

The effect of sward type and simulated grazing system on ecosystem services of grassland on a sandy soil

N.J. Hoekstra^{a,*}, M.J.J. Hoogsteen^{b,c}, J. de Wit^a, E.A. Lantinga^b, N. van Eekeren^{a,b}

^a *Louis Bolk Institute, Kosterijland 3-5, Bunnik, the Netherlands*

^b *Farming Systems Ecology Group, Wageningen University and Research Centre, Droeendaalsesteeg 1, Wageningen 6708 PB, the Netherlands*

^c *Ministry of Agriculture, Fisheries, Food Security and Nature, Bezuidenhoutseweg 73, The Hague 2594 AC, the Netherlands*

ARTICLE INFO

Keywords:

Climate change adaptation and mitigation
Forage production
Habitat provision
Soil quality

ABSTRACT

Grasslands, which cover approximately 67% of the world's agricultural land, deliver a wide range of ecosystem services. This study aimed to quantify the effects of sward type and grazing system on grassland ecosystem services—specifically forage production, climate regulation, water retention, and habitat provision—on a sandy soil in the Netherlands. A field experiment established in 2011 compared four sward types: *Lolium perenne* without fertiliser (Lp0), *L. perenne* or *Festuca arundinacea* with 250 kg N ha⁻¹ yr⁻¹ (Lp250 and Fa250), and a low-input mixture of *L. perenne*, *F. arundinacea*, and *Trifolium repens* receiving only 50 kg N ha⁻¹ yr⁻¹ in the form of organic manure (LFT50). These four sward types were subjected to three simulated grazing systems: continuous grazing (CG, 4 cm stubble height, high cutting frequency), lenient/regenerative grazing (LG, 10 cm stubble height and high cutting frequency), and rotational grazing (RG, 4 cm stubble height and low cutting frequency). Results from 2013 to 2016 showed distinct trade-offs among ecosystem services. Rotational grazing maximised herbage yield, continuous grazing enhanced forage quality and clover content, and lenient grazing tended to be most beneficial for soil carbon and water retention. Lp0 consistently underperformed across all services, indicating that extreme extensification may reduce multifunctionality. Lp250 and Fa250 supported high productivity and climate regulation but were less beneficial for habitat quality. LFT50 offered a balanced performance across forage production and quality, and biodiversity, particularly under CG, though water retention and climate regulation was less than anticipated. Overall, no single sward type or simulated grazing strategy combination optimised all ecosystem services simultaneously. These findings underline the need for integrated grassland management approaches that balance productivity with ecological sustainability under varying land-use objectives and environmental conditions.

1. Introduction

The concept of ecosystem services has been valuable in raising attention to the importance of economically neglected outputs of agricultural systems (provisioning and cultural services) and of system functions that are essential for maintaining the provision of these outputs (regulating and habitat services), particularly since the formalisation in the Millennium Ecosystem Assessment (MEA, 2005). By offering an approach to measure and value these services, the concept of ecosystem services and related valuation concepts like True Cost Accounting, can facilitate more balanced decision making by e.g. policy-makers, consumers and farmers (FAO, 2023).

67% of world's agricultural land is covered by grasslands (FAO, n.d.)

that provide a wide range of ecosystem services. Zhao et al. (2020), however, show that ecosystem services research with a focus on grassland is limited. Based on a review by (Isselstein and Kayser, 2014), we selected 'forage production', 'climate regulation', 'water retention' and 'habitat quality' as the most relevant ecosystem services to be assessed in our experiment on grassland management systems.

Forage production in terms of dry matter production is an indicator related to food production. Agriculture faces the challenge of an increasing global demand for food against a backdrop of the urgency to maintain production systems and make them more resilient to a changing and ever more volatile environment. Besides productivity, the quality of the produced forage in terms of protein content and digestibility is equally important as this determines the efficiency of the

* Corresponding author.

E-mail address: n.hoekstra@louisbolk.nl (N.J. Hoekstra).

<https://doi.org/10.1016/j.agee.2026.110548>

Received 11 July 2025; Received in revised form 12 May 2026; Accepted 21 May 2026

Available online 31 May 2026

0167-8809/© 2026 The Authors. Published by Elsevier B.V. This is an open access article under the CC BY license (<http://creativecommons.org/licenses/by/4.0/>).

conversion into meat and milk (Isselstein and Kayser, 2014; Tonn et al., 2021).

Climate change is one of the most urgent current changes affecting agriculture. An important aspect of climate regulation in grassland systems is its function as a potential carbon sink, through a build-up of soil organic matter (SOM) (Iepema et al., 2022). SOM changes in grassland systems depend on the input and decomposition of new and existing organic matter. Organic matter inputs may occur in the form of animal manures, decaying plant roots and aboveground plant tissue, whereas decomposition rates depend on climatic factors (temperature, moisture) and edaphic factors (soil texture and microbial activity) and the chemical composition of the organic material (Jackson et al., 2017). In addition to carbon sequestration, root development and SOM content have an important impact on other ecosystem services, including water regulation (Deru et al., 2018).

Increasing demand for agricultural products has led to an intensification of agriculture which is accompanied by a reduction in habitat services, aboveground (Rigal et al., 2023) as well as belowground (Tsiafouli et al., 2015). An important aspect of habitat services in the Netherlands is the availability of food sources (i.e. earthworms, insects) for meadow birds. Moreover, in spring, tall, dense swards can form a major impediment for meadow-bird chicks foraging insects (Devereux et al., 2004), which makes sward biomass or sward height in spring an important indicator for habitat quality (De Haas et al., 2019). Additionally, in this period, it is important to minimise the level of disturbance in terms of defoliation events, as this is associated with low nest survival (Grüebler et al., 2012).

There are multiple ways in which the deliverance of these grassland ecosystem services can be affected, among which grassland species composition, fertilisation and grazing management. In terms of species composition and fertilisation, productive grasslands in western Europe are dominated by perennial ryegrass (*Lolium perenne* L., Lp) which is suitable for both grazing and cutting, has high nutritional value and palatability and high yield potential (Isselstein et al., 2005). Productivity of perennial ryegrass is highly reliant on organic and inorganic fertilisers. Generally, there is a positive effect of fertilisation on forage production and N content, and a small positive effect on root biomass production has been reported (Cougnon et al., 2017; Tomaškin et al., 2013). However, the effect on SOM build-up is less straightforward, as fertilisation not only affects the amount but also the quality of the plant material. A synthesis by Conant et al. (2017) showed that soil C sequestration rates were generally positive for both organic and inorganic fertilisers, but the sequestration rates were much higher for organic fertilisers because of increased application rates of organic matter.

Species with a higher root and stubble biomass, such as *Festuca arundinacea* (Fa) (Cougnon et al., 2017) could increase the potential for climate regulation and water retention, through an increase in SOM content and improved soil structure. Additionally, its deep rooting system allows Fa to explore deeper water reserves and contributes to a good adaptation to dry conditions (Fariaszewska et al., 2020; Wilman et al., 1998; Surault et al., 2007; Poirier et al., 2012). However, forage quality in terms of digestibility is generally lower for Fa than for Lp (Cougnon et al., 2014).

Inclusion of white clover (*Trifolium repens*, Tr) has many potential benefits in terms of its attractiveness for pollinator insects, earthworms and a positive effect on forage quality in terms of digestibility and protein content (De Haas et al., 2019; Harris & Ratnieks, 2022). Also, because of symbiotic N fixation the production level of grassland can be maintained at lower inputs of (artificial) fertiliser (Lüscher et al., 2014). However, white clover root biomass is relatively low compared to grasses (De Haas et al., 2019; van Eekeren et al., 2009a). Combining multiple plant species has been reported to have positive effects on ecosystem services due to diversity benefits for productivity (Finn et al., 2013; De Haas et al., 2019; Zetterlind et al., 2025), root distribution and biomass, which may increase SOM formation and storage (Lange et al.,

2015; Bai and Cotrufo, 2022).

While there is an increasing body of research on the effects of grazing on ecosystem services, the focus is mainly on the effect of grazing pressure (in relation to overgrazing) (Ren et al., 2018; Maestre et al., 2022; Tuo et al., 2024) while the effect and difference between different grazing systems is much less researched and is mainly focussed on forage production and quality, and not on other ecosystem services (e.g. Parsons and Chapman, 2000). The main parameters defining grazing systems are the rotation interval (time between two consecutive grazing events) and the stubble height to which the swards are grazed (which is a result of stocking rate and duration; i.e. grazing pressure) (Visscher et al., 2011). Rotational grazing (low stubble height, relatively long rotation interval) and continuous grazing (low stubble height, short or virtually absent rotation interval) are the two systems that are most commonly used in practice in productive grazing systems (Visscher et al., 2011). The longer rotation intervals associated with rotational grazing generally result in higher aboveground production than for continuous grazing (Parsons and Chapman, 2000; Hoekstra et al., 2019). White clover performs relatively well in continuous grazing systems due to reduced competition for light with grasses (Schwank et al., 1986). At the same time, high defoliation frequency and intensity have been associated with both lower (Dawson et al., 2000) and higher (Ennik et al., 1980; Deinum, 1985) root biomass, all closely linked to the availability of stored carbohydrates, root/shoot ratio and tiller density.

There is increasing attention for regenerative grazing. This grazing system is characterized by the stocking of large herds of livestock on multiple small paddocks for short periods (Morris, 2021). Based on this, the “Pure-graze” or lenient strip graze system has been developed, which is primarily aimed at grazing with dairy cows (for which feed quality is more important than for beef). This system is characterised by high pre-grazing herbage mass, short grazing duration (less than one day) and high post grazing stubble height (Visscher et al., 2011). One of the aims of this system is to increase the input of plant residues to the soil (higher grazing losses from senescing stubble material). However, the high stubble height may negatively affect the quality and production of the grazed forage and also negatively affect the performance of white clover (Schils and Sikkema, 2002).

While there is some information on the independent effects of sward type and grazing system on ESS, there is very little on the combined effect, even though interactions are likely. For example, Maestre et al. (2022) reported more positive effects of grazing in species rich areas. In order to design grassland systems that maximise the different ecosystem services, we need more information on the performance of different sward types in interaction with grazing systems. Our objective was to quantify the effect of sward type (combination of species composition and fertilisation), simulated grazing system and their interaction on the ecosystem services of grassland focusing on forage production, climate regulation, water regulation, and habitat provision on a sandy soil. To this end we designed an experiment with four sward types (Lp0: Lp without fertiliser, Lp250: Lp with organic and inorganic fertiliser, Fa250: Fa with organic and inorganic fertiliser and LFT50: a mixture of Lp, Fa and white clover with only organic manure) which were subjected to three simulated grazing systems (rotational grazing, continuous grazing and lenient (regenerative) grazing). We formulated the following hypothesis regarding the effect of sward type and simulated grazing system on ecosystem services:

(i) We expect forage productivity to be highest for Lp250 and Fa250 and lowest for Lp0, due to the lower fertilisation rate in the latter. In LFT50, reduced fertilisation is expected to be partly compensated for by the symbiotic N fixation by white clover. Forage quality, in terms of protein content and digestibility, is expected to be highest for Lp250 and LFT50 because of the positive contribution of white clover. The relatively long regrowth interval under RG is expected to increase forage production compared with CG and LG. In contrast, forage quality is expected to be highest under CG due to the short regrowth interval and low stubble height. Finally, the LFT50 sward type is hypothesised to

perform best under the CG grazing system, as CG is expected to promote the proportion of white clover in the sward; (ii) We expect SOM levels, and consequently climate regulation, to be highest in Fa250 and Lp250 due to their high productivity and strong root development, particularly in Fa250, and lowest in Lp0. Similarly, SOM levels are expected to be highest under LG because of the greater input of decaying stubble material to the soil; (iii) We expect water regulation, in terms of water holding capacity and water infiltration, to improve with increasing SOM, as described above. In addition, sward types and grazing systems that stimulate earthworm abundance (see below) and root development (particularly Fa250 and Lp250) are expected to enhance water infiltration; (iv) We expect white clover to have a positive effect on earthworm abundance, particularly in the LFT50 x CG combination, thereby benefiting both soil biodiversity and the food availability for meadow birds. Maximum sward height in spring is expected to be greatest in highly productive sward types and grazing systems (Lp250 and Fa250, RG), which may negatively affect the mobility of meadow bird chicks. Long regrowth periods under RG are also expected to increase sward height, however, this negative effect may be partly offset by the reduced frequency of disturbance during spring.

