

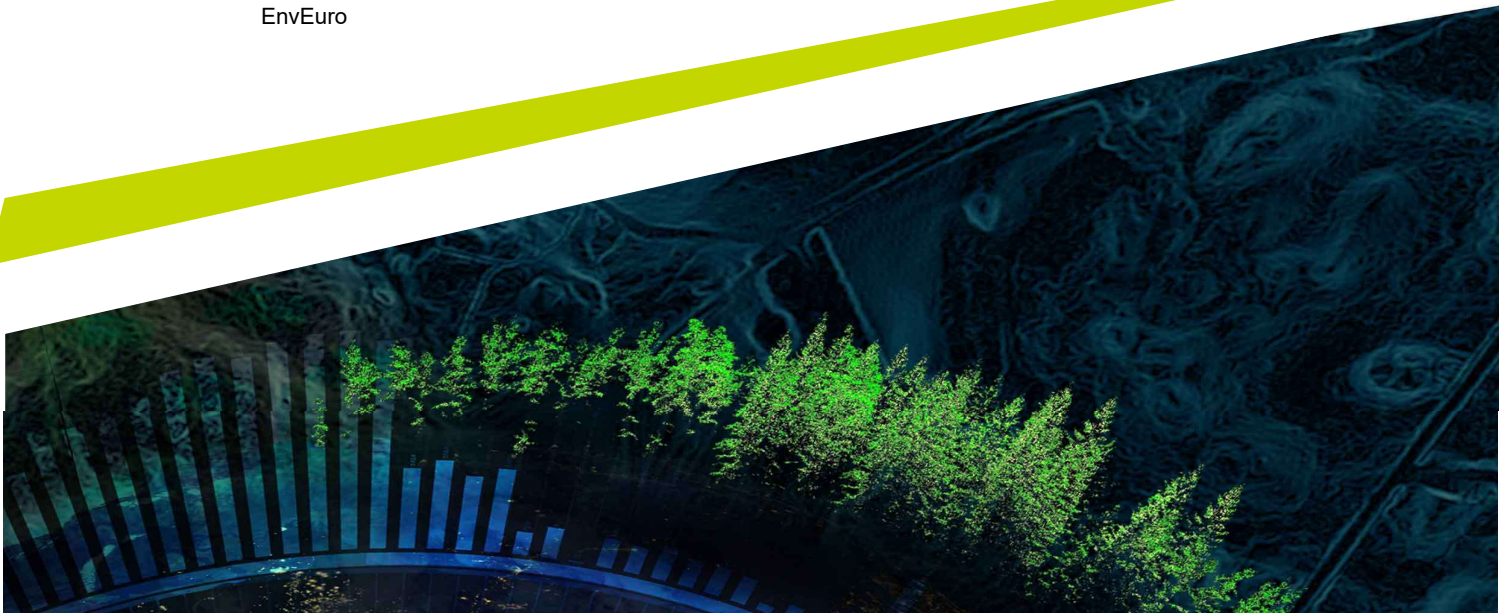


# Forest fertilization does not cause any long-term effects on tree growth or vegetation composition

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Independent project • 30 credits  
Swedish University of Agricultural Sciences, SLU  
Faculty of Natural Resources and Agricultural Sciences  
EnvEuro



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**Keywords:** density, ground vegetation, height, stem radial growth, legacy nitrogen effect, soil total N and C/N ratio, tree

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## Abstract

Forest fertilization is considered one of the most effective management options to improve forest productivity. In Fennoscandia, nitrogen (N) fertilization has been practiced for over 50 years and is normally added in the form of granules of  $\text{NH}_4\text{NO}_3$  at least 15 years prior to the final harvest. In addition to the intended effect of increasing tree productivity, N addition will induce changes in other parts of the forest ecosystem. Such effects include changes in the community composition of plants, insects, and soil fungi. Although many of these effects appear short-lived there are concerns that forest fertilization will induce long-term changes in soil nutrients and associated plant productivity. This study aims to investigate the long-term residual effects of commercial forest fertilization on tree growth and vegetation composition. The experiment was conducted in two regions of Sweden: the northeastern area of Uppsala and the Skinnskatteberg municipalities. It involves 36 forest stands, with half previously fertilized and the other half were unfertilized during the previous forest rotation. It is worth mentioning that these stands were prior to this study clearcut (5-14 years ago). The two forest types were used to evaluate effects of past fertilization. Most data were collected in August 2022, including tree heights, collection of trees cross-sections, vegetation records, and soil samples collection. However, the analysis of the soil data and stem radial growth measurements were conducted at the beginning of 2023. Although tree density, soil N and C/N ratio could affect tree stem radial growth and C/N ratio could influence tree height and density, past fertilization did not affect the radial and vertical growth of trees. Further, there were no differences between fertilized and unfertilized stands in terms of soil N, C/N ratio or on ground vegetation composition. These results suggest that the long-term effects of N fertilization are small or absent, with no major long-term effects on soil N content or vegetation distribution and productivity. The results highlight the resilience and sustainability of forests and provides useful insights for the rational use of fertilization and conservation of forest ecosystems. The forest manager should focus on planning and nutrient management to better balance forest development and promote more sustainable forestry practices.

*Keywords:* density, ground vegetation, height, stem radial growth, legacy nitrogen effect, soil total N and C/N ratio, tree

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# 1.Introduction

It is well known that forest plays an important role for ecosystem functioning and human well-being globally. With the development of society, people's dependence on forests has also increased, including but not limited to logging activities. As a result, global forest loss has steadily increased in recent decades (Wade et al. 2020). This trend not only threatens the existence of forests but also risks destroying the economic benefits derived from them. This is because the forest has both direct and indirect economic value, including the ability to provide raw materials for paper-making and job vacancies for local people (Berg 2021). Therefore, there are strong incentives to develop sustainable management systems that can ensure the long-term functioning of forests (da Gama e Silva 2022).

In Swedish forest management, the practice of reforestation following harvesting has been established as a legal requirement to promote sustainable forest development (*Swedish Forestry Act* 2022). As a result, the forests in Sweden are predominantly characterized by even-aged stands (Berglund 2021).

