

ABSTRACTS

PAPERS

DRIVERS OF CHANGE IN BIODIVERSITY ACROSS A MULTI-HABITAT UK SURVEY.

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This presentation will report on the extensive multi-habitat botanical survey that was carried out in 2009 as part of DEFRA's Terrestrial Umbrella (TU) project. Over 560 vegetation quadrats (2 x 2 m) were surveyed across 112 sites encompassing upland and lowland heathland, M19 bogs and sanddunes. We also revisited 23 of the sites surveyed by Stevens *et al.* (2004) and surveyed 112 plots at the TU experimental sites (Ruabon, Budworth, Thursley, Wardlow, Newborough, Pwllperian). In addition to the vegetation data recorded, soil cores, moss tissue and plant tissue were sampled and extensive laboratory analysis encompassing tissue C, N, P, soil C and N, mineralisation potential, moss chlorophyll fluorescence (Fv/Fm) and PME activity is nearing completion.

A significant pattern of species richness reduction as a function of increasing atmospheric nitrogen deposition was found across all habitats surveyed with a rapid loss of species at low N deposition levels ($r^2=0.36$). The multi-habitat results compare well with earlier data from Stevens *et al.* (2004) and are consistent across all habitats when analysed independently. Losses in both higher plant species and sensitive moss and lichen species were recorded, in contrast with gains in graminoid cover as modelled N deposition increased. For example, in M19 bogs, there was a strong positive relationship between *Eriophorum vaginatum* cover and N deposition ($r^2=0.43$).

A SURVEY OF BIOINDICATORS AS TOOLS FOR INFERRING NITROGEN DEPOSITION DRIVEN CHANGES IN HEATHLAND ECOSYSTEMS

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Defra policy relevance:

- When fully analysed, results from this study will provide field scale evidence of the link between N deposition and plant diversity in heathland ecosystems.
- Evidence of the impacts of N critical load exceedance on biogeochemical cycling and ecosystem functioning will also be provided, scaling up what is known about responses under controlled experimental conditions to the wider, national scale.

Findings from the heathland UKREATE manipulation experiments indicate that certain parameters (e.g. *Calluna* growth responses, lower plant abundance, soil and litter enzyme activity, foliar N content) provide reliable responses to enhanced N deposition. The robustness of these indicators has been investigated within non-experimental, spatial surveys of heathland systems across the UK along an N deposition gradient. Results from earlier, small-scale (unpublished) regional surveys indicated a relationship between N deposition and plant species richness and cover. The current survey investigates this relationship further in conjunction with interacting drivers such as climate (e.g. annual rainfall, temperature), site attributes (aspect, altitude, soil pH) and habitat management. In addition, it aims to evaluate the relationship between modelled N deposition and a range of biochemical indicators of N deposition load.

For lowland heathland systems, preliminary results show a weak relationship between N deposition and changes in plant functional groups. Grass abundance is positively related to increasing N deposition levels ($r^2=0.159$). Conversely, lichen cover is seen to decline with increasing levels of N deposition ($r^2=0.492$). For the soil parameters, weak (non-significant) relationships are discernable between N deposition and soil C:N ratio ($r^2=0.063$), total soil N ($r^2=0.071$) and C ($r^2=0.066$) contents. No clear relationship is evident for either extractable NO_3 ($r^2=0.0487$) or NH_4 ($r^2=0.3346$) concentrations. In view of these provisional results, it is evident that the functional diversity of heathland systems is threatened by elevated rates of N deposition. Lichens and grasses appear to be the most robust vegetational indicators of N driven change. N driven changes to soil chemical processes are not observable at this stage.

This paper will consider how much of this ecological change can be attributed to N deposition and incorporate climate, topographical data and changes in base cation and sulphur deposition in the analysis. Concurrent changes in soil pH, moss tissue N and mineralisation potential are also considered.

**SPATIAL TRENDS IN NITROGEN CONTENT IN *CLADONIA PORTENTOSA*;
POTENTIAL TO USE AS A BIOMONITOR FOR ATMOSPHERIC NITROGEN
DEPOSITION**

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Lichens react as one of the most sensitive and receptive species to atmospheric pollution and are suggested to be good biomonitors for nitrogen deposition. We present results from a survey on the species *Cladonia portentosa* from 74 locations in Scotland, the Netherlands and around the Baltic Sea ranging in nitrogen deposition from 3-48 kg N ha⁻¹yr⁻¹. Field sites were visited between 2005 and 2009 and focussed on heathland and coastal dune grassland vegetation.

The nitrogen content of *Cladonia portentosa* appeared to be a suitable biomonitor of the nitrogen deposition levels. A comparison with EMEP deposition data showed that *Cladonia* reflects the deposition history of the last 3–6 years. With increasing nitrogen load, we observed a shift from lichen-rich short grass vegetation towards species-poor vegetation dominated by grasses. In coastal dune grasslands, species richness decreased only for lichens at acid sites. Critical loads for acid, dry coastal dunes might be lower than previously thought, in the range of 4-6 kg N ha⁻¹ yr⁻¹ wet deposition.

THE VALUE OF EPIPHYTES ON TWIGS AND TRUNKS AS INDICATORS OF NH₃ CONCENTRATIONS ACROSS THE UK

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DEFRA policy relevance:

- The objective of this project is the formation of a bio-monitoring scheme that can be used across the UK to evaluate the potential risk from atmospheric NH₃ pollution.
- During the research, lichen species currently defined as being N-sensitive will be reviewed, and a UK specific list formulated.

The composition of lichen communities on acid-barked trees respond to increasing atmospheric NH₃ concentrations through the loss of acidophytic and the increase in nitrophytic species (Fрати *et al.*, 2007). The Lichen Acidophyte Nitrophyte (L_{AN}) scoring method is based on the species composition of epiphytic lichen communities. Initial field studies display a good correlation between L_{AN} scores and atmospheric NH₃ concentrations (Wolseley, *et al.*, 2009), and indicate the method can be utilised for bio-monitoring purposes.

Comparative research on lichen communities in the western (oceanic climate) and eastern (continental climate) regions of the UK (Wolseley, *et al.*, 2006) highlight that potential discrepancies may occur in the scoring system as a result of a climatic gradient. In addition, shortlists of acidophytic and nitrophytic species with clumped species are frequently used, which can potentially skew results.

Epiphytic lichens and bryophytes are being surveyed on *Quercus* and *Betula* trunks and twigs at 29 sites across the UK, covering a range of atmospheric NH₃ concentrations, in oceanic and continental climatic conditions. Initial assessments were undertaken on surveyed sites to identify the strength of the relationship between L_{AN} values of epiphytes and atmospheric NH₃ concentrations at these sites. The effect of species clumping upon the scoring system, and its subsequent influence on the relationship of the L_{AN} score with NH₃ was also evaluated.

Fрати, L. *et al.* (2007). *Environmental Pollution* 146: 311-316

Wolseley, P.A. *et al.* (2006). *The Lichenologist* 38 (2): 161-176

Wolseley, P.A. *et al.* (2009). In: Sutton, M.A. *et al.* (2009). Atmospheric Ammonia. pp 101-108

NITROGEN IMPACTS IN FIXED SAND DUNE GRASSLAND: RESULTS FROM A GRADIENT SURVEY AND IMPLICATIONS OF AMMONIA POINT SOURCES

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This study has relevance to Defra policy as:

- It tests a range of bio-indicator measures across a nitrogen gradient in fixed sand dune grassland – a BAP priority habitat.
- It calculates local impact of an ammonia point source on an SAC upwind of the point source.

In combination with other selected semi-natural habitats, a gradient survey was conducted to test bio-indicator measures of nitrogen impacts on fixed sand dune grassland (NVC community SD 11/SD8). This is a de-calcified older dune community, with soils typically >4 cm thick and over 100 years old. Recent work in the Baltic suggests that acidic dune grasslands are much more sensitive to N than calcareous systems, with critical loads perhaps as low as 4 kg N/ha/yr wet deposition (Remke et al. 2009). This study tests effects on species richness, N mineralisation and moss enzyme assays for dune grasslands within a deposition range of 5 - 17 kg/ha/yr, including some sites on the continent to improve replication at the lower and upper ends of the gradient.

A second study has looked at the specific input of ammonia to a sand dune SAC from a point source that is downwind of the site. Typically such point sources are assumed to have negligible impact, but careful analysis of ammonia levels at this site show that the point source contributes to exceedance of the Critical Level (at 1 ug/m³) even though the site is 800 m upwind of the prevailing winds. While background total N deposition at the site is very low, the additional input from the point source contributes an additional 30%, bringing total N deposition to within the critical load range.

RECOVERY OF PLANT SPECIES RICHNESS DURING LONG-TERM EUTROPHICATION OF A SPECIES RICH GRASSLAND

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Eutrophication (nutrient enrichment of habitats) is considered to be one of the main causes of plant diversity decline worldwide. Several experiments have shown a rapid loss of species in the first years after eutrophication started. However, little is known about species richness dynamics in the long term. Here, we use a 50-year fertilization field experiment to show that species richness initially declined, but started to recover after approximately 25 years. In the fertilized treatment, the period of decline was associated with a strong divergence of the plant trait community from the control, reflecting a shift to a plant community adapted to nutrient rich conditions. During the subsequent period of diversity increase, the trait community remained stable. These results show that plant species richness can at least partially recover after the initial diversity decline caused by eutrophication. However, our data also suggest that for this recovery to occur, two criteria must be met: 1) species adapted to the nutrient-rich conditions are part of the regional species pool, and 2) these species are able to colonize the area.

