Norwegian University
of Life Sciences

Master's Thesis 2023   30 ECTS
Faculty of Environmental Sciences and Natural Resource Management

Impact of Forest Management History on Fine Root Dynamics and Mycorrhizal Colonization in Norwegian Boreal Forests

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Acknowledgements

Abstract

This thesis examines the influence of forest management history on fine root traits in Norwegian boreal spruce forests, focusing on the role of fine roots in carbon sequestration. The study was conducted across various forest sites in southeastern Norway, incorporating contrasting forest management histories – near-natural forests (NN) with minimal human influence and mature forests with a history of clear-cutting (CC). The research aimed to assess the impact of forest management practices on fine root specific root length (SRL), biomass distribution, and mycorrhizal associations.

The methodology included soil sampling and fine root analysis using scanning and statistical techniques. Despite initial hypotheses suggesting significant differences in fine root traits between NN and CC forests, the findings revealed minimal variations attributable to past forest management practices. Neither microclimatic nor macroclimatic factors, including forest structure, significantly influenced these traits.

Contrary to expectations, SRL was found to be similar across both forest types, challenging the hypothesis that management practices in CC forests might significantly influence SRL of fine roots. Although variations in fine root biomass distribution across soil layers were noted, these were not directly associated with forest types. Furthermore, the anticipated increase in root tip abundance and mycorrhizal associations in NN forests, attributed to greater tree species diversity and more complex ecosystem structures, was not observed.

The study contributes to the understanding of below-ground ecological dynamics in boreal forests and underscores the need for further research to unravel the complex interactions affecting root traits. It highlights the importance of considering fine roots in forest carbon cycling and ecosystem health assessments. The findings also suggest that mature forests, regardless of their management history, possess an inherent resilience in maintaining fine root characteristics vital for carbon sequestration.

Keywords: Boreal ecosystems, boreal forest, forest management, root ecology, fine roots, carbon cycling, carbon sequestration, soil carbon storage, forest soil, specific root length, root tips, mycorrhiza

Contents

Introduction

The boreal forests of Fennoscandia have undergone significant ecological and management changes over centuries. The invasion of Norway spruce (Picea abies; hereafter referred to as spruce) in the late Holocene marked a pivotal shift in this landscape. Today, spruce is the predominant tree species in mesic to moist soil conditions, constituting about 39.1% of Norway's productive forest area (Statistisk sentralbyrå, 2023a; Øyen & Nygaard, 2020). Its dominance plays a vital role in supporting a diverse array of flora, fauna, and fungi, thereby anchoring the region's rich biodiversity.

Understanding the implications of human intervention in this ecosystem is of paramount importance, especially considering Norway's extensive forestry history, which spans over five centuries (Storaunet & Rolstad, 2020). Historical records reveal that wood exports from the region have been occurring for at least 500 years. The forestry practices in Norway experienced a significant transformation in the 1940s. Prior to this period, selective cutting was the norm, focusing on harvesting the largest trees due to their higher market value. However, this approach was gradually superseded by stand-based forest management strategies, predominantly emphasizing clear-cutting (Storaunet & Rolstad, 2020).

Today, approximately 60% of Norwegian forests have been subjected to at least one cycle of clear-cutting, drastically altering the forest landscape. This has resulted in a mosaic pattern of even-aged forest stands, each representing different successional stages. These stages range from recently clear-cut areas to mature forests. Remarkably, only a mere 1.7% of the forest remains untouched by any form of forestry activity, highlighting the extensive impact of human endeavors on these long-established forest ecosystems (Storaunet & Rolstad, 2020).

Boreal forests, constituting 10-15% of Earth's land area, are significant for their role in the global carbon cycle, holding approximately one-third of the world's forest carbon. These forests are vital in absorbing atmospheric carbon dioxide and are crucial for climate change mitigation (Ameray et al., 2023; Peichl et al., 2023). In Norway, a critical debate centers around the more effective carbon storage approach: actively managed forests or protected old-growth forests. While forest protection might enhance carbon sequestration in boreal ecosystems, some advocate for sustainable forestry practices. They argue that using harvested timber as a substitute for fossil resources, coupled with the higher carbon uptake of younger trees, could offer greater environmental benefits (Flugsrud et al., 2016).

In boreal forest ecosystems, understanding the dynamics and functionality of roots, especially fine roots, is essential for assessing forest health, productivity, and carbon sequestration (Cudlin et al., 2007). Trees in these ecosystems absorb carbon dioxide from the atmosphere, storing it in their biomass, including above and below-ground structures such as roots. Fine roots are characterized by their high physiological activity, smaller diameter and higher abundance and turnover rate compared to coarse roots, playing a pivotal role in water and nutrient uptake, as well as in the carbon exchange from trees to soil (Clemmensen et al., 2013; Finér et al., 2007; Leppälammi-Kujansuu et al., 2014). This process is crucial for the formation of soil organic matter, thereby enhancing the forest's ability to sequester carbon (Germon et al., 2020; Prescott & Grayston, 2023).

Forest management activities that affect fine root biomass and ectomycorrhizal fungal communities can significantly influence the health and functioning of forest ecosystems (Prescott & Grayston, 2023). The interplay of fine root dynamics, including morphology such as specific root length and root tip abundance, plays a substantial role in the global carbon cycle, accounting for approximately 22% of terrestrial net primary production (McCormack et al., 2015).

Specific Root Length (SRL) is a key indicator of forest productivity, health, and carbon sequestration. SRL, which measures the length of root per unit of biomass, indicates the efficiency and capacity of roots in resource uptake and carbon storage. A high SRL, characterized by longer and thinner roots, suggests that trees need to extend further to access nutrients, indicative of lower nutrient availability in the environment. Conversely, a low SRL, marked by shorter and thicker roots, implies that nutrients are more readily available in the environment (Cudlin et al., 2007; Ostonen et al., 2007).

The abundance of root tips in forests suggests the presence of mycorrhizal associations, where a hyphal sheath surrounds each mycorrhizal root tip. These associations are crucial for nutrient acquisition in boreal forests with typically scarce resources. These symbiotic relationships between fungi and plant roots enhance nutrient and water uptake, contributing to increased carbon sequestration. Thus, the abundance of root tips can indicate the extent of mycorrhizal relationships and their impact on carbon sequestration (Freschet, Pagès, et al., 2021; Peng et al., 2022).

Examining fine root morphology is pivotal in understanding carbon sequestration in boreal forest ecosystems. Despite being relatively understudied, these characteristics are critical in evaluating forest health, productivity, and carbon sequestration potential. Their study is essential for effective forest management and conservation, particularly for climate change mitigation strategies (Freschet, Roumet, et al., 2021).

Launched in 2021, the EcoForest project is an interdisciplinary initiative funded by the Norwegian Research Council, directly addresses these critical topics. As an interdisciplinary initiative, it aims to deepen our understanding of how modern forestry management practices impacts biodiversity, carbon storage, and ecological processes in boreal forests. It involves researchers from the Norwegian University of Life Sciences (NMBU), University of Oslo (UiO), Norwegian Institute of Bioeconomy Research (NIBIO), and Norwegian Institute for Nature Research (NINA).

