industrial co-products, and former food products as alternatives to conventional feed supplements. Depending on regional availability, integrating local protein sources (e.g., rapeseed meal, pulses) together with co-products and former food products can support pig production with markedly lower soybean meal inclusion, although complete substitution is rarely possible without alternative high-quality protein supplies and precise amino-acid balancing.

This shift both reduces competition between animal and human food supply chains and strengthens territorial food sovereignty (de Quelen *et al.*, 2024; Shurson *et al.*, 2025).

At the same time, precision feeding and amino acid supplementation tailored to these diets optimize nutrient use, limiting nitrogen and phosphorus losses (Mosnier *et al.*, 2011). Rooted in an agroecological framework, this transition also enriches production environments: providing diverse forages for free-range or organic poultry enhances biodiversity, improves welfare, and reduces reliance on synthetic inputs (Dumont and Bernués, 2014). With its roots in an agroecological framework, this transition enriches production environments and is becoming increasingly relevant for free-range and organic poultry production, including egg production, where outdoor access and diverse outdoor areas are key features. Providing diverse forage and managed outdoor areas can enhance on-farm biodiversity, provide behavioural stimuli and reduce reliance on synthetic inputs (Dumont and Bernués, 2014). However, outdoor access can also increase exposure to hazards associated with wildlife and the environment (e.g. parasites and pathogens), so appropriate biosecurity and range management are required to limit health and food safety risks. This includes managing the interface with wild birds, which is recognised as an important pathway for the introduction of avian influenza.

These low-input systems, despite their environmental benefits, often result in lower productivity, which can lead to higher emissions per kilogram of product due to reduced feed conversion efficiency and slower growth (Van Wagenberg *et al.*, 2017). To address this, the pig sector is testing nutritional and genetic strategies such as optimizing feed efficiency, improving manure management, using low-protein diets, and selecting for nitrogen use efficiency (Kyeariakis *et al.*, 2011; Lautrou *et al.*, 2022; Rauw *et al.*, 2023). According to Kasper (2024), however, the success of these measures depends on carefully managing sanitary, social, and economic trade-offs to avoid adverse side effects from intensification.

Conclusion

A cross-cutting analysis of animal production systems suggests that their long-term sustainability depends on integrating grassland valorisation, efficient use of local resources, and agroecological principles. Permanent grasslands contribute to ecosystem multifunctionality, temporary ones restore soil fertility in crop rotations, with multi-species mixtures enhancing system resilience. Together, these elements form key levers for strengthening both environmental robustness and economic viability of livestock farming.

The transition toward circular systems, precision feeding, and greater use of local co-products offers a promising pathway, provided trade-offs between productivity, animal welfare, and environmental performance are carefully managed. Climate change, particularly intensifying heat stress and drought, further reinforces the need for innovative adaptation strategies in forage production, grazing management, and dietary formulation. Sustainability cannot be achieved through technical innovations alone. It requires the coordinated involvement of scientists, farmers, policymakers, and society at large to design, test, and implement adaptive governance models that address local realities while engaging with global food security and environmental challenges. Beyond technical solutions, social organization and governance are enabling conditions for scaling grassland-based innovations. Labor availability, collective action for pasture and alpine grazing management, coordination between farms and advisory services, and policy instruments rewarding ecosystem services strongly influence adoption, persistence

and impact. Embedding technical options into territorial strategies (e.g., support to service herds grazing in a targeted manner across different areas to prevent shrub encroachment where farm numbers decline) can therefore deliver simultaneous environmental and socio-economic benefits.

Looking ahead, priority research should embed grassland multifunctionality and resource circularity into policy instruments, create incentives for the lasting adoption of agroforestry and other agroecological practices, and examine governance and equity conditions in the transition. Building such a science–policy–society dialogue will be essential for the sustainable transformation of livestock production.

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Protein efficiency and grassland performance on Northwest European dairy farms

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Abstract

Ruminants can convert grassland biomass into valuable human-edible protein and other nutrients through milk and meat. In this context, grassland-based dairy systems can play a key role in improving protein self-sufficiency. The European North Sea region as an important area of permanent grassland and intensive dairy farming offers considerable potential for sustainable protein production. This study evaluates the protein conversion efficiency of grassland-based dairy farms. A total of 49 dairy farms across five countries in Northwest Europe (Belgium, France, Germany, The Netherlands, and Sweden) were investigated. Data were collected via interviews with farm managers covering farm structure, grassland management, animal performance, and feed composition. From this, human-edible protein conversion ratios (hePCR) were calculated. Biomass samples were collected and botanical assessments of up to five grasslands per farm conducted to evaluate forage nutritive value and plant species diversity on the farm-level. Farms were categorized based on their hePCR (<1.0 for low, >1.0 for high) and relationships derived with environmental, biodiversity and farm-performance data. The study highlights that there is no trade-off between net protein production from dairy milk and dairy farm performance measures.

Keywords: dairy farming, feed, food competition, protein conversion efficiency

Introduction

Ruminants are able to transform grassland biomass into high-value, human-edible protein in the form of milk and meat (Wilkinson, 2011). Consequently, grassland-based dairy systems have the potential to play a key role in improving protein self-sufficiency. A key advantage of such systems is their capacity to reduce reliance on food-competing feed by making use of resources that cannot be consumed directly by humans, such as grassland biomass. In this context, the food efficiency of livestock systems and the broader issue of feed–food competition have gained increasing attention (Barbieri *et al.*, 2022). One promising indicator for evaluating this efficiency is the human-edible protein conversion ratio (hePCR), which quantifies the balance between human-edible protein input and output within the system (Wild *et al.*, 2025). The European dairy sector accounts for approximately one-fifth of global milk production (Eurostat, 2025), with the North Sea region distinguished by large proportions of permanent grasslands and highly specialized dairy farming systems (Huyghe *et al.*, 2014). Despite this high potential for sustainable protein production, little is known about the protein efficiency at farm level in this region.

Materials and methods

A total of 49 dairy farms located in five Northwestern European countries (Belgium, France, Germany, The Netherlands, and Sweden) were included in this study. These farms are characterized by high milk production intensity, with milk contributing, on average, $84.2 \pm 14.0\%$ to total farm income. The average herd size was 119 ± 77 dairy cows, and 43 of the farms kept Holstein–Friesian breeds. The study consisted of two components: First, a structured interview with the farm manager, which covered topics including farm structure, grassland management practices, animal performance and feed rations. Second, a field-based grassland survey on up to five grassland fields per farm. In one or two 2×2 m subplots per field, the number of different plant species was counted. This species richness (SR) was then averaged over all investigated fields per farm. Species richness therefore is given per 4 m^2 . Hand-plucked biomass samples were collected from these subplots by cutting the upper third of the vegetation from ten hand-grabs, which together formed a composite sample. These samples were dried at 60°C for 48 hours and ground to 1 mm. Crude protein (CP), acid detergent fibre (ADF), crude ash (CA) and crude fat (CL) and the enzyme-soluble organic matter (ESOM) were analysed by Near-Infrared Reflectance Spectroscopy (Phoenix 5000, Bluesun Sci, USA), with each sample scanned twice. Metabolisable energy (ME, $\text{MJ kg}^{-1} \text{ DM}$) was calculated as follows: $\text{ME} = 5.51 + 0.00828 \times \text{ESOM} - 0.00511 \times \text{CA} + 0.02507 \times \text{CL} - 0.00392 \times \text{ADF}$. The coefficient of variation (CV) of ME served as indicator for production stability per farm. Annual feed qualities offered to dairy cows (kg DM year^{-1}) during lactation (305 days) and the dry period (60 days) were multiplied by their crude protein (g (kg DM)^{-1}) and the corresponding human-edible protein fraction. CP values came from feed analyses and standard feed tables (LfL, 2021). The human-edible protein conversion ratio (hePCR) was calculated per farm using feed ration data according to the following formula: $\text{hePCR} = \sum_n (\text{kg CP feedstuff}_i \times \text{human-edible proportion of the CP from feedstuff}_i) / (\text{kg CP milk} \times \text{human-edible proportion of the CP from milk})$, where n is the number of feedstuffs used per farm per year, CP is the crude protein concentration, and i represents each feed type, following the methodology of Wild *et al.* (2025). Farms were categorised based on hePCR into two groups (low, high), with thresholds of <1.0 considered net protein production ($n = 22$) and >1.0 as net protein loss ($n = 27$). To characterise milk production performance, the total amount of energy corrected milk (ECM) per cow and year was calculated, along with the amount of ECM per cow and year produced from grass using a residual method. ECM per hectare of grassland was then determined. Also, the proportion of milk produced from concentrates, maize silage, and arable land (including all fed crops growing on arable land, except ley-grass) have been calculated accounting for energy requirements of maintenance and performance similar to Wild *et al.* (2025). An analysis of variance (ANOVA) was conducted with hePCR category (low vs. high) as the influencing variable on milk production performance, concentrate feed, SR and CV ME. Post hoc comparisons of means using t-tests were then applied to compare group means.

Results and discussion

ANOVA showed that higher inputs of human-edible concentrate feed were linked to increased total energy-corrected milk (ECM) yields (Table 1). However, the common practice of evaluating dairy system productivity solely based on annual milk yield per cow is increasingly criticised due to concerns related to environmental impact, animal welfare, food security, and the economic dependence of smallholder farmers on the feed industry (Knaus, 2016; Peyraud and Peeters, 2020). Consequently, some studies advocate prioritizing milk production per hectare of grassland—referred to as grassland performance—over per-cow productivity. This approach is typical in pasture-based dairy systems such

Table 1. Estimated means $\pm$ SE of parameters of interest as affected by the hePCR categories (low: <1 ; high: >1).

Parameter	low hePCR (<1)	high hePCR (>1)
ECMtot (kg cow ⁻¹ year ⁻¹)	8982 $\pm$ 519a	9942 $\pm$ 469a
ECMgr (kg cow ⁻¹ year ⁻¹)	4210 $\pm$ 315a	2867 $\pm$ 336b
ECMgr (kg ha ⁻¹ GL)	8655 $\pm$ 1410a	7123 $\pm$ 1323a
Concentrate feed (g kg ⁻¹ ECM)	1289 $\pm$ 19a	230 $\pm$ 17b
Proportion milk from concentrates (%)	23 $\pm$ 4a	39 $\pm$ 3b
Proportion milk from maize (%)	11 $\pm$ 4a	27 $\pm$ 3b
Proportion milk from arable land (%)	39 $\pm$ 4a	68 $\pm$ 4b
Species richness (4 m ²)	7.1 $\pm$ 0.4a	6.9 $\pm$ 0.4b
CV ME	0.03 $\pm$ 0.01a	0.03 $\pm$ 0.01a

Superscript lowercase letters indicate significant differences within a row ($p < 0.05$). hePCR: human-edible protein conversion ratio, ECMtot: total energy-corrected milk per cow, ECMgr: energy-corrected milk from grassland, GL, grassland; ECM, energy corrected milk; CV ME, coefficient of variation of ME.

as those in New Zealand and Ireland (Peyraud and Peeters, 2020; Knaus, 2016). Consistent with this, farms with lower inputs of human-edible concentrate feed in our study exhibited higher milk yields from grass per cow and per hectare (Table 1); even though the latter could not be statistically confirmed, a trend has been observed. In addition, high hePCR farms not only use more concentrates but also show greater share of milk produced from maize and arable land (Table 1). Overall, the biodiversity level was generally rather low, but SR was slightly higher for low than high hePCR farms in the North Sea region. This might be due to overall lower regional biodiversity in coastal lowlands; Wild *et al.* (2025) also reported lower mean Shannon diversity on farms in northern Germany compared to other regions. No significant difference in the CV of ME was observed between low and high hePCR farms (Table 1) which suggests that grassland herbage was stable irrespective of farm type.

Conclusion

Higher hePCR increased total ECM per cow but lowered the proportion of milk derived from grass, thereby intensifying feed-food competition. Farms with low concentrate inputs moreover produce milk from slightly more diverse grasslands. The sustainability of grass-based dairy farms is therefore highly dependent on external feed resources produced on arable land.

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Regulating shrubs and weeds on mountain pastures by site-adapted management

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Abstract

Mountain pastures host diverse plant species. If a single species becomes too dominant, there is an interest to regulate it—especially, if this herb or shrub is poisonous for grazing livestock or if it reduces biodiversity and fodder quality. Knowledge on regulating such weeds and shrubs is often only passed on orally. We gathered knowledge on relevant weeds and shrubs in mountain pastures and made it accessible online. Therefore, we conducted a qualitative survey among experts from science, advisory services, nature conservation and agricultural practice, and reviewed available literature. We summarized specific measures against 24 unwanted plant species in an online manual. Although no one-size-fits-all solution exists, we identified several key principles to regulate these plants: (1) Early regulation requires less effort; dominant plants need more intensive measures. (2) Dense and competitive swards slow the spread of weeds. (3) Selective herbicides are rare; broad-spectrum ones create gaps quickly recolonized by weeds. (4) Most unwanted plants are either promoted by over- or undergrazing. Thus, site-adapted grazing management, with appropriate livestock type, stocking density and timing, is the most efficient way of regulation. (5) Any regulatory intervention only makes sense if the causes (over- or undergrazing) are eliminated through adapted grazing management. (6) Unwanted plants are persistent and persevering, due to traits such as underground storage organs, high regeneration ability, rapid spread, etc. Successful regulation is not a one-off measure but means to be more persistent than the plants to be regulated.

Keywords: knowledge transfer, mountain pastures, overuse, shrub encroachment, site adapted management, underuse, weeds

Introduction

Pastures in mountain and alpine areas have been shaped by centuries of extensive grazing. Their ecological integrity depends on a delicate balance between intensity of use and site potential. However, changes in grazing intensity, livestock type, and land abandonment have lead to the spread of a few species that outcompete most of other plants. From an agronomical point of view, these species are unwanted because they reduce pasture productivity, and many of them are toxic (e.g., *Aconitum* sp., *Colchicum autumnale*, *Pteridium aquilinum*, *Ranunculus* sp., or *Senecio* sp., *Veratrum album*), unpalatable due to low digestibility (e.g., *Deschampsia cespitosa*, *Juncus* sp., *Nardus stricta*), unattractive due to secondary compounds' concentration (e.g., *Rumex* sp., *Gentiana lutea*, *Mentha longifolia*, or *Hypericum perforatum*), or protected by thorns or spikes (e.g., thistles) or by lignification (i.e., all tall and dwarf shrubs). From a biodiversity conservation point of view, some of these species are desirable as long as they occur at low densities. However, if the mentioned weeds and shrubs become dominant, they overgrow less competitive plant species, and biodiversity is reduced (Zehnder *et al.*, 2020). Thus, both farmers and environmentalists are interested in regulating these unwanted species. While herbicides have been used in the past, their application in mountain areas is increasingly restricted due to environmental concerns and regulatory changes. Therefore, sustainable, site-adapted management strategies are urgently needed. For centuries, the knowledge about weed and shrub regulation has been

passed down in mountain farms from generation to generation. However, due to abandonment and changing working conditions in the mountains, this knowledge is gradually being lost. Therefore, we aimed at retrieving this knowledge, compiling it and making it freely available.

Materials and methods

To gather comprehensive knowledge on the regulation of weeds and unwanted shrubs in mountain pastures, we employed a mixed-method approach combining expert consultation and a literature review. First, a structured survey was sent to 130 professionals involved in alpine forage production and pasture management in Switzerland. The target group included practitioners as well as experts from cantonal agricultural services, advisory institutions, agricultural schools, research organizations, ecological consultancies, and parks. The survey asked participants to report on weeds and problematic shrubs they encounter, describe management practices they have applied, and assess their effectiveness. We received 31 responses, covering 24 different unwanted plant species. The responses varied in detail and scope, reflecting the diversity of experiences and site conditions.

To complement the survey data, we conducted a literature review on both scientific publications (English) and technical reports or local guidelines (German, French, Italian) related to weed control in mountain pastures. The collected information was synthesized into general management principles and species-specific profiles. For each species, we present concrete regulation measures directly applicable on mountain farms. These were published in an online manual, which is freely accessible and designed to be updated as new knowledge becomes available (https://www.agroscope.admin.ch/agroscope/de/home/aktuell/newsroom/2023/06-14_Problempflanzen_Alpggebiet.html). The synthesis emphasized practical relevance, ecological sustainability, and adaptability to site-specific conditions.

Results and discussion

In the online manual, we present specific measures for each of the 24 unwanted plants (i.e., mechanical, biological, chemical, and grazing-based methods). Because there is no one-size-fits-all solution, in the following, we focus on broad lines applicable for all the species:

- (a) Early detection and rapid response The most effective control strategy is early intervention. If unwanted plants are detected and removed at an early stage (i.e., as long as they are isolated individuals), the effort required is manageable. Once these species establish and spread, mechanical or manual removal becomes labour-intensive and costly—or can be almost impossible, especially if the seedbank of the original pasture vegetation is already lost. Regular monitoring of pastures is therefore essential.
- (b) Maintaining a dense and competitive sward Many unwanted species benefit from bare soil or gaps in the vegetation. A dense, well-established sward acts as a natural barrier against invasion. Practices that promote dense sward—such as avoiding overgrazing, especially under wet conditions, reseeding bare patches, and maintaining soil fertility—are crucial for long-term prevention.
- (c) Limitations of chemical control Selective herbicides for the common unwanted species in mountain pastures are rare, and broad-spectrum herbicides often do more harm than good by disrupting the plant community. Moreover, their use is increasingly restricted in mountain pastures, especially in protected areas and water source catchment areas. As a result, chemical control is not a sustainable option for most mountain pastures.

(d) Understanding the role of grazing pressure Most unwanted plants fall into one of two categories: (1) plants promoted by overgrazing (e.g., nitrophilous or trampling-tolerant species, such as *Rumex* sp.); (2) plants favoured by undergrazing or abandonment (e.g., *Pteridium aquilinum*, *Nardus stricta*, shrubs or tall forbs). Identifying the underlying cause is essential for choosing the right management strategy.

(e) Site-adapted grazing management The most effective long-term approach is adaptive grazing management tailored to site conditions. This includes: (1) Appropriate livestock type: Light-footed animals such as sheep, goats or low-productive cattle are better suited for steep or sensitive areas (Pauler *et al.*, 2022). Highly productive animals, in turn, generally overuse the few flat and nutrient-rich sites while underusing less attractive areas (Pauler *et al.*, 2020). (2) Appropriate stocking rate: animal numbers and grazing length should be adjusted regularly based on vegetation carrying capacity. Climate change is already causing changes in forage availability—either more forage due to higher temperatures or less due to drought. (3) Appropriate timing: many unwanted plant species are more palatable in spring than in autumn. Moreover, knowing the exact vegetation cycle of problematic plants, allows effective intervention during the phase when they are most sensitive.

(f) Persistence and long-term commitment Many unwanted plants are highly resilient, with traits such as underground storage organs, high regeneration capacity, and prolific seed production. Successful regulation is rarely achieved through a single intervention. Instead, it requires consistent effort over multiple years, often combining mechanical, biological, and grazing-based methods.

Conclusion

There is no universal recipe for regulating weeds and unwanted shrubs in mountain pastures. However, several key principles have emerged: early intervention, maintaining a dense sward, and above all, implementing site-adapted grazing management. Regulatory measures are only effective if the root causes—typically over- or underuse—are addressed. The compiled knowledge from practitioners and experts provides a valuable foundation for sustainable pasture management and biodiversity conservation in alpine regions.

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Modelling the seasonal dynamics of crude protein content and grazing management practices

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Abstract

Grazing of temporary and permanent grasslands is a major lever for enhancing the protein self-sufficiency of livestock systems, leading to significant savings on nitrogen-rich concentrate feeds. The dynamics of crude protein content in the vegetation were modelled by considering both climatic factors and grazing management practices. This analysis was carried out using grazing cycles from three experimental farms: Le Pin-au-Haras (1452 grazing cycles over the past 29 years), Thorigné d'Anjou, and Trévarez. The grazing year was divided into four practical seasons—Early Growth, Peak Growth, Summer Period, and Autumn Regrowth—based on a growth model for perennial ryegrass (*Lolium perenne*). Multiple linear regressions were conducted for each season to develop predictive equations. Model accuracy was assessed using the Mean Absolute Percentage Error (MAPE), and the models were implemented in a prediction tool in Excel. Model performance varied across seasons. The Early Growth model showed the highest accuracy, while the model for the Summer Period was the least effective. Compared to climatic variables, farming practices had a moderate effect on crude protein content. The influences of potential evapotranspiration, temperature, and rainfall on crude protein (CP) content varied significantly depending on the season.

Keywords: proteins, grazing, seasonal dynamics, modelling

Introduction

The dependence of French livestock farms on protein imports raises many issues related to the environment (deforestation in Brazil), ethics (working conditions of farmers in South America), the economy (price volatility) and geopolitics (competition between importing countries) (Dronne, 2018). Global population growth will lead to increased demand for plant and animal proteins. This increase, combined with climate variability, will put further pressure on plant protein availability (Rouillé *et al.*, 2014). A rich and balanced feed, grass is the main source of protein in herbivorous livestock farming systems. It ensures protein production regardless of how it is used, whether grazed or harvested (green fodder, silage, wrapped silage or hay). An effective use of grass, particularly in grazing, is a major lever in strengthening the farm's fodder and protein self-sufficiency (Pierre *et al.*, 2023). A summary was produced as part of the RMT Avenirs Prairies (Future Grasslands) project, drawing on data on grazing from various experimental sites in western France: the INRAE station at Le Pin au Haras, the Thorigné d'Anjou experimental farm and the Trévarez experimental farm. This work had two objectives: (i) to quantify the contribution of grazed grassland to crude protein production (in kg/ha) over the practical seasons of its use; and (ii) to identify the factors influencing the crude protein content of grazed grass in relation to practices associated with land cover use and climatic conditions affecting grass growth.

Materials and methods

The first step (quantitative component) consisted of compiling a database of descriptors associated with 2514 events related to grazing. This compilation covered just under 29 years of grazing history

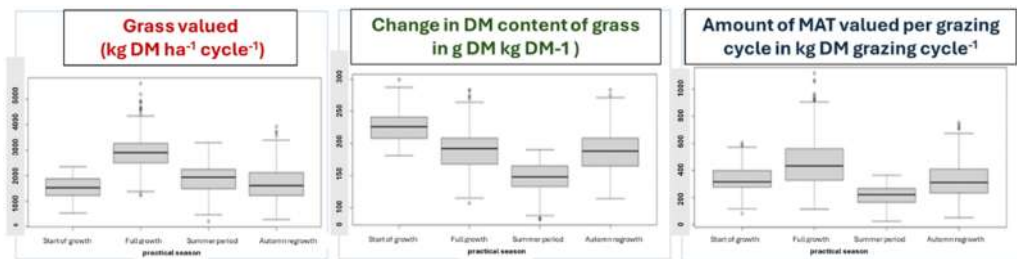


Figure 1. Quantification of grazed grassland over the 4 practical grazing seasons (INRAE du Pin au Haras estate, $n = 1454$ grazing cycles).

at the Domaine du Pin au Haras ($n = 1452$). The rest of the data were based on data from two Casdar projects: Optialibio for the Trévarez farm ($n = 610$) and HerbValo for the Thorigné farm ($n = 160$). The cycles of grassland use for grazing were characterised by various descriptors: dry matter content of grass (g kg DM^{-1}) when the animals entered the pasture, associated vegetation cover height at the start of grazing (in cm, platemeter), height at the end of grazing (in cm, platemeter), cover density (kg DM cm ha^{-1}) and biomass lost (kg DM ha^{-1}) during grazing.

The second stage (simulation) used meteorological data associated with these grazing events, supplemented by various variables relating to practices such as nitrogen fertilisation, regrowth age and residual height after grazing.

Grazing cycles were divided into four main periods defined according to various parameters associated with ryegrass (RGA) growth, using hierarchical ascending classification methods in Rstudio. Multiple linear regressions were performed using Rstudio to model changes in vegetation DM content (g kg DM^{-1}) in relation to practical and environmental factors associated with grazing events. To evaluate the performance of the prediction models, the Mean Absolute Percentage Error (MAPE) was calculated on approximately 1/4 of the cycles removed from the model database. The MAPE expresses the average error of the predictions as a percentage of the actual values, which makes it easy to understand the magnitude of the errors relative to the actual observed values.

The formula used is presented below:

$$\text{MAPE} = \frac{1}{n} \sum_{i=1}^n \left| \frac{y_i - \hat{y}_i}{y_i} \right| \times 100,$$

where n = The total number of grazing cycles, y_i = The actual value of MAT at grazing cycle i , $\hat{y}_i$ = The predicted value of crude protein at grazing cycle i .

Results and discussion

The contribution of grazed grassland to crude protein production was assessed at different sites over the practical seasons of grassland use for grazing. Figure 1 illustrates the results obtained at the INRAE du Pin au Haras site, $n = 1454$ grazing cycles.

For start growth, full growth, summer period and winter regrowth with grazing utilisation levels of about $1.9 \text{ t DM ha}^{-1} \text{ cycle}^{-1}$ and a mean DM content of grazed grass exceeding 180 g DM kg^{-1} DM (median values over the year), grazing contributes to DM production with around $340 \text{ kg of DM ha}^{-1} \text{ cycle}^{-1}$ exported. Based on a herd fed a ration consisting of grazed grass (2.8 t DM LU^{-1}),

PRACTICAL SEASONS	MODELS	Number of variables	Most influential variable	Adjusted R ²	MAPE
Start of growth	$Y = 28,84220 - 0,35643*ETPcumRe - 3,51810*P - ETPmoyRe$	2	P-ETPmoyRe	0,6194	9%
Full growth	$Y = 58,63753 - 0,50723*AgeRe + 0,14909*Nfertcumulée - 3,85264*TmoyRe$	3	TmoyRe	0,5596	13%
Summer period	$Y = 65,744661 - 0,688309*HS.-1 + 0,049672*Nfertcumulée - 6,669675*ETPmoyRe - 4,292115*PluvmoyRe$	4	ETPmoyRe	0,4069	12%
Autumn regrowth	$Y = -30,560240 - 0,836155*HS.-1 + 0,051034*Nfertcumulée + 6,910127*TmoyRe - 0,028921*P - ETPcumul - 17,939230*ETPmoyRe$	5	ETPmoyRe	0,5435	7%

Figure 2. Models of MAT content in grazed grass over 4 practical seasons, and as a function of precipitation (P); ETP Tmoy Nfert, radiation.

maize silage (1.3 t DM LU⁻¹) and concentrated feed ((1,325 t DM LU⁻¹), the contribution of grazed pasture to the protein self-sufficiency of the ration is around 78%.

The change in DM content over the practical seasons was modelled on 442 grazing cycles from the three sites that contributed to this work (Le Pin, Trevarez and Thorigné). Figure 2 shows the regressions obtained for each practical season with their level of significance and the average error associated with the model predictions.

Cumulative mineral fertilisation (Nfertcum) has a positive effect on the MAT content of grazed grass, regardless of the practical season. The age of regrowth and the height at the end of the previous cycle consistently have negative effects. The duration of regrowth and the stage of vegetation reduce the MAT content due to the dilution effect caused by grass growth (Gillet, 1980). ETP during the regrowth phase, whether average or cumulative, always has a negative effect on the DM content of grazed grass. During the summer period, high temperatures during the regrowth phase cause water stress in plants, which also reduces the DM content (Durand, 2007). The difference between rainfall (P) and ETP, whether cumulative during the year or average during the regrowth phase, always has a negative effect on the DM content of grazed grass.

Conclusion

This study quantified the contribution of grazed grassland to DM production over the four practical seasons. The modelling work showed that temperature, ETP and rainfall significantly influence DM variation. Agricultural practices, such as fertilisation and regrowth age, had a less pronounced impact compared to climatic conditions. However, these factors must be qualified, as the multiple linear regression models are not always reliable. The robustness of these models needs validation with grazing cycles from independent sites.

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Assessing the forage contribution of Bird's-foot trefoil in diverse swards under organic management

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Abstract

Forage legumes are crucial for sustainable grassland systems, by enhancing soil fertility by fixing atmospheric nitrogen and reducing fertilizer dependence. Their integration into multi-species swards is vital for both forage productivity and quality in organic farming. This study investigated the effect of different legume species on herbage nutritive value, with a focus on crude protein content throughout the growing season. The experiment involved bird's-foot trefoil, red clover, and alfalfa (lucerne) in various species mixtures, with four harvests per season. Samples from the first three cuts were analysed for crude protein and other quality traits. Legume species significantly impacted crude protein content during the second cut ($P < 0.05$), with values ranging from 148.1 to 179.7 (g/kg). Swards containing bird's-foot trefoil consistently had higher protein levels compared to those with red clover. Although variability was observed between cuts, legume-rich swards maintained both yield and quality. Incorporating underutilized legumes such as bird's-foot trefoil into multi-species swards can enhance forage nutritive value, particularly in organic systems. These findings underscore the potential of diverse legume species to improve protein content and sustainability in forage management.

Keywords: biodiversity, environment, forage, perennial grasses, yield

Introduction

Grasslands are vital agroecosystems in Europe, sustaining ruminant production, supporting biodiversity, and delivering ecosystem services such as nutrient cycling and carbon sequestration (Bengtsson *et al.*, 2019). In organic and low-input systems, where synthetic nitrogen (N) fertilizers are restricted, the role of grasslands is even more crucial for maintaining forage supply and soil fertility (Soussana *et al.*, 2010). Forage legumes play a central role in improving herbage quality through biological N fixation and by increasing CP concentration (Lüscher *et al.*, 2014). Well-established species such as red clover and lucerne (alfalfa) are widely used in European grasslands and are valued for their yield potential and capacity to enhance seasonal forage quality (Eriksen *et al.*, 2014). However, reliance on a narrow group of dominant legumes can reduce system resilience under variable environmental conditions. Less extensively used species, such as bird's-foot trefoil, are gaining attention due to their potential to improve protein utilization and animal performance. Unlike other legumes, bird's-foot trefoil contains condensed tannins that can reduce ruminal protein degradation, enhance amino acid absorption, and mitigate methane emissions (Broderick *et al.*, 2017). Furthermore, its adaptability to marginal soils and persistence under stress conditions make it a promising candidate for diversifying multi-species swards. Against this background, evaluating species-specific effects of legumes on forage protein content can provide practical insights into designing resilient, high-quality swards.

Table 1. Dry matter (DM) yield and crude protein (CP) content

Plant species combination	Annual DM yield (Mg/ha) ($P < 0.05$)	DM yield (Mg/kg)				CP content in the DM (g/kg)		
		Cut I ($P = 0.08$)	Cut II ($P < 0.05$)	Cut III ($P < 0.05$)	Cut IV ($P < 0.05$)	Cut I ($P = 0.65$)	Cut II ($P < 0.05$)	Cut III ($P = 0.25$)
(1) G1+G2/L1	8.96 ^c	3.02 ^{bc}	2.27 ^c	2.08 ^c	1.59 ^d	156.7 ^b	179.7 ^c	183.0 ^{abc}
(2) G1+G2/L2	4.65 ^a	1.92 ^a	1.08 ^a	0.79 ^a	0.86 ^a	145.8 ^{ab}	160.2 ^{abc}	191.7 ^{bc}
(3) G1+G2/L3	6.03 ^{abc}	2.22 ^{abc}	1.85 ^{cde}	1.24 ^{ab}	0.72 ^a	154.7 ^{ab}	159.9 ^{abc}	191.7 ^{bc}
(4) G3+G4+G5/L1	8.33 ^{cde}	3.06 ^c	1.89 ^{cde}	1.80 ^{cde}	1.58 ^{cd}	147.6 ^{ab}	175.1 ^{cde}	191.4 ^{bc}
(5) G3+G4+G5/L2	5.46 ^{ab}	2.06 ^a	1.17 ^{ab}	1.12 ^a	1.12 ^{abcd}	149.0 ^{ab}	151.6 ^{ab}	183.4 ^{abc}
(6) G3+G4+G5/L3	5.75 ^{ab}	2.04 ^a	1.72 ^{bcde}	1.12 ^a	0.87 ^{ab}	142.9 ^{ab}	147.6 ^a	184.7 ^{abc}
(7) G3+G4/L1+L4	7.64 ^{bcde}	2.72 ^{abc}	1.84 ^{cde}	1.71 ^{bcde}	1.38 ^{bcd}	151.2 ^{ab}	167.3 ^{bcde}	194.9 ^c
(8) G3+G4/L2+L4	5.57 ^{ab}	2.25 ^{abc}	1.33 ^{abc}	0.97 ^a	1.03 ^{ab}	139.9 ^{ab}	148.1 ^a	172.9 ^a
(9) G3+G4/L3	6.46 ^{abcde}	2.35 ^{abc}	1.84 ^{cde}	1.25 ^{abc}	1.02 ^{ab}	150.4 ^{ab}	159.9 ^{abc}	191.5 ^{bc}

G1, meadow fescue; G2, perennial ryegrass; G3, tall fescue; G4, *×Festulolium*; G5, timothy; L1, alfalfa; L2, red clover; L3, bird's-foot trefoil; L4, white clover. Different letters indicate significant differences between the treatments ($P < 0.05$)

A better understanding of how underutilized legumes such as bird's-foot trefoil perform compared with traditional species like red clover and alfalfa is crucial for enhancing protein yield and advancing sustainable forage management.

Materials and methods

The field trial was initiated in the summer of 2022 at the Lithuanian Research Centre for Agriculture and Forestry. It was arranged in a randomized complete block design with three replications, with each experimental plot covering an area of 8.3 m². The soil at the site was classified as sandy loam according to its texture, containing 58.4% sand, 28.2% silt, and 13.4% clay. Prior to sowing, the agrochemical properties of the topsoil (0–20 cm) were assessed. The analyses indicated a soil organic matter content of 2.10%, total nitrogen of 0.15%, available phosphorus (P₂O₅) of 129 mg kg⁻¹, and available potassium (K₂O) of 100 mg kg⁻¹. The mean soil pH was neutral at 7.2. The experimental treatments were based on different legume–grass mixtures, in which the dominant legumes accounted for 60% of the sown proportion. This design created variation in species richness, ranging from three to four species per mixture. (Table 1). In the mixtures, grasses were included either as meadow fescue cv. 'Alanta' with perennial ryegrass cv. 'Elena DS' or as a combination of tall fescue cv. 'Monas', *×Festulolium* cv. 'Punia DS', and timothy cv. 'Žolis', each group contributing 40% of the sown composition. These grass bases were then paired with a fixed 60% proportion of one of the selected legumes: alfalfa cv. 'Malvina', red clover cv. 'Arimaičiai', or bird's-foot trefoil cv. 'Gelsvis'. In two treatments alfalfa and red clover were combined with white clover cv. 'Nemuniai'. The swards were managed without mineral nitrogen fertilisation, synthetic pesticides or herbicides. Soil fertility and sward productivity were maintained through biological nitrogen fixation by legumes, consistent with organic management practices. During the establishment year, two cleaning cuts were carried out to suppress weed growth. Forage quality assessments based on chemical analyses were conducted in 2024, as the second year of use provides more representative data on established swards. The chemical composition of forage samples was determined using near-infrared reflectance spectroscopy (NIRS). For data evaluation, a one-way analysis of variance (ANOVA) was applied to assess treatment effects. Differences were considered statistically significant at $p < 0.05$, and all analyses were performed using Statistica 8 software.

Results and discussion

The annual dry matter yield of the grass–legume mixtures varied significantly among treatments ($P < 0.05$), ranging from 4.65 to 8.96 (Mg/ha) (Table 1). Although it is commonly observed that the first cut contributes the largest share to the annual herbage yield, the swards established under the organic farming system exhibited a more balanced yield distribution throughout the growing season, accounting on average for 36.8%, 25.4%, 20.5%, and 17.3% of the annual yield in the first, second, third, and fourth cuts, respectively. Crude protein (CP) content showed distinct seasonal variation across the three harvests. No significant differences were detected among mixtures in the first cut, where CP levels ranged from 139.9 to 154.7 (g kg⁻¹). However, pronounced differences emerged in the second cut, with values spanning from 147.6 and 148.1 in mixtures with red clover or bird's-foot trefoil to 179.7.0 (g kg⁻¹) in the alfalfa-based mixture with meadow fescue and perennial ryegrass. The third cut again revealed no significant differences, but CP levels were generally higher, reaching up to 194.9 (g/kg) in the tall fescue and × *Festulolium* mixture with alfalfa and white clover.

Across harvests, mixtures with alfalfa consistently achieved relatively higher crude protein concentrations, particularly in the second and third cuts. Bird's-foot trefoil also contributed to elevated CP content in the later harvests, whereas red clover mixtures tended to show lower or more variable protein levels. White clover inclusion slightly increased protein content in some treatments (up to 194.9% in cut III), though without a clear effect on yield.

Conclusion

Alfalfa-based mixtures provided the most consistent improvements in crude protein content, particularly in the second and third harvests, while bird's-foot trefoil also showed potential to enhance forage quality. Red clover mixtures produced the lowest yields and tended to have lower protein content compared to alfalfa and bird's-foot trefoil.

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Performance of dairy cows fed protein-reduced concentrates in an organic grass-based feeding system

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Abstract

This study investigated the effects of protein supply on milk production in a herbage-based feeding system over three years. A total of 138 lactation records from 94 Holstein and Swiss Fleckvieh cows on an organic farm were assessed. During winter, cows received hay, followed by daytime grazing from late March and full-time grazing thereafter. Per lactation, cows received 333 kg of concentrate (as fed): 3 kg day⁻¹ until 90 days in milk (DIM), then 1 kg day⁻¹ until 150 DIM. Cows were paired by breed, parity, and calving date and randomly assigned to either a cereal-based concentrate (137 g crude protein kg⁻¹ dry matter) or a protein-rich concentrate (221 g crude protein kg⁻¹ dry matter). Calving occurred from late December to early May. Biweekly milk control records were categorised by lactation stages (early, mid, and late) and analysed using a linear mixed-effects model. In early lactation, cows fed cereal-based concentrate produced less energy-corrected milk (26.8 vs. 28.5 kg, $P = 0.01$), had lower milk fat (38.6 vs. 40.0 g kg⁻¹, $P = 0.049$), and urea concentrations (161 vs. 206 mg l⁻¹, $P < 0.001$). Their somatic cell counts also appeared to be slightly higher ($P = 0.02$).

Keywords: grassland, herbage, Holstein, nitrogen, supplementation, Swiss Fleckvieh

Introduction

The Grassland-based Milk and Meat Production incentive was introduced in Switzerland in 2014 to support site-adapted, herbage-based, and concentrate-reduced ruminant production. The original guidelines stipulate that at least 75% in lowland areas and 85% in mountain regions of the annual cattle ration must consist of fresh or preserved herbage, with concentrate use limited to a maximum of 10%. As part of the programme's ongoing development and simplification, there was a proposal for an alternative scenario allowing only concentrate feeds with a crude protein (CP) content below 12% for dairy cows. The minimum required proportions of herbage and the maximum proportion of concentrates in the annual ration would be eliminated. In this scenario, the protein requirements of the ruminants would primarily have to be met through herbage. Reducing the CP content of the concentrate is expected to decrease nitrogen (N) losses, as there is a strong positive relationship between N intake and N excretion (Yan *et al.*, 2006). Protein-reduced supplementation could therefore contribute to achieving Switzerland's N reduction targets—a 15% reduction in N losses by 2030 compared to the 2014–2016 baseline. Worldwide, a United Nations initiative calls for the halving of N losses by 2030. This study was designed to investigate the effects of a reduced protein supply scenario on the production traits of dairy cows under a herbage-based feeding system.

Materials and methods

The experiment included three calving seasons and therefore lasted approximately 3.5 years. The experiment was carried out on the organic farm Ferme-École de Sorens, located approximately 820 m a.s.l. in Sorens, Switzerland. During the winter-feeding period, the forage of the Holstein and Swiss Fleckvieh cows consisted of hay ($n = 18$; mean $\pm$ SD 5.2 $\pm$ 0.15 MJ net energy for lactation (NEL) and

111 ± 10.7 g CP (kg dry matter (DM))⁻¹) ad libitum. The experimental cows calved from the end of December until the beginning of May. From the end of March onwards, the cows were continuously switched to full grazing (fresh herbage (n = 40): 6.1 ± 0.25 MJ NEL and 178 ± 32.2 g CP (kg DM)⁻¹). During the first 90 days in milk (DIM), all cows received 3 kg of concentrate on an as-fed basis. From 100 to 150 DIM, following a 10-day adaptation period, the cows were given 1 kg of concentrate. In the CERE treatment, all cows were supplied with a commercial cereal-based concentrate (n = 9; 7.8 ± 0.10 MJ NEL and 137 ± 5.3 g CP (kg DM)⁻¹). In the PROT treatment, all cows were fed a mixture of two commercial concentrates, resulting in the following average energy and protein content: 8.0 ± 0.10 MJ NEL and 221 ± 6.6 g CP (kg DM)⁻¹ (n = 9). The cows were paired according to breed, parity (2.5 ± 1.55), and calving date (average calving date per year: between 20 and 28 February ± 35 days). Every 14 days, during the official milk recording, the individual daily milk yield was recorded. At the same time, we collected and mixed one milk sample from two consecutive milkings from each cow. The fat, protein, lactose, and urea contents, as well as the somatic cell counts of these milk samples, were analysed. The energy-corrected milk (ECM) yield was calculated based on fat, protein, and lactose content. For the evaluation, the results of the fortnightly official milk recordings were classified into three lactation stages: early lactation, comprising the 1st to 6th recordings; mid lactation, covering the 8th to 14th recordings; and late lactation, including the 15th up to the last available recording for the standard lactation. A linear mixed-effects model (R Core Team, 2025; packages: *lme4* and *lmerTest*) was used for the evaluation, with treatment, lactation stage, and their interaction as fixed effects. The individual cow was included as a random effect. For the evaluation of the somatic cell count data, we used a generalised mixed-effects model (gamma distribution, logarithmic transformation) from the *lme4* package. The Tukey test was used for pairwise comparisons.

Results and discussion

In total 138 lactation records from 94 dairy cows (56% Holstein and 44% Swiss Fleckvieh) were used for the evaluation. All examined traits (milk yield, ECM yield, milk components, milk urea concentration, and somatic cell count) differed significantly between lactation stages ($P < 0.001$) (these differences are not discussed further in this article, as they are not of primary interest). In early lactation, cows in the CERE treatment produced less milk (27.9 vs. 29.3 kg day⁻¹ (least-squares means), ±0.34 (standard error), $P = 0.001$) and less ECM (26.8 vs. 28.5 kg day⁻¹, ±0.33, $P = 0.001$) than those in the PROT group. This result was expected, as the difference in protein intake was greatest during early lactation. Furthermore, cows seem to respond more strongly to additional protein intake during early lactation (Law *et al.*, 2009). Finally, the cows were fed a higher proportion of hay with lower CP content, compared to fresh herbage, during early lactation. The interaction between treatment and stage of lactation was highly significant for milk and ECM yield ($P < 0.001$). Although small amounts of concentrate were still fed during mid lactation, the differences in milk yield and ECM were no longer significant. At late lactation, when dairy cows no longer received concentrate, milk and ECM yields were similar for both treatments. In contrast to the milk protein content (30.9 vs. 30.5 g kg⁻¹, ±0.29, $P = 0.65$) and lactose content (47.8 vs. 48.0 g kg⁻¹, ±0.14, $P = 0.29$), the milk from CERE cows in early lactation had a significantly lower fat content (38.6 vs. 40.0 g kg⁻¹, ±0.48, $P = 0.008$). In the mid- and late-lactation stages, there were no significant differences between the treatments with respect to the aforementioned milk components. Milk urea content was significantly lower in early lactation in CERE cows than in PROT cows (161 vs. 206 mg/l, ±5.8, $P = 0.001$), most likely due to the different protein contents of the concentrates. During mid and late lactation, milk urea levels were, on average, higher (242 to 270 mg/l; $P < 0.001$ for all pairwise comparisons).

than in early lactation and no longer differed between treatments. This was likely the reason for the highly significant interaction between treatment and lactation stage with regard to milk urea content ($P = 0.001$). According to Glatz-Hoppe *et al.* (2020), the optimal milk urea content ranges between 150 and 250 mg l⁻¹ and indicates a dietary CP intake that approximately meets the animal's requirements. Even when the supplement consisted exclusively of a commercial cereal-based concentrate, the lower reference value for milk urea content was achieved, on average. During mid lactation, the upper threshold was exceeded, on average, as is frequently observed under full grazing conditions. Elevated milk urea content can strain the animal's metabolism (Zhao *et al.*, 2025a) and is associated with reduced nitrogen use efficiency as well as increased nitrogen excretion (Zhao *et al.*, 2025b). Interestingly, somatic cell counts appeared to be slightly higher in the CERE cows than in the PROT cows (87 561 vs. 80 342 cells/ml, ± 7025 (back-transformed values); $P = 0.02$). The potential link between protein supply and udder health needs to be confirmed and warrants further investigation, as protein supply may influence the animal's immune competence (Raboisson *et al.*, 2014).

Conclusion

Our findings show that a commercial cereal-based concentrate containing 12% CP in 88% DM (CERE) results in lower milk yields and milk fat content during early lactation. Such a low-protein concentrate may impair early-lactation performance even under herbage-based feeding systems in cows calving in winter or early spring. By contrast, the lower milk urea content in cows receiving the low-protein concentrate, while within the normal range, indicates lower nitrogen losses. The apparently higher somatic cell counts in the low-protein concentrate treatment require confirmation. Future studies should investigate how a reduced protein supply affects the immune competence of dairy cows.

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Optimising autumn grazing management for white clover swards using light interception and agronomic markers

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Abstract

Maximising spring herbage availability is essential for supporting early-lactation dairy cow production in grazing systems. Autumn grazing management plays a critical role in achieving this, but maintaining sufficient white clover (*Trifolium repens* L.; clover) is also vital. This study investigated the impact of autumn closing date and its influence on clover persistence and productivity in perennial ryegrass (*Lolium perenne* L.; PRG)-clover swards. An experiment was established with four autumn closing dates (29 September, 16 October, 1 and 14 November), each with four replicates. The experimental area had a clover content of 18% prior to the start of the experiment in September 2023. Early closure significantly increased herbage dry matter yield (+631 kg DM ha⁻¹), accumulated from autumn 2023 closure until the spring defoliation on 14 February 2024—but reduced clover content. The later-closed sward supported clover persistence, which was associated with greater light availability at the sward base (+41%). A significant negative correlation ($r = -0.16$, $P = 0.005$) was observed between petiole length and light availability. Agronomic traits responded differently to autumn closing date: petiole length of clover decreased with increasing light availability, while stolon length and number of nodes both increased ($P < 0.05$). Further research is needed to refine autumn management strategies that maintain clover while ensuring sufficient grass availability.

Keywords: autumn closing date, clover persistence, grazing management, spring herbage availability

Introduction

Maximising spring herbage availability is vital in spring-calving, pasture-based systems, as it supports the nutritional demands of early lactation dairy cows, and increases animal performance (Walsh *et al.*, 2023). However, the seasonal nature of perennial ryegrass (PRG) and white clover (clover) growth limits forage availability during winter and early spring, placing greater importance on effective autumn grazing management to ensure adequate herbage availability at turnout. The inclusion of clover in swards improves herbage quality, supports increased milk production, and helps maintain herbage yields while reducing reliance on chemical nitrogen (N) fertiliser inputs. To realise these benefits, sufficient levels of clover are required in the sward (Egan *et al.*, 2018). Maintaining clover is challenging, particularly over winter, when clover growth slows and becomes highly susceptible to canopy shading and competition from PRG (Murray *et al.*, 2022). Autumn closing date influences herbage mass and consequently, light availability at the sward base—an essential factor for stolon development and clover survival (Phelan *et al.*, 2014). Despite these challenges, current grazing recommendations typically treat PRG-clover swards the same as PRG-only swards. This study investigates the effects of different autumn closing dates on clover persistence, sward structure, and early spring herbage production. The objective is to inform practical management strategies that support clover recovery and retention over winter.

Materials and methods

The experiment was carried out at the Teagasc, Animal and Grassland Research and Innovation Centre, Moorepark, Fermoy, Ireland, from September 2023 to February 2024. A plot experiment was established with four autumn closing dates: 29 September (Early), 16 October (Mid-Early), 1 November (Mid-Late), and 14 November (Late), each replicated four times. Individual plots measured 1.5×5 m. The experimental area had a clover content of 18% prior to the commencement of the study in September 2023. Defoliation events were carried out by mechanically cutting the entire plot to a height of 4 cm using an Etesia mower (Etesia Hydro 124D; Etesia UK). Plots assigned to each closing treatment had a simulated grazing at their designated autumn closing date and then remained ungrazed until spring. Herbage mass was measured at the spring defoliation on 14 February 2024 by harvesting the entire plot. Fresh weight was recorded, and a 100 g random sub-sample was dried at 90°C for 16 hours to determine dry matter (DM) content. Sward clover content was assessed prior to each defoliation event by collecting random grab samples across each plot. A 70 g sub-sample was then separated into grass and clover fractions and dried at 90°C for 16 hours to determine the clover DM proportion. Within replicates one and three, five stolons per plot were selected and marked using coloured wire at the oldest leaf-bearing node following defoliation on each closing date. A 1-metre stick, divided into five equal segments, was placed in the centre of each plot, and the nearest stolon to each segment was selected and marked. These stolons were measured at the time of closure, and subsequently every three weeks until 4 December (common measurement date; MD), and then every four weeks until spring defoliation (14 February 2024). Stolon length, number of nodes and petiole length were recorded at each MD. Measurements were taken using a ruler and string to follow the natural curvature of each stolon. Light availability at the sward base was measured at each MD using a 1-metre line quantum sensor (LI-COR., Lincoln, NE, USA) connected to a data logger. All data were analysed using PROC MIXED or PROC CORR in SAS 9.4 (SAS Institute, Cary, NC, USA, 2002). For herbage mass and clover content, the statistical unit was the plot ($n = 16$). Stolon traits were analysed using stolon nested within plot as the statistical unit ($n = 40$). Light availability was analysed at the plot level, with repeated measurements across dates. Fixed effects included autumn closing date; repetition was included as a random effect.

Results and discussion

Autumn closing treatment had a significant effect on DM yield (DMY) ($P < 0.001$; Table 1), measured at the spring defoliation on 14 February 2024. The Late closing treatment produced significantly lower DMY compared to the Early closing treatment, 450 and 1081 ± 46.9 kg DM ha⁻¹, respectively. Each additional day of closure increased yield by 13.7 kg DM ha⁻¹, similar to Looney *et al.* (2022) who reported that each additional day the sward remained closed, herbage mass increased by 16 kg DM ha⁻¹ in spring. However, the Late closing treatment had a greater clover content and survival compared to the earlier closing dates. Clover content increased progressively with later closing dates, with average sward clover content; 5.9% (Early), 5.6% (Mid-Early), 10.9% (Mid-Late), and 13.1% (Late) ($P = 0.05$). Light availability at the base of the sward also varied significantly between autumn treatments ($P < 0.05$). Black *et al.* (2009) reported that in PRG–clover swards, clover contributes relatively little to the upper leaf area of the canopy and is shaded by PRG, resulting in reduced light interception. This highlights clover's greater sensitivity to light competition compared with PRG in dense swards. The Mid-Late and Late treatments had the highest percentages of available light (62.0% and 59.0%, respectively), whereas the Mid-Early and Early treatments showed substantially lower light availability (41.0% and 21.5%, respectively). These findings align with the observation that earlier closing dates,

Table 1. Effect of autumn closing date on dry matter yield, clover content, and clover agronomic traits.

	Autumn treatment				SE	P
	Early	Mid-Early	Mid-Late	Late		
DM yield (kg DM ha ⁻¹)	1081	711	461	450	47.0	0.001
Clover content (%)	5.90	5.60	10.90	13.10	2.40	0.05
Stolon length (cm)	5.03	3.48	5.27	7.36	0.92	0.05
Number of nodes	6.15	4.09	4.87	7.77	0.54	0.001
Petiole length (cm)	0.68	0.83	0.75	0.70	0.04	0.05
Light availability (%)	21.50	41.00	62.00	59.00	11.45	0.01

although resulting in greater herbage accumulation (+631 kg DM ha⁻¹), reduced light availability at the sward base. Petiole length also varied significantly among treatments ($P < 0.05$), supporting the morphological responsiveness of clover to changes in light conditions driven by canopy closure. Light availability was significantly negatively correlated with petiole length ($r = -0.16$, $P < 0.01$; Table 1), although this relationship explained only approx. 3% of the variability, indicating that light availability played only a minor role in petiole elongation in this study. Nevertheless, the trend is consistent with shade-avoidance responses previously reported in clover, where reduced light promoted petiole elongation (Solangaarachchi and Harper, 1987). Stolon length increased from 3.48 ± 0.92 cm in the Mid-Early treatment to 7.36 ± 0.92 cm in the Late treatment ($P < 0.05$), while the number of nodes increased significantly with later closing dates ($P < 0.001$). These responses suggest a greater potential for clonal spread and sward persistence in PRG–clover swards with a later autumn closing date (after 1 November). In contrast, Marcuvitz and Turkington (2000) found that increased shading led to longer total stolon lengths due to greater internode elongation, but this was accompanied by a reduction in the number of nodes, indicating a trade-off between elongation and branching under low-light conditions.

Conclusion

Autumn closing date had a clear impact on both spring yield and clover persistence. Early closure increased spring DM yield (+631 kg DM ha⁻¹) but reduced clover content (−7.5%), while later closure reduced yield yet supported clover recovery. Light availability at the base of the sward over winter is a key factor in supporting clover persistence. These findings highlight the importance of autumn grazing strategy for balancing early spring feed availability with short-term clover recovery.

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Fresh grass intake of dairy cows with daily or weekly rotation, performed with electric or virtual fencing

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Abstract

Daily rotation may increase fresh grass intake and grazing efficiency of dairy cows. However, daily moving of physical fences is labour intensive. Virtual fencing (VF) could support farmers, but its effect on dairy cow performance, particularly in terms of fresh grass intake, remains unknown. In this trial we tested differences in (calculated) fresh grass intake of dairy cows between electric fencing (EF) or VF with daily (D) or weekly rotation (W). Sixty-four lactating dairy cows were randomly assigned to four treatments (EFD, EFW, VFD, VFW; blocked on lactation stage and production parameters). Grass intake was calculated based on energy balance or grass height measurements before and after grazing. Differences were observed between the treatments, with VFD and EFW having a higher intake than VFW. This difference was mainly caused by differences in grass availability between pastures—VFD and EFW were located on a pasture with a numerically higher grass height before grazing than VFW and EFD. Fencing method had no effect on grass intake.

Keywords: grass intake, rotational grazing, virtual fences

Introduction

Grazing instead of feeding in the stable may offer potential benefits, including enhanced biodiversity (Charlton and Rutter, 2005), reduced ammonia losses and lower enteric methane production (Koning *et al.*, 2024). However, grazing is considered challenging due to concerns about reduced dry matter intake, low grazing efficiency, and a low grassland use efficiency compared to harvesting and silage making. Daily rotation may be a solution to increase the efficiency, though physically moving fences each day is labour-intensive. Virtual fencing (VF) could support farmers in this matter, but its effect on dairy cow performance, particularly fresh grass intake, remains unclear. In this study, we compared the (calculated) fresh grass intake of dairy cows that grazed with electric fencing (EF) and VF with both daily (D) or weekly rotation schedules (W).

Materials and methods

The trial was conducted at Dairy Campus, Leeuwarden, The Netherlands (53°10'50.7"N, 5°45'25.2"E). Sixty-four lactating dairy cows were randomly allocated to four treatments: electric (EF) or virtual fencing (VF) with daily (D) or weekly (W) rotation (EFD, EFW, VFD, VFW; 16 animals per group, blocked on lactation stage and production parameters). All cows were in second parity or higher, at least 100 days in milk and experienced with grazing, but were naïve to the virtual fencing system.

Cows grazed during the day and were housed in the stable at night, receiving a partly mixed ration (PMR) at the feeding fence, consisting of grass silage, maize silage, soy meal, and beet pulp. Additionally, each cow received 1 kg of concentrates during milking, plus extra concentrates in automated concentrate feeders according to individual milk yield.

The trial commenced on 23 May 2024, with a 4-day adaptation phase during which all cows grazed in a single pasture together using EF. Subsequently, cows assigned to VF treatments were trained for 4 days using Nofence® collars (Batnfjordsøra, Norway). During training, one virtual boundary was added daily, resulting in full virtual enclosure by day 4.

After training, the main trial started, which lasted four weeks. Cows were rotated either daily (D) or weekly (W), with treatments assigned to different pastures: EFD and VFW were on pasture A3, and EFW and VFD on pasture A4. For daily rotating groups, 0.15 ha was offered per group per day (0.075 ha carried over from the previous day and 0.075 ha new), totalling approx. 0.6 ha per week. Weekly rotation groups received 0.5 ha per group per week.

Grass height was weekly measured using an electric plate meter (Jenquip EC-09), by walking a V-shaped (D) or W-shaped (W) pattern, collecting 60 (D) or 90 (W) measurements per plot. Fresh herbage was sampled weekly and was chemically analysed by Eurofins (Wageningen, Netherlands) for dry matter content, protein and in vitro digestibility. Additional chemical composition was determined via NIRS, and feeding value was calculated according to CVB (CVB, 2016).

Daily data collection included body weight, fat- and protein-corrected milk yield, and group feed intake (adjusted for refusals). Fresh grass intake was estimated using pre- and post-grazing grass height, or calculated via the energy balance, as follows:

Grass intake (VEM) = energy requirements—VEM intake concentrates—VEM intake PMR.

Requirements are based on body weight and production parameters (Van Es, 1975; CVB, 2016). Daily grass intake (kg) was then calculated by dividing VEM from grass by its energy value.

Means were calculated per cow for adaptation, training, and each week of the main trial. A repeated measures model was used in SAS (version 9.4; SAS Institute, Cary, NC, USA) to test the effects of treatment, period, and their interaction on feed intake, milk yield and composition, and body weight. Each model included period as a repeated measure with cow as the subject.

Results and discussion

The average dry matter (DM) yield before grazing was 3059 kg DM ha⁻¹, and 3672 kg DM ha⁻¹ after grazing (mean values across all treatments and the entire study period), resulting in an estimated average intake of 4.9 kg DM per animal per day. Prior to grazing, more herbage was available on pasture A4 (EFW and VFD; 3258 kg DM ha⁻¹) compared to pasture A3 (EFD and VFW; 2859 kg DM ha⁻¹). However, post-grazing residuals were also higher on A4 (2901 kg DM ha⁻¹) than on A3 (2444 kg DM ha⁻¹).

Despite these higher residuals, calculated grass intake based on pre- and post-grazing grass height was still greater on A4 (5.4 kg DM animal⁻¹ day⁻¹) than on A3 (4.4 kg DM animal⁻¹ day⁻¹). Differences between daily and weekly rotation were small; although daily rotation appeared to result in more

Table 1. Animal production characteristics during the trial.

	Treatment				<i>P</i> value		
	VFW	VFD	EFW	EFD	Treatment	Period	T × P
Milk (kg day ⁻¹)	33.0	31.9	33.7	34.5	0.50	<0.01	<0.01
Milk fat (%)	4.49	4.62	4.51	4.48	0.74	<0.01	0.16
Milk protein (%)	3.49	3.61	3.53	3.35	0.01	<0.01	0.39
FPCM (kg day ⁻¹)	34.2	34.0	35.6	36.0	0.59	<0.01	0.79
Body weight (kg)	634	620	634	627	0.89	<0.01	<0.01

VFW, virtual fencing, weekly rotation; VFD, virtual fencing, daily rotation; EFW, electric fencing, weekly rotation; EFD, electric fencing, daily rotation; FPCM, fat and energy corrected milk; T × P, treatment × period interaction.

Table 2. DM intake of the animals (kg DM day⁻¹), based on measured intake (concentrates, partly mixed ration (PMR)) and calculated based on the energy balance (requirements—known intake) approx. (grass).

DM intake	Treatment				<i>P</i> value		
	VFW	VFD	EFW	EFD	Treatment	Period	T × P
Concentrates	5.78	5.55	5.88	6.59	0.50	<0.01	0.07
PMR	13.3	11.2	11.3	12.3	—	—	—
Grass	3.8	5.5	5.6	4.3	<0.01	<0.01	<0.01
Total	22.4	22.0	22.7	22.9	0.90	<0.01	0.02

VFW, virtual fencing, weekly rotation; VFD, virtual fencing, daily rotation; EFW, electric fencing, weekly rotation; EFD, electric fencing, daily rotation; FPCM, fat and energy corrected milk; T × P, treatment × period interaction.

uniform grazing, calculated intake based on grass height was lower under daily rotation (3.1 kg DM animal⁻¹ day⁻¹) than under weekly rotation (6.7 kg DM animal⁻¹ day⁻¹). Due to methodological limitations, statistical analysis of these differences was not possible.

To estimate grass intake based on animal energy requirements, production parameters (Table 1) were used. Except for milk protein content, no significant differences in production characteristics were observed between treatments.

Grass intake calculated via energy balance was highest in treatments VFD and EFW (Table 2); both treatments were located on pasture A4, which also had the highest pre-grazing grass height and corresponding higher grass intake (5.4 kg DM animal⁻¹ day⁻¹). These values aligned closely with intake estimates derived from energy balance. Intake for VFW and EFD was estimated at 3.8 and 4.3 kg DM animal⁻¹ day⁻¹, respectively, which also corresponded well with intake based on grass height measurements.

However, due to considerable variation in grass height within and between pastures, intake estimates based on grass height measurements were deemed unreliable. Calculations based on animal energy requirements, along with recorded concentrate and PMR intake, are considered more accurate. The observed differences in grass intake between treatments, particularly the higher intake in VFD and EFW compared to VFW, can primarily be attributed to differences in forage availability. Unfortunately, forage availability varied more than anticipated across pastures, despite efforts to standardise conditions during the trial: both VFD and EFW grazed on pastures with numerically higher pre-grazing grass height than those used for VFW and EFD. The type of fencing system (electric vs virtual) did not influence fresh grass intake.

Conclusion

The method of fencing had no measurable effect on calculated fresh grass intake. Instead, calculated grass intake was primarily determined by the quantity of available herbage.

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Extensive grazing in bird protection areas—does virtual fencing minimize the trade-offs?

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Abstract

Extensive livestock grazing can preserve semi-natural grasslands, which are an essential habitat for many bird species. However, ground-nesting birds are threatened by trampling. Temporary fencing can safeguard bird breeding success, but installing fences is labour-intensive. Virtual fencing (VF) could facilitate bird protection efforts. Virtual fences are defined on a digital map and transmitted to a GPS-collar on the grazing animal. We tested whether VF reduces cattle trampling damage using artificial bird nests. Over 33 days, 19 heifers were grazed on a 19-ha pasture in a national park at the Baltic coast. The pasture comprised four vegetation types, where we placed one small (100 m²) and one large (300 m²) VF-exclosure and equally sized open plots, respectively. We deployed artificial nests and checked them 5 times during the trial (912 nest inspections). Overall, we found 14 and 62 trampled nests in VF and open plots, respectively. Trampling probability was independent of plot size, within-plot nest position and vegetation type. Trampling was nearly 6 times more likely in open than in VF plots. Consequently, VF can effectively reduce bird nest losses caused by cattle. VF could thus optimize the conservation outcomes of extensive grazing systems while minimizing the workload for biodiversity-friendly grazing management.

Keywords: bird protection, cattle, conservation management, smart farming, trampling

Introduction

Extensive grazing with cattle or other livestock species is an established approach to preserve open landscapes, such as semi-natural grasslands, which are an essential habitat for many bird species. However, livestock grazing comes along with a conservation conflict, as nests of ground-nesting birds are threatened by trampling, which can lead to nest failure (Pakanen *et al.*, 2011; Sharps *et al.*, 2017). Consequently, temporary fences are required to foster the breeding success of bird species nesting within pastures. Installing such fences is labour-intensive and challenging in the agricultural practice. The rising technology of virtual fencing (VF) could facilitate such bird protection efforts (Wätzold *et al.*, 2024). Virtual fences are drawn on a digital map via a mobile device and communicated to a GPS-collar on the grazing animal (Umstatter, 2011). When the animal approaches the fence line, the collar emits audio cues, and an electric cue if the animal does not draw back. We tested whether VF is effective in reducing nest trampling damage by grazing cattle in a national park at the Baltic coast.

Materials and methods

The animal experiment was approved by the State Office for Agriculture, Food Safety and Fisheries Mecklenburg-Western Pomerania, ref. number: 7221.3-1-023/24. Mid-September 2024, 19 pure- or crossbred Fleckvieh heifers belonging to a large organic farm operating on the Darß peninsula at the

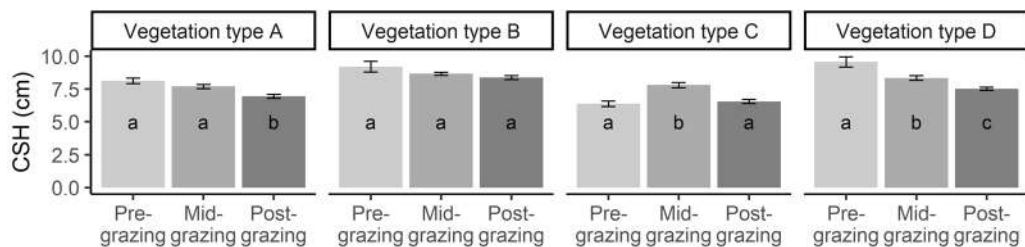


Figure 1. Mean $\pm$ SE of the compressed sward height (CSH, raw data) in the four vegetation types with letters indicating significant ($P < 0.05$) differences between measurement time points within vegetation type after Tukey's HSD P -value adjustment.

German Baltic coast were fitted with virtual fencing collars (Nofence, Batnfjordsøra, Norway). Over 10 days, the animals were habituated to the collars and trained with the technology following Hamidi *et al.* (2022) on a ca. 1-ha section of a 19-ha pasture located in the Western Pomerania Lagoon Area National Park. The heifers were released onto the whole pasture on 27 September 2024 and grazed there until the end of the trial on 30 October 2024.

The pasture was classified based on its general appearance and species composition into four vegetation types: (A) short green grass (mostly *Holcus lanatus*, *Poa pratensis*, *Dactylis glomerata*), (B) *Juncus effusus*-dominated, (C) short dry grass (mostly *Bromus hordeaceus*) and herbs, (D) *Agrostis capillaris*-dominated. These types were distributed in several patches over the pasture. During pre-, mid-, and post-grazing, we measured the compressed sward height (CSH) 150 times per vegetation type with a rising-plate meter (excluding rushes). In each vegetation type, we placed one small (100 m²) and one large (300 m²) virtually fenced plot (exclosures) and equally sized open reference plots. We initially deployed five artificial nests composed of three commercially acquired snail shells (*Helix pomatia*) in the small plots and 10 nests in the large plots. After two weeks, we increased the numbers to nine nests in the small plots and 19 nests in the large plots. Nests were checked for trampling damage (1–3 shells broken) five times during the trial at 7-day intervals (except for the final check, which was done after 4 days) resulting in a total of 912 nest inspections.

We analysed the CSH in a generalised least squares model as a function of the measurement time point (pre-, mid-, and post-grazing) in interaction with the vegetation type, including a variance function structure to account for within-group heteroscedasticity. We analysed the probability of a nest being trampled in a generalised linear mixed effects model with binomial distribution. Plot type (VF or open), plot size, nest position (close to the border or in the plot centre), vegetation type, inspection date and their two-way interactions served as explanatory factors. The random effect accounted for the grouping of nests within plots. The global model was simplified based on likelihood ratio tests and AIC. All analyses were done in R (v. 4.4.1).

Results and discussion

The CSH varied significantly between vegetation types in interaction with measurement time points ($\chi^2 = 44.62$, $df = 6$, $P < 0.001$, Figure 1). In total, we found 14 and 62 destroyed nests in exclosures and reference plots, respectively. The probability of a nest being trampled was independent of plot size, nest position and vegetation type, while the effects of plot type ($\chi^2 = 28.17$, $df = 1$, $P < 0.001$) and inspection date ($\chi^2 = 15.57$, $df = 4$, $P = 0.004$) were highly significant (Figure 2).

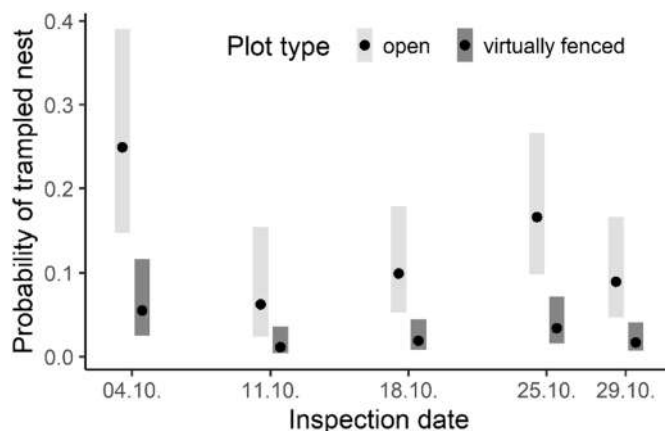


Figure 2. Estimated marginal means ($\pm$ 95% confidence interval) of the probability of an artificial bird nest being trampled by heifers in open and virtually fenced plots during the grazing trial in 2024.

The probability that a nest was trampled in an open plot was 0.12 (SE = 0.03) compared to 0.024 (SE = 0.01) in VF plots (odds ratio 5.68, SE = 1.86, z -ratio = 5.31, $P < 0.001$).

Bird nests have been found to be trampled by cattle in both tall and short vegetation (Pakanen *et al.*, 2011) with the local intensity of cattle activity being an important driver of nest losses (Sharps *et al.*, 2017). Thus, it is not surprising that we did not find a difference in trampling probability between vegetation types despite moderate differences in sward height. The highest trampling probability was recorded at the first inspection date, which is in line with experimental results from Baltic coastal meadows in Finland (Pakanen *et al.*, 2011) and might be related to higher cattle activity and exploration behaviour at the beginning of the grazing period. This highlights the importance of protecting environmental assets or sensitive areas when cattle are released on a new pasture, for example via virtual fencing.

Conclusion

VF effectively reduced bird nest losses caused by cattle trampling under extensive grazing. Similarly, VF could protect occurrences of rare plant species or environmentally sensitive areas. Once economic and legal hurdles have been overcome, VF will be a promising opportunity to optimise the conservation outcomes of livestock systems, without burdening farmers with additional physical work for biodiversity-friendly grazing management.

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Effect of forage conservation on total phenolic concentration and antioxidant activity

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Abstract

Numerous phenolics with antioxidant properties can be found in grassland forage species. Species, vegetative stage and environmental features affect phenolic compounds found in forages. However, effect of forage conservation has not yet been studied. Thus, the objective of this study was to assess the effect of conservation method on total phenolic compounds concentration (TPC, Folin method) and antioxidant activity (AO, DPPH method) of different forages (permanent pasture, sainfoin and cocksfoot). Forages were harvested in May 2023. Biomass from each forage was separated into fresh forage and 4 conserved forages: unwilted and wilted silages, haylage and hay. Total phenolic compounds and AO were significantly higher in fresh sainfoin than in fresh pasture and cocksfoot. Compared to fresh forages, TPC and AO decreased in all wilted forages. For sainfoin and permanent pasture, TPC in unwilted silage was lower compared with fresh forage whereas there was no difference for cocksfoot. For unwilted cocksfoot silage, AO was higher than in fresh cocksfoot whereas for sainfoin and the permanent pasture, there was no difference between fresh forage and unwilted silage. This work provided a valuable report on the reduction in TPC and AO in wilted forages.

Keywords: antioxidants, forage, hay, phenolics, silage

Introduction

Animal health management is a key point for sustainable ruminant production systems. Different secondary metabolites of grassland forage species such as phenolic compounds provide farmers with the opportunity to contribute to the health improvement of their animals with minimal investment. These metabolites have positive animal health effects, especially antioxidant, anti-inflammatory or anti-parasitic effects (Poutaraud *et al.*, 2017). Although phenolic compounds in grassland and forages have recently gained attention, knowledge on their concentrations in forages and factors affecting these concentrations remains limited. Pastures can contain important phenolic concentrations (Maxin *et al.*, 2024). Concentrations in fresh forages would vary with species and stage of growth (Maxin *et al.*, 2020). Environmental conditions may also act on phenolics in plants. Different studies showed that antioxidant activity (AO) in forage was significantly and positively correlated to their phenolic concentration (Amrit *et al.*, 2023; Maxin *et al.*, 2024). To the best of our knowledge, the effects of forage conservation have not been evaluated. Thus, the objective of this study was to assess the effect of the forage conservation method (fresh, unwilted and wilted silages, haylage and hay) on forage total phenolic compounds (TPC) concentration and on AO activity.

Materials and methods

This study was conducted at INRAE Herbipôle research farm at Saint-Genès-Champanelle (45°43'N, 3°1'E; 870 m asl) ([10.15454/1.5572318050509348E12](https://doi.org/10.15454/1.5572318050509348E12)). Three forages were used: permanent grass

Table 1. Effect of the conservation method on total phenolic compounds and antioxidant activity evaluated with DPPH method of three forages.

	Forage conservation methods					SEM	P		
	FF	US	WS	HG	H		F	C	F × C
Dry matter (%)									
Sainfoin	13.8 ^{Cd}	14.9 ^{Bd}	34.1 ^{Ac}	57.3 ^{Ab}	88.0 ^{Aa}	1.08	<0.001	<0.001	<0.001
Pasture	20.8 ^{Ad}	18.8 ^{Ad}	30.1 ^{Bc}	56.9 ^{Ab}	87.9 ^{Aa}				
Cocksfoot	18.3 ^{Bd}	17.4 ^{Bd}	31.1 ^{Ac}	61.8 ^{Ab}	90.2 ^{Aa}				
Total phenolic compounds (mg gallic acid equivalents/g DM)									
Sainfoin	11.8 ^{Aa}	8.6 ^{Ab}	4.2 ^{Ac}	4.9 ^{Ac}	4.3 ^{Ad}	0.21	<0.001	<0.001	<0.001
Pasture	5.1 ^{Ba}	4.0 ^{Cb}	3.8 ^{Bb}	3.8 ^{Bb}	3.1 ^{Bc}				
Cocksfoot	5.0 ^{Ba}	5.3 ^{Ba}	4.4 ^{Ab}	3.4 ^{Bbc}	2.3 ^{Cc}				
Antioxidant activity DPPH (mg trolox equivalents/g DM)									
Sainfoin	23.9 ^{Aa}	22.4 ^{Aa}	9.6 ^{Ac}	11.9 ^{Ab}	7.2 ^{Ad}	0.49	<0.001	<0.001	<0.001
Pasture	9.1 ^{Ba}	7.3 ^{Cab}	6.5 ^{Bb}	6.1 ^{Bb}	3.1 ^{Bc}				
Cocksfoot	10.8 ^{Bb}	13.0 ^{Ba}	9.0 ^{Ab}	5.4 ^{Bc}	2.5 ^{Bd}				

FF, fresh forage; US, unwilted silage; WS, wilted silage; HG, haylage; H, hay. Forage values in the same column with different capital letter superscripts are different ($P < 0.05$); significant differences ($P < 0.05$) between conservation methods within forages are indicated by different lowercase letter superscripts in the same row. F, P value for the effect of forage; C, P value for the effect of the conservation method, F×C, P value for the interaction between forage and conservation method.

dominant pasture (88% Poaceae, mainly *Poa pratensis* L. 1753, *Poa trivialis* L. 1953 and *Holcus lanatus* L. 1753, 3% Fabaceae and 9% other dicotyledon species), sainfoin (*Onobrychis viciifolia* Scop, cv. Lupinella) and cocksfoot [orchardgrass] (*Dactylis glomerata* L., cv. Archibaldi). Plots of sainfoin and cocksfoot were sown on May 2022. The three plots were split into three areas as replicates. All forages were harvested on 30 May 2023 at late bud stage for sainfoin, heading stage for pasture and earing stage for cocksfoot. After cutting, in each of the 9 experimental areas biomass was separated into that for fresh forage and that for four types of conserved forage (unwilted and wilted silages, haylage and hay); hence there was a total of 45 samples. Biomass for unwilted silages (US) were immediately chopped (5 cm) and ensiled. Wilted silages (WS), haylages and hays were made after wilting grass on ground during respectively 6, 24 and 96 hours. Wilted silages were also chopped (5 cm) before ensiling. Silos and hay bales were opened after 4 months. All samples were freeze-dried and ground to less than 1 mm before analysis. Total phenolic compounds concentration was determined by a modification of the method using the Folin-Ciocalteu as described by Maxin *et al.*, 2020. Results were expressed as milligrams of gallic acid equivalents per gram of product dry matter (DM). Antioxidant activity was assessed using the measurement of the free-radical scavenging activity of the DPPH* (2,2-diphenyl-1-picrylhydrazyl). Results for DPPH were expressed as mg trolox equivalents per g DM. Data were analysed by ANOVA model including conservation method ($n = 5$), forage ($n = 3$) and their interaction as fixed effects, and area ($n = 3$) as random effect. Multiple comparisons of means were performed with a Tukey test.

Results and discussion

The achieved DM contents for the different forages are presented in Table 1 and average values were 88.7% for hays, 58.7% for haylages, 31.8% for WS, 17.0% for US and 17.7% for fresh forages. As observed previously (Rufino-Moya *et al.*, 2022; Amrit *et al.*, 2023), TPC concentrations and AO determined with the DPPH method were positively correlated ($r = 0.951$, $P < 0.001$) meaning that phenolics present in the forages contributed significantly to the antioxidant activity.

Total phenolic compounds concentrations and AO were significantly higher in fresh sainfoin (11.8 mg gallic acid equivalents/g DM and 23.9 mg trolox equivalents/g DM) than in fresh pasture and cocksfoot (5.1 and 5.0 mg gallic acid equivalents/g DM and 9.1 and 10.8 mg trolox equivalents/g DM, respectively). Although all values obtained in this study were lower than previous published values (sainfoin: Maxin *et al.*, 2020; pastures: Maxin *et al.*, 2024; cocksfoot: Hauck *et al.*, 2004), they confirmed that sainfoin is a good provider of secondary metabolites. Phenolic compounds and antioxidants highly vary with species, varieties and environmental features (Treutter, 2006; Maxin *et al.*, 2020), which could explain the lower values observed here compared to previous studies.

The conservation method affected the TPC concentration and AO of the three forages. The highest TPC concentrations were observed in fresh forage for sainfoin and the permanent pasture, and in fresh forage and US for cocksfoot. For the AO, the highest values were obtained in fresh and US for sainfoin and the permanent pasture and in US for cocksfoot. Total phenolic concentrations and AO were significantly lower in hay for all forages with on average a reduction of 52 and 71%, respectively, compared to fresh forages. The values for WS and haylages were intermediate between fresh forages and hays. The degradation observed in conserved forages could be explained by losses of leaves, a rupture of plant cells during chopping and/or light and oxygen exposure. Lower values in hays compared to silages observed in the present study disagree with Rufino-Moya *et al.* (2022) that observed higher TPC concentrations and AO values in hay than in silage for sainfoin. However, harvest and storage conditions, especially storage duration were different between both studies. The higher TPC concentration and AO in US for cocksfoot could be linked to its low conservation quality (high butyric and alcohol concentrations and presence of mild, data not shown). In fact, TPC can increase in degraded silage quality due to the stress suffered by plant cells during fermentation process and mould contamination (Alba-Mejia *et al.*, 2023).

Conclusion

The conservation method affected the total phenolic compounds concentration and antioxidant activity in all forages. Compared to fresh forages, they decreased more in wilted forages than in unwilted ensiled forages. *In vivo* studies are now required to assess the impacts on animal health and make some recommendations.

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Variations in phenolic concentrations and antioxidant activity of permanent pastures according to botanical variation

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Abstract

Pastures contain polyphenols with health-promoting abilities in animals, especially antioxidant effects. Knowledge on the phenolic content and antioxidant activity of pastures is limited. The objective of this study was to assess the variations in total phenolic content (TPC, Folin method) and antioxidant activity (AO, ORAC and DPPH methods) of 33 pastures located in 3 areas of France according to their botanical composition. Grass samples were collected during spring 2022 and 2023 at a sum of temperatures of $707 \pm 145^\circ\text{KD}$. Relationships between TPC and AO contents and botanical composition were studied. Total phenolics and AO evaluated with DPPH and ORAC highly varied between pastures. Results showed that TPC and AO decreased when the proportion of Poaceae increased, whereas they increased when the number of dicotyledons species in the pasture increased. Other botanical families seemed to contribute to TPC and AO values. Next step will be to study the involvement of certain species in the variations.

Keywords: animal health, antioxidants, grassland, polyphenols, phenolics

Introduction

Animal health management is a key point for sustainable ruminant production systems. Plants contain secondary metabolites such as vitamins or phenolics that may have different health-promoting abilities in animals, especially antioxidant, anti-parasitic or anti-microbial effects (Poutaraud *et al.*, 2017). However, knowledge on phenolic content and antioxidant activity of pastures and the factors affecting these characteristics remains scarce. We had previously observed a high variability in total phenolic content and antioxidant activity in grassland (Maxin *et al.*, 2024). Environmental conditions may act on phenolics in plants. However, Reynaud *et al.* (2010) suggested also that botanical composition could affect the phenolic content and composition of pastures: their contents would be higher in more diversified pastures and linked to the occurrence of some dicotyledon families, such as Asteraceae and Plantaginaceae. Thus, the objective of our study was to investigate the effect of botanical diversity on phenolic content and antioxidant activity of different permanent pastures located in three areas of France.

Materials and methods

Samples from 33 permanent pastures were collected in 3 different areas of France: in Auvergne-Rhône-Alpes, Haute-Saône and Indre. Samples were collected twice during spring 2022 and 2023 at a sum of temperature of about $707 \pm 145^\circ\text{KD}$. The sum of temperatures was calculated, as the cumulative

Table 1. Total phenolic content, antioxidant activity and proportions of the identified major botanical families in the 33 pastures studied

	<i>n</i>	Mean	SD	IQR	Min	Max
Total phenolics (mg gallic acid equiv./g DM)	64	25.4	6.52	7.3	8.6	42.2
DPPH (mg trolox equiv./g DM)	63	31.0	9.42	11.6	9.4	57.1
ORAC (mg trolox equiv./g DM)	64	153.3	41.81	46.3	51	255
Total identified dicotyledon species	64	10.9	5.41	6.0	2.0	28.0
Poaceae (% contribution in total quadrat biomass)	64	68.9	15.39	23.7	34.4	92.3
Fabaceae (% contribution in total quadrat biomass)	64	7.9	7.49	11.9	0.0	31.3
Asteraceae (% contribution in total quadrat biomass)	64	6.8	7.36	7.0	0.0	41.7
Ranunculaceae (% contribution in total quadrat biomass)	64	6.1	6.34	6.9	0.0	30.7

² IQR, interquartile range.

sum of above zero daily average temperature from 1 February to the sample collection day considered (Theau & Zerourou, 2005). Each year, the botanical composition of selected pastures was determined using 10 quadrats (50 × 50 cm) randomly distributed within the plot. All species growing in the quadrat were identified and their contribution to the total quadrat biomass was estimated using hand recovery method. Contributions from the 10 quadrats were averaged for each plot. Then, biomass contained in each quadrat was harvested and mixed to constitute a representative sample of each pasture. Grass samples were freeze-dried and ground before analysis. Total phenolic content (TPC) was assessed by the Folin–Ciocalteu method (Maxin *et al.*, 2024) and expressed as mg of gallic acid equivalents per g DM. The antioxidant activity (AO) was estimated using two different methods: the measurement of the free-radical scavenging activity of the DPPH* (Maxin *et al.*, 2024) and the measurement of the oxygen radical absorbance capacity (ORAC, Vegepolys Valley Lab, Angers, France). First, the quantitative relationship between TPC, AO and the botanical families was studied by principal component analysis (PCA) using FactoMiner package in R software (version 4.5.1). All variables were active. Then, mixed models (lme4 package) were performed to explain TPC and AO: botanical families and the sum of temperatures were tested, and year and plot were included as random effects. The low collinearity between the sum of temperatures and the botanical families included in the models was verified.

Results and discussion

TPC and AO highly varied between pastures (Table 1). Total phenolic content was significantly and positively correlated to DPPH ($r = 0.88$) and ORAC ($r = 0.96$) values, suggesting that phenolics present in pastures had a significant antioxidant activity. Pastures presented an important botanical diversity: a total of 152 different plant species from 21 botanical families were identified. The most frequently occurring families were Poaceae, Fabaceae and Asteraceae.

The first 3 axes of the PCA represented 52.7% of total variability. The first axis (28.0%) made a distinction between the TPC, ORAC and DPPH values and some botanical families: pastures with higher proportions of Fabaceae, Asteraceae and Plantaginaceae and the higher number of dicotyledon species in plot seemed to have a positive influence on the values. On the contrary, the high proportion of Poaceae and some minor families (Convolvulaceae, Violaceae or Orobanchaceae) seemed to have negative correlation. These results suggest that more diversified pastures would present higher TPC, ORAC and DPPH values, confirming previous results (Reynaud *et al.*, 2010). The second axis (13.3%), less exploratory, discriminated the pastures mainly on the sum of temperatures which would have a positive influence on TPC, ORAC and DPPH values. The best mixed models obtained to explain TPC, ORAC and DPPH were consistent with PCA results (Table 2): an increase in the contribution of

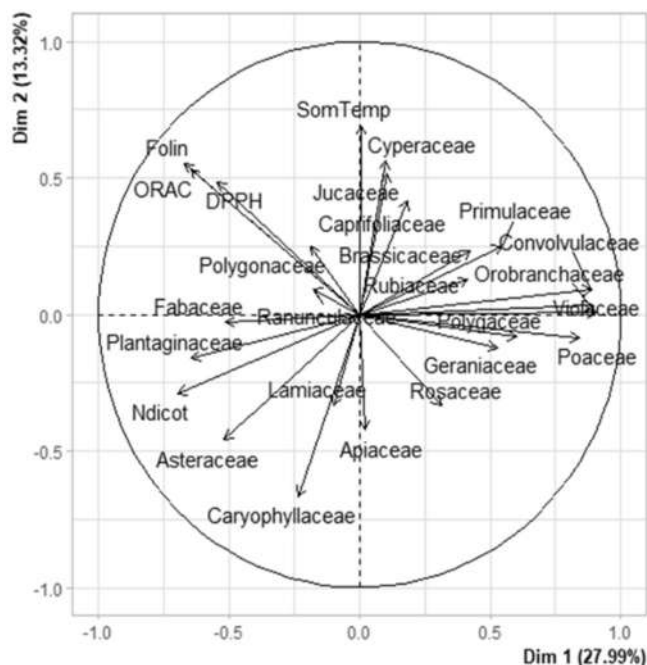


Figure 1. Graph of variables. Principal component analysis with botanical family's proportion, Folin, DPPH and ORAC, and number of dicotyledon species.

Table 2. Best models for total phenolic content (TPC), antioxidant activity determined with DPPH and ORAC methods using the botanical family's proportion and the sum of temperatures.

	Intercept	Covariables ¹						RMSE
		SumTemp	Poaceae	Rosaceae	Plantaginaceae	Apiaceae	Juncaceae	
TPC	24.0 ± 4.51***	0.02 ± 0.006***	-0.15 ± 0.040***	-1.06 ± 0.306***	0.68 ± 0.274*	0.60 ± 0.302*	-0.17 ± 0.081*	2.4
DPPH	29.8 ± 7.28***	0.03 ± 0.009***	-0.18 ± 0.065**	-0.77 ± 0.404*	1.07 ± 0.429*	—	-0.28 ± 0.128*	5.6
ORAC	141.1 ± 31.32***	0.18 ± 0.043***	-0.96 ± 0.297**	-6.21 ± 2.111**	3.48 ± 1.980*	—	-1.37 ± 0.591*	18.5

Values are coefficient ± SE. RMSE, residual means square error; *** $P < 0.001$; ** $P < 0.01$; * $P < 0.05$.

Poaceae, Juncaceae and Rosaceae negatively affected TPC, ORAC and DPPH values whereas Plantaginaceae had a positive effect. Apiaceae had also a positive effect on TPC. Both families, Plantaginaceae and Apiaceae have been already identified as potential antioxidant sources (Gonçalves & Romano, 2016; Thiviya *et al.*, 2021).

Conclusion

The first results of this study enabled to confirm that differences in TPC and AO could be explained by variations in botanical composition. Next step will be to study the involvement of specific species in the variations.

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Specifications of livestock products with origin and quality labels can help protect multifunctional grasslands

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Abstract

Grass-based livestock farming is recognized as a multifunctional way to produce food, by supplying both human-edible proteins and regulating ecosystem services. However, European grasslands are threatened by conversion to crops in lowlands and afforestation in uplands. Livestock products labelled with a PDO (Protected Designation of Origin), whose production relies on local forages resources, have the potential to mitigate this threat. We assessed this potential in the Massif Central, a mountainous area of central France. We studied the specification documents of eight PDO products and one product under 'Label Rouge', a French quality label (7 cheeses and 2 beef). The specification documents include 5 to 12 rules, with an average of approx. 9 for both cheese and beef. Most documents have rules related to the origin of the animal feed (8/9), the proportion of grass in the diet (6/9), and stocking rates (7/9). Some also include fertilization rules (2/9), and one requires that permanent grasslands represent at least 90% of the farm's grassland area. Overall, most specifications contain rules on grassland maintenance and management, confirming that labelled products can contribute to grassland protection in the Massif Central.

Keywords: beef, cheese, dairy, multifunctionality, Protected Designation of Origin, rules

Introduction

Grass-based livestock farming is recognized as a multifunctional way to produce food, by supplying ecosystem services (ES) of different types (Leroy *et al.*, 2024). They provide provisioning ES through human-edible proteins, while providing regulating ecosystem services, such as erosion prevention, water purification or pollination. However, grasslands are threatened by conversion to crops in lowlands and afforestation in uplands. Their multifunctionality is also threatened by increased fertilization and high stocking rate, which reduces the levels of regulating ES (Schils *et al.*, 2022).

Livestock products labelled with a PDO (Protected Designation of Origin), whose production must rely on local forage resources, may mitigate these threats, through securing a local feed source and maintaining traditional farming (Flinzberger *et al.*, 2024). We assessed this potential in the Massif Central, a mountainous area of central France comprising an important share of grasslands (Allart *et al.*, 2024), and where numerous PDO cheeses are present.

Materials and methods

We studied eight PDO products and one product under 'Label Rouge', a French quality label, representing 7 cheeses and 2 beef types (Table 1). We downloaded the specification documents of these products on the website of INAO (Institut National de l'Origine et de la Qualité), the French national institute which validates and protects the labels of origins and quality (<https://www.inao.gouv.fr/>, accessed 16 July 2025). We listed all the management rules that can impact grasslands and their management.

Table 1. Summary table of the management and production rules in the specification documents of nine products with labels of origin (PDO) or quality (Label Rouge), Q: Quantified or semi-quantified, M: Mentioned but not quantified

Specifications	PDO Salers	Label Rouge Bœuf fermier Aubrac	PDO Fourme d'Ambert	PDO Laguiole	PDO Bleu d'Auvergne	PDO Fourme de Montbrison	PDO Cantal	PDO Fin gras du Mézenc	PDO Saint- Nectaire
Type of product	Cheese	Beef	Cheese	Cheese	Cheese	Cheese	Cheese	Beef	Cheese
Animal diet rules									
Feed origin (PDO area)	Q		Q	Q	Q	Q	Q	Q	Q
Specific feeding rules (e.g. only the first cut of hay used)		Q	Q	Q	Q	Q	Q	Q	Q
Concentrate limitation	Q		Q	Q	Q	Q	Q	Q	Q
Ban of GMO feed			M	M	M	M	M	M	M
Minimum duration of grazing		Q	Q	Q	Q	Q	Q	Q	Q
Proportion of grass in the diet	Q		Q		Q	Q	Q		Q
Silage use constraint		Q		Q		Q			Q
No crucifers in the diet			Q		Q	Q			
Green fodder in the diet						M	Q		
Production and animal management rules									
Origin of cattle in the PDO area					Q		Q	Q	Q
Specific production period	Q							Q	
Specified breed		M		M					
Minimum duration of the finishing period		Q							
Milk production limitation				Q					
Land management rules									
Maximum stocking rate		Q	Q		Q	Q	Q	Q	Q
Fertilisation limitation					M		M	Q	Q
Permanent grassland constraint								Q	Q
Environmental rules (e.g. floristic diversity)								M	M
Total	4	6	8	8	10	10	10	11	12

Results and discussion

The specification documents include 4 to 12 management rules, with an average of ~9 rules for both cheese and beef. The rules concern animal feed and diet, animal management and production, as well as land management (Table 1). Most documents have quantitative, or semi-quantitative rules related to the origin of the animal feed (8/9) and the proportion of grass in the diet (6/9). For example, the 'Bleu d'Auvergne' specifies that the 'basic diet of the dairy herd is exclusively made of forage from

the local PDO area' and that the grass in different forms (grazed, hay, or silage) must represent at least 70% of diet of the lactating cows', in terms of dry matter. Two out of seven documents have also quantitative rules regarding farm land cover, for example 90% of the grassland area must be permanent grassland for 'Saint Nectaire'.

All the specification documents provide in addition a list of allowed concentrates. All these rules are justified by the specific taste that feed can provide, in particular through local plant species, the quality of the milk that can be produced, and the preservation of local traditions. Quantitative rules exist in addition for stocking rates (7/9) and fertilization (2/9), they are also motivated to maintain traditional uses. Some rules to maintain local flora also exist for two products, but they are not quantified ('grasslands are used annually in a way that maintain floristic diversity' for 'Fin Gras du Mézenc').

Most of the labels have therefore quantitative or semi-quantitative rules to ensure a large share of grass in cattle diet, which contributes to the maintenance of grasslands in the PDO regions. Direct but unquantified rules related to the preservation of flora are also present, and indirect quantified rules related to the traditional mode of production of labelled products (stocking rate and fertilization) are in addition represented, and they can contribute to improved grassland multifunctionality.

However, it is difficult to compare quantitatively which labels can best promote multifunctionality because, for a given specification, they use distinct indicators or metrics, or group of animals. For example, the maximum stocking density for 'Bleu d'Auvergne' is 1 livestock unit per hectare of grassland (LU/ha), and only applies to dairy cows in the herd supplying milk for PDO cheese, whereas 'Fourme de Montbrison' indicates that the maximum stocking rate is 1.4 LU/ha for the whole farm area, and applies to the whole farm and animal species (i.e. cattle and possibly sheep and goats). It is nevertheless possible to identify labels that are particularly protective of permanent grasslands, which provides higher levels of biodiversity and regulating ES than temporary grasslands (Schils *et al.* 2022). For cheese Saint-Nectaire fixes a percentage of permanent grassland in the grassland areas (90% as mentioned above) and for beef, 'Fin Gras du Mézenc' indicates that animals must be fed on grasslands that are at least 10-year-old, which is higher than the 5 years above which a grassland is considered a permanent one.

Conclusion

Unlike organic products, which can directly contribute to preserving environment through pesticide interdiction, PDO and quality labels were not initially designed to protect the environment. However, in the beef and dairy context of Massif Central, their production involves the maintenance of grasslands, i.e., a semi natural habitat supplying high levels of regulating ES, and host biodiversity. The specification document of PDO and quality labels have thus the potential to support and guide the implementation of sustainable livestock farming practices, through the maintenance of multifunctional grasslands.

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Produce beef from dairy and make the most of grasslands in a low-input system

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Abstract

The use of sexed semen on dairy cows offers the opportunity to cross part of the dairy herd with beef bulls to produce beef from dairy herds. The Tripl'Scotch experiment has been set up on the INRAE experimental farm at Le Pin (Normandy) to produce Angus crossbred animals from a dairy herd made up of 3 breeds: Holstein (Ho), Normande (No) and Jersey (Je). During 4 birth years (2020 to 2023), 102 steers and heifers have been reared with grazed grass and conserved grass silage. The animals are weighed fortnightly and the bodyweight is calculated by linearization at a fixed age to describe the individual growth profiles. Ho and No crossbreeds have the same growth profile, while Je crossbreeds are always significantly lighter. After a 3-months indoor finishing period with excellent grass silage and no concentrate, all the animals are slaughtered at 2-years old, at an average body weight of, respectively for Ho, No and Je crossed Angus, 608, 611 and 532 kg, and 49.0, 49.7 and 48.1% carcass kill out rate. This type of beef production helps to maintain and make the most of unused grasslands for dairy cows and to improve the profitability of the dairy system with beef progeny which are desired by societal demand.

Keywords: beef, crossbred, grazing, growth

Introduction

Traditionally, male calves in dairy systems are of little demand and value and are often sold at 2 weeks of age to produce veal calves or young bulls. This problem is exacerbated in certain specialized dairy breeds, such as the Jersey. The increasing use of female sexed semen (Boichard *et al.*, 2024) offers the possibility of crossing some of the dairy herd with beef bulls to produce crossbred calves that have improved rearing and fattening potential. Some breeders cross with Belgian Blue or Charolais to add value to calves sold at 2 weeks old, while others cross with Angus or Hereford to produce 2-year-old steers and heifers. Angus and Hereford breeds offer many advantages, from ease of calving to early-maturity, good adaptation to a grass-based system, hornless and high meat quality. This last approach was applied in the Tripl'Scotch experiment carried out on the INRAE experimental farm at Le Pin (Normandy) ([10.15454/1.5483257052131956E12](https://doi.org/10.15454/1.5483257052131956E12)) with the objective to produce beef from grasslands which were less productive or too far from the milking parlour. The aim of this article is to present the management and the results obtained after 4 complete cohorts of crossbred animals reared and fattened solely on pasture and grass silage, and slaughtered around the age of 2 years.

Materials and methods

The Tripl'Scotch experiment is implemented in the multi-year Tripl'XL experiment (Delaby *et al.*, 2024). Briefly, Tripl'XL compares the response of 3 breeds of dairy cows, Holstein (Ho), Normande (No) and Jersey (Je) to different times of concentrate allocation during lactation. Cows are managed in a grass-based system maximising grazing (April to Nov) with compact 3-month calving (Feb to

April). The breeding season is divided in two periods, 5 weeks (21 April to 10 June) using pure breed sexed semen and 7 weeks (10 June to 21 July) where Angus bulls are used according to observed heats of cows not yet in-calf. Around 10 (2020) to 40 (2023) crossbred calves (HoX, NoX and JeX) were subsequently born every year in a 7-week calving period. These calves are reared during the first 5 months with the replacement dairy heifers, and are subsequently managed in a single group to the end of the fattening period, 2 years later. After the milk phase (weaning at 73 days on average), they received a ration composed of straw + concentrate (around 400 kg gross) indoor up to mid-September. At 5 months, the males are surgically castrated and the group of cross-bred animals graze for around 2 months. During the 1st winter period, from November to April, the ration is composed of grass silage or big bale haylage (5 to 6 kg day⁻¹, dry matter based—DM). A long grazing period (220 to 240 days) takes place to the end of November. The single group grazes grasslands which are less productive or too far from the milking parlour for the dairy herd. The 2nd winter period (December to March) is the indoor fattening period with grass silage of excellent nutritive value, offered ad libitum.

The animals are weighed fortnightly, each Wednesday afternoon, on diverse calibrated scale Marechalle (Chauny, France), from birth to slaughter and bodyweight (BW, kg) is calculated by linearization at a fixed age. The average daily gain (ADG, g day⁻¹) is obtained by difference of BW at fixed age divided by the period duration concerned. At grazing, during the second year, pre- and post- grazing sward height is assessed with a platometer (80 measures per ha). Before grazing, grass samples are taken with scissors, oven-dried (60°C—48 hours) and analysed for chemical composition and pepsin-cellulase digestibility to estimate the nutritional value of the grass offered (INRA, 2018). During the two indoor periods, daily DM intake is measured at the group level and calculated as an average per animal. The GLM model applied (SAS 9.4, SAS Institute, Cary, NC) on the individual BW, carcass weight (CsW, kg) and ADG for key measurement dates includes the effect of the year of birth ($n = 4$), sex ($n = 2$), dam breed ($n = 3$) and the interaction between sex and dam breed.

Results and discussion

The main results in terms of BW and ADG are presented by breed of dam in Table 1. For all the parameters studied, no interaction between sex and dam breed have been observed. After 4 experimental years, 102 crossbred animals (55 steers, 17/20/18; and 47 females, 14/14/19 Je/No/Ho respectively) were slaughtered at an average age of 712 days (between 635 and 775 days), with no significant difference of age between males and females or breed of the dam.

At birth and at weaning, the JeX animals are significantly lighter than the HoX and NoX (−11 and −16 kg, respectively). After weaning, during the 2 months of grazing, the animals needed 0.15 to 0.18 ha. During the first winter, the individual grass silage intake was on average 6 kg DM per day. At 330 days of age (2nd turnout), HoX and NoX have a similar BW (301 and 294 kg, respectively) and are heavier ($P < 0.001$) than JeX (270 kg).

The grazing area used during the second grazing period increases from spring to autumn as a result of the increase in grass intake associated with BW gain, and the decrease in grass growth. On average, the area required in spring (April to June) was 0.30 ha per animal (310 kg) or 1.0 ha per 1000 kg BW. This rises to 0.55 ha per head (370 kg) in summer, i.e. 1.5 ha per 1000 kg BW. In autumn (from Sept to November), each animal (430 kg) grazes 0.85 ha (2.0 ha per 1000 kg BW). The pre-and post-grazing height was on average 10.5 and 5.5 cm respectively. In average, the chemical composition of the grass offered shows good nutritive value with 164 g of CP, 515 g of NDF, 275 g of ADF and an OM

Table 1. Bodyweight (BW), average daily gain (ADG) and carcass weight (CsW) Holstein (Ho), Normande (No) and Jersey (Je) crossbred cattle.

	Dairy breed (n)			RMSE
	Ho (37)	No (34)	Je (31)	
BW at birth (kg)	44.5 ^a	44.8 ^a	33.1 ^b	6.7
BW at weaning (70 days) (kg)	97 ^a	97 ^a	81 ^b	10.8
BW at 1st indoor winter (240 days) (kg)	223 ^a	217 ^a	200 ^b	21.6
BW at 2nd turnout (330 days) (kg)	301 ^a	294 ^a	270 ^b	31.6
ADG during 2nd grazing (g day ⁻¹)	727 ^a	722 ^a	635 ^b	119
BW at 2nd indoor winter (600 days) (kg)	497 ^a	489 ^a	441 ^b	42.8
ADG during fattening (g day ⁻¹)	1003 ^a	1070 ^a	812 ^b	240
BW at slaughter (700 days) (kg)	608 ^a	611 ^a	532 ^b	49.6
Age at slaughter (days)	707	714	714	22.9
CsW (kg)	298 ^a	304 ^a	256 ^b	25.5
CsW yield (%)	49.0 ^a	49.7 ^a	48.1 ^b	2.01

Adjusted means for year and sex ratio effects. On the same row, two results without common letter are significantly different.

digestibility of 723 g.kg⁻¹ DM. The estimations with the INRA (2018) equations of the UFL and PDI values are 0.92 (1620 kcal NEL) and 90 g (kg DM)⁻¹, respectively. During this grazing season, the ADG is more than 700 g for the HoX and NoX and significantly lower for the JeX (635 g day⁻¹). As expected, the ADG is superior for the steers than the heifers (740 and 650 g day⁻¹, respectively, $P < 0.001$). At the end of November, when animals are back indoors, HoX and NoX weigh 497 kg and 489 kg, respectively (NS) while JeX are approximately 50 kg lighter (441 kg, $P < 0.001$).

During the final winter period (4 months), the individual grass silage intake evolves from 9.8 kg DM at the beginning to 12.2 kg DM during the last two months of the fattening period. The quality of grass silage used was high (150 g CP, 1690 kcal NEL or 0.96 UFL and 75 g PDI/ kg DM) and allowed all animals to be finished without any concentrate. For HoX and NoX, similar BW (608 and 611 kg) and CsW (298 and 304 kg) at slaughter were observed while JeX were significantly lower (532 and 256 kg, respectively). The final carcass yield was also higher for HoX (49.0%) and NoX (49.7%) compared to JeX (48.1%). Almost all the animals met the carcass specifications defined for slaughter, in terms of carcass weight (min-max), conformation (O = or O+) and fatness (3 to 4).

