Quantifying uncertainty in critical loads

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Contents

Report Summary

This report outlines a systematic method for collating empirical evidence from the literature useful for setting critical loads of N deposition, using the example of EUNIS class D (bog, mire and base-poor fen) ecosystems in Europe. The report suggests an analytical method for estimating a critical load and its associated uncertainty. We summarise information available in the body of literature and discuss limitations in current knowledge in the context of critical loads setting. We suggest future work which may be required to improve understanding of empirically-based critical loads.

Background

Air pollution increases the deposition of nutrients and acidifying compounds to ecosystems, which can significantly alter ecological functioning and species composition. The impact of reactive nitrogen (N_R) deposition may be particularly profound in oligotrophic systems such as bogs and mires, which are adapted to low nutrient availability. Critical loads are defined as "the amount of N deposition below which significant harmful effects on specified sensitive elements of the environment do not occur according to present knowledge" (Nilsson & Grennfelt 1988). Critical loads for nutrient nitrogen deposition are set based on expert judgement via consultation of the literature (e.g. Achermann & Bobbink 2003; Bobbink & Hettelingh 2011); the current critical load for raised and blanket bogs is set at 5-10 kg N ha^{-1} yr^{-1} and for valley mires, poor fens and transition mires, 10-15 kg N ha^{-1} yr^{-1} (Bobbink & Hettelingh 2011). Although the concept of 'no harmful effects' has recently been called into question (Payne et al. 2013), critical loads remain the legislative tool to regulate air pollution levels and concentrations in many parts of the world. However, to our knowledge, there has been no systematic, quantitative assessment of the uncertainty in critical loads.

In this study, we synthesise empirical evidence of the effects of N deposition on EUNIS class D (bog, mire and base-poor fen) ecosystems in Europe, with particular reference to the Atlantic biogeographic region and the UK. We use a worked example for N deposition on bogs and mires of the UK and Atlantic region, but the method is applicable to other habitats for which sufficient studies have been conducted. Our primary aim is to re-assess the evidence base for the current critical load, and assess the extent to which it is possible to generate an improved critical load estimate with associated estimate of uncertainty. We discuss limitations of the current evidence base for the purposes of defining critical loads, and briefly describe future advances that are required.

Methodology

Synthesis of evidence

We collated published studies containing empirical information on the effects of N deposition on bog, mire and base-poor fen ecosystems in Europe, with particular reference to the Atlantic biogeographic region and the UK. Literature was collated based on citations within reports from expert workshops on empirical critical loads and dose-response relationships (Achermann & Bobbink 2003; Bobbink & Hettelingh 2011). The studies included: a) monitoring programmes over time, b) spatial surveys which use a space-for-time substitution, and c) N addition experiments, primarily collated from the peer-reviewed literature. The studies varied in terms of their exceedance indicators, which included chemical (e.g. concentrations of N in interstitial water, peat, plant tissue, C:N or N:P stoichiometry) and/or ecosystem (e.g. growth and productivity of moss, vascular plant cover or biomass growth) response indicators. Appendix 1 lists literature cited in Achermann & Bobbink (2003) and Bobbink & Hettelingh (2011) which we considered to be potentially relevant to an empirical synthesis. Due to time constraints, this report utilises information from 30 studies cited within the reports and is not an exhaustive synthesis.

Information extracted from the 30 studies (Table 1, appended document) included: reference; location of study; habitat; approach (1. monitoring over time, 2. spatial gradient surveys and 3. lab, greenhouse and field experiments); number of replicate plots or sites; estimates of background wet and dry N deposition; N range tested; indicators of exceedance and their nature (chemical or ecosystem); deposition level (kg N ha^{-1} yr^{-1}) at which a statistically significant effect was observed; mathematical functional response of exceedance indicator to increasing N deposition; interpretation of findings by the critical loads community (where relevant). Based on these fields it was possible to determine how effective each study was in determining the level of N deposition at which an effect on ecological functioning had occurred. Possible biases or uncertainties are briefly described in Table 1. We attempted to quantify this by attributing each study with a “robustness score” (see Box 1). Some studies were identified as not containing appropriate evidence to contribute to an empirical assessment of the critical load and so were excluded prior to assessing all tabulated fields.

To avoid pseudo-replication, and since effects are likely to be causally linked, there was only one table entry per experimental or observational site (even if there may be more than one measure of exceedence). Conversely, if a study examined effects at multiple sites (but not within the context of a spatial survey), each site was recorded as a separate table entry, since effects were often unequal across sites and each site represents a unique entry, termed “evidence item” throughout this report.