This master thesis is a part of that broader effort and seeks to elucidate the importance of fine roots in carbon sequestration and the impact of forest management practices on this essential process. Furthermore, it specifically explores whether there are notable differences in fine root characteristics between CC and NN forests and examines the potential implications of these differences has on the contribution of tree fine roots to carbon sequestration in soil. I tested following hypotheses:

H1) Specific Root Length (SRL) as an Indicator of Forest Health and Productivity: I hypothesize that spruce fine roots in CC forests will exhibit a lower SRL compared to NN forests. This expectation is based on the active management strategies employed in CC forests, which are specifically tailored to favor spruce trees.

H2) Distribution of Fine Root Biomass in Soil Layers: I anticipate a higher concentration of fine root biomass in the litter-fibric-humic (LFH) layer relative to other soil strata. In CC forests, due to the predominance of spruce, the LFH layer is expected to contain more spruce fine root biomass. Conversely, NN forests, with their diverse tree and vegetation species, are likely to demonstrate greater competition among plants for space in the LFH layer, thereby affecting root biomass distribution.

H3) Root Tip Abundance and Mycorrhizal Associations: My hypothesis is that NN forests will exhibit a greater number of root tips compared to CC forests, indicative of a more prevalent mycorrhizal presence. In NN forests, which typically have greater forest structure

diversity and more complex ecosystem structures, the demand for nutrients might be higher due to the presence of a variety of trees in different stages of growth. This diversity can lead to a more intricate web of mycorrhizal associations, as different species of fungi may associate with different tree species.

Materials and methods

Study site

The study site for the EcoForest project were selected to encompass a diverse range of forested areas within southeastern Norway (Figure 1). Commencing in 2021, the project established twelve distinct locations, each featuring two designated plots that embody contrasting forest management histories. Latitude, longitude, and elevation (m a.s.l) were registered for each plot (Table 1). The first of these plots represents near-natural forests (NN), which have been subject to minimal human influence, maintaining an ecological state close to their natural condition. The second set of plots corresponds to mature, even-aged forests (CC) that have experienced one clear-cutting operations in the past.

Table 1. *List of sites and their geographical position (given in decimal degrees and meters above sea level) as well as mean annual temperature (MAT; °C) and mean annual precipitation (MAP; mm). Macroclimate (MAT and MAP) is modelled, at a 100 x 100 m scale, for the period 2004 to 2014. Forest types are mature previously clear-cut mature forests (CC) and near natural (NN) forests.*

| Nr	Site name (abbr.)	Forest type	Latitude	Longitude	Elevation	MAT	MAP
1	Skotjernfjell (SKO)	CC	60.2413	10.8084	571	2.2	972
		NN	60.2422	10.7960	610	2.3	974
2	Gullenhaugen (GUL)	CC	60.3700	10.7872	591	2.1	854
		NN	60.3526	10.7966	668	1.9	867
3	Hemberget (HEM)	CC	60.9211	12.1889	584	0.6	766
		NN	60.9151	12.2065	581	0.5	764
4	Braskereidfoss (BRA)	CC	60.7476	11.9264	332	2.3	683
		NN	60.7398	11.9285	427	2.1	684
5	Särkilampi (SAR)	CC	60.2005	12.5281	388	2.4	762
		NN	60.1877	12.5080	368	2.4	761
6	Øytjern (OYT)	CC	60.8432	10.4090	663	1.4	819
		NN	60.8389	10.3812	640	1.3	818
7	Tretjerna (TRE)	CC	60.5773	10.2285	520	2.6	821
		NN	60.5836	10.2265	472	2.7	821
8	Halden (HAL)	CC	59.0798	11.5595	197	5.4	1051
		NN	59.0798	11.5465	211	5.3	1056
9	Blåfjell (BLA)	CC	59.7880	10.3865	322	4.8	1049
		NN	59.7831	10.3813	264	4.8	1041
10	Storås (STR)	CC	60.2615	9.7091	432	2.8	884
		NN	60.2591	9.7007	483	2.6	888
11	Marker (MRK)	CC	59.3835	11.7590	178	4.9	960
		NN	59.3601	11.7900	187	4.9	971
12	Langvassbrenna (LAN)	CC	60.2010	10.4980	548	2.6	883
		NN	60.2018	10.4738	607	2.2	868
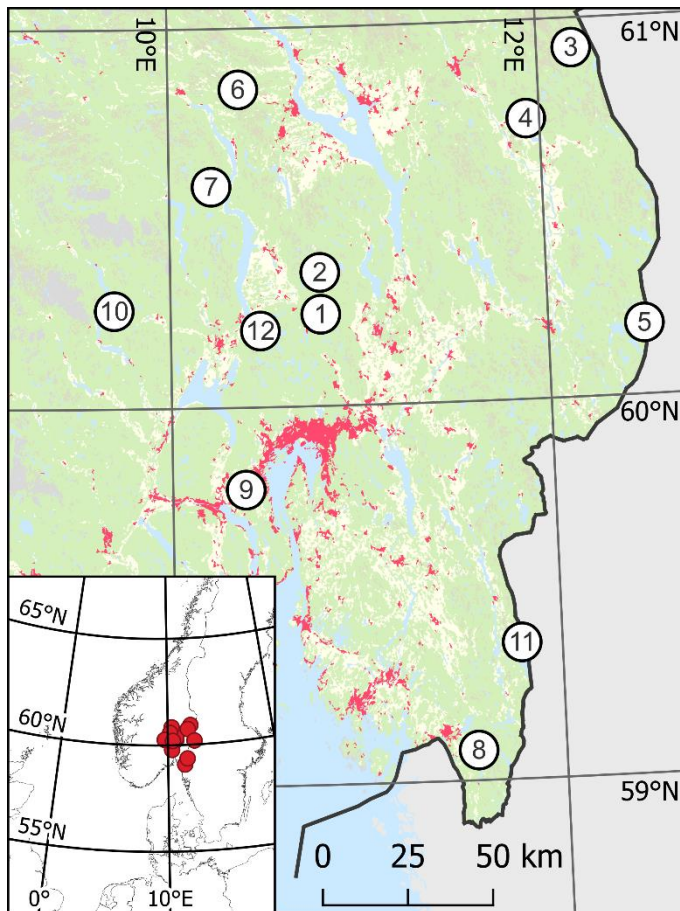
Figure 1. *Map over the study site in Southeastern Norway showing the location of all sites. See table 1 for sitenames and more detailed geographical information (map by Johan Asplund).*

The selection criteria for the plots were defined to ensure comparability between the NN and CC pairs. Proximity was a key factor, with the stipulation that while the NN and CC should be situated near one another, they must not be adjacent. The average distance between NN and CC was 1300 m, with a range from 690 to 3140 m. Uniformity in environmental variables was important; both types of plots were required to share the same site index, vegetation type, general soil characteristics, exposure (aspect), and slope, thereby controlling environmental variability.