2. Materials and methods

2.1. Experimental design

The simulated grazing experiment was established in April 2011 on a sandy podzol (clay content: 5%, sand content 89%, silt content, 5%) at “De Borse Weilanden” in Wageningen, the Netherlands (51°59'34.0"N; 5°39'19.1"E). Before the experiment started, the site had been under arable cropping for more than 40 years, and the preceding crops were spring barley, maize, potato and winter wheat. During the experimental period (2014–2016) reported in this paper, the mean annual temperature was 11.1 °C and the average annual precipitation sum amounted to 854 mm. The largest cumulative rainfall deficit (precipitation minus potential evapotranspiration of a standard field with a growing crop, calculated from April 1st) occurred in July and August 2015 (–180 mm), whereas the rainfall deficit did not exceed –40 mm in 2014 and 2016 (S1).

The experiment was laid out in a factorial design with four sward types and three simulated grazing systems. The four sward types consisted of a combination of species composition and fertilisation, as summarised below:

Lp0. : Perennial ryegrass (*Lolium perenne* L., cvs. Barflip, Barforma and Arsenal) without N fertilisation.

Lp250. : Perennial ryegrass (cvs. see above) fertilised with 50 kg N ha^{–1} yr^{–1} in the form of cattle manure granules + 200 kg N ha^{–1} yr^{–1} in the form of calcium ammonium nitrate (CAN).

Fa250. : Tall fescue (*Festuca arundinacea* Schreb., cvs. Barolex and Bariance) fertilised with 50 kg N ha^{–1} yr^{–1} in the form of cattle manure granules + 200 kg N ha^{–1} yr^{–1} in the form of CAN.

LFT50. : A mixture of perennial ryegrass, tall fescue and white clover (*Trifolium repens* L. cvs. Alice and Riesling), fertilised with 50 kg N ha^{–1} yr^{–1} in the form of cattle manure granules only.

The seeding rates were 30 kg ha^{–1} for grass and 3 kg ha^{–1} for clover. The application of manure granules corresponded to an organic matter (OM) input from cattle manure of approximately 2.2 Mg OM ha^{–1} year^{–1}. Swards under the continuous grazing system received five split applications of CAN, whereas those under rotational and lenient grazing systems received three split applications of CAN (see Hoogsteen et al., 2020 for more details).

The three grazing systems were simulated by mowing the plots at different cutting heights and at different frequencies, representing continuous grazing (CG, 4 cm stubble height and high cutting

frequency), lenient grazing (LG, 10 cm stubble height and high cutting frequency) and rotational grazing (RG, 4 cm stubble height and low cutting frequency) as outlined in Table 1. The management in 2012 and 2013 was similar, see Hoogsteen et al. (2020).

The experiment was laid out in a split-plot design with four replicated blocks consisting of three main plots each, to which the three grazing systems were randomly assigned. Sward type (Lp0, Lp250, Fa250, LFT50) was randomly assigned to four sub plots within these main plots. Hence, the experiment consisted of 12 main plots (4 blocks x 3 grazing systems) and 48 sub plots (see Figure S1.1 for schematic representation of the experimental design). The sub-plot size was 2.75 × 10 m.

2.2. Sampling and measurements

In this paper we present the results from 2014, 2015 and 2016, representing the third, fourth and fifth full growing year of the swards. Measurements to assess the different ecosystem services of the grassland systems were carried out throughout the three monitoring years, to ensure capturing the medium-term effect of the different treatments, which only may become apparent after a few years (see also S1).

2.2.1. Forage production and quality

At each harvest, a 1.5 m × 10 m strip was cut at the assigned stubble height (Table 1) using a Haldrup grass harvester (Haldrup, Løgstør, Denmark) and total herbage biomass was recorded. A sub-sample was collected for dry matter analyses (dried overnight at 70 °C). The total N, P, K, S and digestible organic matter content of the dried and ground herbage was determined using near infra-red spectroscopy (NIRS, Eurofins Agro, The Netherlands).

During two (2014 and 2015) and four harvests (2016), a 50 cm × 50 cm square was cut from the central plot area at the treatment specific stubble height in all LFT50 plots just before the harvest (at harvests when all three grazing systems were cut at the same date). This herbage sample was sorted into clover and grass, and dried at 70 °C for 24 h to determine the proportion of white clover in the swards.

In 2016, before each harvest the sward height was measured with a falling plate meter (3 measurements per plot), and the maximum sward height in spring (before June 15) was calculated.

2.2.2. Soil chemical properties

In October 2016, in each sub-plot a soil sample was taken consisting of 20 soil cores of 0–10 cm depth using a grassland auger with a 2 cm diameter. Soil samples were pooled per sub-plot and part of this soil was sent to Eurofins Agro (Wageningen, the Netherlands) and analysed for N-total, Potential mineralisable N, C-total and SOM using near infra-red spectroscopy (NIRS, Eurofins Agro, the Netherlands). The other part was subjected to Soil Health Analysis (Moebius-Clune et al., 2016) at Cornell University, USA, which included available water capacity, extractable P and K analysis (See S2 for more information).

2.2.3. Soil physical properties

In October 2014 and 2015, the water infiltration was measured on three randomly chosen spots in each sub-plot. To this end, a PVC pipe of 15 cm high (ø of 15 cm) was driven into the soil to a depth of 10 cm, after which 500 ml of water was gently poured into the ring. The time required for the 500 ml water to infiltrate was recorded and the water infiltration rate (mm minute^{–1}) was calculated. On the same days, soil penetration resistance was measured with an electronic penetrometer (Eijkelkamp, Giesbeek, The Netherlands) with a cone diameter of 1 cm² and a 60° apex angle. Cone resistance was recorded per cm of soil depth and expressed as an average value of 20 penetrations per plot in the soil layers of 0–10 cm, 10–20 cm and 20–30 cm.

In October 2015, visual assessments of soil structure and rooting were conducted on 20 × 20 × 10/15 cm soil cubes from the 0–10 cm and 10–25 cm soil layer. Cubes were dug out with a spade and broken in both

Table 1

Overview of the cutting regime of the simulated grazing systems in 2014, 2015 and 2016.

	Simulated grazing system								
	Continuous grazing (CG)			Lenient grazing (LG)			Rotational grazing (RG)		
	2014	2015	2016	2014	2015	2016	2014	2015	2016
Stubble height (cm)	4			10			4		
Average sward height at defoliation (cm)	11	14	11	17	16	17	18	16	18
Number of cuts per year (n)	9	7	8	8	6	8	4	4	4
Average cutting interval (days)	23	26	25	26	30	25	51	49	49

horizontal and vertical direction. Soil structure was assessed by estimating the proportion of soil crumbs, sub-angular blocky elements and angular blocky elements in the cubes, following the method by Peerlkamp (1959) and Shepherd (2000). Rooting score was assessed by scoring visible root density (score 1–10; 1 for no roots and 10 for high root density), with an estimation of the proportion of young roots relative to the total (De Boer et al., 2018). Soil biota activity was assessed by scoring the abundance of soil pores (score 1 – 10; 1 = no visible pores and 10 = above average pore density) (De Boer et al., 2018).

2.2.4. Soil biological properties

In October 2014, earthworms were sampled in two soil blocks (20 cm × 20 cm × 20 cm) per sub-plot. The blocks were transferred to the laboratory where the whole block was broken down and earthworms were hand-sorted, counted, weighed and fixed in alcohol prior to identification to species and classification into functional groups (epigeic, endogeic and anecic species) (Bouché, 1977). Abundance and biomass were expressed per m².

2.2.5. Root biomass and degradability

Roots (both standing and decayed root biomass) are an important component of soil organic matter. Root biomass was determined in July 2013 and March 2014 (previously reported by Hoogsteen et al., 2020). Two soil cores with an 8 cm diameter were collected to a depth of 60 cm in each subplot using a hydraulic auger. Each core was split into 0–10, 10–30 and 30–60 cm soil depth intervals and pooled per sub-plot. The samples were soaked in lukewarm water for a few hours, after which the soil was washed off the roots by extensive rinsing with running water. All non-root organic matter was removed from the sample, and the root material was dried at 70 °C overnight.

In order to get more insight in the possible contribution of this root material to soil organic matter buildup, for the Lp250 and Fa250 treatments, the dried root material was combined over all grazing treatments and stored under cool and dry conditions and OM, C and N content of the root material was determined. Subsequently, this material was used to assess root degradability in a litterbag experiment and a gass-bottle experiment (S3 and S4 for more details).

2.3. Calculations

All individual responses underpinning the different Ecosystem Services (ESSs) were standardized from 0 to 1 using Eq. 1:

$$ESS = [ESS_{RAW} - \min(ESS_{RAW})] / [\max(ESS_{RAW}) - \min(ESS_{RAW})] \quad (1)$$

where, ESS_{RAW} represents untransformed ecosystem service values, following Xu et al. (2024). Based on these standardized individual responses, average ESS indices were calculated for Forage Production (FP_DMY = herbage dry matter yield, FP_quality = N concentration, Organic matter digestibility), Climate regulation (SOM and SOC), Water regulation (Water infiltration 2014 and 2015, Water capacity) and Habitat provision (meadowbird habitat, earthworm parameters, proportion clover). Meadowbird habitat was based on the maximum sward

height in spring (2016) and the number of defoliation events before June 15. As for both parameters, a higher value corresponds to a negative impact on the ecosystem service, the respons was calculated as 1 – ESS. Earthworm parameters included Earthworm abundance, Earthworm biomass, Earthworm individual weight, Epigeic earthworm abundance and number of species. In a final step, the main ESSs were recalibrated to an average respons of 0.5, and the overall mean across all ESS was calculated.

2.4. Statistical analysis

The effects of sward type (Lp0, Lp250, Fa250, LFT50) and grazing system (CG, LG, RG) and their interactions on ecosystem services were determined using general linear mixed effects models. Sward type and grazing system were considered fixed factors, while block and “block × grazing system” (equivalent to plot) were used as random factors. Statistical analyses were performed in R 3.6 (R Core Team, 2020) with the packages lme4/lmerTest (Bates et al., 2015; Kuznetsova et al., 2017). All data were checked for normality, outliers and homoscedasticity to ensure all assumptions for ANOVA were met. When significant effects were detected, data were subjected to post hoc tests (Day and Quinn, 1989) using the emmeans/multcomp packages in R (Hothorn et al., 2012; Lenth, 2019) with Tukey HSD adjustment for multiple comparisons. Pearson correlation analyses was carried out in order to investigate relations between the different ESSs and related response variables.

Additionally, we applied a reflected discriminant analysis (RefDA) to improve our insight into the relationship between the ESSs and the simulated grazing and sward type treatments. RefDA is a principal component analysis reflected by group information giving reflected components (Nierop et al., 1994). In order to investigate the relation with the soil chemical, physical and biological parameters, their correlations with reflected components were computed and visualized in corresponding RefDA biplots.