Conclusion

The Tripl'Scotch experiment demonstrates the possibility of developing “beef from dairy” products, with Ho, No or Je dairy cows, that combine a number of positive aspects. Angus cross-bred animals are able to make the most of grassland with very few concentrates, and produce a carcass weight that meets consumer demand in terms of size and quality of the meat cuts. This co-product is also of interest to dairy farmers as an added value for cross-bred calves, kept for rearing and fattening on the farm or sold to others such as crops farms wishing to reintroduce grassland for agronomic reasons in their crop rotations.

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Grazing area and goat mobility in a Mediterranean forest rangeland using precision livestock tools

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Abstract

Understanding how grazing animals interact with forest–grassland mosaics is critical for the long-term sustainability of Mediterranean rangelands. We investigated seasonal changes in goat grazing patterns and space use within a forest pasture in northern Morocco by combining GPS collar tracking with leg sensors. Monitoring was carried out in spring, summer and autumn, and GPS data were complemented with forage availability measurements to map seasonal grazing ranges. The goats travelled from 8.3 ha in spring to more than 17 ha in summer and autumn, indicating greater mobility when feed resources were scarce. In spring, goats devoted most of the daytime to grazing (48%), whereas in summer and autumn, grazing time dropped, and daily walking distances increased. Energy balance assessments showed surpluses in spring but deficits in the two other seasons, linked to longer movement times and lower forage intake. The study demonstrates the value of precision monitoring tools for capturing fine-scale animal–landscape interactions and for guiding adaptive grazing management.

Keywords: forest rangeland, GPS collar, grazing behaviour, Mediterranean ecosystem, sensor

Introduction

In northern Morocco, forest pasture ensures free forage production for grazing animals. In this region, the livestock farming system is concerned with grazing goats in forest rangelands, ensuring free animal feed all year round (Chebli *et al.*, 2018). Browsing in mountainous forest rangelands of northern Morocco involves additional physical activities for the goats, such as the vertical movement (Chebli *et al.*, 2022) due to the harsh landscape characterized by steep and uneven slopes. These altitudinal movements increase the time and energy required to travel a given distance, which negatively affects livestock productivity and implicitly the household income. This kind of information is difficult to obtain only by using direct observation because observers cannot precisely measure individual animal behaviour, such as movement and activity (Hlimi *et al.*, 2024). Data on animal activities are essential to understand the feeding and grazing behaviour of animals and to identify optimal management strategies. This study aimed to: (i) assess seasonal variations in grazing area; (ii) examine changes in goat mobility and activity patterns; and (iii) evaluate seasonal energy balance using GPS collars and sensors.

Materials and methods

The study was conducted in the forest rangelands of Beni Arouss, in northern Morocco, an area with a typically Mediterranean climate. The vegetation is dominated by holm oak (*Quercus rotundifolia*) and strawberry tree (*Arbutus unedo*), accompanied by shrub species such as *Cistus* spp. and *Erica arborea*. Eight adult, non-pregnant local goats (30 ± 2.6 kg and an age of 36 ± 6 months) were fitted with GPS collars (recording positions at regular intervals) and tri-axial accelerometer leg sensors to monitor activity (Figure 1). Tracking was performed over three seasons: spring, summer, and autumn. GPS data recorded grazing periods during daylight, which were used to map grazing areas and calculate daily



Figure 1. Goat wearing a GPS collar (A) and a leg sensor (B). Downloading GPS (C) and leg sensor (D) data.

walking distances. Activity sensors classified behavior into grazing, walking, or resting. Grazing areas were estimated using kernel density methods to delineate seasonal home ranges. Energy expenditure was calculated from walking distance and activity data. Metabolizable energy intake was estimated from forage availability and diet composition to determine seasonal energy balance. The methods are described in detail by Chebli *et al.* (2022).

Results and discussion

The spatial extent of grazing varied considerably between seasons (Figure 2). Although Figure 2 may give the impression that travel distances are higher in spring, the data clearly show that goats travelled longer daily distances during summer and autumn compared to spring. During spring, forage is abundant and spatially continuous, allowing goats to satisfy their dietary needs within more restricted areas and therefore travel shorter total distances despite high grazing activity. In spring, goats concentrated their activity within a relatively compact grazing area of 8.3 ha. This reflects a lower need to travel in search of feed, as high-quality forage was readily available near resting sites. By contrast, during summer and autumn, the decline in biomass and forage quality forced animals to expand their range considerably, exceeding 17 ha in both seasons. GPS tracking showed that grazing routes became more dispersed, with goats visiting a wider array of vegetation patches. Daily walking distances increased markedly in these periods, indicating a behavioural adjustment to reduced resource availability. This expansion of the grazing footprint during feed scarcity is consistent with patterns reported in other Mediterranean rangelands and has implications for vegetation pressure and regeneration capacity (Rodrigues *et al.*, 2013).

The goats spent 48% of their feeding time browsing (eating) during the spring, in contrast to the summer (27%) and autumn (31%). In summer and autumn, grazing time declined significantly, while the proportion of time spent walking increased. This shift suggests that animals devoted more effort to searching for dispersed and lower-quality feed resources. Periods of prolonged locomotion, combined with reduced intake, contributed to negative energy balances in these seasons (Papachristou, 1997).

The energy balance of the studied goats was positive in spring, but deficits emerged in summer and autumn due to the combined effects of lower intake and higher locomotion costs. These findings echo



Figure 2. Seasonal movement patterns of local goats in Beni Arouss forest rangelands (Northern Morocco).

results from other Mediterranean rangelands where seasonal forage gaps compromise animal nutrition (Chebli *et al.*, 2020).

The integration of GPS tracking and activity monitoring provided a fine-scale picture of goat–landscape interactions across seasons. The observed expansion of grazing areas during feed scarcity underscores the need for adaptive stocking rates and targeted feed supplementation during critical periods. Without such measures, prolonged high mobility and energy deficits could lead to declining animal performance and accelerated vegetation degradation (Schlecht *et al.*, 2009).

Conclusion

Precision Livestock Farming tools proved effective in quantifying spatial and temporal patterns of goat grazing in a Mediterranean forest rangeland. Seasonal differences in grazing area, mobility, and energy balance reflect strong links between forage availability and animal behavior. Management strategies that combine adjusted stocking rates with strategic supplementation during summer and autumn can help sustain both herd productivity and ecosystem resilience. This approach supports data-driven decision-making for the sustainable use of Mediterranean silvopastoral resources.

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Pasture Base Ireland: new insights in temperate grassland production

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Abstract

Farm grassland dry matter (DM) is rarely documented on commercial farms in Europe. PastureBase Ireland (PBI) is a web-based grassland management decision support tool developed in 2013 for Irish grassland farmers. The objectives of using PBI on grassland farms is to focus on optimising grass utilisation across all ruminant sectors, improving farm grassland productivity, promoting sustainable grazing practices and supporting evidence-based grassland decision-making. Annual pasture growth averaged 13.2 t DM ha⁻¹ during the eleven-year period, with 7% variation in DM production (+/-907 kg DM ha⁻¹) between years. This paper investigates the grassland management performance from a long term dataset of grassland farms ($n = 163$) within the PBI database and describes their grassland performance.

Keywords: decision support, grassland management, grass utilisation, PastureBase Ireland

Introduction

Pasture Base Ireland (PBI) (Hanrahan *et al.*, 2017) has a range of grassland support tools available to farmers to assist in short, medium and long-term grassland decisions. Each year, a growing number of farmers are using the application. In 2024, over 142 000 farm covers were recorded, an increase of 43 000 since 2022. There is a clear and continual increase in grassland measurement across the country, which has been aided by the grassland measurement requirement for farmers with a nitrate's derogation plan.

There are over 11 application program interfaces linked from external Agri industry partners linked to PBI, these include Jenquip plate meter, Grasshopper plate meter, dairy Co-ops individual supplier milk inputs, Feed and soil analysis labs etc. On Irish farms white clover is now becoming more widely used, due to reductions in chemical N usage. Several new decision support tools have been developed in PBI to assist strategic decision-making regarding nutrient application, including the 'Nitrogen Planner', nitrogen cycling calculator.

Materials and methods

The current dataset comprised of 163 farms who completed 35 or greater farm covers in PBI from 2014 to 2024. By increasing the level of farm cover measurements on farm, the level of grass production is better quantified. All farms used had cumulative paddock summary report data calculated. Within the database, grazing events per paddock, spring, summer and autumn DM production were calculated, as well as grass allowance per cow, grass silage supplementation and concentrate offered during the season using the methodology described by (Hanrahan *et al.*, 2017).

Results and discussion

Grazing management and grass DM production data from the farms from 2014–2024 are summarised in Table 1. Annual pasture growth averaged 13.2 t DM ha⁻¹ during that period, with 7% variation

Table 1. Seasonal and annual DM production (t DM ha^{-1}) over eleven years (2014–2024) on a sample of Pasture Base Ireland farms ($n = 163$)

Year	Spring	Summer	Autumn	Annual	No of grazing and silage events
2014	1.6	6.4	5.1	13.1	7.1
2015	1.7	6.4	5.5	13.6	7.5
2016	1.4	6.6	5.6	13.6	7.9
2017	2.2	6.5	5.6	14.3	7.9
2018	1.3	5.0	4.9	11.2	6.7
2019	2.1	6.6	5.2	13.9	7.9
2020	2.0	6.4	5.5	13.8	8.0
2021	2.1	6.1	5.7	14.0	8.4
2022	1.9	6.0	4.6	12.5	8.2
2023	1.9	6.0	5.3	13.2	8.1
2024	1.7	5.6	4.9	12.3	8.1

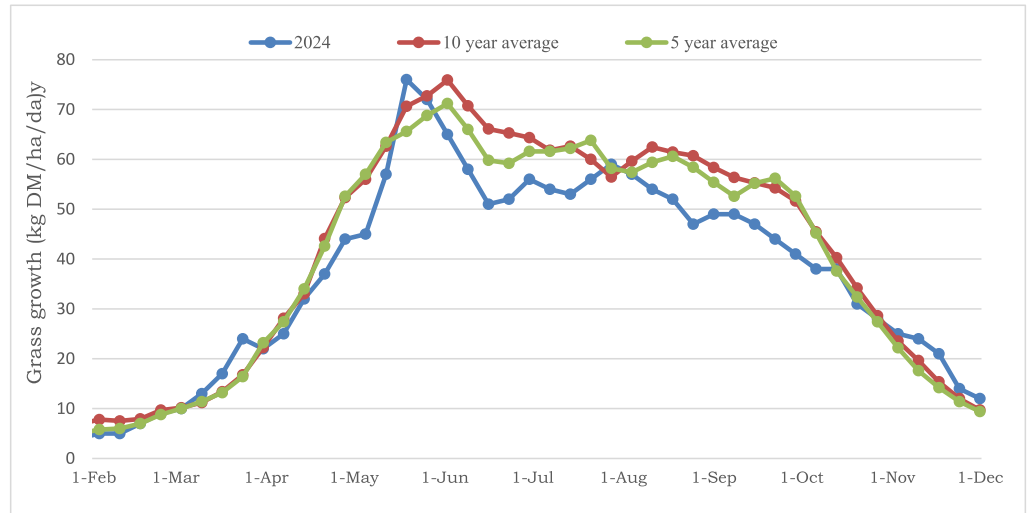


Figure 1. Weekly grass growth on PastureBase Ireland grassland farms (2014–2024).

in DM production ($\pm 907 \text{ kg DM ha}^{-1}$) between years. Spring pasture DM production had greater variation compared with summer and autumn (Table 1).

On average, over the 11 years, spring (Jan–April) pasture DM production was $1.8 (\pm 0.3) \text{ t DM ha}^{-1}$, summer (May–July) DM production was $6.2 (\pm 0.5) \text{ t DM ha}^{-1}$, and autumn (Aug–Dec) DM production was $5.3 (\pm 0.3) \text{ t DM ha}^{-1}$. The average number of days at grass ranged from 274 to 296 days between years, with an overall mean of 285 grazing days. Mean pre-grazing herbage mass declined from $1611 \text{ kg DM ha}^{-1}$ in 2014 to $1474 \text{ kg DM ha}^{-1}$ in 2023, which is in line with recommendations (O'Donovan et al., 2022) and was associated with an increase in the number of defoliation events per year (7.1 to 8.1). Stocking rates have increased over the 11-year period; the average stocking rate on the milking platform has increased from $2.8 \text{ livestock units (LU) ha}^{-1}$ in 2014 to 3.0 LU ha^{-1} in 2024. The level of pasture allocation to the grazing herd has remained constant, at approximately $3.7 \text{ t DM cow}^{-1}$. Supplementation of cows with both conserved forage and concentrate increased over the 11-year period. During the first four years of data collection, supplementation with conserved forage and concentrate averaged $290 \text{ kg DM cow}^{-1}$ and 520 kg cow^{-1} , respectively. During the last four years

of data collection, these figures had increased to 550 kg DM cow⁻¹ and 959 kg cow⁻¹, respectively. The increase in supplementation during the grazing period is due to reductions in cumulative herbage production (particularly during the last three years), a significant decline in chemical N input and challenging climatic conditions including dry summers and wet springs.

Conclusions

Within the dataset used from PBI, annual pasture growth averaged 13.2 t DM ha⁻¹ on Irish grassland farms from 2014 to 2024, with 7% variation in DM production (+/-907 kg DM ha⁻¹) between years. The average number of days at grass ranged from 274 to 296 days between years, with an overall mean of 285 grazing days. The future of grassland systems will depend on their ability to increase and adapt grassland production within the constraints of reducing chemical N fertiliser inputs and adapting to challenging climatic conditions. Conserving adequate feed levels for the winter period is a growing challenge for farmers in recent years due to variations in grass growth. Using available grassland technologies and achieving grazing management targets will be critical to meeting future grassland challenges. PBI will in the future, develop as a European grassland database and decision support system.

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The effect of sward species diversity on enteric methane emissions from spring calving dairy cows

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Abstract

The current experiment compared the enteric methane emissions (CH₄) of spring calving dairy cows in a perennial ryegrass system receiving 250 kg N ha⁻¹ year⁻¹ (PRG), a perennial ryegrass with white clover system receiving 125 kg N ha⁻¹ year⁻¹ (PRGWC) and an 8 species (perennial ryegrass, timothy, meadow fescue, white clover, red clover, alsike clover, plantain and chicory) sward system receiving 125 kg N ha⁻¹ year⁻¹ (MSS). The sward systems were rotationally grazed and managed identically. Dry matter intake (DMI) increased with increasing sward diversity ($P < 0.01$) resulting in greater CH₄ emissions for cows grazing PRGWC compared with PRG ($P < 0.01$). However, CH₄ per kg of DMI was less for cows grazing MSS, therefore they had less CH₄ emissions than the cows grazing PRGWC ($P < 0.01$) and similar emissions to those grazing PRG despite having greater DMI. On a milk solids basis MSS also had less CH₄ emissions compared with PRG and PRGWC ($P < 0.01$).

Introduction

Conventionally, pasture-based dairy systems in north-western Europe have relied on simple monoculture swards of productive grass species such as *Lolium perenne* (perennial ryegrass; PRG). Such swards are capable of high levels of productivity and nutritional value although rely heavily on artificial nitrogen (N) fertiliser. Nitrogen fertiliser use contributes to nitrous oxide emissions and N losses through nitrate leaching. The addition of legumes such as *Trifolium repens* (white clover; WC) to PRG swards can maintain similar levels of productivity to PRG-only swards at a reduced artificial N input through fixation of atmospheric N (Murray *et al.*, 2023). Studies investigating further sward species diversification, particularly through the addition of 'herb' species such as *Plantago lanceolata* (plantain) and *Cichorium intybus* (chicory), have noted a series of potential benefits pertaining to nitrate leaching (Navarrete *et al.*, 2018) and resilience to drought (Grange *et al.*, 2021). However, another major environmental concern in relation to pasture-based dairy systems is enteric methane (CH₄). Within the literature, data on CH₄ emissions produced by different sward types are inconsistent (Dwan *et al.*, 2025; Enriquez-Hidalgo *et al.*, 2014; Jonker *et al.*, 2019; Loza *et al.*, 2021). These inconsistencies may be due to differences in the duration of these experiments. Therefore, in order to determine a more conclusive outcome, the current study investigated the enteric CH₄ emissions of dairy cows grazing PRG, PRGWC and multispecies (MSS) sward systems over a prolonged measurement period.

Materials and methods

The current experiment was undertaken in conjunction with a larger multi-year farm systems study described by (Jezequel *et al.*, 2024). The study consisted of 108 spring calving Holstein-Friesian and Holstein-Friesian × Jersey dairy cows grazing three sward systems: PRG receiving 250 kg N ha⁻¹

Table 1. Effect of sward system on mean daily animal performance and methane emissions (CH_4) from cows grazing perennial ryegrass receiving $250 \text{ kg N ha}^{-1} \text{ year}^{-1}$ (PRG), perennial ryegrass with white clover receiving $125 \text{ kg N ha}^{-1} \text{ year}^{-1}$ (PRGWC) or a multispecies sward receiving $125 \text{ kg N ha}^{-1} \text{ year}^{-1}$ (MSS).

	PRG	PRGWC	MSS	SE	P-value
Milk yield (kg)	15.3 ^a	15.9 ^{ab}	16.6 ^b	0.41	<0.01
Fat (g kg^{-1})	53.9	53.4	52.0	0.85	NS
Protein (g kg^{-1})	39.8	38.9	39.3	0.44	NS
Milk solids (kg)	1.40 ^a	1.45 ^{ab}	1.49 ^b	0.022	<0.01
DMI (kg)	17.1 ^{ax}	18.2 ^{ay}	20.3 ^{bz}	0.42	<0.01
CH_4 (g)	294 ^a	310 ^b	297 ^a	3.8	<0.01
CH_4 g (kg milk solids) ⁻¹	217 ^a	219 ^a	208 ^b	2.9	<0.01
CH_4 g (kg DMI) ⁻¹	17.6 ^a	17.7 ^a	15.4 ^b	0.37	<0.001

DMI, dry matter intake. ^{a,b}Values with not sharing a common superscript are significantly different ($P < 0.05$; Tukey–Kramer adjusted). ^{x,y,z}Values not sharing a common superscript tend to be different ($P < 0.1$; Tukey–Kramer adjusted).

year^{-1} , PRGWC receiving $125 \text{ kg N ha}^{-1} \text{ year}^{-1}$ and MSS receiving $125 \text{ kg N ha}^{-1} \text{ year}^{-1}$. The MSS was sown as a mix of PRG, *Phleum pratense* (timothy), *Festuca pratensis* (meadow fescue), WC, *Trifolium pratense* (red clover), *Trifolium hybridum* (alsike clover), plantain and chicory. The cows assigned to each sward system were balanced for breed, parity, calving date, milk production and bodyweight. The cows were managed in a rotational grazing system with a target post grazing sward height of 4 to 4.5 cm. They were milked in a herringbone milking parlour twice daily, at 07:30 and 15:30, with yield measured daily and composition (fat and protein) measured weekly. Dry matter intake was estimated in July and October using the Ytterbium Oxide technique as described by (Pérez-Ramírez *et al.*, 2012). Methane measurement was carried out using three Greenfeed units (C-Lock). After a one-month training period starting in mid-April, 33, 28 and 36 cows were regular Greenfeed users in PRG, PRGWC and MSS respectively. The data was averaged over 11 consecutive two-week measurement periods from May to October. Statistical analysis was undertaken using the mixed procedure in SAS with individual cow as a random effect and sward system, parity, breed, calving day of the year, period and period by sward system interaction as fixed effects. A first-order autoregressive covariance structure was applied with period as a repeated measure. Multiple comparisons were adjusted using the Tukey–Kramer method.

Results and discussion

Mean daily visits to the Greenfeed units were similar across sward systems ($2.1 \text{ visits cow}^{-1} \text{ day}^{-1}$) and daily concentrate offered was $0.87 \text{ kg DM cow}^{-1} \text{ day}^{-1}$. Daily milk yield and milk solids yield (milk fat+protein) was greater for the cows grazing MSS compared with PRG ($P < 0.01$) while those grazing PRGWC were intermediate but not statistically different to the other two sward systems (Table 1). The cows grazing MSS had greater DMI than cows grazing the other two sward systems ($P < 0.01$) and the cows grazing PRGWC tended to consume more than those grazing PRG ($P = 0.07$). The cows grazing PRG and MSS had similar CH_4 emissions but less emissions compared to those grazing PRGWC ($P < 0.01$). The cows grazing MSS produced less CH_4 per kg of milk solids compared with the cows grazing PRG and PRGWC ($P < 0.01$). There was no difference in CH_4 per kg of DMI (CH_4 yield) between the cows grazing PRG and PRGWC while the cows grazing MSS had lower CH_4 yield compared with the other two sward systems ($P < 0.001$).

Increased CH₄ from grazing PRGWC swards is due to greater DMI compared with PRG. However, this is not the case with MSS, which increased DMI but reduced methane yield. This is likely related to the inclusion of plantain and chicory as these differentiate MSS from PRGWC. Ultimately, grazing MSS reduced the emission intensity of milk solids production and therefore may have potential to reduce the carbon footprint of milk production from grazing dairy systems. However further research is required on the longevity of plantain and chicory in grazed swards as poor persistency would limit their long-term methane mitigation potential.

Conclusion

This study found that PRGWC increased CH₄ emissions from grazing dairy cows compared with PRG swards, although PRGWC did not affect CH₄ on a milk solids or DMI basis. Cows grazing MSS had similar emissions to PRG but produced less CH₄ per kg of milk solids and per kg of DMI compared with cow grazing PRG and PRGWC.

Acknowledgements

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The hidden side of dairy product quality: forage quality analysed through multiple indices

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Abstract

Dairy labels, such as the PDO (Protected Designation of Origin), set criteria on grazing such as the minimum duration per year. Nevertheless, there are no requirements regarding the fodder quality even if it impacts the quality of the milk and can modulate animal health. Forage quality and floristic composition were analysed in 30 permanent grasslands presenting a wide range of Land-Use Intensity (LUI) within farms producing milk for PDO Normandy cheeses. Fodder quality was characterized by Crude Protein content (CP) and pepsin cellulase digestibility (dC), but also by analyses of Non-Structural Carbohydrates (NSC; starch, fructans, sucrose, glucose, fructose, pinitol) and total phenolics. All parameters appeared to vary among grasslands. Forage CP content and dC were positively correlated with LUI. Fodder quality was also linked to the floristic composition as revealed by the positive link between dC and CP and legume content. NSC and total phenolic contents were uncorrelated with classical forage quality indices and therefore provide essential additional information. Grassland management, which influences plant community composition and thus the biochemical quality of forage, should be considered in PDO certification.

Keywords: floristic composition, fodder quality, grasslands, non-structural carbohydrates, phenolics

Introduction

While dairy labels such as PDO (Protected Designation of Origin) specifications include requirements on grazing practices there are no requirements regarding the quality of grassland fodder. However, forage quality can influence both animal health and the taste of milk. Forage quality is generally characterized by crude protein content (CP) and pepsin cellulase digestibility (dC), which are good indicators of nutritional value but do not take into account the quality of energy compounds. Non-structural carbohydrates (NSC), which include soluble carbohydrates (mainly fructans, sucrose, fructose and glucose) and starch, are a rapidly usable energy source that improves rumen health and boosts milk production (Klevenhusen & Zebeli, 2021). Fructans, depending on their type and thus the plant species, can stimulate the growth of rumen beneficial bacteria (King *et al.*, 2024). Forages may also contain large amounts of polyols (mainly pinitol) which contribute not only to their energetic value (Lowry & Kennedy, 1995) but also to animal health through their antioxidant and anti-inflammatory properties (Pandi *et al.*, 2022). Then, phenolic compounds can improve milk yield and flavour as well as animal health (O'Connell & Fox, 2001). We evaluated the content of NCS, starch, pinitol and total phenolic compounds in forage from grasslands involved in Normandy dairy PDOs, in order to assess the relevance of their consideration when analysing forage quality. We then sought to identify the factors, among LUI and floristic composition, determining the forage biochemical composition.

Materials and methods

This study was conducted on permanent grasslands (at least 5 years old) grazed by dairy cows in 30 farms involved in a PDO Normandy cheeses (Camembert de Normandie, Livarot, Pont-l'Évêque and

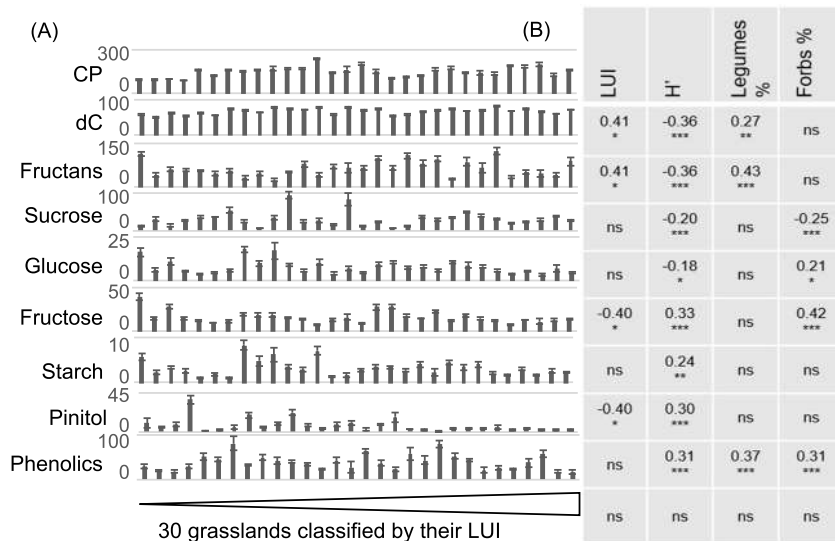


Figure 1. Forage quality indices (CP, crude protein in g kg^{-1} ; dC, pepsin cellulase digestibility in %, fructans, sucrose, glucose, fructose, starch, pinitol, and phenolics contents in $\text{mg (g dry matter)}^{-1}$) of the 30 grasslands (A) and their correlations with LUI (land use intensity; $n = 30$), H' (Shannon–Weaver diversity index) and the % of legumes and forbs in the plant community ($n = 120$) (B). Pearson's r and P -value threshold: * $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$; ns, not significant.

Neufchâtel). For each grassland, a land use intensity (LUI) index was calculated on the basis of grazing intensity, fertilization and mowing frequency (Blüthgen *et al.*, 2012).

Fodder samplings (at least 100 g of dry matter harvested 5 cm above the ground on at least $5 \times 0.4 \text{ m}^2$ quadrats) and vegetation surveys (species cover in four 1 m^2 quadrats) were carried out in June and July 2024 on a $10 \times 10 \text{ m}$ plot located in a floristically homogeneous area of each of the 30 grasslands. CP and dC were analysed by an independent laboratory (LANO) with Kjeldahl method (ISO 5983) for CP and according to Aufrère *et al.*, (2007) for dC. For water-soluble carbohydrates (WSC), pinitol and total phenolic compound analyses, hot water-soluble extracts were obtained by a succession of three water extractions. For WSC and pinitol analyses, extracts were analysed by HPLC with a refractometric detection using a Sugar-Pak column (Waters, Mildford, MA, USA) for sucrose, fructose and glucose + pinitol, and a Carbomix column (Sepax Technologies, Newark, USA) for fructans. Glucose content was determined by glucose oxidase—peroxidase kit (GOPOD reagent, Megazyme, Wicklow, Ireland) and the value was subtracted to the 'glucose + pinitol' concentration obtained by HPLC to determine pinitol concentration. Starch was determined using the Total Starch Assay kit (Megazyme). Polyphenols were measured using the Folin–Ciocalteu reagent using gallic acid as standard (Boizot & Charpentier, 2006). Statistical analyses were performed using "R" (version 4.4.1). Pearson's correlation tests were used to detect significant correlations among NSC, starch, pinitol and total phenolic compounds with LUI ($n = 30$), floristic diversity H' , percentage of legumes and forbs ($n = 120$).

Results and discussion

The two classical quality indices CP and dC increased with LUI (Fig. 1), and were negatively linked with floristic diversity (Fig. 1B).

Digestibility and CP are known to be improved by management intensity (Gardarin *et al.*, 2014; Berauer *et al.*, 2020). In contrast, most NSC contents were unrelated to LUI, with the exception of glucose and starch, present in small quantities, whose levels decreased as LUI increased (Fig. 1B). NSC contents were strongly related to H', either positively (glucose, fructose and starch) or negatively (fructans and sucrose). These relationships are at least partially linked to the floristic composition and in particular to the proportion of forbs, which appeared to be negatively linked to fructan contents and positively linked to sucrose and glucose contents. The variability of pinitol and phenolic compound contents among the 30 grasslands appeared very high (Fig. 1A). Phenolic content, unrelated to LUI, H' and functional group proportion (Fig. 1 B), appeared strongly linked to one plant species relative cover, *Agrostis stolonifera* (data not shown, $r = 0.40$, $P < 0.05$).

Conclusions

NSC and total phenolics contents are not correlated with conventional energy value indices, therefore they provide a better assessment of forage quality. As they are well known to influence animal health (O'Connell & Fox, 2001; Pandi *et al.*, 2022) they are valuable complementary indices of forage quality. The relationship between floristic composition, particularly *A. stolonifera* cover, and indices such as phenolic compound content, invites us to explore quality at the level of different plant species. Since grassland management influences plant community composition and thereby the biochemical quality of forage, it should be considered in PDO certification, given its impact on milk quality and the likely repercussions on cheese characteristics.

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Plant species identity affects earthworm abundance in managed grasslands

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Abstract

Multispecies swards can match the productivity of perennial ryegrass monocultures in managed grasslands under reduced levels of nitrogen fertilisation. However, the effects of multispecies leys on soil life—another important aspect of sustainable agriculture—are not well understood. In this study we quantified the effect of plant diversity on the abundance of earthworms in the soil. We established a field experiment on a sandy soil in The Netherlands. Grassland communities were composed of systematically varying proportions of one up to six species according to a simplex design. The plant species represented three functional groups (FGs): grasses (*Lolium perenne*, *Festuca arundinacea*), legumes (*Trifolium pratense*, *Trifolium repens*) and herbs (*Cichorium intybus*, *Plantago lanceolata*). Swards received a nitrogen (N) application rate of 150 kg N ha⁻¹ year⁻¹; additional perennial ryegrass monoculture plots received 300 kg N ha⁻¹ year⁻¹ (Lp300). We measured earthworm abundance at the end of a three-year grassland period. We did not find grassland species diversity effects on earthworm abundance. Mineral fertiliser increased the number of earthworms in *L. perenne* monocultures, but the *T. pratense*, *T. repens* and *P. lanceolata* monocultures did not differ significantly from Lp300.

Keywords: earthworms, herbs, LegumeLegacy, legumes, multispecies leys, nitrogen

Introduction

Beef and dairy farmers are challenged by the need to reduce fertiliser inputs to prevent excess nutrient losses to the environment. One measure could be to diversify grasslands and include nitrogen (N) fixing legumes so that fewer external inputs are necessary. Multispecies swards can outperform perennial ryegrass monocultures in managed grasslands in terms of productivity (Baker *et al.*, 2022). However, the effects of multispecies leys on soil life—another important aspect of sustainable agriculture—are not well understood. Here, we studied the effect of plant species identity and interspecific interactions on earthworm abundance. Earthworms are seen as ecosystem engineers and are known to improve soil structure and recycle nutrients. We conducted a three-year field experiment and used Diversity-Interactions (DI) models to study the plant species identity effect and the potential interspecific interactions. Additionally, we compared a *Lolium perenne* (Lp) monoculture that received 150 kg N ha⁻¹ year⁻¹ and an equi-proportional six-species multispecies sward to a reference Lp monoculture that was fertilised at a rate of 300 kg N ha⁻¹ year⁻¹ of nitrogen fertiliser.

Materials and methods

A three-year field experiment was set up in The Netherlands (51°37′1.6″N 5°00′18.3″E) on a sandy soil (1% clay, 10% silt, 86% sand, 2.7% OM, pH 4.8) in October 2019. Thirty-four grassland communities were randomly assigned to one of 52 plots. The grassland communities were randomly

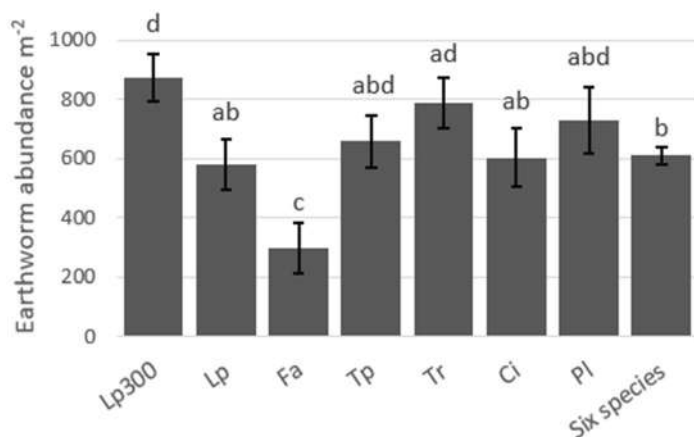


Figure 1. Estimated earthworm abundance m^{-2} . Lp300 = *L. perenne* at $300 \text{ kg N ha}^{-1} \text{ year}^{-1}$, Lp = *L. perenne* at $150 \text{ kg N ha}^{-1} \text{ year}^{-1}$, Fa = *F. arundinacea*, Tp = *T. pratense*, Tr = *T. repens*, Ci = *C. intybus*, PI = *P. lanceolata*, Six species = equi-proportional Lp-Fa-Tp-Tr-Ci-PI mixture. Error bars represent standard errors. Different letters indicate a significant difference between communities ($P < 0.05$).

assigned to plots of $4 \text{ m} \times 10 \text{ m}$. The communities were composed of systematically varying proportions of one up to six species according to a simplex design. Species belonged to three different functional groups, i.e. grasses (*Lolium perenne* = Lp, *Festuca arundinacea* = Fa), legumes (*Trifolium pratense* = Tp, *Trifolium repens* = Tr) and herbs (*Cichorium intybus* = Ci, *Plantago lanceolata* = PI) (Moral *et al.*, 2023). Species proportions of the multispecies communities were based on the seeding rate in monoculture for a species with proportion is 1. All plots received $150 \text{ kg N ha}^{-1} \text{ year}^{-1}$. In addition, an extra perennial ryegrass monoculture community, receiving $300 \text{ kg N ha}^{-1} \text{ year}^{-1}$ (Lp300, $n = 5$), was included as the reference monoculture system for beef and dairy farming in The Netherlands. In April 2023, in each plot, two blocks of dimensions $20 \times 20 \times 20 \text{ cm}$ were dug out. Earthworms in the soil blocks were carefully counted and determined to species level. Diversity-Interactions (DI) models were used to jointly predict the effect of species richness, species proportion and species identity on earthworm abundance by using the DImodels package (version 1.3.3.9001) in RStudio (version 4.4.0). The basis of a DI model is the following linear regression model: $y = \text{Species identities} + \text{Species interactions} + \text{Error}$ (Moral *et al.*, 2023). Significant differences were studied using the `compare_communities` function in the DImodels packages in RStudio.

Results and discussion

Earthworm species richness was low in all plots, ranging from two to four species, which was expected on this sandy soil with an arable history. The most abundant species was *Lumbricus rubellus*, which was unexpected since this is an epigeic species, whereas endogeic species were expected to be the most abundant (van Eekeren *et al.*, 2014). Earthworm abundance was strongly influenced by plant species identity, but there were no significant effects of interspecific interactions. This means that the equi-proportional six-species mixture performed as would be expected based on the weighted average of the constituent species.

There was a significant difference between the Lp monocultures, with a higher earthworm abundance in the Lp300 community compared to the Lp monoculture that received $150 \text{ kg N ha}^{-1} \text{ year}^{-1}$ (Fig. 1), suggesting that an increase in mineral fertiliser results in more earthworms in the soil in Lp monocultures. Earthworms are known to do well in nitrogen-rich environments (van Vliet *et al.*,

2007). This was also reflected in the high earthworm abundance in the clover monocultures (specifically in Tr), which are known to enrich their surroundings with nitrogen fixed from the atmosphere, ultimately creating nitrogen-rich soil environments. It was unexpected that Pl also showed a similar earthworm abundance as the Tp community (Fig. 1), since previous studies found that earthworm biomass and survival was lower in microcosms with Pl compared to those containing Tp due to lower rhizodeposition of Pl (Eisenhauer et al., 2009). Lowest earthworm abundance was found in the monoculture of tall fescue (Fa) (Fig. 1). Earthworms prefer a lower C/N ratio of their food—depending on the ecological group, this varies between 8.8 and 12.4 (van Vliet et al., 2007). Although we did not measure the C/N ratio of the above-ground biomass in our experiment, it is reasonable to argue that Fa is harder to decompose for earthworms due to the generally lower digestibility of Fa compared to Lp. Additionally, Fa has thicker roots which could be less preferred by earthworms (Deru *et al.*, 2012).

Conclusion

Earthworm abundance was influenced by the plant species identity but not by interspecific interactions between different plant species. Thus, mixtures did not perform better than what was expected based on the weighted average of the identity effects of the constituent plant species. The lowest earthworm abundance was found in Fa monocultures. Mineral fertiliser increased the number of earthworms in Lp monocultures, but the Tp, Tr and Pl monocultures did not differ significantly from Lp300. Here, we only highlighted one aspect of healthy soils in terms of soil life but it is important to take more aspects into account to make overarching conclusions on the impact of grassland species on soil life in managed systems.

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Valuable veal: rethinking dairy calf rearing through participatory research in Walloon grass-based dairy systems

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Abstract

The low market value of dairy calves, particularly males, but also surplus females, remains a major challenge for specialized dairy farms, raising concerns about animal welfare or antibiotic use. Alternatives that integrate meat production within dairy systems are still poorly explored. To address this gap, a group of ten dairy farmers formed an operational group (OG) to investigate innovative and viable rearing pathways for dairy calves. These pathways range from early fattened calves to steers or heifers. All systems emphasize grass-based feeding, using cows' milk and pasture to reduce costs and enhance meat quality for higher market value. To define the OG's programme, semi-structured interviews were conducted to identify farmers' research priorities and practical concerns, which will guide participatory on-farm experiments in the next phase. The main issues identified by the members of the OG include feeding strategies, optimal slaughter age, and consumer acceptance. This contribution presents the composition of the OG and outlines their topics of interest. It highlights the need for context-specific production pathways that combine economic viability with improved animal welfare and environmental performance.

Keywords: beef from dairy, crossing, dairy calves, veal production

Introduction

Selective breeding for high milk production has reduced meat traits in dairy calves, resulting in low market value particularly for males and surplus females, raising animal welfare concerns, as some calves are euthanized at birth or receive suboptimal care (Haskell, 2020). In Wallonia, 90% of male calves are sent off-farm before 30 days to intensive white veal systems, which are criticised for restricting natural behaviours, providing nutritionally limited diets, and prompting routine antibiotic use (EFSA, 2023). Alternatives exist, including rosé veal production with grass- and grain-based feeding, or rearing calves to maturity as bulls or steers. However, these approaches raise important questions regarding animal welfare, economic viability, workload, and what is practically feasible for farmers (Hopkinson, 2017). In this paper, we describe the farm characteristics and the main topics of interest of the operational group "GO Veaux" focused on the valorization of dairy calves.

Materials and methods

In March 2025, field visits were conducted on each of the Walloon dairy farmers that had expressed interest in joining the GO Veaux operational group during its construction phase (April–August 2024). Farmers were informed about the project through articles in the specialised press and by direct contact via a snowball effect. Selection was based on farmers' motivation and the requirement that they were already engaged in rearing dairy calves. Semi-structured interviews were carried out with the ten selected farmers to document their current practices for valorizing dairy calves and to identify their main topics of interest, with the aim of proposing on-farm trials in the second phase of the project within a participatory research framework (Balta and Fourest, 2022). Interviews were

Table 1. Description of the participating farmers.

Farm	A	B	C	D	E	F	G	H	I	J
Type of production	Conv	Org	Org	Org	Org	Org	Org	Org	Conv	Conv
Type of herd(s)	Mix., meat	Dairy	Dairy, meat	Mix.	Mix., meat	Dairy, mix.	Dairy	Mixed	Dairy, meat	Dairy, meat
Dairy cows breed	Norm	Hol	Hol, Jer, BS	Fle	Jer, Fle	Hol, Fle	Hol	Fle, Hol, Mt	Jer	Hol
Dairy cows (n)	50	85	50	140	40	50	95	20	20	100
Milk prod.	4000	7000	7500	5500	4000	8500	7500	4000	5000	9500
Milk transf.	No	Yes	No	No	Yes	Yes	No	Yes	Yes	No
Crossing and sex	xBB-M	xMt-M	No-M/F	xBB-M	xBB/Lim-M	xBB-M	xBB-M/F	No-M	No-M	xWag-M/F
Calves (n/year)	All	All	9	All	All	3	10	All	All	10
Milk feed	Dis. milk	With cows	With cows	With cows	With cows	Dis. milk	With cows	With cows	Skim. milk	Pow. milk
Castration	No	No	No	No	No	No	No	No	Yes (M)	Yes (M)
Age (months)	18–24	4–14	24–36	1	4–5	8	4	5–8	15–24	30–36
Destination	Merchant	Direct sale	Direct sale	Fattening	Direct sale	Direct sale	Fattening	Direct sale	Direct sale	Direct sale

Conv, Conventional; Org, organic; Mix., mixed; Norm, Normande; Hol, Holstein; Jer, Jersey; Fle, Fleckvieh; BS, Brown Suisse; Mt, Montbéliarde; BB, Belgian Blue; Lim, Limousin; Wag, Wagyu; Milk prod., Milk production (kg cow⁻¹ at 305 days); Milk transf., Milk transformation; Dis. milk, Discarded milk; Skim. milk, Skimmed milk; Pow. milk, Powder milk.

transcribed with the support of Whisper (OpenAI) and analysed through thematic content analysis.

Results and discussion

Participating farmers showed considerable diversity in their approaches to valorizing dairy calves, including breed crossing, sex, milk feeding, and castration practices, as well as differences in overall production systems (Table 1). Notably, seven of the ten farms were organic, and eight raised either mixed breeds or both dairy and meat breeds. In some cases, calves were finished off-farm; however, eight farmers reared their animals through to slaughter, with seven of them marketing the meat directly.

Fifteen main topics were identified from the analysis of the interviews (Fig. 1). In farmers' statements, each theme was often connected to one or more other themes. For example, one farmer asked: "What is the relationship between the animal's slaughter age and the taste of the meat?", which is a question linking two themes. The hypothetical answers suggested by farmers frequently led to further questions. For example: "How would potential changes in feed and slaughter age affect the production costs on my farm?"

Conclusion

The diversity of contexts and practices currently employed by farmers to valorize surplus dairy offspring supports the relevance of an operational group (OG) dynamic for research and development. This configuration enables farmers and researchers to learn collaboratively in real-world, contextualized settings. Fourteen main topics of interest were identified by OG members, with a particular focus on optimal slaughter age, feeding strategies at different stages of calf development, and consumer perception of meat quality. In the next phase of the project, farmers will conduct individual on-farm trials and share their experiences, while researchers will provide support to enhance practices.

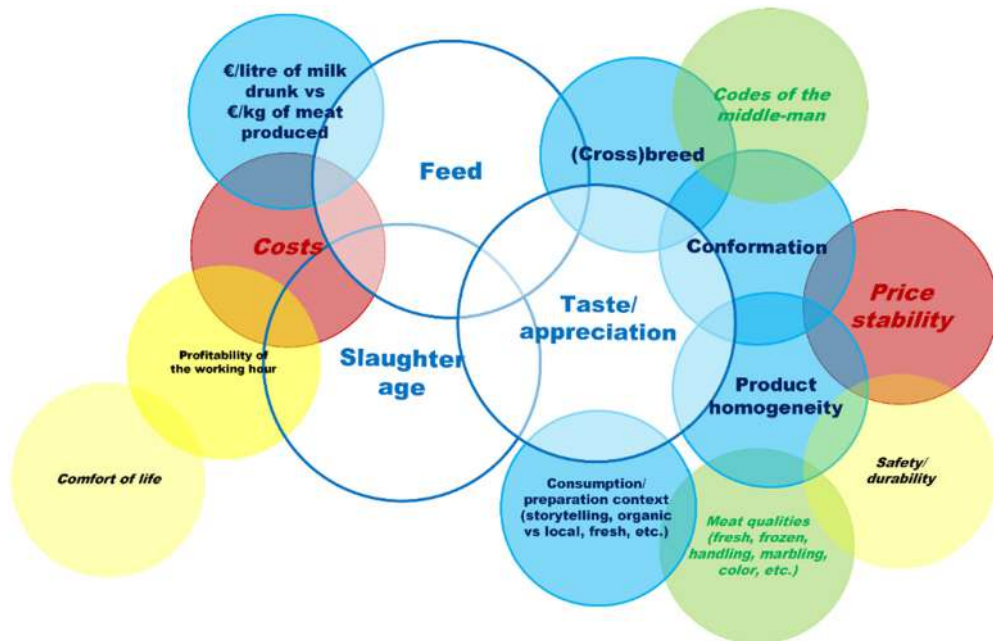


Figure 1. Summary diagram of the shared interests of the OG members regarding trials to be carried out. From the outside to the centre of the diagram: from the most peripheral to the most prominent; From left to right: from production costs (inputs) to profits from livestock farming (outputs); In the blue circles: the three most salient topics; In the blue and red discs: the seven common topics; In the green discs: the two frequently mentioned topics; In the yellow discs: the three topics extrapolated from farmers' global statements.

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The role of ruminant livestock in a sustainable food system: a Mediterranean perspective

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Abstract

Based on the role of ruminants in a sustainable food system, processing human inedible feed, transforming it into food for human consumption and nutrients for plant production, but with environmental trade-offs, a set of criteria for sustainable ruminant production in the Mediterranean is developed: (1) in terms of land use, livestock feed should not be produced on land that can be used to produce food directly for human consumption or that could be better used for nature conservation or carbon sequestration; (2) in terms of nutrient cycling, legumes should be used, due to their capacity for nitrogen fixation, and the capacity of animal production systems to provide nutrients for plant production should be maximized; (3) in terms of climate and biodiversity, livestock production should be associated with adequately managed forestry, ensuring forest regeneration and carbon sequestration and reducing fuel load; (4) in terms of soil, soil tillage should be avoided, thus leading to an increase in soil organic matter and the corresponding carbon sequestration. As a system fulfilling these criteria, in the case of beef, it is proposed to combine grazing the suckling herd on sown biodiverse legume rich permanent pastures in *Montado* agro-forestry with optimised intensive fattening.

Keywords: agroforestry, biodiversity, carbon sequestration, nutrient cycling, ruminants, sustainability

Introduction

It is not yet clear what the full set of global mitigation options is that could align agriculture and the agri-food system with the Paris Agreement (Ivanovich *et al.*, 2023; Springmann *et al.*, 2018; Zurek *et al.*, 2022). Within agriculture, the livestock production sub-sector is, by itself, a significant threat to maintaining human activity within ecological limits (planetary boundaries) (Li *et al.*, 2024), with GHG emissions of 5.6–7.5 GtCO₂e year⁻¹, of which ruminants account for 64% to 78% (Herrero *et al.*, 2016). However, it is a sector of great nutritional, economic, and social importance, with a total global economic market value of animal production between 1.61 and 3.3 trillion USD, compared to 2.57 trillion USD for crop production (Schrobbach *et al.*, 2023), and employing 1.3 billion people in production and retail (Herrero *et al.*, 2016). Ruminant production represents 65.8% of this economic value, of which 56.1% corresponds to cattle (Schrobbach *et al.*, 2023).

The foundation of ruminant meat production (and of small ruminants for milk) is pastureland (Greenwood, 2021), which covers 40.5% of the Earth's land area excluding Greenland and Antarctica (Carrascosa *et al.*, 2024). Pastures provide multiple ecosystem services (e.g., regulation of hydrological and nutrient cycles, carbon sequestration, animal/human health) (Schils *et al.*, 2022; D'Ottavio *et al.*, 2018), are habitats of high biodiversity and natural value, and represent about one-third of the global terrestrial biomass carbon reservoir (Bai & Cotrufo, 2022). Yet their role in sustainability is often overlooked (Poux & Aubert, 2022). Climate mitigation scenarios commonly propose reducing both ruminant numbers and pasture areas in favor of reforestation (Duru *et al.*, 2021), even though,

paradoxically, strategies that also value biodiversity often call for maintaining pasturelands (Pörtner *et al.*, 2021).

There are currently no global, European, or national certifications for animal production that ensure the absence of competition with direct human food production, carbon sequestration, and nature conservation, while maximizing circularity opportunities and reducing greenhouse gas emissions. Here, we work towards developing a certification meeting these requirements, focused on ruminant production in the Mediterranean, establishing its conceptual basis and the sustainability criteria to be met.

Conceptual basis

The key role of ruminants in a sustainable agri-food system arises from their unique digestive system that lets them convert cellulose-rich feeds—like rough forages and agri-food by-products, inedible for humans—into meat, milk, and manure. Thus, the common belief that ruminants are less efficient than monogastric animals is misleading (Herrero *et al.*, 2013).

Ruminants should be fed in ways that minimize competition with food for direct human consumption—both in feed and land use. Studies estimate that up to 35% of global protein needs could be met by animals fed only on non-competing feed sources (Van Zanten *et al.*, 2018). Pastures should also avoid occupying land better suited for carbon sequestration or nature conservation (Hayek *et al.*, 2024; Searchinger *et al.*, 2023).

Ruminants' unique digestive system produces methane. Their climate impact can be reduced not only by lowering methane emissions but also by enhancing carbon sequestration—through improved pasture management, wildfire prevention via grazing, and better tree health in silvopastoral systems, provided forest regeneration is maintained.

Carbon and nitrogen cycles must be addressed together. Today's agri-food system depends heavily on synthetic nitrogen from the Haber-Bosch process. Legumes are the only natural alternative, fixing nitrogen through symbiosis with rhizobia. While edible legumes should be promoted on fertile land, less fertile areas are better suited for forage legumes—ideally used in grazing systems—which also offer higher nitrogen fixation potential. By feeding on forage legumes, ruminants help transfer biologically fixed nitrogen into the food system—via meat, milk, and manure for crops (Poux & Aubert, 2022). The Mediterranean landscape faces unique challenges: poor, degraded soils (Carmo & Domingos, 2021), mismatches between rainfall and plant needs, and high fire risk—exacerbated by climate change. In this context, integrating pastoralism with forest management is crucial. Well-managed grazing helps reduce fire-prone vegetation, while continuous forest cover must be avoided to manage fire risk, water availability, and aquifer recharge.

Proposed sustainability criteria

Based on the preceding analysis, we propose a set of criteria for sustainable ruminant production in the Mediterranean:

(1) in terms of land use, livestock feed should not be produced on land that can be used to produce food directly for human consumption or that could be better used for nature conservation or carbon

sequestration; (2) in terms of nutrient cycling, legumes should be used, due to their capacity for nitrogen fixation, and the capacity of animal production systems to provide nutrients for plant production should be maximized; (3) in terms of climate and biodiversity, livestock production should be associated with adequately managed forestry, ensuring forest regeneration and carbon sequestration and reducing fuel load; (4) in terms of soil, soil tillage should be avoided, thus leading to an increase in soil organic matter and the corresponding carbon sequestration.

These criteria are specific to the Mediterranean area due to their focus on issues which are particularly relevant for the Mediterranean: potential for the use of biodiverse legume mixtures in pastures, wildfire risk, soil organic matter loss due to excessive soil tillage, under unfavourable edaphoclimatic conditions.

As a system fulfilling these criteria, in the case of beef, it is proposed to combine the grazing of the suckling herd on sown biodiverse legume rich permanent pastures in Montado agro-forestry with optimised intensive fattening.

Acknowledgements

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Integrating dual-purpose with single-purpose crops for forage and grain production in Sweden

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Abstract

There is increasing interest in enhancing the integration of livestock and crop production within Europe's agri-food systems. Dual-purpose crops exploit the natural plasticity and phenology from one establishment to produce both forage and grain. The practical feasibility in northern European cropping systems may be constrained by environmental conditions and high land-use intensity. However, integrating dual-purpose crops with single-purpose crops could enhance the possibilities of forage and grain production within a crop sequence. Field experiments exploring different two-year crop sequences are ongoing at four sites across Sweden. The treatments include combinations of spring and winter annual cereals with forage crops e.g. spring barley, winter wheat, rye, annual and perennial ryegrass, maize, clover and lucerne. The dual-purpose crop combinations are compared to a traditional crop production system of cereals harvested for grains. Four treatments are tested per site in a randomized complete block design. The evaluation includes phenological development, agronomic performance and feed quality. Initial observations from the first crop cycle indicates that inherent phenology and plasticity of the annual and the winter annual crops, enables the utilisation of dual-purpose cropping systems across a north-south gradient in Sweden.

Keywords: boreal agriculture, dual-purpose crops, forage, grain, integrated crop and livestock system (ICLS), northern European agriculture

Introduction

Integrated crop and livestock systems (ICLS) can help reduce environmental impact of our food production without loss of profitability and yield by closing nutrient and energy cycles locally (Taube *et al.*, 2024; Ryschawy *et al.*, 2017; Lemaire *et al.*, 2013). Knowledge of regional adaptation for producing crops for both feed and food is key to make ICLS efficient, yet practical guidelines are often lacking (Garreth *et al.*, 2020). For successful ICLS there is need for robust farm practices that can handle yearly fluctuation in growing conditions.

A concept that could further aid farmers integrate crop and livestock production is dual-purpose crops, described by McGrath *et al.* (2021) as annual crops (e.g. cereals) utilised for both forage and grain harvest by altering the natural plasticity and phenology. The feasibility of such concept in northern Europe may be questionable, due to environmental conditions, relative small-scale farming and high land-use intensity. Perhaps, integrating dual-purpose crops with single-purpose crops producing both forage and grain can increase overall system benefits (e.g. filling feed gaps, productivity, economic outputs, nutrient use efficiency and ground cover). For instance, an experiment conducted in Wiscon-

Table 1. Overview of field experiment treatments, showing crop sequences, intended harvests, and locations.

Treatment	Crops sequence	Harvest year 1	Harvest year 2	Location
A	Spring barley → winter wheat	Threshing barley	Threshing wheat	Lönntorp, Götala, Lövsta
B	Spring barley → triticale	Threshing barley	Threshing triticale	Röbäcksdalen
C	Hybrid rye+Westewold ryegrass	Multiple mowing	Thresh rye	All
D	Grain maize+lucerne	Threshing maize	Multiple mowing	Lönntorp
E	Spring wheat+English ryegrass+clover	Threshing wheat	Multiple mowing	Götala, Lövsta
F	Silage maize+hybrid rye	Cut whole maize	Threshing rye	Lönntorp, Götala, Lövsta
G	Spring barley+hybrid rye	Threshing barley	Cut whole rye	Röbäcksdalen
H	Spring barley+hybrid rye	Cut whole stand	Threshing rye	Röbäcksdalen

→ indicates the subsequent crop after harvest, and + indicates intercropping.

sin, USA, evaluated rye (*Secale cereale*) and maize (*Zea mays*), showing that rye provided high-quality forage without compromising maize yield (Glaze-Corcoran *et al.*, 2023).

This explorative study presents preliminary results from field experiments of different crop sequences producing forage and grain within a two-year crop rotation in four different regions of Sweden. Evaluation of the conducted field trails include agronomic practises, yield, feed quality, and phenotypic morphology. Methodology of the ongoing experiment and preliminary results and observations of the first year are presented in this paper.

Material and methods

Four distinct locations in Sweden were selected for this experiment, Lönntorp (55°66'N, 13°10'E), Götala (58°22'N, 13°29'E), Lövsta (59°49'N, 17°48'E) and Röbäcksdalen (63°81'N, 20°24'E). Mineral fertilizer was used up to 220N, 30 P, 100 K, & 20S kg ha⁻¹ year⁻¹. Prior to sowing with a seed drill the fields were ploughed and harrowed. Plot sizes differed per location and ranged from 14 to 90 m². At each location the treatments were replicated four times within a randomized complete block design. The experiment was established spring of 2025 and will be finalized autumn 2026. In an adjacent field, a parallel crop sequence will be established in spring 2026 that finalizes during the autumn 2027.

Some flexibility in agronomic management per site was permitted to optimize the success of the proposed crop sequences; all locations were instructed to record their management actions in detail. Prior to each harvest the phenological stage of all crops was estimated using the BBCH scale (Meier, 1997). Fresh weight (FW) and dry weight (DW) were determined for all plots. From these harvests dry samples were stored for future feed quality analysis.

Treatments A (spring barley (*Hordeum vulgare*) followed by winter wheat (*Triticum aestivum*)) and B (spring barley followed by winter triticale (×*Triticosecale*)) are characterized by consecutive years of grain production. All other treatments produced both grain and forage and consisted of intercropping winter hybrid rye with annual ryegrass (*Lolium multiflorum*) (C), maize with lucerne (*Medicago sativa*) (D), wheat with perennial ryegrass (*Lolium perenne*) and red clover (*Trifolium pratense*) (E), maize with winter hybrid rye (F), and spring barley with winter hybrid rye (G and H). A detailed overview of all treatments is given in Table 1.

Forages were harvested at the heading stage (BBCH 50) when multiple harvests were planned, and at the dough stage (BBCH 85) when only a single harvest was intended. Grain harvesting occurred at full maturity (BBCH 89).

Results and discussion

The findings presented here are based solely on observational data collected in 2025, and no additional data sources were yet available for analysis. Treatment C established a productive forage ley in the first year across all sites, with biomass contributed by both rye and ryegrass. Yield is expected to vary by location but should be sufficient, as rye serves a flexible role within the ICLS concept. Ryegrass and clover established as undersown crop in spring wheat (treatment E) that was threshed at a similar time to other cereal crops in the area. Treatment G resulted in a good establishment of undersown winter rye and was harvested at same time as treatment B, indicating that undersowing rye did not affect spring barley development. Pending yield and feed quality analyses, these initial observations appear promising. Treatment F showed site-specific differences in crop establishment. At Götala and Lövesta, rye was sown simultaneously with maize: at Lövesta, rye failed as maize developed, whereas at Götala, rye continued to grow and hampered the maize development. At Lönnstorp, rye was undersown a month after maize, allowing normal maize growth while limiting rye establishment. At the same time, lucerne (Treatment D) established well, undersown with maize one month after the maize was sown. Seasonal differences, agronomic management and crop adaptation to local growing conditions need to be evaluated further, and agronomic experiments addressing species-specific issues may be needed. All treatments in this experiment proposed a dual-purpose utilization of crops by mechanical harvesting for both grain and forage. It is relevant to consider animal grazing into the proposed crop sequence, as system benefits of an ICLS may be enhanced further. A large-scale grazing experiment is needed to investigate responses of both crops and livestock.

Conclusions

The preliminary observations indicate potential utilization of dual-purpose crops in northern European integrated crop and livestock systems, due to the inherent phenology and plasticity of annual cereals. This experiment demonstrates proof of concept from a crop perspective, and it lays groundwork for future dual-purpose crops application in Northern Europe.

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Effect of forage species and conservation on the content of minerals with nutritional interest for ruminants

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Abstract

To assess effects of forage species and conservation methods on the content of minerals, 3 plots were used: sainfoin (SF), permanent pasture (PP) and cocksfoot [orchardgrass] (OG). Each plot was divided into 3 areas giving 9 experimental areas, in which 5 conservation methods were compared: green forage (GF), unwilted (UWS) and wilted (WS) silages, haylage (HG) and hay (H), giving 45 samples in total. Mineral contents were analysed for 11 elements (P, Ca, Mg, K, Na, Cl, S, Zn, Cu, Mn, Fe). Contents were higher in SF than in OG for P, Ca, Mg, Zn and Cu, while they were lower for Na, Cl and Mn. Variations between conservation methods were observed mainly in SF. Compared with GF in SF, contents increased for Ca, Zn in silages, and decreased for Ca, Mg, S and Mn in H. K content in GF decreased in UWS and increased in WS, HG and H. Although the contents of minerals varied in conserved forages, that was strongly species-dependent.

Keywords: forage, conservation, macro minerals, trace elements, ruminants

Introduction

Minerals are essential for production, health or reproduction of ruminants (INRA, 2018). Forages can provide a significant proportion of the minerals supplied by the ration, but their mineral contents vary according to plant species, growth stage and cut cycle (INRA, 2018). Additional variations could occur in conserved forages, but studies about variations between green and conserved forages are limited. This trial aimed to evaluate the effect of forage species and conservation method on the mineral content.

Materials and methods

This study was conducted at INRAE Herbipôle research farm (Saint-Genès-Champanelle, France). Three neighbouring plots were used: *Onobrychis viciifolia* Scop. (sainfoin, SF), permanent pasture (PP) and *Dactylis glomerata* L. cocksfoot [orchardgrass] OG). The SF and OG were sown on May 2022. Each plot was split into 3 areas used as randomized repetitions giving 9 experimental areas, which were harvested on 30 May 2023 at the first cut, and at late bud–early flower stage of growth for SF and heading stage for PP and OG. The PP field was grass dominant with *D. glomerata* L., *Poa pratensis* L., *Holcus lanatus* L. For each experimental area, biomass was separated for the 5 conservation methods: green forage (GF), unwilted (UWS) and wilted (WS) silages, haylage (HG) and hay (H), giving a total of 45 samples. Biomass for UWS were immediately chopped and ensiled. Wilted silages, HG and H were made after wilting forage on ground during 6, 24 and 96 h, respectively. Silages were conserved in PVC cylinder silos; HG bales were made by wrapping biomass in plastic layers and H bales were round. Conserved forages were sampled approximately 19 weeks after their preparation. All samples were forced-air oven-dried 72 h at 60°C then ground to 1 mm. Seven macro minerals (P, Ca, Mg, K, Na, Cl and S) and four trace-elements (Zn, Cu, Mn and Fe) were analysed. All elements were analysed with Inductively Coupled Plasma Atomic Emission Spectrometry (ICP-OES) except

Cl which was analysed by argentometric titration on mineralized samples. Data were analysed by a multi-way mixed ANOVA model using R with area as random factor, and forage species, conservation methods and their interaction as fixed factors. Multiple comparisons of means were performed with the Tukey test.

Results and discussion

The effect of forage species was strong and affected significantly all minerals ($P < 0.001$, Figure 1) except Na. Regardless of conservation method, contents were higher in SF than in OG for P, Ca, Mg, Zn and Cu, while they were lower for Na, Cl and Mn. The Na contents were only affected by the forage species and were 0.13, 0.32 and 0.28 g (kg DM)⁻¹ for SF, PP and OG, respectively (SEM = 0.02, $P < 0.001$; data not shown in Figure 1). For K, S and Fe, we observed an interaction between the forage species and conservation methods. The K and S contents were higher in SF than in OG for all conservation methods except in GF for K and in H for S. The Fe content was also higher in SF than in OG, but only for WS and HG. Although the differences between conservation methods were smaller than the differences between the forage species, the conservation methods affected the content of all elements except Na, with more pronounced variations for SF. For SF, compared with GF, content increased in UWS by 11% for Ca and by 20% for Zn while it decreased by 11% for K. The K content also increased in WS, HG and H compared with GF by 12%. Finally, for SF, contents decreased in H compared with GF by 18% for Ca, 20% for Mg, 22% for S and by 28% for Mn. The increase of Ca and Zn in silages would likely relate to the loss of volatile compounds and gases during fermentation as suggested by Schlegel *et al.* (2018). The reductions in the contents of Ca, Mg and Mn in hay, could be due to the loss of leaves during drying and the fact that these elements are mainly located in the leaves (Fleming, 1963). The reduction in the S content and the increase in the K content we observed in H for SF contrasted with the similar contents of these elements in hay and green forage observed by (Schlegel *et al.*, 2018) for a grass dominant pasture for both elements and a reduction in hay observed by (Meschy *et al.*, 2005) for K. However, the reduction in the S content was in accordance with that observed by (Meschy *et al.*, 2005) on average, on pastures composed of grasses and/or legumes that could suggest that the S losses during haymaking could be specific to legumes. The K increase we observed in SF for wilted forages (H, HG and WS) is consistent with a likely higher concentration of K in stem than in leaf in some legumes (Whitehead *et al.*, 1985) and a loss of leaves during drying on the ground.

Conclusion

Although the forage species was an important source of variation in the forage mineral content, the forage mineral contents could also significantly vary according to the conservation methods for several elements, especially for sainfoin, with an increase in silages and a decrease in hay. The clear dependency of the mineral contents on the conservation methods according to the species suggests that, based on current knowledge, farmers should analyse their forages to adjust their mineral supplementation.

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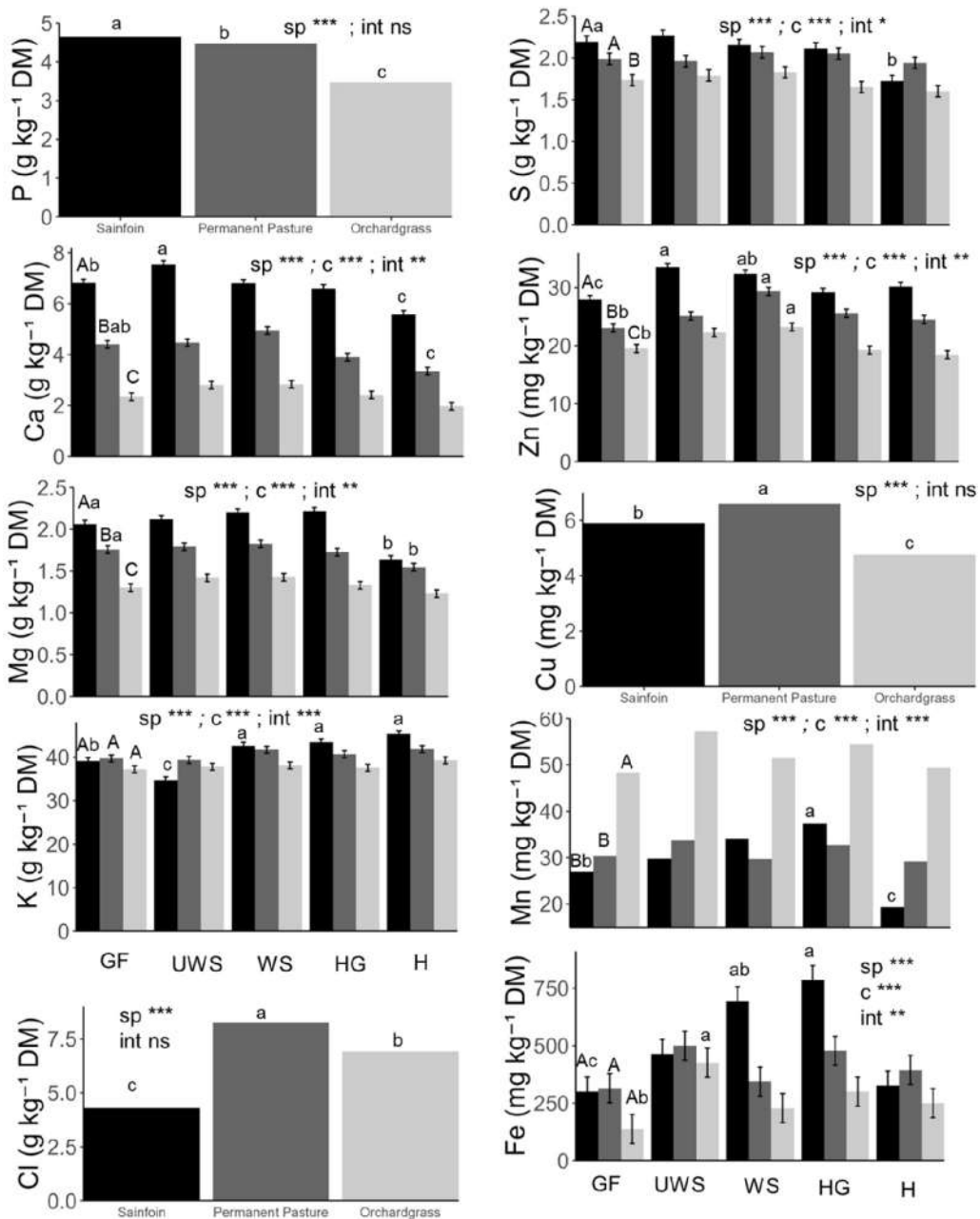


Figure 1. Macro-minerals (g kg⁻¹ DM) and trace-elements (mg kg⁻¹ DM) contents in forages from three forage species and five conservation methods (sp, species; c, conservation method; int, interaction). ****P* < 0.001; ***P* < 0.01; **P* < 0.05; ns, not significant. Capital letters: result of post-hoc comparisons of means between species. Lower-case letters, result of comparisons of means between conservation method. Post-hoc Tukey tests were based on the means of the interaction when *P* interaction < 0.05, and on the means of the forage species and of the conservation method otherwise. Mn contents were log transformed for the analyses.

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Incorporation of different levels of alfalfa-based dehydrated silage pellet in laying hens' diet

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Abstract

Alfalfa-based silage pellet (ABSP) meal has been previously identified as a locally available alternative protein source that could partially replace soybean meal in organic poultry diets. Forty 32-week-old Novogen Brown Light hens were randomly assigned to four dietary treatments with ABSP inclusion levels of 5, 10, 15 or 20% in a standard diet over 35 days. The results of the study showed that feed intake was similar regardless of the incorporation level. A linear effect of ABSP inclusion level was observed on yolk colour, yolk-to-albumen ratio ($P < 0.01$), as well as on the ω -3 fatty acid content and the ω -6/ ω -3 ratio in yolk fat ($P < 0.05$). In conclusion, ABSP can be included at levels of 15–20% in laying hen diets without negatively affecting laying performance, while improving certain aspects (yolk colour, PUFA) of yolk quality.

Keywords: egg quality, *Medicago sativa*, optimal substitution level, organic laying hens, silage

Introduction

Nowadays, the majority of conventional protein sources used in European livestock feed are derived from imported soybeans, primarily produced in the Americas. This reliance on external protein sources presents a significant risk to Europe's social, economic, and environmental sustainability (Tallentire *et al.*, 2018). In organic poultry farming, sourcing suitable organic feed ingredients is particularly challenging due to constraints related to production systems, animal welfare standards, and the availability and quality of organic feedstuffs (Bostan *et al.*, 2019). Among potential alternatives, leaf silage of *Medicago sativa* (alfalfa/lucerne) remains underexplored in poultry nutrition. Given the increasing demand for sustainable and locally sourced feed ingredients in organic farming, alfalfa-based silage pellets may offer a promising alternative to imported soybean meal. There is limited research on the optimal inclusion rate of this ingredient in poultry diets and its potential effects on laying performance and the physical-chemical characteristics of eggs in organic laying hens. Grass and alfalfa silage's substitution has been successfully tested previously (Abbas *et al.*, 2024). The present study aims to evaluate the optimal inclusion level of alfalfa-based silage pellets in the diet of organic laying hens, with a focus on laying performance and the physical and chemical quality parameters of the eggs.

Materials and methods

Alfalfa-based silage pellets were produced from early vegetative stage (about four weeks of regrowth) forage crop harvested at the Center for Agronomic Technologies (CTA, Modave, Belgium). The forage mixture consisted of 75% alfalfa, 10% white clover (*Trifolium repens*), and 15% cocksfoot (orchard grass) (*Dactylis glomerata*). The forage was mowed and pre-dried in the field. Two days after harvesting, the forage was baled (one bale) and wrapped with five layers of plastic film. The bale was unwrapped

Table 1. Effects of different inclusions of alfalfa-based silage pellets in the diet on production performance and physicochemical parameters of eggs in organic laying.

Parameter	5%	10%	15%	20%	SEM	<i>p</i> value	Linear	Quadratic
Initial weight (g)	1874	1874	1874	1873	17.1	0.99	—	—
Final weight (g)	1918	1898	1914	1873	7.01	0.63	—	—
ADGW(g day ⁻¹)	3.0	2.4	2.6	0.65	0.79	0.25	—	—
ADFI (g day ⁻¹)	123.9	110.9	113.1	118.1	9.04	0.49	—	—
FCR	2.09	1.95	2.14	2.04	0.16	0.66	—	—
Egg weight (g)	62.4a	61.0b	61.0b	61.2b	0.25	0.001	0.001	0.001
Egg mass (g)	62.36	61.02	62.38	61.18	0.071	0.65	—	—
Laying rate (%)	95.14	93.42	95.42	93.65	0.16	0.65	—	—
Yolk colour	8.08 a	8.36 b	10.00 c	10.83 d	0.20	0.01	0.001	NS
Yolk:albumen	0.43 a	0.40 a	0.44 ab	0.48 b	0.21	0.01	0.01	0.05
C18:3 FA	1.39 a	1.42 a	1.63 ab	1.80 b	0.09	0.001	0.05	NS
W3 FA	3.46 a	3.42 a	3.68 ab	3.94 b	0.14	0.03	0.01	NS
W6:W3 FA	6.50 a	6.25 ab	5.90 c	5.47 c	0.17	0.001	0.001	NS

^{a, b} Means without a common letter are significantly different at the 0.05 level; ADGW, average daily weight gain; ADFI, average daily feed intake; FCR, feed conversion ratio; FA, fatty acids.

seven months later. The fodder was then manually sorted to remove fibrous fractions and dried in a cold-air dryer until reaching a dry matter content of approximately 80%. The fodder was then pelletized into pellets approximately 2 cm in length and 5 mm in diameter. Finally, the pellets were ground using a flat-die grinder. The experiment involved four treatment groups and two replicates per treatment, with 5, 10, 15 and 20% ABSP substituted on a fresh-feed basis for a standard organic commercial diet of similar grain size. The diets were distributed *ad libitum*. A total of forty 32-week-old Novogen Brown Light organic laying hens were used in the trial, which lasted for five weeks. Production performance parameters were calculated following the methodology described by Moula *et al.* (2010). Three eggs from each block were chosen, and the fatty acid profile was analysed from the fat content after weighing 2 g egg yolk into a 50 ml Falcon tube and adding 40 ml chloroform/methanol (2:1, v/v) according to Douny *et al.* (2015). Statistical analysis was performed using mixed (when individual data available) or general linear model (SAS, 2023) based on the following structure:

$Y_{ij} = \mu + \alpha_i + \beta_j(\alpha_i) + \gamma_i + \varepsilon_{ijk}$, where Y_{ij} is the studied variable, μ the overall mean, α_i the i -th treatment effect ($i = 1-4$), $\beta_j(\alpha_i)$ the j -th replication effect ($j = 1-2$) nested in the i -th treatment effect, γ_i the i -th time, and ε_{ijk} the normally distributed residual error.

Results and discussion

The inclusion of ABSP at levels ranging from 5 to 20% had no significant effect on the final body weight, feed intake, feed conversion ratio or laying rate of the hens (Table 1). However, a significant difference in egg weight was observed among the four dietary groups ($p < 0.001$), with the highest average egg weight recorded in the group receiving the lowest level of ABSP supplementation. The egg mass was similar among the treatments.

Incorporating up to 20% ABSP did not reduce total consumption. It was likely that the treatment applied to the alfalfa silage allowed for such ingestion. The egg weight was significantly different among the groups, favouring the group that received a low incorporation rate (5%) of ABSP ($p < 0.001$). In contrast, the egg mass was not significantly different among the treatments; findings that are consistent with those reported by Laudadio *et al.* (2014) and Wüstholtz *et al.* (2017), but different from those

reported by Mourao *et al.* (2006), who reported that body weight, egg production, and egg weight in young hens were negatively affected by a diet containing 15% alfalfa meal. The present results also contrast with those of Laudadio *et al.* (2014), who observed no significant changes in egg weight when alfalfa was incorporated into the diet at levels ranging from 4 to 16%. These results differ from those reported by Englmaierová *et al.* (2019), who observed a decrease in laying rate and an increase in feed conversion ratio when 4% dehydrated alfalfa was included in laying hen feed. In agreement with Karsten *et al.* (2010) and Carrasco *et al.* (2016), a highly significant difference in polyunsaturated fatty acids was observed with increasing levels of ABSP, notably a linear increase in C18:3 ω -3 and in the ω -6/ ω -3 ratio ($p < 0.01$).

Conclusions

The inclusion of ABSP in the diet, at levels ranging from 5% to 20%, did not affect feed intake, feed conversion ratio, hen laying rate, or egg physical quality. However, it improved the ω -3 and ω -6/ ω -3 fatty acid ratio in eggs, while reducing egg weight. Based on the findings of this study, ABSP meal appears to be a promising feed ingredient for organic poultry farming. An inclusion rate of at least 15–20% can be recommended for organic laying hens without compromising performance, while enhancing egg quality.

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Forage production for sustainable ruminant systems in a context of climate change

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Abstract

Climate change affects the production and distribution of grass throughout the year. In this paper, we review and discuss ways in which grass-based systems can cope with climate change. While grass growth may be reduced in spring and summer, new opportunities for grass use may arise in autumn and winter. Advanced plant phenology means that harvesting must be brought forward, which can be challenging during periods of frequent rainfall. Several strategies can be employed to adapt existing resources or develop new ones. Grassland management should take advantage of the diversity of grasslands between plots that are more or less productive. Consideration could be given to introducing climate-resilient species (such as resource-conserving grasses, Mediterranean legumes and diverse plants like chicory), cereal-legume intercrops, summer catch crops and C4 plants. Developing agroforestry is also a way of providing additional forage resources during periods of low grass growth. In terms of the forage and livestock system, the current and future climate means that more forage stocks will need to be built up to overcome dry summer periods. Alternatively, structural changes will need to be made to reduce stocking rates.

Keywords: adaptation, climate change, forages, grasslands, ruminant

Introduction

Grass-based ruminant systems are widely recognised as favouring the agroecological transition in livestock production. They offer several advantages, including reduced reliance on external inputs (such as pesticides, fossil energy and water) and increased ecosystem services (e.g. carbon storage, biodiversity, product quality and animal welfare). Although less critical than for crop production, the impact of climate change on livestock production is expected to become substantial after 2040 due to lower water availability, the increasing impact on forage production and quality, and the high risk of spreading animal diseases (European Environment Agency, 2024). Furthermore, the observed temperature increase over the European land area is higher than the global average. According to emission scenarios, the temperature increase is expected to reach 2.5–3°C by 2050. In this short review paper, we summarise the main trends in forage production in the context of climate change, as well as the most promising strategies for adapting forage production and management.

Main trends in forage production in the context of climate change

Climate change affects seasons and plant phenology. Tree observations in Germany revealed that the phenological seasons changed between 1961 and 2020, with winter duration decreasing from 120 to 101 days and spring and autumn increasing from 96 to 102 days and 59 to 73 days, respectively, while summer duration remained almost unchanged (Deutscher Wetterdienst, 2025). Regional forage management models for the Massif Central in France (SIDAM, 2023) predict a one-month advance in both grazing and early cutting by 2050 compared to 2000. A retrospective study of the evolution

of hay quality harvested at INRAE Marcenat (1000 m asl) between 1979 and 2010 (271 samples) confirmed a decrease in quality (Deroche *et al.*, 2020). The cutting date was advanced by only 6 days, despite the fact that, based on the sum of temperatures at cutting, it should have been advanced by 19 days to maintain the same stage of maturity. Consequently, the crude fibre content of the hay increased significantly by $24 \text{ g (kg DM)}^{-1}$, whereas the crude protein content decreased significantly by $25 \text{ g (kg DM)}^{-1}$, and the *in vivo* organic matter digestibility decreased by 0.018 g g^{-1} . Weather conditions and the occurrence of three- to four-day periods without rain in spring may limit the ability to sufficiently advance the cutting date of hay to maintain its quality.

Thus, climate change is already affecting grass production and quality, with implications for livestock production. A meta-analysis of the effects of climate change on the yield and quality of European pastures (Dellar *et al.*, 2018) shows that pasture yields will increase in areas that become warmer and wetter, particularly in the north and in parts of the Alps and continental Europe (by up to 80%), whereas they will decrease in areas that become warmer and drier, particularly in southern Europe and parts of continental Europe (by up to 40%). Generally, plant protein concentration is likely to decrease. Different functional groups react differently, implying future changes in pasture composition. Another meta-analysis concluded that a decline in forage quality with rising temperature could lead to a 0.9% increase in methane production with a 1°C temperature rise, and a 4.5% increase with a 5°C rise (model average, Lee *et al.*, 2017).

The strategies for adapting forage production and management

The first is to adapt grazing and harvesting practices to the seasonal pattern of grass growth. Generally, grazing and harvesting management involves balancing the quantity and quality of grass (Baumont *et al.*, 2014). Adapting forage calendars to climate change will primarily involve using grass earlier in spring and later in autumn when weather conditions and soil bearing capacity enable it, as well as creating stocks for summer gaps.

A second strategy is species diversification within and between plots. It is well established that species diversity provides multiple services in sown and permanent swards. Functional composition explains the annual pattern of grassland production and quality (Michaud *et al.*, 2014). While diversity does not affect the negative relationship between biomass yield and the nutritive value of forage, it can stabilise forage quality throughout the year (Huyghe *et al.*, 2008). Thus, species diversity and diversity of sward types bring flexibility to grass management. Species richness secures yield in drought conditions as species asynchrony increases the temporal stability of mixtures (Haughley *et al.*, 2018). Exploring the genetic resources of Mediterranean and semi-arid zones could help temperate grasslands adapt to climate change, as only a small proportion of interspecific diversity is utilised in grassland mixtures (Lüscher *et al.*, 2022). However, the trade-off between species' adaptation to drought and forage feed value must be considered, as drought-adapted species are usually of less nutritive quality.

A third strategy involves producing additional forage resources. This can be achieved by intercropping cereal and legume mixtures (Maxin *et al.*, 2017). The species proportions in the mixtures, the seeding rate, the growth stage at harvest, and the utilisation method (grazing or silage) offer a wide range of forage resources, yielding between 4 and 10 t DM ha^{-1} , with the nutritive value increasing with the proportion of legumes and early growth stage. Catch crops can also produce forage during the off-season (summer and winter) (Maxin *et al.*, 2020). A large diversity of plants (and plant mixtures) can be used, including annual legumes such as berseem (*Trifolium alexandrinum* L.), crimson clover

(*Trifolium incarnatum*) and vetch (*Vicia sativa*), as well as phacelia (*Phacelia*) and C4 plants such as foxtail millet (*Setaria italica*), teff grass (*Eragrostis tef*) and sorghum (*Sorghum bicolor*), with yields ranging from 2 to 3 t DM ha⁻¹ and up to 10 t DM ha⁻¹ for sorghum. Another option is to integrate agroforestry into livestock forage systems. Trees and woody species can overcome grass shortages during dry periods as they are more drought-resistant. Tree leaves can have an interesting feed value (Bernard *et al.*, 2020), but with significant differences between species (Mahieu *et al.*, 2021).

Conclusion

Climate change impacts forage production and its intra- and inter-annual variability. In the short and medium term, there are many adaptation solutions to secure forage systems, that can be combined to cope with climate change. In the long term, redesigning livestock systems may be necessary, including reducing stocking rates and changing animal breeds or species. Research should focus more strongly on: (i) the animal-grass dynamic, i.e. selecting and managing animals for grass and grass for animals; (ii) managing compromise with risks and searching for equilibrium; and (iii) developing appropriate tools that integrate grass, animal and system issues.

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The effect of location and nitrogen fertiliser rate on annual herbage production in red clover silage swards

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Abstract

Red clover (*Trifolium pratense* L., RC) in silage swards can produce high herbage yields with reduced chemical nitrogen (N) application rates; however, retaining sufficient RC content to maintain production is challenging. The objective of this study was to examine the effect of location and N rate on annual herbage production and RC content of perennial ryegrass (*Lolium perenne* L., PRG)–RC swards across two years. There were three N fertiliser rates (Low; 0 kg N ha⁻¹, Medium; 75 kg N ha⁻¹ or High; 150 kg N ha⁻¹) and plots were managed under a three-cut protocol at two locations in Ireland. Location 1 (Moorepark, Co. Cork) was a free-draining acid brown earth soil of sandy loam/loam texture. Location 2 (Grange, Co. Meath) was a brown earth of imperfectly drained clay loam. Location significantly effected DMY and RC content ($P < 0.001$); Moorepark had a lower DMY than Grange ($13\,855 \pm 250.1$ and $15\,683 \pm 250.1$ kg DM ha⁻¹, respectively) but a greater RC content (613 g (kg DM)⁻¹ and 305 ± 11.2 g (kg DM)⁻¹, respectively). As N rate increased RC content declined; 525 ± 11.8 g (kg DM)⁻¹ (Low), 460 ± 11.8 g (kg DM)⁻¹ (Medium) and 392 ± 11.8 g (kg DM)⁻¹ (High). There was no impact of N rate on DMY in Moorepark ($13\,855 \pm 370.2$ kg DM ha⁻¹), while in Grange, the Low rate had the lowest yield ($14\,155 \pm 370.2$ kg DM ha⁻¹) compared to the Medium and High ($16\,447 \pm 370.2$ kg DM ha⁻¹) ($P < 0.01$). Location can have a significant impact on both DMY and RC content, while the response to chemical N fertiliser can vary across location.

Keywords: herbage production, location, nitrogen, red clover

Introduction

Red clover (*Trifolium pratense* L., RC) inclusion in perennial ryegrass (*Lolium perenne* L.; PRG) silage swards maintains herbage production and quality at reduced rates of nitrogen (N) fertilisation due to its ability to biologically fix atmospheric N (Clavin *et al.*, 2016). Incorporation of RC can therefore aid in meeting chemical N reduction targets (Egan *et al.*, 2024); however, poor persistence of RC is a key management challenge (Marshall *et al.*, 2017). Nitrogen fertiliser is widely reported to increase herbage production although clover content of PRG-legume swards is reduced under high N application which results in reduced biological N fixation (Kristensen *et al.*, 2022). Previous studies have investigated moderate spring N application to increase PRG-RC sward herbage production and avoid repercussions on RC content (Clavin *et al.*, 2016). However, varying effects of N fertiliser (Kristensen *et al.*, 2022) and soil N mineralisation (Geisseler *et al.*, 2024) on herbage production and RC content across different soil types and locations has not yet been considered. The objective of this study was to examine the effect of location and N rate on annual herbage production and RC content of PRG-RC silage swards across two years.

Materials and methods

A plot study was established (with a seeding rate of 7 kg ha⁻¹ RC and 24 kg ha⁻¹ PRG) in May 2022 at two locations in Ireland. Location 1 was Moorepark in Fermoy Co. Cork (52°9'N; 8°16'W),

Table 1. The effect of location and year on annual herbage production and red clover content averages across three N rates

	Location		SEM	Level of significance			
	Moorepark	Grange		Location	Year	Location×year	N rate×location
Year 1							
DMY (kg DM ha ⁻¹)	16 683	18732	279.6	<0.001	<0.001	NS	<0.01
RC content (g (kg DM) ⁻¹)	692	435	11.2	<0.001	<0.001	<0.001	NS
Year 2							
DMY (kg DM ha ⁻¹)	11 028	12635	279.6				
RC content (g (kg DM) ⁻¹)	535	175	11.2				

Values represent the mean averages across three N rates±SE. $P > 0.05$ =NS (non-significant).

a free-draining acid brown earth soil of sandy loam/loam texture. Location 2 was Grange in Dunsany, Co. Meath (53.52°N, 6.66°W), a brown earth of imperfectly drained clay loam. Moorepark had a soil index 4 for Morgan's soil test for phosphorus (P) (11.6 mg l⁻¹) and potassium (K) (192 mg l⁻¹) on average over the trial period, with a soil pH of 6.8. Grange had a soil index 3 for P (7.2 mg l⁻¹) and K (138.2 mg l⁻¹), with a soil pH of 6.5. (Index 1 to 4; 1 = deficient, 4 = no response to nutrient application) (Alexander *et al.*, 2008). There were three N rates (0, 75, 150 kg N ha⁻¹; Low, Medium, High, respectively) and three repetitions of each treatment. Plots were managed under three cut silage protocol (May, July and August/ September) across both years (2023 and 2024). A BCS 740 two-wheel tractor with a scythe cutter bar (BCS, Abbiategrosso, MI, Italy) was used to cut plots to 6 cm. Phosphorus (35 kg ha⁻¹) and K (375 kg ha⁻¹) were applied at both locations annually, with additional P and K applied as per soil test results (Alexander *et al.*, 2008). Granulated lime was applied at both locations in 2024 (1250 kg ha⁻¹). Dry matter yield (DMY) was determined by weighing fresh harvested material and drying a 100 g sub sample (90°C for 16 h) to determine the DM content. Sward RC content was determined by separating a 200 g subsample into PRG and RC portions and drying (90°C for 16 h) to determine the DM proportion of each. Data were analysed using PROC MIXED in SAS 9.4 (SAS Institute, Cary, NC, USA, 2002) with location, N rate, year and associated interactions included as fixed effects, with repetition included as a random effect and plot as subject.

Results and discussion

Annual DMY reduced from year 1 ($P < 0.001$) (17 707 ± 250.1 kg DM ha⁻¹) to year 2 (11 831 ± 250.1 kg DM ha⁻¹) across both locations. There was also a significant effect of location (P 0.001) with a lower DMY at Moorepark (13 855 ± 250.1 kg DM ha⁻¹) than Grange (15 683±250.1 kg DM ha⁻¹). There was a significant interaction between location and year ($P < 0.001$) on RC content, Grange had a greater reduction in RC content compared to Moorepark (23 and 60%, respectively) (Table 1).

Reduced yield and clover proportion of PRG-RC mixed swards across successive harvest years is frequently reported and is linked to poor persistence of RC (Marshall *et al.*, 2017). Differences in RC content across locations is likely caused by varying pedoclimatic properties, as noted by Kristensen *et al.* (2022). Soil temperature (Frame *et al.*, 1998) and moisture content (Sprent, 1976) is important for RC growth and N fixation. The higher average soil temperatures in Moorepark compared to Grange (10.2 and 9.6°C, respectively) and increased annual rainfall (1076 and 868 mm, respectively) may have been more favourable for RC growth. Increased herbage production and reduced RC content observed in Grange may also be attributed to greater soil N content associated with higher clay content soils (Geisseler *et al.*, 2024).

There was a significant location by N rate interaction on DMY ($P < 0.01$), with Moorepark producing $13\,855 \pm 370.2$ kg DM ha⁻¹ across the 3N rates, while the Low N treatment in Grange had the lowest yield ($14\,155 \text{ kg} \pm 370.2$ DM ha⁻¹) compared to Medium and High ($16\,447 \pm 370.2$ kg DM ha⁻¹). The lack of effect in Moorepark could be attributed to greater N fixation associated with a higher RC contents (Kristensen *et al.*, 2022) for the Low N rate 675 ± 16 g (kg DM)⁻¹, compared to the Medium and High N rates 608 ± 16 and 557 ± 16 g (kg DM)⁻¹, respectively. Where in Grange there was a significantly lower sward RC content of 376 ± 16 , 313 ± 16 and 226 ± 16 g (kg DM)⁻¹ for Low, Medium and High N, respectively, likely caused by higher soil N (Geisseler *et al.*, 2024) and lower soil temperature and rainfall (Frame *et al.*, 1998; Sprent, 1976). The reduced RC content as a result of increased N rate ($P < 0.001$) was consistent with findings from Kristensen *et al.* (2022).

There was also a significant N rate by year interaction on DMY ($P < 0.001$); there was no difference between N rates in year 1 ($17\,707 \pm 370.2$ kg DM ha⁻¹), while in year 2 the Low N had the lowest yield ($10\,270 \pm 370.2$ kg DM ha⁻¹) compared to Medium and High ($12\,612 \pm 370.2$ kg DM ha⁻¹). The increased N response in year 2 can be explained by the reduced RC content across years as frequently reported in RC studies (Kristensen *et al.*, 2022; Marshall *et al.*, 2017).

Conclusion

Different soil types and weather conditions across locations have a significant effect on herbage production and RC content of PRG-RC silage swards. Moderate application of N fertiliser can be strategically used to optimise herbage production of PRG-RC silage swards at reduced RC content; however, the effect of N on DMY varies across location and consideration is required.

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Exploring the role of grassland flora in animal health: perceptions and knowledge among European researchers

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Abstract

A common assumption is that grassland floral diversity benefits animal health, even though knowledge remains unclear. This study aimed to assess European researchers' knowledge of the links between grassland flora and animal health. In 2024, we collected 66 responses from 14 countries through an online questionnaire containing 5 sections: geographical area; profession and grassland expertise; perceptions of grasslands' contributions to animal health; identification of positive or negative effects for 31 and 28 species, respectively. A K-means clustering identified 3 respondent profiles: (i) 'animal' ($n = 19$) with expertise in animal nutrition/health, in lowlands and agroecological concerns; (ii) 'agronomy' ($n = 14$), with expertise in agronomy and ecology in piedmont/mountain areas; (iii) 'animal welfare' ($n = 19$), with average expertise focused on grazing conditions and resource access. The 'animal' group had the fewest 'don't know' responses and clearly identified well-known beneficial species, such as sainfoin or chicory. The 'agronomy' group had the most 'don't know' responses, despite their botanical expertise, suggesting more cautious responses due to an unstable nature of knowledge. It frequently cited antioxidant/anti-inflammatory properties. Toxic species were well identified at low rates by all groups. Overall, knowledge of flora-health links is low, highlighting the need for clearer, interdisciplinary insights on this emerging area of interest.

Keywords: animal health, flora diversity, grasslands, survey

Introduction

The contribution of grasslands to the agroecological transition of ruminant farming is now well documented, with the exception of one priority: integrated management of animal health. Some grassland species, including trees, contain secondary metabolites with antioxidant or anthelmintic properties (Hoste *et al.*, 2005; Poutaraud *et al.*, 2017; Maxin *et al.*, 2020) and then could contribute to this management. Nevertheless, farmers' perceptions about these properties/practices seem more complex and difficult to establish (Petit *et al.*, 2020; Rose *et al.*, 2024; Di Blasi *et al.*, 2024). Some of these works have suggested that these perceptions could be linked to the scientific knowledge mobilized by advisors/facilitators and the fields of expertise from which it originates. However, to date, there are no studies establishing the level of knowledge of researchers from different disciplines (animal science, animal health, botany, ecology, agronomy, etc.) and their potential impact on technical advice and farmers' perceptions. The aim of our work was to carry on an exploratory survey to understand how European researchers link grassland flora and animal health in relation to their expertise. Our hypothesis was that the greater the botanical expertise, the more accurate the knowledge of the health risks and benefits associated with grassland flora would be.

Materials and methods

In 2024 we issued an online questionnaire in 6 languages and collected 66 responses from 14 countries. The questionnaire consisted of 5 parts: geographical area; profession and expertise in grasslands;

perception of the contributions of grasslands to agroecology and animal health. In the last part, respondents were first given a list of 31 plant species and asked to identify their level of knowledge for each species by clicking on one or several beneficial effects: don't know, none, anti-inflammatory/antioxidant, anti-parasitic, digestive/renal/cardiovascular function. They were then given a second list of 28 plant species and asked to identify their level of knowledge by clicking on one or several negative effects: don't known, none, digestive and metabolic disorders, toxicity, photosensitivity. Eighteen species were proposed for both beneficial and negative effects (*Medicago sativa*, *Onobrychis viciifolia*, *Lotus corniculatus*, *Vicia sativa*, *Lathyrus pratensis*, *Achillea millefolium*, *Plantago lanceolata*, *Matricaria chamomilla*, *Cichorium intybus*, *Taraxacum officinale*, *Sanguisorba minor*, *Centaurea nigra*, *Rumex acetosa*, *Cirsium arvense*, *Allium vineale*, *Juncus effusus*, *Phragmites australis*, *Urtica dioica*), 13 only for beneficial effects (*Lolium perenne*, *Phleum pratense*, *Dactylis glomerata*, *Anthoxanthum odoratum*, *Thymus serpyllum*, *Acer campestre*, *Castanea sativa*, *Salix caprea*, *Corylus avellana*, *Morus alba*, *Quercus robur*, *Hedera helix*, *Parthenocissus quinquefolia*), and 10 only for negative effects (*Ranunculus acris*, *Hypericum perforatum*, *Jacobaea vulgaris*, *Hypochaeris radicata*, *Robinia pseudoacacia*, *Prunus laurocerasus*, *Taxus baccata*, and chestnuts, acorns, ivy roots).

To test our hypothesis, we carried out a K-means clustering analysis of the answers given to questions on expertise (11 questions). The profiles were then compared in terms of geographical area, level of study, perceptions of the role of grasslands in the agroecological transition / animal welfare and frequency of citations of beneficial or negative effects associated with the proposed species.

Results and discussion

We established 3 respondent profiles (Table 1): (i) 'animal' ($n = 19$) with a higher proportion of respondents with expertise in animal nutrition/health/reproduction, located in lowland areas, MSc level, and concerned about the contribution of grasslands to agroecology; (ii) 'agronomy' ($n = 14$) with a higher proportion of respondents with expertise in agronomy (nutrient cycle, carbon storage) and ecology (flora of hedges and grasslands, water quality), located in piedmont/mountain areas, PhD level; 'animal welfare' ($n = 19$) composed by respondents with an average level of expertise, but concerned about welfare of grazing animals, in terms of access to resources (grass and water) and grazing conditions (hedges, shelters, paths).

The 'agronomy' group, despite its expertise in flora, had the highest 'don't know' response rates on the effects of grassland species, while the 'animal' group had the lowest rates. This result could validate the lack of established knowledge on this subject in botanical literature, and the growing interest in animal sciences for certain species mentioned in the questionnaire (sainfoin (*Onobrychis viciifolia*), narrow-leaved plantain (*Plantago lanceolata*), birdsfoot trefoil (*Lotus corniculatus*), chicory (*Cichorium intybus*)). Despite decreasing rates from the animal group to the agronomy group, the results converge on the level of knowledge of toxic effects, which are generally low, but most often correctly identified (e.g. common ragwort (*Jacobaea vulgaris*), buttercup (*Ranunculus acris*), St John's wort (*Hypericum perforatum*), robinia (*Robinia pseudoacacia*)). This is also partly true for the beneficial effects of species that have been studied and proposed as a mixture for sowing (grasses for digestive functions, sainfoin/chicory/birdsfoot trefoil for antiparasitism), but also for less consensual species (sweet vernal grass (*Anthoxanthum odoratum*) and ivy (*Hedera helix*) for antiparasitism, wild garlic (*Allium vineale*) for multiple effects). The 'agronomy' group stood out from the other two groups by a high 'antioxidant and anti-inflammatory' response rate for many species.

Table 1. Rates of cited grasslands in researchers' profiles species according to their supposed beneficial health effects

Species	% of response within a group														
	Don't know			none			Anti-inflammatory/ antioxidant			Antiparasitic			Digestive/renal/ cardio-vascular		
	We	An	Ag	We	An	Ag	We	An	Ag	We	An	Ag	We	An	Ag
<i>Lolium perenne</i>	63	53	50	16	10	22	0	10	22	0	0	0	22	10	29
<i>Phleum pratense</i>	63	60	64	10	10	14	0	5	0	0	0	0	31	11	22
<i>Dactylis glomerata</i>	47	53	64	21	21	22	0	5	0	0	0	0	31	10	22
<i>Anthoxanthum odoratum</i>	68	53	71	0	0	14	37	37	50	42	42	50	22	22	0
<i>Medicago sativa</i>	53	47	50	5	5	14	26	26	36	10	0	0	0	0	0
<i>Onobrychis vicifolia</i>	47	31	50	0	0	0	10	10	80	47	47	44	0	0	7
<i>Lotus corniculatus</i>	63	47	80	0	0	7	26	12	42	31	16	14	0	5	0
<i>Vicia sativa</i>	74	78	80	5	0	7	16	16	80	5	0	0	0	10	0
<i>Lathyrus pratensis</i>	74	63	71	5	0	14	5	10	80	0	5	0	0	5	0
<i>Achillea millefolium</i>	68	42	64	0	0	7	23	23	80	5	5	0	5	0	0
<i>Plantago Lanceolata</i>	42	26	50	0	0	7	21	26	36	42	31	0	5	0	0
<i>Matricaria chamomilla</i>	63	60	64	0	0	7	23	23	80	10	10	7	0	5	0
<i>Cichorium intybus</i>	42	42	80	0	0	7	31	37	50	31	21	22	5	4	22
<i>Taraxacum officinale</i>	53	47	50	0	0	0	19	19	80	5	5	22	0	5	0
<i>Sanguisorba minor</i>	79	68	80	0	5	0	21	21	29	5	5	7	0	4	7
<i>Centaurea nigra</i>	79	31	80	0	5	7	5	0	0	10	21	36	0	5	7
<i>Rumex acetosa</i>	63	53	80	5	16	14	21	21	29	5	5	0	0	5	0
<i>Crisum arvense</i>	63	60	73	5	21	7	10	10	14	5	0	0	0	0	7
<i>Allium vineale</i>	47	53	64	0	10	0	26	26	42	31	42	36	0	5	0
<i>Juncus effusus</i>	79	79	85	10	16	7	10	10	14	0	0	0	0	5	0
<i>Phragmites australis</i>	31	79	92	10	21	0	5	5	7	0	0	0	0	5	0
<i>Urtica dioica</i>	53	47	50	0	5	14	25	25	80	0	10	0	10	22	22
<i>Thymus serpyllum</i>	63	63	64	0	5	0	21	21	80	10	10	14	10	0	7
<i>Acer campestre</i>	74	30	85	10	5	7	10	10	14	0	0	0	5	10	7
<i>Castanea sativa</i>	60	78	80	10	5	7	21	21	29	0	5	7	0	5	0
<i>Salix caprea</i>	63	53	71	5	5	7	19	19	19	5	5	7	0	0	7
<i>Corylus avellana</i>	63	79	80	5	5	7	5	0	0	16	5	0	5	5	0
<i>Morus alba</i>	74	31	80	5	10	7	26	31	42	5	0	0	0	0	0
<i>Quercus robur</i>	79	74	71	5	10	0	21	26	36	5	5	7	0	5	7
<i>Hedera helix</i>	79	25	86	10	16	7	0	10	14	21	21	36	0	0	0
<i>Parthenocissus quinquefolia</i>	68	79	85	10	16	7	16	16	22	0	0	0	5	0	0

We, animal welfare profile ($n = 19$); An, animal profile ($n = 19$); Ag: agronomy' profile ($n = 14$).

Conclusion

The level of knowledge of the links between grassland flora diversity and the effects on animal health is low and disparate between fields of expertise, including among a research audience. This could contribute to the research questions they explore, the technical advice and farmers' perceptions of the grassland contribution to animal health. They call for a clarification of knowledge on this subject, which is of growing interest to livestock farmers in Europe.

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Assessing the reasons, costs and impact on ecosystem services of replacing permanent grasslands in quality label area

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Abstract

Grasslands play a major role in quality label products in France and notably in the Massif Central. However, to adapt their systems to climate change and pests, some farms are considering modifying or diversifying their forage systems, in particular by replacing permanent grassland with other forage or crops. The aim of this study was to evaluate the reasons that lead farmers to replace grasslands with other forage, and to assess the economic cost of replacing grassland and changes in ecosystem services (carbon sequestration and floristic diversity). To achieve this, 15 farms in the Massif Central were surveyed to identify the motivations behind permanent grassland replacement within their forage systems and to perform a partial budget analysis. A typology of grassland replacement practices was established, classifying the 15 farms into 6 types. This typology was associated with replacement costs and the quantification of ecosystem services: only two types showed an economic loss upon replacement but the loss of ecosystem services resulting from the conversion of permanent grasslands was significant. This work enriches the debate on the role of permanent grasslands in certified agricultural systems and on the issue of valuing ecosystem services for farmers.