The initial focus of the study was on forest dominated by spruce, with a specified site index (H40) of G17, which refers to the expected height of the dominant trees at a reference age of 40 years and is a common metric for assessing forest productivity. However, data on site index was not available for NN stands and had some variation among the CC stands. Additionally, the terrain of the selected plots had to be even and devoid of hydrological features such as streams or water bodies, which could introduce additional ecological

dynamics. While it was intended for these plots to be free of thinning processes, ditching, or significant pest attacks, this was not always achievable. Consequently, some plots, such as Blåfjell, exhibited signs of significant pest attacks.

For the clear-cut plots (CC), certain conditions were mandated to ensure that the forests were indeed representative of managed lands. A minimum basal area of 20 m²/ha was required, providing a quantitative measure of forest density.

Conversely, the near-natural plots (NN) were required to demonstrate minimal signs of human-induced alterations. Most importantly no signs of a clear-cut. This encompassed an absence of dead wood removal, something that used to be common practice in managed forests but one that can substantially alter the habitat and available resources for forest biota.

Within each of the 24 plots, 6 subplots were established using a randomized design (figure 2). From these subplots, soil samples were collected. Prior to this study, samples from two of these subplots per plot had been analyzed in early 2023 as a part of the master thesis of Martina Vårdal. For the present investigation, I have analyzed samples from one additional subplot from each plot, incorporating basal area of living spruce (m²/ha) and microclimatic conditions such as soil and ground temperature. These variables have not been previously included in the dataset. Consequently, the dataset for this study will comprise data from 3 subplots from each of the 24 plots.
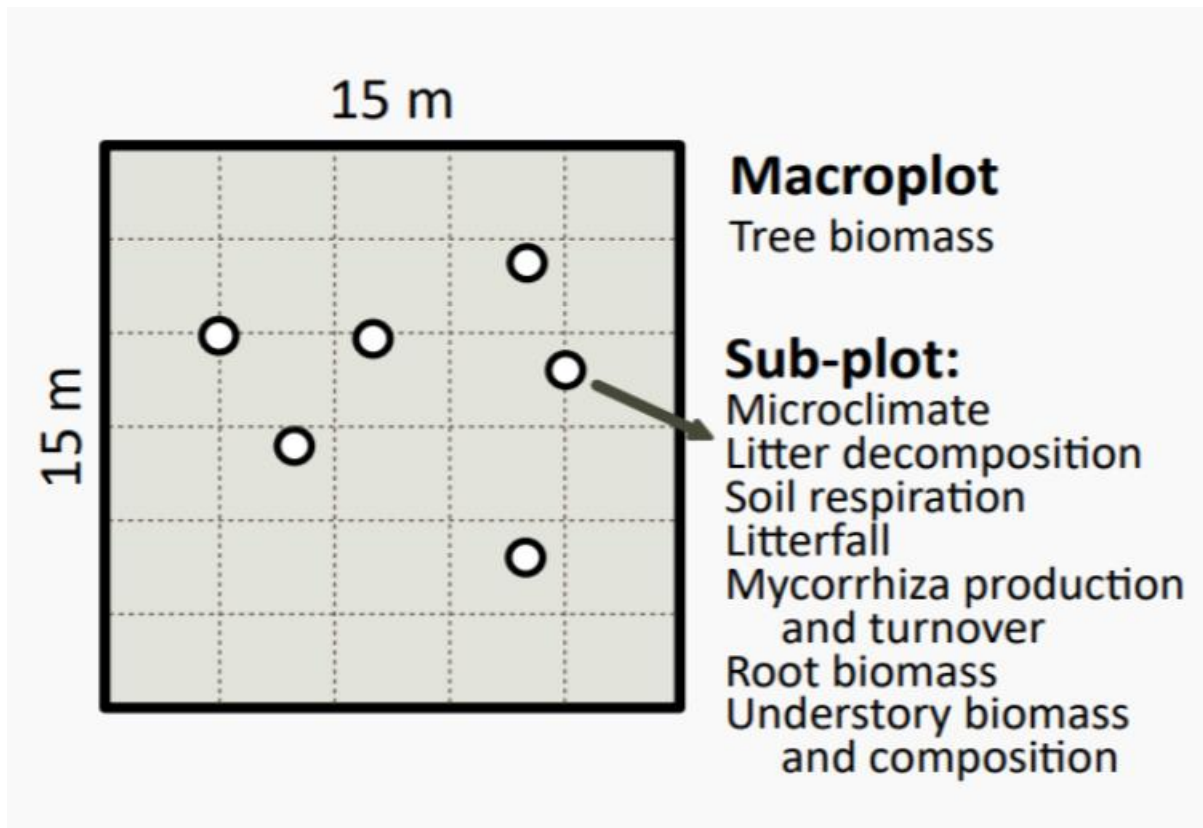
Figure 2. *Illustration of a randomized design showing a plot of 15 x 15 m scale with 6 subplots with scale of 1 x 1 m randomly placed within. In the subplots, variables such as microclimate, litter decomposition, soil respiration, litterfall, mycorrhiza production and turnover, root biomass, and understory biomass and composition were registered or to be determined by the end of the EcoForest project (illustration by Johan Asplund, slightly modified by thesis author).*

Sampling of soil

Soil cores were collected in each of the six subplots in June 2022 by use of cylindrical augers ($\emptyset$ = 6.6 cm). The protocol for these cores included the collection of the entire soil profile starting from the LFH (litter-fibric-humic) horizon, extending into the mineral soil to a minimum depth of 5 cm, thus ensuring a comprehensive soil sample consisting of both the humus layer and the upper mineral soil.

Despite the intentions of the protocol, there were instances where the samples did not conform to the expected standard. Some cores fell short of including the 2-5 cm range of the mineral soil layer. Nevertheless, these imperfect samples were not excluded from the dataset but were incorporated to maintain the integrity and continuity of the collected data.

To ensure the preservation of their characteristics, the soil core samples were promptly frozen after the field sampling. The sorting process is time-consuming, and immediate freezing helped maintain their integrity until they could be processed and sorted.

After defrosting, the soil cores were sectioned into specified layers for in-depth analysis. Standard practice consisted of the identification of three layers. In the case of deeper samples, a fourth layer was discerned. These segments included the surface LFH layer, the upper 0-2 cm of the mineral soil, the intermediate 2-5 cm of the mineral soil, and the lower rest mineral (LRM) layer, encompassing soil below the 5 cm depth.

The total length and weight of the soil cores were measured, as were the length and weight of each individual layer. To estimate the dry matter weight, representative samples from each soil layer were taken. All roots were removed from these samples before they were dried at 105 °C for approximately 24 hours.

