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Unpalatable plants facilitate tree sapling survival in wooded pastures

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Summary

1. In endangered wooded pasture ecosystems established tree saplings are frequently found in spatial association with protective structures, suggesting nurse effects. This associational resistance is thought to be a driving force behind tree regeneration in wooded pastures. Experimental evidence for associational resistance is, however, scarce. We studied the effects of unpalatable plants on tree sapling performance in a wooded pasture ecosystem in Switzerland and tested the associational resistance hypothesis experimentally.

2. We transplanted 600 *Picea abies* saplings of two size classes (5 cm and 10 cm) in plots: (i) with unpalatable plants (either *Gentiana lutea* or *Cirsium acaule*, representing a chemically and mechanically defended species, respectively); (ii) with one of both unpalatable plants clipped to the ground; and (iii) without any unpalatable plants. Grazing intensity was measured once in each plot and tree sapling performance was followed for 2 consecutive years.

3. Grazing intensity was significantly higher in plots without unpalatable plants, and significantly higher in plots with *Cirsium* than with *Gentiana*. Correspondingly, sapling survival (21·5% and 10·6% after 1 and 2 grazing years, respectively) was significantly higher near unpalatable plants, and significantly higher in plots with *Gentiana* than with *Cirsium*. Large tree saplings survived significantly better than small ones, and depended less on the unpalatable plants for survival.

4. Except for saplings planted near *Gentiana*, sapling removal by cows and horses was the major cause of death, the extent varying between unpalatable species and treatments.

5. The growth in height of the saplings was unaffected by the treatments.

6. *Synthesis and applications.* This study shows that unpalatable plants can enhance tree regeneration in wooded pastures. Sapling survival was significantly higher near unpalatable plants where grazing intensity was significantly lower. Protection against cattle is particularly important for small saplings. These results have important management implications for the endangered and disappearing wooded pastures in western Europe. Transplanting tree saplings near unpalatable plants could be an alternative reforestation technique in intensively grazed wooded pastures. On the other hand, removal of unpalatable plants and shrubs might prevent or slow down undesired tree encroachment in less intensively grazed areas.

Key-words: associational resistance, cattle, *Cirsium acaule*, *Gentiana lutea*, grazing, *Picea abies*, tree regeneration

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Introduction

An increasing number of studies have indicated that positive interactions among plant species may drive community function and structure in various ecosystems.

Recent examples come from ecosystems such as high mountain vegetations (Nunez, Aizen & Ezcurra 1999; Choler, Michalet & Callaway 2001), deserts (Munzbergova & Ward 2002), forests (Arevalo & Fernandez-Palacios 2003) and pastures (Callaway, Kikvidze & Kikvidze 2000). Such spatial associations suggest facilitative effects, as one species directly or indirectly ameliorates the environment of the other. Understanding such interactions between plants helps to clarify the structure, diversity and dynamics of plant communities and ecosystems (Castro, Zamora & Hodar 2002). So far, most evidence for these facilitative interactions between plants comes from descriptive studies, and empirical evidence is rare but needed, especially for grazed ecosystems (Callaway, Kikvidze & Kikvidze 2000).

Wooded pastures are traditional semi-natural ecosystems, extensively grazed and logged, with natural regeneration of both grassland and woodland. Today, they are among the most endangered ecosystems in Europe, threatened by both intensification and abandonment of management. Wooded pastures have probably been developed by humans since Neolithic times (Pott & Hüppe 1991) but others suggest a natural origin via now extinct large herbivores as *Bos primigenius* and *Equus ferus* (Vera 2000). Remnants of this once-abundant system are nowadays well known for their high conservation values, and serve as examples for nature development projects in western Europe, where large herbivores are increasingly reintroduced in former agricultural areas in order to increase biodiversity (Olff *et al*. 1999). While an understanding of the mechanisms regulating biodiversity maintenance is a prerequisite for sustainable management in these systems, it is often lacking.