3. Results

3.1. Forage production and quality

3.1.1. Herbage dry matter yield (DMY)

There was a highly significant ($P < 0.001$) interactive effect of sward type × year on DMY (Fig. 1a, Table S5 and S6). On average, DMY increased in the order Lp0 < LFT50 = Lp250 < Fa250 ($P_{ST} < 0.001$). For Lp0 and Lp250, DMY was not significantly affected by year and DMY of Fa250 was slightly lower in 2016 compared to the two previous years. However, LFT50 showed a strong decrease in DMY from 10.9 ton ha⁻¹ yr⁻¹ in 2014 (similar to Fa250 and significantly higher than Lp250) to 6.2 ton ha⁻¹ yr⁻¹ in 2016 (significantly lower than Lp250).

DMY was also highly significantly ($P < 0.001$) affected by the interactive effect of grazing system × year (Fig. 1b). On average across years, DMY was 5.4, 7.3 and 9.5 ton DM ha⁻¹ yr⁻¹ for grazing system LG, CG and RG, respectively.

There was a highly significant ($P < 0.001$) sward type × grazing system interactive effect on DMY (Fig. 1c). For sward types Fa250 and Lp250, DMY was significantly higher for RG compared to LG and CG. For

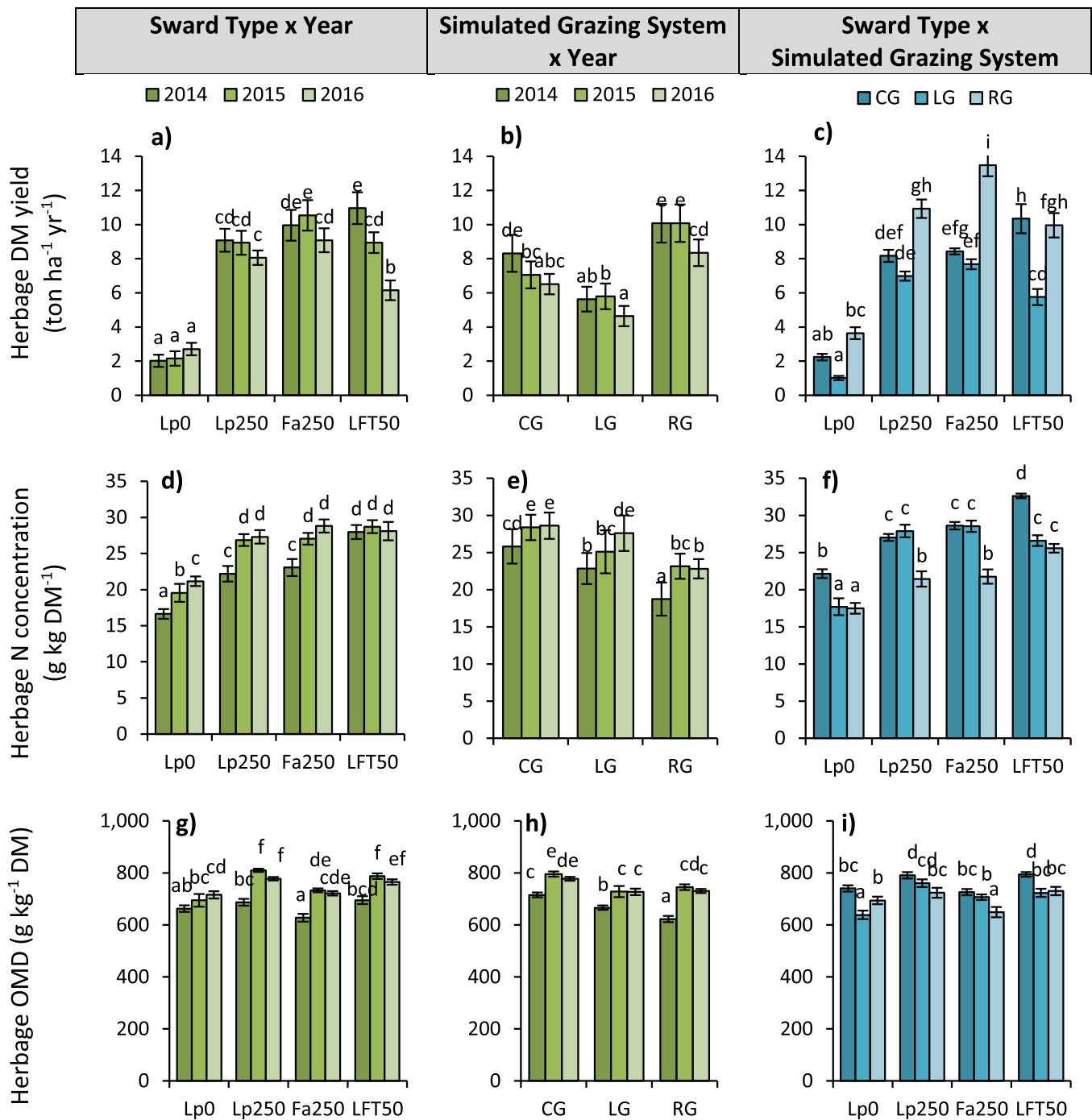


Fig. 1. The interactive effect of sward type (Lp0 = *Lolium perenne* without fertilisation, Lp250 = *Lolium perenne* with fertilisation (Manure + CAN), Fa250 = *Festuca arundinacea* with fertilisation (Manure + CAN), LFT = mixture of Lp, Fa and *Trifolium repens* (Manure only)) by year (a, d, g), simulated grazing system (CG = continuous grazing, LG = lenient or regenerative grazing, RG = rotational grazing, see Table 1) by year (b, e, h) and sward type by grazing system (c, f, i) on the cumulative herbage dry matter yield (ton ha⁻¹ yr⁻¹) (a, b, c), herbage N concentration (g kg⁻¹ DM) (d, e, f) and herbage organic matter digestibility (g kg⁻¹ DM) (g, h, i). Error bars represent one standard error (n = 16 for b, e, h, n = 12 for rest). Bars with the same letters do not differ significantly from one another at P < 0.05 (Tukey's HSD). Treatment means are reported in Table S6.

LFT50, DMY for CG was significantly higher than LG.

3.1.2. Herbage N concentration

There was a significant ($P < 0.001$) sward type x year interaction on the herbage N concentration (Table S5, Fig. 1d). For all years, N concentration was lowest for Lp0 (on average 19 g kg⁻¹ DM) and highest for LFT50 (on average 28 g kg⁻¹ DM across all years). For LFT50, the N concentration remained stable across the years, whereas for the other

sward types, N concentrations increased from 2014 to 2016.

There was a significant ($P < 0.01$) grazing system x year interaction on N concentration (Fig. 1e). On average N concentration was highest for CG (27.6 g N kg⁻¹ DM), followed by LG (25.2 g N kg⁻¹ DM) and lowest for RG (21.6 g N kg⁻¹ DM). The difference between CG and LG was not statistically significant in 2016, when the N concentration under LG was relatively high.

For sward type Lp0 and LFT50, the N concentration was higher for

CG compared to LG and RG, whereas for Lp250 and Fa250, the N concentration was lower for RG compared to CG and LG, resulting in a significant sward type x grazing system interaction ($P < 0.001$, Fig. 1f).

3.1.3. Herbage organic matter digestibility (OMD)

There was a significant ($P < 0.001$) sward type x year interaction on herbage OMD (Table S5, Fig. 1g). In 2014, there was no effect of sward type on herbage OMD (on average $668 \text{ g kg}^{-1} \text{ DM}$). In 2015 and 2016, herbage OMD was lowest for Lp0, followed by Fa250 and highest for LFT50 and Lp250.

There was a significant ($P < 0.001$) grazing system x year interaction on OMD (Fig. 1h). On average, OMD was highest for CG ($763 \text{ g kg}^{-1} \text{ DM}$), followed by LG ($707 \text{ g kg}^{-1} \text{ DM}$) and lowest for RG ($699 \text{ g kg}^{-1} \text{ DM}$), but the difference between LG and RG was only statistically significant in 2014, when herbage OMD was relatively low.

There was a significant ($P < 0.001$) sward type x grazing system interaction on OMD (Fig. 1i). For Lp0 and LFT50, OMD was similar or lower for LG compared to RG, whereas for Fa250 and Lp250 the opposite was true.

3.1.4. Sward height in spring and disturbance

There was a significant sward type x grazing system interaction ($P < 0.001$) for the maximum sward height in spring (2016) (Fig. S10). The maximum sward height in spring was lower for Lp0 (17.7 cm) compared to the other sward types (22.8 cm) and was lower for CG (15.8) compared to LG (22.2) and RG (26.6 cm). The number of defoliation events before June 15 (indicator of disturbance) was 1 for RG, 2 for LG and 3 for CG.

3.1.5. Clover proportion

The proportion of white clover in the LFT50 sward ranged from less than 10% in October 2016 to 58% in July 2014 (Fig. 2). There was a significant effect of grazing system ($P < 0.05$, CG > LG) and Year ($P < 0.01$, 2014 > 2015 and 2016) on the proportion of clover (Table S5).

3.2. Soil chemical properties

There was a highly significant ($P < 0.001$) effect of sward type on soil organic C and SOM in the 0–10 cm soil layer measured in October 2016 (Table 2). Concentrations were lowest for Lp0 and highest for Lp250 and Fa250, with LFT50 in between. There was a significant ($P < 0.05$) effect of grazing system on soil organic carbon, where LG was higher than CG, with RG in between.

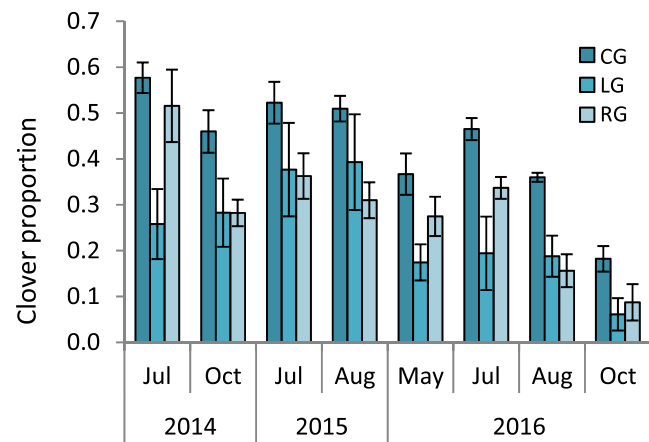


Fig. 2. The effect of simulated grazing system (CG = continuous grazing, LG = lenient or regenerative grazing, RG = rotational grazing, see Table 1) on the clover proportion in herbage DM in the LFT50 swards during selected harvests in 2014, 2015 and 2016. Error bars represent one standard error ($n = 4$).

Table 2
The effect of sward type ($n = 12$) and simulated grazing system ($n = 16$) on soil chemical properties at 0–10 cm depth in 2016. The F-values and their statistical significance (P-values) for the effect of sward type (ST), simulated grazing system (GS) and their interaction are presented.

