Changes to ecosystem properties brought about by changing plant communities: impact of *Pteridium aquilinum*-control/heathland restoration treatments on soil properties

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Introduction

It is well known that soils can be influenced by the plant species that grow in then and if this is so then it can be predicted that where a dominant plant species invades and dominates a given place for a long-time it will change the soil properties (Jenny 1980). We can also speculate that once this species is removed it will either have effected a permanent change to the soil properties or there will be a reversal of the changes enforced during that species' occupancy. Essentially, this might be considered as part of autogenic change or within the concept behind the facilitation model of ecological succession (Connell & Slatyer, 1977). The obvious place to consider such species-induced impacts on soils and their potential reversal is where a weed species invades and dominates the ecosystem for a long-period and then is controlled by human action. The impact of management on such ecosystem processes was implicit in the reworking of Jenny's word model (Marrs & Bradshaw, 1993).

Here, we test the effects of changing the species complement of an ecosystem on selected soil properties within an experimental model system. The test site was an area of land which had been colonized by bracken (*Pteridium aquilinum* (L. Kuhn), a fern species that has a worldwide distribution (Marrs & Watt, 2006). In many parts of the world, and certainly within Great Britain, bracken is considered a serious weed problem for agriculture, forestry and nature conservation (Varvarigos & Lawton, 1991; Pakeman & & Marrs, 1992). Indeed, there has been a series of policies implemented within agri-environment schemes in Great Britain to reduce bracken infestation and to create earlier-successional communities, usually heath, moorland or acid-grassland

The experiment started in 1993 and at that time the site was covered in dense bracken fronds and had a deep litter layer (ca, 30 cm, Ghorbani *et al.*,2006); furthermore, the underlying vegetation was extremely species poor "consisting of a few scattered *Calluna* bushes and little else” (Le Duc *et al*, 2007). As bracken was originally a woodland species, reproduces rarely by spores and there is evidence that clones can exist on a given site for millennia, it is possible that bracken has been present in this area since woodland clearance, almost certainly for centuries, and known historical evidence indicates at least 100 years (N Taylor, pers.comm.). Therefore, the starting position were soils that had had a dense bracken cover for a considerable period. In 1993, a large-scale experiment was started designed to reduce the bracken infestation and develop a plant community with a greater conservation interest (Le Duc *et al*., 2000, 2003, 2007; Alday *et al*., 2013a,b). The treatments applied have been maintained for a twenty-year period and by 2003 there was evidence of alternative states being produced in some of the treatments (Alday *et al*., 2013a). This experiment, therefore, is an ideal model system for testing whether management-induced changes in soil properties have occurred and their magnitude.

We hypothesised that (1) bracken control treatment and associated changes in vegetation enforced by vegetation restoration would change in some soil properties; (2) that such change would include reductions in total soil carbon and nitrogen and increases in soil pH and available nutrients and soil biological activity, and (3) any change would be related to the severity of treatment inpact. These hypotheses were tested by assessing for treatment effects within the formal experiment using general mixed-effects modelling and testing for significance using Bayesian inference.

Methods

The experiment is located at Hordron Edge site in the Peak District, Derbyshire (Latitude and Longitude; 53°23’N, 1°41’W) within the North Peak Environmentally Sensitive Area

(ESA). The site is grazed by sheep at a low stocking density, which is determined by the ESA agri-environment scheme prescriptions (ca. 0.5 sheep ha^{-1}; Pakeman et al., 2000). Experimental treatments were first applied in 1993 and treatments were applied continuously until 2012 when they stopped. The vegetation and soil change reported here, therefore, assesses change in soil properties with respect to treatment during this 20-year period. Full methodological details are available in Le Duc et al. (2003).

The experiment used a randomized block, split-split-plot design; there are three blocks (10x40m) with the main plots (10 x 36m) initially (1993-2003) receiving one of six bracken control treatments (no treatment (the experimental control), cut once per year, cut twice per year and three treatments including the herbicide asulam (applied by knapsack at 4.4 kg ai ha^{-1}, 11 litres Asulox in 400 litres water ha^{-1}; Manufacturers, Bayer CropScience PLC and United Phosphorus PLC). The three herbicide treatments were: (1) a single spray treatment in 1993; (2) a single spray treatment in 1993 followed by a single cut in 1994, and (3) a single cut in 1993 followed by a single spray treatment in 1994. Vegetation restoration treatments were then applied to sub-plots (10 x 18 m) and sub-sub-plots (10 x 5 m). The sub-plot treatments tested sheep grazing *versus* no sheep grazing (fenced *versus* unfenced) and the sub-sub-plot treatments received one of three *Calluna* seeding treatments (no seeding, seed applied in brash, and seed applied in litter). All treatments were applied randomly within each experimental stratum, with 108 sub-sub-plots.