It has been proposed that facilitation by grazing-resistant plants, also known as associational resistance (Bakker *et al*. 2004), associational avoidance (Milchunas & Noy-Meir 2002) and defence guilds (Atstatt & Odowd 1976), is one of the processes driving tree establishment in wooded pastures (Olff *et al*. 1999; Bokdam & Gleichman 2000). Various studies (Rousset & Lepart 1999; Kuiters & Slim 2003; Vera 2000; Smit *et al*. 2005) report spatial associations between young trees and unpalatable plants, possibly indicating establishment of trees via facilitator species, but empirical evidence for associational resistance in these systems is scarce (Pages & Michalet 2003; Tirado & Pugnaire 2003). Furthermore, factors affecting associational resistance between tree saplings and unpalatable plants are largely unknown.

Plants have evolved various traits against herbivory, such as chemical, morphological and phenological defence mechanisms, and these vary in their effectiveness (Crawley 1983). As a consequence, the survival probability of spatially associated tree saplings may depend on the effectiveness of the facilitator's defence strategy and vary among facilitator plants with different defence mechanisms.

Sapling size affects the probability of being grazed by herbivores, with small tree saplings less likely to be discovered by herbivores and therefore less likely to be damaged (Rao *et al*. 2003). Consequently, small tree saplings may need less protection against browsing than larger tree saplings. Alternatively, large tree saplings may be easier to detect and hence avoided (when not palatable to herbivores), in contrast with small tree saplings which may be grazed accidentally together with surrounding, preferred vegetation. Thus, associational resistance may depend on the size of the tree sapling.

Tree saplings growing near unpalatable plants may be protected effectively against cattle grazing but could suffer more from competition with their facilitators (for light, nutrients, water, etc.) than tree saplings growing in open swards or grassland (Berkowitz, Canham & Kelly 1995; Rousset & Lepart 2000). Accordingly, growth of protected tree saplings may be reduced as a result of competition with their facilitators, leading to a trade-off between protection against grazers and being limited in growth by unpalatable plants.

In this field study we tested the proposed facilitative effects of a physically and chemically defended unpalatable species on tree sapling performance. We transplanted *Picea abies* (L.) Karst. saplings of two size classes in plots with and without unpalatable plants, and in plots with unpalatable plants removed to ground level. Tree sapling performance was followed throughout two grazing seasons. In accordance with the associational resistance theory, we hypothesized that tree saplings planted in plots with unpalatable plants would have higher survival but reduced growth rates. Furthermore, we expected to find differences in protective effects of the two unpalatable species and different performances of the two size classes of tree saplings.

Methods

STUDY AREA

This study was conducted in the Swiss Jura Mountains. The climate is generally continental with Atlantic influences. Mean yearly temperature varies between 3 °C and 5 °C. Annual precipitation lies between 1400 mm and 2000 mm in this region (with a peak in summer) but soils are considered to be relatively dry because of the high permeability of the underlying calcareous bedrock. Our study site (6°12′N, 46°32′E) is a wooded pasture of approximately 12 ha at an altitude of 1340 m a.s.l. It is a typically species-rich grassland of the *Mesobrometum* type, with scattered individuals or small groups of *P. abies* trees and a mean tree cover of *c*. 40%. The study site is situated in the Pré-aux-Veaux, an extensively grazed wooded pasture area (102 ha) belonging to the larger Parc Jurassien Vaudois. The area is typically grazed from early June until early October according to a rotation system, with three to four grazing rotations per year, each lasting approximately 10 days. The average herd consists of *c*. 58 heifers, occasionally including five to 10 horses at the end of the grazing season. Because of the extreme hot weather conditions during the first year (2003), the third grazing rotation was extended and fences between paddocks were opened in order to enlarge the grazed area (in this case 110 ha grazed for 33 days).

Naturally occurring large vertebrate herbivores in the site are rarely seen but include roe deer *Capreolus capreolus* L. and mountain hare *Lepus timidus* L.