Whilst a number of studies included potentially important interacting treatments (e.g. deposition of other nutrients, temperature, CO_2), we collated information on the effects of N deposition alone.

Box 1. Robustness score (0-5; 0 – low robustness, 5 – high robustness). The score was attributed based on entries to Table 1.

Sources of bias and/or uncertainty:

i. **Quality of estimate of N deposition/application rate**. Can N deposition/application rate be quantified? Has background deposition been accounted for? Are both wet and dry deposition quantified? Is the estimation of deposition consistent across sites and/or monitoring periods? (see Sutton *et al.* 2003).
ii. **Control for confounding sources of variation**. Studies are more robust if they control for sources of variation other than N deposition. This is critically important in observational studies which examine change over space or time, where other variables may also be changing (e.g. climate, land-use, deposition of other nutrients). The robustness of either spatial or temporal gradients relies on a consistent or comparable method of estimation of deposition at the different points in space or time. Application rate(s) tested. Experimental studies should test for the effects of a range of application rates (i.e. > 1 treatment). Ideally, the first level of treatment should reflect realistic deposition scenarios.
iii. **Length of study**. Studies are considered more robust if they are conducted over longer time periods, more in keeping with critical load decision-making.
iv. **Nature of exceedance indicator**. Ecosystem responses are preferred to chemical responses as they indicate change to the system beyond uptake of nutrients.

Scoring:

0-1: No N deposition rate reported, but inferred; other environmental factors and sources of environmental change were not controlled for. In experimental studies, no quantification of background deposition.

2: Varying methods of N deposition estimate across sites/time. Methods of deposition estimate not given.

3: Study well-designed but statistical analyses and/or reporting lacking. Effect detected at first treatment level, meaning critical load falls somewhere between control and first level treatment. Perhaps only wet N deposition quantified. Short (i.e. <2 growing seasons) study may increase likelihood of type II error (failure to reject null hypothesis). Lab experiments rather than field experiments where response may not be replicated in ‘real world’ situations. May measure chemical responses only.

4: Study well-designed and reported but effect detected at first treatment level. May measure chemical responses only.

5: Very valuable study for determining critical loads, with no obvious problems.

Statistical analysis to derive critical loads

We proposed a statistical analysis of the evidence presented in Table 1 whereby the probability of “significant harmful effects” is modelled as a function of N deposition using logistic regression. Using studies with a robustness score of 3 or above (with studies scoring lower having insufficient information to be included in any analysis), each evidence item was summarised into a binary variable (0 – no observed effect in exceedance indicator; 1 – observed effect in exceedance indicator) linked to the reported N deposition level. Critical loads have previously been set based on evidence from a broad range of exceedance indicators. By converting information in each evidence item to this binary response, we were also able to pool information from a variety of exceedance indicators.

An appropriate model to analyse these data where the response variable is binary (i.e. ‘no effect’/’effect’) and the explanatory variable is continuous (i.e. N deposition rate) is the logistic regression model. Here the explanatory variable predicts a probability score that the response variable will be in state 0 or 1, and it uses the logistic function

$$F(x) = \frac{1}{1 + e^{-(\beta_0 + \beta_1 x)}}$$

where *x* is the independent variable and the function *F(x)* is constrained to have only values between 0 and 1 so it can be used to predict a probability score (see Figure 2 for an example of this type of curve). Logistic regression is one example of a generalised linear model – these differ from ordinary linear regression in that they use error distributions other than the Normal – and in this case the binomial distribution is appropriate (see e.g. Nelder & Wedderburn 1972 for more details) and the model is fitted using a maximum likelihood optimisation. The model fitting process also should include an assessment of the adequacy of the chosen model, such as described below.

Model fit evaluation

The Wald test statistic can be used to determine whether a parameter (i.e. the effect of the independent variable on the dependent variable) is significantly different from zero (see Fox 1997 for formulae). This helps determine whether the independent variable has significant explanatory power in the model (i.e. if N deposition rate explains a significant amount of variation in the binary response). The likelihood ratio test and lagrange multiplier test are also commonly applied to determine the significance of a parameter in logistic regression models.

The observed data can be partitioned, probabilities and standard errors generated for the observed data and then plotted for comparison with the model-fitted logistic curve. This allows a visual inspection of how closely the model fits the observed data (see Crawley 2005). The Hosmer-Lemeshow[1] test for logistic regression models may also be applied to assess quantitatively the ‘goodness-of-fit’ by comparing observed and model-predicted event rates. However, on small datasets, these test statistics may not be powerful and it might be more informative to compare model fits through bootstrapping (see below).