Keywords: ecosystem service, grassland, partial budget

Introduction

Grasslands cover approximately 70% of the world's agricultural land (FAO, 2023). These are highly diverse ecosystems ranging from steppes to temperate grasslands. In France, in 2020, forage crops accounted for 47% of agricultural land, 88% of which was grassland (INSEE, 2024). While grasslands play an important role in feeding herbivores, they also provide other ecosystem services (Michaud *et al.*, 2020). However, there has been a decline in these areas over the past several years: the decrease reached 33% for permanent grasslands between 1960 and 2010 (Puydarrieux and Devaux, 2013). Permanent grasslands are defined as areas covered with grass for more than five years, including semi-natural grasslands that are rich in floral diversity. This decline has continued in recent years due to the effects of severe weather conditions and the presence of pests (vole). The central Massif is a region of France with extensive grasslands, where several PDOs rely on permanent grasslands and their diversity to promote cheese and meat products. Maintaining these areas and grasslands in general is therefore a major challenge for quality-certified sectors, but also in terms of preserving a wide range of ecosystem services. The aim of this study is to analyse the reasons for and economic consequences of replacing grassland, considering the services provided by grasslands, namely carbon storage and floristic diversity.

Materials and methods

Farm surveys were conducted in the central Massif on dairy and beef farms. The selection criteria for farms were the diversity of structures and the involvement in the conversion of permanent or temporary grassland (completed or planned, and conversion or overseeding). To this end, the PDO

sectors provided a list of farmers, which was supplemented by contacts from farmers concerned by these issues in the field. Of the 21 farms proposed, 15 were surveyed. The other farms could not be surveyed because they did not meet the selection criteria (4), did not respond (1), or refused the interview (1). The surveys were conducted in April 2025 using an interview guide with open-ended and closed-ended questions. The interview guide was structured in four parts: presentation of the farm, description of the replacement project and motivation, costing of the replacement, and evaluation of the project.

To quantify the replacement cost, the partial budget method was implemented on each farm (Kay *et al.*, 2011). This involves quantifying the change carried out by replacing a grassland (calculating the additional costs, additional income, and reduced costs and income on the farm). The partial budget was calculated for one year for replacement with a crop (crop duration) and averaged over five years for replacement with temporary grassland in order to accurately calculate the cost of change in management of the grassland. The mixtures used for temporary grassland replacement are mainly commercial mixtures (non-local species) and adapted to the expectations of livestock farmers (between 5 and 12 species per mixture). A typology based on grassland replacement situations was performed. For each type of grassland, partial budget values were mentioned, as well as a quantification of floristic diversity and carbon sequestration services. Floristic diversity was estimated based on a typology of grasslands in the central Massif (Gaillot *et al.*, 2020), and the difference in diversity between the two situations was represented by a slider. Carbon sequestration was quantified using the CarSolEL tool (Chambaut *et al.*, 2021) for both situations and was represented by a slider.

Results and discussion

The study covered 15 farms in the central Massif, including 12 with PDO status, three organic farms, and one conventional farm. Fourteen farms had a dairy herd with an average of 65 dairy cows and an average milk production of 7030 l VL⁻¹ year⁻¹. Four farms had suckler herds, three of which were breeders with an average of 35 cows and one was fattening animals. The average size of the plot covered by the analysis was 4.3 ha. All the farmers surveyed were seeking to increase their self-sufficiency in fodder or concentrates on the farm, and four main reasons were given for ploughing up grassland: (1) unsatisfactory yield (9 farmers), with 5 farmers citing problems with drought or voles, (2) decline in pasture quality (5 farmers), (3) improvement of existing crop rotation on the farm (4 farmers), and rearrangement of crop rotation (3 farmers).

The typology performed according to grassland replacement practices showed 6 different types: 4 that described changes in permanent grasslands (more than 10 years) and 2 in less than 8 years old permanent grasslands. The replacement costs for these six types of grassland were positive for four types and negative for two types, with systematic favourable changes for young permanent grasslands intended to evolve and included in rotations. However, the economic gain was only significant when the yields of the original grassland were very low. Among the economically unfavourable changes is the overseeding of permanent grasslands. The partial budgets were calculated considering working time and material costs based on French references, but different results may arise in other countries.

Considering ecosystem services, a loss of floristic diversity was observed for types where the initial grassland was permanent. The difference in carbon storage variation was highly variable depending on the type. There was no difference in storage variation between the initial temporary grassland and the replacement situations. The positive effect on carbon sequestration observed for the conversion of

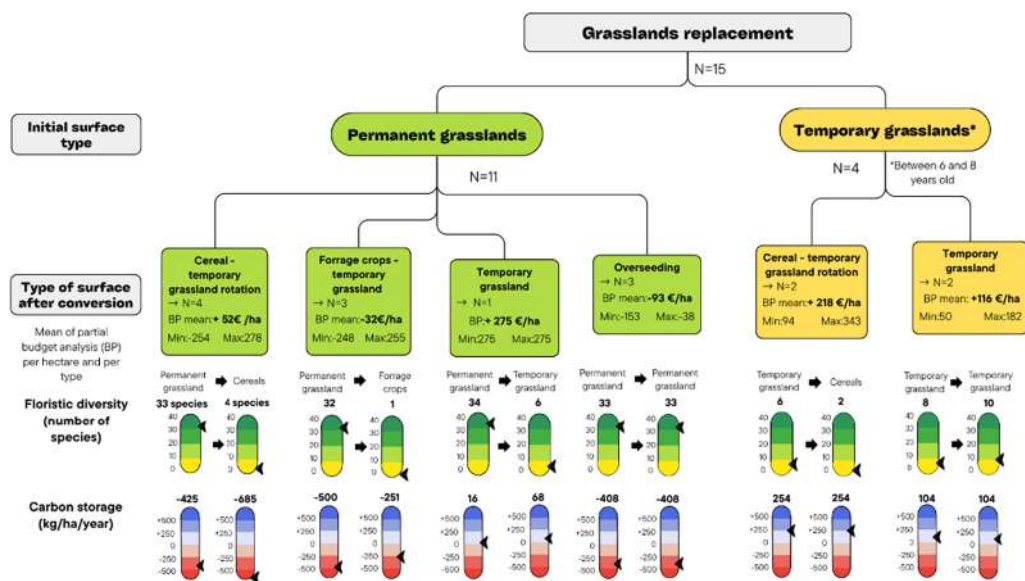


Figure 1. Typology of grassland conversion situations and representation of associated change costs and effects on provided services.

permanent grassland to temporary grassland were related to the use of permanent grassland (low use in the system). However, ecosystem services have not been considered in terms of economic cost.

Conclusion

This work has made it possible to quantify the replacement of grasslands, particularly for permanent grasslands. In four out of six cases, the replacement of grassland was favourable, but the gain remains small. On the other hand, when the ecosystem services provided by grasslands are considered, this shows a loss of services in the event of their replacement. This work highlights the importance of considering the services provided by grasslands in agricultural discussions, particularly within the chain sectors. More broadly, it calls into question the economic value of these services to farmers with a view to maintaining permanent grasslands.

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The potential of radar data for estimating forage quality in ley fields in Sweden

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Abstract

Assessment of pre-harvest forage quality is a key strategy for meeting the nutrient requirements of livestock. In this context, remote sensing-related models are an alternative to ensure continuous monitoring of the forage quality. Radar sensors such as the C-band Sentinel-1 (S-1) offer a potential data source that is not dependent on the weather conditions affecting optical remote sensing data. In this study, S-1 radar data were used to estimate forage quality variables from ley fields. Data collection was carried out at five different sites in Sweden over two growing seasons (2023 and 2024). A Random Forest model was used to estimate Neutral Detergent Fibre (NDF), using S-1 variables and rainfall data as predictor features. Various combinations of characteristics were tested on 70% of the dataset corresponding to 185 points. The trained model performance was validated using 30% of the dataset. The most accurate estimate of NDF was characterized by a correlation coefficient of 0.69 and a mean square error of approximately 41 g (kg dry matter (DM)⁻¹). The approach developed could be a potential solution to the lack of optical data in Nordic countries.

Keywords: forage quality, grasslands, leys, NDF, radar, Nordic countries

Introduction

In Northern Sweden, leys occupy more than 75% of arable land. Efficient ley management is a major challenge, as it depends heavily on the compromise between maximum availability of forage biomass in the field and high nutritional quality. Therefore, accurate information on forage quality is useful to help farmers to make decisions to harvest at the optimum time. Destructive sampling is generally used to estimate grass biomass and quality. The samples collected must then be analysed in the laboratory to measure variables, such as digestibility, crude protein and neutral fibre content (NDF), which have an impact on animal production. These measures are costly, time-consuming and effort intensive. Owing to remote sensing data potential to characterize the biophysical properties of vegetation, optical data are used for forage monitoring. However, this potential is limited by the presence of clouds, which contaminate pixel information. To address the weather-related limitations of optical sensors, several studies have focused on the use of radar data (Ali *et al.*, 2017). The launch of the Sentinel-1 (S-1) satellites, which operate in the microwave domain, enables robust approaches for monitoring grasslands using open access radar data with high temporal and spatial resolutions. The present study fits into this same context. We have explored the potential of radar data acquired by S-1 for estimating the quality of forage crops in ley fields in Sweden.

Materials and methods

To link radar data to forage quality and to cover the diversity of pedo-agroclimatic contexts in Sweden, in situ measurements were collected in five different counties of Sweden, namely Västerbotten, Jämtland, Uppsala, and Västra Götaland over two years (2023 and 2024), and Gotland only in 2024. At each site, two conventionally fertilized fields were selected and three harvests per year were planned. The fertilization amounts and times are different from one field to the other depending on the field management strategy. Before each harvest, three field missions were undertaken. The selected ley fields are botanically composed of grass (e.g. timothy, meadow fescue or ryegrass), legumes (e.g. red and white clovers or lucerne) and weeds. The leys' age can be up to 4 years, and they are mostly dominated by grass. One of the two fields in Gotland is irrigated and one of the fields in Västra Götaland is organic ley with a high percentage of clover. During each mission, to avoid working in areas with extremely dense vegetation or sparse vegetation, two 10-m Sentinel-2 pixels were selected based on their Normalized Difference Vegetation Index (NDVI) values where two 5-m long strips per pixel were harvested with a lawnmower. After harvesting, samples were weighed before and after oven-drying for 48 hours at a temperature of 60°C. Dried samples were analysed in a commercial laboratory using NIRS. The analysed NDF values varied between 348 and 687 g (kg DM)⁻¹. Processed C-band S-1 data delivered by Google Earth Engine platform were used to average the backscattering coefficients in VV and VH polarizations and the incidence angle per field and per date. The polarization ratio (PR = VH/VV) was computed per field. The final dataset, which contains 265 points, joined the in-situ measurements with the all available and nearest S-1 dates. In addition, precipitation data delivered by the Swedish Meteorological and Hydrological Institute (SMHI) were used. The gap between the S-1 acquisition dates and the sampling dates raised the question of which date to use for precipitation. For each field, the cumulative precipitation was computed to the date of S-1 acquisition and the date of sampling using different day-windows (3, 7, 10, 15 and 31 days). In the present study, random forest (RF) regressor was selected as a machine learning model to estimate NDF. The dataset was randomly split into 70% for model training and 30% for validation. The model's features can be related to the radar information and the sensor configuration backscattering coefficients in VV or VH, PR, incidence angle, the relative orbit number and its direction in addition to the precipitation accumulated to the date of S-1 and the sampling date using the different day-windows. To evaluate the established model performance different statistical metrics are used such as the mean absolute error (MAE), root mean square error (RMSE), bias and Pearson correlation coefficient (*r*).

Results and discussion

For each field and sampling date, we retrieved the NDF values using the trained RF model. To evaluate the model's performance and validate it, we compared the NDF values estimated using the model with the average in-situ NDF per field and sampling date and computed MAE, RMSE, Bias and *r*. Figure 1 illustrates the statistical metrics of the validation, as a function of the combination of features. The MAE values ranged from 33 to 48 g/kg of dry matter (DM). The RMSE values ranged between 41 and 57 g (kg DM)⁻¹, respectively. In addition, the bias values ranged between -10 and -2 g (kg DM)⁻¹, and correlation coefficients ranged from 0.08 to 0.69. The lowest accuracy characterizes the use of VV-backscattering coefficient as a feature (MAE = 48.16 g (kg DM)⁻¹, RMSE = 56.77 g (kg DM)⁻¹, bias = -3.29 g (kg DM)⁻¹ and *r* = 0.08). The use of VH and PR improved NDF retrieval accuracy where the RMSE decreased 6% and 3% relative to VV-RMSE, respectively. The statistical metrics of combination 7 (Figure 1) highlight the improvement of the NDF estimation with the introduction of radar acquisition data such as the incidence angle, the relative orbit number and its direction with

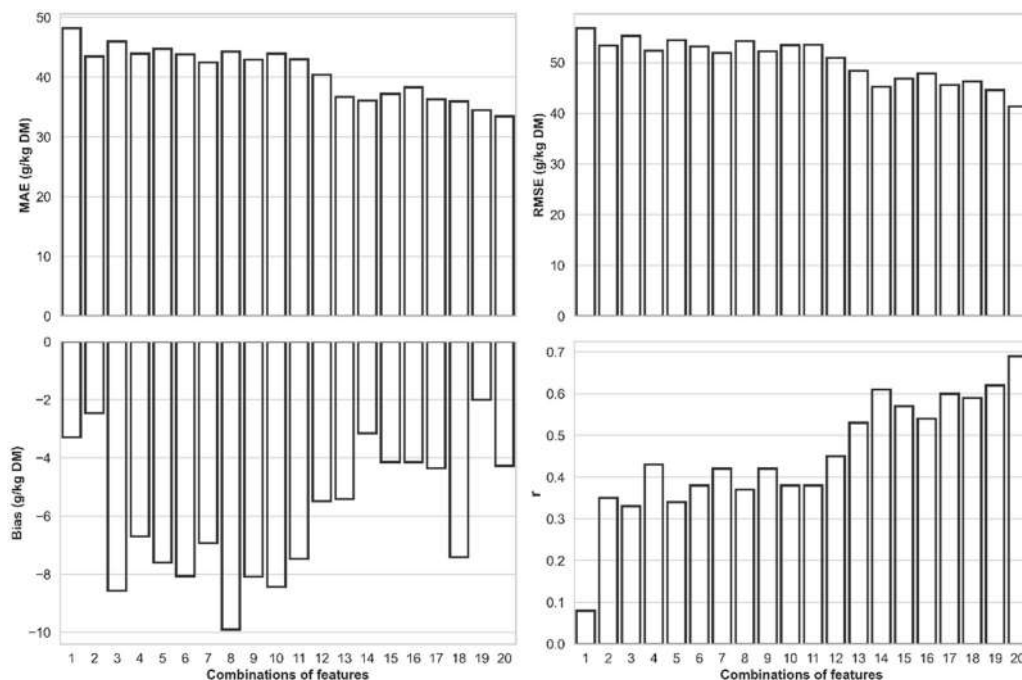


Figure 1. Combinations of features for NDF estimations 1, VV; 2, VH; 3, PR; 4, PR and incidence angle; 5, PR, incidence angle and orbit (relative number and direction); 6, PR, incidence angle and VH; 7, PR, VH, incidence angle and orbit (relative number and direction); 8, PR, incidence angle, VH and VV; 9, PR, incidence angle, VH, VV and orbit (relative number and direction); 10, PR, incidence angle, VH, VV and orbit (relative number); 11, PR, incidence angle, VH, VV and precipitation (S-1); 12, combination of 7+precipitation at the date of S-1 acquisition; 13, combination of 12+3-day cumulative precipitation to the date of S-1 acquisition (S1-3Days); 14, combination of 12+(S1-7Days); 15, combination of 12+(S1-10Days); 16, combination of 12+(S1-15Days); 17, combination of 12+(S1-31Days); 18, combination of 17+precipitation at the date of sampling; 19, combination of 14+precipitation at the date of sampling; 20, combination of 19+7-day cumulative precipitation at the date of sampling.

the VH and PR data. The correlation coefficient reached 0.42 and the MAE decreased to $42.45 \text{ g (kg DM)}^{-1}$. Although the inclusion of radar acquisition characteristics improved the accuracy, the precipitation data contributed to increasing the R to reach 0.69 and reducing the MAE, RMSE and Bias to 33.43 , 41.34 and $-4.27 \text{ g (kg DM)}^{-1}$, respectively. These metrics correspond to the consideration of the cumulative and the precipitation at the dates of S-1 and the sampling in tandem with the radar data as features. The results highlight the potential of VH and PR radar variables as descriptors of plant dynamics, in contrast with the VV backscattering coefficient, which is mainly influenced by soil-related factors rather than crops (Baghdadi *et al.*, 2017). The consideration of the incidence angle in combination with PR slightly improved the correlation coefficient of the NDF estimations. Since the incidence angle is linked to the sensor configuration, adding the orbits' relative data increased the correlation coefficient and simultaneously decreased the RMSE and MAE values. The radar signal is impacted by soil and vegetation water content; in the absence of irrigation, precipitation can govern the variability of soil and vegetation moisture. Consequently, including precipitation and its cumulative values as features enhanced the accuracy of NDF estimations.

Conclusions

In the present study, NDF estimation was performed using a random forest algorithm with different feature combinations including Sentinel-1 and precipitation data. The results demonstrate the potential of radar data for estimating NDF, highlighting its ability to overcome the weather dependency of optical data. In further steps, the potential of microwave data will be explored to estimate other forage quality variables such as organic matter digestibility and crude protein and using other weather variables such as temperature.

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A multi-stakeholder approach to discuss agroecological challenges in grazing-based systems

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Abstract

Within the project Grazing4AgroEcology, 31 Agricultural Knowledge and Innovation System (AKIS) meetings were held in eight European countries. The aim was to discuss the possibilities and limitations to adopting the best-practices captured. These multi-stakeholder AKIS meetings were moderated by trained facilitators using pre-defined methods. A two-step process was carried out: (1) identifying and prioritising the most relevant challenges and innovations at country level, (2) discussing them to co-create solutions to improve the current state and to identify associated policies, both on national and international level. The most-mentioned challenges were knowledge transfer and best practices exchange on manure management, technical grazing challenges, the mindset of stakeholders, biodiversity and animal health. The facilitators evaluated the meetings through a structured questionnaire. In general, the meetings were positively assessed. These findings indicate that a suitable methodological approach was applied to facilitate discussion and co-creation.

Keywords: agroecology, AKIS meeting, grassland, grazing, participatory research

Introduction

The implementation of innovation in agriculture is complex, requiring both technical and social competences from multiple fields of expertise. Innovations often exist in fragmented form among various stakeholders, who may hold differing perspectives or priorities. This can lead to misunderstandings and hinder progress. In grazing systems—in which economic, environmental, ecological, cultural, and social dimensions intersect—innovation typically demands collaboration across research, industry, practice, and policy. A multi-stakeholder approach is essential to align stakeholders, foster interaction, facilitate knowledge exchange and support the adoption of innovation. Multi-stakeholder meetings were organised throughout Europe within the framework of the Horizon Europe Thematic Network Grazing4AgroEcology (G4AE) to discuss the possibilities and limitations to adopting the best-practices captured and provide recommendations for further optimisation and transfer of innovations in the field of agroecology in grazing-based ruminant farming systems.

Material and methods

Within the project Grazing4AgroEcology (2022–2026), a total of 31 Agricultural Knowledge and Innovation System (AKIS) meetings were held until July 2025 in eight European countries (France, Germany, Ireland, Italy, The Netherlands, Portugal, Romania and Sweden). A diverse network of

Table 1. Overview of the main characteristics and of the evaluation of each type of AKIS meeting for prioritisation meetings (type 1), national meetings (type 2) and international meetings (type 3).

Type of meeting	Meeting duration (hours)	Number of participants	Number of participating stakeholder categories	Meeting evaluation				
				Overall impression	Networking	Knowledge transfer	Participants involvement	Atmosphere
1	3.5 ± 1.3	16.0 (7–40)	11 (5–8)	4.6 ± 0.5	5.0 ± 0.0	4.4 ± 1.1	4.5 ± 0.5	4.9 ± 0.4
2	4.0 ± 1.8	13.0 (6–136)	8 (2–13)	4.4 ± 0.5	4.4 ± 0.9	4.6 ± 0.6	4.5 ± 0.6	4.9 ± 0.3
3	6.3 ± 1.6	24.0 (11–257)	10 (6–15)	4.6 ± 0.5	4.9 ± 0.4	4.4 ± 0.8	4.4 ± 0.5	4.7 ± 0.5
Total	4.4 ± 1.9	16.0 (6–257)	9 (2–15)	4.5 ± 0.5	4.7 ± 0.7	4.5 ± 0.8	4.5 ± 0.6	4.8 ± 0.4
<i>P</i> value	0.012	0.077	0.159	0.558	0.100	0.944	0.913	0.606

For meeting duration and aspects of meeting evaluation (Likert scale from 1 = poor to 5 = excellent) mean ± SD are given, others are medians (minimum–maximum).

stakeholders participated in the meetings, including farmers, advisors, NGOs, researchers, and policymakers. Details about the grazing-related systems covered by Grazing4AgroEcology are provided in Fracchetti *et al.* (2026). AKIS meetings were moderated by trained facilitators using pre-defined methods. A two-step process was carried out: (1) identifying and prioritising the most relevant challenges and innovations at country level during simplified brainstorming sessions, (2) discussing them to co-create solutions to improve the current state and to identify associated policies in structured meetings. In the second step, the meetings could be either national or international. Here, individual innovations or challenges were discussed. The facilitators evaluated each meeting through a structured questionnaire of 78 questions in six categories (general information, participants, materials, methods, evaluation and follow-up). This included, among others, details on the duration, preparatory materials, discussion formats, and evaluations of the process of the meetings, expressed on a 5-point Likert scale. Differences due to the type of meeting were investigated by a Kruskal–Wallis test, followed by multiple comparisons according to Bonferroni in case of a significance ($P < 0.05$) of the main effect.

Results and discussion

The main agroecological challenges in grazing systems concerned knowledge transfer and best practice exchange on manure management, technical grazing issues, stakeholder mindset, biodiversity, and animal health. Manure management challenges focused on reducing inputs, handling clover manure management, and adapting to changing regulations. Technical challenges included grass growth and intake, grazing systems and management, and economics. Biodiversity-related issues involved increasing multispecies swards and legumes—sometimes linked to climate change—managing biodiverse plots, and implementing biodiversity incentives. In animal health, concerns centred on negative grazing effects such as tympanites. Most topics aligned with the five agroecological pillars of animal systems according to Dumont *et al.* (2013): improve animal health, reducing inputs, reducing pollution, enhancing diversity within animal production systems and preserving biodiversity. Stakeholder mindset was a complementary aspect, emphasising the need to sustain farmer motivation, autonomy, value, and awareness of management practices. The type of meeting affected only the meeting duration, which was on average 4.4 ± 1.9 hours (Table 1).

In this regard, the international AKIS meetings with a duration of about 6 hours were found to last longer than the prioritisation and national AKIS meetings ($P = 0.01$ and $P < 0.01$, respectively), which had lengths of around 3.5 or 4 hours. All other investigated features were unaffected by the type of meeting. In general, the meetings were positively assessed. None of the aspects that described the evaluation of the meeting was impacted by the type of meeting, as perceived by the facilitators. Farmers, researchers, advisors and educational institutions were the most strongly represented category

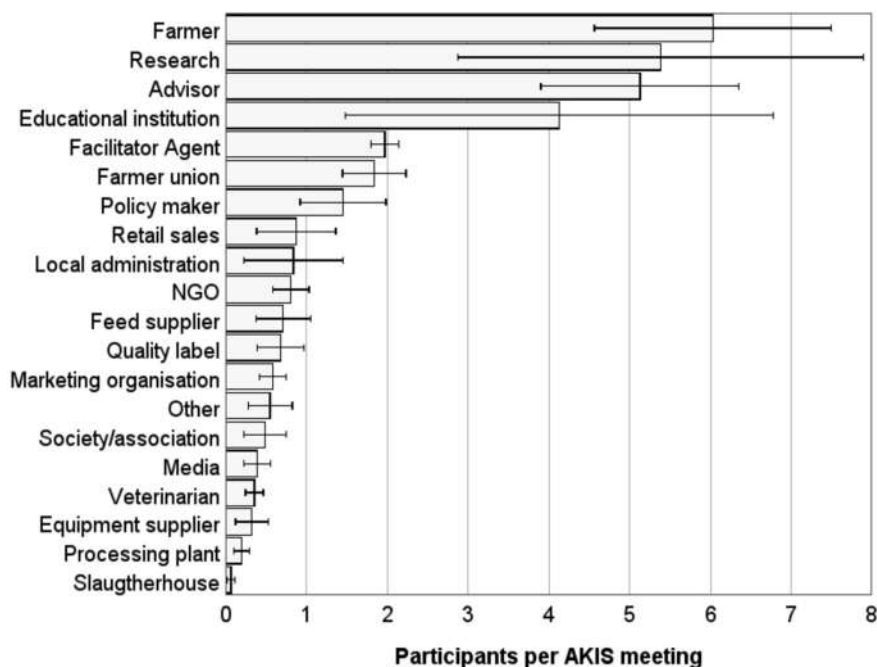


Figure 1. Mean $\pm$ SE of the participants per AKIS meeting and stakeholder category.

at the meetings (Figure 1). The mean numbers of 16 participants per meeting from nine stakeholder categories are well in line with the values exhibited by the multistakeholder discussion groups held within the Horizon 2020 Thematic Network Inno4Grass (Peratoner *et al.*, 2019). However, the large variation observed for the single meetings shows that context-specific adaptations were implemented by the organisers.

Conclusion

A multi-stakeholder approach using different types of AKIS meetings was applied to discuss agroecological challenges in grazing-based systems. The type of meeting did not affect the evaluation of the meetings, which were positively assessed. This suggests a suitable methodological approach for all meeting types to promote discussion and co-creation. Apart from meeting duration, there were no differences in characteristics of different meeting types.

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Effect of sward type on the estimation accuracy of yield-dependent variables through multispectral measurements

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Abstract

Models predicting forage yield based on multispectral measurements seldom take the botanical composition into account. This study investigates the impact of a simplified assessment of the botanical composition based on the yield proportion of functional groups on the accuracy of predictive models based on multispectral data ($n = 480$) to estimate dry matter yield, protein yield, and energy yield of permanent meadows in South Tyrol (NE Italy). Using a two-year database, we developed distinct predictive models employing multispectral proximal sensing mimicking the Sentinel-2 bands. Forage samples were visually classified into three different sward type compositions (rich in grasses/rich in forbs/balanced). We further built a random subset of 121 samples for each sward type to be used to develop sward-type-specific models. Seven modelling strategies using Partial Least Squares Regression have been developed. Similar accuracy was achieved for global and sward type-specific models, but the highest accuracy was obtained for single sward type-specific models, depending on the dependent variable.

Keywords: botanical composition, yield proportion, machine learning, multispectral data

Introduction

Forage yield, as well as parameters combining the information of yield and forage quality, such as crude protein yield or energy yield, are among the most important factors for grassland farmers. The choice of harvest time typically involves balancing forage quality and yield, because the crude protein concentration declines with sward age (Michaud *et al.*, 2011). Conventional methods for estimating yield are labour-intensive and often insufficient in fully capturing spatial heterogeneity. Multispectral remote and proximal sensing offers a scalable and efficient solution, but many models do not take the influence of species composition on spectral signals into consideration. We expected grasses, forbs, and legumes to possess distinct structural and spectral profiles. This study explores whether accounting for these functional-group proportions can enhance the prediction accuracy of dry matter yield, metabolisable energy yield, and protein yield using multispectral data derived from hyperspectral signatures resampled to Sentinel-2 bands.

Materials and methods

The study took place at nine experimental sites (90 plots in total, ranging in elevation between 725 and 1716 m asl), located on different meadows, where the effect of management on forage quality and yield of permanent meadows in South Tyrol (NE Italy) was investigated. Their mowing frequency ranged from 2 to 4 cuts year⁻¹, coupled to a total nitrogen input from 0 to 200 kg total N ha⁻¹ year⁻¹, resulting in a species richness of vascular plants between 22 and 52 per plot. Data were collected at plot level and, just before each cut, the sward type was visually assessed based on the yield proportion

of grasses, forbs and legumes: G (rich in grasses, >70% grasses), B (balanced, 50–70% grasses), and F (rich in forbs, grasses <50%, legumes <50%). Observations on swards rich in legumes were removed from the dataset as they were very infrequent. Multispectral reflectance data were captured just before mowing using a portable spectroradiometer (RS-3500, Spectral Evolution, Haverhill, MA, USA) and then resampled to the 13 spectral bands of Sentinel-2. Forage samples were cut at a stubble height of 5 cm and dried at 60°C to a constant weight then analysed for crude protein concentration (CP). Metabolisable energy (ME) was estimated according to Arrigo et al. (2021). Crude protein yield (CPY) and metabolisable energy yield (MEY) were calculated as the product of dry matter yield (DMY) and CP or ME, respectively. Our final dataset consisted of 480 observations (124 of G, 121 of B, and 235 of F). Seven modelling strategies were implemented: (1) Global-Global_Equal_prop: global model trained on all 84 observations (28 for each sward type) and validated on 37 observations of all sward types in equal proportions; (2) Global-Global_diff_prop: global model trained on 84 observations and tested on 37, both based on the sward type proportions present in our complete dataset (F = 49%, G = 26%, B = 25%) (3) Global-individual_84: general model of 84 samples trained with equal sample sizes per sward type and validated separately on 37 observations per type; (4) Individual-Individual (balanced): models trained and tested within each sward type separately; (5) Global-individual_250: the models were trained on 250 samples consisting of 70% of the individual datasets for each sward type and tested on each sward type (30%); (6) Global_Global_all_data (proportional): global model trained on all available data with proportional sward-type representation, validated on a mixed test set; (7) Global_individual_all_data: models fitted and validated all on available data across the entire dataset. We followed two approaches to data set creation. As the B category had the limiting sample size ($n = 121$) and we wanted to keep equal observations for each sward type to prevent the sample size from affecting the accuracy, we sampled 121 observations also from the G and F categories using stratified sampling based on experimental field and growth cycle (approaches 1–5). For approaches 6 and 7, the whole dataset ($n = 480$) included different proportions of the sward types. Partial Least Squares Regression (PLSR) models were developed to predict DMY, CPY and MEY (observed range: 0.1–6.6 Mg ha⁻¹, 81.8–524.5 kg ha⁻¹ and 0.8–61.2 GJ ha⁻¹, respectively) from the Sentinel-2 spectral bands. Data was split into a training (70%) and a testing set (30%). On the training set, the optimal number of components for each PLSR model, yielding the lowest RMSE, was determined through three repeated 5-fold cross-validations, each using different random fold subsets. The model was then fitted to the complete training set, which was used to predict the test set observations. To reduce the effect of the limited dataset size and the variability caused by random selection of training and test set observations, each modelling strategy was repeated 30 times with different random subsets. All analyses were conducted in R (version 4.3.1) using the *caret* and *pls* packages.

Results and discussion

DMY prediction using Sentinel-2-like multispectral bands had R^2 values ranging from 0.50 to 0.71 based on training approach and sward type. Sward type-specific models for B and F had the best accuracy in terms of R^2 and RMSE; type G always had the worst performance (Table 1). The constant performance of global mixed-type models (Models 1, 2, 6) with minimal accuracy loss ($R^2 = 0.60$ – 0.67) in comparison to the most accurate sward type-specific models supports their usage in situations when the composition of the sward is uncertain. The model performance was, in general, much lower for CPY ($R^2 = 0.26$ – 0.53). The highest R^2 -values (0.53) were achieved by types B and F in the global-individual-all-data approach. Again, for B and F, sward type-specific models increased the accuracy of CPY, but not for G. F exhibited the lowest accuracy for the sward type-specific models, whereas B had the highest. Similar trends were detected for metabolisable energy yield (MEY), with type B always

Table 1. Mean R^2 and RMSE for DMY, CPY, and MEY for models adopting different training–testing setups

Model Nr.	Model	Sward type	DMY		CPY		MEY	
			R^2	RMSE	R^2	RMSE	R^2	RMSE
1	Global-global_equal_prop		0.60	0.72	0.37	81.0	0.35	6553
2	Global-global_diff_prop		0.61	0.65	0.33	85.9	0.47	5847
3a	Global-individual_84	B	0.58	0.69	0.39	74.2	0.51	5844
3b		G	0.50	0.87	0.44	90.4	0.44	7571
3c		F	0.66	0.59	0.26	76.1	0.38	5527
4a	Individual-individual	B	0.63	0.65	0.52	67.8	0.57	5504
4b		G	0.53	0.81	0.51	90.0	0.45	7468
4c		F	0.70	0.56	0.29	75.4	0.49	5064
5a	Global-individual_250	B	0.64	0.66	0.52	66.7	0.59	5412
5b		G	0.53	0.82	0.48	91.4	0.49	7272
5c		F	0.71	0.56	0.33	73.3	0.50	4977
6	Global_global_all_data		0.67	0.65	0.52	74.4	0.54	5813
7a	Global_individual_all_data	B	0.64	0.65	0.53	68.4	0.63	5333
7b		G	0.56	0.79	0.49	86.3	0.46	7084
7c		F	0.70	0.60	0.53	70.0	0.55	5379

DMY, dry matter yield (t ha^{-1}); CPY, crude protein yield (kg ha^{-1}); MEY, metabolizable energy yield (MJ ha^{-1}); R^2 , coefficient of determination; RMSE, root mean squared error; G, rich in grasses; B, balanced, F, rich in forbs.

showing the highest accuracy and G the lowest, with intermediate accuracy levels between DMY and CPY. For the sward type-specific models, the highest accuracy was achieved by B, and the lowest by G.

Conclusion

Overall, the results demonstrate that Sentinel-2-like multispectral data can provide reliable predictions of forage yield and yield-related traits in permanent meadows. While type-specific calibration often leads to the highest accuracy, balanced or fully trained datasets can deliver similarly strong results, making them practical for operational applications where detailed botanical composition is not known in advance.

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Tannin content in sainfoin (*Onobrychis viciifolia*) as a response to mixture composition

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Abstract

Onobrychis viciifolia Scop. (s Perly (OV1) or Visnovsky (OV2), sainfoin) is a forage legume regaining interest in sustainable grassland systems due to its multiple agronomic and ecological benefits. Beyond nitrogen fixation and pollinator support, sainfoin is particularly valued for its tannins, which possess anthelmintic properties that benefit livestock health and reduces methane emissions. As secondary metabolites involved in stress response, tannin levels may be influenced by plant intraspecies competition within mixtures. This study evaluates how sainfoin proportion in grass-legume mixtures affects its tannin content across two varieties, (Perly, OV1; Visnovsky, OV2), under field conditions. Linear modelling revealed a significant negative relationship between legume proportion and tannin content, indicating that lower sainfoin cover—implying higher competition from grasses—may stimulate tannin production. OV2 consistently showed lower tannin levels. Total yield was also negatively related to legume cover, especially in OV1, supporting the interpretation of competition-driven stress responses. These findings underline the importance of mixture composition in functional forage quality.

Keywords: forage quality, grass, legume mixtures, sainfoin, tannins

Introduction

Mixed grass–legume swards offer multiple agronomic and ecological benefits (Mueller-Harvey *et al.*, 2019), including atmospheric nitrogen fixation (Malisch *et al.*, 2017), high yields and forage quality. Sainfoin is highly palatable, and its tannin content has been shown to have a positive effect, providing anthelmintic properties that promote animal health (Manolaraki *et al.*, 2010). Because tannins are involved in plant stress responses (Barbehenn *et al.*, 2011), their concentration may vary with interspecific competition in mixtures. Despite sainfoin's potential, there is limited understanding of how interactions in mixtures affect its tannin expression, particularly under varying proportions in grassland systems. This study aims to investigate the effects of the proportion of sainfoin on its tannin content when grown in mixture with grasses and considering variety-specific responses.

Materials and methods

The field experiment was established in 2024 as part of the LEGENDARY Horizon 2020 project, on an arable, moderately deep soil, located in Switzerland at 440 m asl. The seed mixtures were targeting sainfoin proportions at establishment of either 100, 60 or 30%. Two varieties, Perly (OV1) or Visnovsky (OV2), were used. Grasses were *Arrhenatherum elatius* and *Festuca arundinacea*. The design followed a randomised complete block with four replicates. A visual assessment was performed to identify the actual species proportions. Sainfoin proportions varied widely across the field plots: OV1 averaged 82% (50–99.8%), OV2 averaged 74% (5–100%). Due to the inconsistency between the sown and the achieved proportions, sainfoin proportion of each plot was used as explanatory variable (Figure 1).

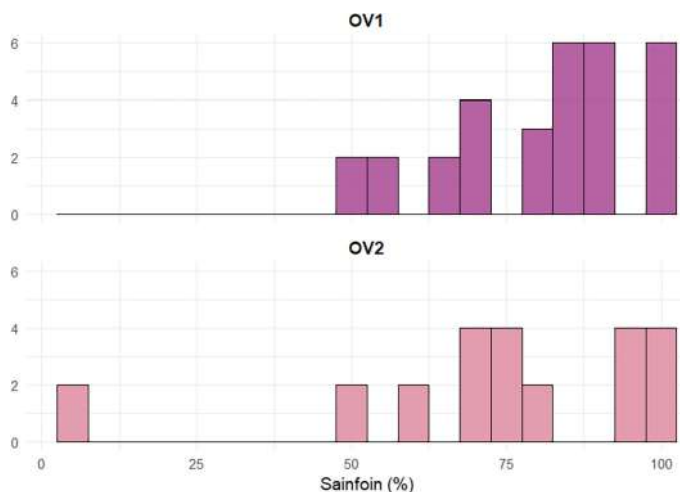


Figure 1. Histogram displaying the proportion of sainfoin by variety: Perly (OV1), Visnovsky (OV2).

In Summer 2025, five individual plants of sainfoin were collected per plot, at a height of 5 cm and at BBCH 65, just before the first harvest. Samples were dried (50°C, 48 h) and ground (0.5 mm sieve). Total yield was recorded in each plot at harvest.

Extraction methodology was based on Yuliana *et al.* (2014), by adding 10 ml of 75% methanol solution to 500 mg of dried (50°C) vegetal grounded material, and using sonication and centrifugation. Total phenolic compounds and tannins were quantified following the Folin–Ciocalteu method (Makkar, 2003). All readings were conducted in duplicate.

R software was used to analyse the data. A linear model was used to study the relation between sainfoin proportion and tannin concentration; for that, the variable was scaled to centralise data.

Results and discussion

The relationship between sainfoin proportion and tannin concentration exhibited differing trends between varieties (Figure 2). The linear model results (Table 2), indicates a significant negative effect of sainfoin proportion on tannin concentration ($p = 0.017$), especially in the OV1 variety. It suggests that tannin production is decreased at higher sainfoin proportion because of lower stress due to competition by less grasses.

Additionally, OV2 variety had significant lower tannin content overall compared to OV1 ($p < 0.001$), indicating variety differences in tannin production, which is in concordance with Malisch *et al.* (2025). Furthermore, the interaction between sainfoin proportion and variety was not statistically significant ($p = 0.18$), despite the apparently steeper slope for OV1 in Figure 2. Table 2 shows higher sainfoin proportions significantly ($p = 0.007$) decrease the yield (since grass often contributes more to yield). Eventually, plots with lower legume proportion (and thus higher grass proportion) showed both higher yields and higher tannin concentrations in sainfoin. This suggests that inter-specific competition may induce the synthesis of tannins as a stress response. This aligns with previous studies indicating that secondary metabolite production in legumes can be modulated by biotic stress (Barbehenn *et al.*, 2011).

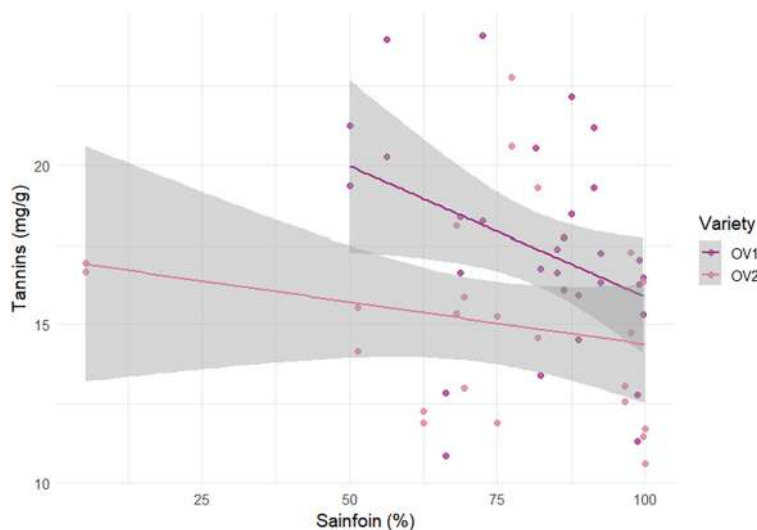


Figure 2. Tannin content linear regression model in sainfoin by variety.

Table 1. Summary of results from linear regression models of sainfoin proportion and variety on tannin content and yield.

	Predictor	Estimate	SE	<i>p</i> -value
Tannin content (g (kg DM)^{-1})	Intercept (OV1)	17.51	0.31	<0.001
	Sainfoin proportion (scaled)	-1.12	0.44	0.017
	Variety OV2	-2.51	0.47	<0.001
	Proportion $\times$ OV2	0.72	0.52	0.180
Yield (kg DM m^{-2})	Intercept (OV1)	0.31	0.23	0.179
	Sainfoin proportion (scaled)	-0.92	0.32	0.007
	Variety OV2	-0.58	0.34	0.107
	Proportion $\times$ OV2	0.72	0.38	0.068

p values <0.05 indicate statistical significance.

Since mixture composition may affect tannin production, this should be a factor to take into consideration for variety testing, since sainfoin plants have a high interest due to the tannin they contain and, according to this finding, mixture proportions can influence this. Future studies could benefit from a wider range of varieties and more balanced mixtures, to perform a more robust model. These results suggest that mixture composition influences both biomass yield and forage phytochemical quality.

Conclusion

Overall, these findings suggest that mixture composition not only affects biomass production but also the phytochemical quality of forage. These findings are relevant for designing multi-species grasslands targeting both yield and functional quality.

Acknowledgement

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Considering the impacts of temporary grasslands on the land use ratio of dairy cow farms

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Abstract

Ruminants allow the use of agricultural land not suitable for growing staple crops. However, intensive ruminant production systems often rely on feedstuff produced on arable land. The land use ratio (LUR) refers to the human-edible plant protein potential (PPP) on the land area used for feed production divided by the human-edible animal-sourced proteins (ASP) on the same land area. We applied the LUR concept on 86 Swiss dairy farms to demonstrate the effects of temporary grasslands. We first analysed how the proportion of temporary to total grassland used by the dairy farms affected PPP, ASP and the LUR. Second, we calculated PPP and the LUR of all farms for the underlying crop rotation with and without temporary grasslands. The results indicate that the greater the proportion of temporary to the total grassland area used by the farms was, the higher were PPP, ASP and the LUR. Incorporating temporary grassland in the underlying crop rotation scenario used to calculate the PPP reduced the LUR proportionally. Our findings highlight the importance of accurately considering the role of temporary grasslands in the context of feed-food competition.

Keywords: animal sourced protein, crop rotation, feed, food competition

Introduction

Ruminants allow the use of agricultural land not suitable for crop production as they transform fibres inedible for humans into animal sourced food. However, high-yielding ruminant production systems often rely on feedstuff produced on arable land and may thus compete with staple crop production (Ineichen *et al.*, 2023). This may be the case when concentrates or maize are being fed, but it also applies to temporary grassland, where roughage, typically grass or grass-clover, is grown on arable land. To assess the land-use efficiency of livestock production systems, van Zanten *et al.* (2016) developed the concept of the land-use ratio (LUR). Here, we investigate the impact of temporary grasslands on the LUR of Swiss dairy cow farms. We analysed the influence of the proportion of temporary grassland to total grassland used by the farms on the productivity of animal sourced protein (ASP) and the LUR at farm level, as temporary grassland often yields more dry matter and protein per m² than permanent grasslands. We further demonstrate the effects on the LUR, if temporary grassland was incorporated in the model crop rotation to calculate the plant protein potential (PPP) of the area used for dairy production.

Materials and methods

Data of 73 conventional and 13 organic Swiss dairy farms located in the lowland ($n = 35$), the hill ($n = 22$) and mountainous zone ($n = 29$) according to the Swiss regulation on agricultural production zones was collected for 2021. The LUR was calculated according to equation 1:

$$\text{LUR}_{\text{protein}} = \frac{\sum_{i=1}^n (\text{LU}_i \times \text{mPP})}{\text{ASP}} \quad (1)$$

Table 1. SD of the random effects in the linear mixed-effects model to assess the impact of temporary grassland on human-edible animal-sourced protein (ASP), human-edible plant protein production potential (PPP) in grams of crude protein (CP) per m² of 86 Swiss dairy farms.

Dependent variable	Effect	SD
ASP (g CP per m ²)	Zone	4.82
	Production form	6.62
	Residual	6.55
PPP (g CP per m ²)	Zone	13.60
	Production form	13.71
	Residual	12.46
Land-use ratio	Zone	0.27
	Production form	0.23
	Residual	0.36

where LU is the land (m²) used to produce feed i ($i = 1, \dots, n$) and mPP is the maximal human edible plant protein yield (g CP m⁻²) on that land. It was calculated applying a model crop rotation designed to maximize plant protein production, depending on soil and climatic suitability on each site. If the land was unsuitable for crop production due to climatic or soil characteristics, the mPP for the respective feed (grassland) was zero. For bought feeds, assumptions about climatic and soil suitability for crop production were made. The sum of all products ($Lu_i \cdot mPP$) is referred to as PPP (g CP). It was divided by the human edible ASP (g CP of milk and meat output) on the land used for feed production to get the LUR. A value above one indicates that the land used for dairy production would have yielded more human edible protein if used for staple crop production. Details are described in Ineichen et al. (2023). However, slight modifications were implemented. The model crop rotation in the BASE scenario consisted of wheat, soy, potatoes and rapeseed for ‘warm’ Swiss climate conditions, whereas soy was replaced by faba beans for ‘cold’ conditions. In the GRASS scenario, two years of temporary grassland were added to the model crop rotation and thus mPPP was reduced to 2/3 of the BASE scenario. Crop yields of organic farms were reduced to 0.61 (rapeseed), 0.69 (wheat) 0.70 (potatoes), 0.74 (soy) and 0.83 (fava beans) of conventional crop yields (Zumwald et al, 2019). Further, land use of replacement stock (heifers and calves) was computed based on standard rations and yields of the respective crops. Differences in the amino acid profile of plant and animal sourced proteins and hence the protein quality for human nutrition were not considered. A linear mixed-effects model using the *lmerTest* package was applied in R Studio version 4.3.3 (R Core Team, 2025) for the BASE crop rotation scenario with the proportion of temporary grassland in the total grassland as fixed effects. As the production zone (lowland, hill, mountain) and form (organic, conventional) were not the main focus of the analysis, they were treated as random factors. Normality of residuals was checked visually. The effect of the crop rotation scenario was not subject to inferential statistics, as the GRASS scenario decreases PPP of all farms proportionally and the effects in the mixed model remain the same.

Results and discussion

The random effects production form (organic or conventional) and zone (lowland, hill, mountain) both explained variation in the magnitude of the remaining model residual for all dependent variables (Table 1). Hence, they are important elements to model the impacts of the use of temporary grassland on land-use efficiency under Swiss conditions.

As hypothesized, the human edible ASP increased with the proportion of temporary to total grassland used by each farm (Table 2). This may be explained by increased productivity of temporary grassland

Table 2. Impact of the fixed effect temporary grassland (TG) on human-edible animal-sourced protein (ASP), human-edible plant protein production potential (PPP) and the land-use ratio (LUR). Temporary grassland is given as the proportion to total grassland used on 86 Swiss dairy farms.

Dependent variable	Effect	Estimate	SE	p-value
ASP (g CP m ⁻²)	Intercept	24.09	5.636	0.061
	TG	12.14	2.864	<0.001
PPP (g CP m ⁻²)	Intercept	21.06	12.780	0.225
	TG	19.86	5.476	<0.001
Land-use ratio	Intercept	0.79	0.238	0.057
	TG	0.36	0.159	0.024

on a given site, but it is not necessarily causal. There may be other effects interlinked with the proportion of temporary grassland present on the farms that influence ASP productivity, such as increased use of maize.

Temporary grassland is mostly suitable for crop production. Thus, we observed a strong increase in PPP with an increasing proportion of temporary grassland (Table 2). As the increase for PPP was higher than for ASP, the LUR (as the ratio of PPP/ASP) also increased.

In the BASE scenario, the PPP was 36.69 ± 21.269 g CP m⁻² and the LUR was 1.06 ± 0.322 . As temporary grassland in crop rotations may have positive impacts such as improved soil fertility, weed and pathogen suppression, nitrogen input substitution, water regulation or increased yield of subsequent crops (Hoeffner *et al.*, 2021), two years of temporary grassland were incorporated in the model crop rotation to calculate the PPP in the GRASS scenario. As the farms would still use the same land area for dairy production, the ASP was not influenced but PPP decreased by 33% to 24.46 ± 14.180 g CP m⁻². Consequently, the LUR also decreased proportionally by 33% to 0.70 ± 0.322 . Mottet *et al.* (2017) have pointed out that approximately 35% of the world's permanent grassland could be used for crop production. Here, the PPP of permanent grassland was also considered. However, transforming permanent grassland into cropland may have serious negative impacts on ecosystem services, such as on soil carbon storage or biodiversity (Schils *et al.*, 2022). Trade-offs with ecosystem services must be considered when assessing the potential to maximize land-use efficiency to the detriment of grassland, be it temporary or permanent.

Conclusion

Relying on temporary grasslands to feed dairy cows is linked to increased productivity of ASP per m² but even stronger to increased land-use competition (LUR). If the benefits of temporary grasslands for ecosystem services are considered in the underlying crop rotation to compute the PPP, the LUR decreases.

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Zootechnical performances and methane emissions of permanent grasslands and by-products-based dairy to beef systems: ex-ante analysis and first year of systemic experimentation

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Abstract

In the Belgian Ardennes, 70% of the land is permanent grassland, and over 90% of the agricultural area is dedicated to feed production, with suckler cows being the primary output. However, beef production faces criticism due to its climate impact and low contribution to food security. The SPoT project addresses these concerns by exploring the sustainability of transitioning to mixed crop-livestock systems driven to optimize sustainable food production. Three long-term organic experimental systems with varying proportions of permanent grassland (70, 30 and 0%) were set up. Each system employs a similar 6-year crop rotation focused on food production, including cereals, cereal-legume associations, rapeseed, potatoes, and vegetables. To enhance beef production using grass and the available byproducts (bran, rapeseed cake, grade-outs, etc.) and mitigate climate impact, the fattening of dairy cross-bred bulls is considered in the systems with permanent grasslands. After two years of experimentation, this contribution will outline the opportunities for such animal production and highlight the challenges arising from its implementation relying on the systemic experiments.

Keywords: dairy-to-beef, feed, food, meat production, methane

Introduction

The European Green Deal highlights the growing societal expectations placed on agriculture, calling for a significant transformation of current production systems. The Walloon Central Ardenne region is characterized by a high density of suckler cow herds, which make use of local grasslands. However, these systems are increasingly scrutinized for their environmental footprint and the competition they pose between animal feed and human food through direct and indirect land-use.

In response to these challenges, assuming that circular crop-livestock systems (CLS) can support climate neutrality while optimizing food production, a systemic trial is currently underway, combining two innovative approaches. The first innovation prioritizes the use of arable land for direct human consumption, while livestock are fed exclusively with forage from permanent grasslands and agricultural by-products or downgraded crops. The second innovation consists of using crossbred dairy-to-beef males (CBDTB) for beef production, as suggested in Mertens et al. (2023). We proposed to test the feasibility and the interest of these two innovations on two CLS with different permanent grassland share. In this paper we focus on the zootechnical performances and the greenhouse gas emissions linked to the fattening of dairy-origin bulls, based on an *ex-ante* analysis, and on the performance observed during the initial years of experimentation.

Materials and methods

Two organic ICLS models were developed and implemented in Libramont (Belgium). These systems vary in the proportion of permanent grasslands (PG) and tilled land (TL): S1: 70% PG/30% TL, S2: 30% PG/70% TL. Both systems are implemented on the same experimental site. Each of the six crops is established in three replicates on plots that are subdivided into two equal parts, one for each system (S1 and S2). The crop plots have an average area of 0.31 hectares. Permanent grasslands are managed without true replication, but are spatially juxtaposed between system 1 (13.6 ha) and system 2 (2.8 ha). Each cropping system follows a six-year crop rotation including rapeseed, spelt, potatoes, onions, oat-pea mix, and rye-lentil. Livestock, raised under organic label, are CBDTB. Their numbers are determined based on an estimated grassland productivity of 7 t dry matter (DM) ha⁻¹, resulting in the annual purchase of 19 calves in S1 and 6 in S2. Calves are bought when 2–4 weeks old. They receive automated milk feeding for three months indoors in addition to downgraded crops and *ad libitum* hay. Animals were weighed every 21 days, and daily supplementation was recorded. In addition, indoors from November to April, animals were fed wrapped silage *ad libitum*, and grazing was monitored using a herbometer and quadrat sampling.

An *ex-ante* environmental assessment of the systems was conducted using the “DECiDE” tool (<https://decide.cra.wallonie.be/fr>). In this analysis, the primary crop outputs were considered edible for humans, leaving only by-products and downgraded crops for animal feed. Slaughter weight was set at 600 kg live weight at 21 months for S1 and S2. Methane emissions, expected to be the main source of greenhouse gases, were estimated by age group (0–6 months, 6–12 months, 12–24 months), based on intake capacity and the formula: $Y_m = 9.75 - 0.05 \times \text{Digestibility rate}$.

During the first two years of experimentation (2023–2024), crop yields were measured and analysed for technological and nutritional quality, leading to downgrading of some products, and sorted to quantify food production. By-products and downgraded crops were weighed and assessed for feed quality using near-infrared spectrometry. As half of the tilled land was still undergoing organic conversion, feed from these areas was substituted with equivalent quantities of commercial organic concentrates. Methane emissions were measured using a mobile Automated Head Chamber System, also known as GreenFeed(r). Each group had access to the system for 21-day periods, receiving up to 0.6 kg day⁻¹ (S1) and 1.2 kg day⁻¹ (S2) of concentrates. Data were averaged monthly when more than 30 valid visits (≥ 2 min) were recorded.

Results and discussion

After two years of trials, the actual availability of feed exceeded initial expectations, largely due to a higher-than-anticipated rate of crop downgrading—especially among cereals and crop associations (see Table 1). In S1, 95% of the available feed was successfully utilized. In contrast, feed consumption in S2 was limited, primarily due to a reduced number of animals (only 3 fattened instead of the planned 6) and the organic farming constraint that restricts concentrate use to a maximum of 40%.

Overall, animal performance aligns well with initial assumptions (see Fig. 1), with no significant differences observed between the two systems, despite the higher concentrate feeding rate in S2. Inter annual variation was observed. The average final weight of 2024–2025 cohort was 19 kg lower than the one from 2023–2024 cohort ($p = 0.017$). The impact of the bluetongue outbreak during summer 2024 and the subsequent vaccination campaigns in 2024 and 2025 remains unclear, warranting further years of experimentation for a more robust evaluation. Nutritionally, the combination of grass

Table 1. Observed production (in 10³ kg) of feed from crops

	Production in 2023–2024		Production in 2024–2025	
	S1	S2	S1	S2
Spelt downgraded (pre-germination)	1.57	1.97		
Downgraded rye (pre-germination, ergot)	2.00	1.87	1.60	1.09
Rapeseed cake	0.46	0.95		
Flour of mixed rapeseed grade-outs (2023+2024) and oat-pea (specific weight too low, 2024)			2.34	2.03
Commercial concentrates (non-organic downgraded spelt and oat-pea, rapeseed cake, potatoes grade-outs).	5.31	5.02	2.81	2.41

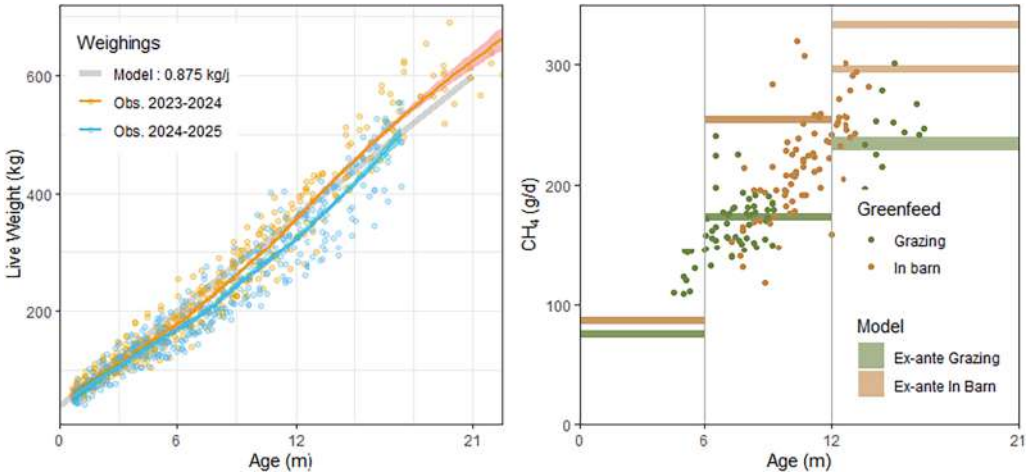


Figure 1. Weight (kg live weight) and methane emission rate ($\text{g animal}^{-1} \text{ day}^{-1}$) in function of the age of the crossbred dairy bulls (mainly Belgian Blue×Holstein) in barn (green) and during grazing period (brown). Each point is single animal measurement. The two modelled emissions between 12 and 21 months represent the expected impact of different byproducts quantities in S1 (bottom) and in S2 (top).

and crop by-products offered in the current systems does not provide a well-balanced ration for growing cattle, as the diet is consistently too high in protein and/or too low in energy, particularly affecting performance during the post-weaning phase (4 to 7 months). Once animals adapt to grazing, growth performance improves.

Methane emissions increased with age and body weight but showed no significant differences between the two systems. Grazing emissions were consistent with model predictions, while emissions recorded indoors were lower than expected. A small but statistically significant difference of 23 g CH₄ day⁻¹ was observed between barn and grazing conditions using a linear model. However, further data across the full age range are needed to validate this trend.

Conclusion

After two years of systemic trials, the first performances of dairy to beef crossbred bulls in two feed-no-food scenarios are described with contrasted share of permanent and tilled land. Zootechnical and methane emissions observations are compared with assumptions made in the *ex-ante* analysis. First performances observed are in agreement with the assumptions but questioned the performances of

these systems when less food crops will be downgraded into feed. These first results illustrate the key role of ruminants to recycle downgraded crops into foods with high nutritional and added value.

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The sum of temperatures as a decision-making tool for the management of permanent grasslands

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Abstract

Maximizing forage yield and nutritive value in grasslands is becoming challenging, particularly in the face of climate change. The sum of temperatures (i.e. growing degree days) is used as a proxy for estimating forage yield and quality, but the validity of this index at large scale remains unclear. We conducted an experiment in 23 intensively managed Swiss permanent grasslands in 2017, 2018 and 2019. At each site, 12 plots of 1 × 5 m were established and harvested after either 2, 4, 6, or 8 weeks from the start of the growing season with 3 replicates for each harvest time. Forage yields, crude protein and fibre contents were assessed, which allowed estimating digestibility, net energy for lactation (NEL) and protein digestible in the intestine (PDIN). Despite variability in the data, we found a strong positive relationship to the sum of temperatures for forage yield, but a strong negative relationship for digestibility, crude protein, NEL and PDIN. Our data suggest that in Switzerland, mowing between 650 and 750°C days provides the best trade-off between forage yield and nutritive value. Our findings show that the sum of temperatures could be integrated in decision-making tool for site-adapted management of permanent grasslands.

Keywords: digestibility, forage yield, mowing, nutritive value, Switzerland

Introduction

Considering the economic importance of intensively managed permanent grasslands, it is important to ensure high forage yield and nutritive value through optimized management. During the first growth cycle of temperate grasslands, both yield and nutritive value are mainly controlled by temperature, since precipitation and soil moisture remain relatively high early in the season (Perotti *et al.*, 2021). The use of the sum of temperatures as a proxy for forage yield was assessed by previous studies conducted in France (Ansquer *et al.*, 2004). However, the relevance of this index as a proxy for forage nutritive value remain unclear. The goal of our experiment was therefore to highlight the relationship between the sum of temperatures and both forage yield and nutritive value using data collected in intensively managed permanent grasslands in Switzerland.

Materials and methods

Our experiment was conducted for 3 years (2017, 2018, 2019) in 23 intensively managed permanent grasslands covering a wide gradient of pedoclimatic conditions, botanical compositions and altitudes (350–1000 m asl). At each site, 12 plots of 1 × 5 m were established and harvested after either 2, 4, 6, or 8 weeks from the start of the growing season, with 3 replicates for each harvest time. The grasslands were classified into 4 types (A, AB-BA, bA-Ab, Bb-bB) based on the dominance of the six main functional types of grasses (Cruz *et al.*, 2010). Grasslands classified as 'A' are dominated by *Lolium multiflorum*, *Lolium perenne* and *Poa trivialis*, 'AB-BA' by *L. perenne*, *Poa pratensis* and

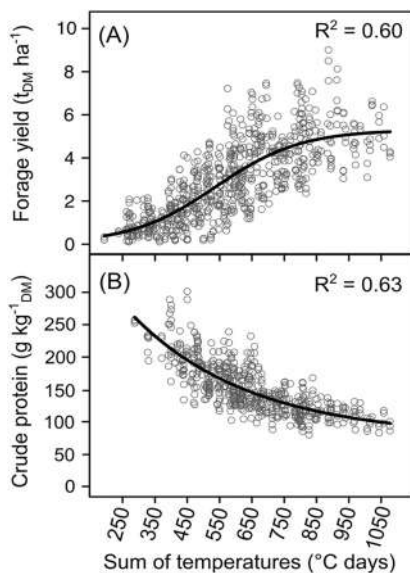


Figure 1. Relationships between the sum of temperatures and (A) forage yield and (B) crude protein.

Dactylis glomerata, 'bA-Ab' by *L. perenne*, *P. trivialis*, *L. multiflorum*, and *P. pratensis* and 'Bb-bB' by *P. trivialis*, *D. glomerata*, *P. pratensis*, *L. perenne*, *Agrostis capillaris* and *Elymus repens*. Forage yields, crude protein and fibre contents were assessed, which allowed estimating organic matter digestibility (OMD), net energy for lactation (NEL) and protein digestible in the intestine (PDIN) according to the formulas described in Daccord *et al.* (2021). The evaluation of the sum of temperatures (SumT) was adapted from Theau and Zerourou (2008). Briefly, SumT was calculated by cumulating daily mean temperatures starting on January 1st and constraining temperatures in the range between 0 and 18°C. Linear, logistic and exponential regression models were tested to express each forage variable as a function of SumT, selecting the model that best fitted the data. Significance of the regressions was assessed using ANOVAs with R (version 4.5.1).

Results and discussion

Forage yield was positively correlated with SumT (Fig. 1) during the first growth cycle, with a logistic model best fitting the data. Mowing between 650 and 750°C days provided between 3.69 and 4.4 t DM ha⁻¹ on average, which corresponds to a satisfactory yield level for permanent grasslands. The data suggest that to maximize forage yield during the first growth cycle the first cut should occur later, at SumT = 950°C days. This would provide an additional 0.7 t DM ha⁻¹, i.e., 5.06 t DM ha⁻¹ in total. It is important to note that the botanical composition of grasslands and the year had little influence on the linkages between forage yield and the sum of temperature. Indeed, the same forage yield was achieved only 50°C days earlier (i.e., few days) during the drier spring of 2018. Furthermore, the same yield was attained 100°C days earlier for grasslands type A and 100°C later for grasslands Type Bb-bB, the latter never producing more than 4.3 t DM ha

Table 1. Equations of the regression analysis between forage yield, nutritive value and digestibility and the sum of temperatures with respective values within the 550–950°C range.

Unit		Equation	R^2	Sum of temperatures (°C days)				
				550	650	750	850	950
Yield	t DM ha ⁻¹	$y = 5.28 / \{1 + e^{-0.0076(x-539)}\}$	0.60	2.75	3.69	4.40	4.83	5.06
CP	g (kg DM) ⁻¹	$y = 75.9 + 405.1e^{(-x/369.3)}$	0.63	167	146	129	116	104
OMD	%	$y = -0.028x + 95.248$	0.69	79.8	77.0	74.2	71.4	68.6
NEL	MJ (kg DM) ⁻¹	$y = 4.87 + 2.32 / \{1 + e^{0.0056(x-746)}\}$	0.60	6.6	6.3	6.0	5.7	5.4
PDIN	g (kg DM) ⁻¹	$y = 48.9 + 270.6e^{(-x/375.3)}$	0.63	111	97	86	77	70

CP, crude protein; OMD, organic matter digestibility; NEL, net energy for lactation; PDIN, protein digestible in the intestine. All models are highly significant with $P < 0.001$.

The sum of temperatures was a significant predictor of crude protein (Fig. 1, exponential model), OMD (Table 1, linear model), NEL (logistic model) and PDIN in forage (exponential model). Mowing intensively managed permanent grasslands between 650 and 750°C days corresponds to forage protein content ranging from 146 to 129 g kg_{DM}⁻¹, digestibility ranging from 77 to 74%, NEL ranging from 6.3 to 6 MJ (kg DM)⁻¹, and PDIN ranging from 97 and 86 g (kg DM)⁻¹, which is in line with the recommendations of optimal forage quality for ruminant nutrition (Agroscope, 2021). Planning the first cut later, at 950°C days, would, on average, reduce protein content to 104 g (kg DM)⁻¹, digestibility to 69%, NEL to 5.4 MJ (kg DM)⁻¹ and PDIN to 70 g (kg DM)⁻¹. The botanical composition of grasslands and the year had no significant impact on the linkages between forage nutritive value and digestibility and the sum of temperatures.

Overall, our results suggest that in Switzerland the optimal cutting time providing the best trade-offs between forage biomass production and digestibility and nutritive value during the first growth cycle in intensively managed permanent grasslands should occur between 650 and 750°C days. These results are quite different from those obtained in France by Theau and Zerourou (2008), which suggested a first harvest between 800 (early hay harvest) and 1200°C days (flowering hay). It is important to stress, however, that these latter authors only considered forage yield as target, while here we considered both yield as well as forage nutritive value and digestibility to estimate an optimal mowing time as a function of the sum of temperatures.

Conclusion

Findings from our study show that farmers must clearly define their priority (i.e., forage yield or nutritive value and digestibility) to decide on the ideal cutting time for a given forage yield and quality threshold. While mowing between 650 and 750°C days offers the best compromise between forage yield and nutritive value, farmers aiming to maximize yield should consider mowing up to 950°C days at the first growth cycle, and farmers aiming to maximize forage nutritive value should consider mowing at 650°C days. The botanical composition of the grasslands can also be considered for improved decision. Overall, findings from our study demonstrate that the sum of temperature appears as a valuable decision-making tool for site-adapted management of permanent grasslands across altitudes and pedo-climatic conditions.

Acknowledgement

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Effects of a biological silage additive on characteristics of a grass-legume silage

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Abstract

This study aimed to evaluate the effects of the biological silage inoculant Josilac classic, which contains the lactic acid bacteria (LAB) strains *Lactobacillus plantarum* and *Pediococcus pentosaceus*, on the chemical composition, fermentation characteristics, and aerobic stability of grass-legume silage. The evaluation was conducted using an in vitro mini-silo setup in accordance with DLG guidelines after 90 days of fermentation. LAB-inoculated grass-legume silage retained significantly higher levels of dry matter (DM) and DM corrected for volatiles (DMc), crude protein (CP), and water-soluble carbohydrates (WSC). It also exhibited a significantly higher total DM content and reduced DM losses after the 90-day storage period. The additive significantly improved key fermentation parameters, including a lower pH, increased total acid concentration, and higher levels of individual acids, particularly lactic acid. Simultaneously, it significantly reduced undesirable fermentation products, such as ammonia-N, alcohols, and butyric acid. Targeted inoculation not only accelerated acidification but also effectively suppressed aerobic microbial populations, thereby enhancing the aerobic stability of the silage.

Keywords: aerobic spoilage, bacteria strains, dry matter loss, forage preservation, grasses

Introduction

Despite implementing measures to prevent atypical fermentation, such as ensuring adequate compaction, maintaining optimal moisture content (30–35% DM), and minimizing oxygen exposure during storage, undesirable fermentation products and DM losses still frequently occur, particularly under farm-scale ensiling conditions. The application of LAB can help ensure controlled fermentation, reduce the risk of spoilage, and promote the production of high-quality silage with good aerobic stability (Carvalho *et al.*, 2020; Muck *et al.*, 2018). The objective of this study was to evaluate the effects of a biological silage inoculant containing the LAB strains *L. plantarum* and *P. pentosaceus* on the chemical composition, fermentation characteristics, and aerobic stability of grass/legume silage, with a focus on mixed forage systems where the combined activity of these strains has not been extensively characterized.

Materials and methods

The first cut of a perennial ryegrass (*Lolium perenne* L.), red clover (*Trifolium pratense* L.), and timothy (*Phleum pratense* L.) mixture was harvested using a mower-conditioner, wilted for 8–12 hours, and chopped under farm conditions by a forage harvester to a particle length of 2–3 cm. Five samples of the chopped forage were collected to establish baseline values for nutritive content, DM, pH, buffer capacity, and yeast and mould counts. Two forage piles, each weighing approximately 20 kg, were prepared and assigned to the following treatments: Control ©, water application at a rate of 4 ml/kg of fresh forage); and LP, application of a combination of *L. plantarum* LSI NCIMB 30083 1k20736, *L. plantarum* L2560 NCIMB 30084 1k20737, *P. pentosaceus* P11 DSM 23689 1k1011, *P. pentosaceus*

Table 1. Effect of inoculant treatment on the fermentation of ensiled grass-legume sward at day 90 of storage

Variable	C	LP	P
DM, g kg ⁻¹	336.3 ^b	345.2 ^a	0.004
DM*, g kg ⁻¹	348.3 ^b	355.7 ^a	0.010
Crude protein, g (kg DM) ⁻¹	151.1 ^b	157.0 ^a	0.019
WSC, g (kg DM) ⁻¹	17.7 ^b	31.2 ^a	0.013
NDF, g (kg DM) ⁻¹	490.7 ^a	477.0 ^b	0.004
ADF, g (kg DM) ⁻¹	295.1 ^a	282.6 ^a	0.069
Lactic acid, g (kg DM) ⁻¹	56.52 ^b	70.89 ^a	0.001
Acetic acid, g (kg DM) ⁻¹	14.99 ^a	13.89 ^a	0.292
Butyric acid, g (kg DM) ⁻¹	0.47 ^a	0.18 ^b	0.001
Propionic acid, g (kg DM) ⁻¹	0.45 ^a	0.43 ^a	0.771
Alcohols g (kg DM) ⁻¹	11.36 ^a	7.04 ^b	<0.0001
pH	4.40 ^a	4.27 ^b	0.0007
N-NH ₃ fraction, g (kg total N) ⁻¹	46.28 ^a	32.51 ^b	0.0005
DM loss, g kg ⁻¹	64.95 ^a	42.33 ^b	0.0004
Yeast, log ₁₀ CFU g ⁻¹	1.99 ^a	1.06 ^b	<0.0001
Moulds, log ₁₀ CFU g ⁻¹	1.74 ^a	1.00 ^b	<0.0001
LAB, log ₁₀ CFU g ⁻¹	7.23 ^b	8.84 ^a	0.0008
Clostridia, log ₁₀ CFU g ⁻¹	1.57 ^a	1.00 ^b	0.0001

* Dry matter corrected for volatiles.

P6 DSM 23688 1k1010, and the enzyme xylanase at 4 ml of suspension per kg of fresh forage to achieve a target inoculation rate of 6.0×10^5 cfu (g forage)⁻¹. The treated forage was packed into laboratory-scale silos (3 l glass jars) at a density of approximately 200 kg DM/m³ (using 1.6–1.7 kg of fresh forage per mini-silo). The jars were sealed with caps to prevent aerobic exposure and stored for 90 days to allow fermentation. The DM content was corrected for the loss of volatile compounds during oven drying. Statistical analysis was performed using the PROC GLM procedure in SAS 9.4 software. The model included treatment as the main fixed effect, with statistical significance declared at $P \leq 0.05$.

Results and discussion

The mean DM content was 366.0 g kg⁻¹ and CP and WSC concentrations were 153.4 and 90.1 g (kg DM)⁻¹, respectively. The herbage had a moderate buffer capacity (31.84 meq (100 g DM)⁻¹) and a pH value of 5.95. The calculated fermentability coefficient (FC) was also moderate (38.9). Consequently, this grass-legume crop was considered moderately difficult to ensile. The number of epiphytic lactic acid bacteria was 5.09 log₁₀ cfu g⁻¹ of pre-ensiled fresh forage. Moulds and yeasts were detected at levels of 4.20 log₁₀ cfu g⁻¹ and 5.24 log₁₀ cfu g⁻¹, respectively.

Inoculation resulted in several significant improvements in the quality of grass-legume silage compared to the control (Table 1). The inoculated silage retained 2.1–2.6% more DM and DMc ($P < 0.05$), along with 3.9% higher CP and 76% higher WSC content than the untreated control ($P < 0.05$). It also exhibited significantly greater overall DM retention, as reflected by higher total DM yields from the mini silos and 6.5% lower DM losses over the storage period ($P < 0.05$). Fermentation quality was markedly improved, demonstrated by a 0.13-unit reduction in pH ($P < 0.05$), increased total acid

Table 2. Aerobic stability results

Variable	C	LP	P
pH at the end of AST (14 days)	8.64 ^a	4.94 ^b	<0.0001
Fresh weight loss during AST, %	4.18 ^a	2.74 ^b	0.0007
Yeast after AST, log ₁₀ CFU g ⁻¹	7.34 ^a	4.85 ^b	<0.0001
Moulds after AST, log ₁₀ CFU g ⁻¹	9.00 ^a	5.45 ^b	<0.0001
Aerobic stability, h	186 ^a	256 ^b	0.001
<i>t</i> _{0.05} = 2.306; Error df = 8			

concentrations, and a 25.4% increase in lactic acid content ($P < 0.05$). In contrast, undesirable fermentation products such as ammonia-N, alcohols, and butyric acid were reduced by 23.9%, 38.0%, and 2.6-fold, respectively ($P < 0.05$). LAB are widely used as silage additives due to their ability to enhance lactic and acetic acid production, promote rapid pH decline, prevent organic matter decomposition, inhibit harmful microorganisms, and delay spoilage (Wang *et al.*, 2018; Fabiszewska *et al.*, 2019; Guo *et al.*, 2023). However, limited information is available on the combined effects of specific LAB strains. This study aimed to evaluate the impact of this novel multi-strain LAB–enzyme combination on silage fermentation dynamics and aerobic stability, providing new insights into optimizing microbial inoculant formulations for improved silage preservation. Furthermore, microbial analysis revealed a 22.3% increase in LAB populations, accompanied by reductions of 36.3, 42.5 and 46.7% in clostridia, mould and yeast counts, respectively ($P < 0.05$), indicating improved microbial stability and silage hygiene.

The treated silage exhibited 34.4% less fresh weight loss compared to the control ($P < 0.05$) (Table 2). Additionally, it maintained a pH that was 3.7 units lower at the end of the test ($P < 0.05$), along with 42.5% and 46.7% reductions in mould and yeast counts, respectively ($P < 0.05$). Most notably, the LP-treated silage demonstrated 2.9 days longer aerobic stability ($P < 0.05$), indicating enhanced resistance to spoilage under aerobic conditions.

Conclusion

Overall, the LP inoculant effectively improved fermentation quality, minimized dry matter and nutrient losses, suppressed yeast and mould growth, and extended the aerobic stability of grass–legume silage. It is acknowledged that the study did not include a comparison with other commercial inoculant products; however, the primary objective was to evaluate the efficacy of a specific multi-strain LAB–enzyme formulation (LP) under controlled conditions.

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Case study on behaviour and welfare of mature Norwegian Red heifers using virtual fencing

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Abstract

This study investigated the adaptation of ten Norwegian Red heifers to Monil virtual fence (VF) collars and assessed their welfare through heart rate monitoring. Over three days in a 4500 m² pasture, VF boundaries confined heifers to a designated section. Daily analysis of VF learning evaluated the frequency of auditory (#Audio) and electrical (#Elect) cueing. Heart rate data were analysed in 10-minute windows: before (Pre-Event), during (Event), and after (Post-Event) VF cueing. Analysis focused on the inter-beat interval (MEANIBI) and an estimate of heart-rate variability (RMSSD). Significant variations were observed in the daily number of cues (First-Day: 5.80 and 3.30, Second-Day: 6.30 and 4.30, Third-Day: 1.20 and 1.10 day⁻¹; $P=0.01$ and $P=0.01$ for #Audio and #Elect, respectively). The #Audio and #Elect were similar on the first and second days, with fewer on day three, suggesting animals adapting to VF cues. The MEANIBI and RMSSD (Pre-Event: 728.31 and 9.64, Event: 697.30 and 8.69, Post-Event: 756.33 and 9.90 ms; $P<0.01$ and $P=0.09$ for MEANIBI and RMSSD, respectively) indicated short-term stress during cueing, with comparison of Pre-Event and Post-Event windows showing no difference. In conclusion, heifers successfully navigated VF cues and exhibited short-term stress.

Keywords: dairy cattle, heart-rate variability, Monil, polar heart rate monitors

Introduction

A virtual fence (VF) is defined as a structure that acts as an enclosure, a barrier, or a boundary without the use of physical objects in the landscape to control animal spatial behaviour and movements (Umstatter, 2011). Virtual fencing of animals works by emitting a warning signal (auditory, vibration, or light) as the animal approaches the boundary of the pre-defined grazing area. If the animal persists in moving towards exiting the grazing area despite the warning signal, an electrical cue is administered. For such systems to be ethically justifiable, animals equipped with VF devices must learn to associate the non-aversive warning signals the impending electrical cue (Lee *et al.*, 2009; Nyamuryekung'e *et al.*, 2023).

Research indicates that animals cope better, learn more effectively, and experience reduced stress levels when there is predictability in their living environment (Bassett & Buchanan-Smith, 2007). However, in the context of using VF for livestock management, both the auditory and the electrical cues can induce stress in animals (Lee *et al.*, 2008). Several studies have examined the physiological indicators of the stress response in animals subjected to virtual fencing, particularly focusing on chronic stressors. Another approach involves monitoring stress immediately following the cue of the VF collars, categorized as acute stress. Measuring heart rates could offer valuable insights into this acute stress response (von Borell *et al.*, 2007).

Hence, the study aimed to investigate mature heifers learning to use the Monil virtual fence collar system, and to evaluate the acute stress of the animals using a heart rate monitor.

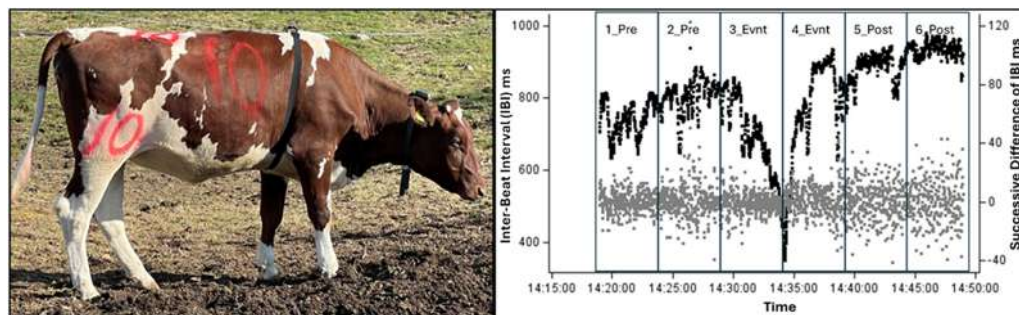


Figure 1. (Left) Heifer equipped with a Monil virtual fence collar and a Polar heart rate monitor. (Right) Scatter plot of Polar heart rate data during a cueing event, displaying inter-beat intervals (black dots; primary y-axis) and successive differences of inter-beat intervals (grey dots; secondary y-axis) used for calculating heart rate variability. The x-axis shows two 5-min segments representing measurements before (1_Pre, and 2_Pre), during (3_Evt and 4_Evt), and after (5_Post and 6_Post) virtual fence cueing.

Material and methods

Ten mature Norwegian Red heifers that were naive to VF from the same herd, were fitted with Monil virtual fence collars ($n = 10$) and Polar heart rate monitors ($n = 9$). For the heart rate sensors, specific areas on the animals were shaved before placing an elastic belt with sensor electrodes around their girth, with contact gel used for optimal sensor contact. The study took place in a 4500 m² area with a physical fence in Asker, Akershus, Norway, from May 11 to 14, 2023. During the deployment, VF collars confine the animals to a designated grazing area from 10:00 to 19:00 h.

Utilizing CSV files from the Monil system, variables extracted included counts of auditory (#Audio) and electrical cues (#Elect), as well as the ratio of electric to auditory cues (Elect:Audio) for each animal daily, reflecting their cueing load over the three days. Heart rate data were processed with Polar Pro Trainer 5 Equine Edition software. The heart rate data of each heifer were then synchronized to its cue timestamps from the VF collar. A 10-minutes window (composed of two 5-minutes segments) surrounding each cue (Event window 3–4, Figure 1) was analysed, along with control windows (Pre-Event and Post-Event; windows 1–2 and 5–6, respectively, Figure 1), to evaluate the average inter-beat interval (MEANIBI) and heart rate variability as the square root mean of the sum of the squares of difference between successive inter-beat intervals (RMSSD) (von Borell *et al.*, 2007).

Using SAS 9.3 software (SAS Institute, Cary, NC, USA), the PROC MIXED procedure was employed, which incorporated both fixed and random effects to analyse all variables. Cueing per animal per day (#Audio, #Elect, and #Elect:#Audio) was assessed using a one-way ANOVA to evaluate the effects of days ($n = 3$), treating Animal ID ($N = 10$) as a random factor.

For the derived variables from the heart rate monitor (MEANIBI and RMSSD), a one-way ANOVA was conducted to analyse the effects of the windows (Pre-Event, Event, and Post-Event) as fixed effects, with Animal ID ($n = 9$) included as a random factor. A repeated structure was modelled for the 5-min window measurement using an AR(1) covariance statement for each cue event. A significance level of $P < 0.05$ was applied to all statistical analyses.

Results and discussion

The virtual fence data indicated a consistent pattern where each electrical cue was preceded by an auditory cue, allowing animals to adjust their movements and avoid the electrical cue. Daily analyses

showed significant effects over the days for the #Audio (First-Day: 5.80, Second-Day: 6.30, Third-Day: 1.20 day⁻¹; $P = 0.01$) and #Elect (First-Day: 3.30, Second-Day: 4.30, Third-Day: 1.10 day⁻¹; $P = 0.01$), but no effect on the ratio of electrical to auditory cues ($P = 0.57$). Pairwise comparisons revealed no significant difference between the first and second days, while the third day differed significantly from both. These results support the ethical assumption that the device enabled animals to predictably modify their behaviour to avoid cues (Lee *et al.*, 2009; Nyamuryekung'e *et al.*, 2023).

The average time between consecutive heartbeats (IBI) was the reciprocal of the heart rate; hence, a lower IBI indicates a higher number of beats per minute. Measurements of heart rate variability using the RMSSD method have proven to be an effective indicator in assessing the balance between sympathetic (stress) and parasympathetic (relaxed) activity; hence, lower RMSSD values are often associated with sympathetic dominance (von Borell *et al.*, 2007).

For the animal welfare assessment, twenty-two 3-tuple windows were analysed. The MEANIBI revealed significant effects across the window (Pre-Event: 728.31, Event: 697.30, Post-Event: 756.33 ms; $P < 0.01$), whereas RMSSD showed a trend (Pre-Event: 9.64, Event: 8.69, Post-Event: 9.90 ms; $P = 0.09$). Pairwise comparisons indicated that exposure to virtual fence cueing during the Event window led to a decrease in both variables compared to the Pre-Event and Post-Event windows. However, the Pre-Event and Post-Event windows did not significantly differ from each other. These findings suggest that Monil virtual fence collar cueing induced short-term stress in the animals, but they were able to recover and return to their normal condition well within 10 min, aligning with other studies evaluating acute stress using heart rate monitors in virtual fence applications (Anderson, 2007; Eftang *et al.*, 2023).

Conclusions

The findings revealed that the virtual fence system enabled animals to adjust their behaviour and hence minimized the cueing across days. While there was a brief stress response when the collar emitted a cue, heifers recovered to their normal state within 10 minutes.

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Effect of harvesting techniques on nutritional composition of alfalfa tops for organic poultry feed

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Abstract

Alfalfa tops represent a promising, locally produced, and drought-adapted protein source for organic poultry diets, in line with EU Organic Regulation 2018/848. However, their nutritional values vary, for example, crude protein (CP) and crude fibre (CF) content, which are influenced by harvesting method and time. This study investigated the effects of different harvesting techniques on CP and CF content of alfalfa tops (*Medicago sativa*) at the first cut. A field trial (May 2024) compared two techniques (Seco Duplex 900 F (SD) and Topcut (TP)) at two cutting heights (33 cm and 40 cm) in a three-variant×four-replicate design (SD33, TP33, TP40). Statistical analyses (one-way ANOVA, post-hoc Tukey test) revealed that cutting height affected top length ($P < 0.05$). Dry matter (DM) yield differed only between TP40 (1.14 Mg ha^{-1}) and SD33 (1.52 Mg ha^{-1} ; $P < 0.05$). Although CP and CF content showed no differences between variants ($P > 0.05$), CF was clearly reduced, with a tendency for higher CP in tops compared to the whole plant. However, the potential CP content (263 g/kg DM) could not be achieved with the applied methods ($218 \text{ g (kg DM)}^{-1}$; $P < 0.05$). Future research should focus on minimizing CP losses during the practical harvesting process.

Keywords: alfalfa cutting methods, crude fibre, crude protein, organic poultry nutrition

Introduction

In organic poultry nutrition, providing sufficient amounts of essential amino acids—especially sulphur-containing amino acids like methionine—remains a major challenge. Alfalfa (lucerne) (*Medicago sativa*) is a widely available, drought-adapted, native forage crop that is rich in crude protein and contains a favourable amino acid profile, making it an attractive protein feed for organic systems.

Of particular interest is the targeted harvesting of alfalfa tops, i.e., the uppermost parts of the plant. By harvesting only these, the proportion of fibrous stem material is reduced, which can result in higher CP concentrations (target $> 25\%$ of DM) and lower CF contents (target $< 20\%$ of DM) compared to the whole plant (Bellof *et al.*, 2021; Maxa *et al.*, 2023). This approach could significantly increase the value of alfalfa as a protein source for monogastric animals such as poultry.

However, the nutritional quality of alfalfa tops is not determined by plant composition alone; it is strongly influenced by the choice of harvesting method, cutting height, and subsequent handling during the harvest process. Losses of fine leaf material, for example, can occur during harvest and post-harvesting processes and may substantially affect the protein content. Against this background, the present study investigated how different mowing systems and cutting heights influence CP and CF concentrations in alfalfa tops in a practical-scale trial. Additionally, the practical-scale harvested material was compared to manually collected square-metre samples to estimate the theoretical maximum potential for CP and CF content. The prerequisite for the entire process was the industry's demand for a wilted crop with a DM content of at least 30%, which can only be achieved with a conventional

Table 1. Experimental design

	Treatment		
	SD33	TP33	TP40
Technique	Seco Duplex 900F	TopCut collect	TopCut collect
Manufacturer	BB-Umwelttechnik	Zürn Harvesting	Zürn Harvesting
Target cutting height	33 cm	33 cm	40 cm
Replication	4	4	4

harvest with wide placement after cutting, followed by a short field drying period and subsequent swathings.

Materials and methods

The field trial was conducted in May 2024 in Dietersheim (Bavaria, Germany) on a 7.3 ha organically managed field. The alfalfa stand was in its fifth year of use and was harvested at the pre-bud stage (BBCH 55–59). A randomized block design with three treatments and four replicates was used. The treatments consisted of two double-blade mowing systems—Seco Duplex 900F (SD) and TopCut collect (TP)—combined with two cutting heights (33 cm and 40 cm), resulting in three variants: SD33, TP33, and TP40 (Table 1). Due to the planned third mowing system (Combcut, Lyckegård) being technically unsuitable for harvesting alfalfa tops, the trial design was modified. TP40 replaced the missing variant to maintain a randomized block design with three treatments and four replicates, resulting in the exclusion of the SD40 treatment and the loss of a full 2 × 2 factorial design, as the number of plots could not be expanded. The harvest procedure was as follows: (1) mowing was performed according to the treatment specifications, (2) swathing was done with a rotary rake, and (3) round bales were produced from the swaths and collected using a bale grab. Each bale was weighed for yield measurements. Subsamples were randomly taken from both faces of each bale and combined into a composite sample per plot, which was used for nutritional analyses with four replicates per treatment.

Throughout the harvesting process, samples of both the whole plant and the alfalfa tops were randomly collected along the stripes or swaths of each variant for nutritional analysis. To evaluate the potential nutritional value of alfalfa tops independently of practical harvesting losses, manual samples were taken additionally from 1 m² quadrats, cutting at 33 cm above ground to simulate an “ideal” harvest of only the upper plant parts. All samples were analysed for DM, CP, and CF contents (Landwirtschaftliche Kommunikations- und Servicegesellschaft mbH, Niederwiesa, Germany). DM was determined by drying at 40°C. Data were analysed using one-way ANOVA (GLM procedure, SAS Studio 9.4), with Tukey’s test for post hoc comparisons. Statistical significance was set at *P* < 0.05 (*n* = 4 per variant and item).

Results and discussion

The average plant height across treatments was 62.1 cm and this did not differ significantly between variants (*P* = 0.624). However, as expected, cutting height influenced top length: the higher cutting height (TP40) resulted in significantly shorter tops (*P* < 0.05). This effect was less distinct in absolute terms (cm) but became clearer when expressed as a percentage of total plant height (%; Table 2). DM yield differed significantly between SD33 (1.52 Mg ha^{−1}) and TP40 (1.14 Mg ha^{−1}; *P* < 0.05). TP33 did not differ significantly from either of these treatments (1.20 Mg ha^{−1}). This suggest that a 7 cm

Table 2. Effects of harvesting techniques on selected harvest traits of alfalfa tops (LS, means±SE)

Item	Unit	Treatment			SE	P-value
		SD33	TP33	TP40		
Cutting height ¹	cm	31.9 ^b	34.6 ^b	42.1 ^a	1.35	0.001
Top length ²	cm	31.2 ^a	27.4 ^{ab}	19.0 ^b	2.20	0.010
Top length ³	%	49.2 ^a	44.1 ^a	31.0 ^b	2.84	0.004
Alfalfa top yield	DM Mg ha ⁻¹	1.52 ^a	1.20 ^{ab}	1.14 ^b	0.09	0.038
Crude protein	g/kg DM	220	220	215	5.12	0.742
Crude fibre	g/kg DM	197	186	195	14.4	0.851

Cutting height, stubble length, measured from the ground; Top length, calculated, average growth height–average cutting height; The percentage length refers to the proportion of the alfalfa tops in relation to the total plant height. ^{a,b} different superscripts within a row show significant differences between the variants ($P < 0.05$).

difference in cutting height (33 cm vs. 40 cm) had little effect on yield when using the same technique (TopCut); however, a potential interaction between cutting height and technique cannot be excluded. However, the combination of mowing technique and cutting height (SD33 vs. TP40) resulted in a substantial yield difference of 0.38 Mg ha⁻¹. No significant differences were found in CP and CF contents between the three treatments ($P \geq 0.742$). Nevertheless, the harvested tops generally showed a trend towards lower CF and slightly higher CP compared to the whole plant (CP whole plant: 216 g (kg DM)⁻¹, baled material: 218 g (kg DM)⁻¹). CF content in the whole plant was 232 g (kg DM)⁻¹, while it was reduced to 193 g (kg DM)⁻¹ in the harvested tops.

Interestingly, while CP content in the baled tops (SD33, TP33, TP40) averaged 218 g (kg DM)⁻¹, manually harvested square-metre samples showed significantly higher CP concentrations (263 ± 2.68 g (kg DM)⁻¹; $P < 0.001$). Similarly, CF content was significantly lower in the manually harvested samples (173 ± 5.85 g/kg DM) compared to the practical-scale harvested material (193 ± 5.85 g (kg DM)⁻¹; $P = 0.029$). This suggests that during the mechanical harvest, losses of fine leaf material with high CP concentrations likely occurred, particularly during swathing, raking and baling, as observed by Al-Gaadi (2018). On the other hand, stalks may also have been carried into the raked material by the movement of the rake tines through the standing long stubble, with the necessary setting at approximately 4–5 cm above the ground and the pick-up tines of the baler. The results indicate that while cutting height and mowing system had little impact on CP and CF, the post-harvest process (swathing and baling) substantially influenced the nutritional value of alfalfa tops. Future research should focus on optimising these steps to minimise nutrient losses, for example, by testing alternative swathing techniques as a merger to reduce crumbling.

Conclusion

Alfalfa tops have considerable potential as a locally produced, organic protein source in poultry production due to their ability to achieve a relatively high CP content and a marked reduction in CF compared to the whole plant. In this study, the mowing technique—including both the mowing system and cutting height—affect DM yield but had no significant influence on CP and CF concentrations. However, it became evident that the post-harvest process (including swathing and baling) had a much greater impact on the nutritional value of alfalfa tops and can lead to substantial losses of fine, protein-rich leaf material.

Future work should therefore focus on optimising harvesting and post-harvest handling techniques to minimize such losses and fully exploit the potential of alfalfa tops as a high-quality organic feed for poultry diets.

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Dutch dairy farmers' perceptions of cow health during grazing and housing periods

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Abstract

Dairy farmers' perceptions of animal health influence management and may vary with farm characteristics and practices such as grazing and housing. This study assessed how Dutch dairy farmers perceive udder and claw health, reproductive disorders (RD), and metabolic diseases (MD) during grazing and housing, and how these perceptions relate to farm characteristics and management. In 2023–2024, 78 grazing farms were surveyed, with farmers rating disease severity on a Likert scale from 1 (no problems) to 5 (serious problems). Claw and udder health were generally perceived more positively during grazing, whereas RD were rated more positively during housing. Farmers with automatic milking systems perceived MD during housing more negatively than those with a milking parlour. Farmers who reported more grazing tended to rate udder health more positively, whereas those with larger herds rated it more negatively during housing. Farmers with larger herds or more hectares available for grazing perceived RD during grazing as more problematic, while those who reported more grazing hours perceived MD during housing more negatively. Overall, perceptions varied for housing and grazing, but showed weak associations with farm characteristics and management.

Keywords: cow health, farm characteristics, farmers' perception

Introduction

Dairy farmers' perception of animal health plays a crucial role in the early detection and management of health problems on-farm (Dutton-Regester *et al.*, 2020). Since many diseases, such as mastitis, lameness, metabolic disorders (MD) and reproductive disorders (RD), begin subclinical, timely action is important in preventing clinical forms and welfare reduction. Whereas perceptions are strongly linked to the motivation and willingness to take action (Vaarst & Sørensen, 2009), they may change throughout the season. Studies seldom include farmers' perceptions on animal health and, if they do, these studies do not systematically compare if dairy farmers perceive dairy cow health differently during grazing and housing. Therefore, this study aims to explore how dairy farmers perceive cow health during grazing and housing, and how these perceptions relate to farm characteristics and management.

Materials and methods

A longitudinal field study with 78 Dutch dairy farmers was conducted in 2023 and 2024. All farms practised grazing and data for each farm were collected for one grazing season. During a farm visit in the housing period, before the start of the grazing season, a structured face-to-face interview was done with the farmer. Farmers were asked to rate the perceived severity of udder health, claw health, RD and MD on a Likert-scale from one (no problems) to five (serious problems) for the housing and grazing period. Farm characteristics, like number of dairy cows, milking system, grazing area (hectares

Table 1. Descriptive statistics of the Dutch dairy farms

	Mean±SD	Min—Max	No. of farms
Grazing hours (per grazing season)	1643 ± 719	690–3981	78
Grazing days (per grazing season)	186 ± 30	121–283	78
Herd size (No. of dairy cows)	114 ± 54	37–310	78
Grazing area (ha per farm)	34.4 ± 18.6	6–118	78

Table 2. Cow health as perceived by dairy farmers during the housing and the grazing period.

Indicator	Period	1 (%)	2 (%)	3 (%)	4 (%)	5 (%)	P-value
Udder health	Housing	19	37	27	13	4	0.011
	Grazing	26	42	19	12	1	
Claw health	Housing	19	37	27	13	4	0.001
	Grazing	26	51	17	5	1	
Reproductive disorders	Housing	27	42	26	5	0	0.006
	Grazing	15	50	22	12	1	
Metabolic diseases	Housing	28	50	18	4	0	0.240
	Grazing	28	58	13	1	0	

A score of one indicates no problems and a score of five indicates serious problems. Values represent the percentage of farms that perceived cow health at each score level.

available for grazing), presence or absence of grass-clover and herb-rich pastures, were also collected during this farm visit. Farmers recorded the start and end date of the grazing season and hours of grazing per day. All data were analysed in IBM SPSS Statistics 29. A Wilcoxon Signed Rank test was used to assess differences in perceived health between the housing period and the grazing season. For further analyses, scores 1+2 and 4+5 were combined. A Mann–Whitney *U*-test was used to assess differences in health perception and the categorical variables (milking system, presence of grass-clover and herb-rich pasture). Spearman rank correlations (*p*) were used to test for correlations between the health perception and the continuous variables (grazing hours and days, herd size and grazing area). In all tests, *P* < 0.05 was considered significant.

Results and discussion

Nineteen farms (24%) milked with an Automatic Milking System (AMS) and fifty-nine (76%) with a conventional milking parlour (MP). Thirty-seven farms (47%) had grass-clover pastures, whereas thirty-three farms (42%) had herb-rich pastures available for grazing. Further descriptive statistics can be found in Table 1.

Farmers generally perceived udder and claw health as better during grazing compared with housing (*P* = 0.011 and *P* = 0.001, respectively; Table 2), which aligns with previous research demonstrating the beneficial effects of pasture access on these parameters (Arnott *et al.*, 2017; Schuppli *et al.*, 2014). By contrast, RD were perceived more negatively during grazing, which is not consistent with earlier studies reporting less RD in grazing systems (Arnott *et al.*, 2017). Nevertheless, farmers with grass-clover pastures perceived reproductive performance during grazing more positively than those without (*P* = 0.039). Perceptions of MD did not differ significantly between housing and grazing. Comparisons across farm characteristics revealed several patterns. Farmers using an AMS tended to perceive MD during housing more negatively than those using a MP (*P* = 0.009). Farmers who reported more grazing hours or days tended to perceive udder health during grazing more positively (Table 3), while farmers with larger herds perceived udder health during housing more negatively. Farmers with larger herds or greater grazing area tended to perceive RD during grazing more negatively.

Table 3. Correlations (ρ) between management, farm characteristics and farmers' perception of health for housing and grazing

Health	Period	Grazing hours	Grazing days	Herd size	Hectares available for grazing
Udder health	Housing	0.039	-0.001	0.242*	0.128
	Grazing	-0.270*	-0.271*	0.181	-0.006
Claw health	Housing	0.107	-0.043	-0.047	0.063
	Grazing	-0.022	0.037	0.123	-0.025
Reproductive disorders	Housing	-0.005	-0.011	0.197	0.123
	Grazing	0.003	0.011	0.243*	0.255*
Metabolic diseases	Housing	0.260*	0.049	0.031	0.201
	Grazing	-0.115	-0.109	-0.008	0.034

* Significance.

A possible explanation could be that a larger herd or a larger area is associated with a reduced feeling of control by the farmer, and with less opportunity to spend time with individual animals (Lind *et al.*, 2025), although this association was not observed for lameness in a study by Jensen *et al.* (2022). Interestingly, farmers who reported more grazing hours tended to perceive MD during housing more negatively ($P = 0.024$), suggesting that these perceptions may relate more to housing system characteristics than to grazing area per se. All correlations were weak, which may reflect the relatively homogeneous nature of the study population and suggests that other factors influence farmers' perceptions. It is also possible that farmers' perceptions do not accurately reflect the actual health status of their herds. Future research should combine perception data with objective herd health records and management-related risk factors, such as stocking density, feed supplementation, and housing conditions, to better understand how perceptions are formed and how they relate to true health outcomes.

Conclusion

Farmers perceived cow health differently between grazing and housing periods, and these perceptions were only weakly related to herd and farm characteristics. This leaves uncertainty about how closely perceptions reflect actual herd health. Nonetheless, gaining insight into how farmers perceive health risks in relation to their production context can help guide strategies to improve cow health and sustainability in pasture-based systems.

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Characteristics of grazing innovations currently emerging in Europe

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Abstract

Agricultural innovations are strongly affected by the context in which they operate. Within the thematic network Grazing4AgroEcology (<https://grazing4agroecology.eu/>), 121 grazing-based innovations have been identified and described by a network of facilitator agents across eight European countries. For successful innovation transfer, information about the context of origin for each innovation is required, helping adopters to interpret the suitability of those innovations for their own context. The context of origin of the innovations was validated and analysed by an international, interdisciplinary Scientific & Technical Working Group. A Multiple Correspondence Analysis was used to explore patterns among innovations. Farms in the Atlantic North and South zones with high rainfall clustered together, while innovations in the Alpine region on sandy soils and low rainfall were mainly developed by part-time farmers focusing on pasture-beef. Innovations in the Mediterranean South with mild winters were generally easy to implement. Innovations on Atlantic Central farms tended to target global markets. The results suggest that the context of origin of the innovations affect their development and establishment.

Keywords: agroecology, context analysis, innovation transfer

Introduction

Agricultural innovations play an important role in enhancing the sustainability, productivity, and adaptability of farming systems across Europe. In the context of this paper, innovation is understood broadly as any practice or approach that supports one or more of the five agroecological principles pivotal to sustainable animal production: (1) adopting management practices that aim to improve animal welfare; (2) reducing the inputs required for production; (3) reducing pollution of farming systems; (4) enhancing diversity within animal production systems to strengthen their resilience; and (5) preserving biodiversity by adapting management practices (Dumont *et al.*, 2013). However, the success of the innovations depends on the specific socio-economic, environmental, and cultural contexts in which they are developed and implemented. Effective innovation in farming systems is rarely universal; what works in one region (or farm) may not be suitable in another (Wigboldus *et al.*, 2016; Peratoner *et al.*, 2025). Understanding these contextual factors is essential to facilitate the adoption and adaptation of innovations across diverse agroecological zones. This paper presents an overview of

the contexts of origin in which 121 grazing innovations were developed and summarises some of the relationships between their features.

Materials and methods

Within the Horizon Europe-thematic network Grazing4AgroEcology (G4AE, www.grazing4agroecology.eu), 121 grazing-based innovations were collected from the eight participating countries: France, Germany, Ireland, Italy, The Netherlands, Portugal, Romania and Sweden. The innovations were identified by a network of trained facilitator agents operating in these countries. For each case study, information was collected by facilitator agents using a standardised protocol, which included a first visit questionnaire, a practice abstract and a video. The context of origin of the innovation was classified using nine key descriptors, each with predefined levels: (1) main domain of the innovation (improvement of grassland management, animal management, workload reduction, improvement of plant protection, improvement of nutrient cycle, breeding advance, improvement of marketing); (2) agroclimatic zone (Alpine, Atlantic Central, Atlantic North, Atlantic South, Boreal, Continental North, Continental South, Mediterranean North, Mediterranean South); (3) climate (very high rainfall, moderate rainfall, little rainfall, mild winter); (4) soil type (sand, loam, peat, clay); (5) management (pasture dairy, pasture beef, cutting-only, ley farming); (6) technical (difficult, easy, computer-based); (7) finance/investment (low, mid, high); (8) market (global, local-urban, local-rural); and (9) social (full-time farmer, part-time farmer, hobby). An interdisciplinary Scientific & Technical Working Group, representing science and agricultural practice, validated the information and conducted the context analysis to assess the transferability of the innovations across agroecological zones in Europe. A Multiple Correspondence Analysis (MCA) was performed on 117 case studies with a classification of the context of origin based on the above-mentioned descriptors except main domain of innovation (with seven observations belonging to underrepresented levels with <5% of the cases) and Boreal (also <5%). Factor levels with zero variance across all cases and observations with missing values were removed as well.

