The role of afforestation and tree species change in nitrate leaching from forest ecosystems

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Declaration

I confirm that this is my own work and the use of all material from other sources has been properly and fully acknowledged.

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Abstract

Elevated nitrogen deposition, caused by traffic, agriculture and industrial emissions, has driven elevated nitrate (NO_3-N fluxes) from European forest ecosystems since the late 20th century. Here, an assessment of interactions between former land use, throughfall Total Inorganic Nitrogen (TIN) and NO_3-N leaching from forest was first carried out by collating a database from the literature. Leaching of NO_3-N was greater in secondary conifer rotations than in conifer-afforested heathland/grassland ($p < 0.05$). The wide variation in NO_3-N leaching fluxes from afforested arable land highlighted the current lack of understanding of the potential for afforestation to improve or threaten water quality. The conversion of conifer plantations to broadleaf forests is increasing across Europe to meet targets to improve forest resilience to pests, disease, and extreme climate events. Due to the changes in soil biochemical processes, the conifer-to-broadleaf conversion was hypothesised to stimulate the breakdown of N-rich organic matter, accumulated during the first rotation, and lead to elevated NO_3-N leaching fluxes. However, a chronosequence study observed that throughfall dissolved organic carbon and NO_3-N fluxes controlled NO_3-N leaching fluxes across the study area, with no significant differences in NO_3-N leaching fluxes observed between stands at different stages in the conifer-to-broadleaf conversion process. A coupled biochemical-hydrological model was then tested in its ability to simulate NO_3-N leaching fluxes from a mature *Pinus sylvestris* forest long-term, to then simulate land-use change scenarios. However, the model could not be successfully parameterised. Much of the existing knowledge on NO_3-N leaching from forest ecosystems is based on past observed data, which limits evidence-based decision-making for forest nutrient management, particularly under changing forest management, climate change and reductions in N deposition. To support much needed efforts to parameterise detailed biochemical-hydrological models, a framework to improve the parameterisation procedure of such models is proposed.

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Contents

Abbreviations

Biome-BGCMuSo	Biome Biogeochemical Multi-soil-layer model
C:N	Carbon to nitrogen ratio
Cl^-	Chloride
ICP Forests	International Co-operative Programme on Assessment and Monitoring of Air Pollution Effects on Forests.
IFEF	Indicators of Ecosystem Functioning database
Kg-N ha^{-1} yr^{-1}	Kilograms of nitrogen per hectare per year
NH_4-N	Ammonium nitrogen
NO_3-N	Nitrate nitrogen
NO_x	NO_2, NO
PAWS	Planted Ancient Woodland Sites (former native woodland replanted with conifer plantations)
TIN	Total Inorganic Nitrogen
WFD	EU Water Framework Directive

Introduction

1. The significance of nitrogen pollution from forest ecosystems

The leaching of NO_3-N from soils to surface and groundwater bodies threatens public water supplies and damages aquatic ecosystems. Dominant sources of NO_3-N pollution in Europe are agricultural fertiliser applications and sewage effluents (Wang *et al*., 2016; EEA, 2005), accounting for 70 % and 20-30 % of NO_3-N leaching in the UK (Environment Agency, 2019). The EU Nitrate Directive, under the Water Framework Directive (WFD), states that NO_3^- concentrations in drinking water must remain below 50 mg/L (EEC, 1991), equivalent to 11 mg/L NO_3-N. Many areas of high N loading frequently exceed this limit (e.g. Whiteman *et al*., 2023; Stuart *et al*., 2016) and the costs of NO_3-N removal or blending are high (House of Commons, 2018). High levels of NO_3-N in surface waters causes eutrophication and acidification, which is detrimental to ecosystem functioning (Pretty *et al*., 2003; Withers *et al*., 2014). Despite efforts to reduce NO_3-N leaching from soils, research has found that there is a time lag between reductions in fertiliser inputs and the time NO_3-N reaches the water table due to travel through and accumulation of NO_3-N in the unsaturated zone (the zone between the soil zone and groundwater/saturated zone) (Wang *et al*., 2013). The impact of changes in land management on groundwater NO_3-N contamination may, therefore, not be seen for decades (Ascott *et al*., 2016).

Commonly, policy interventions targeting NO_3-N pollution often account for pollution from agricultural activities, sewage, and urban environments (Comber et al., 2013; Zhang et al., 2014), but forest ecosystems are an often-overlooked source of potential

NO_3-N leaching. Leaching of NO_3-N from forest ecosystems was generally assumed to be low, with concentrations of NO_3-N in deep soil solution below the 11 mg/L WFD threshold (Gundersen *et al.*, 2006). Due to low levels of NO_3-N leaching fluxes from forest ecosystems, it was sometimes assumed that groundwater recharge derived from from forested land will dilute NO_3-N-concentrated recharge from agricultural land (Sundermann *et al.*, 2020; Calder *et al.*, 2002). However, evidence shows elevated nitrogen deposition lead to increased NO_3-N leaching from forest ecosystems from the 1980s onwards, particularly in the UK and central Europe (e.g. Vanguelova *et al.*, 2024; Vanguelova *et al.*, 2010; Vanguelova *et al.*, 2007; Macdonald *et al.*, 2002; Dise *et al.*, 1995). Forestry practices such as clear felling and ploughing (e.g. Mupepele & Dormann, 2016; Jerabkova *et al.*, 2011; Nisbset *et al.*, 2011; Gundersen *et al.*, 2006) and unanticipated events, e.g. storms and insect infestations (Olsson *et al.*, 2022; Pitman *et al.*, 2010), can also lead to significant peaks in NO_3-N leaching from forest ecosystems, with concentrations of NO_3-N in soil solution above the 11 mg/L threshold.

Elevated NO_x and NH_3 emissions from traffic, industrial processes and agricultural practices led to elevated wet and dry deposition of nitrogen to forest ecosystems from the 1980s-2000's (EEA, 2020; Vanguelova and Pitman, 2019; Hůnová *et al.*, 2017; Janssens *et al.*, 2010). The implementation of EU and national emission-control policies and subsequent reductions in N emissions led to a decrease in average N deposition inputs continentally, with some reduction in NO_3-N leaching in European forest ecosystems (Schmitz *et al.*, 2024; Schmitz *et al.*, 2019), but local emission sources continue to cause high N deposition inputs in some areas, such as those surrounded by intensive livestock farming (Vanguelova *et al.*, 2024).

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Availability of N historically limited ecosystem productivity (Vitousek *et al.*, 2010). Aber *et al.* (1989) hypothesised that in N-limited ecosystems, there should be a lag between an increase in N deposition inputs and increases in NO_3-N leaching due to the capacity of N-limited forest ecosystems to use and retain N-inputs. Leaching of NO_3-N from forest ecosystems is minimal until the availability of other nutrients becomes limiting (Braun *et al.*, 2010). Research on the relationship between N deposition and NO_3-N leaching from European forest ecosystems established that total throughfall inorganic N (NO_3-N and NH_4-N) was the main predictor of NO_3-N leaching early on (Dise *et al.*, 2009; van der Salm *et al.*, 2007; Dise *et al.*, 1995). Dise *et al.* (1995) observed that NO_3-N leaching from forest ecosystems was minimal below a throughfall TIN <10 kg-N ha^{-1} yr^{-1}, then increased (although with much variation) between 10-25 kg-N ha^{-1} yr^{-1}, and was consistently elevated where throughfall TIN >25 kg-N ha^{-1} yr^{-1} (Dise *et al.*, 1995). Above the 25 kg-N ha-1 yr-1 deposition input threshold, TIN leaching fluxes varied between 9 and 35 kg-N ha^{-1} yr^{-1} (Dise *et al.*, 1995). In a later analysis of the IFEF database, out of 32 forests receiving an N deposition >30 kg ha^{-1} yr^{-1}, 12 displayed TIN leaching fluxes >25 kg-N ha^{-1} yr^{-1}, with the highest observed leaching flux at approximately 39 kg-N ha^{-1} yr^{-1} (Dise *et al.*, 2009). Although the highest TIN leaching fluxes from forests are frequently less than fluxes from agricultural land, which in the UK are estimated to be between 39-88 kg-N ha^{-1} yr^{-1} (Silgram *et al.*, 2001), scenarios where NO_3-N leaching from forests are as high as 39 kg-N ha^{-1} yr^{-1} questions the capability of forest ecosystems to provide “clean” water as an ecosystem service and the potential for groundwater under forests to be used to dilute pollution from agricultural land. Furthermore, as efforts to reduce fertiliser use reduce N inputs from agricultural land, NO_3-N leaching from forest ecosystems may in future equal those from agricultural land.

Clearcutting has been observed to increase NO_3-N leaching to the point where forest fluxes could match those from agricultural land per hectare basis. For example, conventional harvesting led to NO_3-N leaching fluxes of 208 kg-N ha^{-1} over a year (average of 52 kg-N ha^{-1} yr^{-1}) in Beddgelert forest, Wales (Reynolds, 2004). Modelled NO_3-N leaching fluxes from clearcut forests in Sweden were estimated to be up to 50 kg-N ha^{-1} yr^{-1} (Futter *et al.*, 2010). N-saturated forest soils may generate higher levels of NO_3-N leaching post-harvest than N-balanced or N-limited forest ecosystems. In Sweden, forests receiving low N deposition inputs leached <5 kg-N ha^{-1} yr^{-1} during the five years post-clearcut, whereas those receiving high N deposition leached 25-35 kg-N ha^{-1} yr^{-1} (Akselsson *et al.*, 2004). In N-addition experiments in Switzerland, where forests were subjected to treatments for 15 years before felling, forests receiving no additional N displayed peaks in NO_3-N leaching of 5 kg-N ha^{-1} yr^{-1}. In contrast, fertilised forest displayed 20 kg-N ha^{-1} yr^{-1} peaks post-harvest (Schleppi *et al.*, 2017). Forest management practices must be considered within the context of historical air pollution and N accumulation within the ecosystems and soils.

Whether forestry operations significantly impact surface and groundwater NO_3-N concentrations is dependent upon the level of disturbance and the area of land disturbed. Welsh clearcuts where <20 % of a catchment was felled within three years showed minimal impacts on streamwater chemistry, but small catchments <10 ha in size displayed marked effects on streamwater chemistry after a 100 % clearcut (Reynolds, 2004). Finnish catchment-scale studies showed spatial variability in groundwater NO_3-N concentrations, with concentrations increasing in one well but not in another in the three years post-harvest, where 30 % of the catchment was clearcut (Mannerkoski *et* *al.*, 2005). Palviainen *et al.* (2014) observed significant increases in runoff NO_3-N, which were only significant when 30 % of the catchment area was felled. The Finnish and Welsh catchment studies were conducted in areas with high water tables, and the impact of forestry operations was observed in groundwater NO_3-N concentrations less than a decade later. A 5-year study in Nottinghamshire, England, where the water table typically sits at 20 m below the soil surface, noted elevated NO_3-N leaching post-harvest of a Corsican pine stand would not likely impact groundwater nitrate concentrations until 30 years later, due to the unsaturated zone thickness and geology (Calder *et al.*, 2002). Quantification of NO_3-N leaching from forest ecosystems, particularly those subject to disturbance, in areas with deep water tables is needed to understand the potential future N-loading from forests to groundwater.

1.1. The effects of soil NO_3-N leaching on European forest ecosystem health and functioning

In addition to surface and groundwater quality concerns, elevated NO_3-N leaching has detrimental impacts on the health and functioning of forest ecosystems. Elevated N deposition inputs in N-limited systems initially encourage tree growth (Janssens *et al.*, 2010). In an analysis of data from 442 European plots between 1995-2010, N deposition was the driving factor for the increasing tree growth rate, displaying a greater signal than ozone and climate-related indicators (Etzold *et al.*, 2020). Increasing N deposition increased the growth rate in Norway spruce, Scots pine, oak and beech until a tipping point around 30 kg-N $ha^{-1}yr^{-1}$, with variation between species (Etzold *et al.*, 2020). In N-saturated soils, the availability of nutrients, such as phosphorus, magnesium and potassium, becomes limiting to tree growth and, therefore, the capacity of trees to retain N (Sardans et al., 2016; Braun et al., 2017; Braun et al., 2010).

Despite increasing vegetation nutrient demand, NO_3-N leaching reduces soil nutrient availability. Due to the negative charge of NO_3^- anions, NO_3-N leaching results in the leaching of base cations, such as Mg^{2+} and $K^{+,}$ from the soil (Braun *et al.*, 2020; Watmough *et al.*, 2005). NO_3^- mobilises heavy metals, especially Al^{3+}, which are toxic to fine root systems, reducing vegetation capacity for nutrient uptake (Brunner & Sperisen, 2013; Vanguelova *et al.*, 2007b; Vanguelova *et al.*, 2005; Ericsson *et al.*, 1995). Elevated N deposition can lead to shifts in microbial community composition (Tahovská *et al.*, 2020; Zechmeister *et al.*, 2011; Högberg *et al.*, 2007; Nilsson *et al.*, 2007), decreases mycorrhizal species diversity (Suz *et al.*, 2014), and a reduction in the growth of mycelial networks (Nilsson *et al.*, 2007), further reducing the capacity of forest soils to retain NO_3-N. Damage to mycorrhizal communities that facilitate P uptake can not only result in a reduction in P uptake but also N uptake at high levels of N deposition (Braun et al., 2020; De Linder et al, 2018), reducing the capacity of trees and the whole forest ecosystem to retain and recycle N sustainably. Deficiencies in foliar nutrients, indicated through high foliar N:other nutrient ratios, have been observed across Europe (Jonard *et al.*, 2015; Sardans *et al.*, 2016). Foliar nutrient deficiencies resulting from the effects of elevated N deposition and NO_3-N leaching increases tree susceptibility to insect infestations and disease, such as Acute Oak Decline (Brown *et al.*, 2018; Cienciala *et al.*, 2017; Thomas *et al.*, 2003).

2. Drivers of NO_3-N leaching fluxes in European forest ecosystems

European-scale long-term forest monitoring networks have generated data to support our understanding of factors that create variation in NO_3-N leaching fluxes between and within forests. Whilst throughfall total inorganic nitrogen (TIN) concentrations typically explained 46-70 % of variation in average annual total NO_3-N leaching fluxes (Dise *et al.*, 2009; van der Salm *et al.*, 2007; Macdonald *et al.*, 2002; Dise *et al.*, 1995), other factors that explained NO_3-N leaching fluxes included mean annual temperature, annual precipitation, and B horizon pH (van der Salm *et al.*, 2007; Dise *et al.*, 1995). Organic soil C:N ratios of <23 indicated soils with a high potential for NO_3-N leaching (Dise *et al.*, 2009; Gundersen *et al.*, 2009; van der Salm *et al.*, 2007; Gundersen *et al.*, 1998).

2.1. The role of tree species in NO_3-N leaching fluxes

Tree species, tree age and soil type also affect soil NO_3-N leaching fluxes, but there are contradictory studies as to how. Typically, NO_3-N leaching fluxes from broadleaved forests are lower than those from coniferous forests. A study directly comparing conifer and broadleaved stands in 15 different forests found low concentrations of NO_3-N in soil solution below beech forests (<10 mg/L). In contrast, concentrations under spruce stands ranged from 10-40 mg/L (Rothe *et al.*, 2002). Where stands of broadleaf and coniferous species were planted side-by-side at two sites with different land use histories; however, N leaching was higher in oak and beech than spruce despite spruce having the highest throughfall deposition, but N leaching from lime and maple was lower than spruce (Christensen et al., 2010). The greater scavenging ability of coniferous forest canopies can lead to greater soil N content than broadleaf forests under similar deposition gradients (Vanguelova & Pitman, 2019), and the higher water use of coniferous stands can also lead to higher levels of NO_3-N in soil leachate in coniferous forests compared to broadleaf forest in dry areas due to a concentrating effect resulting from lower drainage volumes (Calder et al., 2002; Nisbet et al., 2011). Limited differences in the response of NO_3-N leaching fluxes to elevated throughfall TIN deposition were observed in studies conducted on the ICP Forests Level II long-term monitoring and Indicators of Ecosystem Functioning datasets (van der Salm *et al*., 2007; Macdonald *et al*., 2002). Kristen *et al*. (2004) observed higher deep soil solution concentrations of NO_3-N under broadleaved stands than coniferous stands subjected to the same deposition inputs. Kristen *et al*. (2004) attributed their observation to the tendency for broadleaf forests to grow on nutrient-rich soil types, but also due to the tendency for conifer forests subjected to low N deposition to be located in Scandavian countries where N is limiting and therefore mostly retained by the ecosystem. Comparisons between broadleaf and conifer forests in the Level II and IFEF long-term monitoring datasets have been limited by the underrepresentation of broadleaf sites and have limited representation of broadleaf sites growing in geographical regions subjected to high levels of N deposition (Gundersen *et al*., 2009; van der Salm *et al*., 2007; Macdonald *et al*., 2002).

2.1.1. Mechanisms underlying the influence of tree species on NO_3-N leaching fluxes

Seasonal variations in dry deposition and phenological differences between broadleaf and coniferous species result in differences in throughfall TIN seasonal patterns. Nitrification in forest canopies by phyllosphere microbial communities generates higher concentrations of NO_3-N throughfall than rainwater (Guerrieri *et al*., 2024; Guerrieri *et al*., 2020; Guerrieri *et al*., 2015). The stimulation of canopy nitrification by dry deposition combined with washing dry deposition from the canopy surfaces further affects the balance of NO_3-N and NH_4-N in throughfall (Guerrieri *et al*., 2024; Guerrieri *et al*., 2020; Guerrieri *et al*., 2015). Under elevated deposition, broadleaf forest tends to have higher concentrations of NO_3-N in throughfall, whereas conifer forest has higher concentrations of NH_4-N (Schrijver *et al.*, 2007).

Soil microbial communities that influence NO_3-N leaching vary between coniferous and broadleaved species. Broadleaf trees are more commonly associated with arbuscular mycorrhizae (AM), but coniferous trees are more commonly associated with ectomycorrhizal (ECM) species (Midgley & Phillips, 2014). There is some variation; for example, maple commonly forms AM associations, whereas oak forms ECM associations (Nilsson *et al.*, 2007). Forests with AM associations have greater N leaching than those with ECM associations due to the ability of ECM to quickly capture N released from soil organic matter (Midgley & Phillips, 2014; Zechmeister *et al.*, 2011). Tree species associated with AM produce litter with lower C/N ratios than ectomycorrhizal fungi and are associated with a higher rate of N mineralisation (Midgley & Phillips, 2014). Observations across Europe have found broadleaf stands generally have lower soil C/N ratios than coniferous stands (Gundersen *et al.*, 2009; Cools *et al.*, 2014; Ribbons *et al.*, 2018), explained by differences in organic matter decomposition and litter chemistry (Hobbie *et al.*, 2006; Hansson *et al.*, 2011). The effect of tree species on soil C/N ratio influences soil pH, which drives differences in ammonia oxidising microbial community composition, affecting the rate of soil microbial N transformation processes, including gross and net ammonification and nitrification (Ribbons et al., 2018). Nitrification converts NH_4^+ to NO_3^-, a more mobile form of N and, therefore, the N compound most associated with N-leaching. The stimulation of nitrification has frequently been cited as the mechanism behind elevated N-leaching following periods of elevated TIN deposition and forestry practices (Dise *et al.*, 1995; Stevens & Hornung, 1990). Higher mineralisation and nitrification rates in beech soils compared to Norway spruce soils were thought to be the underlying cause of higher NO_3-N leaching from beech (Oulehle *et al.*, 2010).

There are also tree age effects on NO_3-N leaching. After planting and establishment, NO_3-N leaching fluxes from coniferous forests have been observed to be low initially, due to high demand of N for tree growth and lower plants, but then increased with tree age and canopy development as throughfall deposition increased (Hansen *et al.*, 2007; Rosenqvist *et al.*, 2007; Reynolds, 2004; Emmett *et al.*, 1997) and less N is taken up by trees and understory. The same was observed in oak chronosequences, but NO_3-N leaching fluxes from young oak forests were higher than those from older oak (Hansen *et al.*, 2007; Rosenqvist *et al.*, 2007). An oak chronosequence on clay soils covering 200 years of development observed higher levels of total N accumulation in older stands, but higher levels of NH_4 in soil solution indicated lower rates of nitrification in older stands (Pitman *et al.*, 2013), suggesting lower rates of N transformation could limit NO_3-N leaching.

3. Conifer-to-broadleaf conversion: Policy interest and common forestry practices in the UK

While there is a vast literature base on disturbance, tree species, tree age, and nitrogen deposition as drivers of NO_3-N leaching fluxes, studies specifically investigating tree species change are rare. In the past 30 years, conifer harvesting, conifer restocking and afforestation scenarios have been researched extensively from a water quality perspective (e.g. Broadmeadow *et al.*, 2019; Duffy *et al.*, 2020; Shah *et al.*, 2021). Despite gaining support in policy and practice, the conifer-to-broadleaf conversion process has received little attention in terms of the potential changes in soil biochemical cycling and impacts on water quality.

The UK has 1,566,000 ha of coniferous forest, accounting for 48 % of forested land (Forest Research, 2023). Following timber shortages in the early part of the 20th century, alongside the afforestation of non-forested land, many ancient woodlands were cleared and faster-growing, non-native (species introduced outside its natural distribution range (Brus *et al.*, 2019) conifer species were planted in their place (Kirby & Thomas, 2011). It was reported that 223,000 ha out of the 552,000 ha classified as ancient woodland sites in the UK are now planted with non-native coniferous species (Planted Ancient Woodland Sites (PAWS)) (Woodland Trust, 2011). Policies encouraging the planting of broadleaves rather than conifers where possible have been in place since the 1980s (Forestry Commission, 1985), and the restoration of PAWS has been encouraged by Government policy since 2005 and reaffirmed in recent years (Defra, 2022; Forestry Commission, 2005). There has also been interest and Government support for converting conifer plantations to native broadleaf forests in mainland Europe since the late 20th century (Dedrick *et al.*, 2007; Zerbe, 2002). For example, the Flemish Forest Law stipulates the ecological restoration of forest ecosystems by promoting forest age and species diversification, encouraging forest conversion (Degrave *et al.*, 2006). Motivations to convert conifer plantations to broadleaf forest cited in studies from Germany and Sweden have included improved resilience against pest and disease, climate change and extreme weather events including storm damage (Löf *et al.*, 2023; Kint *et al.*, 2006; Kenk & Guehne, 2001).

The restoration of native woodland supports native biodiversity (Forestry Commission, 1985; Watkins, 1986) and has additional benefits such as enhanced recreational value (Forestry Commission, 1985; Spracklen *et al.*, 2013). A European project focused on conifer-to-broadleaf conversion highlighted the risks of extreme climatic events, disease, soil degradation and associated decreases in growth and timber quality as the main reasons for converting conifer stands (Dedrick *et al.*, 2007). Other incentives to plant broadleaf forests may be to improve water quantity, increasing the resilience of public water supply to the increasing frequency of droughts expected with future climate change; converting broadleaf forests to coniferous forests decreased soil runoff and drainage volumes, so the opposite could be assumed in the reverse change (Komatsu *et al.*, 2008).

The preferred method of conifer-to-broadleaf forest conversion in mainland Europe is a transitional pathway, using continuous cover forestry or thinning and natural regeneration (Dedrick *et al.*, 2007; Zerbe, 2002). Continuous-cover forestry (CCF) approaches are preferred as a way of minimising NO_3-N leaching fluxes from coniferous plantations during harvesting (Webster *et al.*, 2022; Marozas *et al.*, 2018; Mupepele & Dormann, 2016; Weis *et al.*, 2006). However, the conversion of coniferous sites to broadleaf cover in the UK will likely feature large areas of clearcut followed by broadleaved seedlings. Patch clearcutting, often exceeding five hectares at a time, has historically been the preferred method of harvest in British conifer plantations (Reynolds, 2004), primarily due to historical policies and tax restraints in the 20th century that have resulted in unthinned coniferous plantations that are difficult to transform (Helliwell & Wilson, 2012). CCF is heavily encouraged in PAWS restoration guidance

(Woodland Trust, 2020) but may remain impractical in ssome scenarios (Confor, 2016; Harmer & Thompson, 2013), and a woodland manager survey revealed an even split between managers who have or planned to use CCF and those who would use a clear-cut approach to PAWS restoration in the UK (Barsoum & Henderson, 2016).

Due to tree age-related increases in NO_3-N leaching, biochemical modelling of Austrian conifer forests showed that long-term average NO_3-N leaching fluxes from CCF systems can be greater than shelterwood-cut or clearcut systems despite the peak in NO_3-N leaching fluxes caused by harvesting disturbance in the latter two scenarios (Dirnböck *et al*., 2016). Removing conifer trees can reduce deposition inputs and NO_3-N leaching fluxes long-term despite initial peaks (Göttlein *et al*., 2023; Gundersen *et al*., 2006). In the conifer-to-broadleaf scenario, there are several factors which discourage a CCF or shelterwood-cut approach, such as the variable success rates of establishing young broadleaved trees and understorey species under a conifer plantation (Degrave *et al*., 2006; Seiwa, 2007; Kostel-Hughes *et al*., 2005; Ammer *et al*., 2002; Szwagrzyk *et al*., 2001), reduced timber quality of conifers from thinned systems, sawmill processing limitations (Macdonald *et al*., 2010), high wind risk in upland locations (Rayner & Nicoll, 2012), mechanical restraints in certain soil types, and diminishing expertise in skilled forestry practices (Ireland *et al*., 2006). Broadleaf sapling density and height were higher under clearcut conversion sites than under partial cutting in German forests 6-years after harvest (Seliger *et al*., 2021). Economic incentives in the UK still typically favour tree planting over other methods of forest establishment, such as natural regeneration and seeding, further discouraging CCF (Mason, 2020).

In the UK, the history of elevated N deposition has led to an accumulation of nitrogen in the organic soils of coniferous plantations (Vanguelova *et al*., 2024). Accumulated N, combined with the disturbance of clearcutting and planting and higher rates of mineralisation and nitrification in broadleaf soils, may lead to a release of N from the organic layer and an increase in NO_3-N in the years following conifer-to-broadleaf conversion. A study on the conversion of western hemlock to birch observed elevated DOC and total N release from mulch treatments 4-7 years after post-conversion (Pitman & Peace, 2023), but whether this translated into elevated leaching fluxes was not studied. The potential for forestry operations to induce peaks in NO_3-N leaching fluxes is particularly of concern in nitrate vulnerable zones. Long-term monitoring has identified Thetford Forest in the region of East Anglia as that with the highest rates of NO_3-N leaching in the UK in the past 25 years (Vanguelova *et al*, 2024). The region is situated atop the chalk aquifer, which provides 30 % of England's drinking water (Farrell & Whiteman, 2023), and the area is designated as a nitrate vulnerable zone. Solutions to reduce NO_3-N leaching from the land surface are needed. Increasing broadleaf cover to improve water quality is therefore of interest but due to the high N accumulation in the conifer soils, an understanding of the effect of converting stands to broadleaf cover on N cycling is needed to inform management decisions. Thetford Forest was therefore selected as the region to focus research efforts on for this thesis.

4. Case Study: Thetford Forest

Thetford Forest is a commercial plantation dominated by Scots and Corsican pines but with interspersed broadleaved stands, particularly along woodland block edges. The land surrounding the forest is used for intensive pig and chicken farming, and cropland is frequently subjected to fertiliser applications. Atmospheric N deposition across the region is high due to intensive farming activities; the area is situated within a nitrate-vulnerable zone (Environment Agency, 2019). Long-term monitoring since 1996, shows that N deposition inputs have not declined in Thetford, despite the national trend in decreasing N deposition, and evidenced a shift in deposition inputs from NO_x compounds to NH_3 (Vanguelova *et al.*, 2024). NO_3-N leaching from forest soils has historically been high, sometimes exceeding 40 kg-N ha^{-1} yr^{-1} (Vanguelova *et al.*, 2007), with soil NO_3-N concentrations higher at woodland edges than >200 m from the edge (Vanguelova & Pitman, 2019). Typical forestry practices in past decades in Thetford forest have used patch clearcuts, coarse woody debris removal and ploughing of planting lines (Pedley *et al.*, 2023).

Geological cross-sections of the region showed that most of the forest is located in an area with up to 1 m of sandy soil above a ~1 m layer of sand on the chalk. Some pockets of woodland on the outskirts of the forest have a more complex geology, with strips of clay, marl, river terrace deposits or gravel beneath the sand. The groundwater table lies approximately 20-25 m below the land surface.

The region is the driest part of the UK, with the frequency of heatwaves and drought expected to increase with climate change (Chan *et al.*, 2023; Arnell *et al.*, 2021), and the pine has been affected by pathogens, including *Heterobasidion annosum* and *Dothistroma* needle blight (Tew *et al.*, 2021; Tew *et al.*, 2019). Diversification of forest stands is therefore of interest for (1) improved resilience of the forest to climate change and disease, (2) reduce NO_3-N leaching to protect groundwater quality and improve tree health, and (3) increase volume of groundwater recharge and improve public water supply resilience to drought. Whilst the main management objective is the diversification of forest stands through thinning pine and underplanting broadleaves, a direct transition of stands from conifer-to-broadleaf by clearcut and planting is an option in areas where thinning and underplanting is practically or economically infeasible.

5. Knowledge gaps

The gaps in the literature can be summarised as follows:

(1) The extensive literature base highlights land use change, soil type, tree species and tree age as factors affecting NO_3-N leaching from European forest ecosystems. Interactions between these factors remain unclear.

(2) Land-use history affects soil biochemical processes, but the effect of land-use history on the relationship between throughfall deposition inputs and NO_3-N leaching in mature forests has not been quantified.

(3) Extensive research has established the effect of felling practices and restocking plantations with the same species on NO_3-N leaching. Less literature exists exploring the impact of tree species change post-harvest on N cycling processes, and even less exists on the specific scenario of conifer-to-broadleaf conversion. Considering the growing interest in the scenario, exploring how converting conifer plantations to broadleaf forests may affect N cycling processes, especially NO_3-N leaching and nitrification is important.

(4) The quantification of long-term impacts of forest management change on NO_3-N leaching needs investigating to inform management decisions. Whilst field studies can provide insights into short-term impacts, modelling exercises are needed to predict changes long-term. Whilst detailed hydrological-

biogeochemical models have been applied to quantify NO_3-N leaching fluxes from forest ecosystems, such models have not been applied to explore the long-term effects of conifer-to-broadleaf conversion on NO_3-N leaching, or changes in forest management practices generally.

6. Aims and objectives

The primary aims of this thesis are:

(1) To explore interactions between tree species, tree age, soil type, N deposition, and land use change as drivers of NO_3-N leaching.
(2) Investigate the short-term impact of the conifer-to-broadleaf conversion scenario on soil biochemical cycling and NO_3-N leaching fluxes.

The objectives of this thesis are:

(1) To synthesise a database from published literature to investigate the interactions between throughfall deposition, tree species, soil type and land history as predictors of NO_3-N leaching from European forest ecosystems.
(2) To quantify the biochemical and hydrological processes in a chronosequence of sites in the conifer-to-broadleaf conversion process.
(3) Calibrate a biogeochemical model to predict NO_3-N fluxes in coniferous forest ecosystems.

7. Hypotheses

The hypotheses tested in this thesis are:

(1) Land use history and soil type will affect the capacity of forest soils to retain N inputs, with nitrogen-rich soils having a lower capacity to retain N inputs (Figure 1a) and therefore having a stronger relationship with throughfall TIN deposition. Broadleaved species planted on nitrogen-rich soils will have a weaker relationship between throughfall TIN fluxes and NO_3-N leaching fluxes than conifer species.

(2) Conifer-to-broadleaf conversion will stimulate soil N mineralisation and nitrification (Figure 1b), which will lead to increased NO_3-N leaching fluxes from broadleaved sites within the first decade after planting of clearcut coniferous sites (Figure 1c), above the NO_3-N leaching fluxes observed under the mature conifers.

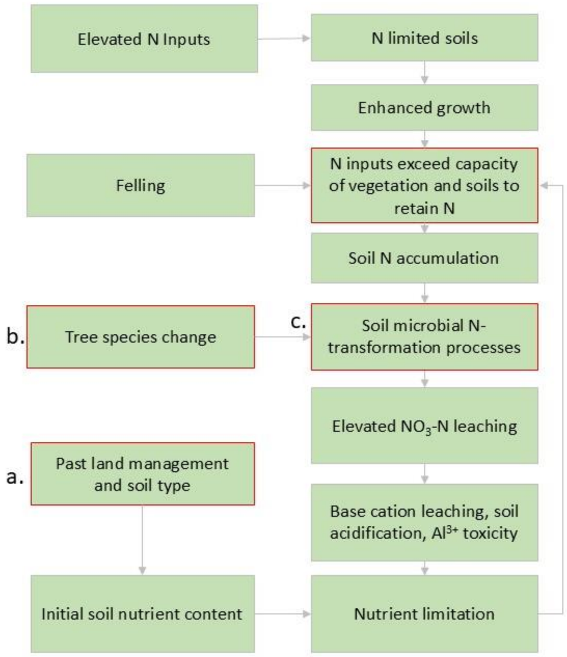


Figure 1: A conceptualisation of NO_3-N leaching from forest ecosystems, driven by deposition, previous land use and forest management changes. Highlighted in red are the components studied in this thesis.

Chapter 2 addresses the first hypothesis from a literature-collated database on NO_3-N leaching fluxes from forests with different former land uses. Chapter 3 addresses the hypothesis 2 through a chronosequence field study, investigating NO_3-N leaching from stands up to 13 years post conifer-to-broadleaf conversion. Chapter 4 attempted to parameterise a model to then apply to study long-term NO_3-N leaching fluxes post conifer-to-broadleaf conversion, but describes several difficulties encountered and then sets out a framework to guide future model users.

References

Aber, J.D., Nadelhoffer, K.J., Steudler, P., Melillo, J.M. (1989) Nitrogen saturation in northern forest ecosystems. *American Institute of Biological Sciences*, 39, 378-386.

Akselsson, C., Westling, O., Örlander, G. (2004) Regional mapping of nitrogen leaching from clearcuts in southern Sweden. *Forest Ecology and Management*, 202, 235-243.

Ammer, C., Mosandl, R., Kateb, H.E. (2002) Direct seeding of beech (Fagus sylvatica L.) in Norway spruce (Picea abies [L.] Karst.) stands - effects of canopy density and fine root biomass on seed germination. *Forest Ecology and Management*, 159, 59-72.

Arnell, N. W., Kay, A. L., Freeman, A., Rudd, A. C., & Lowe, J. A. (2021). Changing climate risk in the UK: A multi-sectoral analysis using policy-relevant indicators. *Climate Risk Management*, 31.

Ascott, M.J., Wang, L., Stuart, M.E., Ward, R.S., Hart, A. (2016) Quantification of nitrate storage in the vadose (unsaturated) zone: a missing component of terrestrial N budgets. *Hydrological Processes*, 30, 1903-1915.

Barsoum, N., Henderson, L. (2016) FCRN024: *Converting planted non-native conifer to native woodlands: a review of the benefits, drawbacks and experience in Britain*. Surrey: United Kingdom.

Braun, S., Schindler, C., Rihm, B. (2017) Growth trends of beech and Norway spruce in Switzerland: The role of nitrogen deposition, ozone, mineral nutrition and climate. *Science of the Total Environment*, 600, 637-646.

Braun, S., Schindler, C., Rihm, B. (2020) Foliar nutrient concentrations of European beech in Switzerland: relations with nitrogen deposition, ozone, climate and soil chemistry. *Frontiers for Global Change*, 3.

Braun, S., Thomas, V. F. D., Quiring, R., & Flückiger, W. (2010) Does nitrogen deposition increase forest production? The role of phosphorus. *Environmental Pollution*, 158, 2043–2052.

Braun, S., Thomas, V.F.D., Quiring, R., Flückiger, W. (2010) Does nitrogen deposition increase forest production? The role of phosphorus. *Environmental Pollution*, 158, 2043-2052.

Braun, S., Tresch, S., & Augustin, S. (2020). Soil solution in Swiss forest stands: A 20 year's time series. *PLoS ONE*, 15.

Broadmeadow, S. B., Nisbet, T. R., & Forster, J. (2019). Trends in surface water chemistry in afforested Welsh catchments recovering from acidification, 1991–2012*. Environmental Pollution*, 247, 27–38.

Brown, N., Vanguelova, E., Parnell, S., Broadmeadow, S., & Denman, S. (2018). Predisposition of forests to biotic disturbance: Predicting the distribution of Acute Oak Decline using environmental factors. *Forest Ecology and Management*, 407, 145–154.

Brunner, I., & Sperisen, C. (2013). Aluminum exclusion and aluminum tolerance in woody plants. *Frontiers in Plant Science*, 4.

Brus, R., Pötzelsberger, E., Lapin, K., Brundu, G., Orazio, C., Straigyte, L., Hasenauer, H. (2019) Extent, distribution and origin of non-native forest tree species in Europe. *Scandanavian Journal of Forest Research*, 34, 533-544.

Calder, I. R. (2004) Forests and water - Closing the gap between public and science perceptions. *Water Science and Technology*, 49, 39–53.

Calder, I. R. (2007) Forests and water-Ensuring forest benefits outweigh water costs. *Forest Ecology and Management*, 251, 110–120.

Calder, I., Reid, I., Nisbet, T., Armstrong, A., Green, J. (2002) *Study of the potential impacts on water resources of proposed afforestation*. England: Loughborough University.

Chan, W. C. H., Arnell, N. W., Darch, G., Facer-Childs, K., Shepherd, T. G., Tanguy, M., & van der Wiel, K. (2023). Current and future risk of unprecedented hydrological droughts in Great Britain. *Journal of Hydrology*, 625.

Christiansen, J.R., Elberling, B., Jansson, P.E. (2006) Modelling water balance and nitrate leaching in temperate Norway spruce and beech forests located on the same soil type with the CoupModel. *Forest ecology and management*, 237, 545-556.

Cienciala, E., Tumajer, J., Zatloukal, V., Beranová, J., Holá, Š., Hůnová, I., & Russ, R. (2017) Recent spruce decline with biotic pathogen infestation as a result of interacting climate, deposition and soil variables. *European Journal of Forest Research*, 136, 307–317.

Comber, S.D.W., Smith, R., Daldorph, P., Gardner, M.J., Constantino, C., Ellor, B. (2013) Development of a chemical source apportionment decision support framework for catchment management. *Environmental Science and Technology*, 47, 9824-9832.

Confor (2016) *PAWS restoration policy review*. Available: https://www.confor.org.uk/media/246406/wsap-recommendations-on-paws-guidance-2016.pdf (Accessed December 2023)

Cools, N., Vesterdal, L., De Vos, B., Vanguelova, E., Hansen, K. (2014) Tree species is the major factor in explaining C:N ratios in the European forest soils. *Forest Ecology and Management*, 311, 3-16.

De Schrijver, A., Geudens, G., Augusto, L., Staelens, J., Mertens, J., Wuyts, K., Gielis, L., & Verheyen, K. (2007) The effect of forest type on throughfall deposition and seepage flux: A review. *Oecologia*, 53, 663–674.

Dedrick, S., Spiecker, H., Orazio, C., Tomé, M., Martinez, I. (2007) *Plantation or conversion – the debate!* Germany: European Forest Institute. Available: https://www.researchgate.net/profile/Heinrich-Spiecker/publication/298017844_Plantation_or_Conversion_-_The_Debate/links/56e53a0b08ae68afa11226c6/Plantation-or-Conversion-The-Debate.pdf (Accessed December 2023)

Defra (2022) *Keepers of time: ancient and native woodland and trees policy in England Government's statement on England's ancient and native woodland and ancient and veteran* *trees*. United Kingdom: UK Government. Available: https://www.gov.uk/search/all (Accessed December 2023)

Dirnböck, T., Kobler, J., Kraus, D., Grote, R., Kiese, R. (2016) Impacts of management and climate change on nitrate leaching in a forested karst area. *Journal of Environmental Management*, 165, 243-252.

Dise, N. B., Rothwell, J. J., Gauci, V., van der Salm, C., & de Vries, W. (2009) Predicting dissolved inorganic nitrogen leaching in European forests using two independent databases. *Science of The Total Environment*, 407, 1798–1808.

Dise, N.B., Wright, R.F. (1995) Nitrogen leaching from European forests in relation to nitrogen deposition. *Forest Ecology and Management*, 71, 153-161.

Duffy, C., O'Donoghue, C., Ryan, M., Kilcline, K., Upton, V., & Spillane, C. (2020) The impact of forestry as a land use on water quality outcomes: An integrated analysis. *Forest Policy and Economics*, 116.

EEA (2005) *Source apportionment of nitrogen and phosphorus inputs into the aquatic environment*. Luxembourg: Office for Official Publications of the European Communities. Available: https://www.eea.europa.eu/publications/eea_report_2005_7/file (Accessed December 2023)

EEA (2020) *Air quality in Europe – 2020 report*. Luxembourg: Office for Official Publications of the European Communities. Available: https://www.eea.europa.eu/publications/air-quality-in-europe-2020-report (Accessed December 2023)

Emmett, B.A., Stevens, P.A., Reynolds, B. (1995) Factors influencing nitrogen saturation in Sitka spruce stands in Wales, UK. *Water, air and soil pollution*, 85, 1629-1634.

Environment Agency (2019) *2021 river basin management plan*. Available: https://consult.environment-agency.gov.uk/++preview++/environment-and-business/challenges-and-choices/user_uploads/nitrates-pressure-rbmp-2021.pdf (accessed 20/01/2021)

Ericsson, T., Göransson, A., Van Oene, H., & Gobran, G. (1995) Interactions between Aluminium, Calcium and Magnesium: Impacts on Nutrition and Growth of Forest Trees. *Ecological Bulletins*, 44, 191-196.

Etzold, S., Ferretti, M., Reinds, G. J., Solberg, S., Gessler, A., Waldner, P., Schaub, M., Simpson, D., Benham, S., Hansen, K., Ingerslev, M., Jonard, M., Karlsson, P. E., Lindroos, A. J., Marchetto, A., Manninger, M., Meesenburg, H., Merilä, P., Nöjd, P., Rautio, P., Sanders, T.G.M., Seidling, W., Skudnik, M., Thimonier, A., Verstraeten, A., Vesterdal, L., Vejpustkova, M., de Vries, W. (2020) Nitrogen deposition is the most important environmental driver of growth of pure, even-aged and managed European forests. Forest Ecology and Management, 458, 117762.

Farrell, R.P., Whiteman, M. (2023) *The Environment Agency Chalk groundwater level monitoring network in England*. In: The Chalk Aquifers of Northern Europe. London: The Geological Society.

Forest Research (2023) *Forestry statistics and forestry facts & figures*. Edingburgh: Forestry Commission. Available: https://www.forestresearch.gov.uk/tools-and-resources/statistics/forestry-statistics/ (accessed 10/02/2021).

Forestry Commission (1985) *Forestry commission 66th annual report and accounts 1985-86*. Edinburgh: Forestry Commission. (Accessed 20/01/2021)

Forestry Commission (1994) *The management of semi-natural woodlands* in Forestry Commission Practice Guides 1-8. Edinburgh: Forestry Commision.

Göttlein, A., Weis, W., Ettl, R., Raspe, S., Rothe, A. (2023) Long-term effects of clearcut and selective cut on seepage chemistry in a N-saturated spruce stand – Case study Höglwald. *Forest Ecology and Management*, 533, 120831.

Göttlein, A., Weis, W., Ettl, R., Raspe, S., Rothe, A. (2023) Long-term effects of clearcut and selective cut on seepage chemistry in a N-saturated spruce stand – Case study Höglwald. Forest ecology and management, 533, 120831.

Guerrieri, R., Lecha, L., Mattana, S., Cáliz, J., Casamayor, E.O., Barceló, A., Michalski, G., Peñuelas, J., Avila, A., Mencuccini, M. (2020) Partitioning between atmospheric deposition and canopy microbial nitrification into throughfall nitrate fluxes in a Mediterranean forest. *Journal of Ecology*, 108, 626-640.

Guerrieri, R., Vanguelova, E. Michalski, G., Heaton, T.H.E., Mencuccini, M. (2015) Isotopic evidence for the occurrence of biological nitrification and nitrogen deposition processing in forest canopies. *Global Change Biology*, 21, 4613-4626.

Gundersen, P., Callesen, I., & De Vries, W. (1998). Nitrate leaching in forest ecosystems is related to forest floor C/N ratios. *Environmental Pollution*, 102, 403-407.

Gundersen, P., Schmidt, I.K., Raulund-Rasmussen, K. (2006) Leaching of nitrate from temperate forests- effects of air pollution and forest management. *Environmental Reviews*, 14, 1-57.

Gundersen, P., Sevel, L., Christiansen, J.R., Vesterdal, L., Hansen, K., Bastrup-Birk, A. (2009) Do indicators of nitrogen retention and leaching differ between coniferous and broadleaved forests in Denmark? *Forest Ecology and Management*, 258, 1137-1146,

Hansen, K., Rosenqvist, L., Vesterdal, L., Gundersen, P. (2007) Nitrate leaching from three afforestation chronosequences on former arable land in Denmark. *Global Change Biology*, 13, 1250-1264

Hansson, K., Olsson, B.A., Olsson, M.m Johansson, U., Kleja, D.B. (2011) Differences in soil properties in adjacent stands of Scots pine, Norway spruce and silver birch in SW Sweden. *Forest Ecology and Management*, 262, 522-530.

Harmer, R., Thompson, R. (2013) *Choosing stand management methods for restoring planted ancient woodland sites*. Edinburgh: Forestry Commission.

Hartwell, S., Morino, K., Nagler, P.L., Glenn, E.P. (2010) On the irrigation requirements of cottonwood (*Populus fremontii* and *Populus deltoides* var.*wislizenii*) and willow (*Salix gooddingii*) grown in a desert environment. *Journal of Arid Environments*, 74, 667-674.

Helliwell, R., Wilson, E. (2012) Continuous cover forestry in Britain. *Quarterly journal of forestry*, 106, 214-224.

Hidy, D., Barcza, Z., Marjanovic, H., Ostrogovic Sever, M. Z., Dobor, L., Gelybó, G., Fodor, N., Pintér, K., Churkina, G., Running, S., Thornton, P., Bellocchi, G., Haszpra, L., Horváth, F., Suyker, A., Nagy, Z., (2016) Terrestrial ecosystem process model Biome-BGCMuSo v4.0: summary of improvements and new modeling possibilities. *Geosciences Model Development*, 9, 4405-4437.

Hobbie, S.E., Reich, P.B., Oleksyn, J., Ogdahl, M., Zytkowiak, R., Hale, C., Karolewski, P. (2006) Tree species effects on decomposition and forest floor dynamics in a common garden. *Ecology*, 87, 2288-2297.

Högberg, P., Johannisson, C., Yarwood, S., Callesen, I., Näsholm, T., Myrold, D.D., Högberg, M.N. (2011) Recovery of ectomycorrhiza after 'nitrogen saturation' of a conifer forest. *New Phytologist*, 189, 515-525.

House of Commons (2018) *UK progress on reducing nitrate pollution*. UK: House of Commons Environmental Audit Committee. Available: https://publications.parliament.uk/pa/cm201719/cmselect/cmenvaud/656/656.pdf (accessed 20/01/2021)

Hůnová, I., Kurfürst, P., Stráník, V., Modlík, M. (2017) Nitrogen deposition to forest ecosystems with focus on its different forms. *Science of the Total Environment*, 575, 791-798.

Ireland, D., Nisbet, T.R., Broadmeadow, M. (2006) *Environmental best practice for continuous-cover forestry*. Bristol: Environment Agency. (Accessed December 2023)

Janssens, I.A., Dieleman, W., Luyssaert, S., Subke, J-A., Reichstein, M., Ceulemans, R., Ciais, P., Dolman, A.J., Grace, J., Matteucci, G.G., Papale, D., Piao, S.L., Schulze, E-D., Tang, J., Law, B.E. (2010) Reduction of forest soil respiration in response to nitrogen deposition. *Nature Geoscience*, 3, 315-322.

Jerabkova, L., Prescott, C.E., Titus, B.D., Hope, G.D., Walters, M.B. (2011) A meta-analysis of the effects of clearcut and variable-retention harvesting on soil nitrogen fluxes. *Canadian Journal of Forest Research*, 41, 1852-1870.

Jonard, M., M., Fürst, A., Verstraeten, A., Thimonier, A., Timmermann, V., Potočić, N., Waldner, P., Benham, S., Hansen, K., Merilä, P., Ponette, Q., de la Cruz, A.C., Roskams, P., Nicolas, M., Croisé, L., Ingerslev, M., Matteucci, G., Decinti, B., Bascietto, M. Rautio, P. (2015) Tree mineral nutrition is declining in Europe. *Global Change Biology*, 21, 418-430.

Kenk, G., Guehne, S. (2001) Management of transformation in central Europe. *Forest Ecology and Management*, 151, 107-119.

Kint, V., Geudens, G., Mohren, G.M.J., Lust, N. (2006) Silvicultural interpretation of natural vegetation dynamics in ageing Scots pine stands for their conversion into mixed broadleaved stands. *Forest Ecology and Management*, 223, 363-370.

Kirby, K.J., Thomas, R.C. (2017) Restoration of broadleaved woodland under the 1985 Broadleaves Policy stimulates ground flora recovery at Shabbington Woods, southern England. *Journal of the Botanical Society of Britain & Ireland*, 7, 125-135.

Komatsu, H., Kume, T., Otsuki, K. (2008) The effect of converting a native broadleaved forest to a coniferous plantation forest on annual water yield: A paired catchment study in northern Japan. *Forest Ecology and Management*, 255, 880-886.

Kostel-Hughes, F., Young, T.P., Wehr, J.D. (2005) Effects of leaf litter depth on the emergence and seedling growth of deciduous forest tree species in relation to seed size. *Torrey Botanical Society*, 132, 50-61.

Kristensen, H.L., Gundersen, P., Callesen, I., Reinds, G.J. (2004) Throughfall nitrogen deposition has different impacts on soil solution nitrate concentration in European coniferous and deciduous forests. *Ecosystems*, 7, 180-192.

Kubin, E. (1998) Leaching of nitrate nitrogen into the groundwater after clear felling and site preparation. *Boreal Environment Research*, 3, 3-8.

Löf, M., Festin, E.S., Szydlo, M., Brunet, J. (2023) Restoring mixed forests through conversion of Norway spruce stands: effects of fencing and mechanical site preparation on performance of planted beech and natural tree regeneration. *European Journal of Forest Research*, 142, 763-772.

Macdonald, J.A., Dise, N.B., Matzner, E., Armbruster, M., Gundersen, P. (2002) Nitrogen input together with ecosystem nitrogen enrichment predict nitrate leaching from European forests. *Global Change Biology*, 8, 1028-1033.

Mannerkoski, H., Finér, L., Piirainen, S., Starr, M. (2005) Effect of clear-cutting and site preparation on the level and quality of groundwater in some headwater catchments in eastern Finland. *Forest Ecology and Management*, 220, 107-117.

Marozas, V., Sasnauskiene, J., Dautartė, A., Gavenauskas, A., Sabienė, N., Armolaitis, K. (2018) Effect of shelterwood cuttings on soil chemical properties in Scots pine (*Pinus sylvestris* L.) forests in Europe’s hemiboreal zone in Lithuania. *Journal of Elementology*, 23, 353-367.

Mason, B. (2020) Encouraging greater use of continuous cover forestry Part 1: Stand and site considerations. *Quarterly Journal of Forestry*, 114, 251-259.

Midgley, M.G., Phillips, R.P. (2014) Mycorrhizal associations of dominant trees influence nitrate leaching responses to N deposition. *Biogeochemistry*, 117, 241-253.

Mupepele, A-C., Dormann, C.F. (2016) Influence of Forest Harvest on Nitrate Concentration in Temperate Streams—A Meta-Analysis. *Forests*, 8, doi:10.3390/f8010005

Nilsson, L. O., Bååth, E., Falkengren-Grerup, U., & Wallander, H. (2007) Growth of ectomycorrhizal mycelia and composition of soil microbial communities in oak forest soils along a nitrogen deposition gradient. *Oecologia*, 153, 375–384. https://doi.org/10.1007/s00442-007-0735-x

Nisbet, T. (2005) *Information note FCIN065: Water use by trees*. Edinburgh: Forestry Commission. Accessible: https://www.forestresearch.gov.uk/research/archive-water-use-by-trees/ (accessed 20/01/2021).

Nisbet, T., Silgram, M., Shah, N., Marrow, K., Broadmeadow, S. (2011) *Woodland for water: woodland measures for meeting water framework directive objectives*. Surrey: Forest Research. Available:
http://citeseerx.ist.psu.edu/viewdoc/download?doi=10.1.1.701.4543&rep=rep1&type=pdf (accessed 20/01/2021)

Olsson, B.A., Bergholm, J., Alavi, G., Persson, T. (2022) Effects of long-term N fertilization on nitrate leaching and vegetation responses in a spruce stand after severe wind damage. *Forest Ecology and Management*, 520, 120422.

Oulehle, F., Hleb, R., Houška, J., Šamonil, P., Hofmeister, J., Hruška, J. (2010) Anthropogenic acidification effects in primeval forests in the Transcarpathian Mts., western Ukraine. *Science of the Total Environment*, 408, 856-864.

Palviainen, M., Finér, L., Laurén, A., Launiainen, S., Piirainen, S., Mattsson, T., & Starr, M. (2014) Nitrogen, phosphorus, carbon, and suspended solids loads from forest clear-cutting and site preparation: Long-term paired catchment studies from eastern Finland. *Ambio*, 43, 218–233. https://doi.org/10.1007/s13280-013-0439-x

Pedley, S. M., Wolstenholme, P., & Dolman, P. M. (2023). Plantation clear-fell patches benefit heathland arthropods. *Ecological Solutions and Evidence*, 4.

Pitman, R., Benham, S., Poole, J. (2013) A chronosequence study of soil nutrient status under oak and Corsican pine with Ellenberg assessed ground vegetation changes. *Forestry*, 87, 287-300.

Pitman, R., Peace, A. (2021) Mulch versus brash: a case study of in situ harvesting residue treatment and its effects on C and nutrients in soil and plant uptake during natural rewilding. *Trees, Forests and People*, 6, 100121.

Pitman, R.M., Vanguelova, E.I., Benham, S.E. (2010) The effects of phytophagous insects on water and soil nutrient concentrations and fluxes through forest stands of the Level II monitoring network in the UK. *Science of the Total Environment*, 409, 169-181.

Rayner, B., Nicoll, B. (2012) *Potential for woodland restoration above the A83 in Glen Croe to reduce the incidence of water erosion and debris flows*. Edinburgh: Forestry Commission.

Reynolds, B. (2004) Continuous cover forestry: possible implications for surface water acidification in the UK uplands. *Hydrology and Earth System Sciences*, 8, 306-313.

Ribbons, R.R., Kepfer-Rojas, S., Kosawang, C., Hansen, O.K., Ambus, P., McDonald, M., Grayston, S.J., Prescott, C.E., Vesterdal, L. (2018) Context-dependent tree species effects on soil nitrogen transformations and related microbial functional genes. *Biochemistry*, 140, 145-160.

Rosenqvist, L. (2006) Afforestation of former arable land in North-Western Europe: Nitrate leaching, carbon sequestration and water recharge. PhD thesis: Swedish University of

Agricultural Sciences. Available: https://www.researchgate.net/publication/30072879_Afforestation_of_former_arable_land_in_north-western_Europe (accessed 20/01/2021).

Rothe, A., Mellert, K.H. (2004) Effects of forest management on nitrate concentrations in seepage water of forests in Southern Bavaria, Germany. *Water, Air and Soil Pollution*, 156, 337-355.

Sardans, J., Alonso, R., Carnicer, J., Fernández-Martínez, M., Vivanco, M.G., Peñuelas, J. (2016) Factors influencing the foliar elemental composition and stoichiometry in forest trees in Spain. *Perspectives in Plant Ecology, Evolution and Systematics*, 18, 52-69.

Schleppi, P., Curtaz, F., & Krause, K. (2017) Nitrate leaching from a sub-alpine coniferous forest subjected to experimentally increased N deposition for 20 years, and effects of tree girdling and felling. *Biogeochemistry*, 134, 319–335. https://doi.org/10.1007/s10533-017-0364-3

Schmitz, A., Sanders, T.G.M., Bolte, A., Bussotti, F., Dirnböck, T., Johnson, J., Peñuelas, J., Pollastrini, M., Prescher, A-K., Sardans, J., Verstraeten, A., de Vries, W. (2019) Responses of forest ecosystems in Europe to decreasing nitrogen deposition. *Environmental Pollution*, 244, 980-994.

