Floral resources in semi-natural grasslands - significance of grazing intensity and spatial variation

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Abstract

Vegetation composition and reproduction of vascular plants were studied in relation to seven spatial structures: *Betula pendula*, *Pinus sylvestris*, *Juniperus communis, Rosa dumalis*, dung pats, grazing rejects and grazed patches. The study was performed in 14 unfertilized semi-natural pastures, with different grazing intensities, in south-central Sweden. Vegetation height differed between sites and between structures and significantly influenced plant reproduction. Intensive grazing decreased the amount of grazing reject and increased the area of grazed patches. Vegetation height and number of fertile shoots were higher in grazing rejects, dung pats and near shrubs than in grazed patches and under trees, indicating that shrubs, but not trees, can function as partial grazing refugees. The results were used to simulate the effects of three different grazing intensities and three different shrub covers on plant reproduction. The simulation showed that grazing intensity was more important for plant reproduction than shrub cover due to the strong effect on the quantity of grazing rejects. Study site was the factor that best explained the variance for plant composition, accounting for 39% of the variation; spatial structurs accounted for 16% of the variation. Trees, shrubs, vegetation height and grazed patches significantly affected plant composition but not dung pats and grazing rejects.

Introduction

Semi-natural grasslands characterized by nutrient-poor soils are among the most species-rich habitats in Europe (Kull & Zobel, 1991; Mortimer *et al.*, 1998; Pärtel & Zobel, 1999). They harbour a large proportion of nationally red-listed species; for example, vascular plants, phytophagous insects, and insects depending on nectar or pollen (e.g. Gärdenfors, 2005). In Europe, the area of semi-natural grasslands has decreased due to intensified or altered land use and pastures and hay meadows have been transformed into arable fields, planted with forest or abandoned (Ihse, 1995; Hodgson *et al.*, 2005; Dahlström *et al.*, 2006).

Regular disturbance to the vegetation, like grazing or mowing, is a prerequisite for species richness in semi-natural grasslands, since it counteracts succession towards tall, species-poor vegetation, scrubland and eventually forest (Vera, 2000). On the other hand, many of the grassland species rely on plants escaping grazing; for example seed predators, nectar/pollen feeders, and many phytophages (Morris, 1967). Moreover, many plant species depend on grazing refugees for their reproduction (Milchunas & Noy-Meir, 2002). Thus, grassland management for biodiversity must aim at a disturbance regime that is intense enough to counteract succession, but weak enough to allow sufficient flowering and reproduction of plants and insects. To obtain such optimal management, type, timing and intensity of management, and the abundance of certain spatial structures that may function as grazing refuges can be manipulated. In this study, we focus on two of these management tools, grazing intensity and spatial structures, and investigate their importance for plant reproduction and species composition in semi-natural grasslands.

In Swedish grasslands, shrubs are among the most common spatial structures that may function as partial grazing refuges, thereby contributing to the spatial heterogeneity of grazing in pastures (Callaway *et al.* 2000; Bakker *et al.*, 2004; Bossuyt *et al.* 2005). For example, *Juniperus* seedlings were shown to have a higher survival under the canopy of mature *Juniperus* and the highest growth rates of seedlings were found at the edge of the canopy (van Auken *et al.*, 2004). Similarly, survival of oak seedlings was highest in thorny shrubs of *Prunus spinosa* where they were protected from grazing (Bakker *et al.*, 2004). *P. spinosa* shrubs can also prevent grazing of other palatable species (Rousset & Lepart, 2002).

In addition to shrubs, dung pats can function as grazing refuges since up to 40 cm of the vegetation around the excreta is avoided by grazing animals (Shiyomi *et al.*, 1998; Jones & Ratcliff, 1983). Dung pats affect the vegetation by increased concentration of nutrients that may influence growth rate, survival and growth form of plants (Bullock & Marriot, 2000).

Temporary grazing refugees can also be created by selective grazing and dietary choices by grazing animals (Rook *et al.*, 2004). Cattle can avoid patches with low forage quality (hereafter called grazing rejects) and alternate between patches with high forage quality, which leads to a mosaic of patches with different grazing pressure in the pasture (Bailey *et al.*, 1998). Palatable species can experience reduced grazing when associated with unpalatable species (Callaway *et al.*, 2000; Bossuyt *et al.*, 2005). Selectivity decreases at higher grazing intensities, which results in a more homogenous sward structure (Jerling & Andersson, 1982; Rook *et al.*, 2004).

Trees and shrubs are long-lived

structures and they influence plant species composition in grasslands. For example, oak trees can be several hundred years old and the canopy of individual trees can cover considerable areas of grass sward (Reyes-López, 2003; Rozas, 2005). Shrubs constitute long-lived features in the grassland, e.g. *J. communis*, that can reach more than 100 years of age (Rejmanek & Rosén, 1992). In contrast to trees, single shrubs influence the vegetation only in small areas below and adjacent to the shrubs (Rejmanek & Rosén, 1992; Marion & Houle, 1996). The effect of trees and shrubs on the grass sward may be due to changes in soil properties, such as soil temperature, nutrients, pH and water content (Dahlgren *et al.*, 1997; Austad & Losvik, 1998; Amiotti *et al.*, 2000; Chambers, 2001).

In general, the effect of bushes on the grass sward diversity is ambiguous. Some studies have demonstrated reduced plant species richness as an effect of increased shrub cover (Rejmanek & Rosén, 1992; Willems & Bik, 1998; Hansson & Fogelfors, 2000; Vera, 2000; Willems, 2001) or increased plant species richness after clearing of shrubs and reintroduced grazing (Barbaro *et al.*, 2001; Rosén & Bakker, 2005). Other studies have shown that abundance of trees and shrubs in semi-natural grasslands may be correlated with species richness of plants (Söderström *et al.*, 2001; Lindborg & Erikssson, 2004), insects and birds (Söderström *et al.*, 2001). Historically, trees and shrubs that occurred in grasslands were used for pollarding, coppicing, and fruit production and were thus important resources in the traditional agricultural system (Peterson, 2005).

In contrast to long-lived trees and shrubs, dung pats can be expected to be too short-lived to leave imprints in the species composition of the grass sward. One exception may be resting places or other areas in which density of excretions is high (White *et al.*, 2001; Kohler *et al.*, 2006).

The aim of this study was to examine the potential for influencing plant reproduction in semi-natural pastures by manipulating either grazing intensity or density of shrubs and trees. This was done by first studying the spatial pattern of grazing and plant reproduction in grasslands with trees and shrubs under different grazing intensities. The spatial pattern showed how different persistent and temporary habitat structures affected plant reproduction and species composition. The field data were then used to model overall plant reproduction per area unit in relation to grazing intensity and density of shrubs. Specifically, we asked the following questions: (1) To what extent is plant reproduction in semi-natural pastures related to spatial variation of grazing? (2) To what extent are grazing and plant reproduction affected by habitat heterogeneity formed by trees and shrubs? (3) How is vegetation heterogeneity related to grazing intensity? (4) To what extent is the production of flowers and fruits affected by manipulation of grazing pressure and density of shrubs, respectively? (5) Which types of persistent and temporary habitat structures affect species composition of the grass sward?

Materials and methods

Study sites

Fourteen unfertilized semi-natural pastures were selected in the County of Uppland, south central Sweden (between 59°44′-60°17′N and 17°20′-18°36′E). The pastures ranged between 5 and 20 ha in size and were grazed by either dairy or meat cattle. They all have a long history of grazing. The vegetation type in all selected grasslands was "dry-mesic species-rich *Agrostis capillaris* type" (Påhlsson, 1994). Scattered trees of *Betula pendula*, *Picea abies* and *Pinus sylvestris* and shrubs of *Juniperus communis*, *Prunus spinosa* and *Rosa* spp. occurred in all grasslands. Common herbs and grasses were *Achillea millefolium*, *Agrostis capillaris*, *Festuca rubra*, *Galium verum* and *Poa pratensis*.

To select study sites with different grazing intensities vegetation height was measured in 25 pastures in July 2005. The pastures were similar with respect to vegetation type, density of trees and shrubs, and productivity. Vegetation height was measured at 1-m intervals in two 50-m transects per site using a rising plate meter (Correll *et al.*, 2003). The pastures were then sorted by mean vegetation height, a rough estimate of grazing intensity, and 14 pastures that represented a gradient from intense to weak grazing were selected for further investigation.

Data sampling

In August, vegetation height was again measured in the transects, and the average August vegetation height was hereafter used as estimate of grazing intensity. To estimate spatial variation of grazing intensity created by the grazers' selectivity, each measuring point in the transects was classified as grazed, grazing reject, or dung pat. To evaluate how grazing and plant reproduction varied between different habitat structures, 15 sampling plots of 50 x 50 cm were randomly placed at each of seven different spatial structures per pasture. The structures were grazing rejects, dung reject, grazed patches, solitary trees of *Betula pendula* and *Pinus sylvestris*, and shrubs of *Juniperus communis* and *Rosa dumalis*. Plots under trees were placed midway between the trunk and the canopy edge; shrub plots, next to the shrub edge. Of *J. communis* shrubs only specimens with low growing branches were chosen because they may function as grazing refuges. Dung pats of 2005 were selected and the plots were placed at the edge of the dung pat. Grazing rejects were defined as patches with non-grazed vegetation, not belonging to any other structure, and grazed plots as patches with apparently grazed vegetation. In the sampling plots vegetation height was measured to estimate the grazing pressure in different structures.

The abundance of all plant species was estimated by presence-absence in the central 10 x 10cm of each sampling plot. All reproductive units of herbs, grasses and sedges were counted in the sampling plots. A reproductive unit was defined for each species as the smallest unit of reproductive organs (buds, flowers, fruits) that could be readily recognized and counted in the field. For most herbs, the reproductive unit was defined as a single bud, flower or fruit. For herbs and for sedges with panicles, cymes, composed umbels or racemes, these were counted as reproductive units. For grasses, the reproductive unit was defined as a panicle.

Differences in density of reproductive units between structures may be due to grazing selectivity (some structures may be avoided by grazing animals) or productivity (in some structures more reproductive units are produced) or both. To separate effects of grazing selectivity and production of reproductive units these parameters were monitored weekly from mid-May to mid-August in one pasture (Åsbergby, 59°44′N and 17°55′E), in ten 50 x 50cm plots per structure (grazed patch, dung pat, rose, juniper, and pine). The approximate number of reproductive units produced since the previous sampling date was estimated by mapping reproductive units in a 10 x 10cm grid in each plot at each date. The sum of all new reproductive units observed during the study period was taken as the total production. Grazing of reproductive units was given by the difference between total production and the number of reproductive units in mid-August. In each plot, vegetation height was measured in August using a rising plate.

Simulating the relative effects of grazing intensity and spatial habitat structures

Grazing intensity was assumed to influence (1) the cover of grazed patches, rejects and dung pats, and (2) the density of reproductive units in these and other habitat structures. Hence, in a homogeneous pasture without permanent spatial structures, the density of reproductive units is:

$$rd_{pasture} = (p_{reject} \times rd_{reject}) + (p_{dung} \times rd_{dung}) + (p_{grazed} \times rd_{grazed}) \qquad \text{(eq. 1)}$$

where p is the proportion cover of the structure (the three proportions sum to 1) in the pasture and rd the density of reproductive units. The overall density of reproductive units was simulated under four different grazing intensities, corresponding to a vegetation height of 3, 5, 7, and 9 cm in August. Simulation was done by using field data to parameterize eq. 1 in the following way: p_{reject} = cover of rejects according to the trend line in Fig. 1, p_{dung} = 0.1 (based on Fig. 1), $p_{grazed} = 1 - p_{reject} - p_{dung}$, rd_{dung} and rd_{grazed} = mean density of reproductive units relative to that in ungrazed rejects (trend lines in Fig. 3), $rd_{reject} = 1$ because rejects were ungrazed per definition.

Of the simulated grazing intensities, 3 cm was assumed to yield the lowest density of reproductive units, and was therefore set as baseline = 1. Weaker grazing was thus assumed to increase the density of reproductive units relative to that baseline. In a pasture with permanent spatial habitat structures a certain proportion of the grass sward will be situated close to, and potentially affected by those structures. The results showed that shrubs but not trees affected density of reproductive units. Hence, the overall density of reproductive units is:

$$rd_{pasture} = (p_{reject} \times rd_{reject}) + (p_{dung} \times rd_{dung}) + (p_{grazed} \times rd_{grazed}) + (p_{shrub} \times rd_{shrub}) \qquad \text{(eq 2)}$$

where p_{shrub} is the proportion cover of grass sward within 0.5 m from shrubs (the four proportions sum to 1). By parameterizing eq. 2 the overall density of reproductive units was simulated for the same four grazing intensities as described earlier, and for two levels of area influenced by shrubs, p_{shrub} = 0.2 and 0.4. rd_{shrub} was parameterized using the average of the trend lines for roses and junipers in Fig. 3. The proportion cover of all structures sum to 1, and when adding shrubs to the pasture, the cover of other structures was reduced in relation to each structure's relative cover. Density was calculated per area grass sward, thus excluding the cover of the shrubs themselves. The other parameters were parameterized as described for eq. 1, and the same baseline (grazing intensity = 3 cm, no shrubs) as before was used.

Statistical treatment

To analyse how mean vegetation height and mean density of reproductive units differed between habitat structures and sites, two-way ANOVA was used across all sites and structures with site as a random factor, structure as a fixed factor, and plot-specific data (15 plots per structure per site) as dependent variables. Significant differences were analysed with post hoc tests with Bonferroni correction for multiple comparisons.

The grazing rejects were selected to be per definition unaffected by grazing, but mean plot density of reproductive units varied from about 60 to 150 between grasslands, mainly depending on species composition and slight differences in productivity. To estimate the relative importance of different habitat structures for plant reproduction at a certain site, such differences were controlled for by using density of reproductive units in the structures relative to that in ungrazed rejects ($\text{density}_{\text{structure}}/\text{density}_{\text{reject}}$). Linear, quadratic and cubic regression was used to find the best curve for the relationship between plant reproduction estimates and site vegetation height.

Species specific production of reproductive units in different spatial structures under different grazing intensities was tested using multivariate analyses across all structures and sites. To allow comparison of species with different definitions of reproductive units, proportions of reproductive units in different structures and sites were used instead of actual numbers. Thus, for each species the total number of observed reproductive units was set as 1 and partitioned among the 14 sites and 7 structures. Species occurring in ≥ 10 plots (64 species, see Appendix 1) were used in the analyses. Data were root arc sinus transformed before analysis, to avoid the variance being a function of the mean (Fowler *et al.*, 1998). Detrended correspondence analysis (DCA) was first carried out to select the most appropriate model, linear or unimodal (Leps & Smilauer, 2003). Since the length of the gradient was 2.8 RDA (linear) analyses were selected. Environmental variables were entered either as dummy variables (site and structure) or as continuous variable (plot vegetation height, Leps & Smilauer 2003). Site vegetation height (measured in transects) was multicollinear with study site and removed from the analyses. Forward selection of environmental variables followed by Monte Carlo permutation tests with 999 permutations were used to test the significance of the environmental variables. Partial constrained ordination (pRDA) was performed to quantify the effects of two groups of environmental variables, site and spatial structure (Borcard *et al.*, 1992).

Variation in plant species composition between habitat structures and sites was tested using multivariate analyses as described for reproduction. Data on plant species abundance were calculated as proportion of plots (x/15) with occurrence per species, structure and site. Only species that had an abundance ≥ 10, i.e. 64 species, were used in the analyses. DCA showed the gradient to be 2.1 and therefore RDA was used. Multivariate analyses were performed using the software Canoco for Windows 4.5 (ter Braak & Smilauer, 2002), and other analyses using SPSS version 13.0.

Results

In absence of shrubs and trees, a mosaic of grazed and ungrazed patches was formed by the grazers by selective foraging and dung deposition. The relative cover of grazing rejects increased with site vegetation height, measured in transects (quadratic $r^2=0.77$, $n=14$, $p<0.001$, Fig. 1), whereas the cover of dung pats was not correlated with site vegetation height (linear $r^2=0.05$, $p=0.5$, Fig. 1). Plot vegetation height varied significantly between sites (two-way ANOVA $F=7.5$, $df=13$, $p<0.001$), structures ($F=62.1$, $df=6$, $p<0.001$), and with the site*structure interaction ($F=4.3$, $df=74$, $p<0.001$). Post hoc tests showed that plot vegetation height varied between spatial structures in the following sequence: grazing reject > dung pat > rose = juniper > pine > birch

= grazed (Fig. 2). Plot vegetation height in junipers and roses were significantly correlated with site vegetation height (linear $r^2>0.46$, $n=14$, $p<0.012$), but no correlation was found for the other structures ($p>0.06$).

Density of reproductive units in plots varied significantly between sites (two-way ANOVA $F=4.8$, $df=13$, $p<0.001$), structures ($F=22.2$, $df=6$, $p<0.001$), and with the site*structure interaction ($F=3.7$, $df=74$, $p<0.001$). Post Hoc tests showed that plot density of reproductive units varied between habitat structures in the following sequence: grazing reject>dung pat=juniper>rose> birch>pine=grazed (Fig. 2). At the plot level, density of reproductive units was significantly correlated with vegetation height (Spearman rank correlation over all sites, $r=0.76$, $n=94$, $p<0.001$, Fig. 2). Site vegetation height was correlated with mean density of reproductive units in grazed patches (linear $r^2=0.36$, $n=14$, $p<0.05$), dung pats (quadratic $r^2=0.42$, $p<0.05$), junipers (linear $r^2=0.65$, $p<0.01$), and roses (linear $r^2=0.35$, $p<0.05$), but no correlation was found for the other structures ($r^2<0.40$, $p>0.14$, Fig. 3A). Density of reproductive units relative to that in ungrazed rejects was significantly correlated with site vegetation height for the same four habitat structures: grazed patches (quadratic $r^2=0.77$, $p<0.01$), dung pats (linear $r^2=0.58$, $p<0.01$), junipers (linear $r^2=0.78$, $p<0.001$) and roses (linear $r^2=0.64$, $p<0.01$; Fig. 3B).

Detailed mapping of reproductive units in one of the pastures showed that mean August density of reproductive units in grazed patches was significantly lower than in dung pats, roses and junipers (one-way ANOVA with Bonferroni correction, $n=10$, $p<0.002$), and that dung pats in turn had higher density than junipers ($p=0.007$, Fig. 4). Mean August density of reproductive units in the different structures was correlated with mean vegetation height in the structures (Fig. 4). The variation in August density between structures did not covary with total density of reproductive units. Instead, compensatory production of reproductive units created the opposite relationship between structures, and grazed patches produced significantly more reproductive units per area unit than dung pats ($p=0.02$, Fig. 4). In grazed patches, only 15% of the observed reproductive units reached flower stage, the rest being grazed at the bud stage. In rejects around dung pats, 80% of the observed reproductive units reached flower stage.

Under the most intense grazing, no grazing rejects occurred (Fig. 1) and in a pasture without shrubs, all reproductive units would be found in grazed patches and around dung pats, at an average density of about 12 per 50 x 50 cm plot (Fig. 3A). Using this value as baseline and simulating density of reproductive units under weaker grazing shows that the density increases from the baseline with a factor of 2.1 up to 7 cm August vegetation, and with a factor of 1.7 from 7 to 9 cm. Weak grazing (9-cm August vegetation) thus yields about eight times higher density of reproductive units than the most intense grazing (Fig. 5). Adding a shrub cover of 0.2 to the model showed that the overall density of reproductive units increased by a factor of 1.14 relative to a pasture without shrubs, at a grazing intensity of 5 cm. The corresponding increase at a shrub cover of 0.4 was about 1.28. The relative effect of shrubs decreased both at higher and lower grazing intensities (Fig. 5).

Applying partial constrained analyses (pRDA) on the frequency of reproductive units of different species showed that study site accounted for 28.5% and spatial structure 15.9% of the variation in plant reproduction between species. In the RDA of plant reproduction, the

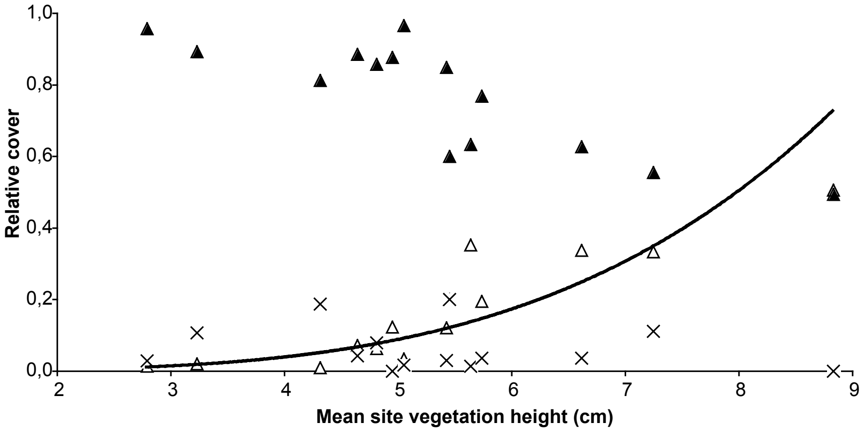

Fig. 1. Relative cover of grazed patches (filled triangles), grazing rejects (open triangles) and dung pats (crosses) in relation to mean site vegetation height in fourteen semi-natural pastures. Cover and vegetation height was measured in two 1*50m transects per site. Regression line is shown for the cover of rejects.

