Grazing by semi-feral cattle and horses supports plant species richness and uniqueness in grasslands

Christoffer Bonavent[1], Kent Olsen[2], Rasmus Ejrnæs[3], Camilla Fløjgaard[3], Morten D.D. Hansen[2], Signe Normand[3], Jens-Christian Svenning[3], and Hans Henrik Bruun[1]

[1]University of Copenhagen
[2]Natural History Museum Aarhus
[3]Aarhus University

October 31, 2022

Abstract

Question: How do naturalistic grazing in contrast to mowing and free succession affect plant community composition and species richness in a temperate grassland grazed by semi-feral cattle and horses?

Location: Mols Laboratory, DenmarkMethods: We investigated grazing exclosures in the rewilding area of the Mols Laboratory, four years after its establishment. We focused on moist to dry grassland vegetation, i.e. excluding scrub and woodland. Each experimental block consisted of five 5 × 9 m plots, representing four fenced treatments, i.e. summer-only grazing, winter-only grazing, full exclosure with annual autumn mowing and full exclosure with passive succession. The matrix was grazed by large herbivores at close-to-natural densities, i.e. regulated bottom-up by the carrying capacity of the area. Hence, the seasonal grazing treatments were grazed at close-to-natural animal density. Quantitative plant community composition was assessed using the point-intercept method in 25 × 25 cm quadrats, supplemented with biomass calibration models based on additional quadrats, in which above-ground plant biomass was harvested after recording and the material sorted to species and weighed. Uniqueness was assessed as the sum of inverse range sizes for constituent species.

Results: We found an appreciably higher plant species richness in grazing treatments than under annual mowing and full exclosure, but only minor differences between seasonal grazing treatments. Uniqueness was highest in year-round and winter-only grazing and lowest in summer-only grazing. The forb:graminoid ratio tended to be high in the winter-only grazing treatment, whereas annual mowing was associated with dominance of graminoids over forbs. Full exclosure plots had accumulation of litter and the lowest species richness. Initial heterogeneity between plots within blocks and a systematic differences between blocks in moist and dry grasslands seemed to swamp treatment effects at this early point after the establishment of the experiment. Data analysis using the biomass estimates derived from the calibration models yielded only minor differences in the patterns described above, when compared to the results obtained using the raw number of intercepts.

Conclusions: Grazing under near-natural conditions is a goal in itself in ecological restoration, but also proposed as an efficient management tool to promote conservation of grassland plants and communities. We found both plant species richness and the prevalence of rarer species (unicity) to be higher with grazing than mowing/abandonment. Similarly, the tendency for forbs to prevail under grazing may translate into

enhanced floral resources for anthophilous insects. Summer-only grazing at low density of large herbivores was not significantly different from winter-only and year-round grazing, but this treatment was much closer to natural grazing than intensive summer grazing typical of agri-environmental practices.

Keywords: biomass estimation, disturbance regime, point-intercept method, rewilding, uniqueness

Introduction

Grazing by large herbivorous mammals is a key process shaping vegetation structure and habitat conditions for plants and other organisms (Bakker et al. 2016; Malhi et al. 2016; Galetti et al. 2018). In European conservation management, there is a strong tradition of aiming at mimicking traditional practices in agriculture and livestock husbandry, e.g. extensive haymaking and summer grazing (Varga et al. 2016). In reality, however, actual conservation management is often strongly constrained by the opportunities compatible with modern high input-high output farm management and agri-environment support schemes (Newton et al. 2012). Either way, conservation management practice is not always rooted in ecological theory and often fails to deliver the desired outcomes for biodiversity (Maxwell et al. 2020; Kindvall et al. 2022). Attempts to apply first principles to grazing management can be comprised under the term "naturalistic grazing", which may be characterized as landscape-scale conservation management, under which grazing as a natural process is seen as an aim in itself, and where human intervention therefore is reduced to a minimum and where herbivore density is not human-controlled, but left to be resource-regulated (Hodder et al. 2005). Although "naturalistic grazing" is considered open-ended with regard to effects on herbivore populations and vegetation, monitoring the effects is crucial to our understanding of how grazing as a natural process works and interacts with other natural conditions and processes.

In large contiguous landscapes, habitat use by large herbivores often shows substantial variation in diurnal, seasonal and between-year patterns. Animal activity tend to be concentrated in certain areas, while large areas may be much more extensively used, e.g. wet areas may be avoided during winter, but preferred in spring and summer (Górecka-Bruzda et al. 2020). Traditional European livestock husbandry had, and continues to have, the growth and survival of domestic animals as its core purpose. Therefore, summer-only grazing on pastures and winter feeding of stabled animals was traditionally the norm in Denmark, in particular for cattle, while some horses have traditionally been left on pastures year-round (Fritzbøger 2004). The pattern of summer-only grazing is strongly reinforced in modern North-European livestock husbandry, in which standard practice is to turn livestock out at very high density (e.g. 800-1000 kg·ha^{-1}) during a short period of intensive grazing at the peak of the growing season (typically May through September or shorter). One way of investigating the resulting impact on vegetation of the annual timing of herbivore activity is to compare areas, to which animal access is restricted to certain parts of the year (Bullock et al. 2001).

Modern European grasslands are often highly grass dominated. The relatively low prevalence of forbs may, however, be a legacy effect of past megafauna extinctions. It has been hypothesized that megafauna once sustained much higher abundance of forbs in grasslands (Bråthen et al. 2021). The shift in dominant growth form has likely been exacerbated by the more recent demise of large herbivores from European landscapes at large, and natural areas in particular. The shift has probably propagated to higher trophic levels, i.e. mega-diverse consumer taxa, e.g. arthropods and fungi (Brunbjerg et al. 2018). Flower-visiting insects have attracted particular attention, partly because this functional group is particularly threatened by both land-use intensification and abandonment, and partly because some anthophilous insect taxa have shown dramatic declines in species richness and abundance (e.g. Hallmann et al. 2017; Warren et al. 2021). The ratio in vegetation of forbs to graminoids has therefore been particularly highlighted, as most forbs have flowers offering resources to anthophilous insects, while graminoids all have wind-pollinated flowers.