TRANSPLANTATION EXPERIMENT

Our experiment was conducted with 600 1- and 2-year-old *P. abies* saplings, which were obtained from a local tree nursery. Seed material originated from a site in the Swiss Jura Mountains at 1400 m a.s.l. near the study site. In autumn 2002 the tree saplings were taken from their plant beds and put in plastic trays (individual pot per tray $3 \times 3 \times 6$ cm) filled with nutrient-rich peaty soil, after the taproots were cut back to 5 cm to stimulate root growth. The saplings over-wintered outside in the trays, protected from severe frost by a thick leaf layer of ±15 cm.

We selected *Gentiana lutea* L. and *Cirsium acaule* Scop. as unpalatable plants. *Gentiana* and *Cirsium*, measuring between 50 and 120 cm and 10 and 30 cm in height, respectively, are both perennial species from calcareous mountain grasslands and occur commonly in wooded pastures in the Swiss Jura Mountains. Cattle generally avoid these plants (C. Smit, personal observation) because they contain toxic elements (*Gentiana*) or are spiny (*Cirsium*). Naturally established tree saplings of *P. abies* have been found to be positively associated with both species, indicating nurse effects (Smit *et al*. 2005).

We randomly selected an equal number of plots (*c*. 200) with *Gentiana* and *Cirsium* or without any unpalatable plant (or shrub, rocky outcrop or tree) within a 1·5-m radius around the plot. The *Gentiana* and *Cirsium* plots contained more than one shoot (ramet) with patch sizes measuring *c*. 30 cm and 20 cm, respectively. Half of the plots with *Gentiana* and *Cirsium* were randomly selected for a clipping treatment: canopies of the unpalatables were removed by cutting the stems at soil level (leaving surrounding vegetation intact). This was repeated periodically during the season because of regrowth of the plants. The cutting treatment was added to test for the potential effects of environmental conditions (litter, soil structure, etc.) other than those relating to changes in the microclimate under the canopy. All 600 plots were at least 1·5 m apart and were distributed evenly over the entire study site in order to avoid spatial clumping.

In May 2003, all 1- and 2-year-old *Picea* saplings were transplanted in the selected plots: (i) near either *Gentiana* or *Cirsium* at ±10 cm from their stem; (ii) near the cut *Gentiana* or *Cirsium*; and (iii) with no unpalatable plant nearby. Each possible combination between unpalatable species (2), treatment (3) and sapling size (2) was repeated 50 times. Tree saplings that died during the 2 weeks following transplantation (29 in total, most probably because of a severe spring frost on 15 May) were replaced with new saplings in the original positions. At the start of the experiment (31 May) the 1- and 2-year-old saplings measured 4·85 cm (± 1·57) and 10·12 cm (± 3·81), respectively (means ± SD). The average vegetation height during the grazing seasons was 5 cm (data not shown) so we assumed that the 1-year-old-saplings (hereafter called small) would be less apparent than the 2-year-old saplings (hereafter called large).

MEASUREMENTS

Grazing intensity was measured after the first grazing rotation period in 2003, by estimating the percentage cover of grazed and cropped shoots or shoot tips in a 40-cm circle around the plot. Shoots of *Gentiana* and *Cirsium* were excluded for this estimation.

Tree sapling performance was followed for two consecutive grazing seasons (2003 and 2004). Measurements on the tree saplings were carried out six times in total by the same person: before the first cattle arrival (i); after each rotation period of the first season (ii, iii and iv); before cattle arrival for the second season (v); and at the end of the second grazing season (vi). Each time presence–absence of the saplings, their height (from ground level to the highest green needle) and survival were recorded. Saplings were recorded as dead when there was no more visible chlorophyll. In the first season, we distinguished between dead standing and removed tree saplings: tree saplings that were not found back at their original planting position (absent) were considered removed by the cows and, hence, dead. Tree saplings that were not removed by the cows but dead standing were recorded as present and dead. We considered removal of tree saplings as the most direct effect of cattle grazing, whereas the performance of non-removed tree saplings could depend on various direct and indirect effects of cattle as well as microenvironmental conditions. In the second grazing season, dead standing and removed saplings could not be distinguished because many rotted away during winter, so only tree sapling survival was recorded.