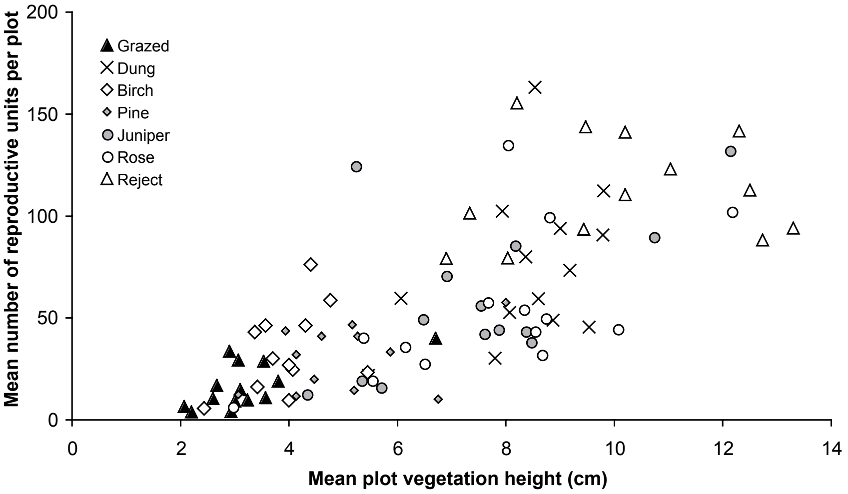

Fig. 2. Mean number of reproductive units in seven habitat structures, in relation to mean vegetation height in the structures, in fourteen semi-natural pastures. Each data point represents one structure in one pasture. Data were sampled in fifteen 50 x 50 cm plots per structure and pasture. For clarity, no error bars are shown.

first two axes accounted for 10% and 7% of the variation, respectively (Fig. 6). The first (horizontal) axis relates to vegetation height, with intense grazing (low vegetation) to the left. The second (vertical) axis relates to presence of shrubs and shows species associated with junipers and roses at the top, and species associated with grazing rejects and grazed patches at the bottom. Trees are found in the left part of the diagram (intense grazing), junipers in the right part, and roses in between (Fig. 6). Vegetation height, grazing reject, dung pat, rose and juniper showed significant association with plant reproduction (Monte Carlo tests, p≤0.05) whereas grazed patch, birch and pine were not associated with plant reproduction (Monte Carlo test, p>0.05). All study sites, except Långalma and Lövsveden, were significantly associated with plant reproduction (Monte Carlo test, p≤0.05).

A number of grass sward species reproduced mainly adjacent to junipers and roses: *Campanula percisifolia*, *Deschampsia flexuosa*, *Galium boreale*, *Pilosella officinarum*, *Veronica officinalis* and *Viola sp.* (Fig. 6). Dung pats and grazing rejects increased the reproduction of *Briza media*, *Cerastium fontanum*, *Festuca rubra*, *Lotus corniculatus*, *Phleum pratense*, *Plantago lanceolata,* and *Trifolium pratense.* No species had high density of reproductive units in grazed patches or near birch or pine.

Species composition patterns were best explained by site, which accounted for 38.5% of the variance, according to pRDA. Spatial structure accounted for 15.8% of the variance. In total 155 plant species were found (on average 62±3 species per site). In the RDA of species composition the first two axes account for 13% and 12%, respectively, of the variation (Fig. 7). The first (horizontal) axis relates to presence of birch and pine, with species associated with those trees to the right. The second (vertical) axis relates to presence of shrubs and shows species

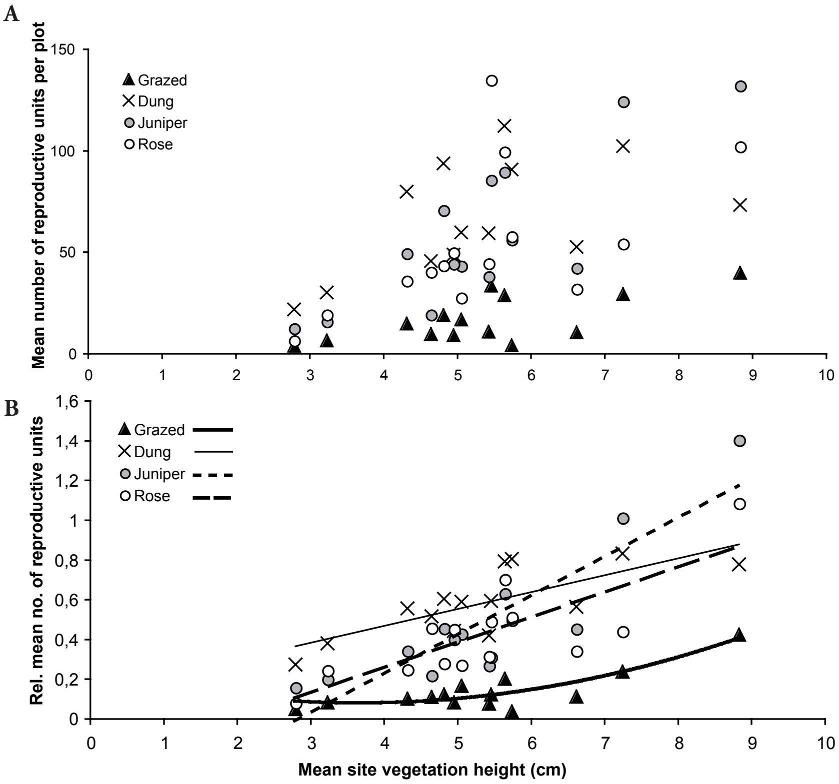

Fig. 3. Mean number (A) and relative mean number (B) of reproductive units per 50 x 50cm plot in four habitat structures in relation to mean site vegetation height in fourteen semi-natural pastures. Relative means show mean for the structure relative to the mean of ungrazed rejects at the site, i.e. density in the structure/density in rejects. Trend lines for best fit are shown (r^2 and significance values are given in text). For clarity, no error bars are shown.

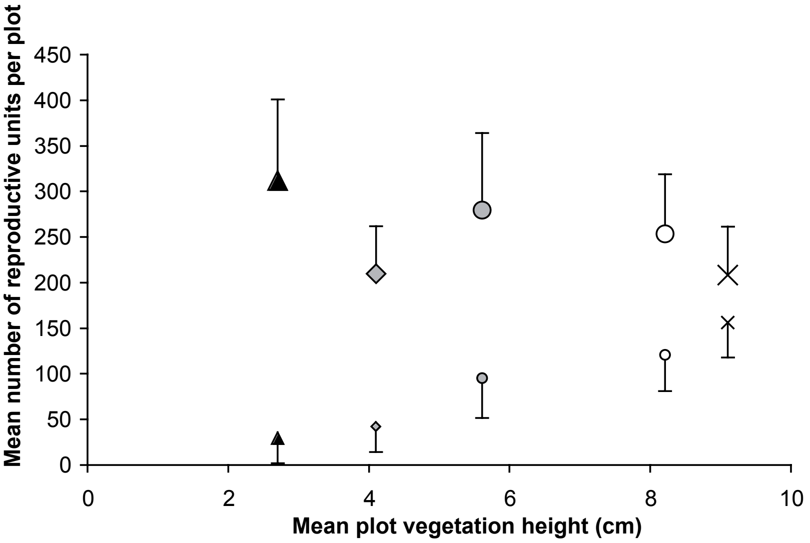

Fig. 4. Mean (S.D.) number of reproductive units per 50 x 50cm plot in four habitat structures (symbols as in Figure 2) in relation to mean plot vegetation height in one semi-natural pasture. Small symbols show number of reproductive units in mid-August, large symbols the total number of reproductive units from mid-May to mid-August. N = 10 plots per structure, see text for explanation.

associated with junipers and roses at the top, and species associated with grazing rejects and grazed patches at the bottom. The environmental variables pine, birch, juniper, rose, grazed patch, and vegetation height significantly influenced species composition (Monte Carlo test, p≤0.001), but not dung pat and grazing reject (both p≥0.05). All sites except Årby and Lagga significantly influenced species composition (Monte Carlo test, p≤0.001, Fig. 5).

A number of species were found to be more abundant near junipers and roses, for example *Anthoxanthum odoratum*, *Campanula rotundifolia*, *Campanula persicifolia*, *Festuca ovina*, *Fragaria vesca*, *Galium boreale*, *Lathyrus linifolia*, *Pilosella officinarum*, *Potentilla erecta*, *Veronica officinalis*, *Vicia sepium* and *Viola* sp. (Fig. 6). Abundance of *Elytrigia repens*, *Alopecurus pratensis* and *Stellaria gramminea* were associated with birch and pine. *Achillea millefolium*, *Cerastium fontanum*, *Leontodon autumnalis*, *Festuca rubra*, *F. pratense*, *Phleum pratense*, *Potentilla reptans*, *Taraxacum* sp. and *Trifolium repens* were associated with the open grass sward, without shrubs and trees. No differences in species composition between grazed patches and rejects could be detected in the RDA diagram.

Discussion

Although several environmental factors may affect the spatiotemporal variation and overall production of flowers, fruits and seeds in semi-natural pastures, this study indicates that grazing of reproductive parts is one of the most important factors. Up to 85% of all plant reproductive units were eaten before fruit maturation, a result that is confirmed by other studies in semi-natural grasslands (Wissman, 2006). The risk of being

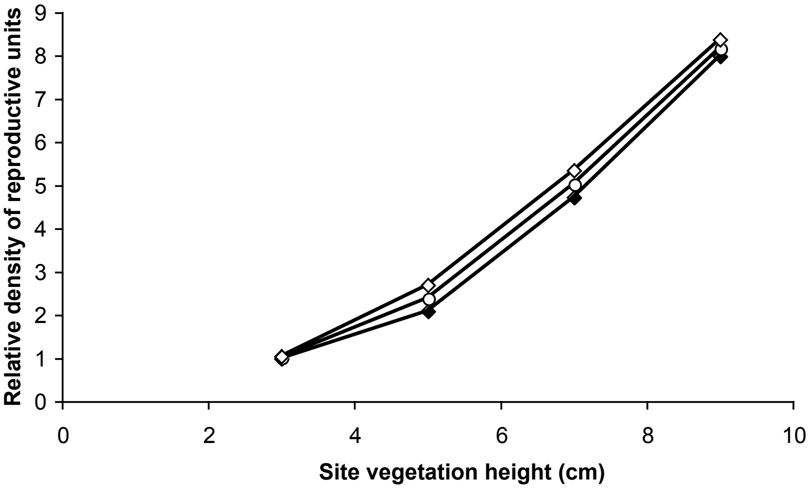

Fig. 5. Simulation of the relative density of reproductive units in relation to grazing intensity (site vegetation height in August) in three model pastures: without shrubs (filled diamonds), and with 20 per cent (open circles) and 40 per cent (open diamonds) cover of grass sward within 0.5 m from shrubs. Simulation of the pasture without shrubs is based on eq. 1 and of pastures with shrubs on eq. 2.

grazed before fruit maturation varied spatially in the pastures, and the spatial patterns of vegetation height and plant reproduction were strongly correlated (cf. Jerling & Andersson, 1982, Hickman & Hartnett, 2002, Pöyrö *et al.*, 2006). The variation was caused by the grazers' foraging behaviour, which created a number of discrete vegetation structures. Shrubs, but not trees, functioned as partial grazing refuges thus forming patches with taller vegetation and higher density of plant reproductive units. Moreover, in areas without shrubs, a mosaic of grazed patches and rejects was formed, partly due to dung deposition. The density of reproductive units in August was on average six times higher in grazing rejects than in grazed patches, four times higher close to dung pats, and three times higher around junipers and roses. Although both dung and shrubs can be assumed to increase soil nutrient levels (Moro *et al.*, 1997, El Bana *et al.*, 2002) and thus the production of reproductive units, any such effects were hidden by strong grazing effects. Grazing decreased August density of reproductive units while increasing the total production of reproductive units, probably by trigging compensatory growth (Belsky, 1986).

Variation in vegetation height and density of reproductive units among structures varied between pastures largely depending on grazing intensity in the pasture. The effects of grazing intensity can be decomposed into three components. First, grazing intensity affected the proportion of grazed and ungrazed patches in the vegetation mosaic. The proportion of ungrazed rejects in August varied from zero in the most intensely grazed pastures in the study, to about 50% area cover in the weakest grazed pasture. Thus, in the studied grazing intensity gradient sward heterogeneity was highest under weak grazing (Jerling & Andersson, 1982; Rook *et al.*, 2004). Grazing rejects may be formed because the grazers avoid areas with lower nutrient value (Bailey *et al.*, 1998), or areas with unpalatable species (Olff & Ritchie, 1998; Rousset & Lepart,

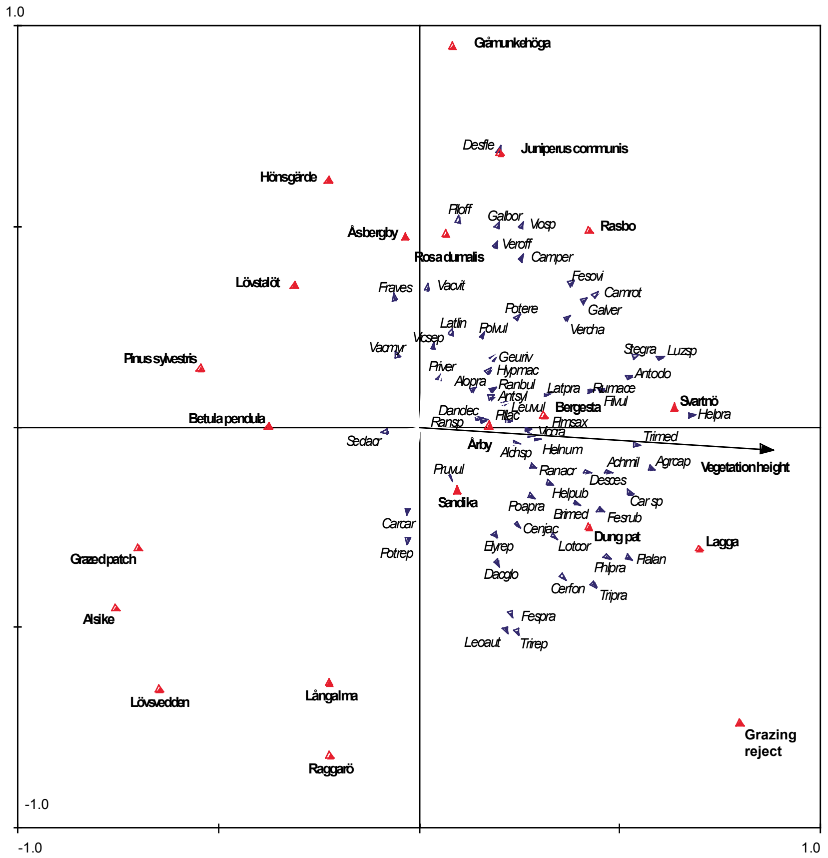

Fig. 6. RDA ordination diagram based on abundance of reproductive units of 64 vascular plant species in 14 semi-natural pastures, each with 7 spatial habitat structures (see text for explanation). Species abbreviations refer to the three first letters of the genus and the species name. See Appendix 1 for full species names.

2003). In this study, however, rejects and grazed patches did not differ in terms of species composition. Other studies in Swedish pastures have shown that rejects are rarely situated on the same places during two consecutive years (Brunsell, 2002), and we therefore suspect that rejects in the studied grassland are mainly caused by more or less random grazing pattern over one grazing season.

Second, grazing intensity affected the proportion of fertile shoots that escaped grazing in grazed patches. The average August density of reproductive units in grazed patches varied from about 10 fertile units per 0.25 m^2 in intensely grazed pastures to about 40 fertile units in pastures with weak grazing. In grazing rejects the density of reproductive units was per definition unaffected by grazing, but varied from about 60 to 150 units per 0.25 m^2 between grasslands, probably depending on species composition and slight differences in productivity. Controlling for such differences by using reproduction relative to that in ungrazed rejects showed that the density of reproductive units in grazed patches varied between 0.1 and 0.4 of the density in rejects, depending on grazing intensity in the pasture.

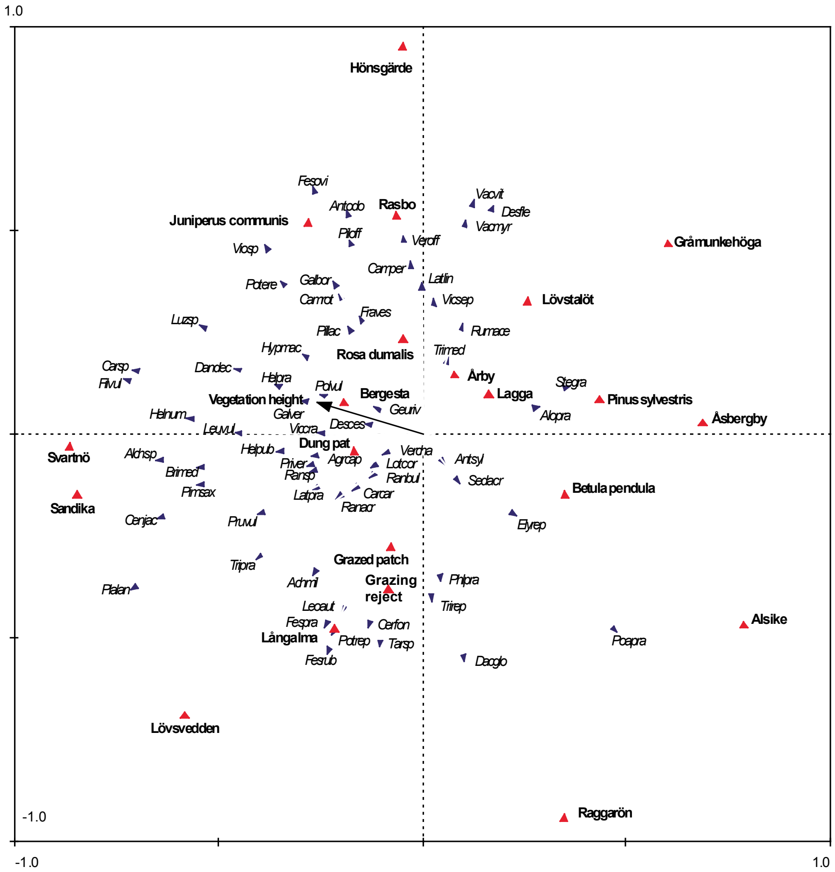

Fig. 7. RDA ordination diagram based on abundance of 64 vascular plant species in 14 semi-natural pastures, each with 7 spatial habitat structures (see text for explanation). Species abbreviations refer to the three first letters of the genus and the species name. See Appendix 1 for full species names.

Third, grazing intensity affected the size and efficiency of grazing refuges around spatial structures such as shrubs and dung pats. The density of reproductive units in refuges next to dung pats was 0.4 of that in grazing rejects in intensely grazed, compared to 0.8 in weakly grazed pastures. The corresponding figures for shrubs (average of junipers and roses) were 0.1 and 1.0. Dung pats constitute grazing refuges that are persistent during at least one summer (Brunsell, 2002). In this study, cover of dung pat refuges was not correlated with grazing intensity, but varied between 5% and 20% independently of grazing. Since density of dung pats can be expected to be a function of cattle density (Bakker, 1989), this lack of relationship may be due to small sampling area.

Grazing intensity had considerably stronger effect on a pasture's production of flowers, fruits and seeds than presence of shrubs. Simulations showed that reduced grazing intensity from 3 cm to 9 cm vegetation height in August resulted in seven times higher density of flowers and fruits in the pasture, mainly because the cover of grazing rejects increased from 0 to about 0.5, but also because four times more reproductive shoots escaped grazing in grazed patches and around dung pats. Presence of shrubs further increased the overall density of reproductive units in the pasture, but only about 15% at the most, at a 20% cover of grass sward close to (within 50 cm of) shrubs. The relative importance of shrubs for plant reproduction was highest at 5 cm grazing intensity. Under weaker grazing a larger proportion of the reproductive units are produced in rejects, dung pats and grazed patches, whereas under more intense grazing even the vegetation around shrubs is grazed. As a result, a 20% shrub cover contributes with only 2% increase of the overall density of reproductive units, both at intense and weak grazing. It should be noted that the simulation estimates plant reproduction in the pasture's area of grass sward, irrespective of the area covered by the shrubs themselves.

Species composition varied between the persistent habitat structures tree, shrub, and grazed patch, but was not affected by the temporary structures dung pat and grazing reject. This result corresponds with other studies in Scandinavian semi-natural grasslands (Rejmanek & Rosén, 1992; Austad & Losvik, 1998). Moreover, plot vegetation height was correlated with species composition, but vegetation height covaried with structure. About 10 grass sward plant species showed an association with shrubs whereas about 10 species occurred mainly in the open grassland, i.e. in the structures grazed patch, reject and dung pat. Production of flowers and fruits, in contrast, was not associated with grazed patches for any of the species, but was strongly associated with grazing rejects, dung pats and shrubs. Only three species showed an abundance association with trees, but reproduction was not associated with trees for any species. Variation in plant species composition and plant reproduction was best explained by study site. Study site includes many different factors that influence plant composition such as historical land-use (Dahlström *et al.*, 2006), historical connectivity (Lindborg & Eriksson, 2004), surrounding landscape (Cousins, 2006), soil conditions (Znamenskiy *et al.*, 2006), geographical differences, which were not measured in this study. Moreover, the variation in plant reproduction between sites is due to the gradient in grazing intensity between the sites.