In 2004, it was apparent that *P. aquilinum* was recovering in all treatments where asulam had been applied (Cox et al., 2007) and these treatments were reverting to a vegetation similar to the untreated controls in the three main-plot treatments where asulam had been applied, and there was no evidence of the creation of a new alternative stable state (Alday et al., 2012, 2013). Accordingly all three original treatments which included asulam were resprayed in August 2004, and followed-up annually until 2012 with an annual spot-spray of asulam delivered to all emergent fronds individually (1ml solution of 6% vol:vol Asulox and water by knapsack sprayer). For clarity, the original treatment codes are retained here.

4.2. Assessing trends in vegetation

In 1993 species composition was measured in selected random sub-plots to ensure that the vegetation and bracken variables were similar at the start. The vegetation composition was monitored in all experiments in June (i.e. before the application of the first cutting treatment) from 1994 to 2007 and 2013. Quadrats (1 m ×1 m) were placed at pre-selected random co-ordinates on 1 m grids within each sub-sub- plot and the cover of all vascular plant, bryophyte and lichen species recorded visually as well as an estimate of bracken litter cover. Within the central 0.25 m2 of the quadrats all fronds were counted and their length measured; mean frond length and frond density (number m^{-2}) were then calculated. Species nomenclature follows Stace (2010) for vascular plants, Atherton et al. (2010) for bryophytes and Dobson (2011) for lichens.

4.3. Assessing the effects of treatment on soil chemistry

In early June 2013, pre-selected, random positions were located in each of the 108 sub-sub-plots. At each location, the surface vegetation was removed from a small area and a surface soil core removed using an Eijkelkamp auger (7 cm diameter, 20 cm depth). Initially the fresh weight of the entire core was measured to estimate bulk density. Thereafter the core was split into two; one was used for analyses carried out on fresh soils, the other was dried and sieved to pass a 2 mm mesh for analyses on dried soils. Soil pH and measures of soil available N nitrogen (NH_4-N and NO_3-N) and P were assessed on fresh soils; soil exchangeable K, Ca, Mg and total N and C were measured on dried soils. Total C and N was measured using a CE Instruments NC2500 Elemental Analyser machine, available N was extracted in 2 M KCL, and both exchangeable cations and available P were extracted in 2.5% vol:vol acetic acid. Analytical methods followed Allen (1989).

At the same time, soil respiration was measured in both of the grazing treatments (grazed *versus* ungrazed) in three of the main-plot treatments (untreated, cut twice per year, asulam). To avoid complications with the sub-sub-treatments only the un-seeded sub-sub-plots were sampled. In total 18 sub-sub-plots were sampled (3 Blocks x 3 main-plot bracken control treatments x 2 sub-plot grazing treatments). On early July 2013 soil respiration was measured using an LCi-portable photosynthesis system (ADC Bio scientific LCi-SD System Serial No.33526). The surface vegetation was removed and collars (enclosed soil volume is 97.5cm^2) were inserted into the ground (Appendix 1, page 24). The soil respiration hood was then placed on top of the collar and CO_2 efflux measured over a 20 minute period using a flow rate of 200 µmols s^{-1}. Within blocks, individual sampling was carried out randomly through the day to minimize any diurnal effects.

In early July 2014, soil samples were collected in a similar manner as above in both of the grazing treatments (grazed *versus* ungrazed) in three of the main-plot treatments (untreated, cut twice per year, asulam). Soil nitrogen mineralization rates were determined using the method outlined in Allen (1989) before and after 21 days incubation at 25°C.

