

Interactive effects of nitrogen deposition, fire and grazing on diversity and composition of low-alpine prostrate *Calluna vulgaris* heathland

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Summary

1. Low-alpine heathlands world-wide have high biodiversity value but are increasingly impacted by the effects of nitrogen deposition as well as fire and grazing. We conducted a 5-year fertilization experiment to examine the interactive effects of these three factors on vegetation diversity and species composition.
2. Factorial combinations of nitrogen addition (0, 10, 20 and 50 kg N ha⁻¹ year⁻¹), burning (burned once or not burned) and grazing (annual clipping, 0% or 12% of current year shoots removed) were applied to internationally important prostrate *Calluna*–*Cladonia* heathland in the Cairngorm Mountains, Scotland. Species diversity (higher plants, mosses and lichens) and percentage cover were recorded annually.
3. Fire had a large effect on vegetation diversity and composition, but both were quick to recover. Species richness recovered within 4 years and vegetation composition was predicted to recover within 7 years. Vegetation composition appeared resilient to the effects of disturbance and there was little invasion by graminoids.
4. Nitrogen deposition interacted with fire. Burned plots showed no significant effect of nitrogen treatment on species diversity, while the diversity of unburned plots was significantly reduced only 1 year after treatment with 50 kg N ha⁻¹ year⁻¹. After 5 years, significant diversity reductions were seen in the 10 kg N ha⁻¹ year⁻¹ treatment. Impacts of nitrogen on species richness were primarily through reductions in lichen diversity.
5. Severe winter browning of *Calluna vulgaris* in plots receiving 50 kg N ha⁻¹ year⁻¹ followed early snowfall in 2002–03. This interaction affected only *Calluna vulgaris* and may have the potential to trigger species composition changes by reducing the dominance of this species.
6. *Synthesis and applications.* Although resilient to small-scale fires, species richness of low-alpine heaths is reduced by exposure to low levels of nitrogen deposition (background + 10 kg N ha⁻¹ year⁻¹). The lichen component of the vegetation is most sensitive to additional nitrogen, although higher plants can be affected via interactions with climate. These data support the current critical load of nitrogen for this community of 5–15 kg N ha⁻¹ year⁻¹ and suggest that lichen diversity could be a useful indicator of nitrogen deposition impacts in alpine habitats. Effective conservation of biodiversity in low-alpine heathland will require action at national and international levels to reduce nitrogen deposition in the many areas where the critical load is currently exceeded.

Key-words: community composition, critical loads, fire, grazing, lichens, montane heathland, N pollution

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Introduction

Despite their perceived 'pristine' nature and remoteness from major urban centres, alpine regions in Europe and elsewhere are becoming increasingly impacted by human activity. Diffuse pollution (including acidification and nitrogen deposition) and poor land management (amongst other drivers) have the potential to cause major impacts on the biodiversity and functioning of alpine ecosystems world-wide. Alpine regions represent significant reservoirs of biodiversity as, globally, the alpine life zone covers 3% of the land area and holds 4% of higher plant species (Körner 1995), while on a European scale the richness of alpine areas is even more marked, with 20% of species occupying 3% of the land surface (Väre *et al.* 2003). In addition, the alpine zone performs a vital functional role in the storage and supply of water to lowland regions, as it contains the headwaters of many major rivers. Up to the present time, studies of the impacts of nitrogen deposition have been focused on lowland and upland habitats, while very little is known about the sensitivity of alpine communities (Bobbink, Hornung & Roelofs 1998). Deposition of nitrogen is thought to be potentially detrimental to alpine vegetation, which has generally developed under conditions of low nutrient availability. The few studies that have been conducted suggest that the cryptogram element of the vegetation may be particularly sensitive (Bobbink, Hornung & Roelofs 1998; Fremstad, Paal & Möls 2005). Nitrogen addition has been shown to cause damage to bryophytes and increased dominance of graminoids in *Racomitrium* heath (Pearce, Woodin & van der Wal 2003; van der Wal *et al.* 2003). This sensitivity has been reflected in the critical load of nitrogen for arctic and alpine heaths, which is currently set at 5–15 kg N ha⁻¹ year⁻¹ (Achermann & Bobbink 2003). Interactions with management factors such as grazing and fire have been shown to have the potential to modify community responses to nitrogen deposition in some systems (van der Wal *et al.* 2003; Barker *et al.* 2004) and so the impacts of nitrogen should not be considered in isolation.

Alpine vegetation in the UK shows evidence of the impacts of both nitrogen deposition and poor land management (heavy sheep grazing and fire; Thompson & Brown 1992). Although generally remote from point sources of pollution, high levels of rainfall and prolonged cover with orographic cloud mean that alpine areas in the UK are exposed to relatively high levels of pollutant deposition. Nitrogen deposition in UK mountain areas currently varies between approximately 7 and 56 kg N ha⁻¹ year⁻¹ (Hall, Heywood & Smith 2004). This is a large range of deposition compared with the rest of Europe and, while conditions in southern Britain are similar to those in continental Europe, the Scottish pollution climate is closer to that in Scandinavia. Grazing has a long history in the alpine regions of Europe and is thought to have caused locally severe community changes and diversity reductions (Ersch-

bamer, Virtanen & Nagy 2003). In the UK, grazing and trampling by domestic herbivores have been blamed for the complete absence of alpine dwarf shrub heaths from areas south of the Scottish Highlands that otherwise appear suitable for this habitat (Thompson & Brown 1992), and continue locally to have detrimental impacts on vegetation composition (Britton, Pearce & Jones 2005). Management of subalpine vegetation, especially heathlands, often takes the form of rotational burning in the UK, and this too can cause damage to communities in the low-alpine zone when fire spreads up from lower altitudes. Strong climatic limitations on growth mean that low-alpine communities may take a long time to recover from such disturbances. While grazing and land management may be locally altered to achieve conservation goals, pollution emissions and impacts occur at a larger spatial scale and must be influenced by application of control policies at a national and international level.

