Do nitrogen-fixing legumes affect soil nutrient levels, plant growth or community properties of an alpine ecosystem?

Siri Lie Olsen

NORWEGIAN UNIVERSITY OF LIFE SCIENCES
Department of Ecology and Natural Resource Management
Master Thesis 60 credits 2010

Do nitrogen-fixing legumes affect soil nutrient levels, plant growth or community properties of an alpine ecosystem?

Siri Lie Olsen

Master thesis 60 credits 2010
Department of Ecology and Natural Resource Management
Norwegian University of Life Sciences

ACKNOWLEDGEMENTS

I wish to thank...

... Ørjan Totland, my main supervisor, for assigning me to this project, for helping me with everything (especially statistics) and for being ambitious on my behalf.

... Sylvi M. Sandvik, my co-supervisor, for helping me in the field, for commenting on my manuscripts and for always having time for me.

... Annie Aasen, lab engineer, for helping me with the soil analyses, as I am a lousy chemist.

... Torbjørn Høitomt, my beloved boyfriend, for helping me in the field, for commenting on my manuscripts and for putting up with me during periods of intense work.

… Jenny Gulbrandsen, my friend and fellow master student, for the good company, for the hot chocolate and for commenting on my manuscripts.

... Linnea, my dog, for keeping me company during endless hours of writing and on long walks to clear my head.

... and finally Finse research station and everyone there for accommodation and hospitality.

Ås, December 15th 2010

Siri Lie Olsen

SUMMARY

Facilitation is an important process in plant communities. Because low nitrogen (N) supply limits plant growth in most terrestrial ecosystems, N-fixing legumes have a great potential as facilitators, as they increase soil N levels (e.g. Thomas & Bowman 1998, Pugnaire & Luque 2001, Halvorson & Smith 2009). Previous studies have shown that the presence of legumes may increase growth and reproduction of adjacent plants (e.g. Morris & Wood 1989, Franco-Pizaña et al. 1996, Thomas & Bowman 1998) and affect species composition and other properties of plant communities (e.g. Thomas & Bowman 1998, Rodríguez-Echeverría & Pérez-Fernándes 2003, Perroni-Ventura et al. 2006).

According to the stress-gradient hypothesis, the outcome of plant-plant interactions shifts from competition to facilitation with increasing abiotic stress (Bertness & Callaway 1994, Brooker & Callaghan 1998, Callaway et al. 2002). This implies that facilitation is the dominant interaction in alpine habitats, which are considered climatically stressful (e.g. Billings and Mooney 1968). Even though legumes are rare in alpine ecosystems (Körner 2003), they have a great potential as facilitators in these habitats where low biomass production and slow decomposition limit the supply of nutrients, including N (e.g. Bliss 1971). However, few studies have examined the role of alpine legumes as facilitators.

In this master thesis I examine how two N-fixing legume species, *Astragalus alpinus* L. and *Oxytropis lapponica* (Wahlenb.) Gay, affect different aspects of an alpine ecosystem in two sites situated along a gradient in abiotic stress at Finse, Norway. The thesis consists of three parts:

In **Part 1** I examine whether soil nutrient levels differ below and outside legumes. My results show that loss on ignition, total C and N content and NH_4^+-N and NO_3^--N levels were higher below than outside *O. lapponica* plants, most likely due to N-fixing by the legumes. There was no difference in soil nutrient levels below and outside *A. alpinus*. The results for *O. lapponica* are in accordance with the majority of literature reporting increased soil N levels below legumes and suggest that *O. lapponica* may positively influence adjacent plants. The percent increase in soil nutrient levels increased with abiotic stress, implying that legume facilitation may also increase with abiotic stress.

In **Part 2** I examine whether leaf density, size, growth rate, reproduction or N content of the alpine herb *Thalictrum alpinum* L. differ with and without the presence of legumes. I found that the leaf density of *T. alpinum* increased with both *O. lapponica* and *A. alpinus*, most likely due to increased clonal growth caused by elevated soil N levels (Part 1). The increase was greatest where abiotic stress was highest, supporting the stress-gradient theory. The lack of response in *T. alpinum* plant size, growth rate and reproduction, as well as a reduction in N content, may be due to increased resource allocation to clonal growth. As the two legume species can facilitate the density of a single target species, they may have the ability to influence the plant community as a whole.

In **Part 3** I examine whether species composition, species richness, species diversity or species evenness differ with and without the presence of legumes. I found that species composition differed and species richness increased with *O. lapponica*, most likely due to increased N availability below the legumes (Part 1). There was no difference in community properties with and without *A. alpinus*. The facilitative effect of *O. lapponica* was found only in the site of highest abiotic stress, supporting the theory that facilitation may increase species richness under stressful environmental conditions (e.g. Hacker & Gaines 1997). Graminoid species richness and cover also increased with *O. lapponica*, but the increase in graminoids was not great enough to exclude herb species, resulting in an increase in total species richness.

Altogether, Part 1-3 clearly show that *O. lapponica* has the ability to influence the alpine ecosystem at Finse. The legumes facilitate other species by increasing soil N levels, which not only increases the leaf density of a single target species, but cascades into community-wide effects altering species composition and increasing species richness. Despite no significant difference in soil N levels, the presence of *A. alpinus* also increased leaf density of *T. alpinum*, but had no community-level effects. Moreover, my study supports the stress-gradient hypothesis, as the facilitative effects were greatest in the most severe end of the stress-gradient.

Thomas & Bowman (1998), who studied *Trifolium dasyphyllum* in the Rocky Mountains, conclude that legumes ”represent an important biotic factor contributing to spatial heterogeneity in species composition (...) of alpine tundra”. I believe that this is true also for the legumes at Finse, at least *O. lapponica*, which I found to alter species composition in the study area. As this only happens on a very local scale, namely in close vicinity of the legumes, the spatial heterogeneity increases, which may again increase total species richness of the ecosystem (Tilman 1982).

A weakness of this study is that I have not conducted any experiments and hence cannot conclude definitely on cause and effect. I have just observed the variation already present. However, based on my findings I can point out interesting trends and make suggestions as to what may have caused them. It would have been interesting to experimentally transplant or sow legumes and study the effects on soil and surrounding vegetation, but this was not within the scope of a master thesis.

Brooker et al. (2008) point out several aspects of facilitation that needs further investigation, including working on several levels of abiotic stress. In this study I examined only two sites (i.e. two stress-levels) for each legume species, but in Northern Norway both *A. alpinus* and *O. lapponica* are distributed from sea level to the middle alpine zone (Lid & Lid 2005), allowing for studies along a much more extensive gradient. A future study could also examine whether other species than *T. alpinum* respond to legume presence with changes in density. Using the data in Part 3, one could select a number of species, some positively and some negatively associated with legumes, and examine how different species and functional groups respond to elevated soil N levels, as well as determine the mechanisms behind any changes in density. This would also increase our understanding of the community-level effects of legumes.

It is important to assess the role of legumes in alpine ecosystems. My study shows that they may enhance spatial heterogeneity and increase species richness, an important quality in itself, but especially in light of the ongoing climatic change. Alpine areas have been predicted to be especially sensitive to global warming (e.g. Guisan et al. 1995), which may lead to homogenization of the alpine vegetation (e.g. Odland et al. 2010). Under such circumstances legumes may play a role in maintaining biodiversity.

REFERENCES

Bertness, M. D. & Callaway, R. M. 1994. Positive interactions in communities. Trends in Ecology and Evolution 9: 191-193.

Billings, W. D. & Mooney, H. A. 1968. The ecology of arctic and alpine plants. Biological Reviews 43: 481-529.

Bliss, L. C. 1971. Arctic and alpine plant life cycles. Annual Review of Ecology and Systematics 2: 405-438.

Brooker, R. W. & Callaghan, T. V. 1998. The balance between positive and negative plant interactions and its relationship to environmental gradients: A model. Oikos 81: 196-207.

Brooker, R. W., Maestre, F. T., Callaway, R. M., Lortie, C. L., Cavieres, L. A., Kunstler, G., Liancourt, P., Tielbörger, K., Travis, J. M. J., Anthelme, F., Armas, C., Coll, L., Corcket, E., Delzon, S., Forey, E., Kikvidze, Z., Olofsson, J., Pugnaire, F., Quiroz, C. L., Saccone, P., Schiffers, K., Seifan, M., Touzard, B. & Michalet, R. 2008. Facilitation in plant communities: The past, the present, and the future. Journal of Ecology 96: 18-34.

Callaway, R. M., Brooker, R. W., Choler, P., Kikvidze, Z., Lortie, C. J., Michalet, R., Paolini, L., Pugnaire, F. I., Newingham, B., Aschehoug, E. T., Armas, C., Kikodze, D. & Cook, B. J. 2002. Positive interactions among alpine plants increase with stress. Nature 417: 844-848.

Franco-Pizaña, J. G., Fulbright, T. E., Gardiner, D. T. & Tipton, A. R. 1996. Shrub emergence and seedling growth in microenvironments created by *Prosopis glandulosa*. Journal of Vegetation Science 7: 257-264.

Guisan, A., Tessier, L., Holten, J. I., Haeberli, W. & Baumgartner, M. 1995. Understanding the impact of climate change on mountain ecosystems: An overview. In: Potential ecological impacts of climate change in the Alps and Fennoscandian mountains (eds. Guisan, A., Holten, J. I., Spichiger, R. & Tessier, L.), Conservatoire et Jardin botaniques, Geneve. Pp. 15-37.

Hacker, S. D. & Gaines, S. D. 1997. Some implications of direct positive interactions for community species diversity. Ecology 78: 1990-2003.

Halvorson, J. J. & Smith, J. L. 2009. Carbon and nitrogen accumulation and microbial activity in Mount St. Helens pyroclastic substrates after 25 years. Plant and Soil 315: 211-218.

Körner, C. 2003. Alpine plant life. Functional plant ecology of high mountain ecosystems. 2nd edition. Springer-Verlag, Berlin. Pp. 344.

Lid, J. & Lid, D. T. 2005. Norsk flora. 7. utgåva. Det Norske Samlaget, Oslo. Pp. 1230.

Morris, W. F. & Wood, D. M. 1989. The role of lupine in succession on Mount St. Helens: Facilitation or inhibition? Ecology 70: 697-703.

Odland, A., Høitomt, T. & Olsen, S. L. 2010. Increasing vascular plant richness on 13 high mountain summits in Southern Norway since the early 1970s. Arctic, Antarctic, and Alpine Research 42: 458-470.

Perroni-Ventura, Y., Montaña, C. & García-Oliva, F. 2006. Relationship between soil nutrient availability and plant species richness in a tropical semi-arid environment. Journal of Vegetation Science 17: 719-728.

Pugnaire, F. I. & Luque, M. T. 2001. Changes in plant interactions along a gradient of environmental stress. Oikos 93: 42-49.

Rodríguez-Echeverría, S. & Pérez-Fernández, M. A. (2003). Soil fertility and herb facilitation mediated by *Retama sphaerocarpa*. Journal of Vegetation Science 14: 807-814.

Thomas, B. D. & Bowman, W. D. 1998. Influence of N_2-fixing *Trifolium* on plant species composition and biomass production in alpine tundra. Oecologia 115: 26-31.

Tilman, D. 1982. Resource competition and community structure. Monographs in Population Biology 17. Princeton University Press, Princeton. Pp. 296.

PART 1. NITROGEN-FIXING LEGUMES DIFFERENTIALLY AFFECT SOIL NUTRIENT LEVELS IN AN ALPINE ECOSYSTEM

ABSTRACT

Low nitrogen (N) supply is a limiting factor for plant growth in most terrestrial ecosystems. N-fixing legumes therefore have the potential to facilitate the growth of surrounding vegetation as they increase soil N levels. This fertilizing effect should be greatest in low-productivity habitats where initial soil N levels are low, for instance in alpine areas. In this study I examined how soil nutrient levels differed below and outside two alpine legumes, Astragalus alpinus *L. and* Oxytropis lapponica *(Wahlenb.) Gay, in two sites situated along a gradient in abiotic stress at Finse, Norway. I found that loss on ignition, total C and N content, as well as* NH_4^+*-N and* NO_3^-*-N levels, were higher below than outside* O. lapponica *plants, most likely due to N-fixing by the legumes. The percent increase in soil nutrient levels was greatest in the site of highest abiotic stress. There was no difference in soil nutrient levels below and outside* A. alpinus*, indicating that* O. lapponica*, but not* A. alpinus*, has the potential to facilitate surrounding vegetation in the study area.*

INTRODUCTION

Facilitation, as opposed to competition, is a positive interaction in which the fitness of one or more species is increased without negatively affecting any of the other species in the interaction (Hacker & Gaines 1997). The positive effect of facilitation may be either a direct amelioration of harsh environmental conditions or an indirect effect through interacting species (Callaway 1995). This includes modification of microhabitats, physical support, negative effects on herbivores, pathogens and competitors, positive effects on microbes and pollinators, transfer of nutrients and soil enrichment (Hunter & Aarssen 1988).

Low soil nitrogen (N) levels limit plant growth in most terrestrial ecosystems. However, some plant taxa, such as the legumes (Fabaceae), compensate for this by living in symbiosis with N-fixing bacteria, particularly within the *Rhizobium*. This gives the legumes a competitive advantage when soil N levels are low (e.g. Vitousek & Field 1999) and may also facilitate the surrounding vegetation, as the presence of N-fixers can increase soil N levels through leaf leaking, root exudates, decomposition of N rich litter (Høgh-Jensen 2006 and references therein) and reduced competition for N (Temperton et al. 2007). N can also be directly transferred between plants through mycorrhiza (e.g. Frey & Schüepp 1992).

The positive effect of legumes on soil N levels has been thoroughly studied in agricultural systems, but has also been found in different types of natural ecosystems, such as deserts (Tiedemann & Klemmedson 1973, Rundel et al. 1982, Rossi & Villagra 2003), alpine areas (Kenny & Cuany 1990, Thomas & Bowman 1998), forests (Myrold & Huss-Danell 2003), volcanic sites (Halvorson & Smith 2009), grasslands (Maron & Connors 1996, Maron & Jefferies 1999, Carino & Daehler 2002), shrublands (Pugnaire & Luque 2001, Rodríguez-Echeverría & Pérez-Fernándes 2003, Perroni-Ventura et al. 2006) and savannas (Belsky et al. 1993, Hagos & Smit 2005, Fornara & Tilman 2008).

The fertilizing effect of legumes should be greatest where initial N levels are low, for instance during primary succession (Halvorson et al. 1991, Bellingham et al. 2001), in disturbed sites (Dancer et al. 1977, Palaniappan et al. 1979) and in other low-productivity habitats. Belsky et al. (1993) note that the difference in "several soil-fertility indices" below and outside the canopy of the leguminous tree *Acacia tortilis* was greater in arid than in more humid sites, and Pugnaire & Luque (2001) found a greater difference in soil N content below and outside the canopy of the leguminous shrub *Retama sphaerocarpa* under low-productivity compared to high-productivity conditions. Their results suggest that biologically fixed N contributes more to the total soil N content in marginal habitats than in more nutrient-rich areas, resulting in a greater difference in soil N content below and outside legumes.

The findings of Belsky et al. (1993) and Pugnaire & Luque (2001) show that the fertilizing effects of legumes is greater in low-productivity sites, implying that legumes may affect soil N levels in alpine areas, where low biomass production and low decomposition rates limit the supply of nutrients, including N (e.g. Bliss 1971, Nadelhoffer et al. 1992). However, to my knowledge only two studies (Kenny & Cuany 1990, Thomas & Bowman 1998) have examined the effect of alpine legumes on soil N levels.

In this study I examine how soil N levels differ below and outside two alpine legumes, *Astragalus alpinus* L. and *Oxytropis lapponica* (Wahlenb.) Gay, in two sites situated along a gradient in abiotic stress at Finse, Norway. If the positive effect of legumes on soil N levels increases with decreasing productivity, as shown by Pugnaire & Luque (2001), the difference in soil N content below and outside legumes should be especially pronounced in alpine areas and increase with increasing abiotic stress. I also examine how other soil variables - dry matter content, loss on ignition, total carbon (C) content, phosphorus (P) levels and pH - differ below and outside legumes and whether the soil variables are related to properties of individual legume plants that may affect soil nutrient levels, such as plant size and the number of root nodules.

Specifically I ask the following questions:

- Do soil N levels (total N, NH_4^+-N and NO_3^--N) or other soil variables differ below and outside legumes?
- Does the difference in N levels or other soil variables below and outside legumes increase with increasing abiotic stress?
- Are soil N levels or other soil variables correlated to legume size or root nodule number?

MATERIALS AND METHODS

Study area and study species

Field work was conducted during the summer 2008 at Finse, southwest Norway (60°36’59’’N, 07°31’23’’E). Finse has a slightly oceanic climate characterized by high precipitation and cool summers (Moen 1998). Mean annual rainfall and temperature are 1030 mm and -2.1 °C, respectively (The Norwegian Meteorological Institute 2010). The study area was located in the middle alpine zone on the southwest slope of Mt. Sanddalsnuten (1554 m.a.s.l.). The bedrock here consists of phyllite with strains of marble (Askvik 2008), which produces soil rich in plant nutrients and lime (Fægri 1967). *Dryas* heath, dominated by the dwarf-shrub *Dryas octopetala*, is the main vegetation type in the study area.

The study was conducted in three sites (10 × 10 m) situated along the slope of Mt. Sanddalsnuten: a low-elevation site (1460 m.a.s.l.), mid-elevation site (1510 m.a.s.l.) and high-elevation site (1554 m.a.s.l.) with a difference in altitude of about 50 m between each site. From the low-elevation site to the top of Mt. Sanddalsnuten the vegetation gradually changes from a continuous, species-rich meadow to an exposed ridge community where patches of open soil and bare rock are frequent. The study species *Astragalus alpinus* was present in the low and high-elevation sites, while *Oxytropis lapponica* was present in the mid and high-elevation sites.

A. alpinus and *O. lapponica* are both perennial, herbaceous legumes. *A. alpinus* grows in many types of alpine plant communities, while *O. lapponica* is restricted to rich, but barren ridges and slopes, as well as *Dryas* heaths (Lunde 1962). In the study area *A. alpinus* is generally found in more favourable microhabitats than *O. lapponica* (pers.obs.). Both *A. alpinus* and several species of *Oxytropis* have been reported to fix nitrogen in arctic and alpine areas (Granhall & Lid-Torsvik 1975, Karagatzides et al. 1985, Johnson & Rumbaugh 1986, Schulman et al. 1988) and should therefore have the potential to increase soil nitrogen levels at Finse.

Study design and chemical analyses

To examine how soil nutrient levels differ below and outside legumes, I collected 40 pairs of soil samples (80 all together). Each pair consisted of one sample from below a legume and one control sample 30-100 cm away from the first sample in the pair and at least 30 cm away from all other legumes. In the low-elevation site I collected 10 soil sample pairs associated with *Astragalus alpinus*, in the mid-elevation site 10 pairs associated with *Oxytropis lapponica* and in the high-elevation site 10 pairs associated with *O. lapponica* and 10 pairs with *A. alpinus*. Soil was collected within a quadrat of 15 × 15 cm to a depth of 15 cm.

I collected the legume plant in each pair by carefully removing legume roots and aboveground biomass from the soil, which was thoroughly mixed and then frozen. I washed the roots and counted the number of nodules and leaves of each legume. Plant material was dried at 60 °C for 24 hours and weighed.

Subsamples of 5 g moist soil were dried over night at 105 °C, and I weighed the dry samples and calculated the amount of dry matter (%). The dry samples were then combusted at 660 °C for 6 hours, and I weighed the ashes and calculated the loss on ignition (%). The remaining soil was dried at 40 °C over night and sieved (2 mm). I analyzed soil NH_4^+-N (mg/kg) content according to the Norwegian Standard 4746 (Appendix 1) and soil P content using the P-Al method (Appendix 2). Soil NO_3^--N (mg/kg) and total C (%) and N (%) content were analyzed by the Soil and Water Chemistry laboratory of the Department of Plant and Environmental Sciences at the Norwegian University of Life Sciences in Ås. Finally I measured soil pH

(InoLab pH 720 pH meter, WTW, Weilheim, Germany) from a suspension of 10 ml dry soil in 25 ml deionized water that had been shaken and left over night.

Primary data is found in Appendix 3.

Statistical analyses

I used analysis of variance (ANOVA) to test whether soil dry matter content (%), loss on ignition (%), total C content (%), total N content (%), NH_4^+-N levels (mg/kg), NO_3^--N levels (mg/kg), P-levels (mg/kg) or pH differed below and outside legumes and if the relationship differed between sites. Data for the two legume species was analyzed separately, as initial tests suggested that they produce contrasting results. All tests were two-way ANOVAs with *site* and *legume presence* as fixed main factors and *pair* as a random nested factor under *site* where this was significant (Table 1). If the interaction between *site* and *legume presence* was significant, I used a paired t-test to compare soil parameters below and outside legumes within each site. Data on NH_4^+-N and NO_3^--N content for *A. alpinus* were ln transformed to fulfill the ANOVA assumptions of normality and equal variances. Analyses were performed using the General Linear Model and Paired t-test procedures in Minitab 15.1.1.0 for Windows.

I used regression analysis to test whether there was a relationship between the different soil parameters and the size (number of leaves and biomass) or the number of nodules of the associated legume plants. First I did a stepwise (forward and backward) selection to see if any of the legume variables were significantly related to each of the soil parameters. I then used the selected models in the regression analyses, with *number of leaves, total biomass* and/or *number of nodules* as independent variables and each of the soil parameters as a dependent variable. Separate analyses were conducted for each species and site. All analyses were performed using the Regression procedure in Minitab 15.1.1.0 for Windows.

RESULTS

Astragalus alpinus

The ANOVA showed no significant difference in any soil variable below and outside *Astragalus alpinus* plants (Table 1), although there was a tendency of slightly higher nutrient levels below the legumes (Table 2). All soil parameters were significantly higher in the low-elevation site, except for dry matter content, which was higher in the high-elevation site.

The regression analyses showed a significant positive relationship between the total biomass of *A. alpinus* and soil dry matter content ($p = 0.006$, $R^2 = 63.8$ %) and between the number of nodules and soil P content ($p = 0.047$, $R^2 = 40.8$ %) in the high elevation site. Otherwise there were no relationships between the size of *A. alpinus* and any of the soil parameters in either site ($p > 0.05$ for all tests).

