Long-Term Effects of Nitrogen (N) Fertilizer and Simulated N Deposition on Boreal Forest Growth

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Licentiate Thesis
Swedish University of Agricultural Sciences
Umeå 2014

ISBN (print version) 978-576-9225-2
ISBN (electronic version) 978-91-576-9226-9

Print: SLU Repro, Uppsala 2014

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Abstract

The studies presented in this thesis aims to increase our understanding of the long-term effects of anthropogenic nitrogen (N) input via fertilization or atmospheric N deposition on tree growth in boreal forest. Firstly, I studied carry-over effects of forest fertilization between tree generations. Growth of 10 years old trees on sites fertilized during the previous tree generation was measured. On sites that were previously fertilized twice (25 and 33 years prior to the study), the young Norway spruce (*Picea abies* [L.] H. Karst.) and Scots pine (*Pinus sylvestris* L.) trees were 24% taller than trees on similar sites that were only fertilized once (25 years prior to the study) or on sites that had never been fertilized. Furthermore, the soil N mineralization rates were 3.7 times higher and the amount of available soil-N 2 times higher on sites that were previously fertilized twice than on other sites.

Secondly, I studied the effects of low annual N addition simulating atmospheric N deposition on tree growth and variables linked to site fertility in two different forest ecosystems, *i.e.* on one *P. abies* dominated site and on one *P. sylvestris* dominated site. On the *P. abies* dominated site tree growth and needle N concentration were higher on plots treated with 12.5 and 50 kg N ha^{-1} $year^{-1}$ than on control plots, which only received background N deposition (1 to 2 kg N ha^{-1} $year^{-1}$). *P. abies* growth increased linearly with 2.5% for every kg of N added, which corresponded to a net sequestration of approximately 19 kg of C per kg N. On the *P. sylvestris* dominated site tree growth and needle N concentration increased only on plots with the 50 kg N ha^{-1} $year^{-1}$ treatment, whereas the low N addition treatments (3, 6 and 12.5 kg N ha^{-1} $year^{-1}$) had no effect on tree growth.

In conclusion, I present data suggesting that commercial forest fertilization can have long-term effects on site productivity and consequently tree growth in the tree generation following the one fertilized. I also show data that supports a positive tree growth response to N addition rates similar to those of atmospheric N deposition. However, further studies are needed of how tree growth in naturally nutrient poor *P. sylvestris* dominated forests respond to low annual N addition rates.

Keywords: annual N addition, carbon sequestration, fertilization, forest growth,

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Dedication

Igor.

Försiktigt, men fort.
Sören

Contents

List of Publications

This thesis is based on the following papers, which will be referred to by Roman numerals in the text:

I From, F., Strengbom, J., Nordin, A. (2014). Residual long-term effects from forest fertilization on tree growth (manuscript).

II From, F., Mörling, T., Lundmark, T., Nordin, A. (2014). Effects of simulated N deposition on tree growth in boreal forest (manuscript).

III Gundale, M.J., From, F., Bach, L.H. & Nordin, A. (2014). Anthropogenic nitrogen deposition in boreal forests has a minor impact on the global carbon cycle. Global Change Biology, 20(1), pp. 276-286.

Paper III is reproduced with kind permission of the publishers (John Wiley and Sons).

The contribution of Fredrik From to the papers included in this thesis was as follows:

I Planned and performed the field work together with the co-authors, analyzed the data and wrote the paper.

II Planned and performed the field work together with the co-authors, analyzed the data and wrote the paper.

III Participated in planning and performing the field work, analyzing the data and writing the paper.

Abbreviations

Table 1. *Abbreviations used in Paper I through III.*

Symbol	Definition
DBH	Diameter at breast height (*i.e.* 1.3 m above ground) (VanSoest *et al.*, 1959)
Dg	Basal-area weighted mean diameter. The diameter corresponding to the mean basal-area of the stand (VanSoest *et al.*, 1959)
DW	Dry weight
H_{100}	The average height (m) at the age of 100 years for the 100 widest (DBH) trees per hectare (ha) (Hägglund, 1981)
N1	Fertilization with 150 kg N ha^{-1}, in Paper I
N2	Fertilization with 150 kg N ha^{-1} applied twice, eight years apart, in Paper I
3N	Addition of 3 kg N ha^{-1} $year^{-1}$, low rate treatment in Paper II and III
6N	Addition of 6 kg N ha^{-1} $year^{-1}$, low rate treatment in Paper II and III
12.5N	Addition of 12.5 kg N ha^{-1} $year^{-1}$, low rate treatment in Paper II and III
50N	Addition of 50 kg N ha^{-1} $year^{-1}$, high rate treatment in Paper II and III

1 Introduction

This thesis aims to explore long-term effects from anthropogenic nitrogen (N) addition on boreal forest growth; particularly in forests dominated by either Norway spruce (*Picea abies* [L.] H. Karst.) or Scots pine (*Pinus sylvestris* L.*).* The thesis focuses on two different categories of N addition: commercial forest fertilization (Paper I) and atmospheric N deposition (Paper II and III), with long-term effects as the common denominator. Forest fertilization can, in this respect, be regarded as intentional N addition while N deposition on the other hand is unintentional. Both intentional and unintentional anthropogenic N additions affect our forests in many ways not yet fully understood.

1.1 Boreal forest growth is nitrogen limited

Most boreal forests are N limited (Tamm, 1991) and will respond with increased growth to N addition (Nohrstedt, 2001). When provided with extra N boreal conifer trees expand their photosynthetic apparatus by increasing the amount of foliage and by increasing the amount of N within foliage (Linder & Troeng, 1980; Tamm, 1991). Fertilizing with N also shifts tree biomass allocation from below to above ground parts (Axelsson & Axelsson, 1986). During the past century anthropogenic N has mainly been incorporated in Swedish boreal forest ecosystems through two major pathways, as atmospheric N deposition and through commercial fertilization (Fig. 1). Nitrogen may also be sequestered by *e.g.* N_2 fixing bacteria in association with *e.g.* Red-stemmed feather-moss (*Pleurozium schreberi* [Bird] Mitt.) (Gundale *et al.*, 2011) or Alder (*Alnus* sp.), however, this thesis focus on the anthropogenic input.

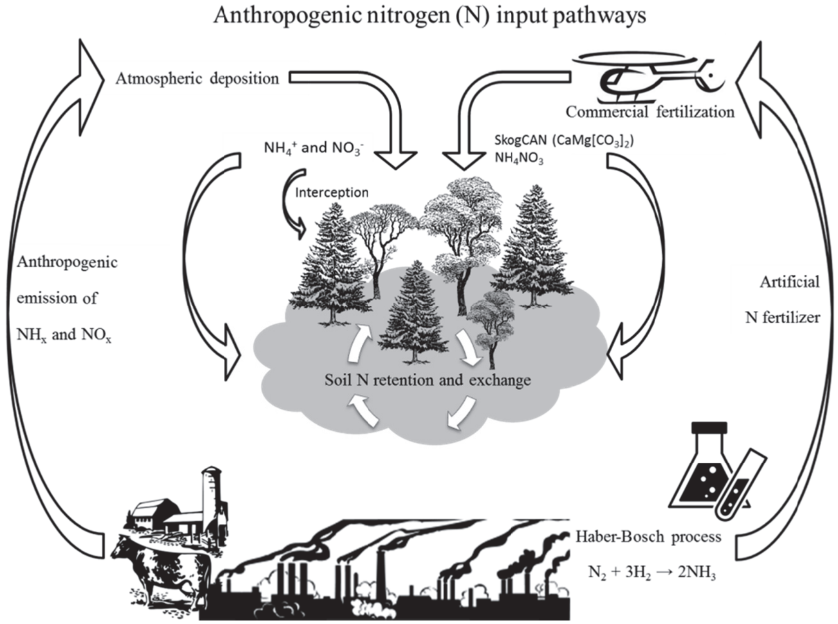

Figure 1. Conceptual model of some anthropogenic N input pathways to boreal forests.