ANALYSES

Differences in grazing intensity between unpalatable species, sapling sizes and treatments were tested using ANOVA. The response variable grazing intensity was $\log 10(x + 1)$ transformed to satisfy the assumptions for normality. We used Tukey HSD tests for comparing pairs of group means of significant variables.

Cox regression was used to analyse and compare the effects of the treatments on survival of the tree saplings over the entire range of the study period (2 years). The start of the experiment was set at 31 May 2003 and measurements were taken at 52, 66, 129, 373 and 498 days. For all the tree saplings that died a 'mid-time' value was calculated, in order to approximate time of death. This was done by taking the average between time of first death observation and the previous census (when the sapling was still alive), resulting in 26, 59, 98, 251 and 436 days.

In order to analyse the effects of the three categorical predictors (treatment, species and sapling size) on sapling survival after one and two grazing seasons, we fitted generalized linear models (GLM) to our data with a binomial error distribution and a logit link function. The predictor variables size (two), species (two) and treatment (three) were added sequentially, including all possible two- and three-way interactions. Chi-square statistics were used for the calculation of the significance of the factors in the models. We used Bonferroni-corrected tests for comparing group means of significant variables and interactions.

With the aim of assessing the importance of sapling removal as cause of death (only after first grazing season), we analysed the ratio of removed to dead saplings (number of removed saplings out of a total of 467 dead saplings) using a GLM with a binomial error distribution and a logit link function. Again, we added the predictor variables size, species, treatment and interactions sequentially, and we used chi-square statistics for the calculation of significance. We applied Bonferroni corrections to tests for multiple comparisons of group means of significant variables and interactions.

We calculated the relative sapling growth by subtracting the initial height from the final height (end of first or second grazing season), divided by the initial height. ANOVA, SS type III, was used to test for differences in sapling growth between treatments and species after the first and second grazing seasons. The response variables were $\log10(x + 1)$ transformed prior to the analysis to improve normality of the errors. Species, treatment and their interaction were used as fixed factors in the models. Only surviving saplings were selected for this analysis, leading to a total of 125 and 61 individuals for the first and second years, respectively.

We used Genstat 7 for the GLM and SPSS release 11·5 for all other analyses.

Results

GRAZING INTENSITY

Results of the ANOVA on grazing intensity showed significant differences between the two unpalatable species and the three treatments, but not between the sapling sizes (Table 1). Tukey HSD tests showed that grazing was generally lowest in plots with unpalatable plants, higher in plots with cut unpalatable plants and highest in plots without unpalatables (Fig. 1). Further, grazing intensity was significantly higher in plots with uncut *Cirsium* than in plots with uncut *Gentiana*.

TREE SAPLING PERFORMANCE

The number of surviving tree saplings declined from 600 at the start of the experiment to 451 (74·1%), 325 (53·4%), 131 (21·5%), 107 (17·6%) and 63 (10·6%) after 52, 66, 129, 373 and 498 days, respectively (Fig. 2). Most saplings (194) disappeared after the third and longest grazing rotation period, and the lowest mortality was during winter (24). Results of the Cox regression

Table 1. Results of ANOVA for the effects of species, size and treatment and interactions on estimated grazing intensity ($\log10(x + 1)$ transformed) in a 40-cm circle around the tree saplings

Source	d.f.	MS	*F*	*P*
Species	1	7·43	26·85	0·000
Size	1	0·07	0·24	0·623
Treatment	2	11·41	41·25	0·000
Species × size	1	0·13	0·49	0·486
Species × treatment	2	1·66	5·98	0·003
Size × treatment	2	0·34	1·23	0·249
Species × size × treatment	2	0·21	0·76	0·469
Error	573	0·28		