The response of vegetation structure to grazing regime will likely involve changes in quantitative plant community composition, with the activities of large herbivores promoting the abundance of certain species, while limiting others. We therefore applied the point-intercept method to quantitatively recording vegetation structure (Jonasson 1988; Godínez-Alvarez et al. 2009; Bonham 2013). Non-destructivity is a virtue of the method, which was desired in the current setup of long-term monitoring plots, also surveyed for other groups of organisms. However, because of differences in plant architecture, the intercept-based abundance does not translate directly to biomass-based abundance. We therefore made calibration models per species and/or functional groups, based on an additional set of quadrats, first subjected to point-intercept recording, next to total harvest and dry-mass estimation per species.

Plant community species richness, or alpha diversity, is of core interest to evaluations of vegetation under contrasted grazing regime, although results may depend on the actual quadrat size applied. From the perspective of gamma diversity in the region or country, however, community unicity - the regional rarity of constituent species - is of higher relevance. One way to evaluate the contribution of individual communities to regional gamma diversity is the 'Sum of inverse range-sizes' (Guerin & Lowe 2015; Ejrnæs et al. 2018), in which constituent species are given decreasing weight with increasing regional occupancy. Also, from the perspective of biodiversity conservation, community unicity may be more relevant than alpha diversity, e.g. even locally species-poor communities may be of high regional conservation value, if they tend to consist of relatively rare species.

Our overarching aim was to assess differences in grassland vegetation structure, community richness and unicity (the prevalence of less widespread species) as a snapshot after four years of naturalistic year-round grazing, as compared to seasonal grazing regimes, to mechanical mowing and to free succession after grazing abandonment and mowing regimes. Specifically, we aimed at investigating:

1) Does plot-scale plant species richness vary between year-round grazing, seasonal grazing (all at naturalistic herbivore density), mowing and passive succession?

2) Does forb to graminoid ratio vary between year-round grazing, seasonal grazing (all at naturalistic herbivore density), mowing and passive succession?

3) Does plant community unicity vary between year-round grazing, seasonal grazing (all at naturalistic herbivore density), mowing and passive succession?

A subordinate aim was the methodological issue of non-destructive assessment of quantitative plant community composition and the sufficiency of the point-intercept method as compared to biomass estimation.

Materials and methods

Study site

The Mols Laboratory is an ecological research station, owned by the Natural History Museum, Aarhus. It is 120 ha located in the glacially shaped hilly landscape of Mols Bjerge at 56.22° latitude and 10.57° longitude. The area covers wide gradients in soil moisture, nutrient status and vegetation openness. Roughly half of the area is covered by open habitats, the other half by scrub and forests, with all types in a mosaic with gradients both between open and canopy-covered habitats and dry and moist habitats. The most frequent open habitat type, as categorized under the European Habitats Directive, was *Species-rich Nardus grasslands* (6230). Despite all research blocks but one (block 70 was not located in a designated habitat type) being assigned to this type of grassland, quite large variation in the species composition and topography between different parts of the area is evident, foremost between hilly glacial gravelly till and sandy marine foreland shaped by the higher sea-level of the Littorina transgression (Atlantic; 6800 - 3900 BCE). This

contrast is presumably particularly linked to hydrology, with the marine foreland being somewhat impacted by exfiltration of groundwater from the hills.

In 2016, as part of a rewilding project, 13 heads of Galloway cattle and 12 Exmoor ponies, supplemented 6 months later by a stallion, were released. Since then, the herds have lived there under near-natural conditions, which means that population size is determined by the carrying capacity, i.e. no supplementary feeding. As population size approaches carrying capacity, food shortage is likely to kick in during the late winter months, at which time weak individuals will die. In order to minimize possible suffering of the animals and to comply with Danish animal welfare legislation, the herds have been continually evaluated, following a scoring protocol based on body condition and behaviour. Individuals failing to meet a set threshold are removed from the area. This so-called "reactive" population management has led to a dynamic development in population size, with the population of large herbivores growing to a total of 44 cattle and 25 ponies in the summer of 2019, and subsequently decreasing to the current level of 12 cattle and 26 ponies. Assuming a standard body mass for cattle of 550 kg and 350 kg for horse, this is equivalent of a drop in large herbivore density from almost 300 kg/ha to 140 kg/ha.

With the purpose of monitoring the effects of the naturalistic grazing regime, 22 randomly selected blocks for permanent vegetation monitoring were established in the spring of 2017. Each block contained four treatment plots, i.e. summer-only grazing (exclosure November through April), winter-only grazing (exclosure May through October), annual autumn mowing (full exclosure with one annual cut during September-October and the thatch removed) and passive succession (full exclosure), all embedded in the matrix of year-round grazing. Animal density was regulated by forage availability of the entire area, which means close to natural levels. Shrubs (but not trees) were initially removed from the mown plots in order to allow cutting by machinery, but not from the other treatments. Fencing did not prevent access to plots by herbivores such as red deer, roe deer and hare, only horse and cattle.

Vegetation recording

In order to select grassland blocks, all monitoring blocks were initially surveyed in order to establish the dominant vegetation type. Out of the initially established 22 blocks, one was discontinued, four were located in closed-canopy forest, four were almost entirely covered by dense scrub and four had scrub-grassland mosaics with too high scrub cover for the point-intercept method to be practically applicable in all treatment plots, leaving nine blocks with mostly open grassland vegetation (Fig. 1).