Oxytropis lapponica

The ANOVA showed that both loss on ignition, total C and N content, as well as NH_4^+-N and NO_3^--N levels, were significantly higher in soil below *Oxytropis lapponica* plants, while there were no differences between sites (Table 1). However, there was a tendency of slightly lower background nutrient levels in the high-elevation site, and the percent increase in soil nutrient levels below legumes was always greater in this site (Table 2). P levels and soil pH did not differ outside and below *O. lapponica*, but were significantly higher in the mid-elevation site compared to the high-elevation site. For dry matter content there was a significant interaction between *legume presence* and *site*, and the t-tests showed that dry matter content was significantly higher outside *O. lapponica* in the high-elevation site ($T = 4.05$, $p = 0.003$), while there was no difference in dry matter content below and outside legumes in the mid-elevation site ($T = 0.34$, $p = 0.74$).

The regression analyses showed that there was a significant negative relationship between the total biomass of *O. lapponica* and dry matter content in the mid elevation site ($p = 0.049$, $R^2 = 40.2$ %). Otherwise there were no significant relationships between the size of *O. lapponica* and any of the soil parameters ($p > 0.05$ for all tests).

Table 1. $F_{d.f.}$ and p-values for ANOVA analyses of dry matter (%), loss on ignition (%), total C content (%), total N content (%), NH_4^+-N (mg/kg), NO_3^--N (mg/kg), P (mg/kg) and pH of soil samples from below and outside the legumes Astragalus alpinus *and* Oxytropis lapponica *in three study sites at Mt. Sanddalsnuten, Finse, Norway summer 2008.* ***Bold*** *letters indicate p-values significant at the 0.05 level.*

Source of variation	**Dry matter (%)**		**Loss on ignition (%)**		**Total C (%)**		**Total N (%)**	
	$F_{d.f.}$	*P*	*$F_{d.f.}$*	*P*	*$F_{d.f.}$*	*P*	*$F_{d.f.}$*	*P*
A. alpinus								
site	$28.56_{1,18}$	<**0.001**	$15.29_{1,18}$	**0.001**	$33.20_{1,36}$	<**0.001**	$23.20_{1,18}$	<**0.001**
A. alp	$1.71_{1,17}$	0.208	$1.48_{1,18}$	0.239	$0.15_{1,36}$	0.702	$0.10_{1,18}$	0.755
site × A. alp	$0.10_{1,17}$	0.752	$0.77_{1,18}$	0.391	$0.11_{1,36}$	0.741	$0.11_{1,18}$	0.743
pair	$6.80_{18,17}$	<**0.001**	$7.25_{18,18}$	<**0.001**	—	—	$2.62_{18,18}$	**0.024**
O. lapponica								
site	$10.40_{1,36}$	**0.003**	$3.34_{1,36}$	0.076	$1.71_{1,36}$	0.199	$1.17_{1,36}$	0.286
O. lap	$6.59_{1,36}$	**0.015**	$7.74_{1,36}$	**0.009**	$6.19_{1,36}$	**0.018**	$6.56_{1,36}$	**0.015**
site × O. lap	$4.21_{1,36}$	**0.047**	$3.42_{1,36}$	0.073	$3.27_{1,36}$	0.079	$3.14_{1,36}$	0.085
pair	—	—	—	—	—	—	—	—

Source of variation	**NH_4-N (mg/kg)**		**NO_3-N (mg/kg)**		**P (mg/kg)**		**pH**	
	$F_{d.f.}$	*P*	*$F_{d.f.}$*	*P*	*$F_{d.f.}$*	*P*	*$F_{d.f.}$*	*P*
A. alpinus								
site	$4.33_{1,36}$	**0.045**	$14.26_{1,36}$	**0.001**	$18.75_{1,36}$	<**0.001**	$11.27_{1,36}$	**0.002**
A. alp	$0.61_{1,36}$	0.440	$0.53_{1,36}$	0.473	$1.29_{1,36}$	0.263	$0.89_{1,36}$	0.351
site × A. alp	$0.25_{1,36}$	0.620	$0.04_{1,36}$	0.838	$0.70_{1,36}$	0.409	$0.97_{1,36}$	0.331
pair	—	—	—	—	—	—	—	—
O. lapponica								
site	$0.03_{1,36}$	0.862	$0.43_{1,36}$	0.517	$12.84_{1,36}$	**0.001**	$9.90_{1,36}$	**0.003**
O. lap	$6.25_{1,36}$	**0.017**	$6.27_{1,36}$	**0.017**	$0.39_{1,36}$	0.535	$3.21_{1,36}$	0.081
site × O. lap	$1.13_{1,36}$	0.294	$0.07_{1,36}$	0.795	$0.62_{1,36}$	0.435	$0.04_{1,36}$	0.851
pair	—	—	—	—	—	—	—	—

Table 2. Amount of dry matter (%), loss on ignition (%), total C content (%), total N content (%), NH_4^+*-N (mg/kg),* NO_3^-*-N (mg/kg), P (mg/kg) and pH in soil samples from below and outside the legumes* Astragalus alpinus *and* Oxytropis lapponica *in the low, mid and high-elevation study sites at Mt. Sanddalsnuten, Finse, Norway summer 2008. All values are mean ± 1 SE. Where there was a significant difference between nutrient levels below and outside legumes, the percent difference (Δ%) is given.*

	Dry matter (%)	Loss on ignition (%)	Total C (%)	Total N (%)
A. alpinus				
Low-elevation				
legumes	42.8 ± 5.2	40.2 ± 8.3	17.9 ± 3.9	1.5 ± 0.3
control	48.0 ± 7.2	34.5 ± 7.2	19.8 ± 3.5	1.4 ± 0.3
Δ (%)	—	—	—	—
High-elevation				
legumes	74.0 ± 1.3	9.2 ± 0.6	3.6 ± 0.3	0.3 ± 0.02
control	76.2 ± 1.2	8.2 ± 0.6	3.8 ± 0.4	0.3 ± 0.03
Δ (%)	—	—	—	—
O. lapponica				
Mid-elevation				
legumes	79.9 ± 0.8	7.4 ± 0.5	2.7 ± 0.2	0.3 ± 0.01
control	80.5 ± 1.6	6.8 ± 0.6	2.4 ± 0.4	0.2 ± 0.03
Δ (%)	—	**7.9**	**11.1**	**9.1**
High-elevation				
legumes	74.0 ± 1.2	9.5 ± 0.8	3.9 ± 0.7	0.3 ± 0.04
control	79.2 ± 0.8	6.8 ± 0.3	2.2 ± 0.1	0.2 ± 0.009
Δ (%)	**7.0**	**39.4**	**76.3**	**54.0**

	NH_4-N (mg/kg)	NO_3-N (mg/kg)	P (mg/kg)	pH
A. alpinus				
Low-elevation				
legumes	2.2 ± 0.6	17.8 ± 4.5	45.1 ± 2.4	6.9 ± 0.08
control	1.7 ± 0.5	17.0 ± 6.2	42.2 ± 1.6	6.9 ± 0.1
Δ (%)	—	—	—	—
High-elevation				
legumes	0.9 ± 0.1	3.8 ± 0.9	37.5 ± 0.5	6.5 ± 0.2
control	0.8 ± 0.1	2.9 ± 0.7	37.1 ± 0.2	6.2 ± 0.2
Δ (%)	—	—	—	—
O. lapponica				
Mid-elevation				
legumes	0.7 ± 0.06	7.0 ± 1.2	38.1 ± 0.5	7.3 ± 0.1
control	0.6 ± 0.1	5.1 ± 0.6	38.1 ± 0.4	7.4 ± 0.1
Δ (%)	**20.0**	**36.5**	—	—
High-elevation				
legumes	0.8 ± 0.06	6.6 ± 0.7	37.1 ± 0.3	7.0 ± 0.08
control	0.5 ± 0.1	0.3 ± 0.7	36.5 ± 0.1	7.2 ± 0.1
Δ (%)	**56.3**	**52.8**	—	—

DISCUSSION

Differences in soil nutrient levels

Loss on ignition, total C and N content and NH_4^+-N and NO_3^--N levels were higher below than outside *Oxytropis lapponica* plants, while there was no significant difference in any of the soil parameters below and outside *Astragalus alpinus*.

The increase in soil NH_4^+-N, NO_3^--N and total N levels below *O. lapponica* at Finse (and the similar, although far from significant, trend for *A. alpinus*) is in line with previous findings of increased soil N levels below alpine legumes: Kenny & Cuany (1990) found increased N levels in soil below *Lupinus* spp. in alpine Colorado, and Thomas & Bowman (1998) found that *Trifolium dasyphyllum* increased soil N levels in the Rocky Mountains. The percent increase in soil N levels below *O. lapponica* was greatest in the high-elevation site, where abiotic stress is highest. This is in accordance with Pugnaire & Luque (2001), who found that the positive effect of the leguminous shrub *Retama sphaerocarpa* on soil N levels increased with abiotic stress in a semi-arid environment in Spain. The greater increase in soil N in the high-elevation site may be due to the tendency of slightly lower background N levels in this site, causing additional N from N-fixing legumes to result in a greater percent increase in total soil N levels.

I did not experimentally examine whether the increased soil N levels below *O. lapponica* actually comes from biological N-fixation. The legumes may simply prefer microhabitats with higher soil N levels. However, Maron & Connors (1996), who studied *Lupinus arboreus* on the Californian coastal prairie, conclude that ”high levels of nitrogen in soil under either dead or live lupines did not result simply from lupines selectively colonizing (or surviving in) nutrient-rich patches”. They found that soil N levels below young legumes were identical to surrounding areas, whereas N levels below adult legumes were higher, indicating that *L. arboreus* improve soil N conditions. Geesing et al. (2000) made the same conclusion based on studies of *Prosopis glandulosa* in semi-arid Texas. In light of the findings of Maron & Connors (1996) and Geesing et al. (2000) I find it likely that the increased soil N levels below *O. lapponica* at Finse is due to biological N-fixation by the legumes.

Loss on ignition and total C content also increased with *O. lapponica* in the study area. There was a tendency of increased loss on ignition below *A. alpinus*, but this was not significant.

The results for *O. lapponica* are in accordance with Maron & Jefferies (1999) and Halvorson & Smith (2009), who found a higher organic matter content in soil below *Lupinus arboreus* and *Lupinus lepidus*, respectively. Fornara & Tilman (2008) conclude that legumes may increase soil C levels by providing N for increased growth of below-ground (and possibly above-ground) biomass. The increase in soil C content I found at Finse could be caused by a similar mechanism, where additional N from legumes increases the biomass of adjacent plants, thereby indirectly increasing C inputs to the soil. The percent increase in loss on ignition and total C content was greatest in the high-elevation site, indicating an increase in facilitation with increasing abiotic stress.

I found no difference in soil P or pH below and outside either legume species. Several studies have shown increased soil P levels below legumes (Rossi & Villagra 2003, Geesing et al. 2000, Gosling 2005), while Thomas & Bowman (1998) found reduced soil P levels below *Trifoilum dasyphyllum* in the Rocky Mountains. Geesing et al. (2000) suggest that the increase in P was due to pumping of nutrients from deeper layers, possibly coupled with the fact that legumes are better at acquiring P from insoluble sources, while Gosling (2005) points to increased litter input. He uses the same explanation for his findings of increased pH. Quite contradictory, Thomas & Bowman (1998) conclude that their reduced P levels are due to the legumes' high P demand. My results contrast all of the above, and further studies are needed to determine how the presence of legumes affects soil P content and pH.

I found few significant relationships between soil nutrient levels and the size or nodule number of *O. lapponica* and *A. alpinus* at Finse. This contrasts the results of Geesing et al. (2000), who found that the increase in N levels below *P. glandulosa* canopies was significantly correlated with trunk diameter. However, it could be that the age of the legume is more important than the size. Pugnaire et al. (1996) found that soil nutrient levels below *Retama sphaerocarpa* increased with shrub age. Both *O. lapponica* and *A. alpinus* are perennial, and it is not unlikely that over several years nutrients may accumulate in the soil below the plants. As for the study of Geesing et al. (2000), trunk diameter could be correlated with the age of the tree and therefore related to soil nutrient levels. However, further studies are needed to determine whether there is a relationship between legume age and soil nutrient levels.

Differences between legume species

While N and C levels increased with the presence of *O. lapponica*, there were no significant differences in soil nutrient levels below and outside *A. alpinus*, despite the fact that *A. alpinus* was nodulated in the study area (pers.obs.) and has been reported to fix N in alpine habitats (Granhall & Lid-Torsvik 1975, Johnson & Rumbaugh 1986). In a previous study I found that the two legume species had the same mean number of nodules, but that *O. lapponica* had a greater biomass (Olsen 2008). In theory a greater biomass could mean more litter, adding more N to the soil, which could explain the differing effect of the two species. However, I found no relationship between legume size and soil nutrient levels, so this seems unlikely.

Another possible explanation is that *A. alpinus* fixes less N than *O. lapponica*, thereby adding less N to the soil. In the relatively favourable microhabitats of *A. alpinus* it may be more beneficial for the plants to acquire N by root-uptake from the soil, as N acquisition through biological fixation is quite costly (Gutschick 1981). Moreover, *A. alpinus* plants may able to acquire a greater portion of their N from the soil because they are better adapted the conditions in the study area than *O. lapponica*. *A. alpinus* is a widespread species in artic and alpine habitats, and the plants at Finse grow nowhere near their altitudinal limit (Lid & Lid 2005). *O. lapponica*, on the other hand, has its altitudinal limit in Norway at 1550 m.a.s.l. (Lid & Lid 2005), meaning that the plants at Mt. Sanddalsnuten are growing in a marginal habitat where biological N-fixation may be an important source of N. I did not measure the rate of N-fixation of the two legume species. However, previous studies indicate that *A. alpinus* has an equal or greater rate of N-fixation than *Oxytropis* spp. (Karagatzides et al. 1985, Schulman et al. 1988), making a difference in N-fixation seem unlikely.

Rather than differences in fixation rates *per se*, the contrasting results for the two legume species may be due to a less pronounced impact of N-fixation on soil N levels in the relatively favourable microhabitats of *A. alpinus*. The low-elevation site, where only *A. alpinus* is found, had much higher soil nutrient levels than the two other sites, and when comparing background soil nutrient contents of *O. lapponica* and *A. alpinus* in the high-elevation site, there was a tendency for slightly higher levels of both loss on ignition, total C and N content, NH_4^+-N and P in the *A. alpinus* samples. This is in line with Schulman et al. (1988), who found higher soil nutrient levels in the habitat of *A. alpinus* compared to two species of *Oxytropis*. In microhabitats with relatively high soil nutrient levels, such as those of *A. alpinus*, the contribution of biologically fixed N may make up too small a part of the total soil

N content to produce a significant difference in soil N below and outside the legume. Thus, the legumes' differing effects on soil N levels may be due to their differing microhabitats.

CONCLUSION

Soil nutrient levels, both N and C, where higher below than outside *Oxytropis lapponica* plants, most likely caused by biological N-fixation by the legumes. The percent increase was greatest in the site of highest abiotic stress, probably due to slightly lower background soil nutrient levels. However, there were few relationships between soil nutrient levels and legume size or nodule number. I found no difference in soil nutrient levels below and outside *Astragalus alpinus*, most likely due to this species' preference for more favourable microhabitats. This means that *O. lapponica*, but not *A. alpinus*, has the potential to influence N budgets in the study area, thereby facilitating other species.

REFERENCES

Askvik, H. 2008. Berggrunnskart Hardangerjøkulen 1416 2, M 1:50000. Norges geologiske undersøkelse.

Bellingham, P. J., Walker, R. L. & Wardle, D. A. 2001. Differential facilitation by a nitrogen-fixing shrub during primary succession influences relative performance of canopy tree species. Journal of Ecology 89: 861-875.

Belsky, A. J., Mwonga, S. M., Amundson, R. G., Duxbury, J. M. & Ali, A. R. 1993. Comparative effects of isolated trees on their undercanopy environments in high- and low-rainfall savannas. Journal of Applied Ecology 30: 143-155.

Bliss, L. C. 1971. Arctic and alpine plant life cycles. Annual Review of Ecology and Systematics 2: 405-438.

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

Carino, D. A. & Daehler, C. 2002. Can inconspicuous legumes facilitate alien grass invasions? Partridge peas and fountain grass in Hawai´i. Ecography 25: 33-41.

Dancer, W. S., Handley, J. F. & Bradshaw, A. D. 1977. Nitrogen accumulation in kaolin mining wastes in Cornwall I: Natural communities. Plant and Soil 48: 153-167.

Fornara, D. A. & Tilman, D. 2008. Plant functional composition influences rates of soil carbon and nitrogen accumulation. Journal of Ecology 96: 314-322.

Frey, B. & Schüepp, H. 1992. Transfer of symbiotically fixed nitrogen from berseem (*Trifolium alexandrinum* L.) to maize via vesicular-arbuscular mycorrhizal hyphae. New Phytologist 122: 447-454.

Fægri, K. 1967. The plant world at Finse, Norway. The University Botanical Museum, Bergen. Pp. 40.

Geesing, D., Felker, P. & Bingham, R. L. 2000. Influence of mesquite (*Prosopis glandulosa*) on soil nitrogen and carbon development: Implications for global carbon sequestration. Journal of Arid Environments 46: 157-180.

Gosling, P. 2005. Facilitation of *Urtica dioica* colonisation by *Lupinus arboreus* on a nutrient-poor mining soil. Plant Ecology 178: 141-148.

Granhall, U. & Lid-Torsvik, V. 1975. Nitrogen fixation by bacteria and free-living blue-green algae in tundra areas. In: Fennoscandian Tundra Ecosystems part 1: Plants and microorganisms (ed. Wielgolaski, F. E.), Springer-Verlag, Berlin. Pp. 305-315.

Gutschick, V. P. 1981. Evolved strategies in nitrogen acquisition by plants. The American Naturalist 118: 607-637.

Hacker, S. D & Gaines, S. T. 1997. Some implications of direct positive interactions for community species diversity. Ecology 78: 1990-2003.

Hagos, M. G. & Smit, G. N. 2005. Soil enrichment by *Acacia mellifera* subsp. *detinens* on nutrient poor sandy soil in a semi-arid southern African savanna. Journal of Arid Environments 61: 47-59.

Halvorson, J. J., Smith, J. L. & Franz, E. H. 1991. Lupine influence on soil carbon, nitrogen and microbial activity in developing ecosystems at Mount St. Helens. Oecologia 87: 162-170.

Halvorson, J. J. & Smith, J. L. 2009. Carbon and nitrogen accumulation and microbial activity in Mount St. Helens pyroclastic substrates after 25 years. Plant and Soil 315: 211-218.

Hunter, A. F. & Aarssen, L. W. 1988. Plants helping plants. BioScience 38: 34-40.

Høgh-Jensen, H. 2006. The nitrogen transfer between plants: An important but difficult flux to quantify. Plant and Soil 282: 1-5.

Johnson, D. A. & Rumbaugh, M. D. 1986. Field nodulation and the acetylene reduction activity of high-altitude legumes in the western United States. Arctic and Alpine Research 18: 171-179.

Karagatzides, J. D., Lewis, M. C. & Schulman, H. M. 1985. Nitrogen fixation in the high arctic at Sarcpa Lake, Northwest Territories. Canadian Journal of Botany 63: 974-979.

Kenny, S. T. & Cuany, R. L. 1990. Nitrogen accumulation and acetylene reduction activity of native lupines on disturbed mountain sites in Colorado. Journal of Range Management 43: 49-51.

Lid, J. & Lid, D. T. 2005. Norsk flora. 7. utgåva. Det Norske Samlaget, Oslo. Pp. 1230.

Lunde, T. 1962. An investigation into the pH-amplitude of some mountain plants in the county of Troms. Acta Borealia A. Scientia 20. Pp. 103.

Maron, J. L. & Connors, P. G. 1996. A native nitrogen-fixing shrub facilitates weed invasion. Oecologia 105: 302-312.

Maron, J. L. & Jefferies, R. L. 1999. Bush lupine mortality, altered resource availability and alternative vegetation states. Ecology 80: 443-454.

Moen, A. 1998. Nasjonalatlas for Norge: Vegetasjon. Statens Kartverk, Hønefoss. Pp. 199.

Myrold, D. D. & Huss-Danell, K. 2003. Alder and lupine enhance nitrogen cycling in a degraded forest soil in Northern Sweden. Plant and Soil 254: 47-56.

Nadelhoffer, K. J., Giblin, A. E., Shaver, G. R. & Linkins, A. E. 1992. Microbial processes and plant nutrient availability in arctic soils. In: Arctic ecosystems in a changing climate: An ecophysiological perspective (eds. Chapin, F. S., III, Jefferies, R. L., Reynolds, J. F., Shaver, G. R. & Svoboda, J.), Academic Press, Inc., San Diego. Pp. 281-300.

Olsen, S. L. 2008. Nitrogenfiksering hos to alpine mjeltarter. Bachelor thesis, Department of Natural Sciences, University of Agder. Pp. 31.

Palaniappan, V. M., Marrs, R. H. & Bradshaw, A. D. 1979. The effect of *Lupinus arboreus* on the nitrogen status of china clay wastes. Journal of Applied Ecology 16: 825-831.

Perroni-Ventura, Y., Montaña, C. & García-Oliva, F. 2006. Relationship between soil nutrient availability and plant species richness in a tropical semi-arid environment. Journal of Vegetation Science 17: 719-728.

Pugnaire, F. I. & Luque, M. T. 2001. Changes in plant interactions along a gradient of environmental stress. Oikos 93: 42-49.

Pugnaire, F. I., Haase, P., Puigdefabregas, J., Cueto, M., Clark, S. C. & Incoll, L. D. 1996. Facilitation and succession under the canopy of a leguminous shrub, *Retama sphaerocarpa*, in a semi-arid environment in south-east Spain. Oikos 76: 455-464.

Rodríguez-Echeverría, S. & Pérez-Fernándes, M. A. 2003. Soil fertility and herb facilitation mediated by *Retama sphaerocarpa*. Journal of Vegetation Science 14: 807-814.

Rossi, B. E. & Villagra, P. E. 2003. Effects of *Prosopis flexuosa* on soil properties and the spatial pattern of understorey species in arid Argentina. Journal of Vegetation Science 14: 543-550.

Rundel, P.W., Nilsen, E. T., Sharifi, M. R., Virginia, R. A., Jarrell, W. M., Kohl, D. H. & Shearer, G. B. 1982. Seasonal dynamics of nitrogen cycling for a *Prosopis* woodland in the Sonoran Desert. Plant and Soil 67: 343-353.

Schulman, H. M., Lewis, M. C., Tipping, E. M. & Bordeleau, L. M. 1988. Nitrogen fixation by three species of leguminosae in the Canadian high arctic tundra. Plant, Cell and Environment 11: 721-728.