Soil parameter	Sward Type ^a					Simulated grazing system ^b					F-value ^c		
	Lp0	Lp250	Fa250	LFT50	CG	LG	RG	ST	GS	STxGS	ST	GS	STxGS
Soil organic C (g kg^{-1})	17.1	21.3	21.3	19.3	18.6	21.0	19.6	22.6	6.3	*	22.6	6.3	*
SOM (g kg^{-1})	35.4	41.3	41.6	38.8	37.9	40.1	37.9	19.4	2.1	1.3	19.4	2.1	1.3
N-total (g kg^{-1})	1.4	1.7	1.6	1.5	1.5	1.6	1.6	11.7	2.6	0.9	11.7	2.6	0.9
PMN (mg kg^{-1}) ^d	46	65	65	59	55	65	57	17.7	3.2	1.6	17.7	3.2	1.6
pH water	6.0	6.2	6.2	5.9	6.1	6.1	6.0	27.7	4.3	*	27.7	4.3	*
Extractable P (mg kg^{-1})	29	37	39	29	33	38	29	52.7	14.0	**	52.7	14.0	**
Extractable K (mg kg^{-1})	114	65	56	46	62	96	53	124.0	62.6	***	124.0	62.6	***

^a Lp0 = *Lolium perenne* without fertilisation, Lp250 = *Lolium perenne* with fertilisation (Manure + CAN), Fa250 = *Festuca arundinacea* with fertilisation (Manure + CAN), LFT = mixture of Lp, Fa and *Trifolium repens* (manure only).

^b CG = continuous grazing, LG = lenient or regenerative grazing, RG = rotational grazing, DMV = dry matter yield

^c Statistical significance is indicated with *** $P < 0.001$; ** $P < 0.01$; * $P < 0.05$; $P < 0.1$. Values within a row followed by the same letters do not differ significantly from one another at $P < 0.05$ (Tukey's HSD)

^d PMN = potentially mineralisable N

All soil parameters related to nutrient availability were significantly ($P < 0.001$) affected by sward type (Table 2). N-total and potentially mineralisable N were significantly lower for Lp0 compared to all the other sward types. pH water and extractable P concentration were significantly higher for Lp250 and Fa250 compared to LFT50 and Lp0. The extractable K concentration was highest for Lp0, and lowest for LFT50.

3.3. Soil physical properties

There was a significant impact of sward type on different parameters associated with soil physical properties, however there was no significant effect of grazing system on any of these parameters (Table 3, S9). The percentage of crumbly soil elements was significantly ($P < 0.05$) lower for Lp0 compared to Fa250 at both 0–10 and 10–25 cm soil depth. The root score and macro pores score at both soil depths were highest for LFT50 followed by Fa250 and lowest for Lp0 ($P < 0.01$). The young root score was significantly lower for Lp0 compared to the other sward types at 10–25 cm soil depth ($P < 0.01$).

Water infiltration was significantly ($P < 0.01$) lower for Lp0 compared to the other sward types in both 2014 and 2015 (Table 3). Water capacity was highest for Fa250, and lowest for Lp0 and LFT50 ($P < 0.01$). There was no significant effect of grazing system on these two parameters.

3.4. Root biomass and degradability

3.4.1. Root biomass

There was a significant ($P < 0.001$) effect of sward type on root biomass in all soil layers (Table S8, see also Hoogsteen et al., 2020). In the 0–10 cm soil layer root biomass for Fa250 and Lp250 was significantly higher than for LFT50. In the deeper soil layers root biomass was higher for Fa250 compared to all other sward types. There was no significant main or interactive effect of grazing system on root biomass.

3.4.2. Root degradability

In the glass-bottle incubation experiment, after 112 days, the C decomposition of the root material was significantly ($P < 0.01$) lower for Lp250 (27.3%) compared to Fa250 (33.8%) (Fig. S3). In contrast, the dry matter decomposition in the in-situ litter bags was significantly ($P < 0.01$) higher for Lp250 compared to Fa250 (Fig. S4a). The decomposition rate was faster for Lp250 during the first 14 days of incubation (Fig. S4b), whereas there was no significant difference between

the two grass species thereafter.

3.5. Soil biological properties

The earthworm density was significantly ($P < 0.001$) affected by sward type, and was higher for LFT50 compared to Lp0 and Fa250 (Table 4). This higher density could be contributed to the epigeic earthworms ($P < 0.001$), whereas there was no effect of sward type on endogeic worms. The earthworm biomass was significantly higher for LFT50 compared to Lp0.

3.6. Ecosystem Services

Responses in ESSs were calculated based on standardised parameters underpinning the individual ESSs (Table 5). Sward type had a highly significant effect on all ESSs. Lp0 showed the lowest standardised score for all ESSs, with the exception of habitat provision (due to its positive effect on habitat quality for meadowbirds and clover). Simulated grazing system had a significant effect on forage production and habitat provision, but its effect on climate regulation and water regulation was marginally significant ($P = 0.05$) and non-significant, respectively. There was no single simulated grazing system that did better for all the selected ESSs, and the same was true when considering the interactive effects of sward type and simulated grazing system (no significant main or interactive effect of sward type on the average ESS, Table 5). For example, the LFT50 sward type under CG simulated grazing system was performing particularly well in terms of forage production and quality and habitat provision. However, for climate regulation and water regulation its performance was average or below. Correlations between the ESSs, their underlying parameters and soil chemical, soil physical, soil biological and habitat parameters are presented in Table S11.

3.7. Differentiation of simulated grazing system and sward type by RefDA

There was a strong differentiation of the simulated grazing systems and sward types based on the ESSs (Fig. 3a, Fig. S12). The first, second and third reflected components explained 39%, 28% and 14% of the variation, respectively, with corresponding group B/T ratios of 0.95, 0.91 and 0.82, respectively. The reflected components showed significant correlations with soil biological parameters soil physical parameters and soil chemical parameters and clover proportion (Fig. 3b and Fig. S12). There was a strong differentiation of sward type Lp0 with the other sward types along the first reflected component, reflecting the

Table 3

The effect of sward type on the physical soil properties (averaged across simulated grazing systems, $n = 12$, standard deviation in parenthesis). The F-values and their statistical significance (P-values) for the effect of sward type (ST), simulated grazing system (GS) and their interaction are presented.

Soil quality and water retention parameters	Year	depth (cm)	Sward Type ^a								F value ^b		
			Lp0		Lp250		Fa250		LFT50		ST	GS	STxGS
Penetration res. (MPa)	'14	0–10	0.73	(0.15)	0.69	(0.14)	0.79	(0.14)	0.69	(0.14)	1.2	2.2	0.1
	'15	0–10	0.84	(0.10)	0.78	(0.08)	0.88	(0.14)	0.85	(0.08)	2.7	1.3	3.3
Crumbs (%)	'16	0–10	47.1	(25.5) ^a	63.3	(15) ^{ab}	70.8	(15.5) ^b	62.9	(19.2) ^{ab}	3.0	*	1.4
	'16	10–25	1.7	(3.7) ^a	1.7	(3.7) ^{ab}	8.8	(8.4) ^b	7.5	(6.9) ^{ab}	4.5	**	0.8
Root score (1–10)	'16	0–10	6.3	(1.2) ^a	7.3	(0.7) ^{ab}	7.8	(1) ^b	7.4	(0.6) ^b	5.5	**	0.7
	'16	10–25	4.8	(1.5) ^a	5.3	(1.4) ^{ab}	6.3	(1.4) ^{bc}	6.5	(0.8) ^c	7.6	***	0.9
Young root score (1–10)	'16	0–10	6.3	(1.4)	6.8	(1.1)	6.8	(1.1)	7.2	(1.3)	1.2	1.7	1.2
	'16	10–25	2.8	(1.7) ^a	4.8	(2.2) ^b	5.0	(2) ^b	5.7	(1.7) ^b	6.6	**	0.4
Macro pores (1–10)	'16	0–10	6.2	(1.1) ^a	7.2	(1.1) ^{ab}	7.3	(0.6) ^b	7.7	(0.5) ^b	6.1	**	1.2
	'16	10–25	4.2	(1.8) ^a	5.4	(1.7) ^{ab}	5.8	(1.2) ^b	6.3	(1.3) ^b	5.4	**	1.1
Water infiltration (mm min ⁻¹)	'14		3.5	(2.9) ^a	19.5	(23.7) ^b	35.1	(40.4) ^b	19.0	(25.5) ^b	14.2	***	0.5
	'15		3.3	(2.5) ^a	12.7	(10.4) ^b	10.2	(7.6) ^b	11.3	(9.1) ^b	5.5	**	1.2
Water capacity (g g ⁻¹)	'16	0–10	0.092	(0.013) ^a	0.098	(0.015) ^{ab}	0.100	(0.015) ^b	0.093	(0.014) ^a	6.0	**	1.4

^a Lp0 = *Lolium perenne* without fertilisation, Lp250 = *Lolium perenne* with fertilisation (Manure + CAN), Fa250 = *Festuca arundinacea* with fertilisation (Manure + CAN), LFT = mixture of Lp, Fa and *Trifolium repens* (manure only)

^b Statistical significance is indicated with *** $P < 0.001$; ** $P < 0.01$; * $P < 0.05$; $P < 0.1$. Values within a row followed by the same letters do not differ significantly from one another at $P < 0.05$ (Tukey's HSD)

Table 4
The effect of sward type and simulated grazing system on earthworm parameters. The F-values and their statistical significance (P-values) for the effect of sward type (ST), simulated grazing system (GS) and their interaction are presented.

	Sward type ^a				Simulated grazing system ^b				F-value ^c			
	Lp0	Lp250	Fa250	LFT50	CG	LG	RG	ST	GS	STxGS		
Earthworm abundance (n m ⁻²)	579	(164) ^a	743	(180) ^{ab}	623	(211) ^b	739	(178)	597	(201)	779	(277)
Earthworm biomass (g m ⁻²)	159	(54) ^a	225	(64) ^{ab}	200	(52) ^b	223	(70)	198	(77)	210	(71)
Earthworm weight (g worm ⁻¹)	0.28	(0.07)	0.31	(0.09)	0.35	(0.11)	0.31	(0.08)	0.34	(0.12)	0.29	(0.09)
Fraction juvenile	0.78	(0.08)	0.82	(0.12)	0.73	(0.16)	0.79	(0.13)	0.72	(0.13)	0.84	(0.09)
Epigeic (n m ⁻²)	122	(64) ^a	165	(87) ^a	108	(65) ^a	168	(91)	168	(132)	182	(137)
Endogeic (n m ⁻²)	457	(154)	578	(178)	511	(227)	571	(184)	425	(144)	597	(201)
Nr of earthworm species	2.6	(0.5)	2.8	(0.7)	2.6	(0.9)	2.7	(0.6)	2.9	(0.9)	2.6	(0.6)

^a Lp0 = *Lolium perenne* without fertilisation, Lp250 = *Lolium perenne* with fertilisation (Manure + CAN), Fa250 = *Festuca arundinacea* with fertilisation (Manure + CAN), LFT = mixture of Lp, Fa and *Trifolium repens* (manure only).

^b CG = continuous grazing, LG = lenient or regenerative grazing, RG = rotational grazing, DMY = dry matter yield

^c Statistical significance is indicated with *** P < 0.001; ** P < 0.01; * P < 0.05; P < 0.1. Values within a row followed by the same letters do not differ significantly from one another at P < 0.05 (Tukey's HSD)

general under-performance of this sward type in all ESSs, associated mainly with low FP_Quality and FP_DMY, which correlated strongly with high available K (Fig. 3a). The second reflected component allowed differentiation between sward type LFT50 and Lp0 (associated with high habitat provision correlated to high clover proportion) and Lp250 and Fa250 with a reversed effect (associated with high climate regulation clustered with high SOM, SOC, N-total and pH). This differentiation was most pronounced for LFT50 under CG. For the sward types LFT50, Fa250 and Lp250, the third reflected component (Fig. S12) showed a differentiation between the RG simulated grazing system (aligned with higher FP_DMY, and lower FP_Quality and water regulation) compared to LG and CG simulated grazing system with a reversed effect.

4. Discussion

Both sward type and simulated grazing system had significant effects on the selected ESSs forage production and quality, climate regulation, water regulation and habitat provision, as discussed below and summarised in Table 5 and Fig. 3.