Field work was carried out in two periods: September 1-16, 2020 and August 2-20, 2021. In the first period, sampling quadrats in treatment plots were put in the periphery of the established circular monitoring plots (see Supplementary materials, Appendix 1) and subjected to point-intercept vegetation recording, after which the above-ground biomass was cut as close to the soil surface as possible, immediately sorted into fractions by plant species (with standing litter as a separate fraction) and dried at 55 @C until constant weight. The resulting data were used to create calibration models per species or functional groups for the prediction of plant species above-ground biomass from non-destructive point-intercept counts. In the second time period, sampling quadrats were located within the circular monitoring plots and surveyed using the same point-intercept method as in the first round. The resulting records were 1) used as-is, 2) subjected to prediction of species' biomass using the regression models of the first round.

Due to time constraints, only six of the nine blocks were included in first round of field work and were thus included in the construction of calibration models (i.e. block numbers 60, 62 and 70 were not sampled). Similarly, mown plots were not sampled, as they were in the process of being mown while the vegetation surveys were carried out. Including newly mown plots would have compromised the reliability of calibration models. In the second year, the field work took place one month earlier, allowing point-intercept recordings in the annually mown plots.

The total sample size for the two periods of fieldwork were: 6 blocks × 4 treatment plots = 24 quadrats in the first period (in 2020) and 9 blocks × 5 treatment plots = 45 quadrats in the second period (in 2021).

Figure 1: Satellite image of the Mols Laboratory property with locations of the 9 sampled blocks marked.

Point-intercept vegetation recording was done using a quadratic frame with a regular 5 × 5 cm point grid within a sampling area of 25 × 25 cm, i.e. 25 regularly spaced sampling points, at each of which a 0.8 cm thick wooden stick was inserted vertically. The frame was elevated above the herbaceous canopy on 50 cm legs. All leaves and stems of plants intercepted by the stick were recorded with species' identity. Dead plant parts were recorded as litter. If the main part of a plant was alive, all its parts was recorded as live. If the main part of the plant was withered, it was recorded as litter. Contact points with bryophytes were also recorded, but were not used in the regression models.

Plant diversity metrics

Species richness per quadrat (alpha diversity) was assessed as one of the simplest metrics of biodiversity.

An index of community unicity was calculated based on species' occupancy within Denmark, i.e. Atlas Flora Danica, AFD (Hartvig & Vestergaard 2015). These data consist of presence and absence records of all vascular plants in 1300 grid cells, each 5 × 5 km, dispersed across the country. Each species i recorded in a quadrat was given a value equivalent to the inverse of its range size and the resulting values summed per quadrat, thus

$\sum_{i=1}^{S} \frac{1}{n\ AFD\ grid\ cells\ with\ species\ i\ present}$, in which S is the number of species in a given quadrat.

The ratio of forbs to graminoids was included as an indicator of floral resources available to anthophilous insects, thus $\frac{n\ intercepts(\text{forbs})}{n\ intercepts(\text{graminoids})}$, or the equivalent for biomass estimates.

Finally, the amount of leaf litter was used as an indication of grazing naturalness.

Statistical analysis of point intercept data

First, an overall model of intercepts per vascular plant species as a function of above-ground biomass was built as a generalized linear model. Next, the residual variation of this model was investigated using analysis of variance with dry mass per point-intercept as the dependent variable and species identity, block and treatment as factors, using data from 24 quadrats recorded i 2020. This was done to investigate the assumption that species identity would explain variation in this ratio. Subsequently, one linear regression model per plant

5

species was built. This was done for plant species with three or more data points, and additionally for genera with more than one species present and for functional groups meeting the same criterion. The functional groups were: Broad-leaved graminoids (leaf blades $>$ 2 mm), narrow-leaved graminoids (leaf blades $<$ 2 mm), *Juncus effusus*-type rushes (for which intercepts with stems were recorded, as they bear no leaf blades), forbs, woody plants (trees and shrubs, incl *Calluna vulgaris* and *Cytisus scoparius*). Initially, separate models were built for forbs with basal leaves, e.g. rosettes, vs. post-and-flag type forbs, but model estimates were very similar, so the two groups were combined to produced a single model with comparable R^2 and lower standard error. A separate model was built for standing dead litter. No data transformations were used. All regressions models were forced through the origin, because otherwise - when predicted biomass was accrued per plot over species - total biomass per plot would gain a spurious strong positive relationship with plot species richness.

Second, for the 45 quadrats sampled in 2021, calibrated biomass values per species per plot were calculated from point-intercept data and summed over species present in plots (total biomass, plus forb and graminoid biomass separately). For each species present in a plot, the best available model was used, i.e. first choice was a species-specific model, second choice a genus-specific model, third choice a model for functional group and, in case none of these were available, a general model based on all point-intercepts was used.

Statistical analysis of treatment data

Generalized linear models (GLM) were used to assess the effect of grazing treatment on the two biodiversity metrics, richness and unicity, and on forb:graminoid ratio and litter amount, while considering block, topographic position and treatment. Block was nested in topographic position, which was either hill or marine foreland. GLM with Poisson errors and log link function was used for species richness and for litter (standing dead intercepts), whereas ordinary Gaussian regression was used for the forb:graminoid ratio.

For litter and for forb:graminoid ratio, the procedure was run for raw intercept counts and for calibrated biomass in parallel.

Results

Biomass calibration model

The initial generalized linear model of harvested dry mass per point-intercept across species and plots showed the expected positive linear relationship, but with much un-explained variation ($R^2 = 0.517$). The subsequent ANOVA showed species identity to be the by far most important factor accounting for the residual variation, explaining 56.4% of the deviance. In contrast, treatment explained less than 2% of the total deviance. Thus, there was a significant degree of variation in mass per intercept across species, justifying the attempt of making single-species calibration models.

