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Predicting effects of N pollutant load on plant species based on a dynamic soil eutrophication indicator

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Cynhaliwyd ymchwil i ddysgu mwy am effeithiau llygredd nitrogen (N) ar laswelltir twyni tywod drwy ddefnyddio model cadwyn, sy’n darogan sut y bydd planhigion yn debygol o ymateb i newidiadau cemegol yn y pridd. Cafodd y model cadwyn ei galibradu gyda’r data a gasglwyd mewn arbrawf a oedd yn cynnwys pori ac ychwanegu N yn Niwbwrch, Sir Fôn. Mi wnaeth y model pridd N14C ddarogan y byddai cynnydd i’w weld yn lefel cynhychiant planhigion ac yn y mewnbynnau carbon C mewn llystyfiant gwyw ar y ddaear wrth i ragor o N gael ei ychwanegu, ac y byddai hyn yn arwain at gynnydd cychwynnol a pharhaol yn y gymhareb C N yn y pridd. Mae hyn yn gwrthgyferbynnu gyda’r rhagfynegiant o leihad yn y gymhareb C N a geir gyda’r model N-dirlawn symlach sy’n cael ei ddefnyddio ar hyn o bryd i gyfrif y lefel o faetholion-N sy’n mynd dros ben y lefel llwyth-critigol. Gwelwyd hefyd bod pob cyfradd o ychwanegiadau N yn creu cynnydd parhaus yn lefel y N sydd ar gael i blanhigion. Drwy ddefnyddio’r modelau cilfach (‘niche’) Multimove ar gyfer rhywogaethau planhigion sy’n nodweddu glaswelltir twyni tywod cafodd y newidiadau hyn yn y pridd eu cysylltu gyda newidiadau yng nghyfoethogiad cyffredinol y fflora gan faetholion, fel y dangoswyd gan sgôr Ellenberg N. Cawsant eu cysylltu hefyd gydag addasrwdd y cynefin ar gyfer rhywogaethau penodol. Dehonglwyd dirywiad yn Addasrwydd Cynefin fel cynnydd yn lefel y risg i’r rhywogaethau. Ar gyfraddau uwch na 30 kg N ha-1y-1 roedd y rhywogaethau mwy sensitif mewn peryg bron yn syth, ond ar gyfraddau llai roedd rhywogaethau yn cael eu peryglu’n ddiweddarach, a pho leiaf oedd yr ychwanegiad N yr hwyraf y deuai’r risg yn amlwg. Ar gyfraddau llai na’r llwyth N critigol ar gyfer twyni calchaidd sefydlog roedd mwy o rywogaethau mesotroffig mewn perygl. Roedd y rhywogaethau hynny yr ystyriwyd eu bod yn ddangosyddion positif o gyflwr cynefin mewn perygl pan oedd cyfraddau ychwanegiad N yn uchel ac isel. Roedd newidiadau mewn Addasrwydd Cynefin o ganlyniad i newid patrwm pori yn cael mwy o effaith dynwaredol ar Addasrwydd Cynefin. Os am feithrin mwy o hyder yn y model cadwyn rhaid mynd i’r afael â’r gwahaniaethau rhwng effeithiau gofodol ac amserol yn sgîl ychwanegiadau N. Byddai’n ddefnyddiol cael rhagor o wybodaeth am effeithiau N ar strwythur llystyfiant a chwymp dail (‘litterfall’) a dylid cynnwys mesuriad gwrthrychol o uchder llystyfiant mewn cynllun monitor, ochr yn ochr â mesuriadau ffloristig. Dangoswyd bod rheolaeth yn allweddol i liniaru effeithiau N er y byddai ymdrechion i symud N drwy dorri neu bori yn annhebygol o wared digon o N i osgoi cyfoethogiad. Gall lleihau uchder llystyfiant arbed rhywogaethau tal cystadleuol rhag bwrw gormod o gysgod dros rywogaethau nodweddiadol twyni tywod sy’n tyfu’n isel ar y ddaear ac yn gorfod cael golau i ffynnu.

2 SUMMARY

The effects of nitrogen (N) pollution on dune grassland were explored using a model chain that predicts how plant species are likely to respond to changes in soil chemistry. The model chain was calibrated to data from an N addition and grazing experiment at Newborough in Anglesey. The N14C soil model predicted increases in plant productivity and plant litter carbon (C) inputs with more N addition, resulting in an initial and persistent increase in soil C/N ratio. This contrasts with predictions of decreasing C/N ratio from the simpler N saturation model currently used to calculate nutrient-N critical load exceedance. All N addition rates also caused persistent increases in plant-available N. Using the MultiMOVE niche models for plant species typical of dune grassland, these soil changes were related to changes in the overall nutrient enrichment of the flora, as indicated by mean Ellenberg N score, and thereby to the habitat's suitability for particular species. Declines in Habitat Suitability were interpreted as increasing risk to the species. At rates above 30 kg N $ha^{-1}y^{-1}$, the more sensitive species were placed at risk almost immediately, but at smaller rates species were placed at risk later on, with an increasing delay with less N addition. At rates lower than the critical N load for calcareous fixed dunes, more mesotrophic species were placed at risk. Species viewed as positive indicators of habitat condition were placed at risk under both high and low rates of N addition. Changes in Habitat Suitability due to changed grazing regime had greater simulated effects on Habitat Suitability. For more confidence in the model chain, differences between the spatial and temporal effects of N addition need to be addressed. More information on the effects of N on vegetation structure and litterfall would be very useful, and objective measurements of vegetation height should be included in monitoring schemes alongside floristic recording. Management was shown to be critical for mitigating the effects of N. Although N removal through grazing or mowing is unlikely to export sufficient N to prevent enrichment, reducing vegetation height can prevent competitive species shading out the more distinctive low-growing, light-demanding dune species.

3 INTRODUCTION

Increased loads of reactive nitrogen (N) due to anthropogenic pollution have been implicated as a major cause of biodiversity loss at global scale (Phoenix, Hicks *et al*., 2006; Bobbink, Hicks *et al*., 2010). However, when individual sites of conservation concern are monitored, N pollution is rarely cited as a factor causing damage. Agencies concerned with environmental protection require predictions of how much damage a given amount of N pollution is likely to cause, for use in assessing the impacts of local development. The current study uses a predictive model chain to assess effects of N pollution on species on a calcareous stable dune grassland site at Newborough, Anglesey.

Effects of N on ecosystems are often assessed in relation to the Critical Load, defined as a "quantitative estimate of an exposure to one or more pollutants below which significant harmful effects on specified sensitive elements of the environment do not occur according to present knowledge" (Nilsson and Grennfelt, 1988). The likelihood of harmful effects at different rates of N deposition has been assessed using artificial additions to field experiments or mesocosms (*e.g*. Ten Harkel and Van der Meulen, 1996); by targeted surveys of a particular habitat along a deposition gradient (*e.g*. Jones, Wallace *et al*., 2004; Remke, Brouwer *et al*., 2009); and more recently by making use of non-targeted but large floristic datasets (Stevens, Smart *et al*., in prep.).

Criteria for defining harmful effects of N on ecosystems have been defined as loss of sensitive species, or chemical changes in indicators such as N concentration in plant tissue, mineral N concentration in soil solution, or N leaching flux. Changes in species composition are more directly related to conservation objectives. The evidence for harmful effects of N in stable dune grasslands was recently reviewed during a revision of critical loads set for European habitats (Bobbink and Hettelingh, in press). Large experimental additions of N (80 kg N ha^{-1} y^{-1} or more) have caused a reduction in species richness (Willis, 1963) or changes in species composition (Boorman and Fuller, 1982). However, experimental additions of more realistic amounts of N to dune grasslands have resulted in no loss of species within the timescale studied (Ten Harkel and Van der Meulen, 1996; Plassmann, Edwards-Jones *et al*., 2009). Targeted surveys of dune grasslands along N deposition gradients show clearer effects on species. In a study of Baltic dune sites with comparatively low rates of N pollution, the cover of the graminoid *Carex arenaria* increased with N deposition rate, and although no direct relationship of N deposition rate with species richness was observed, a decrease in the cover of short, lichen-rich vegetation was considered likely to result in species loss (Remke, Brouwer *et al*., 2009). A survey of dune grassland sites in England and Wales showed a negative relationship of species richness with N deposition rate (Jones, Wallace *et al*., 2004).

The contrast between experimental and survey evidence is probably due to differences in the timescale of N addition. Amounts of extra N applied in realistic experiments are small relative to the cumulative addition of pollutant N since the 19th century. Current N deposition is likely to be well-correlated with historic rates of N deposition, except where local facilities such as chicken or pig farms have been built recently, and so small differences in current deposition rate reflect large differences in cumulative deposition. Cumulative N deposition has been shown to be strongly negatively related to several biodiversity indicators such as high proportional cover of forbs, and low proportional cover of grasses (Dupre, Stevens *et al*., 2010).

Critical Loads can be calculated using a steady-state mass balance approach, in which input fluxes are balanced against export fluxes such as denitrification and an "acceptable" rate of immobilisation into soil organic matter. Cumulative and historic effects are not easily incorporated into the steady-state mass balance approach, but can be considered using dynamic modelling. For example, where the critical load has been exceeded, a model of soil N dynamics can be used to calculate a target load which will allow recovery (defined using a chemical criterion) by a particular date. However, there remains a need to link changes in soil conditions to the occurrence of species of conservation concern.

Capacity for forecasting effects of atmospheric N pollution on biodiversity has recently been developed by linking dynamic models of soil chemistry with regression models of plant species occurrence (de Vries, Wamelink *et al.*, 2010). For example, the Very Simple Dynamic (VSD) soil model (Posch and Reinds, 2009) has been used to drive the GBMOVE set of species niche models (Smart, Scott *et al.*, 2010) to determine changes in Habitat Suitability for Common Standards Monitoring indicator species on a blanket bog site (Rowe, Emmett *et al.*, 2009). Several niche occupancy models of this type are being assessed by the CCE[1] for predicting effects of N pollution on biodiversity at European scale (CCE, 2011), although testing of the approach has so far been limited.

The VSD model uses a simple breakthrough model (Gundersen, Callesen *et al.*, 1998) to predict N accumulation, which essentially increases the total amount of N in soil until a threshold C/N ratio is reached where some of the added N begins to leach. The fact that different soils begin leaching at different C/N ratios can be allowed for by using different threshold values for different soil types (Rowe, Evans *et al.*, 2006), but the VSD model does not simulate variable turnover rates of soil organic matter, or increases in plant litter inputs due to N fertilisation effects. For this reason, the N14C model (Tipping, in prep) was applied in the current study. This model explicitly includes effects on plant production, and variable-sized pools of organic matter with different turnover rates.

The GBMOVE species niche models (Smart, Scott *et al.*, 2010) were developed using multiple logistic regression to fit presence/absence data to environmental factors, and thus predicts the likelihood of a species being present under a given set of conditions. Since the actual likelihood of finding a given species on a site depends also on its occurrence in the local species-pool, and rates of dispersal and extinction, this likelihood of presence is termed Habitat Suitability. The statistical approach used to fitting Habitat Suitability has been re-evaluated since publication of the GBMOVE model, and a version which uses three separate fitting methods (generalised linear models, GLM; generalised additive models, GAM, and multivariate adaptive regression splines, MARS) to make an ensemble forecast has been developed, known as MultiMOVE. By averaging the predictions obtained using these three methods, the uncertainty due to choice of statistical model is likely to be reduced.

Both versions of the species niche models (GBMOVE and MultiMOVE) use mean Ellenberg 'N' score (E_N) as the environmental factor that summarises the effects of nitrogen. Scores were assigned to vascular plant species occurring in Central Europe by Ellenberg (Ellenberg, 1974; Ellenberg, Weber *et al.*, 1992) to represent their occurrence in relation to different environmental gradients. The 'N' score reflects the fertility of a site where the species is likely to be found. However, Ellenberg did not clarify whether E_N represented occurrence in relation to nitrogen availability, or to the availability of other nutrients and production-limiting factors. Several studies have suggested that E_N score reflects the overall fertility and plant productivity of a typical location for the species, rather than exclusively nitrogen availability (Hill and Carey, 1997; Schaffers and Sykora, 2000; Rowe, Emmett *et al.*, 2011). However, site productivity is likely to be correlated with nitrogen availability, at least in nitrogen-limited systems. The mean E_N score for present species provides an integrated indicator of

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[1] CCE: Coordination Centre for Effects of the International Cooperative Programme on Modelling and Mapping of Critical Loads and Levels and Air Pollution Effects, Risks and Trends of the United Nations Economic Commission for Europe

site productivity (Diekmann, 2003). The current study used Ellenberg scores as modified for use in the UK by Hill *et al*. (1999).

A critical step in combining dynamic biogeochemical models with species niche models is to translate the abiotic outputs from the biogeochemical model into a mean E_N score that can be interpreted by the niche model. This step was originally calibrated by relating the mean E_N score in a subset of plots (those with complete soil data) from the floristic dataset provided by the Countryside Survey in 2000 (Firbank, Barr *et al*., 2003) to soil total N and total C concentrations. However, these bulk soil properties only partially explain the observed variation in mean E_N score, presumably because much of the organic matter is rather unreactive and its N content makes little difference to plant N exposure. For this reason, a new measure of available (mineralisable) N was included in the most recent Countryside Survey (Emmett, Reynolds *et al*., 2010). In a pilot study, this measure proved to be a more accurate predictor of mean E_N than bulk soil properties alone (Rowe, Emmett *et al*., 2011).