Implications for conservation

The study shows that adjustment of grazing intensity is the most efficient tool for regulating the resources of nectar, pollen, flowers, fruits and seeds. The density of reproductive units roughly doubled for every 2 cm taller mean vegetation in August. Intensely grazed pastures showed low vegetation heterogeneity since no grazing rejects occurred. Moreover an ungrazed pasture without grazed patches would have reduced vegetation heterogeneity compared to a grazing intensity that creates a mosaic of grazed and ungrazed patches. This corresponds with the general idea of intermediate disturbance creating the highest habitat heterogeneity (Connell, 1978). In the studied pastures this heterogeneity maximum (50% grazing rejects) was reached under the weakest grazing intensity (9 cm vegetation height) in the studied intensity gradient. An August vegetation height of 8 cm indicates unusually weak grazing of Swedish dry-mesic pastures, whereas 3-5 cm is common, and has even been a recommended grazing intensity for grasslands subject to EU management subsidiaries (Overud & Lennartsson, 2004). Thus, from a heterogeneity perspective, this study indicates that the normal grazing intensity in the region's pastures is rather intense.

A 20% cover of vegetation close

to shrubs increased the floral resource by a maximum of 15%. However, the corresponding effect of shrubs on organisms depending on floral resources is more difficult to quantify. For insects depending on pollen or nectar, for example, the resource increase may be about 15% since it does not depend on which structures the resources are found. For sedentary organisms such as phytophages and seed predators, on the other hand, shrubs and other persistent structures may be quantitatively more important than these structures' contribution to the production of floral resources. Such organisms may select plants close to shrubs since the location of rejects and dung pats cannot be predicted at oviposition in the early summer.

For fruit phytophages and seed predators in particular, only mature reproductive units constitute the resource. Thus, density of reproductive units in August may be a proper estimate of a pasture's quality for these organisms, whereas density earlier in the summer, or the total production of reproductive units, is a less relevant estimate. For example, even if the production of buds was high in grazed patches, only 15% reached flower stage before being eaten compared to 80% in dung pats.

In Scandinavia, abundance of shrubs in semi-natural grasslands is often discussed in connection with restoration and management. EU management subsidiaries for high nature value farmland are in Sweden usually accompanied by management directives regarding, for example, grazing intensity, timing of management and prescriptions for increment of grass-sward area and quality by removal of bushes and trees (Jordbruksdepartementet, 2000; Overud & Lennartsson, 2004). Such recommendation may, however, adversely affect grassland biodiversity, unless the importance of spatial variation in different types of pastures is considered.

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References

Amiotti, N. M., Zalba, P., Sanchez, L. F. & Peinemann, N. 2000. The impact of single trees on properties of loess-derived grassland soils in Argentina. *Ecology 81*, 3283-3290.

Austad, I. & Losvik, M. H. 1998. Changes in species composition following field and tree layer restoration and management in a wooded hay meadow. *Nordic Journal of Botany 18*, 641-662.

Bailey, D. W., Dumont, B. & Wallis DeVries, M. F. 1998. Utilization of heterogeneous grasslands by domestic herbivores: Theory to management. *Annales de Zootechnie 47*, 321-333.

Bakker, J. P. 1989. *Nature Management by Grazing and Cutting*. Kluwer Academic Publishers, Dordrecht.

Bakker, E. S., Olff, H., Vandenberghe, C., de Maeyer, K., Smit, R., Gleichman, J. M. & Vera, F. W. M. 2004. Ecological anachronisms in the recruitment of temperate light-demanding tree species in wooded pastures. *Journal of Applied Ecology 41*, 571-582.

Barbaro, L., Dutoit, T. & Cozic, P. 2001. A six-year experimental restoration of biodiversity by shrub-clearing and grazing in calcareous grasslands of the French Prealps. *Biodiversity and Conservation 10*, 119-135.

Belsky, A. J. 1986. Does herbivory benefit plants? A review of the evidence. *American Naturalist* 127, 870-892.

Borcard, D., Legendre, P. & Drapeau, P. 1992. Partialling Out the Spatial Component of Ecological Variation. *Ecology 73*, 1045-1055.

Bossuyt, B., De Fre, B. & Hoffmann, M. 2005. Abundance and flowering success patterns in a short-term grazed grassland: early evidence of facilitation. *Journal of Ecology 93*, 1104-1114.

Brunsell, M. 2002 Betesrator och kärlväxters reproduktion: En studie av betesmarkers heterogenitet i tid och rum. Examensarbete i Naturvårdsbiologi 72, Uppsala.

Bullock, J. M. & Marriot, C. A. 2000. *Plant responses to grazing and opportunities for manipulation*. In: Grazing Management. (Eds. A. J. Rook & P. D. Penning). British Grassland Society, pp. 17-26.

Callaway, R. M., Kikvidze, Z. & Kikodze, D. 2000. Facilitation by unpalatable weeds may conserve plant diversity in overgrazed meadows in the Caucasus Mountains. *Oikos 89*, 275-282.

Chambers, J. C. 2001. *Pinus monophylla* establishment in an expanding *Pinus-Juniperus* woodland: Environmental conditions, facilitation and interacting factors. *Journal of Vegetation Science 12*, 27-40.

Connell J. H. 1978. Diversity in tropical rain forests and coral reefs. *Science 199*, 1302-1310.

Correll O., Isselstein J. & Pavlu V. 2003. Studying spatial and temporal dynamics of sward structure at low stocking densities: the use of an extended rising-plate-meter method. *Grass Forage Science 58*, 450-454.

Cousins, S. A. O. 2006. Plant species richness in midfield islets and road verges - The effect of landscape fragmentation. *Biological Conservation 127*, 500-509.

Dahlgren, R. A., Singer, M. J. & Huang, X. 1997. Oak tree and grazing impacts on soil properties and nutrients in a California oak woodland. *Biogeochemistry 39*, 45-64.

Dahlström, A., Cousins, S. A. O. & Eriksson, O. 2006. The History (1620-2003) of Land Use, People and Livestock, and the Relationship to Present Plant Species Diversity in a Rural Landscape in Sweden. *Environment and History 12*, 191-212.

El Bana, M. I., Nijs, I. & Kockelbergh, F. 2002. Microenvironmental and vegetational heterogeneity induced by phytogenic nebkhas in an arid coastal ecosystem. *Plant and Soil 247*, 283-293.

Fowler, J., Cohen, L. & Jarvis, P. 1998. *Practical statistics for field biology,* 2nd edition. John Wiley and Sons, New York.

Gärdenfors, U. (ed.) 2005. Rödlistade arter i Sverige 2005 - The 2005 Red List of Swedish Species. ArtDatabanken, SLU, Uppsala.

Hansson, M. & Fogelfors, H. 2000. Management of a semi-natural grassland; results from a 15-year-old experiment in southern Sweden. *Journal of Vegetation Science 11*, 31-38.

Hickman, K. R. & Hartnett, D. C. 2002. Effects of grazing intensity on growth, reproduction, and abundance of three palatable forbs in Kansas tallgrass prairie. *Plant Ecology 159*, 23-33.

Hodgson, J. G., Grime, J. P., Wilson, P. J., Thompson, K. & Band, S. R. 2005. The impacts of agricultural change (1963-2003) on the grassland flora of Central England: processes and prospects. *Basic and Applied Ecology 6*, 107-118.

Ihse, M. 1995. Swedish agricultural landscapes - patterns and changes during the last 50 years, studied by aerial photos. *Landscape and Urban Planning 31*, 21-37.

Jerling, L. & Andersson, M. 1982. Effects of selective grazing by cattle on the reproduction of Plantago maritima. *Holarctic ecology 5*, 405-411.

Jones, R. M. & Ratcliff, D. 1983. Patchy grazing and its relation to deposition of cattle dung pats in pastures in coastal subtropical Queensland. *The Journal of the Australian Institute of Agricultural Science 1*, 109-111.

Jordbruksdepartementet 2000. Miljö- och landsbygdsprogram för Sverige. År 2000-2006. Regeringskansliet. Jordbruksdepartementet.

Kohler, F., Gillet, F., Reust, S., Wagner, H. H., Gadallah, F., Gobat, J. M. & Buttler, A. 2006. Spatial and seasonal patterns of cattle habitat use in a mountain wooded pasture. *Landscape Ecology 21*, 281-295.

Kull, K. & Zobel, M. 1991. High Species Richness in An Estonian Wooded Meadow. *Journal of Vegetation Science 2*, 715-718.

Leps, J. & Smilauer, P. 2003. Multivariate Analysis of Ecological Data using CANOCO. Cambridge University Press.

Lindborg, R. & Eriksson, O. 2004. Historical landscape connectivity affects present plant species diversity. *Ecology 85*, 1840-1845.

Marion, C., & Houle, G. 1996. No differential consequences of reproduction according to sex in Juniperus communis var depressa (Cupressaceae). *American Journal of Botany 83*, 480-488.

Milchunas, D. G. & Noy-Meir, I. 2002. Grazing refuges, external avoidance of herbivory and plant diversity. *Oikos* 99, 113-130.

Moro, M. J., Pugnaire, F. I., Haase, P. & Puigdefabregas, J. 1997. Mechanisms of interaction between a leguminous shrub and its understorey in a semi-arid environment. *Ecography 20*, 175-184.

Morris, M. G. 1967. Differences between the invertebrate faunas of grazed and ungrazed chalk grassland I. Responses of some phytophagous insects to cessation of grazing. *Journal of Applied Ecology 36*, 459-474.

Mortimer, S., Hollier, J. & Brown, V. K. 1998. Interactions between plant and insect diversity in the restoration of lowland calcareous grasslands in southern Britain. *Applied Vegetation Science 1*, 101-114.

Olff, H. & Ritchie M. E. 1998. Effects of herbivores on grassland plant diversity. *Trends in Ecology & Evolution 13*, 261-265.

Overud S. & Lennartsson T. 2004. Skötsel och restaurering av betesmarker och slåtterängar. En sammanställning av den regionala naturvårdens kunskaper och erfarenheter. Jordbruksverket, Naturvårdsverket, Riksantikvarieämbetet. Jordbruksverket Rapport 2004:11.

Peterson, A. 2005. Has the generalisation regarding conservation of trees and shrubs in Swedish agricultural landscapes gone too far? *Landscape and Urban Planning 70*, 97-109.

Påhlsson, L. (ed.) 1994. *Vegetationstyper i Norden. (Nordic Vegetation Types).* TemaNord 1994: 665.

Pärtel M. & Zobel M. 1999. Small-scale plant species richness in calcareous grasslands determined by the species pool, community age and shoot density. *Ecography 2*, 153-59.

Pöyry, J., Luoto, M., Paukkunen, J., Pykala, J., Raatikainen, K. & Kuussaari, M. 2006. Different responses of plants and herbivore insects to a gradient of vegetation height: an indicator of the vertebrate grazing intensity and successional age. *Oikos 115*, 401-412.

Rejmánek, M. & Rosén, E. 1992. Influence of colonizing shrubs on species-area relationships in alvar plant communities. *Journal of Vegetation Science 3*, 625-630.

Reyes-Lopez, J., Ruiz, N. & Fernandez-Haeger, J. 2003. Community structure of ground-ants: the role of single trees in a Mediterranean pastureland. *Acta Oecologica-International Journal of Ecology 24*, 195-202.

Rook, A. J., Dumont, B., Isselstein, J., Osoro, K., WallisDeVries, M. F., Parente, G. & Mills, J. 2004. Matching type of livestock to desired biodiversity outcomes in pastures - a review. *Biological Conservation 119*, 137-150.

Rosén, E. & Bakker, J. P. 2005. Effects of agri-environment schemes on scrub clearance, livestock grazing and plant diversity in a low-intensity farming system on Öland, Sweden. *Basic and Applied Ecology 6*, 195-204.

Rousset, O. & Lepart, J. 2003. Neighbourhood effects on the risk of an unpalatable plant being grazed. *Plant Ecology 165*, 197-206.

Rozas, V. 2005. Dendrochronology of pedunculate oak (*Quercus robur* L.) in an old-growth pollarded woodland in northern Spain: tree-ring growth responses to climate. *Annals of Forest Science 62*, 209-218.

Shiyomi, M., Okada, M., Takahashi, S. & Tang, Y. H. 1998. Spatial pattern changes in aboveground plant biomass in a grazing pasture. *Ecological Research 13*, 313-322.

Söderström, B., Svensson, B., Vessby, K. & Glimskär, A. 2001. Plants, insects and birds in semi-natural pastures in relation to local habitat and landscape factors. *Biodiversity and Conservation 10*, 1839-1863.

ter Braak C.J.F. & Smilauer P. 2002. CANOCO Reference manual and CanoDraw for Windows User's guide: Software for Canonical Community Ordination (version 4.5). Microcomputer Power (Ithaca, NY, USA), 500p.

van Auken, O. W., Jackson, J. T. & Jurena, P. N. 2004. Survival and growth of Juniperus seedlings in Juniperus woodlands. *Plant Ecology 175*, 245-257.

Vera, F. W. M. 2000. *Grazing Ecology and Forest History*. CABI Publishing.

White, S. L., Sheffield, R. E., Washburn, S. P., King, L. D. & Green, J. T. 2001. Spatial and time distribution of dairy cattle excreta in an intensive pasture system. *Journal of Environmental Quality 30*, 2180-2187.

Willems, J. H. 2001. Problems, approaches, and, results in restoration of Dutch calcareous grassland during the last 30 years. *Restoration Ecology 9*, 147-154.

Willems, J. H. & Bik L. P. M. 2006. Restoration of high species density in calcareous grassland: the role of seed rain and soil seed bank. *Applied Vegetation Science 1*, 91-100.

Wissman, J. 2006. Grazing Regimes and Plant Reproduction in Semi-Natural Grasslands. Swedish University of Agricultural Sciences.

Znamenskiy, S., Helm, A. & Pärtel, M. 2006. Threatened alvar grasslands in NW Russia and their relationship to alvars in Estonia. *Biodiversity and Conservation 15*, 1797-1809.

Appendix 1. The 64 species (≥10 plots) used in the multivariate analyses. The total number of plots (abundance) and total number of fertile shoots are shown for each species. Underlining refers to abbreviation in Fig. 6 and 7.

Species	Abundance	Fertile shoots
Achillea millefolium	840	498
Agrostis capillaris	1023	7746
Alchemilla sp.	188	1019
Alopecurus pratensis	22	64
Anthoxanthum odoratum	136	1420
Anthriscus sylvestris	26	52
Briza media	69	509
Campanula persicifolia	39	197
Campanula rotundifolia	145	3181
Carex sp.	292	433
Carum carvi	27	161
Centaurea jacea	81	526
Cerastium fontanum	66	2236
Dactylis glomerata	227	625
Danthonia decumbens	39	476
Deschampsia cespitosa	66	298
Deschampsia flexuosa	154	1988
Elytrigia repens	46	127
Festuca ovina	391	6768
Festuca pratensis	166	525
Festuca rubra	741	1903
Filipendula vulgaris	304	338
Fragaria vesca	176	168
Galium boreale	209	209
Galium verum	501	1317
Geum rivale	18	65
Helianthemum nummularium	38	688
Helictotricon pratense	139	896
Helictotricon pubescens	139	311
Hypericum maculatum	24	577
Lathyrus linifolia	32	35
Lathyrus pratensis	103	57
Leontodon autumnalis	52	1357
Leucanthemum vulgare	29	48
Lotus corniculatus	53	725
Luzula sp.	307	401
Phleum pratense	54	358
Pilosella lactucella	25	17
Pilosella officinarum	181	514
Pimpinella saxifraga	95	424
Plantago lanceolata	175	985
Poa pratensis	779	2011
Polygala vulgaris	30	838
Potentilla erecta	81	2523
Potentilla reptans	83	190
Primula veris	34	580
Prunella vulgaris	52	594
Ranunculus acris	80	410
Ranunculus bulbosus	44	56
Ranunculus sp.	74	38
Rumex acetosa	128	109
Sedum acre	11	72
Stellaria graminea	388	20749
Taraxacum sp.	114	0
Trifolium medium	236	1434
Trifolium pratense	221	2097
Trifolium repens	703	4165
Vaccinium myrtillus	41	9
Vaccinium vitis-idaea	33	93
Veronica chamaedrys	474	1656
Veronica officinalis	105	506
Vicia cracca	40	68
Vicia sepium	16	32
Viola sp.	105	355
Sum fertile shoots		78827

Shrub effects on herbs and grasses in semi-natural grasslands - positive, negative or neutral relationships?

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Key Words Functional traits; Grazing; Grazing refuge; Reproduction; *Rosa dumalis*; Seedling establishment

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Abstract

This study investigated how the abundance and reproduction of herbs and grasses relates to presence of *Rosa dumalis* shrubs in three semi-natural pastures in Sweden. Shrubs may affect grassland plants negatively, e.g. by competition, positively, e.g. by serving as grazing refuge, or neutrally. At different distances from *R. dumalis* shrubs, data were collected on abundance and frequency of reproductive shoots of all plant species, and on vegetation height and litter depth. In one grassland, data were collected on seedling density and the frequency of reproductive shoots in presence and absence of grazing. The shrubs functioned as grazing refuges with taller vegetation, deeper litter and higher probability of plant reproduction. The overall number of plant species remained the same at all distances from shrubs. Most species showed a neutral relationship with shrubs. Between 8 and 26% of the species showed a negative pattern to shrubs and 14-30% a positive pattern. Seedling density was negatively correlated with litter depth but peaked at 60-90 cm from shrubs. Establishment of seedlings of small-seeded species was negatively related to shrubs due to thicker litter layer close to shrubs. The observed patterns were compared with different functional traits, such as Ellenberg values, plant height, growth form and Raunkiaer life form. Plant height (from literature) was the trait that best explained plant species' relation to shrubs because tall species were more common close to shrubs. Shrubs increase the heterogeneity in grasslands and intensive shrub clearing may negatively affect biodiversity.

Introduction

Conservation of semi-natural grasslands is of major importance for the conservation of European species and habitats (Rook *et al.*, 2004), due to the rapid decline of these habitats in Europe during the last century (Ihse, 1995; Stanners & Bordeau, 1995), their exceptional species richness (Pärtel & Zobel, 1999; Weibull & Östman, 2003), and the high numbers of red-listed species (Gärdenfors, 2005). Considerable financial resources are allocated from European subsidiary programmes to management of the remaining grassland areas (Kleijn & Sutherland, 2003). Financial support in Sweden is usually accompanied by management directives regarding, for example, grazing intensity, timing of management and prescriptions for increment of grass-sward area and quality by removal of bushes and trees (Regeringskansliet, 2000; Overud & Lennartsson, 2004). Abundance of shrubs in semi-natural grasslands is thus discussed in connection with restoration and management. In contrast to considering shrubs as competitors to the grassland organisms, shrubs may also be viewed as elements increasing the grassland heterogeneity and thereby the diversity of plants, insects and birds (Söderström *et al.*, 2001; Lindborg & Eriksson, 2004).

In addition to these practical aspects of shrubs in grasslands, the relationships between shrubs and grassland plants are also subject to conceptual discussions. Plant community structure and plant interactions may be described by two alternative conceptual models, namely competitional and associational responses, respectively (Callaway, 1995). Models based on competition for resources assume that interactions between species are mainly negative (Brooker *et al.*, 2005), whereas associational responses (Tahvanainen & Root, 1972), or facilitation (Connell & Slatyer, 1977), describe how species are favoured by growing close to other species (Callaway, 1995). For example, shrubs, spiny species, or toxic plants can provide protection from grazing (Rebollo et al., 2002). Several studies have shown that grazing-sensitive plants can gain shelter from herbivory by growing close to spiny or unpalatable plants (Callaway, 1995; Hjälten & Price, 1997; Olff & Ritchie, 1998; Olff *et al.*, 1999; Callaway *et al.*, 2000; Rebollo *et al.*, 2002; Rousset & Lepart, 2003). Shrubs and other deep-rooted plants may also increase nutrient availability for the grass sward, by transferring nutrients from deeper soils to the surface through the leaf litter (Moro *et al.*, 1997; Austad & Losvik, 1998; El Bana *et al.*, 2002). Woody shrubs in arid and semi-arid environments can accumulate wind-borne sediments around their canopy and modify microclimate and soil nutrients (Moro *et al.*, 1997; El Bana *et al.*, 2002; Facelli & Temby, 2002). In temperate areas, tree canopies in wooded hay-meadows can lower temperature and sun radiation compared with open areas (Austad & Losvik, 1998).

The conditions for herbs and grasses may be less favourable close to shrubs than in the open grassland because of deeper litter layer, caused by leaf deposition and reduced grazing intensity (Jensen & Gutekunst, 2003), and reduced availability of light (Rejmánek & Rosén, 1992; Einarsson & Milberg, 1999). If abundant, shrubs may also cause local fragmentation of the grassland, which may result in reduced pollination of grassland herbs (Lennartsson, 2002).

Different plant species may be affected differently by shrubs. Short plants can suffer from light competition close to shrubs (Drews *et al.*, 2004) and can be more grazing resistant than tall species (Diaz *et al.*, 2001) and may therefore be more abundant in the open grassland.