1.2 Anthropogenic nitrogen input in Sweden

N deposition has taken place all over Sweden, to a greater extent in the south than in the north. The opposite is true for forest fertilization; it is done locally, under controlled conditions and mainly in the middle and northern parts of the country. The two anthropogenic N input pathways to forests could be called ‘unintentional’ and ‘intentional’ fertilization. Nitrogen deposition is unintentional in the sense that we have no control over where deposition occurs, reactive nitrogen (N_r) travels in the prevailing wind direction, sometimes far from the source. In contrast, today's commercial forest fertilization is a very precise method and applied N is usually retained in the intended forest stand.

1.2.1 Forest fertilization in practice

In the middle of the 20th century the demand for forest products increased dramatically *cf* Kardell and Lindkvist (2010) and Lindkvist *et al.* (2011). Forest managers and academics predicted dwindling stocks and sought ways to increase forest growth to ensure a sustainable supply of the raw material to the industry. With the discovery of the Haber-Bosch process decades earlier, artificial N fertilizer could be mass produced and was widely used in agronomy. With the increments in yield observed in agriculture for ulterior motives, foresters researched the growth enhancing effects of adding N to forest stands in early fertilization field trials. They found that forest fertilization was as a good method for increasing tree production to meet the growing demand. From these early experiments it was also determined that from low N application amounts (<50 kg N ha^{-1}) the forest growth increments were minor or statistically insignificant (Pettersson, F. 2013[1]). In a Norwegian study, Brantseg *et al*. (1970) concluded that fertilizing with less than 75 kg N ha^{-1} did seldom increase growth significantly in Norwegian *P. abies* and *P. sylvestris* forests. Möller (1978) estimated that only about 55% of the possible growth enhancing effect seen in early fertilization experiments was reached in commercial forest fertilization at that time. Possible reasons for the lower effect were presumed to be uneven fertilizer spreading, abiotic inferiority, stand characteristics and estimation errors in optimal harvest volume. In the modern day this proportion is likely to have increased, since the environment and forest production is treated as being equal under Swedish forestry law, with accurate spreading being of higher importance today than in the 1970's.

The area fertilized annually in Sweden rapidly increased from only a few thousand ha in the early 1960's to about 190 000 ha (about 0.8% of the productive forest area, *i.e.* 23 million ha) in the mid-1970's (Fig. 2) (Anon, 2014). This was followed by a decrease to 30 000 ha in the beginning of the 1990's. The swift decrease in commercial fertilization was likely due to the weak economic climate at that time and the public concern for fertilization causing acidification and eutrophication (Kardell & Lindkvist, 2010; Lindkvist *et al.*, 2011). At the beginning of this century the use of N fertilizer increased and in 2010 about 80 000 ha of forest land was fertilized (Anon, 2014). Following the decline in the global economy in recent years the total area annually fertilized decreased to about 53 000 ha in 2011 and continued to decrease down to 43 000 ha in 2012.

During the commercial forest fertilization upswing in the 1960's, 111.5 kg N ha^{-1}, in form of urea, was often applied (Kardell & Lindkvist, 2010; Lindkvist *et al.*, 2011). As field experiments increased foresters understanding of the relation between different sources of N and its effects on the forest surrounding the controlled experiments, the applied N form was changed to ammonium nitrate (NH_4NO_3) and later SkogCan (ammonium nitrate, about 27% N, with dolomite chalk and Boron), which is still commonly used. Simultaneously the N dose applied was increased to 150 kg N ha^{-1}. In more recent years there have been experimental field trials with organic N fertilizer

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[1] Pettersson, F., personal communication, February 5th, 2013. Forest Research Institute of Sweden.

(about 4% N, dry weight [DW]) produced from sewage sludge (Sahlén, 2006; Sahlén *et al.*, 2011). The effects of organic N is expected to extend over 15 years and early experiments have shown an increased production by 15 to 70% after adding 300 to 2000 kg ha^{-1}.

In commercial forestry N fertilization applied eight to nine years before final harvest increases net present value by about 11% and is considered one of the most profitable management practices in established boreal forests (Simonsen *et al.*, 2010). It is estimated that about 10% of Sweden's forest area has been fertilized since this management practice started (Lindkvist *et al.*, 2011; Näslund *et al.*, 2013). However, in the 1990's private and corporate forest owners could, in accordance with recommendations from the Swedish Forest Agency, fertilize suitable forest stands in northern Sweden with up to 600 kg N ha^{-1} rotation^{-1} but with no more than 200 kg per occasion (*cf* Näslund *et al.*, 2013). Today, the Swedish Forest Agency recommends that no more than 450 kg N ha^{-1} be applied during a rotation period in the northern parts of Sweden. For central Sweden no more than 300 kg is recommended and in southern Sweden a maximum of 150 or no fertilization at all, is advised.

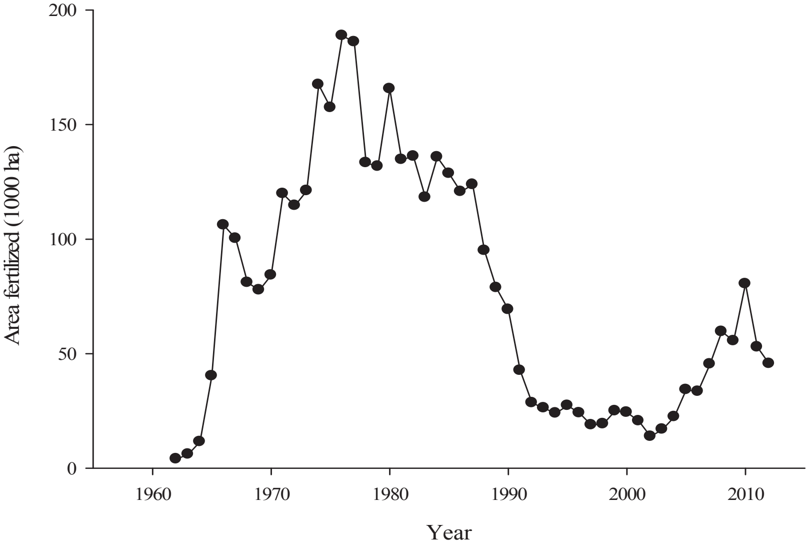

Figure 2. The forest area (in thousands of hectares) annually fertilized with N between 1962 and 2012. From 1986 through 2012 only data from the large-scale forestry sector is included. During this time period, small-scale private forest owners are estimated to have fertilized between 1000 and 3000 ha annually. Based on official national data from the Swedish Forest Agency (Anon, 2014).

1.2.2 Forest fertilization studies

Intentional forest fertilization has been studied in many ways and there seems to be some consensus within the research community - commercial N fertilizer rarely affects the system for more than 10 years (Nohrstedt, 2001; Saarsalmi & Malkonen, 2001). Until the present time, most of the previous Swedish forest fertilization studies have, with a few exceptions (Högbom *et al.*, 2001; Sikström, 2005; Johansson *et al.*, 2013), mainly studied the growth enhancing effects within one forest generation and regularly discarded the notion of effects extending over more than one stand rotation period. However, the current paradigm has been questioned in recent years. Studies have demonstrated long-term effects (>20 years since the last fertilization) on the composition of forest floor species (Strengbom & Nordin, 2008; Strengbom & Nordin, 2012), where normal commercial fertilization (150 kg N ha^{-1}) applied in the preceding rotation period increased the proportion of graminoids and decreased the proportion of heather (*Ericaceae*) species (mainly dwarf shrubs [*Vaccinium sp.*]).

Much of the added N when fertilizing forest is retained in the soil layer (Melin *et al.*, 1983; Melin & Nommik, 1988; Nommik & Larsson, 1989; Nohrstedt, 1990) and a lesser part in the trees. Tops and branches are relatively rich in nutrients (Mann *et al.*, 1988; Smith *et al.*, 1994; Olsson *et al.*, 2000; Harrington *et al.*, 2013), and are generally left after thinning and sometimes after clear-felling. The remaining harvest residues and needles that have been previously enriched by N fertilization may be transferred to the next tree generation after clear-felling. Furthermore, soil N mineralization rates and the amount of available soil nutrients generally increase after final felling and soil scarification (Vitousek & Matson, 1985; Rosén et al., 1996; Lundmark-Thelin & Johansson, 1997). Previously immobilized N may therefore become accessible to the subsequent tree generation when the mineralization rates increases after physical disturbances (*e.g.* soil scarification).