Temperton, V. M., Mwangi, P. N., Scherer-Lorenzen, M., Schmid, B. & Buchmann, N. 2007. Positive interactions between nitrogen-fixing legumes and four different neighbouring species in a biodiversity experiment. Oecologia 151: 190-205.

The Norwegian Meteorological Institute 2010. eKlima. Data from weather station 25840 at Finse, normal period 1961-1990. To be found on the web page http://eklima.met.no. Last visited 02.02.2010.

Thomas, B. D. & Bowman, W. D. 1998. Influence of N_2-fixing *Trifolium* on plant species composition and biomass production in alpine tundra. Oecologia 115: 26-31.

Tiedemann, A. R. & Klemmedson, J. O. 1973. Effect of mesquite on physical and chemical properties of the soil. Journal of Range Management 1: 27-29.

Vitousek, P. M. & Field, C. B. 1999. Ecosystem constraints to symbiotic nitrogen fixers: A simple model and its implications. Biogeochemistry 46: 179-202.

APPENDIX 1. Determining NH_4^+-N using the Norwegian Standard for Water Samples (NS 4746) modified for analyses of soil extracts

Chemicals

- 1M KCl
- Sodium citrate buffer
- Reagent A
- Reagent B
- NH_4^+ standard solution (200 mg/l)

Preparation of chemicals

Sodium citrate buffer: To 600 ml deionized water, 80 g trisodium citrate dihydrate ($Na_3(COH(CH_2COO)_2COO)\times 2H_2O$) was added, and the resulting solution was diluted to 1000 ml with deionized water.

Reagent A: To 1000 ml sodium citrate buffer, 10.0 g salicylic acid and 1.0 g sodium nitroprusside ($Na_2(Fe(CN)_5NO)\times 2H_2O$) were added, and the resulting solution was kept in a dark glass bottle.

Reagent B: 20 g sodium hypochlorite was diluted to 1000 ml with deionized water and kept in a dark glass bottle.

Procedure

To 20 ml 1M KCl solution in a 100 ml glass bottle, 5 g of dry soil was added, and the suspension was left to shake for 1 hour at 150 rpm using a horizontal shaker (KS501 digital, IKA Werke GmbH & Co. KG, Staufen, Germany). Extracts were filtered (Whatman 42), poured into two glass tubes and frozen. In addition, a reference sample without soil was prepared. Half of the extracts were analyzed for NO_3^--N by the Soil and Water Chemistry laboratory of the Department of Plant and Environmental Sciences at the Norwegian University of Life Sciences in Ås. The rest was analyzed for NH_4^+-N as described below.

Each sample was diluted 5 times with 1M KCl. To 3 ml extract, 0.5 ml reagent A and 0.5 ml reagent B were added. Samples were incubated at room temperature, in the dark, for 2 hours, and absorption was measured spectrophotometrically at 655 nm (UV-2101PC spectrophotometer, Shimadzu Scientific Instruments, Columbia, USA). The reference sample was used to zero the instrument.

Standards with a known concentration of NH_4^+ (0 mg/l, 0.1 mg/l, 0.5 mg/l, 1.0 mg/l, 1.25 mg/l and 1.60 mg/l) were prepared and measured in the same way. The resulting standard curve was used to calculate the amount of NH_4^+ (in mg/l) in the samples. The amount of NH_4^+-N in mg/kg could then be calculated.

APPENDIX 2. The ammonium-lactate (P-Al) method for determining plant-available phosphorus (P)

Chemicals

- Al-solution: 0.1M NH_4^+-lactate + 0.4M acetic acid, adjusted to pH 3.75
- 5 % ascorbic acid
- Molybdat reagent
- P standard solution (1 ppm)

Preparation of chemicals

Al-solution: The concentrated Al-solution was diluted 10 times with deionized water to produce Al-solution for extraction (Al_e) and 100 times to produce Al-solution for washing (Al_w).

5 % ascorbic acid: To 100 ml deionized water, 5 g of ascorbic acid was added, and the resulting solution was kept in a dark glass bottle.

Molybdat reagent: To 34 ml of deionized water, 24 ml concentrated H_2SO_4 was added. To 20 ml deionized water, 2.6 g ammonium heptamolybdate tetrahydrate ($(NH_4)_6Mo_7O_{24} \times 4H_2O$) was added. To 20 ml deionized water, 0.07 g potassium antimony (III) oxide tartrate ($K(SbO)C_4H_4C_6 \times ½H_2O$) was added. The molybdat solution and the antimon solution were then added to the sulphuric acid while stirring. The resulting solution was diluted to 500 ml with deionized water and kept in a dark glass bottle.

Procedure

All glass equipment was placed in 0.5M HCl over night and rinsed with deionized water before use.

To 40 ml Al_e solution in a 100 ml glass bottle, 2 g of dry soil was added, and the suspension was left to shake for 1.5 hours at 150 rpm using a horizontal shaker (KS501 digital, IKA Werke GmbH & Co. KG, Staufen, Germany). Filters (Whatman blue ribbon 589/3) were washed two times with Al_w-solution, and the soil suspension was then filtered into a glass tube and placed in the refrigerator. In addition, a reference sample without soil was prepared.

The filtered samples were diluted 5 times with deionized water. To 10 ml diluted sample, 0.4 ml ascorbic acid and 0.4 ml molybdat reagent were added. Samples were incubated at room temperature for 1.5 hours and absorption was measured spectrophotometrically at 880 nm (UV-2101PC spectrophotometer, Shimadzu Scientific Instruments, Columbia, USA). The reference sample was used to zero the instrument. According to the P-Al standard procedure, the samples should be diluted 10 times and incubated for 15 minutes. However, initial analyses suggested that 5 times dilution and 1.5 hours incubation time was appropriate for extracts of alpine soil with a low P content.

Standards with a known concentration of P (0.3 ppm, 0.4 ppm, 0.5 ppm, 0.6 ppm, 0.7 ppm, 0.8 ppm, 0.9 ppm and 1.0 ppm) were prepared and measured in the same way. The resulting standard curve was used to calculate the amount of P (in ppm) in the samples. The amount of P in mg/kg was then calculated according to the following formula:

$$(\text{ppm} \times \text{ml extract} \times \text{dilution} \times 1000\ \text{g}) / (\text{g soil} \times 1000\ \text{ml}) = \text{mg/kg P}$$

APPENDIX 3. Amount of dry matter (%), loss on ignition (LOI) (%), total C content (%), total N content (%), NH_4^+-N (mg/kg), NO_3^--N (mg/kg), P (mg/kg) and pH in soil samples from below (+) and outside (-) the legume species *Astragalus alpinus* and *Oxytropis lapponica*, as well as the number of leaves, number of nodules and biomass (g) of the legumes, in the low (L), mid (M) and high (H)-elevation study sites at Mt. Sanddalsnuten, Finse, Norway, summer 2008. * denotes missing values.

Site	*A. alp*	*O. lap*	dry matter	LOI	C	N	NH_4-N	NO_3-N	P	pH	leaves	nodules	biomass
L	+		70.4	10.0	4.09	0.37	0.26	0.87	36.92	7.0	24	0	0.33
L	-		72.9	6.9	2.44	0.25	0.26	1.49	36.53	6.9			
L	+		63.1	14.3	6.77	0.53	1.24	2.45	37.72	6.9	52	27	0.90
L	-		59.5	14.8	8.54	0.61	1.05	5.08	38.20	6.7			
L	+		52.5	24.1	34.46	2.46	4.25	22.76	59.59	6.8	25	9	0.15
L	-		72.8	7.8	2.98	0.26	0.30	0.62	36.92	7.0			
L	+		17.4	94.9	18.98	1.36	1.77	9.83	42.92	6.7	12	8	0.12
L	-		27.1	59.4	30.93	2.33	2.63	21.41	47.77	6.7			
L	+		40.8	31.7	14.25	1.16	0.95	14.03	40.14	7.3	23	13	0.43
L	-		30.0	51.0	26.38	2.08	3.11	24.16	45.13	7.2			
L	+		39.3	37.6	22.16	1.80	1.84	22.23	46.74	7.2	73	81	1.10
L	-		31.3	48.6	28.55	2.21	4.68	27.30	49.97	7.2			
L	+		26.2	68.0	36.82	2.96	7.03	48.68	54.92	7.0	40	35	0.50
L	-		25.9	63.3	31.95	2.63	1.94	65.47	47.08	7.1			
L	+		28.1	59.0	29.88	2.12	2.56	30.46	48.56	6.7	31	5	0.51
L	-		*	42.9	22.33	1.71	1.13	12.55	41.05	6.9			
L	+		45.9	29.8	14.99	1.06	1.19	10.47	43.84	7.2	7	3	0.20
L	-		75.3	6.8	1.95	0.21	0.30	2.56	35.90	7.6			
L	+		44.1	32.3	15.21	1.30	0.99	16.59	39.34	6.5	66	2	1.11
L	-		37.1	43.2	22.52	1.87	1.67	9.00	43.24	6.1			
M		+	79.4	9.9	3.59	0.28	0.84	2.79	40.13	6.9	11	11	0.30
M		-	70.9	10.3	4.13	0.33	0.91	1.40	36.80	7.0			
M		+	85.3	5.3	1.88	0.20	0.53	4.19	37.26	7.5	17	2	0.39
M		-	83.2	5.7	1.88	0.23	0.35	4.79	36.96	7.7			
M		+	80.6	9.7	3.33	0.30	1.01	4.19	37.76	7.3	21	0	0.73
M		-	79.5	6.2	1.59	0.17	0.25	6.87	36.62	7.6			
M		+	81.6	5.3	1.78	0.20	0.51	5.48	36.82	7.1	14	12	0.17
M		-	82.6	5.6	1.55	0.17	0.34	4.15	40.90	7.1			
M		+	79.3	5.9	2.33	0.24	0.54	4.39	36.98	7.1	52	28	1.01
M		-	73.8	9.6	4.68	0.45	1.23	7.20	39.32	7.0			
M		+	80.0	7.2	3.39	0.31	0.75	8.55	37.41	7.2	21	8	0.44
M		-	82.7	4.8	1.22	0.13	0.27	4.84	37.45	7.5			
M		+	78.9	7.3	2.64	0.26	0.78	6.32	37.73	7.3	50	14	0.84
M		-	78.5	6.1	1.98	0.22	0.70	4.56	37.92	7.5			
M		+	75.6	7.7	2.47	0.27	0.73	6.43	37.01	7.4	21	2	1.33
M		-	83.1	5.9	1.77	0.19	0.38	5.59	37.67	7.8			
M		+	79.8	8.1	3.26	0.34	0.97	14.43	41.97	7.4	40	6	1.22
M		-	83.7	7.3	3.40	0.30	0.69	7.40	39.00	7.6			
M		+	79.0	7.1	2.27	0.23	0.72	12.80	37.75	7.6	37	18	0.68
M		-	87.3	6.6	2.05	0.22	1.04	4.20	38.80	7.8			

H		+	75.9	9.1	3.47	0.33	1.16	8.68	37.91	7.0	42	25	0.78
H		-	76.0	6.8	1.94	0.20	0.22	2.28	36.07	7.2			
H		+	77.5	10.3	3.52	0.31	0.87	8.19	37.31	6.5	73	3	1.22
H		-	79.5	6.0	2.40	0.24	0.39	3.85	36.33	7.2			
H		+	73.8	8.5	3.04	0.30	0.63	7.07	36.52	6.7	40	5	0.69
H		-	78.3	6.8	2.21	0.25	0.39	6.20	36.58	7.8			
H		+	73.6	9.2	3.57	0.34	0.91	6.91	36.84	6.9	69	0	0.96
H		-	78.7	6.1	1.93	0.24	0.61	5.87	37.15	7.3			
H		+	77.4	6.7	2.49	0.25	0.58	7.31	36.71	7.0	92	34	1.82
H		-	78.6	6.8	2.04	0.23	0.37	5.28	36.54	7.5			
H		+	74.7	8.4	3.20	0.27	0.64	6.99	36.51	7.1	49	3	0.53
H		-	78.4	7.2	2.50	0.23	0.58	8.39	36.65	7.3			
H		+	76.8	6.6	2.16	0.20	0.76	2.24	35.84	7.2	36	22	0.87
H		-	85.4	5.3	1.61	0.15	0.51	3.80	36.37	6.8			
H		+	71.1	11.1	4.79	0.37	1.08	4.23	37.36	7.2	32	17	0.30
H		-	78.8	8.0	2.53	0.24	0.49	4.72	36.12	7.3			
H		+	74.4	9.4	3.59	0.33	0.75	5.75	36.94	7.2	57	26	0.99
H		-	79.5	9.0	3.00	0.24	1.35	0.84	37.25	6.2			
H		+	65.1	15.5	9.43	0.70	1.05	8.96	38.66	6.8	69	45	1.40
H		-	78.8	6.1	2.10	0.21	0.50	2.19	36.35	7.0			
H	+		78.6	6.0	1.60	0.15	0.82	0.53	36.69	5.3	22	0	0.34
H	-		78.3	7.5	2.49	0.22	1.10	0.53	36.76	5.1			
H	+		74.1	9.7	3.43	0.28	1.10	3.33	40.92	7.4	26	27	0.41
H	-		75.0	8.5	3.54	0.30	1.03	1.32	37.06	5.8			
H	+		75.0	7.5	2.42	0.22	0.94	2.34	36.00	5.9	45	7	0.57
H	-		78.4	6.4	2.69	0.25	0.89	7.39	36.52	6.8			
H	+		79.6	7.0	2.65	0.21	1.73	1.97	36.86	6.6	70	8	1.10
H	-		78.8	6.9	2.17	0.19	0.68	4.56	38.55	7.0			
H	+		68.2	11.9	4.76	0.39	1.01	6.67	37.34	7.1	6	8	0.16
H	-		69.3	12.4	5.83	0.44	0.87	3.04	37.48	6.3			
H	+		78.5	9.0	4.07	0.26	0.14	0.40	38.97	6.3	56	11	0.90
H	-		78.6	6.9	2.89	0.27	0.53	3.44	36.43	7.1			
H	+		68.3	11.1	4.58	0.37	0.86	6.68	36.79	6.8	9	5	0.15
H	-		74.1	9.4	4.70	0.36	0.71	0.56	37.36	5.7			
H	+		70.6	9.6	3.79	0.32	0.63	4.24	36.59	6.5	29	23	0.31
H	-		70.0	10.8	5.21	0.44	1.76	2.16	37.73	5.0			
H	+		72.5	9.9	4.24	0.30	1.32	3.20	36.86	6.3	53	4	0.34
H	-		79.7	6.6	2.50	0.21	0.51	2.32	36.04	6.2			
H	+		74.8	10.0	4.63	0.38	0.86	8.46	37.99	7.0	31	14	0.39
H	-		79.7	6.9	2.76	0.23	0.31	3.59	36.68	6.9			

PART 2. NITROGEN-FIXING LEGUMES POSITIVELY AFFECT DENSITY, BUT NOT PERFORMANCE, OF AN ALPINE HERB

ABSTRACT

Facilitation is common among plants. According to the stress-gradient hypothesis, plant-plant interactions shift from competition to facilitation with increasing abiotic stress, implying that facilitation is an important process in climatically stressful alpine areas. In these low-productivity habitats, nitrogen (N)-fixing legumes may facilitate neighbouring species by increasing soil N levels. In this study I examined how leaf density, size, growth rate, reproduction and N content of the herb Thalictrum alpinum *L. differed with and without two alpine legumes,* Astragalus alpinus *L. and* Oxytropis lapponica *(Wahlenb.) Gay, in two sites situated along a gradient in abiotic stress at Finse, Norway. I found that leaf density of* T. alpinum *increased with the presence of both legumes species, most likely due to increased clonal growth caused by elevated soil N levels. The increase was greatest where abiotic stress was highest, supporting the stress-gradient theory. The lack of response in* T. alpinum *plant size, growth rate and reproduction, as well as a reduction in N content, may be due to increased allocation of resources to clonal growth. This study shows that the presence of two legume species can increase the density of a single target species, implying that they might have the ability to influence the plant community as a whole.*

INTRODUCTION

Facilitation is a fundamental process in plant communities (Callaway 1995). Positive interactions among plants have been observed in a wide variety of habitats; from deserts, savannas and grasslands to shrublands, forests, wetlands and tundra (Callaway 1995 and references therein). According to the stress-gradient hypothesis, the net outcome of plant-plant interactions shifts from competition to facilitation with increasing abiotic stress (Bertness & Callaway 1994, Brooker & Callaghan 1998, Callaway et al. 2002 - but see Maestre et al. 2005, Bowker et al. 2010). Under harsh environmental conditions any stress reduction due to neighbouring plants should be favourable and may outweigh the negative impact of competition (Callaway 1995, Callaway & Walker 1997). However, recent studies indicate that under *extreme* abiotic conditions, the positive effect of facilitation may decrease as biotic interactions collapse (Michalet et al. 2006, Forey et al. 2010).

Nitrogen (N)-fixing legumes have a great potential as facilitators, since N is a limiting factor for plant growth in most terrestrial ecosystems. Both greenhouse experiments and field studies have shown positive effects of legumes on neighbouring species, both in terms of survival (Espigares et al. 2004), plant biomass (Jefferies et al. 1981, Belsky et al. 1993, Franco-Pizaña et al. 1996, Maron & Connors 1996, Pugnaire et al. 1996, Quinos et al. 1998, Maron & Jefferies 1999, Bellingham et al. 2001, Pugnaire & Luque 2001, Rodríguez-Echeverría & Pérez-Fernándes 2003, Espigares et al. 2004, Gosling 2005), plant N content (Pugnaire et al. 1996, Quinos et al. 1998, Thomas & Bowman 1998, Maron & Jefferies 1999, Bellingham et al. 2001, Lee et al. 2003, Rodríguez-Echeverría & Pérez-Fernándes 2003, Temperton et al. 2007) and flowering (Morris & Wood 1989, Pugnaire et al. 1996).

If the effect of positive interactions increases with increasing abiotic stress, facilitation should be an important process in alpine areas, which are considered climatically stressful environments characterized by low temperatures, strong winds and unstable soils (e.g. Billings and Mooney 1968). In a global study Callaway et al. (2002) found that biomass, growth and reproduction of alpine plants increased when growing close to other plants. Competition was the dominating interaction at intermediate altitude, where the environment is less stressful, while at high altitudes, where conditions are harsher, positive interactions dominated. Choler et al. (2001) found the same pattern in the Alps, and Wang et al. (2008) concluded that facilitation was the dominant interaction in alpine Tibet (but see Moen 1993, Olofsson et al. 1999). The effect also seemed to vary between habitats at the same altitude, with the greatest effect in the most exposed areas (Choler et al. 2001, Callaway et al. 2002, Kikvidze et al. 2005).

As low biomass production and litter decomposition rates limit the nutrient supply in alpine soils (e.g. Bliss 1971, Nadelhoffer et al. 1992), N from N-fixing legumes should have a positive effect on neighbouring plants. To my knowledge only one study have examined how the presence of legumes affects the growth of adjacent plants in alpine areas: Thomas & Bowman (1998), who studied *Trifolium dasyphyllum* in the Rocky Mountains, found higher graminoid and forb leaf N concentrations in legume patches. Biomass of non-legumes did, however, not increase where legumes were present. They concluded that the legumes may have both positive and negative effects on neighbouring plants.

In this study I examine how leaf density, size, growth rate, reproduction and N content of the alpine herb *Thalictrum alpinum* L. differ with and without the presence of two alpine legumes, *Astragalus alpinus* L. and *Oxytropis lapponica* (Wahlenb.) Gay, in two sites situated along a gradient in abiotic stress at Finse, Norway. Klanderud (2005) and Klanderud & Totland (2005) found that addition of nutrients increased growth of *T. alpinum* in the study area, indicating that this species is nutrient limited. In Part 1 I showed that the presence of *O. lapponica* increased soil N levels at Finse and that the increase was greatest in the most stressful end of the gradient. If *T. alpinum* in the study area is indeed nutrient limited, the presence of legumes, at least *O. lapponica*, should have a facilitative effect, and the effect should increase with increasing abiotic stress. I also examine whether growth of *T. alpinum* is related to legume size.

Specifically I ask the following questions:

- Do leaf density, size, growth rate, reproduction or N content of *Thalictrum alpinum* differ with and without the presence of legumes?
- Does the difference in leaf density, size, growth rate, reproduction or N content of *T. alpinum* with and without legumes increase with increasing abiotic stress?
- Are leaf density, size, growth rate, reproduction or N content of *T. alpinum* related to the size of the legumes?

MATERIALS AND METHODS

Study area and study species

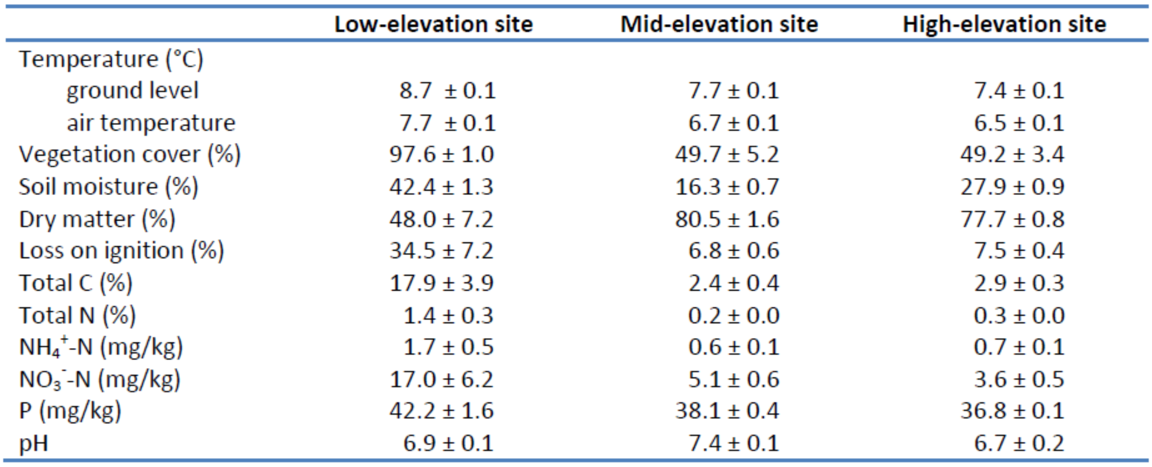
Field work was conducted during the summer 2008 at Finse, southwest Norway (60°36’59’’N, 07°31’23’’E). Finse has a slightly oceanic climate (Moen 1998) with a mean monthly temperature and rainfall during the growing season (May-September) of 4.4 °C and 89 mm, respectively (The Norwegian Meteorological Institute 2010). The study area was located on the southwest-facing slope of Mt. Sanddalsnuten (1554 m.a.s.l.) in the middle alpine zone. The bedrock consists of phyllite with strains of marble (Askvik 2008), and *Dryas* heath, dominated by the dwarf-shrub *Dryas octopetala*, is the main vegetation type in the study area.