In the 24 quadrats recorded i 2020, a total of 60 plant species were found, of which 35 occurred in more than three quadrats. For 22 species, linear regression models of biomass on intercepts yielded a p-value < 0.05 as were accepted as single-species prediction models. Five genera with more than one species present yielded acceptable models, i.e. *Agrostis, Carex, Galium, Festuca* and *Juncus* (*effusus*-type) and, similarly, the four functional types as well as for total biomass and for standing litter. The regression models are presented in Supplementary materials (Appendix 2: Table S1).

Grazing treatments

The total number of species per treatment found across plots differed quite markedly, with full exclosure having the fewest species in total (Table 1). Similarly, the average species density, i.e. number of species per

6

plot (mean alpha diversity), varied being treatments, with winter-only and year-round grazing showing the highest levels (Table 1).

Treatment	? diversity	? diversity
Winter-only grazing	50	11.11
Year-round grazing	49	10.78
Summer-only grazing	42	9.67
Annual mowing	41	8.78
Full exclosure	37	7.78

Table 1: Richness of vascular plants at the ? and ? level for each treatment.

In the second-year data, treatment accounted for a rather small fraction of total deviance as compared to block and topographic position. Only for unicity did treatment overall seem to have an appreciable effect (Table 2). When broken into individual treatment groups, species richness per plot (log transformed) was relatively high in winter-only and year-round grazing, while it was low in full exclosure. When treatment, location and block were considered, the model could account for 64 % of the total deviance in richness between quadrats. Treatment overall accounted for 10 % explained deviance, without being statistically significant.

A similar pattern was seen for unicity, for which winter-only grazing on average had a more than 50 % higher level than full exclosure. Here, the model could account for 61 % of the total deviance, with treatment overall being statistically significant and accounting for 17 % of total deviance.

No statistically significant effects of treatment on the forb:graminoid ratio was found. Nonetheless, we find the tendency for annual mowing to have the lowest ratio noteworthy. For explanatory models based on calibrated biomass estimates, the patterns for forb:graminoid ratio and litter were very similar. The models showed lower explained deviance for litter and higher for the forb:graminoid ratio.

	Treatment intercept and effects					Deviance explained	
	Full exclosure (intercept)	Mow-ing	Rewil-ding	Summer exclosure	Winter exclo-sure	Treat-ment	All vari-ables
Richness (log-link)	2.05***	0.12 ns	0.33*	0.36*	0.22 ns	10.44%	64.18%
Unicity	0.01***	0.0 ns	0.0(p=0.07)	0.0*	0.0 ns	17.08%**	60.9%
Forb:graminoid ratio	0.47 ns	0.01 ns	0.23 ns	0.46 ns	0.32 ns	3.94%	41.97%
Litter (log-link)	4.46***	-0.58***	-0.84***	-1.03***	-0.77***	26.09%***	65.16%
Forb:graminoid ratio (Biomass estimate)	0.77	-0.13 ns	0.7 ns	1.08 ns	0.73 ns	5.73%	52.03%
Litter (Biomass estimate)	16.71***	-7.32*	-9.5**	-10.72**	-8.97**	25.57%***	60.56%

Table 2: *Intercept and biomass based GLM output using various biodiversity related parameters. The full exclosure treatment is used as the intercept, while the other treatments are shown with their effect relative to the intercept value. Deviance is listed as the percentage explained by the GLM model relative to the NULL model for 1) treatment alone and 2) for both treatment, location and block. Levels of significance are: * = p < 0.05, ** = p < 0.01, *** = p < 0.001.*