Sand dunes are a habitat of conservation importance, supporting vascular and lower plant species which typically require high levels of ground-level light and are susceptible to damage by N pollution. Critical loads of N for dune grassland have been set separately for acid and calcareous stable dunes (both EUNIS class B1.4) and for shifting coastal dunes (EUNIS class B1.3) (Bobbink, Braun *et al*., 2010). In the UK, the values now being used for critical loads mapping are 9 kg N ha^{-1} y^{-1} for acid stable dune grassland, 12 kg N ha^{-1} y^{-1} for calcareous stable dune grassland, and 10 kg N ha^{-1} y^{-1} for stable dune grassland of unknown pH (Jane Hall, CEH, pers. com.). A site with calcareous dune grassland was chosen for the current study, partly for reasons of data availability, but also because calcareous dune soils are buffered against acidification and so N effects are principally via nutrient addition rather than acidification.

The aims of the study were:

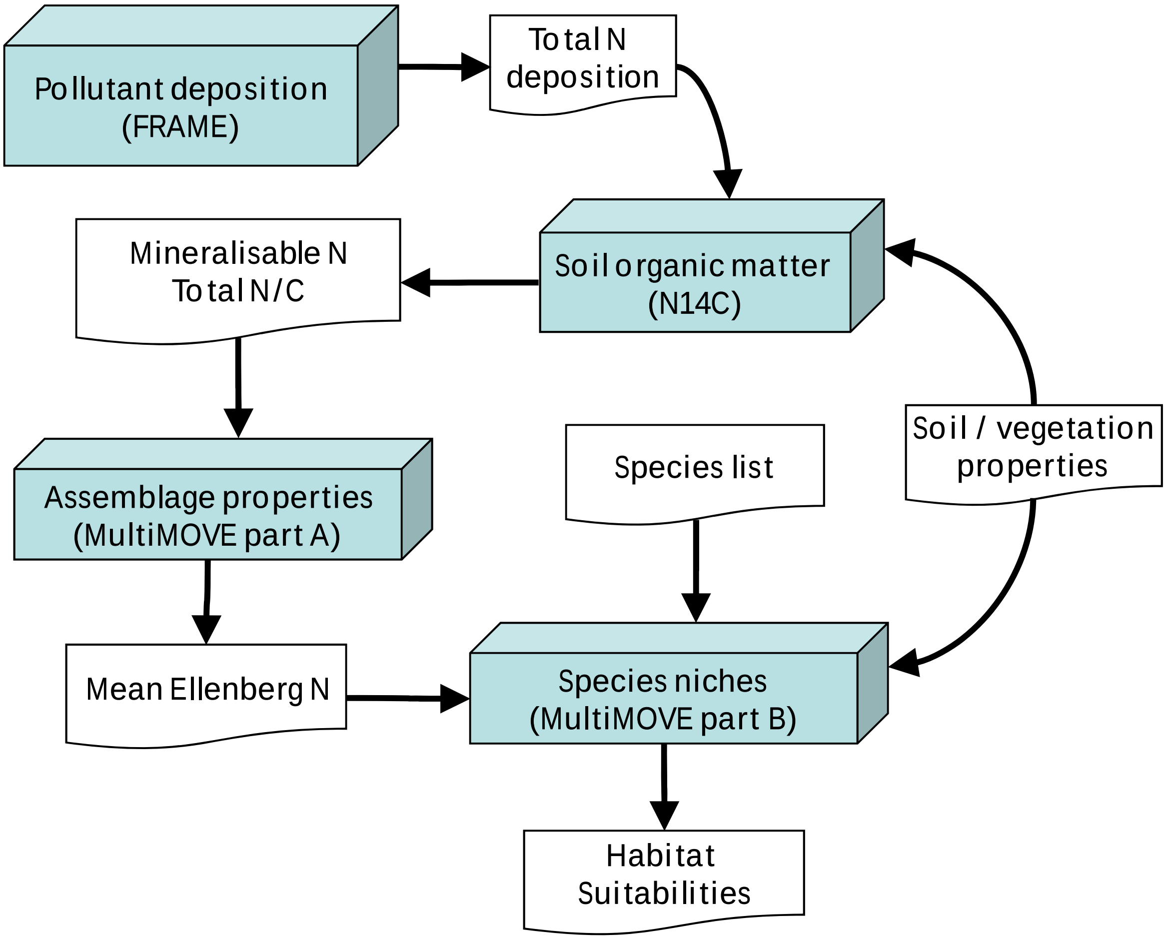
a) to explore the use of mineralisable N as a predictor for mean Ellenberg N score in the dataset from Countryside Survey in 2007;
b) to explore the ability of the N14C model to predict the rate of N mineralisation;
c) to combine the N14C and MultiMOVE models to predict Habitat Suitability for a set of species typical of dune grassland, under different N pollution scenarios;
d) to explore methods for interpreting predicted Habitat Suitability.

4 METHODS

4.1 Niche occupancy model chain

The models used to predict effects of N deposition on Habitat Suitability for dune grassland species, and the quantities passed at each stage, are illustrated in Figure 1. Best estimates for the actual influx of pollutant N were obtained using the FRAME model (Dore, Kryza *et al*., 2009), and other pollution scenarios were derived from these estimates. The N14C model (Tipping, in prep) was used to predict changes in soil mineralisable N from total N deposition and site characteristics. Soil mineralisable N was used to drive predictions of change in the floristic assemblage, as represented by mean E_N score. These changes in mean E_N, together with other site characteristics including vegetation height, were used by the MultiMOVE model to calculate Habitat Suitability for a selected set of species. The stages in the model chain are described in more detail below.

Figure 1. Models (shaded boxes) used to determine effects of N deposition on Habitat Suitability for a set of plant species, showing quantities (unshaded sheets) passed at each stage.



4.2 Newborough experiment

An experiment was set up in 2003 at Newborough (Figure 2) to assess the combined effects of grazing and nutrient addition on plant species composition and soil and vegetation processes (Plassmann, Edwards-Jones *et al*., 2009). Grazing was manipulated using exclosures, resulting in treatments that were ungrazed (by larger herbivores), rabbit-grazed, and rabbit-and-pony grazed. Three levels of N addition (+0, +7.5 and +15 kg N ha^{-1} y^{-1}) were applied in a fully factorial design. Phosphorus addition treatments were also included but are not considered in the current study. Background levels of N deposition at the experimental site are around 11.6 kg N ha^{-1} y^{-1} (Jones, 2010), so the N addition treatments correspond to a total of 11.6, 19.1 and 26.6 kg N ha^{-1} y^{-1}.

Figure 2. Newborough experiment, showing a) nitrogen addition, and b) rabbit-and-pony grazing exclosure. Photos: Laurence Jones.

Results from the Newborough experiment were used to set up the model chain. Perhaps surprisingly, neither inorganic N leaching flux nor mineralisable N as measured in 2009-2010 was significantly affected by N application treatment. Grazing treatments did affect both of these measurements – cessation of grazing by ponies and rabbits decreased inorganic N leaching but increased the N mineralisation rate (Table 1). The decrease in leaching is likely due to the decreased patchiness of N inputs, which can exceed plant and immobilisation demands in excreta patches in the grazed system. Reasons for the increase in mineralisable N are less clear, but two theories emerge from studies in the Netherlands. One is that mineralisation rates in non-agricultural systems are driven by the quantity (and quality) of litter inputs as a substrate for microbial decomposition which are higher in ungrazed systems (Aerts and de Caluwe, 1997; van Wijnen, van der Wal *et al.*, 1999). The second relates to the type of grazer: in ungrazed systems grazing still occurs but by voles rather than larger herbivores. Vole biomass can equal that of larger grazers; they produce small, rapidly decomposable and widely distributed dung, therefore stimulating decomposition rates uniformly (Bakker, Olff *et al.*, 2004). Similar effects of grazing on mineralisation have been observed in UK saltmarsh (Olsen, Dausse *et al.*, 2011) and dunes in the Netherlands (Maarten Schama, pers. comm.).

Table 1. Effects of grazing treatments at Newborough on inorganic nitrogen leaching and mineralisable N. Mean values for N addition treatments.

	Inorganic N leached	Mineralisable N
	kg N ha^{-1} y^{-1}	
Pony & rabbit grazed	1.04	4.65
Ungrazed	0.22	8.16

4.3 Nitrogen deposition rates and scenarios

Actual rates of N deposition at the study site were calculated from measurements of deposition fluxes in 1997-8, which were 3.3 kg NOx-N ha^{-1} y^{-1} and 4.9 kg NHy-N ha^{-1} y^{-1} from atmospheric sources, plus 3.4 kg NHy-N ha^{-1} y^{-1} from a nearby chicken unit (Jones 2010). Deposition from atmospheric sources was rescaled for the period 1840-2000 using historic scenarios generated in the GANE project. Future rates of atmospheric N deposition were estimated by rescaling measured atmospheric N deposition to trajectories based on FRAME model (Dore, Kryza *et al.*, 2009) runs for the Department of Energy and Climate Change's UEP30 scenario (DECC, 2009).

The extra N from the chicken unit was assumed to have been constant after 1972 when the unit was set up. This local N addition was used as the basis of different management scenarios in the current study. These were designed to compare less polluted scenarios (in which emissions from the chicken unit were reduced to zero in 2012) and more polluted scenarios (in which the chicken unit's emissions increased 2-, 4- or 10-fold), with the best estimate for the actual level of N deposition. An additional scenario was added in which N deposition was maintained throughout at the GANE estimate for pristine deposition at the site (Table 2, Figure 3a). As well as these scenarios related to chicken farm emissions, scenarios were run with actual N deposition until 2011 and then simple increments of N load (Figure 3b). These incremental scenarios were used to define individual species' sensitivity (Annexe Table 3), and to illustrate numbers of species at risk (Figure 12).

Table 2. Description of nitrogen pollution scenarios related to chicken farm emissions.

Scenario	Rationale	Calculation
Pristine	Never any N pollution	GANE estimate for pristine deposition (1.8 kg N ha^{-1} y^{-1})
Actual	Best estimate for actual deposition	Measured deposition scaled with GANE/FRAME past/future trajectories
ZeroCU	Zero emissions from nearby poultry unit after 2012	As 'Actual', minus 3.4 kg N ha^{-1} y^{-1} from 2012
CUx2	Small expansion of nearby poultry unit	As 'Actual', plus 3.4 kg N ha^{-1} y^{-1} from 2012
CUx4	Large expansion of nearby poultry unit	As 'Actual', plus 10.2 kg N ha^{-1} y^{-1} from 2012
CUx10	Very large expansion of nearby poultry unit	As 'Actual', plus 33.2 kg N ha^{-1} y^{-1} from 2012

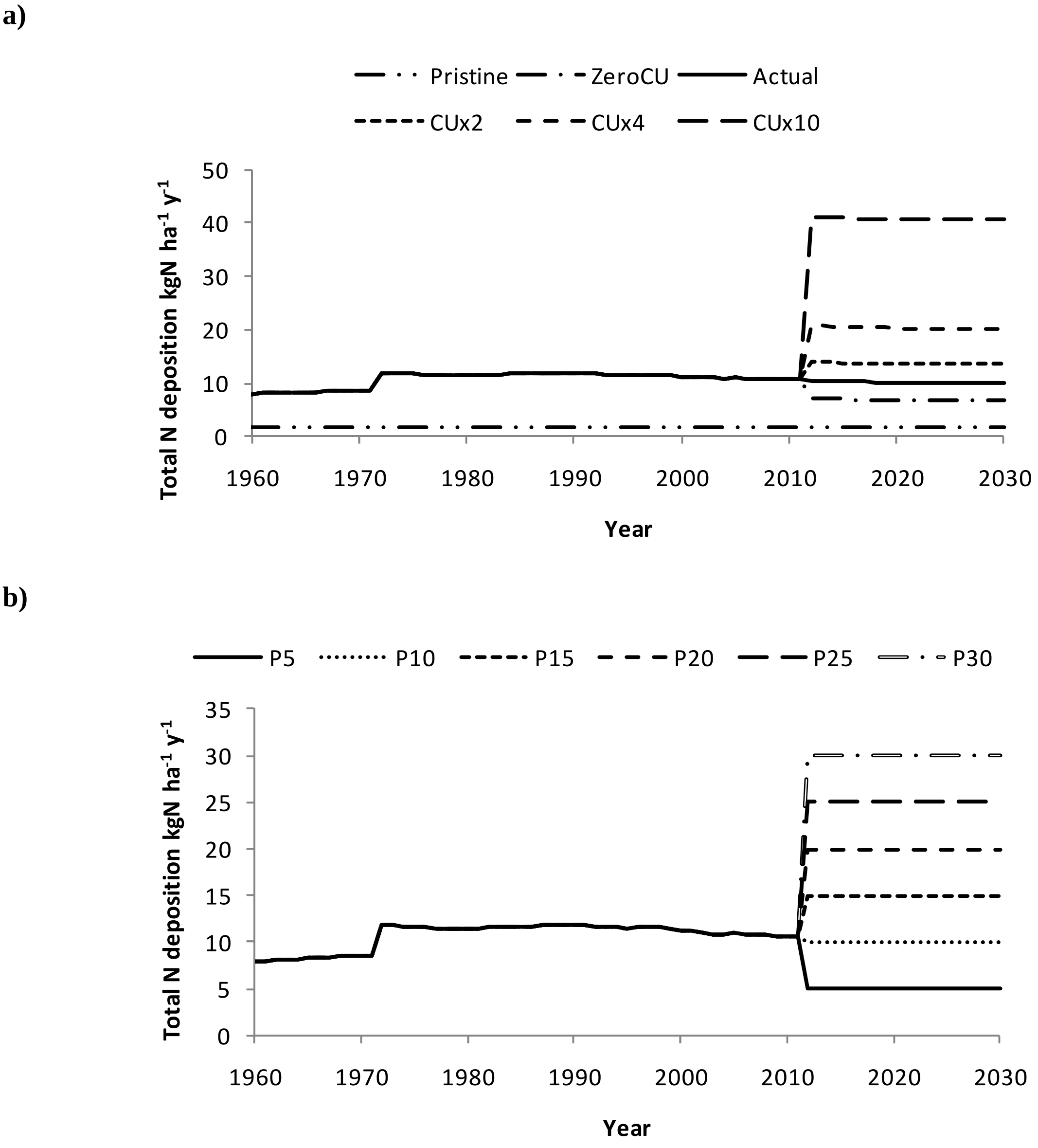


Figure 3. Rate of nitrogen deposition 1960-2030 under the scenarios studied: a) scenarios related to chicken farm emissions; b) simple increments of nitrogen load.