Tall species, in contrast, can be expected to be more common closer to shrubs because they compete better for light and are often sensitive to grazing (Diaz *et al*., 2001). Taxonomic groups, such as monocotyledons and dicotyledons, and life forms (Raunkiaer) respond differently to grazing disturbance (Dupré & Diekmann, 2001, McIntyre *et al*., 1995) and can therefore be expected to react differently to shrubs, if the shrub affects grazing intensity. Plant species with different Ellenberg values (Ellenberg *et al*., 1991) respond differently to grazing; for example, species indicating high light intensities and low soil moisture can increase by grazing and species indicating high nitrogen levels can decrease by grazing (Pykälä, 2005) and may therefore be differently influenced by shrubs.

Shrubs in pastures may also differently affect different plant life stages. For example, seed production may positively respond to shrubs because of protection from grazing (Rebollo *et al*., 2002), whereas seedling establishment may negatively respond due to weaker grazing pressure and deeper litter layer close to shrubs (Lennartsson & Oostermeijer, 2001).

Thus, the response of a plant species will be the combined effect of responses of different life stages, and the responses of different species will exert a combined effect on the plant community. In both cases, the observed relationships between shrubs and grass sward plants may be positive, negative or neutral.

To study grassland plants and their relation to shrubs, *Rosa dumalis* was chosen because it is a common spiny shrub in Swedish grasslands and because it may function both as a grazing refuge and as a competitor. By using field observations and a field experiment with cages we evaluated possible negative and positive relationships between *R. dumalis* shrubs and grasses and herbs to answer the following questions: (1) Is plant species richness related to shrubs in a negative, positive or neutral manner? (2) Does the relationship between plant abundance and distance to shrub differ between plant species and can species response be attributed to different functional traits such as plant height, Ellenberg value, Raunkiaer life form, growth form, taxonomic group, and seed weight? (3) Is plant reproduction and seedling density related to distance to shrubs in a positive, negative or neutral manner? (4) Can the pattern of seedling density and reproduction be explained by the shrub functioning as grazing refuge?

Methods

Study system

The study was performed in three semi-natural pastures in the county of Uppland in central Sweden: Focksta (9 hectares, N59°47′, E17°23′), Stammen (6 hectares, N59°44′, E17°55′) and Åsbergby (17 hectares, N59°44′, E17°55′). All three pastures are situated between forest and arable fields, on sandy gravel. They have a long grazing history and are all assigned as pastures on historical maps from 1850-60. Shrubs of *Juniperus communis, Prunus spinosa* and *Rosa dumalis* are scattered in all three grasslands and the vegetation can be characterised as dry-mesic herb-rich *Agrostis capillaris-Festuca ovina* vegetation (Söderström, 1993; Påhlsson, 1994). Other common herbs and grasses were *Achillea millefolium*, *Festuca rubra*, *Filipendula vulgaris*, *Galium verum*, *Helictotrichon pratense* and *Trifolium repens*. All pastures were grazed by cattle from May to September.

R. dumalis is a multi-stemmed shrub, normally of 1-2 m high. Stems and branches are covered with sharp, hooked spines. Once established, the shrub can expand radially by rhizomes.

The three pastures were examined in August but in different years, Focksta in 2003, Stammen in 2001 and Åsbergby in 2002. Stammen was originally part of another study, with a slightly different sampling design. Except for the RDA analysis (see below) all analyses were performed for each pasture separately.

Data sampling

Solitary *R. dumalis* thickets were randomly chosen, of ca. 1.5 m high and minimum 1 m in diameter, and growing in areas without trees, other shrubs, or stones. Fifty-centimetre-wide transects (22 transects in Focksta, 20 in Stammen, and 22 in Åsbergby) were placed radially from the edge of the thickets into the open grassland. At selected distances from the shrub, data on presence of plant species, presence of reproductive shoots, vegetation height, and depth of the litter layer were sampled. In Focksta and Åsbergby, data were sampled in 20x50 cm plots, placed at eight distances from the shrubs (plot centre at 10, 30, 50, 70, 110, 130, 210, and 310 cm from the shrubs); in total 176 plots per grassland. In Stammen, 25x50 cm plots were placed at six distances from the shrubs (plot centre at 12.5, 37.5, 62.5, 87.5, 125, and 175 cm from the shrub edge), in total 120 plots.

At all sites presence-absence of herbs and grasses was noted in the plots, giving a frequency value of X/22 transects in Focksta and Åsbergby and X/20 in Stammen. Frequency of fertile shoots for each species was measured as presence-absence of fertile shoots only in plots with presence of the species, thus roughly estimating the probability of reproduction in August for the species. In Stammen presence-absence of fertile shoots were also counted in July. The vegetation height was measured per plot using a rising plate (Correll *et al.*, 2003). Litter depth was measured in one central point per plot by using a 0.5-cm-graded stick in Focksta and Åsbergby. In Stammen each plot was divided into ten 10x12.5 cm subplots, and the litter layer was measured in the centre of each subplot. In each subplot, the number of seedlings of herbs and sedges was also counted. Thus, for each observed seedling, an estimate of the litter layer was sampled. Of the seedlings 98% could be identified to species, and a mean litter layer per species was calculated.

In order to separate direct effects of distance to *R. dumalis* shrubs and effects of grazing, five transects in Stammen were protected from grazing using 1x2m coarse-meshed cages. The cages were set out in the spring the year before the study, to obtain an estimate of litter depth (built up by last year's vegetation) in absence of grazing. Vegetation height, litter depth, presence-absence of all plant species and of fertile shoots were measured at six distances, as described earlier for the unprotected transects in Stammen. Nomenclature follows Krok & Almqvist (2001).

Data analyses

First, to outline the general structure of species composition of the three studied grasslands a Detrended Correspondence Analysis (DCA) was carried out to determine the length of the gradient, which is a measure of unimodality of the species responses along an ordination axis (ter Braak & Smilauer, 2002). Secondly, a Redundancy Analysis (RDA) was chosen since the DCA showed that the gradient length for the ordination was less than 1.5 SD, indicating that a linear model would best fit the data. Vegetation height, litter depth and distance to shrub were used as environmental data. Study sites were entered in the analysis as dummy variables (ter Braak & Smilauer, 2002). Only data for species that occurred in more than 10 plots were used in the analysis. The tests were performed using CANOCO 4.5 (ter Braak & Smilauer, 2002).

Mean frequency of herbs and grasses, mean vegetation height, mean litter depth, and mean proportion of reproductive plots and seedling density (in Stammen) were calculated per distance. Correlations between these parameters and distance from shrub were analysed using Spearman-Rank correlation.

To obtain an estimate of each species' relation to shrubs, mean frequencies of occurrence were calculated for half of the distances close to shrubs (plot distances 0-70 cm) and for half of the distances away from shrubs (distances 87.5-310 cm). Splitting the transect at ca. 80 cm from the shrub was motivated by the fact that vegetation height and litter depth decreased with distance to shrub up to approximately 80 cm, and thereafter remained constant. Only species that occurred in at least 10 plots were used, see Appendix 1. The difference between the two distance intervals was analysed using Mann-Whitney U-test. An estimate of the relative shrub association for each species was calculated by dividing the mean frequency value of the 0-70 cm distances by the mean frequency of the 87.5-310 cm distances. Relative shrub association value >1 thus indicates that the species was more common close to shrubs. The relative shrub association values were plotted against plant height, seed weight, and Ellenberg Index for light, moisture, nitrogen and dominance (Ellenberg *et al*., 1991). Average plant height at flowering was taken from Lid (1985) and seed weight mainly taken from Müller-Schneider (1986). For some species, new data on average seed weight were collected by weighing 10-100 (depending on species) mature dry seeds per plant of 15-20 plants per species. Before weighing, the seeds were stored ca. 1 month in room temperature. The species were also categorised in four Raunkiaer life forms: chamaephytes, hemicryptophytes, geophytes and therophytes (Ellenberg *et al*., 1991) and the mean proportions of the different categories were compared for species with relative shrub association >1 and species with shrub association <1 using Mann-Whitney U-test. All species were also ranked from 1 to 4 according to different growth forms (following Lid, 1985): stem with tendrils (1), self-supporting stem (2), creeping stem (3) and rosette form (4) and correlated with relative shrub association. Statistical analyses were performed using SPSS 13.0.

Results

Species richness and vegetation patterns

The average number of vascular plant species per plot was 19±0.4 SE in Focksta (in total 74 species), 29±1.5 SE in Stammen (66 in total) and 19±0.3 SE in Åsbergby (72 in total). Species number was not correlated with distance to shrub in any of the three studied grasslands (Spearman rank correlation, $p>0.16$).

The RDA ordination diagram shows the species frequencies in relation to the three study sites and the environmental variables vegetation height, litter depth and distance to shrub (Fig. 1). In the RDA, the first three eigenvalues showed that the first axis accounted for 0.44 of the variance in species composition, the second for 0.21 and the third axis for 0.16 of the variance. The first axis is positively correlated with Stammen and vegetation height and negatively correlated with Focksta and distance. The second axis is positively correlated with Åsbergby and distance and negatively correlated with Focksta, litter depth, vegetation height and Stammen. The third axis is positively correlated with litter depth, vegetation height and Åsbergby and negatively correlated with distance to shrub and Stammen. Some species (in the RDA diagram found near the arrows for the sites) were clearly correlated with one of the sites (Fig. 1). Ten species in the upper left corner of the graph, for example *Agrostis capillaris* and *Stellaria graminea*, were positively correlated with distance to shrub, thus being more common away from shrubs. A similar number of species in the lower right corner of the diagram, for example *Campanula persicifolia* and *Helictotrichon pratense*, were negatively correlated to distance to shrub, thus showing a positive association with shrubs (Fig. 1). However, most of the species were not related to the distance to shrubs. For example, species in the upper right corner of the diagram, such as *Campanula rotundifolia* and *Luzula campestris*, were correlated with both Åsbergby and Stammen but showed no difference in abundance between plots close to and distant from shrubs.

The effects of shrubs on different species shown in the RDA were confirmed by comparing the mean frequency of each species in the proximate distance interval with the mean frequency in the distant interval. In Focksta, 12% of the species were significantly more common in the proximate interval and 30% of the species in the distant interval. Of the species 58% showed a neutral relationship with shrubs. The corresponding proportions in Stammen were 8%, 14% and 78% of the species, and in Åsbergby 26%, 24% and 50% of the species (Appendix 1). Eighteen species showed a significant response to shrubs in some grassland but not in others, but none of the species showed opposite responses in different grasslands.

Vegetation height and litter depth

In all three grasslands the vegetation was taller and the litter layer deeper close to shrubs than in the open grassland. Thus, both parameters were significantly negatively correlated with distance to shrubs (Spearman rank correlation, p<0.001 in all cases, Fig. 2). In plots protected from grazing no significant correlation with distance to shrubs was found, neither for vegetation height (r=-0.147, n=30 plots, p=0.4, Fig. 2a) nor litter depth (r=-0.278, n=30, p=0.1, Fig. 2b).

Reproduction and recruitment

The reproductive success of most plant species was correlated with distance to shrubs. The mean per species probability of reproduction decreased significantly with distance to shrub in Focksta and Åsbergby (Spearman rank correlation, p<0.001, Fig. 3). The significant correlations were mainly due to higher abundance of fertile shoots within ca. 10-40 cm from the shrubs (Fig. 3). In Stammen the probability of reproduction was significantly higher in shrubs in August (Spearman, r=0.829, n=6, p=0.042, Fig. 3) but not in July (Spearman, r=0.86, n=6, p=872, Fig. 3) indicating that plots distant to shrubs were more grazed than plots close to shrubs. In plots protected from grazing the mean probability of reproduction for each species was close to 1, with a slight but significant positive correlation with distance to shrub (Spearman, r=0.829, n=6 plots, p=0.042, Fig. 3).

The number of seedlings of herbs and sedges per plot in Stammen was significantly negatively correlated with litter depth (Spearman, r=0.621, n=120 plots, p<0.01, Fig. 4a). The mean number of seedlings did not correlate linearly with distance to shrub, but seedling density

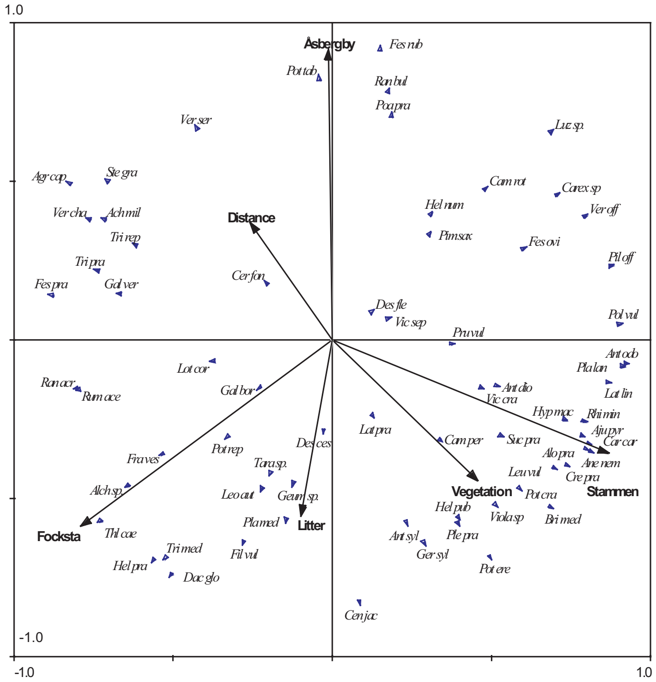

Fig. 1. Redundancy analysis (RDA) ordination diagram of plant species (triangles) in relation to three environmental variables and three study sites (arrows). The length of the arrows represents the explanatory importance of each environmental variable. The first axis is horizontal, the second is vertical. Abbreviations of species refer to underlining in the full names in Appendix 1. The environmental variables are distance to *Rosa dumalis* shrub (cm), litter depth (cm), vegetation height (cm), and the study sites are three semi-natural pastures, Focksta, Stammen and Åsbergby.

showed a distinct peak at a distance of 60-90 cm from the shrub (Fig. 4b). Seedling density at the peak (mean of distances 62.5 cm and 87.5 cm) was ca. 40% and significantly higher than distances both closer to (12.5 cm and 37.5 cm; Mann-Whitney U-test, U=434, p<0.001) and more distant from shrubs (125 cm and 175 cm; U=519, p=0.007). Mean seedling density for the distances 12.5-62.5 cm was, in contrast, not significantly different from seedling density for the distances 87.5-125 cm (U=663, p=0.2). In total 25 species of seedlings were found. Four species were more common near shrubs, one species more common distant to shrubs, and 20 species showed a neutral relationship with shrubs (Appendix 1).

Functional traits

Plant height (from literature) was positively correlated with relative shrub association values in all three grasslands Focksta (Spearman, r=0.455, n=43 species, p<0.01, Fig 5), Stammen (r=0.429, n=64, p<0.001, Fig 5) and Åsbergby (r=0.390, n=45, p<0.01, Fig. 5). In Focksta, but not in Åsbergby or Stammen, relative shrub association values were correlated with Ellenberg Index for light (Spearman, r=-0.319, n=44 species, p=0.035). The relative shrub association values were not significantly correlated with the species' Ellenberg Index for moisture, nutrients or dominance in any of the grasslands. Hemicryptophytes was the most common

Raunkiaer life form (84% of the species), and the mean proportion did not differ significantly between plant species with high shrub association value and plants with low shrub association value in any of the sites (Mann-Whitney U-test, $p>0.06$ for all sites). Moreover for growth form no correlation with shrub association values was found in any of the grasslands (Spearman Rank, $p>0.149$).

The mean frequency of grasses was significantly negatively correlated with distance to shrub in Focksta (Spearman rank correlation, $r=-0.881$, $n=8$ distances, $p=0.004$) and Åsbergby ($r=-0.929$, $n=8$, $p<0.001$) but not in Stammen ($r=-0.543$, $n=6$, $p=0.3$). The significant correlations were mainly due to higher frequencies within 0-40 cm of the shrubs. No correlation between mean frequency of herbs and distance to shrubs was found in any of the three grasslands ($p>0.16$).

Plant species with heavy seeds were able to produce seedlings in deeper litter layer

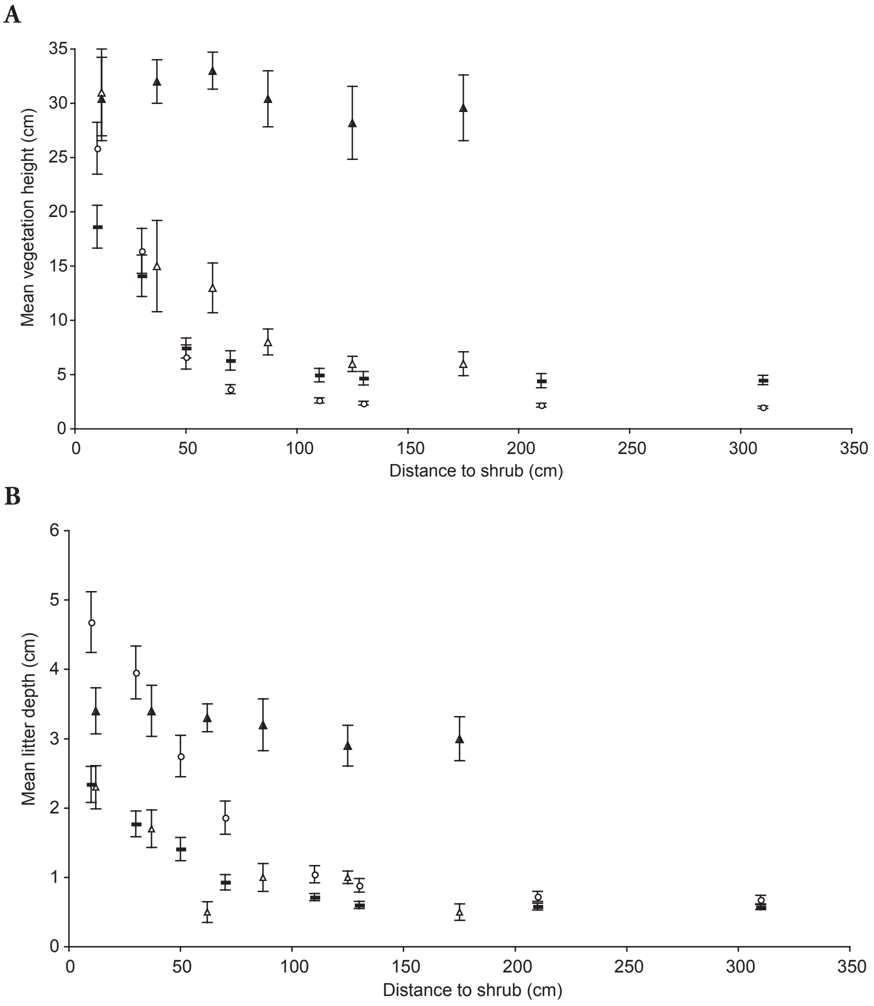

Fig. 2. Mean vegetation height (A) and mean litter depth (B) at different distances to *Rosa dumalis* shrubs, in three semi-natural pastures, Focksta (circles), Stammen (triangles) and Åsbergby (lines). Filled triangles show plots protected from grazing (Stammen). Error bars show one S.E.

than light-seeded species. The mean litter depth per seedling species (see methods) correlated strongly with the seed weight of the species (Spearman rank correlation, r=0.881, n=24 species, p<0.01, Fig. 6). The seed weight was positively correlated with relative shrub association values in Stammen (Spearman rank correlation, r=0.340, n=55 species, p=0.011), but not in Åsbergby and Focksta (p>0.7).

Discussion and conclusions

R.dumalis shrubs in semi-natural pastures did not affect the total richness of plant species, and most of the species, 62±8 % S.E., showed a neutral relationship with the shrubs in terms of abundance. Between 8 and 26% of the species showed a negative and 14-30% a positive relationship with shrubs. This indicates that the net effect of shrubs in semi-natural grassland is neutral or positive rather than negative for most plant species. The different patterns shown by different species could be attributed to a combination of at least three significant underlying mechanisms. First, the shrubs provided partial protection against grazing, which facilitated fruit production and also created taller vegetation and a deeper litter layer around the shrubs. Second, the taller vegetation, and the shrubs themselves, increased the competition for light, as indicated by the fact that positive frequency patterns were found among tall, presumably more competitive plant species, in particular grasses, but not among short species. Third, deeper litter layer around shrubs counteracted the establishment of seedlings of plant species with small seeds. Seedling density of such species showed a negative relationship with the shrubs. In one grassland, Stammen, species with heavy seeds were more often positively related to shrubs in the adult stage than species with small seeds.

In two grasslands, only relationships between species data and environmental data were analysed, but in Stammen

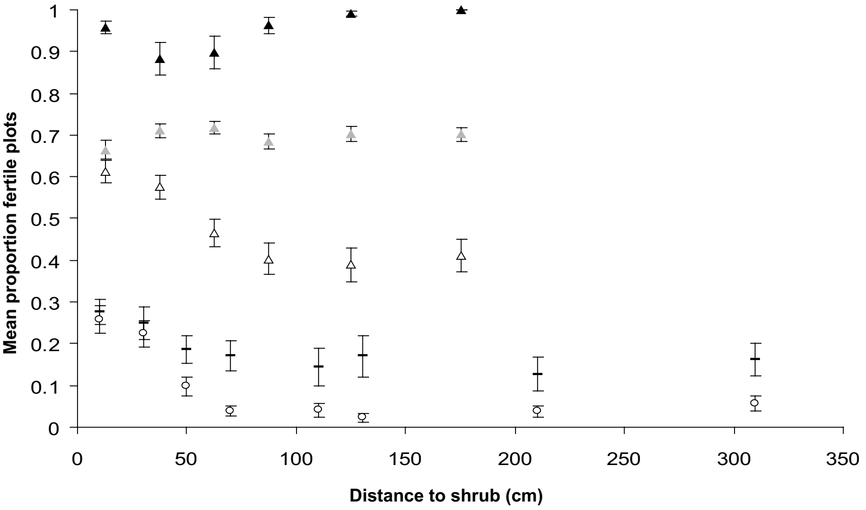

Fig. 3. Mean per species probability of reproduction (see methods) of herbs and grasses in relation to distance from *Rosa dumalis* shrubs in three grasslands, Focksta (circles), Stammen in July (grey triangles) and in August (open triangles) and Åsbergby (lines). Black triangles show plots protected from grazing (Stammen). Error bars show one S.E.