Forest fertilization is a cost-effective way to increase forest productivity (Saarsalmi & Mälkönen 2001). In Fennoscandia commercial fertilization is done by adding  $\text{NH}_4\text{NO}_3$ . It is usually accomplished by aerial dispersal of granulated fertilizers. However, the effects of forest fertilization are not limited to affecting production trees. Compared with the lack or imbalance of nutrients that may exist in a similar natural state, fertilizer application balances the nutrients the vegetation needs in the forest and improves the stand's ability to resist external damage from various pests (Saarsalmi & Mälkönen 2001). It also has long-term effects on the forest, both in terms of diversity and composition of the ground vegetation and growth of trees (Pettersson & Högbom 2004; Olsson & Kellner 2006; Strengbom & Nordin 2008, 2012; From & Nordin 2011). The impacts of nitrogen enrichment on ecosystems and their services are widespread, and goes far beyond influencing tree growth (Manning 2012). In Sweden, it is common to use nitrogen fertilizer in forest ecosystems since nitrogen is typically the most growth-limiting nutrient and nitrogen enrichment can result from nitrogen fertilization (Lundin & Nilsson 2021). Nitrogen is transferred among plants, microorganisms, soil organic matter and soil minerals (Zhu et al. 2015). Accordingly, it can directly or indirectly affect forest productivity and composition of the vegetation.

In recent years, the risk of negative side effects of forest fertilization has been controversial in the public domain. Many people are concerned about the environmental and ecological impacts that can result from fertilizer application, including water pollution, wildlife, plants, and so on (Sullivan & Sullivan 2018; Lundin & Nilsson 2021). Researchers confirmed that the residual effect of nitrogen fertilization is persistent for the entire forest, which is beyond one rotation period in forestry (Strengbom & Nordin 2008; From et al. 2015).

Nitrogen deposition refers to the entry of nitrogen elements into forest ecosystems through gas, dry deposition, and wet deposition. Nitrogen deposition accumulation is due to the global emission of nitrogen oxides. It mainly comes from fossil fuel combustion and agriculture (National Atmospheric Deposition Program 2016). Excess nitrogen can stress forest ecosystems, leading to nitrogen saturation and even losses of nitrogen to other ecosystems, thus leading to many negative conditions, such as soil acidification, eutrophication, etc. (Cao et al. 2019; Sung 2020; Verma & Sagar 2020). For Sweden, the rate of nitrogen deposition differs among regions. The deposition decreases from south to north. Thus, although the level of nitrogen deposition is highest in the south, the rate of nitrogen deposition has decreased in recent years generally (Binkley & Högberg 2016). In addition to nitrogen deposition, the use of nitrogen fertilizers is one of the main causes of nitrogen eutrophication.

According to the current research, anthropogenic nitrogen additions is one of the main drivers behind observed changes in the distribution of vegetation types in Swedish forests. Specifically, species that are favoured by shade and have high nutrient demands have expanded on behalf of nutrient-conservative species (Hedwall et al. 2019). Simultaneously, there are researchers who claimed that the proportion of shade tolerant plant species increases during the succession of boreal forest ecosystems (Jonsson et al. 2021). Moreover, according to Bobbink et al. (2010), it seems like nitrogen enrichment of the boreal forest have not resulted in changes in species richness of understory vegetation. However, it does have an impact on species composition. When nitrogen is added, the response of bryophytes is species-specific. Specifically, dominant species like *Hylocomium splendens* show a decline when the nitrogen addition rate exceeds  $10 \text{ kg N} \cdot \text{ha}^{-1} \cdot \text{yr}^{-1}$ . On the other hand, species that live in more nutrient-rich habitats, such as *Brachythecium* spp, tend to increase. As for vascular plant species, the introduction of N leads to an increase in fast-growing graminoids and herbs, while slow-growing species such as ericaceous dwarf shrubs experience a decline (Bobbink et al. 2010).

Based on the above, the residual impacts of nitrogen fertilization on forests deserve to be studied. Thus, in this paper, the impacts will be further explored and provide references and recommendations for the future sustainability of forests.

In order to evaluate whether the long-term residual effect of past nitrogen enrichment interferes with the distribution and abundance of vegetation in the forest,



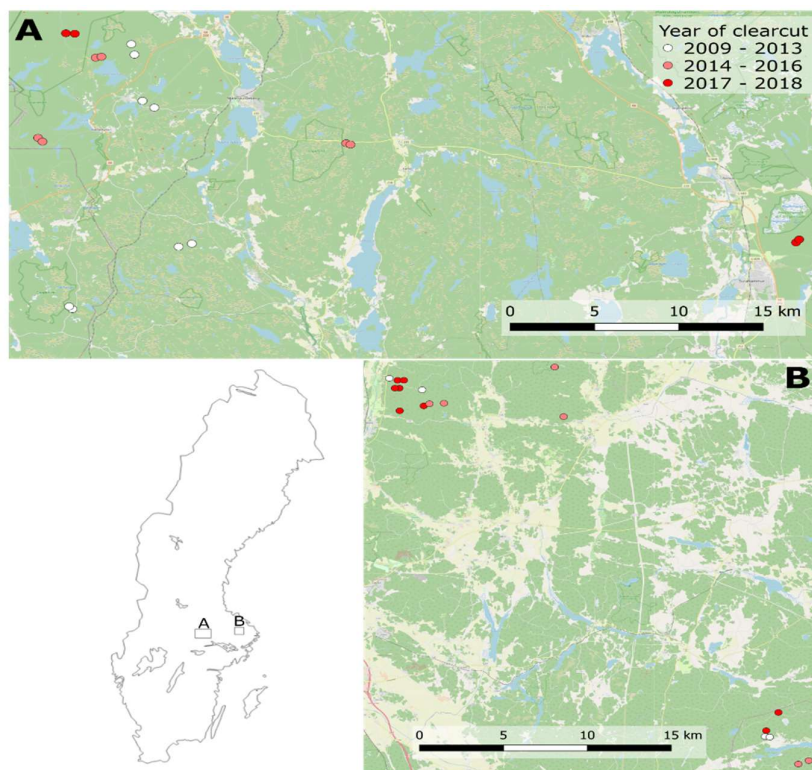
and to study whether it can generate legacy effects on tree growth that extends across forest generations, I compared forest stands, which have been commercially fertilized and unfertilized stands in the previous forest rotation. To evaluate the long-term effects, I compared stands that were fertilized at least 15 years before they were clear-felled. If I cannot find any significant differences between fertilized and unfertilized stands, then it indicates that the effect of forest fertilization is limited to one generation of forests. Conversely, if there are differences, this suggests that effects have carried over to the next generation of forests. In detail, the following hypotheses will be tested:

1. In the forest, past fertilization increases the soil N content, and decreases C/N ratio.
2. Past fertilization induces changes in the abundance of the ground vegetation. The abundance of slow-growing nitrogen-conservative plants, such as *ericaceous dwarf shrubs*, is lower, but as nitrogen enrichment favors fast-growing plants such as *graminoids* and *herbs*, their abundance is higher.
3. Legacy effects of fertilization in the previous forest rotation are indicated by higher growth of the trees (greater tree height and greater stem radial growth).

## 2. Materials and methods

### 2.1 Description of forest stands

The research was conducted in two regions in Sweden: the northeastern area of Uppsala (between 59.79-60.04° N, 17.75-18.19° E) and the Skinnskatteberg region (between 59.68-59.87° N, 15.47-16.28° E), as Fig.1.