Schmitz, A., Sanders, T.G.M., Bolte, A., Bussotti, F., Dirnböck, T., Peñuelas, J., Pollastrini, M., Prescher, A-K., Sardans, J., Verstraeten, A., de Vries, W. (2024) Responses of forest ecosystems in Europe to decreasing nitrogen deposition. In: *Atmospheric nitrogen deposition to global forests*. The United States: Academic Press.

Schrijver, A.D., Geudens, G., Augusto, L., Staelens, J., Mertens, J., Wuyts, K., Gielis, L., Verheyen, K. (2007) The effect of forest type on throughfall deposition and seepage flux: a review. *Oecologia*, 153, 663-674.

Seiwa, K. (2006) Trade-offs between seedling growth and survival in deciduous broadleaved trees in a temperate forest. *Annals of Botany*, 99, 537-544.

Seliger, A., Puffpaff, S., Manthey, M., & Kreyling, J. (2021) Management options for the conversion of allochthonous coniferous forest patches towards more natural species composition in the vorpommersche boddenlandschaft national park, NE Germany. *Baltic Forestry*, 27.

Shah, N. W., Nisbet, T. R., & Broadmeadow, S. B. (2021) The impacts of conifer afforestation and climate on water quality and freshwater ecology in a sensitive peaty catchment: A 25 year study in the upper River Halladale in North Scotland. *Forest Ecology and Management*, 502, 119616.

Silgram, M., Waring, R., Anthony, S., & Webb, J. (2001). Intercomparison of national & IPCC methods for estimating N loss from agricultural land. *Nutrient Cycling in Agroecosystems*, 60, 189-195.

Spracklen, B.D., Lane, J.V., Spracklen, D.V., Williams, N., Kunin, W.E. (2013) Regeneration of native broadleaved species on clearfelled conifer plantations in upland Britain. *Forest Ecology and Management*, 310, 204-212.

Stevens, P.A., Hornung, M. (1990) Effect of harvest intensity and ground flora establishment on inorganic-N leaching from a Sitka spruce plantation in north Wales, UK. *Biogeochemistry*, 10, 53-65.

Stuart, M.E., Wang, L., Ascott, M., Ward, R.S., Lewis, M.A. (2016) *Modelling the groundwater nitrate legacy*. Keyworth: British Geological Survey.

Sundermann, G., Wägner, N., Cullmann, A., von Hirschhausen, C.R., Kemfert, C. (2020) *Nitrate pollution of groundwater long exceeding trigger value: Fertilization practices require more transparency and oversight*. Berlin: Deutsches Institut für Wirtschaftsforschung.

Suz, L.M., Barsoum, N., Benham, S., Dietrich, H.-P., Fetzer, K.D., Fischer, R., García, P., Gehrman, J., Kristöfel, F., Manninger, M., Neagu, S., Nicolas, M., Oldenburger, J., Raspe, S., Sánchez, G., Schröck, H.W., Schubert, A., Verheyen, K., Verstraeten, A. and Bidartondo, M.I. (2014) Environmental drivers of ectomycorrhizal communities in Europe's temperate oak forests. *Molecular ecology*, 23, 5628-5644.

Szwagrzyk, J., Szewczyk, J. (2001) Tree mortality and effects of release from competition in an old-growth Fagus-Abies-Picea stand. *Journal of vegetation science*, 12, 621-626.

Tahovská, K., Choma, M., Kaštovská, E., Oulehle, F., Bárta, J., Šantrůčková, H., Moldan, F. (2020) Positive response of soil microbes to long-term nitrogen input in spruce forest: Results from Gårdsjön whole-catchment N-addition experiment. *Soil Biology and Biochemistry*, 143, 107732

Tew, E. R. (2019*) Forests of the future: ecosystem services in a forest landscape facing significant changes*. PhD thesis: Jesus College, Cambridge.

Tew, E., Coventry, R., Fensom, E., Sorensen, C. (2021). Forest resilience part 2: Practical considerations. *Quarterly Journal of Forestry*, 115, 196-207.

Thompson, R., Humphrey, J., Harmer, R., Ferris, R. (2003) *Restoration of native woodland on ancient woodland sites: Practice guide*. Edinburgh: Forestry Commission. Available: https://www.forestresearch.gov.uk/research/lowland-native-woodlands/restoration-of-lowland-plantations-on-ancient-woodland-sites-paws/#:~:text=The%20aim%20of%20PAWS%20restoration,woodland%20comprising%20site%20native%20species.&text=Conifers%20were%20established%20on%20many,people%20these%20probably%20typify%20PAWS (accessed 20/01/2021)

Thorpe, E. (2011) *Impact of nitrogen deposition on methane- and ammonia-oxidising microbial communities in forest soils*. PhD thesis: University of Essex.

van der Salm, C., de Vries, W., Reinds, G. J., & Dise, N. B. (2007) N leaching across European forests: Derivation and validation of empirical relationships using data from intensive monitoring plots. *Forest Ecology and Management*, 238, 81–91.

Vanguelova, E., Barsoum, N., Benham, S., Broadmeadow, M., Moffat, A. (2007) *Ten years of intensive environmental monitoring in British forests*. Information note FCIN088, UK: Forestry commission.

Vanguelova, E.I., Hirano, Y., Eldhuset, T.D., Sas-Paszt, L., Bakker, M.R., Püttsepp, Ü., Brunner, I., Lõhmus, K., Godbold, D. (2007b) Tree fine root Ca/Al molar ratio – Indicator of Al and acidity stress. *Recent Advances in Woody Root Research*, 141, 460-480.

Vanguelova, E.I., Benham, S., Pitman, R., Moffat, A.J., Broadmeadow, M., Nisbet, T., Durrant, D., Barsoum, N., Wilkinson, M., Bochereau, F., Hutchings, T., Broadmeadow, S., Crow, P., Taylor, P., Houston, T.D. (2010) Chemical fluxes in time through forest ecosystems in the UK – Soil response to pollution recovery. *Environmental Pollution*, 158, 1857-1869.

Vanguelova, E.I., Nortcliff, S., Moffat, A.J., Kennedy, F. (2005) Morphology, biomass and nutrient status of fine roots of Scots pine (Pinus sylvestris) as influenced by seasonal fluctuations in soil moisture and soil. *Plant and Soil*, 270, 233-247.

Vanguelova, E.I., Pitman, R.M. (2019) Nutrient and carbon cycling along nitrogen deposition gradients in broadleaf and conifer forest stands in the east of England. *Forest Ecology and Management*, 447, 180-194.

Vanguelova, E.I., R.P., Benham, S. (2024) *Responses of forest ecosystems to nitrogen deposition in the United Kingdom*. In: Atmospheric nitrogen deposition to global forests. The United States: Academic Press.

Verheyen, K., Lust, N., Carnol, M., Hens, L., Bouma, J.J. (2006) *Feasibility of forest conversion: ecological, social and economic aspects (FEFOCON)*. Brussels: the Belgian Science Policy. Available: https://orbi.uliege.be/bitstream/2268/39490/1/2006_MA04_en.pdf (Accessed December 2023).

Wang, L., Butcher, A.S., Stuart, M.E., Gooddy, D.C., Bloomfield, J.P. (2013) The nitrate time bomb- a numerical way to investigate nitrate storage and lag time in the unsaturated zone. *Environmental Geochemistry and Health*, 35, 667-681.

Wang, L., Stuart, M.E., Lewis, M.A., Ward, R.S., Skirvin, D., Naden, P.S., Collins, A.L., Ascott, M.J. (2016) The changing trend in nitrate concentrations in major aquifers due to historical nitrate loading from agricultural land across England and Wales from 1925 to 2150. *Science of the Total Environment*, 542, 694-705.

Watkins, C. (1986) Recent changes in government policy towards broadleaved woodland. *Area*, 18, 117-122.

Watmough, S. A., Aherne, J., Alewell, C., Arp, P., Bailey, S., Clair, T., Dillon, P., Duchesne, L., Eimers, C., Fernandez, I., Foster, N., Larssen, T., Miller, E., Mitchell, M., & Page, S. (2005). Sulphate, nitrogen and base cation budgets at 21 forested catchments in Canada, the United States and Europe. *Environmental Monitoring and Assessment*, 109, 1–36.

Webster, K.L., Leach, J.A., Hazlett, P.W., Buttle, J.M., Emilson, E.J.S., Creed, I.F. (2022) Long-term stream chemistry response to harvesting in a northern hardwood forest watershed experiencing environmental change. *Forest Ecology and Management*, 519, 120345.

Weis, W., Rotter, V., Göttlein, A. (2006) Water and element fluxes during the regeneration of Norway spruce with European beech: Effects of shelterwood-cut and clear-cut. *Forest Ecology and Management*, 224, 304-317.

Whiteman, M., Farr, G., Haslam, A., Train, C., Kieboom, N., Farrell, R., Davison, P., Williams, H., Tang, S., Hall, J., Jones, L. (2023) *Source apportionment of nitrogen pressures at a Chalk-fed groundwater-dependent wetland*. In: The Chalk Aquifers of Northern Europe. London: The Geological Society.

Withers, P.J.A., Neal, C., Jarvie, H.P., Doody, D.G. (2014) Agriculture and eutrophication: where do we go from here? *Sustainability*, 6, 5853-5875.

Woodland Trust (2011) *The state of the UK's forests, woods and trees*. UK: Woodland trust. Available: https://www.woodlandtrust.org.uk/media/1827/state-of-uk-forests.pdf (accessed 20/01/2021).

Woodland Trust (2011) *The state of the UK's forests, woods and trees*. UK: Woodland trust. Available: https://www.woodlandtrust.org.uk/media/1827/state-of-uk-forests.pdf (Accessed 20/01/2021).

Woodland Trust (2020) *Ancient Woodland Restoration – An introductory guide to the principles of restoration management*. UK: Woodland Trust. Available: https://www.woodlandtrust.org.uk/publications/2020/01/ancient-woodland-restoration-management-principles/ (Accessed December 2023).

Yanai, R.D., Currie, W.S., Goodale, C.L. (2003) Soil carbon dynamics after forest harvest: An ecosystem paradigm reconsidered. *Ecosystems*, 6, 197-212.

Zechmeister-boltenstern, S., Michel, K., Pfeffer, M. (2011) Soil microbial community structure in European forests in relation to forest type and atmospheric nitrogen deposition. *Plant and Soil*, 343, 37-50.

Zerbe, S. (2002). Restoration of natural broad-leaved woodland in Central Europe on sites with coniferous forest plantations. *Forest Ecology and Management*, 167, 27-42.

Zhang, Y., Collins, A. L., Murdoch, N., Lee, D., & Naden, P. S. (2014) Cross sector contributions to river pollution in England and Wales: Updating waterbody scale information to support policy delivery for the Water Framework Directive. *Environmental Science and Policy*, 42, 16–32.

Chapter 2

Former land use, tree species and soil type affect the predictability of nitrate leaching from forests exposed to elevated N deposition

Abstract

Nitrate (NO_3-N) leaching from forest ecosystems is naturally low, but decades of elevated nitrogen deposition from agricultural, industrial and transportation sources in Europe have led to elevated leaching from forests across Europe, threatening surface and groundwater quality. Here, in a systematic review, we collated a European-scale dataset from published literature on factors affecting NO_3-N leaching, including variables not presented in previous analyses of continental-scale databases, such as former and surrounding land use.

A stepwise AIC regression model identified forest type (coniferous or broadleaved), soil type, proximity to agriculture, and throughfall total inorganic nitrogen (TIN) deposition as the most significant predictors of NO_3-N leaching ($R^2 = 0.27$, $p = 0.05$). There were significant associations between former land use categories and soil orders ($X^2=43.59$, $p < 0.001$), and further interrogation of the dataset revealed that NO_3-N leaching was significantly greater in secondary conifer rotations than in heathland/grassland ($p < 0.05$) whilst C:N ratios were greater in heathland/grassland than secondary rotations. Forests in these two scenarios were more likely influenced by the soil nutrient baseline upon planting and external nutrient inputs than in other scenarios. In addition, throughfall TIN deposition and NO_3-N leaching increased with tree age in conifer ecosystems.

The results encourage consideration of optimal rotation periods to limit age-related increases in NO_3-N leaching fluxes and inspire the incorporation of periods of open-field management between forestry rotations to reduce future NO_3-N leaching fluxes. Considering the wide variation in NO_3-N leaching fluxes observed in afforested arable soils, an improved understanding of nutrient cycle interactions is also necessary to predict the potential of afforestation to mitigate NO_3-N leaching fluxes from arable land previously subjected to fertiliser treatments.

1. Introduction

High levels of nitrate (NO_3-N) leaching from soils are a threat to water resources. The EU Water Framework Directive (WFD) states that NO_3-N concentrations in drinking water must be below 11 mg NO_3-N L^{-1} (Wuijts *et al.*, 2022). Elevated NO_3-N leaching fluxes in forest ecosystems, where concentrations of NO_3-N in soil drainage waters have exceeded the WFD threshold, have historically occurred following periods of high nitrogen (N) deposition from surrounding agricultural and industrial activities (Kämäri et al., 1998; Emmett and Reynolds, 1996). Despite continental-scale reductions in NO_x emissions during the past decade, forest ecosystems across Europe carry the legacy of historical deposition. Soil and foliar chemistry show effects of past deposition (Jonard *et al.*, 2015; Schmitz *et al.*, 2019; Gilliam, 2021), especially in areas with intense local NO_x and NH_3 emissions, such as those near industry or livestock sheds and dense road networks (Vanguelova & Pitman, 2019). Prolonged elevated inputs of N leads to the saturation of the forest soil system, a point at which N input is greater than plant and microbial demand and can no longer be assimilated into biomass. Once the N saturation point is exceeded, excess N drains from the soil, mostly NO_3-N (Macdonald *et al.*, 2002). The amount of NO_3-N at risk of leaching from forest ecosystems is reduced through its storage and efficient recycling by soil microbial communities, abiotic immobilisation and vegetation uptake (Gundersen *et al.*, 2006). However, despite a vast evidence base on N deposition effects on forests, the full impact of prolonged periods of elevated deposition on forest processes is not entirely understood. An excess of N availability compared to other essential nutrients leads to an imbalance of tree nutrient status, leading to nutrient deficiency in elements other than N. Significant foliar nutrient deficiencies have been observed in forests across Europe and remain a major concern to tree health (Jonard *et al.*, 2015). High N deposition can limit nutrient uptake, particularly phosphorus (Braun *et al.*, 2010), and mobilise toxic elements such as Al^{3+}, which damages root systems (Gundersen *et al.*, 2006), contributing to excess NO_3-N in soil solution.

Elevated N deposition increases litterfall mass in broadleaf forests but not coniferous forests (Gundersen, 1995). Elevated litterfall fluxes can stimulate the mineralisation of organic matter (Walkiewicz *et al.*, 2021), and in general, broadleaf litterfall decomposition releases more nutrients, such as P, K and Mg than coniferous, creating more nutrient-rich soils (Augusto *et al.*, 2002). Broadleaf forests are typically regenerated on nutrient-rich soils due to their known high nutrient demand, whereas coniferous species are typically planted on acidic, free-drained soils. Broadleaf forests exhibit different C:N ratios in the organic soil and mineral topsoil layers compared to coniferous forests (Cools *et al.*, 2014). Although the exact threshold is disputed, soils with an organic soil C:N ratio of below 25 are considered at greater risk of elevated NO_3-N leaching fluxes (Gundersen *et al.*, 2009). The variation in N cycling processes between coniferous and broadleaved forests may affect soil responses to elevated throughfall total inorganic nitrogen (TIN) inputs (Vanguelova & Pitman, 2019). Based on the Indicators of Forest Ecosystem Functioning (IFEF) dataset, MacDonald *et al.* (2002) reported no effect of tree species on the response to elevated throughfall inputs. Still, continental-scale datasets typically have an underrepresentation of broadleaved species compared to conifers. Analysis of the International Cooperative Programme on Assessment and Monitoring of Air Pollution Effects on Forests (ICP Forests) Level II dataset identified a difference in response between broadleaves and conifers in momentary soil solution NO_3-N concentrations (Kristensen *et al.*, 2004). However, it did not compare fluxes, which could be affected by hydrological differences between the two ecosystem types. Differences in soil solution NO_3-N concentrations were attributed to the tendency of broadleaved trees to grow on nutrient-rich soil types (Kristensen *et al.*, 2004), rather than any exogenous factor. When grown nearby on the same soil type, coniferous forests have displayed greater leaching driven by elevated N deposition, which is explained by large canopy surface areas and better canopy scavenging ability throughout the year (Rothe *et al.*, 2002). Some evidence is available, but interactions between tree species and soil type are poorly understood.

Individual afforestation chronosequence studies show that former land use can also affect the response of forest ecosystems to elevated NH_3/NO_x deposition (van der Salm *et al.*, 2006; Rosenqvist *et al.*, 2010), but systematic comparisons between different types of former land use are needed. Former land use affects soil nutrient availability, organic layer thickness, soil acidity, and carbon and N stocks (Thuille *et al.*, 1999; Ollinger *et al.*, 2002; Thuille & Schulze, 2006). Considering the impact of these factors on the capacity of forests to assimilate N into biomass, former land use is likely to affect the response of forest ecosystems to elevated deposition. With the current interest in afforestation to support nature recovery and sequester and store carbon, to support Government climate change, Net Zero and nature recovery targets (UK Government, 2021; UK Government, 2021b), the knowledge of its relationship to the nitrogen cycle and leaching is sorely needed. The ICP Forests Level II database is dominated by sites where forest was planted on historically forested land (Appendix Figure 1), so the database is unsuitable to test former land use effects on the response of NO_3-N leaching to deposition.

The main aim of this study is to quantify tree species and former land use impacts on NO_3-N leaching in mature European forests, compared to other local scale and climatic factors. We constructed a database from published literature and used it to test the following hypotheses: (H1)forest type (conifer/broadleaf) will have a significant impact on the propensity of the forest to leach NO_3-N as a result of elevated deposition (H2) there will be an interaction between tree species, soil type and former land use on the relationship between throughfall deposition and NO_3-N leaching fluxes, and (H3) this will be associated with differences in organic and mineral topsoil C:N ratios and pH.

2. Methods

2.1. Search strategy and selection criteria

The following databases were searched to identify relevant literature: (1) SCOPUS (Elsevier), (2) Web of Science Core Collection (Clarivate Analytics), (3) Environmental Impact (CABI), (4) Dissertations and Theses Global (Proquest). The search string (Appendix 2) was designed to select literature containing studies on the effect of climatic variables, deposition, soil nutrients and species composition on foliar nutrition, root nutrient uptake and NO_3-N and NH_4-N leaching. Studies conducted on the common European species; *Fagus sylvatica*, *Quercus petraea/robur*, *Betula pendula*, *Picea sitchensis*, *Pinus sylvestris*, and *Picea abies* were selected, representing three conifer and three broadleaved species. Studies conducted on mixed stands, where one of the selected species represented <60 % of the stand, were excluded. The effect of tree species mixes on soil carbon and N stocks, P and K cycling, pH and microbial community structure (Wang *et al.*, 2008; Dawud *et al.*, 2017; Gunina *et al.*, 2016; Rehschuh *et al.*, 2021), could generate different responses of NO_3-N leaching to deposition inputs compared to monocultures and warrants its own full investigation.

The search strings produced 3059 results. Search result titles and abstracts were screened against a predetermined selection criterion with the assistance of Rayyan software, which allows the user to tag accepted and rejected studies and label each with a justification of the decision (Ouzzani *et al.*, 2016). Although many informative controlled field and *ex situ* experimental studies were found (e.g. Ritter & Vesterdal, 2006; Moldan *et al.*, 2018), only observational field studies were included for data extraction to minimise variation caused by other experimental manipulations and condensed timescales. Peer-reviewed studies, student dissertations, and theses were considered. Literature reviews found by the search were screened to identify relevant studies, which were then also included. As the intention was to extract information about individual stand characteristics, studies which analysed large datasets without presenting site-specific data were excluded but were qualitatively compared to the findings presented here. To ensure the dataset developed was an independent test dataset of the data used to establish relationships between throughfall total inorganic nitrogen (TIN) and NO_3-N leaching fluxes previously, studies that used sites within the ICP Forests Level II database were excluded. Of the 3059 results, 2559 were excluded following the screening of titles and abstracts and 500 papers were screened in full. The full paper screening identified 13 papers suitable for data extraction.

The final set of variables with >4 studies was selected for data extraction (Table 1). Soil C:N ratios and soil pH data were extracted as these variables may explain the relationships between the exposure and outcomes. Soil C:N ratio and pH were not considered an exposure or outcome to avoid over-parameterising the relationships between NO_3-N leaching and the other exposures considered. Studies that exclusively reported deep soil solution concentrations without calculating leaching fluxes were excluded as concentration-only data fails to capture the complexity related to hydrological factors.

Table 1: Variables extracted from the literature on NO_3-N leaching in European forests. The n_p indicates the number of included studies that presented information on each variable and the n_s indicates the number of sites with available data, as studies typically presented data from multiple sites.

Exposures	**Outcome**	**Variables to explain exposure/outcome relationships**
Tree species (n_p = 13, n_s = 51) Soil order (n_p = 12, n_s = 43) Land use history (n_p = 13, n_s = 50) Proximity to agriculture (n_p = 0, n_s = 51*) Mean annual temperature (n_p = 13, n_s = 51) Mean annual precipitation (n_p = 12, n_s = 49)	NO_3-N leaching (n_p = 13, n_s = 51) Relationship between NO_3-N leaching and throughfall TIN concentrations (n_p = 13, n_s = 51)	Organic layer C:N ratio (n_p = 4, n_s = 26) Mineral topsoil (0-10cm) C:N ratio (n_p = 5, n_s = 35) Organic layer pH (n_p = 4, n_s = 18) Mineral topsoil (0-10cm) pH (n_p = 9, n_s = 37) Soil texture (n_p = 3, n_s = 10) Stand age (n_p = 11, n_s = 45)

* Proximity to agriculture was calculated for all sites from coordinates and land cover maps.

2.2. Data extraction

Information on authors, citations, journal, publication year, study species, coordinates, study location name and country were recorded for all publications. Where coordinates were not presented, these were estimated from Google Earth based on the study location name, maps presented in the publication and landcover data.

Throughfall TIN and NO_3-N leaching fluxes were extracted for all individual stands studied across the literature base, along with available information on stand characteristics, former land use, soil physical and chemical properties, and climatic variables. Where individual data points were not shown in written form, data was extracted from figures using WebPlotDigitiser 4.2 (Rohatgi, 2018).

2.3. Variables

2.3.1. *Throughfall TIN and NO_3-N leaching*

When collating wet deposition data, throughfall TIN fluxes rather than bulk deposition were used as these were more frequently reported, and this eliminates the complexities caused by variation between species in canopy processes in determining TIN inputs to the forest floor. Throughfall TIN fluxes were extracted. Where throughfall NH_4-N and NO_3-N concentrations were reported separately, these were added to calculate TIN. Mean annual throughfall TIN concentrations and NO_3-N leaching fluxes were typically recorded in kg ha^{-1} yr^{-1}; we converted other units where this standard was not presented.

2.3.2. *Stand characteristics and proximity to agriculture estimation*

Study year and tree planting year information were used to calculate tree age, where not explicitly stated at the time of study. Other characteristics, such as stand density data, were not consistently reported and, therefore, were not considered. Surrounding land use was rarely reported, and proximity to agricultural land was calculated using stand coordinates and the CORINE European Landcover Maps, aligning the monitoring period of each study with the nearest available land cover representation (Copernicus Land Monitoring Service, 1990; Copernicus Land Monitoring Service, 2000; Copernicus Land Monitoring Service, 2006). Agricultural land types used from the European Land Cover Maps were “121: Industrial land”, which included intensive livestock houses, “231: pasture land” and “211: non-irrigated arable land” as other agricultural land types were not expected to have significant effects on NO_3-N leaching. Studies were grouped into three groups: 1990-2000, 2000-2006 and 2006-2010. The 1990, 2000 and 2006 land cover maps were then used to calculate the distance between each study stand and the closest parcel of agricultural land using the NNJoin tool (Tveite, 2016) in QGIS.

2.3.3. Soil characteristics

Inconsistencies in the definition of “organic layer” were reported between studies. Where studies stated “C:N ratio/pH of the organic layer” this was extracted. The humus (OH) layer was selected where studies individually represented the fermentation (OF) and OH layers. Data was also extracted for what studies referred to as the A horizon or the top 0-10 cm of soil below the organic horizon and referred to as “mineral topsoil” throughout this paper. There was insufficient data for the analysis of deeper mineral soils.

2.3.4. Former land use categorisation

A summary of the former land use information presented in each paper was recorded, and key management practices were identified. Former land use categories were then developed to encompass the management practices used across all sites. Five categories were determined: (1) Coppice- stands consisting of woodland that was coppiced at some point in its history but not during the study period, (2) Arable- land formerly used for crops, (3) Heathland or grassland- land formerly managed to restrict tree growth, including with or without the use of grazing (4) Old growth forest- land formerly planted with mature, native forest, felled and replanted with current species, (5) Rotation- second or later rotation of the current species. Few studies provided more detailed descriptions of former land use, and information on differences in, e.g. nutrient management, brash removal, etc, between sites within each category could not be obtained but may contribute to variation in N-NO_3 leaching within categories.

2.3.5. Climatic variables- precipitation and temperature

Mean annual temperature (MAT) and mean annual precipitation (MAP) were recorded when presented. For 2 sites where mean annual precipitation was not presented, data for the coordinates of the sites provided were used to extract climate data from the ERA5-Land monthly average dataset (Muñoz, 2019). The mean daily precipitation per month 1994-2014 was multiplied by the number of days per month, summed for each year, and the mean annual precipitation calculated in mm.

2.3.6. Categories within exposure groups

The broad tree types considered were broadleaved and coniferous. Soil orders considered were cambisols, podzols, inceptisols and alfisols. Quantitative variables were grouped into categories to compare the effect of categorical variables with the effect of quantitative variables. For MAT, sites were subset based on whether they were situated in locations with a MAT of 2-6, 6-9 or 9-12 ^{o}C. For MAP, sites were subset based on whether they were situated in locations that experienced 500-1000, 1000-1500 or

1500-2000 mm MAP. For proximity to agricultural land, sites were subset as 0-250, 250-500, 500-750, 750-1000, >1000 m away from the nearest parcel of agricultural land.

2.3.7. Statistical analysis

The Akaike Information Criterion was applied in a stepwise algorithm to identify a linear regression model with the best combination of exposures as predictors for the whole dataset. The predictors assessed in the stepwise regression were throughfall TIN, tree type, soil order, former land use, and the proximity to agricultural land, MAT and MAP subsets. As the residuals of the output model were not normally distributed, the procedure was repeated with the dependent variable log-transformed. Regression equations were extracted from published analyses of the ICP Forests Level II and IFEF European datasets for comparison.

The data was subset by categories within each exposure to further explore the dataset and the correlation (Pearson's r) between throughfall TIN and log(NO_3-N leaching) was calculated. When correlations were weak ($r < 0.3$, Freedman *et al.*, 1978), the linear model was not applied. In comparisons with $r > 0.3$, resultant linear regression coefficients were compared with analysis of covariance (ANCOVA). Regression diagnostic plots were used to identify whether each model met the assumptions of linear regression. Where assumptions were not met, the dependent variable was log-transformed. To assess the presence of unequal variances between categories, a Bartlett's Test was used, which calculates a test statistic from the logarithm of sample variances and compares to a chi-square distribution. Where variances between categories within exposures were unequal the regression coefficients were compared with a non-parametric ANCOVA using the sm.ancova function in the sm package in R.

Pairwise comparisons of estimated marginal means were conducted on linear models using the emmeans package in R. A chi-squared test of independence was used to identify interactions between exposures, e.g., whether particular soil orders were associated with particular former land uses. Analysis of variance (ANOVA) was used to test for significant differences in pH and C:N ratios between categorical independent variables, using a Tukey's Honestly Significant Differences test to investigate pairwise comparisons. A Kruskal-Wallis test was used where data did not meet ANOVA assumptions, with a Dunn's test to make pairwise comparisons.

3. Results

3.1. Comparison to other datasets

The literature screening process identified 13 papers that provided annual leaching flux and throughfall TIN data at 51 individual sites (Appendix 3), mostly situated in mainland central Europe (Appendix 4). There was a positive but insignificant relationship between NO_3-N leaching (log-transformed) and throughfall TIN across the sites (Figure 1a, $y = 0.03x+0.83$, $p > 0.05$, Adjusted $R^2 = 0.02$). The stepwise AIC algorithm selected "throughfall TIN", "soil type", "forest type" and "proximity to agriculture" as the best combination of predictors for log(leaching), but the model only explained 26.8 % of the overall variation (Table 2). Most analyses of the Level II and IFFE databases found throughfall TIN as the best predictor of NO_3-N leaching, with the models explaining a greater proportion of the variation in NO_3-N leaching than this present study (Table 3). Application of the linear regression equation previously developed from the IFEF and ICP Forests datasets reported by Dise *et al*. (2009) to predict N leaching from throughfall TIN, gave a weak correlation between modelled and observed values in broadleaved (r

= 0.03 for both models) and coniferous (r = 0.19 for both models) forest (Figure 1b and c). In general, modelled values underestimated NO_3-N leaching fluxes from broadleaf and coniferous sites located further away from agricultural land and overestimated NO_3-N leaching fluxes from conifer sites located closer to agricultural land.

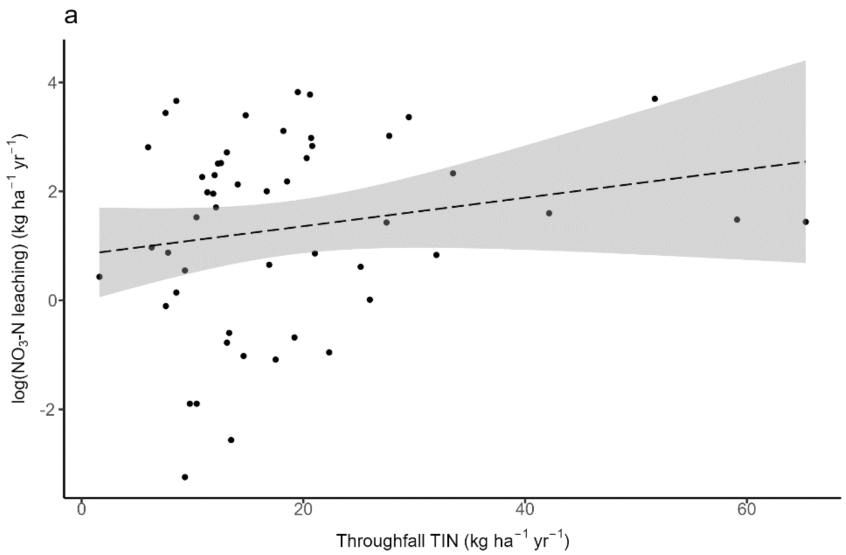
Table 2: The outputs of the linear regression model selected during the stepwise AIC algorithm as the model that best predicted log(NO_3-N leaching). n represents the number of sites in each category.

	n	Estimate	Standard Error	t value	p-value
(Intercept)		0.10053	0.83756	0.120	0.9056
Throughfall TIN	51	0.04808	0.02969	1.619	0.1197
Forest type: Conifer	24	-1.58400	0.72542	-2.184	0.0399
Soil order: cambisol	9	3.16990	1.67351	1.894	0.0714
Soil order: Inceptisol	7	2.87013	1.39848	2.052	0.0522
Soil order: podzol	11	0.69868	0.75749	0.922	0.3663
Proximity to agriculture: 250-500 m	8	-1.91467	1.32860	-1.441	0.1636
Proximity to agriculture: 500-750 m	15	0.16144	0.75508	0.214	0.8327
Proximity to agriculture: 750-1000 m	4	3.18300	1.13721	2.799	0.0105

Table 3: Predictors of NO_3-N leaching fluxes in European forests across three databases.

Dataset	Transformation of dependent variable	Most significant predictors of N-NO_3 leaching	Units for N-fluxes	n	Adjusted R^2	p-value	Reference
Present dataset	log	Throughfall TIN Forest Type Soil Order Proximity to agricultural land	kg ha^{-1} yr^{-1}	42	0.27	0.05	
Evaluation of N and Sulfur		Throughfall TIN	kg ha^{-1} yr^{-1}	65	0.69	<0.001	Dise, & Wright (1995)

Fluxes (ENSF)							
IFEF		Throughfall TIN	kg ha^{-1} yr^{-1}	181	0.62	<0.05	Macdonald *et al* (2002)
IFEF		Throughfall TIN	kg ha^{-1} yr^{-1}	194	0.65	<0.0001	Dise *et al*. (2009)
Level II		Throughfall TIN	kg ha^{-1} yr^{-1}	57	0.39	<0.0001	Dise *et al*. (2009)
Level II		Throughfall TIN Soil C:N ratio MAT	mol ha^{-1} yr^{-1}	55	0.44		Van der Salm *et al* (2007)
Level II		Throughfall TIN	mol ha^{-1} yr^{-1}	57	0.30		Van der Salm *et al* (2007)



b. Using IFEF model

c. Using ICP Forests model

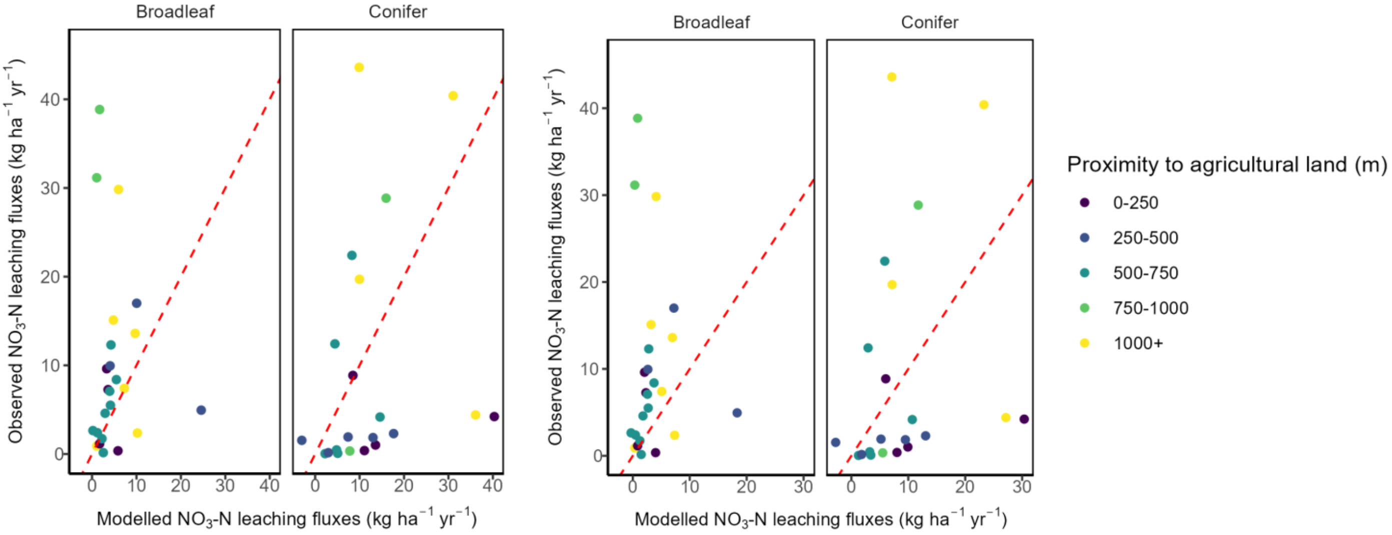


Figure 1: (a) The relationship between the log of NO_3-N leaching and throughfall TIN (both in kg ha^{-1} yr^{-1}) in a dataset of 51 sites across European coniferous and broadleaved forests collated from the literature ($y = 0.03x+0.83$, $p > 0.05$, Adjusted $R^2 = 0.02$), (b) Observed vs modelled values of NO_3-N leaching for the sites in the present literature database, predicted from throughfall TIN using linear models from the IFEF and (c) ICP

Forests datasets (Dise *et al.*, 2009) for coniferous and broadleaf forest. The red dashes represent 1:1 line. The correlation between modelled and observed values was low for both coniferous and broadleaf forest using the IFEF model (r = 0.19 and 0.03 respectively) and ICP Forests model (r = 0.19 and 0.03 respectively).

3.2. Tree Type

Tree type did not significantly affect the strength of NO_3-N leaching response to throughfall TIN inputs (Figure 2, ANCOVA, $F = 0.5$, $p > 0.05$). Dise *et al*. (1995) previously identified three thresholds; little or no leaching below deposition inputs of 10 kg ha^{-1} yr^{-1}, wide variation in response to deposition between inputs of 10-25 kg ha^{-1} yr^{-1}, and significant leaching where deposition is above 25 kg ha^{-1} yr^{-1}. Similar trends were observed in the present dataset (Figure 1a), but several broadleaved sites exhibited elevated levels of leaching (>10 kg ha^{-1} yr^{-1}) below the throughfall TIN inputs = 10 kg ha^{-1} yr^{-1} threshold. Only one coniferous site displayed elevated levels of leaching below this threshold. Where throughfall TIN was above 25 kg ha^{-1} yr^{-1}, one broadleaved site and two coniferous sites did not display elevated levels of NO_3-N leaching. There were no significant differences in the mineral topsoil C:N ratio (Kruskal Wallis, $X^2 = 3.37$, $p > 0.05$), or in the pH (ANOVA, $F=0.57$, $p > 0.05$), between broadleaved and coniferous sites. There were no significant differences in the C:N ratio of the organic layer (Kruskal Wallis, $X^2 = 0.01$, $p > 0.05$) or organic layer pH (ANOVA, $F = 0.12$, $p > 0.05$).

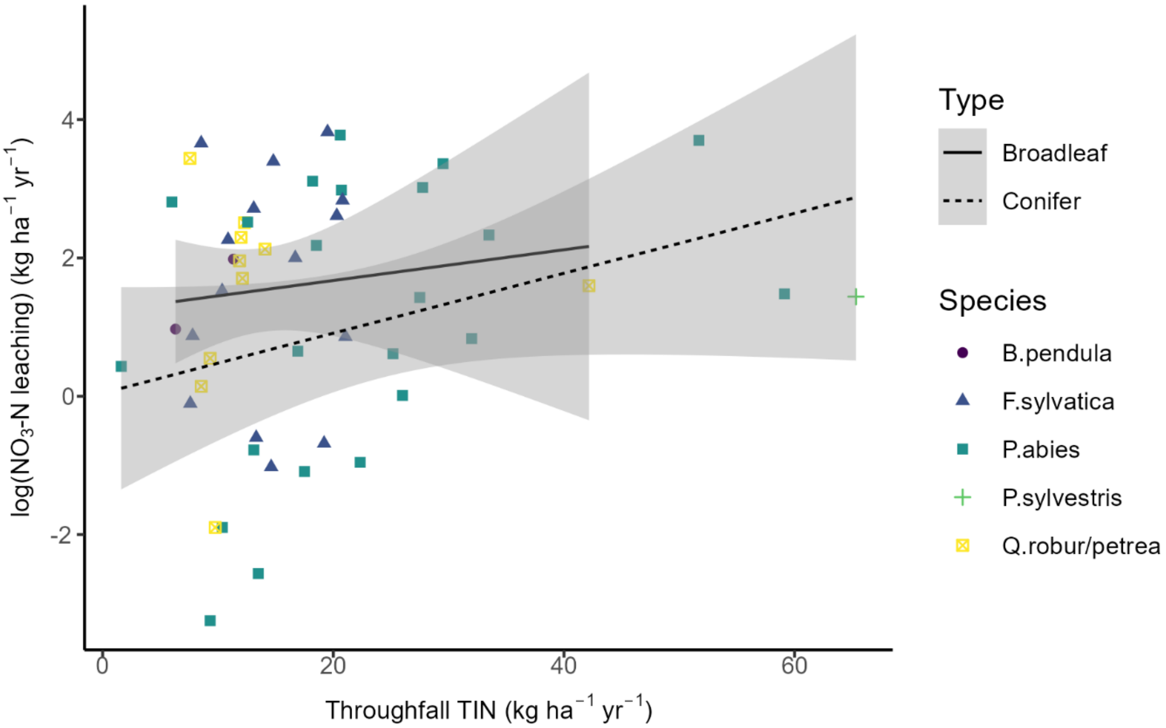


Figure 2: The relationship between throughfall TIN and NO_3-N leaching in broadleaved (n = 27) vs. coniferous sites (n = 24) in a database collated from published literature on European forest ecosystems. The shaded area shows +/- standard error. The response of NO_3-N leaching to throughfall TIN was not significantly affected by tree species ($p > 0.05$).

3.3. Former land use legacy effects and age effects.

Of the 51 individual sites in the dataset, former land use information was available for 50. Forests planted on former arable ($Broadleaf_n = 9$, $conifer_n = 9$) were equally represented by both conifer and broadleaved species. Forests planted on coppice ($Broadleaf_n = 3$, $conifer_n = 0$) and old growth forest ($Broadleaf_n = 10$, $conifer_n = 5$) were more commonly broadleaved species whereas forests planted on former rotations ($Broadleaf_n = 0$, $conifer_n = 4$) and heathland/grassland ($Broadleaf_n = 4$, $conifer_n = 6$) were more commonly coniferous species. There were no significant differences in mean total annual NO_3-N leaching fluxes between broadleaved forests planted on sites with different former land uses (Kruskal-Wallis $X^2 = 3.75$, $p > 0.05$) but the difference between conifers planted on sites with different former land uses was significant (Figure 3a, Kruskal-Wallis $X^2 = 9.3$, $p < 0.05$). Pairwise comparisons found NO_3-N leaching fluxes were significantly lower in arable land afforested with conifers compared to conifer rotations ($p < 0.05$), but no other pairs were significant ($p > 0.05$). There was a significant difference in mineral soil C:N ratios (Figure 3b, ANOVA, $F = 4.13$, $p < 0.01$) but not organic layer C:N ratios (ANOVA, $F = 1.64$, $p > 0.05$) between sites with different former land use. Pairwise comparisons identified significant differences in mineral soil C:N ratios between afforested heathland/grassland and arable land ($p < 0.005$), and heathland/grassland and old growth forest ($p < 0.05$).

When the data was subset by forest type and former land use, some categories did not display a moderate or strong correlation ($r > 0.3$) between NO_3-N leaching and throughfall TIN (Figure 4a). The correlation between NO_3-N leaching and throughfall TIN was not significant ($p < 0.05$) in any subset, likely due to small sample sizes. Due to the absence of a strong correlation in other subsets, linear regression could only be applied to compare the response of NO_3-N leaching to throughfall TIN in broadleaves planted on former heathland/grassland, conifers planted on arable land, conifers planted on former heathland/grassland, and secondary conifer rotations. There was no significant difference in the response of NO_3-N leaching to throughfall TIN between conifers and broadleaves planted on former heathland/grassland (ANCOVA, $F = 0.46$, $p > 0.05$). There was an overall significant difference in the response of NO_3-N leaching to throughfall TIN between conifers planted on rotations, heathland/grassland and arable land (Figure 4b, ANCOVA, $F= 9.58$, $p < 0.001$). Post-hoc pairwise comparisons identified no significant difference between afforested heathland/grassland and arable land (estimate = 7.57, $t = 1.78$, $p > 0.05$) and afforested arable land and rotations ($t = -2.22$, $p > 0.05$), but there was a significant difference between heathland/grassland and rotations ($t = -3.86$, $p < 0.05$).

NO_3-N leaching increased with increasing tree age until 80 years old for conifers (Figure 5a). Throughfall TIN followed a similar pattern with tree age for conifers (Figure 5b). Conifers were, however, not represented after 100 years, unlike broadleaves. Broadleaves did not show obvious age effects, but N leaching was typically low in forests older than 100. One prominent outlier, a 200-year-old broadleaf site planted on a former old-growth forest site with a NO_3-N leaching flux of 45.7 kg ha^{-1} yr^{-1}, was studied by Gundersen *et al*. (2009). The authors considered this location to be a transient phenomenon, located in a site with little forestry management, hypothesising that the unusually high leaching resulted from 50 years of elevated deposition, a period of higher-than-average MAT and MAP, and the multi-structured arrangement of the stand fostering a diversity of microclimates.

Separating land-use legacy effects from tree age effects is challenging as different former land uses were associated with various age groups. Former arable sites were planted with trees mostly less than 50 years of age; former rotations were planted with trees between 50-100 years old; former heathland/grasslands were planted with trees 0-100 years old. Former coppice and old growth forests were planted with trees that were mostly over 100 years old.

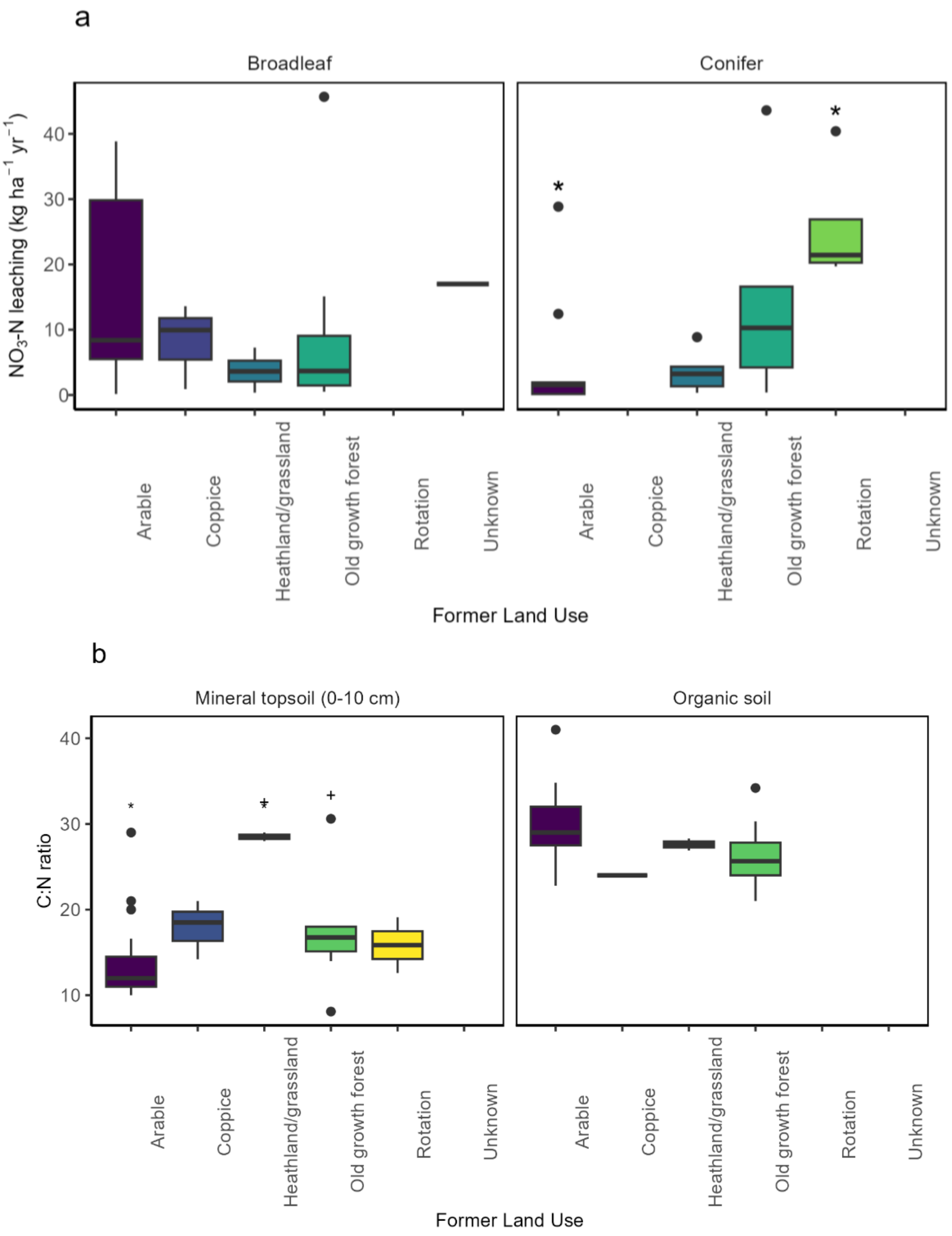
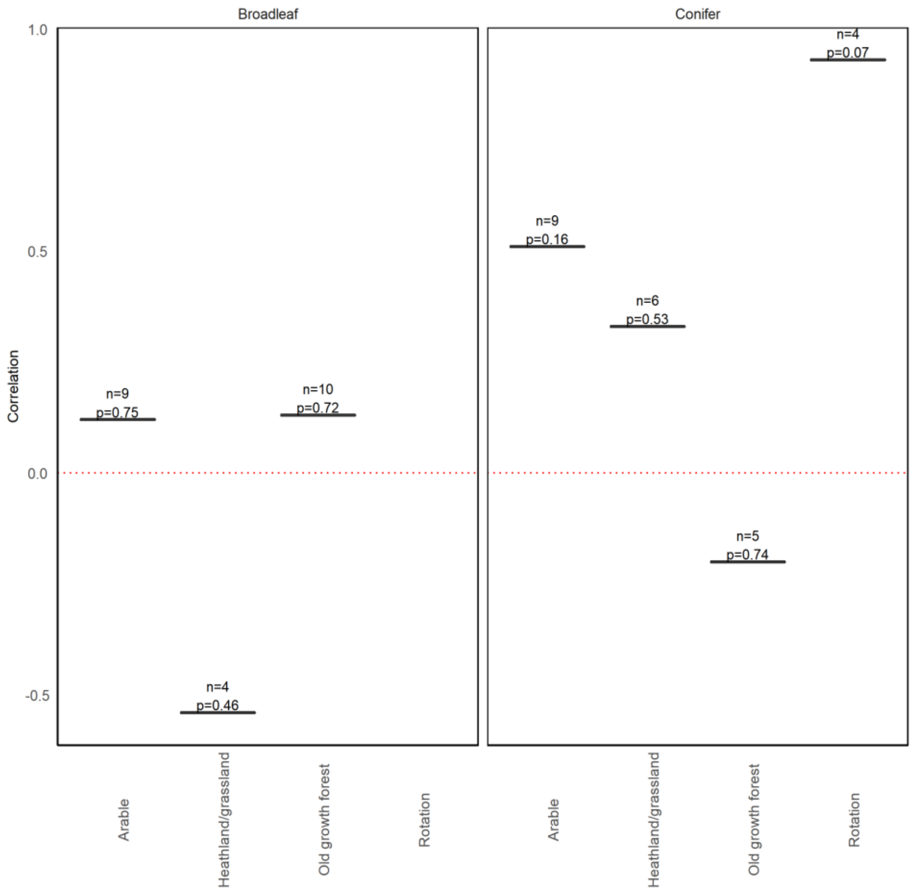


Figure 3: Differences in (a) NO$_3$-N leaching in broadleaf and coniferous forest and (b) organic and mineral topsoil (0-10 cm) C:N ratios of forested sites associated with different land use histories, from a database of 51 sites collated from published literature on European forest ecosystems. The * and + symbols note groups that are statistically significantly to each other different at the $p < 0.05$ threshold.



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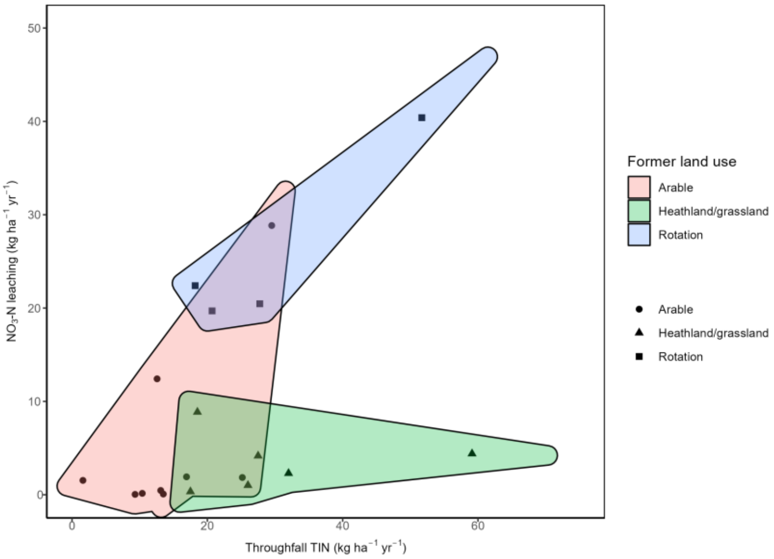


b. Conifers

Figure 4: In a dataset collated from published studies on European forests, (a) the correlation between average annual throughfall TIN and NO_3^--N leaching fluxes in data

subset by forest type and former land use, (b) the relationship between NO_3-N leaching and throughfall TIN on coniferous forest sites only with different land use histories (arable n = 9, heathland/grassland n = 6, rotation n = 4). Polygons highlight the range in throughfall TIN inputs and N-NO_3 leaching in each former land use category.

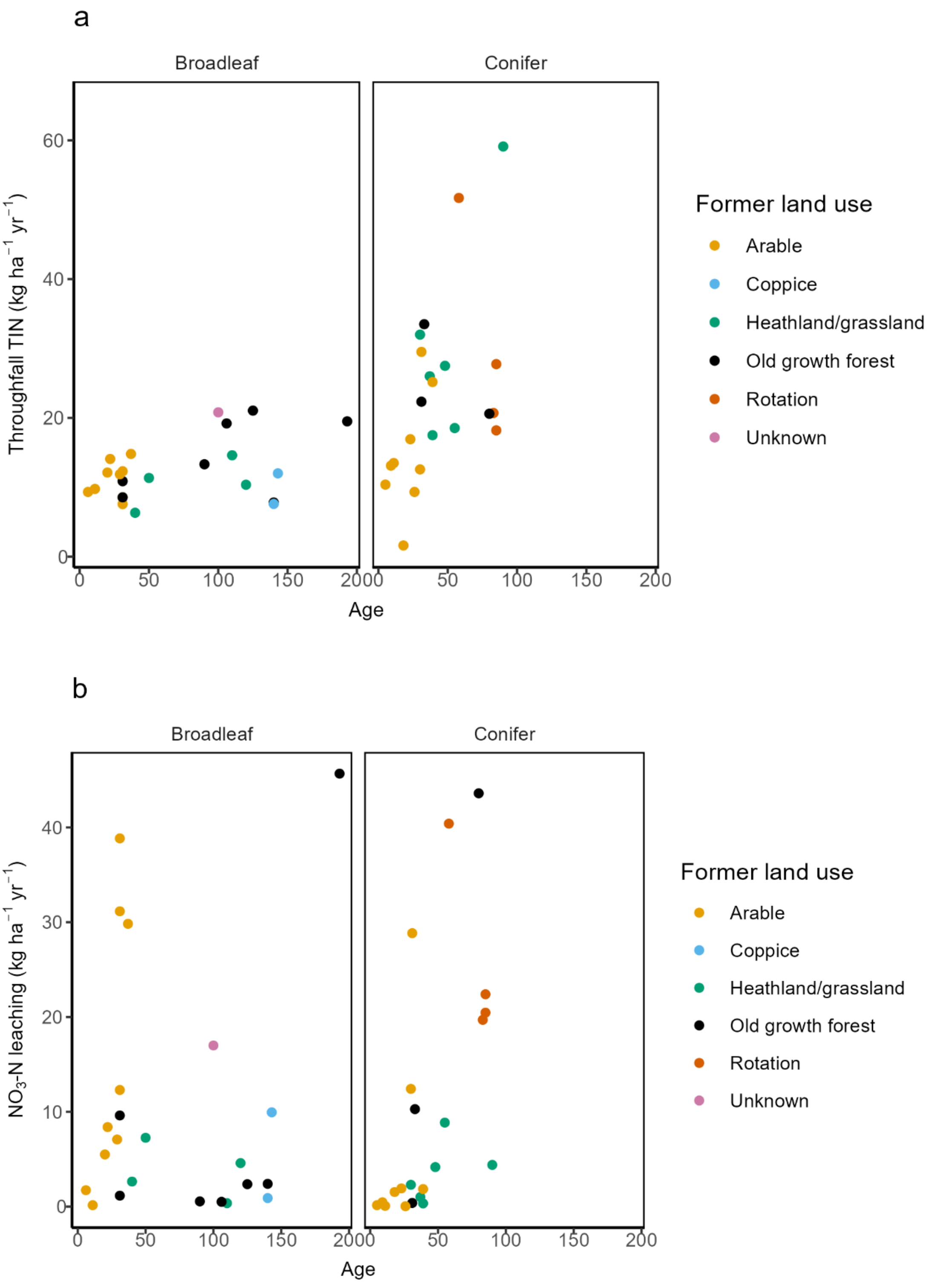


Figure 5: Variation of (a) throughfall TIN and (b) NO_3-N leaching with tree age in broadleaf and coniferous forests with varied land use histories, in a database of 51 European forest sites collated from the literature. Land use histories represented were

former arable land ($\text{Broadleaf}_n = 9$, $\text{conifer}_n = 9$), coppice ($\text{Broadleaf}_n = 3$, $\text{conifer}_n = 0$), old growth forest ($\text{Broadleaf}_n = 10$, $\text{conifer}_n = 5$), rotations ($\text{Broadleaf}_n = 0$, $\text{conifer}_n = 4$) and heathland/grassland ($\text{Broadleaf}_n = 4$, $\text{conifer}_n = 6$).