1.2.3 Past and current atmospheric nitrogen deposition

During the last century global emission rates of oxidized and reduced N increased rapidly due to human activity (*e.g.* the industrial revolution) which resulted in increased atmospheric N deposition rates (Lamarque *et al.*, 2005; Galloway *et al.*, 2008; Reay *et al.*, 2008). As a response to the increasing emission rates the global N deposition rates are predicted to increase further worldwide (Evans, 2001). Anthropogenic N deposition over Europe has long been a widespread phenomenon and according to the European Monitoring and Evaluation Programme (EMEP) model MSC-W the estimated deposition in 2011 was about 1 to 25 kg N ha^{-1} (UNECE, 2013). In Sweden the total deposition of reactive NH_4^+ and

NO_3^- is about 1 to 12 kg N ha^{-1} $year^{-1}$ (Fig. 3). Much of the N deposit over southern Sweden followed with prevailing winds mainly from the UK and Europe's northwest coast; however, although the emissions have decreased in northern Europe no general significant decrease in deposition rates has yet been observed in Sweden (Akselsson, C. 2013[2]).

Some N deposition is natural, *e.g.* formed by lightning in combination with nitrous oxide (N_2O), this natural formation and following deposition cannot easily be separated from the anthropogenic source, but it can probably be assumed that the proportion of natural deposition is a small fraction of the total deposition, especially in southern Sweden.

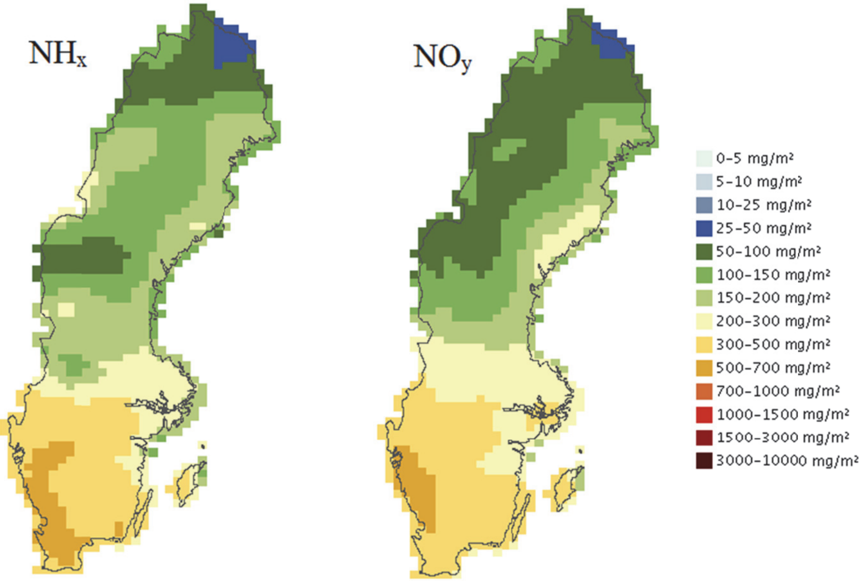

Figure 3. The total amount of reduced (NH_x) and oxidized (NO_y) N deposited over Sweden in 2012. The figure is based on the National survey of atmospheric chemistry data for the Swedish Environmental Monitoring, version AtmosphericChemistry2011.1, developed by SMHI on behalf of the Environmental Protection Agency.

1.2.4 The effect of nitrogen deposition on tree growth

The effect of deposited or unintentional N addition on boreal forest growth has not been studied to the same extent as forest fertilization. There are many indications that the ongoing deposition of N has increased tree production but natural N deposition is in essence hard to study. To exclude natural N

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[2] Akselsson, C., personal communication April 22nd, 2013. Swedish Environmental Research Institute.

deposition, experimental sites have to be enclosed in transparent material, *e.g.* roofing studies (Koopmans *et al.*, 1996; Corre & Lamersdorf, 2004). The roofing method also excludes natural rainfall which must be artificially simulated through expensive irrigation systems. However, this approach offsets the local environment and cannot in a good way substitute natural conditions. Because of these technical challenges, most of the previous studies on the effects of N deposition on tree growth in Europe have been correlative, *e.g.* Solberg *et al.* (2004) and Solberg *et al.* (2009). In these correlative studies the increment in tree growth per kg N (ha^{-1}) is often in the range of 1 to 2%, depending on *e.g.* tree species and site conditions. In a correlative landscape study Thomas *et al.* (2010) suggested that temperate forests sequestered about 60 kg C per kg of N added in above ground biomass.

Another approach is to locate the experiments to an area with low atmospheric N deposition. Under these circumstances, the effects of different N addition rates on tree growth can be studied with minimal disturbance to the natural environment. Most long-term stand-scale empirical studies of annual N addition on tree growth have, however, applied N at much higher rates (>30 kg N ha^{-1}) than is realistic for boreal and temperate forests (de Vries *et al.*, 2009). The C sequestered by trees in these N addition experiments have been shown to increase with about 25 kg per kg of N added (Högberg *et al.*, 2006; Hyvonen *et al.*, 2008).

Under ambient N conditions the proportion of N taken up by above ground vascular plants is usually non-linear (Templer *et al.*, 2012). By reviewing several ^{15}N tracer studies Templer *et al.* (2012) showed that under low N addition rates (≤8 kg N ha^{-1} year^{-1}) most of the N was generally immobilized in the soil layer whereas only a small proportion was sequestered in above ground vegetation. At higher N addition rates the proportion sequestered by above ground vascular plants increased and the proportion retained in the soil layer decreased. The increased sequestration in above ground vegetation may be caused by decreased competition from soil microbes, which have been shown to decrease after N addition (Treseder, 2008). The nonlinear N uptake by trees shown in these previous studies imply that the tree growth increments due to N addition should be nonlinear as well, since at low N addition rates a smaller proportion reaches the above ground vegetation.

1.3 Aim

The purpose of the studies upon which this thesis is based was to increase our understanding of long-term effects of N addition on tree growth in boreal forest ecosystems. To accomplish this, several variables associated with site fertility and tree growth have been measured in the field in areas with low background N deposition. Nitrogen was either applied annually at low rates ($\leq$12.5 kg N ha^{-1} year^{-1}), simulating N deposition in boreal forests and at a higher rate (50 kg N ha^{-1} year^{-1}) similar to previous long-term N addition studies (Paper II and III), or as normal commercial forest fertilization (150 kg N^{-1} occasion^{-1}) in the stand preceding the one studied (Paper I).

Specifically I have explored the following questions:

1. Are there long-term carry-over effects between forest stands following each other in time of commercial N fertilization on tree growth, available soil N and soil N mineralization rates?
(Paper I)

2. What are the tree growth responses to long-term annual N addition at low rates in mature forests dominated by *P. sylvestris* or *P. abies*?
(Paper II and III)

2 Materials and methods

The method section herein is a summarized description of the materials and methods in Papers I through III. I refer to the respective Paper and references cited therein for more detailed descriptions.

This thesis focuses on two economically vital tree species for Swedish forestry, *P. abies* and *P. sylvestris*. The conifer forest stands included in studies I, II and III were all representative of Sweden's boreal forests in terms of soil type, standing stock and abiotic conditions, and met the seven recommendations for forest fertilization developed by the Swedish Forest Agency and the Forestry Research Institute of Sweden in 1988 (Pettersson & Palmér, 1988; Pettersson *et al.*, 1988), namely: (i) Solid ground, (ii) podzolic soil, (iii) Site Index (H_{100}) between T16≤ and ≥G30, (iv) where conifer species constitute ≥80% of total stand basal area, (v) the forest stand is at least suitable for first commercial thinning, (vi) the forest stand should not be harvested within 10 years and (vii) the stand should be healthy and sufficiently stocked.