Results and discussion

Grassland or animal management were addressed by 63% of innovations. About two thirds of all innovations related to pasture dairy farms (69%). The technical content of half of the case studies was easy to implement, whilst one third of them were computer-based. Only 13% of the cases required high financial investments, while 57% of them were characterised by low financial investments. Most of the cases (58%) targeted a local-rural market, around 30% a global market, and a much lower share (13%) addressed a local-urban market. They were mainly developed by full-time farmers (83%), and none by hobby farmers.

The first two components of the MCA explained 25% of the total variability in the dataset (Figure 1). Overall, the MCA highlights the central role of agroclimatic conditions, rainfall, management and market orientation in different case studies. Farms located in the Atlantic North and Atlantic South agroclimatic zones (France and Ireland) were mainly related to high rainfall. Innovations in the Alpine zone, characterised by sandy soils and low rainfall, were mainly developed by part-time farmers and focused primarily on pasture-based beef. Innovations in the Mediterranean South zone like Portugal and Italy with mild winters, tended to be of easy implementation. Farms located in the Atlantic Central zone such as in Germany, Sweden and the Netherlands, tended to target global markets.

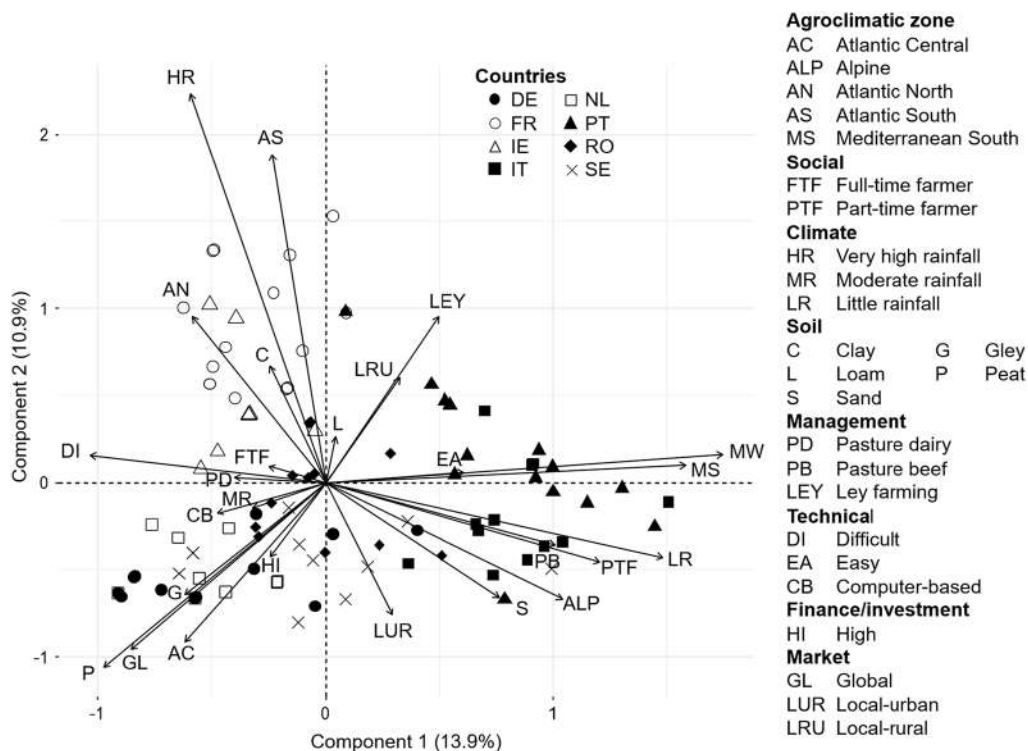


Figure 1. Biplot-ordination diagram showing the results of MCA on the classification of the grazing innovations tapped and classified within Grazing4AgroEcology. Only vectors with percent contribution greater than 0.1 on at least one of the axes are displayed.

Conclusion

The two principal components explained only a part of the total variability and therefore cannot fully describe all correlations. However, the MCA revealed contrasts and patterns, indicating that the context of origin affects the development and implementation of innovations in grazing systems.

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Linking farmers' grazing motivation to farm characteristics and management on Dutch dairy farms

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Abstract

Farmers practice grazing for various reasons, but it is unclear whether their main motivations are related to farm characteristics or management, knowledge that is essential both for informing targeted advisory and policy interventions and for improving our understanding of decision-making in dairy farming. This study aimed to assess whether the categorised main reason (CMR) for grazing—grouped as ‘animal’, ‘economic’ or ‘people’ and developed inductively from farmers’ responses during interviews—relates to farm characteristics and management of Dutch dairy farms. Seventy-eight grazing dairy farms were visited during the 2023 and 2024 grazing season, and farmers were interviewed about their main reason for grazing. Additional data on farm characteristics and management were collected. Chi-square and Kruskal-Wallis tests were used to assess associations between CMR and farm characteristics and management. No significant differences were found between CMR groups in grazing hours and days, herd size, hectares available for grazing, grass-clover and herb-rich pastures or milking system. These results suggest that grazing motivations are diverse and largely independent of farm structure, indicating that advisory and policy strategies should consider individual farmer perspectives.

Keywords: drivers, farm characteristics, farm management, motivation for grazing

Introduction

Farmers perceive several barriers to and drivers for grazing. For Canada, Smid *et al.* (2021) categorised them into different themes: climate, cow welfare, economics, infrastructure and management. In the Netherlands, the main drivers were animal welfare, animal health, images of livestock farming, labour reduction, less need to buy protein, and grazing is the farmer's preferred production system. The barriers to practice grazing were related to variability in grass quantity and quality, climate and available labour (Van den Pol-van Dasselaar *et al.*, 2024). Schuppli *et al.* (2014) showed that animal health is often mentioned as both a driver and a barrier to grazing. Even though there is a good understanding of drivers and barriers, it is unclear if these drivers are related to farm characteristics and/or management. Therefore, this study aims to assess whether a farmer's categorised main reason (CMR) for applying grazing (‘animal’, ‘economic’, or ‘people’) is associated with farm characteristics and management in Dutch dairy farms. These categories were developed inductively from qualitative insights gained during farm visits and structured around recurring themes in farmers' reasoning.

Materials and methods

A longitudinal field study with 78 Dutch dairy farmers was conducted in 2023 and 2024. All farms practised grazing and farm-level data were collected for one grazing season. During a farm visit in

Table 1. Main reasons of grazing of 78 Dutch dairy farmers categorised into the main reasons

Main reason	No. of farms	Categorised main reason
Cost-related	24	Economic
Personal beliefs or preferences	21	People
Animal health	13	Animal
Animal welfare	7	Animal
Public image	6	People
Labour	4	Economic
Soil	2	—
Other	1	—

the housing period, just before the start of the grazing season, a structured face-to-face interview was done with the farmer. Farmers were asked for their main reason to graze and gave diverse answers, resulting in relatively low numbers per category. Only farms reporting main reasons mentioned more than three times were included in the evaluation ($n = 75$). The reasons were categorised into three main reasons (CMR; ‘animal’, ‘economic’ and ‘people’; Table 1). Via the interview several data were collected as well: herd size, milking system, hectares available for grazing and the presence of grass-clover and herb-rich pastures. Farmers recorded the starting and ending date of the grazing season for their dairy cows, as well as the hours of grazing per day. Cow-hours per hectare available for grazing was calculated by multiplying grazing hours by number of milking cows, divided by hectares available for grazing. All data were analysed in IBM SPSS Statistics 29. A chi-square test was used to analyse differences between the CMR and categorical variables (milking system and presence of grass-clover and herb-rich pastures). A Kruskal-Wallis Test was used to analyse differences between the CMR and the continuous variables (grazing hours, grazing days, herd size, hectares available for grazing and cow-hours per hectare). In all tests, a P -value < 0.05 was considered significant.

Results and discussion

Of the 75 farms, 18 farms milked with an automated milking system (AMS) and 57 in a milking parlour (MP). Thirty-five farms had grass-clover pastures and 40 did not, whereas 31 farms had herb-rich pastures and 44 did not. Mean annual grazing hours was 1628 ± 724 , mean annual grazing days 185 ± 30 , mean herd size 115 ± 54 and mean hectares available for grazing 35 ± 19 . Twenty-seven farmers indicated ‘people’ as their CMR for grazing, 20 mentioned ‘animal’ and 28 ‘economic’ (Table 1). No significant differences in CMR distribution were found between farms with an AMS versus MP, nor between farms with or without grass-clover or herb-rich pastures (Table 2). Furthermore, no significant differences were observed between the three CMR groups in terms of grazing hours, grazing days, herd size, hectares available for grazing and cow-hours per hectare available for grazing (Table 3).

This study found no clear association between farmers’ CMR and differences in farm characteristics and management. For example, grazing hours (per year) were similar across CMR groups ($P = 0.182$), and the lack of statistically significant differences could be related to the relatively low sample size ($n = 75$) or due to the categorisation of main reasons for grazing. It is likely that farmers consider multiple, overlapping reasons when deciding to implement grazing (Petit *et al.*, 2022), which may make it difficult to identify clear associations with specific farm characteristics and management. Other factors that could be important, such as type of grazing management and grass intake, were not included in this study. Future research could include these parameters and zero-grazing farms to examine whether

Table 2. Association between categorised main reason and categorical farm characteristics and management

		Animal	Economic	People	<i>P</i> -value
Milking system	AMS	4 (20%)	5 (18%)	9 (33%)	0.360
	MP	16 (80%)	23 (82%)	18 (67%)	
Grass-clover pastures	Yes	8 (40%)	12 (43%)	15 (56%)	0.502
	No	12 (60%)	16 (57%)	12 (44%)	
Herb-rich pastures	Yes	7 (35%)	11 (39%)	13 (48%)	0.639
	No	13 (65%)	17 (61%)	14 (52%)	

Values represent the number of farms per category for the categorical variable (*n* = 75); percentages indicate the proportion of farms per variable and categorised main reason.

Table 3. Association between categorised main reason and continuous farm characteristics and management

	Animal	Economic	People	<i>P</i> -value
Grazing hours (per year)	1529 ± 634	1808 ± 761	1515 ± 735	0.182
Grazing days (per days)	178 ± 32	187 ± 29	189 ± 31	0.591
Herd size	116 ± 56	107 ± 53	124 ± 54	0.397
Hectares available for grazing (per farm)	32.6 ± 16.6	36.5 ± 17.4	34.6 ± 22.0	0.630
Cow-hours per ha available for grazing (h ha ⁻¹)	5886 ± 574	5246 ± 361	5831 ± 497	0.641

Values represent the mean ± SD of each continuous variable for farms, grouped by their categorised main reason for grazing

their perceived barriers to applying grazing are more strongly related to farm characteristics, management practices, or the broader sociological environment

Conclusion

No associations were found between the categorised main reason for grazing (animal, economic, or people) and farm characteristics or management. Despite the relatively small sample size, the results indicate that grazing motivations are diverse and shaped more by individual contexts than by farm structure or practices.

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Factors influencing methane emission measurements when combining data from indoor and outdoor GreenFeed unit environments

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Abstract

This study compared CH₄ emissions and preference between indoor and outdoor environments using 32 Norwegian Red cows divided into two groups. Cows were housed in a barn equipped with an automatic milking system, allowing them free access to pasture. Two GreenFeed units (GEM) were used to measure CH₄ emissions, one placed inside the barn (indoor GEM) and the other on pasture (outdoor GEM). Data were collected across the two groups for two weeks each. When considering the weekly data from the animals that visited either GEM unit the results indicated significantly lower CH₄ emissions measured outdoors (328.6 vs. 301.1 g/day; $P = 0.03$), with higher weekly visits (3.4 vs. 5.5 visits/week; $P < 0.01$) compared to indoors. Analysing the weekly data from the animals that visited both GEM units the findings showed a trend towards lower CH₄ emissions measured outdoors (329.6 vs. 303.5 g/day; $P = 0.09$), with animals visiting twice as often (6.2 vs. 3.2 visits/week; $P < 0.01$) in comparison to indoors. The study suggests that GEM units in different environments can influence CH₄ measurement outcomes, and some animals explicitly avoid visiting the GEM unit. However, for those that do visit both units, the preference is for the outdoor GEM unit.

Keywords: GreenFeed, Norway, pasture

Introduction

Grazing offers several benefits to livestock and the environment. In the livestock sector, grazing may reduce farmers costs for feed, improve animal health and welfare, and provide products (meat and milk) of high nutritional value. For the environment, grazing may improve soil health, increase nutrient cycling, and act as a carbon sink. According to Norwegian regulations, mature cattle must have at least eight weeks of free movement and exercise on pasture during summer. Mitigation strategies for enteric methane (CH₄) emissions from ruminants are in focus, and grazing seems to be a promising approach. Wan *et al.* (2025) provided a meta-analysis which concluded that low grazing intensity was effective in reducing CH₄ emissions. In another review by Buddle *et al.* (2011) forage quality was highlighted as a factor influencing the emissions. Inclusion of white clover resulting in higher forage digestibility, and inclusion of tannins and saponins in the plant species all positively affected CH₄ emissions. Measuring CH₄ emissions from grazing livestock is challenging. The GreenFeed monitoring system (GEM) is used to measure CH₄ from livestock, suitable for use both when the animals are indoors and while grazing. The GEM is a static short-term measurement device used by individual cows integrating airflow, gas concentration, and head position. Animals must be trained to use GEM units (Hammond *et al.*, 2016), especially outdoor units, as indoor GEM units tend to be visited more frequently. The use of multiple GEM units placed in different environments can give insightful understanding of how feed changes during the day might affect methane emissions. However, using multiple GEM units for measurement might be a confounding factor affecting results, if not properly designed. An experiment performed summer of 2023 aimed to compare CH₄ emissions from dairy cows within

the same treatment that utilized two GEM units positioned in indoor and outdoor environments, and to examine the animals' preference for using the two GEM units, and thus to provide a quantified documentation for design of future studies using multiple GEM units.

Materials and methods

The study was conducted at Mære Agricultural School in Steinkjer, Norway (63°56'9"N; 11°23'26"E), utilizing 32 mid to late lactation Norwegian Red dairy cows. The cows were split into two groups, each trained to use the GEM units during a two-week adaptation period, followed by two weeks of recording. During both periods, grass silage was available indoors, limited to 6 kg DM per animal and day, and cows were given access to a pasture with an expected DMI of 50% of daily roughage requirements. Concentrates were available in the milking robot, GEM units, and an indoor concentrate feeder, individually adjusted to pre-experimental requirements, with an expected yield drop in accordance with the breed standard lactation curve (Volden, 2011) and an expected daily roughage intake equal to 12 kg DM grass silage. Pasture intake was calculated as the difference between the net energy requirement and net energy indoor feed intake. The nutrient composition of the indoor diet and the pasture is given in Lardy *et al.* (2025). The pasture was composed of a mixture of timothy (*Phleum pratense*), meadow fescue (*Festuca pratensis*), smooth meadowgrass (*Poa pratensis*), and red clover (*Trifolium pratense*).

In the experiment, the cows had the opportunity to visit the indoor GEM with 4-h intervals, and the outdoor GEM with 3-h intervals. The indoor unit was insulated with a wooden panel to prevent interference from eructation by nearby animals. Access to the outdoor GEM unit was controlled by a chute, which minimized disruption from other animals during visits. The GEM unit manufacturer collected all raw data and validated it by ensuring proper animal head positioning. The exported data from the GEM manufacturer was compiled into daily summaries that represented the average CH₄ emissions and total visit counts for each individual animal. The daily data set was compiled into weekly measurements for each animal in the two groups. In a complete data set (FullData), where all animals visited the GEM units regardless of location, there would be a total of 64 data points per GEM unit. Average daily CH₄ measurements and cumulated visitation counts were computed within the weekly data for each GEM unit. If an animal did not visit a GEM unit, that data point was marked as missing. A second dataset (SubData) included individual animals who visited both GEM units during a week to make sure the indoor and outdoor measurements were paired. The data was analysed using the PROC MIXED procedure in SAS 9.3 software (SAS Institute, Cary, NC, USA). Week was used as a repeated measure, GEM location as a fixed effect, group as a random variable and animal served as the subject and a CS covariance structure applied.

Results and discussion

Analysis of the weekly data shows more data points were recorded for the indoor than outdoor GEM for animals visiting either GEM unit. The dataset comprised 50 data points from the indoor GEM and 30 from outdoor GEM, instead of the expected 64 data points for each, representing 28 and 17 individual animals, respectively. The analysis revealed that CH₄ emissions were significantly lower outdoors (301.1 g/day) compared to indoors (328.6 g/day; Table 1). Although fewer animals visited the outdoor GEM, the frequency of visits was higher (5.5 vs. 3.4 visits per week; $P < 0.01$) compared to indoor visits. In a separate analysis that focused on animals visiting both GEM units each week, the dataset comprised 21 data points out of the expected 64, representing 14 individuals out of 32. This analysis indicated a trend toward lower CH₄ emissions outdoors (303.5 g/day) compared to indoors

Table 1. Average daily methane (CH₄) emissions and the frequency of weekly visits of cows visiting an indoor and outdoor GEM unit

	Variable	GEM location		SE	P
		Indoor	Outdoor		
FullData	Daily CH ₄ (g/day)	328.6	301.1	9.4	0.032
	Visits (No./week) ¹	1.21 (3.4)	1.70 (5.5)	0.15	0.003
	Data points	50	30		
SubData	Daily CH ₄ (g/day)	329.6	303.5	11.9	0.09
	Visits (No./week)	3.2	6.2	0.71	0.0002
	Data points	21	21		

¹ Natural log transformation applied before statistical analysis; back-transformed values included in brackets.

(329.6 g/day; Table 1) and showed that animals visited the outdoor unit twice as often (6.2 vs. 3.2 visits per week) compared to the indoor unit. Hammond *et al.* (2016) indicate that when GEM units are used outdoors, wind can reduce the fraction of CH₄ captured. On the other hand, Renand *et al.* (2013) reported significant differences in CH₄ emissions during day and night. In contrast, Lardy *et al.* (2023) did not see any differences in daily CH₄ emissions between cows grazing day or night. However, a drop in CH₄ emissions was observed between indoor and outdoor measurements, using two GEM units in a setup similar to the current experiment.

Conclusion

The study suggests that the environment of the GEM units can significantly affect measurement outcomes and implies that some animals may actively avoid visiting the GEM units. For those that do visit, there is a preference for visiting the outdoor GEM unit. This research provides a quantified documentation for future studies using multiple GEM units for measurement, highlighting that if not properly designed, a confounding factor might affect the results, especially if cohorts are restricted to a specific GEM unit.

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Assessing livestock distribution evenness using Global Navigation Satellite System tracking for improved grazing management

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Abstract

Uneven grazing distribution is a major challenge in continuous grazing systems. Livestock exhibits selective behaviour depending on different biotic factors and abiotic controls, often resulting in both overgrazed and undergrazed areas within the same pasture. Fencing, herding and attractants—such as supplements or water troughs—can be used to achieve more uniform grazing pressure. Nevertheless, inherent physical elements, such as terrain geomorphology, remain unchangeable and can limit the effectiveness of these management practices. Recently, evenness of grazing distribution has been quantified combining Kernel Density Estimation and Gini indices derived from animal tracking data. Compared to traditional monitoring methods, the approach offers a new paradigm to assess livestock distribution quantitatively and in near real-time. However, because grazing distribution is partially shaped by the spatial characteristics of pastures, it is important to consider the natural upper limit of grazing evenness imposed by the characteristics of each pasture. The present study explores the relationship between grazing distribution evenness and the spatial characteristics of pastures under varying conditions.

Keywords: animal tracking, grazing distribution, landscape heterogeneity, Precision Grazing Management

Introduction

In recent years, Precision Livestock Farming (PLF) has gained increasing attention among researchers, largely due to the decreasing cost of sensor technologies. Currently, PLF tools focus on management of individual animals (Berckmans, 2014), as their development has mostly occurred under controlled conditions (indoor systems), where inputs and outputs are easier to manage compared to rangelands. Precision Grazing Management (PGM) considers the individual animals as components of a more complex system (i.e. the rangeland). Consequently, the main challenge for farmers is to optimally manage spatiotemporal interactions between animals and pastures (Laca, 2009).

According to Holechek (1988), the most influential characteristic in rugged and mountain ranges is slope. The other variable with a large effect on livestock distribution is distance to water. This study evaluated the relationships of the evenness of grazing distribution with slope, water availability, stocking densities, and grazing period duration across different pasture systems.

Table 1. Characteristics of studied farms

Commercial name	Number of paddocks	Number of trackers	Paddock areas (ha)	Type of rangeland	Latitude-Longitude	Herd size (heads)	Species
Fuente del Perro	2	59	19, 22	Dehesa rangeland	38°24'N 4°42'W	73	Cattle
El Palacio	1	29	96	Dehesa rangeland	37°58'N 6°15'W	220	Cattle
Cabañas	1	48	630	Shrub-Grassland mixture	40°33'N 4°45'W	230	Cattle
Mediana de Voltoya	3	81	43, 85, 107	Shrub-Grassland mixture	40°41'N 4°32'W	110	Cattle
Navarrete	2	13	193, 1562	Mountainous woodland	36°58'N 4°34'W	160/2000	Cattle/Sheep
Las Albaidas	1	27	610	Dehesa rangeland	37°54'N 4°50'W	1600	Sheep
Natrus	5	117	8, 13, 37, 76, 143	Forested rangeland	42°06'N 2°28'E	270	Cattle

Materials and methods

Individual cows from seven commercial farms located across various climatic regions of Spain, were fitted with commercial Global Navigation Satellite Systems (GNSS) trackers (Digitanimal, Madrid, Spain), configured with a 30-minute time resolution. A total of fifteen grazing paddocks were studied to evaluate the effect of slope and distance to water (Table 1) on grazing distribution evenness.

The GNSS data preprocessing followed a two-step protocol. First, days with fewer than 24 fixations (equivalent to less than 12 h of location data) and/or gaps of data with more than 3 consecutive hours missing were removed. Subsequently, original trajectories were resampled at 30-minute intervals using the *trajr* package in R software (R Core Team, 2024) to reduce the influence of days and/or collars with higher number of fixations.

Grazing distribution evenness metrics were calculated from resampled GNSS locations based on the methodology proposed by Maroto-Molina *et al.* (2023). The method consists of computing a Kernel Density Estimation (KDE) to smooth the raw GNSS positions, using a 15-metre bandwidth, a regular grid of 25 metre and a Gaussian (quartic) kernel function. KDE values and pixel areas were sorted in ascending order and normalized by dividing each value by the maximum, obtaining normalized values ranging from 0 to 1. A modified Gini index (G) was subsequently calculated from the Lorenz curve defined by the normalized values of KDE corresponding to the normalized values of pixel areas. This Gini index was used as an indicator of the uniformity of grazing distribution (see Maroto-Molina *et al.* 2023, for details).

Data analysis consisted in a Linear Mixed Model developed using *lmerTest* package in R (R Core Team, 2024). Percentage of the area with slopes >10%, maximum distances to water, stocking densities (1 sheep = 0.160 cow) and number of days on pasture were considered as fixed effects. Farm identity was considered as random factor. Significance was declared with $p < 0.05$ and tendency was considered for p values between 0.05 and 0.10.

Results and discussion

On average, the mean value of G in the paddocks studied was 0.604 with a SD of 0.170, a maximum value of 0.874 and a minimum of 0.321. The highest values were recorded at Fuente del Perro farm, showing also the second lowest SD (0.05), which indicates a consistently high evenness of grazing distribution. Conversely, the lowest G value (0.321) was observed at Natrus farm, which also had the highest variability (SD = 0.183).

This phenomenon coincided with terrain characteristics, since Fuente del Perro had the lowest average proportion of area with slopes greater than 10%, while Natrus had the highest. The only fixed variable which had a significant effect, and a negative relationship on G was the slope ($p = 0.046$), which implies lower grazing evenness with increasing proportion of area with slopes greater than 10%. However, no significance or trends were observed for maximum distances from water or number of grazing days ($p = 0.315$). This aligns with Holechek (1988), who suggested that reductions in stocking rates based on distances to water should typically begin at 1.5 km. Only four of the studied paddocks had a maximum distance to water over 1.5 km. A positive tendency was observed between stocking density and G values ($p = 0.098$), which coincides with the results obtained in mountainous rangelands (Hart *et al.*, 1991). The range of stocking densities varied from $0.050 \text{ AU} \cdot \text{ha}^{-1}$ to $5.11 \text{ AU} \cdot \text{ha}^{-1}$ with an average of $1.036 \text{ AU} \cdot \text{ha}^{-1}$, which is common in Spanish rangelands. The number of grazing days showed no relation ($p = 0.935$) with G values. This result may be attributed to the extended grazing periods observed in the study, with an average of 129.2 days. Padrón-Tejera *et al.* (2024) explored the temporal evolution of G values in different scenarios and reported that G values tend to rise quickly during the first grazing days before stabilizing.

Conclusion

Grazing distribution is influenced by factors that can be modified by farmers such as stocking densities, and permanent terrain features, such as slope. Further research is required to explore the influence of other abiotic controls and biotic factors such as vegetation or weather. GNSS sensors operating in real-time can provide the data necessary to monitor optimal livestock distribution in different scenarios.

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Impacts of adaptation and mitigation strategies to climate change in French ruminant systems

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Abstract

Ruminant livestock systems are facing a dual challenge in the context of climate change. They must adapt their practices to cope with increasing climatic hazards while also mitigating greenhouse gas (GHG) emissions to reduce their environmental impact. Through the ClimaTerra project, the Institut de l'élevage and the INOSYS Réseaux d'élevage have tested various adaptation and mitigation strategies across 16 ruminant systems, each one being representative of the diverse pedoclimatic conditions found in France. The work was conducted using modelled case studies, which were deliberately degraded to simulate the effect of climate-related hazards—primarily those observed in 2022. Different combinations of adaptation measures were then analysed to identify technically viable solutions. The economic and environmental impacts, including carbon footprint, of each scenario were quantified. The scenarios explored three main adaptation pathways: forage diversification, optimization of herd management, and extensification. These approaches aim to restore forage autonomy under climate stress. In most cases, GHG emissions were reduced, and farmer income improve compared to a scenario without adaptation practices.

Keywords: adaptation, climate change, economic impact, livestock, mitigation, strategy

Introduction

Ruminant livestock systems are facing a dual challenge in the context of climate change: they must adapt their practices to cope with increasing climate hazards while also mitigating GHG emissions to reduce their environmental impact. In France, climate projections indicate a temperature increase ranging from +1 to +3.9°C by the end of the century (Soubeyroux *et al.*, 2020). The ClimaTerra national project was launched to support farmers in adapting to these changes. One of the projects key actions involved developing reference scenarios that incorporate both adaptation and mitigation strategies for typical agricultural systems. This study focuses specifically on the results related to grassland-dominated ruminant systems.

Materials and methods

The study was based on 16 farming models, each representative of a specific region in France. These 'typical cases' were developed within the framework of the INOSYS-Livestock Network reference system (Charroin *et al.*, 2005). The selection criteria included: representativeness of 5 main French ruminant sectors, geographic distribution, and complementarity with other existing studies.

This article focuses on seven of the 16 cases, chosen for their emphasis on grass-based systems in mountain (non-pastoral) and lowland areas. These include: two dairy cattle systems, one beef system, two meat sheep systems, one goat system and one mixed cattle-sheep system.

By design, the typical cases operate under optimized conditions in an average climate year without hazards. To evaluate the relevance of different adaptation strategies, the climate condition of

Table 1. Description of scenarios

Area (sector)	Average fodder deficit	Scenario 0 (S0)	Scenario 1 (S1)	Scenario 2 (S2)
Lowland systems (dairy cattle, sheep meat, mixed)	15%	Buying fodder. Reduction in cereal sales	Increase in forage area at the expense of cereal area. Grass management changes	Scenario 1+ Change of breed. Modification of the breeding management. Reduction of unproductive animals
Mountain systems (dairy cattle, beef cattle, goats)	39%	Buying fodder	Reducing unproductive animals	Scenario 1+ Optimization of grazing, early mowing

2022—considered representative of the average year in 2050 in terms of temperature—were used as reference. This year is marked by severe summer water deficit and high temperature peaks. For each case, impacts were assessed on forage and crop yields, and in some instances, on herd productivity, using data from 1,500 farms monitored by the INOSYS Livestock Network.

Three scenarios were analysed: (1) short-term reaction (S0), (2) medium-term anticipation (S1), and (3) long-term systemic evolution (S2). In parallel with adaptation strategies, mitigation levers were also identified and evaluated from technical, economic (based on the 2023 economic context) and environmental perspectives using the CAP'2ER[®] tool. While the results are not intended to be generalized, they provide valuable insights into emerging trends and potential pathways for climate-resilient livestock farming.

Results and discussion

The impact of a hot and dry year like 2022 varies significantly between lowland and mountain livestock systems. The forage deficit, measured against herd requirements, was 15% in lowland areas compared to 39% in mountain regions. This disparity is primarily due to more challenging pedoclimatic conditions in mountainous areas and the less resilient forage systems based on permanent grasslands. The different scenarios are described in Table 1. Lowland systems benefit from greater flexibility, as they typically include crops that can be redirected to feed livestock or used to expand forage areas. In contrast, mountain grassland systems, dominated by permanent grasslands, offer limited room for manoeuvre. Some sectors have access to specific adaptation levers. For example, in sheep farming in lowland system, it is possible to increase the grazing capacity by utilizing additional areas outside the farm, such as cover crops, inter-row species, and other temporary grazing options.

The quantified impacts of scenario 2 (S2) compared to scenario 0 (S0) are presented in Table 2 using three key indicators: technical (forage autonomy), economic (income per labour unit (LU) and climate (GHG emission in kg CO₂ eq/ha of agricultural area AA). In lowland systems, the adaptation levers implemented in S2 successfully restored forage autonomy at the farm level in all cases. However, this was not consistently achieved in mountain systems where constraints are greater. Significant changes to the livestock systems, particularly in herd management, led to income improvements ranging from 11 to 74%, with one exception. Additionally, the income levels assessed in S2 is improved compared to S0, this shows that anticipating climate hazards by modifying these practices is a successful strategy. However, the S2 income does not reach those of a situation without climatic hazards in the 2023 economic context, raising important questions about how to sustain farm income in a future climate

Table 2. Quantified impacts of the comparison of scenarios 0 and 2

Zone	Sector	Forage autonomy in S2 (%)	Evolution in percentage of income/LU between S0 and S2	Evolution in percentage of GHG emission (GHG kg CO ₂ eq/ha AA) between S0 and S2
Plain	Dairy cattle	100	+14%	−15%
	Meat sheep	100	+30%	+4%
	Meat sheep	100	+11%	−17%
	Sheep-beef	100	−3%	−5%
Mountain	Dairy cattle	76	+13%	−10%
	Beef cattle	100	+74%	−40%
	Goat	67	+46%	−8%

context, even when systems are adapted. Finally, the environmental impact also improved in all but one scenario, largely due to the reduction of unproductive animals. These findings demonstrate that, within the studied cases, achieving the dual objective of adaptation and mitigation is indeed possible.

Due to a lack of detailed references, the impact on labour could not be quantified. However, it is important to note that the combinations of adaptation and mitigation levers studied tend to increase the complexity of system operations, and their implications for labour should not be overlooked. Finally, the study tested only one combination of hazards, a hot and dry year like 2022. It would be valuable to assess the impact of a different climatic conditions, such as the very wet year of 2023, to determine whether the scenarios explored are robust and adaptable to a wider range of climate events.

Conclusion

The simulations carried out demonstrate that anticipating climate hazards through changes in practices and the evolution of forage and livestock systems can help absorb the impacts of extreme weather events while also contributing to the reduction of greenhouse gas emissions. However, important questions remain regarding sustainability of economic performance in a changing climate, as well as the evolving workload that farmers must manage under increasingly complex systems.

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Enteric methane and milk production from cows grazing annual ryegrass alone or with forage legumes

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Abstract

This study evaluated the effects of reduced nitrogen fertilization and the intercropping annual ryegrass (*Lolium multiflorum* Lam.) with forage legumes—common vetch (*Vicia sativa* L.) and red clover (*Trifolium pratense* L.)—on milk production and enteric methane emissions in grazing dairy cows. Twelve Holstein × Jersey cows were assigned to a crossover design involving two treatments: ryegrass monoculture (RG) or ryegrass-legume mixture (RG+Leg). Methane emissions were measured using GreenFeed® systems; grazing behaviour, milk yield and composition were also assessed. Legume inclusion contributed ~9% of the pre-grazing biomass and cows grazing RG+Leg pastures had lower herbage mass (1549 vs. 1773 kg DM ha⁻¹) and lower herbage allowance (45.8 vs. 51.7 kg DM cow⁻¹) than cows on monoculture ryegrass. Energy-corrected milk (ECM) and methane emissions (g day⁻¹ and g (kg ECM)⁻¹) were not significantly affected by treatment. These results suggest that, under subtropical grazing conditions, reducing nitrogen fertilization combined with the modest inclusion of vetch and red clover does not mitigate enteric methane emissions nor enhance animal performance. Enhanced strategies to increase legume proportion in mixed swards are needed to unlock their potential for sustainable intensification of pasture-based dairy systems.

Keywords: dairy cow, grazing, methane

Introduction

Pasture-based dairy systems are recognised as a sustainable model for milk production, especially when high-quality forages are available (Galloway *et al.*, 2018). While mixtures of forage legumes and perennial grasses have been extensively studied and adopted to improve sward nutritive value, enhance milk yield, and reduce environmental burdens (Peyraud *et al.*, 2009), the integration of legumes into annual grass pastures remains less explored. This study aimed to assess the effects of reducing N fertilisation and introducing intercropping legumes with annual ryegrass on herbage quality, milk production, and CH₄ emissions. We hypothesised that legumes would improve forage quality, milk yield and reduce methane intensity.

Materials and methods

The experiment was conducted in Lages, Santa Catarina, Brazil (27°47'S; 50°18'W; 920 m asl), from September to November 2023. A 4.2-ha experimental area was divided into two subplots (monoculture ryegrass: RG; ryegrass intercropped with legumes: RG+Leg), each subdivided into paddocks for adaptation and data collection.

Treatments involved annual ryegrass (*Lolium multiflorum* Lam.) either as a monoculture or intercropped with common vetch (*Vicia sativa* L.) and red clover (*Trifolium pratense* L.). Ryegrass was sown

Table 1. Herbage characteristics of annual ryegrass (*Lolium multiflorum* Lam.) alone (RG) or annual ryegrass+legumes (RG+Leg)

	RG	RG+Leg
Pre-grazing herbage mass (kg DM ha ⁻¹)	1773	1549
Herbage allowance (kg DM cow ⁻¹)	51.7	45.8
Pre-grazing herbage height (cm) ¹	32.4	28.2
Post-grazing herbage height (cm) ¹	18.2	16.9
Defoliation severity ²	0.44	0.40
Legumes (g (kg DM) ⁻¹)	0.0	89.0

¹ Measured with sward stick.

² Defoliation severity = (pre-grazing herbage height–post-grazing herbage height)/pre-grazing herbage height.

at 40 kg ha⁻¹ and legumes at 6 kg ha⁻¹. Nitrogen application was 150 kg ha⁻¹ for RG and 75 kg ha⁻¹ for RG+Leg. Rotational grazing was employed (four days of occupation), targeting 40–50% defoliation. Twelve early-lactation Holstein×Jersey cows (67 ± 24.6 days in milk) were assigned to treatments using a crossover design (two 12-day periods: eight for adaptation, four for data collection). Cows were supplemented daily with 4 kg of ground sorghum and 160 g of a mineral mix.

Pasture height was assessed before and after grazing in each period by taking 100 measurements per paddock using a sward stick. Pre-grazing herbage mass was estimated from a calibration curve developed with a rising plate meter (Farmworks®, model F200, Palmerston North, New Zealand), based on five sampling points where compressed height was recorded and the aboveground biomass was harvested from 0.1 m² quadrats. The mean pre-grazing compressed height was calculated from 200 readings obtained with the rising plate meter.

Milk yield and composition were evaluated during the last four days of each period. Milk production was recorded daily during both milkings (7 am and 3 pm), using an electronic meter (Waikato Milking Systems, Hamilton, New Zealand). Milk samples from both milkings were collected daily on the same days that milk production was recorded, for subsequent chemical composition analysis. Methane emissions were measured using two GreenFeed® systems (C-Lock, Rapid City, SD, USA). Each cow accessed the system on two separate days—specifically on days 9 and 11 of each experimental period—ensuring gas sampling every 3 h over a 24-h cycle.

All statistical analyses were conducted using the MIXED procedure of SAS (SAS Institute, Cary, NC, USA). The model included the fixed effect of treatment and random effects of period.

Results and discussion

Pre-grazing herbage mass averaged 1773 kg DM ha⁻¹ in RG paddocks and 1549 kg DM ha⁻¹ in RG+Leg paddocks, with daily herbage allowance exceeding 50 kg DM cow⁻¹ in RG and reaching 45.8 kg DM cow⁻¹ in RG+Leg (Table 1).

Daily CH₄ emissions and emission intensity were similar between treatments, averaging 326 g day⁻¹ and 11.3 g (kg ECM)⁻¹, respectively (Table 2). Milk yield, ECM, milk fat and milk protein did not differ between treatments. Contrary to the hypothesis, red clover and vetch inclusion did not enhance milk production or reduce methane emissions. The low legume proportion (approx. 9% DM) likely limited their contribution to forage quality and overall dry matter yield, reducing potential benefits. In previous studies, positive effects have been observed with higher legume contents (≥20% DM) or

Table 2. Methane emissions, milk yield, and milk composition of dairy cows grazing annual ryegrass (RG) or ryegrass+legumes (RG+Leg).

	RG	RG+Leg	rsd	P value
Methane				
g day ⁻¹	330	321	21.4	0.362
g (kg ECM) ⁻¹	11.0	10.6	1.01	0.377
Milk production (kg day ⁻¹)	31.9	29.9	1.23	0.040
ECM production (kg day ⁻¹)	30.9	30.0	1.66	0.393
Milk fat (g kg ⁻¹)	38.5	40.7	3.44	0.282
Milk protein (g kg ⁻¹)	33.4	32.3	1.71	0.273

rsd, residual standard deviation; ECM, energy-corrected milk production, calculated as follows: ECM = kg of milk production × (37.6 × fat (g kg⁻¹) + 20.9 × protein (g kg⁻¹) + 948) / 3138.

more competitive species and adapted management (Lüscher *et al.*, 2014). Although legumes often improve herbage nutritive value and milk output (Dineen *et al.*, 2018), such effects may not materialise when herbage mass is insufficient (Ribeiro-Filho *et al.*, 2005).

Similarly, the absence of a CH₄ reduction effect may be linked to the low content of secondary metabolites in the species used, as the methane-mitigating potential of legumes is often associated with tannins or other bioactive compounds (Niderkorn *et al.*, 2011).

Conclusion

Forage legumes have potential to enhance productivity in pasture-based dairy systems, but require sufficient sward proportion. While methane emissions did not decrease significantly, reducing nitrogen fertiliser by 75 kg ha⁻¹ lowered the environmental footprint. Establishment and persistence under rotational grazing in subtropical conditions remain key challenges.

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Technological development for monitoring pasture quality and supporting grazing management decisions: a review

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Abstract

Effective grazing management depends on accurate and timely information on pasture biomass and nutritive value. This review examines recent advances in sensing technologies for estimating forage quality parameters, particularly crude protein (CP) and neutral detergent fibre (NDF), in pasture-based systems. Emphasis is placed on two complementary indicators: Dry Matter (DM) and Crude Protein (CP). Proximal tools such as rising plate meters (RPM) have evolved to incorporate optical components capable of estimating vegetation indices like the Normalized Difference Vegetation Index (NDVI), which may correlate with forage quality. Meanwhile, satellite-based remote sensing (Sentinel-2) offers broader spatial coverage and access to multiple spectral bands. These allow for the computation of several indices that can be explored using statistical and machine learning (ML) models to predict CP and NDF content. The integration of sensor-derived data with artificial intelligence (AI) represents a promising avenue for developing predictive models and decision-support systems (DSS), improving rotational grazing, supplementation planning, and reducing the occurrence of metabolic and nutritional disorders. This review highlights key findings in the literature and identifies knowledge gaps, particularly in the validation of new technologies across different pasture types and seasons. Future research should focus on combining multi-source data for real-time, on farm applications.

Keywords: fibre, grazing management, pasture crude protein, predictive models, proximal and remote sensing

Introduction

Climate change is widely recognized as one of the greatest challenges of our time, with profound effects on agriculture, food security, and rural economies. These issues, accompanied by a rising global demand for food, require a wide range of adaptation and mitigation strategies including the integration of precision agriculture (PA) technologies, which potential usage in grassland management should be more fully explored and applied (Ali and Kaul, 2025, Meena *et al.*, 2025). PA uses advanced technologies, including proximal and remote sensing, to support data-driven decisions. These tools improve resource efficiency, optimise yields, reduce environmental impacts, and enhance resilience to climate variability (Meena *et al.*, 2025).

Monitoring and estimating pasture availability and quality parameters plays a crucial role in maximizing grass production and utilization (Serrano *et al.*, 2024), allowing better defining fertilization strategies, irrigation schedules, and the frequency of pasture utilization in pasture-based farms (Ali and Kaul, 2025). Key parameters for assessing pasture quality in grazing systems include DM, dry matter

digestibility, metabolizable energy, organic matter digestibility, CP, and water-soluble carbohydrates. Chemical composition, water content, and nutrient concentration are also widely recognized as indicators of nutritive value, with both CP and NDF being the most used chemical measures, namely in animal production systems (Serrano *et al.*, 2024).

PA technologies have great potential for enhancing the accuracy and availability of information on grasslands, but their application remains challenging due to high spatial variability of soil and pasture characteristics, as well as the dynamic and seasonal nature of grass species (Serrano *et al.* 2024). Despite high costs, this variability can be reduced through integrating multiple sensor technologies (Martelli *et al.*, 2025) and, according to Ali and Kaul, 2025, it is possible to combine proximal and remote sensing with machine learning models (ML) for predictive modelling purposes. Research has been carried out on using multiple sensors combined or integrated into the estimation models, however, the integration of pasture DM availability (in kg ha^{-1}) measured by RPM, and pasture quality, measured by active optical sensor (AOS), combined with ML and AI as a tool for supporting management decisions, is still a gap in literature, particularly those related to nutritional and metabolic disorders that can negatively impact herd performance, reproduction (Abbasi, 2017) and lipid-related metabolic disorders (Hayirli and Grummer, 2004).

Overview

As mentioned above, CP and DM are two key parameters for decision-makers in animal production systems, as indicators of pasture availability and quality, with DM expressed in kg ha^{-1} and CP, in % of DM (Serrano *et al.* 2024). Over the last twenty years, many studies have been published on the evaluation of technologies for estimating and monitoring pasture herbage availability, using both proximal and remote sensors. The RPM records a combined measure of pasture sward height and density, referred to as Compressed Sward Height, and is the most frequent measurement used for DM herbage monitoring in pastures (Serrano *et al.* 2024). A holistic precision grass measurement and analysis system to optimize pasture-based livestock production is under development at Moorepark Animal and Grassland Research Centre in Cork in Ireland, using a range of measurement technologies to provide yield and quality data to a cloud-based platform (Murphy *et al.*, 2019). A smart automated RPM with GPS capabilities was used to record Compressed Sward Height, however, more laboratory methods and remote sensing were carried out to validate the decision support system. Duli Zhao *et al.* (2006) assessed CP, NDF and above-ground biomass, using a spectroradiometer in warm season grass pastures to develop and validate algorithms for prediction purposes. Despite the different works developed around the monitoring and prediction models of CP and DM, the integration of the two essential dimensions, pasture herbage availability (related with the compressed height measured by RPM) and pasture quality (related by NDVI measured by optical sensor), in a single ground-based data technology, to obtain a global indicator, CP in kg ha^{-1} , has not yet been carried out (Serrano *et al.*, 2024).

Technological development proposed

Aiming to validate two complementary sensors, Serrano *et al.* (2024) evaluated the possibility of using the RPM and AOS to obtain CP in kg ha^{-1} , as a global indicator. This parameter that simultaneously integrates two essential dimensions (DM in kg ha^{-1} measured by RPM, and pasture quality measured by AOS) was shown to have the potential of a decision-making and management tool. Bearing this in mind, a new sensor that combines the two mentioned technologies (RPM and AOS) is proposed in this work as a possible accessible tool for obtaining CP in kg ha^{-1} , through a ground-based sensor (Figure 1).

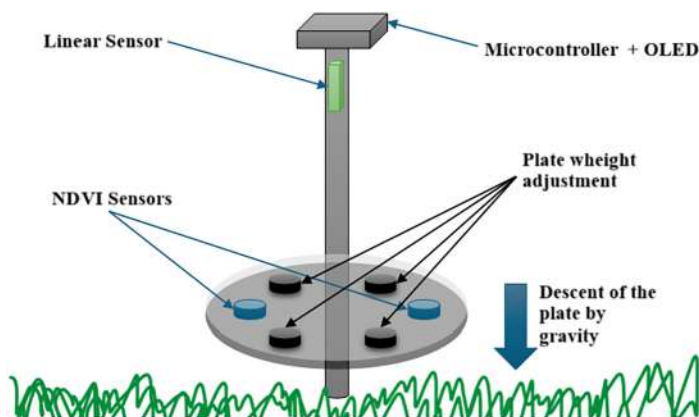


Figure 1. New sensor proposed, integrating RPM and optical sensors.

Given the spatial variability and seasonal changes of pastures, along with differences in operator bias and labour costs across farming systems, long-term studies are needed to validate this new sensor for use in predictive models and DSS. Integrating this technology with multi-source data, AI, ML offers great potential to improve rotational grazing, supplementation planning, and the prevention of metabolic and nutritional disorders in grazing ruminants.

Conclusions

Future research should focus on real-time, on-farm applications, while efforts to raise awareness about the benefits of precision technologies, especially for frequent, accurate measurements of pasture availability and quality, essential to ensuring the economic and environmental sustainability of extensive livestock systems.

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Assessment of *Gliricidia sepium* silage enriched with cassava root for cattle production in Costa Rica

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Abstract

C4 grasses are the primary feed for tropical cattle but require supplementation due to low protein and digestibility. This study evaluated *Gliricidia sepium* silage mixtures with *Manihot esculenta* (cassava) tubers at 0, 10, 20 and 30% substitution. After 50 days, nutritional value, fermentation parameters, and rumen degradability of dry matter (DM) and crude protein were assessed. Cassava inclusion significantly affected fermentation, increasing non-fibrous carbohydrates (34.5 to 46.1% DM) while decreasing crude protein (23.5 to 17.9% DM) and neutral detergent fibre (28.7 to 24.3% DM). Although crude protein concentration decreased, total DM increased, resulting in similar potential protein intake across silages. Cassava enhanced DM solubility and effective degradability, although protein solubility decreased.

Keywords: forage preservation, nutrition, rumen, silage

Introduction

Tropical livestock farming often relies on low-quality pastures, requiring better forage (Barbizan *et al.*, 2020). *Gliricidia sepium*, a high-protein legume (17–22%), is an excellent alternative for silvopastoral systems. However, preserving this resource during periods of scarcity presents a significant technical challenge (Villanueva-López *et al.*, 2015).

Silage preserves forage, but tree legumes often lack the carbohydrates required for proper fermentation (Borreani *et al.*, 2018). Supplementing with energy sources like cassava root, can ensure rapid acidification and preservation, due to their fermentable carbohydrates content. This study aimed to assess the effect of adding cassava root on the nutritional quality and fermentation of *G. sepium* silage, optimizing local resources for sustainable feeding systems.

Materials and methods

The experiment was conducted at the University of Costa Rica and the National University of Costa Rica between July and November 2021. *Gliricidia sepium* leaves and cassava root tubers (CRT) were obtained from commercial farms. The design combined *G. sepium* leaves with four levels of CRT substitution (0, 10, 20 and 30% feed basis). All silages were supplemented with 5% molasses and a farm-made inoculum. Ingredients were chopped, mixed, vacuum-packed in 5.0 kg silos, and stored for 50 days (four replicates per treatment). Afterward, pH and ammonia nitrogen (AOAC, 1998) were measured.

Table 1. Fermentation quality of the experimental silages according to the percentage of cassava root tubers (CRT) and the chemical composition and nutritional value (g (kg DM)⁻¹) of those silages and the feeds used to elaborate them.

Item	CRT	<i>G. sepium</i> leaves	% CRT				SEM	<i>p</i> -value
			0	10	20	30		
pH			4.20 ^a	4.22 ^a	3.90 ^b	3.89 ^b	0.058	0.005
Ammonia nitrogen (%)			16.33 ^a	16.00 ^{ab}	14.00 ^{bc}	13.00 ^c	0.471	0.004
Dry matter	368.0	251.8	244.9 ^c	283.3 ^a	272.9 ^b	280.1 ^{ab}	0.203	<0.001
Crude protein	44.4	267.1	234.8 ^a	183.9 ^b	188.7 ^b	179.0 ^b	0.709	0.002
NFC	837.3	189.2	347.2 ^b	436.2 ^a	428.0 ^a	462.8 ^a	1.349	0.002
NDF	84.0	424.6	287.3	265.7	262.3	243.3	0.908	0.054
Lignin	7.0	106.7	73.7	73.0	67.3	62.7	0.483	0.386
NFC/NDF ratio			1.21 ^b	1.63 ^{ab}	1.66 ^{ab}	1.90 ^a	0.103	0.009

NFC, non-fibrous carbohydrates; NDF, neutral detergent fibre. ^{a,b,c,d} Means with different letters within each row differ significantly ($p < 0.05$).

Dry matter (DM) and crude protein (CP) were determined using AOAC (1998) methods. Neutral detergent fibre (NDF) and acid detergent lignin (ADL) were analysed following Van Soest *et al.* (1991).

Two cannulated dairy cows fed *Cynodon nlemfuensis ad libitum* were used; two replicates per silo were incubated for 0, 2, 4, 6, 8, 12, 24, 48 and 72 hours. *In sacco* degradation coefficients of DM and CP were estimated using Ørskov & McDonald (1979) equations for potential degradability (PD) and effective degradability (ED):

$$PD = a + b(1 - e^{(-ct)}) \quad (1)$$

$$ED = a + bc/(c + k) \quad (2)$$

where t is incubation time, a is the soluble fraction, b is the degradable fraction, c is the degradation rate, and k is the passage rate (5% h⁻¹ and 8% h⁻¹). Data were analysed via ANOVA and repeated measures mixed models using R version 4.3.1. Tukey's test identified significant differences ($p \leq 0.05$) and trends ($0.05 < p \leq 0.10$).

Results and discussion

Table 1 presents the composition of *G. sepium* and cassava root tubers (CRT). CRT improved silage pH and ammonia nitrogen by providing soluble carbohydrates (Borreani *et al.*, 2018). However, ammonia nitrogen exceeded 10% due to protein hydrolysis from low DM. CRT increased DM and Non-fibrous carbohydrates (NFC), enhancing fermentation and digestibility (Borreani *et al.*, 2018). Although CP decreased, levels remained suitable for ruminants (Calsamiglia *et al.*, 2010). NDF reduction occurred without lignin changes, potentially improving intake and energy balance (Barbizan *et al.*, 2020). Consequently, optimal NFC/NDF ratios in CRT silages indicate improved nutrient utilization without harming rumen health (de Melo *et al.*, 2021).

Table 2 presents the rumen degradability characteristics of the experimental silages. CRT addition increased DM solubility, likely due to higher NFC content, which optimizes rumen degradable DM (Barbizan *et al.*, 2020). Conversely, CP solubility tended to decrease, possibly due to the reduction in CP derived from forage (20–30%). Meanwhile, the degradable fraction of DM tended to decrease, while that of CP tended to increase; this suggests a need to reformulate the diet to promote nutrient synchronization (Calsamiglia *et al.*, 2010) and potentially increase rumen microbial protein synthesis

Table 2. Rumen degradability of the experimental silages according to replacement by cassava root tubers (CRT).

Item*	% CRT				SEM	p-value
	0	10	20	30		
DM degradability						
<i>a</i> (% DM)	37.68 ^b	45.61 ^{ab}	53.09 ^a	48.01 ^a	1.900	0.001
<i>b</i> (% DM)	33.94	26.55	23.16	30.09	2.713	0.078
<i>c</i> (% h ^{−1})	0.104	0.117	0.074	0.094	0.015	0.282
ED0.05(% DM)	60.29	63.85	66.69	67.26	2.537	0.246
ED0.08 (% DM)	52.54 ^b	57.71 ^{ab}	59.79 ^{ab}	64.01 ^a	2.045	0.014
CP degradability						
<i>a</i> (% CP)*	58.10	55.45	50.64	49.99	2.285	0.079
<i>b</i> (% CP)*	25.54	29.37	32.92	33.25	2.053	0.068
<i>c</i> (% .h ^{−1})	0.108 ^c	0.146 ^{bc}	0.182 ^{ab}	0.222 ^a	0.011	<0.001
ED0.05(% CP)	75.59	77.22	76.73	76.84	0.685	0.411
ED0.08 (% CP)	72.93	67.54	70.30	68.41	2.404	0.431

DM, dry matter; a, the soluble fraction; b, the potentially degradable fraction; c, the rate of degradation of b; potential degradability (72 h) was calculated as a + b; ED, effective degradability at passage rate of 5% h⁻¹ and 8% h⁻¹; CP, crude protein. ^{a,b,c,d} Means with different letters within each row differ significantly ($p < 0.05$).

(Grubješić *et al.*, 2019). ED showed differences at a passage rate of 0.08% h⁻¹, possibly due to rapidly degradable NFC synchronizing with degradable forage protein (Barbizan *et al.*, 2020).

Conclusion

The inclusion of CRT improved fermentation by reducing pH and ammonia concentrations in silages. Also, it reduced CP while it increased content of non-fibrous carbohydrates. Changes in nutritive value by the addition of CRT increased the solubility and effective degradability of DM of silages. Based on these results, *G. sepium* silages containing 20–30% CRT can be considered valuable feed supplements for tropical cattle.

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Key factors influencing successful rebreeding in compact calving grass-based dairy systems

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Abstract

In pasture-based systems, compact calving of dairy cows is often practised to synchronise the grass growth profile with the herd feed demand. Consequently, the breeding season is short and dairy cow fertility is of major importance. During 14 years, the performance of two grass-based systems, with or without maize silage and concentrate, and including both Holstein (Ho) and Normande (No) dairy cows were compared on the INRAE experimental farm at Le Pin (Normandy). The capability to be in-calf after 13 weeks of insemination on natural heats has been evaluated, comparing the characteristics of the pregnant (Ho 56% and No 72%) and non-pregnant cows. The first group of parameters which influence the success for recalving concerns the previous calving. Dairy cows in-calf calve at 8 (Ho) to 16 (No) days earlier in the year, with fewer major calving problems—notably in No breed, and have more time to re-establish cyclicity before the next breeding season. A second group concerns the quality of the ovarian cycle, assessed by the progesterone profile. The Ho dairy cows in calf have more normal cycles (54 vs 35%). As a consequence, the 21-day submission rate is double (50% vs 25%) for the Ho and No cows in-calf. These factors are expressed differently according to the breed and the feeding system studied. A third group of factors concern the animal performance, as milk and milk solids (fat plus protein) yield expression and body reserve mobilisation. However, the difference between the groups of cows in-calf or not are not significant. Getting pregnant at the right time during a short breeding season is a real challenge for Ho and No “continental” dairy cows, and the above criteria must be favourable to achieve successful rebreeding to increase herd longevity.

Keywords: breed, dairy cow, feeding level, grass system, in-calf success

Introduction

In temperate regions, grass growth is seasonal, with a maximum observed in late spring and less growth in late summer and autumn. Consequently, in pasture-based systems, compact calving of dairy cows is recommended in spring to synchronise the herd feed demand with the grass growth profile (Delaby and Horan, 2021). For a dairy cow to be in-calf during the short breeding season associated with compact calving is a real challenge. All the stages from calving to re-calving are important, and many cows are unable to meet all the requirements in a short period. In light of the “The cow for the system?” experiment (Bedere et al, 2017) undertaken over 14 years on the INRAE experimental farm at Le Pin (Normandy; [10.15454/1.5483257052131956E12](https://doi.org/10.15454/1.5483257052131956E12)), the aim of this study is to describe the characteristics of pregnant and non-pregnant cows, and to highlight the parameters influencing reproductive success in contrasted grass-based systems with compact calving.

Materials and methods

Between 2006 and 2019, two grass-based feeding systems were compared for Holstein (Ho) and Normande (No) dairy cows. Every year, the 35 cows allocated to the High (H) feeding system received

a high quality TMR (% DM—Maize silage (MS) 55%—Dehydrated alfalfa 15% and Concentrate (Cc) 30%) *ad libitum* in winter (Jan to March). On pasture, H cows had access to 0.35 ha/cow and received 4 kg of Cc. The grazing area allows 100 days of grazed grass (mid-April to mid-July), and afterwards, the cows received 5 to 8 kg DM of MS as supplement up to mid-November. The 35 cows on the Low (L) feeding system are fed in winter with a ration composed of 50% grass silage (GS) and 50% haylage (% DM), without any Cc. At grazing, the platform area (0.55 ha/cow) is enough to permit 180 to 200 grazing days (from mid-April to mid-November), always without Cc. Some years, during 3 to 4 weeks in summer, the L group received GS as supplement to compensate for insufficient grass growth. During the dry period, all cows received a mixed ration composed of 65% GS and 35% hay. The 90-day breeding season is from 21 March to 21 June, with artificial insemination (AI) on spontaneous oestrus and heat visually detected (5 times per day—no technological assistance), authorised only after 42 days postpartum. Due to the rule of the 42 days, some cows calving late have an incomplete breeding season (<90 days; Table 1). Individual milk yield (MY) has been recorded twice daily at each milking. Fat and protein content was evaluated on am and pm individual milking samples, 3 times per week. From a week after calving to mid-July, individual cow milk samples were collected on Monday, Wednesday and Friday and stored at -20°C . At the end of each year, the milk progesterone (P4) concentration was determined using commercial ELISA kits. The P4 profiles have been quantified as described previously by Bedere et al (2017). The cows are weighed every week and the body condition score (BCS: 0 to 5) is evaluated every month by two identical experienced assessors.

The objective is here to describe similarities and differences between cows that are in-calf or not after the breeding season. Within breed, some characteristics describing the previous calving (date, conditions), the P4 profiles (normal or not), the breeding process (intervals between calving, breeding season starting, AI's), the lactation performance (total MY or milk solids) and the BCS variation have been analysed with the R 4.4.3 (2019) software according a linear model including the year (Y), feeding system (FS), parity (P), calving difficulty (CD), in-calf (InC) and the simple interactions between InC and FS, P or CD. The binary data (0/1) have been analysed according to a binomial approach, with the same explanatory factors.

Results and discussion

The final database includes 358 Ho and 437 No lactations, with 56.4% and 71.6% pregnant. In both FS and both breeds, cows that become pregnant have an earlier previous calving date (between 8 to 16 days according to groups) than cows that are not in-calf and consequently have more time during the breeding season to recover cyclicity and regular cycles. This has an important consequence for breeding season duration, which is shorter in the case of late calving. The proportion of cows which have access to a full breeding season, particularly for No cows, is less for the groups not in-calf. The interval between calving and cyclicity recovery (39 and 31 days in average for the Ho and No cows) does not differ significantly between FS or InC, but the interval between cyclicity recovery to start of the breeding season (March, 21) is significantly lower for the future empty cows. This shorter interval, here due to late calving date, is known to be unfavourable to the success of the first AI, as the first cycles after the cyclicity recovering, are often less fertile. In the case of No cows only, difficulties at calving, like caesarean or metritis, are significantly more frequent in the not in-calf population (23 and 33% respectively in the H and L FS). For the Ho group only, the proportion of normal cycles was significantly higher in future pregnant cows (> 50%), whereas it was very low for the empty Ho cows in the Low FS group (27%).

Table 1. Characteristics of cows in-calf or not, according to breed and feeding system.

Feeding system	Holstein (<i>n</i> = 358)				In-calf (Pr<)	Normande (<i>n</i> = 437)				In-calf (Pr<)
	High (<i>n</i> = 174)		Low (<i>n</i> = 184)			High (<i>n</i> = 218)	Low (<i>n</i> = 219)			
	In-calf (<i>n</i> = 98)	Not (<i>n</i> = 76)	In calf (<i>n</i> = 104)	Not (<i>n</i> = 80)						
								In calf (<i>n</i> = 160)	Not (<i>n</i> = 58)	
Calving difficulty (%)	23.7 b	24.6 b	13.6 a	19.0 ab	NS	15.2 b	23.1 bc	8.0 a	33.3 c	0.001
Calving day of year	22.4 a	33.1 ab	29.2 ab	37.4 b	0.010	27.6 a	44.0 b	26.6 a	43.3 b	0.001
Breeding season duration (days) (2)	89.0 a	84.7 ab	85.8 ab	80.2 b	0.007	87.3 a	79.2 b	86.2 a	79.3 b	0.001
Complete breeding season (%)*	81.7 a	61.8 b	74.5 ab	61.1 bc	0.009	73.1 a	40.8 b	76.3 a	51.4 b	0.001
Cyclicity to breeding season start (days)	17.7 a	8.1 ab	18.0 a	2.4 b	0.007	19.5 a	6.6 b	24.7	6.8 b	0.001
Calving to 1st AI (days)	84.3 ab	86.4 ab	76.0 a	89.4 b	0.020	81.0 a	73.8 a	78.3 a	78.9 a	NS
Normal profile cyclicity (%)	55.2 a	43.4 a	53.4 a	26.9 b	0.006	64.2 a	67.1 ab	78.6 b	63.8 a	NS
21 days submission rate (%)**	48.7 a	26.1 b	51.3 a	20.0 b	0.001	41.0 b	23.6 a	57.4 c	25.2 a	0.002
21 days submission rate (%)*	55.2 ab	44.1 a	68.3 b	40.7 a	0.006	53.3 a	51.3 a	73.4 b	48.9 a	0.05
Milk yield 44 weeks (kg)	8153 a	8226 a	5962 b	5768 b	NS	6214 a	5863 b	4447 c	4584 c	NS
Max milk yield (kg)	40.2 a	38.8 a	29.2 b	27.4 b	NS	30.0 a	28.0 b	21.7 c	21.7 c	NS
Milk solids 44 weeks (kg)	553 a	560 a	406 b	397 b	NS	456	437	332	340	NS
BCS at calving (pts)	2.80 a	2.80 a	2.75 a	2.60 a	NS	3.40 a	3.55 a	3.05 b	3.20 b	0.030
BCS minimum (pts)	2.10 a	2.00 a	1.70 b	1.65 b	0.066	2.85 a	2.95 a	2.40 b	2.45 b	NS
Max BCS loss (pts)	−0.75 a	−0.85 ab	−1.00 b	−1.00 b	NS	−0.55 a	−0.60 ab	−0.65 b	−0.75 b	NS

Within breed, the letters indicate the difference between the 4 groups and the column In-calf (Pr<) describes the probability of the main effect In-calf or not. Results in italics indicate a significant interaction between FS and InC.

* For each cow breeding season, due to the 42 days post calving.

**21 days submission rate at global breeding season.

As a consequence of all the previous results, the proportion of cows with one AI during the 21 first days of the breeding season is significantly higher (50% vs 25% in average) for the future pregnant cows. This is still true, but less so for this criterion, calculated from the start of the right of AI (at least 42d after calving). Within breed and FS, the milk yield performances are not different between the in-calf and not in-calf groups. Only the No cows not in-calf in the H feeding system produce less than the pregnant cows. Similarly, the BCS and their variations do not differ significantly between pregnant and empty cows of each group of FS.

Conclusion

These results confirm the small effect of the FS on the capability to be in-calf in grass-based dairy systems with compact calving management. Calve early, without calving problems, with a normal cyclicity seems really important to maximise the chance of the cow becoming pregnant. The criterion “day of calving” could be taken into account in selection programmes in order to keep daughters from early pregnant cows.

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Forage species and seasonal cuts affect green biorefinery outputs: mass balance and protein yield

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Abstract

In Norway, green biorefinery is gaining interest due to its potential for domestic feed protein production. This study aimed to quantify the effects of forage species and seasonal cuts on the distribution of dry matter and protein across biomass fractions, and on the yield of green protein concentrate (GPC). A plot experiment was conducted at NIBIO Tuv, Steinkjer (63.9°N, 11.4°E) in 2023 using pure stands of timothy and red clover, in addition to a mixture of timothy, meadow fescue and red clover (70:20:10 seed ratio). Forages were harvested four times in 2024 and processed at lab-scale to separate pulp, brown juice, and GPC. Forage dry matter yields (DMY) and proportions and yields of each fraction, were measured. All samples were analysed for nitrogen content to calculate crude protein (CP). Red clover achieved the highest total DMY (12.3 Mg ha⁻¹) and CP yield (2.76 Mg ha⁻¹), followed by the mixture and timothy. Red clover also resulted in the highest GPC yield (2.15 Mg DM ha⁻¹; 1.16 Mg CP ha⁻¹). Additionally, red clover demonstrated the highest protein recovery efficiency across all cuts. However, since growing red clover in pure stands can be challenging, these findings highlight its importance as a component in forage mixtures to enhance protein extraction in green biorefinery systems.

Keywords: feed protein, green biorefinery, red clover, seasonal variation, timothy

Introduction

Green biorefinery systems can supply domestic, high-quality feed protein. Such systems are especially attractive in the Nordic countries, including Norway, where a strong dependence on imported soybean meal raises concerns about self-sufficiency and supply security. Forage crops can be processed into green protein concentrate (GPC), a valuable feed product for monogastric animals. Previous research highlights that forage species composition and harvesting strategies significantly impact both dry matter yield (DMY) and protein available for biorefinery processing (Stødkilde *et al.*, 2021). In Norway, timothy and red clover dominate grasslands for their adaptability and feed value, while red clover additionally provides biological N fixation and associated soil-fertility benefits. Although studies from other Nordic countries have demonstrated promising results for red clover-based mixtures in biorefinery applications (Santamaría-Fernández *et al.*, 2019), evidence from Norwegian conditions remains limited. The objective of this study was therefore to quantify how forage species and individual harvest cuts (within a four-cut harvest system) affect the mass balance of green biorefinery fractions and the yield of extractable crude protein (CP). By mapping the distribution of the annual harvest across four cuts, this work provides practical information about species choice and harvest management under Norwegian conditions.

Materials and methods

A plot experiment was conducted at the NIBIO research farm Tuv, Steinkjer (63.9°N, 11.4°E), on a silty light clay soil. The three forage stands were established in 2023: pure timothy (*Phleum pratense* L.,

cv. Grindstad), pure red clover (*Trifolium pratense* L., cv. Gandalf), and a mixture of timothy, meadow fescue (*Festuca pratensis* L., cv. Vester), and red clover in a seed ratio of 70:20:10 (by weight). The experiment followed a randomized complete block design with three replicates; each plot measured 10.5 m². All plots received a compound fertilizer (N-P-K; 18-3-15) supplying 181 kg N ha⁻¹ annually. In 2024, forages were harvested four times on the following dates: 25 May, 27 June, 02 August, and 06 September. An internal 8.25 m² area of each plot was used for yield determination and sampling. Plot-level DMY was recorded for each cut and treatment. Subsamples from each harvest were collected and processed as fresh using a lab-scale green biorefinery procedure. Briefly, 750 g of fresh biomass per sample was pressed using a tabletop screw press (Angel 7500, Angel, South Korea) to separate juice and pulp. The press juice was filtered and heated to 85°C, cooled to room temperature, and centrifuged (Eppendorf 5910 Ri, FA-6x250) at 10 000 RCF for 10 min. The supernatant (brown juice) and the precipitate (GPC) were collected. All the fractions were dried at 60°C for 48 hours. Nitrogen (N) content of the forage and of each fraction (pulp, GPC and brown juice) was determined by the Dumas method and CP was calculated as N × 6.25. For each sample, DM and CP mass balances were computed as the proportion of forage DM or CP present in each fraction. Annual yields were obtained by summing forage and GPC yields across the four cuts. Data were analysed with a mixed model (PROC MIXED, SAS). Pairwise comparisons used Tukey's adjustment ($P \leq 0.05$).

Results and discussion

Across all cuts, forage species significantly affected the portioning of DM and CP among biorefinery fractions (Table 1). Red clover allocated markedly less DM and CP to pulp and more to GPC than timothy, with the mixture as intermediate. These findings broadly agree with Damborg et al. (2020), although in contrast to their results, red clover in our study showed higher CP recovery to GPC than to pulp, likely due to differences in processing conditions and harvest maturity. Cut number significantly affected most fractions, and modest forage × cut interactions were seen for DM partitioning in pulp and brown juice. Processing losses were highest for red clover, likely from foam removed during juice filtration.

Table 1. Distribution of DM (g (kg forage DM)⁻¹) and CP (g (kg forage CP)⁻¹) across biomass fractions (pulp, brown juice, GPC) and processing losses.

Item	Timothy	Red clover	Mixture	SEM (<i>n</i> = 12)	<i>P</i> value		
					Forage	Cut	Forage×cut
DM							
Pulp	688 ^a	484 ^c	642 ^b	6.8	<0.001	0.003	0.017
GPC	109 ^c	177 ^a	131 ^b	4.0	<0.001	<0.001	0.093
Brown juice	176 ^b	221 ^a	179 ^b	4.9	0.001	0.005	0.028
Lost	27 ^b	118 ^a	48 ^b	12.2	0.005	<0.001	0.411
CP							
Pulp	633 ^a	366 ^c	520 ^b	12.7	<0.001	0.001	0.230
GPC	256 ^c	425 ^a	342 ^b	6.9	<0.001	0.002	0.068
Brown juice	96 ^c	136 ^a	111 ^b	2.7	<0.001	<0.001	0.194
Lost	15 ^b	74 ^a	26 ^b	11.4	0.023	<0.001	0.919

Mixture, mixture of timothy, meadow fescue, and red clover sown at a 70:20:10 seed ratio by weight. The losses were calculated by difference as 1000 – (Pulp+GPC+Brown juice).

Table 2. Annual yield of dry matter (DM) and crude protein (CP) in forage and green protein concentrate (GPC).

Item	Timothy	Red clover	Mixture	SEM ($n = 3$)	<i>P</i> value
Forage DM yield, Mg ha ⁻¹	7.69 ^c	12.3 ^a	10.2 ^b	0.22	<0.001
CP yield in forage, Mg ha ⁻¹	0.99 ^c	2.76 ^a	1.65 ^b	0.08	<0.001
GPC DM yield, Mg ha ⁻¹	0.75 ^c	2.15 ^a	1.26 ^b	0.04	<0.001
CP yield in GPC, Mg ha ⁻¹	0.25 ^c	1.16 ^a	0.55 ^b	0.02	<0.001

Mixture, mixture of timothy, meadow fescue, and red clover sown at a 70:20:10 seed ratio (by weight).

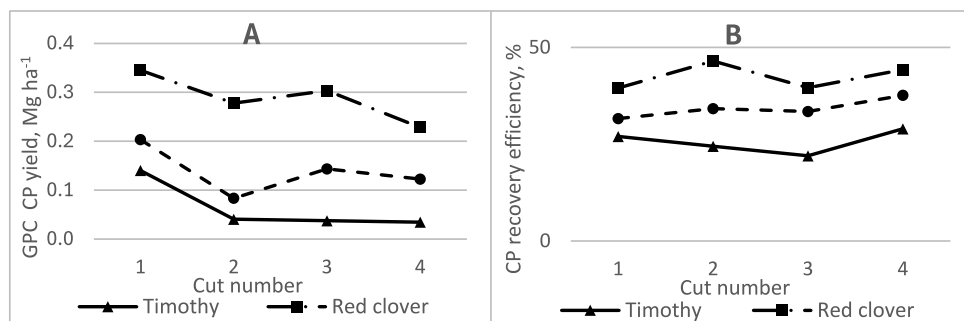


Figure 1. Crude protein (CP) yield (A) in green protein concentrate (GPC) and CP recovery efficiency (B) relative to total forage CP in GPC, for timothy, red clover, and a mixture (timothy, meadow fescue, and red clover) across four cuts.

Annual GPC yields reflected the observed partitioning patterns, and species differences were highly significant for annual forage and GPC yields ($P < 0.001$; Table 2). Red clover delivered the greatest forage DMY (12.3 Mg ha⁻¹) and CP yield (2.76 Mg ha⁻¹), translating to the highest GPC yields (2.15 Mg DM ha⁻¹; 1.16 Mg CP ha⁻¹). The mixture outperformed timothy but was consistently below red clover, indicating clear benefits of including legumes for both biomass and extractable protein. Seasonally, red clover maintained the highest GPC CP yield and CP recovery across the four cuts, with timothy lowest and the mixture as intermediate (Fig. 1). Overall, GPC CP yield declined from the first to the fourth cut for all forages, with a minor mid-season increase (2nd to 3rd cut) for red clover and the mixture, largely reflecting variation in forage yield among cuts. Comparing cuts within red clover, the relationship between GPC CP yield and recovery efficiency likely reflects variation in forage CP content and yield across cuts, rather than a direct trade-off between the two.

Collectively, these results show that red clover maximizes protein recovery into GPC, while grass–legume mixtures offer a pragmatic improvement over pure grass where pure clover stands are agro-nomically challenging.

Conclusion

Species choice was the key driver of green biorefinery performance under Norwegian conditions. Red clover delivered the highest forage and GPC yields, with the mixture consistently intermediate—likely reflecting the legume contribution, though a contribution from meadow fescue cannot be ruled out. The CP yield of GPC varied among cuts, highest at the first. Legume-rich swards with red clover seem most promising for protein recovery in green biorefineries, though other legume species should also be tested.

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Using data-driven grassland tools and fresh grass analyses to reduce dietary crude protein

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Abstract

The Grassland Production Registration project (GPR) followed and supported 20 Dutch dairy farmers, who are also involved in the nationwide project “Koe en Eiwit” (KE). GPR aims to reduce dietary crude protein (CP) levels on dairy farms to 155 g CP per kg DM. Actively using data-driven grassland tools (e.g. the Dutch apps “Grassland Calendar” and “Grass Signal”), combined with weekly fresh grass analyses, is expected to contribute to the insights in grass quality and should make it possible to adjust rations during the grazing season. Analyses showed no differences in CP-level between GPR and KE farms despite a higher proportion of fresh grass in the ration of the GPR farms. Based on this first analysis, it is concluded that data-driven grassland tools can help farmers to reduce the dietary CP level on grass-based dairy farms.

Keywords: data-driven, grassland management tools, reduction of crude protein

Introduction

To mitigate nitrogen emissions from dairy farms to the environment, it is essential to reduce crude protein (CP) in the ration of dairy cows. In certain areas of the Netherlands, the proportion of fresh grass in the ration is relatively high. Due to considerable variation in CP concentration in this grass, it is challenging to reduce dietary CP in grass-based feeding systems. Therefore, understanding how grassland management and grass growth influence CP is crucial. It is not yet possible to accurately predict the CP and dry matter availability of fresh grass based on grassland characteristics, management and temperature. Consequently, to reduce CP of the overall diet (g (kg DM)^{-1}), analysis of the CP concentration in fresh grass is necessary. Access to accurate data and tailored advice on fresh grass quality can help dairy farmers to manage the availability of pasture grass more accurately. This information offers real-time insights into grass quality, like current CP concentration in the grass. This enables farmers to adjust rations accordingly. For example, they might reduce soybean meal and increase maize in the ration to offset high CP in grass. It also helps in fine-tuning fertilisation strategies and determining optimal grazing times. To facilitate data-driven decision-making, newly developed tools (e.g. Grass Signal) are integrated into the grass production registration (GPR) project, with the aim to explore whether GPR and newly developed grass growth tools can help farmers to achieve target CP in the cow rations more effectively.

Materials and methods

The Grassland Production Registration project (GPR) monitored and supported 20 Dutch dairy farmers across the Netherlands in 2024. The GPR project was a spin-off of the national initiative “Koe en Eiwit” (KE), and participating farmers were selected from the KE programme. The KE project aimed to reduce dietary CP on 150 commercial dairy farms to 155 g CP g (kg DM)^{-1} through management strategies (Wind *et al.*, 2024). Strategies were monitored by analysing the Annual Nutrient Cycle

Assessment (ANCA), a model to describe the complete nutrient flow for individual dairy farms (Aarts *et al.*, 2015). However, KE did not address the use of fresh grass—a critical limitation for farmers who practise grazing. To address this gap the GPR project incorporated data-driven grassland tools, such as the Dutch apps ‘Grassland Calendar’ and ‘Grass Signal’ in combination with fresh grass analyses. Within the Grassland Calendar app, farmers recorded grassland characteristics, fertilisation details and dates of harvesting or grazing. Based on these data, Grass Signal provided preliminary predictions for grass yields and CP based on models (Kempenaar *et al.*, 2022).

Weekly fresh grass samples were collected by Eurofins Agro (Wageningen, The Netherlands) on each farm that indicated grazing would begin the following day. Sampling followed a standardized protocol; e.g., grass was sampled on Mondays from pastures scheduled for grazing from Tuesday onward. Samples were directly sent to Eurofins for chemical composition analysis using Near-Infrared Reflectance Spectroscopy (NIRS). Each of the 20 GPR farms was budgeted for up to 28 fresh grass samples, resulting in a total of 557 samples analysed during the season.

As the GPR-farmers had more insights in grass quality, they were able to adjust their cow rations more easily throughout the grazing season, e.g., by increasing maize silage or by reducing soy bean meal. This advantage was not available for the other KE-farmers. To evaluate whether using data-driven grassland tools (including Grassland Production Registration and the related apps) supports farmers in making dietary adjustments to meet CP targets, a comparative analysis was conducted between GPR- and other KE-participants (comparable in soil type or geographical distribution), using statistical analyses (Kruskal-Wallis) in R-studio based on the ANCA. Additionally, the predictability and accuracy of the data-driven grassland tools were assessed using SAS 9.4.

Results and discussion

Between week 14 and week 46, a total of 557 fresh grass samples were collected. The lowest CP concentration ($74 \text{ g (kg DM)}^{-1}$) was measured in week 31, while the highest ($338 \text{ g (kg DM)}^{-1}$) was measured only 5 weeks later in week 36, though on a different farm. The average CP concentration across all samples was $206 \text{ g (kg DM)}^{-1}$. As illustrated in Figure 1, there was considerable variation in CP concentrations across farms, with a general trend that CP increased slightly during the year.

To assess whether the GPR approach supports achieving the target CP in the ration, a comparison was made between the two groups of farmers, based on available ANCA. Unfortunately, ANCA was

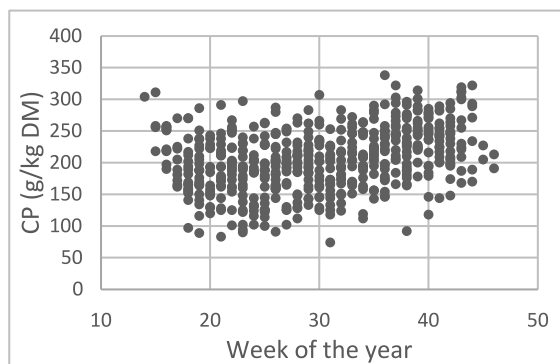


Figure 1. CP-concentrations (g (kg DM)^{-1}) in fresh grass across farms.

Table 1. Characteristics of the Grass Production Registration (GPR) and the reference group (KE).

	GPR	KE	<i>P</i> value
Number of farms	18	108	
Cows per farm (average)	145	142	ns
Percentage of fresh grass in ration ¹	15	10	0.030
Percentage of maize silage in the ration ¹	14	18	ns
Hours grazing (hours total)	1272	830	ns
Intake fresh grass (kg DM cow ⁻¹ year ⁻¹)	1178	963	0.030
CP fresh grass (g (kg DM) ⁻¹)	191	193	ns
CP grass silage (g (kg DM) ⁻¹)	164	168	ns
CP maize silage (g (kg DM) ⁻¹)	67	68	ns
CP in concentrates (g (kg DM) ⁻¹)	184	195	ns
CP byproducts (g (kg DM) ⁻¹)	109	140	0.040
CP in the ration 2024	157	157	ns

¹ DM-basis; % of grass silage (37%), concentrates (28%), and byproducts (5/6%) were comparable across GPR and KE.

not available for all farms, so this analysis includes 18 GPR farms and 108 other KE-farms (as shown in Table 1). Table 1 indicates that the GPR-group had, on average, a higher use of the (dietary) fresh grass. Although a higher proportion of fresh grass in the ration is generally expected to make it more challenging to meet protein targets, both groups achieved comparable CP in their rations. The GPR farmers compensated the higher grass intake by reducing CP in concentrates, and mainly in byproducts (Table 1).

Nine of the 18 farmers achieved the target CP of 155 g CP (kg DM)⁻¹ of the ration. The analysed CP concentrations (g (kg DM fresh grass)⁻¹) were also compared with the CP predicted by the models in Grass Signal. The prediction of CP based on the day of the year had a strong correlation ($R^2 = 0.70$) for the first cut. The correlation between prediction and actual analyses declined in the later cuts ($R^2 = 0.14$).

Conclusion

By using GPR to monitor grass quality, dairy farmers were able to maintain the same average dietary CP despite incorporating a higher proportion of fresh grass in their rations compared to the reference group, by using concentrates and byproducts with lower CP concentrations. While the use of grassland apps demonstrated potential to support grassland management, their predictive accuracy decreased for later cuts. Nevertheless, these tools enhanced farmers' awareness of fresh grass quality and the impact of their management adjustments. Weekly fresh grass sampling contributed to a better understanding of grass quality. According to the participating farmers and advisors, this aspect of the project was seen as beneficial.

Acknowledgement

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The effect of the level of fresh grass on the dietary crude protein level; obstacle or opportunity?

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Abstract

In the 'Koe en Eiwit' (Cow and Protein) project, 150 Dutch dairy farmers are aiming to reduce the level of dietary crude protein (CP) to 155 g per kg of dry matter (DM). One common challenge within the project is that grass-based dairy farms seem to find it more difficult to reduce the level of crude protein in their rations than farms with a smaller proportion of fresh grass. Analysis of 145 Annual Nutrient Cycle Assessment (ANCA) datasets showed that there was no significant difference in crude protein levels between the whole ration and fresh grass. However, differences in the composition of the ration were observed. Grass-based farms can provide valuable insights into strategies for reducing CP levels. On average, farms with a high proportion (22%) of fresh grass in the total ration have a lower CP level in grass silages and a lower proportion of concentrates than farms with a low proportion (2%) of fresh grass.

Keywords: crude protein, dairy farming, fresh grass

Introduction

In 2024, The Netherlands had 13 900 dairy farms (CBS, 2025). Collectively, these farms managed 1.15 million ha of grassland and fodder crops. In 2023, 83% of this area was grassland (Agrimatie, 2025). Therefore, grassland management is one of the most important factors in optimising the Dutch dairy sector's feed rations. Within the 'Koe en Eiwit' project, the average ration contains 48% fresh grass and/or grass silage. As dairy farms are grass-based and weather conditions in the Netherlands are highly variable, it is often said that reducing the crude protein (CP) level, and thus reducing ammonia emissions, with a grass-based ration is very difficult. Reasons given include: (1) the grass already contains a lot of crude protein; (2) the quality of the fresh grass varies greatly; and (3) this leaves only a few options for adjustment to optimise the ration. A previous study on the effect of grass production strategies (Wind et al., 2023) showed that awareness of how grassland management can reduce CP content could be increased. For example, more attention could be given to fertilising and mowing strategies. These strategies, along with the feeding strategy for concentrates, would support the goal of reducing the CP content in feed to $155 \text{ g (kg DM)}^{-1}$ on Dutch dairy farms with grassland. This study therefore focuses on whether there will be an effect in 2024 on the amount of fresh grass in the total ration and the total dietary crude protein content. Additionally, the potential challenges, obstacles, and opportunities of feeding relatively large amounts of fresh grass to achieve lower protein levels in the total ration were examined.

Materials and methods

The Annual Nutrient Cycle Assessment (ANCA) is a tool dairy farms can use to calculate and visualise the cycle and losses of nitrogen, phosphorus and carbon. The tool provides various key figures about crop production, fertilizing, feeding, milk production and environmental performances. These key

figures are based on a large number of calculation rules. These calculation rules are reviewed and further developed annually on the basis of the latest scientific insights. As part of the “Koe en Eiwit” project, participating dairy farms ($n = 150$) submit their ANCA annually. For 2024, we received 145 ANCA to use for monitoring and in-depth research. For the analysis to be performed, the 145 ANCA were divided equally into four representative groups based on the proportion of fresh grass in the total ration. Based on these four groups, statistical significance tests (ANOVA and Kruskal–Wallis) were performed using R-studio ($P < 0.050$ declared as significantly different).

Results and discussion

The four groups of dairy farms are divided based on the proportion of fresh grass in the total feed ration. On average, this is 2, 8, 12 and 22%, respectively. Table 1 shows these groups based on a number of farm characteristics in relation to fresh grass. The analysis shows that, on average, the more intensive farms (farms with a high number of cows and higher number of cows per hectare of grassland) feed the least amount of fresh grass, but this is not statistically significant. The opposite applies to the other characteristics: the higher the proportion of fresh grass, the higher the production of protein grown on-farm, hours of grazing and intake fresh grass. No significant difference was found between the proportion of fresh grass in the ration and the CP level.

The amount of fresh grass, with high level of CP, has to be compensated for in the ration (leaving aside individual farm management effects). Table 2 shows an analysis of the five main feed components. The amount of grass silage has no influence on the percentage of fresh grass in the total ration. However, the lower the amount of fresh grass, the significantly higher amount of corn silage, concentrates and co-products in the total ration.

Table 1. Description of the four different groups based on the average percentage of fresh grass in the total ration.

	Average percentage of fresh grass in total ration				<i>P</i> value
	2%	8%	12%	22%	
Number of cows per hectare	2.12	2.03	1.77	1.63	
Number of farms	36	37	36	36	
Number of cows per farm	163	162	117	122	ns
Hectares of grassland on-farm	57	66	56	69	0.026
Protein grown on-farm (%)	49	59	65	70	<0.001
Hours grazing (hours total)	296	783	954	1560	<0.001
Intake fresh grass (kg DM cow ⁻¹ year ⁻¹)	203	643	1042	1765	<0.001
CP level 2024	157	155	157	156	ns

Including a representation of four characteristics relating to fresh grass in which the groups differ from each other.

Table 2. Overview of the average percentage per feed component of the 4 groups of farms based on the percentage of fresh grass in the total ration

	Average percentage of fresh grass in total ration				<i>P</i> -value
	2%	8%	12%	22%	
Amount of grass silage (%)	35	37	39	36	ns
Amount of corn silage (%)	24	21	16	10	<0.001
Amount of concentrates (%)	30	27	26	26	0.010
Amount of co-products (%)	7	6	5	5	0.040

Table 3. Overview of the average CP-level per feed component of the 4 groups of farms based on the percentage of fresh grass in the total ration

	Average percentage of fresh grass in total ration				<i>P</i> value
	2%	8%	12%	22%	
CP level of fresh grass (g (kg DM) ⁻¹)	198	193	187	190	0.043
CP level of grass silage (g (kg DM) ⁻¹)	172	168	167	160	0.001
CP level of corn silage (g (kg DM) ⁻¹)	68	67	68	68	ns
CP level of concentrates (g (kg DM) ⁻¹)	218	203	190	167	<0.001
CP level of co-products (g (kg DM) ⁻¹)	140	137	153	114	ns

Considering we are focusing on the CP level of the total ration, Table 3 shows the CP level in g (kg DM)⁻¹ of the diet components. The CP level in grass silage is significant lower in more grass-based systems. The CP level of fresh grass is derived from the CP level in the grass silage within the ANCA tool. This means if protein in grass silage is lower the CP level in fresh grass will also be calculated lower. But we see no significant differences of CP in fresh grass, due to correction that is made based on factors such as grazing hours and summer feeding. Although the value of fresh grass is determined using models, purchased products such as concentrated feed and co-products are calculated on the basis of actual values. It is therefore remarkable that the CP level in concentrated feed is significantly lower in the group of farms that have the highest proportion of fresh grass in the ration.

Conclusion

As there is no direct effect of the proportion of fresh grass on the CP level, we can learn from the 2024 dataset from dairy farmers who are able to respond effectively to rations with unpredictable crops. This is an opportunity. These farmers are able to compensate for the high level of protein in fresh grass with lower protein in grass silages and concentrates. There will undoubtedly be obstacles at the individual farm level when it comes to feeding large amounts of fresh grass, but there are also opportunities to learn a great deal from the truly grass-based farms.

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Forage yield and quality of grassland grasses and legumes in the Mediterranean dehesa

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Abstract

The selection of forage species is key to productivity and sustainability in the Mediterranean dehesa. This study evaluated, over three seasons (2021–2024), the dry matter yield (DM), crude protein (CP, g kg⁻¹), and relative forage value (RFV) of four perennial grasses and six annual forage legumes under extensive conditions and without additional reseeding at the Valdesequera experimental farm (Badajoz, south-western Iberia). Highly significant differences were observed among species in DM yield, with *Trifolium subterraneum* ssp. *subterraneum* being the most productive, followed by *Festuca arundinacea*, *Lolium rigidum*, and *Ornithopus compressus* (both with and without testa). *Biserrula pelecinus*, *Dactylis glomerata*, *Phalaris tuberosa* and *Trifolium glomeratum* showed the highest CP values, with no significant differences among them. *Phalaris tuberosa* and *Dactylis glomerata* stood out for their RFV. The species×season interaction was significant for CP and RFV, reflecting sensitivity to climatic variability. Functional persistence was demonstrated by the maintenance of production during the years without reseeding. These results support the relevance of combining adapted grass and legume species to improve the stability and resilience of Mediterranean dehesa pastures under climate change scenarios.

Keywords: annual legumes, biodiversity, forage persistence, Mediterranean pastures, perennial grasses, resilience

Introduction

The improvement of productivity and sustainability in extensive livestock systems of the Mediterranean dehesa depends on the selection of pasture species adapted to poor soils and the pronounced climatic variability of the region (Lüscher *et al.*, 2014). Functional mixtures of annual legumes and perennial grasses enable increased forage yield, stabilise quality, and promote long-term resilience (Shackleton *et al.*, 2024). The multifunctionality of these pastoral systems underscores the importance of integrating animal production, biodiversity, and ecosystem services in the management of the dehesa (Tindale *et al.*, 2023). In this context, the present study evaluates the yield and forage quality of four perennial grasses and six annual legumes over three consecutive seasons under real Mediterranean dehesa conditions (Galea-Gragera & Llera, 2024), with the aim of identifying the species best suited to improve the productivity, quality and resilience of these systems.

Materials and methods

The experiment was conducted over three seasons (2021–2024) at the Valdesequera experimental farm (Badajoz, south-western Iberia). Four perennial grasses (*Dactylis glomerata* (DG), *Phalaris tuberosa* (PT), *Festuca arundinacea* (FA), *Lolium rigidum* (LR)) and six annual legumes (*Trifolium subterraneum* (TS), *T. glomeratum* (TG), *T. cherleri* (TCh), *Ornithopus compressus* without testa (OCS), *O. compressus* with testa (OCC), *Biserrula pelecinus* (BP), *Medicago polymorpha* (MP)) were evaluated, selected from

the official active collection of the Annual Pasture Legume Germplasm Bank of CICYTEX (Galea-Gragera & Llera, 2025). A randomised complete block design was used, sown in 2021 and assessed during 2022–2024, without reseeding. No additional nitrogen fertiliser was applied, and grazing was controlled throughout the trial (Galea-Gragera & Llera, 2024). Dry matter yield (DM, Mg ha⁻¹) was determined by cutting and drying at 65°C to constant weight. Crude protein content (CP, g kg⁻¹) was analysed using the Dumas method (ISO 16634-1:2008) with a LECO analyser (LECO Corporation, 1998). Relative forage value (RFV) was determined according to Van Soest et al. (1991) and Moore and Undersander (2002). Statistical analyses were conducted using ANOVA, and differences between means were evaluated using the LSD test ($p < 0.05$), with R software (v. 4.4.3).

Results and discussion

Highly significant differences ($p < 0.00001$) were observed among the ten evaluated pasture species in dry matter (DM) yield. *Trifolium subterraneum* showed the highest production (40.3 Mg ha⁻¹), significantly exceeding all other species. It was followed by *F. arundinacea* (30.9 Mg ha⁻¹), *L. rigidum* (26.2 Mg ha⁻¹) and *O. compressus* (with and without testa; 26.0–24.4 Mg ha⁻¹), which did not differ significantly from each other. *Dactylis glomerata* (20.4 Mg ha⁻¹) and *P. tuberosa* (15.7 Mg ha⁻¹) showed intermediate yields, whereas *M. polymorpha* had the lowest production (13.6 Mg ha⁻¹). Regarding forage quality, *B. pelecinus*, *D. glomerata*, *P. tuberosa* and *T. glomeratum* had the highest crude protein (CP) contents, with no significant differences among them. According to the classification proposed by Llera et al. (2022) (Figure 1), all grasses fell within the second and third quality categories for both CP and relative forage value (RFV), with DG and PT notable in the second category. Among the legumes, none reached the highest quality category, although BP and TG were placed in the second category for CP and the third for RFV. Both variants of *O. compressus* presented the lowest RFV values across the three seasons.

The species×season interaction was not significant for DM but was significant for CP ($p < 0.01$) and RFV ($p < 0.00001$), reflecting sensitivity to the annual climatic variability typical of Mediterranean dehesa conditions (Lüscher et al., 2014; Galea-Gragera et al., 2024). Sustained production over three seasons without reseeding suggests functional persistence of the selected species a practical advantage for sustainability and cost control in extensive systems. Overall, the results support the use of adapted

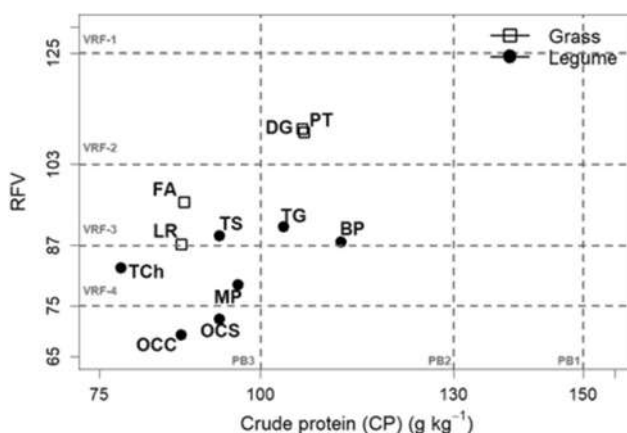


Figure 1. Classification of the evaluated pasture species according to their CP (g kg⁻¹) and RFV, following the proposal of Llera et al. (2022).

grass–legume combinations to enhance productivity, quality and resilience under variable climatic scenarios (Shackleton *et al.*, 2024; Tindale *et al.*, 2023).

Conclusion

The results demonstrate that several grass and legume species showed high productive performance and forage quality under Mediterranean dehesa conditions. *Trifolium subterraneum* stood out for its dry matter yield, while *D. glomerata* and *P. tuberosa* achieved the highest values for forage quality parameters (CP and RFV). The persistence of production over three consecutive seasons without reseeding suggests that these species are well adapted to the local conditions, supporting their use in sustainable, low-input pasture systems. Although no mixtures were tested, the complementary performance observed among species indicates that the combination of adapted grasses and legumes could enhance the stability and resilience of Mediterranean grasslands under increasing climatic variability.

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Multispecies grassland leys enhance energy yields for sustainable livestock systems

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Abstract

Multispecies grassland leys are widely recognized for their ecosystem service provision, including nitrogen fixation, improved soil health, and resilience to climatic stress. Their potential to deliver higher biomass yields has also been well documented. However, a persistent preconception has been that these benefits come at the cost of trade-offs in the optimal harvest time for each species in the mixture in terms of forage quality, thus resulting in a lower forage digestibility overall. Furthermore, the inclusion of herbs in productive grasslands has so far been underexplored. In this study, conducted across 22 sites in contrasting environments (LegacyNet network), we tested monocultures and species mixtures of grasses, legumes, and herbs (with up to 6 species in each mix) under low nitrogen input. Forage quality was assessed using near-infrared reflectance spectroscopy (NIRS). Multispecies mixtures maintained digestibility levels comparable to monocultures and, due to their significantly higher biomass production, had substantially increased metabolizable energy yields per hectare than monocultures. These findings highlight that multispecies mixtures can enhance energy yield without compromising forage quality, reinforcing the potential of multispecies mixtures to support more sustainable livestock production.

Keywords: forage quality, functional groups, herbs, LegacyNet, multi-site experiment, multispecies swards

Introduction

Forage quality is of utmost importance for livestock production, particularly from grasslands, as grasslands account for approximately 78% of global animal feed production area (Mottet *et al.*, 2017). High-quality forage enhances animal performance, reduces reliance on concentrates, and lowers environmental impacts such as methane emissions (Beauchemin *et al.*, 2022), thereby enhancing circularity and improving environmental performance.

Multispecies grasslands have demonstrated potential to reduce the requirements for external inputs (e.g. fertilizer), while improving ecosystem services like weed suppression through mechanisms such as resource-use complementarity (Suter *et al.*, 2021). While legumes are known to enhance crude protein content and digestibility, the broader effects of functional diversity - particularly the inclusion of herbs such as *Plantago lanceolata* L. and *Cichorium intybus* L. on forage quality remains underexplored. Quality of green forages is shaped by complex interactions among species composition, phenology, and environmental conditions. In multispecies swards, the differing phenological stages of individual species mean that a single harvest date inevitably compromises some species' quality, as it reflects an average optimal timing rather than the species-specific optimum achievable in monocultures. Furthermore, these dynamics vary across regions, underscoring the need for multi-site studies to generate robust, generalizable insights. This study compiles data from a coordinated multi-site experiment to assess how increasing species richness—from monocultures to six-species mixtures—affects the nutritional quality of green forage across 22 sites in diverse environments. We investigate how multispecies mixtures influence ME content and total energy yield per hectare, relative to monocultures, under low inputs of N fertilizer.

Materials and methods

This study uses data from the LegacyNet experiment (O'Malley *et al.*, 2023), in which each site established monocultures and mixtures of up to six species (two grasses, two legumes, two herbs; at most sites these were *Lolium perenne* L., *Phleum pratense* L., *Trifolium pratense* L., *Trifolium repens* L., *Cichorium intybus* L. and *Plantago lanceolata* L.) in a completely randomized design. All treatments generally received low to intermediate (mean: 108 kg N ha⁻¹) nitrogen fertilizer input, but unlimited phosphorus (P) and potassium (K) fertilizers. The number of harvests per year varied across sites according to local practice, but was uniform across treatments at any site. Forage quality was analysed in a single laboratory using NIRS (FOSS NIRSystems 5000), calibrated with standard chemical analyses across all species, as well as their mixtures. Energy content was calculated as the annual mean weighted by DM yield across harvests (based on 18 sites with complete harvest quality data), while energy yield was determined by multiplying biomass yield by energy content at each harvest and summing across the year (based on 22 sites with complete biomass data; at four sites, interpolated energy content was used due to partial harvest quality data, which was acceptable given the dominant influence of biomass yield). Plot level contents and yields were modelled as random coefficients linear mixed models using the Diversity-Interactions modelling frameworks (Kirwan *et al.*, 2009). All statistical analyses are preliminary.

Results and discussion

Metabolizable energy (ME) content in the dry matter of monocultures ranged from 10.2 MJ ME kg⁻¹ (SE = 0.1) in *P. lanceolata*, to 10.8 MJ ME kg⁻¹ (SE = 0.2) in *T. repens* (Fig. 1A). The six-species mixture reached 10.4 MJ ME kg⁻¹ (SE = 0.2). For annual energy yield, monocultures across all sites averaged between 85.7 GJ ME ha⁻¹ (SE = 5.4) in *P. lanceolata* to 119.6 GJ ME ha⁻¹ (SE = 7.6) in *T. pratense*, while the average of the six species mixture yielded 121.6 GJ ME ha⁻¹ (SE = 5.0, Fig. 1B), exceeding all monocultures except *T. pratense*.

This study demonstrates that multispecies mixtures can outperform or match monocultures in total ME yield per hectare across a large gradient of environmental conditions. However, this advantage is primarily driven by biomass overyielding due to niche complementarity, asynchronous growth, and efficient resource use among functional groups and species (Suter *et al.*, 2021). The absence of

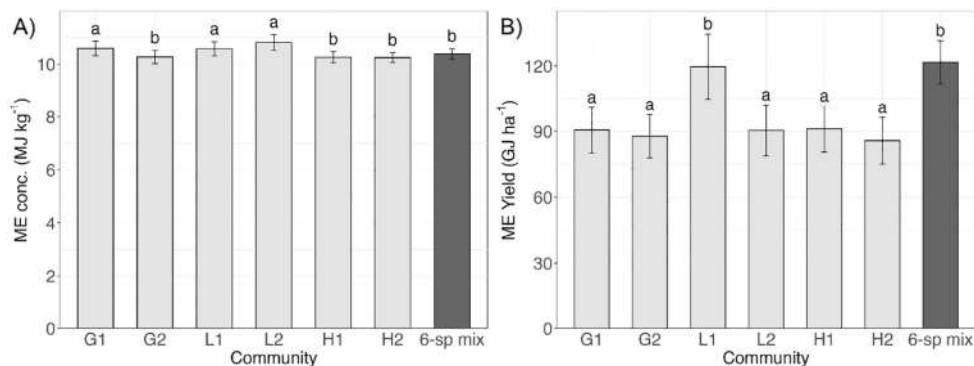


Figure 1. Metabolizable energy (ME) content in MJ per kg dry matter (A) and ME yields in GJ per hectare (B) of monocultures from mostly *Lolium perenne* (G1), *Phleum pratense* (G2), *Trifolium pratense* (L1), *Trifolium repens* (L2), *Cichorium intybus* (H1) and *Plantago lanceolata* (H2), as well as their respective equi-proportional six-species mixtures (6-sp mix). Error bars represent 95% confidence intervals.

synergistic effects on ME content suggests that combining functional groups primarily results in higher biomass yields, while maintaining forage ME content. Interestingly, ME content was consistent across all species in monocultures, suggesting that the harvest time was generally well-chosen—balancing slight quality losses in fast-growing species with sufficient development time for slower-growing ones. Importantly, the wide range of ecosystem services provided by multispecies mixtures—including soil health, nitrogen fixation, weed suppression, and drought resilience—offer benefits that extend beyond forage quality (Finn *et al.*, 2024).

Conclusion

Multispecies mixtures significantly increased total forage energy yields across the multi-site experiment, due to biomass overyielding, without reducing forage energy content. These findings demonstrate their potential to enhance forage-based livestock systems across diverse soil types and environmental conditions, while also delivering other ecosystem services—thus offering a multifunctional alternative to monocultures for forage production.

Acknowledgement

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Opportunities identified by dairy farmers for using autonomously operating robots to harvest and feed fresh grass

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Abstract

A nearly autonomously operating robot is available which, in one working process, can harvest fresh grass, fertilize the paddock, transport the grass, and unload the feed into the feed trough. Ten Dutch farmers were asked about the benefits, drawbacks, implementation and required functions for farm management of such a robot. Farm size varied from 35 to 230 ha and stocking rate at the grazing platform varied from 2.7 to 10.0 cows·ha⁻¹. In general farmers considered this robot a tool that could optimise both grass production and utilisation. They identified opportunities to extend the grazing season in early spring and late autumn by harvesting fresh grass with the robot when circumstances do not suit grazing cows. Grazing infrastructure was mentioned as a key factor for operating the robot. Combining this robot and grazing animals at the same grazing platform is feasible, but the size of the platform and the demand for fresh grass determine the options. To balance the diet, the robot should estimate the nutritional value of the harvested fresh grass.