The study was conducted in three sites (10 × 10 m) situated along the slope of Mt. Sanddalsnuten: a low-elevation (1460 m.a.s.l.), mid-elevation (1510 m.a.s.l.) and high-elevation site (1554 m.a.s.l.), with a difference in altitude of about 50 m between each site. The low and mid-elevation sites were fenced to prevent sheep grazing. The study species *Astragalus alpinus* was present in the low and high-elevation sites, while *Oxytropis lapponica* was present in the mid and high-elevation sites.

Thalictrum alpinum was chosen as the target species for this study as it is widespread in the study area and was found in sufficient numbers in all three sites. *T. alpinum* is a small perennial herb of circumpolar distribution (Lid & Lid 2005), in alpine areas typically occurring in rich heaths and meadows. This is a clonal species producing ramets from below-ground rhizomes (e.g. Steven & Waller 2004). However, individual plants and ramets are hard to distinguish, and I hereafter refer to both as *plants*.

To document differences in abiotic conditions along the slope of Mt. Sanddalsnuten, I measured temperature at ground level (+ 0 cm) and air temperature (+ 100 cm) in each site from July 12th to August 9th 2009 using data loggers (Tinytag Plus, Gemini Data Loggers Ltd., Chichester, UK). In each site I also estimated total vegetation cover (%), measured soil moisture (%) (Moisture Meter HH2, Delta-T Devices Ltd., Cambridge, UK) and collected soil samples (see Part 1). Both temperature, vegetation cover, soil moisture and soil nutrient contents were higher in the low-elevation site than in the mid and high-elevation sites (Table 1). The low-elevation site was also less exposed to wind (pers.obs.). Between the mid and high-elevation sites I found only minor differences in vegetation cover and abiotic conditions. However, the difference in soil nutrient levels below and outside *O. lapponica* was greater in the high-elevation site than the mid-elevation site (Part 1).

Table 1. Temperature, vegetation cover and soil properties of the three study sites at Mt. Sanddalsnuten, Finse, Norway. Data on soil variables and vegetation cover were collected in 2008, temperature and soil moisture data in 2009. Both vegetation cover and all soil characteristics were measured outside legumes. Vegetation cover and soil moisture were measured in 15 × 15 cm sampling quadrats. Temperature is given as daily means at ground level (+ 0 cm) and as air temperature (+ 100 cm) based on 29 days of measurements. All values are mean ± 1 SE.



	Low-elevation site	**Mid-elevation site**	**High-elevation site**
Temperature (°C)			
ground level	8.7 ± 0.1	7.7 ± 0.1	7.4 ± 0.1
air temperature	7.7 ± 0.1	6.7 ± 0.1	6.5 ± 0.1
Vegetation cover (%)	97.6 ± 1.0	49.7 ± 5.2	49.2 ± 3.4
Soil moisture (%)	42.4 ± 1.3	16.3 ± 0.7	27.9 ± 0.9
Dry matter (%)	48.0 ± 7.2	80.5 ± 1.6	77.7 ± 0.8
Loss on ignition (%)	34.5 ± 7.2	6.8 ± 0.6	7.5 ± 0.4
Total C (%)	17.9 ± 3.9	2.4 ± 0.4	2.9 ± 0.3
Total N (%)	1.4 ± 0.3	0.2 ± 0.0	0.3 ± 0.0
NH_4^+-N (mg/kg)	1.7 ± 0.5	0.6 ± 0.1	0.7 ± 0.1
NO_3^--N (mg/kg)	17.0 ± 6.2	5.1 ± 0.6	3.6 ± 0.5
P (mg/kg)	42.2 ± 1.6	38.1 ± 0.4	36.8 ± 0.1
pH	6.9 ± 0.1	7.4 ± 0.1	6.7 ± 0.2

Study design

To examine whether leaf density, size, growth rate, reproduction or N content of *Thalictrum alpinum* differ with and without the presence of legumes, I selected 120 pairs of *T. alpinum* plants. Each pair consisted of one plant growing in close vicinity (<5 cm) of a legume and one growing at least 30 cm away from any legume, but no more than 100 cm away from the other *T. alpinum* in the pair. In the low-elevation site I had 30 *T. alpinum* pairs associated with *Astragalus alpinus*, in the mid-elevation site 30 pairs associated with *Oxytropis lapponica* and in the high-elevation site 30 pairs associated with *O. lapponica* and 30 with *A. alpinus*.

Leaf density: I placed a 15 × 15 cm sampling quadrat around each *T. alpinum* plant and counted all *T. alpinum* leaves within the quadrat, as well as the total number of legume leaves.

Growth: I measured the size of each *T. alpinum* plant, first in the end of July and then again in the end of August, by counting the leaves and measuring maximum length (including petiole) and width of every leaf using a digital caliper. I also counted the number of leaves of each legume associated with a *T. alpinum* plant.

Reproduction: I counted the number of flowers and/or fruits of each *T. alpinum* plant in the end of July and then again in the end of August. In August I also measured the height of the inflorescence, the number of flowers having produced fruits and the number of fruits per flower on all fertile plants. Fruits were dried at 60 °C for 24 hours and weighed. Only 17 out of 240 plants produced fruits this year, which made it difficult to compare fruit number or weight between plants. Instead I categorized each *T. alpinum* plant as either *fertile* or *infertile*, depending on whether it had produced an inflorescence, and analyzed for differences in fertility.

N content: I harvested all *T. alpinum* plants at the end of the growing season, and the plant material was dried at 60 °C for 24 hours. Plant CHN-N content was analyzed by the Department of Animal and Aquacultural Sciences at the Norwegian University of Life Sciences in Ås using gas chromatography. Individual *T. alpinum* plants were too small to be analyzed separately, so in the final analysis 3 plants were analyzed together.

Primary data is found in Appendix 1 and 2.

Statistical analyses

For each *Thalictrum alpinum* plant I calculated plant size in July (W_1) and August (W_2) as (number of leaves × width × length) and mean plant size as ($[W_1 + W_2] / 2$). I also calculated the relative growth rate as ($[\ln W_2 - \ln W_1] / [t_2 - t_1]$) (Hunt 1982) where $[t_2 - t_1]$ is the number of days between the two measurements.

I used analysis of variance (ANOVA) to test whether leaf density, mean size, growth rate or N content of *T. alpinum* differed with the presence of legumes and/or between sites. All tests were two-way ANOVAs with *site* and *legume presence* as fixed main factors. Except for the analysis of N content, I used *pair* as a random nested factor under *site* when significant (Table 2). If the interaction between *site* and *legume presence* was significant, I used a paired t-test – except for N content, for which I used a two-sample t-test – to compare *T. alpinum* parameters with and without legume presence within each site. The data on *T. alpinum* density was ln transformed and plant size data was square root transformed to fulfill the ANOVA assumptions of normality and equal variances. All figures are shown with untransformed data. Analyses were performed using the General Linear Model, Paired t-test and 2-Sample t-test procedures in Minitab 15.1.1.0 for Windows.

I used simple regression to test whether there was a relationship between the leaf density of *T. alpinum* and legume leaf density or between mean *T. alpinum* size, growth rate and N content and the size (number of leaves) of the associated legumes. Only quadrats with legumes were included in the analyses, and separate tests were performed for each site as I expected to see differences between sites of differing abiotic stress. Analyses were performed using the Regression procedure in Minitab 15.1.1.0 for Windows.

I used Pearson's chi-quadrat test to test whether there was a difference in fertility between *T. alpinum* plants growing with and without legumes. First I tested for differences in fertility between sites, then for differences with and without legumes within each site. I also used logistic regression to test whether there was a relationship between fertility and the size of *T. alpinum* or legume size within each site. In this analysis I only included the *T. alpinum* plants growing with legumes and used T. alpinum *size* and *legume leaf number* as predictors. Analyses were performed using the Cross Tabulation and Binary Logistic Regression procedures in Minitab 15.1.1.0 for Windows.

In all tests data for the two legume species was analyzed separately, as analyses in Part 1 suggested that they differ in their effect on soil N levels and hence their potential facilitative ability.

RESULTS

Astragalus alpinus

Leaf density: The ANOVA showed a significant interaction between *site* and *legume presence* for *Thalictrum alpinum* leaf density (Fig. 1A, Table 2). The t-tests revealed that leaf density was almost significantly higher in quadrats with *Astragalus alpinus* in the high-elevation site (T = 2.00, p = 0.055), while there was no difference in leaf density between quadrats with and without *Astragalus alpinus* in the low-elevation site (T = 1.28, p = 0.21). The regression analyses showed no significant relationship between the leaf density of *T. alpinum* and legume leaf density (p = 0.55, R^2 = 1.3 % for the low-elevation site; p = 0.43, R^2 = 2.3 % for the high-elevation site).

Growth: The ANOVA showed that *T. alpinum* was significantly larger (Fig. 1B) in the low-elevation site compared to the high-elevation site, while there was no difference in growth rate (Fig. 1C) between sites (Table 2). Moreover, there was no significant difference in size or growth rate between plants growing with and without legumes. The regression analyses showed no significant relationship between the size ($p = 0.47$, $R^2 = 1.9$ % for the low elevation site; $p = 0.36$, $R^2 = 3.0$ % for the high elevation site) or growth rate ($p = 0.78$, $R^2 = 0.3$ % for the low-elevation site; $p = 0.74$, $R^2 = 0.4$ % for the high-elevation site) of *T. alpinum* and legume size.

Reproduction: The chi-quadrat test showed no difference in fertility of *T. alpinum* growing with and without *A. alpinus* in neither the low-elevation site ($\chi^2 = 0.61$, $p = 0.44$) nor the high-elevation site ($\chi^2 = 0.88$, $p = 0.35$) (Fig. 2). However, the low-elevation site had a significantly higher number of fertile *T. alpinum* plants than the high-elevation site ($\chi^2 = 14.1$, $p < 0.001$). The logistic regression showed no relationship between fertility and *T. alpinum* size ($p = 0.71$, OR = 1.00 for the low-elevation site; $p = 0.08$, OR = 1.00 for the high-elevation site) or legume size ($p = 0.75$, OR = 0.99 for the low-elevation site; $p = 0.49$, OR = 0.96 for the high-elevation site).

N content: The ANOVA showed an almost significant interaction between *site* and *legume presence* for *T. alpinum* N levels (Table 2), but the t-tests detected no significant differences in N levels in *T. alpinum* plants growing with and without *A. alpinus* in neither the low-elevation site ($T = 1.64$, $p = 0.12$) nor the high-elevation site ($T = 1.31$, $p = 0.21$) (Fig. 1D). The regression analyses showed no significant relationship between *T. alpinum* N content and legume size ($p = 0.22$, $R^2 = 18.1$ % for the low-elevation site; $p = 0.63$, $R^2 = 3.5$ % for the high-elevation site).

Oxytropis lapponica

Leaf density: The ANOVA showed that the leaf density of *Thalictrum alpinum* was significantly higher in quadrats where *Oxytropis lapponica* was present (Fig. 1A, Table 2). The increase in leaf density with *O. lapponica* was greater in the high-elevation site (75.2 %) than the mid-elevation site (61.0 %). However, leaf density was significantly higher in the mid-elevation site compared to the high-elevation site. The regression analyses showed no relationship between *T. alpinum* leaf density and legume leaf density ($p = 0.54$, $R^2 = 1.3$ % for the mid-elevation site; $p = 0.55$, $R^2 = 1.3$ % for the high-elevation site).

Growth: The ANOVA showed that *T. alpinum* was significantly larger (Fig. 1B) in the mid-elevation site compared to the high-elevation site, while there was no difference in growth rate (Fig. 1C) between sites (Table 2). Moreover, there was no difference in size or growth rate between plants growing with and without *O. lapponica*. The regression analyses showed no relationship between the size ($p = 0.12$, $R^2 = 8.3$ % for the mid-elevation site; $p = 0.32$, $R^2 = 3.6$ % for the high-elevation site) or growth rate ($p = 0.87$, $R^2 = 0.1$ % for the mid-elevation site; $p = 0.69$, $R^2 = 0.6$ % for the high-elevation site) of *T. alpinum* and legume size.

Reproduction: The chi-quadrat test showed no difference in *T. alpinum* fertility with and without legumes in neither the mid-elevation site ($\chi^2 < 0.001$, $p = 1.00$) nor the high-elevation site ($\chi^2 = 0.1$, $p = 0.79$), and there was no difference in fertility between the two sites ($\chi^2 = 0.98$, $p = 0.32$) (Fig. 2). The logistic regression showed an almost significant negative relationship between *T. alpinum* fertility and legume size in the mid-elevation site ($p = 0.056$, OR = 0.87), but not the high-elevation site ($p = 0.36$, OR = 0.99, and there was no relationship between fertility and *T. alpinum* size in either site ($p = 0.83$, OR = 1.00 for the mid-elevation site; $p =0.46$, OR = 1.00 for the high-elevation site).

N content: The ANOVA showed a significant interaction between *site* and *legume presence* for *T. alpinum* N levels (Fig. 1D, Table 2). The t-tests revealed that the N content of *T. alpinum* was lower in quadrats with *O. lapponica* in the mid-elevation site ($T = 2.21$, $p = 0.041$), while there was no difference in N content with and without *O. lapponica* in the high-elevation site ($T = 1.28$, $p = 0.22$) (Fig. 1). The regression analyses showed no relationship between *T. alpinum* N content and legume size ($p = 0.78$, $R^2 = 1.1$ % for the low elevation site; $p = 0.21$, $R^2 = 19.0$ % for the high elevation site).

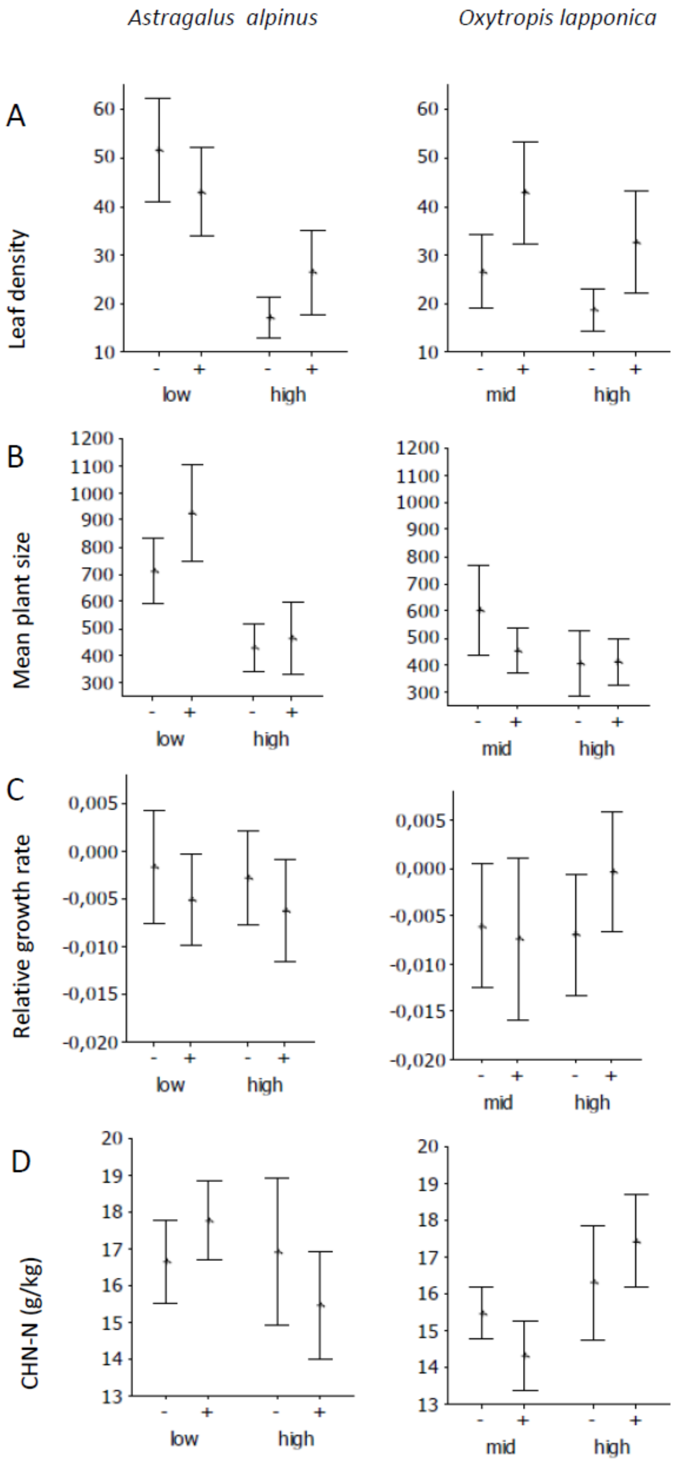


Fig. 1. Leaf density (the number of leaves in each sampling quadrat) (A), mean plant size (number of leaves × width × length) (B), relative growth rate (C) and CHN-N content (D) of Thalictrum alpinum *growing with (+) and without (-) the legumes* Astragalus alpinus *(left) and* Oxytropis lapponica *(right) in the low, mid and high-elevation study sites at Mt. Sanddalsnuten, Finse, Norway, summer 2008. All figures show mean values with 95 % CI.*

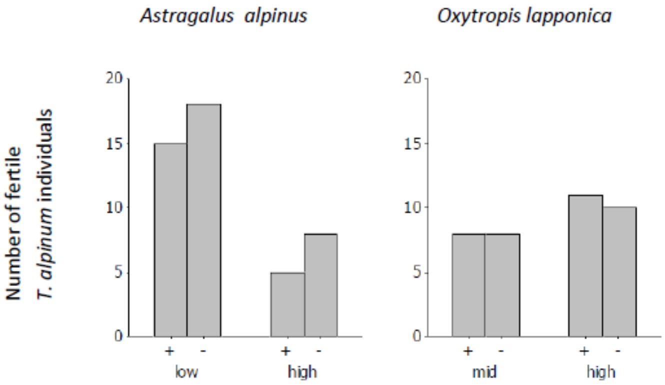


Fig. 2. The total number of fertile Thalictrum alpinum *plants growing with (+) and without (-) the legumes* Astragalus alpinus *(left) and* Oxytropis lapponica *(right) in the low, mid and high-elevation study sites at Mt. Sanddalsnuten, Finse, Norway, summer 2008.*

Table 2. $F_{d.f.}$ *and p-values for ANOVA analyses of leaf density, mean plant size, relative growth rate and CHN-N (g/kg) content of* Thalictrum alpinum *growing with and without the legumes* Astragalus alpinus *and* Oxytropis lapponica *in three study sites at Mt. Sanddalsnuten, Finse, Norway summer 2008.* ***Bold*** *letters indicate p-values significant at the 0.05 level.*

Source of variation	**Leaf density**		**Size**		**Growth rate**		**CHN-N (g/kg)**	
	$F_{d.f.}$	P	$F_{d.f.}$	P	$F_{d.f.}$	P	$F_{d.f.}$	P
A. alpinus								
site	$33.47_{1,58}$	**<0.001**	$26.65_{1,58}$	**<0.001**	$0.18_{1,116}$	0.670	$2.51_{1,36}$	0.122
A. alp	$0.89_{1,58}$	0.349	$3.21_{1,58}$	0.078	$1.78_{1,116}$	0.185	$0.05_{1,36}$	0.824
site × A. alp	$5.64_{1,58}$	**0.021**	$2.54_{1,58}$	0.117	$<0.01_{1,116}$	0.999	$3.95_{1,36}$	**0.054**
pair	$2.23_{58,58}$	**0.001**	$2.21_{58,58}$	**0.001**	—	—	—	—
O. lapponica								
site	$4.79_{1,58}$	**0.033**	$4.45_{1,116}$	**0.037**	$0.81_{1,115}$	0.370	$14.85_{1,36}$	**<0.001**
O. lap	$13.48_{1,58}$	**0.001**	$0.49_{1,116}$	0.486	$0.61_{1,115}$	0.436	$0.00_{1,36}$	0.985
site × O. lap	$0.09_{1,58}$	0.762	$1.12_{1,116}$	0.291	$1.39_{1,115}$	0.241	$4.92_{1,36}$	**0.033**
pair	$1.51_{58,58}$	**0.058**	—	—	—	—	—	—

DISCUSSION

Leaf density of *Thalictrum alpinum* plants increased with the presence of both *Astragalus alpinus* and *Oxytropis lapponica* at Finse. Plant size, growth rate or reproduction of *T. alpinum* did, however, not change with the presence of either legume species, while N content decreased with the presence of *O. lapponica* in one of the study sites.

The increase in *T. alpinum* leaf density with the presence of *O. lapponica* is most likely due to increased N availability, as in Part 1 I found increased soil N levels below *O. lapponica* plants at Finse. This anticipation is supported by the findings of Klanderud (2005) and Klanderud & Totland (2005) that *T. alpinum* respond positively to nutrient addition in the study area. The percent increase in *T. alpinum* leaf density was greatest in the high-elevation site, indicating that facilitation was strongest where abiotic stress is highest, supporting the stress-gradient hypothesis. In Part 1 I found a greater difference in soil nutrient levels below and outside *O. lapponica* in the high-elevation than the mid-elevation site, indicating that the increased facilitation in the site of highest abiotic stress may be due to a proportionally greater improvement of soil conditions, as suggested by Pugnaire & Luque (2001).

While the increase in *T. alpinum* leaf density with *O. lapponica* is probably due to an increase in soil N levels below the legumes (Part 1), the almost significant increase in *T. alpinum* leaf density with *A. alpinus* is more difficult to explain. In Part 1 I found no significant difference in soil nutrient levels below and outside *A. alpinus*, although there was a tendency for increased nutrient content in soil below the legumes. It could be that this tendency was great enough for *A. alpinus* to cause an increase in the leaf density of *T. alpinum* in the high-elevation site, where abiotic stress is relatively high. A sheltering effect of *A. alpinus* seems unlikely, as previous studies have shown that the number of leaves of *T. alpinum* is reduced by the presence of neighbouring plants in the study area (Klanderud 2005, Klanderud & Totland 2005). However, further studies are needed to determine whether the tendency for increased soil nutrient levels below *A. alpinus* is enough to cause the observed increase in leaf density.