4.4. Statistical analysis

All statistical analyses were performed in **R** 3.1.1 (R Development Core Team 2014). As the aim here was to describe the broad vegetation trends that had occurred in the different treatments over time only bracken variables (mean frond length, frond density, bracken cover, litter cover) and aggregated vegetation variables (species richness, Shannon-weiner diversity, and cover of graminoids and dwarf-shrubs) are considered here. The graminoid group included grasses, sedges and rushes and the dwarf shrubs included *Calluna vulgaris*, *Erica tetralix*, *Vaccinium myrtillus*, *V. oxycoccus* and *V. vitis-idaea*). Two analyses were performed: (1) calculation of means and standard errors for each variable were compared for the vegetation at the start of the experiment (1993) and the year after treatment stopped (2013). Second, a description of the changing response of each variable was assessed using Generalised Additive Modelling (GAM) performed using the 'mgcv' package. Only a selection of the outputs are shown, and for the GAMs only one of the asulam-treatments are presented as they all showed similar responses. Full results are available from the senior author.

The responses of each of N mineralisation rate, soil nitrification rate, soil respiration rate, pH, C:N ratio, soil moisture, total N, total C, extractable P, total NH_4 and total NO_3 to *Pteridium*-control and grazing treatments were modelled using linear mixed-effects models, with plot and block specified as random effects in all models. This allowed for any plot- and/or block-level effects resulting from the split-split plot experimental design to be accounted for in the analyses. (Pinheiro & Bstes 2000). All models were implemented in a Bayesian framework using the **MCMCglmm** version 2.16 package (Hadfield 2010). All models incorporated parameter-expanded priors and were run with sampling of every 50th iteration for 1.0×10^6 iterations after a 1.0×10^4 burn-in, resulting in an effective sample size of approximately 2.0×10^4 for each predictor from the posterior distribution. Convergence of all models was assessed through inspection of the trace plots.

3. Results

3.1. Vegetation change over the course of the study

For bracken variables there was little difference in mean frond length at the start of the study and the untreated plots in 2013, although there was a slight increase in the un-grazed plots compared to the grazed ones in 2013 (Fig.1a). Mean frond density and bracken litter cover showed broadly similar patters except there was an increase in 2013 compared to the start. Grazing had little effect on these two variables (Fig. 1b,c). All bracken control treatments had reduced all three bracken variables by 2013; the three treatments involving asulam were most effective, followed by the cut twice yearly and then the cut once yearly treatments (Fig. 1a-c).

Species richness did not change from the start of the experiment and the untreated plots in 2013, but the cover of graminoids and dwarf-shrubs declined (Fig. 1d-f). Species richness and graminoid cover increased where the bracken had been controlled but there were few differences between bracken control treatments in the sheep-grazed treatments; removal of grazing brough about a reduction in species richness and graminoid cover (Fig. 1d,e). Dwarf-Fig. 1d) showed a more idiosyncratic response being greater in the Cut then Spray treatment than the others but variability was also high (Fig. 1e).

The modelled time courses are shown for three variables are shown; frond density to describe changes in the living summer bracken biomass (Fig. 2), bracken litter to describe changes in year-round bracken necromass left on site (Fig. 3) and graminoid cover to describe change in the restored vegetation (Fig. 4). The r^2 values and % deviance explained for these fitted models were 0.33-0.71 and 35%-76% for frond density, 0.37-0.87 and 44%-89% for bracken litter and 0.10-0.60 and 16%-64%.

The bracken frond density showed large fluctuations throughout the study period in the untreated plots; the grazed plots showed an increasing tres with tiem and the ungrazed plots were more stable. The cut twice per year plots showed the fasted decline and this decline was faster in the ungrazed treatment. The cut once yearly treatment also showed a similar decline but there was a marked increase in frond density in the early years with large fluctuations. The asulam treatment showed an initial good reduction in frond density but this was followed by gradual recovery until the treatments were repeated when there was good reduction in frond density with the repeated treatments. The bracken litter cover responses were similar, increasing cover in the untreated plots, fastest reductions in the cut twice yearly plot, a slower reduction the in cut once yearly treatments but a much slower reduction in litter cover after the initial asulam treatment, with recovery until the treatments were applied continuously after which the bracken cover declined. The graminoid cover remained throughout in the untreated plots but increased in all plots where bracken was controlled. The largest and fastest increases were in the sheep-grazed cut once and twice yearly treatments, the asulam-treated plots were slower, and cover was smaller.

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3.2. Effects of treatment on soil properties

The results of the statistical analyses are presented in Table 2, and for each soil chemistry variable an interaction graph is presented with the untreated grazed plot as the model intercept. Significance of treatment effects is assessed in comparison with this treatment, whilst significance of grazing effects is an additional effect.