*Fig. 1: Map of the study area in Sweden (A: Skinnskatteberg, B: The northeastern area of Uppsala). Background map: OpenStreetMap. Sites for data collection are indicated by the dots in panel A and B. The white dots represent the sites where the clearcut was completed from 2009 to 2013, the orange dots represent the sites where the clearcut was completed from 2014 to 2016, and the red dots represent the sites where the clearcut was completed from 2017 to 2018.*

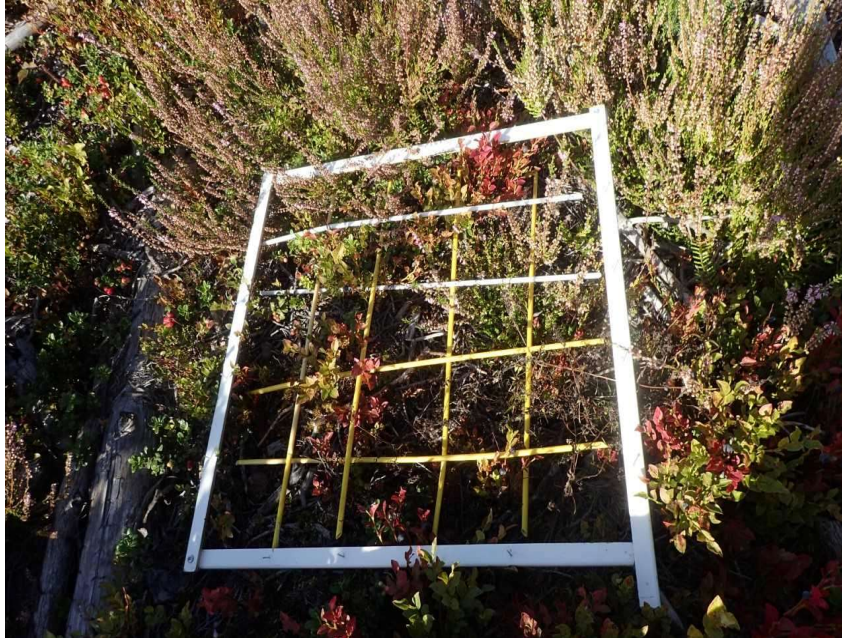
To investigate the long-term residual effects of commercial forest fertilization, I used 18 forest stands that had not been fertilized in the previous forest generation, and 18 stands that had been previously fertilized on one occasion with an N dose corresponding to  $150 \text{ kg N ha}^{-1}$  ( $=15 \text{ g N/m}^2$ ) to improve forest productivity. Nitrogen was added as granulated ammonium nitrate ( $\text{NH}_4\text{NO}_3$ ) at least 15 years prior to the clearcutting. All the forest stands were mixed coniferous forests with some deciduous trees, mainly birch. The tree layer was dominated by either Scots pine (*Pinus sylvestris*) or Norway spruce (*Picea abies*). Then, during the period of 2009-2018, the forests were clearcut and replanted with Norway spruce (*Picea abies*) and Scots pine (*Pinus sylvestris*), the two birch species (*Betula pendula*, *Betula pubescens*) spontaneously appeared on the clearcuts. The vegetation consisted of multiple layers, with mosses and lichens covering the soil, vascular plants growing above them, and the tree layer as the highest layer (refer to Fig.2 for a visual timeline).



Fig.2. Timeline of forest management

## 2.2 Data collection and preparation

In August 2022, most of data were collected within three circular plots with a 10 m radius (main plots) at each forest site. Vegetation was separated into two categories, tree ( $>1.2\text{m}$ ) and ground vegetation ( $<1.2\text{m}$ ). The vegetation composition below 1.2 m was assessed in 6 plots sized 0.5 by 0.5 m within each main plot. To assess vegetation composition, the presence or absence of species within 25 subplots of each of the 0.5 by 0.5 plots (Fig.3 and Fig.4), resulting in a frequency ranging from 0 to 25. All vascular plant species were recorded individually and collectively, while lichens and mosses were recorded only as species groups.



*Fig.3. Photography of vegetation survey subplot grid*

25 soil cores (diameter=2.5 cm) were taken on grounds not directly influenced by soil scarification in each main plot evenly (Fig.4). The depth of soil sample collection was independent across plots, only the organic layer was collected and combined to form one composite soil sample per plot. The samples were stored on ice and then transferred to a freezer. Subsequently, subsamples were obtained by freeze-drying and passing through a 1 mm sieve, their carbon and nitrogen concentrations were determined by an elemental analyser (TruMac CN; LECO). Thus, the C/N ratio could be calculated.

Frozen soil samples were used for pH analyses in the experiment. 25 mL of deionized water was added to each sample, and they were shaken on a shaker at 650 rpm for 30 minutes at room temperature to ensure complete suspension. Later, the mixture was equilibrated with atmospheric CO<sub>2</sub> for 15 minutes before measuring the pH with a pH meter.

The height measurements of the trees (>60 cm tall) were done in smaller subplots to ensure the feasibility of measurement, specifically within circular plots of a radius 3m (with the same centers as the 10 m radius plot) at each stand (Fig.4). Moreover, for the oldest clearcuts (logged between 2009 and 2015), three trees per species in each plot were cut to assess stem radial growth. The decision to sample trees only in the oldest clearings and not include additional data was not intended to improve accuracy but was rather since the trees in the younger clearings had not yet matured sufficiently for tree-ring analysis. The specific years of the trees first felling were also recorded.

After cutting the cross-sections of the trees were brought to the lab for further processing. Afterward, they were carefully dried at room temperature to avoid

being cracked and remove excess moisture to prevent mold formation. They were also sanded with the fine sandpaper to create a smooth flat surface to improve the visibility of the tree rings. Finally, the cross-sections were scanned with a scanner and the growth rates were measured manually from the scanned images by *ImageJ* software.