2.1 Paper I

I revisited previously studied sites where changes in field vegetation as an effect of previous commercial fertilization had been documented (Strengbom & Nordin, 2008; 2012). The correlation between the previous fertilization and the compositional changes in forest floor vegetation prompted me to further investigate whether long-term fertilizer-induced changes in tree growth could be found in the young (10 years) conifer trees. I analyzed tree growth and stand biogeochemical variables in second rotation stands where normal commercial N fertilizer had been applied once (as 150 kg N ha^{-1}, 25 years before our study) or twice (2 x 150 kg N ha^{-1}, 33 and 25 years before our study) with the last N addition about 10 years before final harvest. The experiment was treated as a completely randomized design with the number of factors (k)=1, number of levels (L)=3, and number of replicates (n)=7. The fertilized stands are hereafter noted as sites with treatment N1 (150 kg N ha^{-1}) and N2 (2 x 150 kg N ha^{-1}). The atmospheric N deposition in this region is about 2.5 to 3.5 kg ha^{-1} year^{-1} (Phil-Karlsson *et al.*, 2011). Final felling, soil scarification and regeneration (planting) were performed about ten years before field measurements, with the exception for the forest floor vegetation which was measured in 2008 and 2009. There was no difference in time since clear-cutting between fertilized and unfertilized sites, for a full description of the studied sites see Strengbom and Nordin (2008).

Tree growth was measured as annual height growth increments, *i.e.* distance between nodes, from first visible node through full tree height and DBH was measured with a caliper. I decided to measure tree height growth as annual increments as a similar study from North America showed that a growth difference occurred first after a few years after seedling establishment (Footen *et al.*, 2009). This design was chosen in order to detect at what tree age any potential alteration in growth would occur. In addition to apparent indicators of tree productivity, such as height and diameter, needle N concentration was also measured. Samples were collected from top branches, dried at 70°C for 24h, grinded in a bead mill and analyzed for C and N concentrations.

The ground vegetation was inventoried with the pin-point intercept method (Strengbom *et al.*, 2002) at 200 random points along a 45 m transect in each forest stand in August 2008 and in August 2009. All contacts with each individual species along a plastic pin were counted as it passed through the vegetation, see Strengbom and Nordin (2008; 2012) for details. Abundances of Wavy Hair-grass (*Deschampsia flexuosa* [L.] Trin.) and Bilberry (*Vaccinium myrtillus* L.) were included in Paper I. I considered that the compositional alterations in forest floor vegetation found by Strengbom and Nordin (2008; 2012) could be correlated to changes in soil N turnover. Therefore, soil N mineralization rates and the amount of exchangeable soil ammonium (NH_4^+) and nitrate (NO_3^-) ions were measured. The soil N availability was measured with resin (ion-exchange) capsules, buried just under the mor-layer in the beginning of the growing season (1st through 3rd of June) and retrieved between the 28th and 30th September. The gathered samples were kept in plastic bags and transported to a lab for analyses. The capsules were extracted in 1M KCL (3 x 7 ml) and analyzed using a FIAstar 5012 Analyzer (Tecator, Höganäs, Sweden). Before statistical analyses the replicate samples from the same site were pooled.

Soil mineralization rates of NH_4^+ and NO_3^- were measured with the buried bag technique. Soil samples were collected at the same time as the capsules were buried, half of the samples were sent to a lab for analysis and the other half were put in plastic bags and deposited at the same locations as the resin capsules. The bags were retrieved at the same time as the resin capsules (*i.e.* 28th and 30th September) and sent to a lab for analyses. The soil samples were extracted in 1M KCL and analyzed for NH_4^+ and NO_3^- content (mg g^{-1} DW soil) using an AutoAnalyzer 3 (SEAL Analytical). The soil samples from each site were pooled before statistical analysis.

2.2 Paper II

I analyzed the effect of continuous low N addition on tree growth in two different forest types, one *P. abies* dominated mesic site and one *P. sylvestris* dominated heath site. The studies were conducted at Vindeln Experimental Forest (Unit for field based research, SLU) where N deposition was simulated by manually spreading ammonium nitrate (NH_4NO_3) by hand each spring (early May) before the commencing growth season in amounts corresponding to past and present deposition rates over large parts of Europe (Phil-Karlsson *et al.*, 2011; Wenche, 2012; Schulz *et al.*, 2013) and at a higher rate commonly included in previous annual N addition experiments in the boreal region (Högberg *et al.*, 2006; Hyvonen *et al.*, 2008). A database of N deposition over Europe is presented online by Long-range Transboundary Air Pollution (CLRTAP) for international co-operation to solve transboundary air pollution problems (UNECE, 2013). During this experiment the background N deposition in this area was 1 to 2 kg N ha^{-1} $year^{-1}$ (Phil-Karlsson *et al.*, 2011).

The two forest ecosystems have been fertilized with 3 to 50 kg N ha^{-1} $year^{-1}$ since 1996 in the *P. abies* stand (0.1 and 0.25 ha plots, of which 0.1 ha was included in Paper II and III) and since 2005 in the *P. sylvestris* stand (0.1 ha plots). The treatments in the *P. abies* site were 0, 12.5 and 50 kg N ha^{-1} $year^{-1}$ (k=1, L=3, n=12), and 0, 3, 6, 12.5 and 50 kg N ha^{-1} $year^{-1}$ in the *P. sylvestris* site (k=1, L=5, n=6). For simplicity the treatments are hereafter represented as the amount N applied in kg and the letter N (*i.e.* 3N, 6N, 12.5N and 50N for 3, 6, 12.5 and 50 kg N ha^{-1} $year^{-1}$), or as low N addition rates (≤12.5kg N ha^{-1} $year^{-1}$) and high N addition rate (50 kg N ha^{-1} $year^{-1}$). For a full description of the *P. abies* site refer to Strengbom *et al.* (2002) and to Gundale *et al.* (2011) for the *P. sylvestris* site.

I acknowledge that manually spreading N fertilizer in the beginning of each growing season is not interchangeable with N deposition because of temporal differences and canopy uptake. N uptake via interception in boreal vegetation has, however, been shown to be of minor importance (Boyce *et al.*, 1996; Adriaenssens *et al.*, 2012). An exact replication of natural N deposition is difficult to practically achieve without expensive irrigation systems.

2.2.1 Sample trees, increment cores and WinDendro

Sample trees in Paper II were selected to represent the plot's basal-area weighted mean diameter, Dg (VanSoest *et al.*, 1959), This method was preferred over other methods, such as arithmetic mean diameter, because of the exact relationship between Dg, the number of trees and the total stand basal area (Curtis & Marshall, 2000). I sampled the four trees of each plot that were closest to the sampled plots Dg.

Current year needles were collected from the sample trees from branches facing south. The needles were oven dried (65°C for 24 hours), grinded with a bead mill to a fine powder and analyzed for C and N concentrations. Increment cores were extracted at DBH from sample trees, these increment cores were analyzed in WinDendro 2008 (Regent Instruments, Quebec, Canada). WinDendro is a commonly used software program for dendrochronology, in which annual year tree ring widths can be measured from scanned images of tree cross sections and increment cores (Fig. 4). The basal-area growth and relative growth increment reported in Paper II and the biomass growth in Paper III were calculated with the annual year ring widths obtained in WinDendro.

This non-destructive growth measurement method was chosen over cutting down the trees since the experiment is still ongoing. The increment cores were taken from the same cardinal direction and misshapes, *e.g.* from wind exposure, should thus be approximately evenly distributed over plots and treatments.

The first years after N fertilization the tree growth response varies between stands (Sikstrom *et al.*, 1998; Nohrstedt, 2001), and the diameter growth response to N addition increases progressively during the first years following the fertilization event (Valinger *et al.*, 2000). Hence, when calculating the relative basal-area growth response to N addition (Fig. 9) the first five years of the fertilization was excluded to allow for the initial adaption phase for trees to express a full growth response to N input to pass.

Figure 4. A scanned image of a Norway spruce (*P. abies*) increment core from the unfertilized control plots prepared for analysis with software program WinDendro. Photo: Fredrik From.

2.3 Paper III

The effect of low annual N additions on whole ecosystem biomass production and N allocation were studied further on the *P. abies* sites from Paper II. To estimate the forest floor biomass, allometric equations were established from analyzing harvested above and below ground samples of *V. myrtillus* and *D. flexuosa*. The pin-point intercept method (Strengbom *et al.*, 2002) was used to estimate *Vaccinium* sp. and *D. flexuosa* abundance and with the allometric equations converted this to biomass per unit area. The biomass ha^{-1} of feather mosses *P. schreberi* and Glittering Wood-moss (*Hylocomium splendens* [Hedw.] B.S.G.) was estimated by multiplying the mean shoot density (shoots per unit area) with the mean shoot mass (of collected samples) and again multiplying with the visually-determined coverage (converted to a per ha basis). The density was measured in five 10 cm circles within each plot where the species covered 100% of the bottom layer. The shoot mass was estimated from 25 shoot samples from both species from five locations within each plot. Feather moss coverage was determined by visually estimating percent coverage of ground vegetation in 15 randomly placed 0.25 m^2 rectangles within each plot.