I did not find any relationship between *T. alpinum* leaf density, size, growth rate or N content and legume size. In Part 1 I found few significant relationships between legume size and soil nutrient levels, and if legume size is not related to soil nutrient levels, it is not likely to be related to the growth of *T. alpinum* either.

The increase in leaf density of *T. alpinum* could be due to increased clonal growth. As a clonal species *T. alpinum* may respond to increased soil N levels below legumes by producing more clones, thereby increasing leaf density. The increase in leaf density could also be due to increased establishment. However, Morris & Wood (1989) found that *Lupinus lepidus* decreased seedling survival of adjacent plants at Mount St. Helens, and according to Klanderud (2005) and Klanderud & Totland (2005) seedlings of *T. alpinum* are “rarely found” in the study area. Therefore, it seems more likely that the increase in leaf density is due to increased clonal growth, although I did not examine this directly.

The N content of *T. alpinum* did not change with the presence of either legume species except for in the mid-elevation site, where it decreased with the presence of *O. lapponica*. This is in line with Atkin & Collier (1992), who found that plant N content did not change following nutrient addition. Körner (2003) interprets this as an allocation of N to increased growth rather than increasing tissue N content, resulting in constant tissue N levels. The constant N content of *T. alpinum* in the high-elevation site, as well as the reduced N content with *O. lapponica* in the mid-elevation site, could be due to allocation of resources to increased clonal growth, which may “dilute” additional N, as suggested by Körner (2003). This is, however, not in accordance with most studies of N content in species growing near legumes, reporting an increase in both N content *and* biomass (Pugnaire et al. 1996, Quinos et al. 1998, Bellingham et al. 2001, Rodríguez-Echeverría & Pérez-Fernándes 2003).

I found no difference in *T. alpinum* fertility with and without the presence of either legume species. According to Steven & Waller (2004), *T. alpinum* in general allocates resources to clonal growth rather than reproduction. The observed increase in leaf density, but not fertility, of *T. alpinum* in this study could be due to such an allocation to increased clonal growth. This is also supported by Klanderud (2005), who found that ”nutrient addition (...) had only minor effects on the sexual reproduction of *Thalictrum*”. Increased clonal growth may, however, be seen as an indirect investment in future reproduction, as an increased number of clones can produce more inflorescences (Sandvik & Totland 2000).

If the increased leaf density of *T. alpinum* is indeed due to allocation of resources to clonal growth, this may explain the lack of response in *T. alpinum* size or growth rate. Another possible explanation may be that growing near a legume inhibits *T. alpinum* growth. *T. alpinum* is a small and delicate herb and may be susceptible to competition. Levine (2000) and Franks (2003) found that the presence of a facilitator plant increased survival of target species, but that growth of the same species was significantly reduced by competition with the facilitator. This is also in accordance with Klanderud (2005) and Klanderud & Totland (2005), who found that the removal of neighbours increased the number of leaves of *T. alpinum* in the study area, indicating competitive inhibition of growth. The negative relationship between the fertility of *T. alpinum* and the size of the associated *O. lapponica* plants in the mid-elevation site may also indicate competition. If the growth and fertility of *T. alpinum* is indeed inhibited by competing *A. alpinus* and *O. lapponica* plants, my results support the conclusion of Thomas & Bowman (1998) that legumes have both positive and negative effects on neighbouring species.

The lack of size and growth rate responses of *T. alpinum* may also be due to the timing of the fieldwork. My second measurement was done rather late in the growing season, possibly after the peak in *T. alpinum* growth when the plants had started to die back. This is supported by my findings of very low or negative growth rates. The study was done in an exceptionally warm and dry summer, which may have sped up the dieback. If this is the case, bad timing of measurements could camouflage differences in size and/or growth rates with and without legumes.

CONCLUSION

Leaf density of *Thalictrum alpinum* increased with the presence of *Astragalus alpinus* and *Oxytropis lapponica* at Finse, most likely due to increased clonal growth caused by elevated soil N levels below the legumes. Accordingly, allocation of resources to clonal growth may have caused of the lack of response in *T. alpinum* plant size, growth rate and reproduction, as well as the reduction in N content. However, there were few relationships between *T. alpinum* parameters and legume size. The increase in leaf density was greatest in the high-elevation site, supporting the stress-gradient hypothesis. In this study I have shown that the presence of two alpine legume species can affect the density of single target species. Changes in density of individual species level may in turn lead to community-level changes, implying that the legumes may have the ability to influence the plant community as a whole.

REFERENCES

Askvik, H. 2008. Berggrunnskart Hardangerjøkulen 1416 2, M 1:50000. Norges geologiske undersøkelse.

Atkin, O. K. & Collier, D. E. 1992. Relationship between soil nitrogen and floristic variation in late snow areas of the Kosciusko alpine region. Australian Journal of Botany 40: 139-149.

Bellingham, P. J., Walker, L. R. & Wardle, D. A. 2001. Differential facilitation by a nitrogen-fixing shrub during primary succession influences relative performance of canopy tree species. Journal of Ecology 89: 861-875.

Belsky, A. J., Mwonga, S. M., Amundson, R. G., Duxbury, J. M. & Ali, A. R. 1993. Comparative effects of isolated trees on their undercanopy environments in high- and low-rainfall savannas. Journal of Applied Ecology 30: 143-155.

Bertness, M. D. & Callaway, R. M. 1994. Positive interactions in communities. Trends in Ecology and Evolution 9: 191-193.

Billings, W. D. & Mooney, H. A. 1968. The ecology of arctic and alpine plants. Biological Reviews 43: 481-529.

Bliss, L. C. 1971. Arctic and alpine plant life cycles. Annual Review of Ecology and Systematics 2: 405-438.

Bowker, M. A., Soliveres, S. & Maestre, F. T. 2010. Competition increases with abiotic stress and regulates the diversity of biological soil crusts. Journal of Ecology 98: 551-560.

Brooker, R. W. & Callaghan, T. V. 1998. The balance between positive and negative plant interactions and its relationship to environmental gradients: A model. Oikos 81: 196-207.

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

Callaway, R. M., Brooker, R. W., Choler, P., Kikvidze, Z., Lortie, C. J., Michalet, R., Paolini, L., Pugnaire, F. I., Newingham, B., Aschehoug, E. T., Armas, C., Kikodze, D. & Cook, B. J. 2002. Positive interactions among alpine plants increase with stress. Nature 417: 844-848.

Callaway, R. M. & Walker, L. R. 1997. Competition and facilitation: A synthetic approach to interactions in plant communities. Ecology 78: 1958-1965.

Choler, P., Michalet, R. & Callaway, R. M. 2001. Facilitation and competition on gradients in alpine plant communities. Ecology 82: 3295-3308.

Espigares, T., López-Pintor, A. & Benayas, J. M. R. 2004. Is the interaction between *Retama sphaerocarpa* and its understorey herbaceous vegetation always reciprocally positive? Acta Oecologica 26: 121-128.

Forey, E., Touzard, B. & Michalet, R. 2010. Does disturbance drive the collapse of biotic interactions at the severe end of a diversity-biomass gradient? Plant Ecology 206: 287-295.

Franco-Pizaña, J. G., Fulbright, T. E., Gardiner, D. T. & Tipton, A. R. 1996. Shrub emergence and seedling growth in microenvironments created by *Prosopis glandulosa*. Journal of Vegetation Science 7: 257-264.

Franks, S. J. 2003. Competitive and facilitative interactions within and between two species of coastal dune perennials. Canadian Journal of Botany 81: 330-337.

Gosling, P. 2005. Facilitation of *Urtica dioica* colonisation by *Lupinus arboreus* on a nutrient-poor mining soil. Plant Ecology 178: 141-148.

Hunt, R. 1982. Plant growth curves. The functional approach to plant growth analysis. Edward Arnold, London. Pp. 248.

Jefferies, R. A., Bradshaw, A. D. & Putwain, P. D. 1981. Growth, nitrogen accumulation and nitrogen transfer by legume species established on mine spoils. Journal of Applied Ecology 18: 945-956.

Kikvidze, Z., Pugnaire, F. I., Brooker, R. W., Choler, P., Lortie, C. J., Michalet, R. & Callaway, R. M. 2005. Linking patterns and processes in alpine plant communities: A global study. Ecology 86: 1395-1400.

Klanderud, K. 2005. Climate change effects on species interactions in an alpine plant community. Journal of Ecology 93: 127-137.

Klanderud, K. & Totland, Ø. 2005. The relative importance of neighbours and abiotic environmental conditions for population dynamic parameters of two alpine plant species. Journal of Ecology 93: 493-501.

Körner, C. 2003. Alpine plant life. Functional plant ecology of high mountain ecosystems. 2nd edition. Springer-Verlag, Berlin. Pp. 344.

Lee, T. D., Reich, P. B. & Tjoelker, M. G. 2003. Legume presence increases photosynthesis and N concentrations of co-occurring non-fixers, but does not modulate their responsiveness to carbon dioxide enrichment. Oecologia 137: 22-31.

Levine, J. M. 2000. Complex interactions in a streamside plant community. Ecology 81: 3431-3444.

Lid, J. & Lid, D. T. 2005. Norsk flora. 7. utgåva. Det Norske Samlaget, Oslo. Pp. 1230.

Maestre, F. T., Valladares, F. & Reynolds, J. F. 2005. Is the change of plant–plant interactions with abiotic stress predictable? A meta-analysis of field results in arid environments. Journal of Ecology 93: 748-757.

Maron, J. L. & Connors, P. G. 1996. A native nitrogen-fixing shrub facilitates weed invasion. Oecologia 105: 302-312.

Maron, J. L. & Jefferies, R. L. 1999. Bush lupine mortality, altered resource availability and alternative vegetation states. Ecology 80: 443-454.

Michalet, R., Brooker, R. W., Cavieres, L. A., Kikvidze, Z., Lortie, C. J., Pugnaire, F. I., Valiente-Banuet, A. & Callaway, R. M. 2006. Do biotic interactions shape both sides of the humped-back model of species richness in plant communities? Ecology Letters 9: 767-773.

Moen, A. 1998. Nasjonalatlas for Norge: Vegetasjon. Statens Kartverk, Hønefoss. Pp. 199.

Moen, J. 1993. Positive versus negative interactions in a high alpine block field: Germination of *Oxyria digyna* seeds in a *Ranunculus glacialis* community. Arctic and Alpine Research 25: 201-206.

Morris, W. F. & Wood, D. M. 1989. The role of lupine in succession on Mount St. Helens: Facilitation or inhibition? Ecology 70: 697-703.

Nadelhoffer, K. J., Giblin, A. E., Shaver, G. R. & Linkins, A. E. 1992. Microbial processes and plant nutrient availability in arctic soils. In: Arctic ecosystems in a changing climate: An ecophysiological perspective (eds. Chapin, F. S., III, Jefferies, R. L., Reynolds, J. F., Shaver, G. R. & Svoboda, J.), Academic Press, Inc., San Diego. Pp. 281-300.

Olofsson, J., Moen, J. & Oksanen, L. 1999. On the balance between positive and negative plant interactions in harsh environments. Oikos 86: 539-543.

Pugnaire, F. I., Haase, P. & Puigdefabregas, J. 1996. Facilitation between higher plant species in a semiarid environment. Ecology 77: 1420-1426.

Pugnaire, F. I. & Luque, M. T. 2001. Changes in plant interactions along a gradient of environmental stress. Oikos 93: 42-49.

Quinos, P. M., Insausti, P. & Soriano, A. 1998. Facilitative effect of *Lotus tenuis* on *Paspalum dilatatum* in a lowland grassland of Argentina. Oecologia 114: 427-431.

Rodríguez-Echeverría, S. & Pérez-Fernándes, M. A. 2003. Soil fertility and herb facilitation mediated by *Retama sphaerocarpa*. Journal of Vegetation Science 14: 807-814.

Sandvik, S. M. & Totland, Ø. 2000. Short-term effects of simulated environmental changes on phenology, reproduction, and growth in the late-flowering snowbed herb *Saxifraga stellaris* L.. Écoscience 7: 201-213.

Steven, J. C. & Waller, D. M. 2004. Reproductive alternatives to insect pollination in four species of *Thalictrum* (Ranunculaceae). Plant Species Biology 19: 73-80.

Temperton, V. M., Mwangi, P. N., Scherer-Lorenzen, M., Schmid, B. & Buchmann, N. 2007. Positive interactions between nitrogen-fixing legumes and four different neighbouring species in a biodiversity experiment. Oecologia 151: 190-205.

The Norwegian Meteorological Institute 2010. eKlima. Data from weather station 25840 at Finse, normal period 1961-1990. To be found on the web page http://eklima.met.no. Last visited 02.02.2010.

Thomas, B. D. & Bowman, W. D. 1998. Influence of N_2-fixing *Trifolium* on plant species composition and biomass production in alpine tundra. Oecologia 115: 26-31.

Wang, Y., Chu, C., Maestre, F. T. & Wang, G. 2008. On the relevance of facilitation in alpine meadow communities: An experimental assessment with multiple species differing in their ecological optimum. Acta Oecologica 33: 108-113.

APPENDIX 1: Leaf density of *Thalictrum alpinum* within 15 × 15 cm sampling quadrats and number of leaves, mean leaf length (mm), mean leaf width (mm) and fertility (0 = infertile, 1 = fertile) of individual *T. alpinum* plants measured in July ($_1$) and August ($_2$) growing with (+) and without (-) the legumes *Astragalus alpinus* and *Oxytropis lapponica* in the low (L), mid (M) and high (H)-elevation study sites at Mt. Sanddalsnuten, Finse, Norway, summer 2008. The table also shows legume leaf density and the number of leaves of individual legume plants measured in July ($_1$) and August ($_2$).

			Thalictrum alpinum								legumes		
site	*A. alp*	*O. lap*	leaf density	no. leaves$_1$	length$_1$	width$_1$	no. leaves$_2$	length$_2$	width$_2$	fertility	leaf density	no. leaves$_1$	no. leaves$_2$
L	+		61	4	20.3	7.0	3	24.2	7.8	1	8	2	3
L	-		52	3	22.2	8.3	5	16.6	4.7	1			
L	+		41	4	23.4	10.9	4	21.8	10.8	0	52	22	21
L	-		116	3	23.6	9.2	2	23.8	9.4	0			
L	+		89	3	30.6	10.5	3	38.8	7.9	0	104	95	76
L	-		72	3	39.5	12.8	3	36.0	12.6	0			
L	+		113	2	40.8	18.3	2	41.4	20.2	0	27	15	23
L	-		68	3	17.6	6.1	5	26.7	8.9	1			
L	+		28	5	28.9	9.6	4	27.9	8.2	1	17	11	11
L	-		54	5	14.8	6.4	4	16.9	5.9	1			
L	+		21	2	40.5	12.8	2	36.3	11.4	0	30	25	17
L	-		34	3	23.9	6.5	2	40.5	12.8	0			
L	+		18	3	39.5	15.9	3	35.8	17.3	0	19	15	12
L	-		13	3	37.9	10.7	3	32.2	10.5	1			
L	+		66	6	19.4	11.2	8	18.7	7.9	1	27	12	15
L	-		30	4	26.2	8.3	4	24.9	6.7	1			
L	+		37	4	24.8	9.4	4	23.3	10.3	1	37	10	14
L	-		63	5	18.8	8.5	4	18.2	5.9	1			
L	+		26	2	24.0	12.0	2	20.7	13.5	1	105	103	110
L	-		49	5	15.7	7.7	3	14.9	7.9	1			
L	+		10	5	12.5	6.8	5	11.4	6.6	1	34	17	15
L	-		27	4	12.1	7.2	4	15.0	6.9	1			
L	+		58	5	16.2	7.4	4	17.0	8.5	1	19	14	12
L	-		27	4	20.2	8.1	4	20.7	8.8	1			
L	+		26	7	21.5	10.9	7	22.8	10.7	1	27	16	8
L	-		58	4	19.3	9.5	4	26.5	10.0	0			
L	+		23	2	21.7	10.4	2	24.0	9.5	0	40	40	23
L	-		75	7	14.0	7.7	7	15.0	8.1	1			
L	+		48	4	34.5	11.9	8	27.0	8.8	1	29	7	19
L	-		86	5	21.8	8.0	4	20.5	8.0	1			
L	+		45	6	10.7	6.2	7	10.0	6.0	1	16	11	4
L	-		38	7	8.9	5.0	7	9.3	5.1	1			

L	+		30	3	31.2	5.8	2	29.1	8.3	0	9	9	8
L	-		26	3	21.2	6.3	3	18.9	6.6	0			
L	+		34	3	34.6	5.8	1	29.4	7.0	0	5	5	4
L	-		37	5	10.5	6.4	4	9.5	5.1	1			
L	+		52	5	26.3	9.2	5	25.8	9.0	1	30	7	8
L	-		55	8	24.0	9.8	7	22.7	7.1	1			
L	+		49	5	27.1	11.3	6	28.4	10.1	1	16	5	2
L	-		61	7	14.1	8.1	7	15.3	5.2	1			
L	+		74	5	25.8	10.9	5	23.8	11.2	0	25	7	8
L	-		83	6	14.2	7.7	5	14.8	6.6	1			
L	+		56	6	14.3	7.9	6	13.3	7.9	1	26	17	20
L	-		22	2	30.6	14.8	2	31.5	14.7	0			
L	+		13	4	16.9	6.4	3	15.8	5.6	1	20	17	21
L	-		28	2	12.9	7.1	2	13.5	6.3	0			
L	+		18	2	50.1	15.5	2	40.4	13.6	0	59	18	15
L	-		24	3	22.4	11.7	2	28.7	10.2	0			
L	+		68	5	28.8	9.4	3	37.7	12.8	0	34	26	27
L	-		96	3	24.1	9.4	7	23.0	9.2	1			
L	+		24	3	29.1	11.1	3	29.9	11.3	0	30	13	14
L	-		109	7	20.4	8.4	9	16.9	6.2	1			
L	+		16	3	19.2	10.2	3	22.4	11.3	0	12	12	13
L	-		4	3	25.5	12.7	4	27.4	13.1	0			
L	+		48	3	26.5	12.4	2	26.4	9.1	0	37	13	15
L	-		61	3	25.0	11.1	3	24.1	7.8	0			
L	+		65	2	36.1	8.3	1	27.3	6.2	1	35	24	24
L	-		52	4	12.9	9.5	2	25.7	12.0	0			
L	+		32	2	14.2	7.2	2	14.5	7.9	0	34	21	11
L	-		25	3	18.5	8.9	3	17.7	8.3	0			
M		+	11	2	26.7	7.6	2	21.4	7.6	0	26	26	27
M		-	38	4	22.1	12.3	4	23.5	11.4	0			
M		+	78	4	15.4	6.4	5	12.6	6.7	0	15	8	7
M		-	16	2	24.5	11.8	2	31.6	12.3	0			
M		+	5	1	32.0	8.6	1	34.9	10.7	0	23	23	23
M		-	19	4	15.0	7.3	3	18.3	8.6	0			
M		+	23	1	20.2	9.7	3	29.7	14.1	0	24	13	20
M		-	12	3	30.1	11.2	3	38.9	10.9	0			
M		+	14	5	9.7	6.5	6	9.1	3.9	1	15	15	18
M		-	41	10	13.8	7.5	9	14.0	7.0	1			
M		+	48	8	17.1	6.3	6	11.6	4.0	1	26	4	4
M		-	35	3	22.4	8.5	3	19.9	7.5	1			
M		+	27	3	18.8	7.4	3	20.9	7.4	0	23	19	22
M		-	45	2	9.8	7.2	2	19.8	6.6	0			
M		+	46	4	9.3	4.8	3	9.1	4.4	0	24	15	11

M	-	6	3	19.7	7.3	3	22.1	7.2	0			
M	+	42	5	10.6	6.4	6	9.9	6.0	1	38	14	13
M	-	16	2	12.9	4.9	2	30.3	4.6	0			
M	+	54	4	11.6	6.9	2	8.0	5.6	0	17	17	10
M	-	18	5	18.3	7.9	2	18.7	8.7	0			
M	+	38	2	23.7	7.5	1	23.7	8.7	0	19	17	16
M	-	70	5	13.5	6.1	3	14.8	5.8	0			
M	+	58	2	37.8	9.3	2	27.6	6.4	0	22	22	21
M	-	48	5	9.3	5.6	2	5.3	5.4	0			
M	+	65	2	12.9	6.7	2	16.1	8.9	0	5	3	2
M	-	37	3	15.6	9.4	1	23.7	13.0	0			
M	+	69	7	7.3	4.9	5	5.4	3.0	1	23	12	5
M	-	16	9	20.5	9.2	7	21.8	7.2	1			
M	+	36	2	25.5	11.3	2	25.9	11.9	0	13	13	13
M	-	13	3	18.6	11.5	3	25.4	11.6	0			
M	+	88	8	12.7	6.3	6	8.4	3.7	1	38	17	26
M	-	41	3	13.3	6.1	2	19.3	7.0	1			
M	+	39	5	22.0	9.3	5	20.9	8.1	1	34	13	20
M	-	17	8	20.7	9.7	7	21.6	9.7	1			
M	+	31	1	18.2	12.3	2	21.7	9.0	1	10	8	10
M	-	21	6	16.5	8.2	6	11.5	4.7	1			
M	+	30	3	26.2	7.6	3	26.8	8.3	0	24	24	23
M	-	32	3	41.0	10.9	3	30.9	11.1	0			
M	+	92	6	13.5	6.5	7	10.8	5.5	1	7	3	2
M	-	25	4	37.5	12.8	3	37.0	12.2	0			
M	+	36	3	28.6	11.8	4	21.4	10.4	0	23	21	25
M	-	10	2	9.5	5.5	2	8.1	5.2	1			
M	+	51	3	22.4	7.3	3	26.2	8.0	0	34	21	24
M	-	26	2	24.2	11.3	2	24.6	7.0	0			
M	+	11	3	23.6	8.2	3	25.1	7.9	0	20	20	25
M	-	37	1	13.4	8.7	1	18.5	9.5	0			
M	+	21	3	24.4	6.9	2	26.8	8.1	0	10	10	13
M	-	13	3	14.8	6.6	3	14.1	6.2	0			
M	+	27	2	20.3	7.3	1	15.1	6.3	0	51	23	23
M	-	6	4	23.3	12.1	6	22.9	8.4	0			
M	+	50	2	29.7	7.3	1	36.9	8.7	0	47	24	10
M	-	8	2	22.6	12.1	2	23.3	11.8	0			
M	+	27	4	20.4	7.4	3	19.2	7.4	0	33	8	12
M	-	18	4	11.6	5.4	5	8.0	4.2	0			
M	+	24	2	36.8	13.5	2	32.9	9.1	0	45	36	27
M	-	5	4	8.4	8.7	4	12.1	7.8	0			
M	+	7	3	25.4	9.2	3	27.6	7.9	0	13	11	11
M	-	8	2	24.6	8.2	2	18.2	5.4	0			