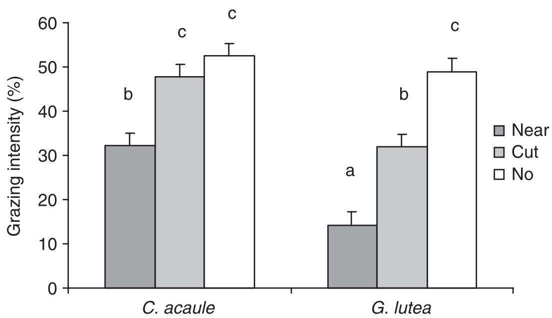

Fig. 1. Mean grazing intensity in 40-cm circles around plots with tree saplings planted (i) ±10 cm from the stems of the unpalatable plants *Cirsium acaule* or *Gentiana lutea* (near; dark bars); (ii) near removed (cut at soil level) unpalatable plants (cut; grey bars); and (iii) with no unpalatable plant nearby (no; white bars). Different letters indicate significant differences in grazing intensities ($P < 0{\cdot}01$, Tukey HSD tests).

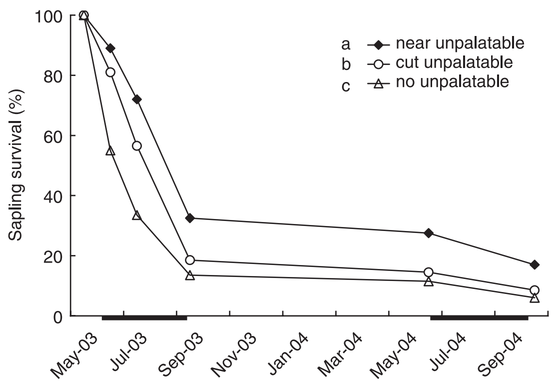

Fig. 2. Percentage survival of tree saplings transplanted near unpalatable plants (black diamonds), near cut unpalatable plants (open circles) and with no unpalatable plants (open triangles), over 2 years. Grazing seasons (black lines) and date of measurement are indicated in the figure. Different letters indicate significant differences between treatments over the entire study period (Cox regression, –2 log likelihood 6190·82, d.f. 2, $P < 0{\cdot}001$).

Table 2. Analysis of deviance for the effects of the predictor variables size, species and treatment and their interactions on sapling survival after one and two grazing seasons (GLM, binomial distribution, logit link)

Source	Survival after 1 year			Survival after 2 years		
	d.f.	Deviance	*P*	d.f.	Deviance	*P*
Size	1	6·22	0·013	1	7·61	0·006
Species	1	2·96	0·085	1	1·07	0·301
Treatment	2	16·16	0·000	2	11·44	0·003
Species × size	1	9·21	0·002	1	5·95	0·015
Species × treatment	2	4·31	0·116	2	7·72	0·021
Size × treatment	2	1·55	0·461	2	8·15	0·017
Species × Size × treatment	2	0·79	0·673	2	3·40	0·182
Residual	590	358·52		575	235·97	
Total	601	399·73		586	281·31	