In the UK the low- to mid-alpine zone occupies around 3% of the land surface and contains the most extensive remaining areas of near-natural plant communities (Thompson & Brown 1992; Nagy 2003). British alpine vegetation consists primarily of dwarf shrub-, moss- and grass-dominated heathlands with significant areas of fell field and snowbed vegetation. The largest and best-preserved areas of alpine vegetation occur in Scotland, while smaller areas are present in England and Wales. The alpine plant communities contain elements of both arctic and alpine floras but are strongly influenced by the oceanic climate. This combination of characters results in a number of plant communities that are either unique to or particularly well represented in the UK. Although the higher plant flora is relatively small (around 224 species recorded above the tree line; Nagy 2003) the lichen and bryophyte floras are rich (700 and 212 species above the tree line, respectively; Fryday 1997; Usher 1997). Alpine areas also support important populations of bird species, such as dotterel *Charadrius morinellus* and ptarmigan *Lagopus mutus*.

Calluna–*Cladonia* heathland (NVC H13; Rodwell 1991) is a climax community co-dominated by prostrate *Calluna vulgaris* (nomenclature for higher plants follows Stace 1991) and terricolous macrolichens, and is almost unique to the UK. This type of vegetation occurs just above the potential treeline and occupies large areas of the low-alpine zone in eastern Scotland (Metcalf 1950; Averis *et al.* 2004). The community has similarities with prostrate dwarf shrub vegetation in Scandinavia, particularly in terms of the lichen component (Averis *et al.* 2004), and is the main habitat for a number of species that are rare in Britain, such as the Red Data Book lichen *Alectoria ochroleuca* (nomenclature for lichens follows Coppins 2002). Although the effects of nitrogen deposition on upland *Calluna* heathlands have been well studied, little is known about how the response of alpine *Calluna* heathland might differ. This community is a climatically controlled

climax vegetation, unlike the upland and lowland heaths, which are successional communities maintained by management. Hence the vegetation is subject to much stronger climatic limitations than that at lower altitude, including a short growing season, high wind speeds and exposure to low winter temperatures, because it occurs in areas with little snow cover. Based on current knowledge of related systems, Bobbink, Hornung & Roelofs (1998) have hypothesized that alpine heaths may be particularly sensitive to nitrogen deposition. In this study we investigated the medium-term effects of nitrogen deposition, burning and grazing, and their interactions, on the species diversity and composition of these alpine *Calluna* heathlands.

Materials and methods

STUDY AREA

The Culardoch experimental site is located on an exposed col at 750 m above sea level, east of the main Cairngorm massif, in the Eastern Highlands of Scotland (3°20'W, 57°4'N; Ordnance survey grid reference No 184985). The vegetation consists of a large area (> 1 ha) of prostrate *Calluna*–*Cladonia* heathland (NVC H13) typical of exposed areas in the low-alpine zone of eastern Scotland. This climax community is dominated by prostrate *Calluna vulgaris* but contains a number of other shrub species, including *Vaccinium* species, *Empetrum hermaphroditum* and *Arctostaphylos uva-ursi*. Lichens, especially members of the genera *Cetraria* and *Cladonia*, are a prominent component of the vegetation, and there are few graminoids apart from occasional tufts of *Festuca ovina*, *Deschampsia flexuosa* and *Carex pilulifera* and few mosses. The site is flat and has a continuous vegetation cover, with *Calluna*–*Cladonia* heathland grading into typical upland *Calluna* heathland (NVC H16) approximately 100 m down-slope. The soil is a subalpine podzol overlying diorite.

The climate in this part of Scotland is cool oceanic, with cold winters and cool wet summers. The mean annual rainfall is 1051 mm, mean minimum air temperature in January –1.9 °C and mean maximum air temperature in July 13.0 °C. The average wind speed is high; the mean hourly wind speed is 6.9 m s⁻¹, with the maximum recorded hourly mean wind speed being 30.1 m s⁻¹ (climate data for on-site automatic weather station, July 2002–June 2005). The background deposition of nitrogen in rainfall was measured using a bulk collector funnel that was emptied fortnightly during the summer (May–October) and monthly during the rest of the year. The total input of nitrogen (oxidized and reduced forms) in the rainfall was estimated to be 10 kg N ha⁻¹ year⁻¹ for the period 2000–04; this compares well with a modelled total deposition (wet and dry) for this habitat of 10.8 kg N ha⁻¹ year⁻¹ during 1999–2001 (NEGTA 2001), as dry deposition is generally low in this wet alpine environment. This level of total nitrogen deposition is relatively low in the UK context.

The site is located on an estate managed primarily for hunting of red deer *Cervus elaphus* and red grouse *Lagopus lagopus scoticus*. The vegetation at this altitude is not actively managed but is located adjacent to more productive areas that are managed by traditional muir burning practices (Moorland Working Group 1998).

EXPERIMENTAL DESIGN

The experiment investigated the influence of three driving variables on the species composition of prostrate *Calluna vulgaris*-dominated heathland. These were: fire, nitrogen deposition and grazing. The three variables were combined in a fully factorial, randomized split-block experimental design with six replicates.