The grazing systems in this experiment were simulated through mechanical defoliation at fixed stubble heights and intervals, rather than by livestock grazing. Consequently, no manure or urine was deposited during grazing events; instead, dried cattle manure pellets were applied uniformly to simulate nutrient return. This method likely reduced the natural spatial heterogeneity in nutrient distribution and sward structure typically observed under actual grazing conditions. Moreover, mechanical cutting led to uniform sward heights within treatments, eliminating the patchiness in vegetation height that often arises from selective grazing and trampling and selective defoliation of i.e. clover. Additionally, we could not take into account potential soil compaction and pugging as a result of grazing. Therefore, the presented results give an indication of the likely response under grazing, but may be more prominent as responses are assumed to be more homogenous under simulated compared to real grazing (Guo et al., 2024).

4.1. Forage production and quality

Herbage dry matter yield is a key ecosystem service in relation to food production. Equally important is forage quality—particularly nitrogen concentration and digestibility—as these factors determine how efficiently grass can be converted into meat and milk. The ESS forage production and quality was strongly affected by sward type and simulated grazing system and their interaction as discussed below.

Sward type had a strong influence on herbage DMY. As hypothesized, DMY was significantly lower in the Lp0 sward compared to the other treatments, due to the absence of fertilization—an effect consistent across all years and simulated grazing systems. The Fa250 sward produced higher DMY than Lp250 across simulated grazing systems, although the difference was statistically significant only in 2015. This may be explained by the superior drought resistance of *Festuca arundinacea* (Fa), attributed to its deep root system (Cougnon et al., 2017; Wilman et al., 1998; Surault et al., 2007), as the summer of 2015 experienced a severe rainfall deficit (up to −180 mm in July and August), whereas deficits in 2014 and 2016 remained below −40 mm (Fig. S1). On average across years and systems, LFT50 yielded similarly to Lp250, suggesting that the inclusion of white clover and tall fescue in LFT50 effectively compensated for the 200 kg N ha⁻¹ yr⁻¹ reduction in fertilizer. This is further supported by soil nitrogen indicators—total N and potentially mineralisable N—which were highest in Lp250 and Fa250, followed closely by LFT50.

However, the herbage DMY of LFT50 declined markedly from 2014 (averaging 10,962 kg DM ha⁻¹) to 2016 (6150 kg DM ha⁻¹), coinciding with a decrease in clover proportion from 0.42 to 0.28 across all simulated grazing systems. In 2016, LFT50 also exhibited the lowest soil pH, phosphorus (P), and potassium (K) levels (Table 2, Fig. S7). The underlying cause of this decline remains unclear. It is uncertain whether it

Table 5

The effect of Sward type (ST) and simulated grazing system (GS) and their interaction on standardised ecosystem services (ESSs).

ESS ⁴	Sward type ¹				Simulated grazing system ²			Sward type x Simulated grazing system												P-value ³		
								Lp0			Lp250			Fa250			LFT50			ST	GS	STxGS
	Lp0	Lp250	Fa250	LFT50	CG	LG	RG	CG	LG	RG	CG	LG	RG	CG	LG	RG	CG	LG	RG			
FP	0.18	0.60	0.58	0.64	0.59	0.41	0.51	0.26	0.05	0.21	0.64	0.57	0.59	0.61	0.55	0.60	0.84	0.45	0.63	***	***	***
DMY	0.16	0.59	0.66	0.59	0.49	0.37	0.64	0.16	0.08	0.25	0.55	0.47	0.73	0.57	0.52	0.90	0.70	0.39	0.67	***	***	***
Quality	0.26	0.61	0.47	0.67	0.68	0.47	0.35	0.47	0.08	0.22	0.73	0.68	0.41	0.62	0.57	0.23	0.90	0.55	0.54	***	***	***
CR	0.23	0.65	0.66	0.46	0.39	0.59	0.52	0.15	0.22	0.32	0.50	0.77	0.66	0.61	0.73	0.63	0.29	0.66	0.44	***	.	ns
WR	0.38	0.54	0.58	0.50	0.50	0.55	0.45	0.40	0.38	0.37	0.61	0.57	0.45	0.55	0.66	0.51	0.42	0.60	0.47	***	ns	ns
HP	0.43	0.43	0.41	0.73	0.54	0.48	0.48	0.47	0.39	0.43	0.49	0.42	0.40	0.42	0.44	0.36	0.80	0.67	0.72	***	***	**
Average	0.30	0.56	0.56	0.58	0.50	0.51	0.49	0.32	0.26	0.33	0.56	0.58	0.53	0.55	0.60	0.53	0.59	0.60	0.56	***	ns	ns

¹ Lp0 = *Lolium perenne* without fertilisation, Lp250 = *Lolium perenne* with fertilisation (Manure + CAN), Fa250 = *Festuca arundinacea* with fertilisation (Manure + CAN), LFT = mixture of Lp, Fa and *Trifolium repens* (manure only).

² CG = continuous grazing, LG = lenient or regenerative grazing, RG = rotational grazing, DMY = dry matter yield

³ Statistical significance is indicated with *** P < 0.001; ** P < 0.01; * P < 0.05; P < 0.1.

⁴ Standardised ecosystem services (ESSs) are based on the standardised averages of the following parameters: **Forage Production (FP)**: DMY, N concentration, OMD, N Yield and DOM; **Climate regulation (CR)**: SOM and SOC; **Water regulation (WR)**: Water infiltration 2014 and 2015, Water capacity; **Habitat provision (HP)**: Meadowbird habitat, Earthworm parameters, proportion clover (average 2014–2016); Meadowbird habitat: the maximum sward height in spring (2016) and the number of defoliation events before June 15. As for both parameters, a higher value corresponds to a negative impact on the ecosystem service, the response was calculated as 1 – EF. Earthworm parameters: Earthworm abundance, Earthworm biomass, Earthworm individual weight, Epigeic earthworm abundance, number of species.

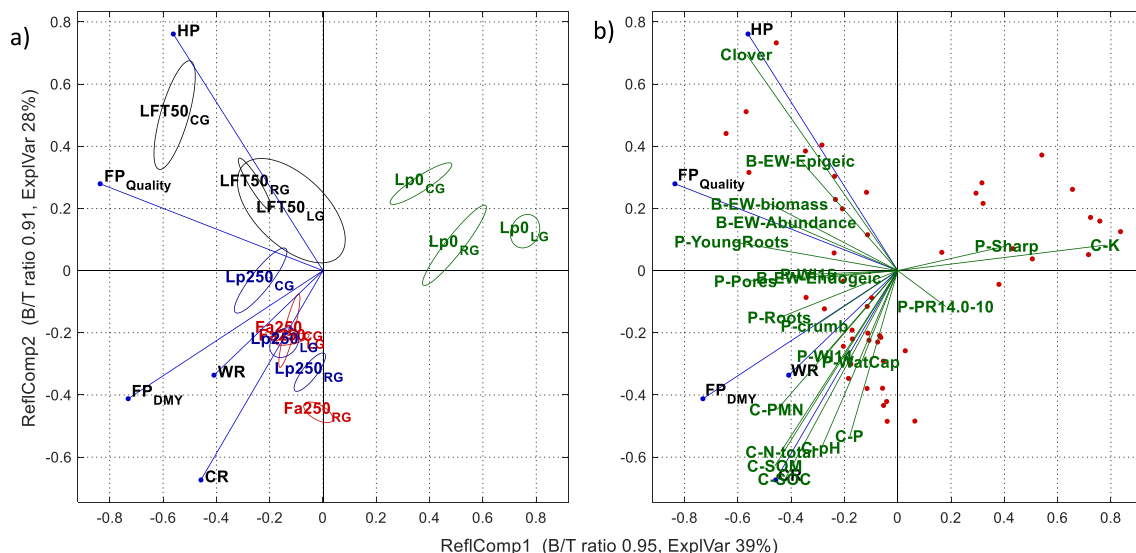


Fig. 3. Differentiation of ESSs of the 12 Sward types x simulated grazing system combinations in a reflected discriminant analysis (RefDA) visualized in bi-plots with a) group means of the reflected components with corresponding standard deviation ellipses and correlations with ESSs and b) reflected component scores (48 red dots) and their correlations with soil chemical (C), physical (P) and biological (B) parameters, and clover proportion (green lines). Bi-plots with the third reflected component are presented in Fig. S12. Abbreviations: Sward types: Lp0 = *Lolium perenne* without fertilisation, Lp250 = *Lolium perenne* with fertilisation (Manure + CAN), Fa250 = *Festuca arundinacea* with fertilisation (Manure + CAN), LFT = mixture of Lp, Fa and *Trifolium repens* (Manure only). Simulated grazing systems: CG = continuous grazing, LG = lenient or regenerative grazing, RG = rotational grazing. Standardised ecosystem services (ESSs) are based on the standardised averages of the following parameters: Forage Production (FP): DMY, N concentration, OMD, N Yield and DOM; Climate regulation (CR): SOM and SOC; Water regulation (WR): Water infiltration 2014 and 2015, Water capacity; Habitat provision (HP): Meadowbird habitat, Earthworm parameters, proportion clover (average 2014–2016); PR = penetration resistance, EW = earthworm, WI = water infiltration, PMN = potentially mineralizable N.

was driven by specific environmental conditions during the monitoring years or indicative of a longer-term downward trend that might have continued had the experiment extended beyond 2016.

The effect of simulated grazing system on DMY varied among the different sward types. For Lp250 and Fa250, DMY was higher under RG compared to CG and LG, due to the longer regrowth period for RG (6–7 weeks) relative to CG and LG (3–4 weeks), consistent with Wilman et al. (1977), who reported a large positive effect on yield when extending the interval from 3 to 6 weeks. In contrast, for LFT50, DMY under CG and RG were similar and significantly higher than under LG. In this case, the

higher yield potential of Lp and Fa under RG was compensated by better clover performance under CG. CG resulted in the highest clover content, while it was lowest under LG. The lower clover content under LG was most likely due to shading by grasses (as a result of greater stubble and/or canopy height), which is known to reduce the number of leaf-bearing nodes and growing points in white clover (Lüscher, 1989; Seresinhe, 1992). Schwank et al. (1986) also identified light competition as the main factor limiting clover proportion in mixed swards. Our findings align with Seresinhe (1992), who reported clover proportions of 30% and 41% under cutting heights of 10 cm and 4 cm, respectively, in

a two-year ryegrass–clover field experiment under similar climatic conditions.

In line with our hypothesis, there was a strong main and interactive effect of sward type and simulated grazing system on herbage N concentration. The absence of fertilizer application in Lp0 resulted in the lowest N concentrations across all simulated grazing systems and years. Across sward types, herbage N concentrations were consistently lowest under RG, which is attributed to the longer grazing interval (Nowakowski, 1962; Hoekstra et al., 2007). During early regrowth, N uptake is driven by N availability, but this becomes diluted as DMY increases with extended regrowth duration (Nowakowski, 1962). For LFT50, N concentration was highest under CG, while LG and RG showed similar levels. This pattern corresponds with the higher clover content observed under CG, as discussed previously. For Lp250 and Fa250, there was no significant difference in N concentration between CG and LG, suggesting that at high fertilization rates, variations in cutting height did not impact herbage quality in terms of N concentration—consistent with the findings of Hoekstra et al. (2007).