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[1] Hosmer, David W.; Lemeshow, Stanley (2000). *Applied Logistic Regression*. New York: Wiley. ISBN 0-471-61553-6.

Estimating uncertainty

In order to quantify uncertainty, the initial step is to define empirically the point below which it can be said that 'no harm' has been detected. Harmful effects can be said to have occurred either at the point which the fitted model estimates a chance of damage that is significantly different from zero (e.g., point 'A' in Figure 1) or at an arbitrarily-decided threshold (e.g. 20% chance that harm has occurred; point 'B' in Figure 1), and both may be useful. In the case of the second method (B) of determining the critical load, a 95% confidence interval can be generated using a bootstrap routine, which can then be used to define a range of values for the critical load (illustrated by the blue bar in Figure 1).

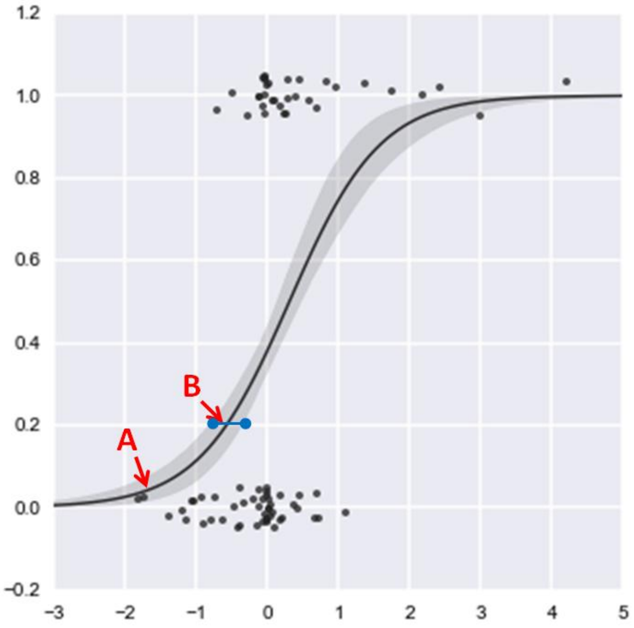

Figure 1. Illustration of logistic regression model. This figure is modified from Waskom (2013) and does not depict data on N deposition and its effect. In the context of this example, the x axis represents level of N deposition and the y axis is probability of 'harmful effect' observed (where observed data only take values of 0 or 1). Points are 'jittered' to improve visual interpretation. The black line gives the logistic regression curve. The 95% confidence interval is shaded in grey. Points A and B illustrate two definitions of 'harmful effects', respectively: when the logistic curve significantly differs from zero, or when line crosses 20% probability.

Results

We collated information from 30 studies, with the dates of studies ranging from 1992 to 2009 (Table 1). 19 evidence items had a robustness score ≥3, and were subsequently translated into a binary variable.

Studies typically focused on two kinds of exceedance indicator: (1) chemical and (2) ecosystem-level responses (Box 2). The indicators monitored most frequently amongst studies were moss height growth, biomass growth, N concentration in plants and species cover change, including changes in biomass of vascular plants.

Box 2. Exceedance indicators in studies examining the effects of N deposition on bog/mire/fen systems

Chemical

- N concentration in peat or water (retention in system)
- Total N (or specific N form) concentration in plant material
- Total N in plant material (N% multiplied by biomass)
- Stoichiometry (C: N and N:P ratios)

Ecosystem

- Moss height growth
- Moss biomass growth
- Moss bulk density
- Capitula morphology
- Survivorship
- Plant species cover change
- Vascular plant cover or biomass change
- Ellenberg (N) scores
- Species extinction
- Decomposition rate and/or litter accumulation
- Microbial biomass and stoichiometry
- Phosphatase activity

In addition to comparisons between treatment and control, occasionally the response to N deposition was described by a mathematical function (i.e. regression slope) for chemical exceedance indicators (e.g. Aaby 1994; Lamers et al. 2000; Tomassen et al. 2003; Wiedermann et al. 2009). No mathematical functional responses were reported for ecosystem indicators in the papers reviewed.