This study has relevance to DEFRA policy since it examines long term effects of nutrient enrichment on species richness and composition of species-rich grasslands.

THE IMPACTS OF ROADS ON THE STRUCTURE AND FUNCTION OF CALCAREOUS GRASSLANDS.

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- This study has policy relevance through its quantification of the magnitude and extent of traffic-derived pollution on roadside plant communities and ecological processes. It will also establish exceedances of critical levels of NO_x at sites of conservation importance across southern England.

There is considerable evidence that anthropogenic induced changes to natural nutrient cycles can have significant impacts on the composition and structure of ecosystems. Vehicle usage has increased approximately 950% since 1950. It is, therefore, important to understand the influence of vehicular traffic on the natural environment. The impacts of changes to calcareous grasslands are particularly important due to their high biodiversity and nature conservation value. This study has used a transect approach to identify patterns of nitrogen dioxide concentrations, plant community composition and soil biogeochemical properties at three calcareous grassland sites. We present evidence that roads are changing species composition in calcareous grasslands by increasing the productivity of grasses relative to forbs, with inter-site variations in environmental factors such as soil pH and pollution levels driving these changes. Although there is the potential for both direct and indirect effects of vehicle emissions on grassland ecosystems, a lack of any consistent relationship between nitrogen dioxide concentrations and soil biogeochemistry indicates that further work is required to identify the mechanisms driving observed change. The number of significant relationships between environmental drivers and above- and below-ground processes highlights the need to consider multiple factors when attempting to understand the influence of anthropogenic changes in naturally heterogeneous landscapes.

BRYOPHYTE RESPONSE TO N DEPOSITION: A NATIONAL SURVEY

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Nearly two decades of simulated atmospheric N deposition applied to UKREATE plots in grassland at Wardlow Hay Cop has shown the common mosses *Rhytidiadelphus squarrosus* and *Pseudoscleropodium purum* are highly sensitive to N pollution. The cover of these mosses was affected to a greater extent than vascular plants, and several core physiological variables including their photosynthetic efficiency and phosphatase enzyme activities were found to be altered in a dose-dependent manner by wet deposition enriched in both reduced and oxidised N. We hypothesised that standard measurements of moss physiology would serve as sensitive bio-indicators of the impacts of enhanced 'natural' N deposition in national-scale surveys, and that common mosses would be ideal monitoring species because of they are widespread and abundant across many habitat types. To test these hypotheses moss samples were collected from a total of 135 sites, encompassing upland and lowland heathland, M19 bogs, sand dunes and acidic grasslands, as part of a multi-habitat botanical survey conducted in the DEFRA Terrestrial Umbrella project in 2009.

Results of phosphomonoesterase (PME) activity, tissue N:P ratios and chlorophyll fluorescence will be reported in this presentation. PME activity was positively correlated with N deposition for *Hypnum jutlandicum* across heathlands and bogs (n = 80 sites, $r^2=0.124$, $P<0.01$). A similar relationship was found for *P. purum* in acidic grasslands (n = 19 sites, $r^2=0.191$, $P=0.062$). We have developed a method for measurement of chlorophyll fluorescence in samples after they have been air-dried and then re-hydrated for approximately a week, and have been investigating the effect of dry storage for different periods of time. The work to date suggests that some moss physiological responses are responsive to national-scale variations in N deposition but other factors also have important impacts on these variables. The advantages and emerging limitations of use of moss physiological variables as bio-indicators of N deposition will be discussed.

**SOLUBLE AMMONIUM IN MOSSES:
ITS VALUE AS A BIOINDICATOR**

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Defra policy relevance:

- A simplified method for measuring soluble ammonium that can be used by the regulatory and conservation agencies as a bioindicator of N deposition.

Soluble ammonium (NH₄-N) in plants is one of the newly recognized methods that can be used as a bioindicator for atmospheric nitrogen concentrations and deposition. Previous studies suggest that the relationship with atmospheric N is more precise for soluble ammonium (NH₄-N) than for total tissue N (%N) in plants.

Although the existing methodology (Loubet *et al.* 2002) is effective it is time consuming and requires the use of chemicals (liquid nitrogen). We report the development of a quicker, simpler and cheaper extraction method. This revised method has been used in several recent bioindicator studies.

We found a very strong ($R^2 > 0.9$) relationship between the NH₄-N concentration in both pleurocarpous mosses and transplanted grass plants and the measured atmospheric NH₃ concentration at different distances from a poultry farm (NH₃ point source). At sites with mixed NH₃ and NO₂ sources (*e.g.* alongside a motorway) the relationship between NH₄-N concentration in mosses and atmospheric NH₃ concentration is less strong ($R^2 = 0.4$).

The amount of NH₄-N found in plants (mainly mosses and grasses) is found to be not only related to the atmospheric N concentration but also to the plant species and N-source.

References:

Benjamin Loubet, Celia Milford, Paul W. Hill, Y. Sim Tang, Pierre Cellier & Mark A. Sutton. 2002. Seasonal variability of apoplastic NH_4^+ and pH in an intensively managed grassland. *Plant and Soil* **238**, 97-110

BIODIVERSITY, VEGETATION GRADIENTS AND KEY BIOGEOCHEMICAL PROCESSES IN THE HEATHLAND LANDSCAPE

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The northwest European heathland landscape with its characteristic communities of nutrient poor and acid soils has a high nature value, because of its locally high biodiversity and the distinct site conditions. In order to conserve and restore the heathlands, numerous restoration projects have been performed, although with varying success. This is partly due to the fact that the key biogeochemical processes distinguishing the various vegetation types within the heathlands are not known in detail.

Therefore, we performed a statistical survey on the main communities and their soil characteristics. In addition, we analyzed the data for key factors determining biodiversity in the heathland landscape. We will present the results from this study, which was based on a dataset of 267 vegetation relevés (classified as EUNIS habitat types) and 22 soil parameters.

The major distinguishing parameter between habitat types is soil acidity explained, while soil moisture content and soil fertility are less important. Acidity related factors as Al^{3+} , Al/Ca-ratio and pH are also strongly correlated to plant diversity in the majority of the habitat types, respectively the species-rich *Nardus* grasslands, the Rhynchosporion communities and the species-rich *Molinia* meadows, indicating that soil acidification is the important factor in loss of plant diversity in these ecosystems. In dry heaths and over the total heathland landscape, biodiversity is negatively correlated with soil NH_4^+ -concentrations. Only in wet heath, nutrient availability, in this case P, is the primary factor in explaining plant diversity.

INCREASED N AVAILABILITY IN A MEDITERRANEAN ECOSYSTEM IN PORTUGAL: LINKING ABOVE- AND BELOW-GROUND CHANGES

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Most studies addressing the effects of increased N availability have been undertaken in northern European ecosystems. Few have been developed in Mediterranean ecosystems. We are undertaking an integrated field study (soil, plants and microorganisms) of the effects of increased N availability in a Mediterranean-type ecosystem in southern Portugal located in Arrábida (PTCON0010 Arrábida/Espichel). The experimental design consists mainly of manipulating N availability (dose and forms). N availability has been modified by the addition of 40 and 80 kg N/ha/year in the form of NH_4NO_3 and 40 kg $\text{NH}_4\text{-N}$ /ha/year (the control plots corresponds to no fertilization). N is added in three equal applications throughout the year corresponding to distinct seasons, which correlate with distinct biological activities (spring, summer and middle autumn/winter). Each N treatment has three replicates (400 m² plots). N additions began January 2007.

The ecosystem was very responsive to short term increased N availability since one year of N additions were sufficient to induce changes in above- and below-ground (soil bacteria and arbuscular mycorrhizal fungi - AMF - spores) communities.

Changes in the plant community involved species appearance and disappearance and/or changes in plant species cover. The treatment which changed plant species the most was the 80 kg N- NH_4NO_3 /ha/y: eighteen 'new' species and ten 'losses' were recorded. Diversity of soil bacterial community (evaluated by TGGE fingerprinting) assessed during spring showed the existence of three main soil clusters formed by: 1) control plots and one of the 40 Kg N- NH_4^+ /ha/y fertilized plots, 2) all the remaining 40 Kg N/ha/y and one of the 80 Kg N- NH_4NO_3 /ha/y and 3) the remaining two plots receiving 80 Kg N- NH_4NO_3 /ha/y. Repeating this analysis on summer, showed no effect of increased N availability. Diversity of soil bacteria may reflect the hierarchy of biological constraints in Mediterranean-type ecosystems:

water and N, i.e., when water is not limiting (spring), soil bacterial communities responded to the N dose; when water became limiting (summer), soil bacterial community did not respond to N. The above-ground shift is also reflected below-ground.

DO PLANTS EVOLVE TO REGULATE THEIR OWN SOIL N SUPPLY?