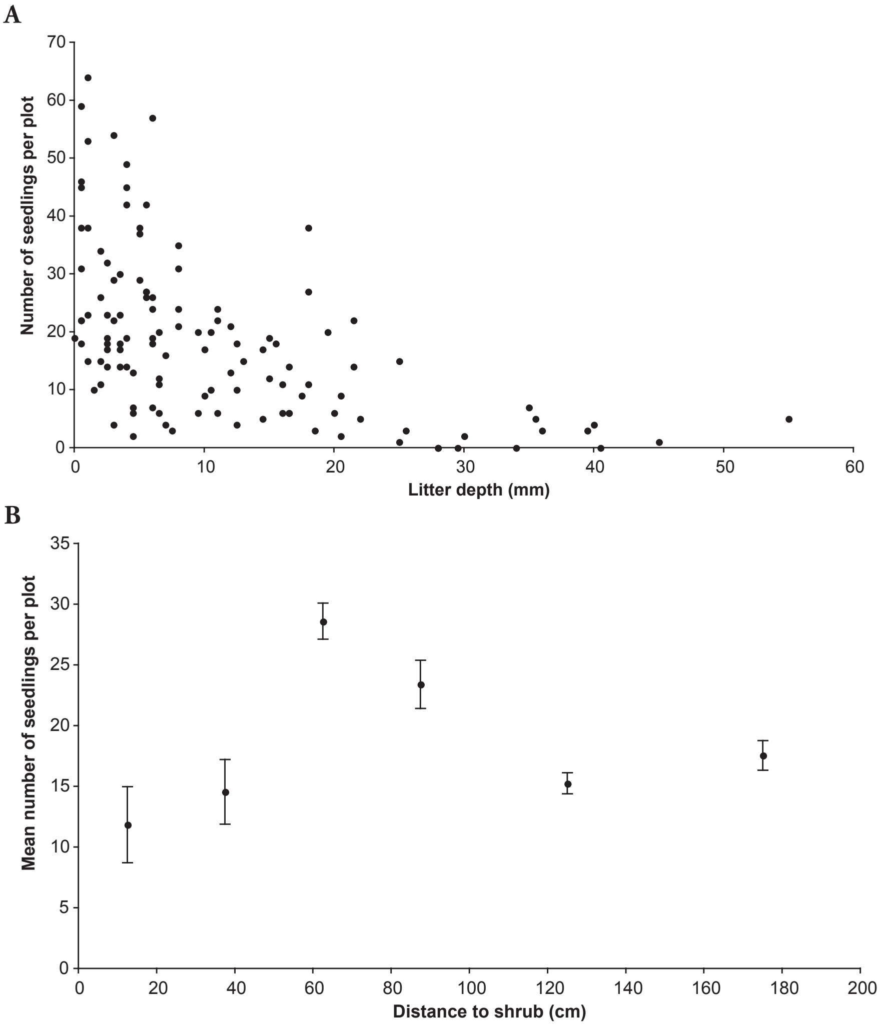

Fig. 4. Number of seedlings per plot in relation to litter depth (A), and mean number of seedlings per plot in relation to distance to *Rosa dumalis* shrubs (B) in a semi-natural pasture, Stammen. Error bars in B show one S.E.

both relationships and mechanisms were studied. In cages protected from grazing the shrubs slightly reduced the reproduction of grassland plants, from 1 to about 0.95 on average for the species. This effect was statistically, but most likely not ecologically significant, and can probably be attributed to competition from the shrubs (Berlow *et al.*, 2003). On the other hand, in presence of grazing the shrubs functioned as grazing refuges (see also Rousset & Lepart, 2003), considerably decreasing the grazing of reproductive shoots in Stammen. The probability of finding reproductive shoots of the occurring species was higher close to shrubs both in Stammen and the other grasslands. In Stammen, the positive effect of the shrub as a grazing refuge thus outweighed the negative effects of competition (observed in the cages) on the reproduction of the studied plant species. The positive relationship between shrubs and reproduction was strongest in

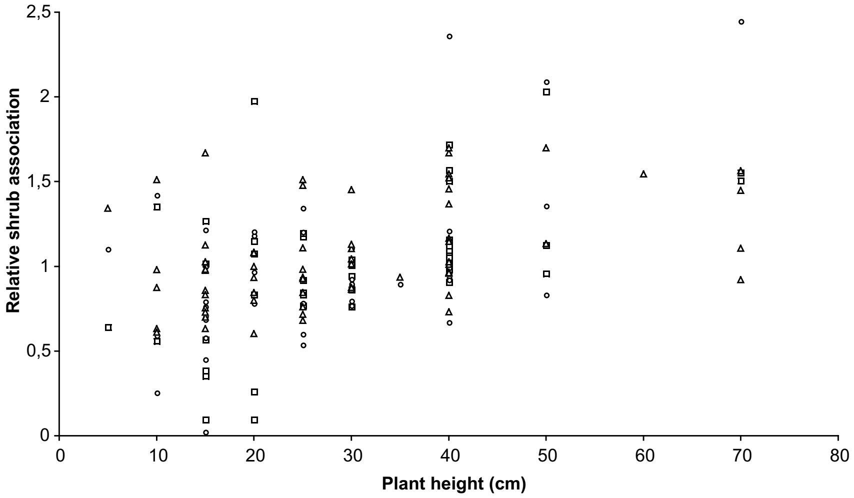

Fig. 5. Species specific plant height (according to literature) in relation to relative shrub association (see text for explanations), in three semi-natural pastures, Focksta (circles), Stammen (triangles), and Åsbergby (lines). For clarity, no error bars are shown.

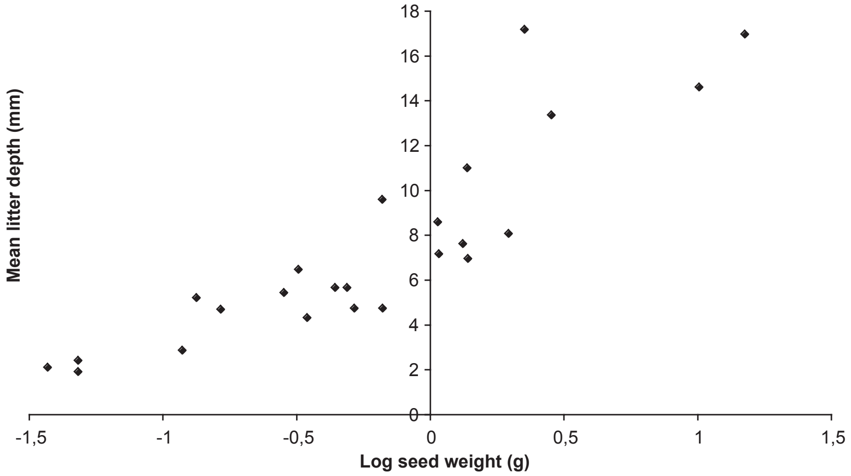

Fig. 6. Species specific seed weight in relation to the mean litter depth of the subplots in which seedlings of the species were found (see text for explanation). Each data point represents one species. For clarity, no error bars are shown.

Focksta, where almost all reproductive shoots were found close to shrubs, and lowest in Stammen. In addition to effects on reproduction, shrubs can also be expected to positively affect the growth and survival of grazing-sensitive species (Diaz *et al.*, 2001), which should ultimately be reflected into a positive relationship between abundance and shrubs of those species. Since the grazing sensitivity of different species is poorly known, no analysis of such patterns could be done in this study. However, several of the species showing the strongest positive relationship with shrubs are considered to be grazing sensitive, for example, *Anthoxantum odoratum* (Hansson & Fogelfors, 2000), *Campanula persicifolia* (Svensson & Glimskär, 1990; Wahlman & Milberg, 2002) and *Deschampsia* *flexuosa* (Wahlman & Milberg, 2002).

Plant height (according to Lid, 1985) correlated with relative shrub association in all three grasslands. There are probably two interacting mechanisms involved in this pattern: (1) tall species may be more grazing sensitive than short species (Diaz *et al*., 2001), and (2) tall species may be more competitive than short species. Dupré & Diekmann (2001) found that low-growing species were more abundant in grazed sites and tall species in abandoned grasslands. In Focksta, competition was further supported by a correlation between relative shrub association and the Ellenberg index for light. If light competition is important, this correlation could be expected for all three grasslands. However, the Ellenberg index scale, ranging from deep shadow (index 1) to full light (9), may be too coarse to accurately detect the small light differences in this study system. Grasses, but not herbs, was positively related to shrubs, which was probably an effect of plant height rather than growth form or life form since the grass species found in this study were among the taller plants. Growth form and Raunkiaer form showed no relationship with relative shrub association.

Establishment of seedlings of small-seeded species was negatively related to shrubs due to thicker litter layer close to shrubs, mainly caused by weaker grazing intensity. Several earlier studies have demonstrated a strong relationship between establishment and litter depth (e.g. Brewer, 1999; Jensen & Meyer, 2001), and Jensen & Gutekunst (2003) also found the relationship to be correlated with seed weight.

If shrubs increase the seed production and decrease the establishment of plant seedlings, the density of seedlings can be expected to be highest at a certain distance from the shrub, a pattern which was supported in this study. In a zone 60-90 cm from the shrub, seedling density was significantly higher than at adjacent distances, both closer to and more distant from the shrubs. Hence the net effect of shrubs on seedling density was positive. The spatial pattern of seedling density was not reflected in the pattern of frequency of adult plants in general, probably because of density dependence (Goldberg *et al*., 2001). The pattern of seedling density may, however, be an important determinant of adult frequency for single species.

The shrub effects on herbs and grasses thus differed between life stages and plant species. In addition, the relative importance of the different mechanisms varied between grasslands and/or years. For example, seed weight explained plant abundance in relation to shrubs in one of the three grasslands, and shrub influence on single species differed in strength (but not direction) between grasslands. The most obvious difference between the sites was the vegetation height (lowest in Focksta, highest in Stammen). The cage experiment indicates that the variation in vegetation height at different distances to shrubs is an effect of grazing intensity, and it is likely that variation in vegetation height between grasslands in a similar manner reflects a between-site variation in grazing intensity. Therefore intensive grazing can be assumed to enhance and weak grazing to reduce the relative importance of all mechanisms related to the shrubs' function as grazing refuges.

Although mechanisms and actual responses were studied in only one grassland, the observed mechanisms well explained the patterns observed in all three grasslands. It is therefore likely that the study demonstrates, directly or indirectly, examples of competitional and associational responses (Callaway, 1995) of grassland plants to *Rosa* shrubs.

Implications for grassland management

In this study, the overall net effect of *R. dumalis* shrubs on the number and general abundance of plant species can be said to be neutral. Most plant species showed a neutral, some a negative and others a positive change in abundance closer to shrubs. Thus the shrubs increase the spatial variation in vegetation composition. The shrubs had a clear positive effect on the reproduction of herbs and grasses, by functioning as grazing refuges. The grazing refuge also had a positive net effect on the seedling density. Although studied in Stammen only, this effect on seedlings was consistent with theoretical predictions and further studies may prove the effect to be more or less general in semi-natural grasslands.

The increased flowering and production of fruits close to shrubs may be important also for other organisms, depending on plant reproduction, in particular insects feeding on pollen, nectar and seeds. For example, many bee species, of which several are red-listed (Gärdenfors, 2005), are considered to be threatened by intensive grazing which reduces the resources of pollen and nectar (Falk, 1991; Williams, 1996; Potts *et al.*, 2003). Moreover phytophagous species depend on grazing refuges for completing their life cycle. For example, both individual butterfly species (Bergman, 2001) and butterfly species richness (Bergman et al., 2004) are affected by shrub cover in semi-natural grasslands. Shrubs in grasslands are also directly used by different organisms, e.g. as substrate for cryptogams (Hallingbäck, 1995; Hallingbäck, 1996) or as nesting sites for birds (Pärt & Söderström, 1999). Such functions are not addressed in this study, but must be taken into account if shrub clearing is considered to improve the conditions for grassland biodiversity.

The heterogeneity created by shrubs in semi-natural pastures can be assumed to be particularly important when grazing is intense. At weaker grazing intensities, the effects of shrubs decrease, in terms of probability of reproduction, vegetation height, and litter layer. In summary, shrubs in semi-natural grasslands contribute to the diversity of a variety of organism groups and this study indicates that obvious negative effects on herbs and grasses are few. Therefore, intensive clearing of shrubs may in most cases have adverse net effects on biodiversity.

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References

Austad, I. & Losvik, M.H. 1998. Changes in species composition following field and tree layer restoration and management in a wooded hay meadow. *Nordic Journal of Botany 18*, 641-662.

Bergman, K.O. 2001. Population dynamics and the importance of habitat management for conservation of the butterfly *Lopinga achine*. *Journal of Applied Ecology 38*, 1303-1313.

Bergman, K.O., Askling, J., Ekberg, O., Ignell, H., Wahlman, H. & Milberg, P. 2004. Landscape effects on butterfly assemblages in an agricultural region. *Ecography 27*, 619-628.

Berlow, E.L., D'Antonio, C.M. & Swartz, H. 2003. Response of herbs to shrub removal across natural and experimental variation in soil moisture. *Ecological Applications 13*, 1375-1387.

Brewer, J.S. 1999. Effects of competition, litter, and disturbance on an annual carnivorous plant (*Utricularia juncea*). *Plant Ecology 140*, 159-165.

Brooker, R., Kikvidze, Z., Pugnaire, F.I., Callaway, R.M., Choler, P., Lortie, C.J. & Michalet, R. 2005. The importance of importance. *Oikos 109*, 63-70.

Callaway, R.M. 1995. Positive interactions among plants. *Botanical Review 61*, 306-349.

Callaway, R.M., Kikvidze, Z. & Kikodze, D. 2000. Facilitation by unpalatable weeds may conserve plant diversity in overgrazed meadows in the Caucasus Mountains. *Oikos 89*, 275-282.

Connell, J.H. & Slatyer, R.O. 1977. Mechanisms of succession in natural communities and their role in community stability and organisation. *American Naturalist 111*, 1119-1144.

Correll, O., Isselstein, J. & Pavlu, V. 2003. Studying spatial and temporal dynamics of sward structure at low stocking densities: the use of an extended rising-plate-meter method. *Grass Forage Science 58*, 450-454.

Diaz, S., Noy-Meir, I. & Cabido, M. 2001. Can grazing response of herbaceous plants be predicted from simple vegetative traits? *Journal of Applied Ecology 38*, 497-508.

Drews, S., Juroszek, P., Neuhoff, D. & Kopke, U. 2004. Optimizing shading ability of winter wheat stands as a method of weed control. *Zeitschrift fur Pflanzenkrankheiten und Pflanzenschutz-Journal of Plant Diseases and Protection 19*, 545-552.

Dupre, C. & Diekmann, M. 2001. Differences in species richness and life-history traits between grazed and abandoned grasslands in southern Sweden. *Ecography 24*, 275-286.

El Bana, M.I., Nijs, I. & Kockelbergh, F. 2002. Microenvironmental and vegetational heterogeneity induced by phytogenic nebkhas in an arid coastal ecosystem. *Plant and Soil 247*, 283-293.

Einarsson, A. & Milberg, P. 1999. Species richness and distribution in relation to light in wooded meadows and pastures in southern Sweden. *Annales Botanici Fennici 36*, 99-107.

Ellenberg, H., Weber, H.E., Düll, R., Wirth, V., Werner, W. & Paulissen, D. 1991. Zeigerwerte von Pflanzen in Mitteleuropa. *Scripta Geobotanica 18*, 1-258.

Facelli, J.M. & Temby, A.M. 2002. Multiple effects of shrubs on annual plant communities in arid lands of South Australia. *Austral Ecology 27*, 422-432.

Falk, S. 1991. A review of the scarce and threatened bees, wasps and ants in Great Britain. Research and Survey in Nature Conservation, No 35.

Goldberg, D.E., Turkington, R., Olsvig-Whittaker, L. & Dyer, A.R. 2001. Density dependence in an annual plant community: Variation among life history stages. *Ecological Monographs 71*, 423-446.

Gärdenfors, U. (ed.) 2005. *Rödlistade arter i Sverige 2005 - The 2005 Red List of Swedish Species*. ArtDatabanken, SLU, Uppsala.

Hallingbäck, T. 1995. *Ekologisk katalog över lavar*. ArtDatabanken, SLU, Uppsala.

Hallingbäck, T. 1996. *Ekologisk katalog över mossor*. ArtDatabanken, SLU, Uppsala.

Hansson, M. & Fogelfors, H. 2000. Management of a semi-natural grassland: results from a 15-year-old experiment in southern Sweden. *Journal of Vegetation Science 11*, 31-38.

Hjälten, J. & Price, P.W. 1997. Can plants gain protection from herbivory by association with unpalatable neighbours?: A field experiment in a willow-sawfly system. *Oikos 78*, 317-322.

Ihse, M. 1995. Swedish agricultural landscapes - patterns and changes during the last 50 years, studied by aerial photos. *Landscape and Urban Planing 31*, 21-37.

Jensen, K. & Gutekunst, K. 2003. Effects of litter on establishment of grassland plant species: the role of seed size and successional status. *Basic and Applied Ecology 4*, 579-587.

Jensen, K. & Meyer, C. 2001. Effects of light competition and litter on the performance of Viola palustris and on species composition and diversity of an abandoned fen meadow. *Plant Ecology 155*, 169-181.

Kleijn, D. & Sutherland, W.J. 2003. How effective are European agri-environment schemes in conserving and promoting biodiversity? *Journal of Applied Ecology 6*, 947-969.

Krok, Th.O.B.N. & Almqvist, S. 2001. *Svensk flora Fanerogamer och ormbunksväxter*. 28th edition. Liber Utbildning.

Lennartsson, T. 2002. Extinction thresholds and disrupted plant-pollinator interactions in fragmented plant populations. *Ecology 83*, 3060-3072.

Lennartsson, T. & Oostermeijer, J.G.B. 2001. Demographic variation and population viability in Gentianella campestris: effects of grassland management and environmental stochasticity. *Journal of Ecology 89*, 451-463.

Lid, J. 1985. *Norsk, svensk, finsk flora*. Det Norske Samlaget.

Lindborg, R. & Eriksson, O. 2004. Effects of restoration on plant species richness and composition in Scandinavian semi-natural grasslands. *Restoration Ecology 12*, 318-326.

McIntyre, S., Lavorel, S. & Tremont, R.M. 1995. Plant Life-History Attributes - Their Relationship to Disturbance Responses in Herbaceous Vegetation. *Journal of Ecology 83*, 31-44.

Müller-Schneider, P. 1986. *Verbreitungsbiologie der Blütenpflanzen Graubündens: Diasporology of the Spermatophytes of the Grisons (Switzerland)*. ETH, Geobotanisches Institut, Zürich.

Moro, M.J., Pugnaire, F.I., Haase, P. & Puigdefabregas, J. 1997. Mechanisms of interaction between a leguminous shrub and its understorey in a semi-arid environment. *Ecography 20*, 175-184.

Olff, H. & Ritchie, M.E. 1998. Effects of herbivores on grassland plant diversity. *Trends in Ecology & Evolotion 13*, 261-265.

Olff H., Vera F.W.M., Bokdam J., Bakker E.S., Gleichman J.M., de Maeyer K. & Smit R. 1999. Shifting mosaics in grazed woodlands driven by the alternation of plant facilitation and competition. *Plant Biology 1*, 127-137.

Overud, S. & Lennartsson, T. 2004. Skötsel och restaurering av betesmarker och slåtterängar. En sammanställning av den regionala naturvårdens kunskaper och erfarenheter. Jordbruksverket Publications 2004:11. Jordbruksverket.

Potts, S.G., Vulliamy, B., Dafni, A., Ne'eman, G. & Willmer, P. 2003. Linking bees and flowers: How do floral communities structure pollinator communities? *Ecology 84*, 2628-2642.

Pykälä, J. 2005. Plant species responses to cattle grazing in mesic semi-natural grassland. *Agriculture Ecosystems & Environment 108*, 109-117.

Påhlsson, L. 1994. *Vegetationstyper i Norden. (Nordic Vegetation Types).* TemaNord 1994: 665.

Pärt, T. & Söderström, B. 1999. The effects of management regimes and location in landscape on the conservation of farmland birds breeding in semi-natural pastures. *Biological Conservation 90*, 113-123.

Pärtel, M. & Zobel, M. 1999. Small-scale plant species richness in calcareous grasslands determined by the species pool, community age and shoot density. *Ecography 2*, 153-59.

Rebollo, S., Milchunas, D.G., Noy-Meir, I. & Chapman, P.L. 2002. The role of a spiny plant refuge in structuring grazed shortgrass steppe plant communities. *Oikos 98*, 53-64.