Keywords: autonomous, automotive, fresh grass, grazing, robot, zero-grazing

Introduction

A nearly autonomously operating robot is available which, in one working process, can harvest fresh grass, fertilize the paddock, transport the grass, and unload the feed into the feed trough. In the Netherlands the first robots operate 24/7 at commercial dairy farms executing zero-grazing. In the Netherlands zero-grazing is executed in different farm setups. Fresh grass, for example, can be offered through either zero grazing alone or in combination with grazing. Farm setup can have implications for the requirements of the robot. The aim of this research was to identify opportunities for such a robot and to clarify the interaction of these opportunities with the farm setup in which robot should be able to operate. In this paper a robot for harvesting and feeding fresh grass is referred to simply as 'the robot.'

Materials and methods

Ten Dutch dairy farmers applying zero-grazing, were asked to reflect on the opportunities of the robot in their on-farm management.

Farmers were asked about required functions, benefits, drawbacks and implementation of the robot. Responses were scored by purpose and topic. Five farmers apply zero-grazing by operating a robot harvesting and feeding fresh grass on their farm. The farms were located all over The Netherlands and vary in farm set up (Table 1). At two farms only zero-grazing was applied. At the other farms grazing was also applied, either as rotational grazing (RG) or compartmented continuous grazing (CCG) (Holshof *et al.*, 2018). Grasslands accessible for either grazing or zero-grazing with the robot varied from 30 up to 200 ha, whereas stocking rate on this area varied from 2.7 up to 10.0 cows ha⁻¹. Zero-grazing by robot was executed in a system comparable to rotational grazing.

Table 1. Farm characteristics from farms executing zero-grazing with or without robot

Farm	Farm size (ha)	Grassland (ha)	Grazing platform (ha)	Cows ha ⁻¹	Cows (ha grazing platform) ⁻¹	Grazing system
A-1	93	75	60	1.9	3.0	—
A-2	230	200	200	1.7	2.0	CCG
A-3	165	95	45	1.3	4.7	CCG
A-4	75	35	35	2.1	4.5	—
A-5	50	50	40	3.4	4.3	CCG
T-1	140	70	110	2.9	3.6	CCG
T-2	64	35	30	1.4	3.0	CCG
T-3	160	135	30	1.4	7.7	RG
T-4	82	69	52	1.7	2.7	CCG
T-5	35	35	35	3.7	3.7	CCG

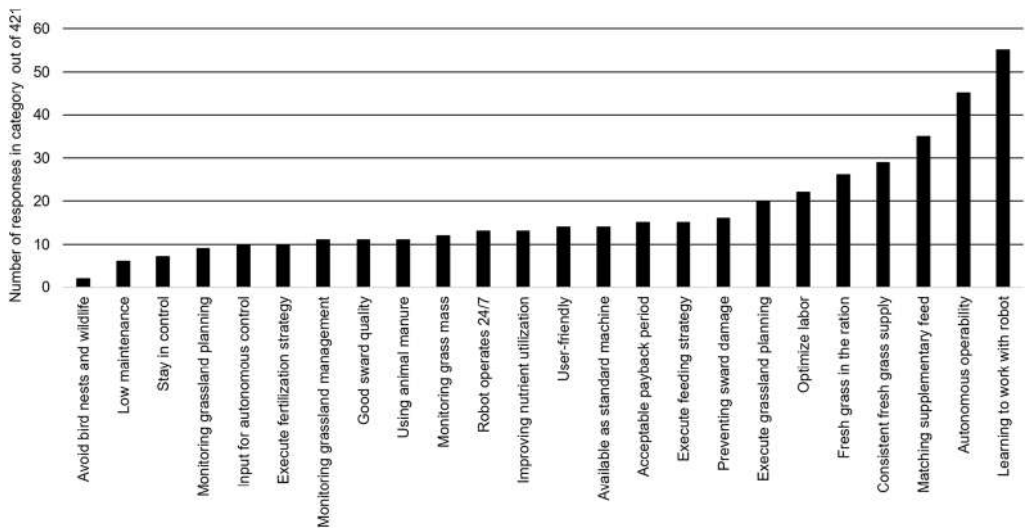


Figure 1. Number of responses of identified purposes, with identified purposes given on the x-axis.

Results and discussion

‘Autonomous operability’ and ‘Learning to work with robot’, were considered the most crucial responses for operating the robot; these two combined accounted for 24% of the answers (Figure 1).

‘Grazing infrastructure’ and ‘Automotive’ were seen as prerequisites for autonomous operability (Figure 2, left wheel). ‘Grazing infrastructure’, farm layout and position and quality of farm roads, is indicated for not only driving the robot, but also preventing sward damage and create optimal routing for harvesting fresh grass. More flexibility in routing and robust operating of the robot are both considered to be realised with the use of more camera and sensor data and the development of interoperability with other robots and activities on the farm. Farmers with a small grazing platform indicate that it is necessary that the robot be allowed to drive on public roads to increase the surface where the robot can harvest fresh grass. With respect to ‘Learning to work with the robot’ (Figure 2, right wheel) farmers identified that operating the robot on the farm requires another way of thinking and working. In general farmers considered the robot a tool to optimise both grass production and utilisation. Opportunities to extend the grazing season in early spring and late autumn by harvesting fresh grass with the robot when circumstances do not suit grazing cows were identified. Combining the robot and

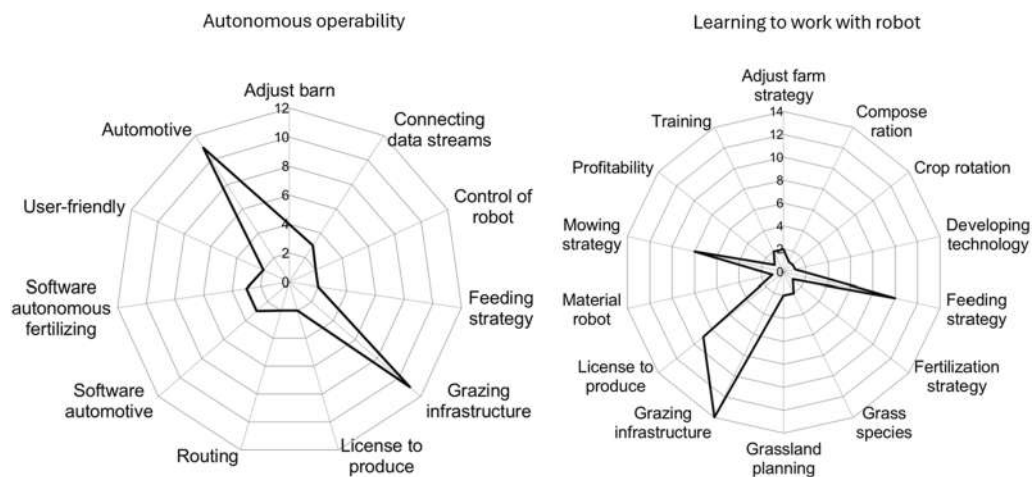


Figure 2. Topics related to autonomous operability (left) and learning to work with robot (right).

grazing animals at the same grazing platform is feasible but the size of the platform and the demand for fresh grass determine the options. Grazing infrastructure, feeding strategy and mowing strategy must be adjusted. Daily tasks change. It has to be learned to trust data and the robot. Software with respect to grassland planning, feeding strategy and fertilizing strategy has to be developed to control the robot to execute these strategies autonomously. Having the robot on the farm to feed cows fresh grass forces the farmers to focus on grassland management and to match fresh grass with additional feeding, resulting in reduction of emissions and improving nutrient utilization. To balance the diet, the robot should estimate the nutritional value of the harvested fresh grass.

Conclusion

Farmers considered the robot a tool to enable the grazing season to be extended. To optimize the use of the robot, its techniques and functionalities, as well as best practices for farm layout and management have to be further developed. By using the robot, the choice is implicitly made to include a minimum amount of fresh grass in the diet daily, which forces the farmer to focus on the grassland to ensure that this grass is available. When zero-grazing with the robot and grazing with animals have to be executed at the same grazing platform, this is considered a challenge, especially when the size of the grazing platform is limited. Farmers indicate that software has to be developed so that the robot can execute grassland management autonomously without farmers having to indicate where the robot has to harvest the required amount of fresh grass from, or how much fertilizer to apply.

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Including forbs in multi-species leys did not affect soil organic carbon and nitrogen stocks

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Abstract

Inclusion of forbs into grassland mixtures is expected to have multiple benefits in animal nutrition, especially if those forbs are rich in secondary plant metabolites. Increased functional group diversity and the presence of deep-rooting forb species, in particular, may also increase soil carbon sequestration in temporary grasslands. In April 2020, we established four temporary grassland mixtures in Frick, Switzerland: grasses, grasses with legumes, grasses with plants rich in essential oils, and grasses with tannin-rich plants. Each mixture was replicated in four randomised plots that were grazed by dairy cows. In March 2020 and February 2024, soil cylinders were taken in the 0–10, 10–20 and 30–50 cm soil layers. Bulk density, soil organic carbon (SOC) and total soil nitrogen (STN) concentrations were measured for stock calculation. Across mixtures, SOC and STN stocks increased from 2020 to 2024 in the 0–10-cm layer as a consequence of increased soil density. Mixture affected neither SOC nor STN stocks or concentrations, but had a minor effect on bulk density in the 10–20-cm layer. The results show that an increase in functional group diversity may not always affect grassland carbon sequestration.

Keywords: multi-species swards, soil organic carbon, temporary grassland

Introduction

Including forbs into temporary grassland mixtures can provide multiple benefits in animal nutrition, especially if those forbs are rich in secondary plant metabolites (Cooledge *et al.*, 2022). Increased functional group diversity and the presence of deep-rooting forb species may also increase other ecosystem functions of temporary grasslands, like sequestration of soil organic carbon (SOC). While root mass and carbon input into soils have been shown to be greater in temporary grasslands containing herbs compared to grass and grass-legume mixtures, their effect on SOC sequestration has seldom been studied (Cooledge *et al.*, 2022).

Accordingly, we assessed the change of SOC and soil total nitrogen (STN) stocks of four temporary grassland mixtures over four years, expecting greater increases in SOC and STN stocks in the two mixtures containing forbs than in the grass and grass-legume mixtures.

Materials and methods

In April 2020 we established four temporary grassland mixtures in Frick, Switzerland, on an organically managed arable field which had previously been part of an arable-ley rotation, and that was ploughed prior to seeding. All seed mixtures contained the grass species *Poa pratensis*, *Lolium perenne*, *Festuca rubra* and *F. arundinacea*. The grass mixture (G) contained only these species; the grass-legume mixture (L) additionally *Trifolium pratense*, *T. repens* and *Medicago sativa*; the tannin-rich mixture (T) *Lotus corniculatus*, *Sanguisorba minor*, *Cichorium intybus* and *Plantago lanceolata*; the essential-oil mixture

(EO) *Achillea millefolia*, *Artemisia absinthium*, *Carum carvi*, *Origanum vulgare* and *Thymus pulegioides*. Each mixture was replicated in four plots, with two subplots per plot, from which biomass was sampled following two different sampling schemes: immediately before stocking, or additionally in the middle of the resting period. The whole area was grazed by dairy cows with an average stocking density of 18 livestock units per hectare and five stocking periods per year. In September 2021, all mixtures received 150 kg ha⁻¹ total nitrogen in the form of slurry. Mixture G was additionally fertilised in spring 2022 and 2023 with 80 kg ha⁻¹ y⁻¹ nitrogen in the form of commercial fertiliser with 12% organic nitrogen concentration.

In March 2020, before ploughing, and in February 2024, soil cylinders (98.17 cm³) were taken in the 0–10, 10–20 and 30–50 cm soil layer at two GPS-positioned pseudoreplicates per subplot. Soil drybulk density (BD) was assessed by weighing the cylinders after drying at 40°C immediately after sampling. Total carbon (TC) and STN concentrations of both years were analysed in the same batch in 2024 by dry combustion (vario Max cube, Elementar, 900°C, 80 s, 80 ml O₂ min⁻¹) and a soil aliquot dried at 105°C for dry matter analysis. SOC concentration was calculated by subtracting the inorganic carbon concentration (500°C ignition loss for 5 h and subsequent dry combustion without O₂ at 900°C, 80 s) from TC. SOC and STN stocks were calculated by SOC/STN stock (Mg ha⁻¹) = (SOC/STN (g kg⁻¹)*BD (g cm⁻³)*height of soil layer (cm))/10.

For each soil layer, mixed effects models with block, year, mixture, sampling scheme and their interactions as fixed effects, and subplot nested in plot as random effects, were fitted to model each dependent variable. In each case, a minimum adequate model was identified with the Akaike Information Criterion corrected for small sample size, retaining the block effect.

Results and discussion

In most cases, the minimum adequate model only contained the year effect (Table 1). In all soil layers, SOC and STN concentrations decreased between the years (on average 0.63 g kg⁻¹ SOC in 0–10 cm, 1.82 g kg⁻¹ SOC in 10–20 cm and 0.47 g kg⁻¹ SOC in 30–50 cm; Table 2). The slight decline may have been caused by ploughing of the field before the establishment of the mixtures as well as yearly variations in SOC/STN levels.

The SOC concentration was similar to other Swiss lowland sites with a high soil clay content of around 40% and a crop rotation including grass-clover leys (Gubler et al. 2019). Bulk density increased during the years, most likely due to the lack of tillage operations and the grazing activity of the cows, as commonly observed for grassland leys in arable rotations (Cooledge *et al.*, 2022). The increase of SOC and STN stocks from 2020 to 2024 in the 0–10-cm soil layer hence was due to the soil compaction effect.

Table 1. Significance (*p* values) of the effect of sampling year on soil organic carbon (SOC) and total soil nitrogen (N) stocks and contents as well as soil bulk density.

Soil layer	SOC stocks	N stocks	SOC content	N content	Bulk density
Upper	<0.0001	0.0154	0.0037	<0.0001	<0.0001
Middle	—	<0.0001	<0.0001	<0.0001	<0.0001
Lower	—	0.2885	0.0929	0.1230	0.0073

Dash, year effect not retained in minimum adequate model.

Table 2. Mean values (SD) of soil organic carbon (SOC) and total soil nitrogen (N) stocks and contents as well as soil bulk density in 2020 and 2024, averaged across mixtures.

Soil layer	Year	SOC stocks (Mg ha ⁻¹)	N stocks (Mg ha ⁻¹)	SOC content (g kg ⁻¹)	N content (g kg ⁻¹)	Bulk density (g cm ⁻³)
0–10 cm	2020	36.46 (2.91)	3.72 (0.28)	29.2 (2.2)	2.98 (0.21)	1.25 (0.058)
	2024	38.75 (1.94)	3.80 (0.17)	28.6 (1.9)	2.80 (0.16)	1.36 (0.052)
10–20 cm	2020	36.15 (3.15)	3.69 (0.28)	28.7 (2.4)	2.93 (0.22)	1.26 (0.090)
	2024	35.53 (2.49)	3.54 (0.20)	26.9 (2.1)	2.68 (0.16)	1.32 (0.047)
30–50 cm	2020	38.27 (4.93)	4.05 (0.43)	14.4 (1.8)	1.53 (0.21)	1.33 (0.069)
	2024	37.47 (4.46)	3.98 (0.56)	13.9 (2.2)	1.48 (0.27)	1.36 (0.065)

The effect of the sward mixtures was only retained in the model predicting bulk density in the 10–20-cm soil layer ($p = 0.068$), with a trend for a higher bulk density in mixture G compared to the other mixtures. The absence of differences in SOC between sward mixtures and the overall decline in SOC and STN concentrations after four years show that there is no clear effect of sward mixture composition on SOC stocks. This is confirmed by the contrasting results obtained in the few studies investigating the development of SOC stocks under temporary grasslands differing in species composition. For example, 4-species mixtures had higher topsoil SOC concentrations compared to 16-species mixtures at nine years after establishment of an experimental grassland on former arable land (Mellado-Vázquez *et al.*, 2016). By contrast, Cooledge *et al.* (2024) found increased soil organic matter concentrations after two years under both a grass-clover and a herbal ley, yet without differences between the two sward types. While many studies have found an increase in root mass and carbon inputs into soil in complex mixtures compared to simple mixtures (e.g. McNally *et al.*, 2015; Mortensen *et al.*, 2025), these effects may not necessarily translate into SOC sequestration, at least not during the time span that temporary grasslands usually occupy in a crop rotation. Differences in rooting depth might, however, have contributed to the trend for greater soil bulk density of mixture G at 10–20 cm depth compared to the other mixtures.

Conclusions

While including forbs into grass and grass-clover leys has previously been shown to increase root biomass, this study demonstrates that it does not necessarily affect SOC stocks over a period of four years, a typical duration for grassland leys. However, the effect of multi-species leys on SOC stocks should also be assessed cumulatively over several crop rotations.

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Application for real-time yield estimation of grassland at a national scale

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Abstract

In the SatGrass research project, a machine learning model was developed using satellite and weather data to estimate dry matter yields on meadows. For the model calibration and validation, ground-truth data were collected bi-weekly at 184 permanent grassland fields throughout Austria between 2018 and 2022. With an R^2 of 0.8 and a mean absolute error of 330 kg dry matter per hectare, a model performance was achieved that is appropriate for implementation as an operational management tool for farmers and national yield monitoring. A backend system with links to satellite and weather data repositories was developed, providing a database of calculated yields for a web-based user interface. Farmers can request the yield on their meadows in real-time with an extrapolation into the near future based on weather forecasts. This approach enables farmers to utilise the tool for planning optimal cut dates based on their expected yield. By aggregating field-related yield data to higher spatial units, it is also possible to monitor yield development regionally and thus create a basis for consulting, statistics, agricultural policy and risk management. Average yields calculated over long periods support the comparative analysis of current developments with long-term trends.

Keywords: dry matter yield model, Earth observation, grassland monitoring, Sentinel-2

Introduction

The transfer of knowledge from research to practice is a crucial, often final stage of many scientific projects as sustainability action. It involves tailoring findings for stakeholders, demonstrating practical examples, or integrating them into observation networks (living labs) that expand the knowledge-base and enhance practical work. In the SatGrass project, knowledge transfer extends to developing an operational management tool that will be presented in this communication.

When research projects result in models that can serve as knowledge-based tools, operational implementation is essential. While calibration and validation—often relying on intensive in-situ data collection and data processing (Klingler *et al.*, 2025)—are standard in research, practical application demands a technical infrastructure for data management, process workflows, and considering user interface and scenarios. Such implementation typically falls outside the scope of research and requires a dedicated follow-up project, often aimed at precision farming (Kushwaha *et al.*, 2024). Operationalizing scientific outcomes is challenging: models must handle exceptional cases, process data efficiently, and ensure high performance, security, and availability.

Materials and methods

By integrating grassland-relevant data, the application presented here provides an information-driven basis to optimize grassland management and to enable monitoring activities. In particular, models combining satellite and weather data determine the growing season by identifying the start of season

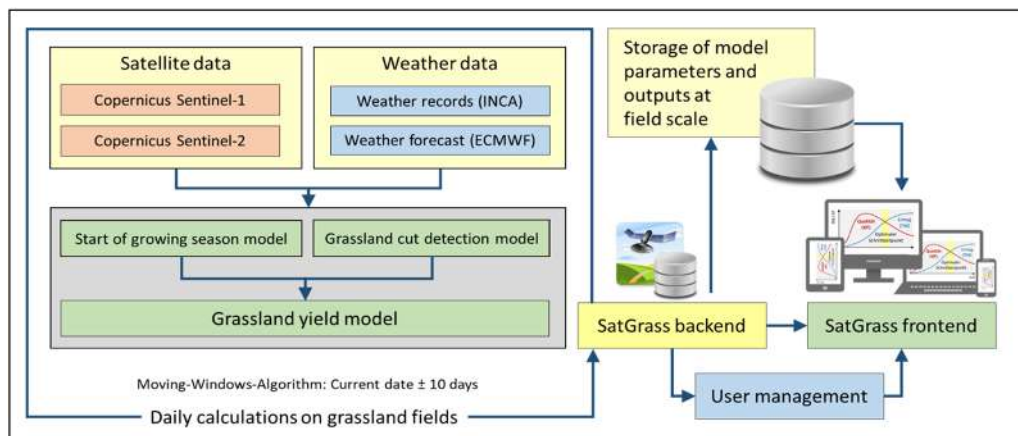


Figure 1. Overview of models, data and their flows in the SatGrass application.

(Dujakovic *et al.*, 2024a) and detecting cutting times (Watzig *et al.*, 2023, Dujakovic *et al.*, 2024b). Within growth periods, satellite and weather data are aggregated and analysed using a neural network (Klingler *et al.*, 2025) to estimate the current grassland dry matter yields, based on an extensive ground truth database of 12,000 yield measurements across Austria (Klingler *et al.*, 2022). Extrapolations into the near future with weather forecasts enable short-term yield projections, that offers users timely and data-driven management options.

In the SatGrass backend, satellite and weather data are extracted for grassland plots using precise polygon geometries. The data are used for daily yield calculations in a moving time window around the current day. The models run automatically, storing their results in a database accessed by the frontend web application. Users can view numerical and graphical outputs of these data on multiple platforms (smartphone, tablet, desktop) for their grassland fields (Figure 1).

Data are stored in PostgreSQL with PostGIS extension. Copernicus Sentinel-1 and Sentinel-2 data are accessed and optimized daily in ZARR format, while weather data are integrated via NetCDF. Observed weather data come from an Austrian weather model (INCA) and from ECMWF weather forecasts. Grassland field polygons are obtained from the Integrated Administration and Control System (IACS). Backend algorithms and models, implemented in C# programming language, connect to the frontend via standardized API. The frontend—developed as a Progressive Web App (PWA) with integrated user management—was developed iteratively through stakeholder workshops and mockup-based design drafts.

Developing an operational application like SatGrass demands transdisciplinary collaboration among grassland experts, data modelers, software developers, remote sensing specialists, and user-experience designers. Integrating these diverse perspectives is both essential and the most challenging aspect of the process.

Results and discussion

Dry matter yield estimation in grassland is an active research field, aiming to close the common knowledge gap in actual yields at the field-level (Reinermann *et al.*, 2020; Ali & Kaul, 2025). SatGrass uses Earth Observation (EO) data with high-temporal and spatial-resolution satellite imagery and weather

data for regional, real-time monitoring of grassland yields at a national scale. With SatGrass, farmers can retrieve yield estimates in kg of dry matter per ha for their fields and compare them with multi-year time series, enabling an accurate assessment of the current situation. The mobile app displays daily yield and provides editable cutting times suggested by the cutting model. Additional features, such as annual yield summaries, harvest diaries, grouping of several fields for yield sums, and result sharing with other users, make SatGrass a comprehensive management tool.

Beyond the mobile app, SatGrass also offers a desktop dashboard, enabling farmers detailed analysis and basic GIS tools to manage their fields. The application is also available to the public to access aggregated yield data across aggregated spatial units (e.g., NUTS regions). These datasets support statistical analysis, agricultural consulting, risk management (e.g., insurance), and policy-making. Multi-year time series further allow for the detection and analysis of climate change-driven trends.

Conclusion

The SatGrass application, launched in March 2026, provides farmers with detailed insights into their grassland fields and corresponding yields. With the planned integration of forage quality modelling, SatGrass will enable farmers to determine optimal cutting times by balancing yield and quality. Field-level yield data can be further refined using a grid-based approach with a resolution of up to 10 metres, unlocking numerous precision farming opportunities—such as tailored, extraction-based fertilizer management. Because SatGrass relies solely on globally available data and requires no additional user input, it can be applied far beyond Austria's borders, following regional model calibration.

Acknowledgements

SatGrass was founded by the Austrian Federal Ministry of Agriculture and Forestry, Climate und Environmental Protection, Regions and Water Management and the Austrian Research Promotion Agency. The operational application is supported by the European Space Agency (ESA) and BMLUK and implemented by SpatialFocus GmbH and EOX IT Services GmbH.

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Impacts of white clover inclusion on intensive grazing dairy systems on a wetland soil

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Abstract

The imposition of reductions to chemical nitrogen (N) fertiliser application limits under the European Union Nitrates Directive (91 676 EEC) have constrained the productivity of perennial ryegrass (PRG; *Lolium perenne* L.) only grazing systems and necessitates the inclusion of white clover (WC; *Trifolium repens* L.) within grazing swards. The current study compared the performance of PRG-only and PRGWC swards within farm systems: a PRG system receiving up to 220 kg N ha⁻¹ year⁻¹, and a PRGWC system receiving up to 125 kg N ha⁻¹ year⁻¹. Pasture measurements were collected pre- and post-defoliation, while milk production data were gathered daily and milk composition fortnightly. Over 2 years, PRGWC produced more pasture herbage (+1062 kg DM ha⁻¹, $P < 0.05$) with reduced chemical N inputs (-72 kg N ha⁻¹, $P < 0.001$) compared to PRG system (11 639 kg DM ha⁻¹ and 191 kg N ha⁻¹). Improved pasture herbage quality in PRGWC resulted in increased milk (+194 kg cow⁻¹) and milk fat plus protein (+12 kg cow⁻¹) production relative to PRG. This study demonstrates the potential benefits of WC incorporation within an intensive pasture-based dairy systems on wetland soils.

Keywords: chemical N, pasture-based dairy system, wetland soil, white clover

Introduction

In response to intensifying pressure to comply with European environmental policies, pasture-based dairy production systems in temperate pastoral regions have transformed from traditional PRG monoculture pastures dependent on high inputs of chemical N fertiliser to systems focused on reducing inputs while maintaining productivity. This has led to a refocus on the incorporation of WC within grazed pastures to provide biologically fixed N (BNF) to the sward, thereby reducing the requirement for chemical N (Humphreys *et al.*, 2017). Incorporating WC and reducing the requirement for chemical N fertiliser applied with PRGWC swards can also contribute to further enhance the sustainability and competitiveness of pasture-based ruminant production systems. However, there has been limited research on the suitability of established PRGWC pastures on wetland soils such as alluvial and gley soils. The objective of this study was to determine the impact of WC inclusion on pasture and animal performance within wetland soils in the border region of Ireland.

Materials and methods

A full lactation (February to November) farm system experiment was conducted at Teagasc Ballyhaise Agricultural College Co. Cavan, Ireland, over 2-years (2023 and 2024). The experiment consisted of two sward type farmlets: PRG receiving up to 220 kg N ha⁻¹ year⁻¹ and PRGWC receiving up to 125 kg N ha⁻¹ year⁻¹. Each farmlet was allocated 26 hectares which were blocked for soil type and drainage class, ensuring all soil types were evenly distributed between farmlets. Pre-grazing pasture

mass was determined by harvesting 3 quadrats (0.5 m×0.5 m) evenly spaced along a diagonal of each paddock using a Gardena hand shears (Accu 60, Gardena International, Ulm, Germany). Pre- and post-grazing sward heights were measured prior to and following each grazing event using a rising platometer (Jenquip, Fielding NZ). Sward WC content was determined using the same methodology as described by Egan *et al.* (2018). Experimental animals, consisting of 140 Holstein Friesian and Holstein Friesian x Jersey dairy cows were randomly assigned between the sward type farmlets based on breed, parity, calving date, previous lactation milk yield, genetic potential (Economic Breeding Index), body weight and body condition score. The milking platform stocking rate was 2.7 cows ha⁻¹ for each farmlet during the experiment. Cows were milked twice daily at 0700 and 1530 h. Milk yield was recorded at each milking for individual animals, while milk solids (MS) yield and milk composition (fat, protein and lactose) were obtained fortnightly. Concentrate supplementation was consistent between treatments, averaging 903 kg cow⁻¹ annually. Data were analysed using PROC MIXED in SAS (SAS, v9.4, SAS Institute, Cary, NC, USA) where milk production data include the effect of year ($n = 2$), sward type ($n = 2$), dairy cow breed ($n = 2$) and their interactions, while pasture production data include the effect of year ($n = 2$), sward type ($n = 2$) and their interactions.

Results and discussion

Mean sward WC content was similar (190 g (kg DM)⁻¹ per year) during both years of the study (Figure 1). White clover swards increased ($P < 0.05$) total pasture production, yielding an additional 1062 kg DM ha⁻¹ compared to PRG (Table 1). This was achieved with reductions ($P < 0.001$) in chemical N fertiliser application. Greater silage conservation was also achieved for PRGWC compared to PRG (1319 and 795 kg DM ha⁻¹ of total pasture herbage production, respectively), although both treatments were not self-sufficient for winter feed (55 and 46% self-sufficiency, respectively). Mean sward nutritive value parameters (ash, crude protein (CP) and neutral detergent fibre (NDF)) were improved for PRGWC pastures. In PRGWC swards, mean CP content was increased to 195 g (kg DM)⁻¹ ($P < 0.001$) and NDF content reduced to 387 g kg⁻¹ DM ($P < 0.001$) in comparison with PRG pastures (188 and 404 g (kg DM)⁻¹, respectively). Organic matter digestibility was similar between sward types (812 and 807 g (kg DM)⁻¹ for PRGWC and PRG, respectively). Cumulative milk and MS yield was increased for PRGWC (+194 and +12 kg cow⁻¹ per year, respectively) compared to PRG with a slight reduction in milk fat content and no difference in protein content observed (Table 1).

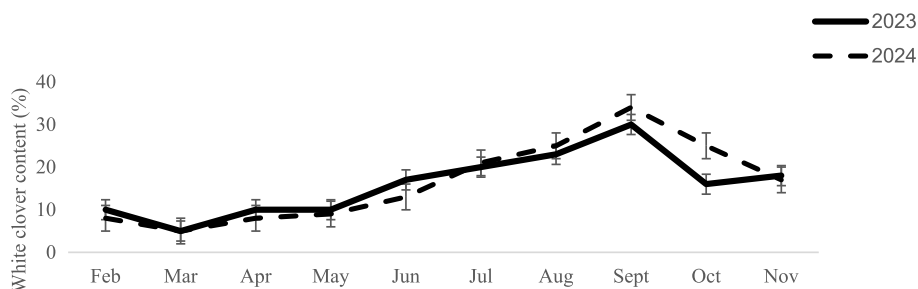


Figure 1. Mean monthly sward white clover (WC) content receiving up to 125 kg N ha⁻¹ annually during the grazing season (February to November) during the two-year experiment.

Table 1. Effect of sward type on pasture-herbage production and animal performance

	PRG	PRGWC	SE	<i>P</i> value
Pasture production (kg DM ha ⁻¹)	11639	12701	365.4	<0.05
Chemical N input (kg ha ⁻¹)	191	119	9.5	<0.001
Milk yield (kg cow ⁻¹)	5024	5218	44.5	<0.01
Fat content (g kg ⁻¹)	54.7	53.9	0.03	<0.05
Protein content (g kg ⁻¹)	37.8	37.7	0.01	NS
Milk solids yield (kg cow ⁻¹)	462	474	4.3	<0.05

No differences in cow body weight or body condition score were observed. The findings of this study highlight the potential of incorporating WC into pasture-based systems on wetland soils. The significant increase in pasture production coupled with a 38% reduction in chemical N application demonstrates the potential of BNF to support pasture productivity within PRGWC swards similar to previous studies (Humphreys *et al.*, 2009; Murray *et al.*, 2024). Moreover, PRGWC pastures produced forage of superior nutritive value which supported increased milk and MS yields. Overall, this study supports the incorporation of WC into pasture-based systems as a viable strategy to reduce nitrogen dependency while maintaining high levels of production on wetland soils.

Conclusion

This study demonstrates that WC incorporation can significantly reduce chemical N applications while improving pasture and animal productivity within an intensive grazing dairy system on wetland soils.

Acknowledgements

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Effect of sward type and nitrogen application rate on sward quality and lamb performance post-weaning

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Abstract

The inclusion of companion forage species, such as herbs and legumes, in perennial ryegrass swards has the potential to improve sward and animal performance in pasture-based systems. This study assessed the effect of sward type and chemical nitrogen (N) application rate on sward quality and lamb performance post-weaning. This experiment had a 2×3 factorial design, consisting of two chemical N application levels, high ($120 \text{ kg N ha}^{-1} \text{ year}^{-1}$) and low ($75 \text{ kg N ha}^{-1} \text{ year}^{-1}$) and three sward treatments, (i) perennial ryegrass- (PRG) only, (ii) PRG+ white clover (*Trifolium repens* L.; WC) and (iii) PRG+WC+Plantain (*Plantago lanceolata* L.; Plan), resulting in six sward treatments. In the post-weaning period, PRG+WC swards had a clover content of 9%, whereas the PRG+WC+Plan sward had a white clover content of 10% and a plantain content of 14%. Sward and lamb performance was monitored for a full production year. In the post-weaning period, a sward type x chemical N application rate interaction was observed, with lambs grazing PRG+WC+Plan with low-N achieving higher average daily gains (ADG) than all the other treatments, and lambs grazing this treatment achieved target slaughter weights earlier than all other treatments.

Keywords: grassland systems, plantain, sheep, white clover

Introduction

Grazed pasture provides the most cost-effective feed for ruminant production systems in temperate regions. Perennial ryegrass forms the foundation of pasture-based sheep production systems in Ireland, characterised by its early season growth and ability to supply high yields of quality forage. However, challenges persist due to the high inputs of chemical-N required for optimum yield and difficulties maintaining sward quality during the post-weaning period. Hennessy (2022) demonstrated that including white clover reduced the requirement for chemical N fertiliser, and Jezequel *et al.* (2024) suggest that diversifying the sward and reducing chemical N inputs can maintain sward yield and nutritive value. Incorporating companion forage species such as white clover and plantain can enhance the quality of perennial ryegrass monocultures post-weaning, positively influencing lamb performance where average daily gains (ADG) are increased, thereby reducing age at slaughter (Woodmartin *et al.*, 2024). White clover can improve sward quality and reduce the fertiliser N requirement through biological fixation of atmospheric N into utilisable forms for the plant. The inclusion of plantain can improve the mineral and trace element content of the sward due to its deep rooting system, with improved N utilization and uptake as a result of the release of bioactive compounds (Nguyen *et al.*, 2022). The objective of this study was to determine the effect of sward type and N application rate on sward quality and lamb performance in the post-weaning period in an intensive pasture-based sheep production system.

Materials and methods

This study was conducted at Teagasc, Athenry, Co. Galway, Ireland for the duration of the 2024 production year as part of a two-year study. A farmlet with a complete randomized 2 x 3 factorial design was used to determine the effect of sward type and chemical N application rate on sward quality and lamb performance post-weaning. Two inorganic fertiliser N application rates, high (120 kg N ha⁻¹ year⁻¹) and low (75 kg N ha⁻¹ year⁻¹), were imposed across three sward types, perennial ryegrass (PRG), perennial ryegrass and white clover (PRG+WC), and perennial ryegrass, white clover and plantain (PRG+WC+Plan), generating six treatment groups for investigation. All farmlets were stocked at 11 ewes ha⁻¹ and managed in a rotational grazing system. Lambs were weaned at an average of 14 weeks of age. A leader follower system was implemented post-weaning, with lambs grazing ahead of the ewes, entering paddocks at a target pre-grazing sward height of 7–9 cm, grazing to a post-grazing height of 6 cm. Ewes entered the paddock immediately after the lambs, grazing to a post-grazing height of 4.5 cm, ensuring optimum conditions for regrowth. The average weaning rate across all treatments was 1.8 lambs/ewe, with an average weaning weight of 28.6 kg. Available herbage was estimated weekly using the rising plate meter. Before each grazing event, two quadrat cuts were taken to a height of 4 cm, one used for dry matter (DM) determination, while the second was separated into species proportions (PRG, WC and Plantain) to determine proportions of companion forage species in each paddock. Lambs were weighed at birth, 6 weeks, 14 weeks, and every 2 weeks thereafter to monitor performance until slaughter, using a portable electronic scale (Pratley, Temuka, New Zealand). Lambs were drafted for slaughter at live weights of 42 kg in June increasing by 1 kg per month until October to achieve a target carcass weight of 20 kg. Lamb performance was analysed using a mixed model in SAS with fixed effects of sward type, litter size, sex and dam parity. Dam was included as a random effect.

Results and discussion

Across treatment groups the average total sward production was 12191 kg DM ha⁻¹ year⁻¹, and the average pre-grazing yield was 1311 kg DM ha⁻¹. In the post-weaning period, PRG + WC swards had a clover content of 9%, while PRG+WC+Plan swards had a clover content of 10% and a plantain content of 14%, giving a total of 24% non-grass companion forages. There was no interaction between sward type and chemical N application rate on lamb performance pre-weaning. McGrane *et al.* (2023) concluded similarly, with sward type having a greater effect in the post-weaning period compared with lamb performance in the pre-weaning period. A sward type×N application rate interaction was observed for lamb post-weaning ADG. Lambs grazing the PRG low-N sward achieved an ADG of +21 g day⁻¹ ($P < 0.05$; compared to the PRG high-N lambs post-weaning). Similarly, lambs grazing the PRG+WC+Plan low-N sward achieved a post-weaning ADG of +35 g day⁻¹ compared to the lambs grazing the PRG+WC+Plan high N sward. There was no difference between lambs grazing the PRG+WC swards at either rate of chemical N. A sward type×N application rate interaction ($P < 0.01$) was observed for lamb age at slaughter, with PRG+WC+Plan lambs achieving target slaughter weights earlier, relative to all other treatments. A sward type×N application rate interaction was observed for lamb lifetime ADG where the PRG+WC+Plan low-N lambs achieved an ADG of 227 g day⁻¹ which was higher than all other treatments. The diversification of pasture systems through the inclusion of herbs and legumes positively influences lamb performance with increased ADG and reduced age at slaughter compared to PRG swards, similar to the conclusions of McGrane *et al.* (2023) and Woodmartin *et al.* (2024). The ability of white clover to support lamb performance at pasture at lower rates of chemical nitrogen application also is similar to that concluded by Hennessy (2022) in dairy systems.

Table 1. The effect of sward type and N application rate on lamb performance

	Sward ¹				N rate			P-value		
	PRG	PRG+WC	PRG+WC+Plan	SEM	High	Low	SEM	Sward	N	Sward×N
Birth weight (kg)	4.6	4.5	4.5	0.09	4.5	4.6	0.08	NS	NS	NS
Pre-weaning ADG ³ (g/day)	246	240	237	4.5	244	238	4.01	NS	NS	NS
Wean weight (kg)	29.2	28.5	28.3	0.47	28.9	28.3	0.42	NS	NS	NS
Post-weaning ADG (g/day)	165 ^a	176 ^a	194 ^b	4.9	168	189	4.3	<0.001	<0.001	<0.05
Lifetime ADG (g/day)	206 ^a	210 ^{ab}	217 ^b	3.9	207	215	3.5	0.06	<0.05	<0.05
Age at slaughter (days)	204 ^a	202 ^{ab}	196 ^b	3.3	203	199	2.9	0.08	NS	<0.01
Slaughter weight (kg)	46.6	47	47	0.3	46.6	47.1	0.25	0.46	0.07	0.43
Carcass weight (kg)	20.7	20.4	20.6	0.14	20.4	20.7	0.12	NS	<0.05	0.06

PRG, perennial ryegrass; PRG+WC, perennial ryegrass+white clover; PRG+WC+Plan, perennial ryegrass+white clover+plantain; N rate, nitrogen rate; ADG, average daily gain

Conclusions

The inclusion of companion forages to the sward, alongside perennial ryegrass offers a potential strategy to improve lamb performance from pasture-based systems at lower rates of chemical N in the post-weaning period.

Acknowledgement

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Impact of nitrogen source, application method, and dose on red clover-grass ley in boreal conditions

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Abstract

Introducing nitrogen-fixing plants like red clover in leys and utilising organic nitrogen (N) fertilisers aims to promote more sustainable fodder production. To increase the cultivation of red clover in northern regions, its cultivation reliability must be improved under hard winter conditions. We investigated the yield response of red clover-grass mixtures to different N fertiliser application methods using the latest commercially available varieties. The field study, conducted in Kuopio, Finland, included three cattle slurry application strategies (injection, trailing hose and solid fraction of slurry) and a control strategy (mineral N) as main plots. Soluble N (sol-N) rate (0, 50 or 100 kg sol-N ha⁻¹ cut⁻¹) was treated as subplots. The trial was on mineral soil with 6.3% organic matter content. Winter damage was observed in spring and dry matter yield and red clover content were measured twice during the growing season. Winter damage was not discovered until the third winter. Two-year results showed that the main plot factor had only a minor effect on dry matter (DM) yield and no effect on red clover content, while the higher sol-N rate increased total DM yield and decreased the red clover content. The yield response differed between cuts.

Keywords: cattle manure, grass, grass-legume, mixture, nitrogen

Introduction

Environmental benefits, along with the national aim to increase N self-sufficiency have increased interest in using N-fixing species in grass leys. In the Nordic region, red clover (*Trifolium pratense*) is the most used perennial legume. However, its cultivation remains at a relatively low level on conventional farms in Finland. One of the main problems limiting wider adoption is the difficulty in predicting yield characteristic as N application rate affects both biomass yield and clover content. Better understanding of N fertiliser application responses with new varieties is therefore needed. This study aimed to increase the persistence of red clover-grass swards and improve N utilisation by optimising the use of cattle manure as a fertiliser.

Materials and methods

A field experiment was established in 2022 in east-central Finland (63°9'N, 27°20'E) on a mineral soil (sandy loam) site with an organic matter content of 6.3%. The ley was established using a seed mixture of timothy (*Phleum pratense* L.) 50%, meadow fescue (*Festuca pratense* Huds.) 25%, and red clover 25%. The experiment followed a split-plot design with four replicates. The main plot factor was four fertiliser application strategies for the 2nd yield: mineral fertiliser only, cattle slurry applied by injection, cattle slurry applied with a trailing hose system and solid fraction of separated cattle slurry. The subplot factor was the rate of soluble N (sol-N) applied per cut: 0, 50 and 100 kg ha⁻¹. All treatments were harvested twice per growing season. The amounts and sources of N applied in each treatment are presented in Figure 1. Phosphorus, potassium, and other nutrients were applied

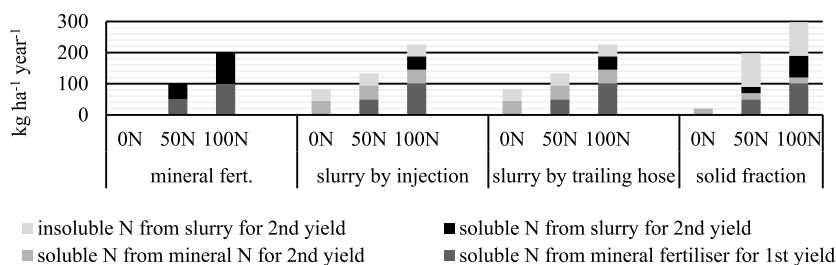


Figure 1. The amounts (kg ha^{-1}) of soluble and insoluble nitrogen (N) from mineral and manure sources in the three soluble N treatments (0 N, 50 N and 100 N kg sol-N ha^{-1}) and in four fertilisation strategies in 2023 and 2024.

uniformly across all treatments according to Finnish fertilisation recommendations to ensure non-limiting conditions for nutrients other than N. At each cut, dry matter (DM) yield and the botanical composition of the harvested DM biomass was determined. Data were analysed using the MIXED procedure of SAS (SAS Institute, Cary, NC, USA), applying analysis of variance (ANOVA). Cuts and years were analysed separately. The main plot, subplot and interaction factor of those was treated as a fixed effect and replicate and replicate $\times$ mainplot as a random effect. Tukey's multiple-comparison test was used for paired comparison and the level of significance was determined at $P < 0.05$.

Results and discussion

The main plot factor, fertiliser application strategy, had only a minor effect on DM yield and no effect on red clover content in both experimental years. In 2023, the drag hose application of slurry resulted in slightly higher average yields compared to mineral fertiliser application alone, while the solid fraction treatment produced slightly lower total yields than the slurry treatments ($P < 0.05$). The difference in total yield between treatments ranged from approximately 400 to 600 kg DM ha^{-1} . No statistically significant differences between fertiliser application strategies were observed in 2024.

In contrast, the rate of sol-N (subplot) significantly influenced total DM yield in both years (Fig. 2). The tested sol-N rates (0, 50, and 100 kg N ha^{-1} per cut) resulted in statistically significant increases in total DM yield at all rates in 2023. In 2024 statistically significant DM yield increase was observed only up to 50 kg sol-N . Corresponding annual yields were 6130, 8090 and 8690 $\text{kg DM ha}^{-1} \text{ year}^{-1}$ in 2023, and 7440, 7950 and 8160 $\text{kg DM ha}^{-1} \text{ year}^{-1}$ in 2024, respectively. The effect of sol-N

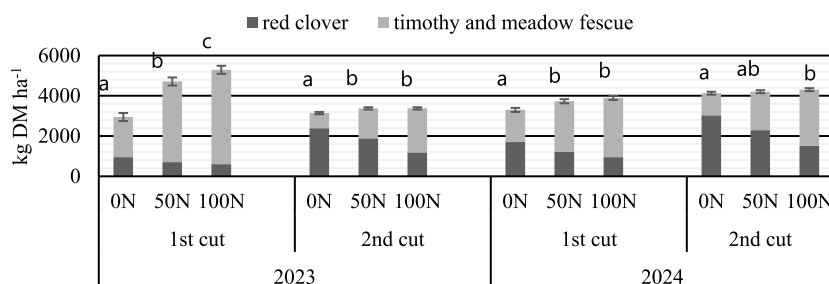


Figure 2. The average dry matter (DM) yields in each soluble nitrogen (N) fertiliser treatments (0, 50 and 100 kg sol-N ha^{-1}) in 1st and 2nd cut for all fertilisation strategies in 2023 and 2024. The bar is divided into different shades according to the plant species composition. Different letters above columns indicate statistical difference ($P < 0.05$) between N treatments on each year and cut. Error bars represent the standard error of mean of total DM yield in each cut.

fertiliser application on total DM yield was primarily driven by yield increases in the first cut. In 2023, the first-cut yield increased significantly at all N application rates due to low clover content (0 N: 32%, 50 N: 15%, 100 N: 11%). In 2024 the clover content (0 N: 52%, 50 N: 33%, 100 N: 24 %) was higher than in 2023, which was reflected in a weaker yield response, that we observed up to the 50 kg sol-N in the first cut. In the second cut, the proportion of red clover in the yield increased noticeably across treatments. As expected, higher sol-N rates reduced clover content in the sward, reflecting the known suppressive effect of mineral N on legume competitiveness. In the second cut, statistically significant but minor yield increases were observed up to the 50 kg N ha⁻¹ (+240 kg DM ha⁻¹) in 2023. In 2024, the second-cut yield was significantly higher only between the 0 and 100 kg N ha⁻¹ treatments (+160 kg DM ha⁻¹). These results suggest that the yield response to N was more consistent and pronounced in the first cut with lower clover content, whereas the second cut showed minor response due to the higher clover content.

It is noteworthy that clover content did not differ between the unfertilised mineral N treatment (true 0 N control) and the 0 N treatments receiving slurry or solid fraction, despite their higher N input for the 2nd yield. The result suggests that the soluble nitrogen in manure was not available during the growth of second harvest due the drought and therefore did not promote grass growth, or that the growth of the first harvest without N fertiliser application determined the clover content and that low N fertiliser application in the second harvest did not change the competitive position of the plants. Also, despite higher total N input in the manure-treated plots compared to mineral fertilised ones, no significant differences in clover content were observed. This indicates that increased insoluble N from organic sources did not suppress clover proportion, while soluble N from any source placed red clover at a competitive disadvantage in the red clover-grass mixture. This finding aligns with previous studies by Kristensen *et al.* (2022) and Nesheim *et al.* (1990).

After the first two winters no winter damage, such as bare spots or patches, was observed. However, winter damage remains one of the primary constraints limiting the broader adoption of clover cultivation under the northern European climate. The third winter (2024–2025) proved severe for the swards, with damage levels ranging between 0 and 40%. Such winter injury may significantly affect the third-year yield and, consequently, the overall productivity of the grassland rotation.

Conclusion

The results showed that the amount of soluble N in fertilisers had a significant effect on both yield level and clover content. In contrast, the insoluble N from manure had no effect on either clover content or yield. The yield response to N fertiliser application clearly declined with increasing clover content in the second cut of the clover-grass mixture, as well as from the first to the second year. Generally, 50 kg of soluble nitrogen in the form of cattle manure or fertilizer was sufficient for fertilizing clover grass. Severe winter damage may decrease sward productivity in the coming years.

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Integration of health services in the forage management on grazing ruminant farms

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Abstract

Integrated animal health is a pillar of agroecology and grasslands represent a lever for agroecological transition in livestock farming. But the place given to animal health in forage management is unexplored. To address this question, we have monitored the forage system management of 8 farms in France. Through two surveys and monitoring during the grazing season, we have qualified grassland allocation in terms of animals, grassland type, timing, and justification. The results show 355 allocations (min 13; max 70) including 268 with one justification and 87 with a combination of two justifications. Seven categories of justifications have been highlighted: using the resource (61% of the justification), preserving grassland potential (16%), making the workload easier (11%), seeking animal performance (6%), supporting behavioural expression (2%), seeking health benefits (3%) and promoting well-being (2%). Five farmers have justified allocation by seeking health services and animal welfare from grassland with variable frequencies (from 1 in 10 to 1 in 80 allocations). We highlight that health services are rarely used to justify grassland allocation. To go further, a yearly analysis, combined with daily grassland use, could allow a better understanding of the integration of health services into forage management.

Keywords: forage system, grazing, health service, ruminants

Introduction

Livestock farming is increasingly challenged for its environmental impacts. Supporting its agroecological transition involves adopting integrated animal health management, a key pillar of agroecology (Dumont *et al.*, 2013; Wezel *et al.*, 2020). Animal feeding is a major entry point for this integration (Lamothe & Savietto, 2017), especially in ruminant systems where grasslands are central. The ecological value of grasslands is well established (Millennium Ecosystem Assessment, 2005), and there is knowledge about the potential health benefits of certain plant species based on the secondary metabolites they contain (antioxidant, anthelmintic) (Poutaraud *et al.*, 2017). However, the role of grasslands in supporting animal health through farmers' use remains underexplored, particularly in forage management. Rose *et al.* (2025) have already identified four profiles based on their knowledge and use of these plant properties for animal health, contrasting with previous work (Petit *et al.*, 2022). We now aim to describe how farmers integrate health services into forage management in terms of grassland allocation.

Material and methods

To better understand the trade-offs that farmers make between the different services expected from grasslands—especially those related to animal health—we monitored the forage system management of eight grass-based ruminant farms in France (Table 1). These farmers, selected from the Rose *et al.* (2025) study, reported using grassland management practices to benefit animal health.

Table 1. Farms location and characteristics

	F1	F8	F9	F13	F17	F26	F28	F30	Mean	SD
Area	IL	IL	IL	O	HS	AURA	AURA	AURA		
Production	suckler cows	dairy cows + cheese	suckler cows	sheep	sheep	suckler cows	dairy cows	dairy cows		
Reproductive female	80	18	40	400	90	60	120	25		
UAA (ha)	130.0	23.0	100.0	92.0	77.0	80.0	176.0	60.0	92.3	45.9
Grassland%UAA (%)	90.0	91.3	70.0	92.4	96.1	100.0	85.2	90.0	89.4	9.0

F, farm; IL, Indre et Loire; O, Ouest; HS, Haute-Saône; AURA, Auvergne Rhône Alpes; UAA, utilised arable area; Grassland%UAA, percentage of grassland in the UAA.

To address this question, we conducted two interviews (February–March and June–July 2024) and relied on grazing season monitoring (March–June 2024) recorded by the farmers themselves. The first interview sought to describe farmers’ seasonal plans for grassland use. In the second, we reviewed their records and discussed the reasons behind each plot allocation. Each allocation was qualified in terms of animal group, type of grassland, timing, and the justifications provided by the farmer. Based on the answers and through a thematic analysis, these justifications were grouped into seven categories: grass resource management, grassland preservation, work organization, animal performance, animal behaviour (to support behavioral expression), animal welfare (to adapt the plot to animal needs), and animal health (to prevent or solve animal health issue).

Given the variability in herd and plot structures, the number of allocations differed across farms. Each allocation was linked to one or two justifications. We then calculated the proportion of each category relative to the total number of justifications per farmer and averaged them across the sample.

Results and discussion

Among the 355 grassland allocations identified across 8 farms (ranging from 13 to 70 per farm), most (268) were based on a single reason, and 87 on a combination of two reasons (Table 2). Most grassland allocations were justified by grass resource management (61%), as farmers prioritized grass growth monitoring and rotational grazing. The second most frequent reason was grassland preservation (16%), often mentioned due to unusually wet conditions. The other explanations were facilitating workload (11%), improving animal performance through diet (6%), supporting animal behaviour (2%), seeking health benefits (3%), and promoting animal welfare (2%). Only five farmers mentioned health or welfare benefits, and their frequency varied widely (from 1 in 10 to 1 in 80 allocations).

Although farmers claim to use grassland diversity to promote animal health (Rose *et al.*, 2025), this is not strongly reflected in their daily grazing management while: (i) there are few farmers who have

Table 2. Percentage of justification given by farmers for each category and category means

Farm	F1	F8	F9	F13	F17	F26	F28	F30	Mean	SD
Nb of grassland allocation	40	69	70	56	13	28	44	35	44.4	19.8
Nb of justification	45	74	90	79	24	30	48	52	55.3	23.6
Justifications										
Resource management (%)	60.0	71.6	70.0	58.2	45.8	50.0	85.4	44.2	60.6	14.3
Grassland preservation (%)	15.6	17.6	24.4	0	0	33.3	10.4	23.1	15.5	11.7
Work organization (%)	15.6	6.8	5.6	19.0	20.8	6.7	0.0	17.3	11.6	7.6
Animal performance (%)	0	0	0	16.5	25.0	0	4.2	1.9	5.9	9.5
Animal behaviour (%)	0	1.4	0	5.1	0	0	0	9.6	2.0	3.5
Animal welfare (%)	4.4	1.4	0	1.3	0	6.7	0	0	1.6	2.5
Animal health (%)	4.4	1.4	0	0	8.3	3.3	0	3.8	2.5	2.9

justified grassland allocation for animal health, behaviour, and welfare, and (ii) when they did it represents few situations in the seasonal forage management. Most of the justifications are related to productive dimensions (use of the resource, grassland longevity, animal performance). This result can be explained by the relative novelty of the concept of integrated animal health management, especially within the analytical framework of forage system management that mainly focuses on the production and consumption flows.

We hypothesize that health-related services from grassland diversity are integrated through a systemic, time-based decision structure operating at both annual and seasonal scales (Sheath and Clark, 1996). Forage system management, defined as the interplay between biophysical conditions and decision-making guiding farm actions (Duru and Hubert, 2003), requires a dual-level analysis combining long-term (annual) planning with short-term (daily) grassland use. In practice, farmers seem to classify grasslands by plant composition, environment, and expected services, then allocate animal groups annually based on their physiological status. Grazing management throughout the season is primarily driven by grass growth dynamics. Therefore, to go further, we propose to analyse how farmers plan their grazing season (first interview) in order to identify the main management rules (Coléno, 2002) regarding grassland type, timing of allocation, and target animal group.

Conclusion

This exploratory study reveals that health-related factors are rarely cited as reasons for grassland allocation in daily farm management. These results encourage a reconsideration and expansion of the forage system concept and the framework within which to analyse it, shifting from a solely production-focused approach to one that also centres on multiple services, including animal health. Achieving this shift calls for analyses carried out across different time scales. Further investigation into this area would enhance our understanding of how health services contribute to grassland management at the farm scale.

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Effect of botanical composition on forage quality prediction using multispectral data

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Abstract

Previous research has shown the potential of multispectral data to predict forage quality parameters of permanent meadows. Usually, models are trained on paired measurements of the forage quality parameter and the corresponding spectral signature, without considering the botanical composition of the samples. This study investigates if the botanical composition, categorised as sward type (rich in grasses, balanced and rich in forbs), depending on the yield proportion of functional groups (grasses, forbs and legumes), affects the performance of models used to predict crude protein, neutral detergent fibre, acid detergent fibre and metabolisable energy content of forage. Partial Least Squares Regression was used to develop predictive models linking forage quality parameters to multispectral data, obtained through resampling hyperspectral measurements of a field spectroradiometer to match Sentinel-2 bands. Three approaches were compared: (1) a global model trained on all sward types and validated on a sample including all types, (2) a global model trained on all sward types but validated on each sward type separately, and (3) separate models trained and validated on specific sward types. Results suggest that type-specific models slightly improve prediction accuracy over a general model, highlighting the role of botanical composition in improving forage quality predictions from multispectral data.

Introduction

Spectral data have shown a high potential in predicting forage quality parameters using various modelling techniques (Wijesingha *et al.*, 2020). In most cases, modelling is performed on a global set of data, not considering the variability in reflectance that could arise from differences in botanical composition of the sward, especially in relation to the functional groups of grasses, forbs and legumes. In this study, we investigate if building sward type-specific models based on values obtained from a field spectroradiometer and resampled to match the thirteen Sentinel-2 bands yields better results in comparison with a global model built mixing all sward types.

Materials and methods

Data collection was performed in 2023 and 2024 at nine experimental sites from two different projects (Peratoner *et al.*, 2020, 2021) in which investigations were made of the effect of grassland management, among others, on forage quality and the botanical composition of permanent meadows with cutting frequencies of two or three cuts per year, in South Tyrol (NE Italy) at altitudes between 725 and 1714 m asl. In order to link forage quality parameters to spectral signatures of the forage, coupled measurements of both were taken. First, hyperspectral data of forage was recorded with a handheld field spectroradiometer (RS-3500, Spectral Evolution, Haverhill, MA, USA) measuring reflectance from 350 to 2500 nm with a resolution of 1 nm from a height of 120 cm. The glass fibre optic had an opening of 25° and was held at 90° from the ground, resulting in a measured area of a circle with a diameter of 53 cm. For each forage sample, three scans were taken in the same position with an

integration time of 40 ms, which were then arithmetically averaged. Measurements were taken under clear sky conditions between 10 am and 2 pm. Then, at the same point the spectral measurements were taken, the forage sample was collected using a metal frame of 0.5×0.5 m and cutting at a height of 4 cm. Before cutting, the simplified botanical composition was determined by visual assessment of the yield proportion of the functional groups of grasses, forbs and legumes (Peratoner & Pötsch, 2019) and categorised as rich in grasses (G, >70% grasses), balanced (B, 50–70% grasses) and rich in forbs (F, <50% grasses, legumes <50%). Forage samples were dried at 60°C until weight constancy. Crude protein (CP), neutral detergent fibre (NDF), and acid detergent fibre (ADF) were analysed by NIRS (NIRSystem 5000, FOSS Hillerød, Denmark) and metabolisable energy (ME) was estimated based on crude ash, CP and ADF (Daccord *et al.*, 2021). The sampling campaign resulted in 481 samples of the following sward types: 122 balanced (B), 124 rich in grasses (G), and 235 rich in forbs (F). To prevent the number of observations from affecting the accuracy of the prediction, we used the same number of observations for each sward type. As the limiting sward type was B with 122 observations, the same numbers of samples were randomly selected from the G and F subsets using stratified sampling based on experimental site and growth cycle, and the selected observations were used for modelling. Three different modelling approaches were compared: (1) global-global model: the model is trained and tested on a mixed sample consisting of equal proportions of the different sward types, (2) global-individual model: the model is trained on a sample consisting of equal proportions of the different sward types and tested on the individual sward types separately, (3) individual-individual model: models were trained and tested on each sward type separately. Before building models, the hyperspectral dataset was resampled to match the wavelengths of the thirteen Sentinel-2 satellite bands using a spectral response function in the *hsdar* package in R. Predictive models for the different quality parameters were built using Partial Least Squares Regression using the thirteen Sentinel-2 band values as independent variables. The sward type-specific datasets (B, G, and F) were always split into a training set (70%, 84 samples) and a testing set (30%, 38 samples) using stratified splitting based on experimental field and growth cycle. For global datasets, training and testing sets contained equal proportions of each sward type. 28 observations from each sward type were randomly sampled from the sward type-specific training set, totalling 84 observations for the global training set. 13 observations from each sward type specific test set were randomly selected and combined in a global test set (39 observations). When building predictive models, the training set was used to determine the optimal number of components for each model. Models with 1 to 10 components were fitted, and a fivefold cross-validation was repeated three times using different fold subsets each time. The number of components yielding the lowest root mean square error (RMSE) was selected. Then, a model was trained on the training set and applied to predict the test set observations. The coefficient of determination R^2 was calculated from the observed and predicted values. The entire procedure was repeated 30 times, with training and testing data randomly reassigned in each iteration, and final performance was computed as the mean R^2 across all iterations. All modelling was done in R software (version 4.3.1) using the *caret* and *pls* packages.

Results and discussion

Overall results indicate that the different forage quality parameters were predicted with varying accuracy in terms of R^2 (Fig. 1). Generally, CP was predicted best, followed by NDF, ADF and ME. The latter was very poorly predicted, showcasing very high standard deviations. There seems to be a slight amelioration for the individual models built on each sward type separately compared to those built on a mixed sample, especially for NDF and ADF. For CP, the performance of the two model types was very similar. Interestingly, there is a big difference in the prediction between the sward types, with G

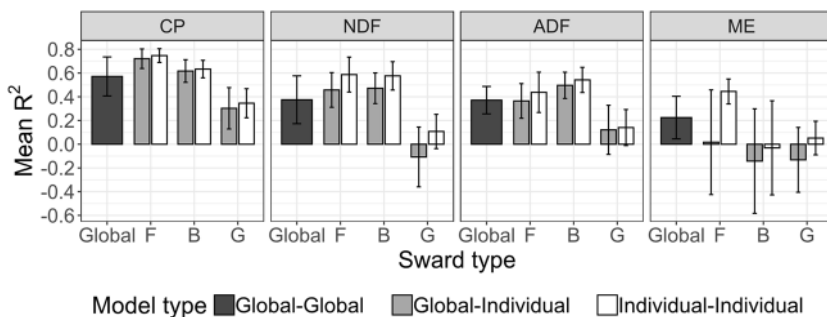


Figure 1. R^2 of predictive models for forage quality parameters built using different modelling scenarios: (a) training and testing on a global dataset including all sward types in equal proportions, (b) training on a global dataset and testing on the individual sward types, (c) training and predicting on individual sward types. F, rich in forbs (grasses <50%, legumes <50%); B, balanced (50–70% grasses), G, rich in grasses (>70% grasses).

being predicted way worse than F and B for all quality parameters. This could be explained by the phenology of grasses, which, especially in later growth stages, display a high stem-to-leaf ratio. Therefore, it could be that either a) spectra are not that efficient in reflecting the quality of stemmy material or b) the longitudinal orientation of stems does not allow the spectra to capture them properly. This lack of accuracy in predicting G is also reflected by the performance of the global-global models. Mean R^2 lies between the performances of individual models for F and B (higher) and for G (lower).

Conclusion

Especially for CP, NDF and ADF, the results showed specific patterns in how predictive performance varied with sward type composition and model typology. However, due to the small sample size and the resulting high standard deviations, these patterns should be validated using a larger dataset.

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Grasslands in Madonie Regional Park: assessment of productivity

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Abstract

Grasslands are well-known for their key role as a source of feed in agro-zootechnical systems. They also provide ecosystem services for environmental stability and conservation of resources. In Europe, grasslands cover more than one third of the agricultural area, but they have been declining for more than 50 years. In the Madonie Regional Park (Sicily, Italy), grassland ecosystem services are progressively affected by the combined effects of agricultural and tourism abandonments and the impact of climate change. In this area, the grazing intensity related to the wild ungulates has produced damage on natural pastures. Based on this, a study was carried out with the aim to monitor the natural pastures at different altitudes in the Madonie Regional Park. Field surveys were conducted to collect biomass samples and determine their dry matter content. The preliminary results show a good level of plant biodiversity and highlight a promising biomass production, despite the modification in floristic composition related to climate change. The application of correct management plans could support the ecological recovery and allow the agro-sylvo-pastoral systems to be sustainable.

Keywords: ecosystem services, grasslands, Madonie Regional Park, phytopastoral characterisation

Introduction

Grasslands cover more than one third of the European agricultural land (FAO, 2006). However, these ecosystems have declined due to land abandonment, reduced management, and the limiting conditions of climate and soil (Queiroz *et al.*, 2014). In this context, the Madonie Mountains (Sicily, Italy) represent one of the most important biodiversity hotspot in the Mediterranean basin (Raimondo *et al.*, 2004). However, this area has undergone a significant decline over the last decade, which has been caused by climate change and an increase in wildlife. This study aims to characterise the productive performance of mountain grasslands within the Madonie Regional Park by assessing forage yield at seven representative sites.

Materials and methods

The study was conducted in the Madonie Regional Park, situated in northern Sicily. It covers an area of approximately 1800 km² and presents various landscapes, including forests, pastures and permanent crops. Seven representative sites were selected to capture the environmental heterogeneity in terms of altitude, exposure and floristic composition. At each site, three replicates were established within the exclusion cages measuring 3 × 5 m were installed. The experimental design extended two consecutive growing seasons (2023–2025). During the first season (2023–2024), one hay cut was carried out in May 2024. The same annual hay cut was carried out again in May 2025 during the second season (2024–2025). Additionally, the experimental protocol was expanded in the second season to include three management regimes corresponding to different phenological stages: early utilisation (G1, November 2024), intermediate utilisation (G2, April 2025), and late utilisation (G3, May 2025).

Data collection was carried out at the time of cutting for each treatment. Measurements included vegetation height, visually estimated percentage cover and above ground biomass. Biomass was manually harvested, at a cutting height of 5 cm above ground surface, dried at 65°C to a constant weight and weighed to determine dry matter (DM) yield. An additional assessment of post-cutting regrowth was carried out at the end of the spring growing season in June 2025 (T2), including morpho-structural measurements. This assessment was carried out on all treated plots, with three replicates at each of the seven sites. A preliminary analysis of the collected data was carried out. Descriptive statistics were calculated for all measured variables, and the results are presented as mean values accompanied by their standard error (SE).

Results and discussion

An analysis of total biomass production (t DM ha^{-1}) was conducted. During the 2024 and 2025 growing seasons, the results revealed variations between the seven areas in the study. In 2024, productivity levels were found to be lower overall, with values ranging from a minimum of 0.7 t DM ha^{-1} in area 4 to a maximum of 3.5 t DM ha^{-1} in area 1 (Fig. 1). In 2025, a great increase in production was observed across most regions, with values reaching up to 5 t DM ha^{-1} . Area 1 was the only exception, whose productivity exhibited a decline in comparison with the previous year (Fig. 1). This overall increase in productivity is consistent with the different precipitation regimes recorded during the two growing seasons. Only 27.78 mm of rainfall was recorded during 2023–2024, whereas 58.86 mm was measured during 2024–2025. This provided more favourable soil moisture conditions for forage growth.

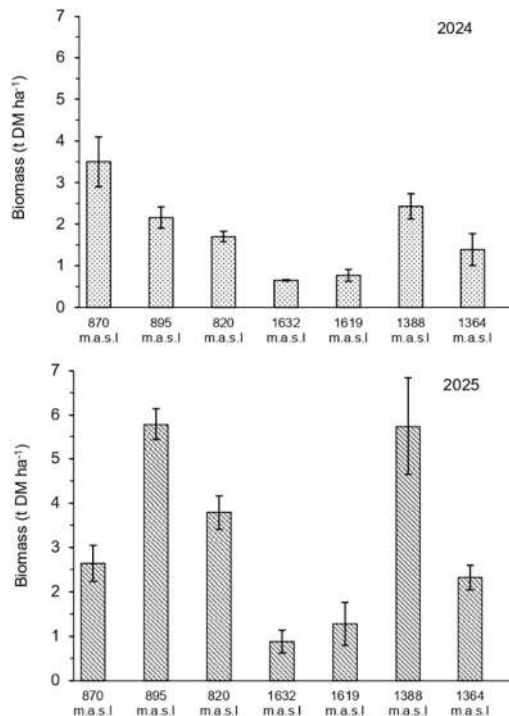


Figure 1. Biomass production was evaluated across an altitudinal gradient and different experimental sites during the 2024 and 2025 growing seasons. The vertical bars represent the standard error of the mean ($\pm$ SE).

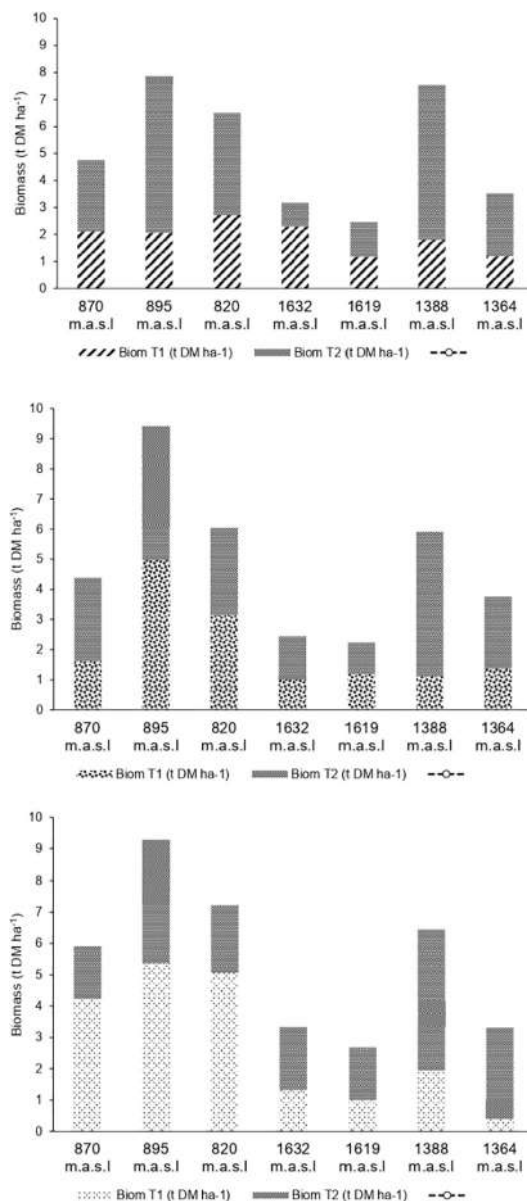


Figure 2. Total forage production (t DM ha⁻¹) obtained at first (T1) and second (T2) cut across study areas (1–7) along an altitudinal gradient (m asl). The three figures correspond to the different management systems: G1 (top figure), G2 (middle figure) and G3 (bottom figure). The altitude values on the x-axis correspond to the sampling sites.

Analysis of the three management strategies reveals that total biomass production (T1+T2) varied between areas and was affected by the type of management strategy. Generally, areas 1, 2, 3 and 6 had the highest productivity in all conditions, whereas areas 4, 5 and 7 showed lower yields, being located at higher altitudes.

Total biomass forage production at the first cut (T1) varied depending on the use and the area of the study. For early use (G1), the first cut yielded low amounts ranging from 1.1 to 2.7 tonnes of dry matter

per hectare. However, this favoured high production at the regrowth cut (T2), particularly in the areas 2, 3 and 6. Middle use (G2), involving mowing in April, increased yields at the first cut (T1), reaching values close to 5 t DM ha⁻¹ in area 2 and exceeding 3 t DM ha⁻¹ in area 3. Conversely, production at the T2 was lower than that observed in G1 management. Late use (G3), involving cutting in May, ensured the highest yield at the first cut (T1) in almost all areas. However, this favoured a reduction in production at the regrowth cut (T2), highlighting a negative impact on the biomass.

Conclusion

The results highlight the influence of the cutting period on the seasonal productivity of pastures. Early and intermediate utilisations are the most balanced in terms of both overall yield and regrowth capacity. Late utilisation determines limited vegetative recovery.

Acknowledgements

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Comparison of the quality of organic and pasture-based raw milk

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Abstract

The EU 'Farm to Fork' Strategy has a target of 25% organic farming by 2030. Organic farms must follow strict standards, including 100% organic, GMO-free feed and limited use of antibiotics, chemical fertilisers, herbicides, and pesticides. While some pasture-based dairy farmers could transition to organic production, many stick with conventional feed due to cost, habit, or profitability. This study compared milk quality from organic and conventional pasture-based farms located in the same area with similar size, livestock, and management. Weekly bulk tank milk samples were collected, along with samples of grazing grass, hay, and concentrate feed. Results show that, under similar feeding and production conditions, organic milk contains more protein, lactose, and non-fat dry solids than milk from conventional pasture-based farms.

Keywords: green deal, organic production, pasture-based milk, sustainability

Introduction

The concept of quality for consumers of animal products has evolved to include new criteria relating to animal ethics and welfare, environmental impact, as well as traditional attributes such as nutritional value, taste, and flavour (Luykx & van Ruth, 2008). In general, milk consumers assume that a grass-based production system is more natural and better for animal welfare than the grain-rich diets, which are typical of intensive systems. Consequently, grass-based feeding system has received greater attention in conventional farming in recent years (Magan *et al.*, 2021). Organic milk production meets these requirements, as well as others, such as ensuring that feed is of organic origin and limiting the use of therapeutic treatments. Both organic and pasture-based systems are managed similarly. For example, in both systems, the animals spend a significant amount of time in pastures, whereas in intensive systems, they are kept confined. Several studies have compared organic milk production with the conventional intensive production system (Manuelian *et al.*, 2022), as well as grass-based and intensive production farms (Abou el qassim *et al.*, 2022). However, to our knowledge, no studies have compared milk production in organic and grass-based systems. The aim of this study was to evaluate the quality of milk produced by organic dairy farms comparing it with that produced by conventional pasture-based farms.

Materials and methods

The study was conducted on three dairy farms located close to each other on the Spanish Atlantic coast in 2023 and 2024. All of the farms had similar climatic, agricultural and management conditions. One farm uses a conventional pasture-based production system, while the other two are certified organic farms. All farms had Friesian cows with similar characteristics in terms of size (34 ± 3.2 lactating cows),

Table 1. Nutritive value of feed provided to the organic and conventional pasture-based farms.

System	Organic	Pasture-based	rsd	P
Grass				
Dry matter (g kg ⁻¹)	176	238	78.7	0.273
Organic matter (g (kg DM) ⁻¹)	867 ^b	910 ^a	26.8	0.042
Crude protein (g (kg DM) ⁻¹)	157	131	23.5	0.136
Net energy (Mcal (kg DM) ⁻¹)	1.43	1.42	0.073	0.935
Grass hay				
Dry matter (g kg ⁻¹)	872	890	34.4	0.584
Organic matter (g (kg DM) ⁻¹)	814	920	25.4	0.808
Crude protein (g (kg DM) ⁻¹)	119	117	64.5	0.974
Net energy (Mcal (kg DM) ⁻¹)	1.31	1.30	0.029	0.650
Feedstuff				
Dry matter (g kg ⁻¹)	886	893	7.9	0.385
Organic matter (g (kg DM) ⁻¹)	933	927	4.2	0.229
Crude protein (g (kg DM) ⁻¹)	175	197	9.5	0.085
Net energy (Mcal (kg DM) ⁻¹)	1.86	1.86	0.031	0.870

DM, dry matter; rsd, residual standard deviation.

stocking rate (1.8 ± 0.35 livestock units ha⁻¹), concentrate consumption (2135 ± 372 kg cow⁻¹ year⁻¹), and yield (6010 ± 875 l cow⁻¹ year⁻¹). The management and feeding practices employed on each farm were similar. The cows were milked twice a day. During the milking sessions, concentrate was supplied according to the lactation stage and production level. After milking, hay was provided *ad libitum* in the barn for two to three hours. Finally, the cows grazed in multi-species pastures for at least 18 hours each day. Samples of feed and tank milk were regularly taken from all farms during the spring. Dry matter intake was estimated using the NRC (2001) theoretical daily dry matter intake equation. The dry matter, ash, crude protein and net energy for lactation were determined using NIRS. Milk samples were analysed by MilkoScan to determine fat, protein, lactose, solids-non-fat, and urea concentrations. Statistical analysis was performed using the R package (R Core Team, 2021). Duncan's test was used to compare systems when significant differences were found.

Results and discussion

Table 1 shows the nutritive value and net energy content of feeds provided. All feeds were similar between production systems except the organic matter of grass in the pasture-based system was higher than the organic system. However, this is not relevant to the overall results. There were no differences in dry matter intake (18.1 ± 0.52 kg day⁻¹) and milk yield (19.7 ± 2.87 kg day⁻¹) between the treatments.

Milk composition is shown in Table 2. There were no differences in milk fat concentration between the treatments. However, organic milk had higher ($P < 0.05$) protein, lactose, and solid-non-fat contents, and a lower ($P < 0.05$) urea concentration than milk from pasture-based cows. Several studies have reported compositional differences between organic and conventional milk (Średnicka-Tober *et al.*, 2016), particularly with regard to the fatty acid profile. However, protein and lactose concentrations are influenced more effect of breed and diet than by organic or conventional systems (Schwendel *et al.*, 2015). The observed differences could then be attributed to the use of organic feed and veterinary treatments. However, other factors, such as genetics or milk yield may also be important. In this study the same breed (Friesian) and similar management were compared.

Table 2. Milk composition according to the organic and conventional pasture-based farms.

	Organic	Pasture-based	rsd	P
Fat (g kg ⁻¹)	36.4	40.6	9.17	0.233
Protein (g kg ⁻¹)	32.9 ^a	32.0 ^b	0.98	0.010
Solid-non-fat (g kg ⁻¹)	87.9 ^a	85.5 ^b	2.06	0.006
Lactose (g kg ⁻¹)	47.2 ^a	46.0 ^b	0.58	0.048
Urea (mg dl ⁻¹)	219 ^b	297 ^a	80.5	0.016

rsd, residual standard deviation. ^{a,b} Different letters in the same row indicate significant differences between treatments.

Conclusion

Differences were found in the protein, lactose and solids-non-fat content of milk from organic sources, as well as a lower urea concentration. However, these differences do not appear to affect milk yield, which was similar in both organic and pasture-based production systems. Therefore, these differences may be due to the type of feed rather than the management system.

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A self-assessment tool for farmers to assess their performance with respect to grazing and agroecology

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Abstract

Despite their best intentions, farmers often do not know whether their way of practising grazing is well balanced to meet the required productive and environmental goals. The EU-funded project Grazing4AgroEcology has therefore developed an integrated self-assessment tool (G4AE-SA tool, including clearly identified indicators) that shows the farm performance within five agroecology pillars relevant for grazing-based farming systems: (1) adopting management practices that aim to improve animal welfare; (2) reducing the inputs required for production; (3) reducing pollution by optimising the biogeochemical functioning of farming systems; (4) enhancing diversity within animal production systems to strengthen their resilience; (5) preserving biodiversity in agro-ecosystems by adapting management practices. In 2024, the G4AE-SA tool was used within a network of 120 Partner Farms across eight countries to assess their current performance. The results point out strengths and weaknesses, assisting farmers and other relevant actors, like advisers, to reflect on the farm's performance and to identify an action plan for further improvement. For the first time, European farmers can objectively assess the overall farm performance on five pillars of agroecology, revealing both the positive impacts of grazing and areas for improvement. This capability will empower farmers to support transition towards sustainable, pasture-based farming systems.

Keywords: animal welfare, biodiversity, diversity, Grazing4AgroEcology, inputs, pollution

Introduction

Practitioners and policymakers increasingly acknowledge grazing as a management strategy capable of supporting food production, animal welfare and the delivery of ecosystem services in parallel (Bengtsson *et al.*, 2019). However, trade-offs between these objectives are seldom made explicit at farm scale. Farmers therefore benefit from practical tools that translate agroecological principles into measurable, farm-specific indicators to guide reflection and change. This paper introduces the Grazing4AgroEcology Self-Assessment Tool (G4AE-SA tool), designed to be pragmatic and farmer-centred: its indicators are grounded in practice, and its outputs are directly linked to tailored recommendations. We present and discuss first results of the G4AE-SA tool from 2024, based on its application on 120 G4AE Partner Farms across eight countries, and explore how these outcomes can inform on-farm optimisation and advisory strategies.

Materials and methods

The G4AE-SA tool assesses five key agroecological pillars relevant to grazing systems. Each pillar includes sub-pillars and measurable farm-level indicators that can be evaluated by farmers, either independently or with adviser support:

- (1) Animal welfare—including farm infrastructure, animal nutrition, and health management practices (20 indicators);
- (2) Reducing inputs—focusing on minimising external feed inputs, optimising nutrient use, maximising herbage utilisation, and improving soil management (35 indicators);
- (3) Reducing pollution—centred on optimising fertilisation, grassland and soil management, pest regulation and waterway protection (20 indicators);
- (4) Enhancing diversity—promoting diversification of production, marketing strategies, and income sources (22 indicators);
- (5) Biodiversity—preserving habitats and maintaining flora and fauna (28 indicators).

Indicators of the tool focus on concrete actions and observable outcomes and are scored on a normalised 0–100% scale. Typical outputs of the tool are performance scores per indicator, sub-pillar and pillar. The tool was implemented on the G4AE Partner Farm Network (PFN) during the first half of 2024, comprising 120 grazing-based livestock farms (both organic and conventional) in eight European countries (France, Germany, Ireland, Italy, the Netherlands, Portugal, Romania, Sweden). Results were stored locally. Each farmer received a language-specific personalised report containing the farm's profile, performance results, and tailored recommendations from Facilitator Agents ensuring that the feedback was directly relevant and actionable. In line with PFN agreements, only aggregated results are publicly disclosed. Finally, a user survey was conducted to capture farmers' and advisers' perceptions of the tool's strengths, weaknesses, and improvement needs, and its potential to support farm optimisation and advisory strategies.

Results and discussion

Figure 1 captures the aggregated status of the PFN in 2024. It shows high mean scores for biodiversity, reduced inputs and enhanced diversity ($\approx 80\%$), intermediate scores for animal welfare ($\approx 60\%$) and lower scores for pollution reduction ($\approx 50\%$). The strong performance in biodiversity and reduced inputs reflects widespread adoption of habitat-friendly practices and input efficiency. High diversity scores indicate active use of varied production strategies. Pollution reduction emerges as the main priority, requiring optimised nutrient cycles, improved monitoring and, in some cases, infrastructure upgrades or revised manure handling. As the PFN is not representative of all farms in the eight G4AE countries, direct comparison between countries was avoided due to contextual differences in farm types, priorities and indicator relevance. Some questions had limited applicability in certain regions, reinforcing the need for within-country interpretation and peer learning rather than cross-country ranking.

The user survey showed that the G4AE-SA tool met the expectations of farmers and FAs, particularly by supporting benchmarking between farms, fostering discussions with peers, and enhancing familiarisation with agroecological concepts. It increased the awareness of the users of specific practices and brought attention to aspects that might otherwise have been overlooked. As the tool was largely oriented towards dairy farming, sheep farmers perceived it as insufficiently tailored to their production. Overall, the G4AE-SA tool supported informed decision-making and targeted optimisation on the Partner Farms. To further increase the impact of the tool, future development should: (i) integrate

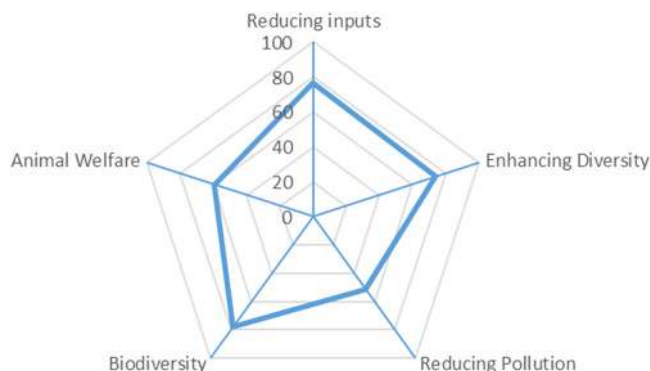


Figure 1. Grazing4AgroEcology Partner Farm Network aggregated pillar scores in 2024 (%).

more grassland functions and ecosystem services; (ii) introduce benchmarking across countries and the option to monitor progress over time; and (iii) connect outputs to advisory packages and financing mechanisms.

Conclusions

Applied to 120 Grazing4AgroEcology Partner Farms across eight countries, the G4AE-SA tool proved most effective as an indicative instrument within a country or peer group, where shared baselines and local advisory support enable realistic improvement pathways. It structures farm information, highlights concrete priorities, and delivers language-specific reports that farmers and advisers can turn into targeted interventions. Results of the tool also foster dialogue among farmers, advisers and value-chain actors. The tool supports farmer-led optimisation towards sustainable, pasture-based systems and informs advisory and policy responses.

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High intraspecies variability in tannin content of big trefoil and salad burnet

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Abstract

When applied in grazing settings, tannin-rich forages can reduce methane emissions from enteric fermentation. Previous studies have shown that tannin-rich forages such as big trefoil (BT; *Lotus pedunculatus*) and salad burnet (SB; *Sanguisorba minor*) are effective at doing so. However, the link between tannin content and their bioactive properties remains unclear. Establishing this link could allow for improvement of these species to be higher yielding and more bioactive as forage species for livestock farmers. We have therefore characterised the polyphenol profile of around 160 individual plants of either species across accessions, analysing tannin concentration, polymer size, mean degree of polymerisation (mDP), and flavanols. BT had a relatively high content of condensed tannins ranging from 5.1 to 28.6 g (kg DM)⁻¹, with an mDP between 9.8 and 17.1, while SB had a high content of hydrolysable tannins ranging from 20.7 to 64.0 g (kg DM)⁻¹, demonstrating great intra- and interspecies variability. Currently a subset of these plants, selected for their variability in tannin concentrations, is screened *in vitro* for their antimethanogenic potential.

Keywords: bioactivity, forages, methane, ruminants, tannins

Introduction

The incorporation of tannin-rich forages in mixed grasslands can promote ruminant health and productivity while simultaneously reducing the environmental footprint of livestock production systems by enhancing N utilisation and lowering enteric methane (CH₄) emissions (Mueller-Harvey, 2006). Tannins effectuate their anti-methanogenic properties by forming complexes with feed components such as proteins and fibre, rendering them inaccessible for methanogenic microbes in the rumen (Bhatta *et al.*, 2013); by sequestering minerals vital for their proliferation (Scharenberg *et al.*, 2007); and by altering enzymatic activity (Adamczyk *et al.*, 2017). Thus, tannins increase the share of post-ruminal digestion rather than fermentation of dietary proteins in the ruminant leading to lower enteric CH₄ production while also promoting N utilisation (Aboagye & Beauchemin, 2019). However, despite well-documented effects of tannins on CH₄ production (Verma *et al.*, 2022), results vary widely, limiting their potential for large-scale implementation. Evidence suggests, however, that bioactivity is linked to tannin structure and content which in turn is closely related to plant species and tissue (Malisch *et al.*, 2015). However, while comparative studies between species are comparatively frequent, very few studies have analysed the intraspecific variability, especially studies that include the important structural characteristics. A homogeneously high tannin concentration would, however, be a prerequisite to obtain predictable bioactive effects in the field. Our study has hence analysed around 160 plants of both BT and SB for their tannin concentration and structure. The two species have previously been shown to significantly reduce CH₄ production *in vitro* which was attributable to their distinctive tannin profiles (Verma *et al.*, 2022).

Materials and methods

A greenhouse trial was conducted using multiple accessions of BT and SB. Two plants were sown per 5-l pot in August 2023. In total, 335 plants were analysed, comprising 176 BT and 159 SB individuals. The BT individuals represented three accessions: LOT29 (80 plants), NSL448192 (54 plants), and NSL448193 (42 plants). The SB individuals represented five accessions: DELAR (38 plants), PI462229 (32 plants), SANG1 (38 plants), SANG10 (34 plants), and SANG6 (17 plants). Plants were harvested simultaneously when the majority of the plants were about to enter flowering stage. Samples were processed for polyphenol extraction and analysis after freeze drying. Using the method explained in Engstrom *et al.* (2014) extracts were screened for their polyphenolic composition and detailed tannin structural features, including prodelphinidin percentage (PD %), galloyl content, and mean degree of polymerisation (mDP) (Engström *et al.*, 2014). Statistical analysis was performed using the software R.

Results and discussion

Both species showed a high variability in tannin content, with BT containing primarily condensed tannins (CT) ranging from 5.1 to 28.6 g (kg DM)⁻¹ and SB containing primarily hydrolysable tannins (HT) ranging from 20.7 to 64.0 g (kg DM)⁻¹ (Figure 1). We observed a large coefficient of variation across individuals of the same accession for total tannin content; however, this variability was much greater in BT (31.6–35.3%) than in SB (12.0–18.6%).

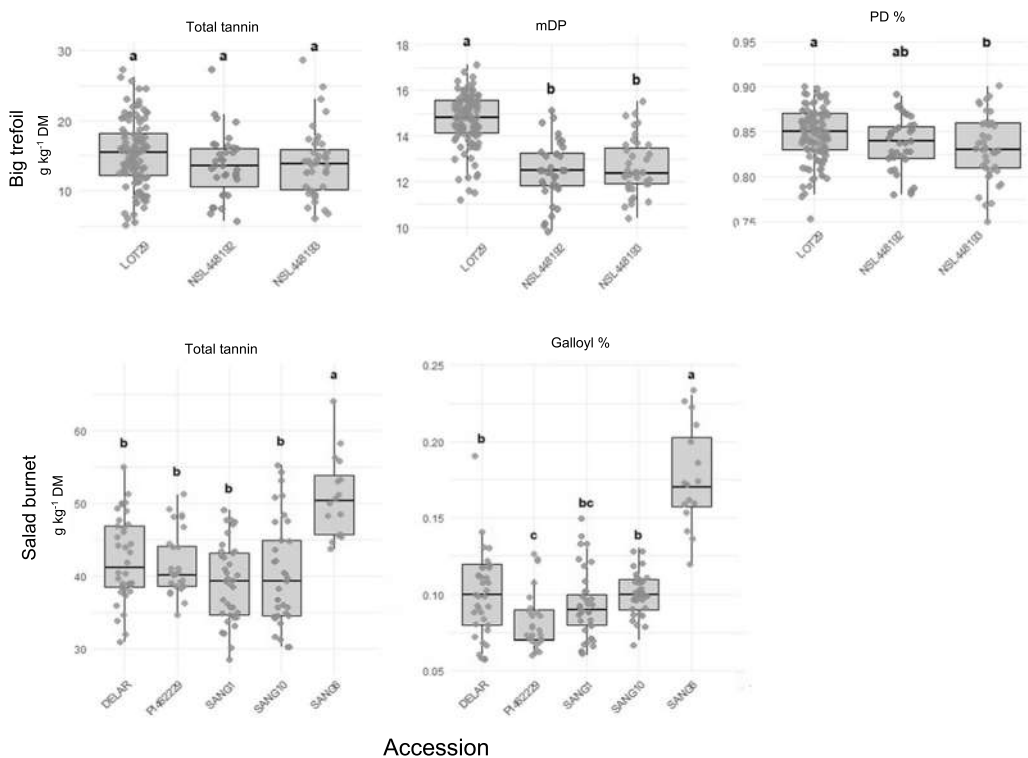


Figure 1. Total tannin content (g (kg DM)⁻¹) and structural composition in different accessions of big trefoil and salad burnet. Different lower-case letters above boxplots indicate statistically different accessions ($P > 0.05$, Tukey test). mDP, mean degree of polymerisation; PD, prodelphinidin.

In the case of BT, LOT29 demonstrated the highest CT content although this was not significant. LOT29 did, however have a significantly higher mDP and PD% (Figure 1). Since greater PD% has been shown to be correlated with greater bioactivity (Huyen *et al.*, 2016), these results thus suggest LOT29 to be a great choice of accession for further breeding. As for SB, SANG6 showed a significantly higher HT content than the other cultivars, while also SANG6 had a significantly higher % of galloyl (Figure 1). Galloyl is a radical of gallic acid, which has shown to significantly reduce methane emissions without compromising animal performance (Aboagye *et al.* 2019), suggesting that the tannin profile of SANG6 may act as an optimal baseline to which breeding for greater bioactivity can be pursued.

Conclusion

In terms of total tannin content, a great variability between individuals of the same accession was observed. However, tannin composition was more distinct amongst accessions, suggesting bioactivity may also differ amongst them. The results from the ongoing *in vitro* antimethanogenic analyses will therefore establish whether this variability will be linked to their bioactivity. The current study thus offers potential for breeding a bioactive forage with stable, and appropriate tannin concentrations that balance digestibility and methane emissions.

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Determining grass harvest time using organic matter digestibility improves economics on dairy farms

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Abstract

Organic matter digestibility (OMD) is a key indicator for grass quality. Although OMD varies among fields and years at the same maturity stage, farmers typically determine their time of harvest based on phenological maturity. The objective of this study was to determine whether this practice is still the most viable based on modelled income over feed cost (IOFC). Hybrid ryegrass (*Lolium × hybridum*, Hausskn.) and permanent grassland were cut weekly from two weeks before to two weeks after heading. Dry matter yields and OMD were measured; potential DM intake and energy-corrected milk yield were calculated for two scenarios: 8 kg concentrate per cow per day or none. IOFC was defined as milk revenue minus the cost of concentrate and forage per cow per day. Harvesting before heading resulted in an IOFC increase of €0.90 to €2.0, compared to harvesting at heading. Increasing yields at later maturity did not offset the decline in OMD and milk output. IOFC for second cuts were €2.0 to €6.0 lower than first cuts. In conclusion, harvest time should be optimized economically, and not solely based on maturity. The findings highlight the potential of precision farming technologies to determine the ideal harvest window.

Keywords: harvest time, IOFC, organic matter digestibility

Introduction

Grasslands cover over 60% of the world's agricultural area, contributing to the primary feedstuffs in ruminant diets (FAO, 2025). Consequently, utilizing grass efficiently is essential. A key indicator for grass quality is OMD (Beecher et al., 2015), positively influencing milk yield, milk efficiency, dry matter intake, and methane yield (Álvarez, 2021; Rinne et al., 2002). The OMD typically peaks at the beginning of the growing season, declining with ongoing maturation of the grass (Álvarez, 2021). In contrast, grass yields increase over time, lowering costs per kg of grass produced. However, OMD varies among fields, farms, and years at the same maturity stage (Nordheim-Viken and Volden, 2009). Yet, harvest time is often based on phenological maturity. This study reviews whether this practice is still economically viable. The objective of this study was to define the ideal time of harvest based on modelled IOFC.

Materials and methods

The study analysed two cuts of hybrid ryegrass and one cut of permanent grassland. Samples were taken between 4 May and 4 July 2023 on fields of the educational and research centre Hofgut Neumühle in Münchweiler an der Alsenz, Rhineland-Palatinate, Germany (49°32'18"N, 7°52'60"E). Grass was harvested weekly over 5 weeks, starting 2 weeks prior to the main harvest and ending 2 weeks after the main harvest, which occurred at the beginning of heading of the grassland or ryegrass. Grass was mowed at 100 mm cutting height, wilted, baled, and weighed to determine the yield. Five samples

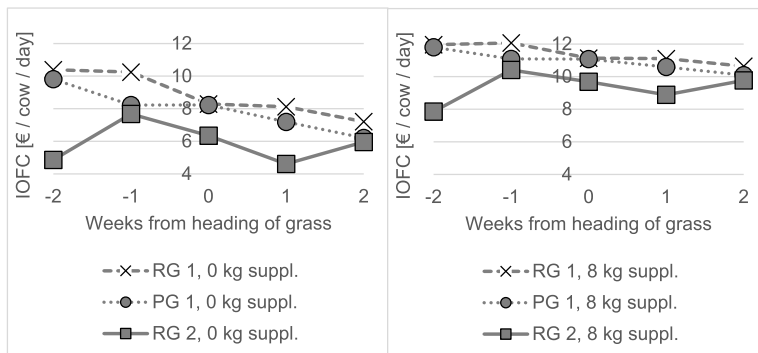


Figure 1. Weekly changes of modelled IOFC (Income over feed cost) per cow per day for ryegrass 1st cut (RG 1) and 2nd cut (RG 2), and permanent grassland 1st cut (PG 1) with (right) or without (left) 8 kg of concentrate supplementation. On the x-axis, 0 determines the time of heading.

per strip were analysed through laboratory near-infrared spectroscopy. Dry matter (DM) yield was calculated using the mean DM content of those samples.

Utilizing OMD measurements of the samples, potential DM intake and energy-corrected milk yields were calculated for two scenarios: no concentrate or 8 kg of concentrate supplementation per cow per day, based on formulas published by Álvarez et al. (2020) and Álvarez (2021). Milk income was calculated using the modelled milk yield and the 2023 average milk price (BLE, 2025). Concentrate costs were set at 0.335 € kg^{-1} , the average price for dairy concentrate feed in 2023 (MLR, 2025). Grass costs were derived from KTBL (2018), adjusted for yield. The IOFC was calculated by subtracting the forage and concentrate costs from the milk income.

Results and discussion

Results varied between ryegrass and permanent grassland, and among different cuts in ryegrass (Figure 1). For the first cut of ryegrass, the ideal timing was a week before heading for the model using 8 kg of concentrate. After that time, there was a decline in IOFC, resulting in a €0.9 lower IOFC at the time of heading. For the model without supplementation, the ideal harvest time would have been earlier, resulting in a €2.1 difference in IOFC compared to harvest at heading. In both cases, increasing yields and associated lower costs per kg of grass could not make up for the decreasing OMD and potential milk output. For the second cut of ryegrass the ideal harvest time was at one week before heading, for both, with or without supplementation. Unlike the first-cut ryegrass, the IOFC increased in the second cut at later harvest stages, where DM yields increased without a remarkable decline in quality. The IOFC in the second cut is about €2.0 lower with concentrate supplementation and between €2.0 and €6.0 lower without any supplementation. This is explained by lower yields and therefore higher costs associated with the second cut. Permanent grassland showed a similar trend to ryegrass with a stronger decline in IOFC. The decrease in OMD was more prominent in permanent grassland, resulting in lower potential milk yield.

Conclusion

The determination of the ideal time of harvest should be considered from an economic point of view. With different OMD values at heading from year to year and field to field, decision making based on maturity stage is not accurate enough. Precision farming technologies can have an economic benefit in finding the right time of harvest based on yield and feed quality.

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Applying an ecosystem approach to the national carbon balance of Sahelian (agro)pastoral systems: insights from the CaSSECS Project

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Abstract

Pastoral and agropastoral livestock systems in the Sahel are often criticised for their high greenhouse gas (GHG) emission intensities, with regards to their comparatively low productivity. The CaSSECS project challenges this perception by adopting a systemic approach to assess the national carbon (C) balance of these ecosystems in Burkina Faso, Niger and Senegal. Combining field measurements of C sequestration and GHG emissions from animals with the GreenFeed system and the FAO's GLEAM-i tool, the study shows that, when contextualized, these ecosystems exhibit a net negative C balance reflecting their potential for C sequestration. The direct measurements of emissions with GreenFeed combined with representative feed rations from the Sahel are 15–24% lower than the estimates generated by the FAO's GLEAM-i tool. Additionally, improved livestock feed strategies can reduce CH₄ emissions yield by up to 35%. Regarding C storage, soil C sequestration potential ranges from 0.15 to 0.25 Mg C ha⁻¹ year⁻¹, depending on soil type and vegetation density. National-level results confirm the existence of a net C sink in all three countries. Therefore, these results argue for a reassessment of the environmental impact of Sahelian livestock systems, support the integration of localised data into national climate strategies, and should help to recognise pastoral systems in climate financing mechanisms. Maintaining soil/tree health is essential to maintain this positive role for the climate.

Keywords: agropastoral systems, carbon balance, climate change, GHG, livestock, Sahel

Introduction

Pastoral and agropastoral livestock farming in the Sahel is often accused of contributing significantly to national GHG emissions and, consequently, of influencing climate change (Balehgn *et al.*, 2021; FAO, 2023). Additionally, Sahelian livestock is criticised for its low feed efficiency, as local breeds produce less meat or milk than Western breeds. However, this perception is shaped by the method used to assess the impact of livestock farming (emissions per litre or per kg produced), which overlooks the multifunctional role of Sahelian livestock within the ecosystem, and is further hindered by a lack of reliable measured data. Therefore, the CaSSECS project proposes an ecosystem approach to pastoral lands which considers the interactions among soils, plants, and animals that drive GHG emissions and carbon sequestration (Assouma *et al.*, 2019a). Additionally, the project has invested in contextualising numerous measures regarding animal emissions and the ecosystem's C storage capacity. In an original way, this study evaluates the C balance at national level from the perspective of identification of ecosystem-based mitigation strategies that strengthen productivity and resilience while reducing the net C footprint.

Materials and methods

To assess the C balance of (agro)pastoral livestock farming in the Sahel (Burkina Faso, Niger and Senegal) the CaSSECS project applies a straightforward equation that calculates the difference between GHG emissions (produced by soil, plants and livestock) and changes in C storage in the soil and plants (Assouma *et al.*, 2019b). If the C balance is negative, this means that the system contributes to storing C and therefore has a positive impact on the climate. If the C balance is positive, the system is an emitter, and therefore has a negative impact on climate.

Regarding the data used to calculate C balance, two complementary approaches were used: (i) collection of *in situ* data on major GHG fluxes and variations in soil and woody biomass carbon stocks; and (ii) application of the FAO's Global Livestock Environmental Assessment Model (GLEAM) to upscale, at the national level, total GHG emissions from pastoral livestock systems in each country. Carbon balance was assessed in relation to the atmosphere, whereby all GHG emissions to the atmosphere were accounted for positively, while all carbon storage was negatively accounted for. In this study, the enteric methane (eCH₄) emissions from ruminants, as the main GHG source, were measured using a GreenFeed® (Gbenou *et al.*, 2024a). Carbon accumulation in wood and woody roots was evaluated by observing tree diameters (2009–2015) and using allometric equations specific to the main species in the pastoral area. At soil level, C accumulation was estimated by balancing incoming (organic matter) and outgoing (gaseous) C fluxes in the soil. This method integrates the use of the entire pastoral area, depending on the seasons and the locations where the herds graze. Scaling methods were applied to estimate a national balance sheet for the three countries considered. Remote sensing was used in particular to scale tree storage data. The results presented below are based on 2022 data. The data used are those from the CaSSECS project activities, and others come from the bibliography.

Results and discussion

The ecosystem-based C balance approach adopted in this study has made it possible to identify slightly negative C balances for (agro)pastoral ecosystems on a national scale (Senegal $-0.63 \text{ Mg CO}_2\text{eq ha}^{-1} \text{ year}^{-1}$, Burkina Faso $-0.37 \text{ Mg CO}_2\text{eq ha}^{-1} \text{ year}^{-1}$, Niger $-0.73 \text{ Mg CO}_2\text{eq ha}^{-1} \text{ year}^{-1}$). The results of the assessment presented are conditioned by the high sensitivity to the methodological choices adopted.

Considering the specific cattle enteric CH₄ emissions measurements produced by CaSSECS, scientists were able to demonstrate a 15 to 24% overestimation of GHG emissions indicated by international references (Table 1).

On average, for cattle in the Sahel, emissions are $46.7 \pm 3.34 \text{ kg of eCH}_4 \text{ per Tropical Livestock Unit per year}$ (Gbenou *et al.* 2024a). The CaSSECS project confirmed that eCH₄ remains the primary source of emissions in the livestock sector. Focusing efforts on measures to mitigate eCH₄ emissions therefore remains the most effective approach. In this sense, the systems implemented within the

Table 1. National GHG emissions

GHG emissions (Mg CO ₂ eq)	Senegal	Burkina Faso	Niger
Cattle (GLEAM-i)	5 361 810	13 943 953	23 357 491
Cattle (CaSSECS project)	4 052 713	11 747 756	19 912 527

project also allowed the testing of local feed rations and supplements. Feed rations can reduce or optimize eCH₄ production in animals (Gbenou *et al.* 2024b). This improvement can lead to up to a 35% reduction in eCH₄ (g kg⁻¹ DMI). In addition, at the national level, herd size has a significant influence on the amount of eCH₄ produced. Consequently, the larger the herd, the greater the emission quantities.

Soil C storage potential varies greatly. This is due in part to the great diversity of soils present in the different Sahelian subsystems. The storage potential measured in the soil as part of the CaSSECS project ranges from 0.15 to 0.25 Mg of CO₂ ha⁻¹ year⁻¹ (Assouma *et al.*, 2019b). This represents a 45% difference in potential of sequestration. Based on measurements by soil and woody compartment, CaSSECS considers a median value of 0.20 Mg of CO₂ ha⁻¹ year⁻¹ across the three countries. In all these scenarios, the ecosystem is acting as a carbon sink.