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When assessing the impacts of nitrogen deposition on terrestrial ecosystems, it is important to understand the role of plant litter upon the fate of and deposited N. In forest ecosystems C:N ratio is regarded generally as a crucial factor governing the transformation rates of N species in soil. Recently Cresser et al. (2008) suggested that plants, especially deciduous plants, may have evolved in such a way that there is a dynamic match between rate of release of N from their deposited litter and plant N uptake requirements. The fact that litter generally has a high C:N ratio at the time of litter fall facilitates microbial retention of mineralized N during early stages of litter decomposition when plant N requirement is low. As litter decomposition proceeds it is likely that its C:N ratio falls until eventually mineralized N is progressively less and less immobilized, becoming plant available as and when required.

An experiment has been set up to assess net mineral N species production using litter collected from hazel trees in June, August and October 2009 so that the litter possessed a progressively increasing C:N ratio. This was to test the hypothesis that N immobilization in mineral soil/litter mixes would be increased with the litter C:N ratio (i.e. would be greatest for October-collected litter). Equilibration with moist soil was allowed for one week and three weeks prior to extraction of ammonium- and nitrate-N with KCl. The results confirmed the substantial immobilization of both ammonium and nitrate as expected. However the experimental period was insufficient, even although run for 3 weeks, to obtain substantial release of mineral N, and a future experiment will be run over a longer time period. It was also found that the litter:soil ratio significantly influenced the net mineral N species production/immobilization rates.

Policy Implications

The significance of this experiment lies in the fact that N deposition is not seasonal in the same way that net mineral N production in soils is and therefore N deposition causes a dynamic mis-match between soil N availability and plant N uptake requirements. It is suggested that this is the mechanism by which N deposition may be causing changes in biodiversity.

Cresser, M.S., Aitkenhead M.J. and Mian, I.A. A reappraisal of the terrestrial nitrogen cycle – What can we learn by extracting concepts from Gaia theory? *Science of the Total Environment*, **400:344-355 (2008)**.

ROLE OF LITTER IN THE BIOGEOCHEMICAL CYCLING OF C AND N IN AN ACIDIC GRASSLAND ECOSYSTEM

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Litter decomposition rates and processes are key connections in cycling of nutrients, and sustainability and primary productivity within an ecosystem. Continuing high levels of N deposition have altered the dynamics of N cycling, so the role of organic matter has become increasingly important in both anthropogenically altered and minimally managed environments.

A microcosm study has been used to study, for a freely draining acid grassland soil, the role of litter in N species dynamics and its influence on dissolved inorganic nitrogen (DIN), organic nitrogen (DON) and organic carbon (DOC) concentrations and fluxes in drainage water as seasonal temperature changes occur over 7 months. Litter decomposition contributed substantially to NH_4^+ production, which was affected strongly by changes in temperature. NH_4^+ from litter layers was remarkably mobile and raised the extractable NH_4^+ concentrations in subsoils beneath the litter layers. However, when litter was incorporated within subsoils, it markedly reduced the mobility of NO_3^- , especially during winter when NO_3^- is considered relatively more mobile due to reduction in biological uptake. Extractable NH_4^+ showed strong positive associations with water soluble DOC, suggesting a potential role of DOC in NH_4^+ mobility and retention. In contrast, extractable NO_3^- concentrations were correlated negatively with DOC, which may indicate a role for DOC in NO_3^- immobilization by acting as substrate for microorganisms.

Presence of litter significantly altered concentrations and fluxes of DIN, DON and DOC in the drainage water. Concentrations of NH_4^+ increased substantially after freeze-thaw events which facilitated NH_4^+ mobilization. Litter layers contributed sustainably towards DOC and DON in the drainage water. Increase in temperature significantly enhanced DOC and DON concentrations for the control and surface litter treatments; however, the opposite trend was observed for the subsurface litter treatment. Cumulative fluxes indicated that DON formed a significant component of total dissolved nitrogen (TDN); 42, 46 and 62% for control, surface litter and subsurface litter treatment respectively. Cumulative fluxes showed net NH_4^+ retention in each treatment and significantly reduced NO_3^- flux associated with subsurface litter placement.

The dramatic differences between surface- and incorporated-litter treatment effects on N cycling and leaching have significant policy and management implications for the protection of surface waters and ground waters from nitrate accumulation. Moreover

understanding the seasonal dynamics of N cycling is a crucial prerequisite to understanding how N deposition may impact upon plant biodiversity.

FOUNDATION STUDY FOR THE ESTABLISHMENT OF A BIOMONITORING NETWORK FOR NITROGEN DEPOSITION IN MÉXICO

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This study aimed to identify species and biological variables suitable for the establishment of a biomonitoring network for nitrogen deposition in México, in particular in those areas where Pine-Oak woodlands are present.

This project was composed of two different approaches, the first undertaken with simulated increases in nitrogen deposition, and a second one carried out in the field under normal contamination conditions. The first study was carried out in a greenhouse where two common mosses, *Braunia secunda* and *Thuidium delicatulum*, and two bromeliads, *Tillandsia recurvata* and *Tillandsia usneoides*, were grown under four different N treatments 0, 1, 2 and 4 g N m⁻² yr⁻¹ where N was applied as an appropriate solution of NH₄NO₃. In this study *Braunia secunda* was the most tolerant and responsive species to N load. In the second field study we assessed the responses of two common mosses, *Braunia secunda* and *Leptodontium pungens*, to increased N deposition by comparing their physiological responses at two different areas: a highly-polluted site located downwind from México City and a low pollution site located both upwind of México City and further away from the major urban areas. Plant responses to nitrogen were determined by measurement of nitrate reductase and surface phosphatase activities, tissue N and P content and sclerophylly. We also tested the effect of the forest type (*Abies* dominated and *Quercus* dominated) and the season (wet and dry).

From this study, based on the response of nitrate reductase and surface phosphatase activities, we can conclude that *Braunia secunda* is the best and most sensitive indicator of N deposition impacts. Validation of this species and measured variables as useful indicators of N deposition impacts at national scale surveys in México certainly warrants further attention.

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HOW DO CHANGES IN HEATHLAND PLANT COMMUNITY COMPOSITION AFFECT ECOSYSTEM FUNCTION?

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- *Global changes such as climate stress, ozone or nitrogen deposition are responsible for shifts in plant community composition and biodiversity loss. By establishing the link between species composition and ecosystem functioning, this study will permit extrapolation of the wide scale consequences of community-level shifts for the functioning of semi-natural ecosystems, and the implications for ecosystem service provision. The study thus contributes to the science base underpinning predictions associated with policies for pollution control and biodiversity conservation.*

Global changes can affect ecosystem function through direct and indirect pathways, and there are also likely to be interactions between these two. Examples of ‘direct’ effects include changes in plant physiology and soil biogeochemistry, whereas ‘indirect’ effects result from changes in plant species composition and the potential for associated shifts in key ecosystem processes. This study investigates the indirect effects of pollution- or climate-driven changes in plant community composition on the function of heathland ecosystems using both field surveys and mesocosm approaches. The aim of this study is to test whether there are critical thresholds of heathland plant community composition (i.e. tipping points) at which point ecosystem properties switch to resemble conditions associated with monocultures of particular plant species.

Soil cores were collected from three lowland heathland sites (Thursley, Yateley and Frensham) in both spring and autumn (2007-2009). Samples were collected from under three dominant heathland plant species (*Calluna vulgaris*, *Deschampsia flexuosa* and *Ulex minor*) growing either in monoculture or in two or three species mixtures of varying proportions. Physico-chemical characteristics, microbial activities, and nutrient availabilities were measured in all soil cores. In parallel, intact turfs were collected from Thursley Common NNR in 2007. These were used to establish a controlled, mesocosm experiment with differing proportions of the three heathland species listed above.

Data from the field sites show that plant community composition generally has only a small effect on soil properties and processes, relative to the magnitude of differences seen between sites and seasons. An effect of community composition is apparent, although this is driven by complex interactions between species, rather than simple additive effects. Mesocosm soil cores were taken in May 2009, to examine how plant community composition affects key ecosystem processes when differences in topography and microclimate are controlled for.

Analysis to date suggests that plant species, and interactions between species and environmental conditions such as soil moisture content, significantly influence soil properties and processes.

METABOLOMIC INTERROGATION OF MOSSES EXPOSED TO N DEPOSITION REVEALS PLANT RESPONSES AT LOW DOSES AT DIFFERENT SPATIAL SCALES

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Increased nitrogen deposition in semi-natural habitats including heathlands has detrimental effects on vegetation and can lead to habitat change and a loss of biodiversity. Detecting responses to increased available reactive nitrogen in such ecosystems is therefore of great importance. Percent foliar nitrogen has in the past been employed to quantify N accumulation but it provides little insight into physiological responses to nitrogen deposition. In this study the metabolomic fingerprinting method, Fourier-transform infrared spectroscopy (FTIR) was employed to more intensely interrogate metabolic responses of *Sphagnum capillifolium* exposed to simulated nitrogen deposition under field conditions at an experimental site near Edinburgh.