OM digestibility was strongly influenced by sward type, following the pattern Lp250 > LFT50 > Fa250 > Lp0. The low OM digestibility of Lp0 aligns with findings by Valk et al. (1996), who attributed reduced digestibility to a slower degradation rate of cell walls under low N fertilization. Similarly, the lower digestibility of Fa250 compared to Lp is supported by Cougnon et al. (2014). Interestingly, OM digestibility of LFT50 was comparable to that of Lp250, suggesting that N fixation and the highly digestible white clover component (De Haas et al., 2019) compensated for the reduced fertilization rate and the lower digestibility of Fa.

In general, OM digestibility was highest under CG, likely due to shorter rotation lengths, which resulted in the harvesting of less mature forage (Wilman et al., 1977; Wilson, 1994; Groot, 1999). Additionally, the lower stubble height in CG may reduce the accumulation of old and dead plant material in the sward (Mayne et al., 1987; Hernández Garay et al., 2000). In contrast, digestibility was lowest under RG, where longer regrowth intervals lead to more mature, and thus less digestible, forage.

4.2. Climate regulation

The ESS climate regulation in grassland systems is primarily determined by the soil's capacity to sequester carbon through the accumulation of soil organic matter (SOM). However, other metrics, including N₂O emissions from soil (mainly associated with fertiliser application rates) and methane emissions from grazing cows (related to stocking density and feed quality) may be equally important (Soussana et al., 2004; Allard et al., 2007; Rivera Herrera and Chará, 2021), but are not considered as part of the current study).

As hypothesized, the ESS climate regulation was lower in the unfertilised Lp0 sward compared to the fertilised sward types. This aligns with findings synthesized by Conant et al. (2017), which showed positive soil carbon sequestration rates for both organic and inorganic fertilisers, with substantially higher rates for the former. These results suggest both direct and indirect effects of fertilisation on SOM accumulation.

All fertilised sward types (except Lp0) received organic manure inputs equivalent to 2.2 Mg OM ha⁻¹ yr⁻¹, directly contributing to SOM. In addition, Lp0 exhibited lower root count and root biomass (Hoogsteen et al., 2020; Table S8) than Lp250 and Fa250, which also limits organic matter inputs. This supports earlier research showing positive effects of fertilisation on root biomass (Cougnon et al., 2017; Tomaškin et al., 2013). Aboveground biomass and associated litter inputs were also likely lower in Lp0. However, belowground carbon inputs generally contribute more to long-term SOM due to their chemical stability (Bai and Cotrufo, 2022). Additionally, it should be noted that increased fertilization has a range of side-effects both in terms of its production and increased GHG emissions associated with nitrous oxide emissions

(Conant et al., 2017).

Differences in SOM between the fertilised swards were smaller. Despite a higher root score in the 0–10 cm layer for Fa250, this did not result in higher SOM in the topsoil. While this outcome does not support our hypothesis, it is consistent with Hoogsteen et al. (2020), who reported no increase in SOM in Fa swards compared to Lp in the topsoil, despite higher root biomass, but did observe higher SOM accumulation in Fa swards at 30–60 cm depth. This is consistent with the greater rooting depth and associated drought resistance of Fa (Poirier et al., 2012). It is possible that at shallow depth, differences in root biomass were insufficient to impact SOM significantly, and that manure inputs and surface litter played a more dominant role.

The performance of LFT50 for ESS climate regulation was intermediate between Lp0 and the fertilised swards, suggesting that the inclusion of white clover can partially compensate for reduced inorganic fertilisation and thus stimulating plant growth and root development—supporting our hypothesis.

Beyond biomass quantity, the decomposition rate of root material also affects soil carbon dynamics and depends on both root traits and soil conditions. To explore this, we used both an in situ litterbag method and an in vitro carbon decomposition assay to assess degradability of Fa250 and Lp250 root material. As expected, the in situ litterbag experiment showed a faster root dry matter decomposition rate for Lp compared to Fa, but only during the first 14 days. In contrast, the in vitro experiment revealed a significantly faster root carbon decomposition rate for Fa compared to Lp. Since the same root material was used in both assays, the contrasting results likely reflect differences in abiotic (i.e. temporal fluctuations in soil temperature and moisture) or biotic conditions in the field (in situ) rather than root properties alone.

In line with our hypothesis, climate regulation was significantly higher under LG compared to CG. We found no effect of simulated grazing system on root score, consistent with Hoogsteen et al. (2020), who reported no significant simulated grazing effect on root biomass. Whereas generally root growth is reported to be lower at higher defoliation frequency, literature offers mixed results on root growth responses to defoliation frequency (Dawson et al., 2000; Deinum, 1985). Therefore, the increase in SOM under LG in the current study is more likely due to greater aboveground organic matter inputs, resulting from the higher stubble height under lenient grazing. This effect may have been even stronger under real grazing conditions (rather than simulated grazing) due to additional inputs and disturbance from animal trampling. However, the net effect under grazing is hard to assess due to the high spatial variation. Additionally, soil temperature during the growing season was likely lower due to the higher stubble height due to its insulating effect (Cutforth and McConkey, 1997), which may have a positive effect on SOM accumulation.

This study focused on SOM concentrations in the topsoil (0–10 cm). For results on SOM stock changes at deeper soil layers in relation to sward type and simulated grazing system, we refer to Hoogsteen et al. (2020).

4.3. Water regulation

The ESS water regulation was based on the soil's capacity to absorb (water infiltration) and retain water (water holding capacity) and is increasingly important under climate change, which is expected to bring more extreme weather events such as heavy rainfall and drought (KNMI, 2014). There was a strong effect of sward type on water regulation, whereas the effect of simulating grazing system was small and non significant, as discussed below.

As hypothesised, water infiltration rates were lower for Lp0 compared to the other sward types. Infiltration rate was positively correlated with several soil parameters, including soil crumbliness, root density, pH, and SOM (Table S11). Except for pH, all these parameters were lower in Lp0, which likely contributed to its reduced infiltration capacity. These findings are partially consistent with Deru et al. (2018),

who also observed positive relationships between water infiltration and soil crumbliness, rooting density, pH, and SOM. However, unlike their results, we found no significant correlation between water infiltration and either soil pore number or earthworm abundance (Table S11c), suggesting that micropores rather than macropores were the dominant contributors to water movement in our study, consistent with the sandy soil type.

There were no consistent or significant differences in water infiltration-related parameters between Lp250 and Fa250. This indicates that differences in infiltration are mainly attributable to fertilisation level (as seen in the Lp250 vs. Lp0 contrast) rather than species composition. Interestingly, the LFT50 mixture—fertilised with only 50 kg N ha⁻¹ yr⁻¹ as organic manure—performed similarly to Lp250 and Fa250, both of which received 250 kg N ha⁻¹ yr⁻¹. However, due to the design of this experiment, it is not possible to determine whether this performance is due to the presence of white clover or the diversity effect of a mixed sward.

In general, water infiltration rates were lower in 2015 than in 2014, consistent with the dryer conditions in 2015 (Figure S1, Deru et al., 2018). However, despite the different conditions the treatment effects were consistent in both years.

Available water capacity, an indicator of plant-accessible water retention, was highest in Fa250 and lowest in Lp0 and LFT50. This parameter was positively correlated with SOM content, in line with findings by Lal (2020), who showed that SOM increased soil water retention more at field capacity than at the permanent wilting point, thereby improving available water capacity. However, even though the difference in available water capacity was statistically significant, the absolute difference (0.09 – 0.1 g g⁻¹) and hence potential impact on drought resistance is likely to be relatively small.

In contrast with our hypothesis, in the timespan of the experiment and our monitoring (three to five years) there was no significant effect of simulated grazing system on any of the parameters associated with the ESS water regulation. This is in line with Teague et al. (2011), who found no difference between rotational grazing and continuous grazing on water infiltration rate and bulk density. This lack of effect is consistent with the relatively small differences in SOM and root systems observed between grazing treatments. It is important to note that the study used simulated grazing systems, and therefore excluded physical effects such as treading and trampling by livestock. As such, real-world differences in grazing duration or livestock density—factors known to affect soil structure and water regulation—were not captured in this experiment.

4.4. Habitat provision

Within this study we have focussed on habitat provision in relation to meadow birds. It is important to note that this study did not directly assess meadow bird foraging behavior or chick survival, nor did it account for other key ecological parameters influencing meadow bird habitat quality, such as the presence of large grazers (Whittingham and Devereux, 2008). Moreover, there are additional parameters that were not assessed as part of this study, that may contribute to the habitat quality and biodiversity value of grazing systems, including the diversity and abundance of insects and other soil biological groups. In general the ESS habitat provision was highest for sward type LFT50 and for the simulated grazing system CG, as discussed below.

Earthworms are a key component of grassland ecosystems, both as indicators of belowground biodiversity and as a crucial food resource for meadow birds (Onrust et al., 2019). Earthworm abundance and biomass were highest in the LFT50 sward, consistent with previous findings showing a positive influence of white clover on earthworm populations (De Haas et al., 2019; Van Eekeren et al., 2009a). This increase was primarily driven by higher numbers of epigeic earthworms, which dwell in surface litter and are readily accessible to foraging birds (Onrust and Piersma, 2019). In contrast, Lp0—lacking any fertilisation—had the

lowest earthworm biomass and abundance, reflecting the positive effect of (organic) fertiliser application on earthworm populations (Edwards and Lofty, 1982).

The results showed no effect of simulated grazing system on earthworm abundance, which is surprising given the higher input of plant residues from the higher stubble height associated with LG, which could have stimulated the abundance of epigeic earthworms.

Soil penetration resistance values in the top soil layer were not affected by the treatments and remained below the theoretical threshold of 1.1 MPa (Oosterveld et al., 2017), above which the foraging success of meadow birds is restricted.

In addition to food availability, sward structure in spring plays a critical role in chick foraging success. Taller swards impede chick mobility and reduce prey capture rates (Devereux et al., 2004). Maximum sward height in spring was lowest for Lp0 and CG. However, the frequent defoliation associated with CG could pose risks to nesting success by disturbing nests and reducing cover, especially during the breeding season (Devereux et al., 2004).

Furthermore, clover in grasslands benefits flower-visiting insects by providing an abundant source of nectar and pollen (Harris and Ratnieks, 2022), which is an important component for biodiversity in itself, but insects are also part of the diet of meadowbirds (Jansma et al., 2025). The clover in the LFT50 sward type performed particularly well under continuous grazing management.

Overall, the LFT50 sward type, particularly when combined with continuous grazing, offers a balanced management strategy for habitat provision. This was achieved at a comparatively low rate of fertiliser input.

As stated before, in this study we used simulated grazing as opposed to real grazing, resulting in reduced sward heterogeneity (Guo et al., 2024), which is known to affect meadow bird habitat quality (Whittingham and Devereux, 2008). These limitations should be considered when interpreting the implications of the grazing treatments for habitat provision, particularly with respect to species reliant on structurally diverse habitats.

4.5. Grazing systems and sward type to optimize grassland ESSs

On this sandy soil with an arable history, our results show a clear differentiation of sward type and simulated grazing systems in terms of their performance of the different ESSs. However, both Table 5 and Fig. 3 clearly show there is no single sward type x simulated grazing system combination that performs best for all the measured ESSs. This implies that different management choices should be made depending on the prioritisation of ESSs within the farming system.

Generally speaking, the effect of sward type was more pronounced than the effect of simulated grazing system on ESSs. Lp0 underperformed in all ESSs, which was associated with high soil K concentration (excess availability as a result of low uptake due to N-limited plant growth) and poor physical soil structure (high proportion sharp-edged soil elements and penetration resistance and low rooting scores). This suggests that extreme extensification by cutting out all N fertilisation and/or natural N-fixation, has a negative impact on the ESSs of grasslands on sandy soil. This is in line with Van Eekeren et al., (2009b).