With the backward chronological analyses of the annual year ring widths (Fig. 5) the mean plot tree diameters was determined for: the end of the study period (2009), before treatments started (end of 1995) and 14 years before that (1981). *P. abies* biomass for each time point was then estimated with the mean plot tree diameter (Paper II), using Marklund's (1988) allometric equations G3, G12, G15, G16 and G23 for stem, branches, needles, dead branches, stump and the root system. The C content of the studied partitions was estimated to be 50% of the total biomass (DW) and the total tree C ha^{-1} was estimated by multiplying the tree C content with the average number of trees ha^{-1} on the site. The annual increase in C biomass per year was estimated by dividing the total increase in tree C ha^{-1} between 1981 and 1995, and 1995 and 2009 with the length of the treatment period, *i.e.* 14 years. The net change in tree biomass C ha^{-1} during the fertilization period (1995 to 2009) was estimated by subtracting the growth during 1981 through 1995 for each plot. The plot relative change in tree biomass C per unit of N fertilizer added was calculated by subtracting the mean change in the control plots, and then by dividing the annual N addition rates. The net changes in tree biomass C per unit N added was combined with the annual change in understory biomass C during the fertilization to attain a vegetation C sequestration rate per kg N added.

Figure 5. A scanned image of a Norway spruce (*P. abies*) increment core from the high N addition treatment with the time periods studied in Paper III highlighted. The pre-fertilization period (14 years) ranged from end of the growing season in 1981 to the end of the growing season in 1995, the fertilization period (14 years) ranged from the end of 1995 to the end of the growing season in 2009. Photo: Fredrik From.

Besides the biomass estimations the allocation of added N in this ecosystem was assessed. N allocation within different ecological compartments was identified in a field tracer study, in the spring of 2011, 99 atom% ^{15}N-labeled $^{15}NO_3{}^{15}NH_4$ was applied in 15.5 m^2 circular sections surrounding six pre-selected sample trees (Fig. 6). The labeled isotope was applied with a standard watering can the 16th of June and samples from the studied compartments, *P. abies* roots and needles, and forest floor above and below ground vegetation, were acquired on September 5th 2011. Three samples from each plant part were collected from different locations in the 15.5 m^2 plots. Foliar tissue samples from feather mosses, *D. flexuosa*, *V. myrtillus* and *P. abies* were collected from each plot where no label had been applied to assess natural abundance of δ^{15}N and %N for each species. Before analyses, all samples were dried in an oven (65°C) for 24 hours and grinded to a fine powder in a ball mill. A Flash2000 Elemental Analyzer connected to a DeltaV Advantage Isotope Ratio Mass Spectrometer was used to assess the δ^{15}N and %N in the collected samples. The percentage of ^{15}N taken up by the different vegetation pools was estimated by calculating the difference ^{15}N atom% between labeled and unlabeled plants and then by converting the excess atom% in the labelled plants to micromole excess ^{15}N g^{-1} plant DW. This value was then multiplied by the biomass (DW) ha^{-1} for each vegetation pool, and finally by dividing this value with the total molar quantity of label ha^{-1} applied to each plot. See Gundale *et al.* (2014) for a detailed description of the ^{15}N allocation.

Soil nutrient availability was assessed with five ion-exchange resin capsules per plot in 2010. The capsules were buried 5 cm below the humus layer within one week after snowmelt (early May) and collected in November (2010). The capsules were extracted in 1M KCL and analyzed for NH_4^+ and NO_3^- using an Autoanalyzer (Omni Process, Solna, Sweden).

Figure 6. Paper III: preparation before applying $^{15}NH_4{}^{15}NO_3$ in June 16th 2011, Svartberget Experimental Forest, Vindeln. Portrayed are *P. abies* trees with treatment 50 kg N ha^{-1} $year^{-1}$ and researcher Dr. Michael Gundale. Photo: Fredrik From.

2.4 Statistical analyses

The software programs Minitab (v.16), MYSTAT (v.13) and SPSS (v.20) was used for the statistical analyses in Paper I through III. For a detailed description of the linear regressions, analyses of variance (ANOVA) and the repeated measure ANOVA see the statistical paragraphs in each Paper. All variables were checked for normal distribution and homoscedasticity, and when these requirements were not met, data was transformed to meet these requirements. The transformation of skewed data was consistently done with log(x+1) for uniformity in Papers I through III. Mean values (untransformed) ±1 standard error (SE) are depicted in figures and tables.

3 Results

3.1 Paper I

Height growth rate (Fig. 7) was greater in the young trees (about 10 years old) grown on sites that had been fertilized twice in the preceding rotation period than trees in control stands. On sites where two previous fertilizations had been applied the trees were about 24% taller than trees on unfertilized control sites ($F_{2.857,\ 25.715}$=5.09, *p*=0.018). Trees on previously fertilized sites (N1 and N2) also had about 15% more N in their current year needles compared to unfertilized controls.

It took two previous fertilizations for effects on soil mineralization and mobile soil N to remain evident 25 years later (Paper I, Table 2). Analyses of resin ion exchange capsules showed that the previously fertilized stands (N2) had about twice the amount of available NO_3^- and NH_4^+ in the soil organic layer than in control stands. The mineralization rates measured with the buried bags showed that the rate was about 3.7 times higher on sites with two previous fertilizations than in the control and sites which were only fertilized once.

A stepwise backward elimination regression analysis of the relation between tree height as response and the abundance of *D. flexuosa*, *V. myrtillus*, the needle N concentration, soil mineralization rates and the amount of mobile soil N, showed that the needle N concentration and the amount of mobile soil N was relevant as explaining factors for mean tree height ($R^2_{(adj.)}$=0.45, *F*=9.14, *p*<0.001).

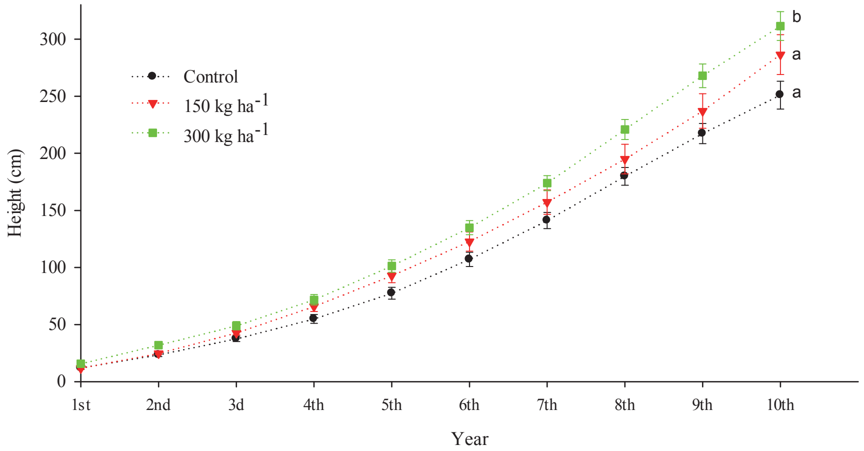

Figure 7. The mean (±1SE) annual tree height on the surveyed sites (n=7) during the first ten years after regeneration. Treatments were the Control (no fertilization), N1 (150 kg N ha^{-1} applied once, 25 years prior to this study) and N2 (150 kg N ha^{-1} applied twice, eight years apart, 25 and 33 years prior to this study). Different lower case letters (a or b) indicate significant post hoc difference between treatments ($F_{2.857, 25.715}$=5.09, p=0.018).

3.2 Paper II

For the high N addition rate (50 kg N ha^{-1} year^{-1}) the *P. abies* and *P. sylvestris* relative basal-area growth (Fig. 8) increased after the first couple of applications and continued to increase for about six years before leveling off at a steadily higher growth rate than the control. The high N addition also increased the needle N concentration in *P. abies* and *P. sylvestris* (Paper II, Table 1). For the lower N addition rates (≤12.5 kg N ha^{-1} year^{-1}) I found contrasts between the two species as no visible effects from N addition on *P. sylvestris* growth or needle N concentration could be detected, whereas both growth and needle N concentration increased in *P. abies*.