Foliar nutrient analyses of *S. capillifolium* tissue only revealed increased percentage nitrogen at the higher N deposition levels of ammonium and nitrate (56 kg N ha⁻¹yr⁻¹). However, FTIR analysis of tissue provided a more entire suite of responses with separation occurring in relation to increasing nitrate addition (from as little as 8 kg N ha⁻¹yr⁻¹). Separation with increasing ammonium deposition was more limited. These results reflect known responses which have shown more pronounced damage in *S. capillifolium* in response to nitrate compared to ammonium.

Preliminary results are also presented for FTIR and foliar N analysis of the moss *Hylocomium splendens* collected during a nationwide survey, from a range of nitrogen deposition scenarios. Regression analyses indicated that the relationship between the metabolomic data and estimated nitrogen deposition was stronger than that between percent foliar N and nitrogen deposition.

These results provide evidence that FTIR can be applied at a regional and national scale to detect changes in bryophytes in response to different forms of N deposition, even at relatively low doses, which are not evident from macronutrient data alone.

LICHENS, AIR QUALITY AND CITIZEN SCIENCE

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A public engagement project to use epiphytic lichens on tree trunks and twigs as indicators of air quality is being supported in the Big Lottery funded OPAL project. During the air survey, from September 2009, on-line data entry is allowing us to gather a large amount of data from contributors across the country about the distribution of nine macrolichens selected as indicators of air quality that are easily identified. Indicator lichens were selected either for their sensitivity to, or tolerance of, nitrogen, and a further group that is becoming widespread in urban areas. Preliminary results will be presented using a simple air quality score that can be compared with air quality data available from modelled and LAQM sources.

PROCESSES INVOLVED IN SURFACE EXCHANGE OF AMMONIA WITH A FERTILISED CORN CANOPY IN NORTH CAROLINA

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In the spring and summer of 2007, an experiment was conducted near Lillington, North Carolina, USA to study the processes of ammonia (NH_3) air-surface exchange in fertilised corn fields. Several air concentrations and flux measurement techniques were used to quantify the emissions and deposition of NH_3 to the vegetation and the soil. Wet chemistry (AMANDA) and photoacoustic spectroscopy (Nitrolux Pranalytica) measurement techniques were used to calculate canopy-scale fluxes (by the modified Bowen-ratio). Canopy sources and sinks were then calculated by inverse modeling of NH_3 , temperature and the turbulence. Additional measurements of NH_4^+ and H^+ in the soil, leaves, stomatal cavities, and leaf surface water, allowed modeling of the soil and total plant cycling of NH_3 . A general overview of the experiment, measurements, and modeling results is presented and the wider relevance to NH_3 in agricultural systems is discussed.

HOW N DEPOSITION AFFECTS CO₂ EXCHANGE ON AN OMBROTROPHIC PEATLAND

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Defra policy relevance:

- This is one of few studies to investigate the effect of oxidised versus reduced N on the CO₂ balance of a peatland ecosystem and will help inform the question, how does N affect the C balance of peatlands.

Human industrialisation has led to large volumes of nitrogen being released into the atmosphere, much of which is then deposited on uplands in the UK. Many of these areas are home to ombrotrophic peatlands which would normally be subjected to low nutrient input. Increasing the available N to these ecosystems has the potential to alter biomass production and change species composition in the ecosystem. This could in turn alter the CO₂ exchange between the land and atmosphere and affect the peatlands status as a carbon store.

At Whim Moss a long term experiment has been running for several years to investigate the affects of nitrogen deposition on an ombrotrophic peatland. The site uses an automated system to simulate wet deposition at different concentrations/doses and with forms of reduced and oxidised N to different plots. The CO₂ fluxes from the plots are measured monthly over a 24 hour period along with meteorological variables and detailed vegetation surveys. These factors are used to create a model of the CO₂ exchange within the plot in relation to environmental conditions which is then used to compare between plots.

This paper will discuss the pattern of CO₂ fluxes over 24 hours and how this is modified by N form and dose for static chambers ~ 38 cm diameter, dominated by *Calluna* with a deep moss (*Sphagnum* and pleurocarpous mosses) understorey.

HOW DO GREENHOUSE GAS EMISSIONS FROM BIOENERGY CROPS DIFFER FROM CONVENTIONAL CROPS?

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Bioenergy is a key component in the UK Government's plans for tackling energy security and climate change; the latter because it is perceived to be 'carbon neutral', i.e. the carbon emitted in using energy is balanced by the carbon absorbed in the growing feedstock. The target is that by 2020, 1 million ha (17% of the total UK arable land) will be available for energy feedstocks, including 350,000 ha for energy crops. However, the effects of miscanthus and short rotation willow on the soil fluxes of N₂O, CO₂ and CH₄ during their growth are not well known. These crops require different management, e.g., they are not limed, require lower N inputs for production, and tend to exhibit greater water-use efficiency than conventional crops. This potentially has a significant effect on soil biogeochemistry and microbial populations and therefore likely also on net GHG emissions from soil.

This study was carried out in Lincolnshire, NE England on adjacent fields of willow (*salix*), miscanthus, wheat (*triticum*) and oilseed rape (*Brassica napus*) on a heavy clay soil. N₂O and CH₄ flux measurements were made monthly using static chambers, CO₂ fluxes were measured continuously by eddy covariance.

Results to date show higher N₂O emissions from soils under conventional crops than bioenergy crops. Average fluxes for the period Jun 2008 to Aug 2009 were 9 and 34 μg m⁻² h⁻¹ N₂O-N from wheat and oilseed rape and 7 and 1 μg m⁻² h⁻¹ N₂O-N from *Miscanthus* and willow, respectively. No clear trend in CH₄ fluxes could yet be identified. These observations are important regarding future land-use and management policies. Controlling factors will be discussed as well as differences between crops and seasonal variations. Additionally controls of N₂O/CH₄ emissions will be presented to assess potential change in future GHG emissions.

Initial results show that N₂O emissions from bioenergy crops may be lower than from conventional crops. This is probably due to the higher use of fertiliser in conventional cropping and annual ploughing. Due to seasonal and interannual variations in the soil and climatic conditions and physiological changes during the circa 20 year live time of the bioenergy crops it is important to continue these greenhouse gas measurements long-term before the on field carbon footprint of the bioenergy crop can be properly evaluated.

HYDROGEN FLUORIDE POLLUTION IMPACTS ON FRUIT TREES IN PESHAWAR

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- Peshawar is the capital of North West Frontier Province (NWFP) of Pakistan. Rapid urbanization and an exponential increase in the number of all types of vehicles have given rise to the deterioration of air quality, especially in the big urban areas of Peshawar. Visible plumes have also been reported from foundries, brick kilns and rolling mills in Peshawar. There are about 400-450 brick kilns around the city.
- The aim of the study was to assess the extent of visible HF injury to fruit trees and other crops in the area.
- Injury to different species was assessed in two surveys, from February to June 2008, and from October 2008 to January 2009. Injury was assessed near to brick kiln fields (BKF), and at three sites at different distances from the brick kilns.
- In the summer, visible injuries to mango, plum and apricot leaves typical of fluoride were observed in the brick kiln area. The mean HF level in the air was higher in BKF than at the other sites during the spring and early summer. The fluoride content of apricot, plum and mango leaves was also significantly higher at BKF compared to other sites.
- The only visible injury recorded during the winter survey was leaf tip burn injury to young wheat plants at BKF area. The fluoride content of spinach and wheat grain of BKF was also significantly higher than at other sites.
- It was concluded that the substantial visible injuries to leaves and fruits of fruit trees at the BKF site were due to hydrogen fluoride emitted from brick kiln factories in the surrounding areas.
- This is the first study carried out on the effects of HF pollution on crops in Peshawar and suggests that air pollution may be having significant effects on the growth and yield of sensitive crops in the areas of brick kiln activity.

GROUND LEVEL OZONE (O₃) AND CROP YIELD IN SOUTH ASIA AND ITS SUBSEQUENT ECONOMIC EFFECT

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This study has relevance to Defra policy as:

- This would aid Defra's participation in the Global Science and Innovation Forum (GSIF), as a part of the strategic framework to improve the coordination of the UK's engagement in international science and innovation.

High concentrations of O₃ (as high as 70 ppb-7 hour means) have been reported in the Indo-Gangetic plains, the most important crop growing region in South Asia. Experimental studies have reported that the current day surface O₃ concentrations in this region frequently result in yield losses of up to 30% for sensitive crops (e.g., wheat, rice, legumes). Future global emission projections suggest that, even with implementation of current emission reduction legislation, South Asia may become one of the most O₃ polluted regions of the world. This suggests that O₃ may pose a potential threat to future food security in South Asia.

A study to estimate economic losses associated with ambient O₃ concentrations and its effect on crops grown in South Asia was conducted based on concentration-based dose-response indices which characterize O₃ dose as 7 hour growing season mean (M7) or accumulated above a threshold of 40 ppb (AOT40) concentrations. The results indicate high yield losses (~10% loss in wheat and 7% for rice) across the region, results that are comparable to experimental evidence. An estimate of the economic loss due to O₃ induced crop yield reductions for the year 2000 suggests a loss of ~4 billion US\$ for four staple crops of the region (rice, wheat, potato and soybean). Results are also presented from application of a socio economic model, developed to assess how these O₃ induced yield losses will affect different sections of the society, i.e. producers (farmers) and consumers across the region.