M	+	132	2	14.6	5.9	2	17.0	5.3	0	17	17	14
M	-	98	5	8.1	4.8	2	9.4	5.0	1			
H	+	3	2	16.1	5.9	2	16.1	5.3	0	93	93	84
H	-	16	3	15.4	6.9	3	15.0	6.2	0			
H	+	10	2	27.7	5.7	1	18.7	1.8	0	34	12	15
H	-	12	9	14.0	6.5	6	12.1	5.7	1			
H	+	13	2	28.0	10.3	5	24.1	10.5	1	90	90	63
H	-	11	4	32.9	13.4	4	26.7	10.9	0			
H	+	5	2	24.7	9.2	2	34.2	7.8	0	169	55	82
H	-	5	3	12.2	6.9	3	13.4	7.5	0			
H	+	8	3	33.2	9.4	3	32.7	10.1	0	244	241	131
H	-	11	3	11.6	6.5	3	15.1	6.2	0			
H	+	120	4	10.5	6.9	3	11.6	7.5	1	102	95	48
H	-	3	3	15.2	8.8	2	16.5	11.1	1			
H	+	47	3	16.9	7.4	3	19.3	7.9	1	86	86	51
H	-	32	2	41.8	12.8	2	30.5	10.7	1			
H	+	22	3	29.4	7.7	2	25.1	8.0	0	173	173	103
H	-	33	4	14.2	5.7	4	9.9	4.2	0			
H	+	35	4	21.3	8.3	4	22.3	8.6	1	149	59	34
H	-	14	4	8.5	5.4	3	8.1	4.7	1			
H	+	31	2	20.0	8.9	2	18.0	8.5	1	75	74	66
H	-	30	3	8.9	5.6	3	10.0	4.5	1			
H	+	20	4	20.8	6.3	4	21.6	6.8	0	117	61	33
H	-	13	4	16.9	7.9	4	19.4	9.2	0			
H	+	15	4	28.5	7.4	3	27.9	7.9	0	75	69	58
H	-	16	3	13.9	5.7	4	10.5	6.1	1			
H	+	19	3	11.9	7.0	3	10.2	7.2	1	62	50	37
H	-	41	5	16.4	6.2	0	0.0	0.0	1			
H	+	107	4	11.7	8.3	3	13.9	9.3	0	52	37	20
H	-	46	2	18.4	7.2	1	11.6	5.2	1			
H	+	75	7	5.6	3.8	7	5.1	4.0	1	64	29	18
H	-	18	9	13.3	6.0	5	8.7	4.7	1			
H	+	28	4	19.1	8.0	4	22.4	8.7	0	72	53	39
H	-	13	3	11.8	6.1	2	5.6	4.1	0			
H	+	57	3	23.9	9.1	3	15.6	6.9	0	72	17	15
H	-	13	2	14.4	9.3	2	17.8	10.1	0			
H	+	21	2	8.4	3.4	2	11.7	4.2	0	33	31	25
H	-	15	2	18.8	8.5	2	21.5	8.7	0			
H	+	54	2	23.5	9.3	2	19.7	8.7	0	32	32	33
H	-	7	3	19.7	7.2	2	17.2	7.1	1			
H	+	29	2	23.1	7.6	3	20.0	4.8	0	44	44	50
H	-	23	3	22.2	7.9	2	25.9	9.1	0			
H	+	35	3	20.1	5.5	3	21.0	5.0	0	114	52	76

H		-	37	2	9.1	7.5	2	10.1	7.3	0			
H		+	9	1	21.7	7.4	3	16.8	7.8	0	42	42	39
H		-	4	4	25.0	14.6	4	21.5	14.8	0			
H		+	37	4	12.4	6.3	4	13.8	7.8	1	20	13	10
H		-	25	4	14.4	7.9	3	18.0	11.1	0			
H		+	18	3	24.6	8.5	3	18.9	9.4	1	65	35	34
H		-	6	3	15.4	9.2	2	21.6	10.8	0			
H		+	5	5	13.6	6.8	6	15.0	7.6	0	30	25	7
H		-	20	3	8.4	5.0	3	7.7	6.0	0			
H		+	33	5	7.2	5.0	3	11.5	7.4	0	126	118	86
H		-	37	4	12.2	6.5	4	8.5	4.9	0			
H		+	13	5	10.7	6.0	3	10.8	7.4	1	65	51	40
H		-	15	3	11.8	5.8	3	15.9	5.7	0			
H		+	48	2	6.3	4.5	2	9.3	5.0	0	89	28	25
H		-	17	3	19.4	10.1	2	25.3	10.5	0			
H		+	41	4	7.5	3.3	4	7.5	3.8	0	92	34	32
H		-	16	3	10.1	5.4	4	6.5	3.8	0			
H		+	18	4	14.2	8.1	4	13.7	8.6	1	34	34	24
H		-	8	3	12.3	5.3	3	19.1	7.4	0			
H	+		20	6	17.0	7.6	4	16.2	8.3	1	29	11	12
H	-		7	3	7.6	5.7	4	6.0	4.5	0			
H	+		7	3	25.2	13.1	3	24.1	10.3	0	38	15	29
H	-		3	2	25.9	12.7	2	32.6	12.5	0			
H	+		24	2	13.5	7.3	2	22.7	7.6	0	56	31	23
H	-		25	7	8.0	5.5	6	6.3	4.1	0			
H	+		11	1	26.1	12.2	1	35.8	11.7	0	21	16	15
H	-		10	3	23.7	13.9	1	29.0	16.5	0			
H	+		45	4	11.5	7.2	4	11.6	7.4	1	58	29	29
H	-		32	7	13.0	7.9	8	12.3	7.7	1			
H	+		5	2	32.7	12.3	2	29.4	11.7	0	54	37	46
H	-		5	1	21.0	11.3	2	16.0	7.0	0			
H	+		4	2	20.3	9.4	2	24.7	9.5	0	35	12	10
H	-		35	2	13.7	7.7	1	13.9	7.0	0			
H	+		30	3	13.4	6.2	1	15.7	9.2	0	33	15	16
H	-		11	1	14.7	12.7	1	26.2	14.3	0			
H	+		48	2	22.4	7.7	2	17.7	6.5	0	53	12	20
H	-		4	2	14.7	6.9	1	21.0	5.9	0			
H	+		12	1	22.0	12.4	2	23.6	12.3	0	34	7	9
H	-		31	2	17.7	8.9	2	19.1	7.6	0			
H	+		9	2	22.3	5.9	2	22.4	5.4	0	25	10	12
H	-		8	3	14.2	11.1	3	15.9	9.6	1			
H	+		23	2	17.5	9.3	2	15.0	7.1	1	37	12	18
H	-		11	5	13.9	7.7	5	13.1	7.5	1			

H	+	47	3	22.2	13.6	3	25.3	13.5	1	8	4	7
H	-	25	2	16.6	10.5	3	12.2	6.8	1			
H	+	17	2	30.5	8.7	2	26.9	8.4	0	68	19	17
H	-	14	6	11.6	6.9	5	11.8	6.6	1			
H	+	42	2	16.6	6.7	1	11.7	7.0	0	35	31	25
H	-	24	3	24.0	8.8	3	16.5	8.7	0			
H	+	9	4	28.4	10.7	3	53.7	12.9	1	10	7	7
H	-	16	5	15.2	7.8	5	14.0	7.8	1			
H	+	75	4	19.4	8.9	4	22.4	8.8	0	43	3	14
H	-	6	3	14.3	10.5	4	20.8	10.5	0			
H	+	8	2	19.6	7.4	2	18.1	7.4	0	74	9	10
H	-	3	3	13.6	7.8	2	21.2	10.0	0			
H	+	15	5	24.3	9.4	5	22.5	9.5	0	39	12	30
H	-	30	4	13.8	7.4	6	17.9	7.2	1			
H	+	49	2	17.3	9.7	2	17.6	7.6	0	38	2	2
H	-	36	4	10.0	4.6	5	13.5	5.7	0			
H	+	37	5	9.5	7.0	4	9.6	6.6	0	30	23	22
H	-	37	4	12.6	8.6	4	14.9	9.7	0			
H	+	20	3	16.4	10.3	3	17.1	9.4	0	37	27	23
H	-	22	2	9.9	5.4	2	10.0	5.6	0			
H	+	42	2	32.2	8.2	1	30.3	5.8	0	37	28	24
H	-	7	1	21.3	11.6	1	26.4	10.0	0			
H	+	22	2	8.7	4.2	1	6.5	3.0	0	35	29	19
H	-	28	2	20.9	10.6	1	24.1	10.5	0			
H	+	16	4	8.6	4.3	4	8.3	4.1	0	190	117	96
H	-	17	2	30.5	8.8	2	17.7	8.8	0			
H	+	5	1	31.2	8.9	1	20.4	9.7	0	28	5	6
H	-	12	6	15.2	6.8	6	13.8	6.4	1			
H	+	4	3	31.6	11.0	3	30.2	9.6	0	62	12	35
H	-	2	2	46.7	12.8	2	49.3	12.5	0			
H	+	19	2	18.6	5.8	1	16.0	7.9	0	37	26	28
H	-	23	1	32.8	9.6	1	23.1	10.0	0			
H	+	108	4	19.0	6.2	2	18.7	5.5	0	16	3	2
H	-	19	2	16.6	8.1	2	15.9	8.3	0			
H	+	15	2	13.1	5.0	2	12.7	4.0	0	5	5	3
H	-	8	2	29.8	9.7	2	27.9	9.6	0			

APPENDIX 2. CHN-N content (g/kg) of *Thalictrum alpinum* growing with (+) and without (-) the legumes *Astragalus alpinus* and *Oxytropis lapponica* in the low (L), mid (M) and high (H)-elevation study sites at Mt. Sanddalsnuten, Finse, Norway, summer 2008.

Site	*A. alp*	*O. lap*	CHN-N
L	+		21.1
L	+		15.9
L	+		17.3
L	+		18.0
L	+		15.7
L	+		18.1
L	+		17.4
L	+		17.6
L	+		18.0
L	+		18.8
L	-		16.4
L	-		19.5
L	-		16.1
L	-		14.8
L	-		18.4
L	-		15.2
L	-		16.7
L	-		14.5
L	-		17.3
L	-		17.6
M		+	15.4
M		+	14.4
M		+	13.8
M		+	14.7
M		+	13.0
M		+	16.7
M		+	15.7
M		+	12.7
M		+	14.0
M		+	12.9
M		-	14.8
M		-	15.8
M		-	14.3
M		-	15.9
M		-	13.8
M		-	16.9
M		-	16.8
M		-	15.3
M		-	15.5
M		-	15.7
H		+	18.9
H		+	17.7
H		+	20.8
H		+	15.7
H		+	18.4
H		+	15.4
H		+	15.3
H		+	16.6
H		+	18.5
H		+	17.2
H		-	20.1
H		-	16.8
H		-	13.4
H		-	14.1
H		-	16.9
H		-	15.9
H		-	15.4
H		-	17.6
H		-	18.9
H		-	14.1
H	+		17.4
H	+		16.4
H	+		15.7
H	+		15.2
H	+		15.6
H	+		14.2
H	+		15.5
H	+		12.3
H	+		19.3
H	+		13.2
H	-		19.0
H	-		17.2
H	-		17.7
H	-		17.2
H	-		13.1
H	-		17.8
H	-		21.9
H	-		11.9
H	-		16.8
H	-		16.5

PART 3. NITROGEN-FIXING LEGUMES DIFFERENTIALLY AFFECT SPECIES COMPOSITION, RICHNESS AND EVENNESS, BUT NOT DIVERSITY, OF AN ALPINE PLANT COMMUNITY

ABSTRACT

Facilitation may increase species richness in plant communities, particularly in habitats with high levels of abiotic stress, such as alpine areas. In these low-productivity habitats legumes are potential facilitators, as they increase soil nitrogen (N) levels through biological N-fixation. In this study I examined how species composition, richness, diversity and evenness differed with and without two alpine legumes, Astragalus alpinus *L. and* Oxytropis lapponica *(Wahlenb.) Gay, in two sites situated along a gradient in abiotic stress at Finse, Norway. I found that species composition differed and species richness increased with* O. lapponica, *most likely due to increased N availability below the legumes, while there was no difference in community properties with and without* A. alpinus. *The facilitative effect of* O. lapponica *was found only in the site of highest abiotic stress, supporting the theory that facilitation can increase species richness under stressful environmental conditions. Graminoid species richness and cover also increased with* O. lapponica, *but the increase in graminoids was not great enough to exclude herb species, resulting in a total increase in species richness. My study shows that* O. lapponica, *but not* A. alpinus, *may contribute to increasing the biodiversity of the alpine ecosystem at Finse.*

INTRODUCTION

Facilitative interactions between plants of different species can cascade into community-wide effects. Hacker & Gaines (1997), Michalet et al. (2006) and Gross (2008) suggest that positive interactions may produce more species-rich communities by facilitating species that might not normally survive and that the importance of facilitation, relative to competition, increases with increasing abiotic stress. This has been confirmed in studies of cushion nurse plants in northern Sweden (Antonsson et al. 2009) and the high-alpine Andes (Cavieres et al. 2002, Arroyo et al. 2003, Cavieres & Badano 2009), shrubs in a Mediterranean shrubland (Holzapfel et al. 2006), the nurse tree *Olneya tesota* in the Sonoran desert (Tewksbury & Lloyd 2001), salt marsh vegetation in New England (Brewer et al. 1997, Hacker & Bertness 1999) and subalpine and alpine meadow ecosystems throughout the northern hemisphere

(Kikvidze et al. 2005). However, Michalet et al. (2006) point out that under *extreme* conditions the positive effect of facilitation on species richness declines, as shown by Cavieres & Badano (2009) and Xiao et al. (2009).

Nitrogen (N)-fixing legumes have the potential to affect plant community diversity, structure and composition by fertilizing the soil in their immediate vicinity (Jacot et al. 2005). Several studies show how the presence of legumes affect the course of succession and the properties of the resulting plant community (Halvorson et al. 1991, Bellingham et al. 2001, del Moral & Rozzell 2005, Li et al. 2010), others how legumes facilitate alien plant species, thereby indirectly changing the native community composition (Maron & Connors 1996, Maron & Jefferies 1999, Carino & Daehler 2002). A few studies also examine how legumes influence the distribution of plants in non-successional native plant communities, with results ranging from positive (Perroni-Ventura et al. 2006) and neutral (Thomas & Bowman 1998, Rossi & Villagra 2003) to negative (Rodríguez-Echeverría & Pérez-Fernándes 2003) effects on plant species richness, diversity and evenness.

Kammer & Möhl (2002) conclude that “species richness in most of the alpine plant communities is stress limited”. This implies that a reduction in stress, for instance due to facilitation by legumes, may increase species richness in alpine areas, which is consistent with the theory of Hacker & Gaines (1997). To my knowledge, only two previous studies have examined how the presence of legumes affects plant community structure in alpine areas: Thomas & Bowman (1998) found that the presence of the legume *Trifolium dasyphyllum* affected the abundance of plant species in the Rocky Mountains, although species richness did not differ between legume and non-legume plots, and Jacot et al. (2005) found that certain species were associated with legumes in the Alps. Clearly, our knowledge of the effect of alpine legumes on community structure is limited.

Fertilization experiments reveal that different plant functional groups respond differently to nutrient addition, with an increase in graminoids as the most common response (e.g. Tilman 1984). However, Rossi & Villagra (2003) found a decrease in graminoids below the canopy of the leguminous tree *Prosopis flexuosa* in a desert habitat in Argentina. To my knowledge, no study has examined the effects of alpine legumes on different plant functional groups.

In this study I examine how species composition, richness, diversity and evenness differ with and without the presence of two alpine legumes, *Astragalus alpinus* L. and *Oxytropis lapponica* (Wahlenb.) Gay, in two sites situated along a gradient in abiotic stress at Finse, Norway. In Part 1 I found increased soil N levels below *O. lapponica* at Finse, and in Part 2 I showed that both *A. alpinus* and *O. lapponica* facilitate the density of a single target species, indicating that the legumes may have the potential to affect plant communities in the study area. I also found that the increase in N, as well as the facilitative effect, was greatest in the most stressful end of the gradient, so if legumes do facilitate species richness at Finse, the effect should increase with increasing abiotic stress, as hypothesized by Hacker & Gaines (1997). Moreover, I examine whether different functional groups respond differently to N addition by legumes.

Specifically I ask the following questions:

- Do species composition, species richness, species diversity or species evenness differ with and without the presence of legumes?
- Does the difference in species composition, richness, diversity or evenness with and without legumes increase with increasing abiotic stress?
- Do different functional groups respond differently to legume facilitation?

MATERIALS AND METHODS

Study area and study species

Field work was conducted during the summer 2009 at Finse, southwest Norway (60°36’59’’N, 07°31’23’’E). Finse has a slightly oceanic climate (Moen 1998) with a mean monthly temperature and rainfall during the growing season (May-September) of 4.4 °C and 89 mm, respectively (The Norwegian Meteorological Institute 2010). The study area was located in the middle alpine zone on the southwest-facing slope of Mt. Sanddalsnuten (1554 m.a.s.l.). The bedrock consists of phyllite with strains of marble (Askvik 2008), and *Dryas* heath, dominated by the dwarf-shrub *Dryas octopetala,* is the main vegetation type in the study area. Other common vascular plant species are *Astragalus alpinus, Bartsia alpina, Bistorta vivipara, Carex rupestris, Carex vaginata, Festuca ovina, Festuca vivipara, Oxytropis lapponica, Potentilla crantzii, Salix herbacea, Salix reticulata, Saussurea alpina, Silene acaulis* and *Thalictrum alpinum*. Nomenclature follows Lid & Lid (2005).

The study was conducted in three sites (10 × 10 m) situated along the slope of Mt. Sanddalsnuten: a low-elevation (1460 m.a.s.l.), mid-elevation (1510 m.a.s.l.) and high-elevation site (1554 m.a.s.l.), with a difference in altitude of about 50 m between each site. The low and mid-elevation sites were fenced to prevent sheep grazing. The study species *Astragalus alpinus* was present in the low and high-elevation sites, while *Oxytropis lapponica* was present in the mid and high-elevation sites.

To document differences in abiotic conditions along the slope of Mt. Sanddalsnuten, I measured temperature, soil moisture and collected soil samples in each of the three sites. I also estimated total vegetation cover. Methods for soil sampling and analyses are described in Part 1, while other measurements are described in Part 2. The low-elevation site was warmer, moister and had higher soil nutrient contents and vegetation cover than the two other sites (Part 2). Between the mid and high-elevation sites I found only minor differences. However, the difference in soil nutrient levels below and outside the legume *O. lapponica* was greater in the high-elevation site than the mid-elevation site (Part 1).

Study design and statistical analyses

I used the same 15 × 15 cm sampling quadrats as described in Part 2 to examine whether species composition, richness, diversity or evenness differ with and without the presence of legumes. In each quadrat (240 all together) I identified all vascular plant species and estimated their cover (%) (Appendix 1). I then used the Excel add-in module Diversity to calculate species richness, species diversity (the Shannon-Weaver diversity index: $H' = -\Sigma\ p_i\ (\ln p_i)$ where p_i is the relative abundance of each species; Ludwig & Reynolds 1988) and species evenness (Shannon's evenness index: $J' = H' / \ln(S)$ where S is the number of species; Ludwig & Reynolds 1988) in each quadrat. In all calculations I removed the legume data from the dataset, as they would automatically bias the results in the legume quadrats (see del Moral & Rozzell 2005).

I used multivariate ordination analysis to test whether species composition differed between quadrats with and without legumes. Preliminary analyses using species cover (%) proved inconclusive, and in the final analyses I categorized each species as present or absent.
First I used detrended correspondence analysis (DCA) with default settings to examine the gradient lengths for the species composition data. The longest gradients were between 1.7 and 2.5 SD units. Lepš & Šmilauer (2003) conclude that redundancy analysis (RDA) is

appropriate when the longest gradient is shorter than 3.0 SD units, so I used RDA to test for differences in species composition. Separate analyses were conducted for each site, as I expected to find differences between sites due to differences in abiotic stress. I used *legume presence* as an environmental variable and *pair* as a covariable in Monte Carlo permutation tests with 999 permutations where the covariable defined the blocks within which to permute. Otherwise default settings were used. Analyses were performed using CANOCO 4.5 for Windows. In all analyses I removed the legume data from the dataset, as they would automatically bias the results in the legume quadrats (see del Moral & Rozzell 2005).

I used analysis of variance (ANOVA) to test whether species richness, diversity or evenness differed between quadrats with and without legumes and/or between sites. I also tested whether species richness and total cover (%) of graminoids and herbs differed with legume presence or between sites. Woody species and pteridophytes were not frequent enough to be included in the analyses. All tests were two-way ANOVAs. In initial analyses I used *site* and *legume presence* as fixed main factors and *pair* as a random nested factor under *site*. *Pair* was, however, not significant and was therefore removed in order to save degrees of freedom. If the interaction between *site* and *legume presence* was significant, I used paired t-tests to compare the different parameters with and without legumes within each site. Data on evenness was arcsin transformed and total cover of graminoids and herbs was square root transformed to fulfill the ANOVA assumptions of normality and equal variances. All figures are shown with untransformed data. Analyses were performed using the General Linear Model and Paired t-test procedures in Minitab 15.1.1.0 for Windows.

In all tests data for the two legume species was analyzed separately, as analyses in Part 1 and 2 suggest that they differ in their facilitative ability.

RESULTS

Astragalus alpinus

The RDA showed no difference in species composition in quadrats with and without *Astragalus alpinus* in neither the low-elevation site (F = 0.76, p = 0.81, % variance in species data explained by the 1st axis = 2.7) nor the high-elevation site (F = 1.28, p = 0.22, % variance in species data explained by the 1st axis = 4.2), and no species seemed to be strongly associated with *A. alpinus* (Appendix 2).

The ANOVA showed no difference in species richness (Fig. 1A), diversity (Fig. 1B) or evenness (Fig. 1C) between quadrats with and without *A. alpinus* (Table 1). The low-elevation site had a significantly higher species richness and diversity than the high-elevation site, while species evenness was the same in both sites.