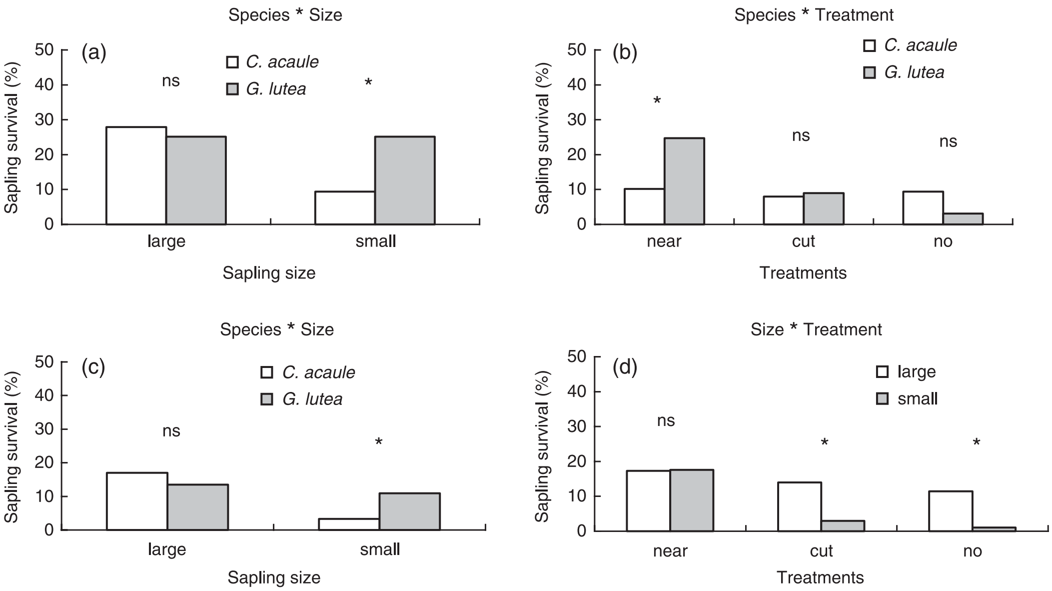

Fig. 3. Directions of the significant interactions for sapling survival after 1 (a), and 2 years (b–d). **P* < 0·05; ns, *P* > 0·05.

showed highly significant differences in sapling survival rates between all three treatments (–2 log likelihood 6190·82, d.f. 2, $P < 0{\cdot}001$). Sapling survival was consistently significantly highest in the plots with unpalatable plants and significantly lowest in the plots without unpalatable plants over the entire period of the study (Wald statistic 30·46, $P < 0{\cdot}001$). The difference between the cut and no unpalatable plots decreased gradually until the end of the first grazing season.

Results of the GLM showed that the factors size, treatment and species × size had significant effects on sapling survival after the first grazing season (Table 2). Overall, survival was significantly higher for the large saplings (26%) than for the small saplings (17%). Further, sapling survival after the first grazing season was significantly different between all three treatments, being highest in plots with unpalatables (33%), lower in plots with cut unpalatables (19%) and lowest in plots without unpalatables (14%) (Bonferroni test, $P < 0{\cdot}05$). The interaction was explained by a significantly lower survival of small saplings in the *Cirsium* plots compared with *Gentiana* plots, while survival of the large saplings was not different between species (Fig. 3a).

The factors size, treatment, species × size, species × treatment and size × treatment significantly affected sapling survival after two grazing seasons (Table 2). Again, survival was significantly higher for the large saplings (14%) compared with the small saplings (7%), and significantly (Bonferroni test, $P < 0{\cdot}05$) higher in plots with unpalatable plants (18%) compared with the plots with cut unpalatables (9%) and without unpalatable plants (6%). The latter two were not significantly different. Figure 3b–d shows the directions of the significant interactions: sapling survival was significantly lower for small saplings in *Cirsium* plots than in *Gentiana* plots, but there was no difference between species for the large saplings (Fig. 3c). Sapling survival was significantly higher in plots with *Gentiana* than in plots with *Cirsium*, but there was no difference between species for the other treatments (Fig. 3b). Further, large saplings survived significantly better than the small saplings in the plots with cut unpalatables and without unpalatables, but not in plots with unpalatable plants (Fig. 3d).