In the assortment of samples I sorted, there was a notable variation. Specifically, three samples (one in Øytjern CC and one in each in the two Halden plots) exhibited LFH layers exceeding 10 cm in length, with the longest measuring 17.4 cm and the shortest just 0.75 cm. Additionally, two samples (same sample in Øytjern CC and one in Storås NN) had a $0 - 2$ cm mineral soil layer that was not the expected 2 cm thickness. Furthermore, three samples (one in each following plots; Braskreidfoss CC, Øytjern CC, and Halden NN) were missing the $2 - 5$ cm mineral soil layer. In contrast, eight samples (one in Skotjernfjell CC, Gullenhaugen NN, Hemberget CC, Tretjerna CC, Marker CC, and Langvassbrenna CC, and finally one in each of the two Blåfjell plots) featured a mineral soil layer that extended beyond 5 cm depth, falling into the category of LRM.

Sorting of roots
The process continued with the careful separation of roots from the soil in all identified layers. This was conducted with a sieve, a pan, and water to ensure that roots were cleanly extracted from the soil. Subsequently, the roots, particularly those from spruce trees, but also from other plant species, were sorted. Spruce roots were further classified by size into three distinct categories based on their diameter: fine roots (<2 mm), medium roots (2-5 mm), and large roots (>5 mm). Only living fine roots were included in this study.

LFH layers exceeding a length of 10 cm were bisected, and only one half was subjected to the fine root sorting process. This step was taken to manage the volume of material and to streamline the sorting process.

Scanning of fine roots

Spruce fine roots, defined as those less than 2 millimeters in diameter, were meticulously prepared for scanning by laying them flat and fully submerged in water within a clear plastic tray. This tray was then positioned on an Epson flatbed scanner (EPSON Flatbed Expression 11000XL 1.8 V3.49, Regent Instruments, Canada) with a resolution of 2400 DPI, which was interfaced with a laptop running the WinRhizo™ software (WinRhizo2013d, Regent Instruments, Canada). WinRhizo is a specialized program that assesses various root parameters, including diameter, length, and the count of root tips and forks. While the software can measure additional characteristics, only the aforementioned parameters were relevant and utilized for this study.

After scanning, all roots were weighed, then dried for approximately 72 hours at 70 °C and weighed again after drying to estimate root biomass.

Statistical analysis

Data points from the study were systematically recorded into a Microsoft Excel spreadsheet, which was then utilized as the dataset for further analysis within RStudio. Within this dataset, specific root length (SRL), measured as root length (in meters) per gram root biomass (m/g), was the sole response variable subjected to log transformation to normalize the data distribution for analytical purposes. Other response variables, maintained in their original scale, included average root diameter in millimeters (mm), root biomass per square meter of soil (g/m^2), and the count of root tips per square meter of soil (n/m^2).

The analysis incorporated a variety of explanatory (independent) variables to elucidate the factors influencing root characteristics. These variables encompassed elements of macroclimate, such as annual precipitation in millimeters (mm) and the mean temperature during the warmest quarter—spanning June, July, August, and September. Macroclimate is modelled at a 100 x 100 m scale and using data from SeNorge which encompasses the period from 2004 to 2014 (Horvath et al., 2019). Microclimate variables included the growing degree days (GDD), calculated as the cumulative number of days with temperatures above 5 °C for both aboveground (air) and belowground (soil) conditions, gathered in July 2022. Both air and soil temperature were recorded using TMS-4 (TOMST s.r.o, Praha, Czech

Republic). The basal area of living spruce trees, measured in cubic meters per hectare (m^2/ha), was also integrated into the analysis as an influential factor.

Notably, data from fine roots collected from subplot 3 in Skotjernfjell within the 2-5 cm mineral soil layer were removed from the final dataset. This exclusion was due to their extreme values, which were identified as outliers with the potential to skew the overall results.

Results

None of the measured root traits showed variation between forest types. However, notable differences were observed in average root diameter, root biomass, and the number of root tips across different soil layers (Table 2, figure 4, 5 & 6). The only trait not exhibiting any differences was SRL (Table 2, figure 3). The average diameter of fine roots decreased with soil depth, with roots in the organic layer being approximately 15% wider than those in the deepest mineral soil layer.

Additionally, significant variation was noted in fine root biomass per surface area across soil layers, suggesting a distinct pattern where root biomass per unit surface area changes with soil depth (Figure 5). However, it should be noted that different soil volumes and masses were sampled and therefore biomass estimates are not straightforward to compare. In terms of the number of fine root tips per surface area, a significant variation across soil layers was also observed, indicating a clear difference in root tip density in different soil layers (Figure 6). This finding emphasizes a notable alteration in the distribution of root tips across various soil depths.

When environmental factors such as micro- and macroclimate or forest structure were included in the analysis, the patterns observed in root traits became less distinct. Moreover, these environmental variables did not significantly impact the measured root traits (Table 2).

Table 2. *ANOVA-table (F [p]-values) derived from linear mixed effects models testing for the effect of forest type and layer on the effect of specific root length, average root diameter, root biomass per soil surface area and number of root tips per soil surface area. For each of the covariates in addition to a model without covariates. Sub-plot nested within plot and site were used as random effect. Significant values are in bold. Specific root length is log-transformed.*

	Specific fine root length (log(g/m))	Average fine root diameter (mm)	Fine root biomass/ Surface area (g/m^2)	Fine root tips/ Surface area (n/m^2)
No covariate				
Forest type	0.93 (0.33)	0.80 (0.373)	0.09 (0.764)	0.54 (0.462)
Layer	0.42 (0.733)	7.97 (**<0.001**)	57.98 (**<0.001**)	81. 26 (**<0.001**)
Forest type: Layer	0.63 (0.590)	0.68 (0.562)	0.31 (0.816)	0.18 (0.908)
Annual precipitation				
Forest type	0.66 (0.420)	0.02 (0.882)	0.44 (0.834)	0.08 (0.780)