A simple linear regression analysis showed that the average relative basal-area growth rate increased by ca 2.5% per kg N added on the *P. abies* site (Fig. 9, R^2=0.66, F=68.76, p<0.001). No satisfying linear relationship for *P. sylvestris* was evident as the model did not properly fit the data in a pure error lack-of-fit test (p-value=0.015<α=0.05) after removing the constant term, which was removed to out rule negative relative basal-area growth response to N addition.

A regression analysis showed that there was a significant linear correlation between the needle N concentration and relative growth for *P. abies*, and as both relative basal-area growth and needle N concentration were unaffected by the low N addition rates this was also true for *P. sylvestris* (Fig. 10). The analyses also showed that the slopes were roughly similar per unit increase in needle N; an increase in needle N concentration by 1 percent unit theoretically increased average relative basal-area growth with about 420% for *P. abies* and 450% for *P. sylvestris*.

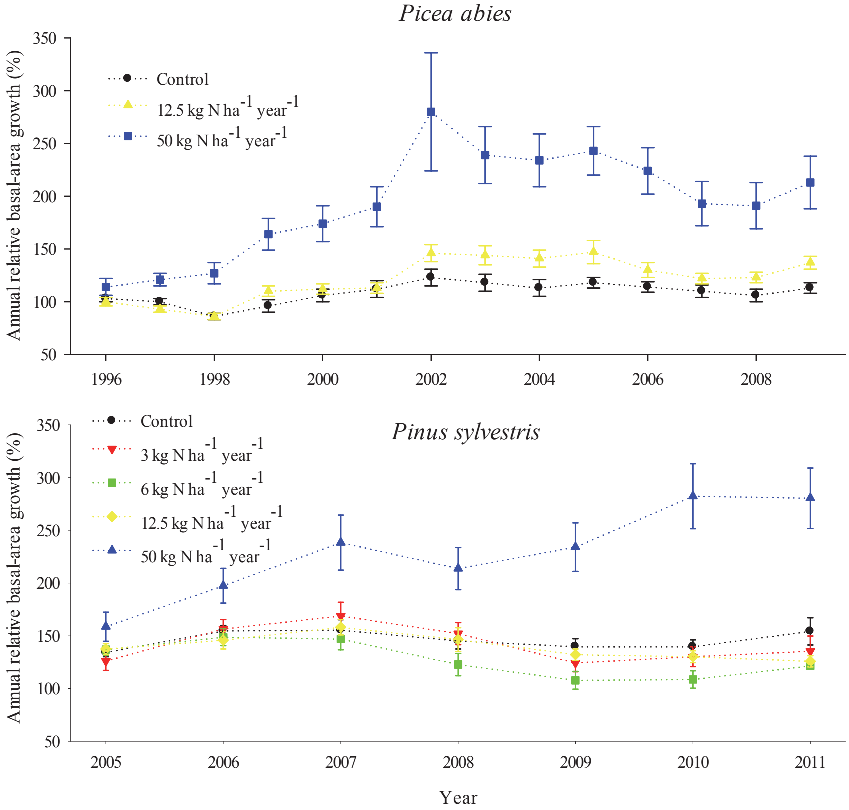

Figure 8. Annual relative basal-area growth (each year's basal-area growth in relation to the average annual basal-area growth over the five-year period preceding the start of N additions) at two N addition experimental sites in boreal forests: plots at the *P. abies* dominated site had been treated with 0, 12.5 and 50 kg N ha^{-1} year^{-1} for 14 years (n=12) at the time of this study, while plots at the *P. sylvestris* dominated site had been treated with 0, 3, 6, 12.5 and 50 kg N ha^{-1} year^{-1} for seven years (n=6). An annual relative basal-area growth of 100% denotes equal growth between the period before fertilization started and the fertilization period.

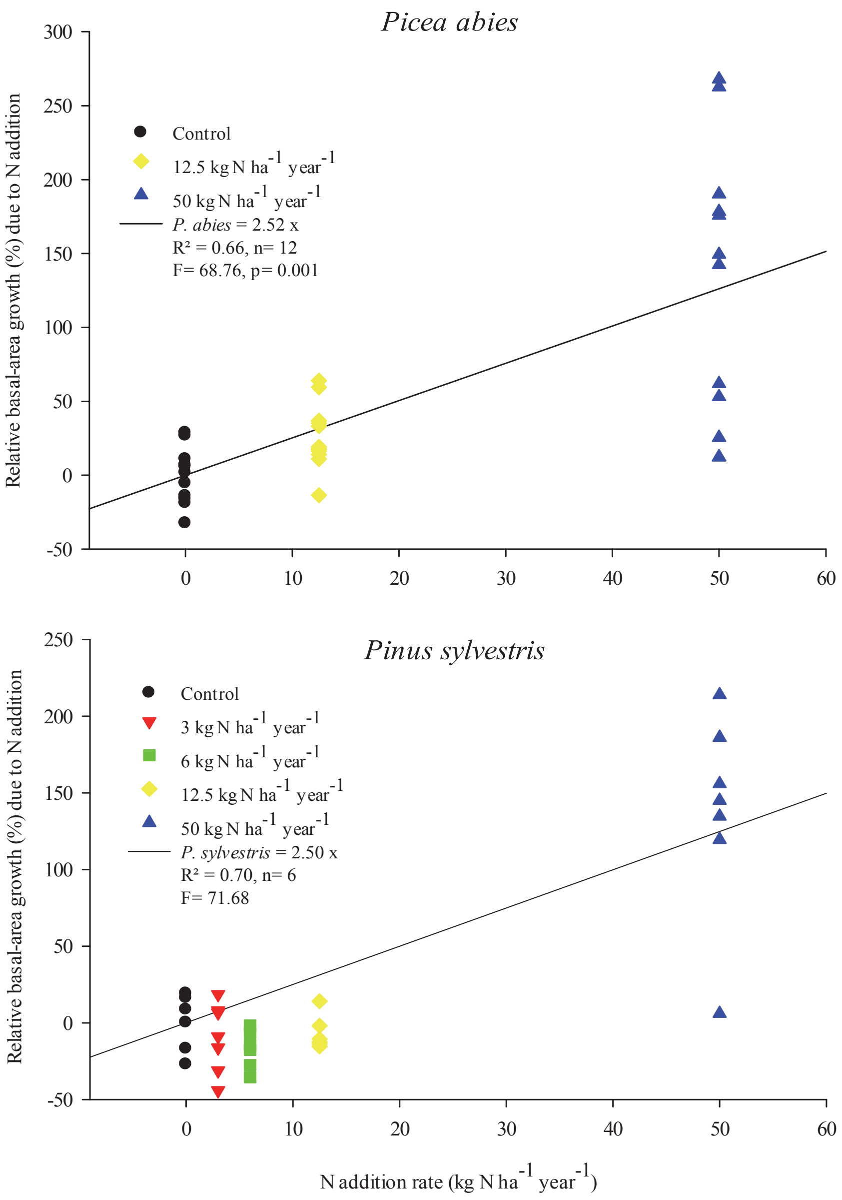

Figure 9. Average plot relative basal-area growth increment per kg N added at two N addition experimental sites in boreal forests after simulated N deposition treatments; plots at the *P. abies* dominated site had been treated with 0, 12.5 and 50 kg N ha^{-1} year^{-1} for 14 years (n=12) at the time of this study, while plots at the *P. sylvestris* dominated site had been treated with 0, 3, 6, 12.5 and 50 kg N ha^{-1} year^{-1} for seven years (n=6). The plots relative basal-area growth increments from N addition were normalized by subtracting the mean relative basal-area growth of the controls. The model did not fit the data properly for *P. sylvestris* (pure error lack-of-fit test p-value=0.015<α=0.05).