4.4 Adapting the N14C model to predict mineralisable N dynamics

The N14C model (Tipping, in prep) was developed to predict effects of N pollution on soil C and N dynamics, under the Defra Critical Loads and Dynamic Modelling project (Evans, Hall *et al.*, 2010). The model has a more detailed treatment of organic matter turnover than the single-pool ‘breakthrough’ N model used in the VSD and MAGIC models, with three organic matter pools with different turnover rates (Figure 4). The N14C model is however highly constrained compared to other detailed soil C and N models, and uses the average age of soil carbon (obtained by ^{14}C dating) to fix the ratio between the two main organic matter pools. The aim was to develop a model sufficiently detailed to reproduce short-term fluctuations in available N in response to changes in N deposition, yet with few enough parameters to allow application to sites where data are sparse.

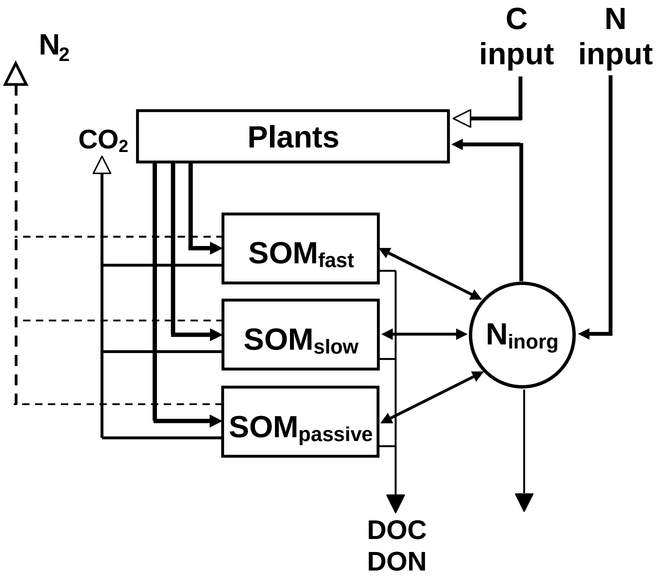


Figure 4. Carbon and nitrogen pools in the N14C model. SOM = Soil Organic Matter, split into three conceptual pools with different turnover rates; DOC = Dissolved Organic Carbon; DON = Dissolved Organic Nitrogen; N_{inorg} = inorganic N that is available for plant uptake and for immobilisation into soil organic matter.

The N14C model is based on annual average fluxes, and predicts changes in pools of C and N that have different turnover rates. The model calculates the inorganic N available per year as the annual release of inorganic N from these N pools, plus annual N deposition, minus denitrification and losses of dissolved organic N. Inorganic N available for leaching is calculated after plant N uptake and any re-immobilisation into soil N stocks. In the original formulation of the model, all available N was taken up by plants until their demand was satisfied, at which point all remaining available N was taken up into soil organic matter until its immobilisation demand was satisfied. This resulted in a predicted rate of inorganic N leaching of zero even after substantial cumulative N additions. This was considered unrealistic, and the measured rate of inorganic N leaching (mean value for all experimental treatments; MLM Jones, unpublished data) was used to calibrate two additional terms in the model that set the maximum proportion of available N that can be taken up by plants, and the maximum proportion of the remaining available N that can be immobilised into soil organic matter.

To compare model outputs with mineralisable N rates measured in the laboratory, it is necessary to consider which soil N fluxes were changed when cores were removed from the soil and incubated. Plant material was removed before incubation, and this took place in the dark, so plant demand may be considered to be zero. Denitrification is likely to have continued, and dissolved organic N was not measured. Hence the measured flux of mineralisable N is comparable to the modelled values for the release of inorganic N from the soil N pools, minus denitrification, minus re-immobilisation into soil N stocks, but with zero plant N uptake. Modelled fluxes are calculated in kg N m^{-2} y^{-1}. Mineralisable N flux was measured over a 28 day incubation in a 3.5 cm diameter soil core from 0-15 cm depth, and was converted to kg N m^{-2} y^{-1} for comparison.

To ensure that the N14C model predicted values for key output variables with reasonable accuracy, several parameters (Table 3) were calibrated using the Generalized Reduced Gradient solver algorithm in Microsoft Excel to minimise an overall error term. This term was calculated as the sum of root square errors between predicted values and four measurements, normalised to a similar scale by dividing by the measured value, with the following weightings:

- Soil total C/N ratio (weighting 10)
- Mineralisable N (weighting 10)
- Inorganic N leaching flux (weighting 1)
- ^{14}C concentration in soil organic matter (weighting 1)
- Soil total C pool (weighting 1)

Table 3. Calibrated N14C parameters.

N14C parameter	**Fitted value**
Maximum N uptake by plants, kg N ha^{-1} y^{-1}	226
Dissolved organic N as proportion of N released from soil organic matter	0.0774
Maximum C/N ratio in plant litter	37.2
Proportion of plant litter C entering "slow" organic matter pool	0.304
Proportion of plant litter C entering "passive" organic matter pool	0.050
Maximum proportion of available inorganic N that can be taken up by plants	0.961
Maximum proportion of remaining inorganic N that can be immobilised into soil organic matter	0.536

The model was calibrated to measurements from the pony-and-rabbit grazed system, since this is the current status of the majority of the site. An attempt was also made to simulate the observed changes to inorganic N leaching and mineralisable N in the ungrazed system, by adjusting (after 2003 when this grazing treatment was imposed) the maximum proportion of available N that can be taken up by plants, and the proportions of plant litter C entering the slow and passive pools. It proved impossible to achieve both an increase in mineralisable N and a decrease in N leaching using these parameters. As discussed in section 0, the measured decrease in leaching in the grazing exclosure is likely due to a decrease in excreta patches, and the increase in mineralisable N may be due to extra litter inputs or to more widely distributed excreta from voles. Further detail could be added to N14C to simulate the effect of input patchiness, but this was considered beyond the scope of the current study. Differences between grazed and ungrazed systems were therefore simulated in the model chain only by changing the height of the vegetation, and not by adjusting the biogeochemical model.

4.5 Predicting mean Ellenberg N from soil N properties

In the original GBMOVE model (Smart, Scott *et al.*, 2010), mean E_N score was related to soil total N and total C concentrations (and also pH and moisture content). Total N and C concentrations change only slowly, and are of limited value as predictors of mineral N leaching (Rowe, Evans *et al.*, 2006), implying that they are poor indicators of N availability. Measurements of mineralisable N have been shown to improve prediction of mean Ellenberg N (Rowe, Emmett *et al.*, 2011).

Mineralisable N was measured in soil cores taken from 699 Countryside Survey plots in 2007 (Emmett, Reynolds *et al.*, 2010), and converted to a mineralisation flux per m^2 per year. Floristic data were also recorded for these plots. The mean E_N score was calculated, based on the species present, without cover-weighing. This mean E_N score was related to abiotic measurements of soil N status using multiple regression. Explanatory terms in this regression were included only if their elimination caused a significant reduction in explained variation and a decrease in model efficiency as measured by Akaike's Information Criterion. Some judgment was used as to which of the variables with significant effect to include as explanatory variables. The main axis of spatial variation in E_N across the UK is from organic, acid, wet soils to mineral, neutral to alkaline, drier soils, with E_N generally decreasing with increased C content. However, one of the principal effects of increased N deposition forecast by the N14C model was an increase in soil C (see Results section), and so using C concentration as an explanatory variate would result in decreased E_N with increased N deposition. This contradicts evidence from recent analysis of large spatial and temporal datasets (Emmett, Stevens *et al.*, in prep.) that larger rates of N deposition are associated with increases in mean E_N. The difference between spatial and temporal variation in soil total C will be discussed later.

The multiple regression model derived from analysis of 2007 CS data to predict the unweighted value of E_N was:

$$E_N = 1.689 + 0.318 * \log_{10}(\text{Nmbl}) + 0.0284 * \text{N/C} + 1.105 * \text{Intensive}$$

where:
Nmbl is the mineralisable N flux in g N m^{-2} y^{-1}; N/C is the soil total N / total C ratio in mg N g^{-1} C, and ‘Intensive’ is a binary term which is set to 1 for more intensively managed agricultural habitats, *i.e.* Broad Habitats 4 (Arable), 5 (Improved Grass) and 6 (Neutral Grass), with the exception of Neutral Grass Priority Habitats i.e. unimproved hay meadows. The term was set to zero for Neutral Grass Priority Habitats and all other habitats. This model explained 70.2 % of the variation in mean E_N in the Countryside Survey 2007 dataset (Figure 5). Using measured data the model underpredicted mean E_N, compared to the measured value, by 0.031 units. To ensure that the predicted mean E_N (and thus predicted suitability for individual species) matched actual current floristics, E_N values were predicted in the current study using the following equation:

$$E_N = 1.689 + 0.318 * \log_{10}(\text{Nmbl}) + 0.0284 * \text{N/C} + 1.105 * \text{Intensive} + 0.031$$

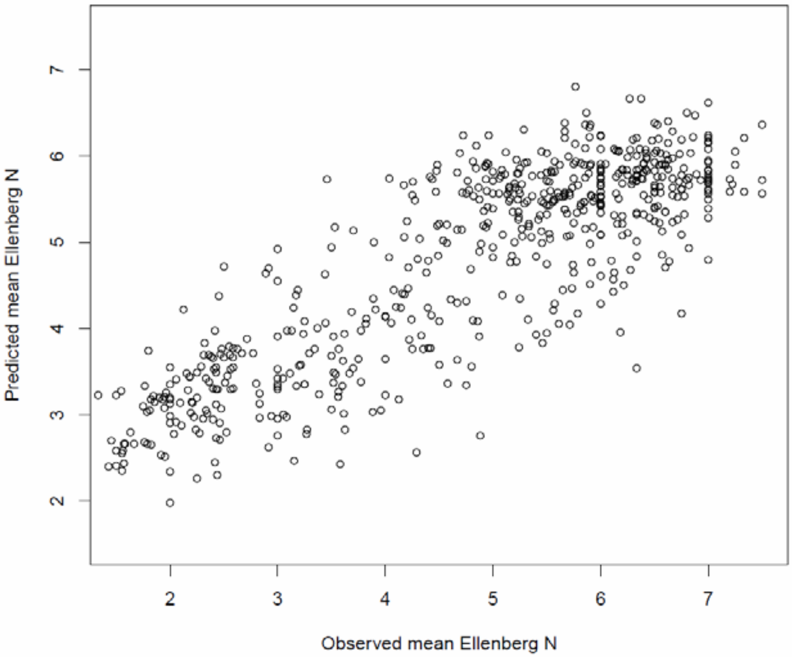


Figure 5. Measured mean Ellenberg N score in the 2007 Countryside Survey dataset plotted against values predicted using the equation E_N= 1.689 + 0.318 * $\log_{10}$(Nmbl) + 0.0284 * N/C + 1.105 * Intensive.

4.6 Predicting Habitat Suitability from mean Ellenberg N and vegetation height

The GBMOVE model of UK plant species’ niches (Smart, Scott *et al.*, 2010) predicts Habitat Suitability for each species as a function of a set of environmental factors, one of which is mean Ellenberg N score. Main effects, quadratic effects and interactions between pairs of factors are included, to represent responses such as a simple increase or decrease with a factor, maximum occurrence at an intermediate level, and changing response to a factor depending on the level of another factor. The GBMOVE niche models were obtained by fitting multiple logistic regression surfaces to the occurrence data in several large floristic datasets from the UK.

Recently the GBMOVE work has been updated, using several different statistical methods (MARS, GAM and GLM) to fit species' response curves. This ensemble approach is more robust than using a single method, and the degree of consistency obtained using the different methods provides a measure of the uncertainty in the Habitat Suitability prediction. The ensemble model, known as MultiMOVE, is implemented in R statistical software, and calculates mean Habitat Suitability and standard deviation, based on the predicted suitability using regression models fitted using the different methods. Different modelling approaches make different assumptions and have different restrictions, some being very flexible while some force a linear response. However, flexible models can be influenced by a small subset of the sample. When modelling a sample of the population it is unknown which approach should be favoured, and therefore the more models used, the greater the understanding of the data and the less likely it is that false inferences will be made.

The probability of occurrence predicted by GBMOVE or MultiMOVE depends on values of environmental factors, but also on the prevalence of the species under optimum environmental conditions. Species which occurred in most quadrats that were near the optimum condition for the species have a greater predicted probability of occurrence than species that occurred in a small proportion of quadrats even when conditions were optimal. To allow comparison between species with differing overall prevalence within the training dataset, Habitat Suitability was calculated as P/P_{max}, where P is the probability of occurrence as predicted by MultiMOVE, and P_{max} is the maximum of the probabilities of occurrence calculated for the species on each of the Countryside Survey plots for which complete soil data are available.