3.4. Soil characteristics

Twelve soil orders were represented in the dataset. Of these 12, there were four orders with sufficient replicates ($n \leq 5$) to fit linear models to. The magnitude of the response to throughfall TIN, from strongest to weakest, was: inceptisols (n=7) > cambisols (n=9) > podzols (n=11). Alfisols (n=15) displayed a very weak negative relationship between N-input and output and a large degree of variation. Of the four soil types with sufficient representation, only inceptisols displayed a strong correlation between NO_3-N leaching and throughfall TIN ($r = 0.65$, $p > 0.05$), with correlations on podzols ($r = 0.12$), cambisols ($r = -0.08$) and alfisols (0.11) all being very weak.

There was no significant difference in leaching fluxes between the four soil orders (ANOVA, $F = 0.80$, $p > 0.05$). There was a significant association between soil orders and different former land uses (Chi-squared, $X^2=43.59$, $p < 0.001$). Podzols were associated with afforested heathland/grassland (n = 6) and old-growth forest (n = 4), cambisols were associated with old-growth forest (n = 4), and alfisols and inceptisols were associated with former arable land (n = 12 and 4 respectively).

There was a significant difference in the C:N ratio of the mineral topsoil (Appendix 5, ANOVA, $F= 4.41$, $p < 0.05$) and mineral soil pH (Kruskal Wallis, $X^2 = 8.42$, $p < 0.05$) between the soil orders. Organic layer pH (Kruskal Wallis, $X^2 = 4.69$, $p > 0.05$) and C:N ratios ($F = 1.24$, $p > 0.05$) did not significantly differ between soil orders. There were no significant differences in the % of sand (Kruskal Wallis, $X^2 = 7.40$, $p = 0.06$) and clay (Kruskal Wallis, $X^2 = 4.64$, $p > 0.05$) between the four soil orders. Leaching and throughfall TIN fluxes were not significantly related to the C:N ratio of the organic soil layer ($p > 0.05$). Throughfall TIN fluxes were significantly related to the C:N ratio of the mineral topsoils ($F = 6.85$, $p < 0.01$). A strong significant interaction between throughfall TIN, former land use and soil order was found in predicting mineral soil C:N ratios (ANCOVA, $F = 7.93$, $p < 0.001$).

3.5. Climatic factors

There was no significant interaction between MAT ($F = 1.84$, $p > 0.05$) or MAP ($F = 2.59$, $p > 0.05$) with throughfall TIN as a predictor of NO_3-N leaching. MAP was a significant predictor of NO_3-N leaching when paired with throughfall TIN ($F = 3.93$, $p < 0.05$), but MAT was not ($F = 2.82$, $p > 0.05$). Throughfall TIN concentrations did not exceed 21.60 kg ha^{-1} yr^{-1} in areas receiving a MAP of 1500-2000 mm.

3.6. Surrounding land use

The interaction of proximity to agricultural land on the NO_3-N leaching response to N-input was insignificant (ANCOVA, $F=1.1$, $p > 0.05$). Whilst there was no significant difference in C:N ratios in the organic layer between sites at different proximities to agricultural land (Kruskal Wallis, $X^2 = 2.46$, $p > 0.05$), the C:N ratios of mineral topsoils were significantly different (Kruskal Wallis, $X^2 = 22.48$, $p < 0.001$). pH was not significantly different in the organic layer (ANOVA, $F = 2.6$, $p > 0.05$) or in the mineral topsoils (ANOVA, $F = 0.99$, $p > 0.05$) between sites at different proximities to agricultural land. Sites situated 500-750 m from agricultural land had low mineral topsoil C:N ratios and a higher organic layer pH, with the difference between organic and mineral topsoil

C:N ratios being greater than observed in soils situated closer or further from agricultural land.

4. Discussion

4.1. Comparison to other datasets

We collated a literature database to improve the understanding of the relationship between NO_3-N leaching and several factors, including throughfall TIN inputs. The studies identified from the literature represented diverse forests where NO_3-N leaching and throughfall chemistry have been investigated, including sites from afforestation chronosequence studies and other independent projects.

Previous studies that utilised the ICP Forests level II and the IFEF databases displayed a stronger relationship between throughfall TIN inputs and NO_3-N leaching, with throughfall TIN able to explain more of the variation in NO_3-N leaching fluxes than in the present dataset (Table 3). Subsetting the dataset collated here by tree type, former land use and soil type revealed the lack of correlation between throughfall TIN and NO_3-N leaching in some groups, suggesting that throughfall TIN is not a main driver of NO_3-N leaching in some forests, particularly in broadleaved forests.

Four potential explanations of the low explanatory power of the models tested in this study can be suggested: (1) the diverse response of NO_3-N leaching in broadleaved forests to throughfall TIN inputs, (2) the representation of forests planted on a wider range of former land uses and therefore different nutrient baselines at planting, (3) the representation of younger forests (< 30 years old) where the response to wet deposition inputs is affected by the nutrient demands of younger trees, (4) the inconsistency of protocols in studies conducted outside large monitoring networks leading to greater variation in water fluxes and soil solution NO_3-N concentrations.

4.2. The absence of tree species effects on NO_3-N leaching and mineral topsoil C:N ratios

The present dataset benefits from a fairly even representation of broadleaf and conifer species. There was an unexpected absence of species interaction in the response of NO_3-N leaching to throughfall TIN. A stronger response of soil solution NO_3-N concentrations and NO_3-N leaching fluxes to elevated deposition has previously been observed in broadleaf compared to coniferous forests (Kristensen *et al*., 2004; Gundersen *et al*., 2009). Explanations of species interactions on the response of NO_3-N leaching to throughfall TIN include the different effects of deposition on litterfall fluxes in broadleaved species. Elevated throughfall TIN has previously been associated with elevated litterfall fluxes (Gundersen 1995) and reduced litterfall C:N ratios (Bergkvist & Folkeson, 1991) in broadleaved forests. Higher levels of leaf litter then stimulate organic matter mineralisation (Walkiewicz *et al*., 2021) and nitrification (Jussy *et al*., 2004). Broadleaved sites planted on former arable land showed a particularly weak correlation between throughfall TIN inputs and NO_3-N leaching and a large variation in NO_3-N leaching fluxes compared to other former land uses. We hypothesise that the previous nutrient management regime of arable lands affected the capacity of afforested soils to retain TIN inputs.

Elevated N deposition may initially have a fertilising effect on forests, promoting tree growth. When elevated deposition persists, available phosphorus eventually becomes limiting, leading to foliar P deficiencies, further exacerbated by damage to mycorrhizal mycelial growth (Sardans et al., 2016; Braun et al, 2010; Braun et al., 2017; Tahovská et al., 2020). Reduced P uptake has also been associated with reduced N uptake capacity (Braun et al., 2020), further increasing potential soil NO_3-N leaching. Due to modern agriculture practices during the 20th century, agricultural soils have been observed to have greater P content than forest soils in Denmark (Rubaek *et al*., 2013). Afforested arable lands, particularly cropland, also had a greater P content than old-growth forests in France (Koerner *et al*., 1997). Therefore, it is a reasonable prediction that the variable baseline nutrient status of afforested arable lands in this dataset affected the forest's capacity to assimilate N into biomass and retain N within the system. This hypothesis is supported by the fact that most afforested arable sites included in this study were on alfisols, a soil type richer in nutrients than podzols, cambisols and inceptisols. The effect of nutrient availability on NO_3-N leaching has been evidenced by a relationship between foliar N:P ratios with elevated NO_3-N leaching to surface water in Czech forests (Oulehle *et al*., 2021) and the reduction of deep soil solution NO_3-N concentrations from 3-5 mg-N l^- to 2-3 mg-N l^- following the application of P and K fertilisers to a Sitka spruce plantation in Wales (Stevens *et al*., 1993). Considering the implications of this observation in species selection for arable land afforestation, further investigation is encouraged to understand relationships between NO_3-N leaching fluxes and other soil nutrients in larger datasets.

Whilst information on NO_3-N leaching fluxes before afforestation was not available for the included study sites, so it is not possible to establish the effect of afforestation on mitigating NO_3-N leaching fluxes from arable land, it is worth noting that the highest observed NO_3-N leaching fluxes from afforested arable sites still remained below 30 kg-

N ha^{-1} yr^{-1}, below the typical NO_3-N leaching fluxes observed in arable lands in Europe. For example, Silgram *et al*. (2001) estimated average NO_3-N leaching fluxes from agricultural land in the UK to be 39-88 kg N ha^{-1} yr^{-1}, and Veltof *et al*. (2014) reported that the highest NO_3-N leaching fluxes from agricultural land in Europe were observed in the Netherlands and Belgium, where fluxes exceeded 50 kg-N ha^{-1} yr^{-1}. However, future improvements in farming practices to optimise fertiliser applications, could eventually reduce NO_3-N leaching fluxes from agricultural land to a level that matches NO_3-N leaching fluxes from some coniferous forest soils. Zhao *et al*. (2022) collated evidence from experimental studies on agricultural fertilisation in four European countries, reporting that at optimal fertilisation rates, mean NO_3-N leaching fluxes from agricultural land was 27 kg-N ha^{-1} yr^{-1}.

Improvements to water quality are frequently cited by stakeholders as benefits of afforestation, and has been considered to be one of the most important ecosystem services provided by forests (Nisbet *et al*., 2021; Lienhoop & Brouwer, 2015), even influencing willingness-to-pay for ecosystem services in forested catchments (Aguilar *et al*., 2018). There is some awareness of the potential negative effects of effects of afforestation on water quality, such as acidification (Dhubháin *et al*., 2009), which are reflected in professional guidance. For example, the UK Forestry Standard highlights the potential for forests to remove excess nutrients from former agricultural land but acknowledges that planting conifer forest in polluted areas could enhance NO_3-N leaching (Forest Research, 2023). The wide variation in NO_3-N leaching fluxes observed in the present study, along with the trend of increasing NO_3-N leaching fluxes with age in conifer sites, highlights the need to consider variation between sites, and the timescale over which ecosystem services are provided by afforested sites.

A significant difference in soil C:N ratios between conifers and broadleaves was expected, considering the previously established control exerted by tree species on mineral topsoil C:N ratios (Cools et al., 2014), but not observed. The effect of former land use on mineral soil C:N ratios was observed, indicating that former land use may have a stronger impact than forest type. However, C:N ratios were slightly lower in broadleaved compared to coniferous forests when planted on old-growth forest sites, suggesting that tree species effects on soil C:N ratios are still detectable in the absence of former land use impacts. The significant interaction between throughfall TIN, soil type and former land use on the effect of mineral topsoil C:N ratios suggest external nutrient inputs and the nutrient baseline at the start of the growth of the forest influences mineral topsoil C:N ratios to a greater degree than internal nutrient cycling in the present dataset. As mineral topsoil pH was only significantly different between different soil types, the chemical properties of the soil do not appear to be directly affected by the tree species growing on the sites in the present dataset.

4.3. NO_3-N leaching in coniferous forest ecosystems

There was a significant difference in the response of NO_3-N leaching to throughfall TIN inputs in arable land, heathland/grassland, and secondary rotations afforested with coniferous species. The significant difference in mineral soil C:N ratios between former land use categories suggests that management practices associated with former land use activities affected the capacity of soils to retain N inputs. Former land use activity affects C:N ratios through effects on N mineralisation and nitrification (Ollinger *et al.*, 2002), and changes in soil horizon structure, especially organic layer thickness (Thuille *et al*., 1999; Thuille & Schulze, 2006). NO_3-N leaching was low in heathlands/grasslands afforested with conifers and high in secondary conifer rotations accompanied by high topsoil C:N ratios in heathlands/grasslands and low topsoil C:N ratios in conifer rotations. As observed in broadleaved species, afforested arable land displayed a high degree of variation in NO_3-N leaching fluxes and mineral topsoil C:N ratios.

Afforested heathlands/grasslands had topsoil C:N ratios mostly above 23, which typically indicates C-rich soils at low risk of elevated NO_3-N leaching (Gundersen *et al*., 2009). To prevent tree establishment, heathlands are sometimes subjected to soil scraping to remove nutrients (Gardiner & Vaughn, 2008), creating N-poor soils. Soil organic carbon stocks and C:N ratios typically increase following the afforestation of heathland (Stevens & Wesemael, 2008; Barcena *et al*., 2014; Smal *et al*., 2019; Tew *et al*., 2021). Low levels of NO_3-N leaching from heathlands/grasslands afforested with coniferous species are expected due to a higher capacity for these soils to retain N in the presence of elevated throughfall TIN inputs. Although different methods of tree removal and brash management can minimise disturbance (Stevens & Hornung, 1990), tree harvesting typically leads to a disruption of ecosystem C and N pools (Finér *et al*., 2003; Gupta & DeLuca, 2012) and loss of nutrients from the soil (Reynolds & Stevens, 1998). This, combined with the possibility that the first rotation of conifers was exposed to prolonged periods of elevated N deposition, may have caused a high initial N status of the soil before replanting, leading to high leaching levels from all sites planted as secondary rotations.

4.4. Age effects in coniferous forest

Whilst it does appear that former land use affects the response to throughfall TIN inputs, the forest age distribution in different land use categories in this dataset makes it difficult to separate the effects of former land use from age-related differences. Afforested arable lands were typically below 50 years of age, secondary rotations were above 50 years of age, and afforested heathlands/grasslands were distributed between 0-100 years of age. Although there were coniferous sites below the age of 50 that displayed throughfall TIN fluxes above 20 kg ha^{-1} yr^{-1}, there were only three sites below the age of 50 that displayed NO_3-N leaching fluxes above 10 kg ha^{-1} yr^{-1}, likely due to the high nutrient demand of younger trees. Van der Salm *et al.*, (2006) observed negligible NO_3-N leaching fluxes in spruce forests aged 0-14 despite throughfall TIN fluxes of up to 14.3 kg ha^{-1} yr^{-1}. The ICP Forests Level II and IFEF databases are dominated by mature forests where a linear relationship is expected between throughfall TIN fluxes and NO_3-N leaching fluxes. Including younger forests in the present dataset may have weakened the relationship between throughfall TIN fluxes and NO_3-N leaching here.

The increase in NO_3-N leaching with tree age in conifers until approximately 80 years old is consistent with Sitka spruce chronosequence studies representing moorlands afforested 0-60 years of age in Wales (Stevens *et al.*, 1993; Emmett *et al.*, 1995). The decrease in NO_3-N leaching in older conifers and broadleaves after 50 years of age is consistent with observations from the ECOFEE dataset of conifers and broadleaves, where a reduction in leaching was apparent after approximately 40 years of age (Gundersen, 1995). Age effects on NO_3-N leaching can be attributed to changes in water fluxes, soil nutrient processes, and canopy cover with tree growth. Canopy development in coniferous forests also increases the scavenging of N compounds from the atmosphere (Schrijver *et al*., 2007). The increase in throughfall TIN with conifer age observed here is likely a dominant cause of increased NO_3-N leaching with conifer age. Further literature published on the Danish afforestation chronosequences included in this present dataset showed reductions in deep soil water drainage fluxes in the first 30-40 years after planting in both conifers and broadleaves, with a recovery in conifer stands by 65+ years (Rosenqvist, 2007). Reducing deep soil water drainage fluxes could lead to a concentrating effect, increasing NO_3-N concentration in soil solution.

4.5. Variation caused by inconsistencies in protocols between studies

Most studies included measured leaching fluxes using a chloride mass balance method to calculate water fluxes and multiplied these by deep soil solution NO_3-N concentrations. Some did, however, use site-specific water flux models, and variation between practices may have caused some of the observed variation in leaching fluxes. Differences between observational methods likely contributed to the higher variability observed here compared to other European datasets where observations are conducted according to a standardised protocol. The lack of reporting of water fluxes made it impossible to distinguish whether biochemical or hydrological processes drive variation in leaching fluxes between species, although this question merits further research.

4.6. Limitations of the dataset

Coniferous forests represented in this dataset were dominated by Norway spruce. However, significant differences in leaching fluxes between coniferous species and broadleaves have been observed. Borken & Matzner (2004) found that Norway spruce had more significant leaching fluxes than oak and beech, whereas Scots pine had lower. Differences between beech, oak and other broadleaved species not included in the present dataset have also been observed (Christiansen *et al.*, 2010).

We attempted to limit publication bias during the selection process by searching for Masters and doctoral theses and by searching for reports mentioned in review articles. The search string also selected studies on NO_3-N leaching, likely to preferentially select studies on sites that exhibited elevated NO_3-N leaching. This could be why most sites had C:N ratios of be 25, which can be considered an indicator of NO_3-N leaching (Gundersen *et al.*, 2009).

4.7. Proximity from agricultural land

N inputs to sites close in proximity to agricultural land are typically dominated by NH_3/NH_4 form rather than NO_3-N (Macdonald *et al.*, 2002). Soils located 500-700 m from agricultural land exhibited significantly lower mineral topsoil C:N ratios and had a broader difference in organic and mineral soil C:N ratios. As NH_4-N deposition has a greater contribution to soil acidity than NO_3-N deposition due to nitrification (Huang *et al.*, 2014), the observation here that the organic layer in these sites was less acidic than organic layers in the other categories indicates there were lower ammonium inputs at these sites, consistent with studies on edge effects on deposition and soil. Deposition, microclimatic gradients, and changes in soil micro- and macro-biota were associated with decreasing soil C and N stocks with increasing distance from the forest edge in a 0-128 m transect study (Remy *et al.*, 2016). Reduced solution NO_3-N and NH_4-N with increasing distance from the forest edge were similarly observed in a 0-200 m transect study of Corsican pine and beech (Vanguelova & Pitman, 2019). The observations in this present study suggest that edge effects are not only visible within forest stands but also within the whole forested landscape, with variations in C and N cycling visible up to 750 m from agricultural land.

4.8. Implications

The findings of this research have several implications for afforestation, forest management, landscape ecology and climate change. We have demonstrated that former land use affects soil C:N ratios and have provided further evidence to suggest that the ability of forest ecosystems to retain N is partially dependent on the availability of other nutrients in the soil. Nutrient availability, dependent on former land management practices, soil and forest type, should be considered when selecting sites and species to plant. The position within the landscape should also be considered during afforestation schemes as proximity to agriculture affects the stratification of C and N in the soil may affect the formation of soil layers, root biomass, and architecture.

We recommend future research prioritisation on: (1) landscape-scale effects on the stratification of nutrients in woodland soils and (2) the role of nutrients such as P, K, Mg and Ca on carbon and N stocks in afforested land and rotation forestry. The latter should be achieved by selecting sites already used for NO_3-N leaching research. Afforestation chronosequence studies on P, K, Mg and Ca exist but were not studied in the same afforestation chronosequence studies used for NO_3-N leaching research, limiting our interpretations.

5. Conclusions

This study collated a database from published literature on N-NO3 leaching, throughfall TIN inputs and site information, providing an independent dataset to test the outputs from continental-scale studies. The poor performance of previous regression models in predicting N-NO3 leaching fluxes from throughfall TIN in the present dataset could be attributed to methodological inconsistencies in NO3-N leaching fluxes, but also the complex land use histories and associated variations in soil nutrients may have contributed. An improved understanding of interactions between different nutrient cycles, particularly nitrogen and phosphorus cycles, is required to better predict the response of afforested arable land to deposition inputs. Such an understanding is necessary to understand the potential of afforestation to mitigate elevated NO3-N leaching from arable soils resulting from past fertiliser use. The higher NO3-N leaching fluxes in secondary conifer rotations compared to afforested heathland/grassland inspire the idea of exploring the incorporation of periods of open-field management between forestry rotations to reduce future NO3-N leaching fluxes, although implications for other nutrient cycles in this scenario would need to be considered.

References

Aguilar, F.X., Obeng, E.A., Cai, Z. (2018) Water quality improvements elicit consistent willingness-to-pay for the enhancement of forested watershed ecosystem services. *Ecosystem Services*, 30, 158-171.

Augusto, L., Ranger, J., Binkley, D., Rothe, A. (2002) Impact of several common tree species of European temperate forests on soil fertility. *Annals of Forest Science*, 59, 233-253.

Barcena, T.G., Kiaer, L., Vesterdal, L., Stefansdottir, H.M., Gundersen, P., Sigurdsson, B.D. (2014) Soil carbon stock change following afforestation in Northern Europe: a meta-analysis. *Global Change Biology*, 20, 2393-2405.

Bergkvist, B., Folkeson, L. (1991) The influence of tree species on acid deposition, proton budgets and element fluxes in south Swedish forest ecosystems. *Ecological Bulletins*, 44, 90-99.

Borken, W., Matzner, E. (2004) Nitrate leaching in forest soils: an analysis of long-term monitoring sites in Germany. *Journal of Plant Nutrition and Soil Science*, 167, 277-283.

Braun, S., Schindler, C., Rihm, B. (2017) Growth trends of beech and Norway spruce in Switzerland: The role of N deposition, ozone, mineral nutrition and climate. *Science of the Total Environment*, 600, 637-646.

Braun, S., Schindler, C., Rihm, B. (2020) Foliar nutrient concentrations of European Beech in Switzerland: Relations with N deposition, ozone, climate and soil chemistry. *Frontiers for Global Change*, 3.

Braun, S., Thomas, V.F.D., Quiring, R., Fluckiger, W. (2010) Does N deposition increase forest production? The role of phosphorus. *Environmental Pollution*, 158, 2043-2052.

Christiansen, J.R., Vesterdal, L., Callesen, I., Elberling, B., Schmidt, I.K., Gundersen, P. (2010) Role of six European tree species and land-use legacy for N and water budgets in forests. *Global Change Biology*, 16, 2224-2240.

Cools, N., Vesterdal, L., Vos, B., Vanguelova, E. Hansen, K. (2014) Tree species is the major factor explaining C:N ratios in European forest soils. *Forest Ecology and Management*, 311, 3-16.

Copernicus Land Monitoring Service, European Union (1990) *European Environment Agency Corine Land Cover 2000.* Available: https://land.copernicus.eu/pan-european/corine-land-cover/clc-1990

Copernicus Land Monitoring Service, European Union (2000) *European Environment Agency Corine Land Cover 2000*. Available: https://land.copernicus.eu/pan-european/corine-land-cover/clc-2000

Copernicus Land Monitoring Service, European Union (2006) *European Environment Agency Corine Land Cover 2006*. Available: https://land.copernicus.eu/pan-european/corine-land-cover/clc-2006

Dawud, S.M., Vesterdal, L., Raulund-Rasmussen, K. (2017) Mixed-species effects on soil C and N stocks, C/N ratio and pH using a transboundary approach in adjacent common garden Douglas-Fir and beech stands. *Forests*, 95.

Dhubháin, A.N., Fléchard, M.C-, Moloney, R., O'Connor, D. (2009) Stakeholders' perceptions of forestry in rural areas—Two case studies in Ireland. *Land use policy*, 26, 695-703.

Dise, N.B., Rothwell, J.J., Gauci, V., van der Salm, C., de Vries, W. (2009) Predicting dissolved inorganic N leaching in European forests using two independent databases. *Science of the Total Environment*, 407, 1798-1808.

Dise, N.B., Wright, R.F. (1995) N leaching from European forests in relation to N deposition. *Forest Ecology and Management*, 71, 153-161.

Emmett, B.A., Reynolds, B. (1996) N critical loads for spruce plantations in Wales: is there too much N? *Forestry*, 69, 205-210.

Emmett, B.A., Stevens, P.A., Reynolds, B. (1995) Factors influencing N saturation in Sitka spruce stands in Wales, UK. *Water, Air, and Soil Pollution*, 85, 1629-1634.

Fichter, J., Dambrine, E., Turpaut, M.P., Ranger, J. (1997) Base cation supply in spruce and beech ecosystems of the Strengbach catchment (Vosges Mountains, N-E France). *Water, air and soil pollution*, 104, 125-148.

Finér, L., Mannerkoski, H., Piirainen, S., Starr, M. (2003) Carbon and N pools in an old-growth, Norway spruce mixed forest in eastern Finland and changes associated with clear-cutting. *Forest Ecology and Management*, 174, 51-63.

Forest Research (2023) *The UK Forestry Standard*. 5th edn. Farnham: Forestry Commission.

Freedman, D., Pisani, R., Purves, R., Adhikari, A. (1978) *Statistics*. 2nd edn. US: Norton.

Gardiner, T., Vaughan, A. (2008) Responses of ground flora and insect assemblages to tree felling and soil scraping as an initial step to heathland restoration at Norton Heath Common, Essex, England. *Conservation Evidence*, 5, 95-100.

Gilliam, F.S. (2021) Response of temperate forest ecosystems under decreased N deposition: research challenges and opportunities. *Forests*, 12, 509.

Gundersen, P. (1995) N deposition and leaching in European forests — Preliminary results from a data compilation. *Water, Air, and Soil Pollution*, 85, 1179-1184.

Gundersen, P., Schmidt, I.K., Raulund-Rasmussen, K. (2006) Leaching of nitrate from temperate forests - effects of air pollution and forest management. *Environmental Research*, 14, 1-57.

Gundersen, P., Sevel, L., Christiansen, J.R., Vesterdal, L., Hansen, K., Bastrup-Birk, A. (2009) Do indicators of N retention and leaching differ between coniferous and broadleaved forests in Denmark? *Forest Ecology and Management*, 258, 1137-1146.

Gunina, A., Smith, A.R., Godbold, D.L., Jones, D.L., Kuzyakov, Y. (2016) Response of soil microbial community to afforestation with pure and mixed species. *Plant and Soil*, 412, 357-368.

Gupta, S.D., DeLuca, T.H. (2012) Short-term changes in belowground C, N stocks in recently clearfelled Sitka spruce plantations on podzolic soils of North Wales. *Forest Ecology and Management*, 281, 48-58.

Hansen, K., Rosenqvist, L., Vesterdal, L., Gundersen, P. (2007) Nitrate leaching from three afforestation chronosequences on former arable land in Denmark. *Global Change Biology*, 13, 1250-1264.

Herrmann, M., Pust, J., Pott, R. (2005) Leaching of nitrate and ammonium in heathland and forest ecosystems in Northwest Germany under the influence of enhanced nitrogen deposition. *Plant and Soil*, 273, 129-137.

Huang, J., Mo, J.M., Zhang, W., Lu, X. (2014) Research on acidification in forest soil driven by atmospheric N deposition. *Acta Ecologica Sinica*, 34, 302-310.

Jonard, M., Fürst, A., Verstraeten, A., Thimonier, A., Timmermann, V., Potočić, N., Waldner, P., Benham, S., Hansen, K., Merilä, P., Ponette, Q., de la Cruz, A.C., Roskams, P., Nicolas, M., Croisé, L., Ingerslev, M., Matteucci, G., Decinti, B., Bascietto, M., Rautio, P. (2015) Tree mineral nutrition is deteriorating in Europe. *Global Change Biology*, 21, 418-430.

Jost, G., Dirnbock, T., Grabner, M.T., Mirtl, M. (2011) Nitrogen leaching of two forest ecosystems in a karst watershed. *Water, Air and Soil Pollution*, 218, 633-649.

Jussy, J.H., Colin-Belgrand, M., Dambrine, É., Ranger, J., Zeller, B., Bienaimé, S. (2004) N deposition, N transformation and N leaching in acid forest soils. *Biogeochemistry*, 69, 241-262.

Kämäri, J., Rankinen, K., Finér, L., Piirainen, S., and Posch, M. (1998) Modelling the response of soil and runoff chemistry to forest harvesting in a low deposition area (Kangasvaara, eastern Finland). *Hydrological Earth System Sciences, 2, 485-495.*

Koerner, W., Dupouey, J.L., Dambrine, E., Benoit, M. (1997) Influence of past land use on the vegetation and soils of present-day forest in the Vosges mountains, France. *Journal of Ecology*, 85, 351-358.

Kristensen, H.L., Gundersen, P., Callesen, I., Reinds, G.J. (2004) Throughfall N Deposition Has Different Impacts on Soil Solution Nitrate Concentration in European Coniferous and Deciduous Forests. *Ecosystems*, 7, 180-192.

Lienhoop, N., Brouwer, R. (2015) Agri-environmental policy valuation: Farmers' contract design preferences for afforestation schemes. *Land use policy*, 42, 568-577.

Macdonald, J.A., Dise, N.B., Matzner, E., Armbruster, M., Gundersen, P., Forsius, M. (2002) N input together with ecosystem N enrichment predict nitrate leaching from European forests. *Global Change Biology*, 8, 1028-1033.

Meesenburg, H., Ahrends, B., Fleck, S., Wagner, M., Fortmann, H., Scheler, B., Klinck, U., Dammann, I., Eichhorn, J., Mindrup, M., Meiwes, K.J. (2016) Long-term changes of ecosystem services at Solling, Germany: Recovery from acidification, but increasing nitrogen saturation? *Ecological Indicators*, 65, 103-112.

Mellert, K.H., Gottlein, A. (2012) Comparison of new foliar nutrient thresholds derived from van den Burg's literature compilation with established central European references. *European Journal of Forest Research*, 131, 1461-1472.

Moldan, F., Jutterstrom, S.E.A-K., Hruska, J., Wright, R.F. (2018) Experimental addition of N to a whole forest ecosystem at Gardsjon, Sweden (NITREX): Nitrate leaching during 26 years of treatment. *Environmental Pollution*, 242, 367-374.

Muñoz, J.S. (2019) *ERA5-Land monthly averaged data from 1950 to present*. Copernicus Climate Change Service (C3S) Climate Data Store (CDS).

Nisbet, T.R., Andreucci, M.-B., De Vreese, R., Högbom, L., Kay, S., Kelly-Quinn, M, Leonardi, A., Lyubenova, M.I., Pol, P.O., Quinteiro, P., Silos, P., Valatin, G. (2021) Forests for water services: A step-by-step guide for payment schemes.Available:

https://cdn.forestresearch.gov.uk/2022/02/pesfor_cost_action_manual_2021_def_singlepages_final.pdf (Accessed 24th September 2024)

Ollinger, S.V., Smith, M.L., Martin, M.E., Hallett, R.A., Goodale, C.L., Aber, J.D. (2002) Regional variation in foliar chemistry and N cycling among forests of diverse history and composition. *Ecology*, 83, 339-355.

Oulehle, F., Goodale, C.L., Evans, C.D., Chuman, T., Hruska, J., Kram, P., Navratil, T., Tesar, M., Ac, A., Urban, O., Tahovska, K. (2021) Dissolved and gaseous N losses in forests controlled by soil nutrient stoichiometry. *Environmental Research Letters*, 16, 064025.

Ouzzani, M., Hammady, H., Fedorowicz, Z., Elmagarmid, A. (2016) Rayyan — a web and mobile app for systematic reviews. *Systematic Reviews*, 5, 210.

Pahlsson, B.A.-M., Bergkvist, B. (1995) Acid deposition and soil acidification at a southwest-facing edge of Norway spruce and European beech in South Sweden. *Ecological Bulletins*, 44, 43-53.

Ranger, J., Nys, C. (1994) The effect of spruce (*Picea abies* Karst.) on soil development: an analytical and experimental approach. *European Journal of Soil Science*, 45, 193-204.

Rehschuh, S., Jonard, M., Wiesmeier, M., Rennenberg, H., Dannenmann, M. (2021) Impact of European beech forest diversification on soil organic carbon and total N stocks - a meta-analysis. *Frontiers in Forests and Global Change*, 4.

Remy, E., Wuyts, K., Boeckx, P., Ginzburg, S., Gundersen, P., Demey, A., van der Bulcke, J., van Acker, J., Verheyen. (2016) Strong gradients in N and carbon stocks and temperate forest edges. *Forest Ecology and Management*, 376, 45-58.

Reynolds, B., Stevens, P.A. (1998) Assessing soil calcium depletion following growth and harvesting of Sitka spruce plantation forestry in the acid sensitive Welsh uplands. *Hydrology and Earth System Sciences*, 2, 345-352.

Ritter, E., Vesterdal, L. (2006) Gap formation in Danish beech (Fagus sylvatica) forests of low management intensity: soil moisture and nitrate in soil solution. *European Journal of Forest Research*, 125, 139-150.

Rohatgi, A. (2018) *WebPlotDigitizer*. Available: https://automeris.io/WebPlotDigitizer (Accessed June 2021)

Rosenqvist, L., (2007) *Afforestation of former arable land in North-Western Europe: Nitrate leaching, carbon sequestration and water recharge.* PhD thesis: Swedish University of Agricultural Sciences. Available: https://core.ac.uk/download/pdf/11695103.pdf

Rosenqvist, L., Hansen, K., Vesterdal, L., van der Salm, C. (2010) Water balance in afforestation chronosequences of common oak and Norway spruce on former arable land in Denmark and southern Sweden. *Agricultural and Forest Meteorology*, 150, 196-207.

Rothe, A., Huber, C., Kreutzer, K., Weis, W. (2002) Deposition and soil leaching in stands of Norway spruce and European beech: Results from the Hoglwald research in comparison with other European case studies. *Plant and Soil*, 240, 33-45.

Rubaek, G.H., Kristensen, K., Olesen, S.E., Ostegaard, H.S., Heckrath, G. (2013) Phosphorus accumulation and spatial distribution in agricultural soils in Denmark. *Geoderma*, 209-210, 241-250.

Sardans, J., Alonso, R., Carnicer, J., Fernández-Martínez, M., Vivanco, M.G., Peñuelas, J. (2016) Factors influencing the foliar elemental composition and stoichiometry in forest trees in Spain. *Perspectives in Plant Ecology, Evolution and Systematics*, 18, 52-69.

Schmitz, A., Sanders, T.G.M., Bolte, A., Bussotti, F., Dirnbock, T., Johnson, J., Penuelas, J., Pollastrini, M., Prescher, A-K., Sardans, J., Verstraeten, A., de Vries, W. (2019) Responses of forest ecosystems in Europe to decreasing N deposition. *Environmental Pollution*, 244, 980-994.

Schrijver, A.D., Geudens, G., Augusto, L., Staelens, J., Mertens, J., Wuyts, K., Gielis, L., Verheyen, K. (2007) The effect of forest type on throughfall deposition and seepage flux: a review. *Oecologia*, 153, 663-674.

Silgram, M., Waring, R., Anthony, S., Webb, J. (2001). Intercomparison of national & IPCC methods for estimating N loss from agricultural land. *Nutrient cycling in agroecosystems*, 60, 189-195.

Smal, H., Ligeza, S., Pranagal, J., Urban, D., Pietuczyk-Poplawska, D. (2019) Changes in the stocks of soil organic carbon, total N and phosphorus following afforestation of post-arable soils: A chronosequence study. *Forest Ecology and Management*, 451, 117536.

Stevens, A., Wesemael, B. (2008) Soil organic carbon stock in the Belgian Ardennes as affected by afforestation and deforestation from 1869 to 2005. *Forest Ecology and Management*, 256, 1527-1539.

Stevens, P.A., Emmett, B.A., Reynolds, B., Norris, D.A. (1993) *Critical loads of N for upland forests in Wales - effects of plantation age*. In: Critical loads: concept and applications. UK: London.

Stevens, P.A., Harrison, A.F., Jones, H.E., Williams, T.G., Hughes, S. (1993) Nitrate leaching from a Sitka spruce plantation and the effect of fertilisation with phosphorus and potassium. *Forest Ecology and Management*, 58, 233-247.

Stevens, P.A., Hornung, M. (1990) Effect of harvest intensity and ground flora establishment on inorganic-N leaching from a Sitka spruce plantation in North Wales, UK. *Biogeochemistry*, 10, 53-65.

Tahovská, K., Choma, M., Kaštovská, E., Oulehle, F., Bárta, J., Šantrůčková, H., Moldan, F. (2020) Positive response of soil microbes to long-term N input in spruce forest: Results from Gårdsjön whole-catchment N-addition experiment. *Soil Biology and Biochemistry*, 143, 107732.

Tew, E.R., Vanguelova, E.I., Sutherland, W.J. (2021) Alternative afforestation options on sandy heathland result in minimal long-term changes in mineral soil layers. *Forest Ecology and Management*, 483, 118906.

Thuille, A., Buchmann, N., Schulze, E-D. (1999) Carbon stocks and soil respiration rates during deforestation, grassland use and subsequent Norway spruce afforestation in the Southern Alps, Italy. *Tree Physiology*, 20, , 849-857.

Thuille, A., Schulze, E-D. (2006) Carbon dynamics in successional and afforested spruce stands in Thuringia and the Alps. *Global Change Biology*, 12, 325-342.

Tveite, H. (2016) *NNJoin*. Available: https://plugins.qgis.org/plugins/NNJoin/ (Accessed June 2021)

UK Government. (2021) *Net zero strategy: build back greener*. UK: HH Associates Ltd. (Accessed December 2021)

UK Government. (2021b) *The England Trees Action Plan 2021-2024*. UK: APS Group. (Accessed December 2021)

Van der Salm, C., de Vries, W., Reinds, G.J., Dise, N.B. (2007) N leaching across European forests: Derivation and validation of empirical relationships using data from intensive monitoring plots. *Forest ecology and management*, 238, 81-91.

Van der Salm, C., van der Gon, H.D., Wieggers, R., Bleeker, A., van den Toorn, A. (2006) The effect of afforestation on water recharge and N leaching in The Netherlands. *Forest Ecology and Management*, 221, 170-182.

Vanguelova, E.I., Pitman, R.M. (2019) Nutrient and carbon cycling along N deposition gradients in broadleaf and conifer forest stands in the east of England. *Forest Ecology and Management*, 447, 180-194.

Veltof, G.L., Lesschen, J.P., Webb, J., Pietrzak, S., Miatkowski, Z., Pinto, M., Kros, J., Oenema, O. (2014) The impact of the Nitrates Directive on nitrogen emissions from agriculture in the EU-27 during 2000–2008. *Science of the Total Environment*, 468-489, 1225-1233.

Walkiewicz, A., Rafalska, A., Bulak, P., Bieganowski, A., Osborne, B. (2021) How can litter modify the fluxes of CO2 and CH4 from forest soils? A mini-review. *Forests*, 12, 1276-1291.

Wang, Q., Wang, S., Huang, Y. (2008) Comparisons of litterfall, litter decomposition and nutrient return in a monoculture Cunninghamia lanceolata and a mixed stand in southern China. *Forest Ecology and Management*, 255, 1210-1218.

Wuijts, S., Fraters, D., Boekhold, S., van Duijnen. (2022) *Monitoring of N in water in the EU*. EU: European Parliament. (Accessed December 2023)

Zhao, J., Pullens, J.W.M., Sørensen, P., Blicher-Mathiesen, G., Olesen, J.E., Børgesen, C.D. (2022) Agronomic and environmental factors influencing the marginal increase in nitrate leaching by adding extra mineral nitrogen fertilizer. *Agriculture, Ecosystems and Environment*, 327, 107808.

Chapter 3

Throughfall dissolved organic carbon and NO_3-N fluxes drive deep soil NO_3-N leaching fluxes despite conversion of conifers to broadleaves in an N-saturated forest

Abstract

In the early 20th century, political motivations in Europe led to extensive afforestation and replacement of broadleaves with non-native coniferous species. Recent interest in converting coniferous plantations to broadleaves has been partly motivated by the latter's perceived enhancement of ecosystem services, although evidence of such improvements is lacking. This study investigated the impact of conifer-to-broadleaf conversion in Thetford Forest, UK, from August 2021-July 2022, measuring deep soil (60-90 cm) NO_3-N fluxes, throughfall chemistry, soil pH (10-30 cm), soil C:N ratios (0-10, 10-30 and 30-90 cm) and net nitrification (10-30 cm) in: (1) mature conifer, (2) former conifer sites planted with broadleaves 0-2 years ago, (3) sites converted to broadleaf 5-8 years ago, (4) sites converted to broadleaf 10-13 years ago, and (5) mature broadleaf stands.

In mature conifers and at both 5-8 and 10-13 years post-conversion, concentrations of NO_3-N in deep soil solution remained above the 11 mg-N/L threshold for drinking water set in the Water Framework Directive. Whilst mature broadleaves, on average, had lower deep soil NO_3-N leaching fluxes and concentrations than mature conifers, variation between replicates, likely due to edge effects, resulted in no significant differences. Soil C:N ratios at 0-10 cm depth were significantly lower in all broadleaved stands than in mature conifer stands (mature conifer = 18.9, sites planted with broadleaves 0-2 years ago = 12.1, 5-8 years ago = 14.3, 10-13 years ago = 13.6, mature broadleaf = 15.6). Soil C:N ratios at 0-10 cm depth were significantly positively related to deep soil NO_3-N leaching fluxes, and throughfall dissolved organic carbon (DOC) and NO_3-N fluxes, and deep soil NO_3-N leaching fluxes were also significantly positively related to throughfall DOC and NO_3-N fluxes but not throughfall NH_4-N fluxes. However, there were no differences between forest management stages in throughfall DOC and NO3-N fluxes or deep soil NO3-N leaching fluxes. Meanwhile, soil pH and net nitrification rates at 10-30 cm depth were significantly and negatively related, and neither were significantly related to deep soil NO_3-N leaching fluxes.

It is concluded that the effect of throughfall DOC and NO_3-N fluxes on processes in the top 0-10 cm of soil-controlled deep soil NO_3-N leaching fluxes during the study period and an effect of forest management practices were therefore not observed. The expected benefits of converting conifers to broadleaves may not be fully realised in scenarios where external inputs control deep soil NO_3-N leaching fluxes, and continued efforts to understand interactions between nutrient cycles in forest ecosystems are needed.

1. Introduction

Prolonged periods of elevated N deposition in the latter part of the 20th century have led to nitrogen saturation in forest soils across Europe (Dise *et al*., 2009; Macdonald *et al*., 2002) and other parts of the Northern Hemisphere (Zhu *et al*., 2015). Nitrate (NO_3-N) leaching occurs where N inputs exceed the total biological demand of an ecosystem (Aber *et al*., 1989), contributing to eutrophication and acidification of surface and groundwater (Gundersen *et al*., 2006; Wright *et al*., 2001), loss of base cations from the soil (Waldner *et al.*, 2015) and mobilisation of aluminium which causes toxicity to roots (Vanguelova *et al.*, 2005) and nutrient imbalances and deficiencies in trees (Jonard *et al.*, 2015).

Due to their greater atmosphere scavenging ability and a canopy all year round, coniferous forests are typically associated with higher levels of throughfall ammonium than broadleaved forests (Vanguelova & Pitman, 2019). Direct comparisons between conifers and beech forests growing in the same area showed that conifers had higher NO_3-N leaching fluxes, attributed to higher throughfall NH_4-N (Rothe *et al.*, 2002). Despite continuing reductions in air pollution across Europe (Dirnböck *et al.*, 2018; Fowler *et al.*, 2007), localised sources of pollution still contribute to high N deposition (Vanguelova & Pitman, 2019) and increase soil solution N in forest ecosystems in some countries, including the UK (Stevens, *et al.*, 2016). ^{15}N Isotope studies demonstrated that N-inputs to forest soils accumulated in the organic soils in the first 6-9 years, where ^{15}N is either immobilised or cycled between soil and vegetation, and deeper mineral soils become the more dominant sink 8-19 years after addition (Veerman *et al.*, 2020).

Observations in the UK thus point to an accumulated stock of soil N under conifers, which could be disturbed and leached out after forestry operations (Vanguelova *et al.*, 2024). Even in forests unaffected by high N deposition, harvesting and site preparation operations typically increase NO_3-N leaching fluxes from forest soils in the first decade. There are several mechanisms behind elevated NO_3-N leaching after forestry operations, such as soil disturbance, changes in vegetation uptake, and altered rates of soil biochemical processes, particularly increased rates of nitrification (e.g. Olsson *et al.*, 2022; Shah *et al.*, 2022; Jerabkova *et al.*, 2011; Huber *et al.*, 2010). Changes such as reduced interception and decreased transpiration, as a result of stand removal, result in increased soil drainage and groundwater recharge (Kubin *et al.*, 2017; Hedwall *et al.*, 2013) and ultimately in increased groundwater NO_3-N concentrations (Mannerkoski *et al.*, 2005; Neal *et al.*, 2004). However, increased water fluxes can dilute water pollutants, which has been observed in some cases (Schleppi *et al.*, 2017; Hartmann *et al.*, 2016; Calder *et al.*, 2002). Similarly, brash removal/whole tree harvesting can reduce NO_3-N leaching fluxes post-harvest by lowering mineralisation and nitrification (Hedwall *et al.*, 2013; Ring *et al.*, 2001; Emmett *et al.*, 1991). The establishment of ground vegetation following clear-felling is a dominant control on NO_3-N leaching fluxes (Bergholm *et al.*, 2015), with a strong relationship between NO_3-N leaching fluxes and percentage ground vegetation cover (Olsson *et al.*, 2022; George *et al.*, 2017). Brash cover can promote broadleaf tree growth on former coniferous sites; its removal may thus not be beneficial to N leaching in the long term (Pitman *et al.*, 2023). As an example, NO_3-N leaching fluxes can also be affected by former land use, with secondary conifer rotations displaying greater NO_3-N leaching fluxes than heathland afforested with conifers (Chapter 2).

Whilst NO_3-N leaching has been widely studied in the context of afforestation, NO_3-N leaching in a land-use change scenarios, such as planting broadleaves on land formerly covered with coniferous forest, remains unstudied. Conifer-to-broadleaf conversion has been preferred in central Europe since the latter part of the 20th century, such as in Belgium and Germany, where the typical practice for converting storm-endangered Norway spruce stands was block clearfelling followed by oak planting (Kint *et al.*, 2006; Kenk & Guehne, 2001). There is a growing interest in conifer-to-broadleaved conversion in the UK.

The UK has 1,566,000 ha of coniferous forest, accounting for 48 % of forested land (Forest Research, 2023). A small proportion is represented by native Caledonian pine forests (17,900 ha; Evans & Carr, 2023), while the rest are non-native plantations. In response to a national timber shortage following WW1, the UK Government acquired low-grade agricultural land to generate large amounts of conifer timber (Mason, 2007; Raum, 2020). However, by 1939, the planted stock was too immature to meet the needs of WW2. Thus, demand was met by harvesting broadleaves from the private forest estate, removing ancient, coppiced and semi-natural broadleaved woodland and restocking with conifers (Raum, 2020). The preference for conifer planting continued until the 1980s (Forestry Commission, 1985) and, finally, the restoration of coniferous plantations planted on ancient woodland sites became the government policy in 2005 (Goldberg *et al*., 2007; Defra, 2022). There was 223,000 ha of UK conifer woodland with economic incentives to support restoration as of 2009 (Latham *et al*, 2018), while scenario-led modelling in Wales predicted 7480 ha of coniferous forest would be converted to broadleaved woodland by 2030 (Manzoor *et al*., 2019). Following high rates of conifer planting in the 1960s and 1970s and with an optimum rotation period not lasting more than 60 years (Environmental Audit Committee, 2023; Mason, 2007), many conifer plantations across the UK are due to be felled in 2020-2030, presenting an opportunity for large-scale conversions to broadleaved species.

The present study focuses on the conifer-to-broadleaf conversion by clear-felling, followed by the planting of broadleaved seedlings. Continuous-cover forestry (CCF) approaches are preferred as a way of minimising NO_3-N leaching fluxes from coniferous plantations during harvesting (Webster *et al*., 2022; Marozas *et al*., 2018; Mupepele & Dormann, 2017; Weis *et al*., 2006) but economic incentives in the UK still typically favour tree planting over other methods of forest establishment such as natural regeneration and seeding, further discouraging CCF (Mason, 2020). Clear-felling, followed by planting, is likely the main method of converting conifers to broadleaves in parts of the UK.

Restocking clearfelled conifer plantations with broadleaved species changes several components of the forest ecosystem biochemical cycle, including:

(1) litter composition, a factor which drives variation in organic and mineral topsoil carbon and nitrogen stocks, soil pH, and microbial community composition (Cruz-Paredes *et al*., 2023; Feng *et al*., 2022; Ribbons *et al*., 2018; Cools *et al*., 2014; Prescott & Grayston, 2013). Litter quality, primarily litter N concentration, C:N ratio and lignin content, is a controlling factor of decomposition (Pei et al., 2019). The mixing of litters with different quality can stimulate decomposition, for example, mixing deciduous broadleaf with *Larix* litter increased decomposition in a Chinese temperate forest, and *Castanopsis eyrei* leaves decomposed faster when added to *Pinus massoniana* forest, due to the introduction of different fungal communities that facilitated faster decomposition of lignin and hemicelluloses (Zhang et al., 2019; Zhang et al., 2008). Planting broadleaf species on sites with coniferous litter could potentially introduce fungal communities that stimulate faster decomposition than restocking with conifers would.

(2) soil microbial community activity, due to changes in soil temperature and moisture (Brockett *et al*., 2012; Schindlbacher *et al*., 2011) and root exudation. Planting broadleaf species in coniferous soil could stimulate N mineralisation from organic matter by producing C-rich root exudates. Wang *et al*. (2021) reported broadleaf species produced

more carbon-rich root exudates than coniferous species, associated with higher microbial biomass, net N mineralisation and enzyme activity. The experimental addition of glucose led to a significantly higher priming effect in coniferous soil compared to broadleaf forest soil in a 60-day incubation by Liao *et al*. (2024). However, Yuan *et al*. (2018) found oxalic acid stimulated N mineralisation to a greater extent than glucose addition to a spruce and a spruce-fir plantation. Hamer & Marschner (2005) found that fructose and alanine stimulated soil organic carbon mineralisation more than oxalic acid and catechol. Tian *et al*. (2019) found that exudates with lower C:N ratios enhanced N cycling in *Quercus variabilis* soils, but only carbon-rich exudates enhanced N loss in *Pinus massoniana* soils. It is, therefore, reasonable to predict that broadleaf planting on coniferous soils, particularly those that have a history of elevated N deposition and therefore N-rich soils, could stimulate the mineralisation of N from organic matter. The increased availability of mineral N could later increase the rate of soil nitrification.

(3) plant nutrient demands and mycorrhizal associations (Gao *et al*., 2020; Liese *et al*., 2018; Adriaenssens *et al*., 2012; Mellert & Göttlein, 2012; Schulz *et al*., 2011). Whilst coniferous species and some broadleaved species, such as oak, are typically ectomycorrhizal, other broadleaf species have arbuscular mycorrhizal associations (Midgley & Phillips, 2014; Nilsson *et al*., 2007). High N deposition can be beneficial for arbuscular mycorrhizae whilst detrimental to ectomycorrhizae, with arbuscular mycorrhizal better able to mobilise and mineralise N (Ma et al., 2021), so a change from conifer to broadleaf species could introduce mycorrhizae better able to function in N saturated soils. Arbuscular mycorrhizae are associated with higher leaching of NO_3-N than ectomycorrhizae (Midgley & Phillips, 2014; Zechmeister *et al*., 2011). Fungi can also contribute significantly to microbial necromass in temperate forests (Xu *et al*.,

2024), with high N necromass decomposing faster (Beidler *et al*., 2020; Maillard *et al*., 2020). The loss of ectomycorrhizal, coniferous trees and replacement with broadleaf trees, particularly arbuscular species, could cause the death of established ectomycorrhizal mycelia, likely to be high in N content if under high N deposition conditions and therefore associated with faster decomposition.

(4) throughfall chemistry (Verstraeten *et al*., 2023; Schrijver *et al*., 2007). For example, pollen contributes greater amounts of K, DOC, dissolved organic nitrogen and NH_4-N under *Quercus* and *Fagus* than *Pinus* and *Picea* (Verstraeten *et al*., 2023).

The present study aimed to investigate the impact of conifer-to-broadleaf conversion on soil nitrogen cycling processes, focusing on NO_3-N leaching fluxes. The objectives were to:

(O1) Use a chronosequence study design to quantify the short-term effects of conifer-to-broadleaf conversion on NO_3-N leaching fluxes in a temperate forest ecosystem.

(O2) Explore relationships between NO_3-N leaching fluxes and variables, including throughfall chemistry, soil pH, net nitrification, and soil carbon and nitrogen stocks and ratios, across the chronosequence to begin to understand the mechanisms driving NO_3-N leaching fluxes during conifer-to-broadleaf conversion.

The hypotheses were:

(H1) Deep soil NO_3-N leaching fluxes will remain elevated in the first decade after planting broadleaves on former coniferous stands.

(H2) The nitrification rate drives deep soil NO3-N leaching fluxes.

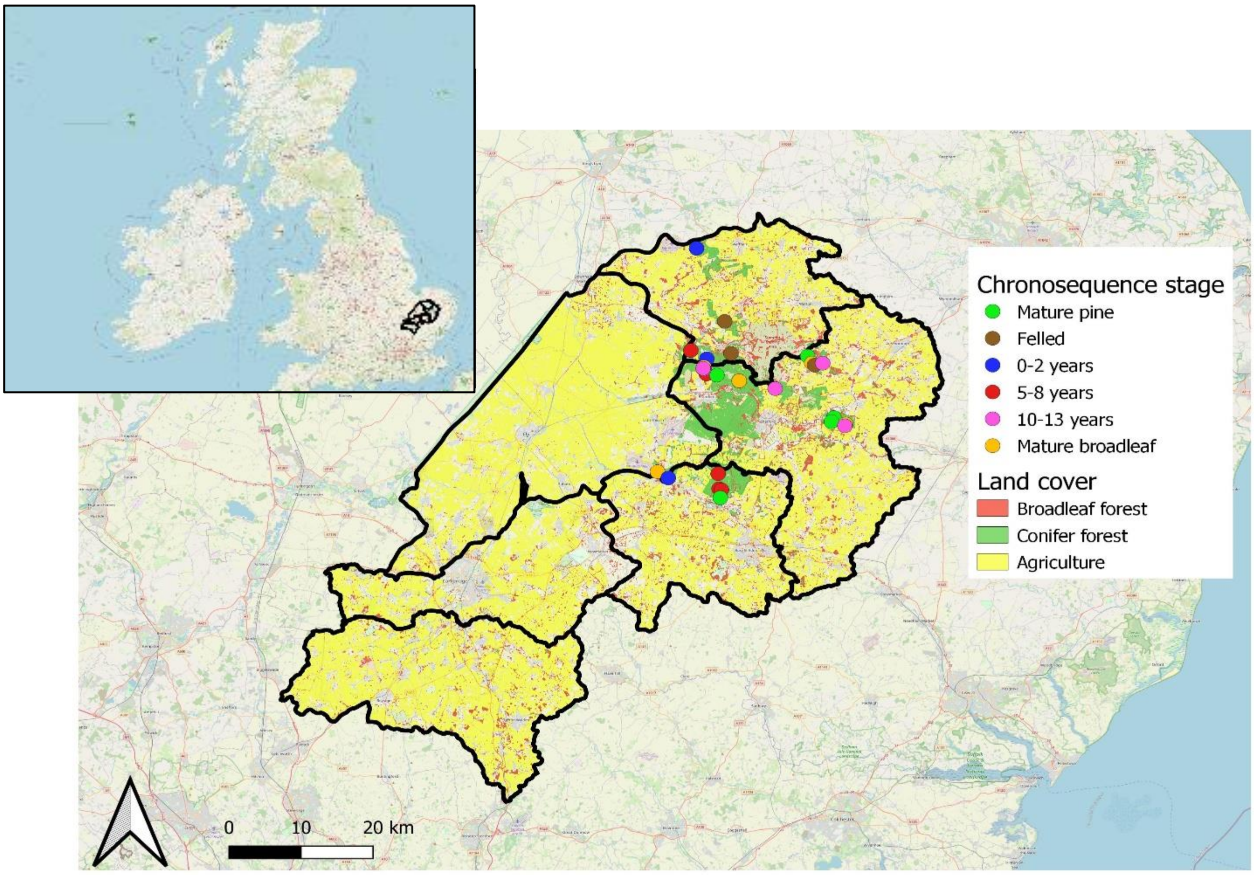
(H3) Deep soil NO_3-N leaching fluxes and nitrification will be related to topsoil pH and C:N ratios.