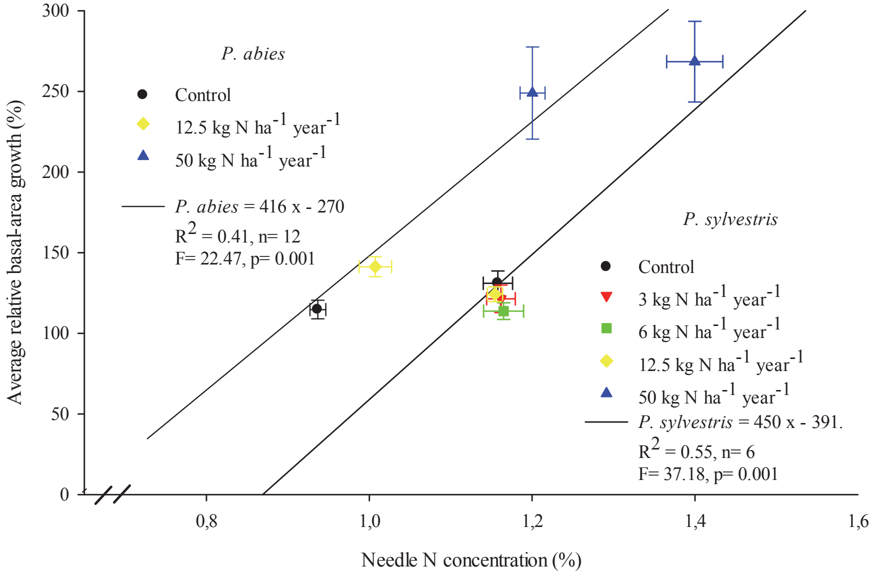

Figure 10. The correlation between needle N concentration (%, DW) and the average relative basal-area growth during the last nine years of N addition for *P. abies* (upper left hand side) and two years for *P. sylvestris* (lower right hand side) after simulated N deposition treatments. Plots at the *P. abies* dominated site had been treated with 0, 12.5 and 50 kg N ha^{-1} year^{-1} for 14 years (n=12), while plots at the *P. sylvestris* dominated site had been treated with 0, 3, 6, 12.5 and 50 kg N ha^{-1} year^{-1} for seven years (n=6) at the time of this study. An average relative basal-area growth of 100% denotes equal growth between the period before fertilization started and the fertilization period. Fertilization started in 1996 and needles were collected in 2009 in the *P. abies* experiment and in the *P. sylvestris* experiment fertilization started in 2005 and needles were collected in 2011.

3.3 Paper III

Nitrogen concentrations in the measured plant tissues were consistently higher in plots with the high N addition rate (50 kg N ha^{-1} year^{-1}) than in the control plots (Paper III, Table 1), whereas the low dose treatment (12.5 kg N ha^{-1} year^{-1}) did not alter the N concentrations significantly. In plots with the high N addition rate the understory biomass was lower than in the control plots (Paper III, Table 2), primarily due to a significant decrease in feather mosses, although, the abundance of *D. flexuosa* increased in response to the high N addition.

Analysis of the resin capsules showed that the amount of mobile soil NH_4^+ and NO_3^- increased in the high N addition plots compared to the control and low N addition plots (Paper III, Fig. 1).

The labeling experiment showed that it was only in the above ground parts of *V. myrtillus* that the N addition treatments affected ^{15}N allocation, for the other fractions no significant changes occurred across treatments. More ^{15}N was allocated to new and old parts of *V. myrtillus* in plots with treatment 50N than in control plots (Paper III, Table 2). After up scaling, approximately one-half of the label applied was detected in the measured plant pools and about as much was found in the humus layer (Paper III, Fig. 2). The largest partition of ^{15}N was located in the feather mosses, where a larger proportion was sequestered in the low N and control plots compared to the high N plots. *D. flexuosa* only sequestered a small proportion of the added label and the amount was higher in N addition plots than in control plots. *V. myrtillus* sequestered about 10 to 13% and *P. abies* sequestered about 7 to 9% of the applied label, and the amounts did not differ across the N addition treatments in any of the two pools.

Biomass C sequestration of *P. abies* increased linearly to increasing N addition rates (Paper III, Fig. 3a), and the total biomass C sequestration of all vegetation pools measured increased with about 16 kg C per kg N added ($R^2_{(adj.)}=0.563$, $p<0.001$, Paper III, Fig. 3b). The net C sequestration in *P. abies* only was about 19 kg per kg N added when not compensating for the changes in the understory biomass (data not shown).

4 Discussion

4.1 Does annual nitrogen addition increase tree carbon sequestration?

Boreal forests are generally thought of as a net C sink (Kurz *et al.*, 2013), and it has been suggested that N deposition is important for the C sequestration in boreal forests (de Vries *et al.*, 2006; Gruber & Galloway, 2008). Nitrogen deposition in combination with increased levels of atmospheric CO_2 and climate change were advocated as explanatory factors for the increased tree growth in British Columbia during the latter part of the 20th century (Wu *et al.*, 2014). Although the long-term effects of N deposition on the ability of forests to function as C sinks have been questioned (Hyvonen *et al.*, 2008; Bala *et al.*, 2013), it has been proposed that climate change, for example, will eventually overshadow the effects from N deposition. Nevertheless, there is a consensus that N deposition usually increases the growth of boreal and temperate forest and will likely continue to do so in the foreseeable future (Solberg *et al.*, 2004; Högberg, 2007; Pregitzer *et al.*, 2008; Sutton *et al.*, 2008; Bedison & McNeil, 2009; de Vries *et al.*, 2009; Laubhann *et al.*, 2009; Solberg *et al.*, 2009; de Vries & Posch, 2011).

In accordance with Wu *et al.* (2014), our data (Paper II and III) show that long-term annual N addition at low rates can increase tree growth, and consequently the C sequestration in *P. abies*. For *P. abies* the rate at which C was sequestered was around 19 kg per kg of N added, although when compensating for the reduced C sequestration in the forest floor vegetation the total biomass accumulation rate was about 16 kg C per kg N. The relationship in Paper III is lower than those reported in previous studies in boreal forests, *i.e.* around 25 kg C per kg of N added (Högberg *et al.*, 2006; Hyvonen *et al.*, 2008). However, the C sequestration per unit N is likely to vary between soil types, tree species and stand ages (Thomas *et al.*, 2010).

Anthropogenic emissions of N compounds are expected to increase during the 21st century (Galloway *et al.*, 2004; Lamarque *et al.*, 2005; Dentener *et al.*, 2006), hence the N deposition rates will continue to increase worldwide (Evans, 2001). As one of the largest biomes in the world, the circumpolar boreal forest region covers about 11% of the global land surface (Sabine *et al.*, 2004), and an increased boreal tree growth rate due to N addition could therefore sequester a large part of emitted CO_2. Although, about 72% of the boreal region receives only ≤3 kg N ha^{-1} year^{-1}, *cf* Gundale et al. (2011), which according to our data would not increase growth in *P. sylvestris*.

The prolonged increase in site fertility and tree growth after fertilization of the previous stand shown in Paper I implies that forest fertilization with N followed by physical disturbance (*e.g.* clear-cutting and soil scarification) will not only increase the C sequestration in the stand subjected to fertilization but also in the subsequent forest stand. Forest fertilization may therefore increase the long-term (>25 years) C sequestration in boreal forests. Further studies are needed to determine whether the increased growth will persist or in time level off.

4.2 Long-term effects of nitrogen fertilization on site productivity

Most of the previous Swedish forest fertilization studies have, with a few exceptions (Högbom *et al.*, 2001; Sikström, 2005; Johansson *et al.*, 2013), mainly studied the growth enhancing effects within one forest generation and regularly discarded the notion of persisting long-term effects (>10 years) (Nohrstedt, 2001). Significant increments in several variables associated with site productivity were found in the subsequent stands 25 years after normal commercial forest fertilization of the preceding stands (Paper I). In general it took two fertilizations with 150 kg N ha^{-1} (N2) of the preceding stand for site alterations to be detectable in the second tree rotation in our study. At the time of our study, the regenerated young trees (about 10 years) growing on sites where the preceding tree stands had been fertilized with 150 kg N ha^{-1} twice were about 24% taller than unfertilized controls (Fig. 7). On these previously fertilized sites increased amounts of mobile soil N and increased soil mineralization rates were also detected. These increments were reflected in the needle N concentrations of the young trees, which was about 15% higher on fertilized sites than on control sites (Paper I, Table 2).