The number of trees per hectare also affects the ecosystem's C storage and sequestration capacity. The balance presented here assumes an average of 8.5 trees ha⁻¹, or a storage capacity of 0.057 Mg CO₂ ha⁻¹ year⁻¹. If the ecosystem loses 50% of its trees, the storage capacity of woody plants decreases, the balance is impacted, and the ecosystem becomes less sequestering (a decrease of 17%). Therefore, the C balance is highly influenced by the storage potential in soils and plants; these variations can shift the ecosystem from a sequestering one to an emitting one. It is therefore important to invest in preserving the quality of soils and woody plants (fighting against soil degradation and the disappearance of woody plants).

Conclusion

The contextualized data collected as part of the CaSSECS project for (agro)pastoral livestock farming in the Sahel, once aggregated, allow us to calculate a negative C balance, i.e., a C sequestration for the ecosystem. In other words, Sahelian pastoral ecosystems are on track to achieve carbon neutrality, with even a slight potential for carbon sequestration. The results advocate for a change in tone to challenge the negative perspective of (agro)pastoral livestock farming, and rather to use the results to defend it as way of production and a lifestyle. Livestock should no longer be viewed solely through the lens of animal emissions and feed efficiency, but rather in relation to their interaction with the broader ecosystem, including in their potential of C sequestration. Moreover, beyond carbon, livestock also plays a very important role in maintaining food security, cultural heritage and the resilience of rural livelihoods. Data used to calculate this balance sheet have already been integrated into the FAO's calculation systems (GLEAM-i). Furthermore, methods and data obtained to calculate this balance must be integrated into the reference data and production methods of Nationally Determined Contributions (NDCs) at the national and international levels to better reflect the real impact of (agro)pastoral livestock systems on Sahelian ecosystems. Considered with all methodological precautions, this C balance calculated in CaSSECS provides context-specific evidence that can support Sahelian countries to sustain pastoral livestock systems and communities, and to protect their ecosystems. The C balance presented here allows us to better understand our systems and to design mitigation options, which will require detailed work with appropriate policies. The result of this study needs to be confirmed through more detailed analysis of the long-term dynamics of the different components of the ecosystem studied here as each of these countries are characterized by strong inter-annual and ten-yearly variability.

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Assessment of perennial ryegrass variety performance on commercial farms over 10 years

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Abstract

Maximising grass utilisation and milk production are two key metrics in pasture-based dairy systems. Perennial ryegrass (*Lolium perenne* L.; PRG) plays a central role in supplying high-quality forage to achieve this and is one of the most widely used grass species on pasture-based farms in temperate regions. Perennial ryegrass varieties are primarily assessed in controlled, grazed, or mechanically defoliated plot trials. However, since 2013, PRG varieties have also been evaluated on commercial farms in Ireland to investigate their performance under real-world conditions. In the current study, 9 varieties selected from the Pasture Profit Index (PPI)—a variety selection tool used by the Irish seed industry—were sown and analysed across commercial farms over a 10-year period. Variety, sward age and year had a significant impact ($P < 0.01$) on herbage production. The highest-yielding variety produced 1538 kg DM ha⁻¹ more than the lowest-yielding variety over the study period. Swards in year 10 yielded 839 kg DM ha⁻¹ more than those in year 1. Variety also had a significant effect ($P < 0.01$) on the number of grazing events per year. These findings highlight the importance of variety selection to sustain high levels of herbage production on commercial farms.

Introduction

Maximising herbage production increases the profitability of pasture-based dairy farms (Fineran *et al.*, 2012). It is recommended that paddocks be reseeded every 12 years to optimise sward production and utilise genetic gain from PRG varieties (Creighton *et al.*, 2012; Tubritt *et al.*, 2021). When reseeding it is important that high performing varieties are chosen as varietal selection can have a major impact on herbage production. There is little data available from commercial farms on PRG varietal performance over long periods. Previously, Byrne *et al.* (2017) and Hearn *et al.* (2023) reported that on-farm evaluation was effective for identifying the most productive PRG varieties; however, these studies only evaluated varieties over a 3- and 7-year period, respectively. Hearn *et al.* (2023) reported varieties that produced the highest dry matter yields in younger swards also performed best in permanent pasture swards. As such, herbage production during the first 4 years post-sowing was a good indicator of herbage production from years 5–7. However, as reseeding is a costly exercise many farmers do not reseed this frequently, and there is a need for longer term evaluation of PRG varieties on commercial farms.

Materials and methods

This study collected data from 108 pasture-based dairy farms in the Republic of Ireland between 2013 and 2024 inclusive using PastureBase Ireland (Hanrahan *et al.*, 2017). Farmers created a profile of each paddock on the farm including data such as area and soil fertility and, as paddocks were reseeded, farmers entered varieties sown and sowing dates. Reseeding was carried out using best-practice methods (Teagasc, 2014) to ensure sward establishment. Subsequently, farmers recorded weekly or bi-weekly herbage mass measurements for each paddock on PastureBase. A minimum of 30 measurements was

required for a farm to be included in the analysis. Farmers were trained on herbage mass estimation (kg DM ha^{-1}) and used their preferred measurement method: rising plate meter, cut and weigh, or visual estimation (O'Donovan *et al.*, 2002).

Each variety used in the analysis has a minimum of 5 replicates per year. The varieties analysed and associated heading dates and ploidy are: Aberchoice (11 June; D), Abergain (4 June; T), Abermagic (28 May; D), Astonenergy (1 June; T), Drumbo (5 June; D), Dunluce (29 May; T), Glenveagh (1 June; D), Kintyre (6 June; T) and Tyrella (4 June; D). Analysis was carried out in SAS version 9.4 (SAS Institute, Cary, NC, USA) using linear mixed models to estimate the association between variety and annual DM production. The fixed effects in the model were variety, year, sward age (defined as measurement year minus sowing year), and associated interactions.

Results and discussion

Variety had a significant effect ($P < 0.001$) on herbage production. The highest yielding variety produced $+1538 \text{ kg DM ha}^{-1}$ compared to the lowest yielding variety (Figure 1). Herbage yield increased with sward age, as 10-year-old swards yielded $+839 \text{ kg DM ha}^{-1}$ more than 1-year-old swards. There is little data to date on the performance of PRG varieties over a period greater than 7 years. It is assumed that herbage yield declines as swards age; however, this was not the case in the current study. There was no interaction between sward age and variety, similar to Hearn *et al.* (2023), who reported little variation in herbage production as swards age. This highlights the importance of choosing high yielding varieties when reseeding. Year also had a significant effect ($P < 0.001$) on herbage yield as there was a $4107 \text{ kg DM ha}^{-1}$ difference between the highest ($15\,739 \text{ kg DM ha}^{-1}$; 2017) and the lowest ($11\,632 \text{ kg DM ha}^{-1}$; 2024) years. A variety by year interaction was also observed, which may reflect differences in how varieties respond to varying climatic conditions.

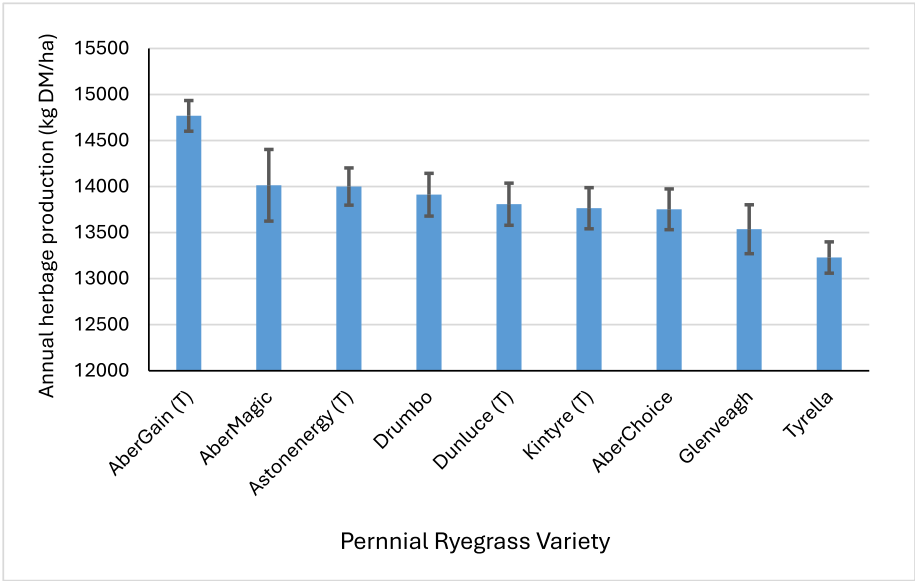


Figure 1. Annual herbage production (kg DM ha^{-1}) of 9 perennial ryegrass varieties over 10 years on Irish commercial farms; error bars represent standard error.

Variety had a significant effect on the number of grazings per year, with an average of 7.3 grazing rotations per year for the 9 varieties. Abergain had the greatest number of grazings (7.7) while Dunluce had the lowest (6.8); this equates to a difference of one grazing rotation between these varieties. Each tonne of pasture dry matter utilised on Irish dairy farms increases profit by €173/ha (Hanrahan *et al.*, 2018); therefore, it is essential to choose varieties with increased herbage production and grazing efficiency. Byrne *et al.* (2018) reported differences in grazing utilisation between varieties is affected by ploidy, proportion of free leaf lamina in the plant, tiller mass and dry matter digestibility. There was also a significant interaction between the number of grazings and sward age due to varieties responding differently to changes in sward age: Drumbo recorded 1.1 fewer grazing events in year 10 compared with year 1 (6.4 and 7.5 events, respectively), whereas Glenveagh recorded 1.1 more grazing events in year 10 compared with year 1 (8.1 and 7.0 events, respectively).

Conclusion

The findings of this study illustrate the importance of choosing high yielding varieties when reseeding as this can increase herbage production and subsequently maximise the number of grazing rotations per year. Additionally, choosing the correct varieties will help to optimise the utilisation of grazed grass and reduce overall production costs.

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The importance of dairy farming for human nutrition: How can we increase protein-efficiency?

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Abstract

A major downside of dairy farming is the use of human-edible protein (HEP) sources for feeding cows, as some dairy farming systems provide less HEP than they consume. We investigated this aspect in an experiment comparing two dairy production systems (high-input and low-input) at the same farm. Based on daily measurements of feed intake and milk production we calculated the daily human edible protein conversion efficiency (HEPCE) for single cows. We used a five-year dataset to investigate the factors affecting the changes of HEPCE over the year. HEPCE was computed for each cow as the ratio between the mean HEP used for feeding and the mean amount of milk protein produced in the 5 days prior to the milk performance tests. For data analysis, we used a linear mixed effects model accounting for the effects of the input system, lactation stage, seasonal effects (month of the year) and of the grazing season. We observed that the high-input farming system was generally less efficient in converting protein, due to the feeding regimes. Access to pasture during summer months was an important driver for increasing HEPCE in the low-input system. Our findings underline how different production systems can help reduce food-feed competition.

Keywords: dairy cows, grazing, human edible protein, human edible protein conversion efficiency, system comparison

Introduction

Dairy farming can provide additional protein for human consumption. However, highly intensified dairy production systems that are heavily reliant on concentrates sometimes use more edible protein than they can produce (Ertl *et al.*, 2015). This inefficiency of providing human edible protein (HEP) is problematic regarding the sustainability of the dairy production sector. There is some evidence that grass-based feeding strategies are more efficient to provide HEP (Ertl *et al.*, 2015), but the changes of human edible protein conversion efficiency (HEPCE) at the individual cow level have seldom been taken into account. Therefore, we investigated the effects of input system, lactation phase, seasonal effects, and of the grazing season in two different dairy production systems on individual cow level to assess factors influencing HEPCE.

Materials and methods

We used data from the project ‘Comparison of dairy farming systems’ taking place at the experimental farm Mair am Hof in Dietenheim/Bruneck (South Tyrol, NE Italy, 890 m asl), where since 2019 a high-input dairy farming system (whole-year indoor housing, high reliance on concentrates and silage) is being compared with a low-input system (grazing during the growing season, no silage fed) on a single farm. Feed intake, the composition of the ration and milk yields were recorded daily, and milk solids and bodyweight changes were recorded on average every 39 ± 7.6 days. Dry matter intake

on pastures was estimated based on the animal's energy needs and biweekly measurements of energy contents of herbage (Zanon *et al.*, 2023). For this paper we use a dataset from February 2019 until January 2024. During this time, the low-input system consisted of 15 Tyrolean Grey cows, the diet of which was pasture herbage (5.5 ± 6.8 kg DM) starting from April until November, hay (7.6 ± 4.6 kg DM) and concentrate (2.2 ± 1.3 kg DM), and their daily milk yield was 21.2 ± 6.0 kg. The high-input system was represented by 15 Simmental cows, with a diet of grass silage (5.2 ± 1.4 kg DM), maize silage (5.7 ± 1.4 kg DM), concentrates (6.9 ± 2.0 kg DM) and hay (2.9 ± 1.1 kg DM); their mean daily milk yield was 31.9 ± 8.3 kg. HEP calculation was based on estimated human-edible proportions of crops and by-products (Wilkinson, 2011) used to feed the cows. The HEP of fibrous feeds (hay, grass silage, maize silage and herbage) is 0, that of cereals and soybeans 0.8 and that of by-products is 0.2. Three different concentrates were fed in the trial: (a) a grain mix (protein content $85 \text{ g (kg DM)}^{-1}$, HEP proportion 0.76); (b) a protein concentrate (protein content $400 \text{ g (kg DM)}^{-1}$, HEP proportion 0.46), with both (a) and (b) being part of the mixed ration, and (c) a concentrate fed via the automatic feeding station (protein content $170 \text{ g (kg DM)}^{-1}$, HEP proportion 0.61), that only the high-input group can access. To be able to measure HEPCE also when no HEP was fed, we computed the amount of HEP used for feeding to produce a kg of milk protein. Thus, a value of less than one means that more HEP is produced in the process than is being fed to the animals; a value greater than one results in a net loss of HEP. To consider the effect of feeding in the days preceding sampling and to reduce the distorting effects of single unusual day values, we used the mean amount of HEP-intake and the mean amount of milk protein produced in the five days prior to the milk performance testing event. For the statistical analysis, we used a linear mixed model accounting for the input system, the period of the year (i.e. outside or during the time in which the low-input group can access pasture), the month of the year and the lactation stage expressed as a class, as well as the interaction terms of input system and the other three variables. We considered the single cows and the experimental year (from 1st February until 31st January of the following solar year) as random effects. Multiple comparisons were performed by Sidak test. HEPCE values were square root transformed to meet the assumptions of normality distribution of residuals and variance homogeneity. Statistical analyses were made with the packages *lme4* (function *lmer*) and *emmeans* in R (4.4.1).

Results and discussion

All investigated factors, as well as the interaction terms of input with month, lactation stage and grazing season were significant ($P < 0.001$ for all terms but grazing season, for which P was 0.002).

Concerning the seasonality, high-input cows showed nearly constant HEPCE values during the whole year, whilst those of low-input cows exhibited a pronounced decrease during the months fully included

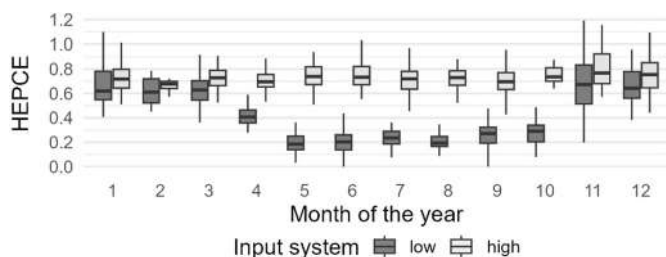


Figure 1. Variation of HEPCE depending on input and the month. March/April and November are the months in which cows start and finish grazing, respectively.

Table 1. Estimated marginal means and 95%-confidence interval (in square brackets) of the estimate of HEPCE depending on input system and the period of the year (outside or during the time between the start and the end of grazing for the low-input system)

Input system	Period of the year	
	Outside the grazing season	During the grazing season
Low	0.47 [0.42–0.53] ^{Ab}	0.37 [0.33–0.41] ^{Aa}
High	0.69 [0.64–0.75] ^{Ba}	0.74 [0.69–0.80] ^{Ba}

Analysis with square-root-transformed data; back-transformed values are shown. Means sharing no uppercase letters within the grazing season and means sharing no lowercase letters within input system do not significantly differ from each other.

into the grazing period (April to October) (Figure 1). Higher HEPCE values were observed for the high-input cows and were detected for all months except February. The high-input system had a HEPCE of 0.69 outside and 0.74 during the grazing season, the difference not being significant (Table 1). In comparison, the HEPCE of the low-input system was 0.47 outside and 0.37 during the grazing season, with the difference between the two values being significant. Our findings underline the importance for dairy cows to be able to feed on herbage from pasture during summer months to improve their HEPCE. Even in the time beyond the grazing season, the less concentrate-dependent low-input system was found to be more efficient in producing HEP, even though both systems were able to generate more HEP than they consumed. Thus, also the high-input system, that is more reliant on concentrates for feeding, is efficiently producing HEP during the lactation period.

Conclusion

Lactating cows in a low-input dairy farming system reliant on less concentrate for feeding, and with pasture access during the growing period, show pronounced differences in HEPCE within years, whilst dairy cows within systems that are strongly depending on concentrates and year-round indoor housing show generally a lower HEPCE and less variation of HEPCE within years. The differences of HEPCE are especially large during summer months due to the integration of pasture herbage into the diet of the low-input system, highlighting the importance of high-quality herbage availability on pasture.

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Influence of management in economic and environmental performance of livestock systems

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Abstract

Greenhouse gas (GHG) emissions from beef production vary significantly depending on livestock production systems (LPS), yet the underlying factors influencing these differences remain poorly defined. Here, we investigate environmental and economic performance simultaneously of cow-calf beef systems in Alentejo, Portugal. Environmental assessment, conducted through a mass balance model, demonstrates substantial variability in GHG emissions (15–124 kg CO₂e (100 g protein)⁻¹). Key influencing factors identified include the LPS type (confined, pasture-based, or mixed), cattle breed, age at slaughter, and both adequacy and upstream environmental impact of supplemental feeding. Grazing-based systems typically achieved lower emissions, provided cattle received appropriate supplementation. Conversely, under suboptimal supplementation during grazing, confined systems were optimal for low to intermediate feed impacts. Slaughtering at weaning became advantageous at higher feed impacts. Optimal emission ranges per LPS were only 15–35 kg CO₂e (100 g protein)⁻¹. Economically, the study explored operational returns (before subsidies) and eco-efficiency (GHG emissions per monetary unit earned). Highest operational returns consistently favoured post-weaning confinement, notably with Angus cattle grazing on sown biodiverse pastures and slaughtered at 11 months. Eco-efficiency outcomes aligned with optimal economic results for Angus and Charolais breeds. Findings underline the importance of meticulous pasture and feed management.

Keywords: Alentejo, beef, eco-efficiency, greenhouse gas emissions, modelling, optimization

Introduction

Beef production faces growing criticism for its environmental impact, especially due to high greenhouse gas (GHG) emissions (de Vries *et al.*, 2015; Poore & Nemecek, 2018). Yet, in regions like Portugal's Alentejo, beef systems are also integral to rural economies and land management (Esgalhado *et al.*, 2020). This underscores the importance of improving the environmental performance of existing livestock systems while ensuring rural development (Herrero *et al.*, 2016). The complexity of beef production, spanning multiple breeds, feeding systems, grazing practices, and slaughter timings, makes it difficult to generalize mitigation strategies. Assessing and improving regional beef production requires analysing the factors that are more relevant at regional level (Lynch, 2019).

Here, we perform an assessment of multiple management options for beef production systems in Alentejo and carry out an economic and GHG assessment to define their performance.

We defined optimal trade-offs depending on breed, pasture type, feed strategy, and slaughter timing. By combining GHG emissions modelling with farm-level economic optimization, this work supports more targeted, realistic pathways for reducing emissions while maintaining economic viability

in Mediterranean beef systems under realistic production constraints. It highlights the trade-offs and synergies between profitability and GHG mitigation, identifies system-specific optimal slaughter ages, and reveals how pasture type, breed selection, and feed adequacy drive both emissions and income. The approach also introduces scenario testing under uncertainty, including variations in feed price, feed impact, and supplementation efficiency.

Materials and methods

This study integrated environmental and economic assessments of cow-calf beef production systems in Alentejo. A mass-balance model (“BalSim”; Teixeira *et al.*, 2019) was used to simulate GHG emissions across multiple livestock production systems (LPS), including pasture-based, confined, and mixed systems. Input data were derived from detailed case studies representing different combinations of breed (Alentejana, Angus, Charolais, Limousin), pasture type (semi-natural or sown biodiverse; Teixeira *et al.*, 2015), feeding strategies and age at slaughter (between weaning and 24 months of age). The model calculated carbon and nitrogen flows to estimate methane (CH₄), nitrous oxide (N₂O), and carbon dioxide (CO₂) emissions over the full production cycle, using AR6 100-year GWP factors (Morais *et al.*, 2025).

The environmental model was coupled with an economic analysis of operational returns (€/kg live weight) prior to subsidy, and an eco-efficiency metric defined as GHG emissions per euro earned (kg CO₂e/€). The “BalSim” model was extended to quantify economic flows and carry out economic cost-benefit assessment. A series of production scenarios was evaluated to test the effect of slaughter age, supplementation adequacy, and upstream feed cost. For each scenario, combinations of management and feeding practices were analysed across realistic feed price and impact ranges. Optimization criteria focused on minimizing emissions while maximizing economic return. Sensitivity analyses explored how changes in feed quality and emission intensity influenced environmental and economic outcomes.

Results and discussion

GHG emissions varied substantially across production systems, ranging from 15 to 124 kg CO₂e (100 g protein)⁻¹. This spread was primarily influenced by LPS type, breed, age at slaughter, and the adequacy and upstream impact of feed. Grazing-based systems consistently showed the lowest emissions when supplementation was nutritionally adequate during grazing. However, when supplementation was suboptimal, pasture systems underperformed, and confined systems could in some cases become environmentally preferable, particularly at low and moderate upstream feed impacts.

Slaughter timing also played a key role in determining emission intensity. While extended grazing often reduced emissions, scenarios with high-impact feed inputs favoured early slaughter (at or near weaning). In these cases, minimizing lifetime feed consumption outweighed the benefits of grazing. Across all systems, the interaction between slaughter age and feed impact revealed a nonlinear relationship, requiring tailored management decisions depending on local feed conditions.

When we minimized emissions by selecting the best LPS for each combination of feeding adequacy during grazing and upstream feed emissions, we found that it was possible to reduce the potential range of GHG emissions to 15–35 kg CO₂e (100 g protein)⁻¹.

From an economic perspective, systems finishing animals in confinement after weaning yielded the highest operational returns, particularly when using Angus steers on sown biodiverse pastures. Slaughtering at 11 months consistently provided optimal economic outcomes, due to favourable weight gains and feed conversion under controlled conditions. These systems achieved returns before subsidies of above 2.5 €/kg live weight.

Eco-efficiency patterns largely mirrored the economic results, especially for Angus and Charolais breeds. In systems with moderate environmental impact and high market value, both emissions per unit of protein and emissions per euro earned were minimized simultaneously. This alignment of economic and environmental performance suggests that system-level optimization is feasible without compromising profitability.

Conclusion

This study shows that strategic adjustments in beef production—especially around feeding and slaughter timing—can substantially reduce emissions without compromising profitability. High-performing systems exist within each LPS type, but their success depends on careful matching of breed, pasture quality, and feed inputs. This integrated assessment provides a practical framework for aligning climate mitigation with farm-level viability in Mediterranean livestock systems.

Acknowledgements

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Biodiversity impacts of European livestock systems: a review of indicators

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Abstract

European livestock production systems can have both positive and negative impacts on biodiversity, depending on their management regime or ecological context. The objective of this study was to systematically review scientific literature to identify studies evaluating the positive and negative impacts of European livestock production systems on biodiversity, across levels of biological organization from populations to communities and ecosystems. The search included peer-reviewed, open-access articles in English (2015–2024), yielding 815 records. After duplicate removal and screening, 96 studies met eligibility criteria. Data were extracted on study features, including livestock type, management regime, systems or practices being compared, biodiversity indicators and overall impact of livestock. The studies represent 26 countries, with France, Spain, and Italy most frequently represented. Most focus on extensive systems, mainly cattle. Studies often compare land uses with or without livestock and differing grazing intensity. Community composition indicators are most common, followed by indicators of ecosystem structure and population abundance. Livestock presence is often associated with positive biodiversity outcomes, or conversely, the absence of grazing with negative effects. In contrast, increasing grazing intensity is generally linked to negative impacts, particularly under high pressure, whereas positive impacts are reported when very low-intensity regimes are compared with moderate regimes.

Keywords: biodiversity indicators, grazing intensity, systematic review

Introduction

The livestock sector represents both a challenge and an opportunity for biodiversity conservation. On the one hand, livestock production is a major driver of environmental pressures, including land use change, habitat degradation, GHG emissions, and nutrient runoff (FAO, 2020). On the other hand, especially in low-intensity systems, livestock can contribute positively to ecosystem management and help maintain structurally and biologically diverse landscapes (Ribeiro *et al.*, 2023). This dual role highlights the sector's potential to shape biodiversity outcomes—either by mitigating its negative impacts or by enhancing its positive contributions. A synthesis of the existing literature on the impacts of livestock on biodiversity is needed to inform such efforts. The main goal of this study is to review the scope and indicators used in studies assessing the impacts of livestock systems on biodiversity.

Materials and methods

The literature search applied a search string comprising four main components: (i) terms related to indicator terminology, (ii) the term biodiversity, (iii) terms representing livestock species and products, and (iv) terms covering the European territory (i.e., countries' names). The search was restricted to peer-reviewed, open-access research articles published in English between 2015 and 2024. A total of 815 records were retrieved from Web of Science ($n = 412$), Scopus ($n = 332$) and PubMed ($n = 71$). After merging results from all databases and removing 286 duplicates, 529 records remained

for title and abstract screening. Of these, 388 were excluded for falling outside the review scope, not covering Europe, not addressing livestock impacts on biodiversity, or not measuring/modelling biodiversity. This screening yielded 141 studies for full-text assessment, of which 96 were retained for data extraction.

Data were extracted on: (1) Study reference, (2) Geographic scope (country(ies) covered), (3) Livestock type (species and production system, e.g., dairy cattle, beef cattle), (4) Management regime (e.g., extensive, intensive), (5) System comparison (systems or practices being compared, e.g., grazed vs. ungrazed, species comparison), (6) Biodiversity indicators, (7) Targeted taxa or biodiversity component, and (8) Overall impact (qualitative assessment of the system’s impact, based on the study main findings, as highlighted by the authors).

Results and discussion

The list of selected publications covers a total of 26 countries and nearly two-thirds (61 out of 96) were published after 2020. Fifty studies focus on a single livestock species, with cattle being the most frequently represented ($n = 26$); 41 studies adopt a multi-species approach, and 5 studies do not specify the livestock species. Across all studies, cattle are the most frequently represented species ($n = 62$), followed by sheep ($n = 44$), and horses ($n = 20$). Most studies focus on extensive management systems ($n = 36$), followed by those comparing extensive and intensive systems ($n = 18$). Unassisted grazing (e.g., rewilding systems, producing environmental services) is examined in 9 studies, while intensive management alone is addressed in 4. Other management types not explicitly linked to intensity (e.g., specialized livestock farms vs. mixed farms) are considered in 15 studies.

Regarding system comparison (Figure 1), most studies compare land uses with or without livestock grazing and regimes under different grazing intensity, followed by comparisons based on livestock type (includes species, breeds and production sectors), farming system, and timing of grazing. Three studies compare diets or livestock products.

The range of reported biodiversity indicators is highly diverse, reflecting the studies’ varied biodiversity concerns.

Most studies use community composition indicators (e.g., species richness, diversity indexes, composition) (Figure 2), followed by indicators of ecosystem structure (e.g., vegetation structure, habitat

Type of comparison		
Livestock presence	50	52%
Livestock intensity	26	27%
Livestock type	15	16%
Farming system	12	13%
Livestock timing	10	10%
Other	3	3%

Figure 1. Main comparison types reported in the reviewed studies. Each study may include multiple types. Numbers on the left show study counts; bars on the right indicate percentages of the total ($n = 96$).

Indicator dimension			
Community Composition	75		78%
Ecosystem structure	29		30%
Species Populations	15		16%
Other	30		31%

Figure 2. Biodiversity dimensions represented by indicators in the reviewed studies. Each study may cover multiple dimensions. Numbers on the left show study counts; bars on the right indicate percentages of the total ($n = 96$).

	Presence		Intensity	
Positive	16	32%	1	4%
Positive & Neutral	9	18%	2	8%
Neutral	4	8%	2	8%
Negative & Neutral	5	10%	2	8%
Negative	7	14%	16	62%
Positive & Negative	9	18%	3	12%

Figure 3. Type of impacts reported by studies comparing livestock-grazed vs. ungrazed areas ('Presence', $n = 50$) and higher vs. lower grazing intensity regimes ('Intensity', $n = 26$). Bars show the percentage of studies reporting each type of impact. Intensity comparisons include only studies conducted under grazing conditions.

richness, ecological infrastructure) and species populations (mainly species abundance). 'Other' indicators include ecosystem function, species traits, scoring systems using proxy variables, and LUC-driven biodiversity loss.

With respect to the biodiversity components targeted by the studies, 48 included variables related to plant communities or species, 17 focused on vertebrates (primarily birds (15 studies), followed by mammals (8) and herptiles (3)), 15 addressed invertebrate communities, and 25 examined overall ecosystem-level variables.

Regarding overall impacts, the analysis of studies comparing livestock presence and grazing intensity reveals two main patterns (Figure 3). First, livestock presence is often associated with positive impacts on biodiversity—or conversely, the absence of grazing with negative effects. Second, higher grazing intensity is predominantly linked to negative outcomes. Positive impacts of livestock presence are reported across a broad range of biodiversity dimensions, including the richness and diversity of plant, invertebrate, and vertebrate communities, ecological infrastructure, and the maintenance of habitat quality for fauna. However, negative impacts are also observed for many of the same components, particularly plant community composition and diversity, habitat quality for target species, and in cases of overgrazing.

In the case of increased livestock grazing intensity, negative impacts are primarily associated with higher intensity levels, affecting multiple biodiversity dimensions. These include reductions in plant and invertebrate diversity, declines in mammal and bird presence and habitat availability, and overall community simplification. In contrast, positive impacts are reported when very low-intensity regimes are compared with moderate grazing levels, including increased plant community diversity and evenness, as well as improved habitat conditions for open-land bird species.

Conclusion

This review provides an overview of the scope of existing research and the types of indicators used to assess the impacts of livestock systems on biodiversity, with grazing presence and intensity emerging as the most common comparison types. While grazing presence and moderate regimes can support biodiversity under certain conditions, higher grazing intensity is generally associated with negative outcomes across multiple dimensions of biodiversity.

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Systematic review of impacts of conventional and pasture-based beef and milk production

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Abstract

Livestock play a vital role in providing nutrition and sustaining livelihoods, especially in rural regions of low- to middle-income countries. However, rising global demands for animal products, driven by population growth, income increases, and urbanization, impose significant environmental challenges, notably greenhouse gas (GHG) emissions and biodiversity loss. However, those impacts are thought to vary substantially across locations and production systems. To address methodological inconsistencies and regional variability, this research conducts systematic meta-analyses following PRISMA 2020 guidelines, focusing separately on European milk and global beef production systems. For milk production, an initial extensive screening identified 86 relevant studies primarily from Italy, Ireland, and Spain, covering intensive, extensive, pasture-based, mountainous, island, and confined systems. Meanwhile, for global beef production, an initial pool of 94 articles from countries like Brazil, the United States, and Italy were reviewed, emphasizing pasture-based and high-grain finishing systems. This research compiles detailed data to characterize and standardize system descriptions, facilitating accurate comparisons and developing a uniform LCA framework that enables comparisons between pasture-based and confined systems. We showed substantial variability in emissions results, which cannot be clearly attributed to the production system. These findings deliver updated insights into livestock sustainability, thereby supporting informed policymaking and sustainable agricultural practices.

Keywords: beef, eco-efficiency, greenhouse gas emissions modelling, optimization

Introduction

Livestock production is both a cornerstone of global food systems and a significant contributor of impacts to the environmental (Ritchie & Roser, 2019). In particular, cattle products face growing scrutiny due to high greenhouse gas (GHG) emissions, extensive land requirements, and potential impacts on biodiversity (Gerber *et al.*, 2013). As global demand for animal-source foods increases, mostly in developing countries, driven by population growth, urbanization, and rising incomes, pressure mounts to assess and mitigate the environmental costs of these systems (Morais *et al.*, 2021; Wisser *et al.*, 2023). While numerous Life Cycle Assessment (LCA) studies have examined livestock impacts, results remain difficult to compare due to heterogeneity in system definitions, methodological choices, and regional contexts. This is especially true for comparisons between pasture-based and confined beef systems, which differ fundamentally in their resource use profiles, productivity levels, and emission pathways. Influential reviews such as Poore and Nemecek (2018) underscore livestock's substantial environmental impact, emphasizing the critical need for systematic assessment across diverse production contexts.

To address this complexity, robust comparative frameworks are needed that standardize system boundaries, functional units, and data processing methods while preserving sensitivity to local variation. This study carried out systematic meta-analyses of beef and dairy production systems. Focusing on global

beef and European dairy, we harmonized system descriptions and LCA assumptions across more than 150 studies, applying strict inclusion criteria and a unified analytical protocol. Special attention was given to the influence of production system type (grazing vs. confined).

The goal was not only to improve the comparability of published LCA results but also to generate a data-rich foundation for regionally differentiated impact assessments. By standardizing inputs and outputs across diverse geographies, this work produced the most comprehensive synthesis to date of emission intensities in beef and milk systems, with a particular focus on how confinement and pasture use shape environmental outcomes.

Materials and methods

This study conducted two systematic meta-analyses following PRISMA 2020 guidelines to compare the environmental performance of grazing and confined cattle production systems. The first focused on global beef systems (Lopreto, 2024), while the second addressed European milk production (Medeiros, 2024). Peer-reviewed LCA studies were identified through structured searches in Google Scholar, Scopus, and Web of Science using keywords such as “Life Cycle Assessment”, “carbon footprint”, “beef production”, “dairy systems”, “grazing”, and “confinement”. After title, abstract, and full-text screening, 94 beef-related and 86 milk-related studies were retained for analysis. Studies were included only if they reported cradle-to-farm-gate emissions per functional unit (typically $\text{kg CO}_2\text{e (kg product)}^{-1}$ or $\text{kg CO}_2\text{e (unit of protein)}^{-1}$), and provided enough detail to classify the system as pasture-based, confined, or mixed.

Besides looking at the results of the studies, we also used the inventories to recalculate GHG emissions using IPCC 2019 GWP100 values, following a procedure set out by Lucić et al. (2025). LCA software openLCA was used to model inventories. All systems were reclassified under unified categories: fully pasture-based, fully confined, or mixed systems. Geographic region, breed, and intensity level were also recorded.

Results and discussion

Across both beef and milk datasets, production system type had a much lower role in explaining variation in GHG emissions than expected. On average, fully pasture-based beef systems had lower emissions per unit of protein than confined systems, but only under conditions of sufficient animal productivity and balanced diets. In low-input grazing systems with poor feed conversion or undernutrition, emissions per unit of output were among the highest observed. Conversely, high-productivity confined systems, particularly those relying on optimized grain-based rations and controlled manure handling, achieved competitive or lower emission intensities than extensive systems, especially in North America and Europe. This underscores that grazing alone does not guarantee lower emissions; management intensity and system design are equally critical.

For milk production, pasture-based systems generally exhibited slightly lower average GHG emissions per kg protein compared to confined systems, though the difference was narrower than in beef. The advantage of grazing was more evident in regions with long grazing seasons and high-quality forages, such as Ireland and northern Spain. However, confined systems with high-yielding animals and efficient feed use showed comparable or even superior performance when emissions were calculated per unit of protein rather than per kg milk. The results highlight the importance of using functionally meaningful units in LCA comparisons. Protein-based metrics proved more effective at capturing true system efficiency than weight-based ones.

Overall, the findings challenge binary narratives around the sustainability of grazing beef and milk systems. While grazing systems often deliver co-benefits like biodiversity support and landscape preservation, their climate performance depends on feed quality, reproductive efficiency, and herd management. Similarly, confined systems are not universally high-emitting; when carefully managed, they can achieve low emissions per unit of output. Better digestibility of grain and higher production efficiency can also reduce emissions. The analysis confirms that meaningful comparisons must go beyond system labels to consider how production practices interact with local conditions and resource constraints.

Conclusion

This study provides a harmonized comparison of GHG emissions from grazing and confined livestock systems, showing that environmental performance depends more on system management and regional conditions than on production type alone. For the global warming potential impact category, while grazing systems can offer climate advantages, especially with adequate nutrition and productivity, well-managed confined systems may perform equally or better in specific contexts. Naturally, for other indicators such as biodiversity, the situation could be different and grazing systems might have shown advantages, but there is a lack of LCA studies that go beyond climate change assessments. These findings emphasize the need for nuanced, context-specific strategies in livestock sustainability policy, grounded in standardized assessment methods and functionally relevant impact metrics.

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Limestone and stocking rate: effects on sheep growth and sward dynamics in a Mediterranean Montado

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Abstract

This study evaluated the effects of two grazing systems—continuous (CS) versus deferred stocking (DS)—and dolomitic limestone application on pasture structure, forage quality, and sheep performance in a Mediterranean silvopastoral system (Montado). A 6-month trial (December 2023–June 2024) was conducted in four 1-ha plots: UC (CS, unlimed), UD (DS, unlimed), TC (CS, limed), and TD (DS, limed). DS produced markedly higher pasture utilisation (animal-days) than CS (3,111 vs 1,281) and liming increased mean sward height (TC 241.2 mm; others 132.8–205.6 mm). Crude protein (CP) declined and neutral detergent fibre (NDF) increased across time but were similar across treatments; CP tended to be slightly higher with limestone (TC, TD). The average daily gain was greater in CS, but the total cumulative liveweight gain per treatment was superior in DS (UD = 433.9 kg; TD = 472.2 kg vs UC = 223.9 kg; TC = 277.9 kg), indicating a more efficient pasture herbage conversion at higher instantaneous stocking pressure. Integrating DS with dolomitic limestone appears to enhance system-level productivity without compromising animal performance, offering a pragmatic pathway for sustainable intensification in dryland pastures.

Keywords: black merino, dolomitic limestone, grazing systems, Montado, sustainability

Introduction

In Mediterranean silvopastoral systems like the Montado, where centuries of livestock grazing have shaped vegetation and ecosystem dynamics, achieving a balance between animal performance and ecological sustainability is critical. The grazing system strongly mediates botanical composition, sward structure, and spatial heterogeneity, which, in turn, influence intake selectivity, diet quality, and long-term resilience (e.g., legume persistence, suppression of low-value species; Papachristou *et al.*, 2016). Deferred stocking (DS) can enhance utilisation and floristics but demands higher stocking rates and management effort, while continuous stocking (CS) favours individual performance through greater selectivity but risks patchy use and degradation (Norman *et al.*, 2010; Ben Salem & Smith, 2008). On acidic or Mn-toxic soils, dolomitic limestone can raise pH, reduce toxicity, and boost pasture growth and quality (Rodrigues *et al.*, 2016; Serrano *et al.*, 2020). However, few studies assess combined grazing system × liming effects on pasture and sheep performance. This study aimed to test whether dolomitic limestone, in combination with either CS or DS, would (i) increase sward height and stabilise nutritive value, and (ii) improve ovine growth—both per head and per unit area—in a Black Merino rearing system.

Materials and methods

The study took place at the University of Évora's Mitra farm, Alentejo, Portugal, from December 2023 to June 2024 in a Montado area dominated by *Quercus rotundifolia* Lam. Four ≈ 1 -ha plots were allocated to: UC (unlimed, continuous grazing CS; approx. 1 Livestock Unit (LU) ha⁻¹), UD (unlimed, deferred grazing—DS; approx. 2.5 LU ha⁻¹), TC (limed, CS), and TD (limed, DS). Twelve geo-referenced sampling points per plot captured sward height (three readings per point) and forage was monthly hand-clipped for CP and NDF determination (AOAC methods). Black Merino weaned females of similar age/body condition (an average age of 3.5 months and an average weight of 26 kg) were randomised to treatments (7, 17, 9 and 17 head in UC, UD, TC and TD), with body weight (BW) recorded tri-weekly. From December to March, all animals received a 26% CP supplement at 1.2% BW to buffer seasonal deficits. Due to atypically warm winter conditions which supported rapid pasture growth, DS paddocks experienced continuous occupation; thus, contrasts reflected differences in instantaneous stocking density more than on/off occupancy. Statistical analyses included repeated-measures GLM ANOVA (treatment, date; Tukey HSD post-hoc tests when $P < 0.05$) and linear regression for individual growth curves. Normality and homoscedasticity were verified using the Shapiro–Wilk and Levene tests.

Results and discussion

Despite a similar overall grazing period, DS accumulated far greater pasture herbage utilisation (animal-days) than CS (3,111 vs 1,281), consistent with the higher instantaneous stocking in UD and TD and more uniform defoliation under short-duration/high-pressure events, aligning with evidence that rotational or deferred pressure reduces spatial heterogeneity typical of selective grazing CS (Papachristou *et al.*, 2016). Liming increased sward height within the same grazing system: TC achieved the tallest mean canopy (241.2 mm) versus UC, TD and UD (205.6, 155.5 and 132.8 mm, respectively), supporting the known liming benefits on nutrient availability and root growth (Rodrigues *et al.*, 2016; Serrano *et al.*, 2020). Seasonally, height rose into late spring as temperature-moisture conditions favoured growth, before senescence. Importantly, amplitude analyses (difference between the 12 highest and 12 lowest points) indicated greater within-plot height dispersion under CS—especially limed CS (TC)—reflecting stronger animal selectivity when grazing pressure is lower and when higher-quality patches persist (Norman *et al.*, 2010). Under DS, amplitudes remained lower and more stable, consistent with tighter utilisation and less opportunity for selective avoidance.

Across all treatments, CP declined and NDF increased throughout the season, with sampling date—but not treatment—driving most of the variation. There was a mild tendency for higher CP in limed treatments (TC and TD), particularly late in the vegetative cycle, consistent with enhanced N supply and improved legume contribution under corrected pH.

Growth regressions showed that UD animals had a significantly different (lower) slope than the other treatments ($P < 0.01$), consistent with the hypothesis that higher pressure reduces per-head performance. Mean ADG patterns reiterated this: CS (UC, TC) tended to deliver higher individual gains, probably due to selectivity at lower grazing pressure (Ben Salem & Smith, 2008).

However, system-level output diverged: the cumulative liveweight gain per treatment was markedly higher in DS (UD = 433.9 kg; TD = 472.2 kg) than in CS (UC = 223.9 kg; TC = 277.9 kg). The limed (TD) delivered the most significant total gain, indicating that modest per-head trade-offs

can be more than counterbalanced by increased throughput where pasture herbage growth permits higher instantaneous stocking rates. These findings align with the sustainable intensification premise for Mediterranean pastures: managing grazing to improve utilisation and regrowth while using soil amendments to enhance baseline productivity (Serrano *et al.*, 2020).

The CS contrast (TC vs UC) highlights a growth response (taller canopy) and higher total gain (277.9 vs 223.9 kg), while under DS, TD outperformed UD in cumulative gain (472.2 vs 433.9 kg). These effects are plausible given improved pH and reduced Mn toxicity, which jointly enhance root activity, water-nutrient capture, and legume abundance (Rodrigues *et al.*, 2016). From a practical standpoint, liming amplifies the benefits obtained by DS, resulting in more biomass growth, more evenly harvested, and a higher yield of lamb per area.

First, atypically warm winter weather enabled a near-continuous presence of animals in DS paddocks, blurring the strict DS vs. CS dichotomy and effectively isolating differences in instantaneous stocking. Second, supplementation (26% CP at 1.2% BW) standardised basal nutrition in early months, reducing between-treatment divergence in quality-driven gains and placing greater weight on differences in pasture quantity and utilisation. These constraints, while pragmatic, mirror real-world management in Mediterranean systems and thus enhance external validity for producers facing inter-annual climate variability (Ben Salem & Smith, 2008). When the objective is maximising individual growth (e.g., finishing lambs quickly), CS at moderate stocking can be advantageous due to higher selectivity. Conversely, if the priority is total output per hectare, a DS approach that concentrates grazing pressure while allowing rest periods—and that is underpinned by corrected soil fertility—can yield larger cumulative gains. The present results quantify this trade-off: DS nearly doubled the total live-weight gain compared to CS, even though individual animals grew marginally slower.

Liming altered sward structure (taller canopies) and nudged late-season CP upward, likely through legume responses, which resulted in higher cumulative gains under both grazing systems. Given the modest cost and multi-year residual effects of dolomitic limestone, integrating periodic liming into Montado grazing plans represents a cost-effective switch for resilience and output (Rodrigues *et al.*, 2016; Serrano *et al.*, 2020). The balance between uniformity and planned selectivity (biodiversity provision) can be fine-tuned through DS timing and recovery windows.

Concluding remarks

Higher instantaneous stocking in DS delivered substantially greater total liveweight gain per hectare than moderate-pressure CS, despite slightly lower per-head performance. Dolomitic limestone increased sward height and tended to support better performance, with the DS-limed combination (TD) yielding the highest cumulative gain. For Montado producers seeking sustainable intensification, pairing DS with periodic dolomitic liming offers a robust route to elevate system-level productivity and ecological stability.

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Effect of twice-daily *Asparagopsis* supplementation on enteric methane emissions and productivity in grazing dairy cows

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Abstract

The methane abatement potential of *Asparagopsis* has not yet been proven in grazing systems, in which the predominant opportunity for supplementation is twice daily during milking. A study was undertaken in Teagasc, Ireland, in which $n = 10$ cows were offered 20 g/day of freeze-dried *Asparagopsis* meal mixed with 2 kg of concentrate (TRT) while $n = 10$ cows received 2 kg of concentrate without *Asparagopsis* (CON) at milking twice daily. Enteric emissions were measured using a GreenFeed unit and milk production was monitored. Preliminary results showed TRT cows produced significantly less methane relative to CON (-20.0%); however, this varied across the study. TRT cows produced 6.7% less milk solids relative to CON. Analysis is still ongoing to determine dry matter intake; however, it is expected that intakes will be lower for TRT cows as refusals of the concentrate and *Asparagopsis* mix meant that TRT consumed 7.4% less concentrate than CON. *Asparagopsis* fed twice daily to grazing dairy cows was effective at reducing methane. Although TRT cows produced less milk solids, this is likely explained, in part, by lower concentrate intake in TRT cows.

Keywords: *Asparagopsis*, dairy cows, grazing, methane, milk

Introduction

Feed additives are effective at reducing enteric methane (CH₄; Hegarty *et al.*, 2021; Fótus *et al.*, 2022); however, most research to date has been undertaken in indoor total mixed ration feeding systems. Research has highlighted challenges in terms of the practical application of feed additives in grazing systems, in which the predominant opportunities for supplementation are twice daily during milking. *Asparagopsis* can have high CH₄ abatement potential, with reductions of 26.7–67.2% reported in the literature (Roque *et al.*, 2019; Alvarez-Hess *et al.*, 2024). The CH₄ abatement potential of *Asparagopsis* has not yet been proven when fed twice daily in grazing systems. The objective of the present study was to determine the efficacy of *Asparagopsis*, pulse fed to grazing dairy cows twice daily at milking, in terms of enteric CH₄ abatement potential and impact on production and animal performance.

Materials and methods

An 8-week study was undertaken in Teagasc, Ireland, in which $n = 10$ cows were manually fed 20 g/day of freeze-dried *Asparagopsis* meal mixed with 2 kg of concentrate (TRT) while $n = 10$ cows received 2 kg of concentrate without *Asparagopsis* (CON) on exit from the milking parlour twice daily. Prior to commencing the study, the animals were divided into homogeneous groups using a balanced randomization procedure based on parity (1, 2 or 3+), breed (Holstein-Friesian or Jersey×Holstein-Friesian), days in milk (DIM; 53.0 ± 19.61 days), mean daily milk yield (23.0 ± 5.38 kg day⁻¹), mean daily milk solids yield (2.1 ± 0.59 kg day⁻¹), body weight (BW; 482 ± 65.2 kg), and mean daily CH₄ production (293.5 ± 50.56 g day⁻¹) and randomly assigned to treatment and control. An 18-day adaptation period took place thereafter to gradually introduce TRT cows to the *Asparagopsis*. Cows

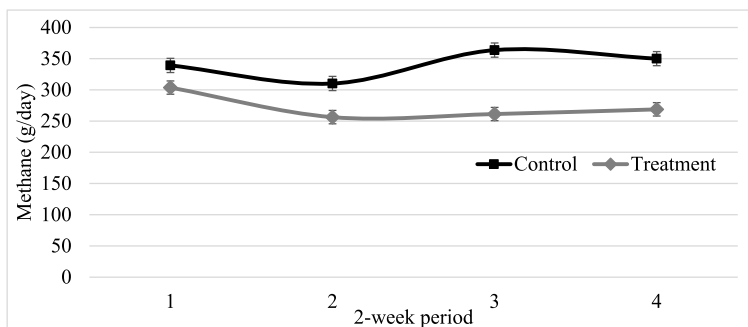


Figure 1. Least square means (standard error bar represents ± 1 SE unit) of enteric methane, as measured using a GreenFeed, of pasture-based dairy cows offered 20 g day^{-1} *Asparagopsis* supplemented twice daily across an 8-week experiment (Treatment), and those without *Asparagopsis* supplementation (Control).

were observed while consuming the control concentrate (CON) and concentrate and *Asparagopsis* mix (TRT) and refusals, if present, were recorded. All cows grazed together in one group, which was stocked at 2.6 cows/ha and managed in a rotational grazing system, with an average rotation length of 21 days. Enteric CH_4 was measured using a pasture-based GreenFeed unit. Body weight and body condition score (BCS) were monitored weekly. Milk yield was recorded daily, while milk solids were recorded weekly.

Statistical analyses were conducted using SAS (proc mixed). To account for diurnal variation in gaseous emissions and GreenFeed visits, spot measurements were adjusted for parity, days in milk, and visit hour (i.e. 01:00 to 02:00, 02:00 to 03:00, etc.) as is outlined by Fresco et al. (2023). Daily emissions for each individual animal were then calculated by averaging spot measurements over a 2-week period. To coincide with the fortnightly CH_4 data, productivity data were also averaged on a 2-week period. Data centred within breed and parity for pre-experimental data were included as a covariate in each respective model. Cow was included as a random effect, while 2-week period was included as a repeated effect. Fixed effects included in the models were treatment, breed, parity and 2-week period.

Results and discussion

Of the 2 kg *Asparagopsis* and concentrate mix offered to TRT cows each day, 541 g was refused. Feed refusals indicate that when *Asparagopsis* is fed within a small quantity of concentrate (i.e., 2 kg/day) that there may be issues in terms of palatability. In a review of *in vivo* *Asparagopsis* studies (Eason and Fennessy, 2025), the amounts of bromoform (mg) per gram of *Asparagopsis* ranged from 1.3 to 7.8 mg/g, which was substantially lower than that in the present study in which the *Asparagopsis* had a mean (standard deviation) bromoform concentration of 18.7 (2.92) mg/g, which was analysed retrospectively once the study had concluded due to time constraints. The bromoform concentration of *Asparagopsis* may therefore also be a contributing factor to the high feed refusals in TRT cows. Despite feed refusals, and the relatively small quantity of *Asparagopsis* offered (20 g/day), analysis showed TRT cows produced 20.0% less CH_4 ($P = 0.003$) relative to CON; however, this varied across the study (Figure 1). Reductions in CH_4 were similar to the -24.4% reported by Alvarez-Hess et al. (2024) in which cows were offered 409 mg/day of bromoform fed in two equal feeds. There are inconsistencies in previous research on the impact of *Asparagopsis* supplementation on milk production, with studies reporting a reduction in milk production often noting a corresponding decrease in concentrate DMI (Alvarez-Hess et al., 2024). In the present study, TRT cows produced 6.7% less milk solids ($P =$

0.079) relative to CON. Analysis is still ongoing to determine DMI; however, it is expected that intakes will be lower for TRT cows as refusals of the concentrate and *Asparagopsis* mix meant that TRT consumed 7.4% less concentrate than CON. Although TRT cows produced less milk solids, this is likely explained, in part, by lower concentrate intake in TRT cows. There was no effect of *Asparagopsis* supplementation on BW or BCS.

Conclusion

The findings from the present study highlight the CH₄ abatement potential of *Asparagopsis* when pulse fed twice daily to grazing dairy cows. Further research should be undertaken to establish the impact of *Asparagopsis* supplementation on dry matter intake and productivity, in addition to measures to minimise feed refusals. This was the first study in which *Asparagopsis* meal was pulse fed twice daily to grazing dairy cows; the findings presented herein are therefore to establish proof-of-concept, and to guide future research on *Asparagopsis* supplementation in grazing animals.

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Impact of grazing compared to a total mixed ration system on dairy cow milk production

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Abstract

Total mixed ration systems (TMR) provide dairy cows with a balanced ration for high levels of milk production. In temperate regions, with long grass-growing seasons, pasture-based systems offer a low-cost alternative. This study aimed to quantify the milk production responses and performance profiles of dairy cows grazing either grass-only (GRASS) or grass-white clover (CLOVER) swards full-time, compared to cows fed a TMR. A farm systems experiment was conducted over seven grazing seasons (2015 to 2021). Fifty-four spring calving dairy cows were assigned to one of the three treatment groups each February for the entire lactation. The GRASS and CLOVER treatments were rotationally grazed. Over the seven-years, TMR supported the greatest daily milk yield and milk solids yield (25.2 and 2.11 kg cow day⁻¹, respectively), CLOVER was intermediate (21.6 and 1.81 kg cow day⁻¹, respectively) and GRASS was lowest (20.5 and 1.74 kg cow day⁻¹, respectively). The GRASS and TMR cows achieved peak milk production on experimental week 9 (27.0 and 30.3 kg cow⁻¹, respectively), while CLOVER achieved peak milk yield on experimental week 12 (28.1 kg cow⁻¹). After peak yield, the weekly decline in milk production was more gradual in TMR cows (1%) than in GRASS and CLOVER cows (2.5%).

Keywords: clover, grass, milk production, total mixed ration

Introduction

Total mixed ration (TMR) diets can support superior levels of milk production from a fully nutritionally balanced diet. An alternative to a TMR-based diet is to exploit grazed grass, which offers a low-cost alternative. In temperate regions, pasture-based milk production is favoured because grassland generally occupies the areas not suitable for the production of human-edible crops (Hennessy *et al.*, 2020). Pasture-based dairy production systems are characterised by a long grazing season in which grazed grass makes up the majority of a dairy cow's diet, which is the cheapest and most economically efficient feed for ruminant production. Agricultural policy is putting pressure on farmers to ensure that food production is carried out in a sustainable manner. In temperate regions such as Ireland and New Zealand, greenhouse gas (GHG) emission ceilings have been announced to reduce the emissions associated with agriculture. Pasture-based dairy production is more environmentally sustainable compared with TMR-based systems due to fewer inputs and lower methane emissions (O'Neill *et al.*, 2012). However, in response to the need to feed a growing global population, a number of limitations exist for grassland farmers, which include fluctuations in herbage quality, grass utilisation and access

Table 1. Comparison of milk production and quality from dairy cows consuming total mixed ration (TMR) of grazing grass-only (GRASS) or grass-clover (CLOVER) pastures.

	TMR	GRASS	CLOVER	SE	<i>P</i> -value
Milk yield (kg cow ⁻¹ day ⁻¹)	25.2 ^a	20.5 ^b	21.6 ^c	0.40	<0.001
Milk fat (g kg ⁻¹)	49.1	49.3	48.9	0.45	NS
Milk protein (g kg ⁻¹)	35.6 ^a	37.1 ^b	36.9 ^b	0.19	<0.001
Milk solids yield (kg cow ⁻¹ day ⁻¹)	2.11 ^a	1.74 ^b	1.81 ^c	0.031	<0.001
Cumulative milk yield (kg cow ⁻¹)	6973 ^a	5824 ^b	5927 ^b	92.7	<0.001
Cumulative milk solids yield (kg cow ⁻¹)	590 ^a	489 ^b	507 ^c	5.6	<0.001

NS, not significant.

to additional land. Therefore, any increase in production will come from either increasing herbage production on the current land area, supplementary feeding, or conversion to a TMR-based system. The incorporation of white clover into grazing systems can increase herbage and animal production above that of a grass-only sward. Nonetheless, few comparisons exist within the literature that have simultaneously compared milk production from grass-only and grass-clover swards to that of a TMR diet; thus, the objective of this study is to quantify the milk production responses and profiles from dairy cows consuming a pasture-based diet, with or without white clover, compared to a TMR diet.

Materials and methods

A full lactation farm systems experiment was conducted over seven grazing seasons (2015–2021) at Teagasc, Moorepark, Fermoy, Co. Cork, Ireland. Each year, in February, 54 spring calving dairy cows were balanced on mean calving date (16th February $\pm$ 5.3), lactation number (2.8 ± 0.26), 2-week pre-experimental milk (24.2 ± 1.99 kg) and milk solid (MS) (2.17 ± 0.195 kg) yields, and randomly assigned to one of three treatment groups. These were: (1) total mixed ration (TMR), (2) grass-only swards receiving 250 kg N⁻¹ (GRASS) and (3) grass-clover swards receiving 150 kg N⁻¹ (CLOVER). Cows on the TMR treatment were housed indoors full time, and fed a diet composed of grass silage (4.5–9.0 kg DM), maize silage (4.5–9.0 kg DM) and concentrate (7.4–8.0 kg DM) in which the composition varied between years based on the grass and maize silage quality. The TMR was fed using roughage intake control systems and was fed out daily using a Keenan diet feeder (Keenan, Borris, Carlow, Ireland) at a 10% refusal rate. The pasture-based treatments were rotationally grazed and stocked at 2.74 LU/ha each year. The target pre-grazing herbage mass for the GRASS and CLOVER cows was 1300–1500 kg DM and the target post-grazing sward height was 4–4.5 cm. Botanical composition was measured prior to each grazing for the CLOVER swards, by cutting and separating samples into grass and legume fractions, followed by drying to determine the DM proportions. The pasture-fed cows were allocated 17 kg DM cow⁻¹ of herbage daily, along with an individual concentrate allocation of 0.89 kg DM cow⁻¹. Absolute milk yields were automatically recorded daily (Dairymaster Causeway, Co. Kerry, Ireland) and milk composition (fat and protein) was measured weekly by infrared spectrophotometry (Milkoscan 203 (DK-3400), Foss Electric, Hollerød, Denmark). Data were analysed using PROC MIXED in SAS 9.4 software (SAS Institute, Cary, NC, USA). Fixed effects included in the model were treatment, year, week, lactation number and associated interactions. Individual cow was the experimental unit, and week was included as a random effect.

Results and discussion

Over the seven years, TMR supported the greatest ($P < 0.001$) daily milk yield and milk solids yield (25.2 and 2.11 kg cow day⁻¹, respectively), CLOVER was intermediate ($P < 0.001$; 21.6 and 1.81 kg

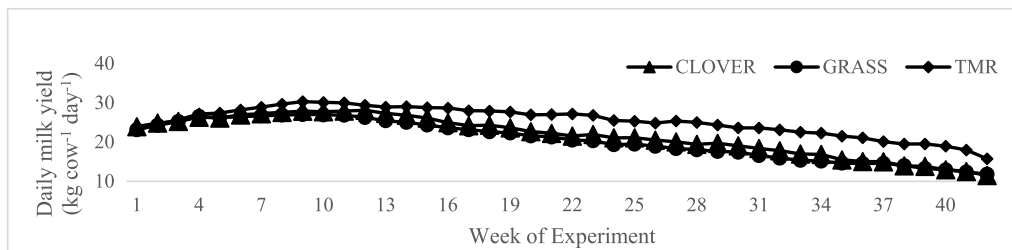


Figure 1. Comparison of three dairy cow diets for average daily milk yield.

cow day⁻¹, respectively) and GRASS was lowest ($P < 0.001$; 20.5 and 1.74 kg cow day⁻¹, respectively; Table 1). The GRASS and TMR treatments achieved peak milk production on experimental week 9 (Figure 1; 27.0 and 30.3 kg cow⁻¹, respectively), while CLOVER achieved peak milk yield on experimental week 12 (28.1 kg cow⁻¹). After peak yield, the weekly decline in milk production was more gradual in TMR cows (1%) than in GRASS and CLOVER cows (2.5%). Therefore, milk yield remained more persistent later into lactation for the TMR cows compared with the GRASS and CLOVER cows which were exposed to fluctuations in herbage quality throughout the grazing season.

The greater ($P < 0.001$) cumulative milk and milk solids production (Table 1) supported by the TMR diet was as expected and is likely a reflection of the higher and more consistent quality of the fully nutritionally balanced TMR diet compared to that of the pasture-based diets (Bargo *et al.*, 2002). At an average annual sward clover content of 21%, the CLOVER treatment produced an additional 18 kg milk solids each year on average compared to that of the GRASS cows, which is typically attributed to a combination of two key factors: increased dry matter intake (DMI) and higher-quality herbage (Egan *et al.*, 2018). Thus, in response to increasing milk production, the incorporation of white clover is a viable option to improve milk output from dairy farms in temperate regions. Furthermore, a TMR system can significantly increase milk production over that of a grass-only diet; however, the environmental and economic consequences of such feeding systems require further investigation.

Conclusions

In the current study, cows fed a TMR diet produced more milk and milk solids than those consuming grass-only or grass-white clover herbage. This was attributed to the more consistent quality of the TMR ingredients and the greater proportion of concentrate fed compared with pasture-based feeding. Adding white clover to a grass sward increased milk solids production by 18 kg per cow each year on average and enabled a significant reduction in chemical nitrogen fertiliser application, while maintaining the same stocking rate of 2.74 LU ha⁻¹.

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Development of a clover sub-model to improve the MoSt GG model and its prediction capabilities

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Abstract

The MoSt GG model was developed in Ireland to predict daily perennial ryegrass (PRG) growth and nitrogen (N) dynamics on grassland farms and is used as a research and decision support tool. Initially, it only simulated PRG and did not reflect the increasing use of white clover (WC) on grassland farms in Ireland. To address this, a clover sub-model was developed and integrated into the existing model. The new model simulates biological N fixation (BNF) based on WC biomass, soil N availability and weather, and accounts for species-specific growth traits, seasonality and interspecies competition. The effects of WC on soil N turnover, herbage N content and N excretion by cows are considered. Preliminary simulations over 21 years (2003–2023) using weather data from Teagasc Moorepark showed that under zero-N fertiliser application, PRG-WC swards produced 36% more annual herbage than PRG-only swards, despite an 8% reduction in PRG-yield contribution, which shows the model's capacity to simulate the contribution of BNF to yield and inter-species competition. At 150 kg N ha⁻¹, PRG-WC swards outperformed PRG-only swards receiving 250 kg N ha⁻¹ by 5%, showing the well-known potential of WC to substitute synthetic N without compromising yield at similar total N inputs.

Keywords: biological nitrogen fixation, grass, clover swards, grassland modelling

Introduction

Since 2019, the Moorepark St Gilles Grass Growth (MoSt GG) model (Ruelle *et al.*, 2018) has been used to predict daily perennial ryegrass (PRG; *Lolium perenne* L.) growth (kg DM ha⁻¹) on-farm, based on local weather and farmers' management practices (nitrogen (N) fertiliser application, pre- and post-grazing sward height). In the model, N dynamics are predicted and can be used to follow soil organic and mineral N content as well as to predict annual N leaching and gaseous emissions. The EU Nitrates Directive imposes regulations on N fertiliser use to protect water quality and to limit N₂O emissions, and further restrictions on N fertiliser application are expected in the coming years. This presents a challenge: how can Irish dairy farmers maintain milk production and profitability while reducing their reliance on synthetic N fertilisers? One solution is the inclusion of white clover (WC; *Trifolium repens* L.) in PRG swards. White clover can provide an additional source of N for plant growth through biological N fixation (BNF). Given the increasing adoption of PRG-WC based systems, it is essential to adapt the MoSt GG model to accurately predict growth of PRG-WC swards.

Material and methods

A WC sub-model has been developed and integrated into the MoSt GG to simulate PRG-WC swards. The original model contained a PRG, N, water and a soil sub-model. The new sub-model adds BNF, WC biomass dynamics and inter-species competition. The key inputs remain unchanged, with two main additions: (1) a binary parameter for WC presence/absence and (2) the initial WC percentage

Table 1. Impact of sward composition and N fertiliser application rate (kg N ha^{-1}) on annual herbage yield, N fixed through biological N fixation, average crude protein (CP) content, and number of cow grazing days

	PRG			PRG-WC		
	0	150	250	0	150	250
Annual herbage yield (kg DM ha^{-1})	7703	11 164	13 013	10 549	13 689	15 109
Annual N fixed (kg N ha^{-1})	—	—	—	82.3	62.5	46.8
Average CP content (kg N (kg DM)^{-1})	120	156	178	171	189	201
No. cow grazing days (day ha^{-1})	491	708	820	672	866	950

in the sward, which accounts for the variability between paddocks for WC establishment potential. In the model, PRG and WC compete for soil mineral N. Perennial ryegrass relies only on soil mineral N uptake, including that supplied by fertiliser, while WC balances two N sources, with the amount of atmospheric N fixed increasing as soil mineral N availability decreases, as demonstrated previously (Ledgard *et al.*, 2001). Biological N fixation depends on WC biomass, soil mineral N availability and environmental conditions. Growth and environmental response functions (potential growth, radiation use efficiency, temperature) are shared but parameterised differently. The original outputs have been kept and include daily growth, biomass partitioning, soil N and water dynamics, and N losses through leaching. New outputs have been added, including BNF, WC growth and N content, and sward composition. A series of simulations were run to evaluate the predicted impact of mineral N fertilisation on PRG-WC swards under grazing management using 21 years (2003–2023) of Moorepark weather data. The soil parameters were identical and consisted of a sandy loam soil with 6% organic matter, a depth of 100 cm, and a paddock area of 1.0 ha. A one-day grazing event happened each time the sward reached a height of 10 cm and all the biomass between 10 cm and the residual height, set at 4 cm, was removed. The number of cows was automatically adjusted for each grazing event to match the available herbage to a requirement of 16 kg DM per cow per day. Three mineral N fertiliser rates (0, 150 and 250 kg N ha^{-1} ; 0 N, 150 N and 250 N) and two sward compositions were compared (PRG monoculture and PRG-WC mix with an initial WC proportion of 10%). One-way ANOVA and *t*-tests ($\alpha = 0.05$) were conducted to test the statistical significance of the differences observed between treatments.

Unless specified, differences between treatments were statistically significant ($p < 0.05$). Annual DM yield increased as N input increased for both sward types. The yield response to additional N was similar for PRG and PRG-WC swards from 0 to 150 kg N ha^{-1} (+23 and +21 kg DM (kg N)^{-1} , respectively), whereas from 150 to 250 kg N ha^{-1} PRG had a greater yield response than PRG-WC (+18 vs +14 kg DM (kg N)^{-1} , respectively) (Table 1).

The PRG-WC swards yield consistently outperformed PRG at each N level (Figure 1A). This is due to the simulated BNF and complementarity of WC growth optimum, which compensates for the decline in summer growth of the PRG component of the sward. Seasonally, the PRG-WC swards reach peak growth later than PRG (week 24 *vs* 17) at 150 N, showing the effect of the species-specific parameters replicating the different timing. As expected, the yield partitioning (Figure 1A) shows that with increasing N fertiliser application, sward WC component declines (Enriquez-Hidalgo *et al.*, 2018). Although the difference was not statistically significant ($p > 0.05$), the PRG component of the sward was always lower in the PRG-WC mix than in the PRG monoculture at same N rate, and the gap declines as N rate decreases (−1.6% at 0 N, −3.6% at 150 N and −4.4% at 250 N). As the

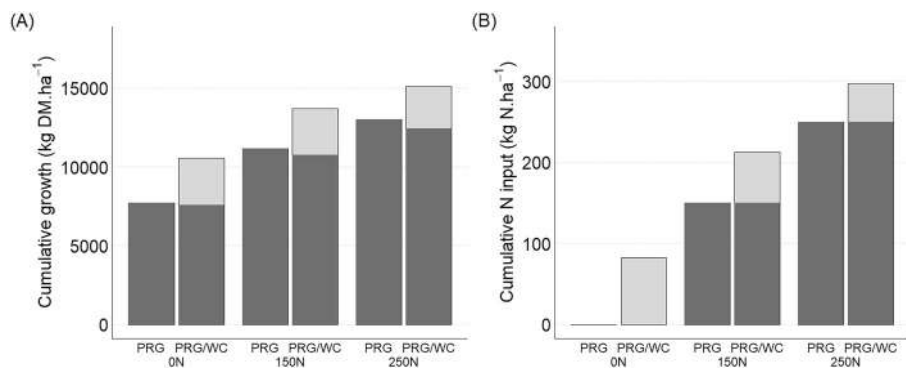


Figure 1. Effect of sward composition and N fertiliser application level on (A) annual cumulative growth (PRG in black; WC in grey), (B) annual cumulative N input (sum of N fertiliser applied (black), and N fixed through BNF (grey)).

N-response of PRG is greater than that of WC, its growth rate outpaces that of WC. This reduces WC access to light, further reducing growth. The simulated BNF matches previous experiments (Burchill *et al.*, 2014) and declines with higher N rate due to lower sward WC content and a direct inhibition of BNF by mineral N (Ledgard *et al.*, 2001) (Figure 1B). In PRG-WC swards, BNF contributes 100% of N inputs at 0 N, 29% at 150 N and 16% at 250 N (Figure 1B), which simulates the reduced reliance on BNF as mineral N increases due to its high energetic cost. At 150 N, PRG-WC swards appear more efficient than PRG swards as they produce 91 kg DM (kg N)⁻¹ input (59 kg DM (kg N)⁻¹ input if considering the fixed N) while PRG swards produce 52 kg DM (kg N)⁻¹ input. The crude protein content is higher in PRG-WC swards and in a range found in previous experiments (e.g. Enriquez-Hidalgo *et al.*, 2018) (Table 1). Both swards show a summer decline in crude protein content, partly due to the dilution of N as fibre content increases. These outputs are well reflected in the variation of grazing days between treatments, where higher yields support more grazing days and higher growth rates allow for a faster recovery (Table 1).

Conclusion

A new sub-model was developed for the MoSt GG model to simulate growth on PRG-WC swards. Evaluation shows that it can simulate BNF and predict herbage yield, quality, and seasonality in line with expectations.

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Effect of silage species on milk production and methane emissions in late-lactation dairy cows

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Abstract

The objective of this experiment was to investigate the effect of silage species on dry matter intake (DMI), milk production and methane emissions in late-lactation, spring-calving dairy cows. Twenty-eight dairy cows were blocked based on pre-experimental milk production, parity and breed and randomly assigned to one of two treatments. The dietary treatments consisted of mechanically harvested pasture by day and either perennial ryegrass (PRG) silage (GS) or PRG-red clover silage (GRCS) at night. All cows were supplemented with 2.7 kg DM day⁻¹ of concentrate. The study consisted of two covariate weeks followed by seven weeks of data collection. Forage DMI was recorded using roughage intake control feeders, and methane emissions were measured using a GreenFeed unit. Cows fed GRCS tended to have greater total DMI ($P=0.06$) and had greater milk yield ($P=0.02$) when compared with cows fed GS. There was no effect of diet on milk protein concentration, fat concentration, or milk solids yield. Cows fed GRCS had similar methane emissions but had lower methane per unit of DMI ($P=0.04$) when compared with cows fed GS. Within pasture-based systems, offering PRG-red clover silage to late-lactation dairy cows at night has the potential to maintain milk production without affecting methane emissions.

Keywords: dry matter intake, methane, pasture, red clover

Introduction

In Ireland, agriculture is responsible for 38% of greenhouse gas emissions (EPA, 2025). In this context, there is an increased interest in incorporating legumes into perennial ryegrass (PRG; *Lolium perenne* L.) swards due to their reduced reliance on synthetic nitrogen (N) fertiliser. In particular, producing red clover (*Trifolium pratense* L.) is receiving growing attention owing to the usually high crude protein (CP) concentration of red clover silages. The effect of feeding grass-red clover silage on dairy cow production has been widely studied in indoor systems where its potential to improve dry matter intake (DMI) and milk production has been demonstrated (Steinshamn, 2010). Regarding methane emissions, the effects of grass-red clover silage reported in previous studies were inconsistent (Gidlund *et al.*, 2017; Bica *et al.*, 2022; Weiby *et al.*, 2025). Furthermore, these studies were conducted using silage diets, which do not reflect pasture-based dairy systems, predominant in Ireland. Therefore, the objective of this experiment was to investigate the effect of silage species on milk production and methane emissions in late-lactation, pasture-based dairy cows.

Materials and methods

This experiment was conducted at the Teagasc, Animal and Grassland Research and Innovation Centre, Moorepark, Fermoy, Co. Cork, Ireland, from September until early-November 2024. Twenty-eight Holstein Friesian and Holstein-Friesian × Jersey dairy cows averaging (mean±SD) 191 ± 15 days in

Table 1. Effect of silage species on dry matter intake, milk production, milk composition and methane emissions¹

Item	GS	GRCS	SEM	P-value
Total DMI, kg day ⁻¹	17.0	18.1	0.42	0.06
Milk yield, kg day ⁻¹	14.0	15.4	0.40	0.02
Protein, g kg ⁻¹	40.6	40.7	0.38	0.87
Fat, g kg ⁻¹	60.7	59.1	1.30	0.37
Milk solids yield, kg day ⁻¹	1.43	1.51	0.04	0.14
Methane emissions, g day ⁻¹	360	358	8.94	0.88
Methane/DMI, g kg ⁻¹	21.1	19.7	0.47	0.04
Methane/milk solids, g kg ⁻¹	258	246	9.65	0.39

GS, perennial ryegrass silage; GRCS, perennial ryegrass-red clover silage; SEM, standard error of the mean; DMI, dry matter intake.

milk, 17.0 ± 2.8 kg of milk yield and 495 ± 50 kg of body weight were blocked based on breed, parity and pre-experimental milk production and randomly assigned to one of two dietary treatments. The two diets consisted of fresh pasture during the day, composed predominantly of perennial ryegrass, supplemented with either PRG silage (GS) or PRG-red clover silage (GRCS) at night. Both silages were made from secondary growths during July 2024. The pre-cutting herbage masses were 3861 and 4759 kg DM ha⁻¹ for GS and GRCS, respectively, with the GRCS comprising 689 g (kg DM)⁻¹ of red clover. Cows were housed in a free-stall barn with free access to feed through roughage intake control (RIC) feeders (Hokofarm). All cows were offered mechanically harvested fresh grass by day, distributed at 0800 h and 1100 h; the two silages were fed at 1530 h and cows were offered 1.75 kg DM day⁻¹ of a 120 g CP (kg DM)⁻¹ concentrate at milking and 0.91 ± 0.27 kg DM day⁻¹ on average of the same concentrate in the GreenFeed unit (C-Lock). The study consisted of a 2-week covariate period followed by 7 weeks of data collection. Individual milk yields were recorded daily, milk composition weekly and milk solids yield (fat yield + protein yield) was then calculated. Forage DMI was recorded daily using RIC feeders, and daily methane emissions were measured using a GreenFeed unit. Data were analysed using a linear mixed model within the MIXED procedure of SAS version 9.4. Diet, week, parity, breed and the interaction between diet and week were included in the model as fixed effects. The diet by week interaction was removed from the model if non-significant ($P > 0.10$). Cow nested within block was included as a random effect, with a covariate adjustment applied for each cow (except for intake and methane measurements). The repeated effect of the model was week. Diet effect was considered as significant at $P \leq 0.05$ and as showing tendencies at $0.05 < P \leq 0.10$.

Results and discussion

Cows fed GRCS tended to have greater total DMI ($+1.1$ kg DM day⁻¹; $P = 0.06$; Table 1) and had increased milk yield ($+1.4$ kg day⁻¹; $P = 0.02$) compared with cows fed GS, in agreement with Halmemies-Beauchet-Filleau *et al.* (2014). The greater DMI observed when feeding red clover silage has been attributed to its faster rates of digestion and passage when compared with grass silage (Kuoppala *et al.*, 2009). Similarly to Vanhatalo *et al.* (2006), there was no effect of diet on milk protein or fat concentrations; however, milk solids yield was also unaffected by silage species in the current experiment, despite a numerical increase observed when feeding GRCS.

In the current study, cows fed GS or GRCS had similar methane emissions and methane per unit of milk solids, averaging 359 g day⁻¹ and 252 g kg⁻¹ respectively across both diets. However, cows fed GRCS had lower methane per unit of DMI when compared with cows fed GS (19.7 vs. 21.1 g kg⁻¹,

respectively; $P=0.04$). The effect of red clover silage on methane emissions is equivocal (Gidlund *et al.*, 2017; Bica *et al.*, 2022; Weiby *et al.*, 2025), which could be attributed to several factors such as red clover proportion in the silage, silage maturity, harvesting date and proportion of silage in the overall diet. In previous experiments, silage was usually the sole forage component of the diet, whereas silage was offered only at night in the current experiment, with red clover representing 30% (on a DM basis) of the GRCS diet. Further research is needed to understand the mechanistic effect of red clover silage on methane emissions.

Conclusions

Feeding PRG-red clover silage to late-lactation dairy cows improved milk yield without affecting methane emissions. Further research is required to investigate the effect of PRG-red clover silage on N excretion, along with an in-depth study of the physiological mechanisms occurring when feeding grass-red clover silage within pasture-based systems.

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Virtual fencing for young dairy heifers in northern Sweden

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Abstract

The aim of the study was to assess the impact of virtual fencing (VF) on animal behaviour, performance and welfare of young dairy heifers during a northern Swedish summer. Thirty heifers (126–294 days old) were allocated into two balanced groups ($n = 15$): (I) control group managed with electric fencing, and (II) VF group. Heifers were fitted with VF collars prior to acclimatisation on pasture. Following this, a 105-day trial began, comprising three 35-day experimental grazing periods, each including a learning phase (days 1–6) and a behavioural sampling phase (days 33–34). Only data from the VF group were assessed for VF interactions, with age used as the treatment factor ($n = 5$). For behavioural comparisons, VF and control groups were evaluated. Data on body weight, cortisol accumulation in hair samples, biomass availability, and collar data reliability are still being processed. The paper only addresses data from period 1, and preliminary results demonstrated that heifers exhibited a rapid learning curve to the virtual boundaries, with escape events decreasing sharply after the learning phase. Age had no significant effect on VF interactions. Behavioural analysis after the learning phase revealed no significant differences in lying or grazing observation between the groups.

Keywords: animal welfare, GNSS, livestock precision farming, virtual borders

Introduction

Northern Sweden has mild summer temperatures with prolonged daylight, providing favourable conditions for grazing. Although there is considerable interest in expanding pasture utilisation in the region, the labour and costs associated with monitoring animals and fencing of grazing areas pose significant challenges to maintain competitive pasture-based livestock production. In this context, precision livestock farming tools such as virtual fencing (VF) may offer a solution by reducing labour requirements and enhancing management flexibility. The technology uses GNSS-equipped neck collars with integrated telecommunications functions. While the design and functionality vary among companies, the main components typically employ audio cues (AC) and electrical pulses (EP) ranging from 1500 to 3000 V (<0.1 J) to establish boundaries and containment zones instead of traditional physical fences. Despite several recent studies investigating the use of VF, some aspects remain unclear. For instance, the technology's performance under a range of environmental conditions needs further investigation to ensure reliability (Fuchs *et al.*, 2025). Northern Sweden often experiences cold, cloudy summer days, which could potentially affect collar performance. Another relevant aspect is the suggested animal age limitation for using the collars, as there is limited literature on how VF affects young cattle. Recently published data indicated no impairment of animal welfare, with cattle learning to associate AC with EP and subsequently avoiding EP by turning at the virtual boundary in response to AC (Campbell *et al.*, 2019; Hamidi *et al.*, 2022; Fuchs *et al.*, 2024). However, the age of cattle tested in previous VF experiments has varied greatly, and, limited scientific published data on the use of VF

with young cattle below six months of age. Addressing both aspects, we aimed to study the impact of VF on the animal behaviour, performance and welfare of young dairy heifers during a northern Swedish summer.

Materials and methods

The study involved 30 Nordic Red dairy heifers (186 ± 51.1 days of age; 222 ± 55.7 kg body weight) and was conducted from May to September 2025 at the Röbäcksdalen experimental farm of the Swedish University of Agricultural Sciences in Umeå ($63^{\circ}45'N$, $20^{\circ}17'E$). Handling of animals in the trial was approved by the Swedish Ethics Committee on Animal Research (Dnr. A 6-2021+5.8.18-03514/2025). Prior to the experiment, the heifers were weighed and fitted with a VF neck collar (Monil, v2.0, Norway, 2024; standard size GNSS-equipped neck collars with solar panel battery). Subsequently, the animals were divided into two balanced groups ($n = 15$) based on age and body weight: (I) control group managed with physical electric fencing, and (II) VF group. Both paddock areas were enclosed with physical fences to ensure safety due to their proximity to the highway. On the day the heifers were assigned to their paddocks, a five-day acclimatisation period was implemented. Following this, a 105-day experiment began, consisting of three 35-day experimental grazing periods. Each period included a learning phase (LP; days 1–6) and a behavioural sampling phase (days 33–34). In the VF group, virtual boundaries and cue functions (audio and electrical) were activated on day 1 (start of the LP), whereas collars in the control group operated in tracking-only mode. In experimental period 1, both groups had access to full paddocks (6.8 ha each). For experimental periods 2 and 3, each paddock was divided into two equal sections (areas A and B; 3.4 ha each). Thus, in period 2, the heifers grazed in area A, and in period 3, they grazed in area B until the end of the trial. This arrangement was designed to assess whether the heifers underwent a learning phase each time the VF boundary was moved.

Fencing interactions were evaluated throughout the experiment. For this specific analysis, only data from the VF group were used, considering the total number of AC, EP, and escape events, with animal age as the treatment factor ($n = 5$): Age 1 (221–295 days), Age 2 (161–220 days), and Age 3 (128–160 days). Data were analysed separately for LP and the after LP using PROC GLM in SAS.

During the behavioural sampling phase, scan sampling was performed every 10 minutes over 8 hours on two consecutive days, following a cross over design by two trained observers (inter-observer reliability: ICC = 0.904). An ethogram containing 19 behaviours was used, and all behaviours were recorded as a group, with individual animals not identified. Behaviours from both days were then combined and expressed as percentages of the total observations for control and VF groups. The four most frequently observed behaviours (grazing, lying down ruminating, lying down, and lying down curled up) were analysed using a Wilcoxon rank-sum test with experimental group (control or VF) as the fixed factor.

Results and discussion

Data on body weight, hair cortisol, biomass utilisation, and VF collar reliability from all periods are still being processed; therefore, this paper presents only VF interaction and behaviour results from period 1. During the six days of the LP, all 15 heifers in the VF group recorded 440 AC, 208 EP, and 30 escape events. After the LP, heifers recorded 2,521 AC, 129 EPs, and only 1 escape event. Daily averages showed a reduction of up to 91.1% in EP and nearly 100% in escape events. This study aligns with others showing that VF technology does work as heifer escape events and EP decreased over time (Fuchs *et al.*, 2025; Confessore *et al.*, 2024). Age did not affect ($P \geq 0.495$) how the heifers interacted

with the VF, either during the LP or afterwards. There is limited literature on animals under six months of age and their ability to learn VF technology, but this study provides novel insights suggesting that age may not be a significant factor in the learning process. Initially, we expected younger heifers to either (a) not understand the sound cues and therefore fail to reduce EP and escape events, or (b) interact less with the VF, potentially learning from the older ones; however, preliminary results did not support these hypotheses.

Behavioural observations were conducted after the LP to investigate whether heifers managed with VF differed in behavioural patterns compared to those managed with electric fences. Preliminary results showed no significant differences ($P \geq 0.72$) between control and VF heifers in the observed frequency of grazing, lying down ruminating, lying down, and lying down curled up at after the LP. Time spent ruminating and lying down is often considered a useful behavioural indicator of positive welfare in cattle (Wagner *et al.*, 2018; Olmos *et al.*, 2009). Our findings align with previous studies with cattle at different ages, which also reported no significant effects of VF on animal behaviour (Campbell *et al.*, 2019; Confessore *et al.*, 2024). Preliminary results suggest that VF did not compromise animal behaviour evaluated after learning phase under the conditions of this study.

Conclusion

Conclusions indicate that learning to respond to VF was not influenced by the age of the animals, as all heifers demonstrated a clear learning curve with reduced electrical pulses and escape events. No behavioural differences were observed between VF and control groups, suggesting that VF did not compromise animal behaviour in this study.

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The effect of plantain inclusion in perennial ryegrass-white clover swards on herbage yield and milk production

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Abstract

Irish dairy production systems are characterised by high utilisation of grazed pasture which typically consists of perennial ryegrass (*Lolium perenne* L.) and white clover (*Trifolium repens* L.). The addition of companion 'herb' forages such as plantain (*Plantago lanceolata* L.) could potentially improve animal and sward performance over traditional swards. A two-year grazing experiment comparing perennial ryegrass-white clover (WC) and perennial ryegrass-white clover-plantain (PL) swards was conducted in Teagasc Moorepark, Ireland in 2023 and 2024. This study investigated the effect of sward type on herbage production and animal performance. Seventeen dairy cows grazed rotationally each year from May in year 1 and April in year 2 to November. The average plantain proportion during the experiment was 43% and clover content was 16.8 and 9.4% for the WC and PL treatments, respectively. Treatment had no effect on milk production (336 and 340 kg MS cow⁻¹ for WC and PL, respectively). There was also no difference in herbage production in either year (11 341 and 11 356 kg DM ha⁻¹ for WC and PL, respectively). This study positively shows that the addition of PL to a WC sward can result in similar milk and herbage production; however, the persistency of plantain remains a potential challenge.

Keywords: herbage yield, milk production, plantain, white clover

Introduction

Pasture-based dairy production in Ireland typically consists of perennial ryegrass and white clover swards that are intensively grazed to utilise as much grazed pasture in the diet as possible. The inclusion of a third forage species, like plantain, has received renewed interest as it has previously been cited that improved animal and sward performance can be achieved by grazing these diverse swards over more traditional ones (Grace *et al.*, 2019). The lower persistency of certain herb species is a known disadvantage of these swards (Gilliland, 2022), however, recent studies have shown plantain can persist under intensive grazing for up to 4 years (Hearn *et al.*, 2024). This experiment's objective was to quantify how the addition of plantain to our traditional sward would affect milk and herbage production by comparing perennial ryegrass-white clover and perennial ryegrass-white clover-plantain swards under an intensive grazing system.

Materials and methods

A dairy farm systems experiment was conducted at Teagasc Moorepark, Ireland in 2023 and 2024. Year one of the study took place from May to November 2023, while year 2 took place from April to November 2024. The experiment had two treatments: a perennial ryegrass-white clover sward (WC) and a perennial ryegrass-white clover-plantain sward (PL). Both were sown in 2022 at a total seeding rate of 35 kg seed ha⁻¹, the WC treatment was sown with 30 kg ha⁻¹ perennial ryegrass and 5 kg ha⁻¹ of white clover, and the PL treatment was sown with 27.5 kg ha⁻¹ perennial ryegrass, 5 kg ha⁻¹

Table 1. Comparison of milk and herbage production from grass-white clover (WC) and grass-white clover-plantain (PL) swards

	WC	PL	S.E.	P-value
Milk yield (kg cow ⁻¹ day ⁻¹)	20.3	21.0	0.53	NS
Milk fat (g kg ⁻¹)	5.41	5.32	0.11	NS
Milk protein (g kg ⁻¹)	3.79	3.75	0.05	NS
Milk solids yield (kg cow ⁻¹ day ⁻¹)	1.84	1.86	0.04	NS
Cumulative milk solids yield (kg cow ⁻¹)	336	340	8.78	NS
Cumulative herbage production (kg DM ha ⁻¹)	11 341	11 356	275.27	NS

white clover and 2.5 kg ha⁻¹ plantain. Thirty-four spring calving dairy cows were selected and were balanced based on parity, calving date and pre-experimental milk production gathered during the 3 weeks prior to commencement of the study and were randomly allocated to one of the two treatment groups ($n = 17$). Both treatments were stocked at 3 cows per hectare in a closed farmlet system. Cows were allocated a daily herbage allowance of 17 kg DM ha⁻¹ and a concentrate allocation of 1–2 kg per cow per day. Swards were rotationally grazed, typically on a 21-day rotation when a target pre-grazing yield of 1400 kg DM ha⁻¹ was reached. The target post-grazing height was 4 cm. Both treatments received equal amounts of nitrogen fertiliser of 180 kg ha⁻¹ year⁻¹. Prior to each grazing event pre-grazing herbage mass was measured by harvesting two strips from each paddock using an Etesia mower. Cumulative herbage mass was recorded and calculated using PastureBase, an online grass management tool. Sward species composition was recorded prior to each grazing using a method described by Egan *et al.* (2018). Milk yield was recorded daily (Dairymaster, Causeway, Co. Kerry, Ireland) and milk composition was measured twice weekly using MilkoScan 203 (Foss Electric, Hillerød, Denmark). Data were analysed in SAS using Proc Mixed. Grassland analysis was conducted with fixed factors treatment, rotation, year, and associated factors. Milk production was analysed using treatment, week and year as fixed factors, with cow a random factor.

Results and discussion

The annual average plantain content for the PL treatment during 2023 and 2024 was 51.5 ± 5.5 and $34.4 \pm 4.5\%$, respectively. In 2024 plantain content was more variable between paddocks ranging from 26.9 to 55.9%, compared to 33.4–51.9% in 2023. White clover content was $9.4 \pm 2.1\%$ for the PL treatment while the WC treatment had a higher white clover content of $16.8 \pm 2.0\%$ during the experimental period. Weed content for both treatments was 3%. Perennial ryegrass-white clover swards are persistent and typically remain in situ for 12 years as part of a reseeding programme (O'Donovan *et al.*, 2017). Potential decreases in plantain content with associated increases in weed content lessen the viability of plantain swards in the long term. Total pasture production was similar for both treatments at 11 341 and 11 356 kg DM ha⁻¹ year⁻¹ for the WC and PL swards, respectively. Surprisingly, although white clover content was higher in both years for the WC treatment, this difference in sward composition did not result in extra herbage production. Hearn *et al.* (2024) showed that the main driver of productivity of these diverse swards was reliant on the presence of white clover; however, as neither sward averaged over the recommended 20% clover content for the year (Egan *et al.*, 2018), differences in additional nitrogen available to the WC sward were perhaps minimal.

Daily milk yield did not differ between swards as shown in Table 1. The PL treatment had a higher daily yield throughout the experimental period, but the higher constituents achieved with the WC treatment meant there was no difference in cumulative milk solids production over the course of the

experiment. Similarly, Navarrete *et al.* (2018) found no difference in milk production with the addition of plantain to perennial ryegrass-clover swards in a 9-month study. However milk production increased from cows grazing swards containing plantain as reported by Jezequel *et al.* (2024) in a three-year farm systems under similar climatic conditions to this study. In a meta-analysis by Nguyen *et al.* (2022) increased milk yield in late lactation from plantain containing swards was attributed to increased digestibility of the sward. Feed intake and sward nutritive quality measurements were obtained during experimentation but have not been analysed to date.

Conclusion

No differences in milk or herbage production were found over the two-year experimental period when comparing WC and PL swards of the same age under intensive grazing. Additional years of experimentation are required to further quantify how changing sward species contribution could affect herbage and milk production. This study shows positive effects of maintaining herbage production and animal performance with the inclusion of plantain into perennial ryegrass-white clover swards; however, the persistence of plantain within the sward remains the biggest challenge moving forward. The results indicate that if proposed environmental benefits from greater species diversity are achieved without reducing animal/herbage production, then this represents a net benefit for grassland systems. Further investigations are required to assess such effects in the longer term.

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Grazing as a sustainable management option: a case study of two Northern Italian livestock farms

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Abstract

The use of agricultural land as meadows is widespread in Italian dairy farms although grazing is rarely practised. Two dairy farms in Northern Italy, A (3.58 Livestock Unit, LSU ha⁻¹) and B (1.77 LSU ha⁻¹) were evaluated for grass productivity and feasibility of grazing for heifers and dry cows. A life cycle assessment analysis was conducted to quantify environmental impact. Grass production was 4.83 ± 1.52 t Dry Matter (DM) ha⁻¹ for A and 3.92 ± 0.48 t DM ha⁻¹ for B. Grass quality, assessed with NIR spectroscopy, showed for farm B higher protein (13.9 ± 1.79 g kg⁻¹ vs. 12.3 ± 1.83 g kg⁻¹ on DM) and lower NDF (43.4 ± 3.34 g kg⁻¹ vs. 46.7 ± 2.70 g kg⁻¹, on DM). Farm enteric methane emissions increased with grazing ($+3.85$ g kg⁻¹, average value of the two farms), due to greater energy expenditure for animal movement. The environmental impact of the farms did not show differences with grazing, even if a difference was observed in the impact of the diets (as kg CO₂ eq annual ration⁻¹). In farm A emissions for heifers and dry cows decreased from 639 to 462 and from 1040 to 752 respectively. In farm B emissions for heifers and dry cows dropped from 300 to 214, and 339 to 233, respectively. From an environmental standpoint, grazing is a viable option with promising results.

Keywords: dairy cow, grass quality, grazing, life cycle assessment, methane emission

Introduction

The presence of grasslands within Italian dairy farms, intensive and extensive, is extremely common. These meadows are often used solely for fodder production but not for direct animal grazing. The main objective of this study is to assess, through scenario analysis, how the inclusion or exclusion of grazing affects the environmental impact of milk production.

Materials and methods

The study was conducted from March to September 2022 on two dairy farms in Lombardy, Northern Italy, with contrasting management systems (Table 1).

Farm A has a larger herd, higher milk yield, and no grazing. Farm B produces less milk and practises eight-month grazing (March–October) for heifers and dry cows. During the grazing season, meadows were sampled on both farms to assess biomass yield through field measurements, and samples analysed by NIR spectroscopy to assess forage quality. Grazing intake was estimated using the Vasquez-Smith (2000) formula. Data were collected to a standardised template, covering housing, milk production, and cropping, to conduct a Life Cycle Assessment. Environmental impacts were assessed with SimaPro (EF 3.1, v1.01), simulating current scenario: no grazing for Farm A, eight months grazing for Farm B, and the reversed scenario. Although the simulation involved only non-lactating animals, the environmental impact was expressed per kg of FPCM produced at farm level. Diets for heifers and dry

Table 1. Main characteristics of the two farms involved in the study

	Unit	Farm A	Farm B
Dry cows	number	93	20
Heifers	number	332	50
Farm Livestock Units	LSU	975	221
Farm Livestock Units per UAA	LSU ha ⁻¹	3.58	1.77
Breed	name	Holstein	Fleckvieh
Milk production	kg FPCM day ⁻¹	29.4	16.2
UAA	Ha	270	134
Grassland	% on UAA	40	55
Grazing period	Months year ⁻¹	0	8
Animal category grazing	category	No grazing	Heifers and dry cows

FPCM, fat and protein corrected m; UAA, utilised agricultural area.

Table 2. Average annual values of meadow nutritional characteristics assessed using NIR spectroscopy.

	DM (g kg ⁻¹ on fresh matter)	CP (g kg ⁻¹ on DM)	NDF (g kg ⁻¹ on DM)	ADF (g kg ⁻¹ on DM)	Ash (g kg ⁻¹ on DM)
Farm A	22.0 ± 2.45	12.3 ± 1.83	46.7 ± 2.70	24.2 ± 2.87	7.28 ± 1.54
Farm B	19.9 ± 2.42	13.9 ± 1.79	43.4 ± 3.34	20.3 ± 2.98	7.56 ± 1.53

cows were based on measured pasture yield and quality and evaluated in CPM Dairy for nutritional balance and nitrogen and phosphorus excretion.

Results and discussion

Table 2 shows the characteristics of the meadows used in ration simulations. Grass on DM from Farm B had a higher protein content ($13.9 \pm 1.79\%$ vs. $12.3 \pm 1.83\%$ on Farm A) and a lower NDF concentration ($43.4 \pm 3.34\%$ vs. $46.7 \pm 2.70\%$ on Farm A). Average Farm A biomass production was 4.83 ± 1.52 t DM ha⁻¹ year⁻¹ and 3.92 ± 0.48 t DM ha⁻¹ year⁻¹ on Farm B.

Table 3 reports annual feed intake per animal across scenarios for both farms. Rations were unchanged, except for replacing hay and meadow silage with fresh grass from direct grazing. All other feed components were supplied when animals returned to the barn.

The environmental impact associated with the milk production was similar across both farms and scenarios. For Greenhouse Gas emissions, Farm A reported 1.97 kg CO₂ eq kg FPCM⁻¹, calculated following International Dairy Federation (2010), in the indoor scenario and 1.98 kg in the pasture scenario; Farm B showed a slight decrease from 2.23 to 2.20 kg CO₂ eq kg FPCM⁻¹ with grazing. Biogenic climate change impacts also showed minimal variation (Farm A: 0.93 kg CO₂ eq kg FPCM⁻¹ indoor vs. 0.94 pasture; Farm B: 1.44 kg CO₂ eq kg FPCM⁻¹ in both). These values, referred to FPCM, remained stable because only non-lactating animals were simulated, while total milk production of the herd was assumed constant across scenarios. The main contributor to biogenic climate change is enteric methane (Opio *et al.*, 2013), which increased with grazing due to higher feed intake needed to meet increased energy demands from physical activity (O'Neill *et al.*, 2011). Emissions were indeed higher in the grazing scenarios: +4.0% for Farm A and +3.5 % for Farm B. A further contributing factor is the different ration composition, with more structural fibre and lower digestibility, leading to increased acetic acid production in the rumen, a methane precursor (Moraes *et al.*, 2014). A marked difference was observed in the climate impact of the diets (kg CO₂ eq annual ration⁻¹

Table 3. Annual feed supply and daily dry matter intake (DMI) estimates per animal in the two feeding scenarios

		Feed	Production	Ration 12 months indoor (kg DM year ⁻¹ head ⁻¹)	Ration 4 months indoor+8 months pasture (kg DM year ⁻¹ head ⁻¹)
Farm A	Dry cow	Corn meal	Imported	642	642
		Mineral integration	Imported	143	143
		Meadow hay/pasture	Farm produced	2505	2441
		Sorghum silage	Farm produced	1314	1314
		DMI (kg day ⁻¹ head ⁻¹)		12.4	11.7
	Heifers	Corn meal	Imported	321	321
		Soya meal	Imported	64.0	64.0
		Meadow hay / pasture	Farm produced	1606	1659
		Corn silage	Farm produced	1369	1369
		DMI (kg day ⁻¹ head ⁻¹)		9.90	10.1
Farm B	Dry cow	Meadow silage	Farm produced	657	876
		Meadow silage/pasture	Farm produced	1971	2847
		DMI (kg day ⁻¹ head ⁻¹)		9.60	12.6
	Heifers	Meadow silage	Farm produced	438	803
		Meadow silage/pasture	Farm produced	2190	2433
		DMI (kg day ⁻¹ head ⁻¹)		9.60	10.6

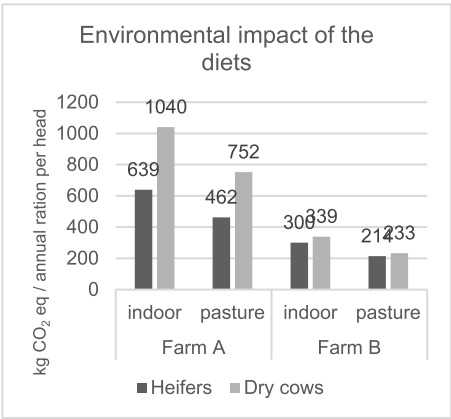


Figure 1. Environmental impact of diets under the two scenarios.

head⁻¹), as shown in Figure 1, with generally lower values in the grazing scenarios. This likely reflects the lower management intensity of pastures compared to tilled forage systems.

Conclusion

Grazing emerges as a promising management option with beneficial environmental effects, especially regarding dietary impact. Including pasture in the feeding strategy reduced the environmental footprint of diets without compromising the overall farm impact per unit of milk. Both farms already produced forage on-site, enhancing system circularity; an effect that could be even greater in farms relying on external forage. Better use of existing grasslands, seasonally or permanently, can thus be a viable, sustainable strategy for dairy operations

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Application of Near Infrared Reflectance Spectroscopy for evaluation of fatty acids content in herbage forage

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Abstract

Forages are a cost-effective feed for ruminants and provide an important natural source of fatty acids (FA). The FA profile determines the quality of derived food products. The FA profile in forages exhibits high variability due to factors such as species, growth stage and management practices. Near infrared reflectance spectroscopy (NIRS) was evaluated as an alternative method to gas chromatography for the quantitative analysis of FA in herbage samples grown in Galicia (NW Spain). Spectral data (400 to 2500 nm) were collected from a sample set ($n = 375$) comprising: *Trifolium incarnatum*, *T. michelianum*, *T. resupinatum*, *T. vesiculosum*, *T. pratense*, *T. repens*, *Medicago sativa*, *Ornithopus sativus*, *Lolium perenne* and a mixed grass-legume sward. Chromatographic data was regressed on spectral data to develop NIRS calibrations for FA composition (C12:0, C14:0, C15:0, C16:0, C16:1, C17:0, C18:0, C18:1n9c, C18:2n6c, C20:0, C18:3n6, C20:1, C18:3n3, C21:0, C22:0, C22:1n9, C20:3n3, C20:4n6 and C24:0) and sum of total, saturated, unsaturated and polyunsaturated FA. NIRS calibrations obtained were deemed suitable for predicting major FA components and FA groups, with coefficient of determination in cross-validation (R^2) ranged from 0.70 to 0.90 (RPD: 2.01 to 3.26). In conclusion, NIRS analysis proves to be a useful tool for the FA profile prediction in the forage species studied.

Keywords: feed quality, forage, lipids, NIRS

Introduction

Herbage forages provide an important natural source of fatty acids (FA) in ruminant diets whether used as cut fodder or grazed pastures. Dairy cows' feeding has an influence on the FA profile of milk, being known that higher proportions of herbage in dairy cow diets increases the content of polyunsaturated fatty acid (PUFA) in milk (Dewhurst *et al.*, 2006). Consequently, the role of FA in dairy cow nutrition, particularly PUFA, has gained increasing attention due to their impact on milk quality and therefore in human health (Chen and Liu, 2020).

In recent years, consumers are increasingly demanding milk from herbage-based systems, as these enhance beneficial FA profiles, supporting the production of dairy products with healthier FA composition (Alothman *et al.*, 2019). However, the FA concentrations in forages vary considerably depending on species, growth stage, and management practices. This variability necessitates accurate quantification methods to develop reliable and efficient tools that can support research activities, optimize herbage management decisions, and facilitate breeding programmes for improved herbage FA profile.

Near Infrared Reflectance Spectroscopy (NIRS) offers a fast and high-throughput alternative to reference methods like gas chromatography (GC) for quantitative FA analysis in forages. Moreover, NIRS

was noted as a unique green analytical technology (Czaja and Engelsens, 2025), aligning with sustainable practices by minimizing chemical reagent use and reducing waste which highlights its eco-friendly application. As an indirect technique, NIRS requires calibration development using reference GC data. The objective of this study was to evaluate the feasibility of NIRS for predicting FA profile of herbage from Galicia (NW Spain).

Materials and methods

The samples were obtained from multiple experiments conducted at CIAM (Galicia, NW Spain) during two years. This study analysed herbage samples of whole plant ($n = 292$) and morphological fractions of legumes ($n = 83$) from winter crops and swards. The sample set ($n = 375$) were: *Trifolium incarnatum*, *T. michelianum*, *T. resupinatum*, *T. vesiculosum*, *T. pratense*, *T. repens*, *Medicago sativa*, *Ornithopus sativus*, *Lolium perenne* and mixed grass-legume sward. The samples were harvested at different dates during spring under varying management systems: for grazing and for ensiling (direct cut and wilted).