2. Methods

2.1. Site description

The experimental sites used for this study were located in Thetford Forest in the East of England, UK (Figure 1). Thetford Forest is a commercial plantation dominated by Scots and Corsican pine but with interspersed broadleaved stands, particularly along woodland block edges. The land surrounding the forest is used for intensive pig and chicken farming, and cropland is frequently subjected to fertiliser applications. Atmospheric N deposition across the region is high due to intensive farming activities; the area is within a nitrate-vulnerable zone (Environment Agency, 2019). A Scots pine stand at Thetford, which has been monitored since 1996 as part of the ICP Level II network (Vanguelova *et al*., 2007), shows elevated levels of NO_3-N in deep soil solution, with concentrations above the accepted drinking water standard set out in the Water Framework Directive (Vanguelova *et al*, 2010). Thetford experiences an average annual rainfall of 600 mm, annual temperature of 11.3 ^{o}C and an average nitrogen deposition of 14.7 kg ha^{-1} a^{-1} (Vanguelova & Pitman, 2019). Thetford Forest frequently experiences droughts, which cause pulses of NO_3-N leaching upon rewetting. The sites were in the Cam, Ely Ouse, Little Ouse and Thet management catchment (Figure 1). The soil texture across all sites was sandy. The site overlies the chalk aquifer, which is heavily abstracted for public and agricultural water supply (Appendix 6); protecting the groundwater is thus of stakeholder interest.

Figure 1: The location of the chronosequence sites across Thetford Forest, East England. Land cover was obtained from UKCEH 2015 landcover map.



A chronosequence of sites was selected to observe the changes in NO_3-N leaching fluxes in the first 10 years after converting conifers to broadleaves. The chronosequence consisted of (1) five mature pine stands, including the long-term monitoring site, (2) three sites felled in September 2021, February 2022 and March 2022, using conventional harvest with 10 % retention, (3) three sites planted with broadleaves 0-2 years ago, (4) five sites planted with broadleaves 5-8 years ago, (5) five sites planted with broadleaves 10-13 years ago, (6) five mature broadleaf sites, planted before 1950 to allow comparison with data in Vanguelova & Pitman (2019). Sites replanted with naturalised or native broadleaf species were preferentially selected, but non-native broadleaved species (Italian alder, red oak) were included where sufficient replicates could not be located. Monocultures were preferentially selected to avoid the influence of mixed species effects. Further details of all sites are provided in Appendix 7. The most distant sites were located 37 km apart.

2.2. Sample collection and analysis

2.2.1. Throughfall monitoring

At all sites older than 2 years, five throughfall collectors were constructed in July 2021. At each site, a 20 m buffer zone was left between the edge of the forest and the first sampling point. Sampling points were situated along a transect directed towards the centre of the stand, spaced 20 m apart. White PVC funnels (d = 25 cm) were mounted using a wooden stake 1 m above ground level and connected via a PVC pipe to a 5L plastic jerry can, which was buried in the soil or covered with ground vegetation where dense roots made it difficult to dig, to protect from direct sunlight. Throughfall samples were collected monthly until March 2022 for 19 sites and until July 2022 for 12 sites.

Throughfall samples were kept in a cooler box with ice packs until transported to a refrigerator in a laboratory within a week of collection and stored at 4 ^{o}C until analysis, as recommended by the ICP Forests manuals for measuring deposition. Throughfall samples were aggregated by site each month, filtered through a 45 mm filter, and analysed for Cl^- and NO_3^- with a Dionex DX 500 Ion Chromatography System with AS 40 autosampler. Samples from August 2021-July 2022, except for January 2023, were also analysed at the Forest Research, Alice Holt laboratory for pH by pH meter, NH_4-N colourimetrically, and DOC and total N by Carbon analyser (Shimadzu 5000, Osaka, Japan), following ICP protocols (Cools & De Vos, 2022). Total throughfall N flux from August 2021 to July 2022 was calculated as the sum of all months.

Throughfall volume was also measured at the time of sample collection. Where throughfall volume could not be accurately measured due to overflowing containers, a linear regression-based model was developed from the observed data to predict throughfall volume from rainfall volume for each stage individually in the winter and summer months. Missing throughfall volume observations were then estimated from rainfall volume data extracted from the Met Office HadUK-Grid dataset (Hollis *et al.*, 2019).

Throughfall was not collected at the felled sites or sites planted 0-2 years ago due to the absence of a canopy. Rainfall volume from the Met Office HadUK-Grid dataset and the concentration of chloride in rainfall collected at the open ICP monitoring site for monitoring bulk rainfall deposition were used to calculate water fluxes for recently felled and planted sites.

2.2.2.Soils solution monitoring

Lysimetric methods and rhizon samplers fail to yield sufficient soil solution during the summer months in Thetford. Therefore, a deionised water plus centrifugation-based method was used to obtain soil solution NO_3^- and Cl^- concentrations in deep soil samples. Soil samples were collected monthly from August 2021 until March 2022 for 9 sites and until July 2022 for 17 sites but were not sampled in November and December 2021. The soil was sampled at 0.6-0.9 m depth (below the rooting zone) at four sampling points at each site. Sampling points were located adjacent to the first four throughfall collectors. Soil was stored in a cooler box with ice packs until transported to a refrigerator in a laboratory within a week of collection and stored at 4 °C until analysis.

Soil samples were aggregated by site each month, and 36 g was added to 20 ml of deionised water and vortexed to suspend ions held in the soil. The soil was separated from the solution by centrifuging at 4000 rpm at 4 °C for 60 minutes. The solution was then filtered through a 45 mm filter analysed for Cl^- and NO_3^- with a Dionex DX 500 Ion Chromatography System with AS 40 autosampler. Final Cl^- and NO_3^-concentrations were calculated by adjusting for soil water concentrations. Soil water concentrations were calculated using the gravimetric method, oven drying 20 g of soil at 70 ° C for 48 hours until a constant weight was achieved and reweighing.

2.2.3.Calculation of leaching fluxes

NO_3-N leaching fluxes are a product of soil water fluxes and NO_3-N concentrations. The chloride mass balance method is commonly applied to calculate soil drainage fluxes based on the primary assumption that chloride is a conservative element in soils (Svensson *et al.*, 2012). The method assumes the only input of chloride is from deposition and that the system is in a steady-state condition, i.e. there have been no significant changes in chloride fluxes over time (Crosbie *et al.*, 2018). There have been no historical anthropogenic inputs to Thetford Forest, meeting the first assumption. Inaccuracies in the method occur in mountainous terrain due to variations in infiltration and runoff (Guan *et al.*, 2009). Still, East Anglia is a flat region, so that was not a concern here. The chloride mass balance method has been applied to investigate NO_3-N fluxes post-felling in forest ecosystems. Still, changes in vegetation cover and site preparation processes are transient between pre-felling and post-felling steady states and can contribute up to 24 % of ion variance (Guan *et al.*, 2013). Due to the changes in site topology during the transient period, particularly from planting seedlings in depressions, water fluxes were not calculated for the felled and 0-2 year broadleaf sites.

For all other sites, water fluxes for each month were calculated through the equation:

Water flux (mm) = (concentration of chloride in throughfall/concentration of chloride in soil solution) * throughfall volume in mm

For each site, leaching fluxes (kg-N ha^{-1}) were calculated for each month by multiplying water fluxes (mm/ha) by the concentration of NO_3-N in deep soil samples (mg/L). Leaching fluxes were presented in kg-N ha^{-1} by dividing leaching fluxes in mm mg ha^{-1} L^{-1} by 1000. Annual leaching fluxes were calculated as the sum of leaching fluxes for each month.

2.2.4. Soil C:N ratios, nitrification, pH, bulk density and organic layer depth

In July 2021, soil was sampled at four points per site aligned with the location of the throughfall collectors. The interest here was to investigate the movement of N down the soil profile, and due to the lack of layer formation in 0-2-year-old sites, the soil was sampled by depth rather than horizon. The soil was sampled at three soil depths: (1) 0-0.1 m, where present leaf litter was brushed aside, but the Oh layer was included in sampling, (2) 0.1-0.3 m, (3) 0.3-0.9 m. The depth of soil was highly variable within stands. Where the chalk layer was reached at less than 0.4 depth, an alternative sampling point was located within the stand. Soil was transported to the laboratory and stored at 4 °C until analysis. The samples were oven-dried for 48 hours, passed through a 5 mm sieve, mixed with a spatula, then 0.25 g was weighed per sampling point and analysed for total carbon, nitrogen and the C:N ratio using a Leco^{TM} CHN elemental analyser. In February 2022, bulk density was determined at two sampling points per site, at 0.1-0.3 and 0.3-0.9 cm soil depths, by digging a pit and hammering a metal cylinder horizontally into the respective layers. The soil in each cylinder was oven-dried for 48 hours and weighed, and the bulk density was calculated by bulk density = weight of oven-dry soil in g/volume of the cylinder in cm^3. Due to limited site access, bulk density at the two sites in January and February was not measured. Still, an average bulk density of the mature coniferous sites studied was used. All of the felled sites were sampled for total carbon and nitrogen before felling, and they are grouped in the analysis of C:N ratios as mature pine. Soil carbon and nitrogen stocks at 0.1-0.3 and 0.3-0.9 depths were calculated from the % of carbon and nitrogen multiplied by the bulk density and soil layer width.

An in situ incubation described by Carter & Gregorich (2006) was used to determine the nitrification rate. The incubation method allowed the observation of the change in NH_4-N and NO_3-N under field conditions without root uptake and deposition inputs. In July 2021, at each site, four clear PVC cylinders (d = 5cm, l = 10 cm) with rubber caps were planted vertically at four points in the soil, with the top of the cylinder at 0.1 cm depth. Four small holes (<1 cm diameter) were punched in each cap to allow gas exchange. At the time of plantation, soil samples were collected at 0.1-0.3 m depth adjacent to the cylinders and kept at 4 ^{o}C until analysis. The soil in the cylinders was collected in August 2021. All samples were analysed using the KCl extraction method. For each sampling point, 20 g of soil was used to determine soil water content by oven drying for 48 hours, and 40 g of soil (passed through 1 mm sieve) was added to 200 ml of 1M KCl solution. The soil in KCl solution was shaken on a horizontal shaker for one hour, filtered and analysed within two days of extraction with a Skalar^{TM} San^{++} continuous flow analyser. Some output values (16 %) were below the limit of detection (LOD). For the calculations, values below the LOD were considered equal to LOD/2, as recommended by the EPA (EPA, 2000). NH_4 and NO_3 concentrations were adjusted for soil water content, converted to NO_3-N/NH_4-N, and multiplied by bulk density and soil depth to present results in kg ha^{-1}. The increase in NO_3-N and sum of NH_4-N concentrations during the incubation period was considered the rate of nitrification and mineralisation, respectively (Δ kg ha^{-1}).

In January 2022, additional soil samples were collected at two points at 0.1-0.3 m depth and aggregated, and pH (H_2O) was measured in the laboratory using a pH probe. The depth of the soil organic layer (O_F, O_H) was also measured at four sampling points per site.

2.2.5. Statistical analysis

To identify potential relationships between all variables, a correlation matrix was created to visualise relationships to investigate further (Appendix 8). The relationship between variables with a Pearson’s r value of >0.4 was tested with a linear regression. Variables that did not meet the assumptions for a parametric test were log-transformed. Pairwise comparisons of estimated marginal means were conducted on linear models using the emmeans package in R. Mean water fluxes, deep soil solution NO_3-N concentrations and leaching fluxes were compared between each chronosequence stage for each month using ANOVA. Mean C:N ratios and C and N stocks for each depth at the site were calculated, and then a mean for each stage was determined. Mean nitrification rates, mineralisation and O-layer thickness were determined for each site, and then a mean for each stage was calculated. All variables were tested for the assumptions for a parametric test by the visualisation of residual diagnostic plots in R. Those that did not meet the assumptions were log-transformed. The difference in C:N ratios, C and N stocks, rate of nitrification and mineralisation, O-layer thickness, pH and bulk density between stages was then compared using ANOVA with a significance threshold of $p<0.05$, looking for the overall significance of "stage" as an explanatory variable and using Tukey's Honest Significant Differences to conduct pairwise comparisons.

3. Results

3.1 Patterns in NO_3-N leaching fluxes, water flues and mean NO_3-N concentrations

There was an overall significant difference in the total observed deep soil water flux between stages (ANOVA, $F = 4.64$, $p < 0.05$), though pairwise comparison did not identify significantly different stages. Overall, mean deep soil solution NO_3-N concentrations were not significantly different between stages from July 2021-2022 ($F = 2.09$, $p > 0.05$). There was no significant difference in the total observed NO_3-N leaching flux between stages from July 2021 to July 2022 (Table 1, ANOVA, $F = 1.96$, $p > 0.05$). The three mature conifer sites studied for the year had total NO_3-N leaching fluxes of 60.29, 38.76 and 27.38 kg-N ha^{-1}. The three mature broadleaves studied for the year had total NO_3-N leaching fluxes of 10.74, 11.84 and 29.73 kg-N ha^{-1}.

There were differences in seasonal patterns between stages in NO_3-N leaching fluxes, deep soil drainage and deep soil solution NO_3-N concentrations (Figure 2). Mature pine displayed the highest peak in deep soil solution NO_3-N concentrations and NO_3-N leaching fluxes. Stands 5-8 years old displayed the highest peak in water fluxes. Deep soil solution NO_3-N concentrations increased in mature broadleaf stands between May and July 2022, whereas concentrations remained stable in all other sites. Water fluxes from mature stands peaked a month later than those with young trees.

Table 1: The mean total NO_3-N leaching fluxes, deep soil drainage, and mean soil solution NO_3-N concentrations from stands at different stages in the conifer-to-broadleaf conversion process, between August 2021 and July 2022.

	Mature pine	0-2 years	5-8 years	10-13 years	Mature broadleaf	p
Total annual NO_3-N leaching (kg-N/ha)	42.14	-	22.15	22.12	17.43	0.2
Total annual drainage (mm/year)	176.98	-	193.65	125.6	130.56	0.03
Mean soil solution NO_3-N (mg-N/L)	30.88	23.64	15.34	19.4	12.46	0.18

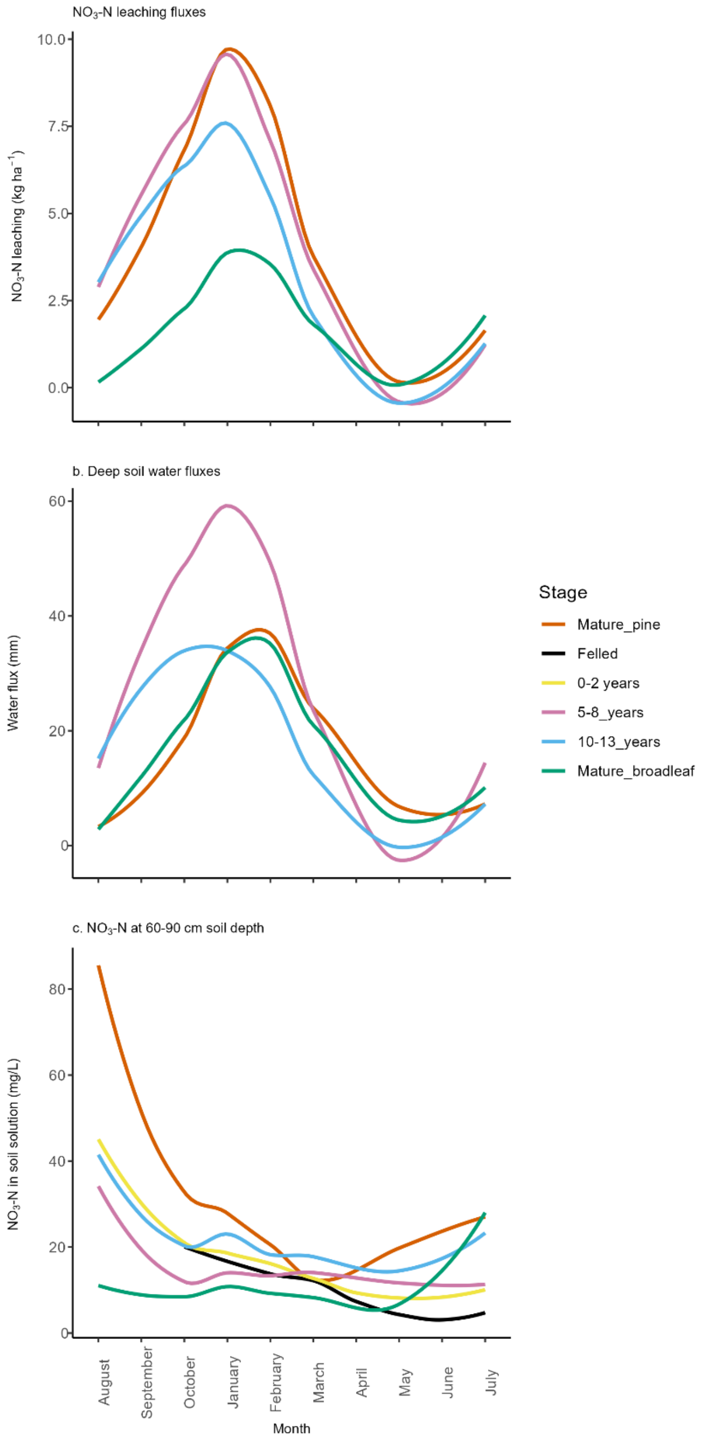


Figure 2: (a) Total NO_3-N leaching fluxes (kg ha^{-1} month^{-1}), (b) total deep soil drainage fluxes (mm), and (c) mean deep soil solution NO_3-N concentrations (mg/L), between August 2021 and July 2022, in stands at different stages in the conifer-to-broadleaf conversion process.

3.2 Throughfall chemistry

There was no overall significant difference in the total observed throughfall NO_3-N fluxes (ANOVA, $F = 2.68$, $p > 0.05$), throughfall NH4-N fluxes ($F = 2.16$, $p > 0.05$), throughfall nitrogen fluxes (TN, $F = 0.33$, $p > 0.05$) and throughfall DOC fluxes ($F = 3.87$, $p = 0.056$) between stages from July 2021-July 2022. However, there were differences in seasonal patterns between stages in throughfall chemical fluxes (Figure 3). Sites planted with broadleaves 5-8 and 10-13 years ago had elevated levels of throughfall NH_4-N fluxes from August to October 2021. Throughfall NO_3-N fluxes peaked in February for all broadleaves, but peaked in March for mature pine and sites planted 10-13 years ago. Sites planted 10-13 years ago had the greatest peak in throughfall DOC fluxes in September 2021, coinciding with a peak in NH_4-N. Mature pine throughfall NH_4-N fluxes peaked when throughfall DOC fluxes reached their lowest point.

No formal measurement of ground vegetation species composition was conducted, but visual assessment showed that most mature broadleaf and coniferous sites had few understory shrubs. Broadleaves had little or no vegetation cover, whereas conifer sites had grass, bracken or bramble. The 0-2 year sites were all covered in short grasses only. The 5-8 year sites had full grass coverage, except for two sites with dense bracken and brambles, taller than the trees. The 10-13 year sites either had grass coverage or dense bracken. Percentage ground vegetation cover measurements at selected sites showed no significant differences between stages (data not shown).

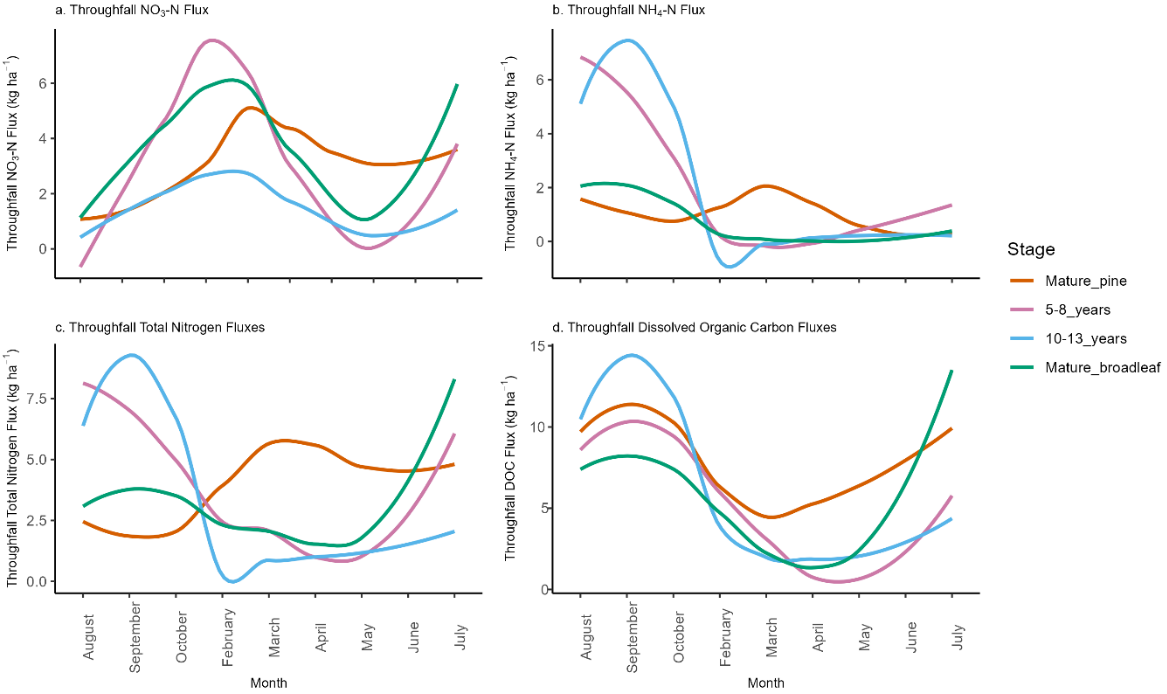


Figure 3: Mean (a) throughfall NO_3-N fluxes (kg ha^{-1} month^{-1}), (b) throughfall NH_4-N fluxes (kg ha^{-1} month^{-1}), (c) throughfall total N fluxes (kg ha^{-1} month^{-1}), and (d) throughfall DOC fluxes (kg ha^{-1} month^{-1}), between August 2021 and July 2022, in stands at different stages in the conifer-to-broadleaf conversion process.

3.3 Soil C and N cycling

The rates of nitrification and mineralisation were not significantly different between the stages (Figure 4, F = 1.93, $p > 0.05$ and F = 1.27, $p > 0.05$) or species (F = 3.42, $p > 0.05$ and F = 2.19, $p > 0.05$). The organic layer thickness and pH at 10-30 cm soil depth were also not significantly different between the stages (F = 1.00, $p > 0.05$ and F = 1.58, p = 0.22). Sites planted 0-2 years ago had no organic layer present, and there was little variation in the thickness of the organic layer between mature pine stands.

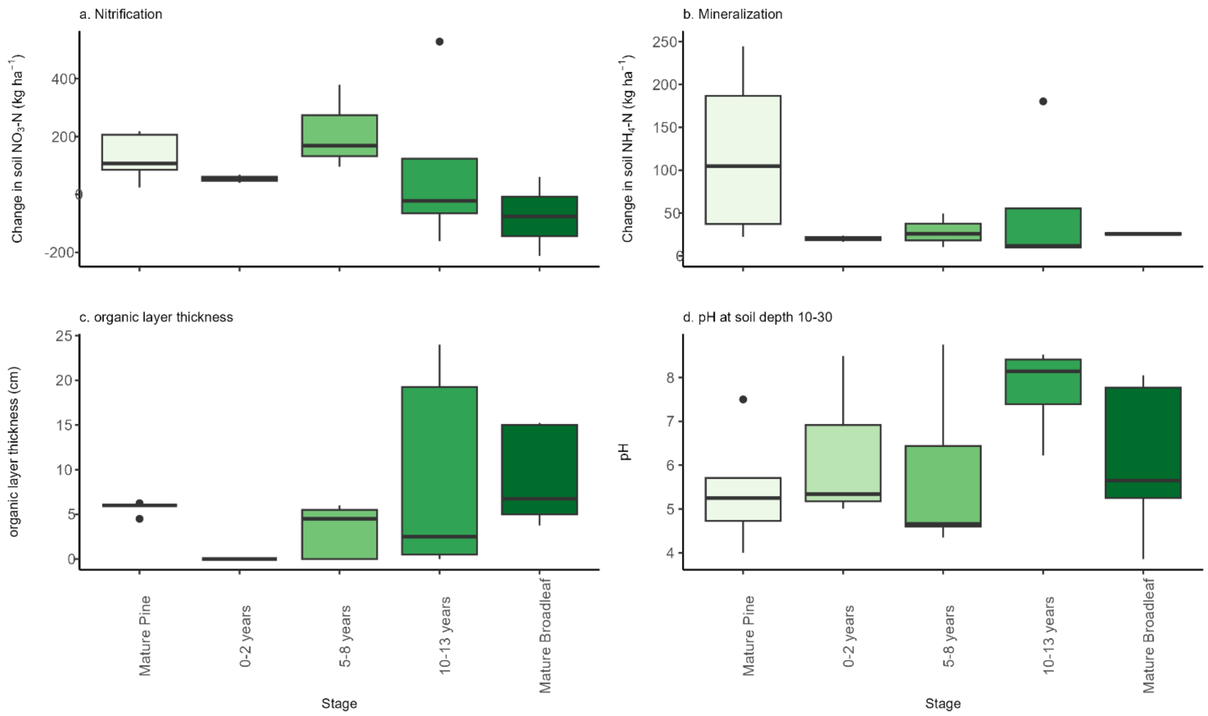


Figure 4: In stands at different stages in the conifer-to-broadleaf conversion process:(a) The rate of change in NO_3-N extracted by the KCl extraction method at 10-20 cm soil depth between July and August 2021 (*in situ* nitrification), (b) the sum of NH_4-N extracted by the KCl extraction method at 10-20 cm soil depth in July and August 2021 (*in situ* mineralisation), (c) organic layer thickness (cm), (d) pH of soil at depth 10-30 cm.

The C: N ratio at soil depth 0-10 cm significantly differed between stages (Figure 5, F = 17.66, $p < 0.01$). The 0-10 cm C:N ratio of sites planted 0-2 years ago (C:N = 12.1) was significantly lower than that in mature pine (C:N = 18.9, $p < 0.01$) and mature broadleaved sites (C:N = 15.6, $p < 0.05$). Mature broadleaf sites planted 5-8 years ago (CN = 14.3) and sites planted 10-13 years ago (C:N = 13.6) were significantly lower than that in mature pine stands ($p < 0.01$), but were not significantly different to each other.

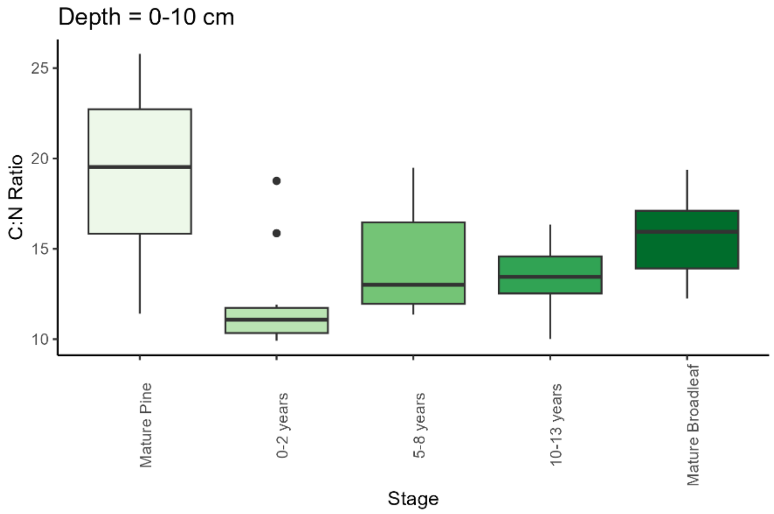


Figure 5: The C:N ratio of the organic soil layer in mature pine (Mean = 18.9, n = 10), former pine sites replanted with broadleaved species 0-2 years ago (Mean = 12.1, n = 3), 5-8 years ago (Mea = 14.3, n = 5), 10-13 years ago (Mean = 13.6, n = 5) and mature broadleaved sites (Mean = 15.6, n = 5).

There was no significant difference in the C:N ratio at soil depths 10-30 and 30-90 cm depth between the stages or the total C and total N stocks (F = 2.14, p = 0.08 and F = 1.95, p > 0.05). In soils at 30-90 cm depth, there was an increase in N stocks 10-13 years after replanting sites with broadleaved species, although the increase is not quite significant (p = 0.07).

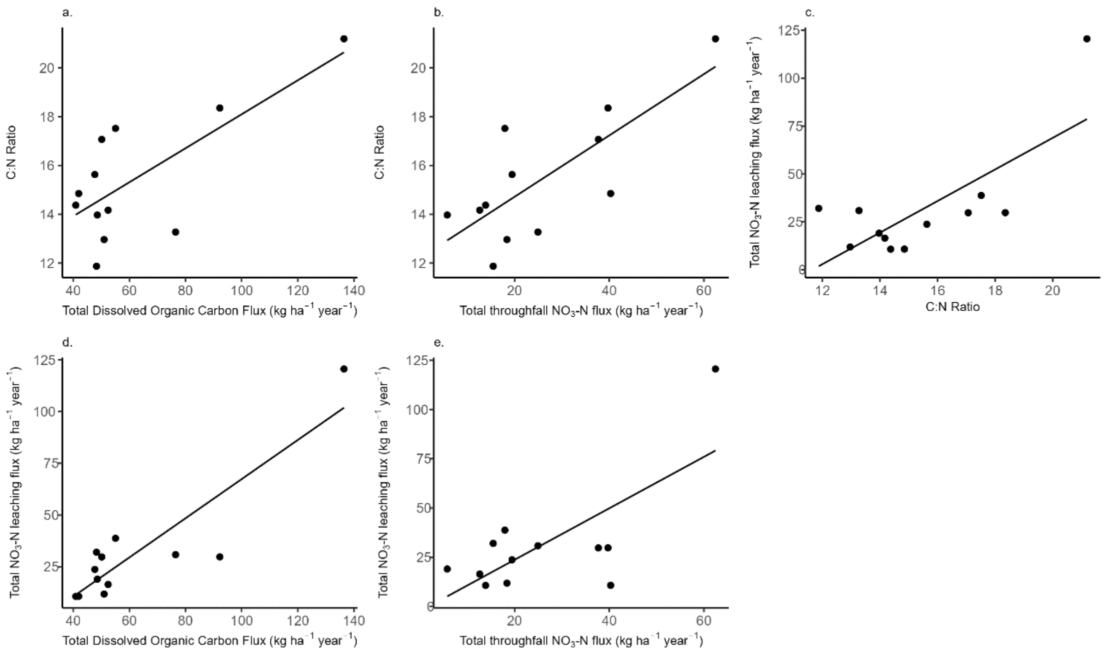


Figure 6: For 12 sites, including Scots pine sites and broadleaf sites of varied species and ages, monitored for one year (August 2021-July 2022) in Thetford Forest: the C:N ratio of soils at 0-10 cm depth and (a) the total flux of DOC in throughfall, (b) the total flux of NO_3-N in throughfall, (c) the total flux of NO_3-N in deep soil solution. The total flux of NO_3-N in deep soil solution and: (d) the total flux of DOC in throughfall, (e) the total flux of NO_3-N in throughfall, are also displayed. All relationships displayed are significant ($p < 0.05$).

3.4 . Relationships between soil C, N, pH, nitrification, throughfall chemistry and deep soil NO_3-N leaching fluxes

A positive but insignificant relationship existed between the nitrification rate and total annual throughfall NH_4-N ($F = 1.88$, $p > 0.05$). Other deposition inputs were similarly not related to nitrification. Nitrification was not significantly related to annual average throughfall pH ($F = 0.56$, $p > 0.05$) or total annual throughfall NO_3-N ($F = 0.02$, $p > 0.05$) or DOC flux ($F = 0.18$, $p > 0.05$). Soil C:N ratios at depth 0-10 cm were positively related to throughfall DOC fluxes (Figure 6a, F = 11.34, $p < 0.05$), but deep soil C:N ratios at 10-30 and 30-90 cm were not (F = 0.58 and 0.11, $p > 0.05$). Soil C:N ratios at 0-10 cm depth were also positively related to throughfall NO_3-N fluxes (Figure 6b, F = 14.12, $p < 0.01$). Soil C:N ratios at 10-30 and 30-90 cm depth were not significantly correlated with throughfall NH_4-N (F = 0.04 and 0.05,, $p > 0.05$), throughfall NO_3-N (F = 0.31 and 0.41, $p > 0.05$), or throughfall TN (F = 0.02 and 0.3, $p > 0.05$).

There was no significant relationship between the nitrification rate and C:N ratios of soil at 0-10 cm depth (F = 0.2, $p > 0.05$), but there was a significant negative relationship between the nitrification rate and C:N ratios of soil at 10-30 cm depth (Figure 7a, F = 6.45, $p < 0.05$) and 30-90 cm depth (F = 15.17, $p < 0.01$). There was also a significant negative relationship between nitrification rate and soil pH at 10-30 cm depth (Figure 7b, F = 6.33, $p < 0.05$). A significant, positive relationship existed between nitrification rate and organic layer thickness (Figure 7c, F = 7.79, $p < 0.05$). The mineralisation rate was not significantly related to C:N ratio (F = 1.6, $p > 0.05$) or pH (F = 0.51, $p > 0.05$) at 10-30 cm soil depth, nor organic layer thickness (F = 1.17, $p > 0.05$).

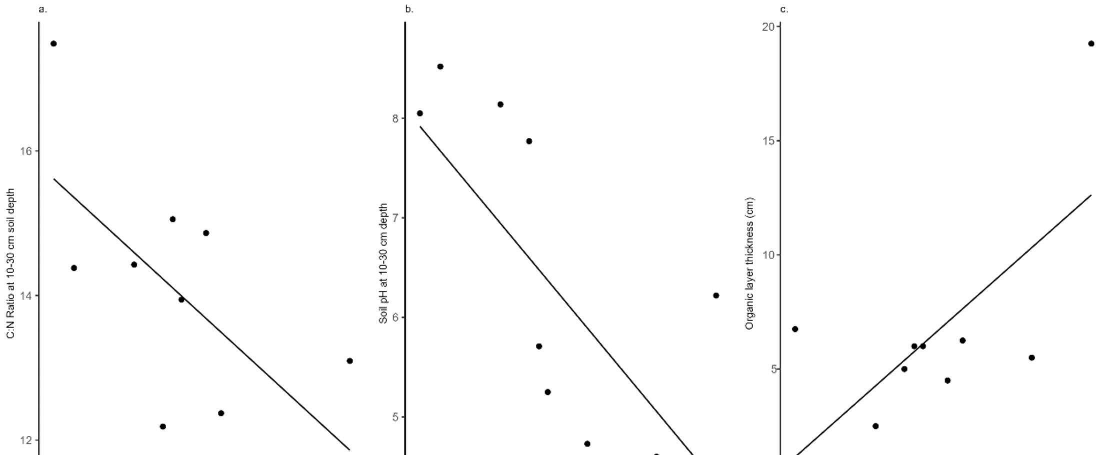


Figure 7: The (a) C:N ratio at 10-30 cm soil depth, (b) soil pH at 10-30 cm soil depth, and

(c) organic layer thickness against the change of NO_3-N during an *in situ* soil incubation between July-August 2021, at Scots pine sites and broadleaf sites of varied species and ages, in Thetford Forest. All relationships displayed are significant ($p < 0.05$).

There was no significant relationship between deep soil NO_3-N leaching fluxes and the nitrification rate in the upper soils ($F = 0.02$, $p > 0.05$). NO_3-N leaching fluxes were unrelated to soil pH at 10-30 cm depth ($F = 0.56$, $p > 0.05$). NO_3-N leaching fluxes were, however, significantly related to soil C:N ratios at 0-10 cm depth (Figure 6c, $F = 11.89$, $p < 0.01$), but not 10-30 cm depth ($F = 0.08$, $p > 0.05$) or 30-90 cm ($F = 0.05$, $p > 0.05$).

There was a significant relationship between NO_3-N leaching fluxes and throughfall DOC fluxes (Figure 6d, $F = 36.29$, $p < 0.0001$), throughfall NO_3-N fluxes (Figure 6e, $F = 10.21$, $p < 0.01$), throughfall TN fluxes ($F = 19.68$, $p < 0.01$), and throughfall total inorganic N fluxes (TIN, $F = 15.43$, $p < 0.001$), but not throughfall NH_4-N fluxes ($F = 2.934$, $p > 0.05$). There was no significant interaction between species and throughfall total TIN fluxes as a predictor of NO_3-N leaching ($F = 0.53$, $p > 0.05$).

Adding “Stage” as a predictor alongside throughfall TIN, where NO_3-N leaching flux was the outcome variable, improved the R^2 from 0.57 to 0.64, but the low number of replicates observed for the full year limits conclusions.

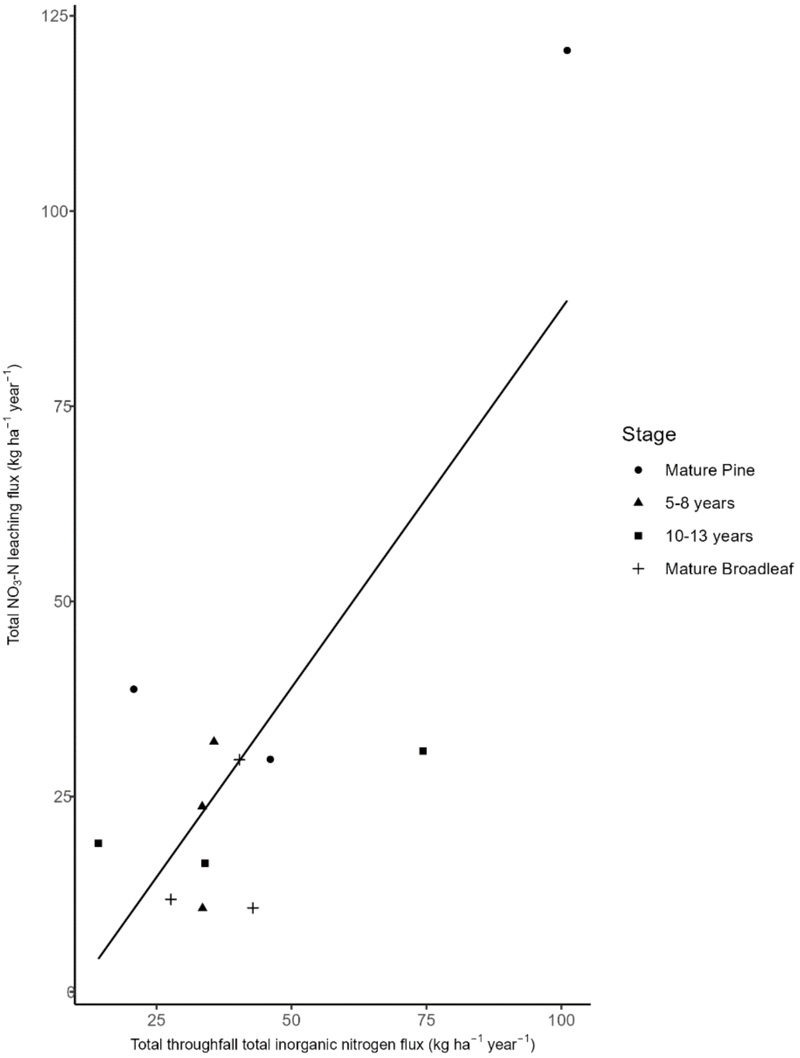


Figure 8: the relationship between total throughfall TIN flux and NO_3-N leaching flux between August 2021 and July 2022 at sites at different stages of the conifer-to-broadleaf conversion process.

4. Discussion

4.1. Soil C:N ratios, throughfall chemistry and deep soil NO_3-N leaching fluxes

Clear-cutting in coniferous plantations is often followed by a rapid increase in NO_3-N leaching fluxes within one year (Jerabkova et al., 2011). Three clearcut sites were included in the present study; the sites labelled as 0–2-year-old broadleaf sites were planted after a minimum of a two-year fallow period as this is the typical regime followed by local forestry practice. Whilst 0-2-year sites displayed elevated concentrations of NO_3-N in deep soil solution, it is possible that the present study did not capture the peak NO_3-N concentrations typically caused by felling. However, the focus here is on changes in soil nutrient cycling following the planting of broadleaves on coniferous rotations rather than the previously well-studied effects of clearfelling on NO_3-N leaching fluxes.

The first hypothesis stated that deep soil NO_3-N leaching fluxes will remain elevated in the first decade after planting broadleaves on former coniferous stands. The reduced soil C:N ratios at 0-10 cm depth in the planted broadleaved sites indicated high decomposition rates (Eastman et al., 2021), suggesting the release of nutrients from organic materials. However, no significant differences in NO_3-N leaching fluxes between the stages were observed despite a significant, positive relationship between NO_3-N leaching fluxes and soil C:N ratios at 0-10 cm depth. The significant relationships between throughfall DOC and NO_3-N fluxes, deep soil NO_3-N leaching fluxes and C:N ratios at 0-10 cm suggest that variation in throughfall chemistry between stands drove NO_3-N leaching fluxes rather than the direct effects of changes in forest management practices on soil processes. As throughfall chemistry was not significantly different, NO_3-N leaching fluxes were also not significantly different between stages.

Throughfall DOC and NO_3-N fluxes were positively related to NO_3-N leaching fluxes. Pitman *et al*. (2010) observed that deep soil NO_3-N concentrations reflected changes in throughfall DOC fluxes in a temperate Scots pine site on sandy soils. Whilst DOC in wet deposition can originate from many biogenic and anthropogenic sources (Zeng *et al*., 2023; Iavorivska *et al*., 2016), considering the location and ecosystem type studied here, the main sources were likely pollen and plant debris. Though, domestic sources and tyre and asphalt wear (Zeng *et al*., 2023; Iavorivska *et al*., 2016) may have influenced the conifer stand where the highest throughfall DOC was observed due to it being situated between a road that experiences heavy traffic and a domestic holding. Here, across the study area, DOC in throughfall was highest in September 2021 and began to increase again during the summer of 2022. Throughfall DOC fluxes have been related to total fruiting biomass in deciduous species (Verstraeten *et al*, 2018), and pollen production in forests across Europe (Verstraeten *et al*., 2023), aligning with the seasonality in throughfall DOC fluxes observed here. Microbial communities found on pollen from broadleaved deciduous species transform NO_3-N to NO_2-N (Verstraeten *et al*., 2023; Verstraeten *et al*., 2021), but NO_2-N in throughfall was negligible here (data not shown). Although throughfall samples here were kept cool and analysed within a week of collection, it is possible that any NO_2 oxidised into NO_3 by the time of analysis. Notably, the broadleaved sites included were still neighboured by mature coniferous stands in all cases, and some pollen inputs may, therefore, still be coniferous. The throughfall NO_3-N fluxes here may thus also be considered an indicator of the effect of pollen on throughfall and the positive relationship between throughfall NO_3-N flux and NO_3-N leaching fluxes, a product of the relationship between throughfall DOC flux and throughfall NO_3-N flux. Whilst elevated throughfall DOC and NO_3-N fluxes can also be attributed to caterpillar infestations of the forest canopy, the NO_3-N and DOC fluxes here were still lower than those reported under infested canopies by Pitman *et al*. (2010).

Solinger *et al*. (2001) found that throughfall DOC concentrations were related to forest floor and mineral soil DOC solutions. Whist soil solution DOC concentrations were not measured here, the soil C:N ratios measured can facilitate predictions of the fate of DOC once it has entered the soil via throughfall. Low C:N ratios indicate high nitrogen availability for plants, which can lead to lower allocation of carbon in roots and less available carbon for microbes (Kopáček *et al.*, 2013). At lower C:N ratios, carbon associated with soil organic matter is reassimilated by soil microbial communities, where it is either released as CO_2 or enters the stable carbon pool, reducing the flux of DOC in soil solution (Gmach *et al.*, 2018). The significant positive relationship between throughfall DOC flux and soil C:N ratio in the upper 0-10 cm of soil indicates a higher availability of carbon dissolved in soil solution with greater DOC inputs, which could leach to the lower soil layers. However, DOC can also bind to iron and clay-rich mineral soil particles, a process more likely to occur at a lower soil pH (Jagadamma *et al.*, 2012; Mayes *et al.*, 2012; Jardine *et al.*, 1989). Kammer *et al.* (2012) reported that most DOC entering the soil from the litter layer was retained in the top 5 cm by both physio-chemical and biological processes, and Sleutel *et al*. (2009) found that DOC fluxes in soil solution decreased with mineral soil depth due to adsorption processes. It is not possible to predict how much DOC would have been adsorbed to soil particles in the 0-10 cm layer as pH was not measured at this soil depth, though this may be a missing link to fully explain the relationship between throughfall DOC fluxes and NO_3-N leaching fluxes. It is hypothesised that an interaction between throughfall DOC inputs and the pH-dependent physio-chemical retention of DOC in the 0-10 cm layer determines carbon availability for soil microbial communities in the lower soil layers.

4.2. Net nitrification and mineralisation

The net nitrification values observed were within the range of those reported by Ambus *et al*. (2006), who employed an *in situ* incubation method in coniferous and broadleaved forests, verifying the experimental method used here. Soil moisture and pH were previously established as the most important controls on potential nitrification rate in soils at Thetford Forest (Thorpe, 2011). Still, soil moisture in July and August was not different between the chronosequence stages here. Here, in the 10-30 cm soil layer, sites with a higher soil pH at 10-30 cm depth had low or negative net nitrification values at the same depth, one explanation for which could be greater assimilation of NO_3-N, which could be supported by greater DOC availability in soil solution due to the higher pH. The negative and lower net nitrification values were also associated with higher C:N ratios in the same soil layer, indicating greater availability of DOC in soil solution and a low N environment with greater competition for nutrients like NO_3-N. DOC availability and pH have been positively related to soil respiration rate in mineral temperate forest soils (Klimek *et al*., 2020). Microbes utilise DOC in producing metabolites (Campbell *et al*., 2022), and the production of amino acids and enzymes, which require N (Liao *et al*., 2024; Zhao *et al*., 2021). Soils with high C:N ratios tend to have higher immobilisation rates (Bengtsson *et al*., 2003). Falkengren-Grerup *et al*. (1998) also observed a negative relationship between net nitrification and C:N ratio in soils below 5 cm depth, and Christenson *et al*. (2008) observed a negative relationship between soil C:N ratio and net nitrification in both organic and mineral soil horizons, in temperate forest soils. The low C:N ratios at higher nitrification rates could be related to higher rates of organic matter decomposition and release of N.

Some studies have shown the opposite effect of pH on nitrification, such as Ste-Marie & Pàre (1999), who observed increasing pH had a positive effect on nitrification, and DeForest & Otuya (2020), who found experimental liming of deciduous forest soils increased rate of nitrification. However, the pH effect on nitrification rates depends on the ammonia-oxidising microbial community structure. Nitrification is conducted by chemoautotrophic ammonia-oxidising bacteria and archaea, which oxidise ammonia to fix inorganic carbon (Norman *et al.*, 2015). Acid-adapted nitrifiers, particularly *Nitrosospira* spp., can dominate over the typical nitrifer, *Nitrosomonas* spp., at lower pH (Killham, 1994). Previous research at Thetford Forest identified both present in Corsican pine and beech stands, but *Nitrosospira* spp. were dominant (Thorpe, 2011).

Net nitrification in mature broadleaf forests and broadleaf stands planted 10-13 years ago had the lowest, often negative values of net nitrification. In addition to immobilisation, other possible explanations include the seasonality of nitrification or gaseous N_2O emissions from the soil. Peak nitrification may have occurred earlier or later in the year than observed. Trap *et al.* (2009) reported that heterotrophic bacteria were responsible for net nitrification in 40-75-year-old forests, and fungi were responsible for net nitrification in 100-year-old forests. Net nitrification was, however, dominated by autotrophic bacteria in young beech forests (Trap *et al.*, 2011). It is, therefore, possible that peak net nitrification is more likely to occur following litterfall in autumn when fungal activity is highest or before the growing season in the spring in mature broadleaf forests. Schütt *et al.* (2014) showed temperature dependency of net nitrification only in spruce mineral soils, not beech mineral soils. Thus, it would not necessarily be expected that peak net nitrification will occur during the hottest part of the year in the mature broadleaved stands measured here. Pure and mixed beech stands in Northern France displayed peak net nitrification rates in mineral topsoils during May and February, respectively (Aubert *et al*., 2005). NO_3-N may also have been released from the soil in gaseous form following nitrification. Nitrification has been established as a primary source N_2O and NO gaseous emissions from forest soils (Kester *et al*., 1997). The experimental method allowed the release of gaseous compounds from the incubation cylinders, which may account for the negative net nitrification values observed in mature and 10-13-year-old broadleaves. N_2O emissions from deciduous soils are generally higher than from coniferous soils (Ambus *et al*., 2006), with N_2O dominating over NO in beech forests (Schindlebacher *et al*., 2004). Schütt *et al*. (2014) observed positive net nitrification rates in winter months and negative net nitrification in spring-early summer during six-week *in situ* incubations of the A horizon of beech soils in Germany. The flux of gaseous N compounds is higher during summer; NO fluxes peaked in July in a spruce stand in Germany (Medinets *et al*., 2019). It is possible that net nitrification rates were high in mature broadleaved stands here, but the conversion of NO_3 and emission of N_2O and NO mean that only measuring the change in NO_3 did not capture actual nitrification rates.

4.3. NO_3-N leaching fluxes

Thetford Forest typically experiences dry conditions in the summer, resulting in periods of no NO_3-N leaching followed by pulses in NO_3-N leaching during the wetter months (Vanguelova et al., 2007). The observation period was relatively dry in eastern England (UKCEH *et al*., 2022); a more extended study period covering eventual wetter periods could capture greater peaks in NO_3-N leaching from the converted broadleaved stands. Marks *et al*. (2004) reported that once accounting for evapotranspiration, excess rainfall for the area is, on average 155 mm yr^{-1}. Modelling by Mansour & Hughes (2004) reported that groundwater recharge under chalk in East Anglia was, on average, 300 mm yr^{-1}. Considering the high transmissivity of chalk (Marks *et al.*, 2004b), soil drainage fluxes under open land would be expected to be near the groundwater recharge figure. Therefore, the calculated recharge rates of 126-194 mm during the study period for forested land are within a reasonable range. Interception by ground vegetation is unlikely to have contributed to variation in water fluxes between stands. In May and July 2021, observations suggested no significant differences between stages in percentage ground cover (data not shown).

Questions remain about the long-term impact of converting conifers to broadleaves, as the study only captured the first decade. Here, throughfall DOC and NO_3-N fluxes controlled NO_3-N leaching fluxes rather than forest management changes, but the causes of variation in throughfall DOC and NO_3-N across the study area cannot be explained. In an area with less variable throughfall DOC, it is possible that effects of forest management changes on NO_3-N leaching fluxes could emerge. Converted broadleaved stands older than 13 years were absent in the study area, as converting to broadleaves is a recent interest in the UK. Some studies on conifer clear-felling and replanting show peaks in NO_3-N leaching fluxes within one to three years, with NO_3-N leaching fluxes decreasing from four years post-felling (Berghom *et al.*, 2015; Hedwall *et al.*, 2013; Huber *et al.*, 2010; Mannerkoski *et al.*, 2005; Neal, 1998). Other studies show a slower response to clear-felling, with the peak observed 4-8 years later (Jerabkova *et al.*, 2011; Futter *et al.*, 2010; Ring, 1995). Chapter 2 and other studies (Emmett *et al.*, 1995) showed increasing NO_3-N leaching fluxes from conifer forests >10 years old and that the previous land use could affect the retention of NO_3-N inputs in forest soils. The long-term impact of conifer-to-broadleaf conversion requires ongoing monitoring.

4.4. Implications for water quality

NO_3-N leaching from agricultural land in the UK has previously been estimated as between 39-88 kg N ha^{-1} yr^{-1} (Silgram *et al.*, 2001). Here, whilst total nit NO_3-N rate leaching fluxes in the observation period were 13.06 kg ha^{-1} yr^{-1} from mature broadleaved stands and 42.14 kg ha^{-1} yr^{-1} for conifers, the peak in NO_3-N leaching fluxes that likely occurred following the disturbance of harvesting could not be observed using the chloride mass balance method as the scenario did not meet the assumptions of the chloride mass balance method. The three mature conifer sites studied for the whole year had total NO_3-N leaching fluxes of 60.29, 38.76 and 27.38 kg-N ha^{-1}, with the highest likely influenced by its position between a road that experiences high traffic volumes and a domestic holding. The three mature broadleaves had total NO_3-N leaching fluxes of 10.74, 11.84 and 29.73 kg-N ha^{-1}, with the highest likely influenced by its position at the edge of the forest next to an arable field, potentially subjected to fertiliser application, and the two sites with lower NO_3-N leaching fluxes situated deeper in the forest. The lack of a significant difference in NO_3-N between mature conifers and broadleaves was likely due to the category variation caused by proximity to pollution sources. All NO_3-N leaching fluxes from mature broadleaf sites remained below the UK range for agricultural land.

Whilst 42.14 kg ha^{-1} yr^{-1} is relatively small in comparison to NO_3-N leaching fluxes from agriculture, it is high in comparison to other European forest ecosystems, where it is uncommon for NO_3-N leaching fluxes to exceed 20 kg ha^{-1} yr^{-1} (Dise & Wright, 1995). In mature broadleaf forests, NO3-N concentrations in deep soil solution remained below the water framework directive threshold of 11 mg-N/L. Still, they were above the threshold in all other chronosequence stages, particularly in mature conifers (33.74 mg-N/L) and 10-13-year-old broadleaves (21.92 mg-N/L). The study area is surrounded by intensive livestock farming and crop production with heavy use of fertilisers, causing surface waters to have poor quality (Environment Agency, 2019). It is located in a Nitrate Vulnerable Zone (Environment Agency, 2024), and farmers are subject to regulations on using nitrogen fertilisers and storing organic manures (Department for Food and Rural Affairs, 2021).

Groundwater NO_3-N concentrations have been less studied in forestry operations than in soil fluxes. A study in the Afon Hafren catchment in Wales showed groundwater NO_3-N concentrations began to decrease three years after repeated major felling events ceased in the area (Neal *et al.*, 2004). Groundwater NO_3-N concentrations remained elevated seven years after clear-cutting in the Kangasvaara catchment but peaked three years after the same operations in the Iso-kauhea catchment, Finland (Mannerkoski *et al.*, 2005). The geological conceptualisation of the study area showed it is situated on the chalk aquifer, where the depth to groundwater is approximately 20-30 m. Substantial amounts of NO_3-N from past agricultural fertiliser use are stored in the UK's unsaturated zone (area between soil and the groundwater table), with 70 % residing in the chalk (Ascott *et al.*, 2016). Due to storage and travel through the unsaturated zone, the effect of forestry practices on NO_3-N leaching fluxes from the soil is unlikely to be observed in the groundwater for decades. However, the percentage of land covered under productive forestry in the Little Ouse and Thet catchment is low compared to agricultural land, and forestry is therefore unlikely to contribute significantly to the total N load to the unsaturated zone of the catchment.

5. Conclusion

NO_3-N leaching fluxes were studied in the context of converting coniferous plantations to broadleaved forests, an increasingly popular land-use change scenario in the UK. Observations provided little evidence of changes in biochemical processes in the first decade after conversion, with significant relationships between deep soil NO_3-N leaching fluxes, throughfall DOC and NO_3-N fluxes and soil C:N ratios at 0-10 cm depth suggesting external inputs drove NO_3-N leaching rather than forest management changes during the observation period. Further research on the conifer-to-broadleaf conversion scenario on water quality over a longer period, in different regional climates, and on various soil types is needed to fully understand the potential impact on water quality. Measuring parameters such as ground vegetation biomass, mycorrhizal communities, and isotope methods are needed to explain better the observations and movement of accumulated N in conifer plantations upon land-use change.

References

Aber, J. D., Nadelhoffer, K. J., Steudler, P., Melillo, J. M. (1989) Nitrogen Saturation in Northern Forest Ecosystems. *Bioscience,* 39, 378-386.

Adriaenssens, S., Staelens, J., Wuyts, K., Samson, R., Verheyen, K., Boeckx, P. (2012) Retention of Dissolved Inorganic Nitrogen by Foliage and Twigs of Four Temperate Tree Species. *Ecosystems*, 15, 1093–1107.

Ambus, P. (1998) Nitrous oxide production by denitrification and nitrification in temperate forest, grassland and agricultural soils. *European Journal of Soil Science*, 49, 495–502.