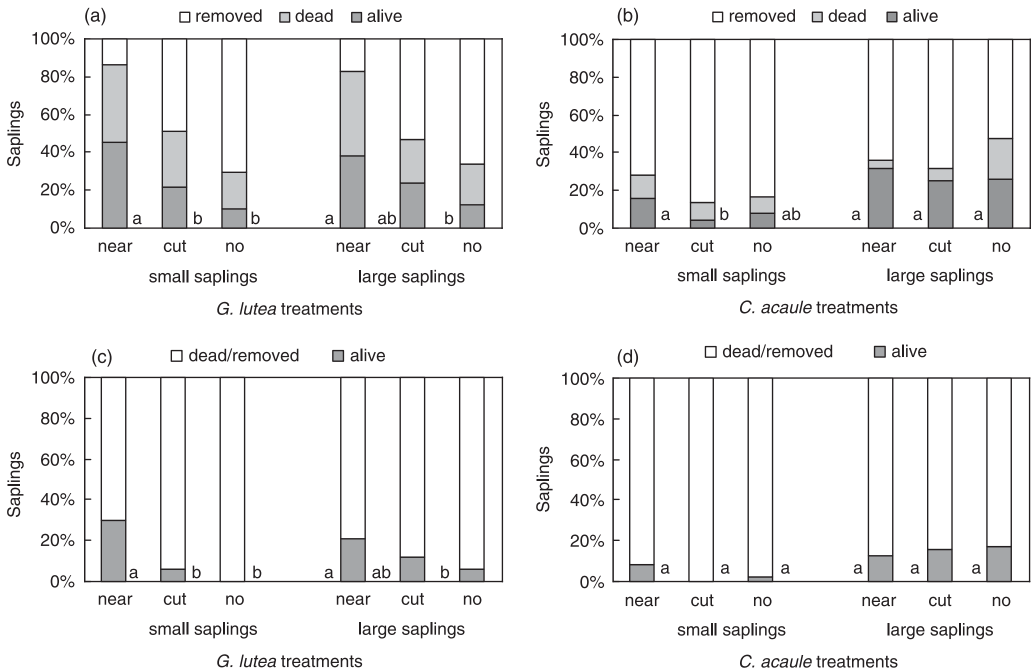

Fig. 4. Percentage of removed, dead and living tree saplings after one (a, b) and two (c, d) grazing seasons for each combination of treatment and size and separately for *Gentiana lutea* (a, c) and *Cirsium acaule* (b, d) treatments. Different letters indicate significant differences between treatments for each species per sapling size ($P < 0{\cdot}05$, Bonferroni tests).

The ratio of removed to dead saplings was significantly affected by unpalatable species, treatment and their interaction (deviance ratios, respectively: 35·47, P = 0·001; 6·36, P = 0·002; 5·78, P = 0·003). The ratio removed to dead saplings was higher for *Cirsium* (0·87) than for *Gentiana* (0·56), and Bonferroni tests revealed that this ratio only differed significantly between *Gentiana* treatments, being significantly lower in the plots with *Gentiana* (0·26) compared with plots with cut (0·66) and without *Gentiana* (0·77). Therefore, except for saplings in plots with *Gentiana*, removal by herbivores was the most important cause of death.

SAPLING GROWTH

On average, the height growth of the saplings after 1 and 2 years was 3% (± 26 SD) and 11% (± 33 SD), respectively, but we did not find significant effects of species, treatment or their interaction. Hence no evidence was found for competition effects of unpalatable plants on the tree saplings, but sample sizes of 126 and 62 may have been too small to detect a potential difference after 1 and 2 years of growth.

Discussion

ASSOCIATIONAL RESISTANCE

In this study, we found clear empirical evidence for the existence of associational resistance in wooded pastures, confirming suggestions based on descriptive studies (Smit *et al*. 2005). Over a period of 2 years, overall survival of *P. abies* saplings was significantly higher when planted near unpalatable plants than when planted near cut unpalatable plants or in the open (without any unpalatable plants). These findings correspond with a significantly lower grazing intensity near unpalatable plants. Hence, we propose that avoidance of the unpalatable plants by cattle caused the observed differences in sapling survival. As the final survival of the *Picea* saplings did not reveal significant differences between the cutting and the control treatments, it appears that the effects of soil and microenvironment did not play an important role in sapling survival in this study.

DIFFERENCES BETWEEN THE TWO UNPALATABLE SPECIES

Although we found an overall facilitative effect of the unpalatable plants for the survival of the transplanted tree saplings, effects of *Gentiana* were stronger than of *Cirsium* for both sapling sizes (Fig. 4). Several hypotheses may be raised for these observed differences. First, *Gentiana* is more apparent and therefore easier to detect and avoided by herbivores than *Cirsium*. Secondly, cattle may avoid *Gentiana* more than *Cirsium* because of a difference in unpalatability. We did not observe cattle eating *Gentiana*, except for its flowers. In contrast, young leaves of *Cirsium* were quite frequently grazed, indicating higher palatability of the physically defended *Cirsium*. Thirdly, the biotic and abiotic environments of the two species may differ. The larger *Gentiana* could offer better protection against direct sunlight and drought, creating a more shaded and moist environment for the tree saplings. In addition, although not observed in this study, plant species composition around both unpalatable plants could vary, thereby affecting the attractiveness of the site to cattle and the performance of the tree saplings. Finally, soil conditions (depth, nutrients, moisture) may differ between the two species, although height of *Picea* saplings did not differ between unpalatable species.