The minimum value at which ecosystem effects were observed was 2.5 kg N ha^{-1} yr^{-1}, and this was based on a linear regression between N deposition and a chemical exceedance indicator (N concentration in capitula and stems of *Sphagnum fallax*) (Tomassen et al. 2003; cited in Bobbink & Hettelingh 2011). In the context of critical loads setting, this evidence has been interpreted as a harmful effect (Bobbink & Hettelingh 2011) though it remains debated whether chemical indicators do denote harmful ecosystem change. In the regression analysis, the overall relationship between N deposition and N concentration was significant, but it is not possible to determine whether significant difference had occurred between pairs of treatment levels (e.g. between control and the first treatment level). Of the studies surveyed, the second lowest deposition at which an effect was reported was 8 kg N ha^{-1} yr^{-1} (Wiedermann et al. 2009).

14 evidence items were excluded where robustness scores were less than 3 (cf. Table 1), primarily due to poorly-estimated or unknown rates of background N deposition, and where there was potential for other confounding effects, leaving 19 evidence items from 12 papers (Table 2). Of the experimental-addition studies with sufficient evidence to be useful in determining critical loads, there were three fundamental, sometimes related problems: i) a high deposition rate as the first level of treatment, ii) an effect detected at the first treatment level and iii) no gradient in treatments (only one treatment level). As such, when the result of the evidence item was translated into a binary variable (effect/no effect) all except one reported an effect at the specified rate of deposition

(Table 2). Therefore, there was little information to constrain the N deposition level at which *no* harmful effects occur through the proposed logistic regression approach. Furthermore, it is often unclear whether control plots themselves have been subject to detrimental effects of atmospheric deposition, and thus the comparison is not between an 'affected' and 'unaffected' system.

Table 2. Summary of effects observed at given N deposition rate

Reference	**Location**	**Effect (0/1)**	**First level of N deposition where effect observed (kg ha^{-1} yr^{-1})**	**Lowest estimated background deposition (kg ha^{-1} yr^{-1})**
Aerts et al. (1992)	N Sweden	1	20.6	2
Aerts et al. (1992)*	S Sweden	0	49	9
Berendse et al. (2001)	Finland	1	34	4
Berendse et al. (2001)	Sweden	1	38	8
Berendse et al. (2001)	Switzerland	1	48	18
Berendse et al. (2001)	Netherlands	1	69	39
Gunnarsson & Rydin (2000)	S Sweden (Akhultmyren)	1	10	7.2
Gunnarsson & Rydin (2000)	S Sweden (Kopparasmyren)	1	10	7.2
Gunnarsson & Rydin (2000)	N Sweden (Luttumyren)	1	10	4.2
Norbakken et al (2003)	Norway	1	12.9	7.9
Bragazza et al. (2004)	Pan-Europe	1	10	1
Phuyal et al. (2008)	Scotland	1	64	8
Tomassen et al. (2003)	Netherlands	1	2.5	0
Wiedermann et al. (2009)	N Sweden (LD)	1	15	2
Wiedermann et al. (2009)	S-N Sweden	1	8	2
Breeuwer et al. (2009)	Sweden	1	40	0
Limpens et al. (2003)	Netherlands	1	40	0
Heijmans et al. (2001)	Netherlands	1	50	15
Redbo-Torstensson (1994)	Sweden	1	11	0.6

*Observational study. Height growth and productivity of *Sphagnum* increased with applications of P but not N; due to previous high background deposition of N it was considered that the system had become P limited and was therefore not responding to subsequent additions of N.

To illustrate the analysis and circumvent the problems associated with lack of information for 'no effect', 90 zero cases were added to the dataset (values where N deposition and effect were zero). Modelling this pseudo-dataset using logistic regression demonstrated that N deposition had a significant, positive effect on observation of damage (*P* value <0.001; Figure 2). In this example, the confidence intervals are artificially narrow because of the addition of non-observed, non-varying zero values (Figure 2).