The fire treatment was applied as a single event at the beginning of the experiment in November 1999. Each block (8 × 9 m) was divided into two subblocks (3 × 9 m, separated by a 2-m buffer strip), one of which was randomly chosen to have the vegetation removed by burning with a blowtorch. Burning removed all of the above-ground vegetation on the plots, leaving only the remains of the largest woody stems; the majority of the litter layer was left intact. This treatment simulated the effect of low-intensity burns, which occur on prostrate *Calluna* heathland when management fires spread from adjacent subalpine heaths. The fuel load on prostrate *Calluna* heathland is too low to support a higher intensity fire. The remaining treatments were nested within the burned or unburned subblocks.

The nitrogen treatment comprised four levels of nitrogen addition: 0, 10, 20 and 50 kg N ha⁻¹ year⁻¹ in addition to the estimated background level of 10 kg N ha⁻¹ year⁻¹. This range included deposition levels commonly found in the UK and Europe, with 50 kg N ha⁻¹ year⁻¹ plus background being slightly above the upper end of the current range. Nitrogen was applied in six equal monthly doses during the growing season for the higher plants (May–October) as a solution of NH₄NO₃ in 1 L deionized water, starting in May 2000. Treatments were applied using a watering can; the additional water supply was equivalent to an extra 5.3 mm rainfall year⁻¹. The concentration of the nitrogen solutions ranged from 13.48 mM N for the 10 kg ha⁻¹ year⁻¹ addition to 66.8 mM N for the 50 kg N ha⁻¹ year⁻¹ addition.

As grazing pressure on prostrate *Calluna* heath is both spatially and temporally patchy, and applying nitrogen solutions to vegetation was thought to have the potential to attract herbivores, it was decided to use grazing exclosures on all plots and to clip the vegetation by hand to simulate grazing. Plots were clipped once year⁻¹ at the beginning of August using secateurs, and the clippings removed. Clipping was applied randomly across the plot and not restricted to any particular species; approximately 50% of the current year growth was removed from any individual shoot. A standard volume of shoots (one for burned and one for unburned plots) was used as a guide to ensure an even application

of the clipping across plots; this was based on previous tests outside the experimental area to relate volume of shoots to proportion of current year's growth removed. The proportion of the current year's growth that had been removed from each plot by the clipping treatment was measured annually in autumn using the method of Grant, Hamilton & Souter (1981). The mean proportion of current year growth removed for 2000–04 was 12%.

In total there were 16 treatment combinations with six replicates of each, giving a total of 96 plots. Each plot had a treated area of 1.5×0.75 m and was surrounded by a 20-cm wide buffer strip. The plots were fully enclosed within individual wire mesh grazing exclosures, with 5-cm mesh suitable for excluding red deer, sheep *Ovis aries* and mountain hares *Lepus timidus*.

Following establishment of the experiment in November 1999, the species composition (including all higher plants, mosses and non-saxicolous lichens) was recorded annually in May on a fixed 50×50 -cm quadrat within each plot. Quadrat position was permanently marked to ensure accurate relocation each year. Composition was recorded as percentage cover to the nearest 1% or 0.5% for species with cover less than 1%. A quadrat divided into 400 squares was used to increase the accuracy of the visual cover measurements by giving a grid to guide estimates. All higher plants and mosses were determined to species level, but some lichen species that could not be determined on morphological characters alone were recorded as species groups, as accurate determinations would have required chemical tests and removal of samples from the plot. This was the case for *Cladonia ciliata*, *Cladonia arbuscula* and *Cladonia portentosa* (recorded under the name *Cladonia 'arbuscula'*) and also for patches of *Cladonia* basal squamules where there were no podetia present. In May 2003, following an early snowfall event the previous autumn, damage to *Calluna vulgaris* was also recorded on each plot as the percentage cover of dead shoots.

DATA ANALYSIS

Species richness and cover data and the cover of damaged *Calluna vulgaris* were analysed using analysis of variance for individual years or repeated measures analysis of variance for the whole period using Genstat 7.2 (Genstat 7 Committee 2003). Species composition data (percentage cover of all higher and lower plant species) for the whole community over 5 years was analysed using a multivariate method known as principle response curves analysis (PRC) in Canoco for Windows 4.5 (ter Braak & Šmilauer 2002). This technique was developed specifically for analysis of community response data from designed experiments sampled repeatedly through time, and was chosen as it provides clear visualization of community composition trajectories in comparison to a control treatment. It is described in detail in van den Brink & ter Braak (1998, 1999). Monte Carlo permutation tests were used to test the significance of the resulting axes. PRC shows the changes in community composition through time with respect to a control treatment. In this analysis the $0 \text{ kg N ha}^{-1} \text{ year}^{-1}$ unburned treatment was selected as the control because these plots were closest to the situation outside the experimental area. Initial analysis showed that the clipping treatment had little or no effect on total community composition and so the analyses presented here include only burning and nitrogen treatment combinations, to aid the clarity of data presentation.