The effect of O₃ on crops is strongly influenced by dose modifying factors that are known to affect plant sensitivity (e.g. species type, phenology and environmental conditions). To accommodate these factors new flux-based risk assessment methods have been developed in Europe. The European stomatal ozone flux model for wheat has been re-parameterised for South Asian (and specifically Indian) conditions. Preliminary results are presented showing the variation in evolution of both AOT40 and *stomatal* flux indices over the wheat growing season for selected sites across the Indo-Gangetic plain; these results reflect the importance of considering spatial variation in the meteorological conditions and crop phenology in relation to influencing O₃ sensitivity of crops in South Asia.

QUANTIFYING OZONE EFFECTS ON CROPS – IMPLICATIONS FOR FOOD SECURITY

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Defra policy relevance:

- In the coming decades, protection of security of food supplies will be one of the most important issues for governments around the world.
- Two recent Royal Society Reports have highlighted increasing ozone pollution as a threat to the security of food supplies.
- Recent progress with establishing flux-based response functions for effects on the quantity and quality of crop yield will be presented together with methodology for estimating effects at the UK, European and global scales.

With the world population predicted to increase to 9 billion people by 2050, security of food supplies is one of the most important challenges for this century. Food security may be usefully conceptually divided into three major components: Food availability (production, distribution, and exchange), food access (affordability, allocation, and preference), and food utilization (nutritional value, social value, and food safety). Stability of food supplies is often also included as a fourth component, to acknowledge that food security varies seasonally and inter-annually in many places. The key components of the food system that ozone pollution interferes with are the productivity of crops, the nutritional value and the stability of food supplies, and it is these effects that will be discussed in this presentation.

Current global yield losses are estimated to be between 7-12% for wheat, 6-16% for soybean, 3-4 % for rice and 3-5% for maize (Van Dingenen et al., 2009), with global economic losses estimated to be in the range \$14-\$26 billion. In terms of economic value, eight of the nine crops with the highest production in Europe in 2007 (FAOSTAT, faostat.fao.org) are sensitive or moderately sensitive to ozone, including wheat, potato, sugar beet, oilseed rape and tomato (Mills et al., 2007). Typical effects include premature senescence and reduced yield quantity and quality. All assessments made so far have been based on either the 24h mean ozone concentration or AOT40. We present here recent progress made by the ICP Vegetation of the LRTAP Convention¹ on developing more biologically sound flux-based methods for assessing the risk of ozone damage to vegetation, together with maps showing where effects are already occurring in Europe in areas where the flux method predicts that crops are at risk of damage (Mills et al, in press).

Mills, G., Buse, A., Gimeno, B., Bermejo, V., Holland, M., Emberson, L. and Pleijel, H. 2007. A synthesis of AOT40-based response functions and critical levels for agricultural and horticultural crops. *Atmospheric Environment*, 41 2630-2643.

Mills, G., Hayes, F., Simpson, D., Emberson, L., Norris, D., Harmens, H., and Büker, P. (2010). Evidence of widespread effects of ozone on crops and (semi-)natural vegetation in Europe (1990 – 2006) in relation to AOT40 – and flux-based risk maps. *Global Change Biology*. In press.

¹ International Cooperative programme on Effects of Air Pollution on Natural Vegetation and Crops of the Convention on Long-range Transboundary Air Pollution.

Van Dingenen, R., Dentener, F. J., Raes, F. Krol, M. C., Emberson, L., Cofala, J. et al. (2009). "The global impact of ozone on agricultural crop yields under current and future air quality legislation." Atmospheric Environment 43(3): 604-618.

FROM EXPERIMENTS TO POLICY: FLUX-EFFECT RELATIONSHIPS FOR SEMI-NATURAL VEGETATION

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This study has relevance to Defra policy as:

- Ozone fluxes are related to changes in vegetation biomass
- Significant biomass loss can occur for sensitive species within northern Europe
- Effects of ozone on roots and above-ground biomass indicate potential reductions in carbon sequestration
- The outcome of the revision of the Gothenburg Protocol is likely to have a direct effect on UK air pollution policy.

The indicators used in the Gothenburg Protocol to protect (semi-)natural vegetation were based on AOT40 (accumulated hourly ozone concentration over the threshold of 40 ppb). Scientific research has developed further and currently the accumulated ozone flux via plant stomata (Phytotoxic Ozone Dose above a threshold of Y, POD_Y , previously described as $AF_{st}Y$) is considered to provide a biologically sound method for describing observed effects on vegetation. It is calculated from the effects of climate (temperature, humidity, light), ozone, soil (moisture availability) and plant development (growth stage) on the extent of opening of the stomatal pores on leaf surfaces through which ozone enters the plant. This work investigated whether it is appropriate to use flux-based methods for (semi-)natural vegetation in the current revision of the Gothenburg Protocol. Results from discussions at the Workshop on Flux-based assessment of ozone effects for air pollution policy (Ispra, Italy, 2009) are presented, where it was agreed that for (semi-)natural vegetation critical levels for indicator species of three permanent grassland types should be considered separately: (a) Productive grasslands that are intensively managed and grazed; (b) Grasslands of high conservation value with low management and little/low fertilizer input; and (c) Natural unmanaged ecosystems (excluding forests). Following additional modelling, results were presented at the Task Force meeting of the ICP Vegetation in Tervuren, Belgium, 1-3 February, 2010 where decisions were made on flux-based critical levels. The data sources, analyses and decision-making processes are described in this presentation.

Flux-effect relationships have been derived for individual species using data from the solardomes at CEH Bangor, open-top chambers at Newcastle, and open-top chambers in Switzerland -Liebefeld. For each dose-response function, the method of Fuhrer et al (1997) was used to determine relative yield/biomass, whereby the effects in each experiment were calculated relative to the absolute yield/biomass at 0 flux in that experiment, calculated by linear regression. The data from all experiments were then combined and subjected to linear regression analysis.

Dose-response functions accepted for establishing critical levels, and their recommendations for use are presented, together with other dose-response functions of interest but not accepted for further use in establishing critical levels. The results show the potential impacts on some species of semi-natural vegetation including indicator species for reductions in carbon sequestration and effects on biodiversity.

OZONE AND NITROGEN EFFECTS ON SOIL MICROBIAL COMMUNITIES

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This study has relevance to Defra policy as:

- Ozone and nitrogen pollution are related to changes in vegetation biomass and species composition of grasslands.
- These pollutants have been shown to negatively affect root growth.
- There is potential for these effects to change soil microbial communities, carbon turnover rates and carbon sequestration.

A field experiment investigating the combined effects of elevated ozone (O₃) and nitrogen deposition (N), on above-ground productivity and species composition of subalpine grassland was established at 2000 m above sea level in the central Swiss Alps by the Air Pollution and Climate Group, Agroscope Research Station ART, Zurich, Switzerland in 2003 (Bassin, Volk et al. 2007). The plots included in this study consist of monoliths extracted from a species-rich *Geo-Montani-Nardetum* pasture, exposed in a free-air O₃-fumigation system to one of two concentrations of O₃ (ambient and 1.6 times ambient) and N additions of 0kg/ha/yr or 50kg/ha/yr. Soil cores of 15cm depth were taken from the monoliths in May 2009 and results from the following investigation are presented.

Nitrogen exposure may affect the soil chemistry directly, or the combined pollutants effect on the vegetation root systems may change the physical properties of the soil, and/or the C inputs to the soil, through either leaf litter, or rhizodeposition. There are many dynamic pools of C within the soil system, in which the turnover rates differ according to variables including temperature, moisture, and the microbial population. As vegetation provides the main source of carbon to the soil, a change in the quantity or quality of C input will influence the diversity, size and functioning of the soil microbial population.

The root and soil carbon system is extremely complex and fluxes between C pools are difficult to analyse. This investigation uses a three-pronged approach with the aim of describing the belowground effects of two co-occurring pollutants on a species rich grassland. Soil chemical analysis is combined with radioisotope labelling of soil C turnover (Community Level Physiological Profiling CLPP), and microbial diversity analysis is preliminarily addressed using Terminal Restriction Fragment Length Polymorphism (T-RFLP).

Bassin, S., M. Volk, et al. (2007). "Nitrogen deposition but not ozone affects productivity and community composition of subalpine grassland after 3 yr of treatment." New Phytologist **175**(3): 523-534.

NITROGEN DEPOSITION AS A MODIFIER OF PLANT RESPONSE TO OZONE

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- Nitrogen deposition may modify the effects of increasing tropospheric ozone on plant communities. Our evidence suggests that negative responses to ozone are exacerbated under conditions of high N availability. This highlights the importance of considering interactions between pollutants in the evaluation of critical levels (or loads) for semi-natural ecosystems.

During the last century, concentrations of ozone in the troposphere have risen considerably as a result of anthropogenic activities. Ozone is an important phytotoxic air pollutant, with current ambient concentrations high enough to cause visible O₃ injury and detrimental effects on plant growth in many parts of the world. Some evidence exists to suggest that environmental factors such as soil fertility and nitrogen deposition may influence plant sensitivity to ozone. An experiment was, therefore, established in 2009 to examine the effects of these two global change issues, in combination.