Layer	0.26 (0.858)	0.83 (0.477)	0.60 (0.614)	2.15 (0.092)
Annual precipitation	0.05 (0.833)	0.51 (0.488)	0.36 (0.558)	0.01 (0.919)
Forest type: Layer	0.41 (0.745)	0.35 (0.789)	0.89 (0.448)	0.11 (0.953)
Forest type: Annual precipitation	0.52 (0.473)	0.01 (0.962)	0.42 (0.527)	0.14 (0.714)
Layer: Annual precipitation	0.29 (0.830)	1.69 (0.172)	0.47 (0.704)	0.51 (0.677)
Three-way interaction: Annual precipitation	0.36 (0.785)	2.01 (0.116)	0.84 (0.474)	0.15 (0.930)
Mean temperature during warmest quarter				
Forest type	0.02 (0.882)	0.21 (0.647)	0.69 (0.409)	0.54 (0.466)
Layer	0.83 (0.477)	2.68 (**0.050**)	1.83 (0.145)	0.19 (0.902)
Mean temperature warmest quarter	0.42 (0.527)	0.64 (0.427)	0.62 (0.432)	0.19 (0.672)
Forest type: Layer	0.35 (0.789)	0.52 (0.668)	0.36 (0.779)	0.38 (0.771)
Forest type: Mean temperature warmest quarter	0.01 (0.962)	0.16 (0.695)	0.49 (0.486)	0.62 (0.432)
Layer: Mean temperature warmest quarter	1.69 (0.172)	2.38 (0.072)	1.48 (0.222)	0.40 (0.755)
Three way interaction: Mean temperature warmest quarter	2.01 (0.116)	0.46 (0.714)	0.35 (0.787)	0.41 (0.742)
Air temperature				
Forest type	0.01 (0.928)	0.01 (0.969)	0.80 (0.384)	0.48 (0.493)
Layer	0.63 (0.599)	2.84 (**0.040**)	0.23 (0.876)	0.28 (0.840)
Air temperature	0.26 (0.619)	0.00 (0.998)	0.08 (0.784)	0.13 (0.725)
Forest type: Layer	0.42 (0.740)	1.08 (0.359)	0.63 (0.596)	0.17 (0.918)
Forest type: Air temperature	0.00 (0.968)	0.00 (0.962)	0.73 (0.404)	0.61 (0.438)
Layer: Air temperature	0.68 (0.564)	1.86 (0.140)	0.60 (0.613)	0.86 (0.463)
Three-way interaction: Air temperature	0.42 (0.742)	0.92 (0.435)	0.53 (0.660)	0.21 (0.889)
Soil temperature				
Forest type	0.04 (0.834)	0.75 (0.395)	0.75 (0.395)	0.23 (0.633)
Layer	0.60 (0.614)	2.24 (0.086)	2.24 (0.086)	3.24 (**0.021**)
Soil temperature	0.06 (0.816)	0.03 (0.863)	0.03 (0.863)	0.04 (0.853)
Forest type: Layer	0.89 (0.448)	0.35 (0.788)	0.35 (0.788)	0.02 (0.996)
Forest type: Soil temperature	0.01 (0.939)	0.71 (0.407)	0.71 (0.407)	0.37 (0.545)
Layer: Soil temperature	0.74 (0.532)	0.20 (0.897)	0.20 (0.897)	0.28 (0.841)
Three-way interaction: Soil temperature	0.86 (0.463)	0.28 (0.841)	0.28 (0.841)	0.03 (0.992)
Basal area				
Forest type	0.55 (0.459)	0.18 (0.669)	0.08 (0.776)	0.08 (0.783)
Layer	0.74 (0.529)	2.51 (0.061)	2.67 (**0.050**)	8.54 (**<0.001**)
Basal area	1.82 (0.180)	2.30 (0.137)	0.59 (0.446)	0.11 (0.745)
Forest type: Layer	2.02 (0.114)	2.82 (**0.041**)	0.07 (0.976)	0.53 (0.658)
Forest type: Basal area	0.18 (0.675)	0.02 (0.877)	0.05 (0.820)	0.19 (0.667)

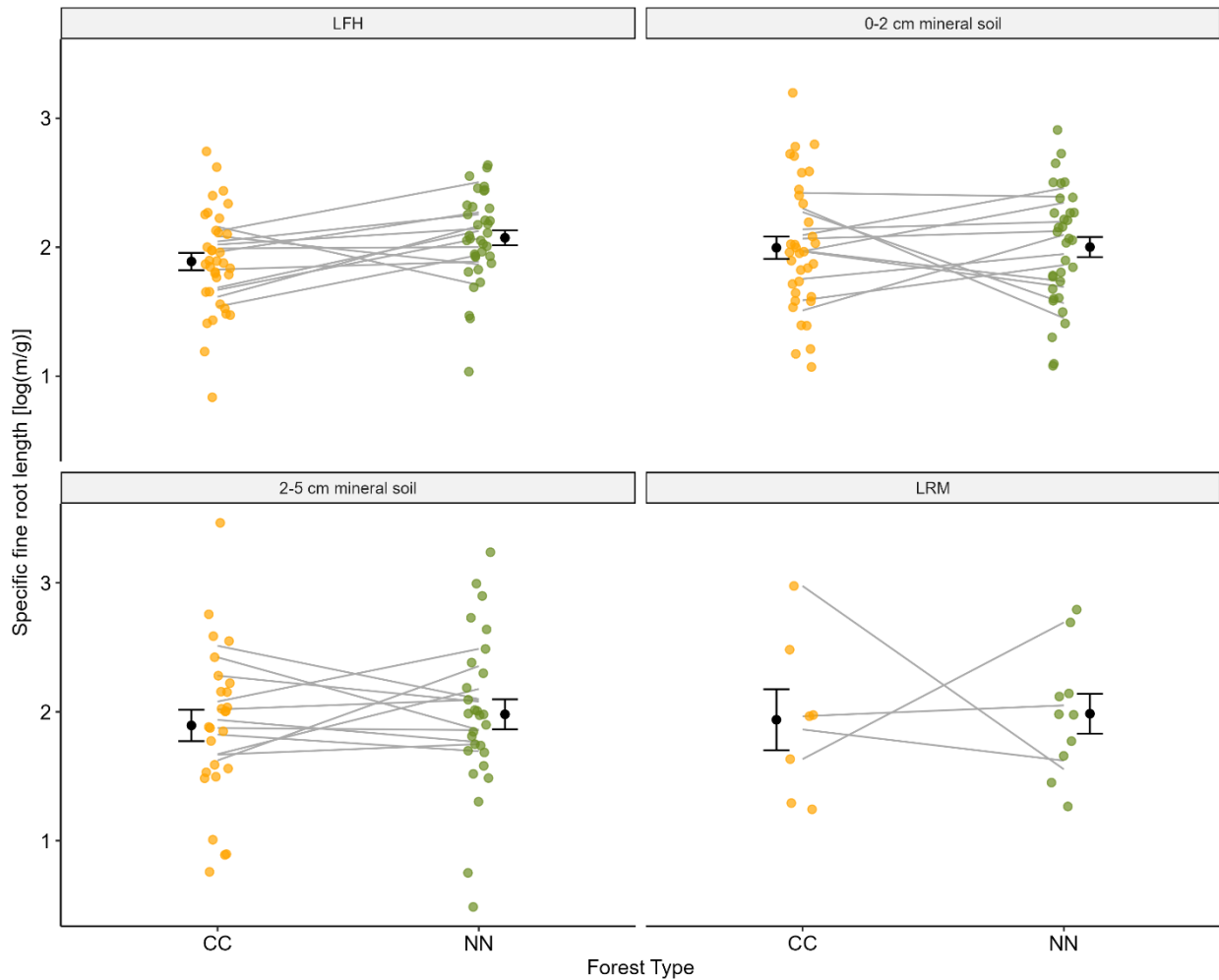| Layer: Basal area | 0.95 (0.421) | 1.49 (0.221) | 0.02 (0.995) | 0.91 (0.434) |
| Three-way interaction: Basal area | 1.64 (0.182) | 2.53 (0.060) | 0.12 (0.946) | 0.42 (0.736) |