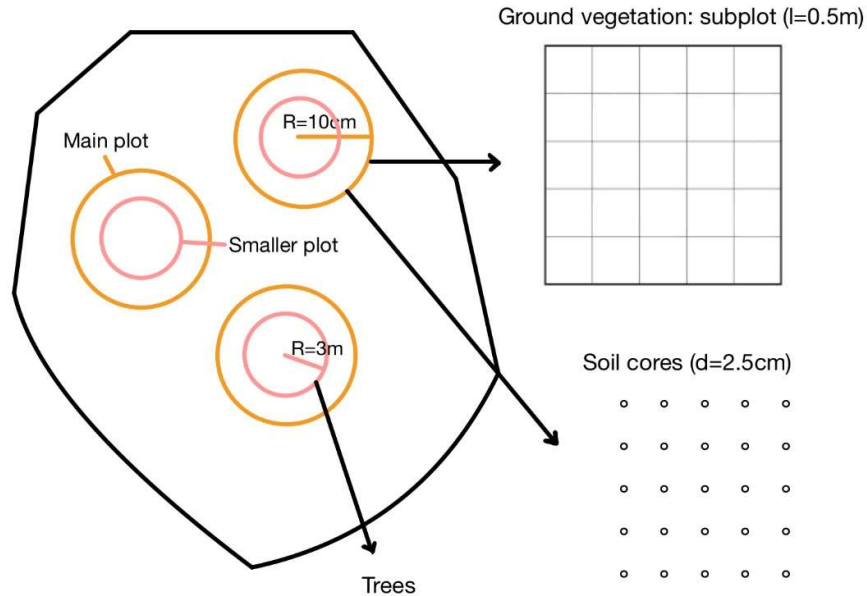


Fig. 4: Visual experimental setup in one site. The orange circle represents the main plot ( $r=10\text{m}$ ), where recorded ground vegetation and sampled soil. The pink circle represents the smaller plot ( $r=3\text{m}$ ), where the height of the trees were measured and cross sections were taken.

## 2.3 Statistical analysis

In this study, the analyses were conducted at the main plot level, with each stand consisting of three main plots. The data from the subplots within each main plot were aggregated and treated as representative of the respective main plot. The study had three replicates in each site. The 36 stands were divided into two groups, with half of the stands subjected to fertilization and the other half serving as the control without fertilization. All analyses were performed using the *R*.

To examine the differences in tree growth (stem radial growth and height) among different tree species, *one-way analysis of variance* (ANOVA) was conducted, followed by *Tukey's honestly significant difference* (HSD) test for pairwise comparisons between each pair of tree species.

The diversity of ground vegetation species was quantified using the Shannon index (H) at the main plot level. The Shannon index is widely used to decide species abundance and richness in ecological studies. The Shannon diversity index can be calculated as:  $H = -\sum p_i * \ln(p_i)$ , where  $p_i$  is the proportion of the entire community made up of species (Es'haghi Rad et al. 2009). Meanwhile, *Principal Component Analysis* (PCA) was used to describe ground vegetation composition. Two-sample t-tests were used to compare soil conditions (soil pH, N, C/N ratio), ground vegetation characteristics (species number and Shannon index), tree height and density between the fertilized and unfertilized forests.

In the analysis, the average height of trees in each plot was used, which was calculated by summing the heights of all trees in the plot and then divided them by the number of trees. Additionally, the density of trees in each plot was calculated as the total number of trees.

Besides, linear models were developed to investigate the impact of clearcut year on pH levels, the association between pH levels and tree growth, the relationship between tree density and height, and the effect of soil N, and C/N ratio on both tree growth and density. Additionally, the influence of clearcut year on soil N, and C/N ratio was also evaluated.

### 3. Results

In the study area, the tree layer, defined as vegetation above 1.2m, consisted of *Betula pendula*, *Betula pubescens*, *Picea abies* and *Pinus sylvestris*. The understory vegetation, defined as vegetation below 1.2m, consisted of vascular plants, mosses and lichens, with the vascular plants dominated by grasses and ericoid plants. Because *Betula pendula* and *Betula pubescens* both belong to birch, they were grouped together and referred to as *Betula* sp.

The soil collected from the area was acidic, with pH levels ranging from 3.5 to 5.0. The average N content was 1.05 kg N/m<sup>2</sup> (=1050 g N/m<sup>2</sup>) and the average C/N ratio was 29.96. Based on the results, there was no considerable difference in pH between fertilized and unfertilized ( $p_1=0.72$ ,  $df_1=105.8$ ). And clear-cutting year was analyzed that it was not highly associated with soil pH ( $p_1=0.2$ ).

To test the hypothesis that past fertilization increases soil quality, a comparison was made between the soil N and C/N ratio of clearcuts in fertilized and unfertilized forests. However, the levels of N and C/N ratio in the soil were not substantially different in the fertilized and unfertilized forests ( $p_1=0.74$ ,  $df_1=105.91$ ;  $p_2=0.97$ ,  $df_2=104.36$ ). Moreover, neither soil N concentration nor C/N ratio was correlated to time since clear-cutting (Table 1).

The ground vegetation was heavily dominated by common vascular plants. *Deschampsia flexuosa* (77.5%), *Vaccinium vitis-idaea* (64.7%), *Vaccinium myrtillus* (60.8%), *Calluna vulgaris* (40.6%) and *Pinus sylvestris* (19.6%) were the most abundant species. Moss cover corresponded to 74.3%, while the share of lichens was 11.0%. My results showed that there was no significant difference in species number between fertilized and unfertilized stands (Fig.5;  $p=0.2$ ,  $df=106$ ). Similarly, there was no difference in Shannon diversity index between fertilized and unfertilized stands (Fig.6;  $p=0.56$ ,  $df=106$ ). In other words, there were no signs of legacy effects from past fertilization on ground vegetation species richness or Shannon diversity index.

Potential differences in plant community composition were assessed by PCA. The study also investigated the effects of time since clear-cutting and fertilization on the community composition of ground vegetation. The results showed that there was no difference in plant composition that could be linked to past fertilization or time since clear-cutting (Fig.7 and 8).

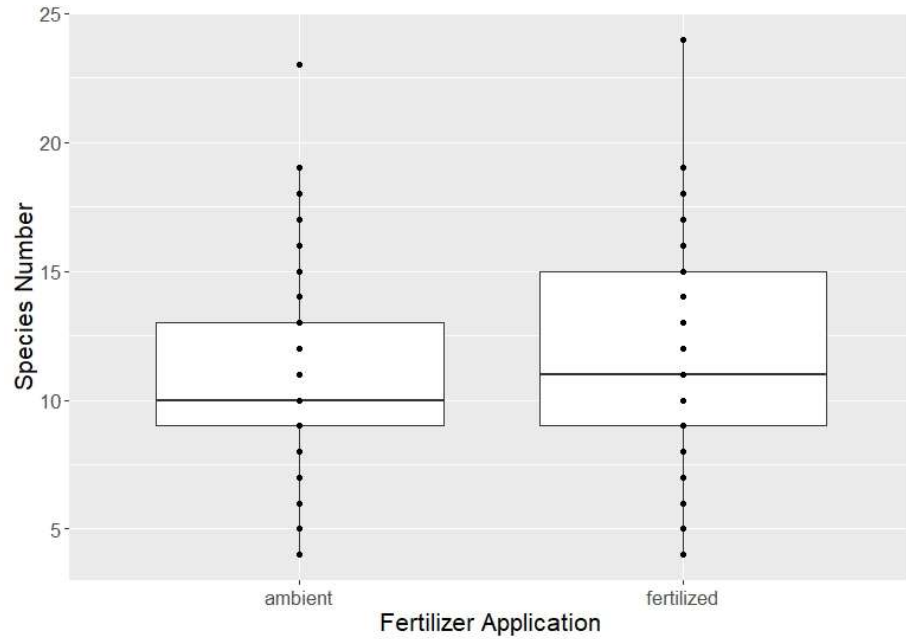


Fig.5. Box plot showing species numbers (richness) of ground vegetation in fertilized and unfertilized plots. The line inside each box corresponds to the median value.