Results

SPECIES RICHNESS

Two of the treatments had significant effects on the total species richness of the community (Table 1 and Fig. 1), of which the strongest was burning, which initially removed all the vegetation cover from the burnt plots and reduced species richness to close to zero in

Table 1. Repeated-measures ANOVA probabilities for effects of burning, clipping and nitrogen addition on total species richness, lichen species richness, *Calluna vulgaris* cover, non-*Calluna vulgaris* shrub cover and lichen cover in *Calluna*–*Cladonia* heathland over 5 years of treatment. Significant treatment effects are shown in bold

Factor lichen cover	d.f.	P-value				
		Total species richness	Lichen species richness	<i>C. vulgaris</i> cover	Non- <i>C. vulgaris</i> shrub cover	Total
Burning	1	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001
Nitrogen	3	0.013	< 0.001	0.521	0.435	0.187
Clipping	1	0.689	0.403	0.568	0.650	0.512
Burn \times nitrogen	3	0.015	0.048	0.616	0.365	0.361
Time	4	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001
Burn \times time	4	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001
Clip \times time	4	0.029	0.045	0.848	0.828	0.668
Nitrogen \times time	12	0.573	0.059	< 0.001	0.414	0.491

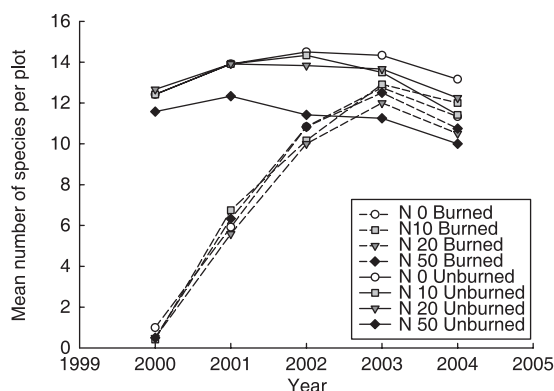


Fig. 1. Effect of four levels of nitrogen addition (0, 10, 20 and 50 kg/ha year⁻¹) in combination with burning on the change in mean species richness through time in *Calluna-Cladonia* heathland. For clarity, the effect of the (non-significant) clipping treatment is not shown and means are for clipped and unclipped plots combined.

May 2000 (6 months after treatment). However, species were quick to recolonize the plots and by 2003 (3.5 years after treatment) there was no significant difference ($P > 0.05$) in the number of species per plot between burned and unburned treatments. Nitrogen addition also had a significant effect on total species richness (Table 1 and Fig. 1). A significant decline in species richness was first detected in 2002 (after two seasons of nitrogen addition) in the 50 kg N ha⁻¹ year⁻¹ treatment compared with the 0 kg N ha⁻¹ year⁻¹ treatment (means of 11.12 and 12.66 species plot⁻¹, respectively; $P < 0.05$). There was also some evidence of a downward trend in species richness in the 20 kg N ha⁻¹ year⁻¹ treatment. Burning and nitrogen-addition treatments exhibited a significant interaction (Table 1). In unburned plots, species richness was already significantly reduced in the 50 kg N ha⁻¹ year⁻¹ treatment in 2001 (mean 12.33 species) compared with the other three treatments (all mean of 13.91 species) after only one season of nitrogen addition, although the change was small. Species richness continued to decline over the following 3 years in the nitrogen-treated plots (Fig. 1). By 2004, after 4 years of nitrogen addition, species richness was significantly reduced ($P < 0.05$) in both the 50 and 10 kg N ha⁻¹ year⁻¹ treatments (means of 10.00 and 11.41, respectively) compared with the 0 kg N ha⁻¹ year⁻¹ treatment (mean 13.16), while richness in the 20 kg N ha⁻¹ year⁻¹ treatment (mean 12.25) was also reduced, although not significantly so. In burned plots there was no significant difference in species richness between the nitrogen treatments after 4 years of addition, although there was a downward trend in the 20 and 50 kg N ha⁻¹ year⁻¹ treatments.

The effect of the treatments on species richness was also analysed for higher plants and lichens separately. For higher plants only burning had a significant effect on species richness ($F_{1,5} = 21.87$, $P = 0.005$). Lichen species richness showed the same pattern as described for the whole data set above (Fig. 2 and Table 1), with a significant reduction ($P < 0.05$) in diversity observed

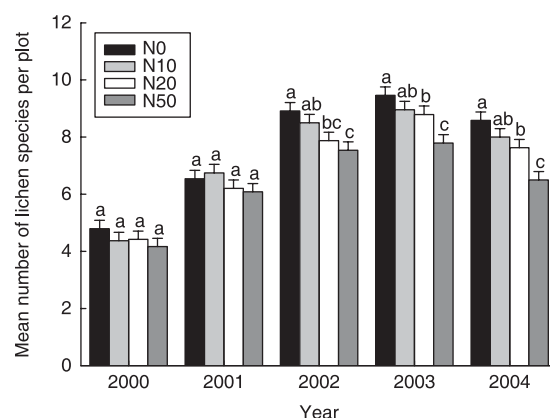


Fig. 2. Effect of nitrogen-addition treatments (0, 10, 20 and 50 kg/ha year⁻¹) on mean lichen species richness. Means within years not sharing the same letter are significantly different ($P < 0.05$); error bars show the standard error of the mean.

in unburned 10 kg N ha⁻¹ year⁻¹ plots after 4 years (mean of 7.5 species) compared with control plots (mean of 9.25 species). No significant effect was seen in burned plots. Thus it was apparent that it was the lichen component of this community that drove the diversity responses to nitrogen addition.