A relatively large proportion of added N when fertilizing forests is retained in the soil layer and a lesser part is sequestered in the trees (Melin *et al.*, 1983; Melin & Nommik, 1988; Nommik & Larsson, 1989; Nohrstedt, 1990). It is common that the soil mineralization rates and the amount of mobile soil N increases after physical disturbances such as soil scarification and clear-cutting, generally due to increased activity of soil microbes (Vitousek & Matson, 1985; Lundmark-Thelin & Johansson, 1997), clear-cutting especially has been shown to mobilize large amounts of formerly immobilized N (Rosén *et al.*, 1996). Previously added N retained in the soil stratum may therefore be available to regenerated trees after soil preparation following final harvest as mineralization rates increases. In accordance with these previous studies on the interactions between soil disturbance and N mobility, the soil mineralization rates were increased on sites that were fertilized twice in the preceding tree rotation than on unfertilized control sites. In addition to the increased mineralization rates, the amount of mobile soil N absorbed by ion-exchange capsules was also higher in previously fertilized sites than in unfertilized control sites. The size of the plant available N pool has been correlated to the amount of N in tree needles (Binkley & Reid, 1985), and a close positive relationship between a high needle N concentration and tree growth has been documented (Bauer *et al.*, 1997; Iivonen *et al.*, 2006). The data in Paper I confirm this positive relationship in a multiple regression analysis between tree growth and the amount of mobile soil N and the needle N concentration. In addition to the changes in variables associated with stand productivity, a fertilizer induced shift in the composition of the forest floor vegetation has been documented on the sites studied in Paper I (Strengbom & Nordin, 2008; Strengbom & Nordin, 2012), and it appears that the commercial fertilization of the preceding tree stands has enhanced the site productivity in the long term (>25 years).

Currently there are only a few high quality studies in this particular subject that are published, one from North America (Footen *et al.*, 2009) and three from southern Sweden (Högbom *et al.*, 2001; Sikström, 2005; Johansson *et al.*, 2013), and there seems to be some discrepancy regarding long-term carry-over effects from forest fertilization. Footen *et al.* (2009) found that the fertilization of the previous tree stand did increase the growth of second rotation Douglas fir (*Pseudotsuga menziesii* [Mirb.] Franco), whereas Johansson *et al*. (2013) in a study of scarification and pre-harvest fertilization did not detect a higher growth in *P. sylvestris* seedlings that grew on sites with previous N addition than seedlings on unfertilized sites. However, our study highlights that it may take more than a few years before a growth-enhancing effect of previous fertilization can be discerned. Also, the ecosystems in these studies are different and only by few means comparable.

In Sweden about 10% of the managed forest land area has been subjected to fertilization in the period from 1960 to 2010 (Lindkvist *et al.*, 2011; Näslund *et al.*, 2013). In order to make informed forest management decisions it is important to elucidate which forest stands that are likely to be more susceptible to long-term alterations due to N addition in the preceding tree rotation period since, for example, present growth models for predicting yield might need adjustment due to residual growth effects of previous fertilization.

4.3 Long-term effects of annual nitrogen addition in two diverse forest sites

To my knowledge, no long-term stand-scale experiments, other than the one described herein, with N addition rates close to N deposition rates over boreal forests have been reported (de Vries *et al.*, 2009). Previous studies relating tree growth to N deposition have mostly been indirect, correlating current deposition to growth (Solberg *et al.*, 2004; Högberg, 2007; Pregitzer *et al.*, 2008; Sutton *et al.*, 2008; Bedison & McNeil, 2009; de Vries *et al.*, 2009; Laubhann *et al.*, 2009; Solberg *et al.*, 2009; de Vries & Posch, 2011). Paper II and III present experimental data that partly confirm these previously correlative studies on the relationship between boreal coniferous tree growth and low annual N additions. Annual N additions increased the amount of mobile soil NH_4^+ and NO_3^- in the *P. abies* high N addition plots compared to the low N and control plots (Paper III, Fig. 1). A close relationship between available soil N and tree needle N concentration is often observed, *e.g.* Binkley and Reid (1985), in accordance, the needle N concentration was higher in the N addition plots than in control plots for *P. abies*. Moreover, in Paper III the N concentrations were higher in all vegetation pools studied (*P. abies*, *V. myrtillus*, *D. flexuosa* and feather mosses) in the high N addition plots compared to the control plots.

As N uptake by above ground vegetation often is nonlinear (Templer *et al.*, 2012), with N uptake by trees being higher at higher N addition rates (Nadelhoffer *et al.*, 2004), I hypothesized that the tree growth response to increasing N addition would be nonlinear as well. Although, in *P. abies* there was no evidence of a nonlinear growth response to N addition as relative basal-area growth increased by about 2.5% per kg N added (Fig. 9). In terms of C sequestration (Paper III), about 19 kg of C per kg N added (total C accumulation in biomass was 16 kg and 19 kg for *P. abies* only). The field tracer experiment also showed that the amount of ^{15}N taken up by *P. abies* did not differ between the N addition treatments as an equal part (7 to 9%) was sequestered by the 12.5N, 50N and control trees (Paper III, Fig 2). This suggests that the N uptake by *P. abies* is linear, otherwise a larger proportion of the added ^{15}N would have been sequestered by trees on high N plots, although, the N allocation in *P. abies* could also have shifted after fertilization.

However, I did not find a linear relationship for *P. sylvestris* as the growth was not altered by the low N addition rates, this may be a question of N sequestration. In a study of two long-term fertilization experiments, Ladanai et al. (2007) found that in *P. abies* stands the trees were a major sink for added N, whereas in *P. sylvestris* stands the soil was the dominant N sink. The soils were not all homogenous between sites and the authors stressed that the results should not be generalized. More precise studies of the N uptake have been done in the present study experimental forest. In a study based on shifts in natural abundance of ^{15}N in bryophytes, dwarf-shrubs and trees at the same *P. sylvestris* site as in the present study, Gundale *et al.* (2011) showed that it was only in the high N addition plots that trees took up a part of the added N and allocated it to above ground parts. The nonlinear N uptake and allocation to above ground parts on the relatively nutrient poor *P. sylvestris* site is in accordance with a recent study on symbiotic C to N exchange in ectomycorrhizal fungi by Näsholm *et al.* (2013). In a nutrient poor *P. sylvestris* ecosystem, not far from the sites studied in Paper II and III (ca 10 km), a ^{13}C and ^{15}N pulse-label experiment showed that soil microbes and mycorrhizal fungi sequestered a relatively large proportion of the added N whereas only a small proportion could be detected in the tree canopies. Näsholm *et al.* (2013) also noted that the N allocation shifted to the tree canopies when N fertilizer had been applied beforehand. Moreover, a meta-analysis of 82 N addition studies showed a 15% decrease in soil microbial biomass in response to N addition (Treseder, 2008). N fertilization may therefore diminish the impact of soil microbes on N immobilization in nutrient poor ecosystems.

Further studies are needed to elucidate where in the two ecosystems added N is retained and whether the nonlinear relationship between tree growth and N addition on the *P. sylvestris* site will change into a more linear relationship with time.

5 Conclusions and recommendations

In this thesis I have made two major findings: 1) that commercial forest N fertilization according to normal practice can have residual effects on site fertility and tree growth that stretches across tree rotation periods, and 2) that annual additions of N caused a positive linear growth response in *P. abies*. In *P. sylvestris* the low N addition rates did not increase growth, whereas high N addition rates increased the growth and needle N concentration.

I recommend that long-term effects of forest fertilization should be considered in forest management practice. I also recommend that the tree growth response to annual N addition at low rates receives further attention by researchers. The difference in growth response to the low N addition rates between *P. abies* and *P. sylvestris* especially calls for further attention.

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Acknowledgements

I would like to thank my supervisors Annika, Tommy, Torgny and Tomas for all their invaluable guidance. I also thank Ann for helping me with the lab analyses and field work. I am grateful for the support I have received from co-workers Linda and Petter. I also thank Allan and Linda for proof reading of this thesis.

I thank Gillis and Kerstin for everything. I give a big bark to Igor for telling me when it is time to leave the computer and go outside for a bit of fresh air. An even bigger bark goes out to Elin for all her love and support.