Rejmánek, M. & Rosén, E. 1992. Influence of colonizing shrubs on species-area relationships in alvar plant communities. *Journal of Vegetation Science 3*, 625-630.

Regeringskansliet. 2000. The Environmental and Rural Development Plan for Sweden 2000-2006.

Rook, A.J., Dumont, B., Isselstein, J., Osoro, K., WallisDeVries, M.F., Parente, G. & Mills J. 2004. Matching type of livestock to desired biodiversity outcomes in pastures - a review. *Biological Conservation 119*, 137-150.

Rousset, O. & Lepart, J. 2003. Neighbourhood effects on the risk of an unpalatable plant being grazed. *Plant Ecology 165*, 197-206.

Stanners, D. & Bordeau, P. 1995. Europe's Environment. The Dobris Assessment. European Environment Agency, Copenhagen.

Svensson, R. & Glimskär, A. 1990. Vegetation changes following fertilization and changed management. *Department of Ecology and Environmental Research, SLU, Report No 38*.

Söderström, B., Svensson, B., Vessby, K. & Glimskär, A. 2001. Plants, insects and birds in semi-natural pastures in relation to local habitat and landscape factors. *Biodiversity and Conservation 10*, 1839-1863.

Söderström, P. 1993. Ängs- och hagmarker i Uppsala län. 1993:3. Länsstyrelsen Uppsala län.

Tahvanainen, J.O. & Root, R.B. 1972. The Influence of Vegetational Diversity on the Population Ecology of a Specialized Herbivore, Phyllotreta cruciferae (Coleoptera: Chrysomelidae). *Oecologia 10*, 321-346.

ter Braak, C.J.F. & Smilauer, P. 2002 *CANOCO Reference manual and CanoDraw for Windows User's guide: Software for Canonical Community Ordination (version 4.5).* Microcomputer Power, Ithaca, NY, USA.

Wahlman, H. & Milberg, P. 2002. Management of semi-natural grassland vegetation: evaluation of a long-term experiment in southern Sweden. *Annales Botanici Fennici 39*, 159-166.

Weibull, A.C. & Östman, Ö. 2003. Species composition in agroecosystems: The effect of landscape, habitat, and farm management. *Basic and Applied Ecology 4*, 349-361.

Williams, I.H. 1996. *Aspects of bee diversity and crop pollination in the European Union*. In: The conservation of bees. (Eds. A. Matheson, S.L. Buchmann, C.O. Toole, P. Westrich & I.H. Williams). Academic Press.

Appendix 1. Mean frequency (%) of adult plant species at two distance intervals from *Rosa dumalis* shrubs in three pastures, Focksta, Stammen, and Åsbergby and mean frequency of seedlings of herbs and sedges (%) in one pasture, Stammen. Only species occurring in 10 or more plots are analysed; n = total number of plots with occurrence of the species (see text for explanation of experimental setup). Underlining refers to abbreviations in Fig. 1. Bold figures indicate significant differences between the two distance intervals (Mann-Whitney U-test, p<0.05).

		Focksta adults			**Stammen adults**			**Åsbergby adults**			**Stammen seedlings**	
		% plots ± SE	% plots ± SE		% plots ± SE	% plots ± SE		% plots ± SE	% plots ± SE		% plots ± SE	% plots ± SE
Species	n	10-70 cm	110-310 cm	n	12.5-62 cm	87.5-175 cm	n	10-70 cm	110-310 cm	n	12.5-62 cm	87.5-175 cm
Achillea millefolium	158	**80 ± 4**	**100 ± 0**	73	**46 ± 8**	**89 ± 3**	152	**81 ± 4**	**92 ± 3**	49	43 ± 14	53 ± 9
Agrostis capillaris	162	**88 ± 4**	**100 ± 0**	52	41 ± 7	56 ± 6	164	**88 ± 4**	**99 ± 0**			
Ajuga pyramidalis				14	14 ± 4	19 ± 2				23	31 ± 4	28 ± 7
Alchemilla sp.	101	**54 ± 5**	**79 ± 5**	37	30 ± 5	35 ± 2	40	38 ± 5	28 ± 4	35	27 ± 9	41 ± 0
Alopecurus pratensis				41	45 ± 4	35 ± 12						
Anemone nemorosa				33	44 ± 3	41 ± 13						
Antennaria dioica				10	17 ± 10	39 ± 24						
Anthoxanthum odoratum	45	**60 ± 5**	**27 ± 4**	96	78 ± 4	82 ± 3	88	**74 ± 5**	**49 ± 5**			
Anthriscus sylvestris	18	**50 ± 4**	**6 ± 2**	14	28 ± 7	8 ± 4				23	**44 ± 12**	**19 ± 6**
Briza media	16	29 ± 3	29 ± 3	42	56 ± 6	61 ± 7						
Campanula persicifolia	15	43 ± 4	11 ± 2	18	**42 ± 10**	**8 ± 5**	19	**47 ± 4**	**13 ± 2**	19	22 ± 14	31 ± 6
Campanula rotundifolia	69	57 ± 5	44 ± 5	73	70 ± 6	58 ± 8	121	84 ± 5	75 ± 5	23	27 ± 19	42 ± 3
Carex sp.	62	44 ± 5	42 ± 5	78	52 ± 9	53 ± 9	109	**55 ± 5**	**69 ± 5**			
Carum carvi	36	**21 ± 3**	**54 ± 5**	25	67 ± 0	72 ± 20						
Centaurea jacea	49	41 ± 5	46 ± 5	41	35 ± 2	41 ± 2				37	33 ± 2	39 ± 2
Cerastium fontanum	49	**22 ± 4**	**50 ± 5**	31	48 ± 4	67 ± 11	40	**12 ± 3**	**47 ± 5**	24	37 ± 10	52 ± 10
Crepis praemorsa				20	93 ± 7	40 ± 23						
Dactylis glomerata	72	50 ± 5	50 ± 5	19	**58 ± 4**	**21 ± 8**	17	**54 ± 4**	**17 ± 2**			
Deschampsia cespitosa	18	30 ± 4	15 ± 3	11	33 ± 7	40 ± 12	15	40 ± 3	35 ± 3			
Deschampsia flexuosa	29	78 ± 5	3 ± 1	17	52 ± 16	11 ± 11	60	**73 ± 5**	**20 ± 4**			
Festuca ovina	53	25 ± 4	41 ± 5	75	51 ± 5	81 ± 2	80	60 ± 5	73 ± 5			
Festuca pratensis	74	50 ± 5	73 ± 5				63	62 ± 5	43 ± 5			
Festuca rubra	86	57 ± 5	63 ± 5	67	**50 ± 3**	**74 ± 2**	148	81 ± 4	88 ± 3			
Filipendula vulgaris	136	84 ± 5	86 ± 4	92	73 ± 3	80 ± 3	120	78 ± 5	73 ± 5	53	60 ± 17	50 ± 6
Fragaria vesca	67	**63 ± 5**	**42 ± 5**	17	62 ± 17	19 ± 13	40	**64 ± 5**	**27 ± 4**	22	38 ± 4	18 ± 3
Galium boreale	104	78 ± 5	67 ± 5	64	71 ± 10	55 ± 9	110	**82 ± 5**	**63 ± 5**			
Galium verum	149	83 ± 4	86 ± 4	82	75 ± 2	68 ± 13	148	84 ± 4	84 ± 4	41	43 ± 7	37 ± 8
Geranium sylvaticum	7	33 ± 2	25 ± 2	9	39 ± 15	11 ± 11						
Geum rivale/urbanum	35	39 ± 5	23 ± 4	17	43 ± 12	13 ± 3	24	**57 ± 4**	**29 ± 3**	12	**46 ± 23**	**4 ± 4**
Helianthemum nummularium	10	58 ± 3	25 ± 2	14	38 ± 17	29 ± 0	25	70 ± 4	55 ± 4			
Helictotricon pratense	145	**76 ± 5**	**89 ± 3**	71	71 ± 3	69 ± 5	87	66 ± 5	49 ± 5			
Helictotricon pubescens	59	**60 ± 5**	**26 ± 4**	52	59 ± 7	43 ± 9	49	**50 ± 5**	**27 ± 4**			
Hypericum maculatum				27	**83 ± 8**	**29 ± 4**	13	**36 ± 3**	**11 ± 2**	14	21 ± 8	38 ± 7
Lathyrus linifolia				40	74 ± 13	74 ± 4	19	**80 ± 4**	**15 ± 2**	11	**42 ± 45**	**4 ± 4**
Lathyrus pratensis	40	31 ± 5	28 ± 4	32	42 ± 10	29 ± 8	36	40 ± 4	50 ± 5	23	36 ± 7	19 ± 10

Appendix 1. cont.

	Focksta adults			Stammen adults			Åsbergby adults			Stammen seedlings		
Species	n	% plots ± SE 10-70 cm	% plots ± SE 110-310 cm	n	% plots ± SE 12.5-62 cm	% plots ± SE 87.5-175 cm	n	% plots ± SE 10-70 cm	% plots ± SE 110-310 cm	n	% plots ± SE 12.5-62 cm	% plots ± SE 87.5-175 cm
Leontodon autumnalis	35	**23 ± 4**	**50 ± 5**	20	**22 ± 6**	**52 ± 7**						
Leucanthemum vulgare	23	40 ± 3	75 ± 4	50	72 ± 9	56 ± 9	12	13 ± 2	38 ± 3	20	33 ± 5	22 ± 3
Lotus corniculatus	45	32 ± 4	48 ± 5	24	15 ± 7	30 ± 4	26	**17 ± 3**	**38 ± 4**	32	31 ± 7	35 ± 2
Luzula campestris	57	38 ± 5	52 ± 5	69	52 ±10	92 ± 4	103	**53 ± 5**	**76 ± 5**	32	31 ± 4	51 ± 9
Phleum pratense	13	29 ± 3	18 ± 2	18	47 ± 15	13 ± 9						
Pilosella lactucella	14	**9 ± 2**	**34 ± 4**									
Pilosella officinarum	12	25 ± 3	25 ± 3	74	**53 ± 5**	**77 ± 4**	57	**55 ± 5**	**75 ± 5**			
Pimpinella saxifraga	59	46 ± 5	59 ± 5	59	63 ± 6	60 ± 2	82	**45 ± 5**	**63 ± 5**	24	27 ± 4	27 ± 4
Plantago lanceolata	13	58 ± 3	50 ± 3	85	65 ± 6	77 ± 2	39	56 ± 4	53 ±			
Plantago media	26	25 ± 3	40 ± 4	17	13 ± 7	19 ± 7				55	56 ± 17	58 ± 6
Poa pratensis	53	46 ± 5	32 ± 5	40	31 ± 8	58 ± 6	102	**73 ± 5**	**55 ± 5**			
Polygala vulgaris				35	47 ± 7	50 ± 8	18	75 ± 3	75 ± 3	17	27 ± 7	30 ± 10
Potentilla crantzii				17	**67 ± 5**	**14 ± 8**						
Potentilla erecta	37	48 ± 5	36 ± 4	51	83 ± 3	87 ± 7				10	**30 ± 6**	**3 ± 3**
Potentilla reptans	32	79 ± 4	54 ± 4	16	52 ± 10	24 ± 10	23	38 ± 3	58 ± 4			
Potentilla tabernaemontani	15	19 ± 3	28 ± 3				11	33 ± 2	58 ± 3			
Primula veris	14	29 ± 3	29 ± 3									
Prunella vulgaris	18	**9 ± 2**	**47 ± 4**	31	**25 ± 5**	**61 ± 3**	17	33 ± 3	38 ± 3			
Ranunculus acris	90	**42 ± 5**	**65 ± 5**	28	21 ± 6	28 ± 2	50	48 ± 5	48 ± 5	41	**26 ± 4**	**50 ± 3**
Ranunculus bulbosus				11	33 ± 17	28 ± 20	44	**20 ± 4**	**59 ± 5**			
Rhinanthus minor				52	45 ± 10	79 ± 12				17	27 ± 3	30 ± 6
Rumex acetosa	100	**48 ± 5**	**71 ± 5**	33	63 ± 4	59 ± 10	67	49 ± 5	50 ± 5			
Stellaria graminea	121	**49 ± 5**	**89 ± 3**	37	**25 ± 4**	**52 ± 4**	128	70 ± 5	82 ± 4			
Succisa pratensis	11	19 ± 2	50 ± 3	24	47 ± 9	33 ± 13						
Taraxacum sp.	27	22 ± 4	57 ± 5	34	36 ± 5	51 ± 7	13	**3 ± 1**	**38 ± 4**			
Thlapsi caerulescens	86	72 ± 5	54 ± 5									
Trifolium medium	122	76 ± 5	69 ± 5	61	**43 ± 0**	**14 ± 0**	83	58 ± 5	51 ± 5			
Trifolium pratense	52	**19 ± 4**	**53 ± 5**	12	61 ± 9	46 ± 4	42	27 ± 4	39 ± 5			
Trifolium repens	129	**49 ± 5**	**98 ± 2**	43	**33 ± 8**	**62 ± 4**	104	**28 ± 5**	**90 ± 3**			
Veronica chamaedrys	110	58 ± 5	67 ± 5	39	**46 ± 0**	**54 ± 0**	113	58 ± 5	70 ± 5	29	28 ± 3	46 ± 8
Veronica officinalis	12	14 ± 2	29 ± 3	43	58 ± 5	61 ± 3	53	**57 ± 5**	**32 ± 4**			
Veronica serpyllifolia	18	8 ± 2	42 ± 4				28	**15 ± 3**	**44 ± 5**			
Vicia cracca		15 ± 2	10 ± 2	13	42 ± 15	13 ± 7	13	14 ± 3	16 ± 3			
Vicia sepium	14	**54 ± 4**	**4 ± 1**	12	42 ± 15	8 ± 4	31	**48 ± 5**	**17 ± 3**			
Viola sp.	52	54 ± 5	39 ± 5	55	58 ± 6	64 ± 4	47	44 ± 5	30 ± 4			

Effects of spatial structures on reproduction and seed predation of four legumes (Fabaceae) in semi-natural pastures

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Key words: *Apion*, *Bruchus*, grassland, grazing, *Lathyrus pratensis*, *Lotus corniculatus*, *Rosa dumalis*, seed set, spatial heterogeneity, vegetation height, *Vicia cracca*, *Vicia sepium*.

Abstract

The effect of four spatial structures, *Rosa dumalis* shrubs, dung pats, grazing rejects (ungrazed patches) and grazed patches, were studied on the abundance, flowering, fruiting and seed-predation for four legumes: *Lathyrus pratensis*, *Lotus corniculatus*, *Vicia cracca* and *Vicia sepium*. The study was performed in seven pastures with different grazing intensities in south-central Sweden. Rose shrubs and dung pats were avoided by grazing animals and grazing rejects, were therefore created near these structures. The quantity of dung, shrub and grazing rejects increased with decreasing grazing intensity. *L. pratensis* was more abundant in rejects than in grazed patches and the flowering and fruiting was highest in dung rejects. *L. corniculatus* was most abundant in dung rejects and in grazed patches, and the reproduction was highest in dung rejects. *V. cracca* did not differ in abundance between structures but reproduction was higher in rejects than in grazed patches. *V. sepium* occurred almost only in rose shrubs but reproduction did not differ between the structures. Predation rate did not differ between the structures but more pods were available for oviposition in rose shrubs and dung pats than in grazing rejects and grazed patches. Predation rate was influenced by pod length and vegetation height. In conclusion, grazing and regulation of grazing intensity are two important tools when managing pastures for plants and insects.

Introduction

Semi-natural grasslands, i.e., unfertilized, uncultivated pastures and hay meadows are species-rich habitats, especially for plants and insects (Mortimer *et al.*, 1998, Pärtel & Zobel, 1989). These habitats are maintained by regular disturbance such as grazing or mowing, which counteract litter accumulation and reduces dominant plant species and therefore allow many small, less dominant species to coexist (Olff & Ritchie, 1998; Jensen & Gutekunst, 2003). A diverse plant community is in turn beneficial for both abundance and species richness of insects (Mortimer *et al.*, 1998; Woodcock *et al.*, 2005). However, management of semi-natural grasslands for conservation has often focused on vascular plants (WallisDeVries *et al.*, 2002) and most productive grasslands are managed with moderate or intensive grazing to increase plant diversity (Olff & Ritchie, 1998; Prolux & Mazumder, 1998; Pykälä, 2005). The requirement of regular disturbance for plant diversity is contrasted by the need of undisturbed conditions that allow reproduction of invertebrates and their host plants (Morris, 1967; Lennartsson, 2000). For example, arthropod diversity is higher in grasslands with tall vegetation than in grasslands with short swards (Morris, 2000) and species richness of butterflies and moths has been shown to peak in taller vegetation than species richness of vascular plants (Pöyry *et al.*, 2006). When grazing ceases in semi-natural grasslands, populations of phytophagous insects initially increase in response to increased availability of resources such as flowers and fruits (Morris, 1967), but when succession continues and plant species diversity is reduced, arthropod diversity also decreases (Siemann *et al.*, 1998; Siemann *et al.*, 1999). Arthropod diversity is also correlated with plant structural diversity (see Lawton, 1983) and the insect fauna of semi-natural grasslands is affected by grazing due to both changes in plant community composition and in vegetation structure (Mortimer *et al.*, 1998). The effects of grazing differ between invertebrate groups, for example leaf-miner assemblies depend on plant species composition and spider assemblies respond to plant architecture (Gibson *et al.*, 1992).

In general, reproductive success of phytophagous insects can be assumed to depend on where the female deposits the eggs (Brody & Morita, 2000). The female can choose flowers with high seed set within a plant (Lalonde & Roitberg, 1994; Brody & Morita, 2000), vigorous plant individuals (Brody & Waser, 1995; Cariveau *et al.*, 2004), or safe patches within a heterogeneous habitat (Vanbergen *et al.*, 2006). In semi-natural pastures, up to 80% of the flowers and fruits can be grazed (Wissman, 2006) and levels of damage on host plants depend on grazing intensity (Jerling & Andersson, 1982), plant palatability, occurrence of spatial structures that function as partial grazing refugees, for example shrubs (Callaway, 1995; Rousset & Lepart, 2003) and on grazing selectivity. Selective grazing can be due to dietary choices; i.e. patches with low forage quality or unpalatable species are avoided by grazing animals (Bailey *et al.*, 1998; Rook *et al.*, 2004). Grazing animals also avoid vegetation near dung pats (Jones & Ratcliff, 1983; Shiyomi *et al.*, 1998) and near spiny species (Bakker *et al.*, 2004). In grazing refugees, plant reproduction is often higher than in grazed patches (Shiyomi *et al.*, 1998; Bakker *et al.*, 2004). Depending on their abundance in grazing refuges, different plant species may be affected differently by grazing. Accordingly, survival of phytophagous insects may depend on choice of host individuals, i.e. plants in spatial structures with reduced risk for mortality due to grazing.

Here we studied the effects of grazing and different grazing refuges on four legumes (Fabaceae): *Lathyrus pratensis* L., *Lotus corniculatus* L., *Vicia cracca* L. and *Vicia sepium* L. and their seed predators (*Apion* spp. and *Bruchus* spp.). The four legumes differ in growth form and occur in different microhabitats within pastures and can be expected to respond differently to grazing and occurrence of different spatial structures. The four legumes and their seed predators were studied in relation to four spatial structures: *Rosa dumalis* shrubs, dung pats, grazing rejects (ungrazed patches) and grazed patches.

In this study we specifically addressed the following questions: 1). How do the abundances of the four legumes vary between different spatial structures in semi-natural pastures? 2). Do flowering, fruiting and seed set, and seed predation rates, vary between spatial structures and is the effect of structures varying with grazing intensity? 3). Does seed predation rate vary between spatial structures, indicating selective oviposition at the structural level or does seed predation rate vary with host plant characters, indicating plant level selectivity?

Methods

Study sites

The study was performed in seven pastures situated in south central Sweden (59°44'N to 60°15'N and 17°20'E to 18°33'E). All sites were unfertilized semi-natural grasslands with high floristic values included in the national survey of semi-natural meadows and pastures in Sweden (Söderström 1993; Persson, 2005). All sites had scattered trees and shrubs such as *R. dumalis*, *Juniperus communis* and *Prunus spinosa*. Small parts of the pastures were forested and all sites included abandoned arable land. The sites were grazed by either meat or dairy cattle. One locality, Tvärnö, was chosen because it was ungrazed during the study period, but it had been grazed the years before.

Measurements of the vegetation height during the summer (see study design) suggested that grazing intensity was highest in Bergesta and Långalma, intermediate in Rasbo, Lagga and Åsbergby and, beside the ungrazed Tvärnö site, lowest in Hagby (Table 1).

Study species

The four study species differ in growth form and seed production and prefer slightly different environmental conditions. *L. pratensis, V. cracca* and *V. sepium* use tendrils to climb and occur in both open and shrubby habitats (Mitchley & Willems, 1995; Mossberg & Stenberg, 2003). *L. corniculatus* occurs mainly in open habitats and flowers continuously through the summer and the pods have 1-30 seeds (Ollerton & Lack, 1998). *L. pratensis* flowers and set fruits in June-July and the pods can have up to 10 seeds (own data). *V. cracca* flowers in June-August and the pods have on average 4-8 seeds and *V. sepium* flowers in early summer and the pods normally have 3-7 seeds (Mossberg & Stenberg, 2003).