SPECIES COMPOSITION

PRC analysis of the cover data revealed two significant axes (Fig. 3a,b and Table 2). The strength of the relationship of individual species to these axes can be seen in the plots of species weights. For clarity only those species with a weight greater than ± 0.25 are shown. An approximation of individual species' responses to treatments was obtained by taking the exponent of the species' weight multiplied by the regression coefficient of the treatment. It can be seen that axis 1 (Fig. 3a) was primarily associated with the effect of the burning treatment and there was no obvious separation of nitrogen treatments. Burned plots had a reduced cover of the dwarf shrubs *Calluna vulgaris*, *Arctostaphylos uva-ursi* and *Empetrum nigrum* and also of the main lichen species, including *Cladonia arbuscula*, *Cladonia furcata*, *Ochrolechia frigida*, *Cetraria islandica* and *Cetraria aculeata*. Bare ground cover and, to a lesser extent, cover of *Cladonia* basal squamules was increased by burning. The burned plots showed a consistent trajectory back towards the composition of the unburned plots, although this was much slower than seen in the species richness data (Fig. 1). Extrapolation of the trajectory of the burned plots on this axis suggested that they would reach the composition of the unburned plots in 2006, i.e. 6.5 years after the burning treatment was imposed.

Axis 2 (Fig. 3b) also showed a clear separation of the burned and unburned treatments. However, on this axis the burned treatments showed a more rapid return towards the composition of the control treatment, although the data appeared to suggest that they had

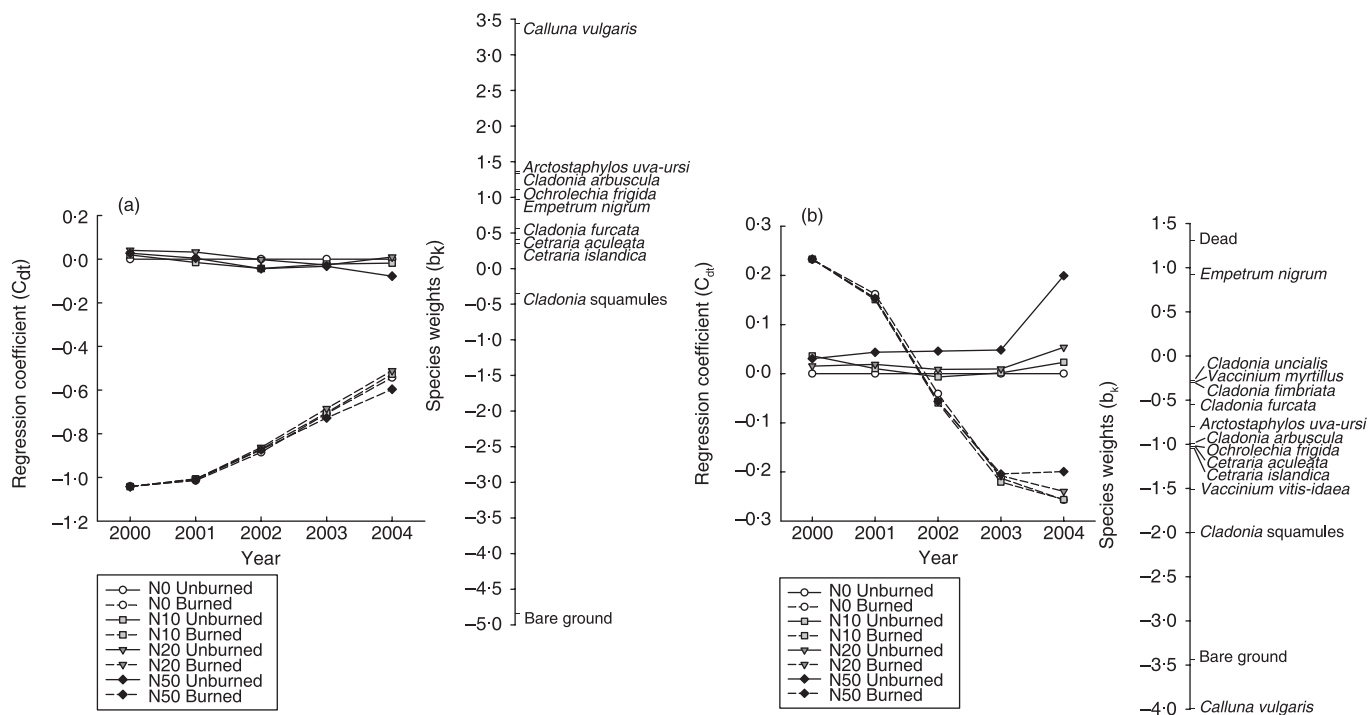


Fig. 3. Principle response curve (PRC) analysis of changes in community composition during 5 years of treatment application relative to a control treatment ($0 \text{ kg N ha}^{-1} \text{ year}^{-1}$, Unburned). See Table 2 for details of axis properties and results of significance tests. (a) PRC axis 1; (b) PRC axis 2.

Table 2. Properties of PRC axes 1 and 2 and results of Monte Carlo significance tests for each axis (2000 permutations)

Parameter	Axis 1	Axis 2
Eigenvalue	0.625	0.039
% variance in species data explained	68.6	4.3
F-ratio	959.632	70.134
P-value	< 0.001	< 0.001

overshot the control composition. This axis also showed that, after 4 years of treatment, during which time there had been very little change in composition relative to the controls on either axis 1 or 2, the unburned plots receiving $50 \text{ kg N ha}^{-1} \text{ year}^{-1}$ began to move away from the other unburned treatments. This change was associated with increased cover of *Empetrum nigrum* and dead *Calluna vulgaris*, while cover of the other dwarf shrubs, lichens and bare ground decreased. In early October 2002 there was a significant fall of snow that covered the plots to a depth of around 50 cm and that was earlier than is typical in this area. In the following May browning of *Calluna vulgaris* shoots was observed and the percentage cover of browned *Calluna vulgaris* on each plot was recorded; other shrub species appeared unaffected. The percentage cover of the damaged *Calluna vulgaris* was seen to be significantly related to nitrogen treatment (Fig. 4), with much greater cover of damaged shoots on the plots that received the $50 \text{ kg N ha}^{-1} \text{ year}^{-1}$ treatment. This interaction between the severe weather event and the nitrogen treatment resulted in the move towards greater dead *Calluna vulgaris* cover in $50 \text{ kg N ha}^{-1} \text{ year}^{-1}$ plots in 2004.