Lp250 and particularly Fa250 were associated with a strong performance in terms of FP_{DMY}, climate regulation and to a lesser extent water regulation, which was related to high soil carbon concentrations, and high N availability in the soil (PMN). LFT50 performed best in terms of habitat performance and forage quality, which was related to the clover proportion and earthworm parameters.

The different simulated grazing systems showed the strongest effects on forage production (both quality and quantity) and habitat provision, whereas the effects on climate regulation and particularly water regulation were small and non-significant. CG resulted in the highest forage quality and habitat provision, RG in the highest forage quantity and LG

performed relatively strong on climate regulation and water regulation.

5. Conclusions

Both sward type and simulated grazing system had significant effects on the delivery of ESSs in grassland systems. Among the simulated grazing systems, CG resulted in the highest herbage quality, LG was most beneficial for climate regulation, and RG supported the greatest forage production. No consistent effects of simulated grazing system on water regulation were observed.

The effects of sward type on ESS were generally more pronounced than the effect of simulated grazing system. The unfertilised *Lolium perenne* sward (Lp0) consistently underperformed across all investigated ecosystem services, indicating that extreme extensification without nutrient input may compromise grassland multifunctionality. In contrast, the fertilised grass monocultures (Lp250 and Fa250) provided high forage yields and contributed positively to most ecosystem services, though they offered limited habitat quality. Fa250 was particularly productive under drought conditions but had lower herbage quality than Lp250. The low-input species mixture (LFT50) delivered a balanced performance across forage quality, climate regulation, and habitat provision, although water regulation outcomes were less favourable than anticipated. LFT50 performed especially well under continuous grazing, which positively influenced clover presence.

Overall, no single sward type or simulated grazing system or combination thereof optimised all ecosystem services. These findings regarding simulated grazing can inform the design of future comparisons of grazing systems. Moreover, these findings highlight the need to balance management objectives and tailor grassland strategies according to the prioritisation of ecosystem services in specific environmental and agricultural contexts. Practical recommendations could include for example that an organic dairy farm on sandy soil, which is heavily reliant on nitrogen supplied by clover, combines a grass-white clover sward (LTF) with continuous grazing (CG) management. In contrast, a conventional farm prone to drought risk may benefit from combining a tall fescue sward (Fa250) with a lenient grazing strategy (LG).

CRedit authorship contribution statement

N. van Eekeren: Writing – review & editing, Methodology, Investigation, Funding acquisition, Conceptualization. **E.A. Lantinga:** Funding acquisition, Conceptualization. **J. de Wit:** Writing – review & editing. **M.J.J. Hoogsteen:** Writing – review & editing, Investigation, Data curation. **Hoekstra Nyncke Jitske:** Writing – review & editing, Writing – original draft, Investigation, Formal analysis.

Declaration of generative AI and AI-assisted technologies in the writing process

During the preparation of this work the authors used ChatGPT in order to improve language and readability of the manuscript. After using this tool/service, the authors reviewed and edited the content as needed and take full responsibility for the content of the publication.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgements

This research was funded by the public private cooperation program “Forage, soil and circular agriculture” via the Top Sector Agri & Food TKI-AF-15102/15284 LWV190195; by Barenbrug Holland, and by the Dutch Dairy Board (In Dutch: Productschap Zuivel) and Culterra. We

wish to express our gratitude to Frans Bakker (WUR, Unifarm), Dorien Bouwmeester, Chris Bisperink and Evert-Jan Dijk for their assistance during the field work and Herman de Boer for facilitating the CO₂ respiration measurements. We would like to thank Dré Nierop for carrying out the reflected discriminant analysis. The authors report there are no competing interests to declare

Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at [doi:10.1016/j.agee.2026.110548](https://doi.org/10.1016/j.agee.2026.110548).

Data availability

Data will be made available on request.

References

- Allard, V., Soussana, J.F., Falcimagne, R., Berbigier, P., Bonnefond, J.M., Ceschia, E., D'hour, P., Hénault, C., Laville, P., Martin, C., Pinarès-Patino, C., 2007. The role of grazing management for the net biome productivity and greenhouse gas budget (CO₂, N₂O and CH₄) of semi-natural grassland. *Agric. Ecosyst. Environ.* 121, 47–58.
- Bai, Y., Crotufo, M.F., 2022. Grassland soil carbon sequestration: current understanding, challenges, and solutions. *Science* 377, 603–608.
- Bates, D., Mächler, M., Bolker, B., Walker, S., 2015. Fitting linear mixed-effects models using lme4. *J. Stat. Softw.* 67, 1–48. <https://doi.org/10.18637/jss.v067.i01>.
- Conant, R.T., Cerri, C.E.P., Osborne, B.B., Paustian, K., 2017. Grassland management impacts on soil carbon stocks: a new synthesis. *Ecol. Appl.* 27, 662–668.
- Cougnon, M., Baert, J., Reheul, D., 2014. Dry matter yield and digestibility of five cool season forage grass species under contrasting N fertilizations. *Grassl. Sci. Eur.* 19, 175–177.
- Cougnon, M., De Swaef, T., Lootens, P., Baert, J., De Frenne, P., Shahidi, R., Reheul, D., 2017. situ quantification of forage grass root biomass, distribution and diameter classes under two N fertilisation rates. *Plant Soil* 411, 409–422.
- Cutforth, H.W., McConkey, B.G., 1997. Stubble height effects on microclimate, yield and water use efficiency of spring wheat grown in a semiarid climate on the Canadian prairies. *Can. J. Plant Sci.* 77, 359–366.
- Dawson, L.A., Grayston, S.J., Paterson, E., 2000. Effects of grazing on the roots and rhizosphere of grasses. In: Lemaire, G., Hodgson, J., de Moraes, A., Nabinger, C., Carvalho, P.Cd (Eds.), *Grassland Ecophysiology and Grazing Ecology*. CABI, Wallingford, UK, pp. 61–84.
- Day, R.W., Quinn, G.P., 1989. Comparisons of treatments after an analysis of variance in ecology. *Ecol. Monogr.* 59, 433–463. <https://doi.org/10.2307/1943075>.
- De Boer, H., Deru, J., Van Eekeren, N., 2018. Sward lifting in compacted grassland: effects on soil structure, grass rooting and productivity. *Soil Tillage Res* 184, 317–325. <https://doi.org/10.1016/j.still.2018.07.013>.
- De Haas, B., Hoekstra, N., Schoot, J.R., Visser, E.J.W., Kroon, H., Van Eekeren, N., 2019. Combining agro-ecological functions in grass-clover mixtures. *AIMS Agric. Food* 4, 547–567.
- Deinum, B., 1985. Root mass of grass swards in different grazing systems. *Neth. J. Agric. Sci.* 33, 377–384.
- Deru, J.G.C., Bloem, J., De Goede, R., Keidel, H., Kloen, H., Rutgers, M., Van den Akker, J., Brussaard, L., Van Eekeren, N., 2018. Soil ecology and ecosystem services of dairy and semi-natural grasslands on peat. *Appl. Soil Ecol.* 125, 26–34. <https://doi.org/10.1016/j.apsoil.2017.12.011>.
- Devereux, C.L., McKeever, C.U., Benton, T.G., Whittingham, M.J., 2004. The effect of sward height and drainage on common starlings (*Sturnus vulgaris*) and northern lapwings (*Vanellus vanellus*) foraging in grassland habitats. *Ibis* 146, 115–122.
- Edwards, C.A., Loft, J.R., 1982. Nitrogenous fertilizers and earthworm populations in agricultural soils. *Soil Biol. Biochem.* 14, 515–521.
- Ennik, G.C., Gillet, M., Sibma, L., 1980. Effect of high nitrogen supply on sward deterioration and root mass. In: Prins, W.H., Arnold, G.H. (Eds.), *The Role of Nitrogen in Intensive Grassland Production*. Institute for Soil Fertility, Haren, The Netherlands, pp. 67–76.
- FAO, n.d. Grasslands, rangelands and forage crops. Available online.
- FAO, 2023. The State of Food and Agriculture 2023 – Revealing the true cost of food to transform agrifood systems. FAO, Rome. <https://doi.org/10.4060/cc7724en>.
- Fariaszewska, A., Ager, J., Van Huylbroeck, J., De Swaef, T., Baert, J., Pecio, A., 2020. Physiological and biochemical responses of forage grass varieties to mild drought stress under field conditions. *Int. J. Plant Prod.* 14, 335–353. <https://doi.org/10.1007/s42106-020-00088-3>.
- Finn, J.A., Kirwan, L., Connolly, J., Sebastià, M.T., Helgadóttir, A., Baadshaug, O.H., Bélanger, G., Black, A., Brophy, C., Collins, R.P., 2013. Ecosystem function enhanced by combining four functional types of plant species in intensively managed grassland mixtures. *J. Appl. Ecol.* 50, 365–375.
- Groot, J.C.J., 1999. Modelling grass digestibility based on morphological and physiological plant characteristics. PhD thesis. Wageningen University, Wageningen.
- Grüebler, M.U., Schuler, H., Horch, P., Spaar, R., 2012. The effectiveness of conservation measures to enhance nest survival in a meadow bird suffering from anthropogenic nest loss. *Biol. Conserv.* 146, 197–203.