Two plant species *Trifolium repens* and *Festuca ovina* were exposed to O₃ under differing levels of N addition to address the question of how soil resource availability (nitrogen status) affects plant response to O₃ in a grass-legume mixture. Mixed species pots were exposed to ozone in a series of 16 open top chambers at Silwood Park (Ascot) for a period of 16 weeks from May to September 2009. Ozone exposure was carried out from 9am-6pm daily, with target concentrations of 0 ppb, 30 ppb, 60 ppb and 90 ppb (resulting in seasonal AOT40 values from 0 – 25,492 ppb.h). In addition, weekly applications of Hoagland's solution with nitrogen levels equivalent to 0 or 50 kg N ha⁻¹ were given to pairs of pots over the course of the experiment. Plant performance was monitored during the exposure period, after which aboveground plant material was harvested, separated into living, senescent or dead material and then oven dried and weighed. Below ground biomass was also measured, following root washing.

Results show a negative effect of O₃ and N on above-ground biomass of *T. repens* and positive effects of both pollutants on the biomass of *F. ovina*. Aboveground biomass reductions of 80.6% (HO₃+N) and 70% (HO₃-N) for *T. repens* indicate greater effects of ozone in high N treatments. Below-ground biomass also showed a significant decrease in ozone treatments (44.6%, HighO₃+N and 35.8% HighO₃-N) compared with filtered air. Overall, results suggest that N deposition may exacerbate the effects of O₃ on legumes, driving changes in plant productivity and community composition.

COMBINED EFFECTS OF OZONE AND DROUGHT ON MESOTROPHIC GRASSLAND COMMUNITIES

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- This investigation evaluates the ecological impacts of rising tropospheric ozone levels on mesotrophic grassland species under differing climates. Results will contribute information in support of Defra's climate adaptation plan and the refinement of ozone critical levels.
- Data collected during this study are also contributing to a flux-based evaluation of ozone risk to semi-natural vegetation at a European scale, thus underpinning the evidence base for (inter)national pollution control policy.

Future climate scenarios include an increase in factors that contribute to tropospheric ozone formation and an increased likelihood of prolonged summer drought. Individually, these stresses reduce plant productivity and growth, and inter-specific differences in sensitivities to ozone and low soil water availability can lead to changes in plant community composition and biodiversity loss. However, little is known about the combined effects of these two important global change drivers at either the species or the community level.

This study involved exposure of model mesotrophic grassland communities to differing levels of ozone, under two levels of simulated rainfall. Mesocosm communities were established in spring 2009 with equal biomass ratios of the following species: *Holcus lanatus*, *Agrostis capillaries*, *Trifolium repens*, *Lotus corniculatus*, *Plantago lanceolata*, *Crepis biennis* and *Hypochaeris radicata*. Ozone fumigation was carried out in 16 open top chambers at Silwood Park (Ascot, UK), for 17 weeks from May to September 2009. Mesocosms received one of four concentrations of ozone (Filtered Air, 30ppb, 60ppb, and 90ppb, resulting in a season-long AOT40 exposure of (0 to 24780 ppb.h), and were subjected to one of two watering regimes. Watering regimes represented either average summer rainfall at Silwood Park over the last 20 years or levels experienced during the summer of 2003, a particularly dry year. Measurements of growth, visible injury, physiology and biochemistry were carried out over the growing season. Aboveground dry biomass was determined, by species, at the end of experiment.

Results indicate species-level differences in responses to ozone and watering regime. Generally increasing levels of ozone exposure resulted in a progressive reduction in aboveground biomass in both well watered and droughted mesocosms. Physiological data suggest that stomatal conductance is lower in water stressed plants, with an associated reduction in ozone flux under drought conditions. Overall, results provide empirical evidence that predicted changes in climate may modify the effects of ozone in some species, as well as emphasising the importance of considering climatic factors in ozone risk assessment and management.

EFFECTS OF OZONE ON METHANE AND CARBON DIOXIDE FLUXES FROM MIRE MESOCOSMS

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Tropospheric ozone poses a significant threat to crop yield and forest productivity of sensitive species, and to ecosystem carbon storage at current levels in Europe, while northern hemisphere background ozone concentrations are expected to increase further during the next decades. Risk assessment of ozone impacts on mires is essential, because (1) knowledge to date on the effects of ozone on mires, and ecosystem functioning in general is limited, (2) mires are a significant store of carbon and an important source of methane (CH₄), and (3) mires are of conservation importance in the UK. The aims of this study are to assess effects of ozone on CO₂ and CH₄ fluxes in peatlands, and to identify the underlying plant, soil and microbial processes.

Mesocosms from a wet heath (Isle of Skye, Scotland) with vegetation dominated by the peat moss *Sphagnum papillosum* and the sedge *Schoenus nigricans* (NVC: M15) were exposed for 2 years to control and elevated levels of ozone in open-top chambers. The control treatment received non-filtered air, whereas the elevated ozone treatments consisted of non-filtered air (NFA) plus 10, 25 and 40/10 ppb. The highest ozone treatment had a target concentration of NFA+40 ppb for 8 hours during the summer and NFA + 10 ppb for 8h per day during winter. In the NFA+10 ppb and NFA+25 ppb treatments, ozone was elevated for 24h per day in both summer and winter. Methane and CO₂ flux measurements, and aboveground plant growth variables were measured at intervals of 6 weeks (summer) or 8 weeks (winter).

Methane emissions were reduced by elevated ozone from the end of the first growing season onwards. Ecosystem respiration was enhanced by elevated ozone during the second growing season, while gross photosynthesis was increased by elevated ozone from the onset of the experiment. The latter may be related to the unexpected higher *Sphagnum* biomass production at elevated ozone. Aboveground sedge growth was not affected by ozone. Potential mechanisms underlying these results will be discussed.

IMPACTS OF OZONE ON A LEGUME-RICH SAND-DUNE COMMUNITY

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A brief report will be provided as to progress with an ongoing OTC study establishing ozone exposure-response relationships for a legume-rich sand dune community ((*Ammophila arenaria*-*Arrhenatherum elatius* grassland-*Geranium sanguineum* sub-community NVC SD9b) of high conservation value. The well-replicated study a range of simulated future ozone climates (present day; future 2030, 2075, 2125). .

Now in the third year of the study, mesocosms subjected to elevated ozone levels are exhibiting dramatic reductions in productivity and marked shifts in species composition; the three dominant legume species (*Ononis repens*, *Lathyrus pratensis*, *Vicia spp.*) being suppressed to the advantage of several aggressive grasses

DEFRA Policy relevance:

Increasing concentrations of tropospheric ozone can decrease productivity and detrimentally alter species composition and biomass of sand-dune grassland communities of high conservation value. This work will assist in mapping and modelling risks posed by ozone and aid policy derivation for the protection of sensitive plant communities.

A FIELD STUDY OF THE EFFECTS OF OZONE ON AN UPLAND GRASSLAND

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Although there have been many studies on ozone and its effects on vegetation, the main focus has been on forests and crops. Very few studies have assessed the effects of ozone on semi-natural vegetation under field conditions. Many semi-natural grasslands in the UK are managed to enhance biodiversity and it is possible that ozone may be a barrier to achieving management objectives in such systems. We present here evidence that exposure to ozone reduces plant species diversity at a site which is managed under the Higher Level Stewardship Scheme (HLS), for creation, maintenance and restoration of species-rich, semi-natural grassland.

The field site, a mesotrophic grassland, is located in the uplands of Northumberland at High Keenley Fell, Allendale, about 18 km from Hexham (NY 7922 5586). The experimental area comprises three replicate treatments. In each treatment a 6 m long pipe releases ozone at a controlled rate to maintain a concentration of 30 ppb above ambient at the vegetation surface 10 m downwind of the release pipe. This is projected to produce a long-time average enhancement of the ozone concentration of 15 ppb, assuming that fumigation occurs 50% of the time. Ozone is generated and released when the wind blows from the SW (between 180°-270°), and when wind speeds are sufficient to ensure dispersion of the gas ($>0.3 \text{ m s}^{-1}$).

36 experimental plots were established, 12 in each of two ozone treatments and a control. The plots were cut in early August of 2007, 2008 and 2009 at the height used by the farmer in the late summer cut. The dry weight of each species was determined. Physical and chemical characteristics of the soil were measured in 2007, before the start of ozone treatments. Flowering of key species was also recorded in the spring of 2009.

There were significant differences in biomass between the transects, which were related to differences in the soil nutrient status. There was also a significant effect of ozone on the biomass of the forb, but not the grass, component of the community in 2008 and 2009. Multivariate analysis based on data for individual species showed that the percentage of the variation that was explained by ozone increased in 2008 and 2009, indicating a pollution-mediated shift in species composition within the forb component of the sward. One of the species that decreased significantly in ozone treatments was *Rhinanthus minor*. This hemiparasitic species has an important role in enhancing diversity in grasslands, and hence the observed effects of ozone on this species could have much wider implications for species diversity in such communities.

IMPACTS OF OZONE ON UK UPLAND MESOTROPHIC GRASSLAND

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An update will be provided on ongoing DEFRA-funded work investigating impacts of present and future (2050) upland ozone climates on productivity and species composition of long-established upland mesotrophic (NVC MG3b) grassland mesocosms. Now in the seventh consecutive year of the study, the results have revealed marked effects of ozone both above- and below-ground on this now scarce upland plant community of high conservation value.