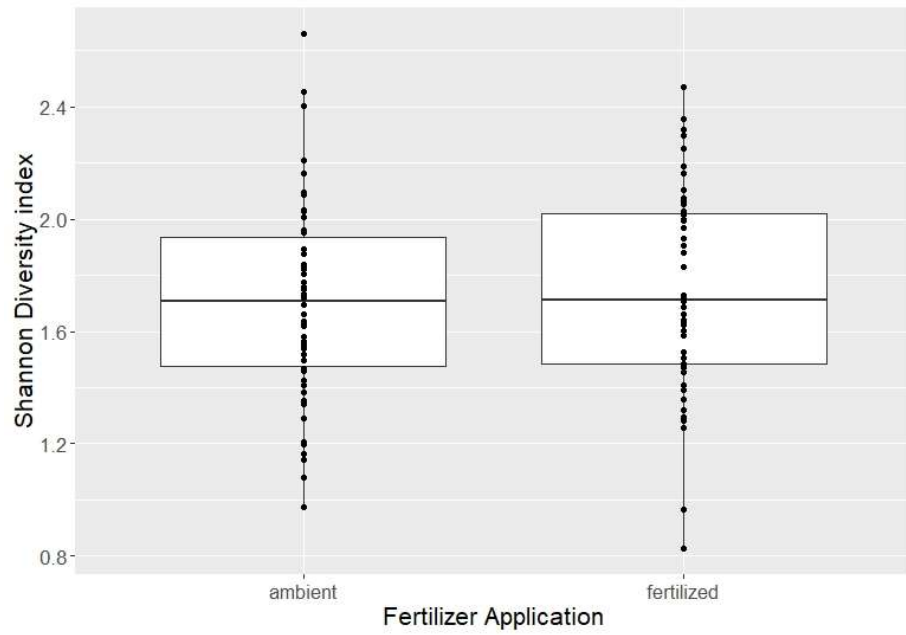


Fig.6. Box plot showing species diversity of ground vegetation in fertilized and unfertilized plots. The line inside each box corresponds to the median value.



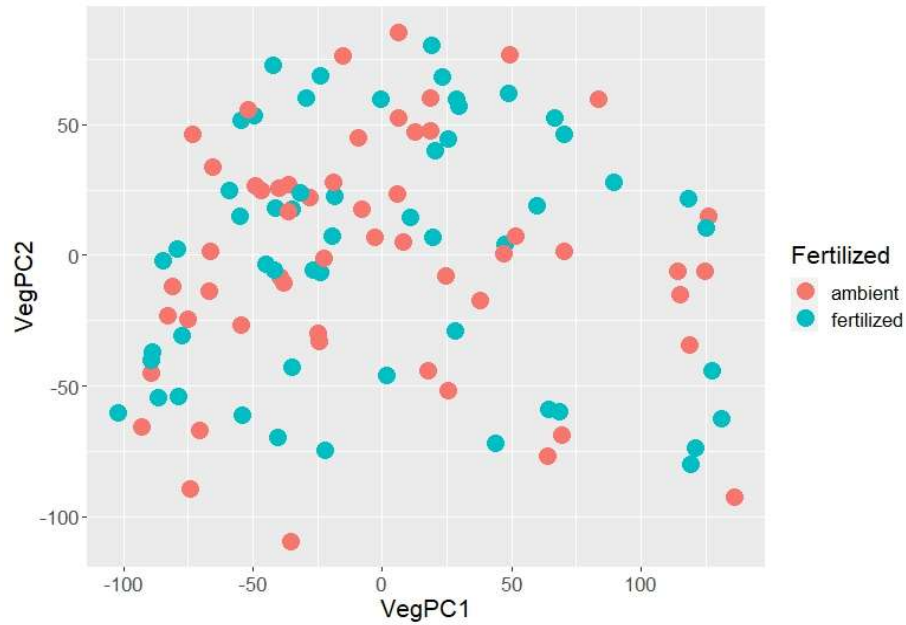


Fig.7. PCA of ground vegetation species composition in response to fertilizer application. The dots represent the community composition of ground vegetation in subplots of unfertilized (red) and fertilized (green) forest stands.

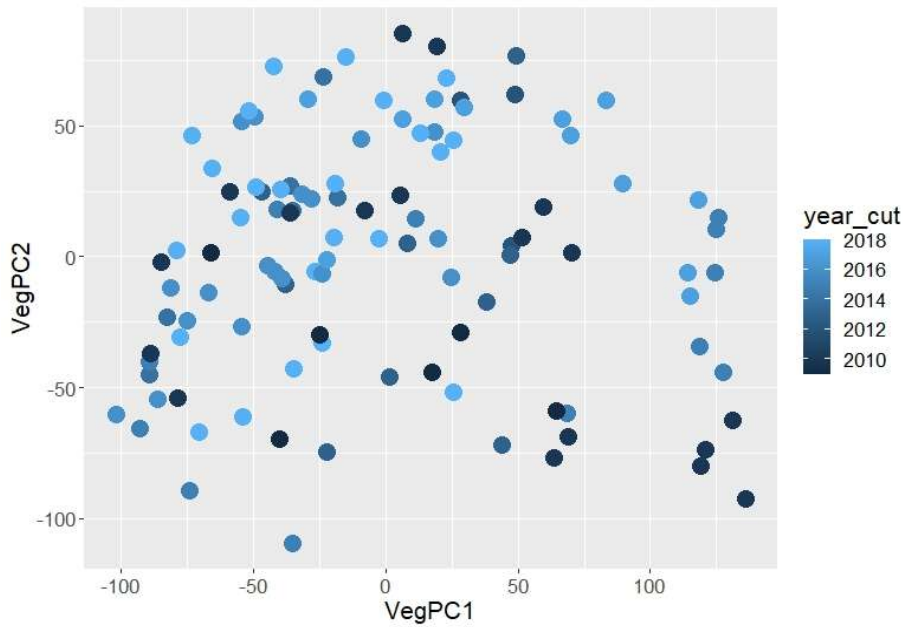


Fig.8. PCA of ground vegetation species composition in response to clear-cutting year. The dots represent the community composition of ground vegetation in subplots of forest stands clearcut at different years. The color gradient represents the year of clearcutting, from dark blue to light blue from 2009 to 2018.