Spectral data of samples set ($n = 375$) were recorded from samples dried and ground through a 1-mm sieve in reflectance mode in the wavelength range from 400 to 2500 nm, using the FOSS NIRSystem 6500. Spectral data collection and chemometric treatment of data were performed using WinISI II software.

FA of the samples were extracted using a modified version of the direct transesterification method developed by Sukhija and Palmquist (1988). The resulting FA methyl esters were detected and quantified by GC coupled with a flame ionization detector, with all data were reported as g (kg dry matter)⁻¹.

The NIRS calibrations were performed by modified partial least squares (MPLS) regression between spectral data and reference values by GC, applying the cross-validation procedure. In order to develop the most accurate calibration models, different combinations of scatter corrections and mathematical treatments were tested. The best-fit calibrations were selected based on standard error (SECV) and the coefficient of determination (R^2) of cross-validation. Additionally, to evaluate calibrations accuracy, we calculated the ratio of performance to deviation (RPD) (Williams, 2001).

Results and discussion

The reference values and NIRS calibration statistics are presented in Table 1, including only parameters with RPD values >2.0 , indicating at least acceptable predictive performance (Williams, 2001). The FA dataset exhibited wide variation, which enhanced the robustness and applicability of NIRS calibrations.

NIRS calibrations obtained were deemed suitable for predicting major FA components and FA groups, with coefficient of determination in cross-validation (R^2) ranged from 0.70 to 0.90 (RPD: 2.01 to 3.26), except for C14:0, C15:0, C16:1, C17:0, C20:3n3, C20:4n6 and C22:0 (R^2 : 0.72–0.78; RPD: 1.71–1.97), and for C22:1n9, C20:1 and C21:0 ($R^2 < 0.50$; RPD < 1.50). NIRS calibrations showed reduced accuracy for minor FAs, likely due to their lower prevalence. The NIRS calibration for C18:3n3 (α -linolenic acid) showed excellent predictive performance, with an RPD value of 3.26, this FA is highly variable in herbage.

Table 1. The reference values ($\text{g (kg dry matter)}^{-1}$) and NIRS statistics of the calibrations for the prediction of fatty acid composition of herbage

Constituent	<i>n</i>	Minimum	Maximum	SD	SECV	R^2	RPD
C12:0	363	0.002	0.230	0.053	0.025	0.77	2.10
C16:0	364	0.057	3.700	0.667	0.295	0.80	2.26
C18:0	371	0.008	0.580	0.119	0.058	0.79	2.05
C18:1n9c	339	0.002	1.133	0.195	0.080	0.83	2.43
C18:2n6c	345	0.096	3.890	0.744	0.370	0.74	2.01
C18:3n3	358	0.366	18.034	3.146	0.967	0.90	3.26
C18:3n6	340	0.002	0.154	0.033	0.013	0.86	2.57
C24:0	373	0.002	0.338	0.072	0.035	0.77	2.05
UFA	351	1.322	20.825	3.560	1.335	0.86	2.67
SFA	367	0.301	5.400	1.009	0.484	0.77	2.08
PUFA	341	1.122	20.572	3.588	1.325	0.86	2.71
TFA	358	1.873	25.784	4.497	1.608	0.85	2.80

SECV, standard error of cross-validation; R^2 , coefficient of determination in calibration and cross-validation; RPD, SD/SECV; FA, fatty acid; UFA, unsaturated FA; SFA, saturated; PUFA, polyunsaturated FA; TFA, total FA.

In general, our results exhibited lower predictive accuracy than those obtained by Foster et al. (2006), potentially attributable to their use of freeze-dried herbage samples, whereas in this work samples were oven-dried at 40°C. The spectral regions key to the NIRS calibrations corresponded to the C–H functional groups, particularly within 2200–2400 nm^{-1} . This range represents second-overtone C–H vibrations in aliphatic chains (–CH₂–, –CH₃ groups) in fatty acid molecules (Shenk *et al.*, 2008).

Conclusion

NIRS spectroscopy demonstrates potential as a rapid method for assessing the fatty acids profile of herbage forage. NIRS could be successfully implemented as a large-scale tool in order to evaluate the fatty acids composition of forage herbage, being an alternative to traditional gas chromatography for fatty acid analysis.

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Methane production potential and phenolic compounds of herbage from alpine summer pastures

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Abstract

Forage from grasslands managed at low intensity, such as alpine summer pastures, often has high concentrations of NDF, which has been shown to increase enteric methane production of ruminants. However, these grasslands are also very diverse and may contain plants rich in phenolic compounds, which can decrease enteric methane production and might partly offset the effect of high NDF concentrations. We quantified methane production potential (MPP), NDF and phenolic compounds (TEP: total extractable phenols, CT: condensed tannins) in the first regrowth of a total of 45 alpine summer pastures in two years. Pastures were characterised by plant community type based on characteristic altitude ('high', 'low') and soil fertility ('fertile', 'marginal'). Plant community had a significant effect on NDF, and, in interaction with year, on TEP, but not on CT or MPP. MPP decreased with increasing TEP concentrations, while NDF was not significantly related to MPP. The results highlight the variability of chemical composition and MPP in summer pastures, which can partly be explained by site conditions, and imply that high fibre concentrations may not necessarily increase enteric methane emissions of animals grazing these pastures.

Keywords: enteric methane production, mountain grassland, phenols, tannins

Introduction

Alpine summer pastures are important habitats of high biodiversity, possess high cultural value and can be an important forage resource. However, extensively managed grasslands often produce forage of lower digestibility and higher fibre concentrations than more intensively managed ones. Low digestibility, and specifically, high concentrations of neutral detergent fibre (NDF), are associated with higher enteric methane production (Eugène *et al.*, 2021). This could unfavourably contribute to greenhouse gas budgets of ruminant livestock systems. On the other hand, several species of alpine summer pastures have been shown to contain relevant concentrations of tannins and other phenols (Jayanegara *et al.*, 2011), which have the potential to reduce methanogenesis in the rumen (Eugène *et al.*, 2021; Jayanegara *et al.*, 2009). Against this background, this study aimed to survey the variability of NDF and phenolic compound concentrations in biomass from a wide range of grasslands on alpine summer pastures in Switzerland, and to establish the relationship between these and the potential for enteric methane production, determined with an *in vitro* method.

Methods

Forage samples were sourced from a network of summer pastures in different regions in Switzerland. Within this network, which was established in 2022, botanical composition, biomass yield and forage quality are quantified at four sites per region. The four sites represent regional plant communities differing in combinations of characteristic altitude ('low', 'high') and soil fertility ('fertile', 'marginal').

Table 1. Effect of year (Y) and plant community type (PC) on methane production potential (MPP) and concentrations of neutral detergent fibre (NDF), total extractable phenols (TEP) and condensed tannins (CT, values logit-transformed)

Response	Estimated mean (SE)								Wald test <i>p</i> values		
	2022				2023				PC	Y	PC × Y
	LF	LM	HF	HM	LF	LM	HF	HM			
MPP (μg ml ⁻¹)	116.4(1.73)	116.1(1.81)	118.5(1.69)	121.1(1.72)	113.3(1.53)	110.9(1.53)	112.8(1.59)	113.7(1.72)	0.2834	<0.0001	0.2632
NDF (g kg ⁻¹)	510(23.9)	550(25.2)	530(22.8)	664(22.8)	488(20.3)	521(20.3)	529(21.0)	583(22.8)	<0.0001	0.0166	0.1857
TEP (g kg ⁻¹)	21.7(2.32)	22.0(2.45)	18.0(2.22)	13.3(2.24)	26.1(1.96)	31.3(1.96)	25.3(2.05)	27.3(2.24)	0.0408	<0.0001	0.0393
logit(CT) (g g ⁻¹)	-5.36(0.22)	-5.29(0.23)	-5.67(0.21)	-5.43(0.21)	-4.90(0.18)	-4.50(0.18)	-4.89(0.19)	-4.28(0.21)	0.1458	<0.0001	0.1678

Plant community types: LF–low-fertile, LM–low-marginal, HF–high-fertile, HM–high-marginal.

This study included spring-grown biomass of 45 sites from twelve regions (33 sites in 2022 and 44 sites in 2023). Biomass NDF concentrations were determined using near-infrared spectroscopy, total extractable phenols (TEP, in gallic-acid equivalents) with a modified Folin–Ciocalteu method, condensed tannins (CT, in leucocyanidin equivalents) with the butanol-HCL-iron method, and methane production potential (MPP, as methane produced per unit gas) with an *in vitro* method, the Hohenheim Gas Test, coupled with gas chromatography, all carried out as described in Jayanegara *et al.* (2011).

The effect of plant community type on MPP, NDF, TEP, and logit-transformed values of CT, was assessed using mixed effects models with plant community type as main effect and site nested in summer pasture area as random effects. To take into account the correlation between NDF and TEP ($r = -0.61$), NDF and logit(CT) ($r = -0.29$), as well as TEP and logit(CT) ($r = -0.68$), MPP was predicted as response of the fixed effects NDF, TEP, logit(CT) and year, and the random effects site nested in summer pasture area, incorporating correlated error structures to account for correlations between NDF and TEP ($r = 0.61$), NDF and logit(CT) ($r = -0.29$) and TEP and logit(CT) ($r = 0.68$).

Results and discussion

Lower values of NDF, CT and MPP and higher values of TEP were found in 2023 compared to 2022 (Table 1). Across years, NDF concentrations were higher in ‘high-fertile’ than in other plant communities. TEP concentrations were higher in ‘low-marginal’ communities than in all other communities in 2022 and higher than in ‘low-fertile’ and ‘high-fertile’ communities in 2023. Together, the interactive effect of plant community type and year explained 44 and 48% of the observed variability in NDF and TEP, respectively. Neither CT nor MMP were affected by plant community type.

MPP decreased with increasing TEP ($p < 0.0001$), but contrary to expectations, neither NDF ($p = 0.18$) nor CT ($p = 0.77$) effects on MPP were significant (Figure 1). While widely used equations predicting MPP per unit digestible organic matter include a positive relationship between NDF and MPP, such a relationship was also weak or absent in some *in vivo* studies (e.g. Pinares-Patiño *et al.*, 2003) as well as other *in vitro* studies (e.g. Raju *et al.* 2011). Among phenolic compounds, it was TEP, and not the CT fraction, that was negatively related to MPP in this study, in line with *in vitro* results by Jayanegara *et al.* (2009).

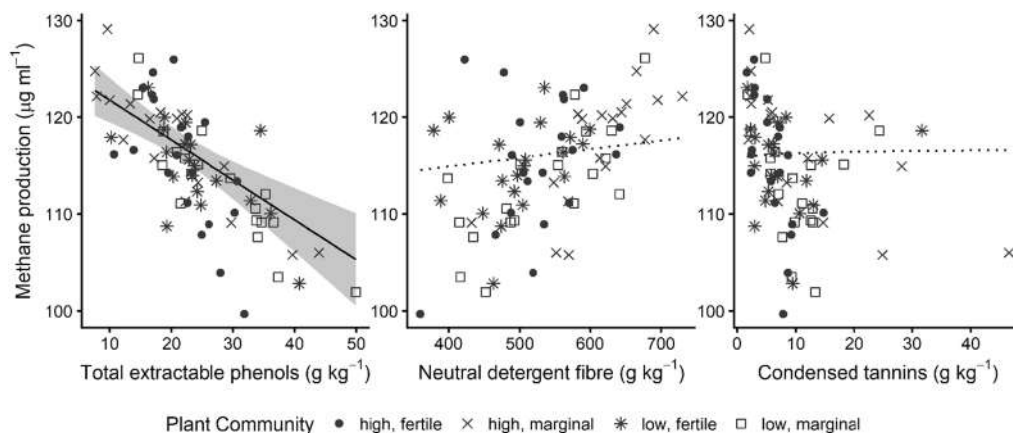


Figure 1. Methane production potential as response to concentration of total extractable phenols, neutral detergent fibre and condensed tannins. Solid line and shaded area: population-level prediction with 95% confidence intervals, averaged over two years. Dotted lines: population-level prediction for non-significant effects.

Conclusion

Chemical composition and MPP varied considerably among grasslands samples from 45 summer pastures. MPP decreased by 14% over the observed gradient of TEP concentrations but, in spite of significant relationships of plant community types with chemical composition, predictability of MPP from plant community type, based on the classification used, was low. On the other hand, the weak relationship between MPP and NDF means that greater plant maturity, which is characteristic of low-intensity grazing system, may not necessarily increase MPP.

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Pasture improvement through endozoochory in southern Portugal

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Abstract

Seed dispersal via the digestive tract of grazing animals (endozoochory) can augment the soil seed bank and enhance pasture composition. This study evaluated biomass production at Herdade dos Cordeiros (Vila Viçosa, Alentejo, Portugal) across three plots: Cordeiros (sown in 2018/19 with a legume-rich mixture), Cabriz (improved through endozoochory from Cordeiros), and Carvalhais (a newly established plot intended for enhancement via endozoochory). Biomass was measured using eight 1 m² grazing-exclusion cages per plot, with three harvest dates: 1 December 2021, 1 March 2022, and 1 June 2022. Dry weights (oven-dried at 105°C for 24 h) were recorded, and botanical composition was categorised into grasses, legumes, and other families. Total biomass was greater in the improved plots, with 1946 kg DM ha⁻¹ in Cordeiros and 2323 kg DM ha⁻¹ in Cabriz, compared with 1239 kg DM ha⁻¹ in Carvalhais. Across the three harvests, the proportion of legumes was highest in Cordeiros (15%), followed by Cabriz (13%) and Carvalhais (12%). Cordeiros also showed a greater proportion of other botanical families. These findings suggest that endozoochory can facilitate pasture improvement, although strategic management of grazing patterns is required to optimise seed dispersal and establishment.

Keywords: biomass production, grazing management, legumes, seed dispersal

Introduction

The principal processes determining the success of grassland restoration are plant establishment and seed incorporation into the soil (de Azevedo *et al.*, 2024). Kaiser (2025) describes five stages of the endozoochorous dispersal process and its influencing factors, and notes that cattle dung possesses several advantageous properties—such as suitable consistency, large surface area, and comparatively low mortality rates of contained seeds—that make targeted seed transfer via grazing cattle promising for practical application. Digestive processes can stimulate the germination of ingested (but undigested) seeds through scarification and the removal of chemical compounds that inhibit germination (Lisboa, 2017). Germination success of ingested seeds tends to be higher in legumes, owing to their hard seed coats, than in grasses (Stiegler *et al.*, 2021). The objective of this study was to evaluate the role of cattle in seed dispersal from a sown dryland permanent pasture, and to assess its effect on the floristic composition of three plots at different transitional stages from natural to improved pasture.

Materials and methods

The study was carried out on three contiguous plots belonging to Sociedade Agrícola dos Cordeiros in Vila Viçosa, Alentejo, Portugal: (i) *Cordeiros* (45.1 ha), sown in 2018/2019 with a legume-rich mixture comprising resupinate clover (*Trifolium resupinatum*), balansa clover (*Trifolium michelianum*), arrowleaf clover (*Trifolium vesiculosum*), crimson clover (*Trifolium incarnatum*), serradella (*Ornithopus*

sativus), and annual ryegrass (*Lolium multiflorum*). It was grazed for two days after seed maturation in summer 2021 by Alentejana cattle; (ii) *Cabriz* (35.4 ha), a natural pasture included in the farm's rotational grazing system for several years, prior to the present study. It was grazed by the same cattle for 42 days following their removal from the *Cordeiros* plot; (iii) *Carvalhais* (58.8 ha), a natural pasture incorporated into the farm during the study year. It was grazed by the same cattle for 59 days after leaving *Cabriz*.

The cows' route was decided by the owner and could not be changed by the research team.

Pasture growth was measured using eight randomly distributed mobile grazing-exclusion cages (1 m²) per plot. Within Carvalhais plot, in addition to the eight mobile grazing-exclusion cages of Carvalhais plot (*Carvalhais-MC*), eight fixed cages (*Carvalhais FC*) were installed to assess pasture herbage growth in the absence of grazing, as cattle had never previously entered this plot.

Three harvests were conducted: 1 December 2021 (autumn production), 1 March 2022 (winter production), and 1 June 2022 (spring production). After each harvest, mobile cages were moved to an adjacent area, and the new cage location was pre-cut to simulate grazing. In *Carvalhais FC*, the cages remained in place throughout the study.

Harvested biomass was botanically separated into grasses, legumes, and other families, then oven-dried at 105°C for 24 h to determine dry matter (DM) production. Legume species present in each plot were identified in the June harvest, coinciding with the flowering of the species, which enabled their correct identification.

Data were subjected to one-way analysis of variance (ANOVA). Where significant differences were detected ($p \leq 0.05$), means were separated using Duncan's multiple range test.

Results and discussion

The most productive plots (Figure 1) were *Cordeiros* and *Cabriz*, while the least productive was *Carvalhais*, which showed similar results for both mobile and fixed cages. Differences in biomass production between the two groups of plots (*Cordeiros* and *Cabriz* versus *Carvalhais*) were highly significant, but only in spring.

Although *Cordeiros* had been improved with a legume-rich mixture, after three years it did not contain significantly more legumes than the other plots (Figure 2). However, it exhibited a higher proportion of species from other botanical families, generally associated with lower nutritional value.

Among the legumes sown in *Cordeiros*, *Trifolium vesiculosum* was particularly noteworthy, persisting three years after sowing and also being identified in *Cabriz*, in some cases in large numbers, suggesting likely endozoochorous dispersal from *Cordeiros*. This species was absent from *Carvalhais*, which received only a single movement of cattle from *Cabriz*; this may not have been sufficient for effective seed dispersal. According to Schaedler *et al.* (2021), maximum germination of ingested seeds occurs on the second and third days after ingestion, whereas seeds remaining in the digestive tract for more than 70 h have a reduced probability of germination. It is therefore suggested that cattle graze for one to two days in the plot dominated by the species targeted for dispersal, followed by one to two days in the plot intended for floristic enhancement.

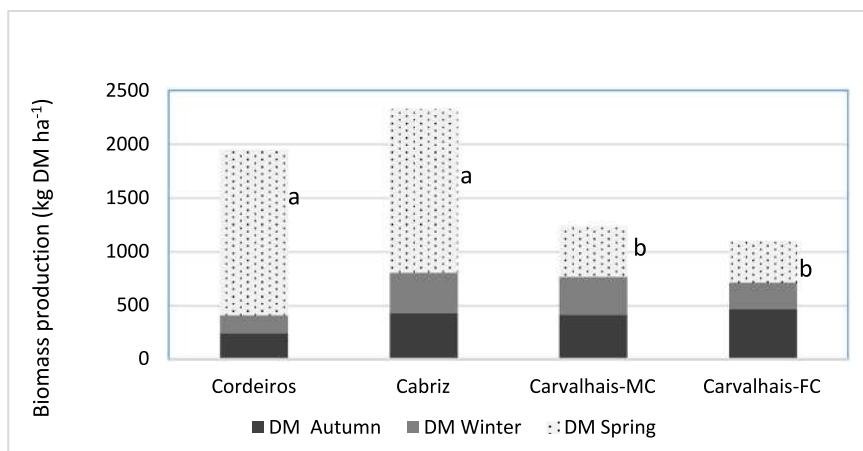


Figure 1. Total biomass production (kg DM ha^{-1}) in Cordeiros, Cabriz, Carvalhais-MC and Carvalhais-FC plots, during autumn, winter and spring. The differences were significant only for DM spring.

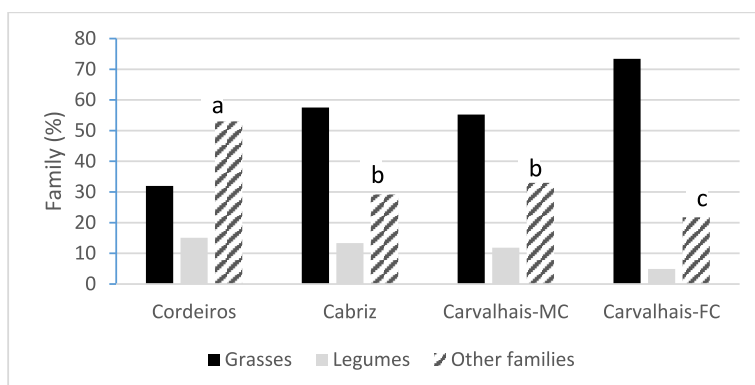


Figure 2. Average annual proportion (%) of each botanical family in Cordeiros, Cabriz, Carvalhais-MC and Carvalhais-FC plots. Significant differences were observed only for other families.

Conclusion

Endozoochory can enhance dryland pastures by transferring desirable species, such as *Trifolium vesiculosum*, between plots. Although the legume-rich mixture sown in *Cordeiros* did not significantly increase legume proportion after three years, dispersal to *Cabriz* highlights the potential of this natural process. Strategically managing livestock movements—grazing for one to two days in the seed source plot after seed maturation, followed by grazing in the target plot—can optimise seed dispersal. This approach offers a low-cost, environmentally sustainable alternative to mechanical reseeding, reducing fuel consumption, soil disturbance, and seed requirements. Further research should evaluate the long-term persistence of dispersed species, their contribution to pasture productivity and nutritional value, and the economic viability of targeted endozoochory as a improvement strategy.

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Effects of cutting management of annual rainfed swards on regrowth and biomass production in southern Portugal

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Abstract

Grazing management strongly influences pasture productivity, persistence, and biomass availability for livestock. This study evaluated the effects of cutting management (stubble height and cutting moment) on regrowth, biomass production, forage availability (quantity removed), and biomass contribution to the soil in an annual-species sown pasture in Alentejo, Portugal, during 2024/25. The moment of cutting was determined by the herbage height (L, 5 cm; M, 7.5 cm; and H, 10 cm), combined with two stubble heights (L, 3 cm; and H, 6 cm), resulting in five treatments: LL, ML, MH, HL, and HH. The LL was considered as an approach to continuous grazing, while the others rotational grazing. A higher stubble residue (6 cm) promoted faster regrowth and greater biomass retention on the soil surface but reduces forage availability for grazing. Overall pasture productivity and forage availability were highest when herbage growth lasted until 10 cm height. Maintaining a higher stubble (greater leaf area) enhanced regrowth and might provide environmental benefits such as improved soil protection and soil organic matter. Conversely, a lower residue increased forage availability. Optimising the effects on soil protection with productivity, the HH treatment offered the most suitable compromise, albeit with a slight reduction in forage availability.

Keywords: grazing environmental benefits, grazing management rainfed annual swards, soil protection, sward stubble

Introduction

Grasslands are a crucial source of forage for livestock production (Bengtson *et al.*, 2019) and play an integral role in agricultural systems for animal husbandry (Chang *et al.*, 2021). They also contribute to rural employment and provide social benefits (Hadjigeorgiou *et al.*, 2005). Beyond their productive role, grasslands deliver important ecosystem services, including biodiversity conservation (Bengtson *et al.*, 2019; Hadjigeorgiou *et al.*, 2005), climate regulation (Bengtson *et al.*, 2019; Zhao *et al.*, 2020), air quality improvement, erosion control, carbon sequestration (Zhao *et al.*, 2020), and recreation (Hadjigeorgiou *et al.*, 2005). Consequently, grasslands fulfil environmental, social, and economic functions.

Optimising these functions requires efficient grassland management to maintain healthy and productive swards. This study aimed to assess how pre-cutting sward height and post-cutting residual height—parameters with both productive and environmental implications—affect grass regrowth, total biomass production, forage availability, residual biomass, and the number of harvests per cycle.

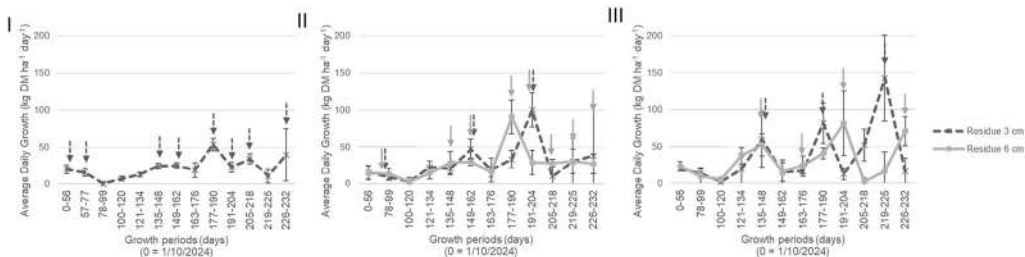


Figure 1. Average daily growth ($\text{kg DM ha}^{-1} \text{ day}^{-1}$). (I) LL treatment (stubble 3 cm); (II) ML (3 cm) and MH (6 cm); (III) HL (3 cm) and HH (6 cm). Arrows indicate the cutting dates.

Materials and methods

The trial was conducted in Crato ($39^{\circ}19'54.4''\text{N}$, $7^{\circ}31'04.8''\text{W}$), Alentejo, Portugal, from 1st October 2024 to the end of the 2024/2025 growing season (June 2025), on a sown pasture (2nd year after sowing, mixture of annual species: 75% grasses and 25% legumes). The meadow species were as follows: *Dactylis glomerata*, *Lolium multiflorum*, *Festuca arundinacea*, *Trifolium subterraneum*, *Trifolium resupinatum*, *Trifolium majus*, *Trifolium michelianum*, *Trifolium incarnatum*, *Trifolium vesiculosum*, *Medicago polymorpha*, *Medicago truncatula* and *Ornithopus sativus*. Three compressed sward heights at cutting—5 cm (low, L), 7.5 cm (medium, M), and 10 cm (high, H)—were combined with two stubble residue heights—3 cm (low, L) and 6 cm (high, H)—in five treatment combinations: LL, ML, MH, HL, and HH. Treatment LL was considered as one that approached continuous grazing (commonly adopted by local farmers) in contrast with the other treatments, considered to be closer to rotational grazing. The experimental design was a randomised complete block with four replicates per treatment. Each plot had a cutting effective area of 1 m^2 , plus a surrounding 0.37 m border.

Cutting was performed with a height-adjustable grass-cutting machine (Stihl RMA239). Harvested biomass was dried at 105°C to constant weight and expressed as kg DM ha^{-1} , considered as “forage available” for animal feed.

Sward height before cut and stubble height (after cut) were measured with a rising plate meter (RPM—Jenquip EC-09) and converted to biomass (kg DM ha^{-1}) using the regression model of Simões *et al.* (2024). RPM estimations were performed weekly, to determine the cutting moment. Harvested biomass and stubble residues were used to calculate average daily growth ($\text{kg DM ha}^{-1} \text{ day}^{-1}$). Total biomass production was obtained by summing residual stubble and “forage available” for animal feed.

Data were subjected to one-way analysis of variance (ANOVA). Where significant differences occurred ($p \leq 0.05$), means were separated using Duncan’s test.

Results and discussion

Average daily growth (Figure 1) showed that leaving a taller stubble residue (6 cm) enhanced regrowth rates. This was particularly evident following cuts at 162 and 148 days, when comparing ML *vs.* MH and HL *vs.* HH, respectively. It also reduced post-cutting growth decline, as observed after cuts at day 204 in ML and MH. These findings align with Smetham (1990), who suggested that regrowth is strongly dependent on the number of leaves retained.

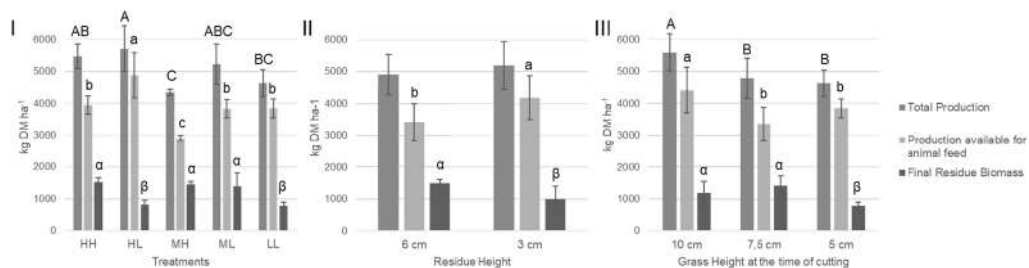


Figure 2. Total production, forage available for animal feed and final stubble residue biomass (kg DM ha⁻¹): (I) treatments, (II) residue height, and (III) height at time of cutting. Duncan's test results for total production (ABC), production available for animal feed (abc) and final residual biomass ($\alpha\beta$).

The number of cuts varied across treatments: LL and MH were cut eight times, HH four times, and ML and HL only three times. Longer recovery intervals in HH, HL, and ML may promote plant vigour and seed production (Carreira *et al.*, 2022).

In terms of biomass available for animal feed (Figure 2), HL achieved the highest value (4882 kg DM ha⁻¹), while total biomass was similar to HH (5710 kg DM ha⁻¹ vs. 5479 kg DM ha⁻¹, respectively). However, HH had a lower proportion of forage available for grazing (3950 kg DM ha⁻¹). MH recorded the lowest forage availability (2898 kg DM ha⁻¹).

Stubble residue height had a clear effect: a lower height (3 cm) produced significantly more available forage (4185 kg DM ha⁻¹), benefiting livestock feeding, whereas a taller residue (6 cm) increased biomass stubble on the soil surface (1487 kg DM ha⁻¹), which may support environmental functions such as soil protection and organic matter accumulation.

Moment of cutting also influenced yields: cutting at 10 cm resulted in the highest total biomass (5595 kg DM ha⁻¹) and forage availability (4416 kg DM ha⁻¹).

Conclusion

Maintaining a taller stubble (as under less intensive grazing) supports faster regrowth and provides environmental benefits, notably improved soil protection. In contrast, shorter stubbles (as occurs with more intensive grazing) enhance forage availability for animal feeding and number of cuts (longer period of grazing). The HH treatment may represent a compromise between productivity and environmental sustainability, although it slightly reduces immediate forage supply. This hypothesis should be tested in further studies.

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Co-innovation pathways in grassland-based livestock systems: a longitudinal analysis from Uruguay

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Abstract

In the Río de la Plata grasslands, beef and wool systems based on native grasslands are key to agricultural production, biodiversity conservation, and the sustainability of family farmers. However, these systems face challenges due to low productivity and income. Understanding their trajectories is essential for supporting viable strategies. This study analysed 10 livestock systems engaged in co-innovation processes in Uruguay (2012–2023), incorporating ecological intensification proposals in grassland and herd management. Changes in the application of strategic, tactical, and decision-support techniques were assessed using the Cow-Calf Management Techniques Index (CCMTI), and temporal trajectories were analysed for three key variables: beef production, trophic efficiency, and net income. Linear mixed models were applied, with year as a fixed effect and farm as a random effect with varying intercepts and slopes. Results showed significant improvements: CCMTI increased in all cases; beef production and trophic efficiency increased significantly ($p < 0.05$); while net income showed upward trends ($p < 0.10$). Findings suggest a more efficient use of pasture energy mediated by management decisions, highlighting co-innovation and ecological intensification as mechanisms to enhance productivity and efficiency.

Keywords: change trajectories, co-innovation, livestock systems, management practices, native grassland

Introduction

The Río de la Plata grasslands constitute one of the largest and most significant sub-humid temperate grassland ecosystems in the world. Livestock production based on cattle and sheep grazing is a core activity and one of the main ecosystem services provided by these grasslands (Modernel *et al.*, 2016). In addition, they support a large number of production units, particularly family-run farms, although these are generally characterized by low productivity and economic returns (Paparamborda *et al.*, 2023). Ecological intensification (Soca *et al.*, 2024), combined with co-innovation processes (Rossing *et al.*, 2010), has shown potential to reverse the situation of low productivity and profitability by redesigning production units and promoting participatory learning, with a central focus on monitoring forage allowance to adjust grazing intensity and the systemic application of strategic, tactical, and decision-support techniques (Soca *et al.*, 2024; Ruggia *et al.*, 2021; Paparamborda *et al.*, 2023). The objective of this study was to examine the trajectory changes of livestock systems that have incorporated technical innovations aimed at ecological intensification, with the goal of facilitating their scaling out. To this end, we applied an adaptation of the methodological approach of Martin *et al.* (2017), which includes: (1) the selection of key variables, and (2) the analysis of the temporal trajectories of these variables using linear and mixed regression models.

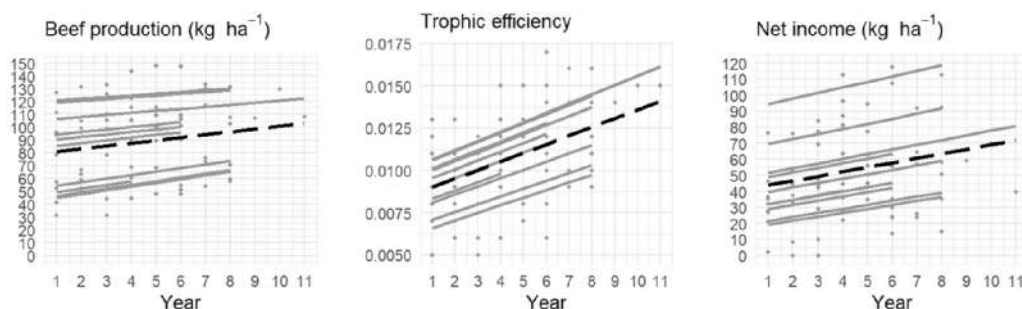


Figure 1. Individual trajectories and average trend of beef production per grazing area, trophic efficiency, and net income per total area for the 10 systems analysed. Grey lines represent individual cases; the black dashed line represents the average trend estimated by a linear mixed model with farms as a random effect.

Materials and methods

We analysed ten livestock systems from the Eastern Hills and Basaltic Ridge of Uruguay (2012–2023) that participated in three co-innovation projects. Each project included monthly visits by an agricultural advisor to reconstruct baseline (BL) economic–productive indicators and collect data during the co-innovation (CI) phase. The systems completed different co-innovation cycles: one with three cycles (since 2012), five with two (since 2017), and four with one (since 2020). Changes in the application of strategic, tactical, and decision-support techniques were assessed using the Cow-Calf Management Techniques Index (CCMTI; Paparamborda, 2017), and trajectories of three key variables were analysed: beef production per grazing area, trophic efficiency, and net income per total area. Variables were measured annually for each farm according to the number of cycles completed. The CCMTI ranges from 0 to 100, with higher values reflecting better implementation of management techniques. Linear mixed models (`lmer()`, `lme4`) were applied with calendar year as a fixed effect and farm as a random effect; analyses were performed in R (v4.4.2) and visualized with `ggplot2`.

Results and discussion

The results showed that the Cow-Calf Management Techniques Index increased in all cases when comparing the baseline with the co-innovation period. The trajectories of beef production per grazing area and trophic efficiency showed significant improvements ($p < 0.05$), while the trajectories of net income per total area showed an upward trend ($p < 0.10$) (Figure 1).

These outcomes were achieved without significant changes in resource use, as proposed by ecological intensification in livestock systems in the Campos region (Paparamborda *et al.*, 2023; Soca *et al.*, 2024): above-ground net primary productivity and stocking rates remained stable (with slight decreases in three cases), the area with improved pastures increased in only two cases, and production costs rose in only four out of ten cases. These changes appear to be related, at least in part, to the generalized increase in the CCMTI and the improvement or maintenance of trophic efficiency, suggesting a more efficient use of pasture energy mediated by strategic and tactical management decisions, as observed in research conducted at the plot scale in the Campos region (Soca *et al.*, 2024). Similar trends were reported by Ruggia *et al.* (2021) in co-innovating farms.

The use of linear mixed regressions allowed modelling individual trajectories and estimating an overall trend, thus capturing longitudinal effects on economic–productive indicators. However, this method may have limitations, as it assumes a linear evolution of these indicators and may not capture the

system dynamics during periods of high climatic variability (Veyssset & Boivent, 2025) or price fluctuations. Therefore, it is important to explore analyses using qualitative approaches or non-linear models in future research.

Overall, these results and their interpretation show that analysing change trajectories is an effective way to assess how systems progress or remain stable toward ecological intensification through participatory co-innovation processes. They also highlight that co-innovation and ecological intensification act as powerful drivers of positive technical change, promoting the sustainability of livestock production systems by improving productivity and efficiency, while preserving the diversity of family farms through better management of native grasslands and more efficient use of energy by the cows.

Conclusions

We presented an approach to analyse the trajectories followed by livestock production systems that adopted ecological intensification through co-innovation. The results demonstrate that these systems improved their key variables mainly through changes in management techniques, which enabled a more efficient use of pasture energy. This trajectory-based analysis proved to be a valuable tool to capture how co-innovation fosters positive technical change, enhancing productivity and efficiency while sustaining the ecological basis of production.

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Modelling monthly pasture productivity with climate and satellite imagery: cues for prediction

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Abstract

Pasture productivity in the Montado faces significant challenges due to climate change, particularly in the semiarid Alentejo region of southern Portugal. Interannual changes in climate partly control pasture productivity, together with local (farm) factors, such as potential solar radiation, elevation, water accumulation potential, and slope. A better understanding of how these different factors interact is crucial for efficiently managing grazing throughout the year. Remote sensing and modelling detect spatial and temporal patterns in productivity. Vegetation indices derived from 10 m resolution satellite imagery were combined with monthly climate parameters from 2016–2022. Data mining and machine learning allowed the identification of areas with highest productivity and driving factors. The most important climate parameters include 3-month temperature (<12°C) and 2-month precipitation (>70 mm). Local factors favour exposed valleys, where water availability and sun exposure are higher, and slopes are less steep thereby minimizing soil erosion.

Keywords: elevation, high-resolution, machine learning, precipitation, temperature

Introduction

The Montado, a mixed agricultural/pastoral ecosystem with low tree density, offers several ecosystem services, such as support to extensive animal production. Understanding and quantifying factors determining grassland productivity in farms with livestock production is paramount for adaptive management by landowners. Water stress can have severe impact on grassland productivity (Muñoz-Gómez *et al.*, 2024) but local variables, such as slope and soil type also play important roles (Mamede, 2024). High resolution remote sensing (RS) plant productivity and topography offer the opportunity to better understand the dynamics of pasture productivity in the Montado. Modelling annual pasture productivity at the farm scale has been achieved using RS products and machine learning (ML) (Mamede, 2024), but landowners need frequent predictions to actively change management by using dynamic models based on constantly updated information. This work encompasses a tentative approach in modelling monthly pasture productivity at the property scale, offering cues for future prediction that allows farm precision and adaptive management.

Materials and methods

The study site corresponds to a 105-ha farm close to Mértola, Alentejo, in southern Portugal (latitude 37.76°N, longitude 7.65°W), with a mean annual temperature during 2017–2022 of 18.1°C and precipitation of 370 mm. The land use is a low-density Montado, comprising natural pastures on which extensive livestock with rotational grazing is practised. The monthly means of the “Plant Phenology Index” (PPI) formulated with RED and NIR bands (Jin and Eklundh, 2014) were obtained

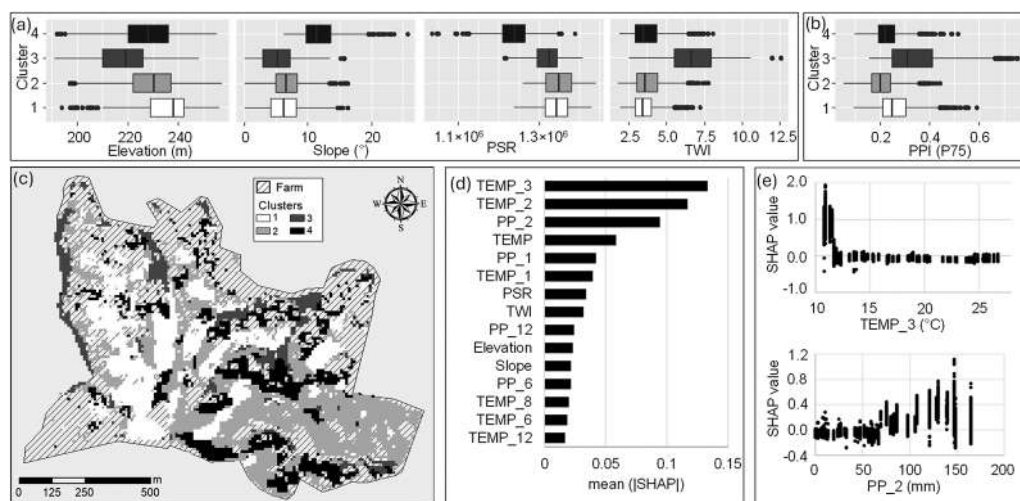


Figure 1. (a) Boxplots showing elevation, slope, PSR, and TWI and (b) 75th percentile of the PPI in the four clusters. (c) Spatial distribution of the four clusters within the farm. (d) Variable importance in pasture productivity represented by the mean of absolute SHAP values. (e) Individual SHAP values for combinations of records/variables for TEMP_3 and PP_2: positive SHAP values indicate positive correlation with productivity, negative indicate negative correlation, and 0 indicates no correlation.

for grassland 10 m-pixels, from 2017 to 2022, from Sentinel 2 RS High-Resolution vegetation phenology and productivity (HR-VPP, 2025). The PPI, taken as a proxy of pasture productivity, has shown good correlation with gross primary production. Topographic variables, including elevation and derived slope, potential solar radiation (PSR), and topographical wetness index-TWI (representing water accumulation potential) were determined from the ALOS PALSAR (2019) and EU-DEM v1.1 (CLMS, 2016) elevation models. Mean monthly precipitation and temperature were extracted from ERA5 Land (C3S, 2022) from 2016–2022. Climate variables were aggregated at distinct time intervals, namely: present month (PP and TEMP); previous 1, 2, 3 6, 8, and 12 months (PP_1 and TEMP_1, and so on). Data were combined in a spatial-temporal dataset, each space/time combination considered a record (386051 records and 19 variables). Spatial changes in productivity were assessed using the 75th percentile (P75) and the standard deviation of the PPI from 2017–2022 and topographical variables. Records were grouped into four clusters using k-means. The ML model XGBoost coupled with Shapley Addictive exPlanations (SHAP) (Li, 2022) was used to model productivity based on topographical and climate variables, as it is ideal for large datasets and shows a better performance when compared to other models (Li, 2022). Model parameter tuning was based on randomly selected records for training (70%; calibration) and testing (30%; validation) until reaching the lowest root-mean-square error (RMSE) highest R2, and lowest overfitting. Data processing and graphics were performed using the open-source software R (R Core Team, 2023) and ArcGIS Pro.

Results and discussion

Cluster analysis shows that the farm can be divided into four main typologies (Figure 1a–c): cluster 1 is located along ridges in the central and W part of the farm, corresponding to highest areas, exposed to sunlight (high PSR), with mild slopes and low TWI; cluster 2 is similar, but dominates the SE area, in the transition between ridges and unshaded valleys, with slightly lower elevation and PSR, and slightly higher slopes and TWI; cluster 3 is found on exposed low and flat valleys, with maximum TWI, and intermediate PSR; cluster 4 is in north facing (low PSR) steep slopes with low TWI. Cluster 3 is the

most productive (Figure 1b), as it gathers all the characteristics favouring plant development: sunlight, higher water accumulation, and low slope minimizing soil erosion. The second highest productive cluster is the cluster 1, with similar topography to cluster 2, except for elevation, which is higher. Differences in PPI in these two clusters might be correlated to additional factors, such as soil type, mineral content, or even management practices, not considered in this study. The best predictive model showed a RMSE of 0.18 PPI units (approx. 6%) and an R^2 of 0.84. The importance of the variables is represented by the mean absolute SHAP value (Figure 1d), showing that 1–3 months climate controls productivity, followed by topography, and by 6–12 months climate. Furthermore, higher productivity is found when TEMP_3 < 12°C and when PP_2 > 70 mm (Figure 1e), indicating climate thresholds beyond which productivity will be affected.

Conclusion

Topography affects pasture productivity favouring exposed valleys with higher water availability. A ML model was developed to predict productivity and identify factors determining it. The most important are the 3-month temperature (<12°C) and 2-month precipitation (>70 mm). Beyond these values, productivity is determined by interaction of all other variables as well as other factors not considered here, such as soil and management.

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Multi-species grasslands sustain on-farm productivity while reducing nitrogen fertilizer inputs

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Abstract

Species-rich grasslands are proposed as a strategy to reduce nitrogen (N) fertilization while maintaining productivity. However, the effectiveness of this strategy in agricultural grasslands remains uncertain, particularly since grasslands with varying species richness are managed differently. Our aim was to compare, in an on-farm context, the biomass yield of three grassland management types varying in species richness and N fertilization levels: (1) intensively managed species-poor permanent grasslands (PG) (2) and multi-species grasslands (MSG) sown with grasses, forbs, and legumes fertilized with 0, 150, and 300 kg N ha⁻¹, and (3) unfertilized highly species-diverse semi-natural grasslands (SNG), at twelve farm locations in the Netherlands over two years. MSG receiving 150 kg N ha⁻¹ produced yields similar to PG fertilized with 300 kg N ha⁻¹. Semi-natural grasslands had the highest species richness but the lowest biomass yields. Transitioning from PG to MSG presents a promising diversification strategy for livestock farmers aiming to reduce N fertilisation, improve sustainability, and maintain productivity. Further increasing in species richness, as in SNG, reduces biomass yield.

Keywords: biological nitrogen fixation, forage yield, sustainability

Introduction

Management of temperate grasslands has focused on enhancing productivity through specialisation and intensification, leading to a reliance on perennial ryegrass (*Lolium perenne* L.) monocultures in the Netherlands. Despite its agronomic qualities, their productivity heavily depends on external nitrogen (N) inputs that pose sustainability challenges, whereby the reduction of biodiversity is an important issue addressed by the dairy industry in the Netherlands. Grassland diversification has been proposed as a strategy to enhance sustainability while maintaining productivity. This can be achieved through extensification towards semi-natural grasslands or the establishment of productive multi-species grasslands, each offering distinct management approaches, ecological and productivity benefits (Allen et al. 2011). In multi-species grasslands, a combination of productive grasses, forbs, and legumes are sown to enhance biodiversity, optimize biological N fixation, and reduce reliance on synthetic fertilizers while maintaining high productivity. Research has demonstrated that species-rich mixtures can sustain high productivity with significantly lower N inputs. However, further on-farm research is needed to assess their viability across diverse conditions. Therefore, this study aimed to compare the biomass yield of three grassland management types—intensively managed species-poor permanent grasslands, multi-species grasslands and semi-natural grasslands—under different N fertilization levels and mixed grazing/cutting management across twelve on-farm locations over two years.

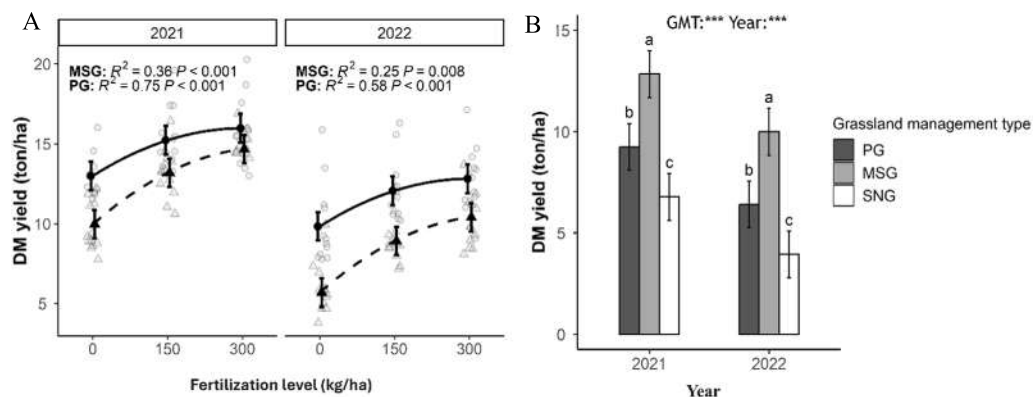


Figure 1. (A) Grassland management type and N fertilization effects on dry matter yield. Solid line and circles represent MSG and dashed line and triangles represents PG. Symbols represent means with 95% confidence interval error bars. (B) Grassland management type and year at zero N fertilization on dry matter yield. Bars represent means with 95% confidence interval error bars. Means with the same letter are not significant different (Tukey test, $P > 0.05$). Annotations represent ANOVA main and interaction effects (*** $P < 0.001$; ** $P < 0.01$; * $P < 0.05$). PG, permanent grassland; MSG, multispecies grassland; SNG, semi-natural grassland.

Material and methods

This on-farm study took place at twelve locations on sandy soils in the eastern part of the Netherlands amounting to a total sample size of 36 fields (for more details on climatic conditions and management see Van Eekeren *et al.*, 2025; Zetterlind *et al.*, 2025). We selected the three contrasting grassland management types in the following way: (1) Intensively-managed permanent grasslands (PG): fields were selected following the definition of permanent grassland (Allen *et al.*, 2011). These fields were conventionally managed with fertilization from both inorganic and slurry manure, and featured a vegetation cover of more than 80% perennial ryegrass and less than 5% legumes; (2) multi-species grasslands (MSG): These fields were intensively managed and sown with a mixture of grasses and a minimum of eight different species of legumes and forbs. These fields were conventionally managed with fertilization mainly as slurry manure; (3) semi-natural grasslands (SNG): These fields were extensively managed with no fertilization, and featured a plant community with a minimum of 15 different plant species per 25 m².

In 2021 and 2022 a plot of 9.0 × 7.2 m² was established at a random location in the farmer-managed fields of PG and MSG, enclosed with fences to exclude grazing and additional nutrient inputs from manure and urine. New plots were established each year. Plots were divided into subplots of 2.4 × 5.2 m² for fertilizer treatments. The fertilizer treatments in these subplots included 0 kg N ha⁻¹ at the centre, with adjacent subplots receiving 150 and 300 kg N ha⁻¹ calcium ammonium nitrate (27% N). In addition to N fertilization, all PG and MSG subplots received 80 kg ha⁻¹ P₂O₅ and 300 kg ha⁻¹ K₂O to ensure these nutrients were not limiting factors for forage growth. The SNG plots did not receive any fertilization. PG and MSG were mowed five times per year, while SNG were mown two times per year.

Results and discussion

The dry matter yields significantly varied among grassland management types, years, and N fertilization treatments (Fig. 1A). In 2021, dry matter yields were larger for MSG compared to PG under 0 to 150 kg N ha⁻¹ fertilization, while this difference disappeared at 300 kg N ha⁻¹. Dry matter yields

in multispecies grasslands receiving 150 kg N ha⁻¹ fertilization and permanent grasslands receiving 300 kg N ha⁻¹ were not significantly different. In 2022, dry matter yields were larger in MSG compared to PG under similar fertilization levels. The dry matter yields under zero N fertilization varied among the three grassland management types (Fig. 2B). MSG provided the largest dry matter yields followed by permanent grasslands and lastly by semi-natural grasslands in both years. The average dry matter yields were significantly larger in 2021 than in 2022 ($P < 0.001$). Figure 1. (A) Grassland management type and N fertilization effects on dry matter yield. Solid line and circles represent MSG and dashed line and triangles represents PG. Symbols represent means with 95% confidence interval error bars. (B) Grassland management type and year at zero N fertilization on dry matter yield. Bars represent means with 95% confidence interval error bars. Means with the same letter are not significant different (Tukey test, $P > 0.05$). Annotations represent ANOVA main and interaction effects ($***P < 0.001$; $**P < 0.01$; $*P < 0.05$). PG, permanent grassland; MSG, multispecies grassland; SNG, semi-natural grassland.

Conclusion

We found that the multi-species grassland MSG sward enabled the N fertilizer requirements to be more than halved, and with 150 kg N ha⁻¹ year⁻¹ it produced yields similar to intensively managed species-poor permanent grasslands (PG) fertilized with 300 kg N ha⁻¹. While the SNG treatment hosts the greatest number of plant species, this comes at the cost of reduced biomass yield. Overall, we conclude that transitioning from PG to MSG is a promising diversification strategy for livestock farmers aiming to reduce N fertilization, improve sustainability, and maintain productivity.

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Sustainable Angus crossbreed beef in biodiverse pastures: enhancing productivity and reducing global warming potential

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Abstract

The connection between carbon flows and cattle production is very uncertain. Here, we assess the Global Warming Potential and circular economy opportunities of beef cattle production at Herdade da Pecena, Alentejo, Portugal. We found that extensive use of permanent sown biodiverse pastures (SBP) enriched with legumes enhances soil organic matter and carbon sequestration. SBP achieved high dry matter (DM) forage productivity (12 638 kg DM ha⁻¹ year⁻¹), outperforming natural pastures (NP) in 32%. Additionally, applying organic matter from intensive units further boosts yields, with treated NP reaching up to 9555 kg DM ha⁻¹ year⁻¹ compared to the regional average of 2833 kg DM ha⁻¹ year⁻¹. The study highlighted a low carbon footprint of 16 kg CO₂e per kg of live weight, at the low end of the range internationally (12–91 kg CO₂e kg⁻¹). This result was due to the use of food industry by-products and corn silage in feed, as well as feed-efficient pasture systems. The finishing of steers lowers the carbon intensity compared to selling weaned calves, due to dilution of maternal impacts. These findings show that well-managed pasture systems can enhance productivity and reduce greenhouse emissions in beef production, while also supporting soil fertility.

Keywords: biodiverse sown pastures, carbon footprint, cattle production, circular agriculture, pasture-based systems, soil organic matter

Introduction

Agriculture is a significant source of greenhouse gas (GHG) emissions, accounting in Portugal for 10% of the national total, mainly methane and nitrous oxide (APA, 2018). The livestock sector, particularly ruminants, is central due to methane from enteric fermentation and manure (Gerber *et al.*, 2013). Although milk and beef consumption is stable, national production does not meet demand (INE, 2023). Increasing domestic production with improved environmental performance is crucial. Ruminants, able to convert cellulose-rich, non-edible feeds into food, also enable circular economy practices (Mottet *et al.*, 2017). Well-managed grazing supports soil health and carbon sequestration, especially with sown biodiverse pastures (SBP). Assessing how circular practices, feeding strategies, and management can reduce GHG emissions is essential.

This work, conducted between October 2019 and May 2021 at Herdade da Pecena, a 1100-hectare farm in Portel (Alentejo) managed by Best Farmer, tested whether SBP could boost forage productivity for grazing and, with maize silage in feedlots, lower Angus beef Global Warming Potential (GWP). We also evaluated how circular approaches—namely applying organic fertilizer from a dairy unit to crops

and pastures—could reduce emissions. GWP was assessed through a Life Cycle Assessment (LCA) approach.

Materials and methods

The farm includes a Holstein-Friesian dairy unit (providing organic fertilizer), a pasture-based suckler herd (Blonde d'Aquitaine and crossbred cows bred with Black Angus bulls), and a projected feedlot for 5000 crossbred Angus steers. On-site crops include ryegrass, triticale, and maize silage for animal feed.

Methods were structured into three components: (1) installation and management of SBP; (2) monitoring pasture productivity and quality in SBP and natural pastures (NP), with (NP1) and without (NP2) organic fertilisation (OF); (3) GHG emissions assessment of extensive and feedlot units.

Pasture productivity and quality were evaluated through intra-annual (January, March, May) and inter-annual (2018–2021) monitoring. Assessments included above-ground primary productivity and animal intake via biomass comparison between grazed and ungrazed areas. Measurements covered: biomass sampling (kg DM/ha), functional group composition (legumes, grasses, forbs), species abundance (%), and ground cover (%). The GWP was assessed with LCA, accounting for the full beef unit—including breeding cows, bulls, heifers, and weaned calves—and feeding regime. LCA was applied to both the extensive (pasture) and projected feedlot units, benchmarked against an operating feedlot from the same producer.

GWP was expressed in kg CO₂e per kg live weight (LW) at farm gate or transfer to feedlot. Emission flows included direct enteric and manure emissions, plus indirect upstream impacts from concentrates (“external feed”) and forage production, covering fertiliser and field operations. LCA used Terraprima’s Online Tool (<https://tool.terraprima.pt/en/login>) with direct farm data. Emissions were modelled using National Inventory Reports (APA, 2016, 2018, 2020) and IPCC guidelines (IPCC, 2006).

Results and discussion

Results of pasture productivity assessments show clear differences between management strategies. SBP achieved the highest productivity in 2020, reaching 12 638 kg DM ha⁻¹ (Table 1), surpassing NP with and without OF. All treatments exceeded the Alentejo regional reference for natural pastures (2833 kg DM ha⁻¹; Abreu *et al.*, 2005), likely due to high baseline soil fertility at Herdade da Pecena from historical OF applications.

The superior SBP productivity in 2020 is linked to species composition and soil enrichment (Teixeira *et al.*, 2011). SBP showed greater legume abundance, enhancing nitrogen content and nutritive value.

NP had limited legumes across years, though by 2021 NP2 showed an increase (up to 15%), indicating small improvements can occur in low-input systems over time.

Regarding the GWP, the LCA for the extensive beef production unit identified enteric fermentation as the primary source of GHG emissions (58%), followed by emissions from manure in pasture (25%). Emissions associated with external feed (9%) and forage crops (6%) were notably lower, while the import of replacement heifers accounted for 11%. The GWP of weaned calves was 21 kg CO₂e kg⁻¹ LW, reduced to 19 kg CO₂e kg⁻¹ LW when accounting for carbon sequestration from SBP.

Table 1. Evolution of annual primary productivity (PP) and legumes (Leg) abundance in SBP and in NP, between 2019 and 2021.

	SBP		NP1		NP2	
	PP (kg DM ha ⁻¹)	Leg (%)	PP (kg DM ha ⁻¹)	Leg (%)	PP (kg DM ha ⁻¹)	Leg (%)
2019	4319	48	8226	1	7600	4
2020	12638	43	7256	<1	8511	<1
2021	8358	20	8359	0	6225	15

In the projected feedlot phase, the GWP of finished steers was estimated at 16 kg CO₂e kg⁻¹ LW. The strongest contributor were the embodied emissions in the weaned calf itself (76%), that is during the extensive phase. External feed contributed 14% of the emissions. Compared to international benchmarks (12–91 kg CO₂e kg⁻¹, Poore & Nemecek, 2018), the system sits at the lower end, and midrange within some European estimates (13–25 kg CO₂e kg⁻¹, Berton *et al.*, 2017, Bragaglio *et al.*, 2018).

Conclusions

This work shows that integrating beef and dairy production systems through pasture management and circular economy practices can reduce the GWP of beef production. The implementation of SBP, combined with the use of organic residues from the intensive units, contributed to increased pasture productivity and carbon sequestration. The carbon footprint of both weaned calves and finished steers was on the lower side of the range for global benchmarks, highlighting the climate mitigation potential of the strategies adopted at Herdade da Pecena.

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Session 5.
Grasslands and rural land policy tools

Innovation systems and strategic communication approaches for grassland technology adoption

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Abstract

Grassland-based agriculture underpins the productivity, sustainability and competitiveness of livestock systems across the world. Despite sustained investment in grassland research, adoption of proven practices and technologies at farm and industry level remains uneven. This paper examines how agricultural researchers focusing on grassland can better embed their findings into action by working more effectively within Agricultural Knowledge and Innovation Systems (AKIS). Drawing on Irish and European experience including advisory services, innovation support services, Operational Groups, Living Labs, the Teagasc Signpost Programme (teagasc.ie/environment/climate-change-air-quality/signpost-programme/), PastureBase Ireland (pasturebase.teagasc.ie) and the Irish Grassland Association (www.irishgrassland.ie) this paper analyses how enabling environments, co-design and feedback loops accelerate adoption. Following established evidence that adoption decisions are frequently rational responses to risk, complexity and context (Vanclay, 2004), the paper emphasises the need to align technical solutions with behavioural and institutional realities. To match the innovation challenge, approximately two thirds of the manuscript is dedicated to AKIS and innovation-system mechanisms, and one third to strategic communication as a critical enabler of change.

Keywords: advisory services, AKIS, co-design, grassland, innovation support, Living Labs, Operational Groups, strategic communication

Introduction

Grassland systems form the backbone of ruminant livestock production across much of Europe, delivering food, ecosystem services and rural livelihoods while contributing to climate mitigation and biodiversity outcomes. In Ireland, grass-based livestock production provides a comparative advantage; nevertheless, the agri-food sector faces increasing pressure to deliver measurable improvements in greenhouse gas mitigation, water quality and biodiversity. Grassland research has generated strong evidence on grazing management, pasture measurement, nutrient-use efficiency, sward composition and emerging digital technologies. Yet the existence of robust research outputs does not guarantee widespread uptake. Research impact ultimately depends on whether knowledge is mobilised, contextualised and embedded within the institutions and decision processes that shape farm and industry practice.

Agricultural Knowledge and Innovation Systems (AKIS) provide a systems lens for understanding why some innovations diffuse rapidly while others stall, even when the technical case is strong (SCAR AKIS, 2019; SCAR AKIS, 2023). From this perspective, innovation is rarely linear; it is produced through interaction between researchers, advisors, farmers, industry actors and policy-makers. Weak linkages between these actors can result in fragmented messaging, misaligned solutions, low trust and inadequate support for implementation. For grassland researchers, achieving impact therefore requires engagement with the wider innovation system co-designing solutions, strengthening feedback loops,

and ensuring that advisory services, policy levers and market incentives collectively create an enabling environment for adoption.

This paper is a qualitative synthesis structured as a scientific discussion paper. It integrates: (i) published literature on innovation systems, advisory services and technology adoption; (ii) European policy and practice guidance on AKIS and advisory services (e.g., SCAR AKIS reports and policy briefs); (iii) Irish case evidence from national initiatives and platforms that link research with practice, including the Teagasc Signpost Programme, PastureBase Ireland and the Irish Grassland Association; and (iv) lessons from feedback-intensive industries (e.g., healthcare and software) where iterative design, user testing and continuous improvement are institutionalised. The analysis focuses on identifying actionable mechanisms that grassland researchers can use to embed findings into farm and industry action.

The results are organised around key AKIS functions that repeatedly emerge as determinants of adoption: advisory capacity and trust; innovation support and brokerage; multi-actor co-design within a triple-helix logic; structured experimentation through Operational Groups and Living Labs; enabling environments created by policy and industry alignment; digital routes for embedding research in decision workflows; and feedback loops that connect farm and industry experience back to research agendas. Together, these functions form the practical architecture through which grassland research is converted into action.

AKIS actors and roles in grassland innovation

Within grassland systems, AKIS is best understood as the practical ‘delivery system’ for turning evidence on grazing management, pasture measurement, nutrient efficiency, sward composition and digital decision support into routine decision-making. Knierim and colleagues emphasise that AKIS analysis is valuable precisely because it maps the organisational diversity and the communication pathways through which knowledge is produced, shared and used, and because it helps to identify weak or missing links between research, advisory services and practice (Knierim *et al.*, 2015). For grassland researchers, this highlights that impact depends not only on generating robust results, but on deliberately working through the institutions and intermediaries that farmers engage with when making operational and strategic choices.

Table 1 summarises the main AKIS actors and the specific roles they play in embedding grassland research into action. A core implication is that researchers influence adoption not only through publications, but through participation in the relationships and routines that connect evidence to decisions especially those mediated by advisory services.

Advisory services and innovation support services

Advisory services are repeatedly identified as the most influential intermediary between research and practice because they combine technical competence with trusted relationships. Advisors interpret research, match it to farm objectives, and provide the ‘implementation infrastructure’ coaching, troubleshooting, sequencing of changes, and monitoring that turns knowledge into routine practice. Importantly, advisory engagement can transform adoption from a one-off decision into a managed process, where farmers experiment, reflect and adjust. This matters in grassland systems where benefits often depend on cumulative management decisions across seasons (e.g., grazing rotation, measurement frequency, nutrient planning).

Table 1. Roles of key actors within grassland AKIS (adapted from SCAR AKIS, 2019).

AKIS actor	Primary role in embedding grassland innovation	Irish/EU examples (web reference)
Researchers	Generate evidence; co-design; evaluate outcomes; support learning	Teagasc (www.teagasc.ie); Universities
Advisory services	Translate research; tailor to farm context; coach implementation; reduce risk	Teagasc Advisory (www.teagasc.ie); private advisory
Innovation support	Broker partnerships; facilitate multi-actor projects; sustain networks	EIP-AGRI support; AKIS facilitators
Farmers	Define problems; test innovations; share peer learning; provide feedback	Discussion groups; Signpost farms (teagasc.ie)
Industry	Scale solutions; align incentives; invest in tools and supply chain capability	Processors; input and tech firms
Government/policy	Create enabling policy; fund advisory/innovation; set targets and standards	CAP AKIS requirements; national agencies

Innovation support services (e.g., facilitators, brokers, network coordinators) complement advisors by enabling collaboration across organisations and disciplines. They are particularly valuable for ‘system innovations’ that require multiple actors to change together such as climate mitigation pathways that involve farm practice, processor requirements, financial incentives and measurement frameworks. When researchers engage early with both advisory and innovation support actors, they can design research outputs that are easier to implement (e.g., clear decision rules, practical tools, training materials) and are more likely to be championed within trusted advisory channels.

Grassland innovations such as decision-support tools, pasture measurement platforms, low-emission nutrient practices, and new sward systems are adopted within increasingly pluralistic advisory landscapes, where farmers access advice through a mix of public, private, and collective providers and through multiple modalities, including digital channels. In this context, advisory services are no longer just technical “messengers” but act as innovation intermediaries, helping farmers interpret data, navigate risk and complexity, and connect with multi-actor networks and pathways for change. For grassland researchers, the implication is that embedding technologies into practice requires engaging early with these advisory and innovation-support functions co-designing tools and messages that fit diverse grassland systems and ensuring advisors have the capacity to support implementation rather than relying on linear dissemination models (Klerkx, 2020).

Triple helix, multi-actor co-design and Living Labs

The triple-helix logic linking research, industry and government remains a useful way to frame how incentives and capabilities combine to enable adoption (Figure 1). In grassland systems, however, adoption outcomes improve when the helix is expanded into a multi-actor model in which farmers and advisors are co-design partners rather than end recipients. Co-design reduces the risk of ‘solution misfit’ by ensuring that innovations align with labour realities, infrastructure, time constraints and local norms. It also builds legitimacy because farmers see their practical knowledge reflected in the solution and can anticipate implementation challenges early.

Living Labs provide a structured environment for co-design and experimentation under real conditions. A grassland Living Lab can be conceptualised as a network of farms and partners where measurement, learning and adaptation are continuous. Such settings are particularly valuable for innovations that require iterative adjustment (e.g., integrating multispecies swards into rotations, adopting pasture measurement protocols, and introducing new decision-support tools). Horizon Europe Cluster 6 explicitly invests in approaches that mobilise existing knowledge and accelerate impact (research-and-innovation.ec.europa.eu). Grassland researchers who participate in Cluster 6-type consortia can extend the reach of national evidence, benchmark approaches across regions, and strengthen the portability of solutions.

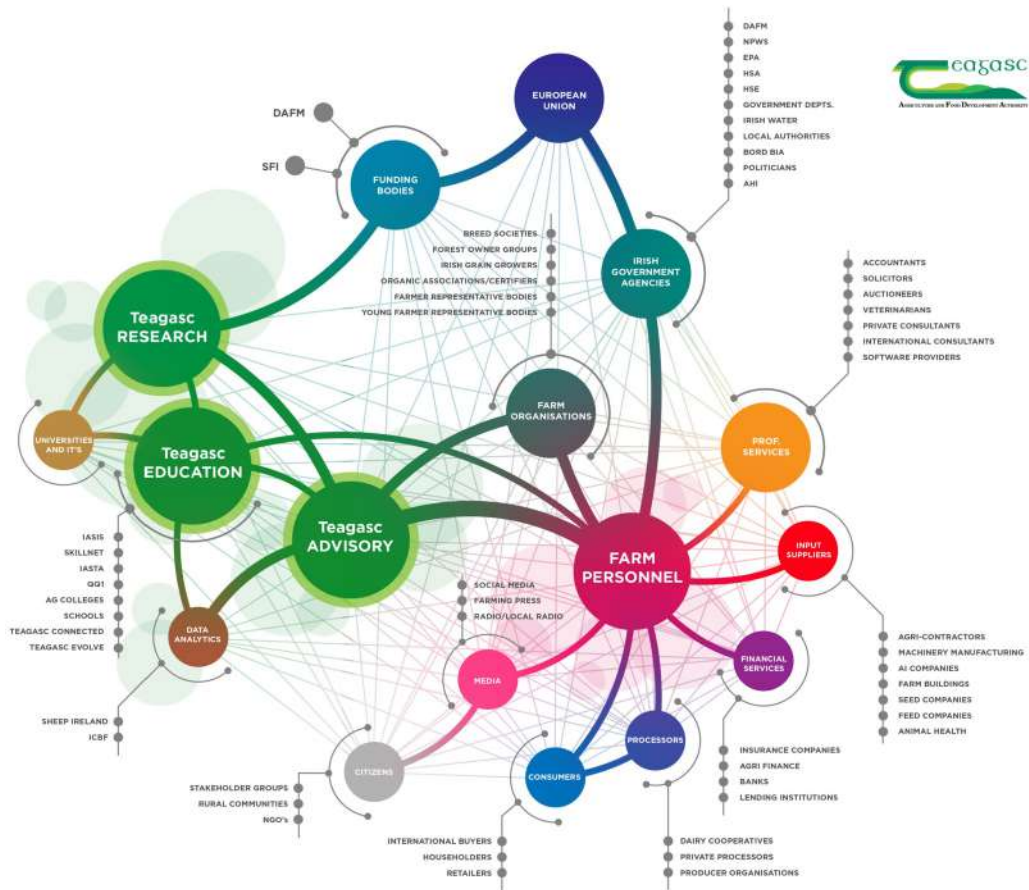


Figure 1. Conceptual diagram of the Irish AKIS illustrating interaction and feedback between research, advisory/innovation support, farmers, industry and policy.

Operational Groups as applied innovation engines

Operational Groups (OGs) under the European Innovation Partnership for Agricultural Productivity and Sustainability (EIP-AGRI; ec.europa.eu/eip/agriculture/) provide a practical mechanism to translate research into applied solutions. OGs bring together multidisciplinary teams including farmers, advisors, researchers and industry to address a specific problem with clear deliverables. For grassland innovation, OGs are well suited to topics where context matters and where on-farm testing is essential (e.g., grazing decision rules under variable growth, nutrient efficiency practices, biodiversity-enhancing swards). Their strength lies in farmer involvement at the problem-framing stage, which increases relevance and improves the likelihood of adoption beyond the pilot cohort.

For researchers, OGs provide an applied pathway to validate research in commercial conditions and to generate usable outputs decision guides, protocols, cost–benefit evidence and training materials. OG governance can also formalise feedback loops by requiring regular reflection with farmer participants and advisors, thereby reducing the risk that research outputs become disconnected from practical constraints.

Thematic networks

Thematic Networks (TNs) offer a specific opportunity to strengthen grassland technology extension by linking strategic communication with adoption mechanisms. Under EU research and innovation programming, TNs are multi-actor projects designed to collect and synthesise existing knowledge and best practices on a defined theme and to translate this knowledge into accessible, user-oriented formats for farmers, advisors and other practitioners (European Commission, 2025). By focusing on knowledge mobilisation rather than primary research, TNs can reduce the ‘last-mile’ problem in innovation—turning scattered evidence into coherent, actionable guidance that is easier to implement on farms.

For grassland researchers, engagement with TNs is therefore a strategic route to impact: researchers can contribute by identifying ‘adoption-ready’ knowledge, co-developing decision rules and measurement protocols, supporting synthesis outputs, and ensuring that communications are aligned with advisory workflows and digital platforms (e.g., pasture measurement tools).

Enabling environments and the role of government and industry

Even highly effective innovations can stall in the absence of an enabling environment. Enabling environments include policy coherence, stable incentives, risk-reduction supports, time and capacity within advisory services, and digital infrastructure that allows farmers to operationalise new decision tools. Government influences these conditions through CAP implementation, funding and accreditation of advisory services, support for innovation projects, and the design of measurement and reporting requirements that signal what ‘good performance’ looks like.

Ireland’s Teagasc Signpost Programme (teagasc.ie/environment/climate-change-air-quality/signpost-programme/) provides a useful case of how enabling environments can be constructed by aligning evidence, advisory support and industry collaboration around climate action. The programme is designed to bring coherence to climate messaging and to prioritise practical, science-based measures for emissions reduction while supporting water quality and biodiversity co-benefits. From an AKIS perspective, its value lies not only in promoting measures, but in establishing a shared narrative, a set of demonstration contexts and a coordinated advisory approach that lowers the ‘coordination burden’ on individual farmers.

Digital embedding of research

Digital technologies can embed research outputs directly into decision workflows, making evidence actionable at the point where decisions are made. PastureBase Ireland (pasturebase.teagasc.ie) is an excellent example of research embedded into a practical digital platform. By standardising pasture measurement and providing decision support, the platform translates research on grass growth dynamics into weekly grazing and feed decisions. Importantly, digital platforms also create data infrastructures that enable feedback: aggregated, anonymised farm data can inform research, benchmarking and advisory improvement, thereby strengthening the research–practice loop.

Mechanisms for accelerating adoption of proven grassland technologies

Table 2 summarises concrete mechanisms that researchers can use to increase the probability that proven technologies are adopted. The mechanisms are grouped across project design, delivery pathways and scaling conditions.

Table 2. Practical mechanisms to accelerate adoption of proven grassland technologies through AKIS.

Mechanism	How it can accelerate adoption	What researchers can do
Embed advisors from project start	Ensures outputs match farm context; increases trust	Co-design trials, tools and training with advisors
Use demonstration farms / Living Labs	Reduces uncertainty; supports peer learning	Design experiments with farmer-defined success metrics
Operational Groups for problem-solving	Creates applied deliverables; supports scaling	Translate findings into protocols and decision rules
Digital decision-support tools	Makes evidence usable at decision time; enables data feedback	Co-develop user-centred features and data governance
Aligned incentives and standards	Reduces financial risk; clarifies 'what good looks like'	Provide evidence to support policy design and KPIs
Structured feedback loops	Improves relevance and usability; builds ownership	Use farmer panels, industry roundtables, iterative evaluation

A recurring barrier to embedding research is the assumption that farmers are 'slow' because of conservatism or lack of information. Adoption research suggests a more nuanced view. Vanclay (2004) argues that adoption decisions reflect rational judgement under uncertainty: farmers evaluate perceived risk, compatibility with their system, labour availability, complexity, and trust in information sources. In grassland systems, where weather variability and biological processes introduce uncertainty, farmers may delay adoption until benefits are observable in contexts similar to their own. This underscores why demonstration, peer learning, and credible economic evidence are often more persuasive than generic messaging.

The Irish Grassland Association (IGA; www.irishgrassland.ie) provides an important example of a voluntary, multi-actor forum where farmers, researchers, advisors, industry and policy stakeholders can share perspectives and address strategic challenges. Such platforms perform key AKIS functions: they build shared problem definitions, create spaces for debate about trade-offs, and support collective sense-making that can influence both practice and policy. For researchers, engaging with the IGA helps ensure that research agendas remain connected to emerging industry needs and that research outputs are communicated through channels that farmers respect.

Strategic communication for improving adoption

Strategic communication is not an add-on to innovation; it is an AKIS capability that shapes whether farmers and other stakeholders understand, trust and act on evidence. This part of the paper focuses on how researchers can contribute to coherent, targeted communication systems that respect the diversity of farming systems and demographics.

Fragmented messaging is a common barrier to adoption. When farmers receive inconsistent advice or shifting policy signals, they often respond by delaying change. Coherent communication requires alignment between the evidence base, advisory recommendations, industry initiatives and government policy. The Teagasc Signpost Programme offers a good example of coherence-building: it seeks to provide a unified set of practical climate measures and consistent messaging across multiple sectors (teagasc.ie/environment/climate-change-air-quality/signpost-programme/). For researchers, coherence means presenting findings as actionable 'packages' with clear assumptions, rather than isolated results.

Farmers are not a homogeneous audience. Differences in enterprise type (dairy, beef, sheep), scale, labour availability, technology access, age profile and values shape both information needs and preferred channels. Effective communication therefore begins with audience research: identifying what

Table 3. Communication channels for grassland innovation: strengths, uses and practical considerations.

Channel	Strengths	Best used for	Implementation notes
Radio	High reach; trusted;	Simple calls-to-action; seasonal reminders	Use consistent spokespeople; repeat key messages
Podcasts	Depth; flexible; niche audiences	Explaining trade-offs; farmer stories; Q&A	Research audience needs first; maintain regularity
Video	Demonstration; visual proof	How-to guidance; showing practice change	Feature farmers; keep short; link to advisory support
On-farm demos	Highest credibility; peer learning	System change; complex innovations	Support demo farmers; capture lessons learned
Digital platforms	Personalised; data-driven	Decision support and benchmarking	Ensure usability; provide training and helpdesk

challenges farmers are prioritising, what language they use to describe problems, and who they trust. Communication plans should also consider ‘moments of decision’ for example, spring grazing set-up, fertiliser planning, or investment decisions, when information is most likely to translate into action.

Ireland provides a useful illustration of how channel strategy must reflect farmer habits. Radio remains a powerful medium for reaching farmers, while podcasts, video and other online content are increasingly used. However, effective use of new channels requires design discipline i.e. planning, testing, and iteration, rather than simply launching content. Table 3 summarises channel strengths and when each is most appropriate.

Peer influence is a strong driver of adoption: farmers often report that they ‘listen to farmers’. Demonstration farmers can therefore be considered a central communication asset. They provide credible evidence of practical feasibility, expose implementation challenges, and enable observational learning that reduces perceived risk. Communication strategies should treat demo farmers as partners, supporting them with measurement tools, advisory backup, and opportunities to share both successes and setbacks honestly. This approach increases trust and reduces the risk of ‘over-promising’.

Other industries provide relevant analogues for feedback-driven communication. In healthcare, clinical pathways and patient information are continually revised based on practitioner and patient feedback, adverse event reporting and outcome measurement. In software, agile development institutionalises iterative user testing, rapid prototyping and continuous improvement. These industries treat communication and user support as part of the product, not separate from it. Grassland innovation can adopt similar practices by establishing regular feedback cycles (e.g., farmer panels, analytics from digital tools, advisory ‘field notes’), and by updating guidance as real-world experience accumulates.

Education systems provide a powerful route for embedding research outcomes and innovation capability. Integrating AKIS concepts, systems thinking, digital literacy and behavioural insights into agricultural curricula equips future farmers, advisors and industry professionals to interpret evidence and participate in co-design. Lifelong learning such as short courses, micro-credentials, discussion-group learning, and blended online formats provides structured ways for farmers and professionals to engage with research continuously. Researchers can contribute by co-developing curriculum modules, supporting teaching farms and Living Lab placements, and producing open educational resources aligned with national priorities.

Conclusion

This paper argues that embedding grassland research into action is fundamentally a systems challenge, not a dissemination challenge. The strongest technical evidence will still underperform if it is not translated, supported, financed, communicated and iteratively refined through the institutions that

shape day-to-day decisions on farms and in the wider agri-food industry. An AKIS lens therefore helps to explain why adoption can be uneven even when innovations are 'proven': uptake depends on the strength and coherence of the relationships between research, advisory services, farmers, industry and policy (SCAR AKIS, 2019; SCAR AKIS, 2023).

For grassland researchers, the most actionable implication is to design research for adoption from the outset. This means partnering with advisors and innovation support services early to co-define research questions, success criteria and practical deliverables; prioritising outputs that fit decision workflows (e.g., decision rules, protocols, seasonal planning tools, and user-centred digital features); and using multi-actor governance to ensure that solutions remain feasible under real farm constraints. Operational Groups and Living Labs provide practical architectures for this approach because they institutionalise co-design, on-farm experimentation and rapid learning, and they convert research outputs into 'useable knowledge' that farmers and advisors can apply with confidence.

Creating an enabling environment is equally important. Policy coherence, stable incentives, accessible advisory capacity, and digital infrastructure reduce adoption risk and lower the coordination burden on individual farmers. Government has a key role through CAP implementation, funding of advisory and innovation supports, and the establishment of clear performance signals. Industry also matters: processors and supply-chain partners can accelerate adoption when they align market requirements, measurement approaches and supports. The Teagasc Signpost Programme (www.teagasc.ie) illustrates how industry-led collaboration can strengthen coherence around climate action, provide trusted demonstration contexts and prioritise practical mitigation pathways at farm level.

Feedback loops are a non-negotiable feature of high-performing innovation systems. Grassland technologies and practices must be refined against farmer experience to ensure they are beneficial, viable and easy to implement. Digital platforms such as PastureBase Ireland (pasturebase.teagasc.ie) show how research can be embedded in tools that support decisions while generating data that informs benchmarking, advisory improvement and future research. At a strategic level, the Irish Grassland Association (www.irishgrassland.ie) demonstrates the value of multi-actor fora where farmers, researchers, advisors, industry and policy-makers can share perspectives, surface emerging constraints and align priorities for grassland development.

Strategic communication is the complementary capability that turns AKIS architecture into adoption at scale. Communication must be coherent across research, advisory, industry and policy; targeted to the diversity of farm systems and demographics; and delivered through channels that farmers recognise and trust. In the Irish context, radio remains influential, while podcasts, video and digital content are growing in importance. However, channel selection should be driven by audience insight and evaluated for reach and behavioural outcomes. Demonstration farmers and peer storytelling are particularly powerful because farmers often 'listen to farmers', and because visible on-farm implementation reduces uncertainty and makes trade-offs tangible.

Finally, education and lifelong learning should be treated as strategic infrastructure for innovation. Embedding AKIS concepts, systems thinking, data literacy and behavioural insight into agricultural education curricula will build the next generation of farmers, advisors and industry professionals who can participate in co-design and interpret evidence in context. Lifelong learning through discussion

groups, short courses and blended formats provides structured opportunities for farmers and advisors to make sense of evolving evidence and to update practice over time.

Taken together, the pathway to greater grassland impact is clear: strengthen the ‘delivery system’ around research through advisory and innovation supports; institutionalise co-design through Operational Groups and Living Labs; create enabling environments through aligned policy and industry signals; build feedback loops that continuously improve solutions; and invest in coherent, targeted communication and education. By working in this way, grassland researchers can contribute directly to resolving major challenges facing the agri-food sector productivity, climate action and environmental performance while accelerating the adoption of proven technologies and practices.

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Potentials and problems of grasslands in instruments to realize green transition in Denmark

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Abstract

The green transition of agriculture is challenged by lack of targeted, effective, simple and clear policy instruments to facilitate effective implementation, and evaluation of outcomes. This paper shows how grasslands could be integrated as measures to reduce GHGs from agriculture in Denmark. Grassland systems were defined with estimated annual (1) soil carbon (C) storage, (2) emissions of nitrous oxide (N₂O) from fertilizers, crop residues, and indirect sources, and (3) emissions of carbon dioxide (CO₂) from liming, and their sum subtracted from the analogous sum for a “reference” crop rotation with soil C storage set to zero. Grassland systems with prevailing soil C storage were grass for seed, catch crops, intermediate crops and perennial energy crops, with annual “net effect” (difference from reference) of 2.9, 0.9, 0.5 and 1 Mg CO₂eq ha⁻¹, respectively. Not all latest knowledge was integrated, and alongside its static nature, the method requires continuous validation to reduce greenwashing of the net effect, while transfer to other regions requires incorporation of local empirical data.

Keywords: biomass, carbon, emission, measure, multi-factor, nitrogen

Introduction

Globally, increasing data show the pivotal role of grasslands in achieving eco-environmental, and societal targets due to their botanical, physiological, agronomic and cultural traits tailored by humans for centuries, and for contributing significantly to soil C storage (Shang *et al.*, 2024). Leveraging these developments remains a methodological challenge for a green transition aiming for European climate neutrality in 2050. This paper shows an approach used in science-based policy advice for C accounting of grasslands—potentials and problems.

Materials and methods

Denmark is taken as a case study, where grasslands have historically been crucial for cattle feed, particularly for dairy cows. The grassland area has not decreased much over time despite shifts towards silage production for indoor feeding and cereal crop rotation, partly due to increased milk production and grass for seed. Cereal crop rotation was set as a reference system receiving only synthetic N fertilizer, as in Denmark the organic N fertilizer is not changing much due to agricultural measures. Grasslands in the context of the agricultural economy of Denmark were represented by five systems: catch crops, intermediate crops, fallow in rotation, perennial energy crops in rotation, and grass for seed (Table 1). For each system and according to the IPCC approach, emission factors (EFs) were either empirically found or mostly tier-1 (global) values. Liming emits CO₂ and was considered, whereas use of fossil energy was not, as it is allocated to the energy sector. Negative emission, i.e., soil C storage, was estimated from studies and reports. All flows for the 5 agricultural systems were converted in CO₂eq. ha⁻¹ year⁻¹ and summed in a net effect relative to the reference system.

Table 1. Description of the five different grass-based systems used in the study.

Catch crops	<i>Lolium spp.</i> , <i>Brassica spp.</i> , Crops grown between main crops to retain soil nitrogen; with-/out legumes; on sandy-loamy soil
Intermediate crops	Crops grown between main crops to improve soil fertility
Fallow in rotation	Cultivated land intentionally left unsown/uncultivated for one/more growing seasons to allow soil recovery and nutrients
Perennial energy crops	<i>Miscanthus spp.</i> harvested annually; <i>Salix spp.</i> , harvested once every 2–3 years; <i>Populus spp.</i> every 2–10 years; life span 10–30 years; Crops grown for bioenergy and improving soil organic matter; on sandy-loamy soil
Grass for seed	<i>Lolium spp.</i> , <i>Trifolium spp.</i> ; Grass cultivated specifically to produce and harvest viable seeds; harvest is 2nd or 3rd year.

See Adamsen and Manevski (2025) for further details.

Table 2. Net annual effect for grassland systems plausible in Denmark.

		Absolute	Difference from reference system (minus sign means higher absolute value, i.e. higher emission since the last row value is a sum, so called “net effect”)					
		Reference crop rotation	Catch crop– non–legume	Catch crop– legume	Inter– mediate crop	Fallow in rotation	Perennial energy crop	Grass for seed
(1) Fertilizer	kg N ha ^{−1} year ^{−1}	172	17–25	50	0	172	52	2
(2) Plant residues decomposition	kg N ha ^{−1} year ^{−1}	62	−40	−100	−20	0	14	−7
Indirect N ₂ O emissions (NH ₃ + NxOy)	kg N ha ^{−1} year ^{−1} = (1) * 6.2% + (1) * 1.2%	12	0	0	0	13	8	3
(3) Nitrogen leaching	kg N ha ^{−1} year ^{−1}	57	12–45	12–45	14	45	34–51	29
(4) Liming	kg C ha ^{−1} year ^{−1}	24	0	0	0	24	0	0
(5) Carbon storage	kg C ha ^{−1} year ^{−1}	0	270	270	135	0	180	790
Conversion to kg CO ₂ -eq ha ^{−1} year ^{−1}								
Direct N ₂ O from (1)	(1)*1%*416	716	71–104	208	0	716	217	8
Direct N ₂ O from (2)	(2)*1%*416	258	−167	−416	−83	0	58	−29
Indirect N ₂ O from (1) (NH ₃ + NxOy)	(1)*6.2%*1%*(44/28)*265 + (1)*1.2%*1%*(44/28)*265	53	0	0	0	54	16	11
Indirect N ₂ O from (3)	(3)*0.81%*416	192	40–152	40–152	47	152	115–172	96
Liming emissions (4)	(4)*(44/12)	88	0	0	0	88	0	0
Carbon storage (5)	(5)*(44/12)	0	990	990	495	0	660	2897
Sum/net effect		1308	934–1079	822–934	459	1010	1066–1123	2983

Emission factors (%) are from IPCC (2019), the Danish national emission inventory (Nielsen *et al.*, 2024, updated with data up to 2022) and other papers (see Adamsen and Manevski, 2025). Multipliers 265 and 416 are global warming potentials of, respectively, N₂O and CO₂ over 100 years.

Results and discussion

Soil C storage for intermediate crops was half of that for catch crops (0.26 Mg C ha^{−1}; Table 2; assumed from earlier estimates), slightly lower than 0.4 Mg C ha^{−1}, i.e., 12% of 3.3 Mg C ha^{−1} annually from catch crop embedded in the soil C pool after 30 years (Jensen *et al.*, 2022).

Annual soil C storage for grass for seed was assumed as half that for “productive grassland” as 1.58 Mg C ha^{−1} (imprecisely reported by Eriksen *et al.*, 2013), compared to 0.49 Mg C ha^{−1} (Olesen *et al.*, 2013). Either way, recent field studies from Denmark show conversion of cereal cropping to a crop rotation with 1/3 grass-clover to add 5.3 Mg C ha^{−1} with a new steady state after 20 years, equating to ca. 0.26 Mg C ha^{−1} year^{−1} (assuming linear increase, which is not true; Jensen *et al.*, 2024). Increased grassland proportion from 1/3 to 2/3 in the rotation resulted in 3.8 Mg C ha^{−1} with no sign of steady-state after 17 years, corresponding to 0.224 Mg C ha^{−1} year^{−1} (Jensen *et al.*, 2022). Conversion to

permanent grassland yielded $10.4 \text{ Mg C ha}^{-1}$ after 35 years, without assessment for a new steady state condition due to lack of data but nevertheless equating to $0.297 \text{ Mg C ha}^{-1} \text{ year}^{-1}$; Jensen *et al.*, 2024). The soil C storage values shown in Table 1 are overestimated for catch-/intermediate crops and for grass for seed. Also, related to steady state, the period of management change should be considered when annual C storage rates are estimated (Shang *et al.*, 2024).

Noteworthy is that time was not explicitly considered in the calculations: the effects are based on field experiments from different sites across Denmark or Europe, with different durations and often different field history. This also means carry-over effects are also not considered.

About the EFs (Table 2), tier-1 values are inaccurate (Yona *et al.*, 2022) as the IPCC was developed for global assessment of the scientific basis of climate change; yet most governments use them. For instance, 1% EF for N fertilizer was discriminated by the IPCC as 1.6% (uncertainty 1.3–1.9%) for synthetic and 0.6% (uncertainty 0.1–1.1%) for organic fertilizer (livestock manure), opposite to the new respectively values for Denmark of 0.3 and 1.2% (Petersen *et al.*, 2023). IPCC-based methods require continuous empirical validation, which needs research funds. Transfer to other regions is problem for many countries without “entrepreneurial” universities. Alternative could be a “quantitative storytelling” (Saltelli and Giampietro, 2017) to flag the existence of ‘uncomfortable knowledge’ usually avoided in policy discussions and control the quality of different solutions.

Conclusion

Plenty of empirical evidence for many processes involved in the C and N cycles of grasslands exists, but it is not necessarily integrated in grassland C and N profiles for policy instruments, either due to lack of methods to scale-up or simplify variability (both spatial and temporal), or simply lack of willingness to change agricultural systems. For the Danish case, a significant gap of knowledge remains for C allometry of grasslands regardless of their definition.

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Transforming grassland science for climate action: Design of a carbon farming curriculum

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Abstract

Grasslands play a pivotal role in carbon farming due to their botanical and agronomic traits, contributing significantly to soil carbon storage. To leverage this potential, the agricultural workforce must be equipped with tailored knowledge and skills. This work presents the first professional curriculum on carbon farming, developed in response to European sectoral needs and structured around the carbon cycle. It integrates scientific understanding, practical training, and business models to support rural entrepreneurs and green innovators in certifying carbon removals. The curriculum is designed for academic and vocational learners at European Qualifications Framework (EQF) level 5, combining three pathways: VOOC, MOOC, and VET. It is currently in the co-creation and pilot-testing phase with project partners, including universities, vocational institutions, farmer organizations, and advisory services, and will be freely accessible online once finalized. Communicating this work at EGF ensures early engagement, expert feedback, and strengthens the curriculum's relevance and impact for advancing carbon literacy and climate-smart agriculture across Europe.

Keywords: agroforestry, carbon cycle awareness, fertilization, nitrogen, soil, training

Introduction

Permanent grasslands cover 34% of EU farmland and store up to 90% of their carbon below-ground, and they also deliver higher, more stable biomass yields and improved soil C and N than annual crops as global demand intensifies (Chen *et al.*, 2022). Yet Europe's permanent grassland area continues to decline. Meeting this challenge calls for deeper insight into C processes, improved sink estimates, conservation of native grasslands, and scalable restoration strategies. Despite the recognized potential of grasslands for C sequestration, dedicated C-farming curricula remain scarce at both academic and professional level; grasslands must receive increased attention in both vocational and higher education. This paper presents a learner-centred curriculum on C-farming, grounded in social-efficiency principles, aiming to equip practitioners and educators with practical, climate-smart skills that serve broader societal and environmental goals.

Materials and methods

We designed the first professional Carbon Farming (C-Farming) curriculum to address skill gaps using EU frameworks such as EQF level 5, ECTS, micro-credentials, and Europass Digital Credentials. It is currently in co-creation and pilot-testing with universities, vocational institutions, farmer organizations, and advisory services across Europe, and will be freely accessible online once finalized. The curriculum combines three pathways: VOOC, MOOC, and VET for structured programmes and is organized into three units. Unit 1 covers the carbon cycle and soil carbon sequestration strategies

Table 1. Overview of the C-Farming curriculum modules and topics, with an emphasis on grasslands.

Module	Topic	Main content	Application to grassland systems
Module 1: Introduction to Carbon Farming	Carbon farming concept and importance; carbon cycle, sequestration and storage; nature-based solutions	Definition, EU Green Deal, CAP 2023–2027, climate mitigation; Carbon sequestration, terrestrial carbon cycle processes; Carbon links to N, P, water cycles; importance of carbon storage	Emphasizes soil carbon dynamics typical in permanent grasslands
Module 2: Sustainable Agricultural Approaches	Sustainable practices and carbon farming	Agroecology, conservation agriculture, agrobiodiversity, regenerative and organic agriculture, IPM, integration, circularity, energy efficiency	Includes grassland-friendly practices like regenerative grazing
Module 3: Carbon Farming Techniques	Carbon sequestration techniques; ghg emissions reduction methods	Agroforestry, integrated agrivoltaics, reduced tillage/inputs, cover crops, rotations, grazing, biochar, manure management; nitrification inhibitors, feed management, water and fuel efficiency	Covers re-vegetation, reduced tillage, and pasture management
Module 4: Markets, Policies and Incentives	Carbon trading and incentives	CAP 2023–2027, compliance and voluntary markets, carbon credits, EU certification, eligibility, digital tools	Discusses how grasslands can generate credible carbon offsets
Module 5: Soil Carbon MRV	Digital tools and MRV	Carbon impact tools, data management (GDPR), MRV systems	Remote sensing and soil monitoring methods used in grasslands
Module 6: Net Positive Regenerative Business	Building regenerative agriculture business	Net positive concept, transformation steps, risk management, ESG and CSR frameworks	Supports business models based on regenerative grazing, silvopastoral systems, native seed production, and carbon credit farming on permanent grasslands
Module 7: Leadership in Agriculture	Net positive and resilient leadership	Future of literacy, systems thinking, regenerative leadership, collaboration, storytelling	Fosters systems thinking for grassland restoration, climate-resilient grazing design, and multi-stakeholder coordination for long-term landscape regeneration.

(e.g., permanent grasslands, agroforestry) based on current science (Cai *et al.*, 2025). Unit 2 is learner-centred, focusing on best-known practices (Liu *et al.*, 2023). Unit 3 introduces business modules on MRV, carbon credits, and avoiding greenwashing. The curriculum targets five groups: farmers and agribusinesses (TG1), students and professionals (TG2), agricultural educators (TG3), sustainability trainers (TG4), and institutions involved in education and innovation (TG5).

Results and discussion

The curriculum (Table 1) addresses the urgent need for transformation in agriculture by promoting approaches to carbon farming that are net-positive in terms of C-storage and net-negative in terms of GHG emissions.

While it emphasizes ecosystem regeneration and long-term carbon accumulation (net-positive), it also integrates practices and tools aimed at achieving measurable reductions in greenhouse gas emissions (net-negative). Grassland management is placed at the centre of this dual strategy, serving as a key leverage point for climate mitigation and adaptation through soil-based solutions, biodiversity support, and resilient land use. It prepares future professionals to implement, monitor, and verify carbon

farming practices, contributing directly to EU climate targets with a consistent emphasis on grasslands. Table 1 serves as a baseline, with the syllabus to be validated through a multi-phase approach. This begins with a needs analysis targeting both university and non-university learners, followed by refinement through universities' prior experience with vocational training and integration of best practices and lessons learned from earlier initiatives. Input will also be collected from agricultural educators, practitioners and stakeholders across the food and farming system—whose activities influence the C cycle—via structured templates and open-ended surveys. This engagement will help ensure relevance to learners' real-world needs, practical alignment, and consistency with educational standards, while grounding the curriculum in real-world practices that affect carbon flows.

Conclusion

By integrating scientific insights, practical tools, and context-specific pedagogy—with a strong emphasis on grassland systems—the C-Farming curriculum prepares a new generation of agricultural professionals and stakeholders to adopt climate-smart practices. It offers a structured and replicable educational model that strengthens certification pathways, employability, and climate resilience. Continuous needs analysis, filtered through stakeholder input, scientific updates, and policy alignment, will ensure the curriculum evolves with emerging priorities and remains responsive to real-world challenges.

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Species-rich grasslands in intensive dairy farming: effects on herbage production and farmland birds

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Abstract

Increasing the number of plant species in grassland swards can improve herbage biomass production and enhance faunal biodiversity. Our project further tests these relationships in grasslands used for dairy production in Northwestern Europe, a region characterized by intensively managed grasslands and facing declining farmland bird populations and increasing occurrence of droughts. Data were collected from 20 dairy farms on sandy soils in Belgium, Germany, The Netherlands, and Sweden. Per farm, two grassland types (less and more diverse swards) were compared. Biomass was sampled 3–6 times per year, and biodiversity was assessed using botanical surveys and passive acoustic monitoring. Results from the first year of data collection indicate that species-rich fields produced slightly more herbage biomass than species-poor fields during the whole growing season. Grazing was associated with lower farmland bird richness, and a positive correlation between plant diversity and bird richness was observed only in Germany where the botanical composition between fields differed more.

Keywords: agricultural biodiversity, farmland birds, grasslands

Introduction

Intensively managed, species-poor grasslands have become widespread in Northwestern European dairy farming, but they negatively impact ecosystem services and biodiversity (Rigal *et al.*, 2023; Schils *et al.*, 2022). Experimental studies show that intensively managed species-rich swards can have equal or better herbage production (Dardonville *et al.*, 2020) while effects on bird diversity are mixed (Hoekstra *et al.*, 2023), yet on-farm data remain scarce. This study investigates production and bird richness of species-rich grasslands across the North Sea region in relation to sward plant diversity, management, and country.

Materials and methods

We present data collected on 20 dairy farms on predominantly sandy soils in Belgium, Germany, the Netherlands, and Sweden during one growing season (April to October 2024; Table 1). Each farm had two permanent grassland fields: one classified as species-poor (F1 in Table 1), typically composed of high-yielding ryegrass-dominated or grass-clover swards, and one classified as species-rich (F2 in Table 1), containing a higher number of forb and legume species (Table 1). Both fields were managed similarly in terms of cutting frequency and fertilization per farm. For each farmer's harvest, the herbage biomass (Dry Matter Yield, DMY) was sampled in six 0.25-m² plots by manual cutting at a height

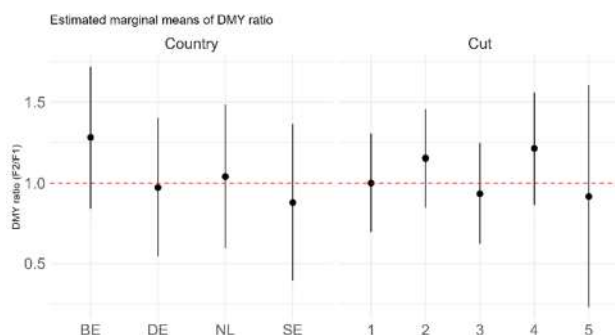


Figure 1. Mean dry matter yield ratio of species-rich (F2) to species-poor (F1) fields across countries and cuts with 95% CI. Values above 1 indicate higher yields in F2; values below 1 indicate higher yields in F1.

of 6 cm and dried at 60°C for 48 hours. Per farm, the DMY ratio (F2/F1) was calculated for each harvest (cut). Seasonal patterns were analysed using a linear mixed-effects model with cut and country as fixed effects and farm nested within country as random effect (lmer, R package *lme4*). Estimated marginal means for each cut and 95% confidence intervals were obtained using the *emmeans* package with Kenward-Roger degrees-of-freedom correction. Between April and June 2024, bird sounds were recorded using passive acoustic monitoring by audio recorders placed in the middle of the field (minimum distance between two recorders was 75 m, median was 500 m). Recording took place between 1 hour before and 3 hours after sunrise. Recordings from 34 fields on 17 farms were legible for analysis. Farmland birds were detected using the BirdNET 2.4 model with 0.5 confidence score (precision and recall being 95 and 23%, respectively). We used linear mixed effects models (lmer, *lme4* package) to assess whether farmland bird species richness (total number of species detected) was associated with plant diversity (Shannon–Wiener index), management (grazing (yes/no), fertilization (total amount of N, kg ha⁻¹), age of the field since the last ploughing (years), or field size (ha). The model included an interaction between plant diversity and country, and grazing as main effects, with farm identity as a random intercept. Competing models were compared using Akaike's Information Criterion (AIC), and fixed-effect significance was assessed with Satterthwaite's degrees of freedom (*lmerTest*). Country-specific slopes for the plant diversity–bird richness relationship were estimated using *emmeans*, with planned contrasts to compare each country's slope to the overall mean.

Results and discussion

Across all farms and cuts, species-rich fields produced on average 1.04 ± 0.12 (95% CI: 0.80–1.29) times the herbage biomass of species-poor fields. By country (Figure 1), Belgium showed the highest average ratio (1.28), while Sweden showed the lowest (0.88), which may be explained by species-rich fields in Sweden being more extensive than in the other countries. However, none of these country-level differences were statistically significant at the 5% level. The seasonal pattern (averaged across countries) was modest, with ratios ranging from 0.92 to 1.21 across cuts (Figure 1). For farmland bird species richness, model comparison using AIC indicated that the best-fitting model (AIC = 166.5) included plant diversity, its interaction with country, and grazing presence. This model outperformed the null model (AIC = 189.3) and all other candidate models which included age, size and fertilization levels. The model explained 39% of the variance in farmland bird richness through fixed effects (marginal $R^2 = 0.39$), and 76% of the total variance when including farm-level differences (conditional $R^2 = 0.76$). There was a significant negative effect of grazing on farmland bird species richness across countries: grazed fields hosted on average 3.62 fewer species than ungrazed ones ($p = 0.034$).

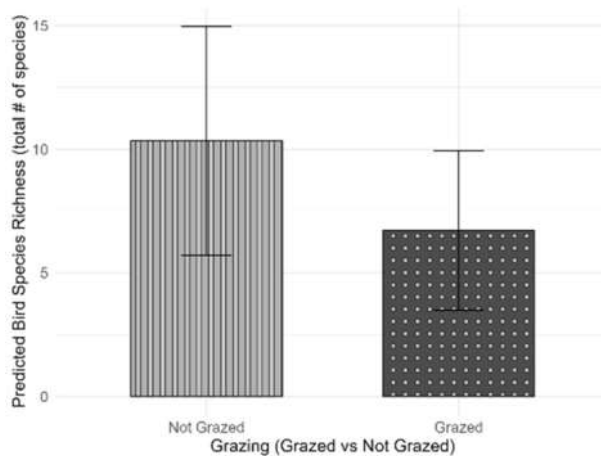


Figure 2. Predicted farmland bird species richness in grazed vs. ungrazed fields, based on a mixed-effects model (fixed effects: plant diversity, country×diversity interaction, grazing; random effect: farm).