Examples of the output from the MultiMOVE model are shown in Figure 6. The plot for *Cladonia rangiformis* shows reasonable agreement among the different curve fitting methods above a mean E_N score of around 2, although the fits differed below this point. There were few samples in the training dataset with such low EbN, and the differences between the models demonstrates the low sample number and the greater uncertainty below this point. In the plot for *Carlina vulgaris*, the MARS fit showed little responsiveness to mean EN score, presumably because this fitting method favoured other explanatory variables such as canopy height. Looking at MultiMOVE fits for all the species with models, there was no consistent tendency for one method to fit greater or lesser Habitat Suitability. Using the average of the fitted curves reduces uncertainty due to the choice of fitting method, and is likely to provide a good estimate of the true response, although we would still expect high uncertainty where the sample size is low.

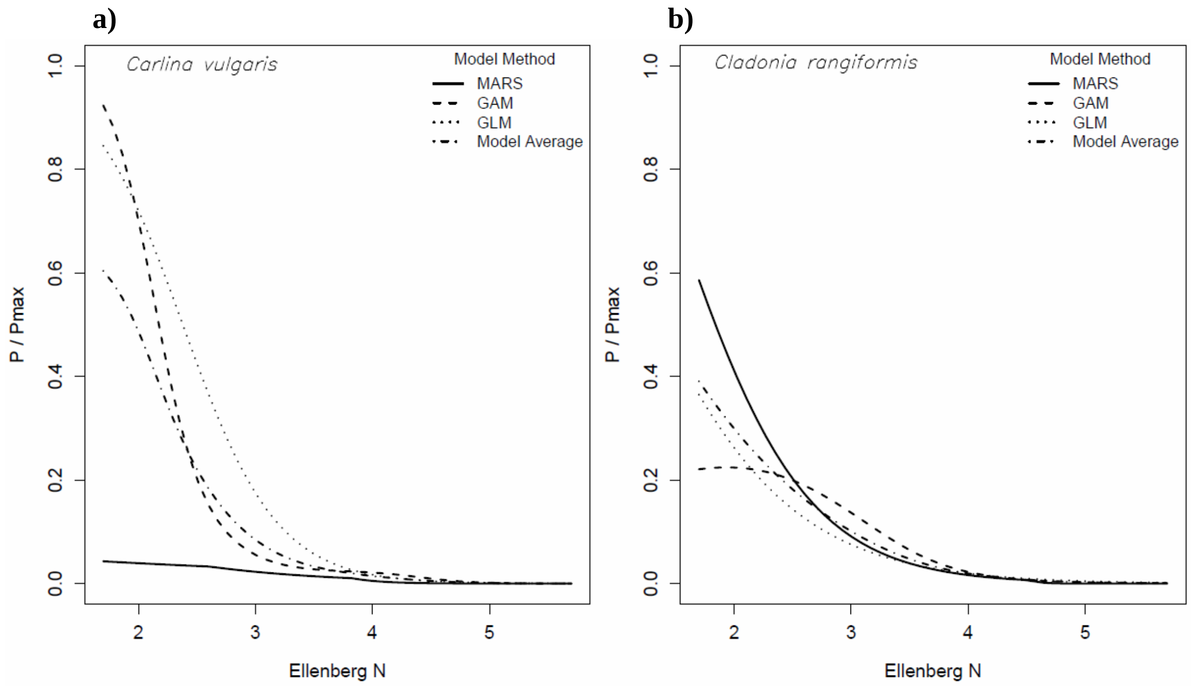


Figure 6. Habitat Suitability for a) *Carlina vulgaris* and b) *Cladonia rangiformis* as predicted by MultiMOVE in relation to unweighted mean Ellenberg N score, for measured values of other abiotic and/or floristic variables at Newborough. Habitat suitability is assessed as P/Pmax *i.e.* probability of occurrence / maximum probability of occurrence in all Countryside Survey plots for which complete soil data are available. Three separate predictions are made using different statistical methods, and can be seen as indicating the uncertainty range due to restrictions and assumptions made in the model method.

The MultiMOVE model uses a proxy measure for vegetation height derived from species composition, since the measurements of vegetation of height in the training dataset were considered insufficiently reliable. Typical heights for all UK species have been defined using an ordinal scale (Grime, Hodgson *et al*., 1988), and MultiMOVE uses the mean of the values on this scale for present species as an indicator of vegetation height. This allows height to be estimated from floristic data where measured height is not available. However, since measured vegetation heights were available from the different grazing treatments at Newborough, and since the absence of consistent changes in species composition meant that all grazing treatments had similar estimated height, a conversion from measured height to Grime's scale was developed. By fitting the median height in each of Grime's classes to the number of the class (Figure 7), the following relationship was obtained:

Grime height score = Maximum(1, 1.17*Ln(Height) - 1.22)

where Height is the measured vegetation height in cm. Mean measured height values used in the current study were 23.2 cm for ungrazed plots and 6.2 cm for grazed plots (Laurence Jones, pers com.) corresponding to height scores of 2.47 and 1.0, respectively.

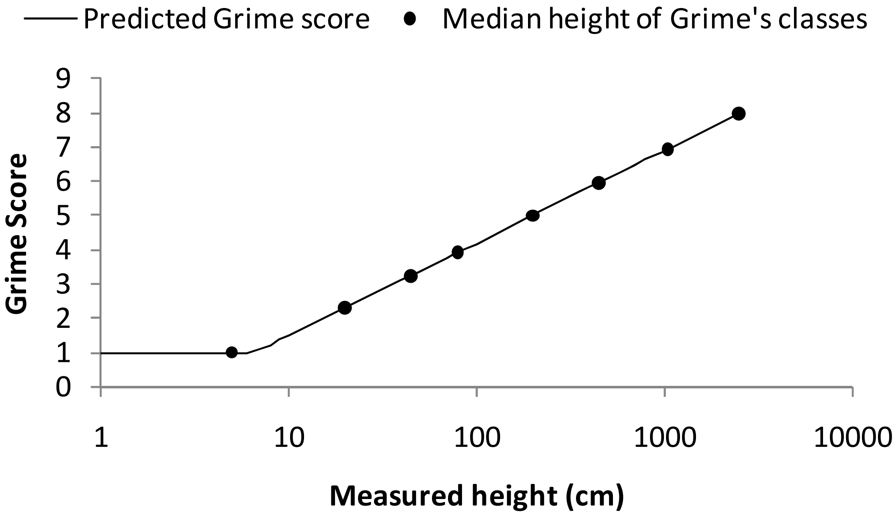


Figure 7. Relationship between measured height and Grime's ordinal height scale (adapted from Grime, Hodgson *et al.*, 1988).

4.7 Selecting species for study

Species were selected for inclusion in the study in two stages, first identifying those which were associated with the habitat on the study site, and then selecting those for which a MultiMOVE niche model exists. Species considered associated with the habitat were those which:

a) have been recorded on the study site;
b) are listed as typical species or as negative condition indicators in the Common Standards Monitoring (CSM, JNCC, 2004) guidance for calcareous fixed dune grassland; or
c) were shown to be affected by N pollution in either lowland acid grassland or lowland calcareous grassland in a study of broad-scale surveillance datasets (Emmett, Stevens *et al.*, in prep.), and are likely to occur in dune grassland.

The latter category was established because it was thought useful to examine species with known susceptibility to N pollution, even though this susceptibility had not been formally evaluated within the specific habitat of dune grasslands. To reduce the set of susceptible lowland grassland species to those likely to occur on dune grassland, only those species where habitat descriptions in Stace (1991) mentioned "sand", "dune", or "coastal" / "sea" were included (Annexe Table 1).

Of the species selected as suitable for study (**Annexe Table 2**), only four were considered to indicate negative conditions, due to their inclusion as negative site condition indicators (*Arrhenatherum elatius*, *Crataegus monogyna* and *Prunus spinosa*) or their positive response to N pollution (*Hylocomium splendens*). Of the remaining 90 species, 33 were listed as typical species in CSM guidance for the habitat (JNCC, 2004), and 13 have been shown to be negatively affected by N pollution (Emmett, Stevens *et al.*, in prep.).

5 RESULTS

5.1 Does N14C reproduce observed soil properties at the site?

Nitrogen addition treatments at Newborough had no significant effect on either soil total C/N ratio or mineralisable N, which limits the amount of testing of the N14C model that can be carried out. Model parameters were calibrated to minimise deviation from measured values, with strong weighting for the soil total C/N ratio, and for mineralisable N content. It is therefore unsurprising that predictions using the actual deposition scenario matched measurements for these properties (Figure 8a & b). Smaller weightings were used to assess error in predictions of inorganic N leaching flux and of total soil C. The N14C model was not able to simultaneously achieve an accurate prediction of total soil C and total soil N (Figure 8c & d). The model did however reproduce measured inorganic N leaching flux with reasonable accuracy (Figure 9).

The trajectories of change under the different scenarios reflected the amount of N added – for example, mineralisable N flux was elevated in the actual scenario compared to the pristine scenario, and progressive increases in N load increased the mineralisable N flux (Figure 8b). However, increasing N load also increased soil total C (Figure 8c), which lead to some perhaps counterintuitive effects on total C/N (Figure 8a). The effect of additional N can be seen clearly in the most extreme scenario (CUx10): a stimulation of plant growth leads to greater litter inputs, and since even litter produced by N-rich vegetation has a greater C/N ratio than soil organic matter, this leads to an initial increase in soil C/N. After some 20 years, other effects of the extra N (decreased litter C/N and increased immobilisation into organic matter) result in a decline in C/N, but it takes around a further 30 years for the C/N ratio to return to the value at the start of the extra addition. The same process takes place in the less extreme scenarios, but over a longer timescale, and none of the other scenarios resulted in a return to the C/N ratio which existed at the start of extra N additions within the timescale considered. This long lag also explains why soil under the pristine scenario had lower C/N than in the actual scenario – the stimulation of C inputs caused by industrial and agricultural emissions of N since the 19th century still outweighs the extra N added to the soil by pollution.

Predicted changes in inorganic N leaching flux corresponded to rates of N addition (Figure 9) until around 2130 when leaching increased abruptly in the most extreme scenario, due to saturation of plant N uptake capacity.

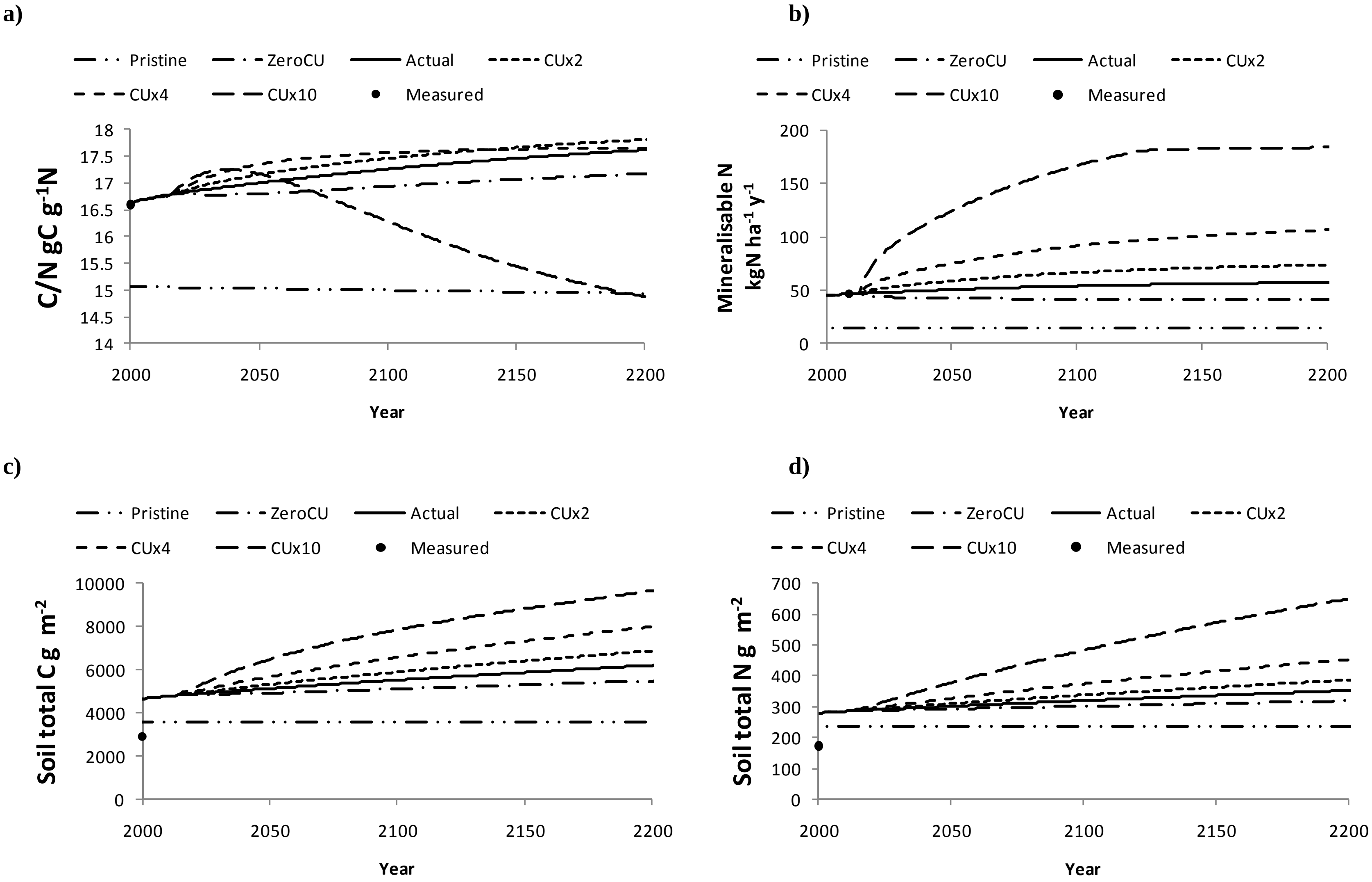


Figure 8. Measured soil properties at Newborough, together with trajectories of predicted values under different future scenarios: a) total soil C/N; b) mineralisable N; c) total soil C; d) total soil N.