Ambus, P., Zechmeister-Boltenstern, S., Butterbach-Bahl, K. (2006) Sources of nitrous oxide emitted from European forest soils. Biogeosciences, 3, 135-145.

Ascott, M. J., Wang, L., Stuart, M. E., Ward, R. S., Hart, A. (2016) Quantification of nitrate storage in the vadose (unsaturated) zone: a missing component of terrestrial N budgets. *Hydrological Processes*, 30, 1903–1915.

Aubert, M., Bureau, F., Vinceslas-Akpa, M. (2005) Sources of spatial and temporal variability of inorganic nitrogen in pure and mixed deciduous temperate forests. *Soil Biology and Biochemistry*, 37, 67–79.

Beidler, K.V., Phillips, R.P., Andrews, E., Maillard, F., Mushinski, R.M., Kennedy, P.G. (2020) Substrate quality drives fungal necromass decay and decomposer community structure under contrasting vegetation types. *Journal of Ecology*, 108, 1845-1859.

Bengtsson, G., Bengtson, P., Månsson, K.F. (2003) Gross nitrogen mineralisation-, immobilisation-, and nitrification rates as a function of soil C/N ratio and microbial activity. *Soil biology and biochemistry*, 35, 143-154.

Berg, B., & Matzner, E. (1997) Effect of N deposition on decomposition of plant litter and soil organic matter in forest systems. *Reviews*, 5, 1–25.

Bergholm, J., Olsson, B. A., Vegerfors, B., Persson, T. (2015) Nitrogen fluxes after clear-cutting - Ground vegetation uptake and stump/root immobilisation reduce N leaching after experimental limiting, acidification and N fertilisation. *Forest Ecology and Management*, 342, 64–75.

Blom, J.M. (2009) *Sweet chestnut (Castanea sativa Mill.) Mycorrjizas: the composition of their communities over a wide geographical range in Europe*. Thesis: Bangor University.

Brockett, B. F. T., Prescott, C. E., Grayston, S. J. (2012) Soil moisture is the major factor influencing microbial community structure and enzyme activities across seven biogeoclimatic zones in western Canada. *Soil Biology and Biochemistry*, 44, 9–20.

Brzostek, E. R., Dragoni, D., Brown, Z. A., Phillips, R. P. (2015) Mycorrhizal type determines the magnitude and direction of root-induced changes in decomposition in a temperate forest. *New Phytologist*, 206, 1274–1282.

Buchmann', N., Gebauer, G., Schulze, E.-D. (1996) Partitioning of ^{15}N-labeled ammonium and nitrate among soil, litter, below- and above-ground, biomass of trees and understory in a 15-year-old *Picea abies* plantation. *Biogeochemistry*, 33, 1-23.

Calder, I., Reid, I., Nisbet, T., Armstrong, A., Green, J. (2002) *Study of the potential impacts on water resources of proposed afforestation*. England: Loughborough University.

Campbell, T.P., Ulrich, D.E.M., Toyoda, J., Thompson, J., Munsky, B., Albright, M.B.N., Bailey, V.L., Tfaily, M.M., Funbar, J. (2022) Microbial communities influence soil dissolved organic carbon concentration by altering metabolite composition. *Frontiers in Microbiology*, 12, 799014.

Carter, M., Gregorich, E.G. (2006) *Soil sampling and mtehods of analysis*. Boca Raton: CRC Press.

Christenson, L.M., Lovett, G.M., Weathers, K.C. (2008) The influence of tree species, nitrogen fertilisation and soil C to N ratio on gross soil nitrogen transformations. *Soil Science Society American Journal*, 73, 638-646.

Cools, N., De Vos, B. (2020) *Part X: Sampling and Analysis of Soil*. In UNECE ICP Forests Programme Co-ordinating Centre: Manual on methods and criteria for harmonised sampling, assessment, monitoring and analysis of the effects of air pollution on forests. Germany: Thünen Institute of Forest Ecosystems.

Cools, N., Vesterdal, L., De Vos, B., Vanguelova, E., Hansen, K. (2014) Tree species is the major factor explaining C: N ratios in European forest soils. *Forest Ecology and Management*, *311*, 3–16.

Crosbie, R.S., Peeters, L.J.M., Herron, N., McVicar, T.R., Herr, A. (2018) Estimating groundwater recharge and its associated uncertainty: Use of regression kriging and the chloride mass balance method. *Journal of Hydrology*, 561, 1063-1080.

Cruz-Paredes, C., Tájmel, D., & Rousk, J. (2023) Variation in Temperature Dependences across Europe Reveals the Climate Sensitivity of Soil Microbial Decomposers. *Applied and Environmental Microbiology*, 89, e0209022.

DeForest, J.L., Otuya, R.K. (2020) Soil nitrification increases with elevated phosphorus or soil pH in an acidic mixed mesophytic deciduous forest. *Soil Biology and Biochemistry*, 142, 107716.

Department for Food and Rural Affairs (2022) *Keepers of time: ancient and native woodland and trees policy in England Government's statement on England's ancient and native woodland and ancient and veteran trees*. Available: www.gov.uk/defra (Accessed December 2023)

Dirnböck, T., Pröll, G., Austnes, K., Beloica, J., Beudert, B., Canullo, R., De Marco, A., Fornasier, M. F., Futter, M., Goergen, K., Grandin, U., Holmberg, M., Lindroos, A. J., Mirtl, M., Neirynck, J., Pecka, T., Nieminen, T. M., Nordbakken, J. F., Posch, M., Forsius, M. (2018) Currently legislated decreases in nitrogen deposition will yield only limited plant species recovery in European forests. *Environmental Research Letters*, 13, 125010.

Dise, N.B., Rothwell, J.J., Gauci, V., van der Salm, C., de Vries, W. (2009) Predicting dissolved inorganic nitrogen leaching in European forests using two independent databases. *Science of the Total Environment*, 407, 1798-1808.

Dise, N.B., Wright, R.F. (1995) Nitrogen leaching from European forests in relation to nitrogen deposition. *Forest Ecology and Management*, 71, 153-161.

Eastman, B. A., Adams, M. B., Brzostek, E. R., Burnham, M. B., Carrara, J. E., Kelly, C., McNeil, B. E., Walter, C. A., Peterjohn, W. T. (2021) Altered plant carbon partitioning enhanced forest ecosystem carbon storage after 25 years of nitrogen additions. *New Phytologist*, 230, 1435–1448.

Emmett, B. A., & Reynolds, B. (1995) Factors influencing nitrogen saturation in sitka spruce stands in Wales, UK. *Water, Air and Soil Pollution*, 85, 1629-1634.

Emmett, B. A., Anderson, J. M., Hornung, M. (1991) The controls on dissolved nitrogen losses following two intensities of harvesting in a Sitka spruce forest (N. Wales). *Forest Ecology and Management*, 41, 65-80.

Emmett, B. A., Cosby, B. J., Ferrier, R. C., Jenkins, A., Tietema, A., Wright, R. F., Emmett, B. A., Cosby, B. J., Ferrier, R. C., Jenkins, A., Tietema, A., Wright, R. F. (1997) Modelling the ecosystem effects of nitrogen deposition: Simulation of nitrogen saturation at a Sitka spruce forest, Aber, Wales, UK. *Biogeochemistry*, 38, 129-148.

Environment Agency (2019) *Check for Drinking Water Safeguard Zones (SgZs) and Nitrate Vulnerable Zones (NVZs) map.* Available: https://environment.data.gov.uk/farmers/ (Accessed November 2023).

Environment Agency (2024) Nitrate vulnerable zones (NVZ) 2017- combined (Final designations). Available: https://www.data.gov.uk/dataset/e156a108-f8c2-498e-aa3f-11ff78f40eee/nitrate-vulnerable-zones-nvz-2017-combined-final-designations (Accessed March 2024)

Environmental Audit Committee (2023) *Seeing the wood for the trees: the contribution of the forestry and timber sectors to biodiversity and net zero goals*. Available: www.parliament.uk. (Accessed December 2023).

EPA (2000) *Guidance for Data Quality Assessment: Practical Methods for Data Analysis: EPA QA/G9: QA00 Update*. Washington: Office of Environmental Information.

Evans, J., & Carr, A. (2023) Predicting the current and future distribution of pine woodland specialist plants in the Cairngorms National Park. *British and Irish Botany*, 5, 180-195.

Falkengren-Grerup, U., Brunet, J., Diekmann, M. (1998) Nitrogen mineralisation in deciduous forest soils in south Sweden in gradietnts of soil acidity and deposition. *Environmental Pollution*, 102, 415-420.

Feng, J., Li, Z., Hao, Y., Wang, J., Ru, J., Song, J., & Wan, S. (2022) Litter removal exerts greater effects on soil microbial community than understory removal in a subtropical-warm temperate climate transitional forest. *Forest Ecology and Management*, 505, 119867.

Forest Research (2023) *Forestry statistics 2023*. Edinburgh: Forestry Commission.

Forest Research (2023) *The UK Forestry Standard The governments' approach to sustainable forest management*. Edinburgh: Forestry Commission.

Forestry Commission (1985) *Forestry commission 66th annual report and accounts 1985-86*. Edinburgh: Forestry Commission. (Accessed 20/01/2021)

Forestry Commission (2023). *Regional Woodland Restoration Innovation Fund.* Available: https://www.gov.uk/guidance/regional-woodland-restoration-innovation-fund (Accessed November 2023)

Fowler, D., Smith, R., Muller, J., Cape, J. N., Sutton, M., Erisman, J. W., Fagerli, H. (2007) Long term trends in sulphur and nitrogen deposition in Europe and the cause of non-linearities. *Water, Air, and Soil Pollution: Focus*, 7, 41–47.

Futter, M. N., Ring, E., Högbom, L., Entenmann, S., Bishop, K. H. (2010) Consequences of nitrate leaching following stem-only harvesting of Swedish forests are dependent on spatial scale. *Environmental Pollution*, 158, 3552–3559.

Gao, L., Cui, X., Hill, P. W., Guo, Y. (2020) Uptake of various nitrogen forms by co-existing plant species in temperate and cold-temperate forests in northeast China. *Applied Soil Ecology*, 147, 103398.

Gebauer, G., Melzer, A., Rehder, H. (1984) Nitrate content and nitrate reductase activity in Rumex obtusifolius L. I. Differences in organs and diurnal changes. *Oecologia*, 63, 136-142.

George, R.M.O., Hansson, L.J., Ring, E., Jansson, P.E., Gärdenäs, A.I. (2017) Nitrogen leaching following clear-cutting and soil scarification at a Scots pine – a modelling study of a fertilisation experiment. *Forest Ecology and Management*, 385, 281-294.

Gmach, M.R., Cherubin, M.R., Kaiser, K., Cerri, C.E.P. (2018) Processes that influence dissolved organic matter in the soil: a review. *Scientia Agricola*, 77, e20180164.

Goldberg, E., Kirby, K., Hall, J., Latham, J. (2007) The ancient woodland concept as a practical conservation tool in Great Britain. *Journal for Nature Conservation*, 15, 109–119.

Göttlein, A., Weis, W., Ettl, R., Raspe, S., Rothe, A. (2023). Long-term effects of clearcut and selective cut on seepage chemistry in a N-saturated spruce stand – Case study Höglwald. *Forest Ecology and Management*, 533, 120831.

Guan, H., Huston, J., Ding, Z., Love, A., Simmons, C.T., Deng, Z. (2013) Principal component analysis of watershed hydrochemical response to forest clearance and its usefulness for chloride mass balance applications. *Water resources research*, 49, 4362-4378.

Guan, H., Love, A. J., Simmons, C. T., Hutson, J., Ding, Z. (2010) Catchment conceptualisation for examining applicability of chloride mass balance method in an area with historical forest clearance. *Hydrology and Earth System Sciences*, 14, 1233–1245.

Guerrieri, R., Templer, P., & Magnani, F. (2021). Canopy Exchange and Modification of Nitrogen Fluxes in Forest Ecosystems. *Physiological Processes*, 7, 115-137.

Guerrieri, R., Vanguelova, E. I., Michalski, G., Heaton, T. H. E., Mencuccini, M. (2015) Isotopic evidence for the occurrence of biological nitrification and nitrogen deposition processing in forest canopies. *Global Change Biology*, 21, 4613–4626.

Gundersen, P. (1991) Nitrogen deposition and the forest nitrogen cycle: role of denitrification. In *Forest Ecology and Management*, 44, 15-28.

Gundersen, P. (1995). Nitrogen deposition and leaching in european forests-preliminary results from a data compilation. *Water, Air, and Soil Pollution*, 85, 1179-1184.

Gundersen, P., Schmidt, I.K., Raulund-Rasmussen, K. (2006) Leaching of nitrate from temperate forests - effects of air pollution and forest management. *Environmental Research*, 14, 1-57.

Gurmesa, G. A., Wang, A., Li, S., Peng, S., de Vries, W., Gundersen, P., Ciais, P., Phillips, O. L., Hobbie, E. A., Zhu, W., Nadelhoffer, K., Xi, Y., Bai, E., Sun, T., Chen, D., Zhou, W., Zhang, Y., Guo, Y., Zhu, J., Fang, Y. (2022) Retention of deposited ammonium and nitrate and its impact on the global forest carbon sink. *Nature Communications*, 13.

Hamer, U., Marschner, B. (2005) Priming effects in different soil types induced by fructose, alanine, oxalic acid and catechol additions. *Soil Biology and Biochemistry*, 37, 445-454.

Hartmann, A., Kobler, J., Kralik, M., Dirnböck, T., Humer, F., Weiler, M. (2016) Model-aided quantification of dissolved carbon and nitrogen release after windthrow disturbance in an Austrian karst system. *Biogeosciences*, 13, 159–174.

Hedwall, O., Grip, H., Linder, S., Lövdahl, L., Nilsson, U., & Bergh, J. (2013) Effects of clear-cutting and slash removal on soil water chemistry and forest-floor vegetation in a nutrient optimised Norway spruce stand. *Silva Fennica*, 47, 933.

Heklau, H., Schindler, N., Eisenhauer, N., Ferlian, O., Bruelheide, H. (2023) Temporal variation of mycorrhisation rates in a tree diversity experiment. *Ecology and Evolution*, 13, e10002.

Hollis, D., McCarthy, M., Kendon, M., Legg, T., Simpson, I. (2019) HadUK-Grid: A new UK dataset of gridded climate observations. *Geoscience Data Journal*, 6, 151-159.

Huber, C., Aherne, J., Weis, W., Farrell, E. P., Göttlein, A., Cummins, T. (2010) Ion concentrations and fluxes of seepage water before and after clear cutting of Norway spruce stands at Ballyhooly, Ireland, and Höglwald, Germany. *Biogeochemistry*, 101, 7–26.

Iavorivska, L., Boyer, E.W., DeWalle, D.R. (2016) Atmospheric deposition of organic carbon via precipitation. *Atmospheric Environment*, 146, 153-163.

Jagadamma, S., Mayes, M.A., Phillips, J.R. (2012) Selective sorption of dissolved organic carbon compounds by temperate soils. *PLOS ONE*, 7, e50434.

Jardine, P.M., McCarthy, J.F., Weber, N.L. (1989) Mechanisms of dissolved organic carbon adsorption on soil. *Soil Science Society of America Journal*, 53, 1378-1385.

Jerabkova, L., Prescott, C. E., Titus, B. D., Hope, G. D., Walters, M. B. (2011) A meta-analysis of the effects of Clearcut and variable-retention harvesting on soil nitrogen fluxes in boreal and temperate forests. *Canadian Journal of Forest Research*, 41, 1852–1870.

Jonard, M., Fürst, A., Verstraeten, A., Thimonier, A., Timmermann, V., Potočić, N., Waldner, P., Benham, S., Hansen, K., Merilä, P., Ponette, Q., de la Cruz, A. C., Roskams, P., Nicolas, M., Croisé, L., Ingerslev, M., Matteucci, G., Decinti, B., Bascietto, M., Rautio, P. (2015) Tree mineral nutrition is deteriorating in Europe. *Global Change Biology*, 21, 418–430.

Kammer, A., Schmidt, M. W. I., Hagedorn, F. (2012) Decomposition pathways of 13C-depleted leaf litter in forest soils of the Swiss Jura. *Biogeochemistry*, *108*, 395–411.

Kenk, G., Guehne, S. (2001) Management of transformation in central Europe. *Forest ecology and management*, 151, 107-119.

Kester, R.A., Meijer, M.E., Libochant, J.A., Boer, W.D., Laanbroek, H.J. (1997) Contribution of nitrification and denitrification to the NO and N_2O emissions of an acid forest soil, a river sediment and a fertilised grassland soil. *Soil Biology and Biochemistry*, 29, 1655-1664.

Killham, K. (1994) *Soil Ecology*. Cambridge: Cambridge University Press.

Kilpeläinen, J., Aphalo, P. J., Barbero-López, A., Adamczyk, B., Nipu, S. A., Lehto, T. (2021) Are arbuscular-mycorrhizal alnus incana seedlings more resistant to drought than ectomycorrhizal and nonmycorrhizal ones? *Tree Physiology*, 40, 782–795.

Klimek, B., Chodak, M., Niklińska, M. (2021) Soil respiration in seven types of temperate forests exhibits similar temperature sensitivity. *Journal of Soils and Sediments*, 21, 338-345.

Kopáček, J., Cosby, B.J., Evans, C.D., Hruška, J., Moldan, F., Oulehle, F., Šantrůčková, H., Tahovská, K., Wright, R.F. (2013) Nitrogen, organic carbon and sulphur cycling in terrestrial ecosystems: linking nitrogen saturation to carbon limitation of soil microbial processes. *Biogeochemistry*, 115, 33-51.

Kubin, E., Křeček, J., Palán, L. (2017) Effects of Forest Practices on Water Resources Recharge in the Boreal Climate. *Environmental Processes*, 4, 509–522.

Latham, R., Hall, J., Perry, S., Goldberg, E., Reid, C. (2018) *Guidelines for the Selection of Biological SSSIs Part 2: Detailed Guidelines for Habitats and Species Groups Chapter 2a Woodlands, Wood Pasture and Parkland, and Veteran Trees*. Peterborough: Joint Nature conservation Committee.

Liao, C., Bao, Y., Cheng, X. (2024) Shifts in microbial mechanism regulate carbon priming effect under two simulated root exudate and nitrogen addition in coniferous and broad-leaved forest soils. *Plant and Soil*.

Liese, R., Lübbe, T., Albers, N. W., Meier, I. C. (2018) The mycorrhizal type governs root exudation and nitrogen uptake of temperate tree species. *Tree Physiology*, 38, 83–95.

Ma, X., Zhu, B., Nie, Y., Liu, Y., Kuzyakov, Y. (2021) Root and mycorrhizal strategies for nutrient acquisition in forests under nitrogen deposition: A meta-analysis. *Soil Biology and Biochemistry*, 163, 108418.

Macdonald, E., Gardiner, B., Mason, W. (2010) The effects of transformation of even-aged stands to continuous cover forestry on conifer log quality and wood properties in the UK. *Forestry*, 83, 1–16

Maillard, F., Schilling, J., Andrews, E., Schreiner, K.M., Kennedy, P. (2020) Functional convergence in the decomposition of fungal necromass in soil and wood. *FEMS Microbiology Ecology*, 96, fiz209.

Mannerkoski, H., Finér, L., Piirainen, S., Starr, M. (2005) Effect of clear-cutting and site preparation on the level and quality of groundwater in some headwater catchments in eastern Finland. *Forest Ecology and Management*, 220, 107–117.

Mansour, M.M., Hughes, A.G. (2004) *Application of numerical modelling to investigate recharge to the chalk aquifer beneath thick till deposits in East Anglia*. Keyworth: British Geological Survey.

Manzoor, A.S., Griffiths, G., Rose, D. C., Lukac, M. (2021) The Return of Wooded Landscapes in Wales: An Exploration of Possible Post-Brexit Futures. *Land*, 10, 59.

Manzoor, S. A., Griffiths, G., Latham, J., Lukac, M. (2019) Scenario-led modelling of broadleaf forest expansion in Wales. *Royal Society Open Science*, 6, 190026.

Marks, R.J., Lawrence, A.R., Humpage, A.J., Hargreaves, R. (2004) *Recharge through till: developing a methodology for estimating groundwater recharge with examples from two case studies in East Anglia*. Keyworth: British Geological Survey.

Marks, R.J., Lawrence, A.R., Whitehead, E.J., Cobbing, J.E., Mansour, M.M., Darling, W.G., Hughes, A.G. (2004b) *Chalk recharge beneath thick till deposits in East Anglia*. Keyworth: British Geological Survey.

Marozas, V., Sasnauskienė, J., Dautartė, A., Gavenauskas, A., Sabienė, N., Armolaitis, K. (2018) Effect of shelterwood cuttings on soil chemical properties in scots pine (*Pinus sylvestris* L.) forests in Europe’s hemiboreal zone, in Lithuania. *Journal of Elementology*, 23, 353–367

Mason, B. (2020) Encouraging greater use of continuous cover forestry. *Quarterly Journal of Forestry*, 115, 46-54.

Mason, W. L. (2007). Changes in the management of British forests between 1945 and 2000 and possible future trends. *Ibis*, 149, 41–52.

Mayes, M.A., Heal, K.R., Brandt, C.C., Phillips, J.R., Jardine, P.M. (2012) Relation between soil order and sorpstion of dissolved organic carbon in temperate subsoils. *Soil Science Society of America Journal*, 76, 1027-1037.

Medinets, S., Gasche, R., Kiese, R., Rennenberg, H., Butterbach-Bahl, K. (2019) Seasonal dynamics and profiles of soil NO concentrations in a temperate forest. *Plant and Soil*, 445, 335–348.

Mellert, K. H., & Göttlein, A. (2012) Comparison of new foliar nutrient thresholds derived from van den Burg’s literature compilation with established central European references. *European Journal of Forest Research*, 131, 1461–1472.

Midgley, M.G., Phillips, R.P. (2014) Mycorrhizal associations of dominant trees influence nitrate leaching responses to N deposition. *Biogeochemistry*, 117, 241-253.

Mupepele, A. C., & Dormann, C. F. (2017) Influence of forest harvest on nitrate concentration in temperate streams-a meta-analysis. *Forests*, 8, 5.

Neal, C., Reynolds, B., Neal, M., Wickham, H., Hill, L., Williams, B. (2004) The impact of conifer harvesting on stream water quality: the Afon Hafren, mid-Wales. *Hydrology and Earth System Sciences*, 8, 503-520.

Neal, C., Reynolds, B., Wilkinson, J., Hill, T., Neal, M., Hill, S., Harrow, M. (1998) The impacts of conifer harvesting on runoff water quality: a regional survey for Wales. *Hydrological Earth Systems Science*, 2, 323-344.

Nilsson, L. O., Bååth, E., Falkengren-Grerup, U., & Wallander, H. (2007) Growth of ectomycorrhizal mycelia and composition of soil microbial communities in oak forest soils along a nitrogen deposition gradient. *Oecologia*, 153, 375–384.

Norman, J. S., Lin, L., Barrett, J. E. (2015) Paired carbon and nitrogen metabolism by ammonia-oxidising bacteria and archaea in temperate forest soils. *Ecosphere*, 6.

Olsson, B.A., Bergholm, J., Alavi, G., Persson, T. (2022) Effects of long-term N fertilisation on nitrate leaching and vegetation responses in a spruce stand after severe wind damage. *Forest Ecology and Management*, 520, 120422.

Olsson, M. O., & Falkengren-Grerup, U. (2003) Partitioning of nitrate uptake between trees and understory in oak forests. *Forest Ecology and Management*, 179, 311–320.

Palviainen, M., Finér, L., Laiho, R., Shorohova, E., Kapitsa, E., Vanha-Majamaa, I. (2010) Phosphorus and base cation accumulation and release patterns in decomposing Scots pine, Norway spruce and silver birch stumps. *Forest Ecology and Management*, 260, 1478–1489.

Pei, G., Liu, J., Peng, B., Gao, D., Wang, C., Dai, W., Jiang, P., Bai, E. (2019) Nitrogen, lignin, C/N as important regulators of gross nitrogen release and immobilisation during litter decomposition in a temperate forest ecosystem. *Forest Ecology and Management*, 440, 61-69.

Pickles, B. J., Genney, D. R., Potts, J. M., Lennon, J. J., Anderson, I. C., Alexander, I. J. (2010) Spatial and temporal ecology of Scots pine ectomycorrhizas. *New Phytologist*, 186, 755–768.

Piirainen, S., Finér, L., Mannerkoski, H., Starr, M. (2007) Carbon, nitrogen and phosphorus leaching after site preparation at a boreal forest clear-cut area. *Forest Ecology and Management*, 243, 10–18.

Pitman, R.M., Vanguelova, E.I., Benham, S.E. (2010) The effects of phytophagus insects on water and soil nutrient concentrations and fluxes through forest stands of the Level II monitoring network in the UK. *Science of the Total Environment*, 409, 169-181.

Pitman, R. M., Vanguelova, E. I., Benham, S. (2023) Soil change and broadleaf tree growth 10 years after wood ash and brash co-application to a clearfelled lowland conifer site in Britain. *Forestry*, 97, 76-93.

Prescott, C. E., Grayston, S. J. (2013) Tree species influence on microbial communities in litter and soil: Current knowledge and research needs. *Forest Ecology and Management*, *309*, 19–27.

Raum, S. (2020) Land-use legacies of twentieth-century forestry in the UK: a perspective. *Landscape Ecology*, 35, 2713–2722.

Raum, S., Potter, C. (2015) Forestry paradigms and policy change: The evolution of forestry policy in Britain in relation to the ecosystem approach. *Land Use Policy*, *49*, 462–470.

Reuter, R., Ferlian, O., Tarkka, M., Eisenhauer, N., Pritsch, K., Simon, J. (2021) Tree species rather than type of mycorrhizal association drive inorganic and organic nitrogen acquisition in tree-tree interactions. *Tree Physiology*, 41, 2096–2108.

Ribbons, R. R., Kepfer-Rojas, S., Kosawang, C., Hansen, O. K., Ambus, P., McDonald, M., Grayston, S. J., Prescott, C. E., Vesterdal, L. (2018) Context-dependent tree species effects on soil nitrogen transformations and related microbial functional genes. *Biogeochemistry*, 140, 145–160.

Ring, E. (1995) Nitrogen leaching before and after clear-felling of fertilised experimental plots in a *Pinus sylvestris* stand in central Sweden. *Management Forest Ecology and Management*, 72, 151-166.

Ring, E., Högbom, L., Nohrstedt, H. O. (2001) Effects of brash removal after clear felling on soil and soil-solution chemistry and field-layer biomass in an experimental nitrogen gradient. *The Scientific World Journal*, 1, 457–466.

Rothe, A., Huber, C., Kreutzer, K., Weis, W. (2002) Deposition and soil leaching in stands of Norway spruce and European beech: Results from the Hoglwald research in comparison with other European case studies. *Plant and Soil*, 240, 33-45.

Rowland, C.S., Morton, R.D., Carrasco, L., McShane, G., O'Neil, A.W., Wood, C.M. (2017) *Land Cover Map 2015* (25m raster, GB). NERC Environmental Information Data Centre (Accessed July 2021)

Rusterholz, H. P., Studer, M., Zwahlen, V., Baur, B. (2020) Plant-mycorrhiza association in urban forests: Effects of the degree of urbanisation and forest size on the performance of sycamore (*Acer pseudoplatanus*) saplings. *Urban Forestry and Urban Greening*, *56*, 126872.

Schindlbacher, A., Rodler, A., Kuffner, M., Kitzler, B., Sessitsch, A., Zechmeister-Boltenstern, S. (2011) Experimental warming effects on the microbial community of a temperate mountain forest soil. *Soil Biology and Biochemistry*, 43, 1417–1425.

Schindlbacher, A., Zechmeister-Boltenstern, S., Butterbach-Bahl, K. (2004) Effects of soil moisture and temperature on NO, NO2, and N 2O emissions from European forest soils. *Journal of Geophysical Research*, 109, D17302.

Schleppi, P., Curtaz, F., Krause, K. (2017) Nitrate leaching from a sub-alpine coniferous forest subjected to experimentally increased N deposition for 20 years, and effects of tree girdling and felling. *Biogeochemistry*, 134, 319–335.

Schrijver, A.D., Geudens, G., Augusto, L., Staelens, J., Mertens, J., Wuyts, K., Gielis, L., Verheyen, K. (2007) The effect of forest type on throughfall deposition and seepage flux: A review. In *Oecologia*, 153, 663–674.

Schrijver, A.D., Geudens, G., Augusto, L., Staelens, J., Mertens, J., Wuyts, K., Gielis, L., Verheyen, K. (2007) The effect of forest type on throughfall deposition and seepage flux: a review. *Oecologia*, 153, 663-674.

Schuaub, M., Vesterdal, Vos, D.B., Ukonmaanaho, L., Fleck, S., Schwärzel, K., Ferretti, M. (2021). *Forest Monitoring to assess Forest Functioning under Air Pollution and Climate Change*. Switzerland: Swiss Federal Research Institute WSL.

Schulz, H., Härtling, S., Stange, C. F. (2011) Species-specific differences in nitrogen uptake and utilisation by six European tree species. *Journal of Plant Nutrition and Soil Science*, 174, 28–37.

Schütt, M., Borken, W., Stange, C. F., Matzner, E. (2014) Substantial net N mineralisation during the dormant season in temperate forest soils. *Journal of Plant Nutrition and Soil Science*, 177, 566–572.

Shah, N.W., Baillie, B.R., Bishop, K., Ferraz, S., Högbom, L., Nettles, J. (2022) The effects of forest management on water quality. *Forest Ecology and Management*, 522, 120397.

Silgram, M., Waring, R., Anthony, S., Webb, J. (2001). Intercomparison of national & IPCC methods for estimating N loss from agricultural land. *Nutrient Cycling in Agroecosystems*, 60, 189-195.

Simon, J. (2023) Relevance of organic vs. inorganic nitrogen in intra- and interspecific competition of seven central European tree species. *Trees - Structure and Function*, 37, 1583–1591.

Sleutel, S., Vandenbruwane, J., De Schrijver, A., Wuyts, K., Moeskops, B., Verheyen, K., De Neve, D. (2009) Patterns of dissolved organic carbon and nitrogen fluxes in deciduous and coniverous forests under historic high nitrogen deposition. *Biogeosciences*, 6, 2743-2758.

Solinger, S., Kalbitz, K., Matzner, E. (2001) Controls on the dynamics of dissolved organic carbon and nitrogen in a Central European deciduous forest. *Biogeochemistry*, 55, 327-349.

Ste-Marie, C., Pàre, D. (1999) Soil, pH and N availability effects on net nitrification in the forest floors of a range of boreal forest stands. *Soil Biology and Biochemistry*, 31, 1579-1589.

Stevens, P.A., Payne, R.J., Kimberley, A., Smart, S.M. (2016) How will the semi-natural vegetation of the UK have changed by 2030 given likely changes in nitrogen deposition? *Environmental Pollution*, 208, 879-889.

Survila, G., Varnagirytė-Kabašinskienė, I., Armolaitis, K. (2021) Deep soil ploughing for afforestation: a review of potential impacts on soil and vegetation. *Baltic Forestry*, 27, 590.

Svensson, T., Lovett, G. M., Likens, G. E. (2012) Is chloride a conservative ion in forest ecosystems? *Biogeochemistry*, 107, 125–134.

Tew, E.R., Vanguelova, E.I., Sutherland, W.J. (2021) Alternative afforestation options on sandy heathland result in minimal long-term changes in mineral soil layers. *Forest Ecology and Management*, 483, 118906.

Thorpe, E. L. (2011) *Impact of nitrogen deposition on methane-and ammonia-oxidising microbial communities in forest soils*. Thesis: University of Essex.

Tian, K., Kong, X., Yuan, L., Lin, H., He, Z., Yao, B., Ji, Y., Yang, J., Sun, S., Tian, X. (2019) Priming effect of litter mineralisation: the role of root exudate depends on its interactions with litter quality and soil condition. *Plant and Soil*, 440, 457–471.

Trap, J., Bureau, F., Akpa-Vinceslas, M., Decaens, T., Aubert, M. (2011) Changes in humus forms and soil N pathways along a 130-year-old pure beech forest chronosequence. *Annals of Forest Science*, 68, 595–606.

Trap, J., Bureau, F., Vinceslas-Akpa, M., Chevalier, R., Aubert, M. (2009) Changes in soil N mineralisation and nitrification pathways along a mixed forest chronosequence. *Forest Ecology and Management*, 258, 1284-1292.

UKCEH (2022) *Hydrological Summary for the United Kingdom*. Available: www.hydoutuk.net/latest-outlook/ (Accessed December 2023)

Vanguelova, E. I., Pitman, R. M. (2019) Nutrient and carbon cycling along nitrogen deposition gradients in broadleaf and conifer forest stands in the east of England. *Forest Ecology and Management*, 447, 180–194.

Vanguelova, E., Barsoum, N., Benham, S., Broadmeadow, M., Moffat, A. (2007) *Ten years of intensive environmental monitoring in British forests*. Information note FCIN088, UK: Forestry commission.

Vanguelova, E.I., Benham, S., Pitman, R., Moffat, A.J., Broadmeadow, M., Nisbet, T., Durrant, D., Barsoum, N., Wilkinson, M., Bochereau, F., Hutchings, T., Broadmeadow, S., Crow, P., Taylor, P., Houston, T.D. (2010) Chemical fluxes in time through forest ecosystems in the UK – Soil response to pollution recovery. Environmental pollution, 158, 1857-1869.

Vanguelova, E.I., Nortcliff, S., Moffat, A.J., Kennedy, F. (2005) Morphology, biomass and nutrient status of fine roots of Scots pine (Pinus sylvestris) as influenced by seasonal fluctuations in soil moisture and soil. Plant and soil, 270, 233-247.

Vanguelova, E.I., R.P., Benham, S. (2024) *Responses of forest ecosystems to nitrogen deposition in the United Kingdom*. In: Atmospheric nitrogen deposition to global forests. The United States: Academic Press.

Veerman, L., Kalbitz, K., Gundersen, P., Kjønaas, J., Moldan, F., Schleppi, P., van Loon, E. E., Schoorl, J., Wessel, W., Tietema, A. (2020) The long-term fate of deposited nitrogen in temperate forest soils. *Biogeochemistry*, 150, 1–15.

Verstraeten, A., Gottardini, E., Vanguelova, E., Waldner, P., Bruffaerts, N., Nussbaumer, A., Neumann, M., Clarke, N., Hansen, K., Rautio, P., Ukonmaanaho, L. (2018) Establishing a link between pollen dispersal, seed production and throughfall dissolved organic carbon

(DOC) flux in temperate forests. In: 7th ICP Forests Scientific Conference: European forests in a changing environment – air pollution, climate change and forest management, Riga, Latvia, 21-23 May 2018. Accessed: https://sc2018.thuenen.de/fileadmin/sc2018/17_Verstraeten_ICP_Forests_SC2018.pdf

Verstraeten, A., Bruffaerts, N., Cristofolini, F., Vanguelova, E., Neirynck, J., Genouw, G., De Vos, B., Waldner, P., Thimonier, A., Nussbaumer, A., Neumann, M., Benham, S., Rautio, P., Ukonmaanaho, L., Merilä, P., Lindroos, A. J., Saarto, A., Reiniharju, J., Clarke, N., Gottardini, E. (2023) Effects of tree pollen on throughfall element fluxes in European forests. *Biogeochemistry*, 165, 311-325.

Verstraeten, A., Gottardini, E., Bruffaerts, N., Neirynck, J., Genouw, G. (2021). In Vitro Nitrogen Transformations by Pollen From Temperate Tree Species. *Research Square*, preprint.

Waldner, P., Thimonier, A., Graf Pannatier, E., Etzold, S., Schmitt, M., Marchetto, A., Rautio, P., Derome, K., Nieminen, T. M., Nevalainen, S., Lindroos, A. J., Merilä, P., Kindermann, G., Neumann, M., Cools, N., de Vos, B., Roskams, P., Verstraeten, A., Hansen, K., Minaya, M. (2015) Exceedance of critical loads and of critical limits impacts tree nutrition across Europe. *Annals of Forest Science*, 72, 929–939.

Wang, Q., Xiao, J., Ding, J., Zou, T., Zhang, Z., Liu, Q., Yin, H. (2021) Differences in root exudate inputs and rhizosphere effects on soil N transformation between deciduous and evergreen trees. *Plant and Soil*, 458, 277–289.

Webster, K.I., Leach, J.A., Hazlett, P.W., Buttle, J.M., Emilson, E.J.S., Creed, I.F. (2022) Long-term stream chemistry response to harvesting in a northern hardwood forest watershed experiencing environmental change. *Forest Ecology and Management*, 519, 120345.

Weis, W., Rotter, V., Göttlein, A. (2006) Water and element fluxes during the regeneration of Norway spruce with European beech: Effects of shelterwood-cut and clear-cut. *Forest Ecology and Management*, 224, 304–317.

Wright, R. F., Alewell, C., Cullen, J. M., Evans, C. D., Marchetto, A., Moldan, F., Prechtel, A., Rogora, M. (2001) Trends in nitrogen deposition and leaching in acid-sensitive streams in Europe. *Hydrology and Earth System Sciences*, 5, 299-310.

Xu, S., Song, X., Zeng, H., Wang, J. (2024) Soil microbial necromass carbon in forests: A global synthesis of patterns and controlling factors. *Soil Ecology Letters*, 6.

Yuan, Y., Zhao, W., Zhang, Z., Xiao, J., Li, D., Liu, Q., Yin, H. (2018) Impacts of oxalic acid and glucose additions on N transformation in microcosms via artificial roots. *Soil Biology and Biochemistry*, 121, 16–23.

Zechmeister-boltenstern, S., Michel, K., Pfeffer, M. (2011) Soil microbial community structure in European forests in relation to forest type and atmospheric nitrogen deposition. *Plant and Soil*, 343, 37-50.

Zeng, J., Han, G., Zhang, S., Xiao, X., Li, Y., Gao, X., Wang, D., Qu, R. (2023) Response of dissolved organic carbon in rainwater during extreme rainfall period in megacity: Status, potential source, and deposition flux. *Sustainable Cities and Society*, 88, 104299.

Zhang, P., Tian, X., He, X., Song, F., Ren, L., Jiang, P. (2008) Effect of litter quality on its decomposition in broadleaf and coniferous forest. *European Journal of Soil Biology*, 44, 392–399.

Zhang, W., Yang, K., Lyu, Z., Zhu, J. (2019) Microbial groups and their functions control the decomposition of coniferous litter: A comparison with broadleaved tree litters. *Soil Biology and Biochemistry,* 133*,* 196-207.

Zhao, C., He, X., Dan, X., Zhao, J., Huang, X., Cai, Z., Meng, H., Zhang, J. (2021) Specific dissolved organic matter components drive the assembly of a core microbial community in acidic soil of ammonium-preferring plants. *Catena*, 207.

Zhu, X., Zhang, W., Chen, H., Mo, J. (2015) Impacts of nitrogen deposition on soil nitrogen cycle in forest ecosystems: A review. *Acta Ecologica Sinica*, 35, 35–43.

Chapter 4

A Biome-BGCMuSo framework for simulating nitrate leaching fluxes from forest ecosystems

Abstract

In addition to experimental and observational field studies, biochemical and hydrological models facilitate the long-term study of land management changes on nutrient and water cycling processes. Coupled nutrient-hydrological models enable the simulation of NO_3-N fluxes, but such models are complex, requiring parameterisation of both the nutrient and hydrological components. A multi-layer biogeochemical model was applied to model NO_3-N fluxes from an N-saturated ecosystem.

Two parameterisation procedures were used to simulate deep soil NO_3-N fluxes from an N-saturated temperate Scots pine forest in Biome-BGCMuSo. First, parameterising just the ecophysiological input file did not generate a parameter set with close alignment to observed values. Second, parameterisation of the ecophysiological and soil input files initially appeared to generate a parameter set closer to observed values when aggregated on an annual timestep ($R^2 = 0.32$). However, upon further interrogation, it was found through the hydrological and respiration outputs that the best-performing parameter set led to the model simulating tree mortality events during the study period, which did not happen in reality. Four parameters were identified as those which exerted the greatest control on NO_3-N leaching fluxes; leaf

C:N, root distribution, canopy light extinction coefficient and the fraction of N in rubisco.

From the trial-and-error process, a framework is developed to guide future Biome-BGCMuSo users on quality-checking model outputs. As the four parameters control the key plant physiological processes that feed into the hydrological and soil components of the model, the framework suggests performing model parameterisation using respiration-related variables even when the desired outcome is NO_3-N leaching fluxes. The framework further suggests using respiration and hydrological outputs as a check against unexpected mortality and advises against using the model for scenarios without 10+ years of observed data for more multiple variables.

1. Introduction

Prolonged periods of elevated N deposition have resulted in N-saturation in temperate forest soils (Schmitz *et al.*, 2024). Nitrogen saturation leads to elevated levels of deep soil nitrate (NO_3-N) leaching, causing eutrophication and acidification in surface and groundwaters, negatively impacting aquatic biodiversity and drinking water quality (Xie *et al.*, 2024; Gundersen *et al.*, 2006). In the latter part of the 20th century, NOx species dominated N emissions, typically from industrial activities and traffic (RoTAP, 2012). Whilst NO_x emissions have declined since then, NH_3 emissions have become dominant, particularly in areas close to intensive livestock farms (Vanguelova *et al.*, 2024; Forsius *et al.*, 2021; Carnell *et al.*, 2018).

Analyses of European-scale datasets found that the wet deposition of total inorganic nitrogen (TIN) was the best predictor of NO_3-N leaching in forest ecosystems, with empirical models performing well in predicting NO_3-N leaching from throughfall TIN (Dise *et al*., 2009; van der Salm *et al*., 2007; MacDonald *et al*., 2004). In N-limited forest ecosystems, increasing TIN deposition may initially positively affect forest growth (Etzold *et al*., 2020), enabling N retention by the forest ecosystem. However, prolonged periods of elevated TIN deposition can reduce tree growth and nutrient imbalances, reducing resilience against pests and diseases (Etzold *et al*., 2020).

Elevated NO_3-N leaching in forests is commonly observed after disturbance events, including harvesting (Jerabkova *et al*., 2011), site preparation and planting (Mannerkoski *et al*., 2005; Kubin, 1998), insect outbreaks (Pitman *et al*., 2010), and storm damage (Olsson *et al*., 2022). The short-term effects of disturbance have been observed in many studies, but long-term effects on land use and tree species change are rare. Recent findings suggested that former land use affects the capacity of forest soils to retain N inputs (Chapter 2), possibly due to former land use impacts on soil nutrient availability. Complex physical process-based biochemical models can be applied to investigate the impact of changes in forest management over several decades, but coupling with hydrological models is required to investigate NO_3-N leaching, which is both a hydrological and biochemical process. Whilst in mature forests, throughfall TIN is the key driver of NO_3-N leaching, hydrological changes have been connected to changes in NO_3-N leaching during the first few decades of forest growth (Rosenqvist *et al*., 2010; Hansen *et al*., 2007). Rewetting after prolonged drought periods stimulates pulses of NO_3-N leaching fluxes (e.g. Leitner *et al*., 2017).

Thus, modelling NO_3-N leaching fluxes requires a model capturing biogeochemical and hydrological cycling.

Many models capable of simulating NO_3-N leaching exist and have commonly been applied in the context of agricultural land (Bell *et al.*, 2021; Cammarano *et al.*, 2021; de Filippis *et al.*, 2020). Models developed in the field of hydrology, such as INCA-N (Wade *et al.*, 2002) and SWAT (Laurent & Ruelland, 2011; Arnold *et al.*, 1998), were produced to simulate outputs at a catchment scale and closely tie the modelling of nutrient flows through a system with hydrological fluxes. However, INCA-N and SWAT include only basic modelling of soil nutrient cycling processes. For example, INCA-N assumes that organic matter availability does not limit mineralisation (Whitehead *et al.*, 1998) and does not account for interactions between the nitrogen cycle and the carbon cycle (Futter *et al.*, 2010). The INCA model suite includes some simplified modelling of groundwater flow and transport. However, unless coupled with a more detailed nutrient model (e.g. Pohlert *et al.*, 2007), such hydrological models may not capture the nutrient changes associated with changing forest management.

Biochemical models developed in soil science capture more of the complexity in soil nutrient cycling but with less development of or no inclusion of the hydrological component. The Model of Acidity and Dissolved Organic Carbon (MADOC) combines models developed to model soil acidification, carbon cycling and nitrogen dynamics between soil and vegetation (Rowe et al., 2015; Posch and Reinds, 2009; Michalzik, et al., 2003). MADOC has been applied to model the effect of declining acid deposition on dissolved organic carbon concentrations in soil water (Sawicka *et al.*, 2017). However, MADOC does not include a representation of hydrological processes (Rowe *et al.*, 2014). The Model of Ecosystem Retention and Loss of Inorganic Nitrogen (MERLIN) model is a mass balance model that simulates nitrogen dynamics within an ecosystem (Tietema *et al.*, 2002), containing detailed modelling of plant and microbial dynamics (Nieder *et al.*, 2011; Cosby *et al.*, 1997). However, seasonal variation is not captured using the MERLIN model as simulations are performed on an annual time step (Futter et al., 2010). Models capturing both hydrological and biochemical processes include the Denitrification Decomposition (DNDC) and Biome-biogeochemical multi-soil-layer (Biome-BGCMuSo) models, but each considers biochemical processes differently. For example, when using the DNDC model, users can specify tree nitrogen uptake by setting the rate of ammonium and nitrate uptake (Norman *et al.*, 2008), but Biome-BGCMuSo calculates N uptake based on foliar N content (Running & Gower, 1991). To our knowledge, the level of detail captured in Biome-BGCMuSo and DNDC in both the biochemical and hydrological components is not captured elsewhere, but the detail can make such models difficult to parameterise.

Building on previous exploration of changes in soil NO_3-N leaching in the first decade years following conifer-to-broadleaf conversion (Chapter 3), the parameterisation of a biochemical model here would allow its later application to predict NO_3-N fluxes in the first 100 years following harvesting and conversion to broadleaved species. Therefore, only models that included detailed modelling of the processes expected to be affected by conifer-to-broadleaf conversion (see Chapter 1) were considered so the selected model could capture the impact of tree species changes on nitrate leaching fluxes. The present study benefitted from data collected at the stand scale, so catchment-scale models were deemed unsuitable. Only models capable of simulating nitrate leaching fluxes on a monthly time step were considered due to the desire to investigate seasonal patterns. Biome-BGCMuSo was therefore selected as the model to use during the present study.

The present study aimed to (a) test the ability of a coupled biochemical-hydrological model to simulate NO_3-N leaching fluxes from a *Pinus sylvestris* stand in Thetford Forest, England, (b) develop a systematic methodology to apply coupled biochemical-hydrological models to simulate soil NO_3-N fluxes. The questions addressed were:

- Can a coupled soil water and nitrogen cycling model simulate monthly NO_3-N leaching fluxes and seasonal patterns in forest ecosystems?
- What are the key model parameters govening nitrogen leaching fluxes at the study site and can a coupled soil water and nitrogen cycling model capture decadal patterns in NO_3-N leaching fluxes?

2. Methods

2.1. Site description

Thetford Forest is a commercial plantation in East Anglia, England, dominated by Scots and Corsican pine. The land surrounding the forest is used for intensive pig and chicken farming, and cropland is frequently subjected to fertiliser applications. Atmospheric N deposition across the region is high due to intensive farming activities; the area is within a nitrate-vulnerable zone (Environment Agency, 2019). Thetford experiences an average annual rainfall of 600 mm, an annual temperature of 11.3 ^{o}C and an average deposition of nitrogen deposition of 14.7 kg ha^{-1} a^{-1} (Vanguelova & Pitman, 2019). Thetford Forest frequently experiences droughts, which cause pulses of nitrate leaching upon rewetting.

An ICP Forests Level II plot is situated in the southeastern part of Thetford forest (Figure 1, Vanguelova *et al*., 2007). The monitoring plot is dominated by Scots pine planted in 1969 and has been managed by routine thinning, which is in line with typical management practice. The soils are classified as arenosol and are underlain by chalk. The depth of the soil varies dramatically across the plot, up to 1 m in some areas and less than 0.2 m in others.

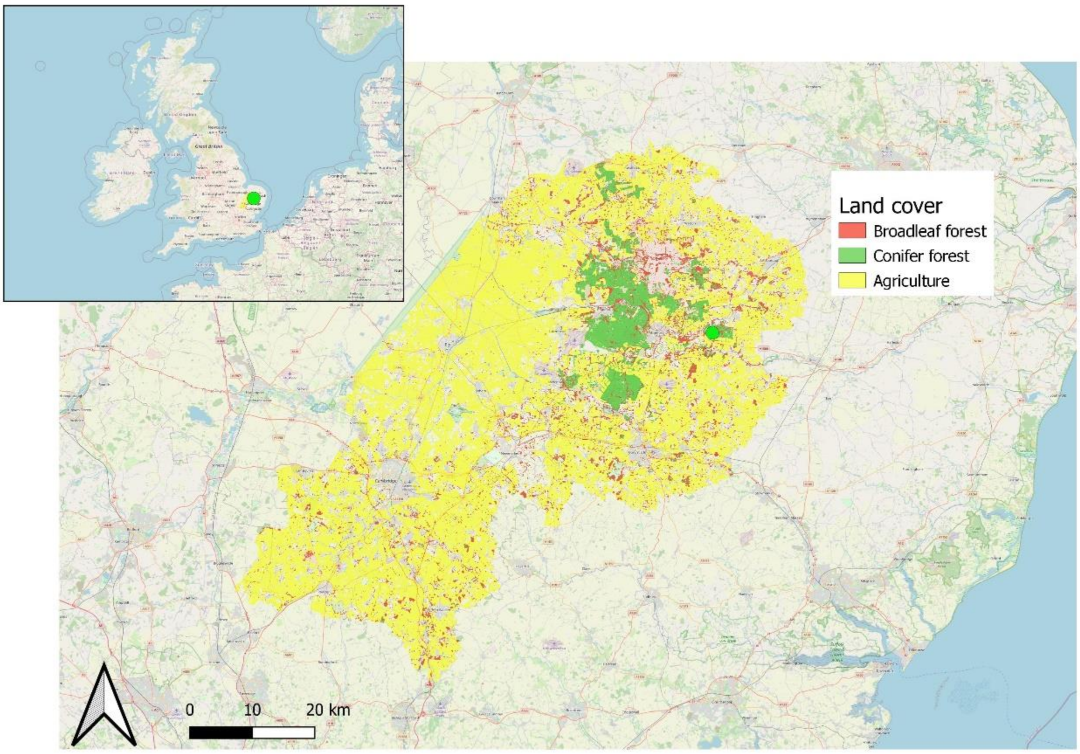


Figure 1: The location of the ICP Forests Level II plot in Thetford Forest, UK (green point), with surrounding land cover information for the catchment provided by the UKCEH Landcover Map 2015.

2.2. Long-term monitoring of nitrate leaching fluxes- field and laboratory methods

Following strict ICP Forests protocols, throughfall and soil solution samples have been collected four weekly since 1996 by Forest Research, UK. Soil solution was collected using 12 Prenart^{TM} suction cup lysimeters planted in shallow (<30 cm, n = 6) and deep (>60cm, n = 6) soils, with a vacuum of 70 kPa applied every four weeks. Soil water samples were bulked into two samples per depth for analysis. Soil solution samples collected at 60 cm depth have shown elevated nitrate levels in deep soil solution for many years, with concentrations above the accepted drinking water standard in the Water Framework Directive (Vanguelova et al., 2007). Throughfall samples were collected from 10 collectors designed according to World Meteorological Organisation requirements, situated in representative locations in the plot, mounted 1 m above the ground on wooden stakes. Throughfall samples were bulked into two samples per collection period for analysis. Soil water and throughfall samples were analysed for NO_3-N and Cl^- using a Dionex^{TM} ion chromatography machine and NH_4-N with a continuous flow analyser. NO_3-N leaching fluxes were then calculated using a chloride mass balance:

$$\text{Total deep soil water flux per month} = Cl^-_{(tf)}/Cl^-_{(ss)} * \text{total monthly rainfall (mm)}$$

$$NO_3\text{-N leaching flux} = \text{water flux} * \text{deep soil solution } NO_3\text{-N concentration}$$

Where tf = throughfall and ss = deep soil solution.

2.3. Long-term monitoring of foliar and litterfall carbon and nitrogen concentrations

As part of the routine monitoring schedule at the Level II site, Forest Research conducted foliar sampling every two years from 1996 to 2021, following the ICP Forests protocol for foliar sampling. Five trees of varying sizes were selected, and samples were obtained from the sun-exposed branches by tree climbers, alternating between the four aspects of each sampling period to minimise tree damage. One-year and zero-year needles were separated, and samples were oven-dried at 70 °C for 48 hours before grinding the needles for chemical analysis.

Litterfall was collected monthly from 1996 to 2021 by Forest Research, following the ICP Forests protocol for collecting litterfall, using 10 funnel collectors of 650 mm diameter mounted 500 mm above the ground. Litterfall was sorted into its parts (e.g. needles, twigs, cones, etc.) and weighed, and each part for all 10 collectors for the 12 sampling periods per year was bulked for chemical analysis. Samples were ground and analysed for chemical analysis. Total carbon and nitrogen content for foliar and litterfall samples were determined with a FlashEA NC Analyser.

Forest Research calculated mean concentrations of needle N and C of the five selected trees yearly for foliar samples. As the concentrations did not vary greatly between zero-year and one-year needles, only observations from one-year needles were used to assess model outputs. For litter samples, the N and C concentrations from the representative bulked samples were assumed to indicate the total amount of N and C in litter per year. I converted foliar and litter N and C to kg m^{-2} for comparison against model outputs, using the total mass of fallen needles per year. As fallen needles were collected monthly, mass lost due to decomposition was assumed to be minimal and, therefore, suitable to use in calculations of foliar nutrients per m^2.

2.4. Model description

Biome-BGCMuSo simulates carbon, nitrogen and water fluxes through an ecosystem in ten individual soil layers (Garcia-Prats *et al*., 2018; Hidy *et al*., 2012). Previous studies have shown good model performance of Biome-BGCMuSo for simulating soil water content, soil temperature (Garcia-Prats *et al*., 2018), transpiration, GPP and NPP

(Srinet *et al*., 2023; Chiesi *et al*., 2016; Mao *et al*., 2016; Nunes *et al*., 2014; Chiesi *et al*., 2007; Schmid *et al*., 2006), LAI (Srinet *et al*., 2023), above-ground biomass carbon pool (Cienciala *et al*., 2006), total ecosystem respiration (Mao *et al*., 2016), and more, specifically in forest ecosystems. Model performance was nearly equal in moist vs dry (R^2 = 0.80 vs 0.79 respectively) forest ecosystems (Srinet *et al*., 2023) and has been applied under different land-use scenarios, including investigating GPP under afforestation (Huang *et al*., 2021) and soil organic carbon stocks in different land-use categories, including forest (Sever *et al*., 2021). Its application to model N leaching from forest ecosystems has been limited. Ye *et al*. (2016) modelled N leaching from a forest in the Tibetan Plateau but could not validate the model outputs for N leaching due to a lack of data availability. Still, the model performed well in simulating other variables, such as GPP observations.

Much of the theoretical basis of the Biome-BGC models originated in the Forest-BGC model (Pietsch et al., 2005). Running & Coughlan (1988) and Running & Gower (1991) described the fundamental processes underlying Forest-BGC, which were adopted into Biome-BGCMuSo, and are summarised here and in Figure 2. Forest-BGC was developed to be especially sensitive to Leaf Area Index (LAI) as remote sensing products commonly measure LAI, thus generating an opportunity to apply the model over a wide spatial scale (Running & Coughlan, 1988). LAI defines the structure of the canopy. The canopy energy, water and carbon characteristics are defined in the model, soil processes are inferred from canopy processes, and nitrogen pools are calculated as proportional to carbon pools. The Beer's law equation uses meteorological inputs of daily short-wave radiation, LAI and the canopy extinction coefficient to calculate canopy average radiation. Canopy interception of rainfall is proportional to LAI, and intercepted water is rerouted to the evaporation compartment when air temperature and net radiation are high enough. Rainfall that is not intercepted enters the soil module and then passes through equations that determine canopy water stress based on leaf water potential and canopy stomatal conductance. Transpiration is calculated using the Penman-Montieth equation with fixed aerodynamic resistance and is then subtracted from the soil water compartment. Later developments improved the simulation of the soil water balance, including the option to simulate water balance using the Richards equation (Hidy *et al.*, 2016).