Seed predators on legumes can be monophagous, i.e. they depend on one host species, oligophagous, i.e. they depend on a small group of host species, or generalists. *Apion loti* is monophagous and oviposits exclusively on *L. corniculatus* (Gønget, 1997). *Apion cerdo* and *Apion craccae* are oligophagous on *Vicia* species, but in Sweden they mainly feed and oviposit on *V. cracca* (Gønget, 1997). *Apion subulatum* is oligophagous on *Lathyrus* species and predates mainly on *L. pratensis* seeds (Gønget, 1997). The seed beetle *Bruchus atomarius* is a generalist and oviposits on *Vicia sepium,* *Lathyrus linifolius* and *Lathyrus vernus* (Freude *et al.*, 1981, Östergård & Ehrlen, 2005). *B. atomarius* is the main seed predator of *V. sepium* in the study region (Lenoir & Pihlgren, 2006). *Bruchus loti* oviposits on *L. corniculatus* and on *Lathyrus* species (Freude *et al.*, 1981). Adult weevils feed on the host plant and fertile females search young pods and lay their eggs between the seeds (Gønget, 1997). After 4-6 days the larvae hatches and penetrates a seed which it consumes and then pupates within (Gønget, 1997). When the beetle emerges it leaves the pod through a hole in the pod wall (Gønget, 1997). Both weevil and seed beetle larvae can be parasitized by Hymenoptera.

Study design

Four 10 x 1m transects were located in areas with high abundance of the four host plants within each site. The transects were also located to include *R. dumalis* shrubs, dung pats, grazing rejects and grazed patches. Each transect was divided into forty 50 x 50cm sampling plots. The vegetation height was measured with a rising-plate meter (Correll *et al.*, 2003) in each sampling plot at two occasions: in June and in August. The spatial structure in each sampling plot was described as one of the following structures: shrub reject, dung reject, grazing reject or grazed patch. The spatial structures were described both in June and August and the changes in cover were calculated in % per site: (sum plots with structure x in June - sum plots with structure x in August)/ sum plots with structure x in June. In the ungrazed Tvärnö site plots were described as dung reject, grazing reject or shrub reject since no grazed patches occurred and therefore no changes due to grazing could be detected. The term reject will hereafter be used for shrub reject, dung reject, and grazing reject collectively.

The total number of shoots (no difference was made between grazed and ungrazed shoots) per plot was counted for *L. pratensis*, *V. cracca* and *V. sepium*. For *L. corniculatus* frequency was measured as number of subplots (10 x 10cm) in the sampling plot with presence (one leaf or more) of *L. corniculatus* since the growth form made it difficult to distinguish separate shoots. For each species the number of flower heads and pods was counted in each sampling plot at three occasions, in June, July and August, and summarised per plot. Number of shoots, flower heads and pods per plot were then calculated as shoots/m^2, flower heads/shoot, and pods/shoot respectively. Mature pods were collected in small paper bags and dried. In the laboratory, each pod was examined with respect to pod length, number of ovules, aborted seeds and developed seeds, using a compound microscope. Seed set was counted as: (number of developed seeds + aborted seeds) / total number of ovules per pod. Each seed was examined for seed predators and classified as predated or unpredated. For each pod seed predation rate was calculated as number of predated seeds/number of developed seeds. The seed predators were determined to species level according to Gønget (1997) and Freude *et al.* (1981). Number of seed predators parasitized by Hymenoptera was counted but the parasites were not determined to species level. The pod data were used to calculate a mean for each plot.

Statistical treatment

Data on plant abundance, flowering and fruiting was analysed for the number of sites each species was present in (see Table 2 & 3). Data on pod characters were analyses for the number of sites and structures where pods could be collected from at least five different plots per structure and site. For *L. pratensis* data on pod characters were analysed for four sites and three structures; dung pat, grazing reject and rose shrub. For *L. corniculatus* data were analysed for two sites and between dung pats and grazing rejects. For *V. cracca* pods could only be compared for dung pats between five sites. For *V. sepium* pod characters were compared between threes sites and between rose shubs and grazing rejects. The variation between structures and sites in number of shoots/m^2, flower heads/shoot, pods/shoot, pod length, seed set, seed predation rate, and parasite frequency was analysed with ANCOVA using structure as fixed factor, study site as random factor and vegetation height measured in June as a covariate. Data variables were tested for normality using Kolmogorov-Smirnof tests and variables that were not normally distributed were log (Y+1) transformed prior to the analyses.

Data variables on pod length, seed set, seed predation and parasite frequency were analysed with ANCOVA with structure as fixed factor, site as random factor and vegetation height as covariate. Parameters that were counted as proportion were root arcsine transformed and the other parameters were log (y+1) transformed prior to analyses.

All significant data variables were further analysed with two-way ANOVA with site and structure as factors followed by post hoc tests for differences between means, with Bonferroni correction for multiple comparisons. All statistical analyses were performed in SPSS 14.0.

Results

Spatial structures and abundance

Spatial structures were influenced by grazing and all rejects decreased in area between June and August and grazing rejects decreased more than dung rejects and rose shrubs, especially in intensively grazed sites (Table 1). Vegetation height per plot depended both on site, structure and on the interaction between site and structure (Table 2) and vegetation height was higher in rejects than in grazed patches. Species abundance for all species, except *V. cracca* (Table 2, Fig 1), was influenced by structure and differed between dung rejects, rose shrubs, grazing rejects and grazed patches (Table 2, Fig. 1 & 2). *L. pratensis* was the most abundant species and the abundance was higher in dung rejects than in grazing rejects and grazed patches, and abundance in rose shrubs was higher than in grazed patches (Fig. 1). *L. corniculatus* had highest frequency in dung rejects and was absent in rose shrubs (Fig. 2). The frequency was also negatively correlated with vegetation height at plot level (Pearson correlation, N=1031, r=-0.111, p<0.001). No correlations between vegetation height and abundancee of the other species were found. The abundance of *V. sepium* was highest in rose shrubs (Fig. 1). The abundance of *V. cracca* did not differ between structures but tended to be higher in dung rejects and in grazing rejects than in grazed patches (Fig. 1). Thus, the general pattern was that the studied species were more abundant in one or more rejects than in grazed patches although *L. corniculatus* was also abundant in grazed patches.

The effects of different rejects varied between sites as the interaction between structure and site was significant for three of the plant species (not *L. corniculatus*). For *L. pratensis* and *V. cracca* the effect of dung reject, grazing reject and rose shrubs varied between sites, but grazed patches had the lowest abundance at six of seven sites (data not shown). For *V. sepium* the effect of dung reject and grazing rejects varied between sites but abundance was highest in rose shrubs and lowest in grazed patches at all sites (data not shown).

Furthermore the abundances of all four study species were influenced by study site alone (Table 2). *V. cracca* had significantly higher abundance in one site compared with the other six sites (Table 3), but for the other species abundances appeared to vary randomly between sites (Table 3).

Flowering and fruit set

Flowering (flower heads/shoot) differed between structures for all species, but *V. sepium* (Table 2). In general, flowering was more frequent in rejects than in grazed patches although the importance of different rejects varied between species. Flowering of *L. pratensis* was more frequent in dung rejects, rose shrubs and grazing rejects than in grazed patches (Fig. 3A) and it was positively influenced by tall vegetation (Table 2). Flowering of *L. corniculatus* was more frequent in dung rejects and grazing rejects than in grazed patches (Fig. 2) and *V. cracca* flowered more frequently in rejects than in grazed patches (Fig. 3B). The interaction between structure and site was significant for *L. pratensis* and *V. sepium*, i.e. the effect on flowering of different types of structures varied between sites (Table 2). Although the effects of rejects varied between sites, grazed patches had lower flowering than rejects in all sites for both *L. pratensis* and *V. sepium* (data not shown).

Thus, both abundance and flowering were in general positively associated with different rejects, but for fruiting (pods/shoot) the effects of structures were less uniform. Structures alone significantly affected the number of pods/shoot for two species (Table 2). Fruiting was higher in grazing rejects and dung rejects than grazed patches for *V. cracca* (Fig. 3B). For *L. pratensis* mean values for pods/shoot appeared to vary between structures (Fig. 3A), but did not differ significantly, probably due to the large variations between sites (Table 3). For *L. corniculatus* pods/m^2 varied between structures and sites (Table 3) and dung rejects and grazing rejects produced a higher number of pods than grazed patches (Fig. 2). *V. sepium* did not differ between structures or sites in terms of pods/shoot (Fig. 3C). The effect of vegetation height was more obvious, with a significant positive effect on fruiting for *L. pratensis*, *L. corniculatus* and *V. cracca* (Table 2).

Seed set and seed predation

For seed predators the available resource, i.e. number of pods or seeds per m^2, could be expected to influence the oviposition choices. Since dung rejects and rose shrubs provided the best grazing refugees (Table 1) with numerous pods (Fig. 3) they should be attractive patches for oviposition. Predation rate did not differ between structures alone for the investigated plant species (Table 2) but for *V. sepium* the interaction between site and structure was significant and predation rate was either highest in rose shrubs or in grazing rejects depending on site.

Vegetation height significantly affected seed set and predation rate for *L. pratensis* (Table 2). Seed set was higher in short than in tall vegetation (Pearson correlation, N=489, r=-0.223, p<0.001, Fig. 4A). Predation rate was also higher in short than tall vegetation (Pearson correlation, N=454, r=-0.125, p=0.007). For *V. cracca*, seed set depended on vegetation height (Table 2) and the highest seed set was found in tall vegetation (Pearson correlation, N=167, r=0.163, p=0.036, Fig. 5), but no effect of vegetation height on predation rate was found. Predation of *L. corniculatus* was neither affected by pod length nor vegetation height. For *V. sepium*, predation rate varied with vegetation height (Table 2) but no positive or negative correlation could be found. For *L. pratensis*, pod length was positively correlated with predation rate (Pearson

Table 1. Mean vegetation height (cm ± SE) per study site in June and August measured in the sampling plots. Changes in % cover of the four structures between June and August. Data are sorted by mean vegetation height in August.

	Vegetation height (cm)		**Change in %**			
Study site	**June**	**August**	**Grazing reject**	**Dung reject**	***R. dumalis***	**Grazed patch**
Bergesta	5.8 ± 0.3	3.3 ± 0.2	-85	-57	-53	29
Långalma	5.1 ± 0.3	3.9 ± 0.2	-67	-27	-26	13
Rasbo	5.5 ± 0.2	4.0 ± 0.2	-37	-31	-34	20
Lagga	5.2 ± 0.2	5.2 ± 0.3	-42	-48	-59	21
Åsbergby	6.2 ± 0.3	5.2 ± 0.3	-54	-37	-42	44
Hagby	8.3 ± 0.4	6.1 ± 0.3	-28	8	-15	28
Tvärnö	7.3 ± 0.3	9.1 ± 0.3	0	0	0	0
Mean			-52	-32	-38	25

Table 2. Parameters tested with ANCOVA with structure as fixed factor, site as random factor and vegetation height a covariate for *Lathyrus pratensis*, *Vicia sepium* and *Lotus corniculatus*. *Vicia cracca* parameters (dung pats) were tested with ANOVA with site as fixed factor and vegetation height as random factor. *F*-values are shown and significant values are bold. ***= p<0.001, **=p<0.01. *=p<0.05.

	Site	Structure	Interaction	Veg. height	df site	df struc.	df inter.	df veg.
Plot vegetation height	**3.1 ***	**8.3 *****	**5.8 *****		6	3	17	
L. pratensis								
Shoots/m^2	**8.1 *****	**4.2 ***	**4.6 *****	0,4	6	3	17	1
Flower heads /shoot	**3.3 ***	**8.0 *****	**2.8 *****	**29.8 *****	6	3	17	1
Pods/shoots	**2.9 ***	1,0	**3.1 *****	**10.4 *****	6	3	17	1
Pod length	1,1	0,1	**2.6 ***	1,6	3	2	5	1
Seed set (%)	2,2	1,2	2,2	**6.3 ***	3	2	5	1
Sum predated seeds (%)	**41.6 *****	0,4	0,7	**7.1****	3	2	5	1
Apion subulatum (%)	2,0	0,6	0,6	0,1	3	2	5	1
Bruchus loti (%)	**18.3 ****	0,2	2,0	2,8	3	2	5	1
Hymenoptera (%)	1,5	0,5	**2.5 ***	0,04	3	2	5	1
L. corniculatus								
Shoot/m^2	**7.4 *****	**3.5 ***	1,6	**9.4 ****	5	3	15	1
Flower heads /shoot	1,0	**6.0 ***	1,8	0,1	4	3	4	1
Pods/shoots	**8.0 ***	**4.0 ***	1,4	**4.7 ***	4	3	4	1
Pod length	1,0	0,001	2,3	0,4	1	1	1	1
Seed set (%)	1,9	0,2	0,8	0,5	1	1	1	1
Apion loti (%)	0,3	0,1	1,7	0,8	1	1	1	1
Hymenoptera (%)	5,4	1,7	0,4	0,5	1	1	1	1
V. cracca								
Shoots/m^2	**13.4 *****	2,5	**3.6 *****	3,0	6	3	17	1
Flower heads /shoot	0,5	**5.4 ****	**1.7 ***	2,6	6	3	16	1
Pods/shoots	1,9	**3.4 ***	1,6	**4.5 ***	6	3	16	1
Pod length	**6.0 *****			0,2	4			1
Seed set (%)	**5.5 *****			**7.3 ****	4			1
Apion cerdo (%)	**12.4 *****			2,9	4			1
Hymenoptera (%)	1,8			0,9	4			1
V. sepium								
Shoot/m^2	**3.3 ***	**17.6 *****	**2.2 ***	2,0	4	3	11	1
Flower heads /shoot	**3.4 ***	0,9	0,3	1,5	4	3	9	1
Pods/shoots	2,1	2,4	0,5	1,2	4	3	9	1
Pod length	12,6	0,7	1,0	0,02	2	1	1	1
Seed set (%)	9,6	10,2	0,8	2,6	2	1	1	1
Bruchus atomarius (%)	0,03	0,01	**43.4 *****	**7.7 ****	2	1	1	1
Hymenoptera (%)	0,0	0,0	**4.3 ***	2,1	2	1	1	1

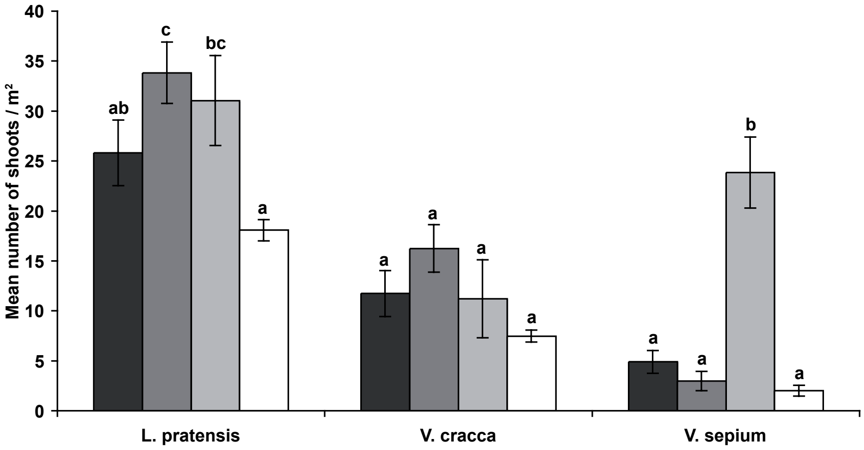

Fig. 1. Mean number of shoots per m^2 ± SE in June for *Lathyrus pratensis*, *Vicia cracca* and *Vicia sepium* shown for four spatial structures: grazing reject (dark grey), dung reject (grey), *Rosa dumalis* (light grey) and grazed patch (white). Mean values with different letters show significant differences, $p<0.05$, between structures, species are analysed separately.

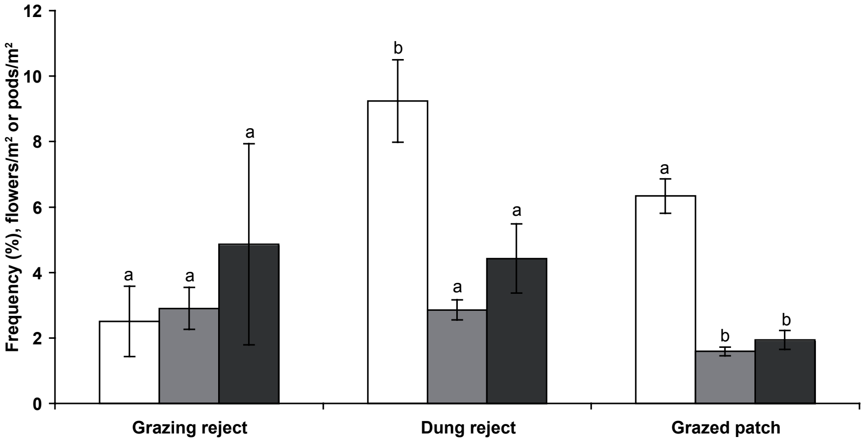

Fig. 2. Mean frequency (% area ± SE) of *Lotus corniculatus* (white bars), mean number of flower heads/m^2 ± SE (grey bars) and pods/m^2 ± SE (dark grey bars). No *L. corniculatus* plants were found in rose shrubs. Means noted with different letters show significant differences between structures at the 0.05-level respectively for abundance, flowering and fruiting.

correlation, N=489, r=0.166, $p<0.001$, Fig. 4B). No correlations for the other three species were found. Pod length did not differ between sites and structures (Table 2) but pod length correlated with seed set and number of seeds per pod for all species (Pearson correlation, $p<0.001$ in all cases).

In general *L. pratensis* and *V. sepium* seeds were more predated (in total 47 ± 2% versus 43 ± 4%) than *L. corniculatus* and *V. cracca* seeds (20 ± 5% versus 11 ± 2%).

Table 3. Mean ± SE shoots/m^2, flower heads/shoot and pods/shoot shown for the seven study sites, sorted by mean site vegetation height with intensively grazed sites to the left and low-intensively grazed sites to the right. Means noted with different letters indicate significant differences between sites at the 0.05 level.

	Bergesta	**Långalma**	**Rasbo**	**Lagga**	**Åsbergby**	**Hagby**	**Tvärnö**
Lathyrus pratensis							
Shoots/m^2	32 ± 3 **ac**	39 ± 4 **c**	22 ± 3 **d**	8 ± 1 **b**	5 ± 1 **b**	23 ± 2 **ad**	37 ± 4 **c**
Flowers/shoot	0.2 ± 0.03 **abce**	0.04 ± 0.01 **d**	0.3 ± 0.1 **be**	0.1 ± 0.04 **cd**	0.3 ± 0.1 **abce**	0.3 ± 0.04 **be**	0.2 ± 0.04 **acd**
Pods/shoot	0.2 ± 0.04 **abcd**	0.01 ± 0.01 **c**	0.4 ± 0.1 **d**	0.2 ± 0.1 **abcd**	0.04 ± 0.04 **abc**	0.1 ± 0.02 **bc**	0.6 ± 0.3 **ad**
Lotus corniculatus							
Frequency (%)	15 ± 2 **a**	3 ± 1 **bc**	9 ± 1 **d**	6 ± 1 **cd**	0.5 ± 0.2 **b**	0.3 ± 0.2 **b**	
Flowers/m^2	2 ± 0.2	1 ± 0.2	2 ± 0.3	2 ± 0.3			
Pods/m^2	1 ± 0.3 **a**	1 ± 1 **a**	3 ± 1 **a**	6 ± 1 **b**			
Vicia cracca							
Shoots/m^2	2 ± 1 **a**	32 ± 4 **d**	8 ± 1 **abc**	8 ± 1 **bc**	11 ± 2 **c**	6 ± 1 **abc**	2 ± 0.5 **ab**
Flowers/shoot	0.3 ± 0.2	0.2 ± 0.03	0.2 ± 0.1	0.2 ± 0.1	0.4 ± 0.1	0.5 ± 0.1	0.3 ± 0.1
Pods/shoot	0.5 ± 0.3	0.04 ± 0.02	0.4 ± 0.2	0.4 ± 0.2	0.1 ± 0.1	0.4 ± 0.1	0.5 ± 0.3
Vicia sepium							
Shoots/m^2	5 ± 2 **a**		1 ± 1 **a**		9 ± 2 **b**	3 ± 1 **a**	12 ± 2 **b**
Flowers/shoot	0.02 ± 0.02		0		0.2 ± 0.1	0.2 ± 0.1	0.1 ± 0.1
Pods/shoot	0.1 ± 0.1		0.3 ± 0.2		0.6 ± 0.2	0.4 ± 0.1	0.2 ± 0.1

Discussion

The studied spatial structures can be seen as temporary or persistent and the duration of grazing refugees partly depend on grazing intensity. Dung pats can last for one or two grazing seasons and when the dung pats are decomposed they are overgrown by vegetation and the rejects can be grazed (White *et al.*, 2001). Grazing rejects occurred mainly in the early summer, especially in intensively grazed pastures (Table 1), and they rarely last more than one grazing season (Brunsell, 2002). Although short lived, dung pats and to a lesser degree grazing rejects affected plant abundance. The higher abundances in dung rejects might be due to increased growth rate following the higher nutrient availability near dung pats (Shiyomi *et al.*, 1998, Bullock & Marriot, 2000). The higher abundances in grazing rejects are probably an effect of larger plant individuals and not to establishments of new plant individuals. Rose shrubs are long-lived structures and, as expected, they strongly affected plant abundances. The abundance of *V. sepium* was highest in rose shrubs, and the strong association to shrubs may be due to the climbing growth form, sensitivity to intensive grazing and high solar radiation. In contrast, rose shrubs had an overall negative effect on *L. corniculatus*, both on abundance and reproduction. *L. corniculatus* was also negatively affected by tall vegetation and short species are often less competitive than taller species and may therefore be negatively affected by both shrubs and tall vegetation (Stephenson *et al.*, 1988; Diaz *et al.*, 2001). Grazed patches were common and covered large areas of the studied pastures (43-72% in June and 56-86% in August) and a random patch is probably more likely to be grazed than rejected over time and grazing tolerant species may increase in abundance. *L. corniculatus* was the only species with relatively high abundance in grazed patches, and short species are often more resistant to grazing than tall species (Diaz *et al.,* 2001; Cingolani *et al.*, 2005). Although not studied here, many other species, e.g. grasses and rosette species, may be positively affected by grazed patches (Dupre & Diekmann, 2001; Svensson & Glimskär, 1990; Hanson & Fogelfors, 2000; Wahlman & Milberg, 2002).