To study stem radial growth, 150 *Pinus sylvestris*, 128 *Picea abies* and 128 *Betula* sp. were sampled. Additionally, the height of 407 trees was measured among them. There were no significant differences in stem radial growth or height between unfertilized and fertilized stands for *Pinus sylvestris* and *Picea abies*, as well as in stem radial growth for *Betula* sp. However, a marginal significance was observed in the height difference between the unfertilized and fertilized stands for *Betula* sp. (Fig.9 and 10, Table 2). Likewise, when analyzed for mixed species, there was no apparent height variation between fertilization and unfertilized ( $p=0.74$ ,  $df=102.54$ ).

To test this, ANOVA was used to analyze the tree species. The results revealed that growth differed among the three species (stem radial growth:  $F=5.58$ ,  $Pr=0.01$ ; height:  $F=18.67$ ,  $Pr=1.76e-08$ ). To further test which tree species were actually different from each other, *Tukey's HSD* tests were performed (Table 3). There were no significant differences in the height between *Pinus sylvestris* and *Picea abies*, and in the stem radial growth between *Pinus sylvestris* and *Betula* sp. But *Betula* sp. grows significantly taller and faster than *Picea abies*. Additionally, *Betula* sp. grows taller than *Pinus sylvestris*, while *Pinus sylvestris* grows faster than *Picea abies*. In brief, the stem radial growth and height of *Picea abies* and *Betula* sp., the height of *Pinus sylvestris* and *Betula* sp., as well as the stem radial growth of *Pinus sylvestris* and *Picea abies* were markedly different.

Stem radial growth and height of trees were not correlated with soil pH ( $p_1=0.68$ ,  $p_2=0.59$ ). Regarding soil nutrients, Table 4 provided information indicating weak negative effects of total N and C/N ratio on stem radial growth. Besides, in terms of height, only the C/N ratio showed a weak negative correlation. The low R-squared values suggested that total N content in the soil was a poor predictor of the trees' stem radial growth. When the total N content in the soil increases, there might be a tendency for a decrease in the trees' stem radial growth. Similarly, an increase in the C/N ratio could be associated with a decrease in both stem radial growth and height of the trees.

Besides, tree density did not significantly differ after fertilization ( $p=0.38$ ,  $df=104.29$ ), but it had a strong positive correlation with average tree height ( $p=6.64e-09$ ,  $k=0.26$ ,  $R\text{-squared}=0.28$ ). Furthermore, there was a negative correlation between soil C/N ratio and tree density (Table 5), with low R-squared value.

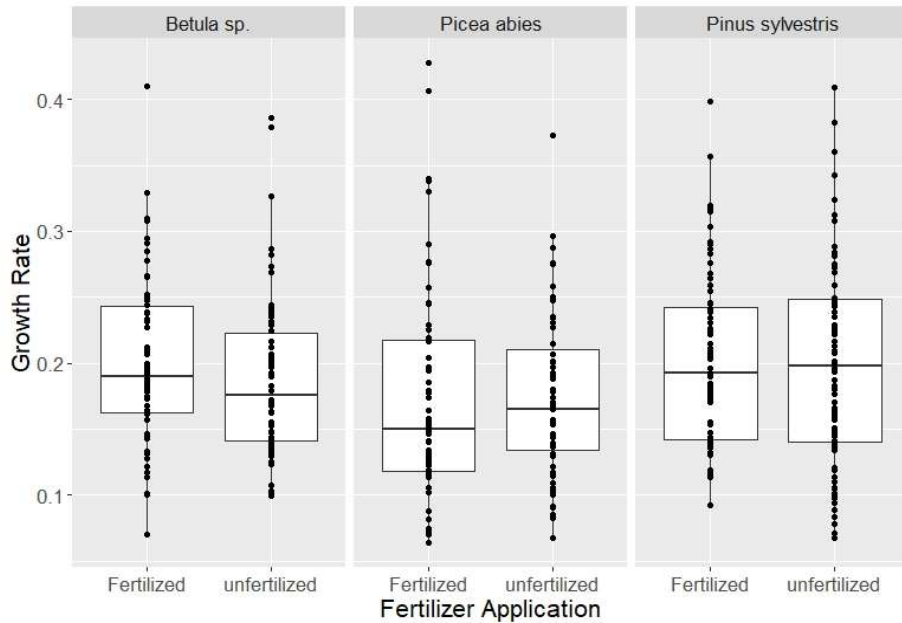


Fig.9. Box plot showing growth rates (stem radial growth) (cm) of different tree species under fertilized and unfertilized conditions. The line inside each box corresponds to the median value.

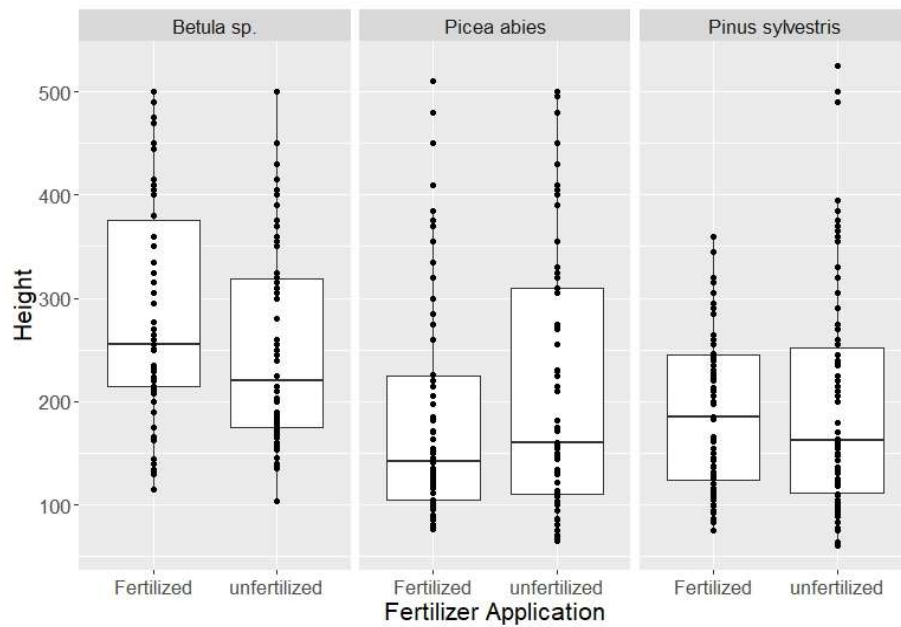


Fig.10. Box plot showing heights (cm) of different tree species under fertilized and unfertilized conditions. The line inside each box corresponds to the median value.

Table 1: Results of linear model between clear-cutting year and total N, C/N ratio.