The rate of photosynthesis is calculated from the CO_2 diffusion gradient from leaf to air, canopy conductance and canopy mesophyll conductance, which is a product of nitrogen, light and temperature functions derived from the meteorological data. The carbon available for leaf growth is calculated from the amount of photosynthate and the leaf:root partitioning ratio where photosynthate is the only limiting factor, otherwise, leaf growth may be constrained by water and nitrogen availability. Leaf growth then calculates LAI for the next timestep. Once carbon is allocated to above or below-ground pools, growth respiration is calculated as a fraction of the carbon pools from coefficients based on rates of the biochemical reactions that produce plant molecules. Litterfall and decomposition are then calculated once carbon allocation to different plant consituents are known, with decomposition determined by litterfall inputs, soil water, and temperature. N availability is defined by LAI and maximum defined leaf N concentration, with root N content defined as a percentage of leaf N.

The mobilisation of N through the decomposition of plant tissues is calculated as a percentage of C release.

Previously, soil N pools were not separated into N-forms. However, as described by Hidy et al. (2016), in the most recent Biome-BGCMuSo versions, ammonium and nitrate pools are calculated separately. Ammonium and nitrate pools are calculated on a layer-by-layer basis and, in addition to decomposition, are also affected by microbial processes, plant uptake, leaching and atmospheric deposition, unless the soil layer is below the rooting zone, in which case plant uptake and certain microbial processes are assumed not to occur. An empirical function defines leaching from the user-supplied parameter of "soluble N subject to mobilisation", soil ammonium and nitrate content, percolation and diffusion (Hidy et al., 2016).

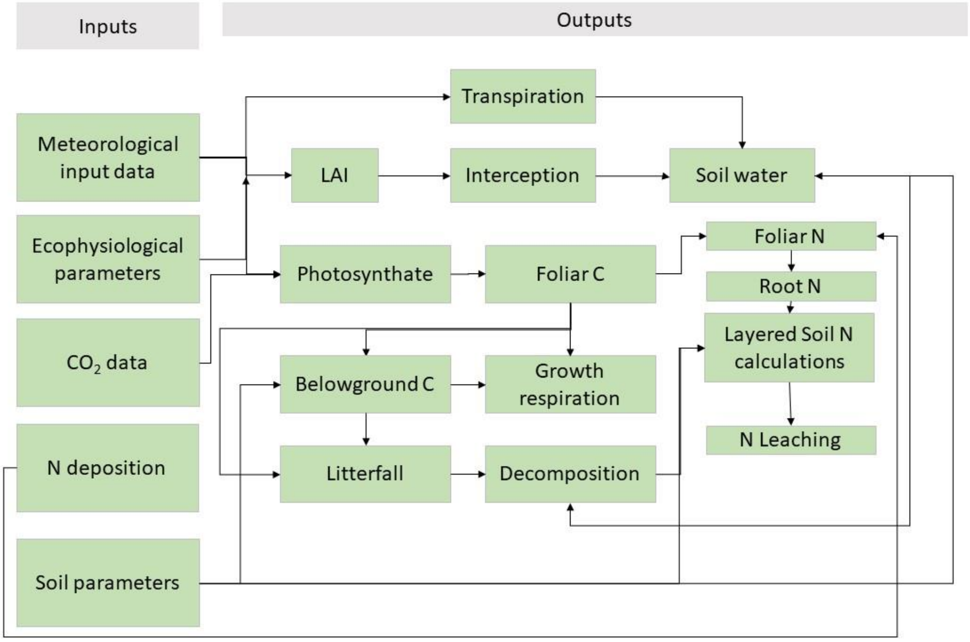


Figure 2: Conceptual diagram of the processes in Biome-BGCMuSo.

2.5. Input data

The input files for Biome_BGCMuSo are summarised in Table 1. Appendix 10 and 11 define the parameters and the literature sources used.

Table 1: The input files and source of data for Biome-BGCMuSo simulations.

File	**Description**	**Sources**	**Optional/ Mandatory**
Initialisation	Locates model inputs, selects methods for e.g. hydrological proceedures		M
Meteorological	Daily observations of temperature, precipitation, solar radiation and day length for each day in the study period	Level II data, MTCLIM model,	M
Ecophysiological parameters	Parameters related to the dominant plant species in the study site such as required temperatures for growth and annual leaf and fine root turnover	Observed Level II data, literature (White *et al.*, 2000), model documentation, parameterisation proceedures	M
Soil parameters	Parameters related to the physical soil properties and rates of soil biochemical activities in the study site.	Observed Level II data, literature (White *et al.*, 2000), model documentation, parameterisation proceedures	M
Ambient CO_2	Annual CO_2 concentration (ppm)	Biome-BGCMuSo demonstration files	O
N deposition	Annual N deposition (ppm)	Level II data	O

The Level II forest monitoring site is paired with an open field plot with an automatic meteorological station. The meteorological station provides a continuous dataset of daily temperature and precipitation. The open field plot also provided data on bulk wet N deposition, which was used to generate the N deposition file. The observed dry N deposition was not available for the entire study period, so it was not used. The model was also run with a constant average wet + dry N deposition value to test whether dry deposition could potentially affect model outputs. As recommended in the Biome-BGCMuSo documentation (Hidy *et al.*, 2021), the MTCLIM model was used to calculate daily solar radiation from a site information file and generate a file in the format required for Biome-BGCMuSo. The CO_2 file used was provided in the Biome-BGCMuSo demonstration materials. Where data was available, ecophysiological and soil parameters were set using observed values from the Level II forest monitoring site. Values from White *et al*. (2000), who collated parameter values from the literature base, were used for some ecophysiological parameters. Parameters were subjected to a sensitivity analysis where neither observed nor literature-recommended values were available.

2.6. Sensitivity analysis

Biome-BGCMuSo allows the user to set 199 parameters to control physiological and soil biological and physical processes. A sensitivity analysis was performed to identify the parameters with the greatest influence on soil NO_3-N leaching fluxes to limit the number of parameters that needs parameterising. For the sensitivity analysis, ecophysiological and soil parameters were varied individually between a minimum and maximum value (Appendix 12). Ecophysiological parameters and soil parameters were tested individually for their effect on soil NO_3-N leaching outputs for 2015-2019. The effect of each parameter value on the mineral N leaching output was visualised, and those seen to affect the output were selected for the parameterisation procedure. Parameters without observational constraints and those with no apparent effect on model outputs were set as the example value suggested in the BIOME-BGCMuSo 6.2 User’s Guide

2.7. Parameterisation and assessment of model performance

Twenty-nine soil parameters and 13 ecophysiological parameters were selected from the sensitivity analysis were subjected to the parameterisation procedure (Figure 3).

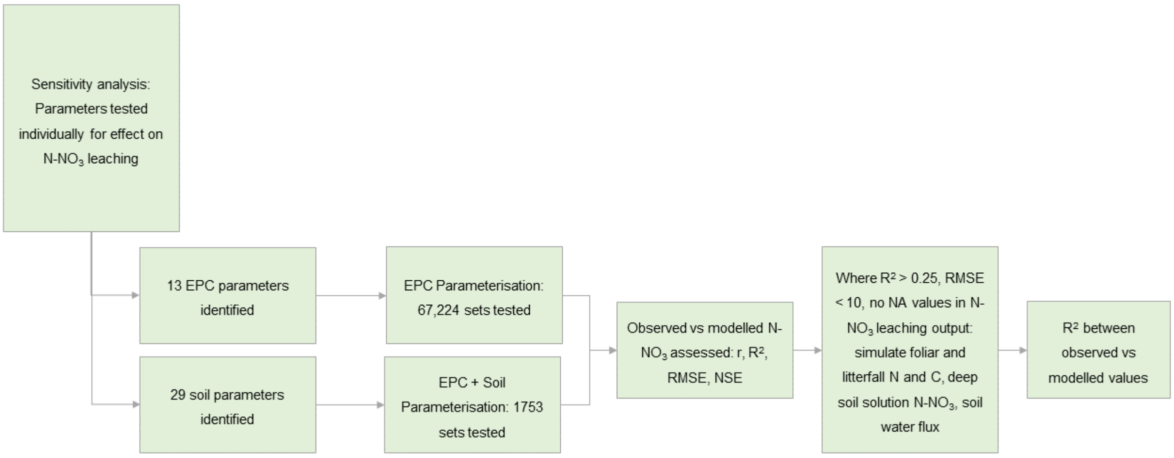


Figure 3: The parameterisation procedure followed to find suitable parameters for Biome-BGCMuSo to simulate NO_3-N leaching fluxes from the Level II plot at Thetford Forest.

The musoMonte() function in R was used to run a parameterisation procedure on the selected ecophysiological parameters. The musoMonte() function selected values within a set range (Appendix 10 and 11) for each model run from a conditional multivariate uniform distribution. musoMonte() was set to run 100,000 iterations, testing 100,000 parameter sets, but the computational resource available only managed to complete 67,224 runs. Ideally, for the 13 parameters tested in this EPC parameterisation procedure, more runs are needed to explore the full parameter space. The need for the model to run from a .exe on Windows limited the options for more intensive computational resources, and whilst the ability to alternatively run the model on a Linux system was available, difficulties were encountered with the R versions and the ability to install the required packages on the JASMIN computing cluster. Each model ran output mineral N leaching daily from 1996-2020.

An equivalent function to musoMonte(), capable of simultaneously running a parameterisation procedure on the ecophysiological and soil parameters, has not yet been developed for Biome-BGCMuSo. A new function was, therefore, written to perform the parameterisation procedure on the ecophysiological and soil parameters file simultaneously. However, this function did not benefit from years of development by experienced software developers and was, therefore, less computationally efficient. Given computational constraints, the simultaneous parameterisation of both ecophysiological and soil parameters were tested with 1753 unique parameter combinations, with parameters randomly selected within a set range for each parameter, outputting mineral N leaching on a daily timestep from 1996-2021.

For both parameterisation procedures, mineral N leaching was aggregated to monthly N leaching and compared against the monthly values of NO_3-N leaching. Parameter sets that generated leaching-fluxes with NA values were removed from the analysis. Pearson's correlation, R^2 , RMSE and Nash-Sutcliffe Efficiency were calculated to assess the alignment between monthly observed and modelled values. Observed and modelled outputs were also aggregated into yearly outputs to test the model's capability to capture yearly averages and totals rather than capture seasonal variability.

2.8. Simulation of other output parameters

The parameter sets which achieved an R^2 >0.25 and an RMSE < 10, where NO_3-N leaching fluxes were simulated for the full 25-year period without producing NA values, were selected for further investigation. Soil water drainage fluxes (60-90 cm depth soil layer) and deep soil NO_3-N concentrations were simulated individually to assess the model's capability to capture biochemical processes vs hydrological processes. Foliar C and N content, needle litterfall C and N content, potential evapotranspiration (PET), and canopy interception were also simulated to assess the model's capability to simulate above-ground processes. A linear model was used to assess model performance between observed and simulated foliar and litterfall C and N content. Observed data was unavailable for PET and interception, but monthly outputs were visualised alongside monthly rainfall to assess whether outputs were within a realistic range. Canopy interception of rainfall was converted from the total mass of water intercepted to the percentage of total rainfall intercepted. The variables of growth respiration and leaf area index were simulated to troubleshoot the model.

3. Results

3.1. Simulation of N leaching fluxes

3.1.1. Ecophysiological parameterisation

The model runs with only the ecophysiological parameter set failed to identify a parameter set capable of simulating NO_3-N fluxes near observed values (Figure 4). The model did capture the temporal trends, with simulated peaks in NO_3-N leaching coinciding with observed peaks, but the mean simulated NO_3-N values were often greater than the observed values by at least 10 kg-N ha^{-1} yr^{-1} (e.g. 2000, 2009). A low correlation between observed and modelled values was seen (Figure 5, $r = 0.15$, $R^2 = 0.02$). The model drastically overestimated peaks NO_3-N fluxes from 2017-2020 when observed values were low due to periods of low rainfall and slight reductions in deposition inputs.

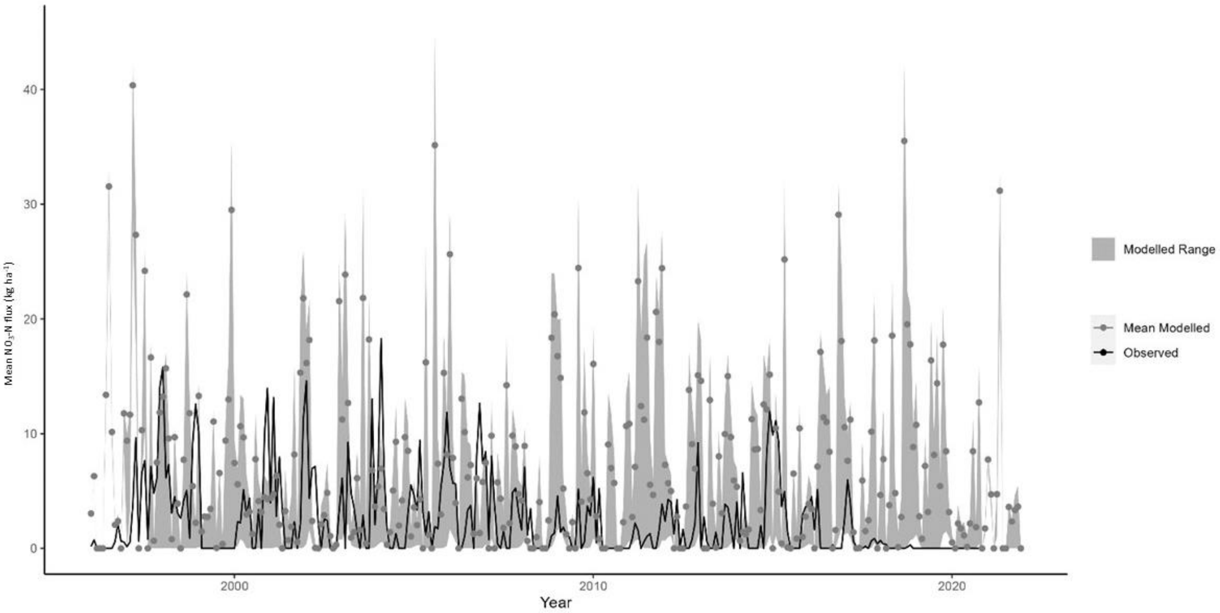


Figure 4: Observed monthly NO_3-N leaching fluxes from a Scots pine stand in Thetford Forest (solid line), the range of simulated monthly NO_3-N leaching fluxes (shaded) and the mean simulated NO_3-N leaching fluxes (grey points) from 67,224 model runs, generated by the Biome-BGCMuSo model during the ecophysiological parameterisation procedure.

The highest adjusted R^2 achieved by the parameter set when simulating monthly N-leaching fluxes in the ecophysiological parameterisation procedure was 0.02 (r = 0.16, RMSE = 7.68, Nash-Sutcliffe Efficiency (NSE) = -0.28). The highest R^2 was achieved when yearly-aggregating total N-leaching fluxes in the ecophysiological parameterisation procedure was 0.12 (r = -0.39, RMSE = 23.09, NSE = -1.60). Out of the 67,224 parameter sets tested, 3024 parameter sets yielded implausible parameter combinations that produced model errors, such as negative soil carbon stocks.

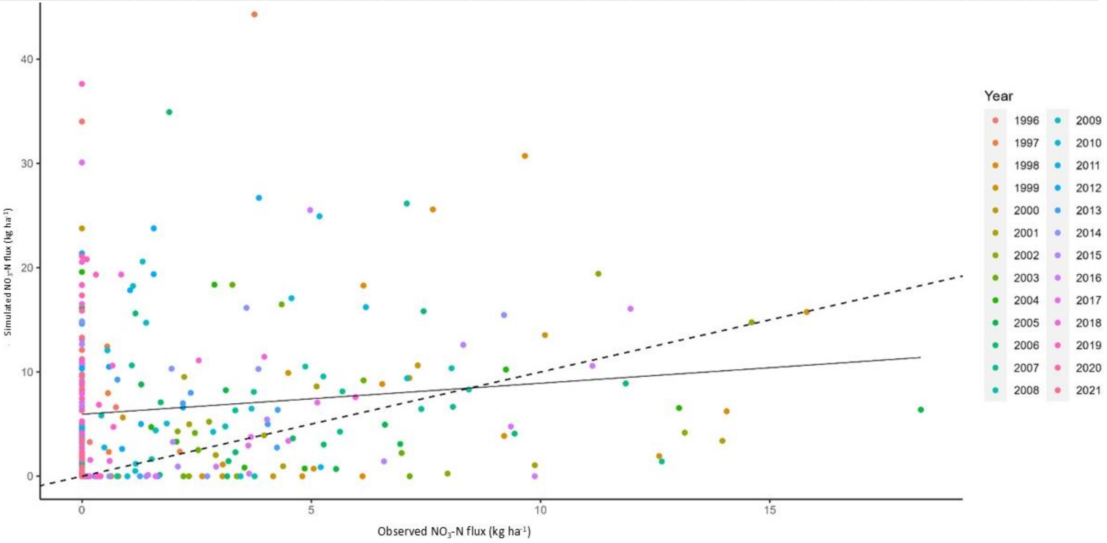


Figure 5: The relationship between observed vs simulated monthly NO$_3$-N leaching fluxes (solid line) from a Scots pine stand in Thetford Forest. Simulated values were generated by the Biome-BGCMuSo model during an ecophysiological parameterisation procedure where 67,224 parameter sets were tested. The dotted line represents the 1:1 line.

3.1.2. Combined ecophysiological + soil parameterisation

Despite the lower number of parameter sets tested, the parameterisation procedure on the ecophysiological and soil parameters generated parameter sets that enabled better model performance than during the ecophysiological parameterisation alone (Figure 6). The temporal patterns generated by the model aligned well with the observed patterns, and lows in NO$_3$-N leaching fluxes were often captured. The mean of NO$_3$-N leaching fluxes simulated across all parameter sets aligns closely with observed leaching fluxes before 2010 but less so post-2010.

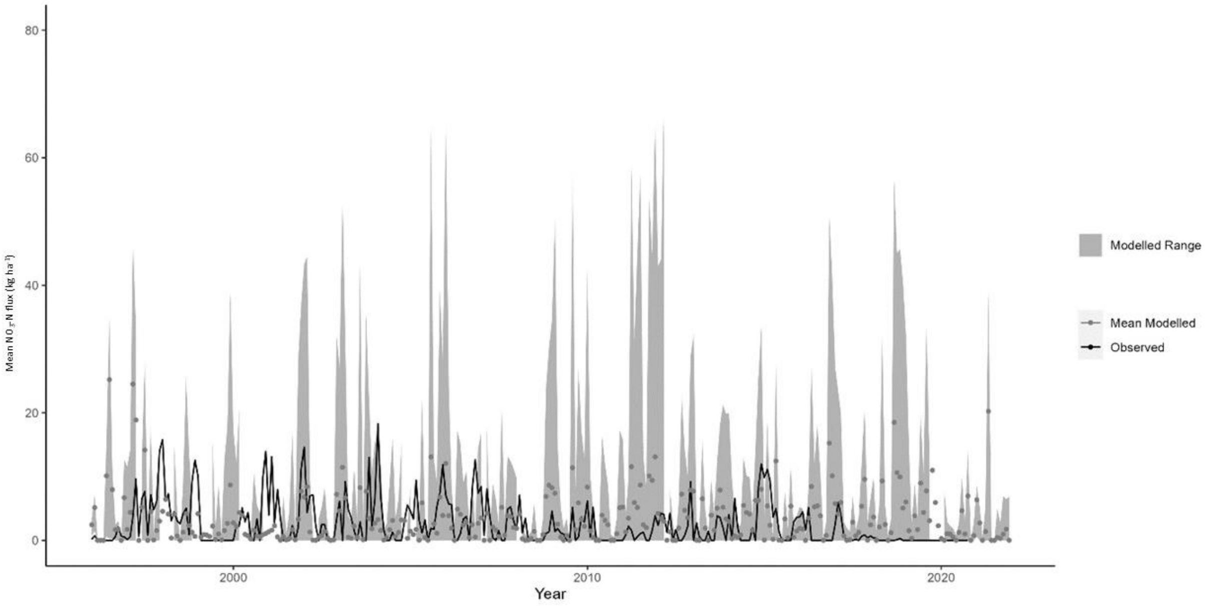


Figure 6: Observed monthly NO_3-N leaching fluxes from a Scots pine stand in Thetford Forest (solid line), and the range of monthly NO_3-N leaching fluxes simulated by the Biome-BGCMuSo model during the ecophysiological and soil calibration procedure (shaded area) where 1753 parameter sets were tested.

The highest adjusted R^2 achieved by a parameter simulating monthly N-leaching fluxes in the ecophysiological + soil parameterisation procedure was 0.16 (r = 0.44 , RMSE = 9.13, NSE = -1.06). The highest R^2 was achieved when simulating yearly-aggregated total N-leaching fluxes in the ecophysiological + soil parameterisation procedure (R^2 = 0.32, r =-0.59 , RMSE = 15.42, NSE = -1.22). The higher R^2 obtained when simulating yearly rather than monthly fluxes suggests that the model may be more suitable for investigating long-term trends.

3.2. Diagnosing model fidelity

3.2.1. Comparison of the most suitable parameter sets

Eight parameter sets met the criteria of an $R^2 > 0.25$ and an RMSE < 10 whilst simulating NO_3-N leaching without NA values 1996-2021 (Table 2).

Table 2: The parameter values of the parameter sets (A-H) that simulated NO_3-N leaching fluxes with an adjusted R^2 with observed values of >0.25 and RMSE < 10.

FILE	**VARIABLE**	**VALUE**							
		A	B	C	D	E	F	G	H
EPC	Transfer growth period as fraction of growing season	0.55	0.55	1	0.55	1	1	1	0.55
	Base temperature	2.67	8	0	8	8	8	8	0
	Annual leaf and fine root turnover fraction	0.447	0.447	0.447	0.447	0.447	0.447	0.447	0.447
	Whole plant mortality fraction in vegetation period	0.075	0.05	0.1	0.1	0.025	0.05	0.05	0
	C:N of leaves	23.17	29.086	20.21	29.09	14.3	17.26	32.04	29.086
	Canopy water interception coefficient	0.07	0.07	0.03	0.07	0.07	0.01	0.05	0.07
	Canopy light extinction coefficient	0.63	0.467	0.467	0.63	0.467	0.63	0.63	0.467
	Fraction of leaf N in rubisco	0.15	0.2	0.125	0.2	0.175	0.15	0.175	0.15
	Maximum stomatal conductance	0.003	0.003	0.005	0.007	0.001	0.003	0.003	0.005
	Maximum depth of rooting zone	2.375	0.5	1.125	1.125	1.125	0.5	1.75	3
	Root distribution parameter	0.6	1	0.6	0.4	0.6	0.4	0.4	0.8
	VWC ratio	1	1	0.8	0.8	1	0.8	5.2	0.8
	VWC ratio 2	0.4	0.4	0.7	0.7	0.4	0.7	1	0.7
	Maximum senescence mortality coefficient of above-ground plant material	0.05	0.025	0.075	0.05	0	0.1	0.4	0.025
SOIL	NH4 mobilen proportion	0.25	1	0	0	0	0.125	0	0
	Parameter 3 for tscalar function of decomposition	275	275	300	250	300	250	300	275

	Parameter 4 for tscalar function of decomposition	0	50	25	0	0	0	50	12.5
	Soil temperature for decomposition	10	-15	-2.5	-15	-8.75	3.75	-15	10
	e-folding depth of decomposition rate's depth scalar	1.1	1.4	0.8	0.5	1.1	0.8	1.4	1.1
	Net mineralisation proportion of nitrification	0.75	1	0.75	0.75	1	0.75	0	1
	Maximum nitrification rate	0.767	0.767	0.417	0.23	0.88	0.417	1	0.417
	For pH scalar function of nitrification 1	0.5	0.4	0.1	0.7	0.9	0.2	0.7	0.9
	For pHscalar function of nitrification 2	0.031	0.137	0.157	0.03	0.07	0.137	0.094	0.2
	WFPS for scalar of nitrification calculation	0.2375	0.175	0.175	0.1125	0.2375	0.5	0.625	0.3
	Respiration fractions for fluxes between compartments (l1s1)	0.775	0.1	0.1	0.55	0.775	0.1	0.325	1
	Respiration fractions for fluxes between compartments (l2s2)	0.325	0.1	0.55	0.55	0.1	0.55	1	0.55
	Respiration fractions for fluxes between compartments (l4s3)	0.1	0.325	0.1	0.1	0.775	1	0.325	0.1
	Respiration fractions for fluxes between compartments (s1s2)	1	0.55	0.775	0.1	0.55	0.1	0.325	1
	Respiration fractions for fluxes between compartments (s2s3)	0.55	1	0.775	0.325	1	0.55	0.1	1
	Respiration fractions for fluxes between compartments (s3s4)	0.1	0.55	0.775	0.55	0.775	0.325	0.1	0.1
	Rate constant scalar of labile litter pool	0.55	0.55	0.775	1	0.775	0.55	0.55	0.1
	Rate constant scalar of cellulose litter pool	0.1	1	0.55	0.775	0.55	0.55	0.1	0.1
	Rate constant scalar of lignin litter pool	0.0575	0.0575	0.1525	0.01	0.2	0.0575	0.01	0.1525
	Rate constant scalar of physical fragmentation of coarse woody debris	0.001	0.0055	0.055	0.00775	0.00325	0.00775	0.00775	0.0055
	Limit of first stage evaporation	1.5	0.5	1	1.5	1	1	0.5	1
	Soil stress function	0.1	0.55	0.775	0.775	0.55	0.775	0.325	0.325
	Parameter: evaporation reduction	0.1	0.325	0.775	0.55	0.1	0.775	1	1
	1 for diffusion calculation (tipping)	1	0.75	0.625	0.625	0.875	0.75	1	0.5
	2 for diffusion calculation (tipping)	35	30	35	40	40	35	32.5	37.5
	3 for diffusion calculation (tipping)	10	1	5.5	7.75	1	7.75	3.25	3.25

Of the eight parameter sets subjected to further investigation, the R^2 between observed and modelled values of mean annual deep soil solution NO_3-N leaching was >0.20 for five, but the relationship between observed and modelled total annual soil drainage fluxes was poor for all ($R^2 < 0.1$, Table 3).

Table 3: The adjusted R^2 of linear regressions comparing observed vs modelled values of six variables simulated by Biome-BGCMuSo, using eight different parameter sets (A-H). Bold font indicates variables where the relationship between observed and modelled values was significant ($p < 0.05$).

Variable	**R^2**							
	A	**B**	**C**	**D**	**E**	**F**	**G**	**H**
Total Annual N leaching	**0.32**	**0.26**	**0.27**	**0.26**	**0.31**	**0.32**	**0.27**	**0.30**
Mean annual deep soil solution NO_3-N	**0.24**	**0.18**	**0.31**	**0.34**	0.01	**0.26**	**0.27**	**0.03**
Total annual soil drainage	**-0.03**	-0.04	-0.03	0.01	-0.02	0.006	-0.03	0.01
Litter needle N	-0.07	-0.04	-0.05	-0.06	**0.41**	-0.03	-0.06	0.14
Litter needle C	0.009	**0.35**	-0.13	-0.12	0.16	-0.012	-0.12	<0.01
Foliar N	0.17	-0.03	**0.35**	0.25	-0.07	0.44	**0.43**	-0.03

Generally modelled total annual NO_3-N leaching flux values for all parameter sets were underestimated pre-2010, but following reductions in observed values post-2010, modelled values became closer to observed and later overestimated NO_3-N leaching fluxes (Appendix 13). Model output did not vary when a constant N deposition value was used instead of the input file with varying annual wet N deposition, despite observations previously suggesting deposition is a large driver on NO_3-N leaching in Thetford Forest.

Simulated patterns in total deep soil drainage generally followed patterns in observed values of total annual rainfall and deep soil drainage but with over or underestimating peaks. The poor drainage simulation may be linked to the estimation of rainfall interception, which was generally poor. Visualisation of simulated rainfall interception showed that in all eight model runs, the model predicted a drop in interception to zero during the study period, indicating the complete loss of a canopy. However, the year in which this event occurred varied between parameter sets. Prior to the loss of a canopy, parameter sets A and E were the only ones that estimated a mean annual interception of >30 % (Table 4).

Table 4: Mean rainfall interception, while the model still predicted the presence of vegetation in eight model runs with different parameter sets.

Parameter Set	Years Interception >0	Mean Interception (%)
A	1997-2005	47
B	1997-2018	8.75
C	1997-2012	2.4
D	1997-2007	7.72
E	1997-2019	31.57
F	1997-2004	1.73
G	1997-2007	5.58
H	1997-2019	16.25

3.2.2. Mortality events in the simulated Biome-BGCMuSo model runs

The visualisation of simulated growth respiration and LAI from the eight parameter sets showed that in all runs, the model simulated a mortality event where growth respiration dropped to zero, shortly followed by a complete reduction in LAI. A widespread mortality event was, however, not observed. The year the mortality event occurred varied for each parameter set, but it generally happened between September and December, except for one in July 2002 (Table 5).

Table 5: The date/day of the year at which growth respiration dropped to zero in eight model runs of Biome-BGCMuSo with different parameter sets.

Parameter set	**Cessation of growth respiration date**	**Yday**
A	16/11/2005	320
B	23/09/2017	266
C	29/09/2009	272
D	10/11/2005	314
E	12/12/2018	346
F	20/07/2002	201
G	04/12/2006	338
H	25/10/2018	298

The mortality events caused an increase in needle litter N content in the outputs of parameter sets A, C, D, F and G to values at least four times greater than those observed. The mortality events caused a decrease in simulated needle litter C content and living needle foliar N content in all parameter sets. The mortality-driven changes in simulated litter and foliar C and N content sometimes brought simulated values into better alignment with observed values, giving R^2 values >0.30 between model and observed values for some parameter sets (Table 3), which could be misinterpreted if the litter and foliar variables were studied in isolation.

3.2.3. Impact of model parameterisation refinement

To further explore parameters that may potentially be responsible for the simulated mortality events, further parameterisation procedures were tested (Figure 7). The main hypotheses for the cause of the mortality events were (1) nitrogen limitation driven by high simulated foliar N content and (2) climatic triggers interacting with N cycling calculations, resulting in limited movement of photosynthetic products. To test this, the parameterisation of the VWC ratios in the stress and senescence section of the EPC files (Table 2) was removed, and the values fixed to those suggested in the Biome-BGCMuSo documentation. The parameterisation procedure with the EPC parameters was repeated for 43,000 runs. However, the parameter sets that achieved the highest R^2 between modelled and simulated NO_3-N leaching still encountered the issue of simulating mortality events.

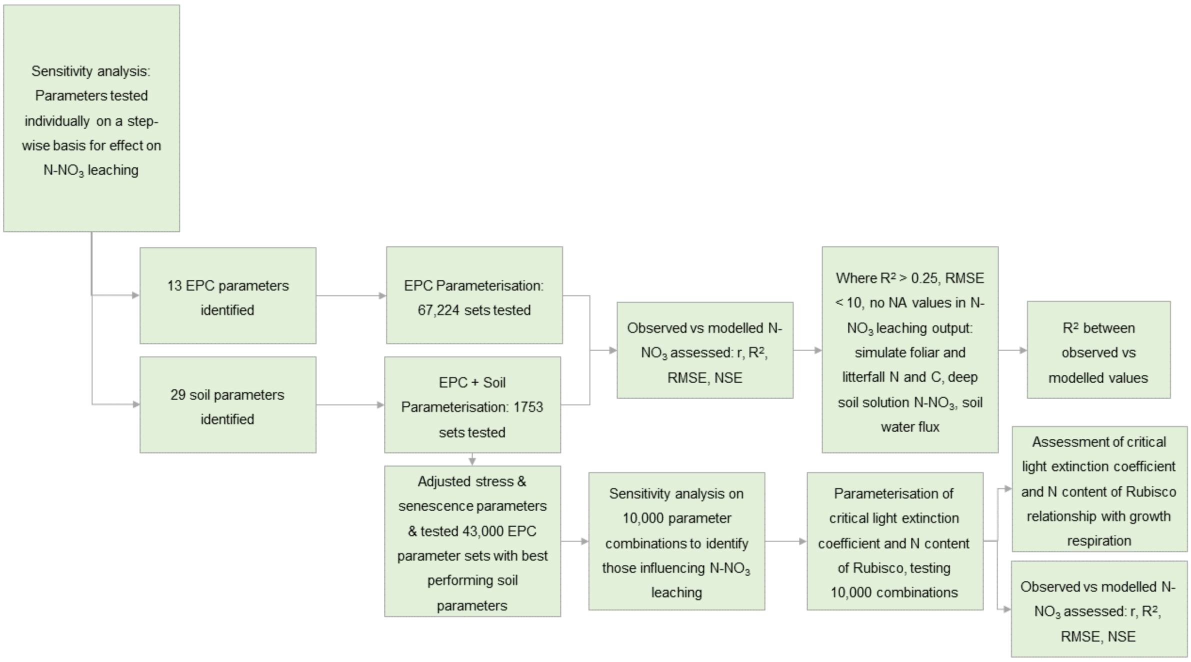


Figure 7: The calibration procedure workflow with additional steps to identify the cause of simulated mortality events.

The updated EPC parameterisation procedure outputs were subjected to a second sensitivity analysis to determine the most influential parameters. Rather than using only one observation year to identify the effect of each parameter on NO_3-N leaching, the second sensitivity analysis used the full study period. The canopy light extinction coefficient, leaf C:N ratio, root distribution parameter and fraction of leaf N in rubisco were identified as those with the dominant control on N-leaching fluxes (Figure 8). As the foliar C:N ratios were within the range of observed values and the root distribution parameter likely exerted its control on N-leaching fluxes via its role in calculating below-ground water transport, rather than affecting foliar N content, these two parameters were not subjected to further parameterisation. The canopy light extinction coefficient and fraction of leaf N in Rubisco were subjected to further parameterisation as described below.

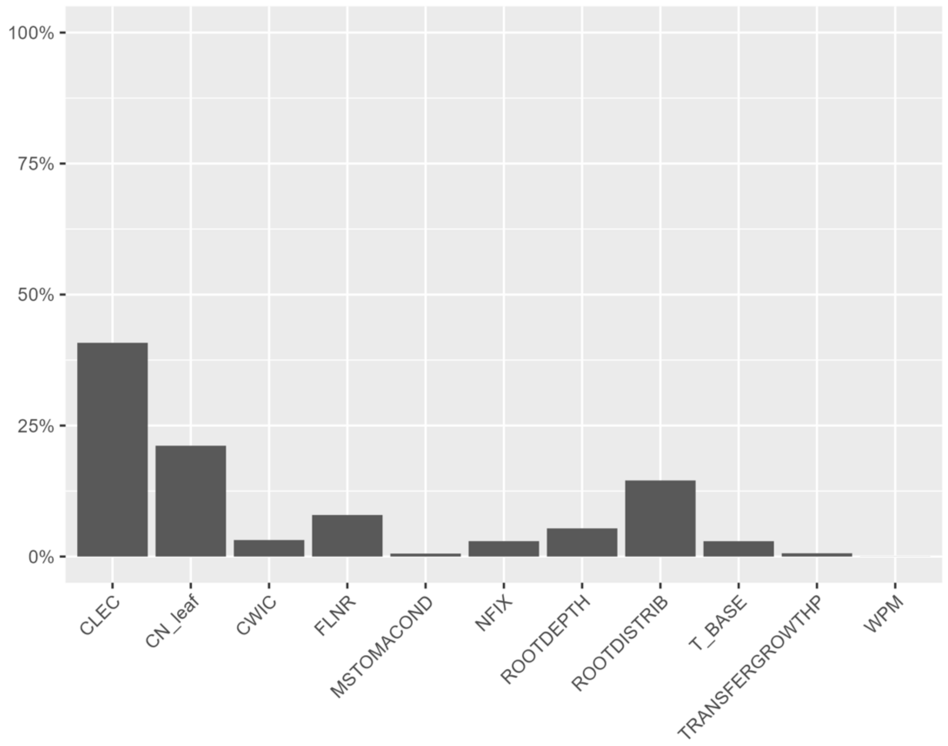


Figure 8:The effect of each parameter tested the EPC parameterisation procedure on simulated NO_3-N leaching.

In the first and second parameterisation procedures using the EPC and EPC + Soil parameters, respectively, the canopy light extinction coefficient and fraction of leaf N in Rubisco parameters varied between 0.1-0.8 and 0.1-0.2 respectively, being close to the values for canopy light extinction coefficient suggested by White *et al*. (2000) for

Pinus spp., of 0.43-0.64, and the value of fraction of leaf N in rubisco suggested in the Biome-BGCMuSo user guide of 0.28 as a value for temperate, mature *P.sylvestris* was not available. The model was run 10,000 times for the next parameterisation procedure, varying the canopy light extinction coefficient between 0.5-1 and the fraction of leaf N in Rubisco between 0.01-0.2. Mean growth respiration was zero when the fraction of leaf N in Rubisco was below 0.03 (Figure 9b). Mean growth respiration was positively correlated with Rubisco N content (Pearson's $r = 0.95$, $p < 0.001$). Critical Light Extinction Coefficient did not display a clear relationship with growth respiration (Figure 9a, Pearson's $r = -0.016$, $p > 0.05$), but the ratio of the Critical Light Extinction Coefficient:Rubisco N content was negatively correlated to growth respiration (Figure 9c, Pearson's $r = -0.84$, $p < 0.001$), particularly beyond a threshold of approximately 20. Mean daily deep soil NO_3-N leaching was negatively correlated with mean daily growth respiration (Figure 9d, -0.92, $p < 0.001$).

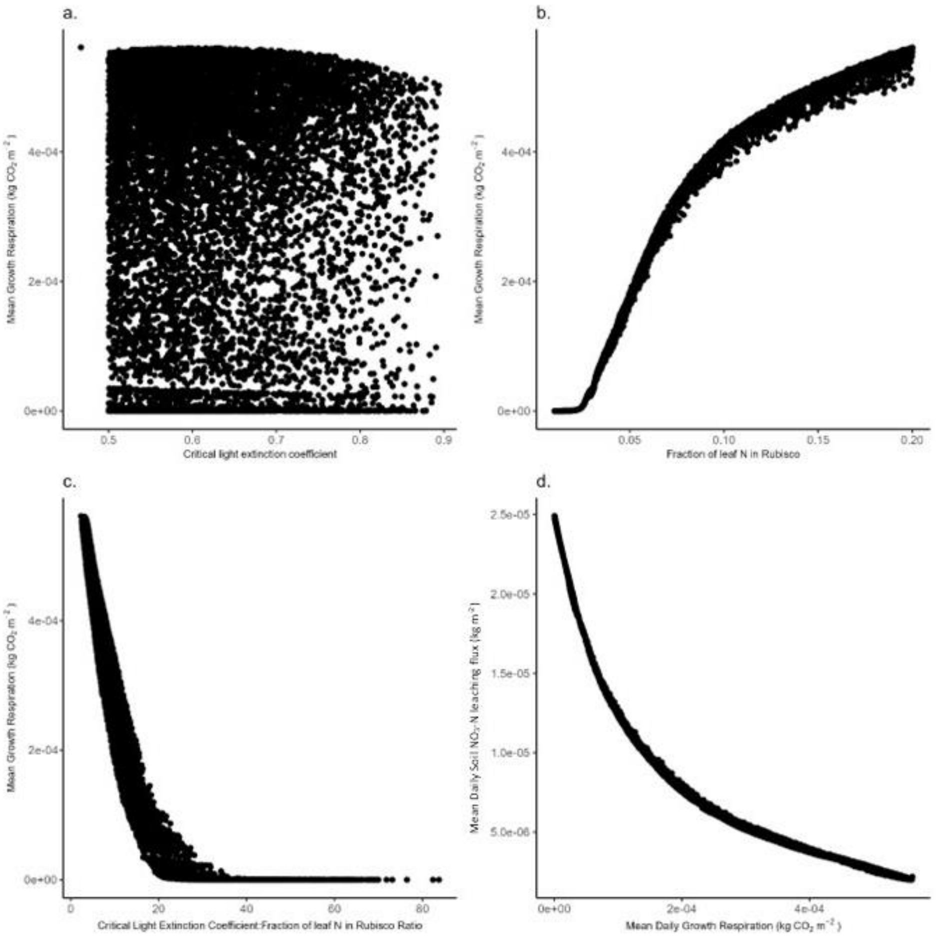


Figure 9: The relationship between mean daily growth respiration and (a) the value of the critical light extinction coefficient parameter, (b) the value of the fraction of leaf N in Rubisco parameter, (c) the ratio between the critical light extinction coefficient parameter and the fraction of leaf N in Rubisco parameter, and (d) mean daily deep soil NO_3-N leaching flux, generated from 10,000 model runs.

The observed mean total annual NO_3-N leaching flux from the study plot in Thetford Forest was 28.41 kg-N ha^{-1}, which, if daily NO_3-N leaching fluxes were constant over a year, would equal 7.78 x 10^{-6} kg-N m^{-2}. Where the mean daily NO_3-N leaching was 7.78

x 10^{-6}, the fraction of leaf N in rubisco values was between 0.05 and 0.06 (Figure 10a). The fraction of leaf N in rubisco was 0.05-0.06, mean daily growth respiration was between 0.00017-0.00025 kg-CO_2 m^{-2} (Figure 10b).

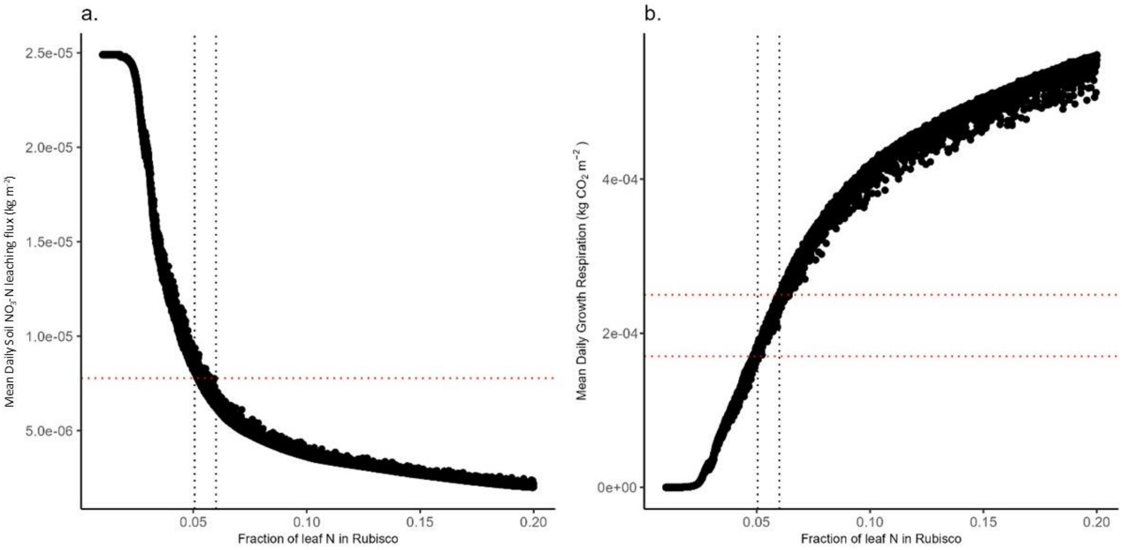


Figure 10: Estimation of (a) the the fraction of leaf N in rubisco based on observed NO_3-N leaching fluxes and (b) the mean daily growth respiration simulated where the fraction of leaf N in rubisco was 0.05-0.06.

4. Discussion

Biome-BGCMuSo is a complex model with a large number of parameters. Studies have previously achieved moderate success in modelling soil carbon storage in forest ecosystems when performing parameterisation procedures on above-ground variables such as GPP (Srinet *et al*., 2023), net ecosystem exchange, actual evapotranspiration (Schmidt *et al*., 2006), soil depth and nitrogen fixation (Cienciala & Tatarinov, 2006). In this chapter, rather than selecting a small number of parameters to parameterise, a partially systematic method was deployed to select the best parameter set for simulating NO_3-N leaching fluxes from a temperate Scots pine plantation.

Parameterisation of the ecophysiological parameters alone did not generate parameter sets that capture NO_3-N leaching fluxes monthly or annually. In the present study, we attempted a parameterisation procedure on both input parameter files, the implementation of which was heavily constrained by high computing demand and limited options for high-performance computing resources. When assessed on an annual time step, the procedure implemented appeared to generate a parameter set which simulated NO_3-N leaching fluxes with some, although limited, alignment to observed values. Several factors make simulating NO_3-N leaching fluxes from Thetford Forest difficult. Observed NO_3-N leaching patterns showed that periods of drought in the region resulted in soil nutrient flushing events upon rewetting (Vanguelova *et al.*, 2007). In the period 1996-2004, N deposition in the area was dominated by NO_x compounds, but the success of policies to reduce NO_x emissions with the simultaneous intensification of agricultural activity in the study region led to a shift towards NH_3 dominance and an increase in dissolved organic nitrogen (Vanguelova *et al.*, 2024). Due to the difference in the response of forest ecosystems to NO_x deposition vs NH_3 deposition, the misalignment in NO_3-N leaching values between observed and model outputs here was initially thought to be due to the effect of switching to NH_3-dominance in deposition inputs on observed values.

However, upon visualisation of the hydrological outputs, it became apparent that the best-performing parameter sets ($R^2 > 0.25$) were simulating mortality events, which were not observed. Two hypotheses for the simulated mortality events were explored:

(1) The occurrence of mortality primarily during winter months and following periods of temperature drops or low precipitation suggested unsuitable parameterisation of the ecophysiological stress and senescence parameters, (2) the high content of N in the foliar N pool and low return of foliar N to the soil pools via litterfall suggested mortality due to N limitation and unsuitable parameterisation of vegetation N processes. Further adjusting the stress and senescence parameters still produced model outputs with mortality events. A study simulating GPP under drought in Mediterranean systems successfully reduced model sensitivity to drought by parameterising the maximum stomatal conductance and cuticular conductance parameters (Chiesi *et al.*, 2007), but in the second sensitivity analysis performed here, stomatal conductance and cuticular conductance parameters were not identified as the most influential to NO_3-N leaching outputs. The second hypothesis of simulated N-limitation is likely the cause of unrealistic model outputs here.

The second sensitivity analysis identified the foliar C:N ratio, root distribution parameter, canopy light extinction coefficient and fraction of N in rubisco as the most influential parameters on NO_3-N leaching outputs over the entire study period. As the parameterisation of foliar C:N ratios were based on the range of observed values, and the root distribution parameter is connected more to below-ground processes, only canopy light extinction coefficient and fraction of N in Rubisco were explored further as those potentially causing N limitation-induced mortality. Further exploration identified the positive relationship between the fraction of N in rubisco and growth respiration, the negative relationship between growth respiration and canopy light extinction coefficients, and growth respiration and NO_3-N leaching. From the observed leaching values, a suitable fraction of N in Rrubisco was estimated as 0.05-0.06, much lower than the range tested in the EPC and EPC + Soil parameterisation procedures. Given computational restraints, further refinement of this parameter set is subject to future work, which may improve model performance. The percentage of foliar N in rubisco is not commonly measured but can be estimated from the equation: (maximum rate of carboxylation * specific leaf area * foliar C:N)/ 7.16 (White *et al*., 2000). Since White *et al*. (2000) published values for Biome-BGC's ecophysiological parameters based on an extensive literature review, there have been extensive studies on carboxylation in *Pinus sylvetris* (e.g. Stangl *et al*., 2022; Vermeulen *et al*., 2015; Beeck *et al*., 2009), and an updated review to aid in model parameterisation is therefore warranted. Growth respiration where fraction of N in rubisco was 0.05-0.06 was estimated as 0.00017-0.00025 kg-CO_2 m^{-2} day^{-1}. Observations of growth respiration in *P. sylvestris* have varied between studies but have generally been higher than 0.00025 kg-CO_2 m^{-2} day^{-1}. For example, Chan *et al*. (2017) reported values between 0.001296-0.00864 kg-CO_2 m^{-2} day^{-1}, and Zha *et al*. (2004) reported values between 0.000266-0.00266 kg-CO_2 m^{-2} day^{-1}. Improved parameterisation of the N fraction in Rubisco is needed to simulate accurate model outputs. An understanding of parameters that interact with the fraction of N in Rubisco would also be beneficial for a more targeted parameterisation approach.

The present study is the first to apply a process-based model to simulate NO_3-N leaching fluxes at Thetford, and potentially the first that has attempted to apply a process-based model centred around ecophysiological processes to simulate NO_3-N leaching fluxes from a temperate forest ecosystem. The failure to parameterise the model using observed NO_3-N leaching fluxes was ultimately related to the reliance of soil biochemical and hydrological processes for accurate simulation of ecophysiological processes. The interception simulation relies on accurate internal calculations of LAI (Running & Coughlan, 1988). Leaf Area Index is one of the most important internal calculations of Biome-BGCMuSo, and it is calculated from the carbon available for leaf growth, nitrogen availability, and water availability (Running & Gower, 1991). The calculated LAI is then used for photosynthesis, respiration, evaporation and soil variables (Running & Gower, 1991). Estimates of LAI, including those measured at Thetford Forest, have typically shown *P. sylvestris* to have an LAI of 1.5-5 m^2 m^{-2} (Goude *et al.*, 2019; Bequet *et al.*, 2012; Flechard *et al.*, 2011; Roberts *et al.*, 1982). In the outputs of the EPC + Soil parameterisation procedure, simulated values of LAI were typically below 1.5-5 m^2 m^{-2} even before mortality occurred, and interception was typically underestimated. Patterns of simulated deep soil drainage typically followed observed values but were either underestimated or overestimated, likely due to the inaccuracies in modelled interception.

Based on detailed diagnosis of model inaccuracies, a framework for generating parameters when simulating NO_3-N leaching fluxes in temperate pine forest is proposed. Users are advised to follow the guidelines below for operating Biome-BGCMuSo or other ecophysiological-focused models for the simulation of soil variables:

1. Apply to sites with sufficient long-term data. A minimum of 10 years of observed data is advised. The unexpected mortality events in the present case study generally did not occur until 9+ years into the study period. Had only 9

years of observed data been available, the outputs may have been misinterpreted.

2. Calibrate based on the variables related to the fundamental processes represented rather than additional variables later added to the model during further development. Parameterisation should be conducted using physiological outputs such as growth respiration or gross primary productivity. In Biome-BGCMuSo, physiological outputs underpin the whole model, whereas the soil-related outputs, such as NO_3-N leaching, could be considered secondary products. Parameterisation based on NO_3-N leaching fluxes here was unsuccessful due to a need to improve parameterisation of the photosynthesis-related parameters before others.

3. Conduct a sensitivity analysis over multiple decades rather than a few years. For example, the first sensitivity analysis in this study, performed only for 2015-2019, selected multiple parameters influential to NO_3-N leaching fluxes while asecond sensitivity analysis performed over the 25 year observation period identified four parameters that could be subjected to a calibration procedure requiring less computational power.

4. Understand interactions between parameters. In such a complex model, parameter interactions are likely to occur, and here, some interaction between

the fraction of N in Rubisco and the canopy light extinction coefficient on the growth respiration outputs was observed.

5. Implement a calibration procedure for both input parameter files.

6. Ensure the drivers behind model simulations align with physical understanding. Here, the growth respiration and leaf area index are visualised to confirm and identify the simulated mortality events occurring in simulations using the parameter sets that achieved the best performance according to the chosen objective functions. in years where NO_3-N leaching fluxes aligned with observed values, NO_3-N leaching in the simulation was driven by mortality-induced increases in litterfall N inputs, whereas the observed NO_3-N leaching fluxes were not.

7. Resolve detected issues by sequentially checking that the model is simulating processes accurately at each stage of the model workflow. Visualisation of LAI and growth respiration checks that the model correctly simulates vegetation growth. Checking the hydrological parameters, particularly interception, ensures that LAI calculations have passed into the next stage of the calculations correctly. Visualisation of the foliar and litterfall nutrients checks that the flux of nutrients between canopy and soil pools is simulated accurately.

If Biome-MGCMuSo can be successfully parameterised to simulated NO_3-N leaching fluxes, potential applications are not just land-use change scenarios. Tree species change requires an additional ecophysiological parameterisation procedure to enable accurate observations of a new tree species. Application to investigate future leaching fluxes under future climate scenarios, where land use remains unchanged, would only require the generation of new meteorological and carbon dioxide input files, a simpler procedure than changing ecophysiological and soil parameters.

5. Conclusions

An ecophysiological-based coupled biochemical-hydrological model was applied to simulate NO_3-N leaching fluxes from a temperate *P. sylvestris* forest located in a region with high N deposition inputs and prone to periods of heatwaves and drought. The model did not simulate NO_3-N leaching fluxes close to observed values. This was partly attributed to two parameters critical to the photosynthesis calculations at the study site. The parameter ranges for these parameters were taken from the User Manual given the lack of robust observational constraints for temperate, mature *P.sylvestris*. The parameterisation of the two key parameters could be improved by measurement of carboxylation rate at the study site. The suggested framework identifies the importance of parameterising such a model using observations of the variables that underpin the model's functioning, in this case, respiration. The model may successfully model NO_3-N leaching fluxes if the parameterisation of ecophysiological processes are improved. The model could, therefore, best be applied at sites where both CO_2 fluxes and NO_3-N leaching fluxes have been measured over 10+ years. Once validated, the model has the potential to test the response of NO_3-N leaching to climate, air pollution and land-use change scenarios.

References

Arnold, J.G., Srinivasan, R., Muttiah, J., Williams, J.R. (1998) Large area hydrologic modeling and assessment part I: model development. *Journal of the American Water Resources Association*, 34, 73-89.

Beeck, M.O.D., Gielen, B., Jonckheere, I., Samson, R., Janssens, I. A., Ceulemans, R. (2009) Needle age-related and seasonal photosynthetic capacity variation is negligible for modelling yearly gas exchange of a temperate Scots pine forest. *Biogeosciences Discuss*, 6, 9737-9780.

Bell, V. A., Naden, P. S., Tipping, E., Davies, H. N., Carnell, E., Davies, J. A. C., Dore, A. J., Dragosits, U., Lapworth, D. J., Muhammed, S. E., Quinton, J. N., Stuart, M., Tomlinson, S., Wang, L., Whitmore, A. P., Wu, L. (2021) Long term simulations of macronutrients (C, N and P) in UK freshwaters. *Science of the Total Environment*, 776, 145813.

Bequet, R., Kint, V., Campioli, M., Vansteenkiste, D., Muys, B., Ceulemans, R. (2012) Influence of stand, site and meteorological variables on the maximum leaf area index of beech, oak and Scots pine. *European Journal for Forest Research*, 131, 283-295.

Brunner, I., & Sperisen, C. (2013). Aluminum exclusion and aluminum tolerance in woody plants. *Frontiers in Plant Science*, 4, 172.

Cammarano, D., Basso, B., Holland, J., Gianinetti, A., Baronchelli, M., Ronga, D. (2021) Modeling spatial and temporal optimal N fertilizer rates to reduce nitrate leaching while improving grain yield and quality in malting barley. *Computers and Electronics in Agriculture*, *182*, 105997.

Carnell EJ, Thomas IN, Tomlinson SJ, Leaver D, Dragosits U. (n.d.). *The spatial distribution of ammonia, methane and nitrous oxide emissions from agriculture in the UK 2018 Annual Report to Defra.* Wallingford: UKCEH, Available: https://nora.nerc.ac.uk/id/eprint/529855/1/N529855CR.pdf (Accessed March 2023)

Chan, T., Berninger, F., Kolari, P., Nikinmaa, E., Hölttä, T. (2018) Linking stem growth respiration to the seasonal course of stem growth and GPP of Scots pine. *Tree Physiology*, 38, 1356–1370.

Chiesi, M., Chirici, G., Marchetti, M., Hasenauer, H., Moreno, A., Knohl, A., Matteucci, G., Pilegaard, K., Granier, A., Longdoz, B., Maselli, F. (2016) Testing the applicability of BIOME-BGC to simulate beech gross primary production in Europe using a new continental weather dataset. *Annals of Forest Science*, 73, 713–727.

Chiesi, M., Maselli, F., Moriondo, M., Fibbi, L., Bindi, M., Running, S. W. (2007) Application of BIOME-BGC to simulate Mediterranean forest processes. *Ecological Modelling*, 206, 179–190.

Cienciala, E., Tatarinov, F. A. (2006) Application of BIOME-BGC model to managed forests: 2. Comparison with long-term observations of stand production for major tree species. *Forest Ecology and Management*, 237, 252–266.