All *Pteridium*-treatments were found to significantly increase soil pH from <4 in the untreated control to a maximum of ~4.6 in the twice-yearly cut treatment, whilst the opposite response was found for soil C:N, with the greatest value of ~17.5 being found in the untreated control compared to values of <17 being found in all other treatments. Removal of grazing had no significant additional effect on soil pH, though a significant further reduction of C:N to ~16 in the cut twice-yearly plots.

Soil moisture was greatest in the untreated control than were any of the *Pteridium*-treatments had been applied, with the two cutting treatments resulting in the greatest reductions. Only the cut twice-yearly plots showed an additional effect of grazing removal, with soil moisture content showing a significant increase over the cut twice-yearly grazed plots.

Soil mineralization and nitrification rates were significantly reduced in the cut twice-yearly and asulam-treated plots when compared to the untreated control. In contrast, soil respiration rates were much greater in the *Pteridium*-treatments than in the control plots; there were no significant additional effects of grazing removal on soil mineralization, nitrification or respiration rates.

Total N was significantly lower under all bracken treatments in the grazed plotts, though there was no additional effect of grazing-removal on the assaultasulam or either of the cutting treatments. However, there was a significant reduction in total N in the untreated plots after grazing-removal, whilst there were significant increases found in the cutasu and asucut plots after grazing removal.

Total C was found to be significantly lower than the control for all bracken treatments in the grazed plots. Removal of grazing pressure resulted in a significant increase in only the asucut treatment, though there was an almost significant reduction found in the untreated plots, with no significant effects found inane of the other treatments.

Extractable P was found to be significantly lower than the untreated control in only the asulam, cut 2x and cut 3x treatments under grazing. There was a significant additional increase in extractable P in the asulam-treated plots with removal of grazing. There were no additional effects of grazing-removal found for any of the other treatments.

There were significant reductions in NH_4 found in the cutasu, cut2x and cut3x treatments when compared to the untreated control under grazing, whilst grazing-removal resulted in an additional significant reduction in NH_4 in only the asulam treatment.

Compared to the untreated control, total available NO_3 was found to be significantly reduced under all bracken-treatments in the grazed plots, whilst the only additional effect of grazing-removal was found in the untreated plots; no additional effects were found for any of the other treatments.