The biodiversity metrics assessed featured high within-group variation from the treatment means (Fig. 2-5). This is a indication that local environmental conditions and legacy effects of initial plant communities prior to setting up exclosures strongly influence plant communities, which was also reflected in the high proportion of deviance explained by location and block in GLMs. The deviance is especially high for the forb to graminoid ratio, which is in accordance with field observations, where some quadrats featured an almost complete dominance of either forbs or graminoids.
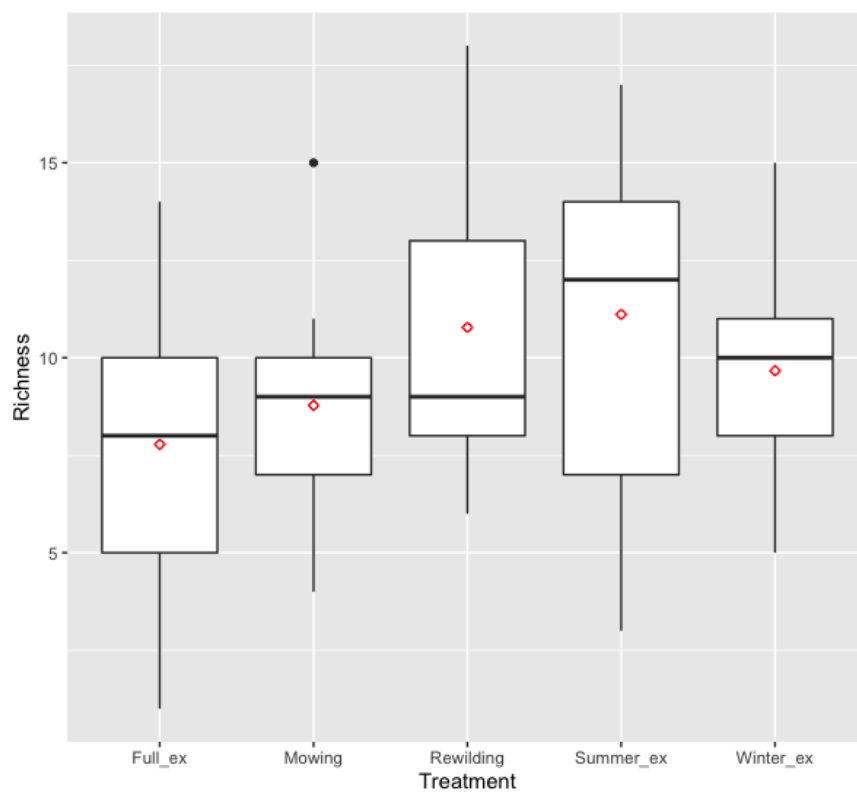
Figure 2: Boxplot of richness by treatment. Mean values are indicated by the red dots, the black bars show the median values and the white boxes contain all values between the lower and upper quartiles. Year-round grazing and Winter-only grazing are statistically significantly different from Full-exclosure.
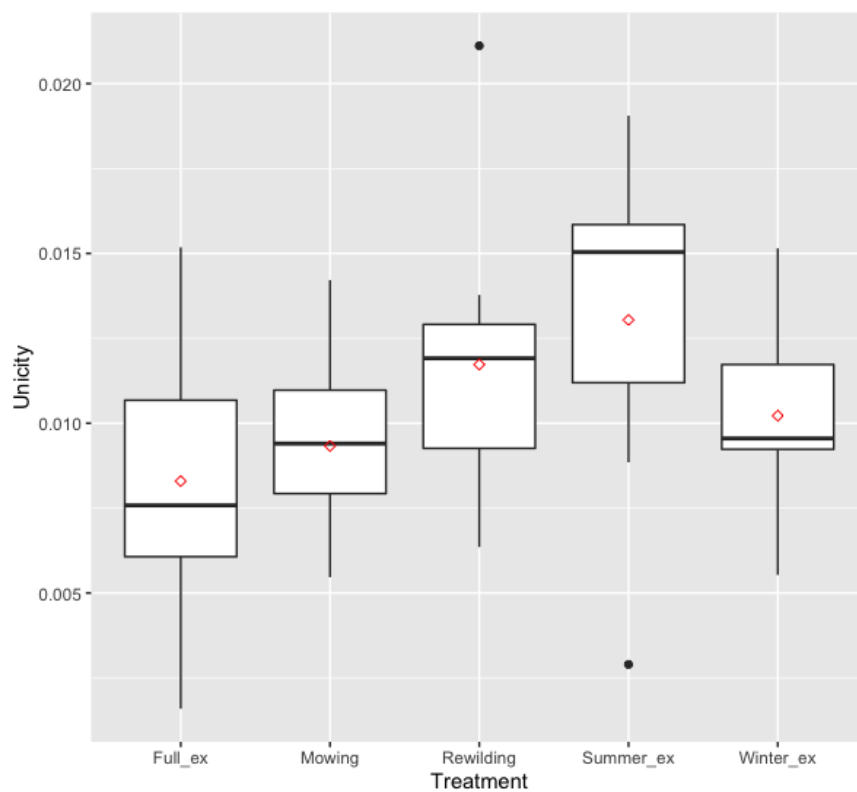
Figure 3: Boxplot of unicity by treatment. Mean values are indicated by the red dots, the black bars show the median values and the white boxes contain all values between the lower and upper quartiles. Winter-only grazing is statistically significantly different from Full-exclosure.
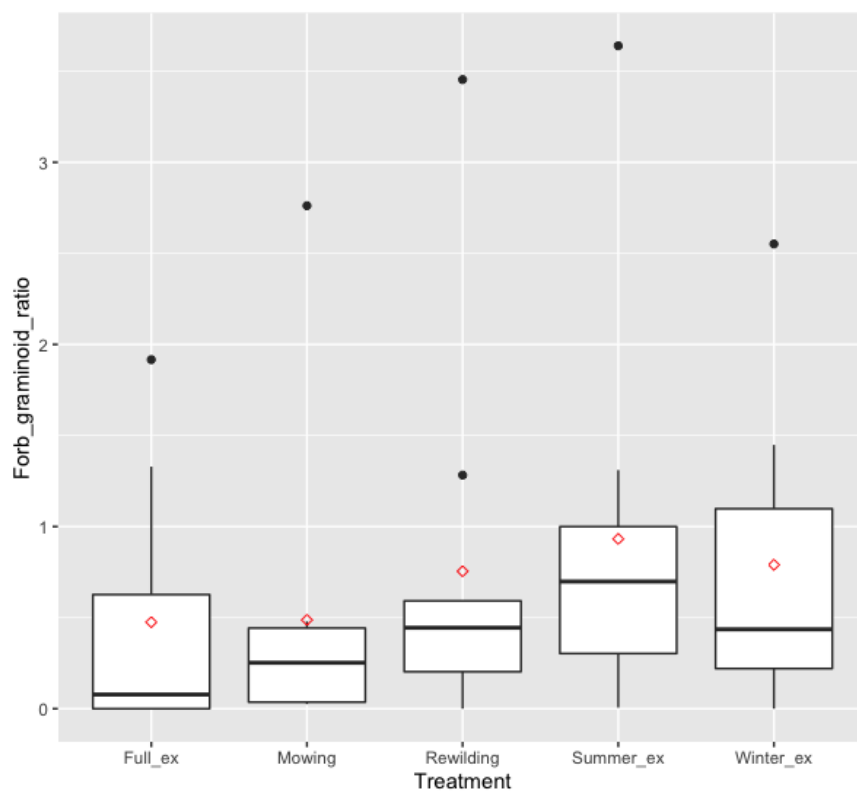
Figure 4: Boxplot of the forb to graminoid ratio by treatment. Mean values are indicated by the red dots, the black bars show the median values and the white boxes contain all values between the lower and upper quartiles. Differences between treatments are not statistically significant.
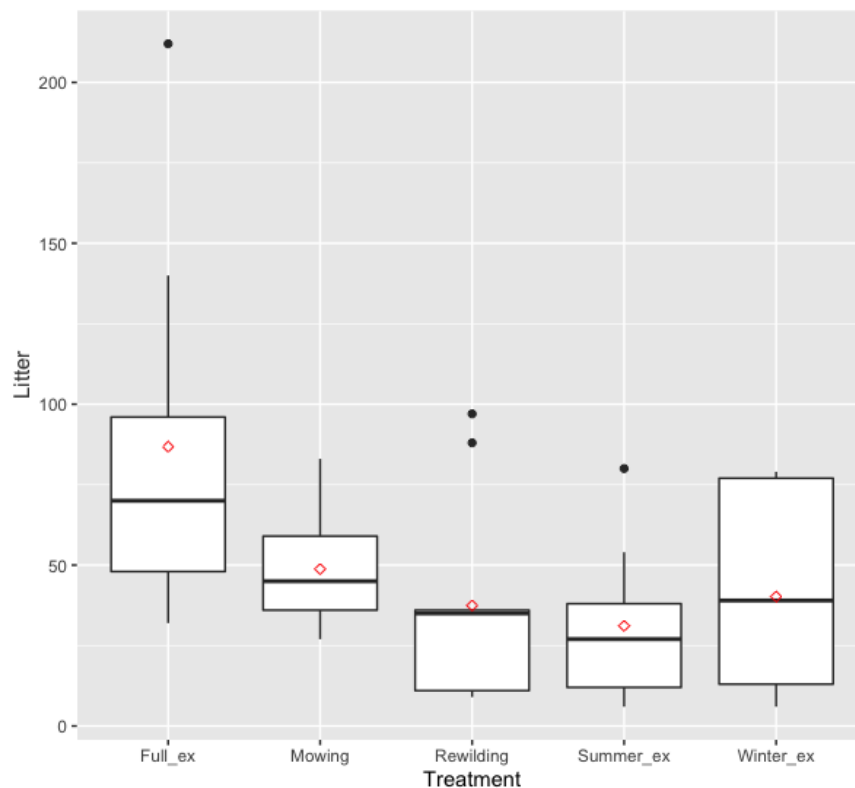
11