Soil parameter	Slope	R-squared	F-statistics	p-value
Total N	2.47	0.02	3.32	0.07
C/N ratio	1.4e-01	0.01	2.58	0.11

Table 2: Results of t-tests for stem radial growth and height of different tree species under fertilized and non-fertilized conditions

Types of trees	Tree growth	t	df	p-value
<i>Betula</i> sp.	stem radial growth	0.89	103.53	0.38
	height	1.96	125.79	0.05
<i>Pinus sylvestris</i>	stem radial growth	0.24	147.21	0.81
	height	-0.54	139.96	0.59
<i>Picea abies</i>	stem radial growth	-0.29	120.9	0.77
	height	-1.19	124.79	0.24

Table 3: Results of multiple comparisons of stem radial growth and heights among three tree species (Tukey's HSD test).

Species group	Tree growth	Mean difference	95% confidence interval		p
			Lower Bound	Upper Bound	
<i>Picea abies</i> ~ <i>Betula</i> sp.	stem radial growth	-0.02	-0.04	-0.001	0.04
	height	-66.08	-97.07	-35.09	0.0000024
<i>Pinus sylvestris</i> ~ <i>Betula</i> sp.	stem radial growth	0.01	-0.01	0.02	0.81
	height	-71.04	-100.92	-41.15	0.0000001
<i>Pinus sylvestris</i> ~ <i>Picea abies</i>	stem radial growth	0.03	0.01	0.05	0.004
	height	-4.96	-34.78	24.87	0.92

Table 4: Results of linear model between soil N, C/N ratio and the mixed tree growth.

Soil parameter	Tree growth	slope	R-squared	F-statistics	p-value
Total N	stem radial growth	-0.04	0.01	6.39	0.01
	height	-0.001	0.01	2.43	0.12
C/N ratio	stem radial growth	-0.004	0.04	19.43	1.34e-05
	height	-0.02	0.09	11	0.001

Table 5: Results of linear model between soil N, C/N ratio and density.

Soil parameter	Slope	R-squared	F-statistics	p-value
Total N	-0.001	0.003	0.72	0.4
C/N ratio	-0.03	0.06	7.56	0.01

## 4. Discussion

Above all, it can be seen from the results that past nitrogen fertilization did not improve soil nutrients status in the form of %N and C/N ratio in the following forestry cycle, nor did it affect ground vegetation, which are contrary to the original hypothesis. Compared to previous findings, the legacy effects of past fertilization are generally weak or completely lacking from the study sites included here and this is an interesting and important result.

The study conducted by From et al. (2015) was done in the middle boreal zone in central Sweden, hence latitude is higher than my study. The primary focus of their study was to compare tree growth, needle nitrogen concentration, and the availability of mobile nitrogen in a 10-year-old forest stand. Fertilized and control groups were selected from sites that had received fertilization once or twice during the previous forest rotation, as well as unfertilized sites. According to the findings of their study, previous fertilized two-times resulted in an increase in tree height. However, those fertilized once were not found to be significantly different from those that were not fertilized. This is consistent with the results of my study. They also observed that fertilization during the previous forest rotation had long-term residual effects on the nitrogen dynamics of the forest stand, surpassing the influence of a single rotation. However, the result differs from the findings of my study, which indicated that fertilization did not have an impact on tree growth. The research by Strengbom & Nordin (2012) was also conducted in central Sweden. The research compared the effects of clear-cutting with nitrogen fertilization, clear-cutting without fertilization, mature forest with nitrogen fertilization, and mature forest without fertilization. The results revealed that clear-cutting combined with nitrogen had a significant impact on plant community composition, particularly in forests initially exposed to nitrogen addition and subsequent clear-cutting disturbance. The effects varied among plant functional groups. In disturbed forests with nitrogen fertilization, there was an increase in the abundance of graminoids and a decrease in the abundance of understory shrubs. Strengbom & Nordin (2008) did the research in central Sweden. They compared the effects of two times fertilized during the previous rotation with unfertilized forests. The results showed that the fertilized forests exhibited denser ground vegetation, lower species evenness and reduced biodiversity. The abundance of understory shrubs decreased, whereas grasses and certain nitrophilous herbaceous plants increased in abundance. Their study also indicated a preference for moss species that thrive on woody debris and leaf litter, while *Hylocomium splendens* and ground-living lichens were less favored. These findings contrast with my research results, as fertilization in the previous rotation did not affect the abundance and composition of ground vegetation after clearcutting and replanting.

Given these diverse findings, it is evident that the effects of past fertilization on tree growth and ground vegetation were influenced by various factors, including specific site characteristics (coordinates of latitude and longitude), as well as the fertilization pattern. My research results do not support significant impacts of fertilization on tree growth and ground vegetation as the other researchers, but it emphasizes the need for further studies to explore the complex relationships within forest ecosystems. I will discuss the factors below that could contribute to these differences and propose directions for improvement.

## 4.1 Selection of experimental area

The forest area in my study belongs to the hemi-boreal forest in Sweden, while earlier studies examining potential long-term effects of forest fertilization have been conducted in the middle-boreal zone, which is located between southern boreal to northern boreal zones (Jonsson et al. 2021, fig.1), like Strengbom & Nordin's study in 2008. In addition to differences in basic latitude and longitude, the characteristics of forest vegetation in the region are also different. The area I studied belongs to the transition zone between boreal and temperate zones. The vegetation types in the transition zone are richer in species, with higher diversity and potentially higher biomass, both in terms of ground vegetation and trees, compared to other areas. At the same time, for the climatic conditions, the general temperature in the middle-boreal area is relatively low, so coniferous forests may be more predominant in this region. In the hemi-boreal region, the climate is warmer, so there may be a high proportion of deciduous forests. In short, because of different locations, the soil properties are different too. Then the effect produced by the fertilizer application is also different. Furthermore, since the short-term effects of nitrogen addition on forests vary by site (Jørgensen et al. 2021), the long-term effects may also vary by site selection.

## 4.2 Clearcutting interference

During the forest rotation cycle studied, forest clearcutting was carried out, resulting in significant changes in the ecosystem. Research has shown that clearcutting could have negative impacts on both above- and below-ground biota in the forest. For example, the fungi that maintain soil health may be disrupted, which can further lead to the breakdown of nutrient cycling in the soil and ultimately affect the growth of vegetation (Hartmann et al. 2012; Hasby 2022). Additionally, studies have confirmed that soil structure could be damaged as a result of clearcutting, which can disturb the soil organic matter and nutrient cycles to varying degrees

(Siebers & Kruse 2019). Therefore, it can be argued that clearcutting may offset the nutrients required for plant growth, which could explain why there was no significant difference in growth before and after past fertilization.