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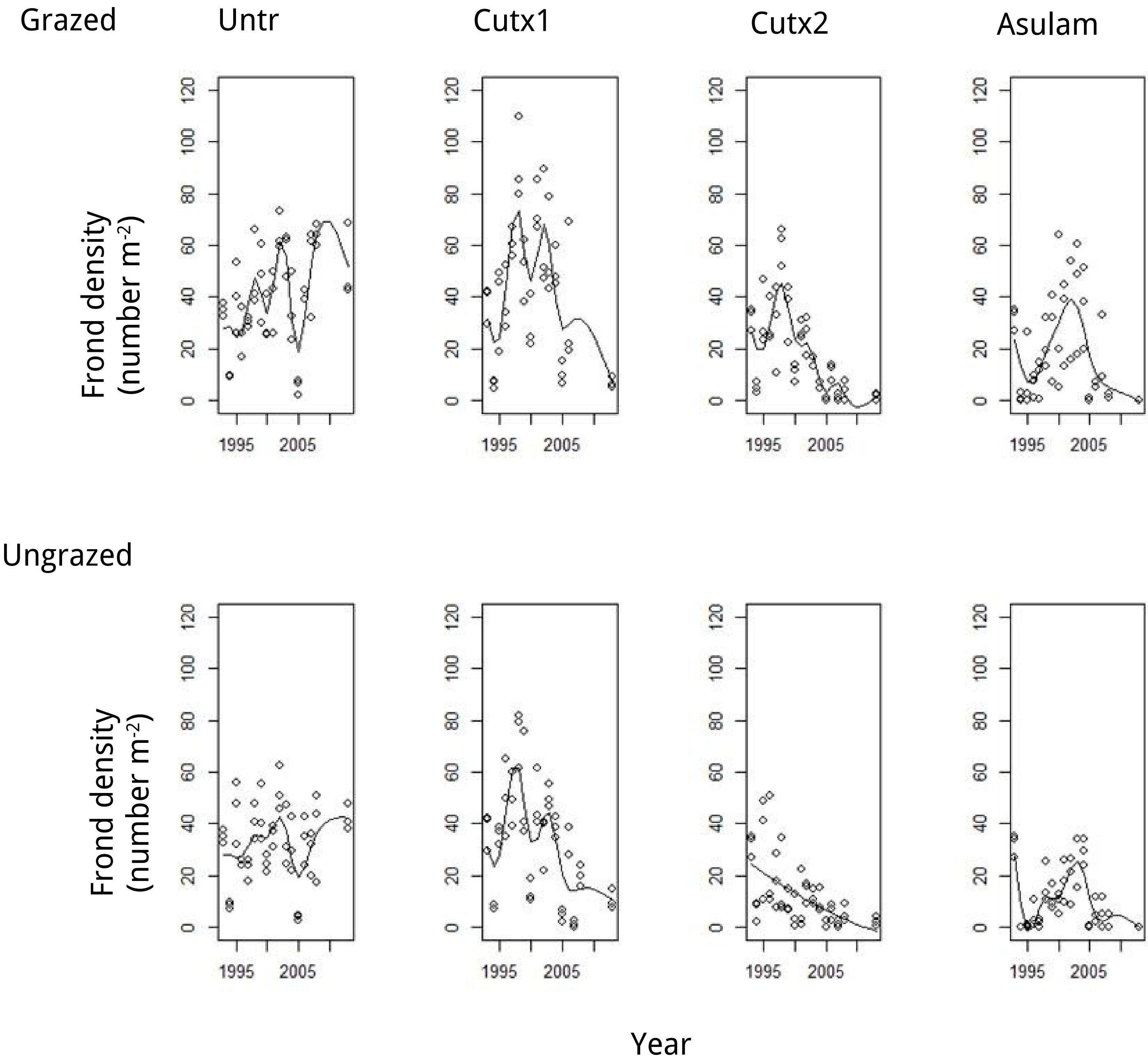
Figures

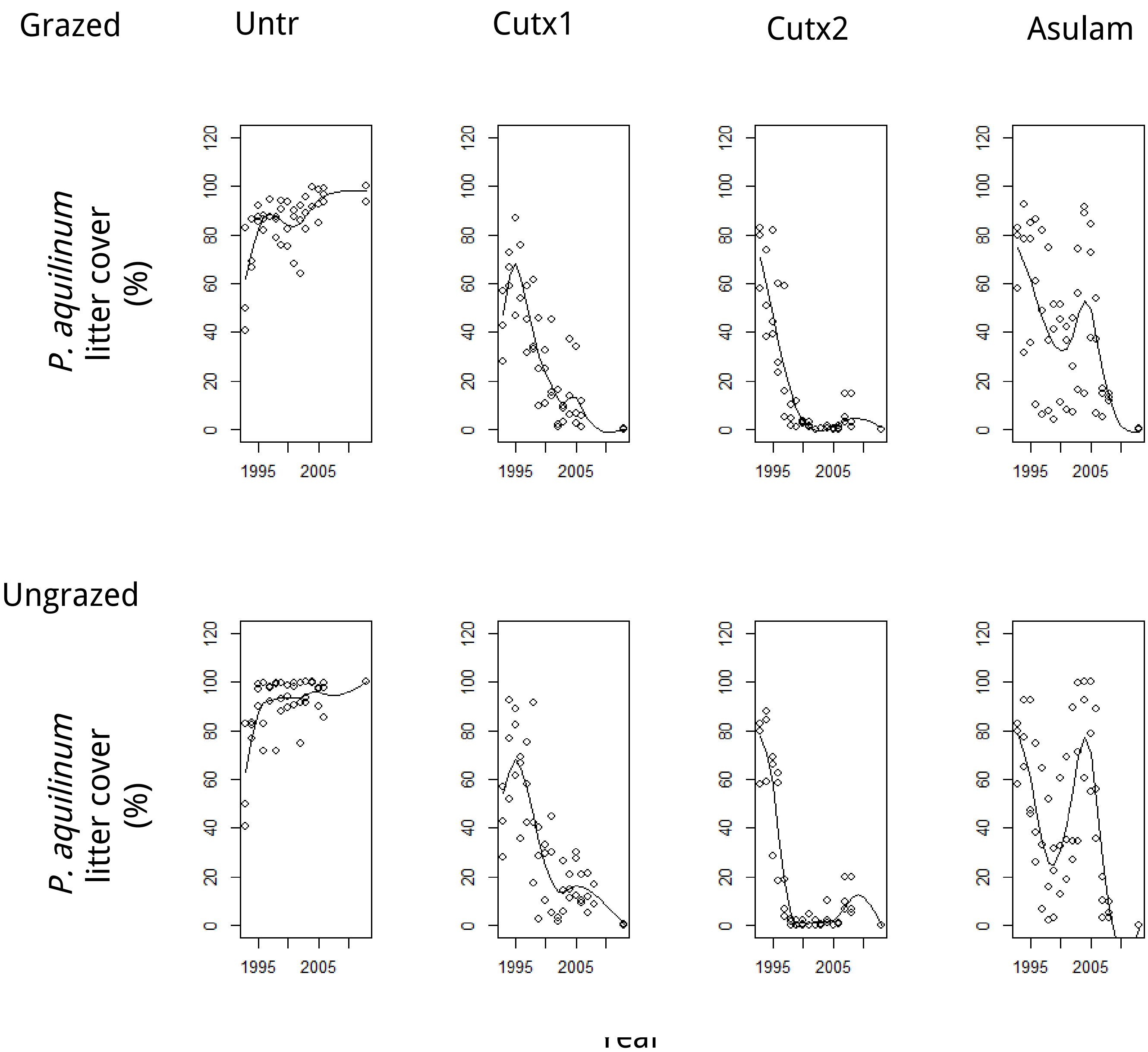
Figure 1. Comparison of some of changes in vegetation between the start of the study in 1993 (pre-treatment) and after the final application of bracken-control and sheep grazing treatments in 2013. (a) Bracken frond length, (b) Bracken frond density, (c) Bracken litter cover, (d) species richness, (e) Graminoid cover, (f) Dwarf-shrub cover. Bracken treatments are coded: Untreated (Untr), Cut once/yr (Cutx1), Cut twice/yr (Cutx2), Asulam (Asulam), Asulam + Cutting (SprCut), Cutting plus Asulam (CutSpr) – note all asulam treatments were retreated with asulam in 2004 and followed up annually with spot-spraying thereafter until 2012; sheep grazing (blank bars), no sheep-grazing (filled bars). Mean values±SE are presented.