Figure 5: Boxplot of litter by treatment. Mean values are indicated by the red dots, the black bars show the median values and the white boxes contain all values between the lower and upper quartiles. All other treatments are statistically significantly different from Full-exclosure.

Discussion

Grazing by semi-feral horses and cattle under near-natural conditions was found to support higher plant species richness and higher prevalence of less widespread species (unicity) in grassland vegetation, as compared to annual mowing and to full exclosure of large herbivores. Also, we saw a tendency for the prevalence of forbs over graminoids to be higher, which may result in increased floral resource availability to anthophilous insects. At first glance, this result may seem trivial, since temperate grasslands as an ecosystem are inherently dependent on the disturbance regime maintained by the activities of megafauna (Janis et al. 2002; Pärtel et al. 2005; Kuneš et al. 2015). It is nevertheless, in our view, an important result, as it highlights the detrimental effect on grassland plant diversity of the now widespread grazing abandonment or mechanical biomass harvest. Since wild grazers, such as red deer, roe deer and hare, had free access to the fenced plots and were observed in the area, our full exclosure treatment mimics the standard conditions in most temperate European landscapes, where populations of wild herbivores are kept at very low densities, i.e. the maximum acceptable to agriculture and silviculture (Fløjgaard et al. 2022). In accordance, we found accumulation of litter in full exclosure plots. Thick layers of litter changes light and microclimate conditions at the soil surface, deteriorating conditions for plant recruitment and survival (Jensen & Gutekunst 2003) and reducing the richness of arthropod assemblages (van Klink et al. 2015). Although grazing and haymaking may share more similarities than dissimilarities (Pykälä 2000), our results indicate that annual autumn mowing does not support the same plant species richness and abundance of forbs as grazing.

One more reason not to over-emphasize differences in vegetation response between grazing treatments ob-

tained from exclosure plots is that larger grazed landscapes are likely to encompass areas used more and less intensively by the large herbivores at different times of the year. Even if it would be possible to experimentally demonstrate plant community properties typical of either year-round, winter-only and summer-only grazing, it may be hypothesized that all these characteristics would be created or maintained in the landscape at large by such animal behaviour, thereby promoting overall habitat heterogeneity. More specifically, summer-only grazing might be a natural grazing regime in naturally nutrient-rich meadows, which are flooded during winter, and that winter-only grazing is a natural regime in grassland, heathland and open woodland on nutrient-poor higher grounds. We also acknowledge that our study only takes a snapshot of effects that may depend on legacy effects of previous land use (Stroh et al. 2021) and may be transient in the dynamic interaction between herbivore population dynamics and inter-annual variation in weather conditions, such as the irregular occurrence of severe summer droughts (Stampfli et al. 2018).

It should be emphasized that modern European grassland management as an agri-environmental practice often occurs as intense summer grazing at high stocking rates (Fløjgaard et al. 2022). This practice is very different from our summer-only grazing treatment, in which animal densities approach natural densities, which are set by winter forage carrying capacity.