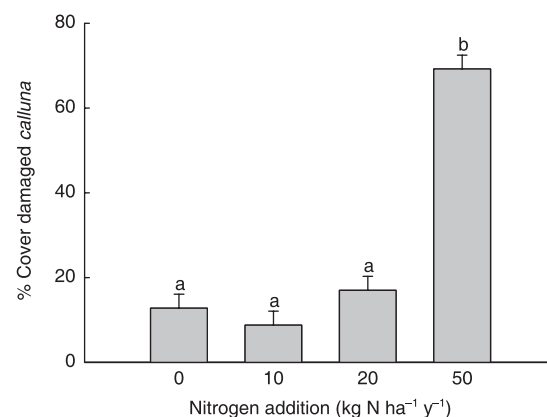


Fig. 4. The effect of nitrogen addition ($0, 10, 20$ and $50 \text{ kg/ha year}^{-1}$) on mean cover of damaged *Calluna* following early snowfall. Letters indicate means that are significantly different ($P < 0.05$) and error bars show standard error of the mean.

Cover changes over the 5 years were also investigated using repeated-measures ANOVA for three main species groups, *Calluna vulgaris*, total dwarf shrubs excluding *Calluna vulgaris* and total lichen cover. The results of these analyses corresponded with the patterns seen in the PRC analysis, with a highly significant effect of burning, time and burning–time interaction on the cover of all three species groups (Table 1). A highly significant nitrogen–time interaction was found for *Calluna vulgaris* cover, with a reduction in cover occurring in the 50 kg N plots in 2004 (Fig. 5a), although, interestingly, there was no compensatory expansion of the other dwarf shrub species (Fig. 5b). Total lichen cover also showed a significant reduction at the highest

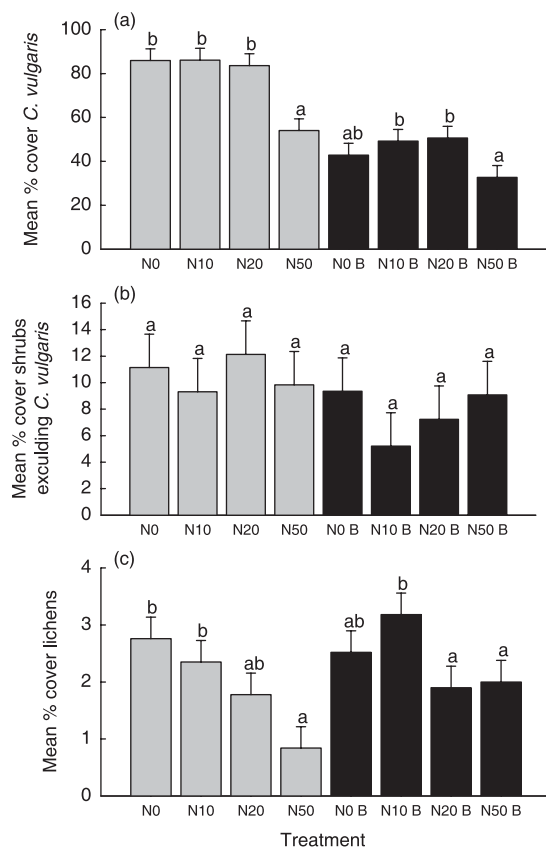


Fig. 5. Effect of burning and nitrogen addition (0, 10, 20 and 50 kg/ha year⁻¹) on the mean cover of (a) *Calluna vulgaris*, (b) dwarf shrubs excluding *Calluna vulgaris*, and (c) lichens in 2004 after 5 years of treatment. Grey bars, unburned; black bars, burned. Within burned/unburned groups, means not sharing the same letter are significantly different ($P < 0.05$); error bars show the standard error of the mean.

Table 3. ANOVA probabilities for the significance of main effects of burning, nitrogen addition and clipping on total lichen cover in *Calluna*–*Cladonia* heathland for each year from 2000 to 2004. Significant treatment effects are shown in bold

Factor	d.f.	P-value				
		2000	2001	2002	2003	2004
Burning	1	< 0.001	< 0.001	< 0.001	0.020	0.407
Nitrogen	3	0.896	0.436	0.037	0.005	0.001
Clipping	1	0.542	0.950	0.881	0.161	0.101

level of nitrogen addition by 2004 (Fig. 5c and Table 3), most notably in the unburned plots. When lichen cover was analysed year by year (Table 3), the effect of the burning treatment gradually declined through time, becoming non-significant by 2004, while a significant effect of the nitrogen treatment first appeared in the third year and became stronger with time. The response of total lichen cover was, however, dominated by the most abundant species. Analysis of individual species cover after 5 years (Table 4) showed that lichen species varied in their response to the nitrogen treatment, with *Cladonia furcata* and *Cladonia 'arbuscula'* showing a significant effect, while *Cetraria islandica* and *Ochrolechia*

Table 4. Variability in individual lichen species' response to treatments. ANOVA probabilities for significant main effects of burning, nitrogen addition and clipping in 2004 after 5 years of treatment. Significant treatment effects are shown in bold

Factor	d.f.	P-value			
		<i>Cladonia 'arbuscula'</i>	<i>Cladonia furcata</i>	<i>Cetraria islandica</i>	<i>Ochrolechia frigida</i>
Burning	1	0.085	0.069	0.152	0.090
Nitrogen	3	0.024	0.016	0.793	0.398
Clipping	1	0.819	0.061	0.736	0.822

frigida did not. Only a limited number of species was analysed in this way, however, as the majority of species were too sparsely distributed.