The ANOVA showed no difference in graminoid or herb species richness (Fig. 2A) between quadrats with and without *A. alpinus*, while richness of both functional groups was higher in the low-elevation site compared to the high-elevation site (Table 2). The total cover of graminoids and herbs (Fig. 2B) followed the same patterns.

Oxytropis lapponica

The RDA showed that species composition differed in quadrats with and without *Oxytropis lapponica* in the high-elevation site (F = 2.19, p = 0.01, % variance in species data explained by the 1st axis = 7.0), but not the mid-elevation site (F = 0.85, p = 0.63, % variance in species data explained by the 1st axis = 2.9). The species most commonly associated with *O. lapponica* were *Carex rupestris*, *Festuca vivipara*, *Salix herbacea*, *Silene acaulis* and *Thalictrum alpinum* (Appendix 2).

The ANOVA showed no difference in species diversity (Fig. 1B) in quadrats with and without *O. lapponica* or between sites (Table 1). Evenness (Fig. 1C) was significantly lower in quadrats with legumes, but there was no difference between sites. For species richness (Fig. 1A) there was a significant interaction between *site* and *legume presence*. The t-tests revealed that in the high-elevation site species richness was significantly higher in quadrats where *O. lapponica* was present (T = 3.56, p = 0.001), while in the mid-elevation site there was no difference in species richness in quadrats with and without *O. lapponica* (T = 0.62, p = 0.54).

The ANOVA showed that graminoid species richness (Fig. 2A) was significantly higher where *O. lapponica* was present and greater in the mid-elevation site than the high-elevation site (Table 2). There was no difference in herb species richness between quadrats with and without *O. lapponica* or between sites. The total cover of both graminoids and herbs (Fig. 2B) was significantly higher in quadrats with *O. lapponica*, but the percent increase was always greater for graminoids (54.6 % vs. 38.9 % in the mid-elevation site and 158.4 % vs. 110.0 % in the high-elevation site). Graminoid cover was higher in the mid than the high-elevation site, while herb cover was the same in both sites.

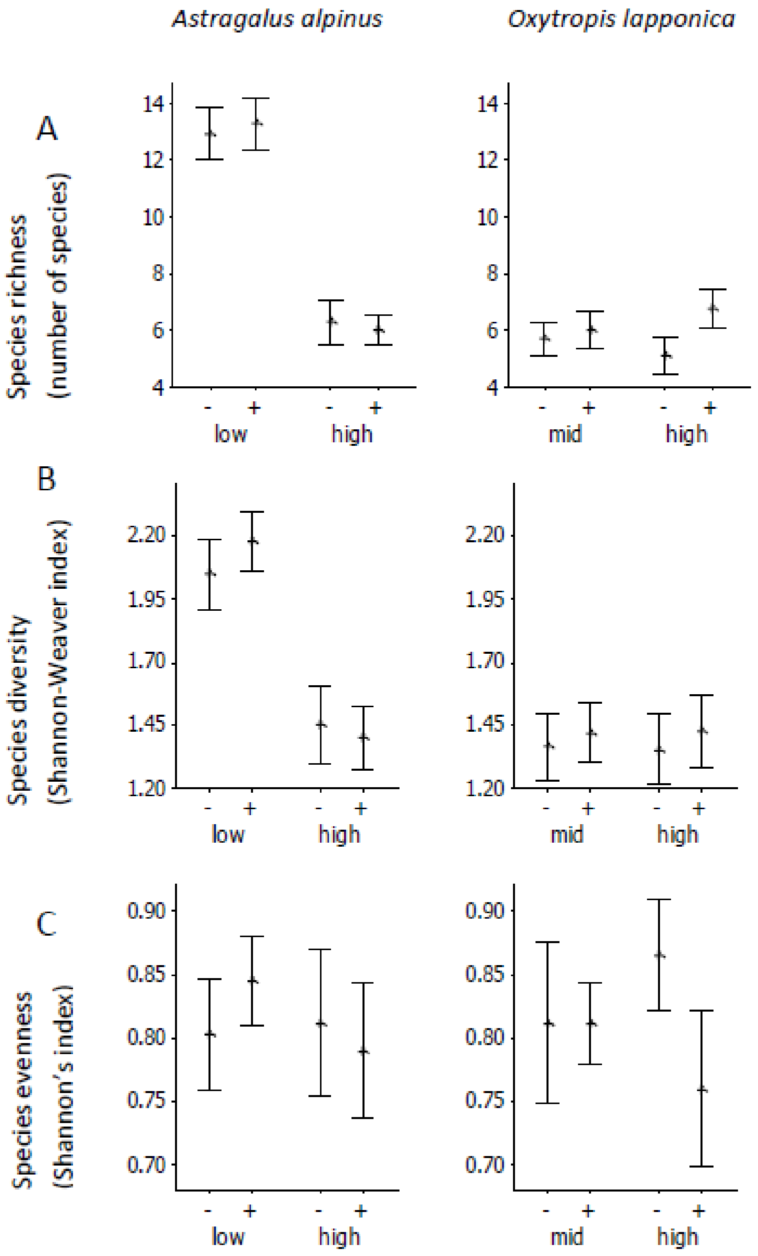


Fig. 1. Species richness (A), diversity (B) and evenness (C) in 15 × 15 cm sampling quadrats with (+) and without (-) the legumes Astragalus alpinus *(left) and* Oxytropis lapponica *(right) in the low, mid and high-elevation study sites at Mt. Sanddalsnuten, Finse, Norway, summer 2009. All figures show mean values with 95 % CI.*

Table 1. $F_{d.f.}$ *and p-values for ANOVA analyses of species richness (number of species), diversity (the Shannon-Weaver diversity index) and evenness (Shannon's evenness index) in 15 × 15 cm sampling quadrats with and without the legumes* Astragalus alpinus *and* Oxytropis lapponica *in three different study sites at Mt. Sanddalsnuten, Finse, Norway, summer 2009.* **Bold** *letters indicate p-values significant at the 0.05 level.*

Source of variation	**Richness**		**Diversity**		**Evenness**	
	$F_{d.f.}$	P	$F_{d.f.}$	P	$F_{d.f.}$	P
A. alpinus						
site	$307.20_{1,115}$	**<0.001**	$108.62_{1,115}$	**<0.001**	$0.37_{1,115}$	0.542
A. alp	$0.02_{1,115}$	0.899	$0.29_{1,115}$	0.592	$0.25_{1,115}$	0.621
site × A. alp	$0.63_{1,115}$	0.427	$1.92_{1,115}$	0.169	$2.08_{1,115}$	0.152
O. lapponica						
site	$0.04_{1,116}$	0.841	$0.00_{1,116}$	0.985	$0.25_{1,116}$	0.618
O. lap	$8.52_{1,116}$	**0.004**	$1.04_{1,116}$	0.309	$6.93_{1,116}$	**0.010**
site × O. lap	$4.05_{1,116}$	**0.046**	$0.02_{1,116}$	0.880	$3.34_{1,116}$	0.070

Table 2. $F_{d.f.}$ *and p-values for ANOVA analyses of species richness (number of species) and total cover (%) of graminoids and herbs in 15 × 15 cm sampling quadrats with and without the legumes* Astragalus alpinus *and* Oxytropis lapponica *in three different study sites at Mt. Sanddalsnuten, Finse, Norway, summer 2009.* **Bold** *letters indicate p-values significant at the 0.05 level.*

Source of variation	**Graminoids**				**Herbs**			
	Species richness		Total cover (%)		Species richness		Total cover (%)	
	$F_{d.f.}$	P	$F_{d.f.}$	P	$F_{d.f.}$	P	$F_{d.f.}$	P
A. alpinus								
site	$233.96_{1,115}$	**<0.001**	$74.25_{1,58}$	**<0.001**	$68.41_{1,115}$	**<0.001**	$56.41_{1,115}$	**<0.001**
A. alp	$<0.01_{1,115}$	0.976	$0.72_{1,57}$	0.401	$0.10_{1,115}$	0.758	$0.53_{1,115}$	0.468
site × A. alp	$0.18_{1,115}$	0.673	$1.49_{1,57}$	0.228	$0.10_{1,115}$	0.758	$0.18_{1,115}$	0.674
pair	—	—	$1.71_{58,57}$	**0.022**	—	—	—	—
O. lapponica								
site	$4.07_{1,58}$	**0.048**	$1.16_{1,116}$	0.284	$3.28_{1,116}$	0.073	$12.73_{1,58}$	**0.001**
O. lap	$14.73_{1,58}$	**<0.001**	$2.92_{1,116}$	0.090	$13.96_{1,116}$	**<0.001**	$20.57_{1,58}$	**<0.001**
site × O. lap	$0.92_{1,58}$	0.341	$1.16_{1,116}$	0.284	$1.08_{1,116}$	0.300	$0.98_{1,58}$	0.326
pair	$2.04_{58,58}$	**0.004**	—	—	—	—	$2.28_{58,58}$	**0.001**

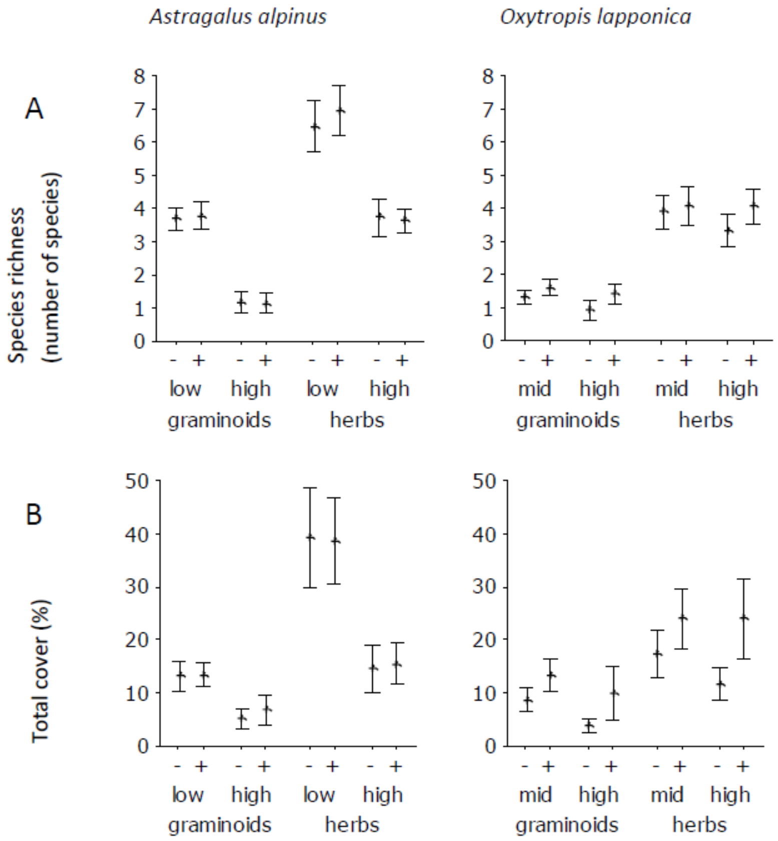


Fig. 2. Species richness (number of species) (A) and total cover (B) of graminoids and herbs in 15 × 15 cm sampling quadrats with (+) and without (-) the legumes Astragalus alpinus *and* Oxytropis lapponica *in the low, mid and high-elevation study sites sites at Mt. Sanddalsnuten, Finse, Norway, summer 2009. All figures show mean values with 95 % CI.*

DISCUSSION

Species composition differed with and without the presence of *Oxytropis lapponica*, and total species richness, graminoid species richness and total cover of graminoids and herbs were higher where *O. lapponica* was present, while species evenness was lower. None of the measured community properties differed with and without *Astragalus alpinus*.

Species composition differed with and without the presence of *O. lapponica* in the high-elevation study site at Finse, while there was no difference with and without *A. alpinus*. In Part 1 I found increased soil N levels below *O. lapponica* plants in the study area, and in Part

2 I found that the herb *Thalictrum alpinum* responded to legume presence with increasing leaf density, suggesting that the change in species composition may be caused by a change in plant density due to increased soil N availability. Tilman (1982) describes how some species respond quickly to fertilization, while others respond more slowly (or not at all), resulting in a change in plant community composition following nutrient addition. Such individualistic responses to nutrient addition have previously been found in arctic and alpine habitats by Shaver & Chapin (1980, 1986), Gough et al. (2002), Madan et al. (2007) and Klanderud (2008), and species-specific density responses to increased soil N levels below legumes may explain the differing species composition with and without *O. lapponica*.

Species richness increased with the presence of *O. lapponica* in the high-elevation site, while there was no difference in richness with and without *A. alpinus*. The results for *O. lapponica* are consistent with Perroni-Ventura et al. (2006), who found an increased species richness below the canopy of two leguminous trees, *Cercidium praecox* and *Prosopis laevigata*, in semi-arid Mexico. Their results showed that species richness was positively related to soil nutrient levels, which were higher below than outside the legume canopies (but see Thomas & Bowman 1998, Rodríguez-Echeverría & Pérez-Fernándes 2003, Rossi & Villagra 2003), and I believe that the increase in soil N below *O. lapponica* (Part 1) caused the increase in species richness in my study.

Grime (1973, 1979) and Tilman (1982) propose a "hump-backed" relationship between productivity and species richness. According to this model, species richness increases with soil nutrient levels up to a certain point as more species are able to survive, and then decreases as some species become dominant and exclude others. Several fertilization experiments have demonstrated such a decrease in species richness following nutrient addition in arctic and alpine areas (Chapin et al. 1995, Gough et al. 2000, Shaver et al. 2001, Gough et al. 2002, Ren et al. 2010, Wang et al. 2010 - but see Fox 1992). In my study the increase in soil fertility below *O. lapponica* was probably so small that more species could establish without any being competitively excluded. Thus, it appears that the presence of legumes facilitated species richness.

The difference in species composition and the increase in species richness with *O. lapponica* was found only in the high-elevation site, indicating that the facilitative effect increased with environmental stress, supporting the theory of Hacker & Gaines (1997), Michalet et al. (2006)

and Gross (2008). This is also in line with my findings in Part 2, where the effect of facilitation on a single target species was greatest in the high-elevation site. In Part 1 I found a greater difference between soil nutrient levels below and outside *O. lapponica* in the high-elevation than the mid-elevation site, indicating that the potential facilitative effect of the legume may be stronger in this site due to a proportionally greater improvement of soil conditions, as suggested by Pugnaire & Luque (2001).

Even though species richness increased with the presence of *O. lapponica*, species diversity did not change, most likely due to the decrease in species evenness. This is in accordance with Thomas & Bowman (1998), who suggest that their observed reduction in species diversity was due to a decrease in evenness (but see Rossi & Villagra 2003). According to Tilman (1982), the increased growth of some species following nutrient addition may result in decreased species evenness. In my study, a reduction in evenness due to the presence of one or more dominant species associated with the legumes may have "cancelled out" the positive effect of increased species richness on species diversity.

The dominant species reducing species evenness are most likely graminoids. I found an increased total cover of both graminoids and herbs where *O. lapponica* was present. However, graminoids seemed to have a greater response to increased soil nutrient levels, as it was the only functional group where species richness increased, and the percent increase in total cover was greater for graminoids than for herbs. This is in accordance with fertilization experiments in arctic and alpine habitats, which often result in a strong increase in graminoids (Fox 1992, Bowman et al. 1993, Theodose & Bowman 1997, Gough et al. 2002, Grellmann 2002, Heer & Körner 2002, Baer et al. 2004, van Wijk et al. 2004, Klanderud & Totland 2005, Klanderud 2008, Wang et al. 2010). However, contrasting the results of Bowman et al. (1993), the increase in graminoids in my study was apparently not great enough to negatively affect the richness of herbs, resulting in an overall increase in species richness where *O. lapponica* was present.

While *O. lapponica* appeared to influence species composition, species richness and species evenness at Finse, *A. alpinus* did not. According to Callaway (1998), positive interactions are species-specific, and not all legumes have been found to be facilitators, as demonstrated by studies showing neutral (Thomas & Bowman 1998, Rossi & Villagra 2003) or negative (Rodríguez-Echeverría & Pérez-Fernándes 2003) effects of legumes on species richness.

Rodríguez-Echeverría & Pérez-Fernándes (2003) suggest that the level of abiotic stress may determine whether or not a legume becomes a facilitator. In Part 1 I found increased soil N levels below *O. lapponica,* but not *A. alpinus,* suggesting that *A. alpinus* may be a poor facilitator. This is most likely because *A. alpinus* grows in less stressful microhabitats with a higher soil nutrient content than *O. lapponica* (see discussion Part 1). Although I did find increased densities of *T. alpinum* where *A. alpinus* was present (Part 2), *A. alpinus* does not seem to influence community properties in the study area.

CONCLUSION

Species composition differed and species richness increased with the presence of *Oxytropis lapponica* at Finse, most likely due to increased soil N availability below the legumes, while none of the measured community properties differed with and without *Astragalus alpinus*. The difference in species composition and the increase in species richness with *O. lapponica* were found only in the site of highest abiotic stress, supporting the theory that facilitation increases species richness under stressful environmental conditions. Species evenness was lower where *O. lapponica* was present, while species diversity did not change, most likely due to an increase in graminoid cover. However, the increase in graminoids was not great enough to exclude herb species, resulting in an overall increase in species richness. My study shows that *O. lapponica*, but not *A. alpinus*, contributes to increasing the biodiversity of the alpine ecosystem at Finse.

REFERENCES

Antonsson, H., Björk, R. G. & Molau, U. 2009. Nurse plant effect of the cushion plant *Silene acaulis* (L.) Jacq. in an alpine environment in the subarctic Scandes, Sweden. Plant Ecology and Diversity 2: 17-25.

Arroyo, M. T. K., Cavieres, L. A., Peñaloza, A. & Arroyo-Kalin, M. A. 2003. Positive associations between the cushion plant *Azorella monantha* (Apiaceae) and alpine plant species in the Chilean Patagonian Andes. Plant Ecology 169: 121-129.

Askvik, H. 2008. Berggrunnskart Hardangerjøkulen 1416 2, M 1:50000. Norges geologiske undersøkelse.

Baer, S. G., Blair, J. M., Collins, S. L. & Knapp, A. K. 2004. Plant community responses to resource availability and heterogeneity during restoration. Oecologia 139: 617-629.

Bellingham, P. J., Walker, L. R. & Wardle, D. A. 2001. Differential facilitation by a nitrogen-fixing shrub during primary succession influences relative performance of canopy tree species. Journal of Ecology 89: 861-875.

Bowman, W. D., Theodose, T. A., Schardt, J. C. & Conant, R. 1993. Constraints of nutrient availability on primary production in two alpine tundra communities. Ecology 74: 2085-2097.

Brewer, J. S., Levine, J. M. & Bertness, M. D. 1997. Effects of biomass removal and elevation on species richness in a New England salt marsh. Oikos 80: 333-341.

Callaway, R. M. 1998. Are positive interactions species-specific? Oikos 82: 202-207.

Carino, D. A. & Daehler, C. C. 2002. Can inconspicuous legumes facilitate alien grass invasions? Partridge peas and fountain grass in Hawai´i. Ecography 25: 33-41.

Cavieres, L., Arroyo, M. T. K., Peñaloza, A., Molina-Montenegro, M. & Torres, C. 2002. Nurse effect of *Bolax gummifera* cushion plants in the alpine vegetation of the Chilean Patagonian Andes. Journal of Vegetation Science 13: 547-554.

Cavieres, L. A. & Badano, E. I. 2009. Do facilitative interactions increase species richness at the entire community level? Journal of Ecology 97: 1181-1191.

Chapin, F. S., III, Shaver, G. R., Giblin, A. E. Nadelhoffer, K. J. & Laundre, J. A. 1995. Responses of arctic tundra to experimental and observed changes in climate. Ecology 76: 694-711.

del Moral, R. & Rozzell, L. R. 2005. Long-term effects of *Lupinus lepidus* on vegetation dynamics at Mount St. Helens. Plant Ecology 181: 203-215.

Fox, J. F. 1992. Responses of diversity and growth-form dominance to fertility in Alaskan tundra fellfield communities. Arctic and Alpine Research 24: 233-237.

Gough, L., Osenberg, C. W., Gross, K. L. & Collins, S. L. 2000. Fertilization effects on species density and primary productivity in herbaceous plant communities. Oikos 89: 428-439.

Gough, L., Wookey, P. A. & Shaver, G. R. 2002. Dry heath arctic tundra responses to long-term nutrient and light manipulation. Arctic, Antarctic, and Alpine Research 34: 211-218.

Grellmann, D. Plant responses to fertilization and exclusion of grazers on an arctic tundra heath. Oikos 98: 190-204.

Grime, J. P. 1973. Control of species density in herbaceous vegetation. Journal of Environmental Management 1: 151-167.

Grime, J. P. 1979. Plant strategies and vegetation processes. John Wiley & Sons, Ltd., Chichester. Pp. 222.

Gross, K. 2008. Positive interactions among competitors can produce species-rich communities. Ecology Letters 11: 929-936.

Hacker, S. D. & Bertness, M. D. 1999. Experimental evidence for factors maintaining plant species diversity in a New England salt marsh. Ecology 80: 2064-2073.

Hacker, S. D. & Gaines, S. D. 1997. Some implications of direct positive interactions for community species diversity. Ecology 78: 1990-2003.

Halvorson, J. J., Smith, J. L. & Franz, E. H. 1991. Lupine influence on soil carbon, nitrogen and microbial activity in developing ecosystems at Mount St. Helens. Oecologia 87: 162-170.

Heer, C. & Körner, C. 2002. High elevation pioneer plants are sensitive to mineral nutrient addition. Basic and Applied Ecology 3: 39-47.

Holzapfel, C., Tielbörger, K. Parag, H. A., Kigel, J. & Sternberg, M. 2006. Annual plant-shrub interactions along an aridity gradient. Basic and Applied Ecology 7: 268-279.

Jacot, K. A., Lüscher, A., Suter, M., Nösberger, J. & Hartwig, U. A. 2005. Significance of legumes for the distribution of plant species in grassland ecosystems at different altitudes in the Alps. Plant Ecology 180: 1-12.

Kammer, P. M. & Möhl, A. 2002. Factors controlling species richness in alpine plant communities: An assessment of the importance of stress and disturbance. Arctic, Antarctic, and Alpine Research 34: 398-407.

Kikvidze, Z., Pugnaire, F. I., Brooker, R. W., Choler, P., Lortie, C. J., Michalet, R. & Callaway, R. M. 2005. Linking patterns and processes in alpine plant communities: A global study. Ecology 86: 1395-1400.