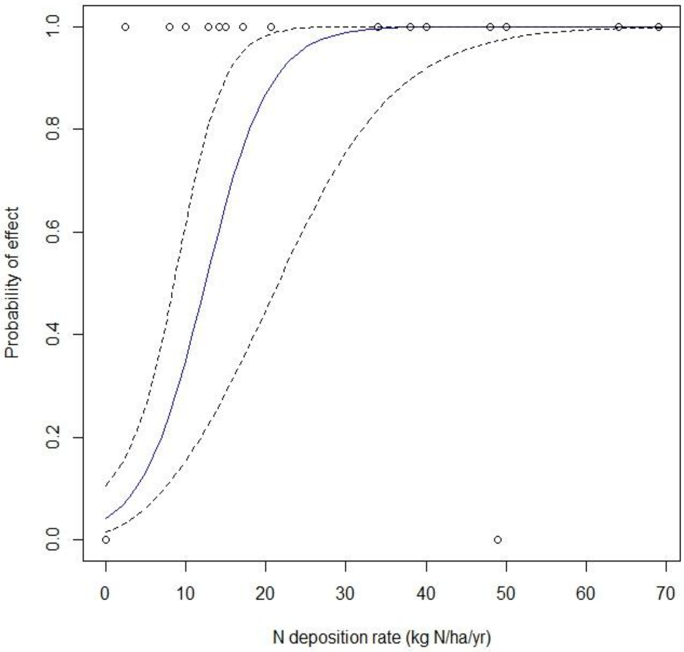

Figure 2. Logistic regression of pseudo-dataset. Points show data for 19 evidence items given in Table 2 and additional 90 zero values. Solid blue line shows relationship modelled by logistic regression. Dotted lines present 95% confidence interval.

Discussion

A broad range of N exceedance indicators are considered in the literature, covering both chemical and biological responses. Most commonly, N concentration in plant tissues and the environment (e.g. soil and interstitial water) is measured and found to increase with increased N deposition rates – as such, it could be said the N deposition affects an ecosystem from the lowest deposition load of N – and therefore that there is no critical load. However, it is debatable whether this change constitutes harm, or potential for harm. The relationship between N in tissues and application rate may saturate (Lamers et al. 2000), indicating the point at which bog communities can no longer accumulate N (i.e. act as filters). Lamers et al. (2000) suggested that a shift from N limited to P-limited systems occurred where N concentration in plant tissue increased and then plateaued with increasing N deposition indicating a saturation of N in the system. Studies have also considered stoichiometric changes (e.g. N:P ratios) which indicate the nature of nutrient limitation. In terms of ecosystem measures, moss height increment, biomass growth and species cover changes are common indicators of N exceedence. These indicators showed more variable responses to N deposition, likely reflecting the more complex mechanisms involved which are reliant on conditions prior to increase in N (i.e. conditioning in the ecosystem), previously extant species and/or previously accumulated N.

Due to the lack of information at low deposition rates, it was not possible to conduct the logistic regression analysis using real data to generate uncertainty around a quantified critical load. This may be possible following further targeted review of evidence available in the literature and including studies published most recently. Evidence of harm (or of no harm) at low N deposition rates will facilitate an improved quantitative understanding of thresholds of change.

Alternative approaches and consideration of additional evidence

It may be possible to make use of absolute values presented in studies, rather than coding by 'effect' or 'no effect'. One example of this is the small meta-analysis of Lamers et al. (2000) which demonstrates the sigmoidal relationship between N concentration in Sphagnum and N deposition rate. This relationship was recently confirmed in a regional survey of 59 peat bogs across Europe (Robroek, in prep). We recommend that similar analyses on other commonly measured indicators, such as biomass increment, be made where possible.

This study considered the simple question of whether and at what rate N deposition affects bog and mire ecosystems. There are several other more detailed questions which are important in the literature but we have not dealt with here. We briefly list these:

- The role of N form - there is some evidence that the effects of N deposition may differ for different forms of N (see Wiedermann et al. 2009b and Risager 1998).
- N deposition may affect different *Sphagnum* species differently, thus generalised interpretations should be viewed with caution (Risager 1998). There is limited evidence on the effects of N deposition on specific species (see Emmett et al. 2011)
- There is evidence for an effect of previous site conditions on N addition – the uptake of additional N may be lower in sites already higher in N deposition (Wiedermann et al. 2009b). Where background deposition has been high, the ecosystem may have become P limited (Aerts et al. 1992; Limpens et al. 2003).
- The effect of wet versus dry deposition may differ (e.g. Sheppard *et al.* 2011).

Conclusions

From the literature surveyed, there is a paucity of information as to how N affects bog and mire ecosystems at low rates of N deposition in Europe. Many experimental addition studies conducted to date have observed effects at the first level of treatment, reducing our ability to determine quantitatively a critical load below which *no* harmful effects are detected. Furthermore, more information is needed about the effects of N deposition over long periods. Indeed for grasslands there is evidence that, on a species-by-species basis there is no detectable lower limit for harmful effects of N deposition (Payne et al. 2013). A review of the most recent literature (post-2009) is required to assess whether there is sufficient additional evidence available to support a quantitative assessment of critical loads and uncertainty.