Specific fine root length

Figure 3. *A comparative analysis of specific root length log transformed of spruce fine roots (<2 mm) across forest types; CC – clear cut (yellow) and NN – near natural (green), and soil layers; litter-fibric-humic (LFH), 0 – 2 cm mineral soil, 2 – 5 cm mineral soil, and lower rest mineral (LRM). Error bars indicate the standard error of the mean. Grey lines indicate the difference between the two forest types within the same site.*
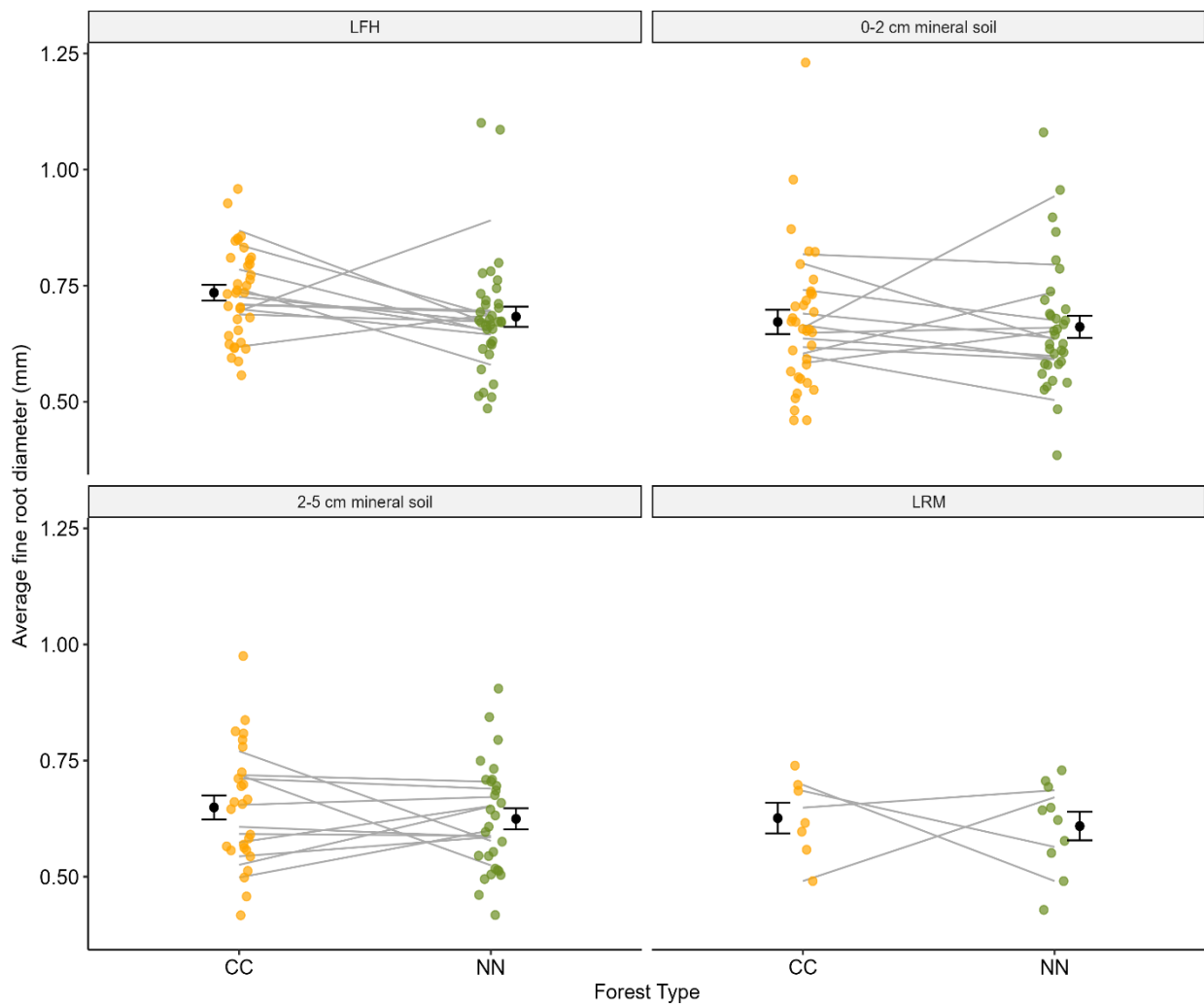
Average fine root diameter

Figure 4. *A comparative analysis of average root diameter of spruce fine roots (<2 mm) across forest types; CC – clear cut (yellow) and NN – near natural (green), and soil layers; litter-fibric-humic (LFH), 0 – 2 cm mineral soil, 2 – 5 cm mineral soil, and lower rest mineral (LRM). Error bars indicate the standard error of the mean. Grey lines indicate the difference between the two forest types within the same site.*
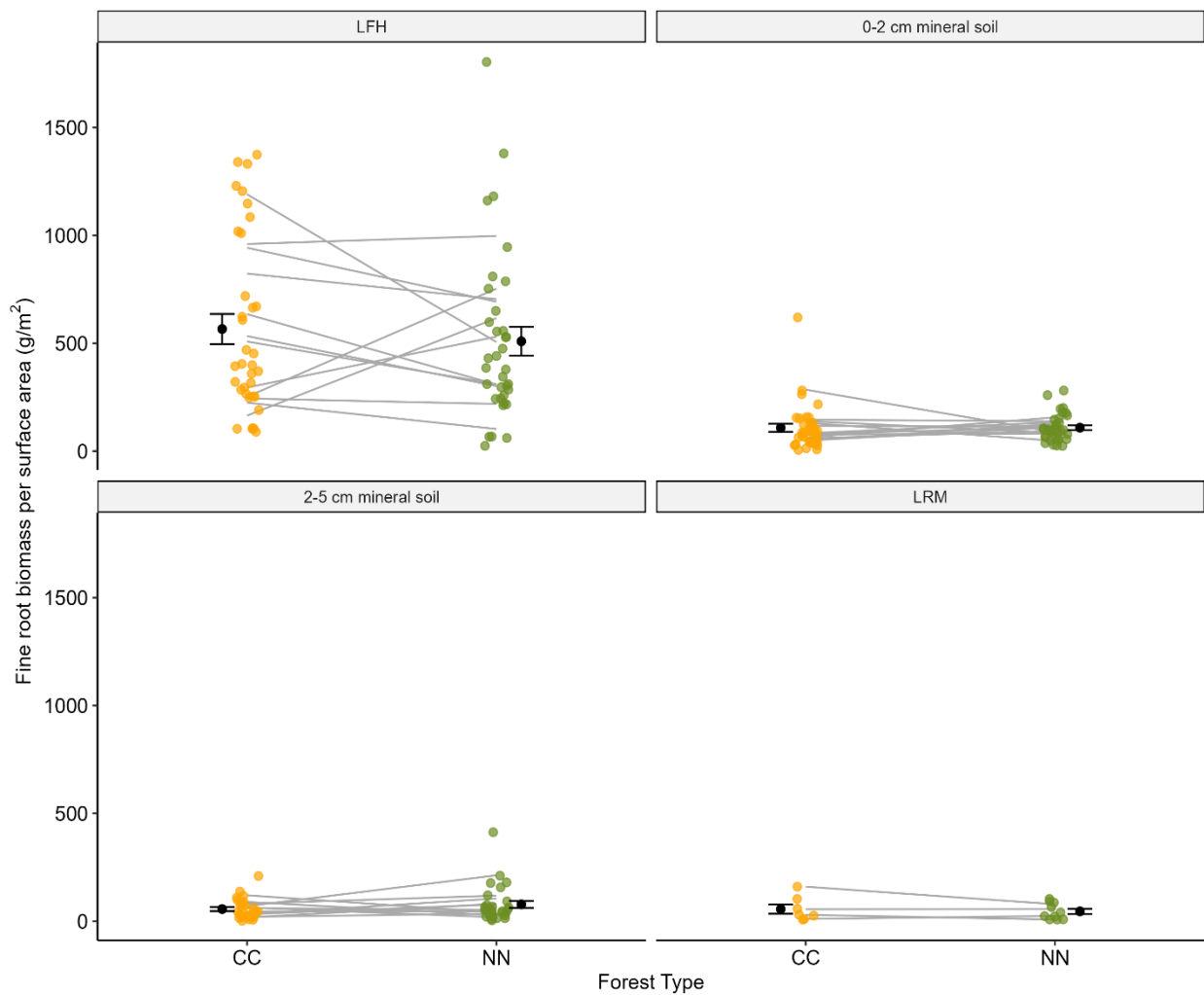
Fine root biomass