Discussion

RESPONSE TO INDIVIDUAL TREATMENTS

Nitrogen deposition on lowland *Calluna vulgaris*-dominated communities is commonly associated with increased graminoid abundance, becoming dominant over dwarf shrubs in the long term (Heil & Diemont 1983; Aerts & Heil 1993; Bobbink, Hornung & Roelofs 1998). The graminoid component of the vegetation at this alpine site was initially very small, although several species were present as isolated individuals, and no evidence of an expansion was seen. This result is similar to that of Power, Ashmore & Cousins (1998), who saw no grass invasion of *Calluna vulgaris*-dominated lowland heathland after 7 years of nitrogen addition; their plots, however, were undisturbed. Transition from shrub to graminoid dominance is generally associated with disturbance, such as heather beetle *Lochmaea suturalis* attacks opening the dwarf shrub canopy (Berdowski & Zeilinga 1987). In alpine heathland the dense mat of dwarf shrubs and lichens covering the ground may reduce the availability of gaps for establishment and, in combination with a low seed input, limit the potential for graminoid expansion. Burned plots, however, had a large proportion of bare ground for several years but still did not see a significant increase in graminoid cover; thus it seems more likely that seed availability is the limiting factor. Species such as *Deschampsia flexuosa* do not form a seed bank (Grime, Hodgson & Hunt 1990) and this would give the dominant dwarf shrubs, with a large long-lived seed bank and/or the ability to spread rapidly through vegetative means, a competitive advantage during regeneration. When nitrogen was added to similar alpine lichen–dwarf shrub vegetation in Norway over a 10-year period (Fremstad, Paal & Möls 2005), *Festuca ovina* was the only higher plant species that responded, showing a slight increase in cover. The initial cover of *Festuca ovina* was higher in that study, however, so the potential for vegetative expansion may have been greater.

While the higher plant component of the vegetation appeared resilient to change and showed only small responses to the nitrogen treatments, the lower plant component of the community was more sensitive to the effects of nitrogen deposition. Lichen cover declined on nitrogen-treated plots, a response that was also reported in the Norwegian study (Fremstad, Paal & Möls 2005) and in studies of heathlands and arctic tundra vegetation (Bobbink, Hornung & Roelofs 1998; Lee & Caporn 1998; Press *et al.* 1998; Gordon, Wynn & Woodin 2001). As there was no obvious change in vegetation structure that might have reduced light availability to lichens or increased competition with higher plants, it seems likely that the reduction was a direct effect of the nitrogen deposition. Lichens and mosses are extremely efficient at retaining applied nitrogen (Curtis *et al.* 2005), and reductions in *Cladonia* growth in response to nitrogen exposure have previously been reported in heathland habitats (Vagts & Kinder 1999). Species reacted individually to the treatments, with *Cladonia furcata* and *Cladonia 'arbuscula'* showing a significant response to the nitrogen treatments while *Ochrolechia frigida* and *Cetraria islandica* did not. Lichen species richness was also reduced and was the main driver of reductions in total community species richness. This reduction in species richness in response to low levels of nitrogen addition appears to be a general response and has been observed in a wide range of communities (Gough *et al.* 2000). This suggests that lichen diversity could be used as an effective indicator of the impacts of nitrogen deposition in alpine lichen-rich communities. What cannot be determined from this study is whether the observed effects of the nitrogen treatments are a result of the total nitrogen load or the concentration of the treatments. Because of practical constraints on the frequency and volume of nitrogen treatment applications, most studies of this type, including this one, expose the vegetation to much higher ion concentrations than would typically be found in rainfall or cloud water. Further work is needed to separate these two effects and to improve understanding of what drives the effects of nitrogen deposition on both higher and lower plants.

The burning treatment had the greatest effect on species richness and composition of the vegetation in the short term. The recovery of the vegetation following this treatment, however, was relatively rapid. Although burning caused a large drop in species richness at the beginning of the experiment, species were quick to recolonize the plots, and species richness had recovered to unburned levels within 4 years. Total cover of lichens and shrubs was still below unburned levels in 2004, but looked likely to recover fully within 7 years if the current trajectory was continued. Species composition data indicated that species from the original species pool were recolonizing, and little invasion of new species occurred despite a high cover of bare ground within the plots for several years. Lichen species quickly recolonized, possibly because of their ability to disperse by wind-blown fragments, but as they are generally

slow growing it is likely that lichen cover will take longer to recover. This relatively direct return towards the initial species composition is in contrast to *Calluna* heaths at lower altitude, where temporary increases of *Vaccinium* species or graminoids often follow management fires (Gimingham 1972). However, such management fires would generally be more intense than burns in the alpine zone, as a result of the higher fuel load present in lower altitude heaths, giving other species the opportunity to establish while the slow-growing *Calluna vulgaris* regenerates.

Although sufficiently large that the majority of the regeneration of higher plants following the fire was from seed rather than from lateral expansion of surrounding individuals (A. Britton, personal observation), the small plot size (1.125 m²) used here may also have contributed to the rapid return of the vegetation to its original composition. Small plots are more sheltered and less exposed to the effects of winter frost heave, which is the predominant cause of winter mortality in seedlings at this altitude (Miller & Cummins 1987), thus allowing vegetation to re-establish more quickly. Where larger areas of alpine heath have been burned, frost heave and exposure may reduce or delay dwarf shrub regeneration and make initiation of erosion likely (Moorland Working Group 1998). While the nature of the experimental site precluded using larger plots, this interaction between plot size, dispersal and physical disturbance, along with fire intensity, must be taken into account when applying results at larger scales to ensure that potential effects are not underestimated (Strengbom, Englund & Ericson 2006).