Reproduction was positively influenced by grazing refugees for three species but no for *V. sepium*. The reproduction of *L. pratensis*, *L. corniculatus* and *V. cracca* was higher in dung rejects than grazed patches

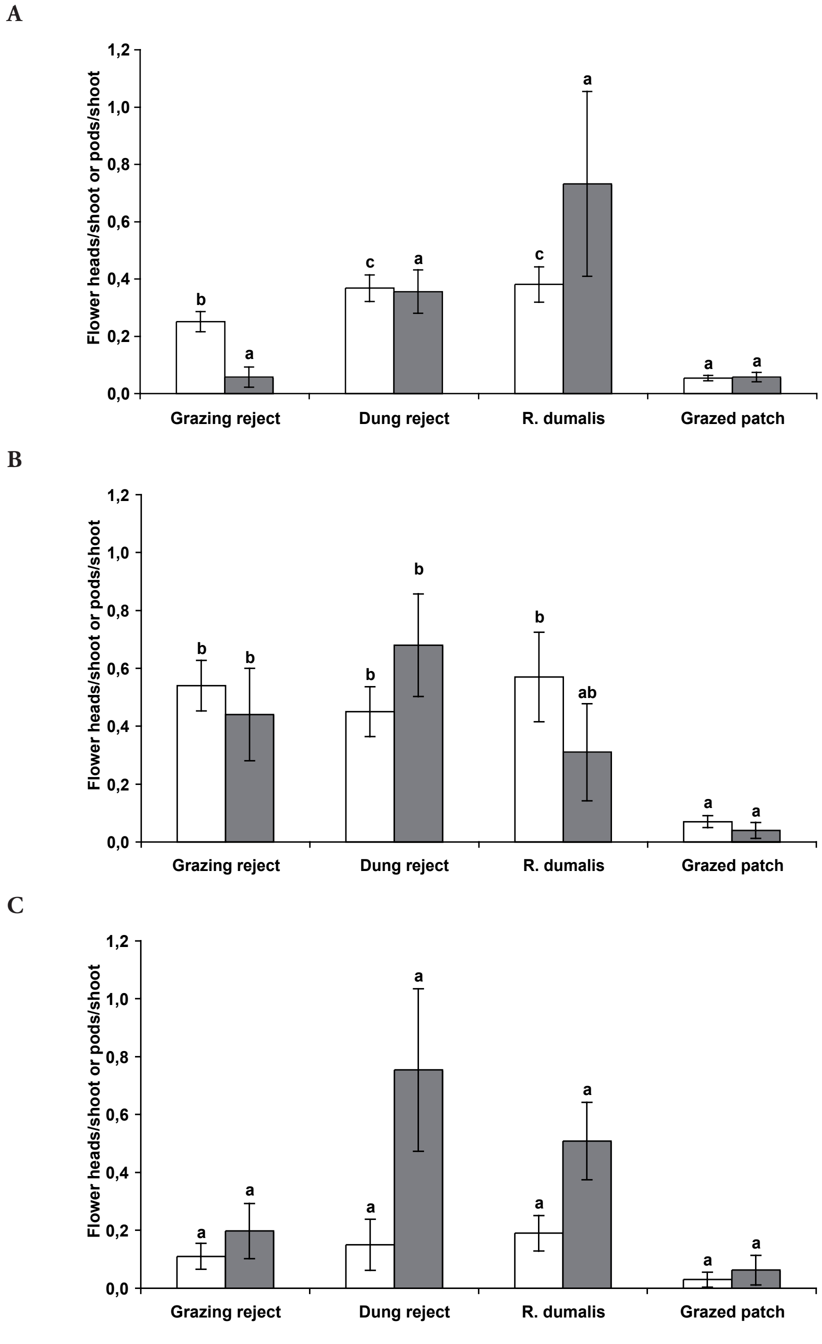

Fig. 3. Mean number of flowering heads/shoot ± SE (white bars) and pods/shoot ± SE (grey bars) for *Lathyrus pratensis* (**A**), *Vicia cracca* (**B**) and *Vicia sepium* (**C**) shown for four spatial structures; grazing reject, dung reject, *Rosa dumalis* and grazed patch. Means noted with different letters show significant differences between structures at the 0.05-level, flowering and fruiting are analysed separately.

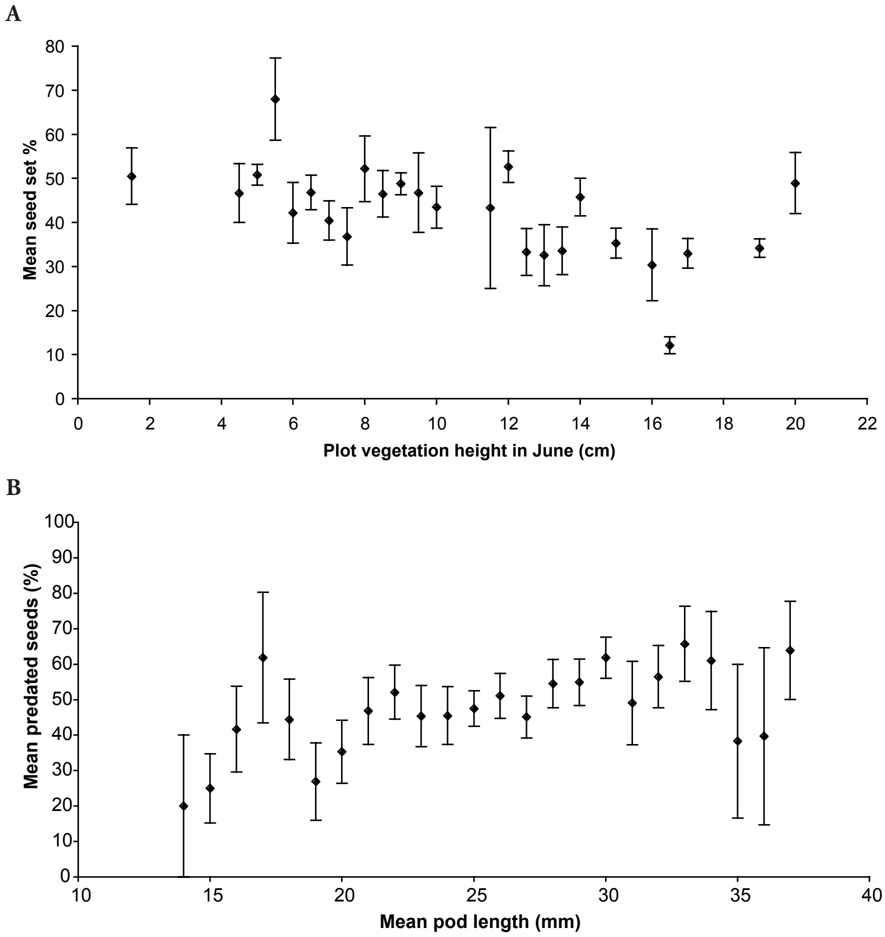

Fig. 4A. Mean ± SE seed set (%) per pod for *L. pratensis* correlated with plot vegetation height in June (cm). **B.** Mean ± SE predated seeds (%) of *L. pratensis* correlated with mean pod length (mm). Both correlations are significant at the 0.001 level.

and the reproduction of *L. pratensis* and *V. cracca* was higher in rose shrubs than in grazed patches. This is probably an effect of grazing avoidance near these structures. For *V. sepium*, flowering and fruiting were not significantly higher in rose shrubs than in other structures. However, the abundance was highest in rose shrubs and the reproduction in terms of number of flowers and pods is probably highest in rose shrubs. Reproduction of *L. pratensis*, *L. corniculatus* and *V. cracca* was positively affected by tall vegetation. Vegetation height was higher in rejects than in grazed patches and it can be difficult to separate the variation between structures from vegetation height.

Beside spatial structures and vegetation height, the abundance and reproduction of all species varied between sites. Differences between sites can be due to factors such as geographic position, soil conditions, local climate and historical land use and for plant reproduction; a major difference was that the sites were selected to represent a gradient from intensively grazed to ungrazed pastures.

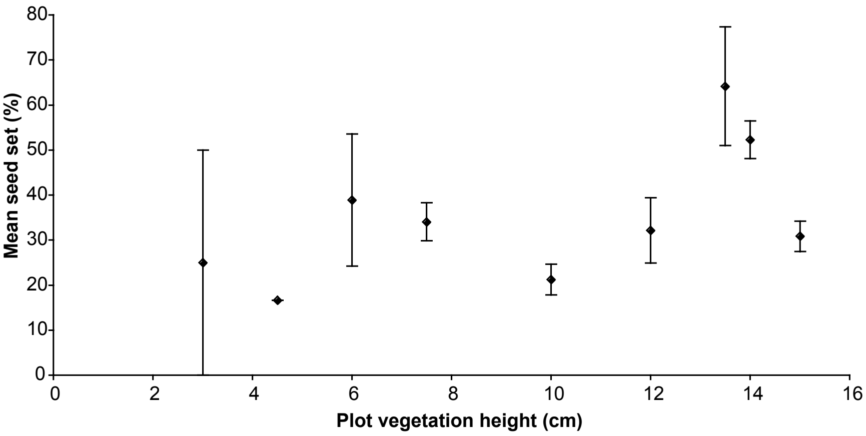

Fig. 5. Mean ± SE seed set (%) for *V. cracca* compared with plot vegetation height (cm). Correlation is significant at the 0.05 level.

Seed set and seed predation

At the end of the summer, pods were mainly found near dung rejects and rose shrubs but predation rate did not vary between the different structures. Almost no pods were found in grazed patches and therefore no comparison with grazed patches could be done for any of the four species. In grazing rejects pods were available in June but in August many pods had been grazed and therefore grazing rejects may be a trap for seed predators. The expected preferences for rejects (with large available seed resources) among seed predators could not be supported (Östergård & Ehrlen, 2005). However, the density of seed predators had a potential to become much higher in rejects than in grazed patches, especially in rose shrubs and dung pats. This indicates that presence of shrubs or lowered grazing intensity in semi-natural grasslands may increase available resources for seed predators and thereby seed predator densities.

Seed set varied with vegetation height for two species. Seed set for *L. pratensis* increased in short vegetation and in contrast, seed set for *V. cracca* increased in taller vegetation. Flowering is higher in tall than short vegetation and flower densities can influence pollinators and thereby seed set and the differences in seed set are probably an effect of pollination and not vegetation height (Cariveau *et al.*, 2004). For *L. pratensis*, predation rate was higher in long pods with many seeds indicating that females chose long pods with many seeds when they oviposit. For *L. corniculatus* no difference in pod length between predated and unpredated pods were found and this result is supported by Ollerton & Lack (1996). For *V. sepium* a positive correlation between pod length and seed predation rate was expected, as found by Lenoir & Pihlgren (2006), but no significant correlation was found. For *L. pratensis* seed predation was higher in short vegetation and this is probably an effect of seed set, which was higher in short vegetation. Vegetation height also reflects host plant height and the quantity of available flowers and pods, factors known to influence seed predation (Brody & Waser, 1995; Cariveau *et al.*, 2004). Seed predators may also manipulate their host plant to ensure fruit set (Brody & Morita, 2000). Moreover, Östergård & Ehrlen (2005) showed that predation by seed predators were mainly determined by the host plant population size and number of flowering host plants.

Conclusions

In conclusion, the abundance and reproduction varied between species and spatial structures. Rejects were important for the reproductive success of vascular plants in semi-natural pastures, especially rose shrub rejects and dung rejects. Reproduction was higher in tall than short vegetation. The density of seed predators had potential to become much higher in rose shrubs and dung rejects than grazed patches but no differences in predation rates were found. Grazing intensity affected the quantity of rejects in the studied pastures. Therefore grazing and regulation of grazing intensity and shrubs densities are powerful tools for managing grassland species, both plants and insects.

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References

Bailey, D. W., Dumont, B. & Wallis DeVries, M. F. 1998. Utilization of heterogeneous grasslands by domestic herbivores: Theory to management. *Annales de Zootechnie 47*, 321-333.

Bakker, J. P. 1989. *Nature Management by Grazing and Cutting* Kluwer Academic Publishers, Dordrecht.

Bakker, E. S., Olff, H., Vandenberghe, C., de Maeyer, K., Smit, R., Gleichman, J. M. & Vera, F. W. M. 2004. Ecological anachronisms in the recruitment of temperate light-demanding tree species in wooded pastures. *Journal of Applied Ecology 41*, 571-582.

Brody, A. K. & Morita, S. I. 2000. A positive association between oviposition and fruit set: female choice or manipulation? *Oecologia 124*, 418-425.

Brody, A. K. & Waser, N. M. 1995. Oviposition patterns and larval success of a pre-dispersal seed predator attacking two confamilial host plants. *Oikos 74*, 447-452.

Brunsell, M. 2002 Betesrator och kärlväxters reproduktion: En studie av betesmarkers heterogenitet i tid och rum. Examensarbete i Naturvårdsbiologi 72, SLU, Uppsala.

Callaway, R. M. 1995. Positive interactions among plants. *Botanical Review 61*, 306-349.

Cariveau, D., Irwin, R. E., Brody, A. K., Garcia-Mayeya, L. S. & der Ohe, A. 2004. Direct and indirect effects of pollinators and seed predators to selection on plant and floral traits. *Oikos 104*, 15-26.

Cingolani, A. M., Posse, G. & Collantes, M. B. 2005. Plant functional traits, herbivore selectivity and response to sheep grazing in Patagonian steppe grasslands. *Journal of Applied Ecology 42*, 50-59.

Correll, O., Isselstein, J. & Pavlu, V. 2003. Studying spatial and temporal dynamics of sward structure at low stocking densities: the use of an extended rising-plate-meter method. *Grass Forage Science 58*, 450-454.

Diaz, S., Noy-Meir, I. & Cabido, M. 2001. Can grazing response of herbaceous plants be predicted from simple vegetative traits? *Journal of Applied Ecology*, *38*, 497-508.

Dupre, C. & Diekmann, M. 2001. Differences in species richness and life-history traits between grazed and abandoned grasslands in southern Sweden. *Ecography 24*, 275-286.

Freude, H., Harde, K. W. & Lohse, G. A. 1981. *Die Käfer Mittlereuropas*. Goecke & Evers Verlag, Krefeld.

Gibson, C. W. D., Brown, V. K., Losito, L. & McGavin, G. C. 1992. The response of invertebrate assemblies to grazing. *Ecography 15*, 166-176.

Hansson, M. & Fogelfors, H. 2000. Management of a semi-natural grassland; results from a 15-year-old experiment in southern Sweden. *Journal of Vegetation Science 11*, 31-38.

Jensen, K. & Gutekunst, K. 2003. Effects of litter on establishment of grassland plant species: the role of seed size and successional status. *Basic and Applied Ecology 4*, 579-587.

Jerling, L. & Andersson, M. 1982. Effects of selective grazing by cattle on the reproduction of Plantago maritima. *Holarctic ecology 5*, 405-411.

Jones, R. M. & Ratcliff, D. 1983. Patchy grazing and its relation to deposition of cattle dung pats in pastures in coastal subtropical Queensland. *The Journal of the Australian Institute of Agricultural Science 49*, 109-111.

Lalonde, R. G. & Roitberg, B. D. 1994. Pollen availability, seed production and seed predator clutch size in a tephritid thistle system. *Evolutionary Ecology 8*, 188-195.

Lawton, J. H. 1983. Plant architecture and the diversity of phytophagous insects. *Annual Review of Entomology 28*, 23-39.

Lennartsson, T. 2000. Management and population viability of the pasture plant Gentianella campestris: the role of interactions between habitat factors. *Ecological Bulletins 48*, 111-121.

Lenoir, L. & Pihlgren, A. 2006. Effects of grazing and ant/beetle interaction on seed production in the legume *Vicia sepium* in a semi-natural grassland. *Ecological Entomology 6*, 601-607.

Morris, M. G. 1967. Differences between the invertebrate faunas of grazed and ungrazed chalk grassland I. Responses of some phytophagous insects to cessation of grazing. *Journal of Applied Ecology 36*, 459-474.

Morris, M. G. 2000. The effects of structure and its dynamics on the ecology and conservation of arthropods in British grasslands. *Biological Conservation 95*, 129-142.

Mortimer, S., Hollier, J. & Brown, V. K. 1998. Interactions between plant and insect diversity in the restoration of lowland calcareous grasslands in southern Britain. *Applied Vegetation Science 1*, 101-114.

Mossberg, B. & Stenberg, L. 2003. *Den nya nordiska floran*. Wahlström & Widstrand, PDC Tangen, Norge.

Olff, H. & Ritchie, M. E. 1998. Effects of herbivores on grassland plant diversity. *Trends in Ecology & Evolution 13*, 261-265.

Ollerton, J. & Lack, A. 1996. Partial predispersal seed predation in *Lotus corniculatus* L (Fabaceae). *Seed Science Research 6*, 65-69.

Persson, K. 2005. Ängs- och betesmarksinventeringen 2002-2004. *Jordbruksverket, 2005:1*.

Proulux, M. & Mazumder, A. 1998. Reversal of grazing impact on plant species richness in nutrient-poor vs. nutrient-rich ecosystems. *Ecology 79*, 2581-2592.

Pöyry, J., Luoto, M., Paukkunen, J., Pykälä, J., Raatikainen, K. & Kuussaari, M. 2006. Different responses of plants and herbivore insects to a gradient of vegetation height: an indicator of the vertebrate grazing intensity and successional age. *Oikos 115*, 401-412.

Pärtel, M. & Zobel, M. 1999. Small-scale plant species richness in calcareous grasslands determined by the species pool, community age and shoot density. *Ecography 22*, 153-159.

Rousset, O. & Lepart, J. 2003. Neighbourhood effects on the risk of an unpalatable plant being grazed. *Plant Ecology 165*, 197-206.

Shiyomi, M., Okada, M., Takahashi, S. & Tang, Y. H. 1998. Spatial pattern changes in aboveground plant biomass in a grazing pasture. *Ecological Research 13*, 313-322.

Siemann, E., Tilman, D., Haarstad, J. & Ritchie, M. 1998. Experimental tests of the dependence of arthropod diversity on plant diversity. *American Naturalist 152*, 738-750.

Söderström, P. 1993. Ängs- och hagmarker i Uppsala län. *Länsstyrelsen Uppsala län, 1993:3*.

Siemann, E., Haarstad, J. & Tilman, D. 1999. Dynamics of plant and arthropod diversity during old field succession. *Ecography 22*, 406-414.

Stephenson, A. G., Johnson, R. S. & Winsor, J. A. 1988. Effects of competition on the growth of *Lotus corniculatus* L. seedlings produced by random and natural patterns of fruit abortion. *The American Midland Naturalist 120*, 102-107.

Svensson, R. & Glimskär, A. 1990. *Vegetation changes following fertilization and changed management.* SLU, Department of Ecology and Environmental Research, 38.

Vanbergen, A. J., Hails, R. S., Watt, A. D. & Jones, T. H. 2006. Consequences for host-parasitoid interactions of grazing-dependent habitat heterogeneity. *Journal of Animal Ecology 75*, 789-801.

Wahlman, H. & Milberg, P. 2002. Management of semi-natural grassland vegetation: evaluation of a long-term experiment in southern Sweden. *Annales Botanici Fennici 39*, 159-166.

WallisdeVries, M. F., Poschlod, P. & Willems, J. H. 2002. Challenges for the conservation of calcareous grasslands in northwestern Europe: integrating the requirements of flora and fauna. *Biological Conservation 104*, 265-273.

White, S. L., Sheffield, R. E., Washburn, S. P., King, L. D. & Green, J. T. 2001. Spatial and time distribution of dairy cattle excreta in an intensive pasture system. *Journal of Environmental Quality 30*, 2180-2187.

Wissman, J. 2006. Grazing regimes and plant reproduction in semi-natural grasslands. Swedish University of Agricultural Sciences.

Woodcock, B. A., Pywell, R. F., Roy, D. B., Rose, R. J. & Bell, D. 2005. Grazing management of calcareous grasslands and its implications for the conservation of beetle communities. *Biological Conservation 125*, 193-202.

Östergård, H. & Ehrlen, J. 2005. Among population variation in specialist and generalist seed predation - the importance of host plant distribution, alternative hosts and environmental variation. *Oikos 111*, 39-46.