EFFECTS OF TREE SAPLING SIZE

Sapling size had significant effects on survival after one and two grazing seasons. Overall, the survival of large saplings was higher than that of small saplings, and large saplings seemed less affected by the treatments (Fig. 3d). This indicates that the first 2 years are the most critical stage for establishment of *P. abies* in wooded pastures. *Picea abies* is browsed by cattle but is not a preferred food source; hence, small saplings are probably browsed by chance together with more tasty surrounding focal species. Larger saplings may be browsed less because they are more obvious to herbivores (Rao *et al*. 2003).

MORTALITY

Sapling mortality is often caused by additive rather than individual processes (e.g. trampling may increase drought stress; Ibanez & Schupp 2001). As most tree saplings planted away from or near to cut unpalatable plants were completely removed and killed by cattle, grazing was the most important factor for sapling mortality. Tree saplings were usually completely uprooted by cattle. Naturally established tree saplings might be less prone to removal by cattle, because of better-developed root systems. As *P. abies* is browsing resistant and grows back after frequent browsing, we may have underestimated the potential of tree saplings to survive browsing and grazing in this study.

SAPLING GROWTH AND COMPETITION EFFECTS

Better growth of the unprotected (cut or no unpalatable plants) vs. the protected tree saplings (near unpalatables) would indicate competitive effects of the unpalatable plants. However, our analyses showed no differences in height of the saplings between treatments or species. These results are in contrast to findings of others studies reporting reduced growth of tree saplings in the proximity of the nurse shrubs (Meiners & Gorchov 1998; Rousset & Lepart 2000). It is possible that in our studied system the competition with unpalatable plants does not differ much from that with vegetation without unpalatables (i.e. mainly grasses). In contrast, the relatively high sapling mortality (other than as a result of removal by cows) near *Gentiana* in particular after the first grazing season could be an effect of increased competition (Fig. 4a). We acknowledge that we did not evaluate competition for light between the treatments. Further, the relatively short study time and low statistical power may well explain the lack of difference in sapling growth between treatments.

DYNAMICS OF WOODED PASTURES

Our findings indicate that unpalatable plants can enhance tree regeneration in wooded pastures and demonstrate the importance of associational resistance for the dynamics of these ecosystems. Given the low survival of unprotected 1-year-old tree saplings, protection against cattle by unpalatable plants facilitates their establishment. Once established, protection against cattle seems to become less important. We found higher survival of older tree saplings, and these established trees will eventually out-compete their nurse plants. At our site, this was demonstrated by the presence of solitary small trees, grown taller than their initial facilitators and surviving the continuous browsing by cattle (Smit *et al*. 2005).

In Mediterranean areas, transplanting tree saplings under naturally existing nurse shrubs is a successful alternative technique for reforestation, offering both economic and ecological advantages above the more traditional large-scale clear-cutting and planting technique (Castro *et al*. 2002). Our results show that transplanting tree saplings near unpalatable plants could also be an alternative technique for reforestation programmes in intensively and overgrazed wooded pastures. On the other hand, removal of unpalatable plants and shrubs might prevent or slow down undesired tree regeneration in extensively grazed areas.

Pastural abandonment and intensification of agricultural management are expected to continue in the near future (Tilman *et al*. 2002), with drastic consequences for many ecosystems. Both intensification and abandonment of grazing may lead to an increase of unpalatable plants in grasslands (Callaway, Kikvidze & Kikvidze 2000) and wooded pastures. This increase of unpalatable plants could speed up the succession to trees in wooded pastures and lead to a rapid closing of the canopy, with an expected loss of biodiversity.

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