Table 1. Mean ($\pm$ SD) values of field characteristics for species-poor (F1) and species-rich (F2) grasslands in each country

	Field type	Shannon index $\pm$ SD	Δ Shannon	Age (years) $\pm$ SD	Δ Age	Size (ha) $\pm$ SD	Δ Size	Total N (kg/ha) $\pm$ SD	Δ N
Belgium	F1	2.92 $\pm$ 0.71		24.2 $\pm$ 42.5		2.76 $\pm$ 2.46		134 $\pm$ 70.6	
	F2	3.42 $\pm$ 0.85	0.50	16.2 $\pm$ 24.5	-8.0	3.21 $\pm$ 4.06	0.45	164 $\pm$ 108	30
Germany	F1	1.35 $\pm$ 0.24		27.4 $\pm$ 29.9		2.62 $\pm$ 1.53		284 $\pm$ 134	
	F2	3.11 $\pm$ 0.62	1.76	50 $\pm$ 22.4	22.6	2.18 $\pm$ 1.29	-0.44	199 $\pm$ 136	-85
The Netherlands	F1	2.57 $\pm$ 1.50		5.2 $\pm$ 3.6		2.91 $\pm$ 2.12		232 $\pm$ 149	
	F2	4.16 $\pm$ 1.54	1.59	3.6 $\pm$ 1.1	-1.6	2.73 $\pm$ 0.85	-0.18	247 $\pm$ 172	15
Sweden	F1	3.55 $\pm$ 0.74		27 $\pm$ 41.4		11.93 $\pm$ 16.0		74 $\pm$ 86	
	F2	5.16 $\pm$ 1.90	1.61	61.4 $\pm$ 39.4	34.4	11.97 $\pm$ 10.4	0.04	0 $\pm$ 0	-74
Average	F1	2.6 $\pm$ 1.18		21 $\pm$ 32		5.06 $\pm$ 8.56		181 $\pm$ 135	
	F2	3.96 $\pm$ 1.47	1.36	32.8 $\pm$ 33.9	11.8	5.02 $\pm$ 6.62	-0.04	153 $\pm$ 147	-28

Δ = mean difference (F2-F1).

(Figure 2). The effect of plant diversity on bird richness was country-specific, being significantly positive in Germany but not elsewhere (estimate = 3.93, SE = 1.24, p = 0.005), which may be explained by the larger plant diversity contrast between species-rich and species-poor fields in Germany (Table 1). This suggests that where grassland plant species richness differences are more pronounced, associated benefits for farmland bird become more detectable.

Conclusion

In 2024, species-rich fields produced slightly more (factor 1.04) herbage biomass than species-poor fields, with no strong evidence of systematic differences across cuts or countries. The wide confidence intervals indicate high variability between farms which may be explained by different contrasts in plant diversity between fields in different countries. Our findings suggest that, under intensive dairy management, species-rich grasslands can achieve herbage biomass yields comparable to species-poor grasslands throughout the growing season. The presence of grazing was associated with lower farmland bird richness across countries, while a positive association between plant diversity and bird richness was observed in Germany, suggesting that biodiversity benefits are context-dependent.

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Mind the gaps: towards better vegetation data for grassland ecosystems

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Abstract

European grasslands rank among the most species-rich ecosystems globally, supporting exceptional plant and animal diversity while delivering key ecosystem services. Yet, these habitats are increasingly threatened by both land-use intensification and abandonment. Effective public policies are crucial for maintaining grassland multifunctionality but face a critical obstacle: the lack of harmonized, high-quality baseline vegetation data. Using Switzerland as a case study, we identify key data gaps and systemic shortcomings that hinder ecological research and policymaking. Despite Switzerland's nationwide farmland biodiversity monitoring programme, its recent implementation leaves a vacuum of long-term reference data. We highlight six priority areas for action: (1) improving the availability and resolution of land-use, soil, and climate data; (2) ensuring full and standardized metadata with consistent taxonomies; (3) enhancing access through dataset indexing and open publication mandates; (4) addressing sampling and observer biases through coordinated, multi-scale surveys; (5) supporting field logistics and fallback site planning; and (6) securing long-term institutional commitment to monitoring. These tools are essential for adaptive policies at national and European scales that sustain grassland biodiversity.

Keywords: baseline data, biodiversity, data harmonisation, environmental policy, land-use

European grasslands are threatened biodiversity hotspots

European grasslands are among the most species-rich ecosystems in the world (Wilson *et al.*, 2012). They emerged from centuries of extensive agriculture by diverse cultural groups who settled once-forested regions (Van Der Knaap *et al.*, 2000). These communities first cleared the climax vegetation—primarily forest—and, through continuous low-level disturbance and facilitated species dispersal, enabled the development of complex grassland mosaics. As a result, semi-natural grasslands became biodiversity hotspots, supporting high plant and animal diversity and providing key ecosystem services such as forage, pollination, and recreation.

Semi-natural grasslands—shaped and maintained through traditional agricultural practices—are increasingly threatened by both land-use intensification and abandonment. Rising global demand for meat and dairy drives intensified fodder production on accessible land and abandonment of less productive sites. This shift undermines the ecological integrity and cultural significance of species-rich grasslands (Bengtsson *et al.*, 2019). In northern Europe, more than 90% of semi-natural grasslands have been lost since the 1930s (Bullock *et al.*, 2011; Pe'er *et al.*, 2014), with similar trends reported for Switzerland (BAFU, 2017). In response, agricultural payment schemes started to be introduced across Europe and in Switzerland beginning in the early 1990s (European Communities, 1985), aiming to preserve these habitats for their biodiversity, ecosystem services, and cultural value.

Table 1. Key challenges in grassland vegetation survey data and their implications

Specific challenge	Implications	Remedy
Data availability: missing or coarse land-use, soil, climate, and microclimate layers	Limits ability to attribute change to environmental drivers	Institutions: develop high-resolution spatial layers. Researchers: interview land managers; collect soil and microclimate data.
Data access: lack of indexing, poor discoverability, undigitized datasets	Prevents data synthesis, reuse, and integration	Institutions: mandate open-access data deposition in journals. Researchers: register and index existing datasets, including legacy data.
Metadata: missing coordinates, plot area, methods; inconsistent taxonomy	Weakens comparability and ecological inference	Institutions: enforce metadata standards and taxonomic alignment protocols. Journals: require full metadata on submission.
Sampling bias: observer effects, inconsistent plot area, under-sampled gradients	Inflates uncertainty; limits generalizability across regions and systems	Institutions: coordinate spatially balanced sampling schemes. Researchers: implement multi-scale, repeated surveys; ensure protocol consistency.
Logistical barriers: difficult terrain, restricted access, private land	Limits monitoring in ecologically important areas	Institutions: allocate dedicated funding for field logistics. Researchers: define and maintain fallback or alternative plots.
Institutional neglect: short-term project cycles; low political prioritization	Undermines continuity and investment in monitoring	Funders and policymakers: allocate a fixed share of research budgets to long-term monitoring infrastructure.

Lack of baseline data hampers interpretation of trends

A major obstacle to evaluating the effectiveness of agri-environmental measures is the lack of harmonized, high-quality vegetation data. This challenge has two main dimensions. First, robust monitoring programmes are absent in many of Europe's most species-rich regions. Such programmes exist in northern Europe, but southern and eastern regions remain largely uncovered (Herzog and Franklin, 2016). Second, there is a widespread lack of baseline data—that is, standardized vegetation surveys that document species composition and abundance before major changes in land-use or biodiversity-related policies. With the exception of the UK, most programmes began around 2000 (Herzog and Franklin, 2016), shortly after the introduction of biodiversity-related payments. Even where older vegetation surveys exist, they are often inconsistently formatted, incomplete, and difficult to access. As a result, valuable information that could at least partially fill this gap remains underutilized. Additionally, the lack of linked land-use, edaphic, and (micro-)climatic variables further undermines interpretability.

Switzerland offers a valuable case study to illustrate these challenges. It is one of the few countries with a large-scale, high-resolution farmland biodiversity monitoring programme (www.all-ema.ch), which evaluates whether agri-environmental policies are effective. However, the programme was only launched in 2015—too recent to provide baseline data needed to assess biodiversity change over decades. Despite a long history of vegetation research, interpreting trends in Swiss grasslands remains difficult. Earlier surveys are unevenly distributed, inconsistently formatted, and difficult to access.

The fragmented and inconsistent state of vegetation data (Table 1) not only hinders ecological research but also weakens the foundation for evidence-based policy making. Without reliable information, it becomes difficult to define meaningful biodiversity targets for ecological focus areas, determine appropriate grazing intensities, or assess multiple ecosystem services in parallel. As a result, policies risk being misdirected—subsidies may be ineffective or poorly targeted, and the multifunctionality of ecosystems may be compromised. Addressing these challenges requires coordinated, long-term institutional efforts. Biodiversity monitoring programmes must be sustained and expanded to ensure continuity in trend detection.

Toward open and standardized vegetation survey data

Existing vegetation data remains non-standardized and inaccessible. Initiatives like the Global Biodiversity Information Facility (www.gbif.org) highlight the potential of open-access platforms for species records. A similar infrastructure is urgently needed for vegetation survey data, linking species lists, metadata, and sampling protocols with land-use and policy information. Though requiring political support, such efforts are essential for evidence-based policy. While institutional reforms are central, researchers can already improve data quality and comparability by providing standardized metadata—such as plot area and species list completeness—and incorporating multiple plot areas to enhance comparability (see e.g. Dengler *et al.*, 2021). In fact, despite progress through initiatives like the European Vegetation Archive (Chytrý *et al.*, 2016), metadata remain underreported. Lastly, analyses need to address survey biases—detection and observer effects—that can be estimated statistically.

Conclusion

Improving vegetation data for grasslands is both a scientific and institutional opportunity. Despite challenges in standardization and access, there is growing momentum toward coordinated, open, and long-term approaches. Rural development programmes and agri-environmental subsidies require a reliable monitoring foundation to be effective. National monitoring, collaborative platforms, and well-documented surveys are key. This effort is more urgent than ever, as climate and land-use pressures intensify across Europe—particularly in southern European countries, where many of the most species-rich grasslands remain under-monitored. Strengthening the data foundations now is essential to ensure that policy and conservation efforts can keep pace with accelerating ecological change.

Acknowledgements

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Selecting study areas representative for the highly diverse growth conditions in the Swiss summering zone

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Abstract

Site-specific management at appropriate stocking rates is essential to maintain ecosystem services of mountain pastures. However, there is no common agreement on what an appropriate stocking rate is. Thus, in 2022, Agroscope launched the Alpine Rangeland Observation Network to create reference values for forage production, forage quality and non-provisioning ecosystem services. In this paper, we present the methodology by which we ensured that the network represents all relevant conditions of the highly heterogeneous Swiss mountain pastures: We (i) mapped the distribution of mountain summer pastures at hectare-level based on Swiss land use statistics and (ii) grouped these pastures by climate region and dominant bedrock type. 14 study areas (i.e. in most cases one summer farm) were selected to cover the most important climate–bedrock combinations, and to be representative in elevation and summer precipitation of the broader climate–bedrock combination(s). Thanks to this selection process, we are able to represent almost 90% of the Swiss summering zone.

Keywords: ecosystem services, mountain pastures, stocking rate, study area selection

Introduction

Transhumant summer farming has shaped Swiss mountain landscapes for centuries. Today, mountain summer pastures, which cover one-third of Swiss agricultural land, host over 400 000 cattle and 200 000 sheep and goats each summer (Federal Office for Agriculture, 2023). Beyond forage production, these ecosystems provide important regulating and cultural services, including biodiversity conservation, carbon storage, and recreation (Pauler *et al.*, 2025). The summering zone is extremely diverse, spanning elevations from 400 to 3000 m a.s.l., and average summer precipitation from 250 to 1220 mm (swisstopo, 2024; MeteoSwiss, 2021). However, global change increasingly threatens their ecological integrity, making sustainable management, such as site-specific stocking rates, essential to sustain the ecosystem services they provide. To improve the basis for sustainable management strategies, Agroscope launched the Alpine Rangeland Observation Network (ARON) in 2022. Developing an assessment framework requires study areas that capture the ecological diversity of mountain summer pastures as a representative basis for measurements. Here, we present the systematic and comprehensive selection process of the ARON study areas that represent the major ecological gradients of the Swiss summering zone.

Materials and methods

We mapped mountain summer pastures at hectare scale using the category “Summer farming areas” of the 2018 Swiss land use statistics (Federal Statistical Office, 2020) and grouped them by climate regions and dominant bedrock types (Begert *et al.*, 2007; Szerencsits *et al.*, 2009). We identified key climate–bedrock combinations and selected study areas (in most cases one summer farm) within

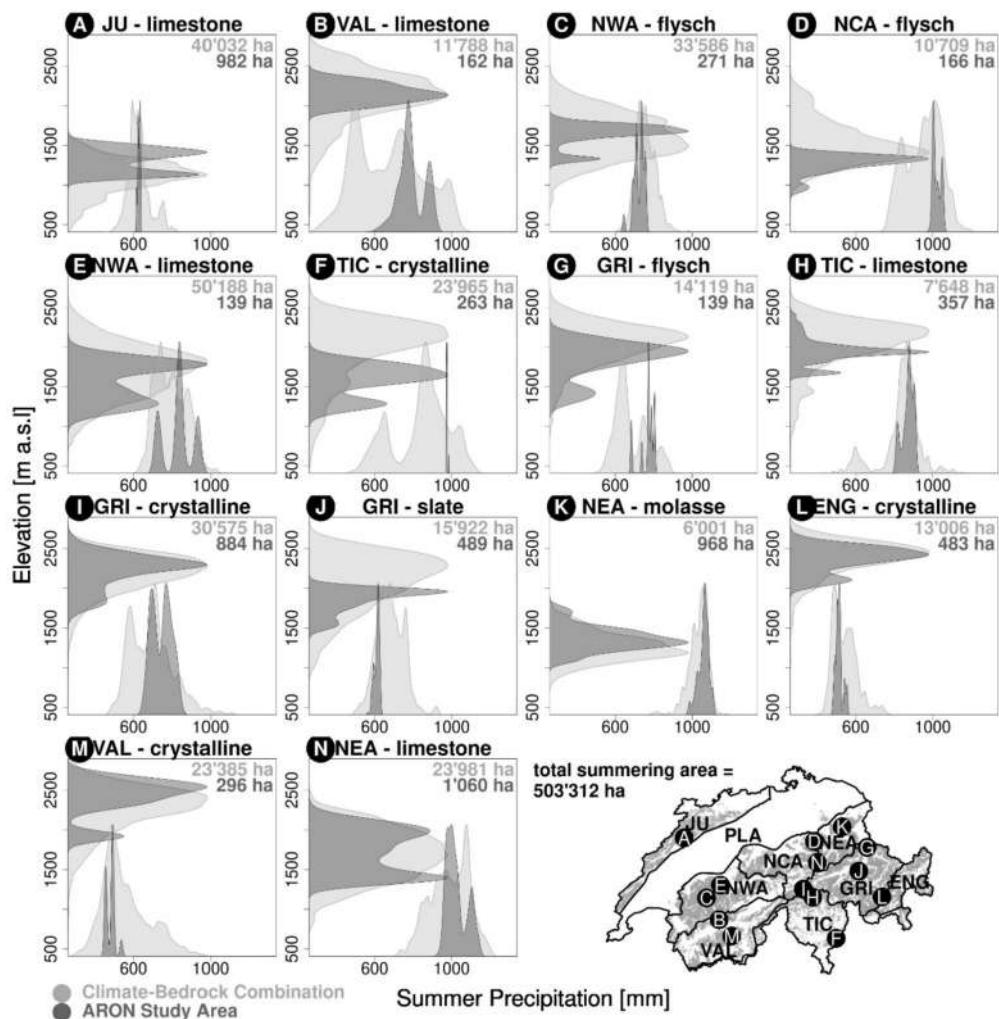


Figure 1. Density plots showing the distribution of elevation (y-axis) and summer precipitation (x-axis, May–September) for the selected climate–bedrock combinations (light grey, over Switzerland) and the corresponding ARON study areas (dark grey). Bottom right: Map of Switzerland showing the summering area in grey, climate regions outlined in black, and the locations of the ARON study areas. (black dots).

them based on representative elevation distributions (swisstopo, 2024) and summer precipitation (1991–2020; MeteoSwiss, 2021). We tested representativeness by comparing the distributions of elevation and precipitation in the study areas against those of their broader climate-bedrock combinations. Candidate areas were further evaluated for slope and vegetation diversity to choose those which best represented the growth conditions of Swiss summer pastures.

Results and discussion

The Swiss summering zone covers 503 312 ha, primarily located in the Alps and the western Jura (Figure 1). The majority (99.1%) is distributed across eight main climatic regions: Northern and Central Grisons (GRI; 23.3%), North-Western Alpine Foothill (NWA; 20.7%), North-Central Alpine Foothill (NCA; 11.7%), Valais (VAL; 10.9%), North-Eastern Alpine Foothill (NEA; 8.9%), Jura (JU; 8.6%), Engadine (ENG; 7.6%) and Southern Alps (TIC; 7.4%). The Swiss Plateau accounts for

only 4,240 ha (PLA; 0.8%) and was excluded from further analysis. From a geological perspective, summering pastures are predominantly situated on limestone (38.9%), followed by crystalline rock (20.1%), flysch (14.1%), slate (6.3%), and molasse (3.6%). Verrucano (2.3%) was excluded due to limited extent, while gravel (14.6%) was not considered separately, as it represents a mix of local bedrock types. Combining climatic regions and bedrock types yielded 20 distinct combinations each covering more than 5000 ha. Based on their extent and relevance, a subset of 17 of these combinations was selected for inclusion in the ARON.

Elevation varies widely by climate region, with the highest areas in the Engadine, Valais, and Southern Alps, and the lowest in the Jura and Prealps. Average summer precipitation (1991–2020) differs substantially, with the North-Eastern Alpine Slope being wettest and the central alpine valleys Engadine and Valais driest. To represent dominant ecological conditions, 14 study areas (139–1060 ha) were selected based on elevation and summer precipitation. The elevation and summer precipitation distributions of the study areas closely correspond to those of the broader climate–bedrock combinations they represent (Figure 1; comparison black and grey density plots). Three areas (B, K, N) span two climatic regions due to their transitional locations.

Conclusion

The systematic selection of 14 representative study areas across key climate–bedrock combinations offers a framework to support the revision of forage yield values and integration of broader ecosystem services into summer pasture management. Together, the study areas cover climate–bedrock combinations that make up over 87% of the summering zone and closely match their respective combinations in terms of summer precipitation and elevation distributions. By capturing the major environmental gradients of Switzerland's summering zone, ARON enables a more accurate, site-specific understanding of pasture productivity under changing conditions.

Acknowledgement

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Assessing the sustainability progress in Romanian grazing farms using the Grazing4AgroEcology farm self-assessment tool

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Abstract

Sustainability research has seen significant advances in recent years, with innovative solutions to reduce human environmental pressures. Recent efforts under the EU Green Deal have focused on reducing the environmental costs of food production. Agroecology has proved to be a promising farming practice that works with nature to meet food security demands. Moreover, grazing systems may bring benefits to both farms and society. This study specifically assesses the sustainability progress of grazing farms in Romania, focusing on their agroecological performance within a national context. It employs the Grazing4AgroEcology (G4AE) farm self-assessment tool, which was developed to assist farmers in self-assessing qualitative indicators across five agroecological pillars: improving animal health, reducing inputs, enhancing diversity, reducing pollution, and preserving biodiversity. The methodological approach combines on-farm data collection with qualitative indicator scoring and comparative analysis between Romanian and EU farms, allowing a spatially grounded understanding of agroecological progress. The assessment revealed that the sampled Romanian farms show notable weaknesses in input efficiency and pollution reduction. Detailed feedback and improvement pathways were discussed with participating farmers. These findings suggest that targeted advisory services, educational programmes, and continued policy support are crucial to strengthen the agroecological transition in Romanian grazing systems.

Keywords: agroecology, farm self-assessment tool, grazing

Introduction

Sustainability research has known important developments in the last decade. It emphasises appropriate innovative solutions such as how to reduce human environmental pressures in a socially acceptable approach. In the EU, there were many initiatives aiming to reduce the environmental costs of food production under the latest EU Green Deal commitments. Agroecology has proved to be a promising practice that can enable farming to work with nature to meet food security demands (Wezel *et al.*, 2009). Grazing can also bring positive effects on the farm economy and also for society (Van den Pol-van Dasselaar *et al.*, 2020).

Consequently, there has been a growing interest in developing different tools to assess the overall farm sustainability. Frameworks like the Sustainability Assessment of Food and Agriculture systems (SAFA), Response-Inducing Sustainability Evaluation (RISE), Multi-attribute Assessment of Sustainability of Cropping Systems (MASC), Land Degradation Assessment in Drylands (LADA), the public goods (PG) etc. vary in terms of their indicators, data requirements, goals, and sustainability aspects (Alaoui *et al.*, 2022). Moreover, the Grazing4AgroEcology thematic network highlighted a lack of farmer-friendly approaches that made the existing frameworks inoperable at the level of farmer knowledge. To address this issue, a self-assessment tool was developed within the G4AE thematic network context.

Materials and methods

The G4AE farm self-assessment tool (Looney *et al.*, 2024) was developed by a broad network of experts who identified and structured qualitative indicators from the scientific literature, aligning them with five core agroecological principles as defined by Dumont *et al.* (2013). These principles and their associated indicators are as follows:

- (1) Improving animal welfare, assessed across five domains: infrastructure quality (7 indicators), nutrition (2), health management (6), animal behaviour (1), and calf welfare and health (4);
- (2) Reducing inputs and pollution, evaluated through indicators related to maximising herbage utilisation (14), reducing feed inputs (5), soil management (6), optimising on-farm organic nutrient use (4), and optimising fertilisation practices (6);
- (3) Enhancing diversity, measured via indicators of production diversity (17), marketing diversity (14), and income diversification (1);
- (4) Preserving biodiversity, assessed through 28 indicators focusing on habitat management.

The indicators were refined through a series of expert consultations and validated through on-farm testing to ensure clarity, relevance, and ease of use. The assessment was conducted through face-to-face interviews between May and July 2024 on a sample of 15 Romanian farms. They are part of the G4AE Partner Farm Network (PFN), which includes 15 grazing-based farms in each of the eight participating countries (Ireland, France, Italy, Portugal, Germany, Sweden, The Netherlands, and Romania), amounting to a total of 120 farms. The average scores were compared with the aggregated averages of farms across the EU.

Results and discussion

The analysis of the sampled Romanian farms reveals critical deficiencies in agroecological performance, particularly in the areas of input use efficiency and pollution reduction (Figure 1). These dimensions are central to sustainable agriculture, as they directly influence environmental outcomes, economic viability, and long-term resource conservation.

An important structural barrier to improved agroecological performance is the low level of formal education. According to Eurostat (2022), a substantial portion of the Romanian farming population has only basic or vocational education, with limited access to ongoing knowledge in sustainable agriculture. This knowledge gap restricts the uptake of innovative and environmentally sound practices, such as precision agriculture, organic methods, and conservation agriculture. Previous research has also consistently demonstrated a strong need for better advisory services (Jitea *et al.*, 2021).

While the study provides valuable insights into current agroecological practices, it is important to acknowledge its limitations. The relatively small sample size limits the generalizability of the findings to the wider population of Romanian grazing systems. Furthermore, the reliance on qualitative indicators, while useful for farmer engagement, introduces a degree of subjectivity that should be considered when interpreting the results. Despite these constraints, the comparative approach employed—contrasting Romanian farm data with EU-level averages—offers a meaningful understanding of regional disparities in agroecological transition. The results underline the need for context-specific support mechanisms, adapted to local socio-economic and educational realities.

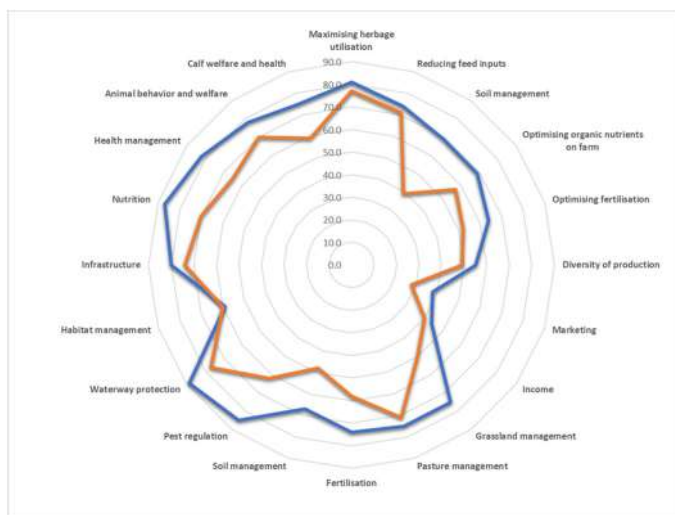


Figure 1. Overall agroecological performance of the Romanian farms (brown line) compared to all EU farms (blue line).

Conclusion

The results highlight serious agroecological flaws in the Romanian farms, especially with regard to pollution control and input usage efficiency. The low level of formal education and restricted access to ongoing training among Romanian farmers are two structural variables that are directly associated with these deficiencies. Adoption of sustainable methods is hampered by a chronic knowledge gap and a lack of advisory services, endangering long-term agricultural sustainability as well as environmental quality. Integrated policy initiatives aimed at improving agricultural education, fortifying extension networks, and encouraging knowledge-based shifts toward more sustainable farming practices are needed to address these issues. These findings emphasize that addressing sustainability challenges in Romanian grazing systems requires more than technical solutions—it demands systemic change supported by education, policy, and institutional collaboration. Integrated policy initiatives should focus on strengthening agricultural education, fortifying extension networks, and fostering participatory learning environments where farmers can exchange knowledge and co-develop agroecological innovations. Future research should expand the sample scope, include longitudinal monitoring to capture progress over time, and integrate quantitative indicators.

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Exploring communication channels tailored to farmers' needs in eight European countries

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Abstract

Agricultural knowledge and innovations frequently fail to reach end-users effectively, resulting in fragmented dissemination and ineffective application by farmers and other stakeholders. This study, part of the Horizon Europe project Grazing4AgroEcology, aims to identify the most efficient communication channels and tools in the grazing sector tailored to farmers' needs across eight European countries (France, Germany, Ireland, Italy, The Netherlands, Portugal, Romania and Sweden). Findings of a web-based survey with 443 responses indicate that access to reliable, up-to-date, and context-specific information is crucial for the successful uptake of agricultural knowledge. The end-users seek ready-to-use knowledge on agronomic and animal husbandry practices, technologies, grazing management and rely on a variety of sources to stay informed. Face-to-face interaction, peer-to-peer learning and internet emerged as the most effective sources of information. A multi-channel approach that also includes consultancy by extension services, digital tools and demonstration farms can further enhance the adoption of best practices. A comparative analysis across countries provides important insights for policymakers and other actors aiming to sharing information within the Agricultural Knowledge and Innovation Systems at both national and international levels.

Keywords: AKIS, communication channel, grazing

Introduction

Agricultural knowledge and innovation are essential in agriculture and rural areas to address challenges such as climate change, environment protection, and circular economy. Improving knowledge flows can accelerate innovation adoption by downstream industry actors such as farmers, small rural industries, and advisors. The Agricultural Knowledge and Innovation Systems (AKIS) should play a key-role in the flow of knowledge to farmers, but their structure varies across countries, with robust and cohesive systems in, e.g., Ireland, Austria and Denmark, and weaker and fragmented systems in, e.g., Greece, Portugal and Romania (EU SCAR, 2019). However, agricultural knowledge and innovations often fail to reach end-users effectively, resulting in fragmented implementation at farm and rural community level (Oost *et al.*, 2020). Furthermore, little is known about the efficiency of dissemination channels in the process of innovation adoption in the downstream industry (Pathak *et al.*, 2019). This study aims to reveal the most efficient communication channels and tools for disseminating agricultural knowledge across eight European countries.

Table 1. Relevance of grazing knowledge to farmers' needs in France (FR), Germany (DE), Ireland (IE), Italy (IT), The Netherlands (NL), Portugal (PT), Romania (RO) and Sweden (SE) (% of respondents).

Agricultural knowledge	FR (<i>n</i> = 41)	DE (<i>n</i> = 61)	IE (<i>n</i> = 65)	IT (<i>n</i> = 61)	NL (<i>n</i> = 56)	PT (<i>n</i> = 30)	RO (<i>n</i> = 55)	SE (<i>n</i> = 54)	<i>p</i> -value
Grazing management—technical issues (e.g. forage management, grazing systems,)	100*	80.3	96.9*	65.6	82.1	83.3	47.3*	85.2	<0.001
Animal husbandry practices and technologies	92.7	83.6	81.5	68.9	89.3	73.3	67.3	81.5	<0.01
Grazing management—environmental issues (e.g. biodiversity, gas mitigation, etc.)	78.0	85.2	83.1	63.9	73.2	76.7	60.0*	88.9	<0.01
Weather forecast	65.9	45.9	80.0*	47.5	39.3	43.3	30.9*	55.6	<0.001
Agronomic practices and technologies	73.2	50.8	52.3	52.5	73.2	63.3	50.9	70.4	<0.05
Market information (e.g. prices)	48.8	57.4	52.3	65.6	33.9*	63.3	60.0	57.4	<0.05
Management of animal welfare	56.1	70.5	56.9	59.0	35.7*	50.0	47.3	64.8	<0.05
Input information (e.g. availability, source, price)	48.8	50.8	61.5	44.3	35.7	53.3	47.3	55.6	0.199
Financial information (e.g. subventions, credit availability)	19.5*	44.3	40.0	27.9	37.5	50.0	70.9*	48.1	<0.001
Warning systems about diseases and pests	26.8	29.5	27.7	24.6	16.1	56.7*	34.5	40.7	<0.01
Wild animal management	12.2	36.1	20.0	18.3	16.1	40.0	27.3	53.7*	<0.001

Asterisks (*) indicate values significantly different from at least one other group ($p < 0.05$, Bonferroni-adjusted post hoc test). In some cases, Bonferroni-adjusted pairwise comparisons found no significant differences despite a significant effect of the country, suggesting the occurrence of small, dispersed differences across countries.

Materials and methods

A web-based survey was conducted from November 2022 to March 2023 in eight European countries (France, Germany, Ireland, Italy, The Netherlands, Portugal, Romania and Sweden) to identify the needs of relevant grassland knowledge and the most efficient communication channels and tools used in the grazing sector by farmers. The questionnaire was translated into local languages and distributed nationally, yielding 423 valid responses in total. The number of responses per country ranged from 30 to 65. Descriptive (percentages) and inferential statistics (Pearson's chi-square test with Bonferroni post-hoc test) were used for data analysis. Statistical significance was set at $P \leq 0.05$.

Results and discussion

Most topics of grazing knowledge were commonly relevant for farmers, despite the emergence of distinct patterns across countries (Table 1). All aspects about grazing knowledge apart from input information were affected by the factor country. Grazing management, both technical and environmental, was a top priority in most countries, except for Romania and Italy. Animal husbandry practices and technologies were consistently important, as were agronomic practices and technologies. Weather forecasting was relevant for Ireland, while Romania and the Netherlands ranked lowest. Knowledge on animal welfare was considered less relevant to farmers' needs in The Netherlands, Romania, and Portugal. Sweden expressed the highest need for knowledge on wild animal management among all countries. Financial information, such as about subsidies and credit options, is in high demand in Romania, while France reported less need for such resources.

Face-to-face interaction, peer-to-peer learning, and internet access were identified as the most effective sources of information (Table 2). In Ireland and Portugal, advisory services were particularly relevant, but France and Italy relied less on them. Traditional methods like phone/SMS and TV/radio are no

Table 2. Ways of acquiring agricultural knowledge relevant to farmers' needs in France (FR), Germany (DE), Ireland (IE), Italy (IT), The Netherlands (NL), Portugal (PT), Romania (RO), and Sweden (SE) (% of respondents).

Ways to acquire agricultural knowledge	FR (n = 41)	DE (n = 61)	IE (n = 65)	IT (n = 61)	NL (n = 56)	PT (n = 30)	RO (n = 55)	SE (n = 54)	p-value
Face-to-face interaction with advisors, researchers, farmers, technical people, etc.	80.5	91.8	93.8	82.0	85.7	83.3	74.5	77.8	0.062
Peer-to-peer learning (learning from peers/colleagues)	90.2	78.7	95.4*	70.5	66.1	76.7	61.8*	88.9	<0.001
Internet	78.0	88.5	64.6	68.9	87.5	83.3	83.6	75.9	<0.05
Printed media (e.g. magazines, newspapers, brochures, professional journals)	63.4	83.6*	61.5	50.8	75.0	40.0	30.9*	75.9	<0.01
Advisory services	14.6*	39.3	69.2*	24.6*	50.0	63.3	32.7	46.3	<0.01
Training/education	36.6	42.6	55.4	39.3	28.6	70.0	43.6	59.3	<0.01
Scientific journals	65.9	67.2	64.6	39.3*	58.9	63.3	47.3	57.4	<0.05
Social media platforms	22.0	31.1	43.1	16.4	32.1	33.3	29.1	31.5	0.090
Phone calls/SMS	9.8	34.4*	26.2	13.1	8.9	23.3	16.4	27.8	<0.01
TV/radio broadcast	7.3	6.6	10.8	27.9*	10.7	13.3	18.2	3.7	<0.01

Asterisks (*) indicate values significantly different from at least one other group ($p < 0.05$, Bonferroni-adjusted post hoc test). In some cases, Bonferroni-adjusted pairwise comparisons found no significant differences despite a significant effect of the country, suggesting the occurrence of small, dispersed differences across countries.

longer widely considered essential. Printed media are still important in Germany, Sweden, and The Netherlands, while being less used in Romania.

Conclusion

Effective knowledge transfer in the grazing sector depends on communication channels tailored to national contexts and farmers' needs. Respondents identified face-to-face interaction, peer-to-peer learning, and internet access as the most effective information sources; however, a multi-channel approach that also incorporates consultancy from extension services, digital tools, and demonstration farms could enhance the adoption of best practices.

Access to reliable, up-to-date, and context-specific information remains essential for effective knowledge uptake. Cross-country comparisons provide valuable insights for policymakers and stakeholders involved in strengthening AKIS at both national and international levels.

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Structural changes in Norwegian dairy farming have led dairy cows away from grassland

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Abstract

Over the past 20 years, the structure of dairy farming in Norway has changed significantly. A survey of 95 dairy farms that had expanded their milk quotas showed an average increase in annual milk delivery of 107 207 l and a rise in milk yield per cow from 6777 to 7463 l. Meeting the demands of larger quotas and herds require more forage, achieved through expanding farmland or increasing yields. However, land expansion lagged herd growth, leading to a 14% increase in livestock density, from 1.27 to 1.46 LU ha⁻¹, and a 25% increase in milk delivery per ha. This intensification also led to greater reliance on purchased concentrate feed. While forage yield per ha tended to increase, the net energy from farm-produced forage per LU decreased from 2982 to 2743 feed units, indicating an 8% reduction in forage contribution to the diet. The concentrate share in the diet rose by 5 percentage points. Despite these changes, herd structure remained stable, with dairy cows consistently comprising 62% of the herds' total livestock. Overall, structural rationalization has resulted in larger, more efficient operations with higher productivity per unit of land and animal, but reduced reliance on forage.

Keywords: dairy farming, diet, forage, intensity

Introduction

The structure of dairy production in Norway has changed markedly over the past two decades. From 2003 to 2024, the number of producers declined by 65%, from 17 769 to 6268, the dairy cow population decreased by 28%, from 282 200 to 202 770 cows, while the total annual quota of milk produced was reduced only by 6%, from 1522 Ml to 1438 Ml. The average herd size remained stable throughout the 1980s and 1990s, largely due to the introduction of a non-tradable, non-leasable milk quota system in 1983. Structural change accelerated following the liberalization of quota trade in 2003 and the introduction of quota leasing in 2009, aimed at improving capacity utilization, particularly in joint farming operations. Additional drivers of change include the mandatory loose housing requirement of cattle and the adoption of automatic milking systems (AMS), both of which have spurred farm expansion and new construction. Prior to 2003, milk quota regulations were primarily used to manage overproduction through government-controlled pricing and reallocation, rather than to facilitate redistribution between farms (Flaten, 2002a). The rationale behind the liberalization of quota trade in 2003 and introducing leasing of base quotas in 2009 was to improve capacity utilization, particularly in joint farming operations. One outcome of the quota liberalization was a doubling of average herd size, from 15.9 cows in 2003 to 32.4 in 2024. Over the same period, data from the Norwegian dairy herd recording system (NDHRS), which includes 98% of all dairy herds, show an average annual increase in milk yield per cow of 95 kg, from 6134 to 8230 kg (TINE, 2025). Higher milk quotas, larger herds, and increased milk yield per cow require more feed. This can be achieved through expanding agricultural land, improving forage yield and quality, or increasing the proportion of concentrate in the diet. According to NDHRS, the dietary net energy contribution from concentrate rose from 37% in 2003 to 44% in 2024. When farmers exit farming, agricultural land becomes

available for others seeking to expand. However, little is known about how farmers adjust herd size, land area and forage production when increasing their total milk delivery. Therefore, the objective of this study was to examine how farm area, livestock density, feeding practices, and milk production per unit of agricultural land are adjusted following a substantial increase in available milk quota.

Materials and methods

Data were collected from the Norwegian Survey of Account Statistics for Agriculture and Forestry (NSASF, 2025), which is equivalent to the EU's Farm Accountancy Data Network (FADN). Inclusion required dairy farms to have increased their milk quota and delivery by more than 40000 L from one year to the next between 2009 and 2022, ensuring a substantial change in production. Of approximately 300 dairy farms in the NSASF, 95 met these criteria. The NSASF database includes records on total milk quota and delivery, average milk yield per cow, livestock numbers by age category, utilized agricultural area (UAA), and estimated grassland yields. Feed purchases were recorded in monetary terms. The physical quantity of concentrate mixtures was estimated using average annual prices per kg for dairy cow feed, delivered at the farms, as published by NIBIO (2025). The quantity of other purchased feedstuffs, such as forages, was also estimated by using the price per kg for concentrate. All purchased feed quantities were converted to feed units for milk (FUm), representing the net energy lactation, related to the energy content of 1 kg of barley. Milk quota and delivery were standardized per unit of utilized agricultural area (UAA). Livestock units (LU) were calculated according to the EU standard, and livestock density was calculated as LU per ha of UAA. For statistical analysis, average values of these variables were calculated across the years before and after milk quota expansion. Data were analysed using the MIXED procedure in SAS (2025), with Time (before or after quota expansion), Region, and their interaction as fixed effects, and Farm ID as random effect. The Regions included Eastern, South/Southwestern (except Jæren), Jæren landscape (South-Western), Western, Central, and Northern. Due to non-constant residual variance in most variables, Box-Cox transformations were applied prior to analysis. Significance testing was conducted on the transformed scale, while predicted means were back transformed for presentation. The interaction between Time and Region was not significant for any of the variables, indicating consistent effects across regions. Therefore, only the main effect of Time (before vs. after quota expansion) is presented. Statistical significance was declared at $P \leq 0.05$ and trends $0.05 < P \leq 0.10$.

Results and discussion

On average, milk quota per farm increased by 113 672 l and milk delivery rose by 107 207 l following quota expansion (Table 1). Quota fill remained stable at 0.94 both before and after expansion, indicating that farmers quickly adapted to the increased quota. Herd size increased by an average of twelve dairy cows, accompanied by an 11-ha expansion in UAA and 16% rise in leased land. In the Norwegian context UAA on dairy farms is grassland, and grassland proportion remained the same after expansion, 0.98 of total UAA. Land expansion was proportionally smaller than the herd increase, resulting in a 14% rise in livestock density (0.18 LU ha^{-1}), and a 23–25% increase in milk quota and delivery per ha of UAA. Farm-produced forage yield showed a tendency to increase ($P < 0.1$), but due to greater rise in herd size and milk output per UAA, the forage proportion in the diet declined by 8% (239 FUm per LU). The concentrate proportion in the diet of dairy cows increased by 5 percentage points, from 0.42 to 0.47. Herd structure remained unchanged, with dairy cows consistently comprising 65% of total livestock. Thus, the observed increase in annual milk yield per cow, 689 l, was achieved through greater concentrate use. An increase in annual milk quota of over 100 000 l and an average herd expansion of twelve cows typically requires investment in new or upgraded cow housing.

Table 1. Effect of milk quota expansion on dairy herd size, utilized agricultural area, feeding, forage production, milk produced per agricultural area and livestock density

Item	Unit	Time		SED	Significance (<i>P</i>) <	
		Before	After		Time	Region
Milk quota	l farm ⁻¹	230290	343962	9116	0.001	ns
Milk delivered	l farm ⁻¹	213288	320495	8698	0.001	ns
Dairy cows	LU farm ⁻¹	31	43	1.1	0.001	ns
Proportion of dairy cows in herd	LU LU ⁻¹	0.63	0.61	0.010	ns	ns
Milk yield	l cow ⁻¹	6777	7464	93	0.001	ns
Utilised agricultural area (UAA)	ha farm ⁻¹	39.5	50.3	3.09	0.001	ns
UAA leased, proportion of total	ha ha ⁻¹	0.45	0.52	0.029	0.001	ns
Grassland proportion of UAA	ha ha ⁻¹	0.98	0.98	0.019	ns	ns
Livestock density	LU (ha UAA) ⁻¹	1.27	1.46	0.045	0.001	0.001
Grassland yield	FUm ha ⁻¹	3713	3938	127.3	0.1	0.001
Concentrate purchase	FUm LU ⁻¹	2132	2375	56.0	0.001	ns
Forage purchase	FUm LU ⁻¹	56.5	124	28.4	0.01	ns
Forage produced on farm	FUm LU ⁻¹	2982	2743	53.3	0.001	0.05
Concentrate proportion in diet dairy cows	FUm FUm ⁻¹	0.42	0.47	0.009	0.001	ns
Milk quota	l (ha UAA) ⁻¹	5767	7112	251.9	0.001	0.001
Milk delivered	l (ha UAA) ⁻¹	5343	6662	221.6	0.001	0.001

l, litres; FUm, feed unit milk; LU, livestock unit, SED, standard error of difference.

Given the high construction cost per cubicle, farmers aim to reduce investment costs by increasing milk yield per cow (Flaten, 2002b). A common strategy during expansion is adoption of AMS, which, while raising capital costs, also supports higher milk yield through more frequent milking which in turn increases feed demand. Additionally, Norway's fragmented and irregular agricultural landscape often makes it costly to acquire or lease additional land. As a result, expanding forage production to match herd growth can significantly raise production costs.

According to Vik *et al.* (2019), the significant structural changes in Norwegian dairy farming were driven less by policy reforms and more by the introduction of AMS, which emerged in the early 2000s. However, investment in AMS was conditional on the access to additional quota, either through private purchase or leasing, alongside an increase in the quota ceiling. Thus, structural rationalization would have occurred even without AMS, primarily a result of political changes to the quota system. Oskam and Speijers (1992) concluded that: "Improved quota mobility by organizing quota trade or leasing will speed up structural development" based on their analysis of milk quota systems across OECD countries. This prediction proved accurate in Norway, where liberalized quota policies contribute to a shift away from traditional grassland-based dairy system.

Conclusion

The structural changes in Norwegian dairy farming have led to increased milk production that relies more heavily on purchased concentrate and less on forage produced from local grassland.

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Impact of grassland conversion on agropastoral habitats of community interest over 40 years in France

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Abstract

Permanent grasslands (PGs) are among the most diverse yet most threatened habitats worldwide. In Europe, PG areas have dramatically declined since the 1970s, primarily due to changes in agricultural practices leading to their conversion into croplands or forests. These conversions have been highly heterogeneous across regions and may have disproportionately affected certain grassland habitat types. Most studies on PG conversion focus only on changes in total PG area, potentially underestimating impacts on specific grassland habitats. To address this gap, we analysed a unique dataset of 19 582 floristic surveys conducted in French PGs between 1980 and 2022 by the National Botanical Conservatories. We assessed the conversion of various PG types—some of which are habitats of community interest (HCI)—between the survey date and 2023. Our results show that 8% of surveyed PGs were abandoned and 6.5% were converted into croplands by 2023, with pronounced regional disparities and losses reaching up to 47% in the Île-de-France region. The overall conversion rate was similar for HCI and non-HCI grasslands, but rates for HCI vs. non-HCI grasslands varied across grassland types. These findings highlight that some grassland types and HCIs are particularly vulnerable to conversion, providing key insights to guide targeted conservation strategies.

Keywords: abandonment, biodiversity conservation, cropland, habitat of community interest, habitat loss, permanent grassland

Introduction

Permanent grasslands (PGs) encompass a wide range of vegetation types that play a crucial role in supporting biodiversity, ecosystem services and resilience. Globally, grasslands are among the most heavily converted and least protected habitats (Hoekstra *et al.*, 2005). In recent decades, they have undergone widespread loss and degradation, primarily driven by changes in agricultural practices (Bardgett *et al.*, 2021). Although many studies emphasize regional disparities in grassland loss, they often overlook impacts of grassland conversion on specific habitat types (Ridding *et al.*, 2015). For instance, grassland types may exhibit different rates and types of conversion (e.g., conversion to crops or abandonment). Additionally, it is unclear whether grassland habitats of community interest (HCI) experience lower conversion rates, notably because they mostly occur within Natura 2000 sites. Comprehensive knowledge on which specific types of grasslands have been most affected by habitat conversion remains limited. This study examines conversion rates of different types of French PGs, including HCI, surveyed between 1980 to 2022, under the hypothesis that grassland loss is not random.

Materials and methods

We used the PRéSur project (Prefiguration of a Monitoring Network for the Conservation Status of Agropastoral Habitats in France) dataset, which compiles detailed phytosociological surveys of agropastoral habitats, including HCI, with a particular focus on PGs. This dataset is a subset of a larger,

Table 1. PRéSur dataset composition by the rate of PGs types and HIC.

Grassland type	HIC (%)	Non-HIC (%)	Sum (%)
Mesophilous meadows	21	10	31
Wet meadows	0.21	19	20
Calcareous grassland	13	1	14
Peaty wet grasslands	10	2	12
Siliceous meadows	10	1	11
Other grasslands	2	10	12
Sum (%)	56	44	100

Other grassland category includes fringe habitats, sandy, mountain, and Mediterranean grasslands, and was structured to ensure an equivalent number of samples per type for robust statistical analysis.

unique, long-term national vegetation database, surveyed over several decades by expert botanists from France’s National Botanical Conservatories. Unlike satellite- or aerial imagery-based land cover products, PRéSur provides fine-scale, expert-validated ground observations of PGs, allowing for precise classification of grassland types and identification of HCI, making it highly suitable for our analysis. We filtered the dataset by retaining only surveys conducted in PGs—defined as open herbaceous areas that are mown or grazed—associated with a spatial accuracy better than 10 metres and located within farmland fields (based on the Land Parcel Identification System data—LPIS). The resulting dataset comprises 19,582 surveys conducted across mainland France between 1980 and 2022. The vegetation surveys were grouped into six grassland types based on their phytosociological classes (Table 1).

We overlaid the 19 582 PG survey locations with three land cover datasets from 2023 to determine their land use in 2023. For 91% of the PG points, land cover was extracted from the LPIS, which provides land cover of georeferenced agricultural parcel data. For the remaining 9%, where land cover could not be determined *via* LPIS, we used the BDTOPO and OSO land cover datasets. We grouped land cover changes into conversions to croplands and forests, excluding other types of conversion (e.g., urbanisation), as they were rare and not of interest to our study. We calculated conversion rates for each grassland type and for HCI and non-HCI grasslands within each type at the national level. We also calculated conversion rates for all grassland type within each French administrative region.

Results and discussion

First, our study confirms that conversion of PGs is still an ongoing trend, due to both cropland expansion and land abandonment. At the national scale, 14.7% of PGs surveyed between 1980 and 2022 were no longer PGs in 2023. Specifically, 6.5% were converted to cropland and 8.2% to forest. The type and rate of conversion varied across French administrative regions. The highest losses occurred in Île-de-France (46.7%) and Centre-Val de Loire (29.6%) mainly due to cropland expansion, and in Provence-Alpes-Côte d’Azur (26.9%) and Occitanie (21.6%) mainly due to abandonment and forest encroachment. These trends are consistent with regional disparities in land use documented by previous studies on French grassland dynamics between 1992 and 2010 (Xiao *et al.*, 2015). Conversions occurred at similar rates even among recently surveyed PGs (between 2015 and 2022), despite conservation efforts and policy measures such as the one aiming to maintain PGs within the Common Agricultural Policy.

Second, our study suggests that the less-frequent grassland types (called ‘other grasslands’) are particularly affected by habitat conversion and that conversion rates varied among grassland types. The highest conversion rates were observed in ‘other grasslands’ (34.5%) and calcareous grasslands (22%), largely

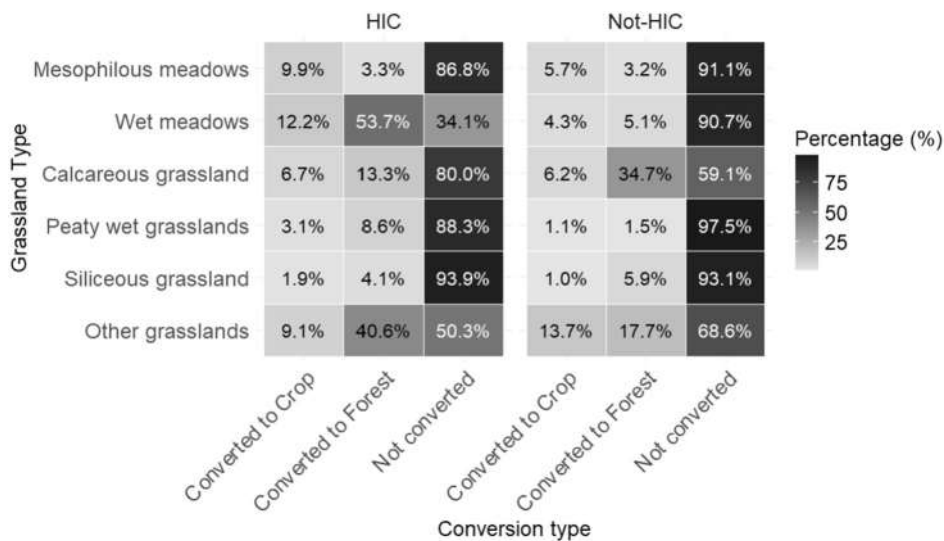


Figure 1. Rates of conversion for HIC PG types and non-HIC PG types.

driven by forest encroachment. This suggests that ‘other grasslands’, although rare in the dataset, may be particularly vulnerable and require greater conservation focus. Calcareous grasslands—among the most species-rich types—typically develop on thin, oligotrophic soils unsuitable for intensive agriculture, making them more prone to abandonment rather than cultivation. By contrast, mesophilous meadows had lower conversion rates (12%), primarily due to conversion to cropland, reflecting their greater agricultural suitability. These findings support our hypothesis that grassland conversion is not random, varying across regions and grassland types.

Finally, our study suggests that HIC grasslands have also been affected by habitat conversion and some HIC grasslands (codes EUR H6220 and H6420; European Commission 2013) have faced an even higher rate of conversion (Figure 1). At the national scale, the total conversion rate of PGs was similar between HIC and non-HIC. Among HIC surveyed in our dataset, the most affected types were Pseudo-steppe with grasses and annuals of the *Thero-Brachypodietea* (HIC 6220; 40% converted to forest) and Mediterranean tall humid grasslands of the *Molinio-Holoschoenion* (HIC 6420; 54% to forest and 12% to crop). Conversion rates within HIC varied also by grassland type. HIC within wet meadows and ‘other grasslands’ showed the highest forest conversion rates (53% and 40.6%, respectively), while non-HIC within calcareous grasslands had the highest forest conversion among non-HIC (34.7%). This suggests that even HIC have been affected by habitat conversion, despite policies, such as Natura 2000, that aim to support the protection of habitats listed under the Habitat directive.

Conclusion

This study shows that conversion rates of French PGs remain high overall, in particular for some grassland types and some HIC, despite existing conservation policies. It also shows that both the rate and type of conversion vary strongly across regions and grassland types. These findings therefore highlight the need for further analyses to identify the drivers of conversion, whether linked to site-specific factors (e.g., accessibility, soil characteristics, topography) and/or ecological attributes (e.g., taxonomic and

functional diversity). Understanding these drivers is crucial to inform effective conservation strategies and prioritizing areas most vulnerable to loss.

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Botanical succession after mowing at mountain pastures of Gorbeia SAC

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Abstract

Mountain pastures are essential agroecosystems for biodiversity conservation and extensive husbandry management. However, the abandonment of this activity, or the change of the livestock species that graze in these systems, favours expansion of the shrub layer, thereby decreasing the area of open pastureland and increasing fire risk. Therefore, mowing is one of the main management actions for these agroecosystems. This study assessed the ecological succession and the changes of the botanical community composition of the mowed sites at the mountain pastures of Gorbeia Special Area of Conservation (SAC) (Basque Country). The sampling was done from 2018 to 2025 at 8 sampling plots (4 mowed sites vs. 4 not mowed sites). Thus, the alterations of the botanical community were studied, and the dominant species of each treatment were identified.

Keywords: clearing, ericaceous, gorse, heathland, pasture management

Introduction

Mountain pastures are agroecosystems that can host a wide biodiversity and offer a great variety of ecosystem services like grass production, carbon sequestration and water regulation (Plantureux *et al.*, 2016), among others. The Basque Country pastures are mainly managed with the objective of avoiding shrubland encroachment, by livestock-grazing and mowing (Ferrer *et al.*, 2001), often combined, since the abandonment of grazing would require a higher frequency of mowing to stop encroachment. Therefore, the objective of this study was to evaluate the influence of mowing on the botanical community and its ecological succession in pastures where extensive grazing is done.

Materials and methods

The study was carried out in the mountain pastures of the Gorbeia Special Area of Conservation (SAC) (Basque Country, Spain) in 2018 and 2025. The landscape is a mosaic of pastures and heathland, which is shaped by the extensive grazing of different species of livestock. In total, 8 plots were sampled: 4 mowed (only once in 2017) and 4 control.

Vegetation was characterized by counting the presence of different botanical functional groups every 0.25 m at each transect of each sampling plot (4 transects/ plot): necromass, gorse (*Ulex gallii*), ericaceous (*Erica cinerea*, *E. vagans*, *E. arborea*, *Calluna vulgaris*, *Daboecia cantabrica*), other woody species, grasses and graminoids, other herbaceous species, ferns, bare soil, and leguminous species.

First, to assess the dissimilarity between treatments the Bray–Curtis index was calculated for the years 2018 and 2025. Then, to identify the groups that contributed the most to the differentiation between treatments, the Similarity Percentage (SIMPER) analysis was done for each year. Lastly, the frequency

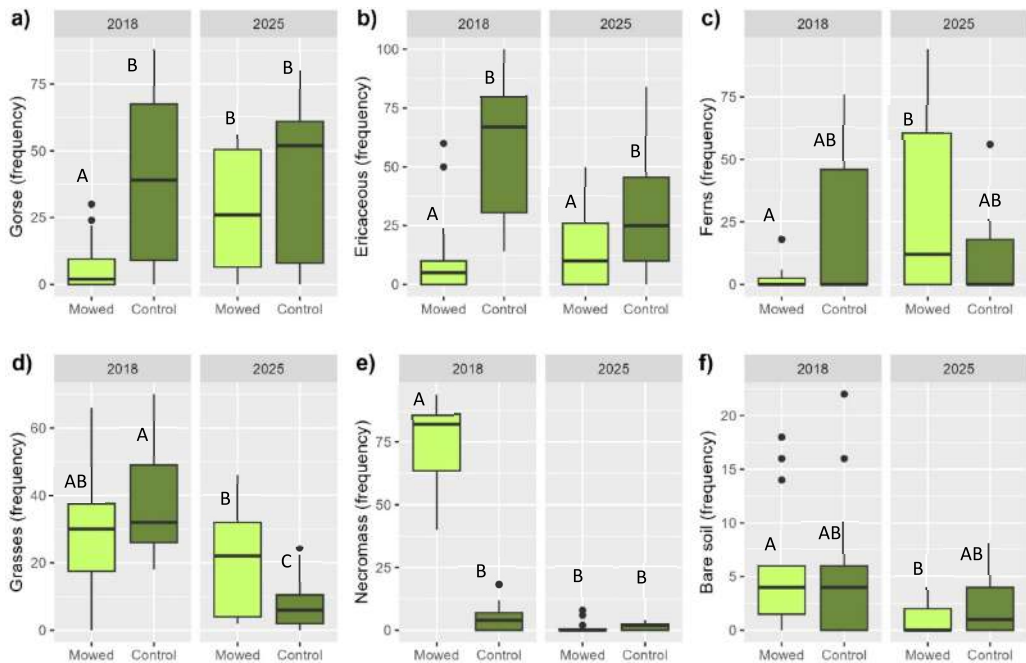


Figure 1. Boxplots for the frequency of gorse (a), ericaceous plants (b), ferns (c), grasses and graminoids (d), necromass (e), and bare soil (f) in mowed (light green) and control (dark green) plots in 2018 and 2025.

of each functional group was compared between both years (2018 and 2025) and between treatments (mowed and control) with a Wilcoxon test.

Results and discussion

The recolonization or revegetation of intervened or perturbed habitats is conditioned by the surrounding plant species (Rogers and Hartnett, 2001). For that reason, the differences between the studied botanical communities decreased with time: while in 2018 the dissimilarity Bray-Curtis index was 0.59, in 2025 it was 0.34. The SIMPER analysis showed that in 2018 the group that contributed most to the botanical community differentiation was necromass (non-cumulative contribution 0.34), more frequent in mowed plots. Ericaceous plants and gorse also influenced community differentiation (0.24 and 0.18, respectively), although they were more frequent in control plots. However, in 2025 the botanical community differentiation was most affected by ferns (0.28), being more frequent in the mowed plots, and followed by gorse and ericaceous plants (0.28 and 0.24, respectively), which were more frequent in control plots. This remarks the recolonization capacity of ferns.

In 2018 gorse frequency was higher in control plots ($W = 54.5$, $p = 0.005$), although by 2025 their frequency in mowed plots had augmented significantly ($W = 57.5$, $p = 0.007$), so there were no significant differences between treatments (Figure 1a). In contrast, ericaceous plants always had a higher frequency in control plots than in mowed plots, both in 2018 ($W = 19$, $p < 0.001$) and in 2025 ($W = 76$, $p = 0.049$) (Figure 1b). The greater growth of gorse compared to ericaceous plants indicated the greater recolonization capacity of the former one, in agreement with Celaya *et al.* (2007). Gorse has the ability to grow rapidly after different types of perturbations like wildfires (Baeza, 2001). However, ericaceous plants, even if they grow more slowly, have a more competitive nature. Therefore, ericaceous

plants are expected to arrive at their initial frequencies becoming dominant over gorse (Celaya et al., 2007) in the mowed plots.

In respect to ferns (Figure 1c), they had similar frequency in 2018 and 2025 in control plots, but they had a significant increase in mowed plots from 2018 to 2025 ($W = 78$, $p = 0.040$). This group has the capacity to grow very rapidly and to colonize zones with no vegetation. Ferns have a strict cycle: they show a maximum coverage in summer and in winter their foliage decomposes (Gil da Costa *et al.*, 2024). This allows the simultaneous growth or conservation of different species.

Grass and graminoid frequency stayed equal in the mowed plots, although in control plots it decreased from 2018 to 2025 ($W = 250$, $p < 0.001$), being smaller in control plots than in mowed plots ($W = 184$, $p = 0.035$) (Figure 1d). This shows how mowed plots can be a great feeding resource for livestock.

Necromass had its maximum values in mowed plots in 2018 ($W = 256$, $p < 0.001$) (Figure 1e), right after the mowing intervention. Necromass increase after mowing was quickly reversed due to its rapid decomposition, leaving sufficient space for the recolonization of other plant species (Prach *et al.*, 1997).

Finally, the amount of bare soil decreased from 2018 to 2025 in mowed plots ($W = 193.5$, $p = 0.010$) (Figure 1f), since after mowing the recolonization by plants occurred rapidly.

Conclusion

At 8 years after mowing, under an extensive grazing management, the botanical communities were still different, even though the ecological succession and the high capacity for recolonization of some groups like gorse led to their homogenization. However, ericaceous plants, which are dominant in Atlantic heathlands, had not yet arrived at their initial frequencies, since they have slower colonization processes. For that reason, the botanical community in mowed sites is expected to continue evolving over the years.

Acknowledgements

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Grassland biodiversity on organic dairy farms in the Lower Bavarian–Upper Austrian uplands

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Abstract

Ecologically valuable grassland habitats of the Lower Bavarian–Upper Austrian border region are increasingly threatened by structural changes in agriculture and the effects of climate change, putting their biodiversity and ecological functions at risk. In this study, using four organic dairy farms as case examples with different grazing and harvesting as well as fodder conservation practices, we demonstrate targeted measures for the management of species-rich grasslands, while simultaneously optimizing grassland use and fodder quality as well as harvest costs. Vegetation monitoring prior to the first cut in 2024 and insect monitoring during three periods in 2024 using Malaise traps, followed by DNA metabarcoding analysis, were conducted on each farm. Statistical analyses were performed using repeated measures ANOVA in R. The results indicated an average plant diversity of 23 species per farm. In total, an average of 478 insect species was recorded per farm over the entire observation period. Diptera were the most abundant, accounting for 58% of all observations, followed by Hymenoptera (14%), Coleoptera (10%), and other taxa. Although no significant differences in insect species numbers were found between observation periods, the initial biodiversity monitoring provided important insights into vegetation and insect diversity on organic dairy farms in the project region.

Keywords: biodiversity, grassland, malaise traps, metabarcoding, management optimization

Introduction

The grasslands in the Lower Bavarian–Upper Austrian region, which stretches across parts of Germany and Austria, are primarily used as fodder for dairy cattle farming. Due to variations in altitude, slope, partly shallow soils, and relatively high annual precipitation, arable farming is only feasible in selected valley areas. As result, grassland dominates land use patterns accounting for 43% of the agricultural area in Upper Austria, which also holds the highest proportion of permanent grassland nationwide (Hintringer *et al.*, 2023), and 34% in Lower Bavaria, with even higher shares in the upland areas of the border region. The diversity of site conditions, grazing practices, and management strategies has led to species-rich grassland stands in many areas. About 65% of Bavarian farms surveyed ($n = 500$) as part of a grassland monitoring project already manage their grassland plots with varying levels of intensity (Heinz *et al.*, 2023). These species-rich grasslands are increasingly threatened by multiple factors. Structural changes in agriculture are resulting in larger farms with more uniform management, while other areas are being extensified or abandoned for afforestation. Additional pressures include climate change and increasing infestations by the common cockchafer (*Melolontha melolontha*). The present study investigated plant and insect diversity on selected organic dairy farms in the region, providing background information to support the further optimization of site-specific management practices on these farms.

Table 1. Organic dairy farm characteristics

	Farm A	Farm B	Farm C	Farm D
Region	Lower Bavaria	Lower Bavaria	Upper Austria	Upper Austria
Average altitude (m asl)	544	696	603	558
Annual precipitation (mm)	1144	1224	986	933
Grassland area (ha)	62	53	15	36
Surveyed grassland sites (of total)	16 (85)	19 (26)	6 (6)	12 (12)
Main fodder conservation method	hay	silage	hay	silage
Number of cuts per year	3–6	2–3	2–4	3–4
Number of dairy cows	50	31	35	52

Materials and methods

The study was conducted on four organic dairy farms located in the Lower Bavarian–Upper Austrian upland border region. All plant species were surveyed on selected grassland areas of the participating farms, and their relative cover or yield contribution was estimated according to the Klapp & Stählin (1936) method, prior to the first cut in 2024. General information on each analysed farm is presented in Table 1.

Two standardized Malaise traps of the “Barták” type, filled with 80% ethanol, were used on each farm to capture and collect insects on grassland sites. The traps were deployed continuously for two weeks during each of the three sampling periods (June, July, and August/September) in 2024. The collected insect samples were identified using DNA metabarcoding by an external service provider (Advanced Identification Methods—AIM GmbH, Leipzig, Germany). In total, 24 samples corresponding to the first week of each sampling period were included in the analysis.

To evaluate differences in the number of captured species across observation periods, a repeated measures ANOVA was performed (R 4.3.3), following a test of normality using the Shapiro–Wilk test.

Results and discussion

The results of the vegetation monitoring on the participating farms, covering between 6 and 19 grassland sites per farm, showed a median plant species richness of 23 species (Figure 1A). Overall, the four farms fall within a favourable median range of approximately 22 plant species that is typical for organic grassland farming systems in Bavaria (Heinz *et al.*, 2023). The majority of recorded species were forbs, accounting for up to 35% of total cover on sites participating in agri-environmental schemes. Among the legumes, white clover was the most common. The number of grass species varied across sites and farms and included species such as orchard grass, perennial ryegrass, meadow foxtail, and others.

Using Malaise traps and the DNA metabarcoding method, an average of 478 insect species were recorded per farm over the entire observation period (Figure 1B). In comparison, a considerably lower total insect species richness of 255 was reported by Panasitti *et al.* (2023) in grassland-dominated regions of southern Germany. In contrast, the farms in our study are organic farms located in an upland region, characterized by a relatively high level of grassland extensification and participation in several agri-environmental schemes. Diptera were the most species-rich group across all farms in our study, accounting for 58% of all recorded species, followed by Hymenoptera (14%), Coleoptera (10%), Lepidoptera (9%), Hemiptera (5%), and other taxa. Each insect taxon plays an important role in plant interactions, such as pollination, herbivory, seed dispersal, or parasitism. In particular, pollinators are crucial for maintaining the diversity of flowering plants, which is characteristic of the

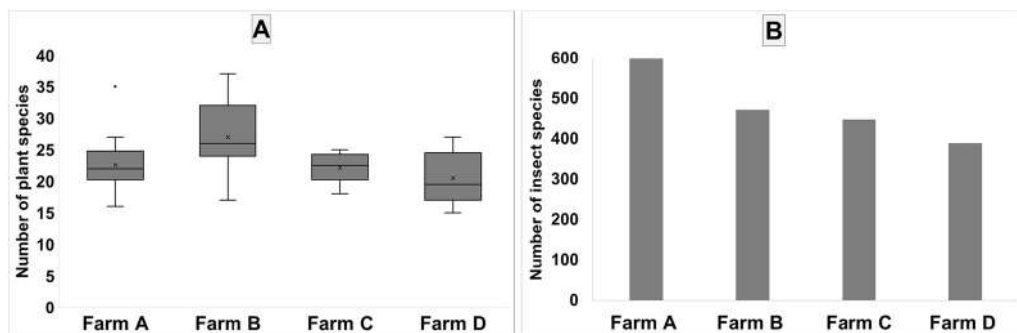


Figure 1. Plant (A) and insect (B) species richness on grassland sites per farm.

less intensive grassland systems found in the studied region. Grassland sites with the highest insect species richness in our study were characterized by a relatively low proportion of grasses (on average 57%) and a high proportion of forbs (on average 25%) in terms of cover or yield contribution. No significant differences in insect species richness were observed among the three sampling periods, based on an analysis conducted independently of farm and Malaise trap identity. This result can also be attributed to external factors, such as weather conditions during sampling and the timing of grassland cuts at specific sites.

Conclusion

Although species-rich grasslands are increasingly threatened by multiple factors, this study demonstrates that the surveyed organic dairy farms located in the Lower Bavarian–Upper Austrian upland border region exhibit notably high species richness in both plant and insect communities on their grassland sites. While the farms already manage their grassland plots with varying levels of intensity, further optimization measures are planned to preserve and potentially increase species richness on selected grassland sites. These measures include, among others, the use of satellite imagery to estimate site-specific dry matter yield in order to identify areas with differing productivity and adapt management practices accordingly. After an adaptation period, species richness will be reassessed to evaluate the effects of the implemented measures.

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Micro-learning to support training for European farming

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Abstract

Micro-learning is short digital modules (<10 min) designed to convey key messages efficiently. As part of the EU-funded ClieNFarms project, 35 modules were co-designed and delivered across 21 European regions to support climate-smart blended training. Developed using Elucidat, and translated via AI into multiple languages, they complemented demonstration events and extended training reach. Hosted on Moodle, modules were grouped into Dairy, Beef, Sheep, Pigs, Arable, and Cross-Cutting topics. Outreach to 29 partners (17 agricultural advisories/consultancies, 12 agricultural educational institutions) increased engagement, with Moodle analytics showing deeper interaction and enrolment. Trainer feedback highlighted practicality, flexibility, and reinforcement of climate-smart concepts. This paper outlines module co-design, implementation, and early outcomes, providing guidance for wider use in climate-smart training.

Keywords: blended learning, climate change, farmer training, grassland management, knowledge transfer, micro-learning

Introduction

Grasslands underpin European climate action, supporting livestock, biodiversity, and sustainable land use. Climate adaptation and mitigation require timely, accessible knowledge transfer. Traditional face-to-face training is valuable but constrained by time and geography.

ClieNFarms developed micro-learning as part of a blended approach, digital modules combined with on-farm events, to support adoption of climate-smart practices across diverse regions. Modules were tailored to local contexts while maintaining consistent messaging across sectors.

Materials and methods

The micro-learning was developed under Task 6.5 of the Horizon Europe funded ClieNFarms project. The Elucidat platform was used as the authoring tool and was selected for its mobile compatibility, Sharable Content Object Reference Model (SCORM) export features, and multilingual interface. The modules were subsequently hosted on a Moodle learning platform to enable structured access and user tracking. Modules were co-designed with regional coordinators, referred to as Innovation Systemic Solution Space managers (I3S), across 21 regions representing a variety of farming systems (e.g. dairy in Ireland, sheep in Romania, olives in Portugal, etc.). The I3S managers identified and prioritised training needs in their local contexts. The I3S managers received online training and support for using the authoring tool and for organising blended training events, ensuring consistent module delivery and effective facilitation. Modules were grouped by system (Dairy, Beef, Sheep, Pigs, Arable, Cross-Cutting) and translated into local languages. Blended learning events combined digital modules with farm walks, briefings, or digital-only sessions. Trainer feedback was collected via a 10-point Likert survey. Moodle engagement statistics were tracked to assess reach and uptake. Partners were engaged via multiple approaches including Sister Project Networks, Direct Outreach, I3S

Manager Outreach, and Existing Collaborations. Outreach targeted both consortium and selected external countries, focusing on agricultural advisory/consultancy and agricultural educational institutions. A partnership was considered established when the organisation formally registered on the Moodle platform to access micro-learning content. Full details of outreach procedures and participation are available in the ClieNFarms Task 6.5 deliverable (Enright *et al.*, 2025).

Results

Thirty-five micro-learning modules were developed. Table 1 shows the frequency of climate-smart techniques addressed in the micro-learning modules for dairy, beef and sheep, as well as a number of cross-cutting solutions. Management of feed, soil nutrient management and breeding were the most frequently covered techniques, while grazing management was less common. Cross-cutting modules are applicable across all grassland enterprises and include hedgerows, anaerobic digestion and agroforestry. The full list of modules is available by scanning the QR code at the bottom of this paper. The micro-learning modules were primarily used as supplementary tools for blended learning events across 21 regions. Survey responses were received from 17 of 21 I3S managers, with eight regions meeting minimum participant attendance. Trainers reported improved on-farm discussions, participant understanding, and content integration. Likelihood of continuing blended learning beyond the project averaged at 60% across regions. Moodle engagement peaked in October, with guest clicks nearly doubling and enrolled user clicks more than tripling following partnership onboarding. Unique enrolled users more than tripled, while unique guest visits also increased, reflecting the transition from guest exploration to enrolled participation. Full historical engagement statistics are available in the ClieNFarms Task 6.5 deliverable (Enright *et al.*, 2025). Outreach established 29 formal partnerships across 17 countries, comprising 17 agricultural advisories/consultancies and 12 agricultural educational institutions. Registration on the Moodle platform enabled access and engagement. Uptake of modules correlated with partnership onboarding particular success driven from collaboration with sister project networks; ClimateSmartAdvisors and ClimateFarmDemo. Detailed tables and analyses are available in the online deliverable (Enright *et al.*, 2025).

Discussion

Early selection of the content-authoring platform and the learning management software is critical for smooth integration, automation, and contributor training. Adequate time and resources for dissemination, along with early partner engagement, increased the uptake and visibility. Alternative feedback approaches, such as interviews or focus groups, may yield richer insights than surveys alone. Maintaining alignment with user needs through continuous engagement ensures relevance and impact. Blended learning provided flexible, cross-sector relevance. Cross-cutting modules (e.g. agroforestry, hedgerows, anaerobic digestion) offered value across dairy, beef, and sheep. It is clear from Table 1 that the most frequently covered climate-smart techniques and farming sectors reflects both the priorities and expertise of the I3S managers in the consortium who co-designed the modules and, broadly, the relative prevalence of farming sectors in Europe. It could be beneficial for future initiatives to carefully select contributors and prioritize techniques based on scientific evidence, practical relevance for farmers and sector representation to ensure maximum coverage and impact.

Survey feedback showed that micro-learning modules reinforced climate-smart practices, supported consistent messaging, and improved participant preparedness. Moodle engagement demonstrated strong interest and visibility, highlighted the success of outreach through sister project networks. Overall, the challenges observed relate to early-stage adoption rather than limitations of the micro-learning approach.

Table 1. Frequency of main climate-smart solutions in the ClieNFarms micro-learning for the three grassland enterprises (dairy, beef and sheep) and cross-cutting solutions.

	Beef	Dairy	Sheep	Cross-cutting
Optimising feed efficiency through grouping cows	1			
Optimising rations	1	1	1	
Managing and optimising concentrate supplementation		1		
Grazing management	1	1		
Soil fertility	1	1		
Breeding/high genetic merit	1	1	1	
Low emissions slurry storage	1	1		
Protected urea	1	1		
Incorporating white clover		1		
Mitigating methane		1		
Reducing number of replacements		1		
Reducing time to finish cattle	1			
Incorporating and managing silviopasture/agroforestry				2
Decarbonising machinery				1
Hedgerows for C sequestration and storage				2
Anaerobic digestion				1
Photovoltaics			1	
12 Steps to reduce gaseous emissions on your farm				1

Conclusions

Micro-learning is effective, scalable tool for climate-smart knowledge transfer. Integration into blended learning enhances flexibility, reinforces key messages, and supports consistent delivery. Sister project networks utilisation maximised outreach success. ClieNFarms demonstrates that co-designed, locally relevant modules complement traditional advisory approaches while reaching broader audiences. Future initiatives should plan extended timelines, early technical decisions, integrated promotion, and multiple feedback mechanisms to maximise sustainability and long-term impact.

Scan the QR code to access the micro-learning and use them as part of your on-farm demonstrations or in your teaching at an agricultural school.



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Stakeholder perception of the relevance of measures promoting species-rich grasslands in the Alps and Carpathians

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Abstract

The maintenance of most species-rich grasslands requires a targeted management intensity, balancing trade-offs between agricultural output and biodiversity conservation. As both management intensification and abandonment lead to biodiversity loss, farmers are pivotal for maintaining and promoting it. However, benefits and challenges related to the management of species-rich grassland may be differently perceived depending on the stakeholder categories. To better understand these differences, a survey was conducted in eight countries/regions of the Alps and Carpathians, in order to provide policy makers with knowledge elements for designing and implementing agricultural and environmental protection policies, taking into account the stakeholders' perception of the relevance of several measures. A survey, yielding more than 2000 responses, was administered to nine stakeholder categories, selected for their direct or indirect involvement in the implementation of new policies designed to protect these habitats. Grazing and the combination of mowing and grazing were highly and equally rated by all stakeholder categories, whilst mowing from centre was the measure perceived as least relevant. For most measures, the perception differed depending on the stakeholder category.

Keywords: agriculture, biodiversity, management, mountain environment

Introduction

When designing and implementing measures aimed at promoting biodiversity in grassland in agriculturally relevant contexts, a common knowledge baseline and a reciprocal understanding of all involved stakeholders are pivotal elements for a constructive and effective dialogue. The Biodiversa+ project Grasslands4Biodiversity (www.grass4b.com) aims to preserve and promote the protection and conservation of grasslands with high biodiversity in the Alps and Carpathians. Indeed, both the intensification of management and land abandonment mostly result in biodiversity loss (Niedrist *et al.*, 2009). Under these circumstances, therefore, farmers and other agriculture-related actors play a key role along with nature conservation-related actors, scientists, policy makers and the general public.

Table 1. Effect of the investigated factors on the perceived relevance.

Source	df	F	P
Intercept	1	22084.0	<0.001
Measure	7	40.5	<0.001
Stakeholder category	8	19.9	<0.001
Stakeholder category×Measure	56	1.5	0.013

df, degrees of freedom, *F*, Fisher's *F*, *P*, probability. The model's pseudo-*R*² (marginal) is 0.50.

The objective of this study is to clarify the expectations and mindset of the stakeholders in order to provide useful information for the development and implementation of agricultural and environmental policies that take into account stakeholder perceptions of the relevance of nine agronomic measures possibly affecting grassland biodiversity.

Materials and methods

A questionnaire was created with Microsoft Forms (with Findmind for Switzerland only) and addressed to nine stakeholder categories: farmers (FAR), farmers' unions and organisations (FUN), farm advisors including veterinarians (ADV), foresters/hunters (FOH), policy makers and administration at the regional level (PMK), scientists from relevant scientific fields (SCI), administrators of protected areas (APA), nature protection organizations (NGO), and the general public (GP). The survey was conducted in eight countries/regions within countries spread across the Alps and Carpathians: NE Italy (South Tyrol), NW Italy (Piedmont/Aosta Valley), Austria, Switzerland, Slovakia, Romania, Ukraine, Poland. Respondents were invited to express their opinion on the relevance of eight important measures used in the participating region/countries to support species-rich grasslands. Respondents were asked to rate each item using a 5-point Likert scale (from 1 = completely unimportant to 5 = very important), allowing also (for the Swiss questionnaire only) mean values between two adjacent levels, and allowing for each question the answer option 'I don't know'. The measures investigated included: (i) grazing (seed dispersal, pollination, fertilisation, trampling), (ii) combination of mowing and grazing, (iii) ensuring low fertilisation levels, (iv) increasing grassland connectivity, (v) late first cut, (vi) mowing height >7 cm, (vii) staggered cut, and (viii) mowing by starting from the field centre. The last three measures are mainly aimed at promoting animal rather than plant diversity. For all cases in which a given stakeholder category within a specific country/region did not reach at least one-third of the target number of responses (50 for farmers and general public, 10 for all other stakeholder categories), those answers were not included in the further analysis. Consequently, due to the limited number of responses, only three stakeholder categories (APA, FAR and SCI) from Ukraine could be included in the analysis. Given the wide variability of the number of responses per combination of measure, stakeholder category and country, means of all available ratings per each combination was computed. The 2027 valid responses resulted then in 528 country-specific and stakeholder-specific mean values ($n = 64$ for APA, FAR and SCI, $n = 56$ for all other stakeholder categories). The resulting dataset was analysed by means of a linear mixed model accounting for the measure, the stakeholder category and their interaction as fixed terms, and the country/region as random. The dependent variable was the relevance rating of measure. This variable, in principle ordinaly scaled, was treated as metric to account for the complexity of the design. Multiple comparisons were made by Sidak. A significance level of 0.05 was used for all tests. All statistical analyses were performed by IBM SPSS Statistics 29.1.1.0 (171).

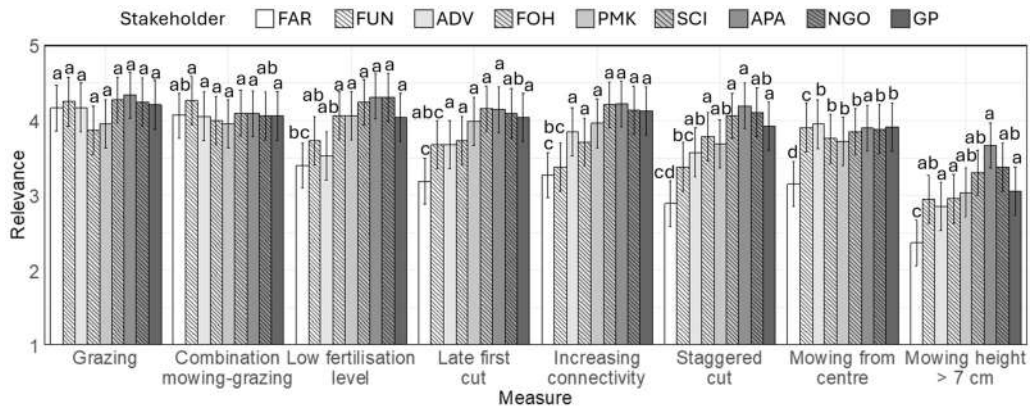


Figure 1. Mean relevance (1 = completely unimportant, 3 = neutral, 5 = very important) of eight measures to promote species-rich grassland applied in eight countries/regions within the Alps and the Carpathians as perceived by nine stakeholder categories (for abbreviations see Materials and methods section). 95%-confidence intervals are shown for comparison of means for different stakeholder categories within measures. Means of different measures within stakeholder categories sharing no lowercase letters significantly differ from each other.

Results and discussion

Both the main terms of the two factors (measure and stakeholder category), as well as their interaction, were found to significantly affect the perceived relevance (Table 1).

This shows that the relevance of the different measures was not equally perceived by the stakeholder categories. Grazing and the combination of mowing and grazing were perceived as highly relevant and were equally rated (around 4) by all stakeholder groups (Fig. 1). For low fertilisation level, increasing connectivity, late first cut, mowing height > 7 cm and staggered cut the relevance was perceived to be highly relevant (around 4) by most stakeholder categories, whilst farmers (and, depending on the measure, also farmers' unions and associations and advisors) rated them on a level near to neutral. Mowing from the field centre was almost universally considered to be of lower importance, being close to neutral for most categories and rated as unimportant by farmers.

Conclusion

This study revealed a complex interaction between grassland management measures and the perceptions of different stakeholders. We found that for the majority of all measures (six out of eight), the perceived relevance depends on the stakeholder category. This suggests that to improve the impact of the grassland protection and conservation measures in the future, it is fundamental to consider different stakeholder groups and their perceptions of grassland diversity as their active support is key to ensuring a successful implementation.

Acknowledgements

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Encyclopedia pratensis, the first on-line encyclopedia dedicated to grasslands and forage crops

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Abstract

To encourage the application of the latest research results, the Encyclopedia Pratensis was launched as part of the Multisward European project. It is currently being expanded as part of the European thematic networks Inno4grass and Grazing for Agroecology. Encyclopedia pratensis is based on international scientific literature and is further enriched by practices from farmers. Encyclopedia pratensis now contains 173 web pages and 1045 articles, as well as different types of documents from national and international projects for grassland farmers, farm advisers, industry professionals, public organisations, universities, the agricultural training and education sectors, and scientific communities. To improve the encyclopedia, an ontology has been adopted as a new classification system for the entire site. A search system allowing multiple positioning access (country, field of innovation, climatic conditions, soil type, etc.) is also being developed. The website navigation will be made more ergonomic so that it can be updated easily without requiring significant maintenance. Finally, to ensure the sustainability of the encyclopedia, consideration is being given to entrusting the French Association for Grasslands and Forages (AFPF) with maintaining and perpetuating the website once the G4AE project has been completed.

Keywords: dissemination, grasslands, forage crops, knowledge transfer, website

Introduction

Grazing-based production systems have the potential to produce high-quality food and benefit the competitiveness of farmers, animal welfare and other ecosystem services that are widely appreciated by society. However, for a variety of reasons, grazing is generally in decline in Europe, which threatens many ecosystem services. One reason for this decline, identified in several projects and EGF conferences (Krause *et al.*, 2018; Schreuder *et al.*, 2022), is the insufficient collaboration on grasslands between farmers, advisers, and scientists in the countries concerned. Consequently, the latest research results are not effectively implemented in practice, and valuable grassland-related knowledge is discovered by practitioners at a late stage. Additionally, information about innovations introduced at farm level is not effectively disseminated to the public in all Member States.

Encyclopedia Pratensis (<https://www.encyclopediapratensis.eu/>) was launched as part of the Multisward European project, which focused on developing multi-species grasslands for multifunctional use. It is an interesting tool for disseminating scientific and practical knowledge on grasslands, and for contributing to bridging the gap between the scientific and practical communities—the overall objective of the European thematic network. This paper presents the development of Encyclopedia Pratensis, its current content and utilisation, and future developments.

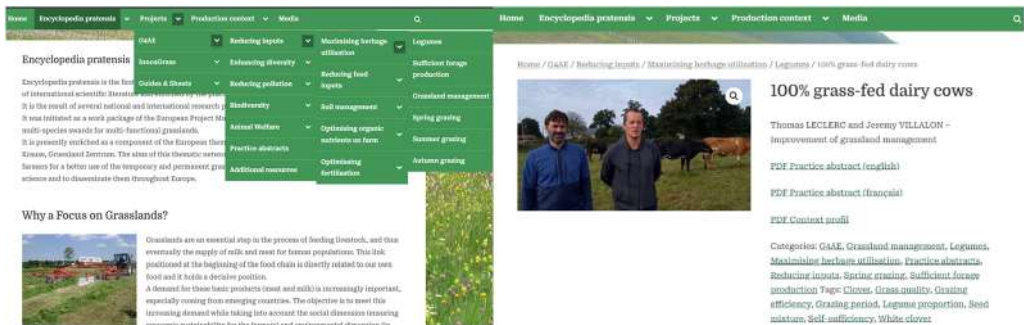


Figure 1. An example of ontology classification in Encyclopedia Pratensis (left), and a case study of '100% grass-fed dairy cows' in the 'Reducing inputs > Maximising herbage utilisation > Legume' category (right).

How was the Encyclopedia Pratensis developed?

Encyclopedia Pratensis is based on international scientific literature and enriched by practices from farmers. Its aim is to become an international reservoir of knowledge relating to agroecology and grasslands. Encyclopedia Pratensis is currently being developed as part of the European thematic networks Inno4grass (<https://inno4grass.eu/en/>) and Grazing for Agroecology (G4AE, <https://grazing4agroecology.eu/>). These networks aim to describe innovative practices implemented by farmers to make better use of temporary and permanent grasslands, consolidate this knowledge with scientific research, and disseminate it throughout Europe.

The current content and utilisation of the Encyclopedia Pratensis

Encyclopedia Pratensis now contains 173 web pages and 1045 articles, as well as different types of documents, such as guides, technical leaflets, innovation analyses, case studies, practice abstracts and farmer portraits. These documents are sourced from national and international projects and are intended for grassland farmers, farm advisers, other industry professionals, public organisations, universities, the agricultural training and education sectors, and scientific communities. These documents are available as HTML pages, PDF files or videos. The HTML content of the website is available in several languages (English, French, German, Dutch, Italian, Polish and Swedish) to facilitate use by farmers in multiple countries.

Thanks to the G4AE project, an ontology of grassland management practices has been created and is being used as a new classification system for the entire website (Figure 1). The main domains of the ontology are 'Reducing Inputs', 'Enhancing Diversity', 'Reducing Pollution', 'Biodiversity' and 'Animal Welfare', in accordance with the five principles proposed by Dumont *et al.* (2013) for designing agroecological animal production systems with a focus on grass-based herbivore systems, grassland management and forage production. Furthermore, a self-assessment tool will enable farmers to enhance their practices and will be linked to relevant resources.

Future development

Several developments are currently underway and will be ready by the end of the G4E project in February 2026. A search system that will allow access to resources through multiple positioning criteria (country, field of innovation, climatic conditions, soil type, etc.) is being developed. Older resources will be reviewed to verify their current accuracy. The website navigation will be made more ergonomic

so that it can be updated easily without requiring major maintenance. The Encyclopedia Pratensis website will be linked with EU-FarmBook (<https://welcome.eufarmbook.eu/>), a Horizon Europe project developing an online platform to gather and share agricultural and forestry knowledge and facilitate access to these resources at a European level.

Finally, a major challenge is ensuring the long-term sustainability of the Encyclopedia Pratensis website once the G4AE project has ended. One option being considered is entrusting the French Association for Grasslands and Forages (AFPF) with maintaining and perpetuating the site once the G4AE project has been completed.

Conclusion

Encyclopedia Pratensis is the first online encyclopedia dedicated to grasslands, forage crops, and grass-based ruminant systems. It contains a significant portfolio of resources from European research projects focused on innovation and action, as well as from thematic networks. Encyclopedia Pratensis has the potential to become an international reservoir of knowledge relating to grasslands and agroecology, and could help to bridge the gap between the scientific and practical communities. A new version of the encyclopedia will be available at the beginning of 2026. It will feature more ergonomic navigation and will be a fluid, clear and user-friendly site that can continue to be easily updated without requiring major maintenance.

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Changes in soil microbial diversity in forage mixtures in Western Europe

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Abstract

Forage mixtures can provide advantages over monocultures in terms of several ecosystem goods and services such as yield, forage quality or resistance to invasibility. But little is known about the influence of these mixtures on soil microbial diversity despite its key roles in soil functioning and plant health. In this work we studied how much the forage sown diversity and management practices impact the diversity of the soil microbial communities (bacteria and fungi) in two locations in Western Europe: in Central France and in Catalonia. We compared grazed and only-cut forage swards. We found different responses depending on the site of the study, with the site in Catalonia showing a negative response to grazing, while the site in Central France showed a positive effect when the mixture was grazed. Richness, abundance and evenness in Central France all showed maximum values near grasses dominance and mixtures of legume and non-legume forbs while in Catalonia different models were selected depending on the diversity index.

Keywords: bacteria, diversity, interaction models, forage mixtures, fungi, next-generation sequencing

Introduction

Next-generation sequencing (NGS) enables us to monitor soil microbial diversity and the changes it experiences due to different environmental conditions. Forage mixtures have been shown to improve the delivery of grassland ecosystem services, such as yield, forage quality and weed suppression. Despite our knowledge on the benefits that forage mixtures provide, little is known about their effect on soil microbial diversity. This work aims to assess the differences in soil microbial diversity caused by plant functional types (PFT) composition of the mixture as well as the effect of direct grazing (cf. cutting) on soil microbial diversity.

Materials and methods

Microbial diversity analysis was carried out in 2023 as part of a larger experiment under the “SUS-FORAGE” project in Theix (Central France) and Solsona (Catalonia). The experiment involved nine species belonging to three different functional groups (grasses, legumes, and non-legume forbs) in

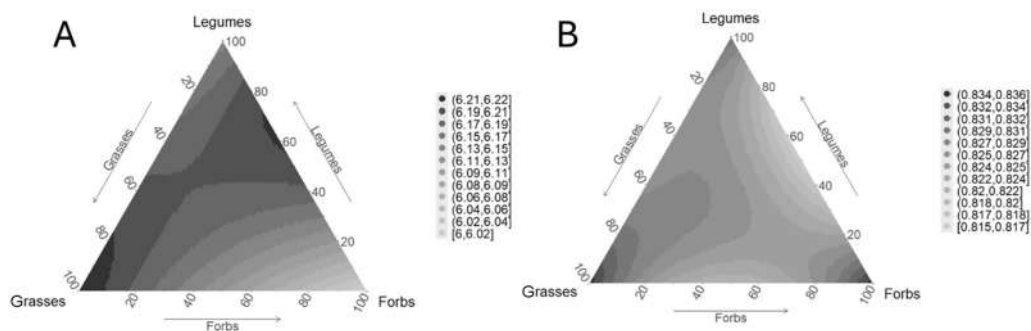


Figure 1. (A) Central France Shannon's diversity index based on the model selected by the lowest AIC (Akaike Information Criterion). (B) Pielou's evenness index for the model applied to Catalonia. In both images the axes represent the proportions of the different functional groups in the experiment. Darker colours represent higher values.

varying proportions and degrees of dominance, following a design similar to Kirwan *et al.* (2014). Two management regimes were considered: grazed and ungrazed. A total of 108 soil samples were collected (54 from Catalonia (27 grazed and 27 ungrazed) and 54 from Central France (27 grazed and 27 ungrazed)) to assess the impact of these practices on soil microbial diversity.

From each plot, 3 composite soil samples were extracted from the top 10 cm soil layer and, after <2 mm sieved, were then frozen and sent to the laboratory for further analysis. The samples were processed and the bioinformatic analyses were performed using the BIOCOP pipeline (Djemiel *et al.*, 2020). Diversity indices (OTU richness, Shannon's diversity index and Pielou's evenness index) for bacteria and fungi were obtained using the *phyloseq* package (McMurdie & Holmes, 2013) and models replicating the diversity interaction model framework were developed in R. Model selection was performed based on the Akaike Information Criterion. Data visualization was performed using the *ggplot2* (Wickham, 2016) and *ggtern* (Hamilton & Ferry, 2018) packages.

Results and discussion

Two different main responses were found regarding plant functional types and bacterial diversity indices. The first one occurred in Theix, where the three plant functional types influenced all three microbial diversity indices. Here, the selected models only included the interaction of legumes and forbs while grasses did not interact with any other plant functional type. The response was the same for all three different diversity indices: the highest diversity values were found in either grass-dominated mixtures or mixtures of legumes and non-legume forbs (Fig. 1A). In Solsona, the selected model varied for each diversity index. Only the legumes had an effect on bacterial richness (negative). For Shannon's diversity index, the selected model was similar to the one in France, but presented a peak in diversity in non-legume forbs dominance. Finally, the Pielou's evenness index showed a complex triple interaction, with peak values in mixtures close to monocultures and the lowest values in mixtures of grasses, non-legume forbs, and legumes and non-legume forbs at around 50% of each (Fig. 1B). The effect of grazing varied depending on the location of the experiment. While in Central France grazing had a positive effect on all three diversity indices, in Catalonia it had a negative effect.

Fungal diversity responses also varied by site. In Theix, fungal richness was not affected by interactions among plant functional types, though higher values were observed with increased grass dominance. In contrast, Shannon's diversity and Pielou's evenness indices showed a three-way interaction, with the

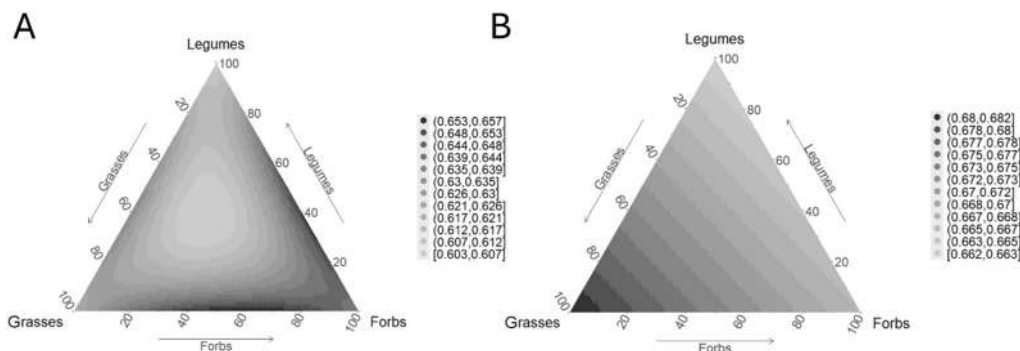


Figure 2. (A) Central France fungal Pielou's diversity index based on the model selected by the lowest AIC. (B) Fungal Pielou's evenness index for the model applied to Catalonia. In both images the axes represent the proportions of the different functional groups in the experiment. Darker colours represent higher values.

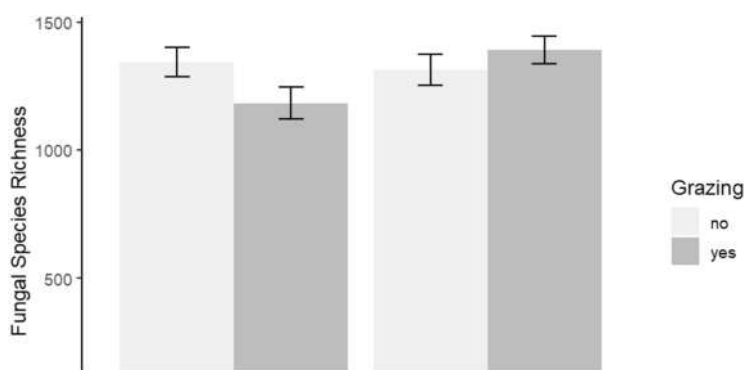


Figure 3. Fungal species richness based on site and the grazing treatment. Darker colours represent mixtures grazed while the light grey colour represents non-grazed mixtures.

lowest values occurring in mixtures with equal proportions of all plant functional types (Fig. 2A). In Solsona, fungal richness and Shannon's diversity were not influenced by plant functional type composition, but were instead modulated by grazing treatment. Pielou's evenness, however, was affected by plant functional types, showing no interaction effects and reaching its highest values in grass-dominated mixtures (Fig. 2B).

Grazing showed a significant interaction effect, enhancing fungal species richness in central France and decreasing it in Catalonia (Fig. 3).

Conclusion

Forage mixtures and soil microbial diversity are intricately linked, with this relationship strongly influenced by site-specific edaphic and climatic conditions. Management practices, particularly grazing, also play a key role, with grazing effects varying by location. Unlike previous studies where mixtures near the centroid often maximize ecosystem functionality, in the case of microbial diversity indices, our results suggest that centroid mixtures tend to exhibit the lowest soil microbial diversity. Further research with larger sample sizes and additional environmental variables is needed to deepen our understanding of these dynamics. Especially the relationship between microbial diversity, ecosystem service delivery and the interaction between the two factors mentioned above and the shown diversity.

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Recovery of lucerne sward after mulching in subsequent regrowth

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Abstract

Perennial forage legumes are a key component of sustainable agriculture systems; however, their utilization as feed can be limited due to the reduced number of livestock in some regions. This study explores alternative uses of lucerne sward by analysing the effect of forage mulching on subsequent sward regrowth. We compared the impact of two dates of cutting and mulching in the first harvest on biomass yield and sward height in the following regrowth. The result showed that lucerne mulching can negatively affect sward recovery, which can influence the benefits of their cultivation compared to standard management. This study contributes to understanding factors affecting the modified management of forage crops regarding the sustainability of the agricultural system.

Keywords: biomass yield, forage management, lucerne, mulching, sward regrowth

Introduction

Perennial forage legumes such as lucerne (*Medicago sativa* L), are a key component of sustainable agricultural systems due to valuable attributes such as biological nitrogen (N₂) fixation and its associated benefit on subsequent crops, improved infiltration of water into soil, and reduced weed pressure (Pietsch *et al.*, 2007). In Czechia, the decline in cattle and sheep herds has significantly reduced the use of grasslands for fodder production, leading to challenges in managing forage crops (Gaisler *et al.*, 2018; Šrédl *et al.*, 2021) where especially the absence of leys in crop rotation and reduced organic fertilization can have negative effects on soil fertility and health. Under this pressure, it is increasingly important to identify management strategies that maintain some proportion of perennial forages in the crop rotation in association with their ecosystem benefits. Harvested forage can be processed into a 'flexible fertility input' through anaerobic digestion or composting and then reapplied to the field, thereby recycling nutrients (Kruijff *et al.*, 2008), but these systems require special facilities as well as being costly. Mulching of lucerne is a more accessible solution and has been identified as a way of enhancing soil N balance by returning symbiotically fixed N₂ to the soil, as the crop can fix 159–230 kg N ha⁻¹ year⁻¹ (Loges *et al.*, 1999). Raza *et al.* (2018) also note that improving water retention and infiltration in soil by mulching is possible by reducing soil evaporation and runoff. For sustainable farming systems that rely on diversified crop rotations, inclusion of perennial forages and closed nutrient cycles for fertility can be beneficial without a direct link to livestock production. However, in contrast with traditional regular cutting, mulching can modify lucerne regrowth and reduce the ecosystem services it offers. This study aims to investigate how different lucerne mulching management practices affect sward recovery and dry matter yield from two field experiments in Czechia.

Table 1. Effect of management practice on lucerne sward regrowth; average compressed stand height (CH, cm) and dry matter yield (DMY, Mg ha⁻¹) in Červený Újezd

Location	Treatment	Date					
		14 May 2025		5 June 2025		17 June 2025	
		Harvest 1		Regrowth		Harvest 2	
		CH	DMY	CH	CH	DMY	CH
Červený Újezd	LF Cut	—	—	—	—	9.5 ^a	43.5 ^a
	LF Mulch	—	—	—	—	8.1 ^b	42.8 ^a
	HF Cut	52.8	6.2	25.4 ^b	50.0	4.9 ^c	33.8 ^b
	HF Mulch	51.8	6.2	20.4 ^a	46.2	3.4 ^d	28.5 ^b
<i>P</i> treatment		0.667	0.999	0.013	0.247	<0.001	<0.001

P, probability of ANOVA, different letters indicate statistical differences between treatments (Tukey HSD test at *P* < 0.05).

Materials and methods

Two field experiments were established on older lucerne swards at two different sites in Czechia: Červený Újezd (410 m asl, third harvest year) and Uhříněves (282 m asl, first harvest year), to explore the effects of management system on lucerne regrowth and biomass yield. Each experiment was set up to compare cutting and mulching at two intensities (two and four times per year in total) resulting in four treatments: low frequency (LF) and high frequency (HF) of cutting or mulching. Both experiments were set up using a randomized block design in four replicates. The plot area was 9 m² in Červený Újezd and 12 m² in Uhříněves. Compressed height (CH, cm) was measured using a rising plate meter on the dates given in Table 1, taking six measurements per plot. Biomass from each plot was harvested, weighed and removed (cut treatments) or mulched in the whole plot area (mulch treatments) where 2 quadrats of 1 m² were harvested per plot to estimate forage production. Mulching was carried out in one single pass using a mulcher (KDL 180 Profi, Stark, Lithuania). Fresh matter yield was measured per plot/quadrat and dry matter yield was calculated from the average dry matter content. The effect of treatments on sward height and dry matter yield was analysed separately for each harvest within each location using analysis of variance (ANOVA, *p* < 0.05) in R Studio (R Core Team, 2021), where all factors were treated as fixed. Significant differences between means were evaluated using the Tukey HSD test at $\alpha = 0.05$. Additionally, a two-way ANOVA was performed to evaluate the combined effects of treatment, site and their interaction on sward productivity.

Results and discussion

The results presented in Table 1 are based on an investigative period spanning May to July 2025, during which the first and second harvest took place, with two further harvests planned for later in the growing season. For high frequency treatments, the sward height recovery was negatively affected after the first mulching in Červený Újezd (Table 1, annual temperature 10°C, precipitation 445 mm) as well as Uhříněves (Table 2, annual temperature 8.3°C, precipitation 584 mm). However, a decrease in dry matter yield in comparison to the HF cut treatment was observed only in Červený Újezd. This reduction of yield is contrary to similar studies done by Raza *et al.* (2014) and Pietsch *et al.* (2007) which reported that utilization system (i.e. cutting or mulching lucerne) did not have a marked influence on yield. As further harvests are completed in this experiment, utilization system will likely continue to exacerbate differences in stand performance. When comparing regrowth after the first and second harvest, in Uhříněves, the HF mulch plots were on average 15.8% taller after the second compared with only 0.9% for the HF cut plots.

Table 2. Effect of management practice on lucerne sward regrowth; average compressed stand height (CH, cm) and dry matter yield (DMY, Mg ha⁻¹) in Uhříněves

Location	Treatment	Date					
		16 May 2025		5 June 2025		20 June 2025	
		Harvest 1		Regrowth		Harvest 2	
		CH	DMY	CH	CH	DMY	CH
Uhříněves	LF Cut	—	—	—	—	7.7 ^a	26.2 ^a
	LF Mulch	—	—	—	—	7.9 ^a	25.9 ^a
	HF Cut	56.7	5.0	22.5 ^b	50.8	3.3 ^b	22.7 ^b
	HF Mulch	56.7	4.9	18.4 ^a	47.3	4.0 ^b	21.3 ^b
<i>P</i> treatment		0.983	0.825	0.005	0.082	<0.001	<0.001

P, probability of ANOVA, different letters indicate statistical differences between treatments (Tukey HSD test at *P* < 0.05).

Productivity was significantly higher in the LF treatments, as root reserve replenishment is enhanced with longer intervals between defoliations and plants have longer to regrow and recover sward height (Teixeira *et al.*, 2007). Similarly, a study by Testa *et al.* (2011), investigating harvest timing of lucerne, found that forage yield was reduced by cutting earlier, as it was in our experiment where the harvest was completed earlier in the growing season.

In comparison of two locations, lucerne stands in Uhříněves were significantly shorter after Harvest 2 than in Červený Újezd.

We conclude that higher frequency of mulching can significantly reduce lucerne regrowth, which may be associated with lower sward height and biomass production. Optimization of the mulching frequency can help with successful adoption of mulching management into systems without livestock.

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