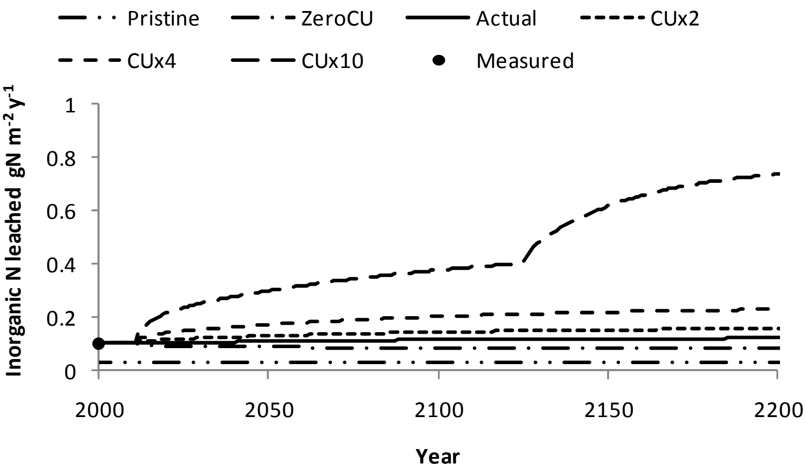


Figure 9. Measured inorganic N leaching at Newborough, together with trajectories of predicted values under different future scenarios.

5.2 Does Ellenberg N predicted from soil properties reflect observed Ellenberg N?

Using the predictive equation for mean E_N derived from the regression of all CS plots for which complete soils data are available, parameterised for abiotic conditions at Newborough as explained in the previous section, resulted in a predicted mean E_N score of 3.60, compared to the observed value of 3.63. For the scenario analyses, predicted N score was adjusted by adding 0.031 units, as described in the Methods section. However, the predicted E_N without this adjustment was remarkably close to the observed value.

5.3 Changes in mean Ellenberg N under different scenarios

Incrementing the rate of N addition generally caused an increase in predicted mean E_N score (Figure 10), which is likely to be associated with a decline in habitat quality. The Pristine scenario was an exception; the greater E_N score predicted for this scenario compared with the Actual scenario seems unlikely to be realistic. In the spatial dataset used to fit the predictive model, mean E_N score is positively correlated with soil N/C ratio. The continual decrease in N/C ratio in all except the Pristine and CUx10 scenarios (due to extra litter production; see Section 5.1) caused mean E_N score to be lower in these scenarios than in the Pristine scenario. The use of N/C ratio and other soil indicators to predict E_N is discussed in Section 6.2.

The most extreme CUx10 scenario resulted in an initial increase in mean E_N due to the large increase in mineralisable N, and an accelerated increase in mean E_N after around 30 years when the soil N/C ratio also began to increase. The change in predicted mean E_N score in 2150 between the CUx10 and the ‘Actual’ scenario was from 3.59 to 3.96 E_N units. This change is sufficient to reduce the predicted Habitat Suitability to near zero for species that are at the edge of their niche, *e.g. Cladonia rangiformis* (Figure 6).

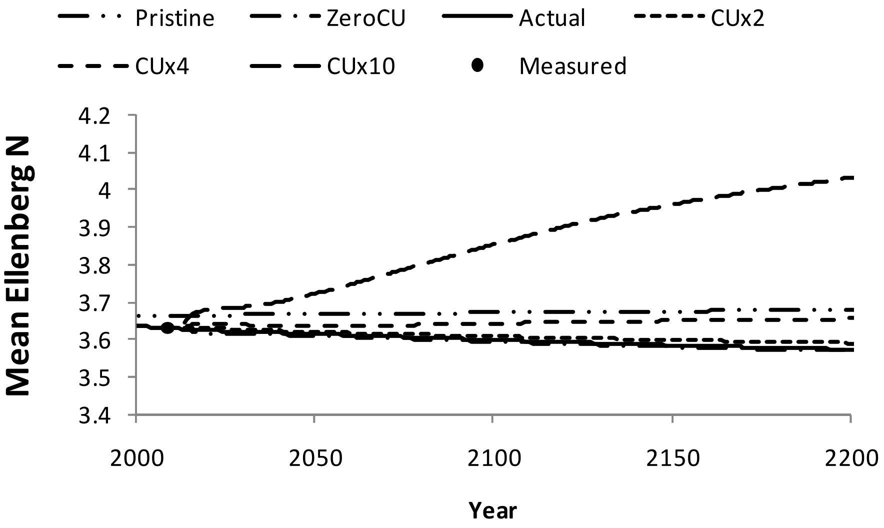


Figure 10. Predicted change in unweighted mean Ellenberg N score under different N pollution scenarios.

5.4 Calculated thresholds for relevant species

Example outputs from the N14C-MultiMOVE model as set up for the Newborough site illustrate possible responses to the scenarios (Figure 11). The coarse-leaved grass *Arrhenatherum elatius* is often a dominant species in ungrazed grassland, and is a negative site condition indicator for dune grassland. Habitat suitability for this species under grazed conditions was not much affected by N addition over the timescale considered, except in the most extreme N addition scenario which caused an increase in Habitat Suitability (Figure 11a). By contrast, in the ungrazed scenario the increase in vegetation height between 2010 and 2020 caused a large increase in Habitat Suitability, which eclipsed the effect of N addition (Figure 11b).

The small shrub *Thymus praecox* is a positive site condition indicator for dune grassland. This species showed fairly stable Habitat Suitability over the timescale considered under grazed conditions, except in the most extreme N addition scenario which caused an initial and continued decline in Habitat Suitability (Figure 11c). Effects of N addition followed a similar pattern in the ungrazed scenario, but the increased sward height reduced Habitat Suitability at all rates of N addition and caused a particularly large reduction at the highest rate of N addition (Figure 11d).

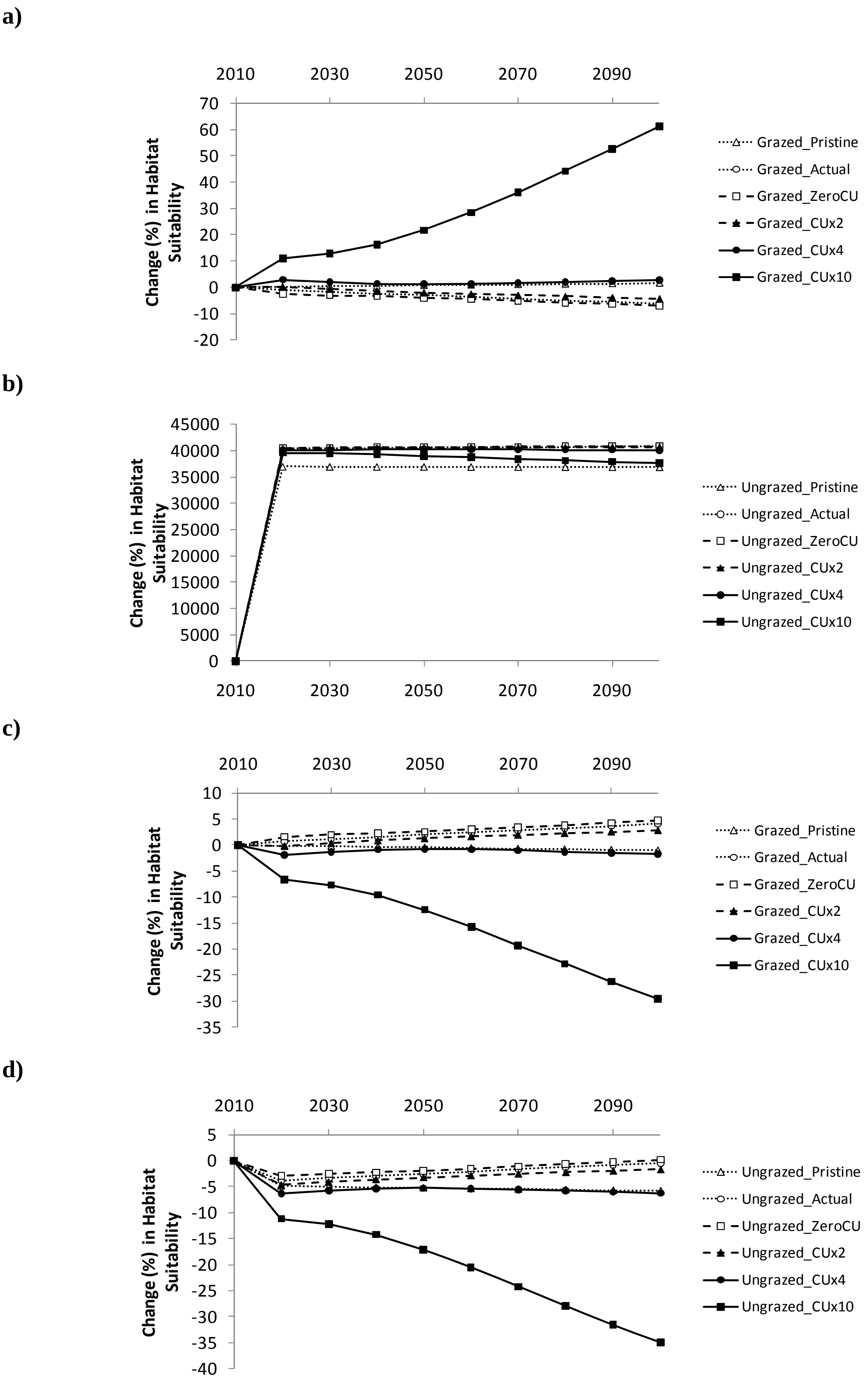


Figure 11. Relative change in Habitat Suitability from 2010 to 2100 under different scenarios for nitrogen addition and grazing, for different plant species: *Arrhenatherum elatius*, a negative site condition indicator for dune grassland, under a) grazed and b) ungrazed scenarios; and

***Thymus praecox*, a positive site condition indicator for dune grassland, under c) grazed and d) ungrazed scenarios.**

The responses of all species selected for study are summarised in Annexe Table 3. Since the change in Habitat Suitability depends on both the amount of N deposition and the time for which this deposition has occurred, a single year (2050) was chosen to compare effects of different N deposition rates. Species varied in the response of Habitat Suitability to N deposition, which declined for several species even at low rates of deposition (*e.g. Tortella tortuosa, Carlina vulgaris, Danthonia decumbens,* and *Salix repens* agg.). Most species were either equally sensitive to N enrichment in grazed and ungrazed systems (*e.g. Salix repens* agg. and *Dicranum scoparium*) or more sensitive in the grazed system (*e.g. Carlina vulgaris* and *Teucrium scorodonia*), but some species were less sensitive in the grazed system (*e.g. Danthonia decumbens* and *Hypericum perforatum*). Some species showed a decline with increasing N deposition in the grazed system, but were not affected by extra N in the ungrazed system since the effect of reduced light availability dominated their response (*e.g. Tortella tortuosa* and *Cladonia rangiformis*). Of the species assessed, 34 responded positively to N enrichment, including the negative indicator *Arrhenatherum elatius* and mesotrophic species such as *Heracleum sphondylium* and *Senecio jacobaea*, but also 12 species that are considered positive indicator species for calcareous dune grassland under Common Standards Monitoring (*e.g. Cerastium fontanum, Ononis repens* and *Trifolium repens*). Several species did not respond to N enrichment under either grazing scenario (*e.g. Prunus spinosa* and *Viola tricolor*).

The number of species at risk (> 5 % decline in Habitat Suitability) increased rapidly with time at rates of N deposition of 35 kg N ha^{-1} y^{-1} or more (Figure 12). Smaller rates of deposition delayed the point at which species became at risk, but cumulative effects were evident, causing a more gradual increase in the number of species at risk with 30 kg N ha^{-1} y^{-1}, and a delayed increase with 25 kg N ha^{-1} y^{-1}. Interestingly, rates of deposition below the current rate also caused increases in the number of species to at risk, after 70 years with 10 kg N ha^{-1} y^{-1} and after 50 years with 5 kg N ha^{-1} y^{-1}. These species were those with strong positive responses to N, and included five Common Standards Monitoring positive indicator species (*Erodium cicutarium, Geranium molle, Odontites verna, Tortula muralis* and *Veronica chamaedrys)* as well as the negative indicator species *Arrhenatherum elatius*.