Cosby, B.J., Ferrier, R.C., Jenkins, A., Emmett, B.A., Wright, R.F., Tietema, A. (1997) Modelling the ecosystem effects of nitrogen deposition: Model of Ecosystem Retention and Loss of Inorganic Nitrogen (MERLIN). Hydrology and earth system sciences, 1, 137-158.

De Filippis, G., Ercoli, L., Rossetto, R. (2021) A spatially distributed, physically-based modeling approach for estimating agricultural nitrate leaching to groundwater. *Hydrology*, 8, 1–25.

Dise, N. B., Rothwell, J. J., Gauci, V., van der Salm, C., de Vries, W. (2009) Predicting dissolved inorganic nitrogen leaching in European forests using two independent databases. *Science of The Total Environment*, 407, 1798–1808.

Environment Agency (2019) *Check for Drinking Water Safeguard Zones (SgZs) and Nitrate Vulnerable Zones (NVZs) map.* Available: https://environment.data.gov.uk/farmers/ (Accessed November 2023)

Etzold, S., Ferretti, M., Reinds, G. J., Solberg, S., Gessler, A., Waldner, P., Schaub, M., Simpson, D., Benham, S., Hansen, K., Ingerslev, M., Jonard, M., Karlsson, P. E., Lindroos, A. J., Marchetto, A., Manninger, M., Meesenburg, H., Merilä, P., Nöjd, P., de Vries, W. (2020) Nitrogen deposition is the most important environmental driver of growth of pure, even-aged and managed European forests. *Forest Ecology and Management*, *458*, 117762.

Flechard, C. R., Nemitz, E., Smith, R. I., Fowler, D., Vermeulen, A. T., Bleeker, A., Erisman, J. W., Simpson, D., Zhang, L., Tang, Y. S., Sutton, M. A. (2011) Dry deposition of reactive nitrogen to European ecosystems: a comparison of inferential models across the NitroEurope network. Atmospheric Chemistry and Physics, 11, 2703–2728.

Forsius, M., Posch, M., Holmberg, M., Vuorenmaa, J., Kleemola, S., Augustaitis, A., Beudert, B., Bochenek, W., Clarke, N., de Wit, H. A., Dirnböck, T., Frey, J., Grandin, U., Hakola, H., Kobler, J., Krám, P., Lindroos, A. J., Löfgren, S., Pecka, T., Váňa, M. (2021) Assessing critical load exceedances and ecosystem impacts of anthropogenic nitrogen and sulphur deposition at unmanaged forested catchments in Europe. *Science of the Total Environment*, 753, 141791.

Futter, M.N., Ring, E., Högbom, L., Entenmann, S., Bishop, K.H. (2010) Consequences of nitrate leaching following stem-only harvesting of Swedish forests are dependent on spatial scale. Environmental pollution, 158, 3552-3559.

Garcia-Prats, A., González-Sanchis, M., Del Campo, A. D., Lull, C. (2018) Hydrology-oriented forest management trade-offs. A modeling framework coupling field data, simulation results and Bayesian Networks. *Science of the Total Environment*, *639*, 725–741.

Goude, M., Nilsson, U., Holmström, E. (2019) Comparing direct and indirect leaf area measurements for Scots pine and Norway spruce plantations in Sweden. *European Journal for Forest Research* 138, 1033–1047.

Gundersen, P., Schmidt, I. K., Raulund-Rasmussen, K. (2006) Leaching of nitrate from temperate forests ⍰ effects of air pollution and forest management. *Environmental Reviews*, 14, 1–57.

Hansen, K., Rosenqvist, L., Vesterdal, L., Gundersen, P. (2007) Nitrate leaching from three afforestation chronosequences on former arable land in Denmark. *Global Change Biology*, 13, 1250–1264.

Hidy, D., Barcza, Z., Haszpra, L., Churkina, G., Pintér, K., Nagy, Z. (2012) Development of the Biome-BGC model for simulation of managed herbaceous ecosystems. *Ecological Modelling*, *226*, 99–119.

Hidy, D., Barcza, Z., Hollós, R., Thornton, P.E., Running, S., Fodor, N. (2021) *User's guide for Biome-BGCMuSo 6.2*. Available: https://nimbus.elte.hu/bbgc/documentation.html (Accessed March 2023)

Hidy, D., Barcza, Z., Marjanovic, H., Ostrogovic Sever, M. Z., Dobor, L., Gelybó, G., Fodor, N., Pintér, K., Churkina, G., Running, S., Thornton, P., Bellocchi, G., Haszpra, L., Horváth, F., Suyker, A., Nagy, Z. (2016) Terrestrial ecosystem process model Biome-BGCMuSo v4.0: summary of improvements and new modeling possibilities. *Geosciences Model Development*, 9, 4405-4437.

Huang, B., Yang, Y., Li, R., Zheng, H., Wang, X., Wang, X., Zhang, Y. (2022) Integrating Remotely Sensed Leaf Area Index with Biome-BGC to Quantify the Impact of Land Use/Land Cover Change on Water Retention in Beijing. *Remote Sensing*, 14, 743.

Jerabkova, L., Prescott, C. E., Titus, B. D., Hope, G. D., Walters, M. B. (2011) A meta-analysis of the effects of Clearcut and variable-retention harvesting on soil nitrogen fluxes in boreal and temperate forests. *Canadian Journal of Forest Research*, 41, 1852–1870.

Kubin, E. (1998). Leaching of nitrate nitrogen into the groundwater after clear felling and site preparation. *Boreal Environment Research*, 3, 3-8.

Laurent, F., & Ruelland, D. (2011) Assessing impacts of alternative land use and agricultural practices on nitrate pollution at the catchment scale. *Journal of Hydrology*, 409, 440–450.

Leitner, S., Minixhofer, P., Inselbacher, E., Keiblinger, K.M., Zimmermann, M., Zechmeister-Boltenstern, S. (2017) Short-term soil mineral and organic nitrogen fluxes during moderate and severe drying–rewetting events. *Applied Soil Ecology*, 114, 28-33.

MacDonald, J. A., Dise, N. B., Matzner, E., Armbruster, M., Gundersen, P., Forsius, M. (2002) Nitrogen input together with ecosystem nitrogen enrichment predict nitrate leaching from European forests. *Global Change Biology*, 8, 1028–1033.

Mannerkoski, H., Finér, L., Piirainen, S., Starr, M. (2005) Effect of clear-cutting and site preparation on the level and quality of groundwater in some headwater catchments in eastern Finland. *Forest Ecology and Management*, 220, 107–117.

Mao, F., Li, P., Zhou, G., Du, H., Xu, X., Shi, Y., Mo, L., Zhou, Y., Tu, G. (2016) Development of the BIOME-BGC model for the simulation of managed Moso bamboo forest ecosystems. *Journal of Environmental Management*, *172*, 29–39.

Michalzik, B., Tipping, E., Mulder, J., Lancho, J. F. G., Matzner, E., Bryant, C. L., Clarke, N., Lofts, S., Esteban, M. A. V. (2003) Modelling the production and transport of dissolved organic carbon in forest soils. Biogeochemistry, 66, 241–264.

Nieder, R., Benbi, D.K., Scherer (2011) Fixation and defixation of ammonium in soils: a review. Biology and fertility of soils, 47, 1-14.

Norman, J., Jansson, P. E., Farahbakhshazad, N., Butterbach-Bahl, K., Li, C., Klemedtsson, L. (2008) Simulation of NO and N2O emissions from a spruce forest during a freeze/thaw event using an N-flux submodel from the PnET-N-DNDC model integrated to CoupModel. *Ecological Modelling*, 216, 18–30.

Nunes, L., Gower, S. T., Peckham, S. D., Magalhães, M., Lopes, D., Rego, F. C. (2015). Estimation of productivity in pine and oak forests in northern Portugal using Biome-BGC. *Forestry*, 88, 200–212.

Olsson, B. A., Bergholm, J., Alavi, G., Persson, T. (2022). Effects of long-term N fertilization on nitrate leaching and vegetation responses in a spruce stand after severe wind damage. *Forest Ecology and Management*, *520*.

Pietsch, S.A., Hasenauer, H., Kučera, J., Čermák, J. (2003) Modelling effects of hydrological changes on the carbon and nitrogen balance of oak floodplains. Tree physiology, 23, 735-746.

Pietsch, S.A., Hasenauer, H., Thornton, P.E. (2005) BGC-model parameters for tree species growing in central European forests. Forest ecology and management, 211, 264-295.

Pitman, R. M., Vanguelova, E. I., Benham, S. E. (2010) The effects of phytophagous insects on water and soil nutrient concentrations and fluxes through forest stands of the Level II monitoring network in the UK. *Science of The Total Environment*, 409, 169–181.

Pohlert, T., Huisman, J. A., Breuer, L., Frede, H.-G. (2007) Integration of a detailed biogeochemical model into SWAT for improved nitrogen predictions—Model development, sensitivity, and GLUE analysis. *Ecological Modelling*, 203, 215–228.

Posch, M., Reinds, G.J. (2009) A very simple dynamic soil acidification model for scenario analyses and target load calculations. Environmental modelling and software, 24, 329-340.

Roberts, J., Pitman, R. M., Wallace, J. S. (1982) A Comparison of Evaporation from Stands of Scots Pine and Corsican Pine in Thetford Chase, East Anglia. *Journal of Applied Ecology*, 19, 859–872.

Rosenqvist, L., Hansen, K., Vesterdal, L., van der Salm, C. (2010) Water balance in afforestation chronosequences of common oak and Norway spruce on former arable land in Denmark and southern Sweden. *Agricultural and Forest Meteorology*, 150, 196–207.

RoTAP. (2012) *Review of Transboundary Air Pollution: Acidification, Eutrophication, Ground Level Ozone and Heavy Metals in the UK*. Wallingford: UKCEH Available: https://nora.nerc.ac.uk/id/eprint/18629/1/N018629CR.pdf (Accessed December 2023)

Rowe, E.C., Tipping, E., Posch, M., Oulehle, F., Cooper, D.M., Jones, T.G., Burden, A., Hall, J., Evans, C.D. (2014) Predicting nitrogen and acidity effects on long-term dynamics of dissolved organic matter. Environmental pollution, 184, 271-282.

Rowe, E.C., Wamelink, G.W.W., Smart, S.M., Butler, A., Henrys, P.A., van Dobben, H.F., Reinds, G.J., Evans, C.D., Kros, J., de Vries, W. (2015) *Field survey-based models for exploring nitrogen and acidity effects on plant species diversity and assessing long-term critical loads*. In: Critical loads and dynamic risk assessments. Nitrogen, acidity and metals in terrestrial and aquatic ecosystems. US: Springer

Running, S. W., Coughlan, J. C. (1988) A general model of forest ecosystem processes for regional applications I. hydrologic balance, canopy gas exchange and primary production processes. In *Ecological Modelling*, 42, 125-154.

Running, S.W., Gower, S.T. (1991) A general model of forest ecosystem processes for regional applications II. Dynamic carbon allocation and nitrogen budgets. *Tree Physiology*, 9, 147-160.

Sawicka, K., Rowe, E.C., Evans, C.D., Monteith, D.T., Vanguelova, E.I., Wade, A.J., Clark, J.M. (2017) Modelling impacts of atmospheric deposition and temperature on long-term DOC trends. *Science of the Total Environment*, 578, 323-336.

Schmid, S., Zierl, B., Bugmann, H. (2006) Analyzing the carbon dynamics of central European forests: Comparison of Biome-BGC simulations with measurements. *Regional Environmental Change*, 6, 167–180.

Schmitz, A., Sanders, T.G.M., Bolte, A., Bussotti, F., Dirnböck, T., Peñuelas, J., Pollastrini, M., Prescher, A-K., Sardans, J., Verstraeten, A., de Vries, W. (2024) *Responses of forest ecosystems in Europe to decreasing nitrogen deposition*. In: *Atmospheric nitrogen deposition to global forests*. The United States: Academic Press.

Srinet, R., Nandy, S., Patel, N.R., Padalia, H., Watham, T., Singh, S.K., Chauhan, P. (2023) Simulation of forest carbon fluxes by integrating remote sensing data into biome-BGC model. *Ecological Modelling*, 475, 110185.

Stangl, Z. R., Tarvainen, L., Wallin, G., Marshall, J. D. (2022) Limits to photosynthesis: seasonal shifts in supply and demand for CO2 in Scots pine. *New Phytologist*, 233, 1108–1120.

Tietema, A., Boxman, A. W., Bredemeier, M., Emmett, B. A., Moldan, F., Gundersen, P., Schleppi, P., Wright, R. F. (1998) Nitrogen saturation experiments (NITREX) in coniferous forest ecosystems in Europe: A summary of results. *Environmental Pollution*, 102, 433–437.

van der Salm, C., de Vries, W., Reinds, G. J., Dise, N. B. (2007) N leaching across European forests: Derivation and validation of empirical relationships using data from intensive monitoring plots. *Forest Ecology and Management*, 238, 81–91.

Vanguelova, E., Barsoum, N., Benham, S., Broadmeadow, M., Moffat, A. (2007) *Ten years of intensive environmental monitoring in British forests*. Information note FCIN088, UK: Forestry commission.

Vanguelova, E.I., R.P., Benham, S. (2024) *Responses of forest ecosystems to nitrogen deposition in the United Kingdom*. In: Atmospheric nitrogen deposition to global forests. The United States: Academic Press.

Vanguelova, E.I., Pitman, R.M. (2019) Nutrient and carbon cycling along nitrogen deposition gradients in broadleaf and conifer forest stands in the east of England. Forest Ecology and Management, 447, 180-194.

Vermeulen, M. H., Kruijt, B. J., Hickler, T., Kabat, P. (2015) Modelling short-term variability in carbon and water exchange in a temperate Scots pine forest. *Earth System Dynamics*, 6, 485–503.

Wade, A. J., Durand, P., Beaujouan, V., Wessel, W. W., Raat, K. J., Whitehead, P. G., Butterfield, D., Rankinen, K., Lepisto, A. (2002) A nitrogen model for European catchments: INCA, new model structure and equations. *Hydrology and Earth System Sciences*, 6, 559–582.

White, M. A., Thornton, P. E., Running, S. W., Nemani, R. R. (2000) Parameterization and Sensitivity Analysis of the BIOME-BGC Terrestrial Ecosystem Model: Net Primary Production Controls. In *Earth Interactions*, 4, 1-85.

Whitehead, P.G., Wilson, E.J., Butterfield, D. (1998) A semi-distributed Integrated Nitrogen model for multiple source assessment in Catchments (INCA): Part 1- model structure and process equations. Science of the total environment, 210, 547-558.

Xie, D., Duan, L., Du, E., de Vries, W. (2024). *Indicators and thresholds for nitrogen saturation in forest ecosystems*. In: Atmospheric Nitrogen Deposition to Global Forests: Spatial Variation, Impacts, and Management Implications, The United States: Academic Press.

Ye, J-S., Reynolds, J.F., Maestre, F.T., Li, F-M. (2016) Hydrological and ecological responses of ecosystems to extreme precipitation regimes: A test of empirical-based hypotheses with an ecosystem model. *Perspectives in Plant Ecology, Evolution and Systematics*, 22, 36-46.

Zha, T., Kellomäki, S., Wang, K. Y., Ryyppö, A., Niinistö, S. (2004) Seasonal and annual stem respiration of scots pine trees under boreal conditions. *Annals of Botany*, 94, 889–896.

Chapter 5

Discussion

The present thesis aimed to first explore the interactions between tree species, tree age, soil type, N deposition, and land use change as drivers of NO_3-N leaching. Whilst several limitations have been encountered, the research here has identified land use change scenarios where the effect on NO_3-N leaching has not previously been studied in detail and understood.

1. Interactions between drivers of NO_3-Nleaching in European forest ecosystems

Chapter 2 identified throughfall TIN deposition, tree species, former land use and soil type as the best combination of predictors of NO_3-N leaching from forest ecosystems. Significant associations were noted between tree species, soil type, former land use, and tree age, complicating conclusions on the influence of each driver on NO_3-N leaching fluxes. Studies that only investigate one factor at a time are valuable to disentangle the magnitude of different drivers (e.g. Rothe *et al*., 2002), but drivers are always confounded. A large-scale systematic study focusing on evaluating multiple sites under suites of factors, e.g. broadleaves planted on alfisol formerly used for arable practices or conifer planted on cambisols formerly managed as heathland, would be an ideal approach to gain a robust understanding of NO_3-N leaching fluxes from forests planted on different soil types with different land use histories.

The ICP Forest Level II long-term intensive monitoring network of forest ecosystems has focused mainly on primary forests or rotations, with poor representation of diverse land-use histories (Appendix 1). Chapter 2 demonstrated that including a more diverse range of land use histories from reviewed literature generated a dataset with a more varied response of NO_3-N leaching to the ICP Forests Level II and IFEF databases. Such intensive monitoring would benefit from covering more afforested arable land and heathland/grassland and forest successional rotations to answer questions on forest management and policy moving forward.

The hypothesis that former land use would affect the N retention capacity of forest soils was not accepted because no significant interaction between throughfall TIN and former land use was observed overall. However, lower N-leaching was observed from heathland afforested with conifers compared to secondary rotations, so former land use is still important to fully understand NO_3-N leaching from forest ecosystems. Chapter 2 identified groups with a poor correlation between NO_3-N leaching and throughfall TIN deposition, which included broadleaf forests, afforested arable soils and forests planted on alfisols. There were associations between these three groups, with afforested arable soils commonly being alfisols and broadleaves more commonly planted on alfisols than conifers. The proposed explanation of the wide variation in NO_3-N leaching in these categories was hypothesised to be related to the availability of other nutrients. Still, there could be other explanations, including those related to the hydrogeology of alfisols.

Podzols are characterised by a spodic horizon, formed following the inhibition of litter decomposition and destruction of silicate and clay minerals, which releases heavy metals, including Al and Fe, by acidic conditions, a process which can take over 4500 years (Sauer et al., 2007; Mokma et al., 2004). Cambisols are soils with little profile differentiation, characterised by a cambic horizon. This horizon has a sandy loam or sandy texture, a soil aggregate structure greater than or equal to 50 % of the soil volume and a thickness greater than or equal to 15 cm (IUSS WRB, 2022). Alfisols, in contrast, are characterised by the presence of an argillic horizon, a B horizon with a base saturation greater than 35 % and a greater percentage of clay than the horizons above it (Soil Survey Staff, 2022; Bockheim et al., 2014). Key forming factors include organic matter accumulation, clay eluviation-illuviation, and A horizon base cation accumulation (Aide, 2021), commonly attributed to historical cover with grasses or deciduous forest (Bockheim & Gennadiyev). Clay eluviation-illuviation was increased in soils with a greater slope in a Danish study (Dalsgaard et al., 1981), highlighting hydrological influences on alfisol development and a potential association between alfisols and sloped areas. The more complex nutrient cycling and hydrogeological processes in alfisols could lead to greater variation in water fluxes and, therefore, a response to NO_3-N leaching fluxes. However, water fluxes were commonly not reported separately in conjunction with NO_3-N leaching fluxes, so discerning the role of water fluxes vs. nutrient concentrations in NO_3-N leaching fluxes from forest ecosystems remains a challenge. Chapter 2 also highlighted the effect of tree age on NO_3-N leaching fluxes, but it was not discerned whether hydrological or nutrient concentration changes were the greater drivers.

2. Conifer-to-broadleaf conversion scenario highlighted the interaction between hydrological and biochemical processes

The role of hydrological fluxes in NO_3-N leaching was again highlighted in Chapter 3, which provides the first observations on the effect of conifer-to-broadleaf conversion on changes in NO_3-N leaching fluxes. The seasonal pattern in NO_3-N leaching fluxes for each chronosequence stage reflected the patterns in deep soil drainage fluxes more so than in deep soil solution NO_3-N concentrations. Whilst deep soil solution NO_3-N concentrations were greater in stands 10-13 years post-conversion than 5-8 years, the lower water flux meant that NO_3-N leaching fluxes were nearly identical. However, based on the observations from Chapter 2, it would be expected that NO_3-N leaching fluxes will increase with tree age, and the full potential of the conifer-to-broadleaf conversion process to increase NO_3-N leaching may not have been captured. These findings lead to the question: at what stages in a forest's lifecycle do hydrological fluxes vs biochemical fluxes drive NO_3-N leaching fluxes? The failed attempt to parameterise a coupled hydrological-biochemical model in Chapter 4 limits conclusions on the long-term impact of the conifer-to-broadleaf conversion on NO_3-N leaching fluxes.

The soil types present in the chronosequence studied here were arenosols and leptosols. Leptosols are defined by the presence of continuous rock starting less than 25 cm from the surface or soil up to 75 cm depth with <20 % volume of fine earth (IUSS, 2022) and form from cambisols (Kowalska *et al.*, 2019). Arenosols are similar to cambisols, with their texture class being loamy sand or sand, consisting of layers <15 cm thick of fine texture and layers with greater than or equal to 40 % coarse fragments (IUSS, 2022). The positive linear relationship observed in the chronosequence study in Chapter 3 between throughfall TIN and NO_3-N leaching aligns with the strong correlation between throughfall TIN and NO_3-N leaching observed for cambisols in Chapter 2. As the study area was typically flat and the soil types typically sandy without clay elements, water movement through the soil profile would be expected to be dominated by downward fluxes.

The rates of nitrification and mineralisation in the 10-30 cm soil layer were not related to throughfall inputs or properties of the 0-10 cm layer, nor were they related to soil NO_3-N leaching fluxes from the deeper soil layer, but were related to the C:N ratio and pH of the 10-30 cm layer. As NO_3-N leaching fluxes were related to throughfall DOC and throughfall NO_3-N inputs and the C:N ratio of soils at 0-10 cm depth, the difficulty in fully explaining the mechanisms driving deep soil NO_3-N leaching fluxes in this study lies with the fact that net nitrification, mineralisation and pH were not measured at 0-10 cm but only 10-30 cm. Soil solution dissolved organic carbon at 0-10, 10-30 and 30-90 cm could also have facilitated a better understanding of the mechanism by which throughfall inputs drove NO_3-N leaching fluxes in the present study.

3. The long-term study of conifer-to-broadleaf conversion was limited by the difficulty in parameterising a coupled biochemical-hydrological model.

Considering the importance of hydrological inputs in driving NO_3-N leaching across Thetford Forest, investigated in this thesis, any model selected to simulate long-term NO_3-N leaching needed to have a strong representation of both hydrological and biochemical processes, hence the selection of Biome-BGCMuSo in Chapter 4. Kiese *et al*. (2011) used the Forest-DNDC model, a similar coupled hydrological-biochemical model, to look at fluxes from German forests, achieving moderate model performance for simulating water seepage ($R^2 = 0.57$), NO_3-N concentrations ($R^2 = 0.52$) and NO_3-N leaching ($R^2 = 0.51$). Forest-DNDC uses daily meteorological data to drive biomass development, N uptake and litterfall (Li *et al.*, 2000) so unlike Forest-BGC and Biome-BGCMuSo, simulation of NO_3-N leaching is not as dependent on the parameterisation of C cycle dynamics alongside N dynamics. Whilst the processes represented in Biome-BGCMuSo may allow it to simulate NO_3-N leaching better following changes in forest management, its tight interactions between N and C cycling were what potentially made it difficult to determine parameters that did not result in tree mortality during the study period.

Identifying the root distribution parameter as individually influential to the Biome-BGCMuSo NO_3-N leaching outputs is important in the conifer-to-broadleaf conversion. The root distribution parameter determines which soil layers meet vegetation transpiration demand (Hidy *et al.*, 2022) and, therefore, affects the calculations of hydrological fluxes. Elevated nitrogen deposition and excess soil N affect root biomass and architecture, but the response differs between arbuscular vs ectomycorrhizal trees (Ma *et al.*, 2021, Linder, *et al.*, 2018). Planting broadleaves, especially arbuscular mycorrhizal species, on former coniferous plantations, which were likely associated with ectomycorrhizas, will likely see the development of broadleaves on soil with a different stratification of nutrients to soils that have been under broadleaf cover, which may affect rooting patterns and biomass. In a three-month greenhouse experiment, the influence of the distribution of nitrogen was tested on the development of willow tree roots, observing significant differences in root biomass and soil water content between treatments reflecting the distribution of nutrients in coniferous vs broadleaf soils (Appendix 14). Connections between rooting patterns, tree growth and health, land use change and soil water fluxes require further field investigations. Changes in fine root biomass and distribution have been previously studied in the reverse scenario of broadleaf-to-conifer conversion, demonstrating higher fine root biomass in broadleaves and stronger responses in fine root plasticity to soil chemistry in broadleaf systems (Förster *et al*., 2021). Validating of field observations of root distribution to the Biome-BGCMuSo root distribution parameter could facilitate NO_3-N simulations following land use change.

4. Recommendations for forest management

Until the long-term effect of conifer-to-broadleaf conversion can be established, alternative systems to reduce NO_3-N leaching from forests should be explored. For conifers, Chapter 2 demonstrated that NO_3-N leaching was lower on former heathlands than on secondary rotations. Harvesting trees in forest ecosystems exposed to chronically elevated N deposition levels can lead to reductions of N in the ecosystem due to export of N with the wood extractionand, therefore, reduce NO_3-N leaching (Gundersen *et al*., 2006). The potential of a forestry rotational system of managing former conifer plantations as heathland for decades before replanting, alongside heathland afforestation to maintain no net loss of forest cover, could, therefore, be explored to mitigate elevated NO_3-N leaching. Such a system would also manage the trade-off between afforestation and maintaining open habitats for biodiversity, such as ground-nesting birds (Forestry Commission, 2016).

Applying soil amendments to reduce elevated NO_3-N leaching following elevated deposition or harvesting has received some attention. Biochar application in forestry can reduce NO_3-N concentrations in forest drainage waters significantly post-harvest

(Lafdani *et al.*, 2020) and reduces N_2O emissions due to improvements in soil physical and chemical properties such as pH and aggregate stability (Li *et al.*, 2018). Biochar application on agricultural soils can reduce NO_3-N leaching by >26 % (Borchard *et al.*, 2019). Amendments could also create more favourable conditions to establish broadleaved trees on conifer sites, for example, wood ash application promoted the growth of oak and birch trees on felled conifer sites (Pitman *et al.*, 2024). Ammendments can also expand the options for species choice. For example, Kazda & Pichler (1998) demonstrated that *Q. robur* was a more suitable species to replace conifers with than *F. sylvatica* in upper Austria due to its better adaptation to the acidic soils in the study area. Applying amendments to increase soil pH could perhaps improve the success of planted *F. sylvatica*. However, increasing soil pH has been associated with increasing the activity of nitrifying ammonia oxidising bacteria (Nugroho *et al.*, 2007; Ste-Marie & Paré, 1999). Biochar ammendments also act as a labile carbon source, and whilst the source is different, here increasing dissolved organic carbon inputs stimulated NO_3-N leaching fluxes across Thetford Forest. Establishing indicators which inform scenarios where labile carbon applications would stimulate vs mitigate NO_3-N leaching fluxes is needed to inform forest management practices.

5. The role of interdisciplinary research in understanding NO_3-N leaching from forest ecosystems

The parameterisation of the coupled biochemical-hydrological model in Chapter 4 was unsuccessful due to the focus on soil biochemical cycling and failure to parameterise two critical physiological parameters, which could be improved from a review of plant physiological literature. Several questions from this thesis centre around connecting biochemical processes to soil physical and geological processes. The variation in NO_3-N leaching observed in forested alfisols could be better understood using physical, process-based soil water transport models such as HYDRUS, and analysing relationships between NO_3-N leaching and the availability of other nutrients. Quantifying the impact of NO_3-N leaching fluxes from Thetford Forest on groundwater, both following prolonged periods of deposition and following forestry operations, will require hydrogeological modelling of NO_3-N transport in the chalk aquifer, including in areas with superficial deposits of clay, which can affect vertical flows. This thesis highlights areas where interdisciplinary collaboration could improve research, providing the next user easily understands each component's outputs.

References

Aide, M. (2021) Soil Genesis of Four Alfisols Established in Oak Hickory Forests along Drainages into the Mississippi River in Southeastern Missouri, USA. Journal of Geoscience and Environment Protection, 09, 124–143.

Bockheim, J. G., Gennadiyev, A. N., Hartemink, A. E., Brevik, E. C. (2014) Soil-forming factors and Soil Taxonomy. Geoderma, 226–227, 231–237.

Borchard, N., Schirrmann, M., Cayuela, M. L., Kammann, C., Wrage-Mönnig, N., Estavillo, J. M., Fuertes-Mendizábal, T., Sigua, G., Spokas, K., Ippolito, J. A., Novak, J. (2019) Biochar, soil and land-use interactions that reduce nitrate leaching and N2O emissions: A meta-analysis. Science of the Total Environment, 651, 2354–2364.

Chen, W., & Peng, S. (2020). Land-use legacy effects shape microbial contribution to N2O production in three tropical forests. Geoderma, 358, 113979.

Dalsgaard, K., Baastrup, E., Bunting, B.T. (1981). The influence of topography on the development of Alfisols on calcareous clayey till in Denmark. Catena, 8, 111-136.

Forestry Commission. (2016) Hockham Thetford Forest: Forest Plan. England: Forestry Commission.

Förster, A., Hertel, D., Werner, R., Leuschner, C. (2021) Belowground consequences of converting broadleaf to conifer forest: Comparing the fine root systems of European beech and Scots pine. Forest Ecology and Management, 496, 119457.

Gundersen, P., Schmidt, I.K., Raulund-Rasmussen, K. (2006) Leaching of nitrate from temperate forests- effects of air pollution and forest management. Environmental reviews, 14, 1-57.

Hidy, D., Barcza, Z., Hollós, R., Dobor, L., Ács, T., Zacháry, D., Filep, T., Pásztor, L., Incze, D., Dencsõ, M., Tóth, E., Merganičová, K., Thornton, P., Running, S., Fodor, N. (2022) Soil-related developments of the Biome-BGCMuSo v6.2 terrestrial ecosystem model. Geoscientific Model Development, 15, 2157–2181.

IUSS Working Group WRB. (2022) World Reference Base for Soil Resources 2022: International soil classification system for naming soils and creating legends for soil maps. Rome: FAO.

Kazda, M., & Pichler, M. (1998). Priority assessment for conversion of Norway spruce forests through introduction of broadleaf species. Forest Ecology and Management, 102, 245-258.

Kiese, R., Heinzeller, C., Werner, C., Wochele, S., Grote, R., Butterbach-Bahl, K. (2011). Quantification of nitrate leaching from German forest ecosystems by use of a process oriented biogeochemical model. Environmental Pollution, 159, 3203-3214.

Kowalska, J. B., Zaleski, T., Józefowska, A., Mazurek, R. (2019) Soil formation on calcium carbonate-rich parent material in the outer Carpathian Mountains – A case study. Catena, 174, 436–451.

Lafdani, E. K., Saarela, T., Laurén, A., Pumpanen, J., Palviainen, M. (2020) Purification of forest clear-cut runoff water using biochar: A meso-scale laboratory column experiment. Water (Switzerland), 12.

Li, C., Aber, J., Stange, F., Butterbach-Bahl, K., Papen, H. (2000) A process-oriented model of N2O and NO emissions from forest soils: 1. Model development. Journal of Geophysical Research Atmospheres, 105, 4369–4384.

Li, Y., Hu, S., Chen, J., Müller, K., Li, Y., Fu, W., Lin, Z., Wang, H. (2018) Effects of biochar application in forest ecosystems on soil properties and greenhouse gas emissions: a review. Journal of Soils and Sediments, 18, 546–563.

Ma, S., Geng, Q., Zhang, H., Bian, C., Chen, H.Y.H., Jiang, D., Xu, X. (2021) Global negative effects of nutrient enrichment on arbuscular mycorrhizal fungi, plant diversity and ecosystem multifunctionality. New Phytologist, 229, 2957-2969.

Mokma, D. L., Yli-Halla, M., Lindqvist, K. (2004) Podzol formation in sandy soils of Finland. Geoderma, 120, 259–272.

Nugroho, R. A., Röliing, W.F.M., Laverman, A.M., Verhoef, A. (2007). Low nitrification rates in acid Scots pine forest soils are due to pH-related factors. Microbial Ecology, 12, 89-97.

Pitman, R., Willoughby, I., & Sansby, R. (2024). Soil change and broadleaf tree growth 10 years after wood ash and brash co-application to a clearfelled lowland conifer site in Britain. Forestry, 97, 76-93.

Rothe, A., Huber, C., Kreutzer, K., Weis, W., & Stichler, W. (2002). Deposition and soil leaching in stands of Norway spruce and European Beech: Results from the Höglwald research in comparison with other European case studies. Plant and Soil, 240, 33-45.

Sauer, D., Sponagel, H., Sommer, M., Giani, L., Jahn, R., Stahr, K. (2007) Podzol: Soil of the year 2007. A review on its genesis, occurrence, and functions. In Journal of Plant Nutrition and Soil Science, 170, 581–597.

Soil Survey Staff (2022) Keys to taxonomy. 13th edn. Washington, D.C,: U.S. Government Printing Office, Ste-Marie, C., Paré, D. (1999) Soil, pH and N availability effects on net nitrification in the forest floors of a range of boreal forest stands. Soil Biology and Biochemistry, 31, 1579-1589.

Van Der Linde, S., Suz, L. M., Orme, C. D. L., Cox, F., Andreae, H., Asi, E., Atkinson, B., Benham, S., Carroll, C., Cools, N., De Vos, B., Dietrich, H. P., Eichhorn, J., Gehrmann, J., Grebenc, T., Gweon, H. S., Hansen, K., Jacob, F., Kristöfel, F., Bidartondo, M. I. (2018) Environment and host as large-scale controls of ectomycorrhizal fungi. Nature, 558, 243–248.

Limitations

Data availability limited this research at several points. In Chapter 2, there were limited data points for some former land use scenarios, such as secondary conifer rotations, and only four soil types could be investigated. The ICP Forests database had a poor representation of afforested land where long term NO_3-N leaching and deposition had been studied (Appendix 1). While the data suggests that former land use affects NO_3-N leaching in conifer ecosystems, strong conclusions can not be made. Long-term monitoring of soil solution in afforested soils is needed.

The field study in Chapter 3 benefitted from multiple replicates of each chronosequence stage and various measured variables, but the study was limited to sandy soil types. There was no representation of forests converted from conifers to broadleaves more than 13 years ago in the study area. . Exploration of the effect of conifer-to-broadleaf conversion on clayey soils is needed to understand whether the practice may be more or less suitable, more or less likely to generate a large peak in NO_3-N leaching on certain soil types.

Chapter 3 highlighted that NO_3-N leaching is sometimes difficult to measure, such as in soils where extraction of soil solution by lysimeters is limited by soil water content. NO_3-N leaching fluxes could not be calculated at the youngest and most recently felled stands, as the chloride mass balance method was unsuitable considering the disturbance and changes in topology. Quantification of NO_3-N losses immediately following harvesting and ground preparation is needed to compare the magnitude of the initial peak to potential future increases in NO_3-N leaching fluxes years after conversion. The variety of soil solution extraction instrumentation and methods to calculate water fluxes, and subsequently NO_3-N leaching fluxes, in the literature extracted in Chapter 2, likely contributed to the wide variation in NO_3-N leaching fluxes and may have masked some relationships. Although these issues still exist in databases such as ICP Forests where, despite guideance on standardised protocols, a variety of lysimetric methods are used.

Chapter 4 suggested that the parameterisation of a coupled biochemical-hydrological model could be improved by calibration with respiration-related variables. The Integrated Carbon Observation System (ICOS) network provides CO_2 measurements from a network of forested sites, adhering to strict protocols, and sometimes ICOS monitoring sites are co-located with ICP Forest Level II plots, which may have long-term NO_3-N leaching flux and deposition monitoring. The suggestion of calibrating the model with respiration variables to then apply to simulate soil biochemical processes is, therefore, not impossible. CO_2 flux was monitored 1996-2005 at Thetford Forest but the data was not readily available for use here. CO_2 fluxes are not available for many forest ecosystems in general. Even if parameterisation of Biome-BGCMuSo had been successful and the model could have been applied to simulate NO_3-N leaching fluxes post conifer-to-broadleaf conversion, validation would have been limited by only one year of observation data (Chapter 3). The forest management change would need to be simulated under multiple air pollution and climate change scenarios to be fully informative to management practices in the region.

Recommendations for future research

Whilst the main recommendation here is to expand the coverage of long-term monitoring to cover different afforestation scenarios, existing data from the ICP Forests database could be used to quantify relationships between throughfall TIN deposition, NO_3-N leaching and the availability of other nutrients such as PO_4 and base cations. Such quantification could assist in predictions of how afforested soils may respond to elevated deposition in areas with local sources of emissions. Quantifying relationships between NO_3-N leaching and above-ground variables, such as foliar nutrition and tree growth, could aid in understanding the contribution of NO_3-N leaching to observed foliar nutrient deficiencies across Europe (Jonard *et al*., 2015).

The conifer-to-broadleaf conversion scenario must be studied in relation to other soil types to guide management decisions. Rates of nitrification need to be measured across all seasons to improve the conceptual understanding of nutrient cycling processes in the conifer-to-broadleaf management scenario, vegetation species composition and biomass changes need to be understood, gaseous N emissions need to be quantified, and the changes in the composition of microbial communities could be better understood. There is already some literature investigating the effect of the conversion scenario on fungal community diversity, with conflicting conclusions. Schauer *et al*. (2023) showed that the alpha diversity of fungi decreased with the conversion of spruce to mixed spruce and beech stands, but Heine *et al*. (2019) showed an increase in fungal species abundance with the conversion of spruce to beech. Higher fungal species diversity would likely increase competition and nutrient demand, reducing NO_3-N leaching.

The application of a coupled hydrological-biochemical model was difficult. Nevertheless, Chapter 4 provided guidelines developed from a long trial-and-error period to streamline future attempts to model soil variables. The model could be tested at a co-located ICOS-ICP Forests monitoring sites.

References

Jonard, M., M., Fürst, A., Verstraeten, A., Thimonier, A., Timmermann, V., Potočić, N., Waldner, P., Benham, S., Hansen, K., Merilä, P., Ponette, Q., de la Cruz, A.C., Roskams, P., Nicolas, M., Croisé, L., Ingerslev, M., Matteucci, G., Decinti, B., Bascietto, M. Rautio, P. (2015) Tree mineral nutrition is declining in Europe. Global change biology, 21, 418-430.

Heine, J., Ottermanns, R., Schäffer, a., Roß-Nickoll, M. (2019) Forest conversion from Norway spruce to European beech increases species richness and functional structure of aboveground macrofungal communities. *Forest Ecology and Management*, 432, 522-533.

Schauer, B., Thorn, S., Blaschke, M., Kudernatsch, T. (2023) Conversion of pure spruce to mixed spruce beech stands: Effects on alpha and beta diversity of multiple taxonomic groups. *Forest Ecology and Management*, 545, 121297.

Conclusions

The present thesis aimed to investigate interactions between tree species, tree age, soil type, N deposition and land use change as drivers of NO_3-N leaching and to explore NO_3-N leaching following the increasingly common practice of conifer-to-broadleaf forest conversion. Chapter 2 identified differences in NO_3-N leaching between heathland afforested with conifers and secondary conifer rotations and high variability in the NO_3-N response of afforested arable land to N deposition inputs (Figure 1a). This observation suggested testing rotation systems where felled conifer rotations are managed as open, heathland habitats whilst planting on heathland to manage NO_3-N leaching at a landscape scale. Tree age effects on throughfall TIN and NO_3-N leaching fluxes were also evident, supporting the conclusions of existing literature.

The conclusions from Chapter 2 have implications for payment for ecosystem service schemes where enhanced water quality is cited as a benefit of afforestation. For example, the variation in NO_3-N leaching fluxes from arable land afforested with broadleaf forest suggests that there is not a full understanding of drivers of NO_3-N leaching from broadleaved forest, and there may be situations in which broadleaved forest is not beneficial to water quality. Chapter 3 also provided evidence to caution against the assumption that planting broadleaf forests will consistently improve water quality, as throughfall DOC and NO_3-N inputs still controlled NO_3-N leaching fluxes, with no effect of the chronosequence stage on either throughfall inputs or NO_3-N leaching fluxes.

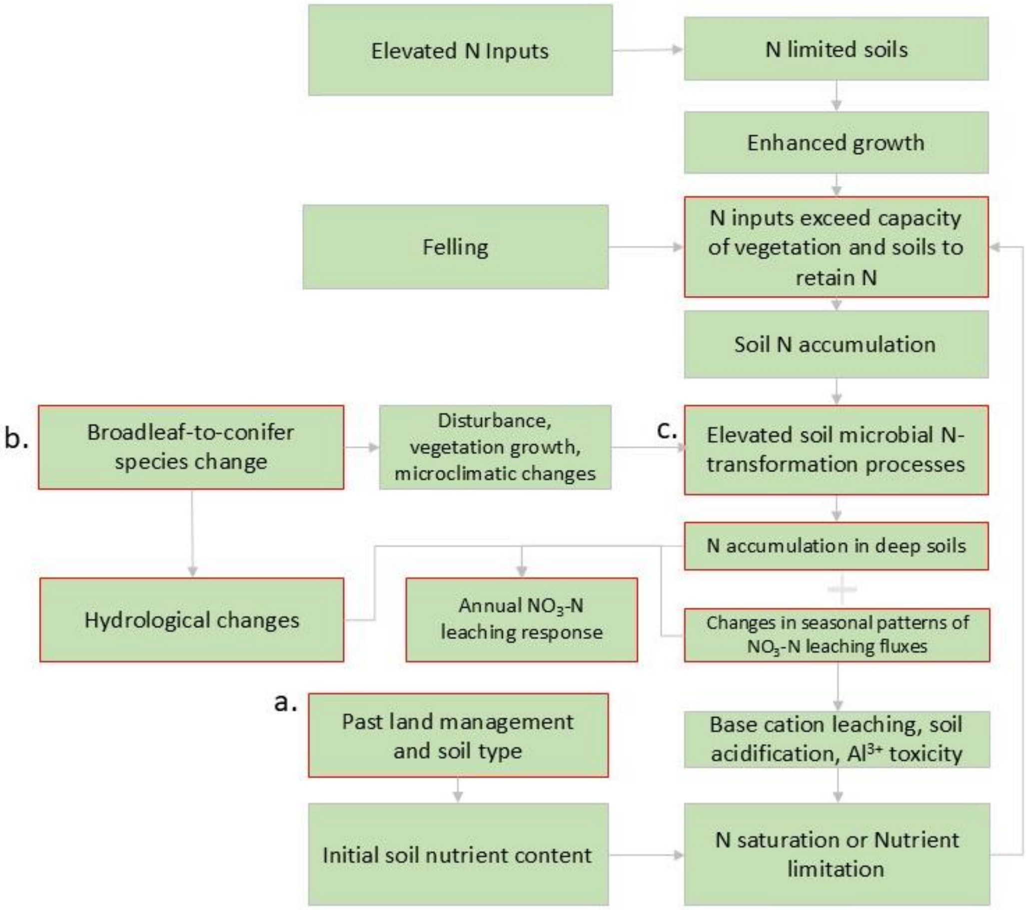
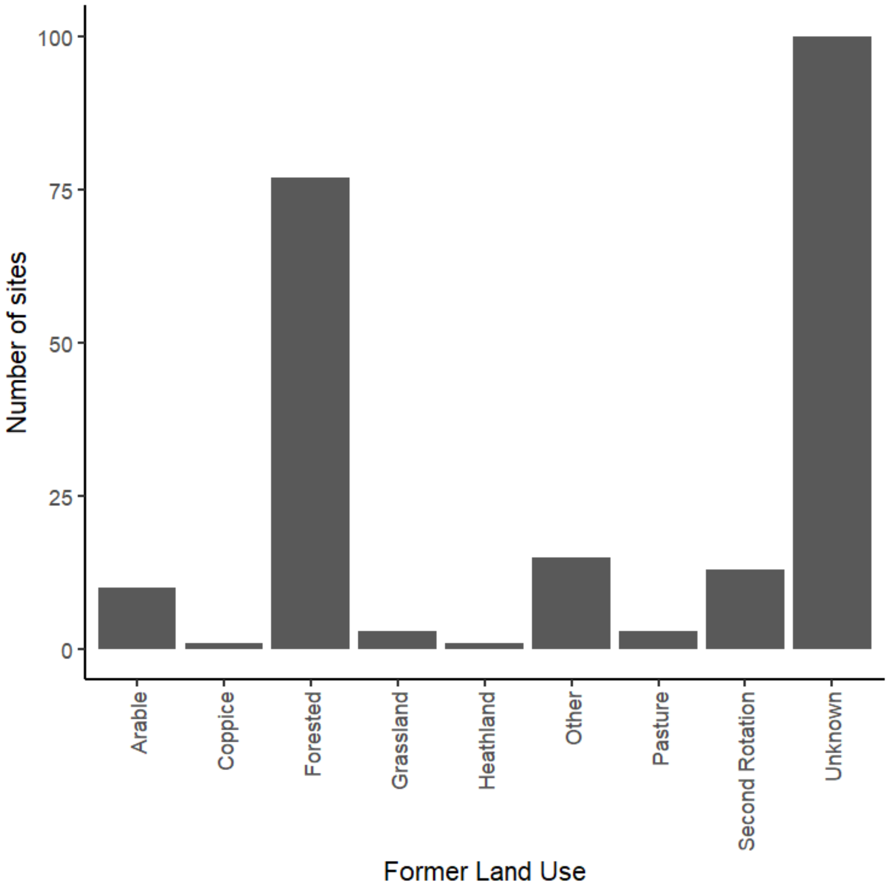


Figure 1: An updated conceptualisation on the response of NO_3-N leaching to land use change.

The long-term impacts of forest management practices are generally only inferred from past observations. The difficulty in applying a coupled biochemical-hydrological model in Chapter 4 to model NO_3-N leaching fluxes from a forest ecosystem limited the long-term projection of NO_3-N leaching fluxes following tree species change. Long-term studies of NO_3-N leaching fluxes from forest ecosystems are rare despite the need to understand NO_3-N leaching in a changing world. NO_3-N leaching from forest ecosystems is a product of decisions made long in the past. Decisions made today will affect nutrient cycling in forests decades into the future, but there is no way to understand NO_3-N leaching in future forest ecosystems without well developed and tested process based models.

Appendices

Appendix 1: The number of forest plots associated with each former land use in the ICP Forests Level II database, where soil solution and throughfall samples have been routinely collected and analysed for chloride, nitrate and ammonium for a minimum of 5 years.



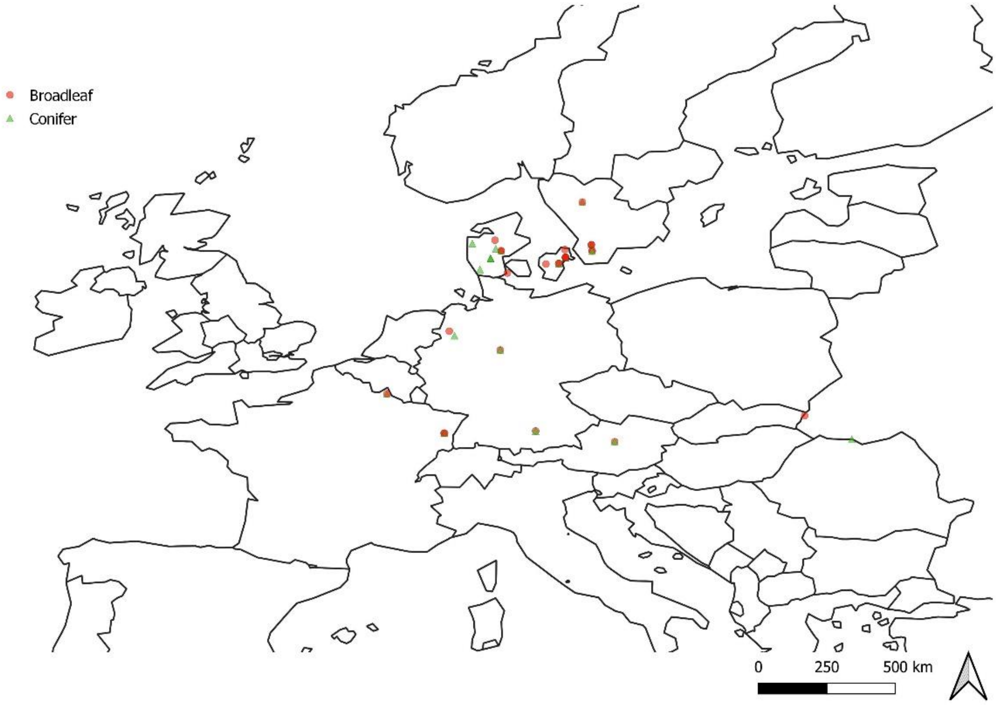
Appendix 2: The search string used for the systematic literature review conducted in chapter 2.

(("Ammonia gas exchange" OR "Tree nutrition" OR "foliar nutrients" OR "forest monitoring" OR "Nitrate uptake" OR "Ammonium uptake" OR "N15 tracer" OR "Nitrogen compounds" OR "Leaf C:N" OR "Leaf C/N" OR "Allocation" OR "C/N ratio" OR "C:N ratio" OR "Leaf N:P" OR "Leaf N/P" OR "N:P ratio" OR "N/P ratio" OR "Leaf N content" OR "Leaf nitrogen content" OR "Root C/N ratio" OR "Root C:N ratio" OR "Root N content" OR "Root nitrogen content" OR "Root N:P ratio" OR "Root N/P ratio" OR "nitrogen leaching") AND ("nitrogen deposition" OR "Wet deposition" OR "Dry deposition" OR "Rainfall" OR "Soil type" OR "Soil texture" OR "Temperature" OR "Soil temperature" OR "Soil water content" OR "Photosynthesis" OR "CO2 concentration" OR "nitrogen leaching" OR "N leaching" OR "Soil nitrogen" OR "Soil nitrogen content" OR "agriculture" OR "agricultural emissions" OR "agricultural land" OR "fertiliser" OR "fertiliser application" OR "Rooting depth" OR "LAI" OR "Leaf area index" OR "Transpiration") AND ("Fagus sylvatica" OR "European beech" OR "Quercus petraea" OR "Sessile oak" OR "Quercus robur" OR "Penduculate oak" OR "Picea sitchensis" OR "Sitka spruce" OR "Pinus Sylvestris" OR "Scots pine" OR "Picea abies" OR "Norway spruce" OR "Silver birch" OR "Betula pendula"))

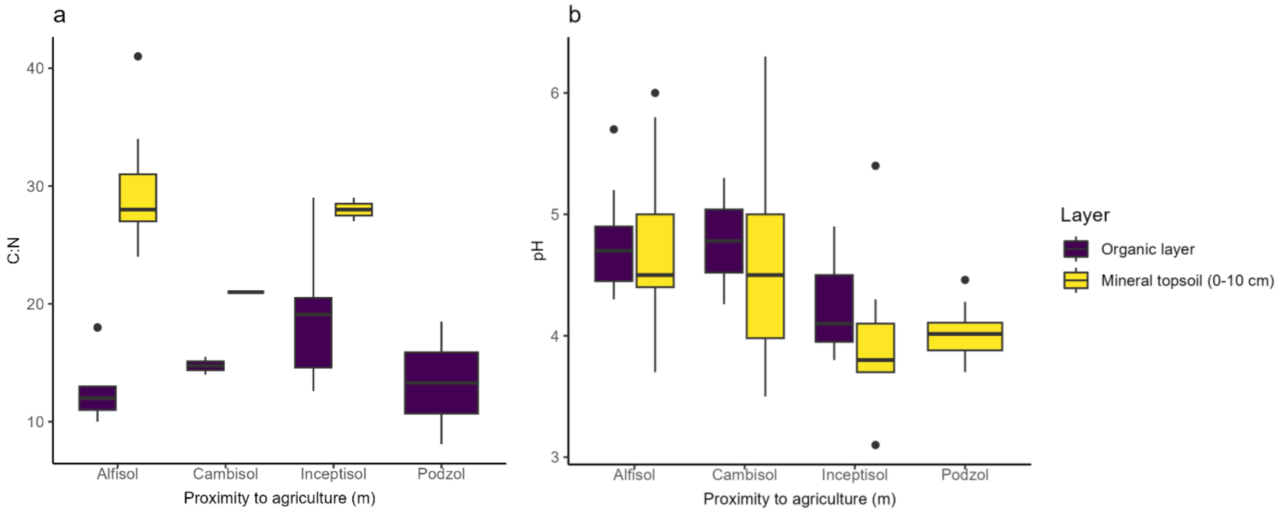
Appendix 3: The studies data was extracted from in the systematic review in chapter 2.

Authors	Title	Pub_ year	Journal
Fichter J, Dambrine E, Turpaut MP, Ranger J	Base cation supply in spruce and beech ecosystems of the Strengbach catchment (Vosges Mountains, N-E France)	1997	Water, air and soil Pollution
Jost G, Dirnbock T, Grabner MT, Mirtl M	Nitrogen Leaching of Two Forest Ecosystems in a Karst Watershed	2011	Water, air and soil pollution
Jussy JH, Colin-Belgrnd M, Dambrine E, Ranger J, Zeller B, Bienaime S	N deposition, N transformation and N leaching in acid forest soils	2004	Biogeochemistry
Ranger J, Nys C	The effect of spruce (Picea abies Karst.) on soil development: an analytical and experimental approach	1994	European journal of soil science
Rothe A, Huber C, Kreutzer K, Weis W	Deposition and soil leaching in stands of Norway spruce and European beech: Results from the Hï¿½glwald research in comparison with other European case studies	2002	Plant and soil
Christiansen JR, Vesterdal L, Callesen I, Elberling B, Schmidt IK, Gundersen P	Role of six European tree species and land-use legacy for nitrogen and water budgets in forests	2010	Global change biology
Herrmann M, Pust J, Pott R	Leaching of nitrate and ammonium in heathland and forest ecosystems in Northwest Germany under the influence of enhanced nitrogen deposition	2005	Plant and soil
Meesenburg, H, Ahrends, B, Fleck, S, Wagner, M, Fortmann, H, Scheler, B, Klinck, U, Dammann, I, Eichhorn, J, Mindrup, M, Meiwes, KJ	Long-term changes of ecosystem services at Solling, Germany:Recovery from acidification, but increasing nitrogen saturation?	2016	Ecological indicators
Bergkvist B, Folkeson L	The influence of tree species on acid deposition, proton budgets and element fluxes in south Swedish forest ecosystems	1995	Ecological bulletins
Pahlsson, B A-M, Bergkvist, B	Acid deposition and soil acidification at a southwest facing edge of Norway spruce and European beech in south Sweden	1995	Ecological bulletins
Oulehle F, Hleb R, Houska J, Samonil P, Hofmeister J, Hruska J	Anthropogenic acidification effects in primeval forests in the Transcarpathian Mts., Western Ukraine	2010	Science of the total environment
Gundersen P, Sevel L, Christiansen JR, Vesterdal L, Hansen K, Bastrup-Birk	Do indicators of nitrogen retention and leaching differ between coniferous and broadleaved forests in Denmark?	2009	Forest ecology and management
Hansen K, Rosenqvist L, Vesterdal L, Gundersen P	Nitrate leaching from three afforestation chronosequences on former arable land in Denmark	2007	Global change biology

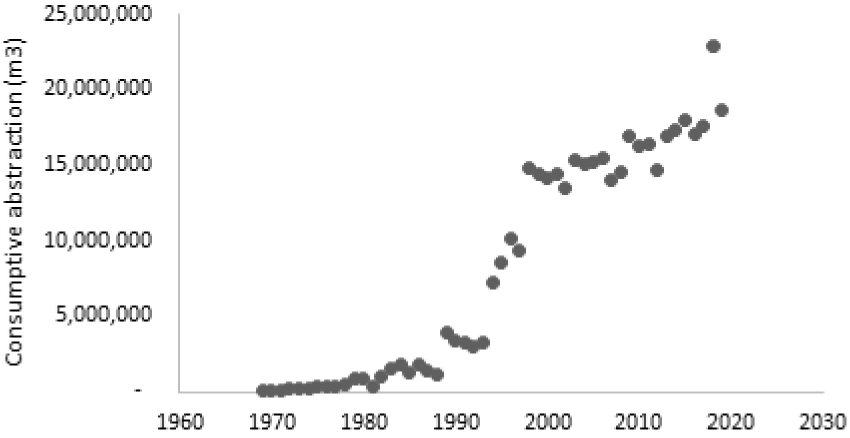
Appendix 4: The location of study sites data was available for in Chapter 2.



Appendix 5: The C:N ratio and pH of organic and mineral topsoils in the study sites extracted in



Appendix 6: Groundwater abstraction in the Little Ouse and Thet catchment. Source: Environment Agency.