Above ground-productivity and community dynamics

Mesocosms subjected to a 2050 ozone climate have shown a steady and continuing decline in the sub-community indicator species and a dramatic decline in the sole legume species. Moreover, as in previous years, exposure to elevated ozone continues to reduce the overall productivity of the meadow. (*ie.* biomass harvested). It is established that the total area of MG3b grassland is now reduced to 750ha, confined to the upland valleys of the North Pennines and Cumbria. Ozone-induced loss of indicator species in this experimental study suggests these rare upland meadows may be under threat from rising background levels of ozone.

Below-ground productivity and community dynamics – preliminary study

During 2009, soil cores were extracted from replicate mesocosms, DNA extracted, amplified by PCR, subjected to DGGE then PCR products isolated and sequenced. 16S and 28S sequences were identified by NCBI BLAST search. The study revealed highly significant effects of long-term ozone exposure on both the size and diversity of bacterial and fungal populations below-ground. The preliminary study highlights the potential scale of, as yet under-studied, below-ground ozone impacts which may have profound consequences for ecosystem functioning.

DEFRA Policy relevance:

Increasing concentrations of tropospheric ozone can decrease productivity and detrimentally alter species composition and biomass of upland grassland communities of high conservation value. This work will assist policy derivation for the protection of ecosystems particularly sensitive to predicted changes in atmospheric composition.

POSTERS

DIVERSITY AND SUSTAINABILITY OF FRAGMENTED HEATHLAND METACOMMUNITIES

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Heathland habitats once extended to some several million hectares in Western Europe, but now their extent is limited to less than 350,000ha. Heathland communities are subject to perturbation due to atmospheric pollution, leading to a reduced competitive ability of ericoids. Atmospheric nitrogen deposition is increasing and may lead to an increase in heathland loss. Through a combination of field and manipulative experiments, this research aims to measure vascular plant, lichen and bryophyte diversity in a range of UK heathlands, and investigate the extent to which this is affected by fragment size, nitrogen pollution, geographical location and local environmental characteristics. The extent to which the fertility of soils has been modified by nitrogen pollution will also be investigated. Preliminary results suggest that there is a relationship between atmospheric nitrogen input and the fertility and diversity of UK heathlands.

OPAL: INSPIRING A NEW GENERATION OF NATURE-LOVERS

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- DEFRA is dedicated to investigating and communicating the importance of the natural environment. OPAL has developed a wide range of local and national programmes to encourage people from all backgrounds to get in touch with nature. In this way, OPAL is facilitating greater understanding and communication between scientists, policy makers, amateur-experts and the public.
- Research carried out at the OPAL Air Centre will provide a comprehensive evaluation of the impact of elevated ozone concentrations, in combination with climate stress and nitrogen deposition, on calcifugous grassland habitats. These data can then provide information on BAP species and habitats at risk from future pollution scenarios and thus contribute to the evidence underpinning emission reduction policies.

The OPAL Air and Climate Research Centres provide the infrastructure for a world class facility for high impact research. Results from studies carried out in the Air Centre's open top chambers will significantly advance scientific knowledge of the impacts of air pollution for biodiversity and ecosystem sustainability. Understanding the relationship between air quality and environmental impacts is not only important scientifically, but is the foundation for public awareness of issues surrounding environmental sustainability. The OPAL team are actively involved in communicating important scientific messages in a fun and interesting way, through its Air Quality Road Shows, Summer Schools and Fun Days. Our activities are aimed at enhancing public knowledge of pollution issues and impacts, and inspiring people to consider the environmental consequences of their everyday actions.

One of the ongoing milestones of the OPAL Climate Impacts project is introduce members of the public to issues surrounding the likely ecological effects of climate change, and the links between human activities, greenhouse gas emissions and climate change. We also use these outreach opportunities to try to inform people about the value of biodiversity and habitats and the role they play in maintaining a healthy and sustainable natural environment. We believe that our activities succeed in helping people to feel more engaged with the natural world around them and enthused to explore it more thoroughly.

One of the key messages we put over during our outreach events is that the health of the environment is important for everyone; if everyone just makes minor changes to their lifestyle it can all add up to become a large benefit. OPAL outreach activities are helping to promote a new generation of environmentalists, with a keen interest in the natural world and issues underlying the maintenance of a healthy natural environment for future generations.

HOW DO CLIMATE CHANGE AND PLANT DIVERSITY AFFECT ECOSYSTEM FUNCTION IN A LOWLAND GRASSLAND?

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- This work contributes to the Government's "Adapting to climate change programme" by contributing to the evidence of impacts of climate change in terms of soil and grassland processes.
- This work will make available information on how plant communities are structured and function together, and how the loss of a functional group through climate change can have a knock-on effect on the ability of the community to function.
- The project will aid predictions of which functional groups or species are most at risk of succumbing to climate change, and whether this would have a profound impact on biodiversity, leading to catastrophic loss of ecosystem functioning.

This experiment combines a climate treatment with three diversity treatments in a full factorial design to investigate how different plant functional groups can mitigate the effect of climate change. Plant functional traits (PFTs) such as specific leaf area combine to give quantifiable 'identities' of plant species which are responsible for their role in ecosystem functioning. We hope to be able to explain how ecosystem processes are affected and how species will adapt in community terms, to accommodate climate change.

Biogeochemical processes such as mineralisation rates are measured to determine the impact of climate change and test whether certain combinations of functional groups are more conducive to mitigating its effect, or whether individual species are disproportionately effective.

Additional experiments, running concurrently with the main experiment, have been set up to investigate the effects of extreme and variable climate change, and the interaction between nitrogen deposition and climate change. In line with recent meta-analyses, increases to plant productivity and an associated decline in species diversity is predicted in response to elevated soil nitrogen availability, an effect which is predicted to be further enhanced by elevated precipitation.

I present some preliminary results from the first two years of the experiment, showing how functional trait averages in communities can affect abundance, and then a description of the work I intend to carry out next using functional diversity indices to describe variance in ecosystem processes.

**BASE AND NON-BASE CATION DEPLETION AND REDUCED C STORAGE
AFTER SIMULATED N POLLUTION IN A SEMI-ARID MEDITERRANEAN
ECOSYSTEM**

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Nitrogen (N) deposition is recognized to affect nutrient dynamics and it has been related to biodiversity loss in grasslands and heathlands across the UK and other temperate areas; however evidences from water-limited ecosystems (such as those of the Mediterranean-Basin) are scarcer. This makes their study important in order to know the impact of N pollution at a global scale. We conducted a one-year seasonal survey within the context of a field fertilization experiment carried out in a semi-arid shrubland from central Spain. Additions of NH_4NO_3 simulated a gradient of N deposition (0, 10, 20 and 50 $\text{Kg N ha}^{-1} \text{yr}^{-1}$) over the background (10-16 $\text{Kg N ha}^{-1} \text{yr}^{-1}$). The surveys lasted from the autumn 2008, one year after the first N addition, to the summer 2009. Soil pH, organic matter, organic C and N, extractable N and P, and base and non-base cations were highly seasonal; this was due to biological (mineralization, nitrification, denitrification and uptake) and non-biological (external nutrient inputs dissolved in rainwater, volatilization and leaching) seasonal processes. Nitrate dominates the inorganic N fraction (~ 5-fold) in soils despite the slight dominance of ammonium in bulk deposition. Inorganic N significantly accumulates in soils after the N addition and the experimental inputs are also related to increased nitrate seasonality. K was dose N-related depleted after using current season nitrate or nitrate seasonality as covariate and its seasonality was reduced with the N supply as a consequence of its depleted soil budget. Mg, Zn or Cu losses were also evident depending on the season. Soil C storage capacity was seasonally reduced by N fertilization with potential implications for the soil to act as a C sink in a context of increased CO_2 emissions and climate change. The potential implication of P limitation on the ecosystem responsiveness to N pollution has also arisen from our experimental evidence. Overall, our data suggest that in the short-term the seasonal nutrient dynamics may overwhelm any soil chemistry alteration related to atmospheric N deposition but the potential implications in the long-term are not so-well understood yet; the ecological relevance of the altered soil chemistry by pollutant N in the context of the high spatial and temporal seasonality that characterize the Mediterranean-type ecosystems should receive attention in future research.

WHAT CONTROLS SPATIAL AND TEMPORAL VARIATIONS IN N SPECIES CONCENTRATIONS IN THE RIVER DERWENT, N. YORKSHIRE?

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The River Derwent catchment in N. Yorkshire has been subject over the past decade to reduced deposition of atmospheric N and also, in lower reaches, to constraints upon rates and timing of fertiliser and manure use and storage following imposition of NVZ status. It may also have benefited significantly from regional reductions in livestock numbers following outbreaks of foot and mouth disease in 2001. Short and long-term benefits may therefore be expected on concentrations of reduced N species in river water. Seasonality in nitrate-N and ammonium-N is distinct along the River Derwent in agriculturally impacted areas over 20 years to 2007, but winter maxima have declined significantly since 2001/02 post NVZ status. High ammonium-N episodes were irregular in areas with high runoff from uplands. Both upland and lowland areas displayed summer peaks in free ammonia. These occurred later than winter ammonium peaks, and it is shown that they are driven mainly by seasonal trends in pH and temperature. Summer ammonia maxima declined after 2001, but occasional high peaks still occurred associated with high ammonium concentrations. These may have various causes, like effluent discharges or the substantial drought in the Yorkshire Dales in 2006. Summer minima in nitrate-N concentrations are now higher than they were, reducing seasonal variations.