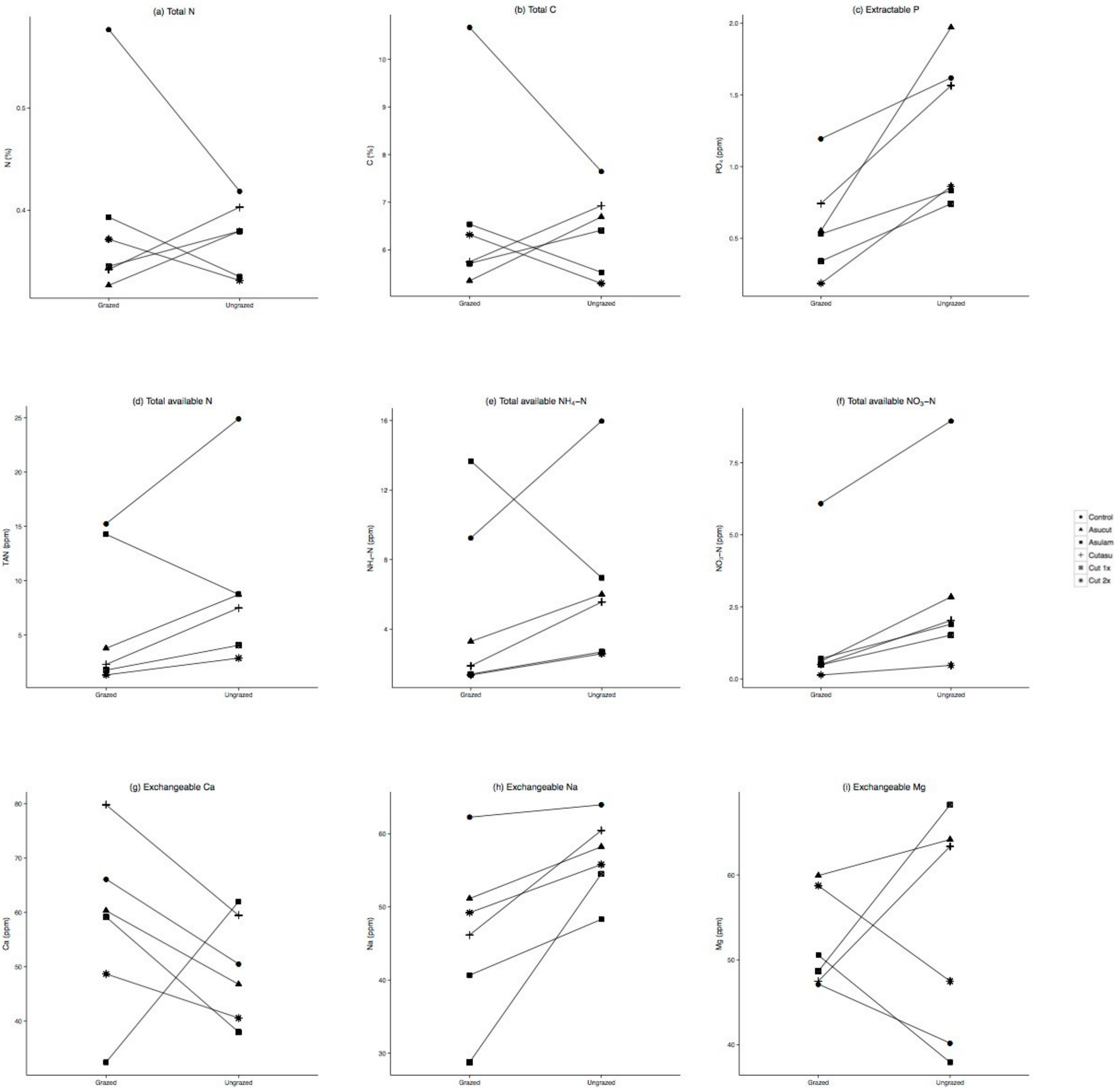
Figure 2. Modelled change in bracken frond density through time (1993-2013) as a result of bracken control and sheep grazing treatments Bracken treatments are coded: Untreated (Untr), Cut once/yr (Cutx1), Cut twice/yr (Cutx2), once in 1993, retreated in 2004 and followed up annually with spot-spraying thereafter until 2012. Raw data are plotted along with the fitted GAM modelled relationship.