Several recent studies have reported a positive effect of year-round grazing on plant species richness and – in particular – the prevalence of rare or threatened species (Köhler et al. 2016; Rupprecht et al. 2016). Temperate European landscapes have seen major habitat changes over recent decades and centuries affecting plant species and communities, including the effective disappearance of first, large wild herbivores and second, free-roaming livestock (Bruun & Fritzbøger 2002; Finderup Nielsen et al. 2021). Naturalistic grazing appears to be key in restoring habitat conditions lost in this long-term land-use change and intensification. Although, in the present study, we were not able to encompass the grassland-scrub-forest ecotone, the heterogeneity arising from naturalistic grazing is probably important to many species showing declining trends in human-dominated landscapes (e.g. Maes et al. 2014).

A tendency for naturalistic grazing to promote forbs over graminoids has recently been reported elsewhere (Henning et al. 2017; Dvorský et al. 2022). Despite graminoids lending their name to the grassland ecosystem, forbs are essential to the phylogenetic and functional diversity of grassland communities, and are particularly important to flower-seeking insects. It has been hypothesized that grass dominance in temperate open biomes is a relatively recent phenomenon (Bråthen et al. 2021) and, no doubt, grass dominance has been promoted by agricultural grassland management (Dengler et al. 2020). Possibly, the change of dominance towards forbs may be an effect of animal trampling as much as their forage preference (Striker et al. 2011). Although differences in forb:graminoid ratio between our treatments were not statistically significant given the variation between plots, we note that the highest levels of forb relative abundance was found in the three grazing treatments. We also note that certain forb species may be sensitive to grazing, but in a larger grazing landscape probably would find habitat in browsing shelter between thorny shrubs or fallen logs.

From a methodical standpoint, our results indicate that the application of the model derived biomass estimates in most cases do not change the overall patterns also revealed by the intercept based analyses. It would therefore seem that the supposed differences in species morphology were, in most cases, not large enough to generate significantly different results in terms of species composition. Furthermore, the application of the calibration models to the number of intercepts constitutes a complicating step in the analysis, due to the error propagation that results from using model estimates in a function. While this does not mean that the model derived results are invalid, it may be both more efficient and accurate to use the intercept based results. It can therefore be argued that the use of biomass calibration models may not be necessary for comparisons to be made between treatments, unless results from methodologically different studies are compared.

Grazing is a natural ecosystem process and, thus, restoring naturalistic grazing using feral animals may be seen as restoration goal in itself, notwithstanding the effects on biodiversity. Nevertheless, the present study demonstrates that naturalistic grazing as a management tool may indeed create habitat for regionally uncommon species of conservation concern and probably enhance resource availability to flower-seeking insects.

Acknowledgements

Supplementary materials

Appendix 1: Fieldwork protocol

The point-intercept method was used to assess plant species' abundance. A gridded quadratic aluminium frame of used to systematize the distribution of points within the 25×25 cm sampling area with 5×5 cm spacing. The frame had two parallel grid layers, made of nylon fishing line, in order to provided a fixed trajectory for the vertical stick, which was a 0.8 cm thick wooden stick. The frame was placed on four aluminum legs, with a height of 50 cm.

The frame was designed to provide 25 intercept points in a 25×25cm sample surface, in order to balance the need to detect a variety of plants and obtain sufficient sample sizes with the available time for fieldwork. The Jonasson (1988) study successfully used a frame with 5×10 cm spacing to model the biomass of dwarf shrub and graminoid plant communities. As the vegetation at Molslaboratoriet is much more diverse and contains many smaller plant species than the vegetation sampled by Jonasson, the spacing was reduced to 5×5 cm, allowing for twice the number of points per area. Another consideration was to use as small an area as possible within the research plots, as the sample surface would be harvested after the point-intercept registration in the first period of fieldwork.

The sample surface was systematically placed in alignment with one of the two corner posts of the vegetation monitoring plot facing the outer fence as shown in fig. 6. Out of these two potential sample surfaces, the one with the least amount of scrub was chosen. If no scrub was present, the sample surface was chosen by coin-toss. The sample surfaces for the first period of fieldwork were placed just outside of the vegetation monitoring circle, as they would subsequently be harvested. The sample surfaces for the second period were placed inside the sample surfaces, following the same principles laid out above.
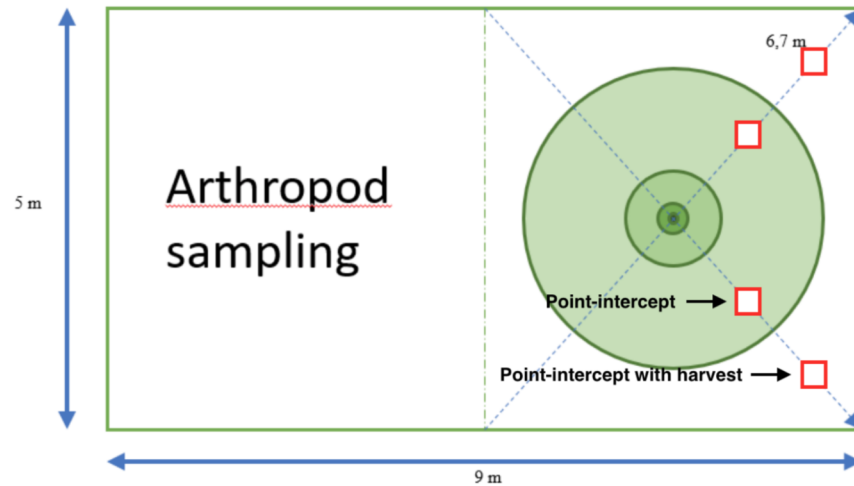
Figure 6: Schematic of the treatment plots with the possible placements of the sample surfaces. Edited from original by Bjarke Madsen & Signe Normand, 2017.

The 25x25 cm. sample surface was delineated with 4 corner pegs. A species list of vascular plants was created for each sample surface, in order to ease the subsequent identification during the point-intercept analysis. The intercept registration was then carried out as described in the materials and methods section. The picture in fig. 6 shows the practical application of the method.