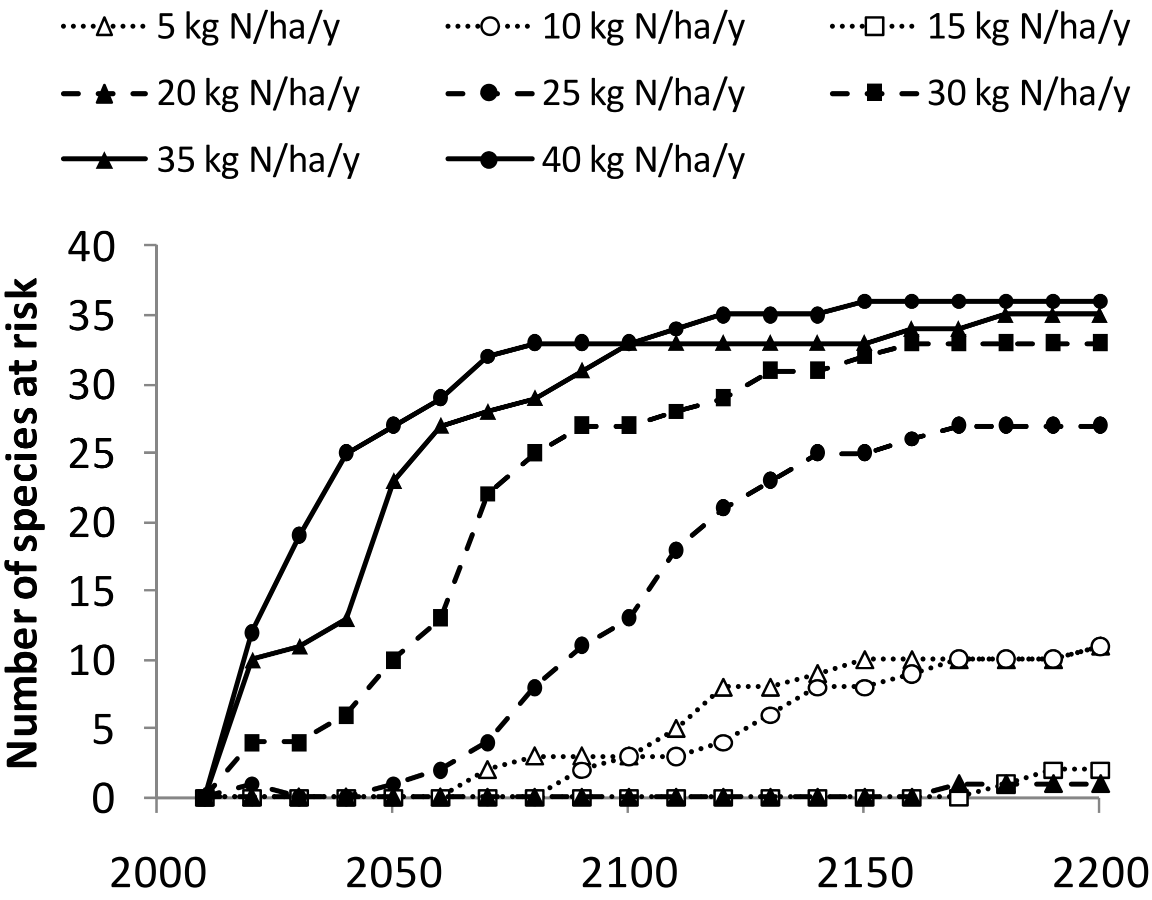


Figure 12. Changes over time in the number of species at risk (i.e. with > 5 % decline in Habitat Suitability) in grazed calcareous stable dune grassland at Newborough under different rates of N deposition.

5.5 Cover change

In view of the delay between changes in Habitat Suitability and changes in plant species presence, it was suggested that changes in cover might be more responsive to changes in Habitat Suitability. It seems logical that a species will decline in cover as the habitat becomes less suitable for it, although evidence for this effect is limited, probably because cover can change from year to year due to a variety of factors (Silvertown, Dodd *et al.*, 1994; Kafer and Witte, 2004). For this reason the decision was taken to base the MultiMOVE model on presence/absence data. Re-fitting the niche models on the basis of cover data would be a major undertaking. An intermediate solution was suggested, in which Habitat Suitability was calculated on the basis of presence/absence data, but transformed into a prediction of cover using an empirical relationship between cover and Habitat Suitability. However, on trying this approach with a test species, the relationship between cover and Habitat Suitability was not clear (Figure 13). It looks possible that there was an increase in the maximum cover attained from Habitat Suitability = 0 to around Habitat Suitability = 0.5, but that in many cases the cover did not reach this maximum. This is presumably because even when Habitat Suitability is large, other factors can decrease the cover of the species. Modelling cover requires further exploration and might be a profitable approach, but it adds additional variation at the patch level that may require additional covariates to explain.

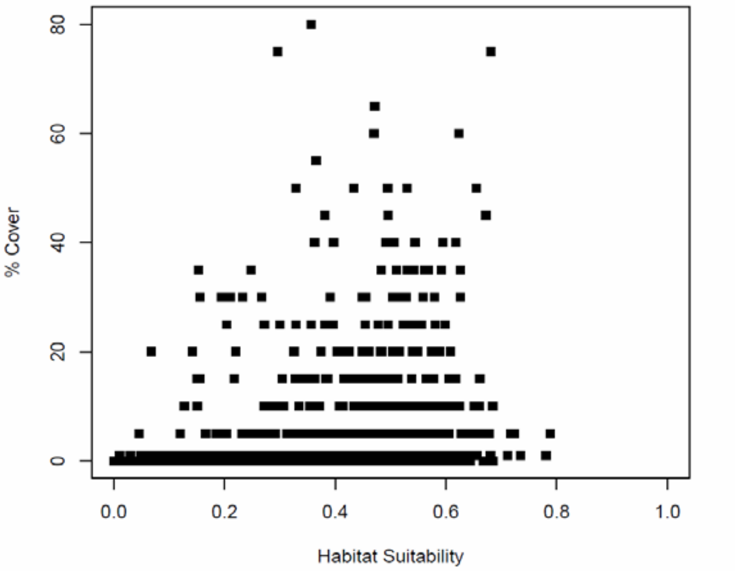


Figure 13. ‚Relationship between Habitat Suitability for *Agrostis stolonifera* as calculated using MultiMOVE for each of the Countryside Survey plots for which complete soil data were available, and visually estimated cover of this species on these plots.

6 DISCUSSION

The performance of the N14C-MultiMOVE model chain will be assessed for individual stages in the prediction, and in terms of the overall prediction of N pollution effects. Implications for conservation management and for critical load setting will also be discussed.

6.1 Predicting N effects on soil chemistry

The biogeochemical effects of N pollution as predicted by N14C contradicted the established simple N saturation model (Gundersen, Callesen *et al*., 1998) which predicts a continual decrease in C/N ratio with added N, until N leaching rises to a point where the extra N is no longer retained and the C/N ratio stabilises. However, stimulation of plant production by N is commonly observed (de Vries, Reinds *et al*., 2006; Reay, Dentener *et al*., 2008) and an increase in soil C/N ratio with N deposition seems plausible. Such an effect was observed in a survey of UK sand dunes {Jones, 2004 #1877, reproduced in Figure 14a}. The N14C predictions of a sustained increase in soil C/N with N addition thus seem likely to be broadly correct. However, the accuracy of the predicted time lag before the soil C/N ratio begins to decline again (due to increased N content of plant litter, and direct N immobilisation) remains uncertain.

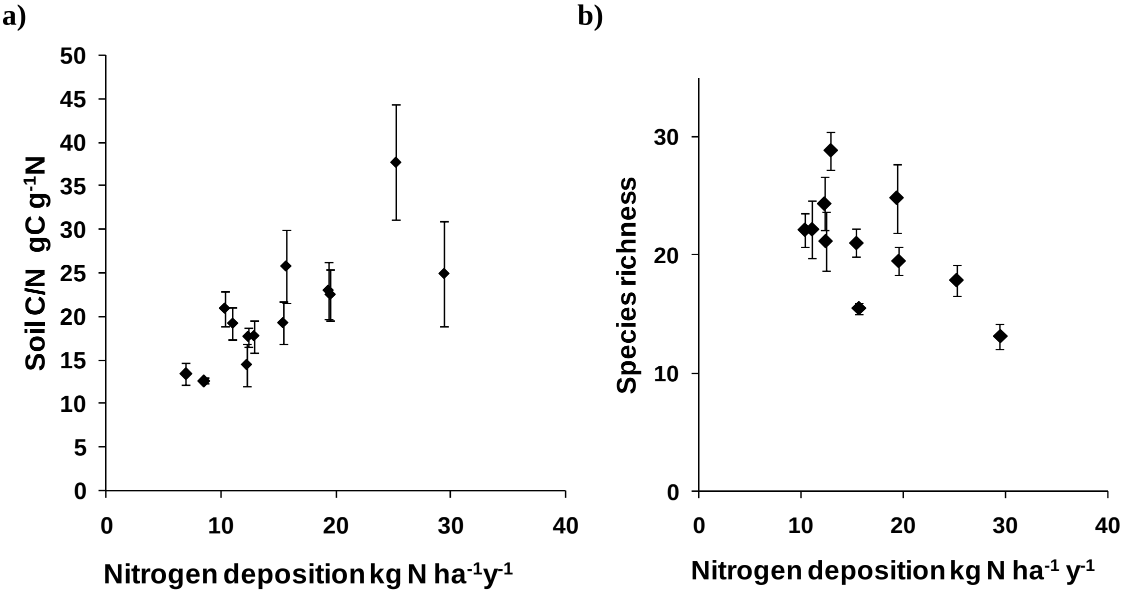


Figure 14. Effect of nitrogen deposition flux on a) soil C/N ratio and b) plant species richness, in a survey of UK dune grassland sites. From (Jones, Wallace et al. 2004).

Effects of N deposition on habitats other than sand dunes were not assessed in the current project, and it remains unclear how widespread an increase in soil C due to extra N might be. However, this seems a likely outcome in ecosystems that are not limited by other factors. Results from the 2007 Countryside Survey suggest that there have been increases in soil C stocks since 1978 in several UK habitats (Emmett, Reynolds *et al.*, 2010), and N deposition may be a driver of this change.

6.2 Predicting soil chemistry effects on mean Ellenberg N score

The first stage of the MultiMOVE model was apparently well able to predict mean E_N score from soil properties. An equation using soil N/C ratio, mineralisable N and a term for management intensity predicted a mean E_N score only 0.03 E_N units different to the value obtained from floristic recording. However, this is only a single test, and the E_N calibration may prove inaccurate when applied to a range of polluted and less polluted sites for the reasons discussed above. The discrepancy is because of the difference between spatial and temporal effects. Considering the UK as a whole, low Ellenberg values are associated with sites with C-rich soils, and with sites with large C/N ratios. However, if the stimulating effect of N on C production is widespread, changes over time on an N-polluted site are likely to result in a long-term increase in soil C content and at least a temporary increase in soil C/N. Thus using these bulk soil properties to predict temporal change in mean E_N score may be inappropriate.

The mineralisable N flux predicted by N14C responded clearly to increased N inputs, and this measure of available N may be more useful as an indicator of temporal change. However, mineralisable N is subject to greater measurement error than soil total C and N, due to the difficulty of maintaining consistent conditions during the incubation (Emmett, Reynolds *et al.*, 2010). This extra error results in this term being automatically downweighted during fitting of the multiple regression when it is used as a predictor for mean E_N. Thus although mineralisable N increased consistently and relatively rapidly with increased N inputs, this term had a comparatively small effect on predicted E_N.

The small changes in E_N with additions of fairly substantial amounts of extra N were unexpected. However, no response of mean E_N to experimental additions of up to 15 kg N / y over 7 years was seen in measured data, so the actual system does not appear to respond rapidly either. The contrast with survey data that show a decrease in E_N with increasing N deposition in many habitats (Stevens, Smart *et al*., in prep.) is presumably due to a difference in timescale. Sites with a history of elevated N deposition are likely to have lost and gained species over time, changing the mean E_N score. Experimental additions of N will have had comparatively minor effects on cumulative N deposition, and there may not have been time for extinction and invasion events to have changed the mean E_N score. For example, by 2010 the difference in total N additions between the Pristine and Actual scenarios was 813 kg N ha^{-1}, considerably larger than the 105 extra kg added between 2003 and 2010 in the most extreme experimental treatment.

The soil indicators used to predict E_N in the current study both had drawbacks. In the case of total N/C ratio, the spatial effect of N/C on mean E_N combined with the decrease in N/C caused by extra N lead to the implication that an increase in N pollution would decrease mean E_N, which is contradicted by survey evidence (Emmett, Stevens *et al*., in prep.; Stevens, Smart *et al*., in prep.). More extreme or more long-term N additions caused an increase in N/C, which helps explain this discrepancy, but N/C may not be an appropriate indicator for short-term studies. In the case of mineralisable N, variability in the measurement reduced its explanatory power (a similar problem arises with using a simple measurement of KCl-extractable N to predict mean E_N; Wieger Wamelink, pers. com.). Nevertheless, the combination of these measurements, together with a term to split more and less intensively managed habitats, explained a large proportion of the variation in mean E_N within the Countryside Survey dataset (Figure 5). It might be possible to achieve greater explanatory power by analysis within smaller habitat groups such as ‘infertile grassland’, but this approach is likely to be limited by the small number of samples in some habitats.

6.3 Predicting effects of mean Ellenberg N score on individual species

Given the small changes in predicted E_N, the small changes in Habitat Suitability for individual species at N deposition rates similar to current rates are understandable. Species varied greatly in their predicted response to N pollution. The amount of N pollution (whether annual flux or cumulative addition) at which the site becomes sufficiently unsuitable for a given species to persist depends on the species. This amount also depends on other environmental conditions, in particular ground-level light availability.

The MultiMOVE outputs can be seen as providing a snapshot in the process of species loss with cumulative N deposition. When N inputs increase in a previously pristine system, highly susceptible species such as those sensitive to low concentrations of gaseous NH_3 will disappear first. As N enrichment continues, further species will be lost in order of sensitivity. The actual extinction event may be triggered by a relaxation of management, or an outbreak of a disease or an insect herbivore, but the effect of cumulative N deposition in decreasing Habitat Suitability is the underlying reason for loss of the species. Thus species showing steeper declines in Habitat Suitability with cumulative N deposition were presumed to be more likely to become extinct on a site than species with shallower declines.

6.4 Interpreting species responses

The responses of the selected species varied, with some responding positively to increased N deposition and /or to increased vegetation height, some responding negatively, and most showing little response to realistic levels of N deposition.

The niche occupancy modelling approach used in the current project provides an important tool for assessing policies for their impacts on biodiversity, as it enables predictions of the effects of anthropogenic drivers on individual species. The presence of particular species is a key component of most operational definitions of biodiversity, whether these be protected species, typical or characteristic species of protected habitats, or unwanted species such as invasive aliens or species typical of other habitats. The EU Habitats Directive includes “typical species” in the definition of favourable conservation status for a habitat (EEC, 1992). It has been proposed to the European Environment Agency that “for each habitat a set of typical species should be identified by each Member State” (Shaw and Wind, 1997), but typical species have not yet been defined for UK habitats. To assess how the changing prevalence of individual species affects concepts such as biodiversity, favourable conservation status, or habitat quality, clear definitions of the relative importance of species as positive and negative indicators within a particular habitat will be necessary.