It may be possible to determine critical 'limits' for certain exceedance indicators (for example, N concentrations in plant tissue), which could demonstrate a saturation limit and a likely shift in ecosystem functioning. This may improve mechanistic understanding, but may not adequately quantify other ecosystem level changes such as changes in species composition and would therefore not necessarily be suitable for establishing critical loads of N deposition. The challenge remains to "benchmark" (i.e. relate a bioindicator result to a non-affected standard) and "intercalibrate" (i.e.

quantitatively relate evidence from a range of bioindicators) such that information from a wide variety of sources can be combined (Sutton *et al.* 2005).

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Appendix 1: Critical Load References

Publications cited in critical loads reports which may contain empirical evidence for the setting of critical loads

Source report	Reference
Berne Report (pp33-37)	Bobbink, R. Et al. (2002) Empirical Nitrogen Critical Loads for Natural and Semi-natural Ecosystems. Update. Background document for the expert workshop held under the UNECE convention on long-range transboundary air pollution, Berne, Switzerland 11-13 November 2002
Berne Report (pp33-37)	Kirkham, FW (2001) Nitrogen uptake and nutrient limitation in six hill moorland species in relation to atmospheric nitrogen depositionin England and Wales. Journal of Ecology 89, 1041-1053.
Berne Report (pp33-37)	Mols et al. (2001) Response of Norwegian alpine communities to nitrogen. Nord. J. Bot. 20: 705-712
Berne Report (pp33-37)	Paal et al. (1996) Responses of the Norwegian alpine Betula nana community to nitrogen fertilization. Can. J. Bot. 75: 108-120.
Berne Report (pp33-37)	Woodin & Sullivan (2001) Biological exceedence of the critical load of nutrient nitrogen in the UK. Report to DEFRA.
Berne Report (pp281-293)	Paulissen, M. Et al. (2002) Differential effects of nitrate and ammonium enrichment on base-rich fen vegetation: preliminary results from Scragh Bog, Central Ireland (BERNE REPORT pp 2981-293)
Berne Report (pp281-293)	Bobbink, R. Et al. (1992) Atmospheric deposition and canopy exchange processes in heathland ecosystems. Env Pollution 75: 29-37.
Berne Report (pp281-293)	Gunnarsson & Rydin (2000) Nitrogen fertilization reduces Sphagnum production in bog communities. New Phytologist 147: 527-537.
Berne Report (pp281-293)	Kooijman, A.M. (1993) Changes in the bryophyte layer of rich fens as controlled by acidification and eutophication/ PhD thesis, Utrecht University.
Berne Report	Nordbakken et al. (2003) Boreal bog plants: nitrogen sources and uptake of recently deposited nitrogen. *Environmental pollution* **126**: 191-200.
Berne Report	Redbo-Torstensson (1994) The demographic consequences of nitrogen fertilization of a population of sundew, Drosera rotundifolia. Acta Bot. Neerl. 43(2): 175-188.
Berne Report	Lamers et al. (2000). Natural nitrogen fails in polluted raised bogs. *Global Change Biology* **6**(5): 583-586.
Berne Report	Van Duren at al. (1997). Nutrient limitations in an extant and drained poor fen: implications for restoration. *Plant Ecology* 133: 91-100.

Emmett et al. 2011 JNCC	Armitage, H.F. (2010) Assessing the influence of environmental drivers on the current condition and recovery potential of Racomitrium heath. PhD thesis, Uni of Aberdeen.
Emmett et al. 2011 JNCC	Bobbink, R. Et al. (2010) Global assessment of nitrogen deposition effects on terrestrial plant diversity: a synthesis. Ecol Apps 20(1):30-59
Emmett et al. 2011 JNCC	Curtis, C. Et al. (2005) Nitrogen saturation in UK moorlands: the critical role of bryophytes and lichens in determining retention of atmospheric N deposition. J. Applied Ecol. 42(3):507-517.
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Emmett et al. 2011 JNCC	DeVries, W. Et al. (2010) Use of dynamic soil-vegetation models to assess impacts of nitrogen deposition on plant species composition and to estimate criticial loads: an overview. Ecol Apps. 20: 60-79.
Emmett et al. 2011 JNCC	Edmondson, J.L. (2007) Nitrogen pollution and the ecology of heather moorland, Manhcester Metropolitan University, Manchester.
Emmett et al. 2011 JNCC	Emmett, B.A. Et al. (2004)Grazing/nitrogen deposition interactions in upland acid moorland. Contract report to Countryside Council for Wales (Contract no. FV-73-03-89B) and the National Assembly for Wales (Contract no. 182-2002) pp96
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