Figure 5. *A comparative analysis of biomass per surface area (g/m²) of spruce fine roots (<2 mm) across forest types; CC – clear cut (yellow) and NN – near natural (green), and soil layers; litter-fibric-humic (LFH), 0 – 2 cm mineral soil, 2 – 5 cm mineral soil, and lower rest mineral (LRM). Error bars indicate the standard error of the mean. Grey lines indicate the difference between the two forest types within the same site.*
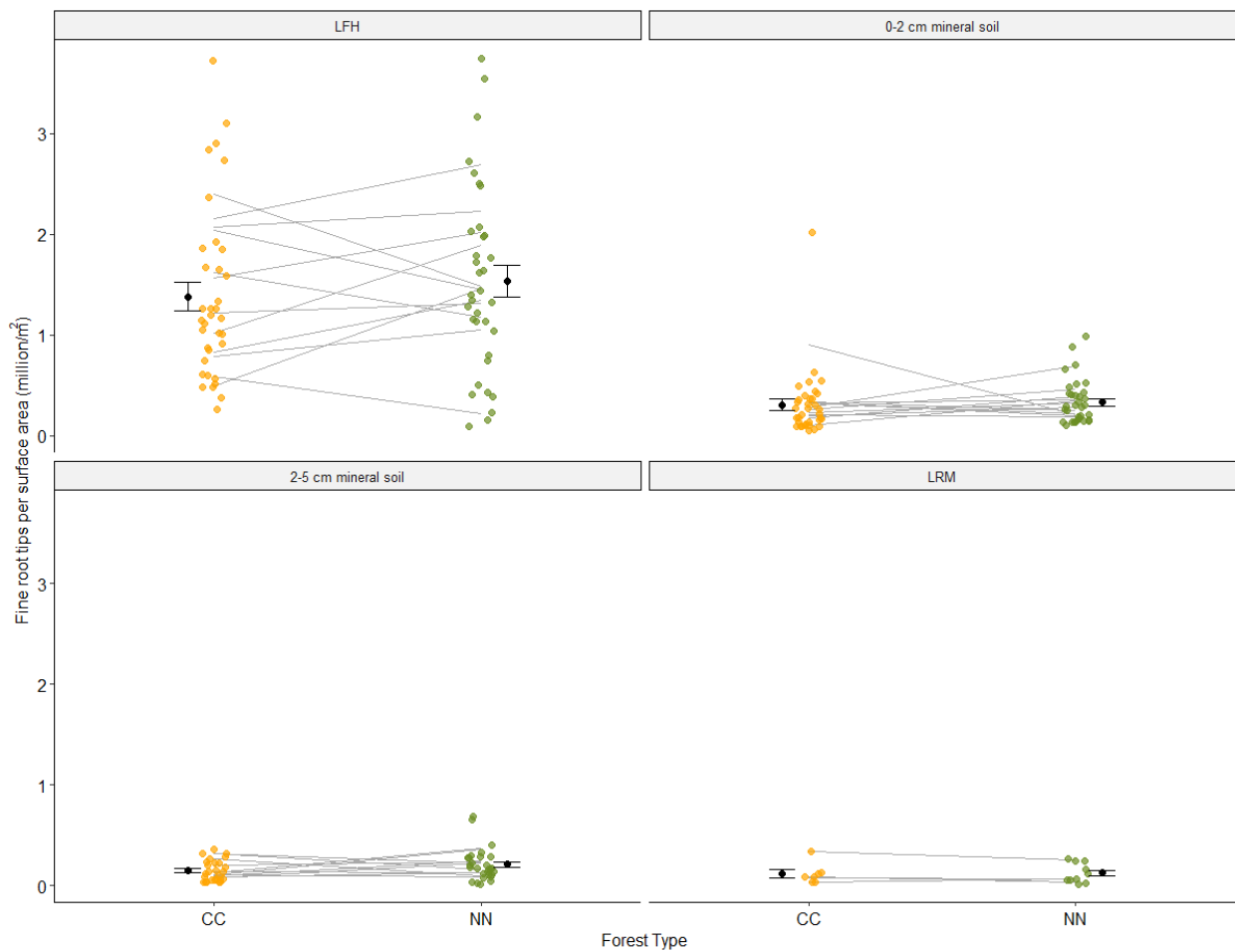
Fine root tips

Figure 6. *A comparative analysis of tips per surface area (million/m^2) of spruce fine roots (<2 mm) across forest types; CC – clear cut (yellow) and NN – near natural (green), and soil layers; litter-fibric-humic (LFH), 0 – 2 cm mineral soil, 2 – 5 cm mineral soil, and lower rest mineral (LRM). Error bars indicate the standard error of the mean. Grey lines indicate the difference between the two forest types within the same site.*

Discussion

The boreal forests of Fennoscandia, largely dominated by spruce, have experienced significant ecological and management changes over the years, mainly due to forestry practices. About 60% of Norwegian forests have been clear-cut at least once, leading to a landscape of even-aged stands, and leaving a minimal portion of the forests in their natural state (Helseth et al., 2022; Storaunet & Rolstad, 2020).