- Guo, T., Guo, M., Pang, Y., Sun, X., Ryo, M., Liu, N., Zhang, Y., 2024. Ungulate herbivores promote beta diversity and drive stochastic plant community assembly by selective defoliation and trampling. *J. Ecol.* 112, 1992–2006.
- Harris, C., Ratnieks, F.L., 2022. Clover in agriculture: combined benefits for bees, environment, and farmer. *J. Insect Conserv.* 26, 339–357.
- Hernández Garay, A.H., Matthew, C., Hodgson, J., 2000. The influence of defoliation height on dry-matter partitioning and CO₂ exchange of perennial ryegrass miniature swards. *Grass Forage Sci.* 55, 372–376.
- Hoekstra, N., Holshof, G., Schils, R., Philipsen, B., Van Reenen, K., Van Houwelingen, K., Van Eekeren, N., 2019. The effect of Kurzrasen and strip-grazing on grassland performance and soil quality of a peat meadow. *Sustainability* 11, 6283. <https://doi.org/10.3390/su11226283>.
- Hoekstra, N.J., Struik, P.C., Lantinga, E.A., Schulte, R.P.O., 2007. Chemical composition of lamina and sheath of *Lolium perenne* as affected by herbage management. *NJAS Wagening. J. Life Sci.* 55, 55–73.
- Hoogsteen, M.J.J., Bakker, E.-J., Van Eekeren, N., Tittonell, P.A., Groot, J.C.J., Van Ittersum, M.K., Lantinga, E.A., 2020. Do grazing systems and species composition affect root biomass and soil organic matter dynamics in temperate grassland swards? *Sustainability* 12, 1260. <https://doi.org/10.3390/su12031260>.
- Hothorn, T., Bretz, F., Westfall, P., Heiberger, R.M., 2012. Multcomp: simultaneous inference for general linear hypotheses. R. Package Version 1–2.
- Iepema, G., Hoekstra, N.J., De Goede, R., Bloem, J., Brussaard, L., Van Eekeren, N., 2022. Extending grassland age for climate change mitigation and adaptation on clay soils. *Eur. J. Soil Sci.* 73, e13134.
- Isselstein, J., Kayser, M., 2014. Functions of grassland and their potential in delivering ecosystem services. *Grassl. Sci. Eur.* 19, 199–214.
- Isselstein, J., Jeangros, B., Pavlu, V., 2005. Agronomic aspects of biodiversity targeted management of temperate grasslands in Europe: a review. *Agron. Res.* 3, 139–151.
- Jackson, R.B., Lajtha, K., Crow, S.E., Hugelius, G., Kramer, M.G., Piñeiro, G., 2017. The ecology of soil carbon: pools, vulnerabilities, and biotic and abiotic controls. *Annu. Rev. Ecol. Syst.* 48, 419–464.
- Jansma, A.P., Hoekstra, N.J., Van den Broek, B., De Kroon, H., Van Eekeren, N., 2025. Linking key performance indicators to insect abundance in agricultural grasslands: effects of plant diversity, farming intensity, soil type and landscape characteristics. *NJAS Impact Agric. Life Sci.* 97, 2529234.
- KNMI, 2014. Climate change scenarios for the 21st century – A Netherlands perspective. Scientific report WR 2014-01. KNMI, De Bilt.
- Kuznetsova, A., Brockhoff, P.B., Christensen, R.H.B., 2017. lmerTest package: tests in linear mixed effects models. *J. Stat. Softw.* 82, 26.
- Lal, R., 2020. Soil organic matter and water retention. *Agron. J.* 112, 3265–3277. <https://doi.org/10.1002/agj2.20282>.
- Lange, M., Eisenhauer, N., Sierra, C.A., Bessler, H., Engels, C., Griffiths, R.I., Mellado-Vázquez, P.G., Malik, A.A., Roy, J., Scheu, S., Steinbeiss, S., Thomson, B.C., Trumbore, S.E., Gleixner, G., 2015. Plant diversity increases soil microbial activity and soil carbon storage. *Nat. Commun.* 6. <https://doi.org/10.1038/ncomms7707>.
- Lenth, R., 2019. emmeans: estimated marginal means, aka least-squares means. R package version 1.3.5.1.
- Lüscher, A., 1989. Ueberwinterung und Frühlingsaufwuchs von Weissklee (*Trifolium repens* L.): Dynamik der Kohlenhydratenreserven und der biologischen N-Fixierung. PhD thesis. Swiss Federal Institute of Technology, Zurich.
- Lüscher, A., Mueller-Harvey, I., Soussana, J.F., Rees, R.M., Peyraud, J.L., 2014. Potential of legume-based grassland-livestock systems in Europe: a review. *Grass Forage Sci.* 69, 206–228.
- Maestre, F.T., Le Bagousse-Pinguet, Y., Delgado-Baquerizo, M., Eldridge, D.J., Saiz, H., Berdugo, M., Gross, N., 2022. Grazing and ecosystem service delivery in global drylands. *Science* 378 (6622), 915–920.
- Mayne, C.S., Newberry, R.D., Woodcock, C.F., Wilkins, R.J., 1987. Effect of grazing severity on grass utilization and milk production of rotationally grazed dairy cows. *Grass Forage Sci.* 42, 59–72.
- MEA, 2005. Ecosystems and Human Well-being: Current State and Trends, 1. Island Press.
- Moebius-Clune, B.N., Moebius-Clune, D., Gugino, B., Idowu, O., Schindelbeck, R., Ristow, A., Abawi, G., 2016. Comprehensive Assessment of Soil Health – The Cornell Framework Manual.
- Morris, C.D., 2021. How biodiversity-friendly is regenerative grazing? *Front. Ecol. Evol.* 9, 816374.
- Nierop, A.F.M., Tas, A.C., Van der Greef, J., 1994. Reflected discriminant analysis. *Chemom. Intell. Lab. Syst.* 25, 249–263.
- Nowakowski, T.Z., 1962. Effects of nitrogen fertilizers on total nitrogen, soluble nitrogen and soluble carbohydrate contents of grass. *J. Agric. Sci.* 59, 387–392.
- Onrust, J., Piersma, T., 2019. How dairy farmers manage the interactions between organic fertilizers and earthworm ecotypes and their predators. *Agric. Ecosyst. Environ.* 273, 80–85.
- Onrust, J., Wymenga, E., Piersma, T., Olff, H., 2019. Earthworm activity and availability for meadow birds is restricted in intensively managed grasslands. *J. Appl. Ecol.* 56, 1333–1342.
- Oosterveld, E.B., Hoving, I., Sikkema, M., Van der Kamp, J., Mettrop, I., Hendriks, R., 2017. Effecten van tijdelijk hoog waterpeil op weidevogels, bodem en grasopbrengst. A&W-rapport 2151. Altenburg & Wymenga, Feanwâlden.
- Parsons, A.J., Chapman, D.F., 2000. The principles of pasture growth and utilization. In: Hopkins, A. (Ed.), *Grass: Its Production and Utilization*. Blackwell, pp. 31–89.
- Peerlkamp, P., 1959. A visual method of soil structure evaluation. *Meded. Landbouwhogeschool. Gent* 24, 216–221.
- Poirier, M., Durand, J.-L., Volaire, F., 2012. Persistence and production of perennial grasses under water deficits and extreme temperatures: importance of intraspecific vs. interspecific variability. *Glob. Change Biol.* 18, 3632–3646. <https://doi.org/10.1111/j.1365-2486.2012.02800.x>.
- R Core Team, 2020. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna.
- Ren, H., Eviner, V.T., Gui, W., Wilson, G.W., Cobb, A.B., Yang, G., Bai, Y., 2018. Livestock grazing regulates ecosystem multifunctionality in semi-arid grassland. *Funct. Ecol.* 32 (12), 2790–2800.
- Rigal, S., Dakos, V., Alonso, H., Auniş, A., Benkő, Z., Brotons, L., Chodkiewicz, T., Chylarecki, P., De Carli, E., Del Moral, J.C., Domşa, C., Escandell, V., Fontaine, B., Foppen, R., Gregory, R., Harris, S., Herrando, S., Husby, M., Ieronymidou, C., Devictor, V., 2023. Farmland practices are driving bird population decline across Europe. *Proc. Natl. Acad. Sci. U. S. A.* 120.
- Rivera Herrera, J., Chará, J., 2021. CH₄ and N₂O emissions from cattle excreta: a review of main drivers and mitigation strategies in grazing systems. *Front. Sustain. Food Syst.* 5. <https://doi.org/10.3389/fsufs.2021.657936>.
- Schils, R.L.M., Sikkema, K., 2002. The effect of cutting height and management system on the performance of a grass/clover sward. Lowland Grasslands of Europe: Utilization and Development. FAO/CIHEAM, Rome.
- Schwank, O., Blum, H., Nösberger, J., 1986. The influence of irradiance distribution on the growth of white clover (*Trifolium repens* L.) in differently managed canopies of permanent grassland. *Ann. Bot.* 57, 273–281.
- Seresinhe, R.T., 1992. Regrowth, productivity and symbiotic nitrogen fixation of white clover (*Trifolium repens* L.) monocultures and mixed swards. PhD thesis. Swiss Federal Institute of Technology, Zurich.
- Shepherd, T., 2000. Visual Soil Assessment, 1. Horizons Regional Council & Landcare Research, Palmerston North.
- Soussana, J.F., Loiseau, P., Vuichard, N., Ceschia, E., Balesdent, J., Chevallier, T., Arrauy, D., 2004. Carbon cycling and sequestration opportunities in temperate grasslands. *Soil Use Manag.* 20, 219–230.
- Surault, F., Veron, R., Chataigner, F., Huyghe, C., 2007. Comportement de prairies mono ou plurispécifiques en année à déficit hydrique marqué. *Fourrages* 192, 507–510.
- Teague, W.R., Dowhower, S.L., Baker, S.A., Haile, N., DeLaune, P.B., Conover, D.M., 2011. Grazing management impacts on vegetation, soil biota and soil chemical, physical and hydrological properties in tallgrass prairie. *Agric. Ecosyst. Environ.* 141, 310–322.
- Tomaškin, J., Jančovič, J., Vozár, L., Tomaškinová, J., 2013. The effect of mineral fertilization on belowground plant biomass of grassland ecosystems. *Acta Univ. Agric. Silv. Mendel. Brun.* 61, 1431–1440. <https://doi.org/10.11118/actaun201361051431>.
- Tonn, B., Komáinová, M., Isselstein, J., 2021. Results from a biodiversity experiment fail to represent economic performance of semi-natural grasslands. *Nat. Commun.* <https://doi.org/10.1038/s41467-021-22309-7>.
- Tsiafouli, M.A., Thébaud, E., Sgardelis, S.P., De Ruiter, P.C., Van der Putten, W.H., Birkhofer, K., Hemerik, L., De Vries, F.T., Bardgett, R.D., Brady, M.V., Björnlund, L., Jørgensen, H.B., Christensen, S., Hertefeldt, T.D., Hotes, S., Hol, W.H.G., Frouz, J., Liiri, M., Mortimer, S.R., Hedlund, K., 2015. Intensive agriculture reduces soil biodiversity across Europe. *Glob. Change Biol.* 21, 973–985. <https://doi.org/10.1111/gcb.12752>.
- Tuo, H., Ghanizadeh, H., Ji, X., Yang, M., Wang, Z., Huang, J., Li, W., 2024. Moderate grazing enhances ecosystem multifunctionality through leaf traits and taxonomic diversity in long-term fenced grasslands. *Science of The Total Environment* 957, 177781.
- Valk, H., Kappers, I.E., Tamminga, S., 1996. sacco degradation characteristics of organic matter, neutral detergent fibre and crude protein of fresh grass fertilized with different amounts of nitrogen. *Anim. Feed Sci. Technol.* 63, 63–87.
- Van Eekeren, N., De Boer, H., Bloem, J., Schouten, T., Rutgers, M., De Goede, R., Brussaard, L., 2009b. Soil biological quality of grassland fertilized with adjusted cattle manure slurries in comparison with organic and inorganic fertilizers. *Biol. Fertil. Soils* 45, 595–608.
- Van Eekeren, N., Van Lie, D., De Vries, F., Rutgers, M., De Goede, R., Brussaard, L., 2009a. A mixture of grass and clover combines the positive effects of both plant species on selected soil biota. *Appl. Soil Ecol.* 42, 254–263.
- Visscher, J., Radersma, S., Van den Pol-Van Dassel, A., 2011. Innovations in grazing systems. Wageningen UR Livestock Research, Lelystad.
- Whittingham, M.J., Devereux, C.L., 2008. Changing grass height alters foraging site selection by wintering farmland birds. *Basic Appl. Ecol.* 9, 779–788.
- Wilman, D., Daly, M., Koocheki, A., Lwoga, A.B., 1977. The effects of interval between harvests and nitrogen application on cell wall components and lignified tissue in perennial ryegrass. *J. Agric. Sci.* 89, 53–63.
- Wilman, D., Gao, Y., Leitch, M., 1998. Some differences between eight grasses within the *Lolium-Festuca* complex under severe water shortage. *Grass Forage Sci.* 53, 57–65.
- Wilson, J.R., 1994. Cell wall characteristics in relation to forage digestion by ruminants. *J. Agric. Sci.* 122, 173–182.
- Xu, F., Li, J., Su, J., Sasaki, T., Lu, X., Wang, Y., Bai, Y., 2024. Understanding the drivers of ecosystem multifunctionality in the Mongolian steppe: The role of grazing history and resource input. *Agric. Ecosyst. Environ.* 359, 108748.
- Zetterlind, B., Janssen, P., Geerts, R., Visser, T., Stip, A., Zwetsloot, M.J., Hoffland, E., Van Eekeren, N., 2025. Multispecies grasslands sustain on-farm forage productivity and quality while reducing nitrogen fertilizer inputs. *Plant Soil*. <https://doi.org/10.1007/s11104-025-08006-0>.
- Zhao, Y., Liu, Z., Wu, J., 2020. Grassland ecosystem services: a systematic review of research advances and future directions. *Landsc. Ecol.* 35, 793–814. <https://doi.org/10.1007/s10980-020-00980-3>.