Policy implications

The influence of long-term changes in N deposition on river water chemistry are an important aspect of minimization of nitrate transfer to surface and ground waters, especially since mobility of ammonium within and from soils has not been adequately considered.

FATE AND IMPACTS OF ACUTE ATMOSPHERIC NITROGEN DEPOSITION IN HIGH ARCTIC TERRESTRIAL ECOSYSTEMS:

A MULTIDISCIPLINARY APPROACH

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- Our work will inform Defra policy as to how N emissions in the UK can impact on remote and pristine ecosystems of exceptional conservation value.
- It will inform how these impacts may influence ecosystem functioning that may feedback to global climate (e.g. ecosystem carbon balance).
- The modelling research will extend and enhance methods that are also being used to simulate the impacts of elevated N-deposition on UK ecosystems as part of ongoing Defra-funded research projects, hence widening the applicability of these models to a broader range of ecosystems.

Anthropogenic atmospheric nitrogen deposition poses a major threat to the structure and function of ecosystems in many regions of the world. Among the most pristine of these are Arctic tundra ecosystems that have previously received very low rates of atmospheric N deposition and have been shown to have some sensitivity to increases in N inputs. However, chronic rates of N deposition in the Arctic are low. Instead, a greater threat may come from acute N deposition events where a significant proportion (~ 40%, and perhaps reaching 80%) of annual atmospheric N input can be deposited as acidic rainfall (pH 4) in less than 1 week. These events result from polluted air masses from industrial Europe that travel to the Arctic with minimal dispersal. To date, however, the impact of these acute N deposition events on Arctic ecosystems remains unknown.

The overall aim of our research is to investigate the impact of acute atmospheric nitrogen deposition in high Arctic tundra. A multidisciplinary approach is being used to investigate the impact of short-term acute nitrogen deposition upon plants, soil microbial community structure, diversity and function and on soil biogeochemical processes and ecosystem carbon balance.

Our research is part of a multidisciplinary European Union project entitled “NSINK” (<http://nsinkproject.group.shef.ac.uk/NSINK/Home.html>), which studies the impact of atmospheric N deposition on N cycling in the high Arctic (Svalbard islands) at different scales: atmosphere, snowpack, glaciers, terrestrial ecosystems (plants, soil) and aquatic ecosystems (rivers and lakes).

In our study, acute N deposition is being simulated using a plot scale N-addition experiment established on tundra at Ny-Ålesund (Svalbard). Initially, ammonium nitrate (pH adjusted

with nitric acid) was added to plots during 2009 to simulate N-deposition events of 0.4 and 4 kg N ha⁻¹ with subsequent ¹⁵N-labelled amendments to occur during the summer, 2010 (0.4, 4 and 12 kg N ha⁻¹). The fate of ¹⁵N will be followed within the soil, soil microorganisms and in plants. The impact of the acute N-deposition events upon plant diversity and activity will be assessed over the following summer growing seasons, whilst impacts upon microbial community structure (bacteria, archaea, and fungi and specific N-cycling functional guilds), and microbial diversity and abundance is being investigated using molecular (DNA)-based approaches. The datasets obtained will be used within biogeochemical models that simulate the impact of both long-term nitrogen accumulation and short-term dynamics associated with acute pollution events upon plant-soil-microbial systems.

CLIMATE SENSITIVITY OF GREENHOUSE GAS FLUXES IN ORGANIC SOILS

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DEFRA policy relevance: DEFRA has highlighted the importance of sustainable management of natural environments. Increasing demands for energy and food supply could threaten peatland environments. Research into the biogeochemistry of these sensitive ecosystems is vital to inform land management policy under a changing climate.

Peatlands cover only 3% of the Earth's terrestrial surface yet they store around 33% of the total soil carbon (C) pool. A large proportion of peatland ecosystems are located in Northern high latitude regions; areas predicted to be particularly sensitive to impacts of climate change. Potential effects, such as warming and drought, may have important implications on the ability of peatlands to sequester C, potentially resulting in net soil C sinks becoming net C sources.

It has been identified that the net greenhouse gas flux from peatlands is primarily determined by slow rates of decomposition as opposed to high rates of primary production. An understanding of how climate change will influence decomposition rates within these ecosystems is vital for the development of specialised ecosystem C and nitrogen (N) models, and for informing future land management policy.

Here we introduce a study examining the role soil nutrient (C, N and P) status plays in regulating decomposition in peatland soils. We will investigate the interactions that emerge between soil nutrient status and other limiting factors, such as temperature and oxygen availability. In particular we will explore the relationship between C:N:P ratios and the temperature sensitivity of decomposition utilising trace gas measurements and stable isotope approaches in nutrient addition experiments. Through this experimental study we look to identify relationships which could be incorporated into current peatland C and N models, thus allowing further parameterisation of these models for temperature response.

A MECHANISTIC STUDY OF THE IMPLICATIONS OF OZONE AND DROUGHT EFFECTS ON VEGETATION FOR GLOBAL WARMING

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This study has relevance to Defra policy as:

- The interaction between ozone and drought may cause alterations in carbon sequestration of unimproved grasslands.
- Ozone driven disruption of stomatal functioning reduces the ability of plants to respond to drought.

Geranium sylvaticum is a characteristic species of unimproved grasslands. Individual plants were established in 2.5 litre pots filled with Humax topsoil and exposed to either well- or reduced-watered conditions. Six plants from each watering regime were exposed to ozone in each Solardome at the Solardome Facility, CEH, Bangor for 14 weeks during the summer of 2009. The experiment was designed to investigate the effects of increasing background ozone in combination with watering regime. The mean 24 hr ozone exposures across the domes ranged between 25 – 95ppb and followed a rural background profile obtained from Keenly Fell, Northumberland. A full destructive harvest was completed after 14 weeks.

There was no significant effect on above ground biomass in either the well- or reduced-watered treatments with increasing ozone. However, root: shoot ratios were significantly reduced with increasing ozone in the well-watered treatment ($p = 0.002$); there was no significant ozone effect in the reduced-watered treatment.

Impairment of stomatal functioning, due to disruption of ABA signalling, is a reported response of some plant species to ozone toxicity. ABA bioassay studies for *G. sylvaticum* showed that for plants in the reduced-watered treatment, by late season stomata were less able to close to exogenous supplied ABA in the high ozone exposure compared to those from the control ozone exposure. The percentage difference between +/- ABA in the well-watered treatment was also significantly reduced with increasing ozone by week 14.

Response of apparently healthy abscised leaves to severe water stress showed that leaves grown in high ozone had significantly higher rates of transpiration than leaves grown in ambient ozone conditions in both the well – and reduced –watered treatments ($p = 0.021$ and 0.005 respectively).

It has been proposed that disruption of ABA signalling may be a result of ozone driven changes in the pH of apoplastic sap. The pH of apoplastic sap of *G. sylvaticum* in the well-watered treatment significantly increased with increasing ozone in both the 1st and 2nd sap aliquots ($p = 0.03$ and 0.036 respectively).

EXPLORING RELATIONSHIPS BETWEEN N DEPOSITION AND PLANT SPECIES DIVERSITY AND COMPOSITION IN A NATIONAL DATASET

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Defra policy and relevance:

- This study identifies a negative relationship between N deposition and species richness in a number of habitats.
- Mechanisms associated with loss of species richness differ between habitats so mitigation of N deposition should be targeted to habitat type.
- CSM indicator species presence measures an important aspect of the condition of interest features on sites designated for their conservation value; this work identifies an impact from N deposition on habitat condition and attempts to identify indicators that could be used for assessment of condition.

We use national scale data from Countryside Survey to explore relationships between N deposition and plant species diversity and composition. The Countryside Survey is based on a series of stratified, randomly selected 1 km² across Great Britain, surveyed in 1978, 1990, 1998 and 2007. Within each 1km square a series of vegetation plots are located to sample different features of the countryside. This gives a large dataset that can be partitioned and analysed in different ways. This work focuses on habitat level analyses so the data is classified to habitat type. Analyses are based on regression between N deposition and the number of species in a quadrat by a mixed model analysis of variance to determine the impact of N deposition on species richness when other potential explanatory variables are taken into account (Type 1 tests). Explanatory variables included N deposition, S deposition, climatic variables and an indicator representing the degree of grazing as fixed effects.

Further work looks at the change in abundance of Common Standards Monitoring (CSM) indicator species in vegetation plots between 1998 and 2007 (as a measure of ‘appropriate diversity’) and attributes changes in abundance to the above drivers of change including N deposition. We have also attempted to identify indicators of N deposition that could be used in CSM monitoring to assess habitat condition.

Analysis showed that species richness was negatively associated with chronic N deposition in infertile grasslands and heathland. Correlations between changes in CSM indicator abundance and other explanatory variables demonstrated an impact from N deposition on condition in these and other habitat types.