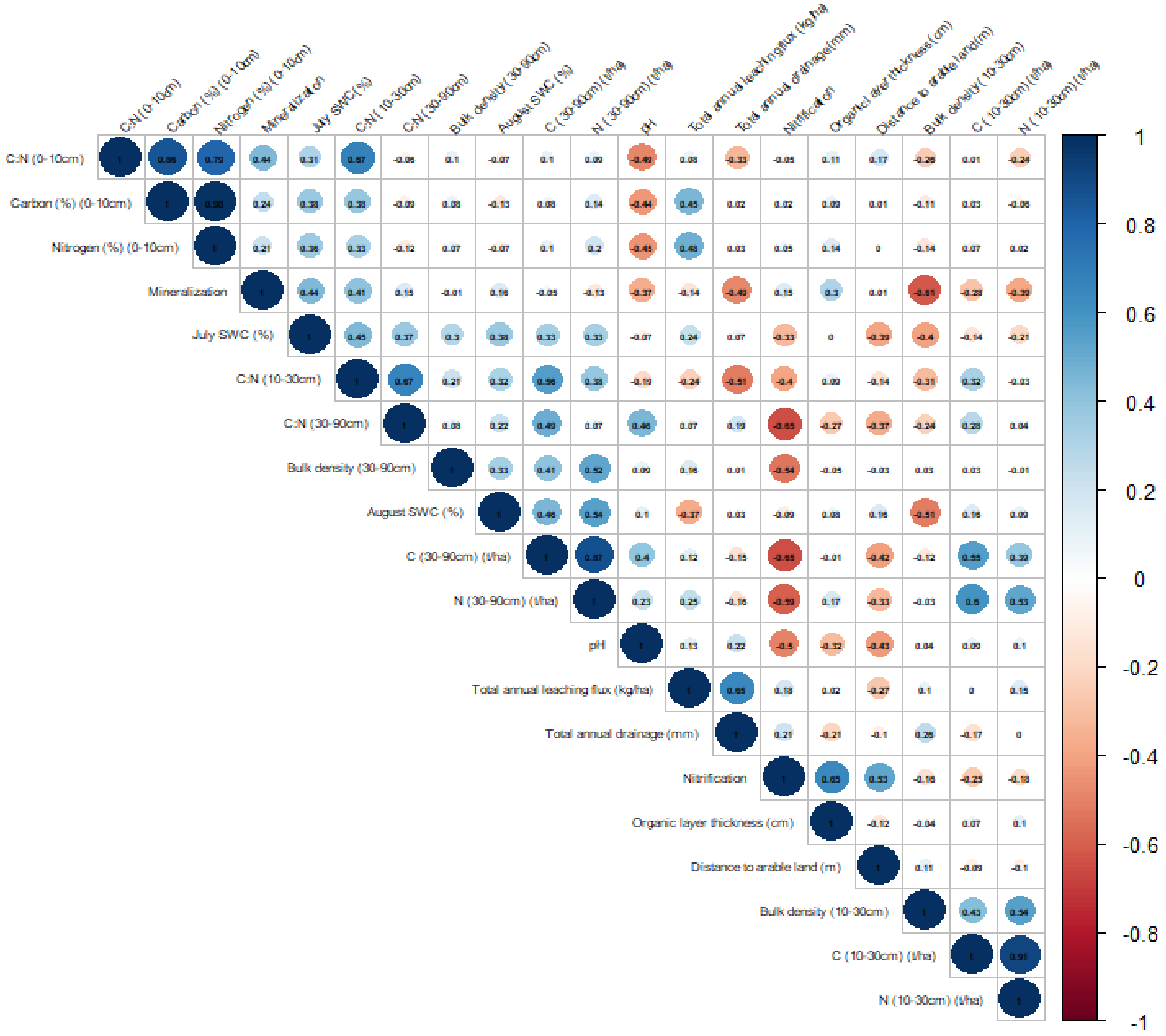


Appendix 7: Details of the field study sites used in Chapter 3.

Stage	Species	Area	Planting year	Measurements
Mature pine	Scots pine	East Harling	1967	Level II site. Throughfall and soil solution collected via lysimeter: 1996-2022. Bulk density. Soil solution via centrifugation Aug 2021-July 2022 Nitrification, mineralisation, soil C and N, pH, O-layer depth
	Scots pine	East Harling	1934	Throughfall + soil solution via centrifugation Aug 2021-July 2022 Nitrification, mineralisation, soil C and N, pH, bulk density, O-layer depth
	Scots pine	Hockham	1965	Throughfall + soil solution via centrifugation Aug 2021-March 2022 Nitrification, mineralisation, soil C and N, pH, bulk density, O-layer depth
	Scots pine	Lynford	1965	Throughfall + soil solution via centrifugation Aug 2021-July 2022 Nitrification, mineralisation, soil C and N, pH, bulk density, O-layer depth
	Corsican pine	Hockham	1983	Soil C and N
	Corsican pine	Croxton	1973	Soil C and N
Mature pine, felled June 2022	Corsican pine	King's Forest	1971	Throughfall Aug 2021-June 2022 Soil solution via centrifugation Aug 2021-July 2022 Nitrification, mineralisation, soil C and N, pH, bulk density, O-layer depth
Felled September 2021	Corsican pine	Hockham	1978	Soil solution via centrifugation Aug 2021-July 2022 Soil C and N, bulk density
Felled January 2022	Corsican pine	Didlington	1973	Soil solution via centrifugation, July 2021 and Feb-July 2022 Soil C and N
Felled February 2022	Corsican pine	Lynford	1969	Soil solution via centrifugation, July 2021 and Feb-July 2022 Soil C and N
Planted 0-2 years ago	Italian alder	Weeting	2020	Soil solution via centrifugation Aug 2021-July 2022 Nitrification, mineralisation, soil C and N, pH, bulk density, O-layer depth
	Red oak	Mildenhall	2021	Soil solution via centrifugation Aug 2021-July 2022 Nitrification, mineralisation, soil C and N, pH, bulk density, O-layer depth
	Sweet chestnut	Didlington	2021	Soil solution via centrifugation Aug 2021-March 2022 Soil C and N, pH, bulk density, O-layer depth
Planted 5-8 years ago	Sycamore	Weeting	2014	Throughfall Aug 2021-July 2022 Soil solution via centrifugation Aug 2021-July 2022 Soil C and N, pH, bulk density, O-layer depth
	Sycamore	King's Forest	2015	Throughfall Aug 2021-July 2022 Soil solution via centrifugation Aug 2021-July 2022 Nitrification, mineralisation, soil C and N, pH, bulk density, O-layer depth
	Beech	King's Forest	2014	Throughfall Aug 2021-March 2022 Soil solution via centrifugation Aug 2021-March 2022 Soil C and N, pH, bulk density, O-layer depth
	Sycamore	King's Forest	2014	Throughfall Aug 2021-July 2022 Soil solution via centrifugation Aug 2021-July 2022

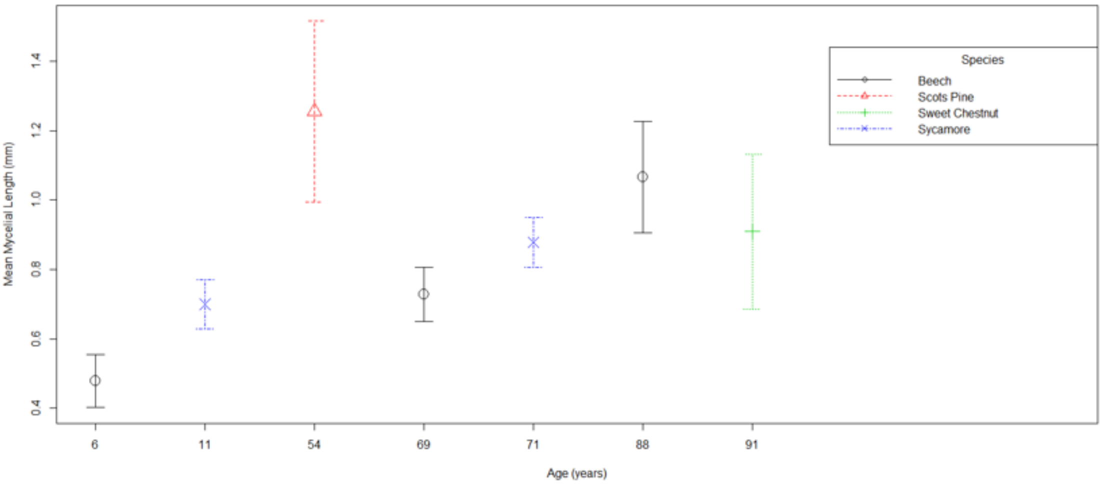
				Nitrification, mineralisation, soil C and N, pH, bulk density, O-layer depth
	Italian alder	Weeting	2016	Throughfall Aug 2021-March 2022 Soil solution via centrifugation Aug 2021-March 2022 Nitrification, mineralisation, soil C and N, pH, bulk density, O-layer depth
Planted 10-13 years ago	Sycamore	Hockham	2010	Throughfall Aug 2021-March 2022 Soil solution via centrifugation Aug 2021-July 2022 Soil C and N, pH, bulk density, O-layer depth
	Sycamore	Croxton	2010	Throughfall Aug 2021-March 2022 Soil solution via centrifugation Aug 2021-July 2022 Nitrification, mineralisation, soil C and N, pH, bulk density, O-layer depth
	Sweet chestnut	East Harling	2011	Throughfall Aug 2021-July 2022 Soil solution via centrifugation Aug 2021-July 2022 Nitrification, mineralisation, soil C and N, pH, bulk density, O-layer depth
	Italian alder	Weeting	2011	Throughfall Aug 2021-July 2022 Soil solution via centrifugation Aug 2021-July 2022 Nitrification, mineralisation, soil C and N, pH, bulk density, O-layer depth
	Sycamore	Weeting	2011	Throughfall Aug 2021-July 2022 Soil solution via centrifugation Aug 2021-July 2022 Nitrification, mineralisation, soil C and N, pH, bulk density, O-layer depth
Mature broadleaf	Sweet chestnut	Mildenhall	1930	Throughfall Aug 2021-March 2022 Soil solution via centrifugation Aug 2021-March 2022 Soil C and N, pH, bulk density, O-layer depth
	Beech	East Harling	1950	Throughfall Aug 2021-July 2022 Soil solution via centrifugation Aug 2021-July 2022 Nitrification, mineralisation, soil C and N, pH, bulk density, O-layer depth
	Oak	Lynford	1950	Throughfall Aug 2021-July 2022 Soil solution via centrifugation Aug 2021-July 2022 Soil C and N, pH, bulk density, O-layer depth
	Beech	Weeting	1952	Throughfall Aug 2021-July 2022 Soil solution via centrifugation Aug 2021-July 2022 Nitrification, mineralisation, soil C and N, pH, bulk density, O-layer depth
	Beech	Hockham	1950	Throughfall Aug 2021-March 2022 Soil solution via centrifugation Aug 2021-March 2022 Soil C and N, pH, bulk density, O-layer depth

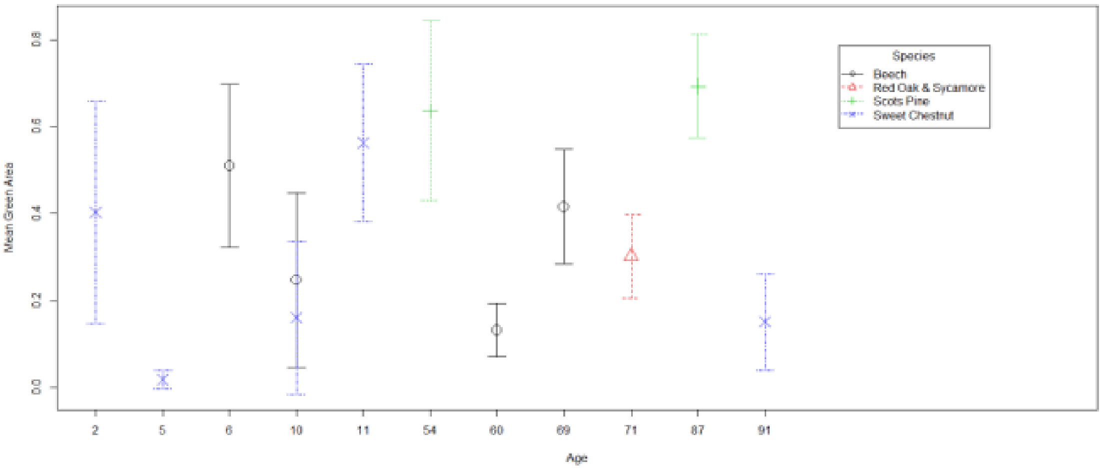
Appendix 8: The correlation matrix created to identify relationships between variables measured in the field study in Chapter 3.



	C:N (0-10cm)	Carbon (%) (0-10cm)	Nitrogen (%) (0-10cm)	Mineralization	July SWC (%)	C:N (10-30cm)	C:N (30-90cm)	Bulk density (30-90cm)	August SWC (%)	C (30-90cm) (t/ha)	N (30-90cm) (t/ha)	pH	Total annual leaching flux (kg/ha)	Total annual drainage (mm)	Nitrification	Organic layer thickness (cm)	Distance to arable land (m)	Bulk density (10-30cm)	C (10-30cm) (t/ha)	N (10-30cm) (t/ha)
C:N (0-10cm)	1	0.86	0.79	0.44	0.31	0.67	-0.06	0.1	-0.07	0.1	0.09	-0.49	0.08	-0.33	-0.05	0.11	0.17	-0.28	0.01	-0.24
Carbon (%) (0-10cm)		1	0.98	0.24	0.38	0.38	-0.09	0.08	-0.13	0.08	0.14	-0.44	0.45	0.02	0.02	0.09	0.01	-0.11	0.03	-0.06
Nitrogen (%) (0-10cm)			1	0.21	0.36	0.33	-0.12	0.07	-0.07	0.1	0.2	-0.45	0.48	0.03	0.05	0.14	0	-0.14	0.07	0.02
Mineralization				1	0.44	0.41	0.15	-0.01	0.16	-0.05	-0.13	-0.37	-0.14	-0.49	0.15	0.3	0.01	-0.61	-0.28	-0.39
July SWC (%)					1	0.45	0.37	0.3	0.38	0.33	0.33	-0.07	0.24	0.07	-0.33	0	-0.39	-0.4	-0.14	-0.21
C:N (10-30cm)						1	0.67	0.21	0.32	0.56	0.38	-0.19	-0.24	-0.51	-0.4	0.09	-0.14	-0.31	0.32	-0.03
C:N (30-90cm)							1	0.08	0.22	0.49	0.07	0.46	0.07	0.19	-0.65	-0.27	-0.37	-0.24	0.28	0.04
Bulk density (30-90cm)								1	0.33	0.41	0.52	0.09	0.16	0.01	-0.54	-0.05	-0.03	0.03	0.03	-0.01
August SWC (%)									1	0.46	0.54	0.1	-0.37	0.03	-0.09	0.08	0.16	-0.51	0.16	0.09
C (30-90cm) (t/ha)										1	0.87	0.4	0.12	-0.15	-0.65	-0.01	-0.42	-0.12	0.55	0.39
N (30-90cm) (t/ha)											1	0.23	0.25	-0.16	-0.59	0.17	-0.33	-0.03	0.6	0.53
pH												1	0.13	0.22	-0.5	-0.32	-0.43	0.04	0.09	0.1
Total annual leaching flux (kg/ha)													1	0.65	0.18	0.02	-0.27	0.1	0	0.15
Total annual drainage (mm)														1	0.21	-0.21	-0.1	0.26	-0.17	0
Nitrification															1	0.65	0.53	-0.16	-0.25	-0.18
Organic layer thickness (cm)																1	-0.12	-0.04	0.07	0.1
Distance to arable land (m)																	1	0.11	-0.09	-0.1
Bulk density (10-30cm)																		1	0.43	0.54
C (10-30cm) (t/ha)																			1	0.91
N (10-30cm) (t/ha)																				1

Appendix 9: The mycelial length, measured from sandbags incubated at 20 cm soil depth for three months, and mean green area of ground cover measurements June 2021, in a subset of the sites studied in Chapter 3. Samples and data analysed by Joshua Deakins.





Appendix 10: Source of ecophysiological parameter values used in the parameter set selected for further investigation. The Biome_BGCMuSo manual provides an example for temperate grassland, not forest. Parameters were switched off and simulated by the model where possible to reduce the number of user-set parameters needed.

Parameter	**Value**	**Source**
Flag: biome type	Woody	Known
Flag: woody type	Evergreen	Known
Flag: photosynthesis type	C3	Known
Yearday to start new growth	1	Biome-BGCMuSo manual
Yearday to end litterfall	364	Biome-BGCMuSo manual
Transfer growth period as fraction of growing season	0.55	Parameterisation StartVal=0.1, endVal=1
Litterfall as fraction of growing season proportion	1	Biome-BGCMuSo manual
Base temperature	2.67	Parameterisation StartVal=0, endVal=8
Minimum temperature for growth	-9999	Model simulated
Optimal1 temperature for growth	-9999	Model simulated
Optimal2 temperature for growth	-9999	Model simulated
Maximum temperature for growth	-9999	Model simulated
Minimum temperature for C assimilation	-9999	Model simulated
Optimal1 temperature for C assimilation	-9999	Model simulated
Optimal2 temperature for C assimilation	-9999	Model simulated
Maximum temperature for C assimilation	-9999	Model simulated
Annual leaf and fine root turnover fraction	0.447	Parameterisation, aligns with values in White *et al.* (2000)
Annual live wood turnover fraction	0.5	White *et al.* (2000)
Annual fire mortality fraction	0.01	No observed fire events
Whole plant mortality fraction in vegetation period	0.075	Parameterisation. StartVal=0, endVal=1. Higher than values in White *et al.* (2000) which set it at 0.005.
C:N of leaves	23.17	Parameterisation. StartVal=14.3, endVal=35. 23.17 aligned with observed values although lower than values in White *et al.* (2000) which were near 30.
C:N of leaf litter	45.0	Observed values between 39-60.
C:N of fine roots	45.0	White *et al.* (2000) suggested 37-82.5 for *Pinus* spp.
C:N of fruit	58.0	Hidy *et al.* 2022
C:N of soft stem	36.6	Hidy *et al.* 2022

C:N of live wood	58.0	White *et al*. (2000) suggested value similar to fine root C:N
C:N of deadwood	730	White *et al*. (2000) suggested 400-1400 for *Pinus* spp.
Dry matter C content of leaves	0.4	Biome-BGCMuSo manual
Dry matter C content of leaf litter	0.4	Biome-BGCMuSo manual
Dry matter C content of fine roots	0.4	Biome-BGCMuSo manual
Dry matter C content of fruit	0.4	Biome-BGCMuSo manual
Dry matter C content of live wood	0.4	Biome-BGCMuSo manual
Dry atter C content of dead wood	0.4	Biome-BGCMuSo manual
Leaf litter labile proportion	0.68	Biome-BGCMuSo manual
Leaf litter Cellulose proportion	0.23	Biome-BGCMuSo manual
Fine root labile proportion	0.34	Calculated from White *et al*. (2000)
Fine root cellulose proportion	0.44	Calculated from White *et al*. (2000)
Fruit litter labile proportion	0.68	
Fruit litter cellulose proportion	0.23	Biome-BGCMuSo manual
Soft stem litter labile proportion	0.68	Biome-BGCMuSo manual
Soft stem litter cellulose proportion	0.23	Biome-BGCMuSo manual
Dead wood cellulose proportion	0.30	White *et al*. (2000)
Canopy water interception coefficient	0.07	Parameterisation. StartVal=0.01, endVal=0.07. White *et al*. (2000)
Canopy light extinction coefficient	0.63	Parameterisation. StartVal=0.3, endVal=0.8. 0.43-0.64 recommended by White *et al*. (2000) for *Pinus* spp.
Potential radiation use efficiency	0.6	N/A for photosynthesis method defined in initialisation file
Radiation parameter 1	0.781	Biome-BGCMuSo manual
Radiation parameter2	-13.596	Biome-BGCMuSo manual
All sided to projected leaf area ratio	2.6	2.37-2.57 recommended for *Pinus* spp by White *et al*. (2000)
Ratio of shaded SLA:sunlit	2	White *et al*. (2000)
Fraction of leaf N in rubisco	0.15	Parameterisation. StartVal=0.1, endVal=0.2. White *et al*. (2000) Biome-BGCMuSo manual
Fraction of leaf N in PEP Carboxylase	0.01	Biome-BGCMuSo manual
Maximum stomatal conductance	0.003	Parameterisation. StartVal=0.001, endVal=0.007. White *et al*. (2000) recommended 0.006
Cuticular conductance	0.00006	Biome-BGCMuSo manual
Boundary layer conductance	0.009	White *et al*. (2000)

Maximum height of plant	19.5	Observed.
Stem weight corresponding to maximum height	160.2	
Plant height function shape parameter	0.5	Near value in Biome-BGCMuSo manual
Maximum depth of rooting zone	2.375	Parameterisation. StartVal=0.5, endVal=3, based on known geology of site.
Root distribution parameter	0.6	Parameterisation. StartVal=0.2, endVal=1.
Root weight corresponding to max root depth	0.4	Near value in Biome-BGCMuSo manual
Root depth function shape parameter	0.5	Near value in Biome-BGCMuSo manual
Root weight to root length conversion factor	1000	Biome-BGCMuSo manual
Growth respiration per unit of C grown	0.3	Biome-BGCMuSo manual
Maintenance respiration in kgC/day per kg of tissue	0.195	Near value in Biome-BGCMuSo manual
Theoretical maximum prop. Of non-structural and structural carbohydrates	0.1	Biome-BGCMuSo manual
Prop. Of non-structural carbohydrates available for maintenance respiration	0.24	Near value in Biome-BGCMuSo manual
Symbiotic + symbiotic fixation of N	0.0048	Biome-BGCMuSo manual
Time delay for temperature in photosynthesis acclimation	0	Biome-BGCMuSo manual
Crop specific parameters	Switched off	Biome-BGCMuSo manual
VWC ratio	1	Parameterisation. StartVal=0.8, endVal = 1.
VWC ratio 2	0.4	Parameterisation. StartVal=0.4, endVal=0.7.
Minimum of soil moisture limit2 multiplicator	0.4	Biome-BGCMuSo manual
VPD: start of conductance reduction	1000	Biome-BGCMuSo manual
CPD: complete conductance reduction	4000	Biome-BGCMuSo manual
Maximum senescence mortality coefficient of above-ground plant material	0.05	Parameterisation. StartVal = 0, endVal = 0.1.
Maximum senescence mortality coefficient of below-ground plant material	0.001	Near value in Biome-BGCMuSo manual
Maximum senescence mortality coefficient of non-structured plant material	0	Biome-BGCMuSo manual
Lower limit extreme high temperature effect on senescence mortality	35	Near value in Biome-BGCMuSo manual. Kunert *et al*. (2021) showed thermal tolerance of temperate conifers is ~38 ^{o}C.
Upper limit extreme high temperature effect on senescence mortality	40	Biome-BGCMuSo manual
Turnover rate of wilted standing biomass	0.01	Near value in Biome-BGCMuSo manual
Turnover rate of non-woody cut-down biomass to litter	0.047	Near value in Biome-BGCMuSo manual. Know decomposition rates are high in Thetford forest.
Turnover rate of woody cut-down biomass to litter	0.01	Biome-BGCMuSo manual

Drought tolerance parameter	17	Seidel *et al*. (2016) tested drought tolerance of Scots pine
Soil water deficit effect on photosynthesis downregulation	0.3	Near value in Biome-BGCMuSo manual
Critical amount of snow limiting photosynthesis	5	Biome-BGCMuSo manual
Limit1 HEATSUM index	20	Biome-BGCMuSo manual
Limit2 HEATSUM index	60	Biome-BGCMuSo manual
Limit1 TMIN index	0	Biome-BGCMuSo manual
Limit2 TMIN index	5	Biome-BGCMuSo manual
Limit1 VPD index	4000	Biome-BGCMuSo manual
Limit2 VPD index	1000	Biome-BGCMuSo manual
Limit1 daylength index	0	Biome-BGCMuSo manual
Moving average	10	Biome-BGCMuSo manual
GSI limit1	0.1	Biome-BGCMuSo manual
GSI limit2	0.01	Biome-BGCMuSo manual
Length of phenophase	10000	Biome-BGCMuSo manual – setting a long first phenophase runs the model without separating logic into 7 phenophases. Allocation parameters must add up to 1.
Leaf Allocation	0.2	Near value in Biome-BGCMuSo manual
Fine root Allocation	0.3	Near value in Biome-BGCMuSo manual
Fruit allocation	0	Near value in Biome-BGCMuSo manual
Soft stem allocation	0	N/A in woody ecosystems – Biome-BGCMuSo manual
Live woody stem allocation	0.1	Near value in Biome-BGCMuSo manual
Dead woody stem allocation	0.1	Near value in Biome-BGCMuSo manual
Live coarse root allocation	0.2	Near value in Biome-BGCMuSo manual
Dead coarse root allocation	0.1	Near value in Biome-BGCMuSo manual
Canopy average specific leaf area	49	Biome-BGCMuSo manual
Current growth proportion	0.37	Near value in Biome-BGCMuSo manual
Maximal lifetime of plant tissue	10000	

Appendix 11: Source of soil parameter values used in the parameter set selected for further investigation. The Biome_BGCMuSo manual provides an example for temperate grassland, not forest. Parameters were switched off and simulated by the model where possible to reduce the number of user-set parameters needed.

Parameter	**Value**	**Source**
Soil depth	1	known
C:N ratio of recaltirant SOM	12	Biome-BGCMuSo manual
NH4 mobilen proportion	0.25	Parameterisation. StartVal = 0, endVal = 0.5
Aerodynamic resistance	107	Biome-BGCMuSo manual
Parameter 1 for tscalar function of decomposition	-9999	Biome-BGCMuSo manual
Parameter 2 for tscalar function of decomposition	1	Biome-BGCMuSo manual
Parameter 3 for tscalar function of decomposition	275	Parameterisation. StartVal=250, EndVal=300.
Parameter 4 for tscalar function of decomposition	0	Parameterisation. StartVal = 0, EndVal = 50
Soil temperature for decomposition	10	Parameterisation. StartVal = -15, EndVal=10.
e-folding depth of decomposition rate's depth scalar	1.1	Parameterisation. StartVal = 0, EndVal=2
Net mineralisation proportion of nitrification	0.75	Parameterisation. StartVal = 0, EndVal=1.
Maximum nitrification rate	0.767	Parameterisation. StartVal=0.3, EndVal = 1.
Coefficient of N2O emission of nitrification	0.02	Biome-BGCMuSo manual
For pH scalar function of nitrification 1	0.5	Parameterisation. StartVal=0.1, EndVal=1
For pHscalar function of nitrification 2	0.031	Parameterisation. StartVal=0.01, EndVal=0.2
For pHscalar function of nitrification 3	5.2	Biome-BGCMuSo manual
For pHscalar function of nitrification 4	0.55	Biome-BGCMuSo manual
For tscalar function of nitrification 1	-9999	Biome-BGCMuSo manual
For tscalar function of nitrification 2	1	Biome-BGCMuSo manual
For tscalar function of nitrification 3	308.56	Biome-BGCMuSo manual
For tscalar function of nitrification 4	71.02	Biome-BGCMuSo manual
WFPS for scalar of nitrification calculation	0.2375	Parameterisation. StartVal=0.05, EndVal=0.3
Lower optimum WFPS for scalar of nitrification calculation	0.45	Biome-BGCMuSo manual
Optimum WFPS for scalar of nitrification calculation	1	Biome-BGCMuSo manual
Minimum value for saturated WFPS scalar of nitrification	0.2	Biome-BGCMuSo manual
Denitrification rate per g of CO2 respiration of SOM	0.1	Hidy et al. (2021)

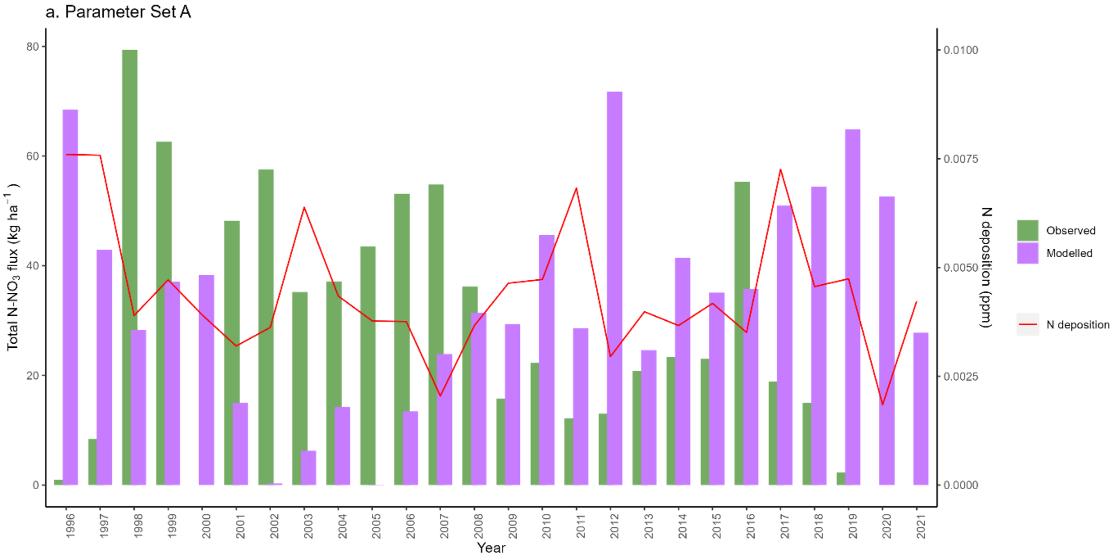
Denitrification related N2/N2O ratio multiplier	1	Hidy et al. (2021)
Critical WFPS value for denitrification	0.55	Biome-BGCMuSo manual
Respiration fractions for fluxes between compartments (l1s1)	0.775	Parameterisation. StartVal=0.1, EndVal=1
Respiration fractions for fluxes between compartments (l2s2)	0.325	Parameterisation. StartVal=0.1, End Val=1
Respiration fractions for fluxes between compartments (l4s3)	0.1	Parameterisation. StartVal=0.1, EndVal=1
Respiration fractions for fluxes between compartments (s1s2)	1	Parameterisation. StartVal=0.1, EndVal=1
Respiration fractions for fluxes between compartments (s2s3)	0.55	Parameterisation. StartVal=0.1, EndVal=1. Matches exact with Biome-BGCMuSo manual
Respiration fractions for fluxes between compartments (s3s4)	0.1	Parameterisation. StartVal=0.1, EndVal=1
Rate constant scalar of labile litter pool	0.55	Parameterisation. StartVal=0.1, EndVal=1
Rate constant scalar of cellulose litter pool	0.1	Parameterisation. StartVal=0.1, EndVal=1
Rate constant scalar of lignin litter pool	0.0575	Parameterization StartVal=0.01, EndVal=0.2
Rate constant scalar of fast microbial recycling pool	0.0575	Biome-BGCMuSo manual suggests 0.07
Rate constant scalar of medium microbial recycling pool	0.014	Biome-BGCMuSo manual
Rate constant scalar of slow microbial recycling pooll	0.0014	Biome-BGCMuSo manual
Rate constant scalar of recalcitrant SOM pool	0.0001	Biome-BGCMuSo manual
Rate constant scalar of physical fragmentation of coarse woody debris	0.001	Parameterisation, startVal=0.001, EndVal=0.01. Matches Biome-BGCMuSo manual
Limit of first stage evaporation	1.5	Parameterisation. StartVal=0.5, EndVal=1.5
Maximum height of pond water	0	No ponds present
Soil stress function	0.1	Parameterisation. StartVal=0.1, EndVal = 1. Biome-BGCMuSo manual suggests 1
Runoff curve number	-9999	Switched off
Fraction of dissolved part of SOIL1 organic matter	0.002	Biome-BGCMuSo manual
Fraction of dissolved part of SOIL2 organic matter	0.002	Biome-BGCMuSo manual
Fraction of dissolved part of SOIL3 organic matter	0.001	Biome-BGCMuSo manual
Fraction of dissolved part of SOIL4 organic matter	0.001	Biome-BGCMuSo manual
Mulch parameter: layer effect	2	Biome-BGCMuSo manual
Mulch parameter: critical amount	0	Biome-BGCMuSo manual
Parameter 1 for mulch function	100	Biome-BGCMuSo manual
2 for mulch function	0.75	Biome-BGCMuSo manual
Parameter: evaporation reduction	0.1	Parameterisation. StartVal=0.1, EndVal=1.
1 for diffusion calculation (tipping)	1	Parameterisation. StartVal=0.5, EndVal=1

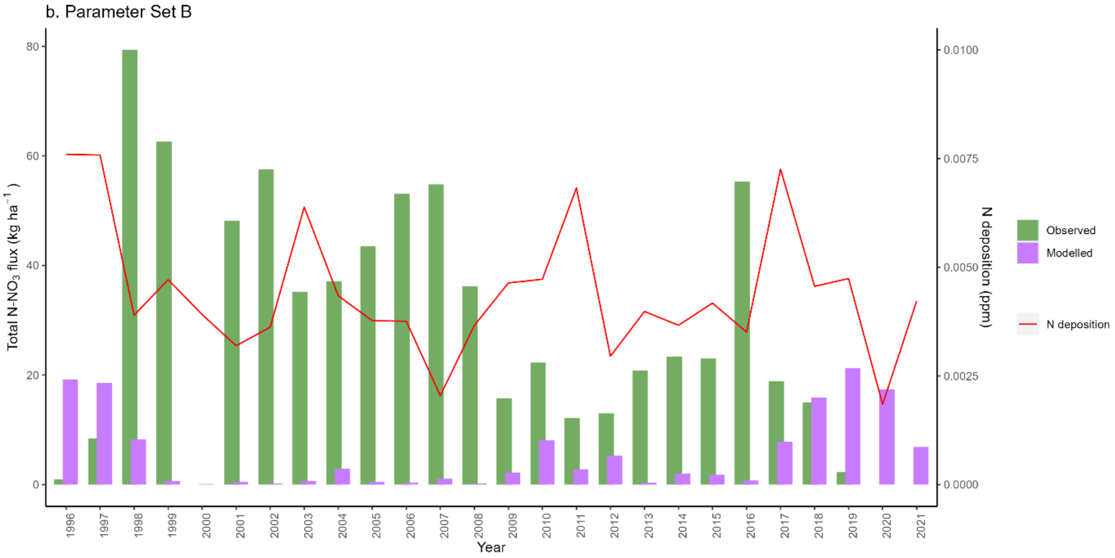
2 for diffusion calculation (tipping)	35	Parameterisation, StartVal=30, EndVal=40. Matches Biome-BGCMuSo manual
3 for diffusion calculation (tipping)	10	Parameterisation. StartVal=1, EndVal=10.
Flag for GW method	0	
Capillary fringe	0.1	Average for this particular geology
Soil CH4 emission bulk density dependence parameter 1	212.5	Biome-BGCMuSo manual
Soil CH4 emission bulk density dependence parameter 2	1.81	Biome-BGCMuSo manual
Soil CH4 emission soil water content dependence parameter 1	-1.353	Biome-BGCMuSo manual
Soil CH4 emission soil water content dependence parameter 2	0.2	Biome-BGCMuSo manual
Soil CH4 emission soil water content dependence parameter 3	1.781	Biome-BGCMuSo manual
Soil CH4 emission soil water content dependence parameter 4	6.786	Biome-BGCMuSo manual
Soil CH4 emission soil temperature dependence parameter	0.01	Biome-BGCMuSo manual
% sand	50	Observed
% silt	30	Observed
Soil pH	5.1	Observed
Soil B measured runoff curve number	-9999	Switched off
Bulk density	-9999	Switched off
SWC content at saturation	-9999	Switched off
SWC at field capacity	-9999	Switched off
SWC at wilting point	-9999	Switched off
SWC at hygroscopic water content	-9999	Switched off
Drainage coefficient	-9999	Switched off
Hydraulic conductivity	-9999	Switched off

Appendix 12: Maximum and minimum sensitivity analysis values used to identify influential parameters before the first parameterisation procedures in Chapter 4.

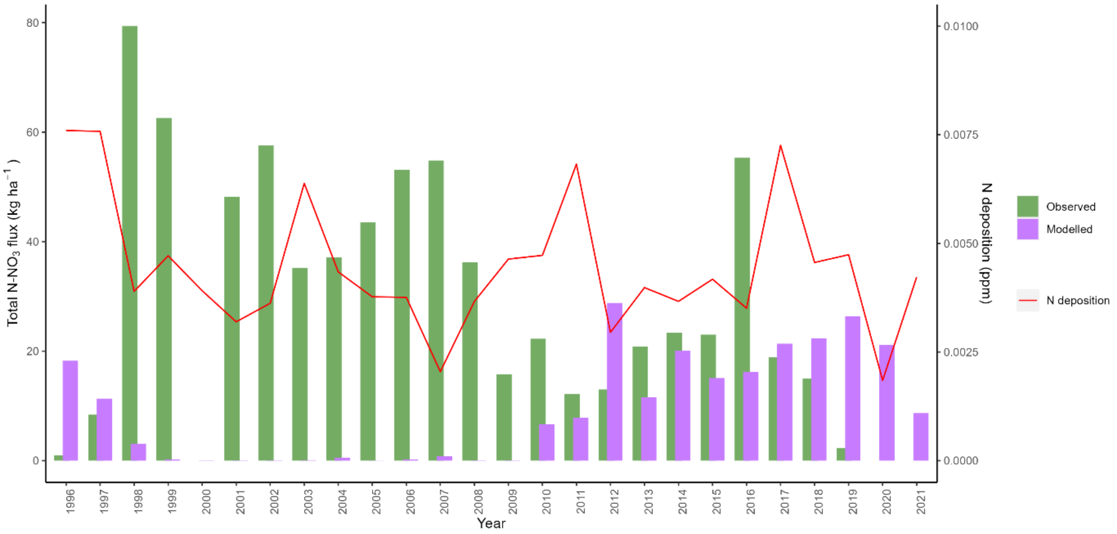
EPC File				Soil File			
Parameter	Start_val	End_val	Steps	Parameter	Start_val	End_val	Steps
11	0.1	1	10	6	0.1	1	5
13	0	9	10	12	250	300	3
22	0.1	0.5	5	13	0	50	5
23	0.1	0.5	5	14	-15	10	5
25	0.01	0.1	5	15	0.5	2	6
26	20	40	20	16	0	1	5
27	20	39	10	17	0.3	1	7
28	30	45	10	19	0.1	1	10
29	30	65	10	20	0.01	0.2	10
30	30	50	10	27	0.05	0.3	5
32	600	850	5	29	0.5	1	5
33	0.1	0.9	10	36	0.1	1	5
34	0.1	0.9	10	37	0.1	1	5
35	0.1	0.9	10	38	0.1	1	5
49	0.1	0.9	10	39	0.1	1	5
50	0.1	0.5	5	40	0.1	1	5
64	0.1	1	5	41	0.1	1	5
65	1	5	5	42	0.1	1	5
66	0.1	2	10	43	0.1	1	5
96	0	1	5	44	0.01	0.2	5
97	0	1	5	45	0.01	0.2	5
114	10	30	20	49	0.001	0.01	5
				52	0.5	1.5	5
				54	0.1	1	5
				63	0.5	1	5
				65	0.1	1	5
				66	0.5	1	5
				67	30	40	5
				68	1	10	5

Appendix 13: Annual observed and simulated N-NO$_3$ leaching fluxes for parameter sets A-H tested in Chapter 4.

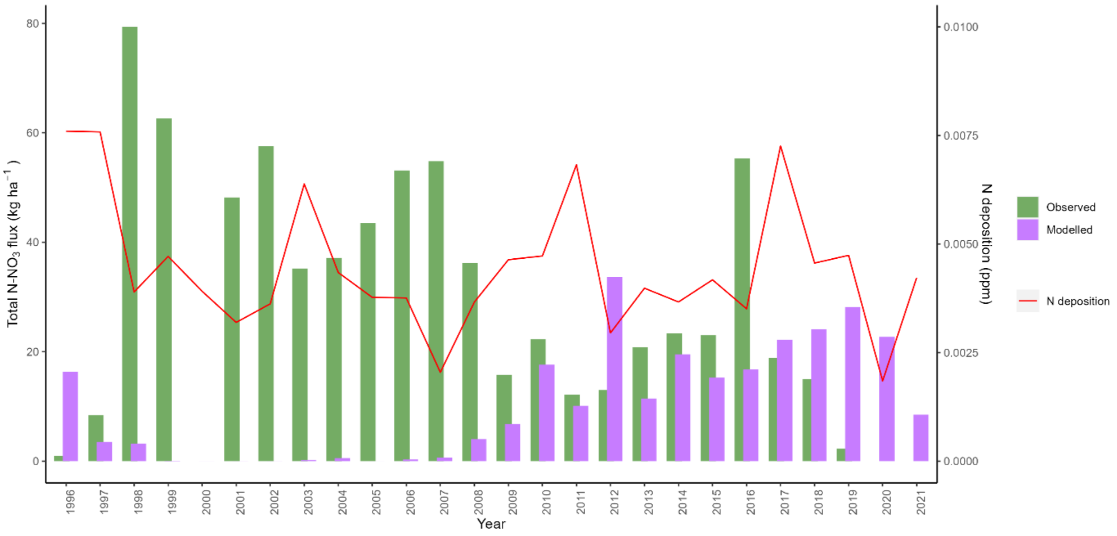




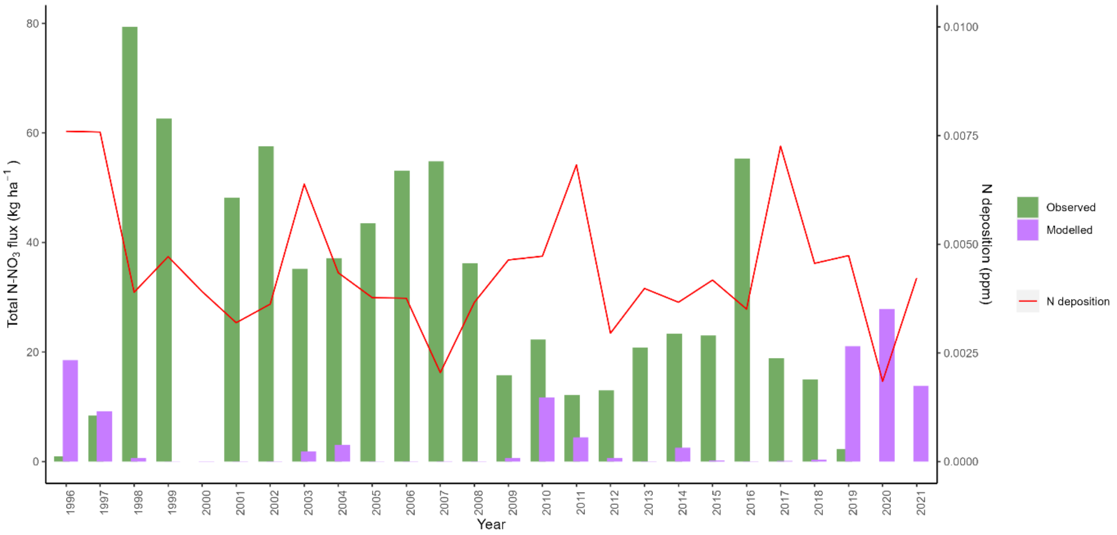
c. Parameter Set C



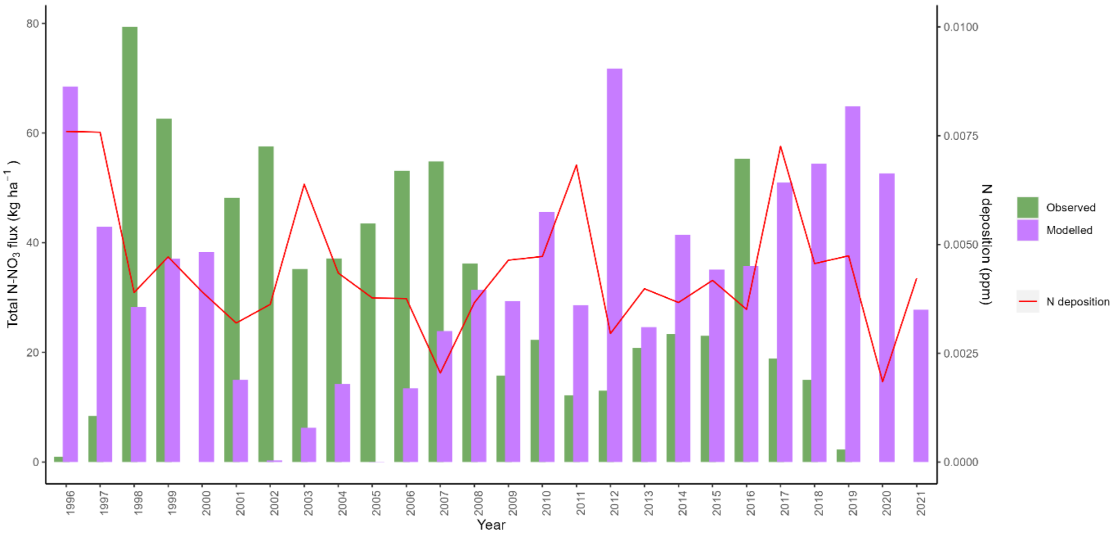
d. Parameter Set D



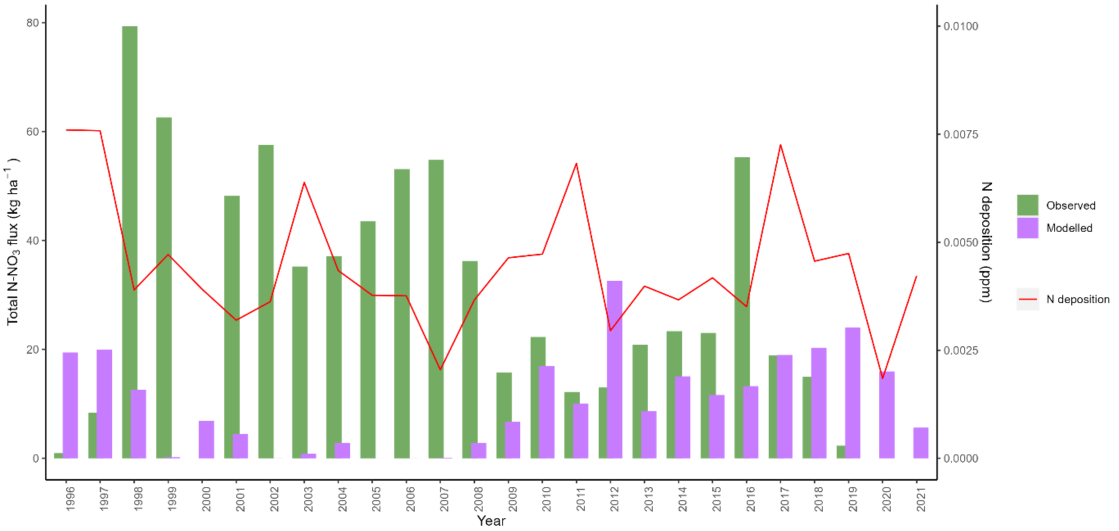
e. Parameter Set E



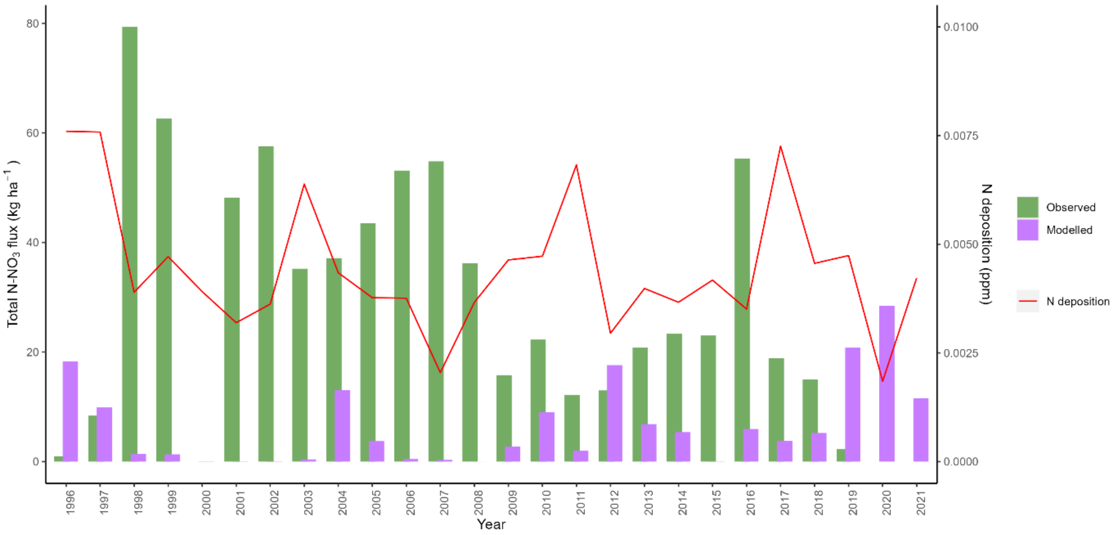
f. Parameter Set F



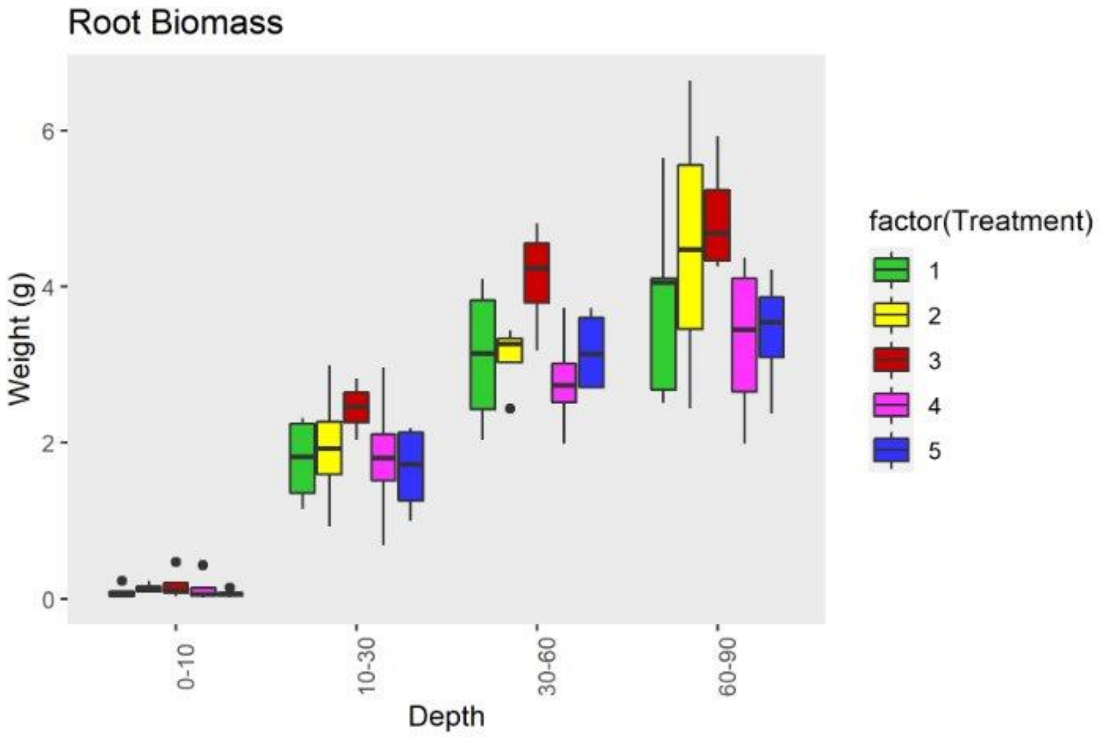
g. Parameter Set G

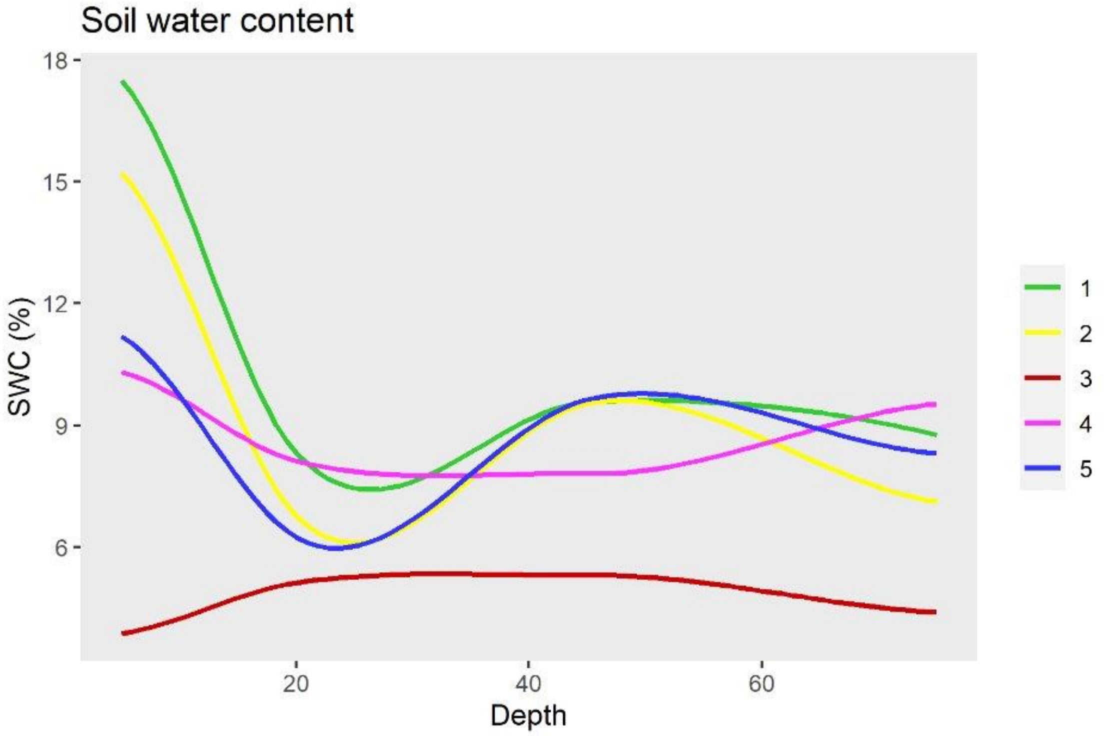


h. Parameter Set H



Appendix 14: The root biomass and soil water content of willow trees at four soil depths, after 4 months growing in rhizotrons (1 m depth) subjected to different NO_3-NH_4 treatments. Treatments 4 and 5 were controls (even or no addition of nutrients). Treatment 1 represented the nutrient stratification in coniferous soils, treatment 2 represented felled conifer soils, and treatment 3 represented the stratification of nutrients in broadleaf soils.





List of trainings attended

1. University of Reading modules:
    a. AP3AE80 Ecosystem Services (Unassessed, Masters level)
    b. APMA96 Plants, Greenspace and Urban Sustainability (Unassessed, Masters level)
    c. GV3ESM Ecosystems Modelling (Unassessed, Year 3 module open to masters students)
    d. MTMG44 Hydrology and Global Environmental Change (Unassessed, Masters level)
    e. GVMEPM Environmental and Pollution Microbiology (Assessed, achieved 75 %, Masters level)
    f. IDM099 Global Environmental Change, Justice and Development (Assessed, achieved 74 %, Masters level)
2. Introduction to programming in R
3. Pathway A- Programming Design, and Introduction to Management and Curation of Scientific Codes.
4. Research data management (BGS)
5. Young Entrepreneurs Scheme 2020
6. SCENARIO: Innovating for Sustainable development 2020
7. Walker Institute: Climate Resilience Evidence Synthesis Training 2020
8. SCENARIO: Writing science for a non-expert audience 2020
9. The Brilliant Club: Teacher Training 2021
10. RRDP's:
    a. How to summarise your research in 3 minutes 1
    b. Diversity and Inclusion: An introduction for doctoral researchers
    c. Aim to be assertive
    d. Presentation skills
    e. How to write a literature review
    f. You and your supervisor
    g. An introvert's guide to influence and impact
    h. Open access and research data management for RCUK-funded students
    i. Preparing to teach program
11. Field Studies Council: Identifying Grasses from Vegetative Features 2023
12. UKCEH: Spatial Analysis In QGIS And R - An Introduction For Environmental Scientists 2024