The problem can be illustrated by considering the conservation value of species that were predicted to be affected by N at Newborough. If inclusion as a Common Standards Monitoring positive indicator species for calcareous fixed dune grassland is taken to be a useful criterion for assessing conservation value, this leads to anomalies, in that 12 of the positive indicator species responded positively to N. Five of these responded sufficiently strongly to undergo enough decline to be ‘at risk’ by 2050 under the reduced N deposition scenarios. At least three interpretations are possible:

1. Nitrogen pollution has little effect on conservation status, since more N pollution leads to the decline in some positive indicator species but an increase in others.
2. The list of indicator species is incorrect, and should be amended to account for N-sensitivity by deleting N-favouring species such as *Ononis repens* and adding N-avoiding species such as *Carlina vulgaris*.
3. The list of indicator species is incompletely specified, and should give higher weighting to species that are scarce or distinctive for the habitat (*e.g. Astragulus danicus*), and lower or zero weighting to species that are common or occur in many habitats (*e.g. Trifolium repens*).

The first interpretation is incorrect, since N deposition rates have been shown to correlate with lower species richness (Jones, Wallace *et al.*, 2004; Emmett, Stevens *et al.*, in prep.; Stevens, Smart *et al.*, in prep.). The second is often advocated by N-pollution specialists, but seems a circular argument. If this route is taken, it would also be necessary to adjust indicator species lists according to sensitivity to ozone, climate change, imperfect management, and other drivers of environmental change. We consider that the third approach is the most valid; indicator species lists for assessing habitat quality should be defined solely according to biodiversity value.

6.5 Overall prediction of N effects

The prediction of N effects on species involves several stages, each of which introduces some uncertainty. The overall confidence that can be attached to each stage of the model chain is summarised in Table 4.

Table 4. Evaluation of stages in the prediction of N effects on ecosystems.

Model stage	Confidence	Evidence and notes
N deposition effects on soil properties (N14C)	Medium	The N14C model reproduces observed increases in C/N with N deposition, in contrast to the VSD model used currently to set critical loads. More testing is needed for confidence in the time-course of future changes.
Soil property effects on mean E_N (MultiMOVE Stage I)	Low	The multiple regression approach explained spatial variation in mean E_N, but temporal change in properties such as %C and C/N is likely to have an opposite relationship with mean E_N. Mineralisable N may be a better indicator of change in mean E_N, but is prone to measurement error. Modelling vegetation height is probably necessary to allow for effects of N on ground-level shading.
Mean E_N effects on species occurrence (MultiMOVE Stage II)	Medium	The niche models are based on large empirical datasets, so accurately reflect species' spatial distribution. More testing is needed for confidence in predicted temporal change, against datasets where temporal change in species presence has been observed.
N deposition effects on species occurrence	Low	Overall confidence can only be as great as confidence in the weakest link in the model chain. Possibilities for testing were limited in the current study by lack of observed species change.

Many attempts have been made to model N effects on soils, using biogeochemical models of varying complexity. The N14C model represents a compromise between highly detailed ecosystem models which may be difficult to parameterise even for sites with many measurements, and very simple models with few parameters but which are limited in accuracy and responsiveness. The study illustrates the need for a model which can predict plant N exposure, and incorporates important processes such as the accumulation of C in soil due to increased plant production. Although further development of the N14C model is required, for instance to incorporate phosphorus limitation on some sites, in general the complexity of this model seems appropriate.

Linking indicators of plant N exposure (soil N/C and mineralisable N) to floristic properties proved reasonably successful. However, predicted changes in soil properties as a result of N addition seem unlikely to produce changes in E_N sufficient to explain species loss (for which there is good evidence, *e.g.* Figure 14b). It may be possible to improve the derivation of exposure indicators from N14C, to make them correspond more closely to factors that explain spatial variation in E_N. However, soil measurements remain limited in their ability to reflect the changes in above-ground competition which are thought to be the immediate cause of many N effects on species. Canopy thickness or standing biomass at certain times of year is likely to affect plants' vigour and reproductive success, and the speed with which gaps close can have major effects on recruitment. The importance of vegetation structure for species prevalence is illustrated in the current study by the comparatively large effect of grazing. To improve predictions from this type of model chain it will be necessary to incorporate predictions of N effects on productivity, and of management effects on standing biomass.

6.6 Implications for Critical Load setting

The definition of Critical Load suffers from the use of the phrase "significant harmful effects on specified sensitive elements of the environment", which could be defined *ad absurdum* as the accumulation of a few extra N atoms in a specified part of a particular species. In practice, Critical

Loads are mainly set using experimental evidence of species loss, although changes in species' vigour without actual loss, and biogeochemical changes such as the onset of N leaching, are also sometimes invoked (Bobbink and Hettelingh, in press). The development of capacity to predict changes in species composition resulting from N pollution is an important step towards relating outputs from dynamic biogeochemical models to the legislative criteria used in conservation assessment (Hettelingh, Posch *et al*. 2009).

When assessing experimental results to determine what the critical load should be, there is inevitably a subjective element in choosing which results to emphasise, and how severe effects need to be before they are considered to be 'damage'. When interpreting species responses derived from survey data, as well as the difficulty of defining which species should be considered for a particular habitat (as discussed in Section 6.4), there is subjectivity in defining the cut-off probability at which a species is considered unlikely to occur. Similarly, when interpreting the outputs from the N14C-MultiMOVE model chain, there is subjectivity in deciding how much of a change in Habitat Suitability is necessary for 'damage' to have occurred. In the current study, a 5 % decline in Habitat Suitability was considered to put a species at risk. It should be borne in mind, however, that this is relative to present-day conditions, and it is likely that N-sensitive species have already become extinct on the site.

Assuming that this criterion for assessing risk to a species is relevant, and that placing a single species at risk can be considered to be causing damage, it is worth considering the plot of species at risk over time (Figure 12) in relation to the critical load. By 2050, all scenarios with addition of 25 kg N ha^{-1} y^{-1} or more had resulted in damage. However, scenarios with rates of N addition lower than the current critical load of 12 kg N ha^{-1} y^{-1} also resulted in damage, by 2090 with 10 kg N ha^{-1} y^{-1} and by 2070 with 5 kg N ha^{-1} y^{-1}. The current critical load and the present-day load at Newborough (also around 12 kg N ha^{-1} y^{-1}) are in the range that caused least damage, i.e. where no species were placed at risk for around 150 years. The simulations used in the current study were run only until 2200, and further extrapolations seem likely to narrow the range of the load that does not cause damage. The current critical load is not inconsistent with this picture.

There is undoubtedly a case for making more use of survey data and modelled projections for setting critical loads. Issues around the subjectivity of setting 'damage' thresholds, the choice of 'sensitive elements' and the timescale over which changes are assessed also apply to interpreting experimental data, which are generally termed empirical (*i.e*. of overriding importance). Recent analyses of survey data (*e.g*. Emmett, Stevens *et al*., in prep.) and the development of multifactor response models such as MultiMOVE represent progress towards acceptance of survey data as also empirical.

6.7 Implications for conservation management

The current study clearly demonstrated the damaging cumulative effects of long-term N additions. Short-term N additions are likely to have comparatively small effects, but if the increase is sustained it will accelerate the process of eutrophication and reduce the time before damage occurs. Thus local and national developments that cause a sustained increase in N inputs should continue to be opposed in the interest of site protection.

The study also showed that while some floristic change can be related to measurable changes in soil chemistry, changes in vegetation structure are of considerable importance. A principal mechanism by which N pollution causes species loss is through increasing productivity and so occluding the soil surface through shading by increased live biomass and increased litter inputs. These factors negatively affect the many species of conservation concern that require good light availability at ground level, whether as adult plants (or animals, Wallisdevries and Van Swaay, 2006) or during recruitment and seedling growth. This implies that more attention should be paid to vegetation structure and litter inputs in monitoring schemes. Vegetation height is often recorded alongside floristic data, but measurements are hard to standardise.

The question of whether active management such as grazing or hay removal can mitigate increased N inputs is controversial. These activities remove relatively small amounts of N from the system, and can only slow the accumulation of N in soil and plants. However, by reducing ground-level shading, grazing or hay removal undoubtedly reduce the immediate effects of N pollution on many sensitive species, and seem likely to allow the long-term persistence of such species as long as the management can be maintained.

6.8 Implications for dynamic modelling of N effects

The N14C forecast of increasing total C/N ratio with extra N addition was opposite to the decrease forecast by the VSD model that is currently used to calculate critical load exceedance for the UK. Evidence of increasing C/N ratio in sand dunes across a surveyed gradient of increasing N deposition (Jones, Wallace *et al*., 2004) suggests that N14C is more accurate, at least for sand dune habitats. Increasing soil C inputs in response to extra N seem likely, at least in systems where productivity is limited by N. More investigation of N deposition effects on plant productivity and soil C/N ratio, in different habitats, would be useful. The N14C model is currently being extended to allow for phosphorus limitation, which may be an important factor in dune habitats (Jones, 2010). The VSD model has also been extended with the addition of new C and N processing routines, and the CCE is investigating whether this new ‘VSD+’ model can be applied across Europe.

The mechanistic detail that is appropriate in a soil C and N model depends on the question being investigated, and also on the likely availability of data for the scales considered. The N14C model is being developed for use when input data are limited, e.g. for modelling critical load exceedance at national scale. The small parameter set also makes the model suitable for assessing change on conservation sites, for which default data (*e.g.* mean values for the soil type) can be used where measurements are unavailable. However, it is clear that the effects of management are not adequately addressed using solely N14C to predict environmental change. Vegetation structure had a large direct effect on Habitat Suitability. The model chain therefore needs more detail on effects of N and management on vegetation structure, and on soil N processes.

The use of mineralisable N as an indicator of eutrophication was justified, particularly since total soil C/N ratio had contradictory effects in spatial and temporal analyses. Plant-available N is difficult to measure with certainty, but mineralisable N is likely to be a more closely-related measurement than is total C/N. Other measurements of plant-available N, such as simple extraction of ammonium and nitrate, net nitrification, or dissolved organic N, could be assessed as alternative indicators. Further investigation of the relationships among C/N ratio, mean Ellenberg N and historical N deposition might reveal different or closer relationships in other habitats. For some species, particularly bryophytes and lichens, direct atmospheric exposure to N is likely to be of greater importance than soil N availability. Gaseous ammonia, in particular, can have direct toxic effects on sensitive lichen species at low concentrations (Cape, van der Eerden *et al*., 2009). Thus it may be useful to include estimates of N deposition rate or of ammonia concentration directly when fitting niche models, as an alternative to using soil N exposure. It seems likely that both cumulative N deposition and current N concentration can affect species occurrence.

7 CONCLUSIONS AND RECOMMENDATIONS

The niche occupancy modelling approach allows current knowledge of N dynamics in ecosystems to be combined with species response models that have a strong grounding in empirical survey data. Applying an example model chain to dune grassland at Newborough illustrated how the approach might be used to identify species at risk and to assess overall effects on the habitat. Weaknesses in the model chain were identified, in particular the partial ability of soil properties to predict changes in floristic properties. A more detailed treatment of vegetation growth is probably required to capture the effects of N on ground level shading, which is a major mechanism by which N affects species prevalence. The ability to predict N deposition effects on soil N availability is improving, however, and the model chain captures some of the effects of increasing N availability on species composition.

The study allows suggestions to be made for improving the prediction and attribution of N effects on sites of conservation importance:

- Responses of soil C/N to N addition, and the use of soil C/N as a driver of species response models, need to be investigated in different habitats.
- Effects of N addition are often mediated by vegetation structure, suggesting that management can mitigate some of the effects of N pollution.
- More long-term datasets showing floristic change would be invaluable for relating such change to N deposition, and to intermediate variables such as vegetation height and soil N characteristics. Consistent and objective methods should be applied to measurements of floristic response, the intensity of drivers such as N deposition or management, and intermediate variables.
- Niche occupancy modelling has the potential to identify species currently at risk on particular sites, which might be a focus of targeted monitoring.
- To interpret changes in many species in terms of damage or protection, clear definitions are required of the importance of individual species as positive or negative indicators of condition in a particular habitat.

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9 ANNEXE

Annexe Table 1. Evaluation of species with negative or positive responses to nitrogen pollution in Lowland Acid Grassland (LAG) or in Lowland Calcareous Grassland (LCG) (Emmett, Stevens *et al*., in prep.) for inclusion in the current study. Evidence for responses was obtained from spatial (S) and/or temporal (T) analyses of the BSBI Vascular plant database (VPD) and/or the BSBI Local Change study (LC). Habitat descriptions were taken from (Stace, 1991).