### 4.3 Short duration of residual effects of fertilization after felling

A model was established in the experimental results to examine the relationship between clear-cutting time and total N and C/N ratio in the soil. According to the model results, it was found that in areas where clear-cutting occurred earlier, the soil had lower N content and C/N ratio. Although the p-value of the model was higher than 0.05, indicating it did not reach statistical significance, it is still worth considering the possibility of this relationship. During the data collection process, it was noted that the clear-cutting time was recorded from 2009 to 2018, while the soil data was collected in 2022. Due to the relatively long-time interval, it is possible that the residual effects of fertilization had already disappeared, which may have caused the effect missing. In other words, the residual effects of fertilization after clear-cutting did not last for a long time. This situation may explain why the model results show lower N content and C/N ratio in areas with earlier clear-cutting records.

### 4.4 Number and dose of fertilizer applications in the previous rotation

In the study by From et al. (2015), they compared stands that had been fertilized twice, once, and unfertilized in the previous rotation cycle. In terms of tree height. The results showed no significant difference in average tree height between stands fertilized once in the previous cycle and the control group. This may indicate that a single fertilization application (corresponding to  $150 \text{ kg N ha}^{-1}$ ) had relatively little effect on tree growth. However, it was noticed that in the trees that were fertilized twice, they were significantly taller than the other groups. This suggests that multiple fertilization may have a positive effect on tree growth in the forest generation that precedes the one fertilized. This may be due to the fact that multiple fertilization provided more nutrients, met the growth needs of the trees, and promoted the increase in their growth rate and height.

Besides, the total N content tested in the soil in this study was  $1050 \text{ g N/m}^2$ , but the fertilization was only  $15 \text{ g N/m}^2$ , so it might be difficult to detect the residual effect of fertilization on the total soil N in the experiment. Therefore, it is difficult

to judge whether fertilization had an effect on the total N content in the soil. In follow-up experiments, it may be considered to increase the amount of fertilization.

In conclusion, the frequency or dose of fertilizer application seems to play an important role in influencing tree growth. Future research should further explore the relationship between fertilizer application times (and dose-response) and tree growth, make further adjustments to analyze the long-term residual effects of nitrogen.

Although the initial hypotheses were rejected, other factors were discovered in the experiment that better explain differences in vegetation growth. I found that soil N and C/N ratio might influence the tree radial growth and the tree height and density might be affected by soil C/N ratio. Besides, tree density could influence the trees' average height. Compared with other influencing factors, the impact of tree density was the most significant. It meant that the higher the density of trees, the higher the average height of trees. From my point of view, when tree density increases, the competitions among the trees became fiercer. High density stimulates vertical growth of trees (Tymińska-Czabańska et al. 2022). The reason for this is because to grow better, the canopy of the tree would become smaller, and the trunk would become longer to compete for sunlight and water. So, in the case of high tree density, the average height of trees in forests will become higher.

Moreover, for the effect of tree species on tree stem radial growth and height, ANOVA analysis showed there was a relationship between tree species and growth, but *HSD* again found that the relationship was insignificant for some individual groups, particularly in the comparison of height between *Pinus sylvestris* and *Picea abies*, and in the comparison of stem radial growth between *Pinus sylvestris* and *Betula* sp. The results were contradictory, so it cannot be determined that tree species is a factor affecting growth, and further experiments are needed in the future to confirm this relationship.

In summary, in future research, the experimental area and site conditions can be changed to be explored. Experiments could include fertilizing more than one time or increase the dose in previous forest rotation to find much clearer result. At the same time, the length of time interval between clearcutting and soil collection needed to be controlled carefully. In addition, a more in-depth analysis of the tree species involved in this study could be conducted to understand the effects of different tree species on the study results. These initiatives help to further refine the findings of this study.



## 5. Conclusion

This paper studied the long-term effects of nitrogen enrichment from past fertilization on forest vegetation growth. The residual effects of fertilization on tree growth and ground vegetation composition were not found, so the effect in this study was limited to one generation of forest.

The results did not support the hypotheses that legacy nitrogen effect changed soil N and C/N, ground vegetation distribution and growth of trees. However, it was found that C/N ratio and density were weakly negatively correlated. In addition, the growth rate of forest trees can be explained by tree density, N and C/N ratio, whereas the height of trees can be explained by C/N ratio. The results of the study help to verify the effect of past fertilization on forest vegetation and provide a reference for potential future forest management to reorient management direction. Past fertilization for these areas cannot make large effect in one forest rotation cycle. Future forest management strategies should focus on other factors, such as soil quality and so on to promote sustainable forest growth and management.

Nevertheless, there are limitations in this study, future studies needed to be expanded to determine if the effects of latitude and longitude are involved, since residual nitrogen not present in the study area might have a stronger impact on forests. The time interval between logging and the next forest cycle soil data collection needs attention to prevent missed impacts. Last but not least, the control group should be considered to increase in the follow-up study. These control groups consist of increasing the frequency and dosage of fertilization in the previous forest rotation, in order to obtain more complete conclusions on the residual long-term effects of fertilizer application. In addition, whether tree species is a factor affecting growth is still unknown.

In short, for the legacy effect of fertilization still needed to be explored in different areas and consider more factors. From the perspective of environmental pollution and fertilization legacy, the experimental results show that fertilization had no long-term impact on forest vegetation. This means that forest ecosystems are able to return to their natural state after long-term fertilization without negative lasting effects. This finding highlights the resilience and sustainability of forests and provides useful insights for rational use of fertilization and conservation of forest ecosystems. It is important to plan rationally, maximizing forest growth benefits.

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## Popular science summary

Forests play a crucial role in the world, offering benefits such as climate regulation, wind protection, and a source of raw materials. In a world of population explosion, forests are becoming increasingly important. One effective way to enhance their productivity is through forest fertilization, commonly using nitrogen in Sweden. At the same time, nitrogen is also increasing. The subject of this study is to explore the residual effects of nitrogen fertilization in forests on tree growth and ground vegetation composition. The study involved two forest rotation cycles and was carried out in the hemi-boreal forest in the northeastern area of Uppsala and the Skinnskatteberg region in Sweden.

By comparing unfertilized stands with those fertilized during the first rotation, analyze whether residual effects of fertilization could disturb the forest ecosystem. The results revealed that the residual effects of nitrogen fertilization had no significant impact on tree growth and ground vegetation distribution. However, forest density, total nitrogen content in the soil, and carbon-to-nitrogen ratio influenced stem radial tree growth. Carbon-to-nitrogen ratio influenced tree height and density. Moreover, the influence of tree species on growth differences remains unclear and requires further investigation.

Although this study suggests that the residual effects of nitrogen fertilization do not significantly affect forest growth, it highlights the importance of considering other factors for promoting sustainable forest growth. This provides valuable insights into forest sustainability for the general public.

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