Figure 7: Closeup of pin insertion into vegetation. Christoffer Bonavent.

In the first period of fieldwork, after the registration of point-intercepts, the above-ground biomass of the sample surface was harvested in the following systematic way.

1. The outline of the sample surface was traced with a kitchen knife, cutting into the soil surface. Parts of rosette plants, graminoids in tufts and large individual plants (such as *Cytisus scoparius* and *Solidago virgaurea*) that extended beyond the sample surface were cut off and were not included in the harvest. Graminoids with numerous and long basal leaves were "combed" so that leaves originating outside of the sample surface perimeter were left out of the harvest, and leaves originating inside the sample surface, but extending outside were included in the harvest.
2. The overall approach to harvesting was to include as much plant biomass and as little mineral soil as possible. Plants with mainly vertical growth were gathered in handfuls and cut of at the base, just above the mineral soil, using a set of Gardena grass shears. Rosette plants were cut just below the rosette, to include as much plant material as possible. Above-ground rhizomes were also harvested, when they were visible and no digging was required. Mosses were also harvested, cutting as close to the top layer of soil as possible. Dead plant material was harvested in the same way as living plant material. The humus layer of topsoil was not included in the biomass harvest.
3. All harvested plant material was put into clearly marked paper bags. If possible, some early sorting of species was performed, in order to ease the later process of sorting the plant material at the species level. Harvested biomass was placed in a fridge as soon as possible after harvesting, for later sorting.

Harvested plant material was hand-sorted to species in the laboratory and placed into separate paper bags. Some plant material, such as small fragments, was not possible to identify to the species level and was simply labelled as "unknown plant material". The bags were then placed in a drying oven and dried at 55° celsius

16

until the weight had stabilized. The dried plant material was stored in the paper bags at room temperature and then weighed, using a scientific scale with an accuracy of 0,01 grammes and a plastic tray, after the last day of fieldwork.

Figure 8: The biomass harvesting procedure. Christoffer Bonavent.

Appendix 2. Calibration models for single species, genera, functional groups, total biomass and standing litter

Model	b	Std. error	r^2	p	Sample size
Species models					
Achillea millefolium	0.05	0.01	0.9	<0.0	9
Agrostis canina	0.06	0.0	0.98	<0.0	7
Agrostis capillaris	0.07	0.01	0.85	<0.0	21
Anthoxanthum odoratum	0.06	0.01	0.92	<0.0	9
Carex nigra	0.12	0.01	0.97	<0.01	4
Carex panicea	0.15	0.01	0.97	<0.0	8
Cytisus scoparius	0.48	0.05	0.97	<0.01	4
Euphrasia stricta	0.24	0.05	0.93	<005	3
Gallium verum	0.07	0.01	0.92	<0.0	7
Hierachium pilosella	0.14	0.01	0.97	<0.0	8
Holcus lanatus	0.05	0.01	0.95	<0.0	6
hypochoeris radicata	0.09	0.02	0.83	<005	5
Juncus conglomeratus	0.13	0.02	0.88	<0.0	7
Juncus effusus	1035	0.15	0.9	<0.0	6
Knautia arvensis	0.12	0.01	0.98	<0.01	4
Luzula campestris	0.02	0.0	0.88	<0.0	9
Plantago lanceolota	0.09	0.01	0.81	<0.0	12
Potentilla erecta	0.08	0.01	0.9	<0.0	10
Rumex acetosa	0.06	0.01	0.94	<0.0	7
Rumex acetosella	0.04	0.0	0.99	<0.0	4
Solidago virgaurea	0.15	0.01	0.99	<0.0	4
Stellaria graminea	0.06	0.0	1.0	<0.01	3
Genus models					
Agrostis	0.06	0.0	0.88	<0.0	30
Carex	0.12	0.01	0.88	<0.0	25
Festuca	0.02	0.01	0.26	<0.01	31
Galium	0.07	0.01	0.91	<0.0	9
Juncus	0.55	0.15	0.54	0.28	13
Functional models					
Broad leaved graminoids	0.07	0.0	0.82	<0.0	89
Narrow leaved graminoids	0.04	0.01	0.41	<0.0	49
Forbs	0.09	0.0	0.77	<0.0	107
Trees and shrubs	0.47	0.04	0.97	<0.0	6
General models					
General	0.07	0.0	0.52	<0.0	264
Withered	0.19	0.03	0.68	<0.0	24

Table 3: Linear calibration models

Out of the total of 60 recorded species in the first period of fieldwork, 35 had the three or more datapoints required to construct a simple linear regression model. Of these, it was possible to create linear models with a p value < 0.05 for 22 species using untransformed data, as shown in table 3. Forcing the intercept through the origin seemed to have positive effects on the R^2 values of the models, which were generally very high, with an average value of 0.92. The slope coefficients (b) of untransformed models ranged from roughly 0.02 to 1.03, however, the 1. quartile was 0.06 and the 3. quartile 0.14, meaning that half the species fall within

18

this much narrower window.

In order to to extend model coverage to species with insufficient data, models for selected genera were constructed. In order for the genera-based models to be accurate, the constituent species must share largely similar morphology. If two or more morphological types in the same genera were sampled, each with more than one species, several models could potentially be constructed, although that was not necessary for this study. As seen in table 3, all models had p values <0.001, with high r^2 values for *Agrostis, Carex* and *Galium*, but not for *Festuca* and *Juncus*.

All morphological type models had p values < 0.001 and, with the exception of "Narrow leaved graminoids" and "Effusus type rushes", all had r^2-values above 0.76, meaning that they would in theory be more accurate than the general model if used for modelling the species that had insufficient data for individual models.