Although the clipping rate of 12% of the current year growth removed that was applied in this experiment is approximately double the amount of biomass removed by grazing on low-alpine heaths in the Scottish Highlands (A. Britton, unpublished data), *Calluna vulgaris* withstood this amount of offtake without obvious damage and vegetation diversity was unaffected. This treatment does not completely simulate the selective effects of grazing herbivores, or the effects of trampling and nutrient deposition which result from their presence, but it does show that the vegetation can withstand a significant level of biomass removal without damage. Effects of trampling and nutrient deposition by grazers may therefore be more important in causing the reductions in alpine dwarf shrub heath extent in areas of high grazing pressure.

INTERACTIONS BETWEEN TREATMENTS AND BETWEEN TREATMENTS AND CLIMATE

The strong interaction seen in this study between burning and nitrogen addition, in terms of effects on community composition, has previously been documented for higher plant species in lower altitude heathland communities subject to a variety of nitrogen-removing management treatments (Diemont 1994; Britton *et al.* 2001; Barker *et al.* 2004). However, for lichens, which

grow on the soil substrate but do not obtain their nutrients from it, the mechanism for this effect is not clear. In unburned plots, reductions in lichen species richness were seen with nitrogen additions as low as $10 \text{ kg N ha}^{-1} \text{ year}^{-1}$, while only non-significant trends towards a reduction were seen in burned plots. Further studies are needed to investigate the reason for this difference.

The severe winter browning and subsequent death of *Calluna vulgaris* in the $50 \text{ kg N ha}^{-1} \text{ year}^{-1}$ plots, which occurred during winter 2002–03 appeared to have the potential to cause significant changes in species composition, which until then had shown little response to nitrogen treatments. The effect of exposure to pollutant nitrogen on dwarf shrub frost and drought tolerance has been the subject of a number of studies (Bobbink, Hornung & Roelofs 1998). However, results have varied. Where the effect of nitrogen on frost tolerance has been measured directly, it has been shown variously to increase tolerance (Caporn, Risager & Lee 1994; Taulavuori *et al.* 1997, 2001), decrease tolerance (Power *et al.* 1998; Caporn, Ashenden & Lee 2000 found a reduction in hardening in spring) or have no effect (Sæbø *et al.* 2001; Power *et al.* 1998 found no effect in early winter). Effects on drought tolerance appear to be more consistent, with most authors finding an increased sensitivity to drought (including winter drought as a result of soil freezing) in plants exposed to elevated nitrogen levels (van der Eerden *et al.* 1991; Fangmeier *et al.* 1994; Lee & Caporn 1998; Gordon *et al.* 1999). Fangmeier *et al.* (1994) have suggested that this increased susceptibility to drought is a result of increased carbon demand in plants exposed to high levels of nitrogen leading to increased stomatal opening and hence increased water loss. Reductions in root : shoot ratios are another potential explanation (van der Eerden *et al.* 1991). Whether the result of reduced freezing tolerance or increased susceptibility to drought, increased winter browning in *Calluna vulgaris* exposed to elevated nitrogen deposition has been reported from both experimental studies and in field situations (e.g. Dutch heathlands; Bobbink, Hornung & Roelofs 1998). In the case of alpine *Calluna* heathlands, which experience particularly severe winter conditions and potentially high rates of evapotranspiration as a result of high mean wind speeds, the interaction between nitrogen deposition and drought and frost tolerance has the potential to be a critical one for determining biodiversity responses to nitrogen deposition.

IMPLICATIONS FOR CRITICAL LOAD ESTIMATION

The responses of vegetation diversity to the levels of nitrogen deposition used in this study support a critical load for low-alpine *Calluna* heaths in the range $5\text{--}15 \text{ kg N ha}^{-1} \text{ year}^{-1}$, which has been proposed for arctic and alpine heaths (Achermann & Bobbink 2003). The lowest nitrogen addition level of $10 \text{ kg N ha}^{-1} \text{ year}^{-1}$

used here corresponds to a total of 20 kg when background deposition is also taken into account. Significant changes in diversity were seen at nitrogen addition levels down to this level, suggesting a lower critical load. Responses were rapid, with all treatment levels showing responses within 4 years. Measurements of soil and vegetation nitrogen and phosphorus contents suggest that plant growth is currently nitrogen limited at this site and soil phosphorus content is relatively high (data not shown), therefore this site should be representative of the most sensitive end of the montane heath spectrum.

CONCLUSION

This study has shown that the biodiversity of low-alpine *Calluna*–*Cladonia* heathlands may be reduced by even low rates of nitrogen deposition. Effects of nitrogen deposition on species richness were manifest mainly in the lichen component of the vegetation, and occurred quickly (within 1 year at the highest deposition rate but also within 4 years at low deposition rates). The value of this habitat is in large part because of the diversity of lichen species that it supports, and the results of this study and similar work in Norway (Fremstad, Paal & Möls 2005) have shown that lichens are the most sensitive component of the community. We would therefore suggest that the monitoring of impacts should focus on lichen biodiversity. Effective conservation of these low-alpine heaths will require action at national and international levels to reduce emissions and deposition of nitrogen as critical loads are currently exceeded in many areas.

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