Klanderud, K. 2008. Species-specific responses of an alpine plant community under simulated environmental change. Journal of Vegetation Science 19: 363-372.

Klanderud, K. & Totland, Ø. 2005. Simulated climate change altered dominance hierarchies and diversity of an alpine biodiversity hotspot. Ecology 86: 2047-2054.

Lepš, J. & Šmilauer, P. 2003. Multivariate analysis of ecological data using CANOCO. Cambridge University Press, Cambridge. Pp. 269.

Li, W.-J., Li, J.-H., Lu, J.-F., Zhang, R.-Y. & Wang, G. 2010. Legume-grass species influence plant productivity and soil nitrogen during grassland succession in the eastern Tibet Plateau. Applied Soil Ecology 44: 164-169.

Lid, J. & Lid, D. T. 2005. Norsk flora. 7. utgåva. Det Norske Samlaget, Oslo. Pp. 1230.

Ludwig, J. A. & Reynolds, J. F. 1988. Statistical ecology. A primer on methods and computing. John Wiley & Sons, Inc., New York. Pp. 337.

Madan, N. J., Deacon, L. J. & Robinson, C. H. 2007. Greater nitrogen and/or phosphorus availability increase plant species' cover and diversity at a high arctic polar semidesert. Polar Biology 30: 559-570.

Maron, J. L. & Connors, P. G. 1996. A native nitrogen-fixing shrub facilitates weed invasion. Oecologia 105: 302-312.

Maron, J. L. & Jefferies, R. L. 1999. Bush lupine mortality, altered resource availability, and alternative vegetation states. Ecology 80: 443-454.

Michalet, R., Brooker, R. W., Cavieres, L. A., Kikvidze, Z., Lortie, C. J., Pugnaire, F. I., Valiente-Banuet, A. & Callaway, R. M. 2006. Do biotic interactions shape both sides of the humped-back model of species richness in plant communities? Ecology Letters 9: 767-773.

Moen, A. 1998. Nasjonalatlas for Norge: Vegetasjon. Statens Kartverk, Hønefoss. Pp. 199.

Perroni-Ventura, Y., Montaña, C. & García-Oliva, F. 2006. Relationship between soil nutrient availability and plant species richness in a tropical semi-arid environment. Journal of Vegetation Science 17: 719-728.

Pugnaire, F. I. & Luque, M. T. 2001. Changes in plant interactions along a gradient of environmental stress. Oikos 93: 42-49.

Ren, Z., Li, Q., Chu, C., Zhao, L., Zhang, J., Ai, D., Yang, Y. & Wang, G. 2010. Effects of resource additions on species richness and ANPP in an alpine meadow community. Journal of Plant Ecology 3: 25-31.

Rodríguez-Echeverría, S. & Pérez-Fernández, M. A. (2003). Soil fertility and herb facilitation mediated by *Retama sphaerocarpa*. Journal of Vegetation Science 14: 807-814.

Rossi, B. E. & Villagra, P. E. 2003. Effects of *Prosopis flexuosa* on soil properties and the spatial pattern of understorey species in arid Argentina. Journal of Vegetation Science 14: 543-550.

Shaver, G. R., Bret-Harte, M. S., Jones, M. H., Johnstone, J., Gough, L., Laundre, J. & Chapin, F. S., III. 2001. Species composition interacts with fertilizer to control long-term change in tundra productivity. Ecology 82: 3163-3181.

Shaver, G. R. & Chapin, F. S., III. 1980. Response to fertilization by various plant growth forms in an Alaskan tundra: Nutrient accumulation and growth. Ecology 61: 662-675.

Shaver, G. R. & Chapin, F. S., III. 1986. Effect of fertilizer on production and biomass of tussock tundra, Alaska, U.S.A.. Artic and Alpine Research 18: 261-268.

Tewksbury, J. J. & Lloyd, J. D. 2001. Positive interactions under nurse-plants: Spatial scale, stress gradients and benefactor size. Oecologia 127: 425-434.

The Norwegian Meteorological Institute 2010. eKlima. Data from weather station 25840 at Finse, normal period 1961-1990. To be found on the web page http://eklima.met.no. Last visited 02.02.2010.

Theodose, T. A. & Bowman, W. D. 1997. Nutrient availability, plant abundance, and species diversity in two alpine tundra communities. Ecology 78: 1861-1872.

Thomas, B. D. & Bowman, W. D. 1998. Influence of N_2-fixing *Trifolium* on plant species composition and biomass production in alpine tundra. Oecologia 115: 26-31.

Tilman, D. 1982. Resource competition and community structure. Monographs in Population Biology 17. Princeton University Press, Princeton. Pp. 296.

Tilman, D. 1984. Plant dominance along an experimental nutrient gradient. Ecology 65: 1445-1453.

van Wijk, M. T., Clemmensen, K. E., Shaver, G. R., Williams, M., Callaghan, T. V., Chapin, F. S., III, Cornelissen, J. H. C., Gough, L., Hobbie, S. E., Jonasson, S., Lee, J. A., Michelsen, A., Press, M. C., Richardson, S. J. & Rueth, H. 2003. Long-term ecosystem level experiments at Toolik Lake, Alaska, and at Abisko, Northern Sweden: Generalizations and differences in ecosystem and plant type response to global change. Global Change Biology 10: 105-123.

Wang, C., Long, R., Wang, Q., Liu, W., Jing, Z. & Zhang, L. 2010. Fertilization and litter effects on the functional group biomass, species diversity of plants, microbial biomass, and enzyme activity of two alpine meadow communities. Plant and Soil 331: 377-389.

Xiao, S., Michalet, R., Wang, G. & Chen, S.-Y. 2009. The interplay between species' positive and negative interactions shapes the community biomass-species richness relationship. Oikos 118: 1343-1348.

APPENDIX 1. Percent cover of all vascular plant species in 15 × 15 cm sampling squares with (+) and without (-) the legumes *Astragalus alpinus* and *Oxytropis lapponica* in the low (L), mid (M) and high (H)-elevation study sites at Mt. Sanddalsnuten, Finse, Norway, summer 2009. * denotes missing values.

Site	*A. alp*	*O. lap*	*Ant dio*	*Ant nip*	*Bar alp*	*Bis viv*	*Cam rot*	*Car atr*	*Car big*	*Car cap*	*Car rup*	*Car vag*	*Cer alp*	*Com ten*	*Des alp*	*Dra nor*	*Dry oct*	*Emp nig*	*Equ var*	*Eri sp.*	*Eup wet*	*Fes ovi*	*Fes viv*	*Gen niv*	*Har hyp*	*Hup arc*	*Jun trif*	*Jun trig*	*Kob myo*	*Leo aut*	*Luz con*	*Luz spi*	*Min bif*	*Par pal*	*Pin vul*	*Poa alp*	*Pot cra*	*Ran acr*	*Rho ros*	*Rum ace*	*Sal her*	*Sal ret*	*Sau alp*	*Sax cer*	*Sax ces*	*Sax opp*	*Sel sel*	*Sil aca*	*Tha alp*	*Tof pus*	*Tri spi*	*Vac uli*	*Vac vit*	*Ver alp*
L	+				1	3						2							3	3	1	4								2												10	15				3	20	10	4				
L	-					2	2	2			3	1							3	6										3				1		2		1				12	20				1	12	10					1
L	+						2	2			3	3							1	1	1	2								1				2			1					2	12				1		9					2
L	-				2	2						5							1			4								3												3	10				1		30	2				
L	+					3				2		6										3								2				1								12	5				2	5	12	1				
L	-				2	2					3	3							1	1		3								3				2				2				2	25				2		12	1		1		2
L	+				1	4					2	5							3	2		2												1	3							3	20				2	40	18					
L	-					3		5			2	3										3												1								7	20				2	30	9	1		5		
L	+				1	1	1			7	1	4							1		1	3																				2	6					7	5	2		5		
L	-				1	1				9		5					15				1	3														1							5				1	22	10	2		1		
L	+				2	1	1				4	2								2	1	2								4				1			2	2				2	30				1		7			4		
L	-					2		3		1	2	2							2			2																1				2	15				1	75	6			3		
L	+				2					2	2	2										3															1						30					25	3					1
L	-				1	2					2	2					18		1		1	2												2			5						25				2		4					
L	+					2				1		5					6				1	2								3				1		2				1		5	12				2	6	10	5		1		
L	-				2	4					1	5										3												1								5	7				3	45	4	2				
L	+					2				3	3	3										2																				1	12				2	40	10	3		2		
L	-					2		4		3		6							1			3										2										2	3				2	1	15	2		3		
L	+					2		2		2	1	6							1			1																				2							4	2				
L	-					2				3	1	10										1																				2					6	1	8	2				
L	+				3	3				5		4							2			2																				2	2				3	20	4					1
L	-				2	5	1			5	3	9					1		4		2							1														2	2					3	6	4				
L	+				3	2			3	5		5					7					3										1		1			3					4	4				4		8	2				
L	-				2	3						8					65					2																				2	4						4	2				

L	+			6	2		3		3	2	7	4					1		2				2						1	7	1	7	10	3			
L	-			4	3				4	2	3					2	1	1	2					2			3		4	10	1	10	6	3			
L	+			4	5					4	4								3		1		6		2			1		9	3		5	3			
L	-				3				2	2	5								1				2			1			2	2	2	9	10			3	
L	+	4		1	3	1			2	2	3							1	2			2			1	2	1		1	7	3	12	6	5			
L	-	2			2					5	2								3										4	3	2	35	10	2		6	1
L	+				4					5	4				7			1											10	4	3	6	7			3	
L	-				6					2	7	2			3			1		1					2		1		8	5		3	6	2		4	
L	+			6	4					8	14				25	1			1						1					7	1	6	6				
L	-			1	3		2		20	2	15				6	2		1											3				5				
L	+				2			2		8	4				45	1					2								4	4	2	1	3				
L	-				3			12	15	2	2								1										2	3	1	5	7	2			
L	+	2		5	2				2	4	5						1		1										10	6	2	10	9	2		3	
L	-			5	3				3	6	6					1			1						1				3	5	2	35	10			2	1
L	+			2	2			2	8		10						2		2						2					7	1		12	4		1	
L	-				2		3			1	5								4										2	4	2	3	8	4		7	
L	+				3				2		7					1			4						8				2	8	3		18			3	
L	-				3				6		4								4										1	1	22	4	12				
L	+			2	5		2		3	4	3			4	3	1		1	3										2	6	2		10	4			
L	-			2	2	1	3			2	5								4										5	8	1	2	10	5			
L	+				2				3	3	10								10		1			2					3				4	3			
L	-				2				2	3	4				18														3	3		2	5	3			
L	+			2	2					1	4								2								2		3	35	1	10	2			12	
L	-			1	2		2			2	5								2							1			5	6	1		7			4	
L	+			3	2		1		2		4								2				2						4	6	1	20	5				
L	-			3	5	1			3	3	8						1		2				2		2				4		3	12	20	1			1
L	+	4					2			3	3					2	2		2				3		2				4	6	1		7	2	4	3	
L	-										2					1			2					2	1				3	10	3		15			4	
L	+	3		2	12	1					4		1						2								1		3	12	1	70	3	3		6	
L	-			1	1					2	2						1		2		1								7	20	3		1	4		2	
L	+		7								5	1				1	3		2		1		3			8			2	10		4	7				
L	-			2	6				3	2	2						2		1						1		1		12	8	1	2	10	2			1
L	+				5	1	2				4					2			2						1				20	20		7	8				
L	-			1	4						5								3						2				3	6		90	10	3			
L	+				3				8	14	3							1	1											2		5	8				

L	-	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*
M	+									25																																						4					
M	-									16					1																						5											12	10					
M	+				2	1				2																		15																					12					
M	-			3	1					5						97																											2						3					
M	+				1					8					2				2			25															3				2					3			3					
M	-									4					1							4																										5	5					
M	+									15																															1					6			12					
M	-									10																																						5					
M	+									10		5																									1				1		1						4					
M	-				2							1			1																					1	1				1		2			9			8					
M	+									7					1																						5				1		2					5	8					
M	-									12					1							2																								1			18					
M	+				8							6																													1					1			8					
M	-				3					7					1																												2			8		2	5					
M	+				4																															2					3		9	1				2	10					
M	-				3							1			3							10																								6			2					
M	+				4					5		1	1		1							5															3		2										10					
M	-				2					8						97																					1						3						3					
M	+									10		2										6																					4						10					
M	-									6																													4							5			7					
M	+				3	1				5						40												14																					9					
M	-				3	1				5						50												6															1					2	8					
M	+				3					7		1										6																										35	20					
M	-				3					5		1			1																																	14	10					
M	+				1					18												12									2												4						9					
M	-				2					18																																				1			15					
M	+				2					12																					2												2						15					
M	-				3					5						3						2																					1					2	5					
M	+				3	1				10						14						7															1									12			12					
M	-									7												22																										55	4					
M	+					1				18		1			2							3															2						5			1		5	20					
M	-									11					1																								1		1					1			10					
M	+									10					1																						20				1		6					25	8					
M	-														4																						5				2		3						5					

M	+						9	1		1											6		7		10	
M	-						6			2									3						6	
M	+			2			20	1		2			3			1	1							50	10	
M	-							4		2			2			2	5								10	
M	+	3		2			12		1							2	4		1					3	14	
M	-						7	1				4					12	3							7	
M	+						10	3		1							10								10	2
M	-			3			10	3		2															5	
M	+						6	1		1							12						5		15	
M	-						3														2				7	
M	+	3					4			1							18								5	
M	-	2		3			5			1							3						2	18	12	
M	+	4					6			2									2				2		7	
M	-			1			4			1															3	
M	+									2			3						5						8	
M	-							1		3			6			1	4						2		3	
M	+						8																	20	13	
M	-			3		6								6			3				2			4	5	
M	+			2			9									1	10				3			2	5	
M	-			2			8								3									4	2	
M	+						6			1													7		5	3
M	-						2			1									2				3			
M	+			3			14							2											3	
M	-			3	1		6				30			5					2						2	
M	+			3	1		5							15											15	
M	-		5	2			12				4									2	5				15	
H	+									2											5	3	1		1	
H	-																		1					6	3	
H	+						1												25					5	2	
H	-			1																			1		3	
H	+												3						12				1	1	3	
H	-	1						2					3				20		3						3	
H	+						8	5					2				2		4					70	1	
H	-																		8		8	2			3	
H	+						10	1					50				2							20	1	

H	-			5										15					1	2	
H	+		1	7	2			1											8	20	
H	-			12								1		3				5	4	3	
H	+			3	6		3					1		2				2	20	8	
H	-		1	3									3					3		6	
H	+			2								17		1		1			25	3	
H	-		2	6								3		4		3				4	
H	+			4	5									3		3			20	5	
H	-		3	5										2		1	1			3	
H	+		3	5						5				3					65	7	
H	-	3	3	4						8				3						5	
H	+		3	5							2	10		1						4	
H	-		3	3					2					2		5				3	
H	+		2	7						30				2		2				3	
H	-		2	4																2	
H	+	2	2	6		1			1		1			3		2			3	4	3
H	-	2	2	7								4		2					1	7	
H	+		5	5		1										1				12	2
H	-		7	6										3		2		2	4	8	
H	+		2	3					1	3	1	1		3					3	10	
H	-		3	5	1					5		1		4		2			25	3	
H	+	2	4		2				1					5					3	6	
H	-		2											4				2		3	
H	+	2	3	3	3				1	2				4		2			18	7	
H	-	5	3											6						3	
H	+		3	12										2		1			25	3	2
H	-		4	6		1								3						2	1
H	+		3	3	2					3	2			2		3				4	5
H	-			7											7					2	
H	+		1					2		5				2				5		4	
H	-			2						2				3						3	
H	+		4		1					2				4						3	
H	-		3							2	2					2			1	5	2
H	+			3										10				2		3	
H	-													20			1	5		2	

H		+			2	2	3											15		4		2	40	3	
H		-																20		2				4	
H		+			3	3							18					9		3		2	2	2	
H		-	1		1								1		1			20		4				2	
H		+			1	6												5						2	
H		-																				3		3	
H		+					4								4			6		3				4	
H		-	4		2					2			1					2				3	15	5	
H		+			3	10	1						5					1					10	3	
H		-			5	4		1										7						3	
H		+	4		1	1							45					2						7	
H		-			4	6	6						4		1		1	1				5		4	
H		+			1				50									2						1	5
H		-					1											1		2				2	
H		+			1	2							3					1		3		2	50	3	
H		-																10			3	2			
H	+												1					4		3		2		3	
H	-						3											4				2		3	
H	+																	6				1	10	2	
H	-																	1						1	8
H	+					2		1										8				1		5	
H	-					1												4						2	
H	+												2					4		2			50	2	
H	-				3								9					4		4				3	
H	+				1		5				1			5				20		3				7	
H	-						1	2						5		1		15		3			2	4	
H	+						1					30						10	2	1				1	
H	-					2												9					15	2	
H	+				2								13					7		3				2	
H	-			2	4	1												2		2			5	7	6
H	+					4	1											18						6	
H	-					4												8				2		3	
H	+		2															8		4		1		12	10
H	-				1								2					1		2				2	2
H	+				2	3												15					5	3	

H	-			6		5	2													1		2			1		9	2
H	+																			6		1					1	
H	-			5	1	4	5						1		12					1	12	1				56	2	
H	+			2											6					6		2			2	3	10	
H	-			6		5	4	2							5		2			2		3				1	5	
H	+			1		5							1		3					6		3					11	
H	-			2		9														1		4				1	9	
H	+					10		3	1											1		4					6	
H	-					8			1											5							4	
H	+	2																		20		3					11	4
H	-			1		1									6					10		3					6	7
H	+			3		2									3					3		3				5	3	
H	-	3		3											8					7		8			1		6	
H	+			2		1		2							2		2			7		2				5	25	
H	-					1						2								11		5			2		1	
H	+					2								30						5		1			1	2	2	
H	-			2				1	1						7	2		1		10			3		1		2	
H	+			2		3														9		1				2	5	
H	-					6											2			7					6		7	
H	+					7														10		5				4	9	
H	-			1		6														3				1	1		7	
H	+			2							2		1							5							8	
H	-	1	2	4		3														4							10	9
H	+																			25		3			3	1	2	
H	-			1				4							3				12								8	
H	+			4		8				3					4		2									12	10	6
H	-			2		2				97												1					1	
H	+			4		5	2													3		2			1	1	7	3
H	-			4	2	4	2													25		2				5	12	4
H	+														6					5						5	3	
H	-	1																1		6						25	5	
H	+							4	1						10					35		4				15	2	
H	-	1				3		3							4					12		1	1			3	4	
H	+													5	3					25		3			1	12	2	
H	-																			18				3	3		1	

H	+		5			5	2			73		10	4		2	
H	-		5							2	1	7		1	8	
H	+		3	1	6							5	4	2	17	1
H	-		2		5			1				2	2		8	13
H	+	1	2		8							3	7		4	5
H	-		3						2			20	2		2	6

APPENDIX 2. Percent quadrats of occurrence and mean % cover ± 1 SE for the 10 most frequent species found with (+) and without (-) the legumes *Astragalus alpinus* and *Oxytropis lapponica* in 15 × 15 cm quadrats in the low, mid and high-elevation study sites at Mt. Sanddalsnuten, Finse, Norway, summer 2009.

	A. alpinus				***O. lapponica***			
	Low		High		Mid		High	
	+	-	+	-	+	-	+	-
Bistorta vivipara								
% occurence	86.7	96.6	46.7	60.0	50.0	53.3	66.7	60.0
% cover	2.7 ± 0.4	2.8 ± 0.3	1.2 ± 0.3	1.8 ± 0.4	1.4 ± 0.3	1.3 ± 0.2	1.6 ± 0.3	1.7 ± 0.3
Carex rupestris								
% occurence	63.3	75.9	46.7	60.0	90.0	80.0	76.7	53.3
% cover	2.5 ± 0.6	1.8 ± 0.3	2.2 ± 0.6	2.3 ± 0.5	9.0 ± 1.1	6.1 ± 0.9	3.7 ± 0.6	2.8 ± 0.6
Dryas octopetala								
% occurence	20.0	24.1	3.3	3.3	6.7	20.0	3.3	—
% cover	3.1 ± 1.7	4.3 ± 2.4	0.1 ± 0.1	3.2 ± 3.2	1.8 ± 1.4	9.4 ± 4.8	1.7 ± 1.7	—
Festuca vivipara								
% occurence	—	3.4	36.7	33.3	30.0	23.3	46.7	26.7
% cover	—	0.03 ± 0.03	1.8 ± 0.6	1.9 ± 0.6	2.3 ± 0.9	1.6 ± 0.8	5.9 ± 2.4	0.9 ± 0.3
Potentilla crantzii								
% occurence	23.3	13.8	—	10.0	43.3	30.0	20.0	20.0
% cover	0.4 ± 0.1	0.3 ± 0.2	—	0.1 ± 0.06	3.0 ± 1.0	1.3 ± 0.5	1.1 ± 0.6	1.0 ± 0.7
Salix herbacea								
% occurence	—	—	96.7	93.3	33.3	20.0	86.7	80.0
% cover	—	—	9.6 ± 1.5	6.7 ± 1.2	0.6 ± 0.2	0.4 ± 0.1	4.3 ± 1.0	4.9 ±1.1
Salix reticulata								
% occurence	83.3	93.1	3.3	3.3	—	3.3	—	3.3
% cover	3.8 ± 0.8	3.8 ± 0.6	0.07 ± 0.07	0.4 ± 0.4	—	0.07 ± 0.07	—	0.2 ± 0.2
Saussurea alpina								
% occurence	93.3	89.7	76.7	53.3	33.3	33.3	43.3	33.3
% cover	10.1 ± 1.6	7.9 ± 1.4	2.3 ± 0.3	1.5 ± 0.4	1.4 ± 0.4	0.8 ± 0.2	1.1 ± 0.3	1.0 ± 0.3
Silene acaulis								
% occurence	66.7	72.4	53.3	33.3	30.0	33.3	60.0	26.7
% cover	10.7 ± 2.9	13.8 ± 4.2	4.5 ± 1.8	3.8 ± 2.0	4.9 ± 2.2	3.9 ± 2.0	12.9 ± 3.6	1.7 ± 0.1
Thalictrum alpinum								
% occurence	100.0	100.0	100.0	100.0	100.0	96.7	100.0	96.7
% cover	7.7 ± 0.7	9.3 ± 1.0	6.1 ± 1.0	4.8 ± 0.6	9.8 ± 0.8	6.7 ± 0.8	4.6 ± 0.7	3.4 ± 0.3