Boreal forests are crucial in the global carbon cycle, holding a significant portion of the world's forest carbon and playing a key role in climate change mitigation (Ameray et al., 2023; Peichl et al., 2023). The debate in Norway centers around the more effective carbon storage approach - whether actively managed forests or protected old-growth forests are better for carbon sequestration. Some advocate sustainable forestry practices, citing the benefits of using harvested timber as a substitute for fossil resources and the higher carbon uptake of younger trees (Flugsrud et al., 2016).

However, this is a narrow view of the forest's role in the carbon cycle, since it centers around the carbon stored above-ground. Roots, especially fine roots, are often an overlooked component of forest ecosystems, and they play a significant role in forest carbon cycling. Spruce have been found to have the highest fine root production among the common tree species in Norwegian forests (Pine, *Pinus sylvestris*, and birch, *Betula pubescens*) and may as a result have an larger impact on the carbon cycling in boreal forests (Hansson et al., 2013).

This is why the study of fine root morphology is crucial in not only evaluating forest health and productivity, but also for carbon sequestration potential. The abundance of root tips suggests the presence of mycorrhizal associations (Soudzilovskaia et al., 2015), which are vital for nutrient acquisition in boreal forests and contribute to increased carbon sequestration.

In this context, understanding the dynamics and functionality of roots, particularly fine roots, is essential in boreal forest ecosystems. Fine roots, characterized by their high physiological activity, play a pivotal role in water and nutrient uptake, and in the carbon exchange from trees to soil, forming a significant part of the soil organic matter and enhancing the forest's carbon sequestration capability (Adamczyk, 2021).

My research indicates that in mature forests, the variations in root traits appear minimally affected by past forest management practices. Contrary to expectations, neither microclimatic

nor macroclimatic factors seemed to significantly affect these traits. In fact, these factors including forest structure (basal area of living spruce), when included in the analysis, made the patterns in root traits less distinct. Furthermore, none of my hypotheses were supported by the results.

My first hypothesis predicted a lower SRL in CC forests compared to NN forests. Contrarily, SRL was found to be similar across these forest types, implying that management practices in CC forests might not significantly influence SRL of fine roots as previously hypothesized. SRL is a valuable environmental indicator, as it has been shown to decrease markedly under conditions such as fertilization and aluminum stress. Additionally, SRL tends to respond negatively to diminished light exposure, higher temperatures, and elevated CO_2 levels (Ostonen et al., 2007). Variations in SRL could signal different nutrient availability across various environments. However, this was not the case in my research.

Regarding my second hypothesis, significant variations were noted in the distribution of fine root biomass across soil layers, with a higher concentration observed in the LFH layer. However, this variation in distribution was not directly associated with forest types. These results suggest a complex interplay between soil layers and fine root biomass, partially supporting this hypothesis, but not confirming the anticipated difference between CC and NN forests.

As for my third hypothesis, I did not find a discernible difference in the number of fine root tips between CC and NN forests. This indicates that the expected increase in root tip abundance and mycorrhizal associations in NN forests, attributed to greater diversity in forest structure and more complex ecosystem structures, were not observed in this study.

The task of sorting fine roots is both intricate and labor-intensive, which accounts for their relatively understudied status and the paucity of associated research. While my findings diverge from previous studies, with the exception of higher fine root biomass in the LFH layer (Børja et al., 2008). Both forest management, or more precisely clear-cutting (Achat et al., 2015; Lacroix et al., 2016), and macro- and microclimate factors have been found to impact fine root biomass in forest soil. There is also research that shows there is a correlation between basal area of trees and fine root biomass (Finér et al., 2007; Helmisaari et al., 2007).

The discrepancies between my results and prior research might be largely attributed to the limited dataset in this study, which comprises only three replicates from each plot. Significant geographical differences among these plots introduce a high degree of local-scale variability that could significantly affect the outcomes. Moreover, the variation in tree spacing and density across different subplots is another critical factor that could influence the results. Previous studies on fine root biomass and dynamics often involved smaller geographic areas, which may explain some of the variation in findings.

This is also further compromised by the differing volumes and masses in the soil samples, which complicates direct comparisons of biomass estimates. Comparing fine root biomass on an area basis is challenging in this scenario. Instead, standardizing fine root biomass by soil depth rather than surface area could yield more accurate results (Finér et al., 2007). However, considering that the depths of the organic layer naturally vary greatly, there is no simple solution to this.

Furthermore, to preserve the integrity of the samples and prevent decomposition or respiration, as detailed in the materials and methods section, the roots were subjected to freezing. It is critical to acknowledge that the freezing and subsequent thawing process renders the roots more vulnerable and prone to damage (Freschet, Pagès, et al., 2021). This increased susceptibility could potentially impact the results negatively.

It should also be noted that two of the replicates were previously sorted and analyzed in an earlier thesis. The data from these two replicates, which included 48 samples, have been merged with an additional replicate comprising 24 samples, which I sorted and analyzed for this thesis. However, incorporating this extra replicate did not enhance the analysis. In fact, it had the opposite effect, leading to less distinct patterns in the findings (Vårdal, 2023). This highlights the importance of consistent sorting and handling of root samples.

Another explanation may be the forest owner structure in Norway. Forest properties and subsequently stands in Norway are usually on a smaller scale compared to other countries (Statistisk sentralbyrå, 2023b), which may eliminate the largest effects of clear-cutting on ecological processes in forests. Moreover, mature clear-cut forest stands have the potential to develop characteristics akin to those of near-natural forests, provided they are given sufficient time to recover and regenerate (Palviainen et al., 2005). This is particularly relevant

considering that the rotation period for forest stands in Norway typically spans approximately 70 to 80 years.

To mitigate these errors, several measures could be beneficial. Expanding the sample size would make the dataset more representative, diminishing the influence of outliers and bolstering the study's statistical strength. Standardizing the sorting and analysis procedures across all researchers would help to minimize variations arising from different handling techniques. Ideally, the same researchers should perform all tasks, including sorting and analysis, to reduce variability due to individual differences. Moreover, adopting blind or double-blind methods, where researchers are unaware of specific sample details, can be an effective strategy to avoid bias in sorting and analysis.

Conclusion

The study reveals that mature forests' fine root traits are only marginally affected by historical forest management, challenging previous beliefs about the influence of micro and macroclimatic factors, as well as forest structure, on these traits. Nevertheless, the study's credibility is somewhat diminished by its limited sample size and inconsistencies arising from different researchers processing the samples, necessitating a cautious approach when interpreting the results.

The discrepancies observed between the study results and the initial hypotheses underscore the intricate nature of below-ground ecological dynamics and the significant influence of various environmental factors on root traits. These findings highlight the need for more comprehensive research to decipher these complex interactions and relationships.

To improve future research accuracy, it is advised to expand the sample size, standardize procedures, and implement blind analysis methods. Such steps would enhance the clarity of research findings and aid in understanding the role of fine roots in forest ecosystems.

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