Figure 3. Modelled change in bracken litter cover through time (1993-2013) as a result of bracken control and sheep grazing treatments Bracken treatments are coded: Untreated (Untr), Cut once/yr (Cutx1), Cut twice/yr (Cutx2), once in 1993, retreated in 2004 and followed up annually with spot-spraying thereafter until 2012. Raw data are plotted along with the fitted GAM modelled relationship.

Figure 4. Modelled change in graminoid cover through time (1993-2013) as a result of bracken control and sheep grazing treatments Bracken treatments are coded: Untreated (Untr), Cut once/yr (Cutx1), Cut twice/yr (Cutx2), once in 1993, retreated in 2004 and followed up annually with spot-spraying thereafter until 2012. Raw data are plotted along with the fitted GAM modelled relationship.


Grazed
Untr
Cutx1
Cutx2
Asulam
Frond density (number m-2)
0
20
40
60
80
100
120
1995
2005
Ungrazed
Year


Grazed
Untr
Cutx1
Cutx2
Asulam
P. aquilinum litter cover (%)
0
20
40
60
80
100
120
1995
2005
Ungrazed
P. aquilinum litter cover (%)


(a) Total N
N (%)
0.5
0.4
(b) Total C
C (%)
10
9
8
7
6
(c) Extractable P
PO$_4$ (ppm)
2.0
1.5
1.0
0.5
(d) Total available N
TAN (ppm)
25
20
15
10
5
(e) Total available NH$_4$–N
NH$_4$–N (ppm)
16
12
8
4
(f) Total available NO$_3$–N
NO$_3$–N (ppm)
7.5
5.0
2.5
0.0
(g) Exchangeable Ca
Ca (ppm)
80
70
60
50
40
(h) Exchangeable Na
Na (ppm)
60
50
40
30
(i) Exchangeable Mg
Mg (ppm)
60
50
40
Grazed
Ungrazed
Control
Asucut
Asulam
Cutasu
Cut 1x
Cut 2x