Species	Evidence	In	Response	Habitat	Incl.
Allium vineale	VPD(S)	LCG	negative	grassy places, rough ground, banks and waysides	no
Anacamptis pyramidalis	VPD(S)	LCG	negative	chalk and limestone grassland, calcareous dunes	yes
Bromopsis erecta	LC(S)	LCG	negative	dry grassland and grassy slopes, especially on calcareous soils	no
Campanula glomerata	LC(S)	LCG	negative	chalk and limestone grassland, scrub and open woodland, cliffs and dunes by the sea	yes
Carex spicata	VPD(T) & LC(S)	LCG	negative	damp grassy places in fields, on banks and waysides and by rivers and ponds	no
Carlina vulgaris	VPD(S)	LCG	negative	open grassland, on usually calcareous but sometimes sandy soils, fixed dunes and cliff tops	yes
Centaurea scabiosa	LC(S)	LCG	negative	grassland, rough ground, cliffs and waysides mainly on calcareous soils	no
Cerastium semidecandrum	VPD(T)	LAG	negative	dry open places on sandy or limestone soils, especially dunes	yes
Cynoglossum officinale	VPD(S)	LCG	negative	rather open ground mostly on sand, shingle or limestone, and waste ground	yes
Daucus carota	LC(S)	LCG	negative	grassy and rough ground, mostly on chalky soil and near sea	yes
Echium vulgare	VPD(S)	LCG	negative	open grassy places, cliffs, dunes, shingle, rough ground, usually on light, often calcareous soils	yes
Geranium columbinum	VPD(S)	LCG	negative	grassy places, banks and scrub, mostly on calcareous soils	no
Lathyrus nissolia	VPD(S)	LCG	positive	grassy places	no
Myosotis ramosissima	VPD(T)	LAG	negative	dry open places on sandy or limestone soils	yes
Ononis repens	VPD(S) & LC(S)	LCG	negative	rough grassy places on well-drained soils, especially coastal	yes
Ornithopus perpusillus	LC(T)	LAG	negative	dry barish sandy and gravelly ground	yes
Rosa micrantha	VPD(S)	LCG	negative	mostly in scrub, often not on calcareous soils	no
Spiranthes spiralis	VPD(S)	LCG	negative	short permanent grassland and grassy dunes	yes
Stachys officinalis	VPD(S)	LCG	positive	hedgebanks, grassland, heaths, avoiding heavy soils	no
Trifolium arvense	VPD(S &T)	LAG	negative	barish ground on sandy soils	yes
Trifolium micranthum	LC(T)	LAG	negative	short turf, especially close-set lawns	no
Vicia lathyroides	VPD(S)	LAG	negative	maritime sand and inland sandy heaths	yes

Annexe Table 2. Evaluation of species for inclusion in the study. MultiMOVE: niche model derived for the species. CSM: listed as a typical species (+) or negative indicator species (–) for calcareous fixed dune grassland in Common Standards Monitoring guidance (JNCC, 2004). Table 2.2.1. Sensitive = significant negative (–) or positive (+) effect of N deposition on the species' distribution demonstrated in (Emmett, Stevens *et al.*, in prep.). Present = recorded in floristic monitoring of the experimental site.

Species	BRC code	MultiMOVE	CSM	Sensitive	Present
Achillea millefolium	9207	yes			yes
Agrostis capillaris	92040	yes	+		yes
Agrostis stolonifera	92039	yes			yes
Aira caryophyllea	92041	yes			yes
Aira praecox	92042	yes	+		yes
Ammophila arenaria	92097	no			yes
Anacamptis pyramidalis	92098	yes		–	no
Anthoxanthum odoratum	920121	yes			yes
Arenaria serpyllifolia	920162	no			yes
Arrhenatherum elatius	920169	yes	–		yes
Astragalus danicus	920207	yes	+		no
Bryum capillare	82094	yes			yes
Campanula glomerata	920315	no		–	no
Campylopus introflexus	820141	yes			yes
Carex arenaria	920344	no	+		yes
Carex caryophyllea	920355	yes			yes
Carex flacca	920376	yes	+		no
Carlina vulgaris	920427	yes		–	no
Centaurium erythraea	9205486	no			yes
Cerastium fontanum	920467	yes	+		yes
Cerastium semidecandrum	920469	yes		–	yes
Cladonia pyxidata	5502912	yes	+		yes
Cladonia rangiformis	5502915	yes	+		yes
Climacium dendroides	820158	yes			yes
Crataegus monogyna	920569	yes	–		yes
Crepis capillaris	920572	yes	+		yes
Cynoglossum officinale	920596	yes		–	no
Dactylis glomerata	920607	yes			yes
Danthonia decumbens	9201915	yes			yes
Daucus carota	9205474	yes		–	no
Dicranum scoparium	820206	yes			yes
Echium vulgare	920670	yes		–	yes
Erodium cicutarium	920745	yes	+		yes
Festuca ovina agg.	920821	yes			yes
Festuca rubra agg.	920824	yes	+		yes
Galium verum	920888	yes	+		yes
Geranium dissectum	920907	yes			yes
Geranium molle	920911	yes	+		yes
Helictotrichon pubescens	920962	yes			yes
Heracleum sphondylium	920968	yes			yes
Holcus lanatus	920983	yes			yes
Homalothecium lutescens	820126	yes			yes
Hylocomium splendens	820346	yes		+	yes
Hypericum perforatum	9201014	yes			yes
Hypnum cupressiforme sens. lat.	820351	yes	+		yes
Hypochoeris radicata	9201020	yes	+		yes
Leontodon autumnalis	9201129	yes			yes
Linum catharticum	9201169	yes	+		no

Species	**BRC code**	**MultiMOVE**	**CSM**	**Sensitive**	**Present**
Lotus corniculatus	9201191	yes	+		yes
Luzula campestris	9201201	yes	+		yes
Myosotis ramosissima	9201320	yes		–	no
Odontites verna	9201361	yes	+		no
Ononis repens	9201377	yes	+	–	yes
Ornithopus perpusillus	9201397	yes		–	no
Peltigera canina	5503836	yes	+		yes
Pilosella officinarum agg.	920976	yes	+		yes
Plantago lanceolata	9201487	yes	+		yes
Pleurozium schreberi	820463	yes			yes
Poa pratensis sens. lat.	9201506	yes			yes
Prunella vulgaris	9201610	yes	+		no
Prunus spinosa	9201617	yes	–		yes
Pseudoscleropodium purum	820507	yes			yes
Ranunculus bulbosus	9201647	no			yes
Rhinanthus minor	9201678	no	+		no
Rhytidiadelphus squarrosus	820533	yes	+		yes
Rhytidiadelphus triquetrus	820534	yes	+		yes
Rubus caesius	9201728	yes			yes
Rumex acetosa	9201734	yes			yes
Rumex acetosella	9201735	yes			yes
Salix repens agg.	9201802	yes			yes
Sedum acre	9201875	yes	+		no
Senecio jacobaea	9201899	yes			yes
Sonchus arvensis	9201952	yes			yes
Sonchus asper	9201953	no			yes
Spiranthes spiralis	9201997	no		–	no
Taraxacum officinale agg.	9202034	yes			yes
Teucrium scorodonia	9202046	yes			yes
Thymus praecox	9202060	yes	+		yes
Tortella tortuosa	820607	yes			yes
Tortula muralis	820614	yes	+		no
Tortula ruralis ruraliformis	820618	yes			yes
Tragopogon pratensis	9202074	yes			yes
Trifolium arvense	9202077	no		–	yes
Trifolium campestre	9202080	yes			yes
Trifolium repens	9202092	yes	+		yes
Veronica chamaedrys	9202168	yes	+		yes
Veronica officinalis	9202173	yes			yes
Vicia hirsute	9202191	yes			yes
Vicia lathyroides	9202194	yes		–	no
Vicia sativa	9202516	no			yes
Viola canina	9202207	yes	+		no
Viola riviniana	9202218	yes	+		yes
Viola tricolor	9202220	yes	+		yes

Annexe Table 3. Threshold values of N deposition rate after 2012 (kg N ha^{-1} y^{-1}) at the Newborough site, at which the Habitat Suitability for species typical of dune grassland falls below 20% (at risk) or below 5% (at serious risk) by 2050, with and without grazing. Species are ranked in order of sensitivity in the grazed system, starting with the most sensitive species *i.e.* those for which Habitat Suitability declines even with zero deposition. CSM = Common Standards Monitoring positive indicator species (+), negative indicator species (–), or not mentioned in CSM (no). Light dom. = no response to N in the ungrazed system, since light effects are dominant.

Species	CSM	Grazed		Ungrazed	
		At risk (< 5% decline)	At serious risk (> 20% decline)	At risk (< 5% decline)	At serious risk (> 20% decline)
Tortella tortuosa	no	0.0	10.8	light dom.	light dom.
Carlina vulgaris	no	0.0	14.5	0.0	0.0
Danthonia decumbens	no	0.0	17.7	71.8	105.7
Salix repens agg.	no	0.0	21.5	0.0	21.5
Dicranum scoparium	no	0.0	26.3	0.0	26.3
Climacium dendroides	no	0.0	26.4	0.0	26.4
Cladonia rangiformis	+	0.0	27.3	light dom.	light dom.
Teucrium scorodonia	no	0.0	29.6	0.0	0.0
Viola riviniana	+	0.0	32.2	0.0	0.0
Thymus praecox	+	0.0	32.4	7.0	42.6
Pseudoscleropodium purum	no	0.0	38.9	0.0	0.0
Linum catharticum	+	0.0	42.4	27.4	92.8
Viola canina	+	0.0	46.1	0.0	46.1
Cladonia pyxidata	+	0.4	48.7	0.4	48.7
Rhytidiadelphus squarrosus	+	0.4	48.6	light dom.	light dom.
Carex flacca	+	0.9	50.2	0.0	0.0
Astragalus danicus	+	1.3	52.2	light dom.	light dom.
Festuca ovina agg.	no	1.6	52.7	54.5	93.9
Hypnum cupressiforme sens. lat.	+	2.0	55.0	9.9	72.8
Pleurozium schreberi	no	3.2	59.6	3.2	59.6
Carex caryophyllea	no	3.3	59.6	light dom.	light dom.
Helictotrichon pubescens	no	3.3	60.1	light dom.	light dom.
Luzula campestris	+	3.9	62.3	light dom.	light dom.
Pilosella officinarum agg.	+	6.3	71.9	244.2	308.0
Prunella vulgaris	+	6.4	72.1	0.0	0.0
Aira praecox	+	8.0	78.7	157.4	227.6
Homalothecium lutescens	no	9.5	84.5	123.0	212.5
Rhytidiadelphus triquetrus	+	14.9	106.4	0.0	0.0
Lotus corniculatus	+	20.0	125.9	41.0	198.6
Aira caryophyllea	no	20.7	129.7	light dom.	light dom.
Crataegus monogyna (g)	–	20.8	130.0	0.0	0.0
Anacamptis pyramidalis	no	28.1	159.2	28.1	159.2

Species	CSM	Grazed		Ungrazed	
		At risk (< 5% decline)	At serious risk (> 20% decline)	At risk (< 5% decline)	At serious risk (> 20% decline)
Galium verum	+	32.9	177.9	0.0	15.0
Veronica officinalis	no	60.3	287.9	0.0	210.7
Agrostis capillaris	+	62.8	297.5	0.0	0.0
Taraxacum officinale agg.	no	86.2	391.5	light dom.	light dom.
Peltigera canina	+	107.6	477.3	0.0	0.0
Campylopus introflexus	no	121.1	531.3	0.0	65.7
Hylocomium splendens	no	227.0	954.9	0.0	0.0
Plantago lanceolata	+	239.8	1005.5	0.0	0.0
Tortula ruralis ruraliformis	no	269.4	1123.5	269.4	1123.5
Hypericum perforatum	no	insensitive	insensitive	0.0	0.0
Bryum capillare	no	insensitive	insensitive	insensitive	insensitive
Myosotis ramosissima	no	insensitive	insensitive	insensitive	insensitive
Ornithopus perpusillus	no	insensitive	insensitive	insensitive	insensitive
Prunus spinosa	–	insensitive	insensitive	insensitive	insensitive
Vicia lathyroides	no	insensitive	insensitive	insensitive	insensitive
Viola tricolor	+	insensitive	insensitive	insensitive	insensitive
Achillea millefolium	no	positive	positive	positive	positive
Agrostis stolonifera	no	positive	positive	positive	positive
Anthoxanthum odoratum	no	positive	positive	positive	positive
Arrhenatherum elatius	–	positive	positive	positive	positive
Cerastium fontanum	+	positive	positive	positive	positive
Cerastium semidecandrum	no	positive	positive	positive	positive
Crepis capillaris	+	positive	positive	positive	positive
Cynoglossum officinale	no	positive	positive	positive	positive
Dactylis glomerata	no	positive	positive	positive	positive
Daucus carota	no	positive	positive	positive	positive
Echium vulgare	no	positive	positive	positive	positive
Erodium cicutarium	+	positive	positive	positive	positive
Festuca rubra agg.	+	positive	positive	positive	positive
Geranium dissectum	no	positive	positive	positive	positive
Geranium molle	+	positive	positive	positive	positive
Heracleum sphondylium	no	positive	positive	positive	positive
Holcus lanatus	no	positive	positive	positive	positive
Hypochoeris radicata	+	positive	positive	positive	positive
Leontodon autumnalis	no	positive	positive	positive	positive
Odontites verna	+	positive	positive	positive	positive
Ononis repens	+	positive	positive	positive	positive
Poa pratensis sens. lat.	no	positive	positive	positive	positive
Rubus caesius	no	positive	positive	positive	positive
Rumex acetosa	no	positive	positive	positive	positive
Rumex acetosella	no	positive	positive	positive	positive

Species	**CSM**	**Grazed**		**Ungrazed**	
		At risk (< 5% decline)	**At serious risk (> 20% decline)**	**At risk (< 5% decline)**	**At serious risk (> 20% decline)**
Sedum acre	+	positive	positive	positive	positive
Senecio jacobaea	no	positive	positive	positive	positive
Sonchus asper	no	positive	positive	positive	positive
Tortula muralis	+	positive	positive	positive	positive
Tragopogon pratensis	no	positive	positive	positive	positive
Trifolium campestre	no	positive	positive	positive	positive
Trifolium repens	+	positive	positive	positive	positive
Veronica chamaedrys	+	positive	positive	positive	positive
Vicia hirsuta	no	positive	positive	positive	positive