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Original Article

The effects of arginine phosphate (ArGrow® Granulat) on growth of Scots pine and Norway spruce seedlings planted in varying soil layer structures simulating site preparation

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Abstract

The pressure to plant seedlings in unprepared instead of prepared soil in forest regeneration is increasing, although seedlings' growth in unprepared soil is poorer. One way to improve seedlings' growth could be to add arginine phosphate to the planting hole at the time of planting. In field studies, abiotic and biotic damage normally disturbs studies which seek to determine growth effects, especially in unprepared soil. The study's aims were (i) to clarify whether it was possible to investigate the differences between varying soil layer structures mimicking site preparation methods and seedling treatments in simulated planting places in partly controlled conditions; and (ii) to investigate the effects of arginine phosphate (arGrow® Granulat) on the growth, especially root growth, of Norway spruce (*Picea abies* (L.) H. Karst.) and Scots pine (*Pinus sylvestris* L.) container seedlings in those simulated planting places. Seedlings of both tree species were planted in 40 l pots sunk partly into the soil in a sandy field. Before planting, pots were filled with soil layers mimicking layers in soil without any site preparation treatment and in spot mounds. After one growing season, the differences in growth parameters between treatments were small in Norway spruce seedlings, and no significant benefits of arginine phosphate were observed. The soil in the spruce pots was fine-textured and contained a thick organic layer. More studies in more common forest soil types suitable for Norway spruce seedlings are needed to be sure about the growth responses. In Scots pine seedlings, adding arginine phosphate granules to the planting holes improved the growth of seedlings at least in the first summer after planting in soil layers mimicking no mechanical site preparation situation, and the growth improvement may even have compensated the mounding effects. In conclusion, mimicking site preparation methods in large pots was a promising method by which the differences between site preparations methods as well as effects of seedling treatments, in our case arginine phosphate, on the growth of conifer seedlings, can be tested.

Keywords: Picea abies; Pinus sylvestris; mechanical site preparation; planting; nutrient concentrations

Introduction

Scots pine (*P. sylvestris* L.) and Norway spruce (*P. abies* (L.) H. Karst.) are the main planted tree species in Northern European forests. Over 95% of planted seedlings are of these tree species in Finland, Norway, and Sweden (Solvin et al. 2021). Today, most seedlings are planted in mechanically prepared soil. Of the mechanical site preparation (MSP) methods, mounding is the most used method in Finland, and 72% of prepared sites were mounded in 2020 (OSF: Natural Resources Institute Finland n.d.). However, there is pressure to reduce or even prohibit the use of MSP due to the environmental reasons, mainly because of rumored effects on natural waters, and social sustainability. In continuous cover forestry, and especially in small caps, to enhance and accelerate the establishment of new tree generation, supplementary-like planting of seedings may be needed on some sites. On these sites, site preparation is impossible or too costly. In unprepared soil, the risk of pine weevil feeding damage (Luoranen et al. 2017, 2022; Wallertz et al. 2018) and competing vegetation (Nilsson and Örlander 1995; Örlander et al. 1996; Johansson et al. 2005) are higher, soil temperatures (Nilsson and Örlander 1999; Heiskanen et al. 2013; Hansson et al. 2018) and air temperatures surrounding planted seedlings are lower (Heiskanen et al. 2013), soil water content can be higher or lower (depending on the weather conditions and planting place in different MSP method) (Nilsson and Örlander 1999; Heiskanen et al. 2013; Hansson et al. 2018), and seedlings have fewer available nutrients (Heiskanen and Rikala 2006; Johansson et al. 2007) than seedlings planted in prepared soil, especially in mounds. The survival potential and growth of seedlings are therefore better in prepared soil than seedlings planted in unprepared soil (Sikström et al. 2020).

Independent of the planting spot structure, the rapid root growth of planted seedlings is important to ensure a good field performance of tree seedlings (Burdett 1990). The larger the root system is, the more effectively the planted seedling can take nutrients and water from the surrounding soil, and their ability to survive under stressful conditions after planting improves

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(Grossnickle 2005). However, the root growth is restricted in unprepared soil for the above unfavorable conditions.

In plants, most nitrogen (N) is stored as arginine (Siddappa and Marathe 2020). Root growth, especially the proportion of fine roots, and mycorrhizal colonization in planted Scots pine and Norway spruce seedlings have been observed to increase when organic N sources are used instead of inorganic N during nursery growing (Gruffman et al. 2012). Using organic N also enhances seedling nutrition (Lim et al. 2021). The effects of applying arginine during nursery growing and at the time of planting may differ. Häggström et al. (2021) studied the effects of arginine phosphate (ArGrow® Granulat, Arevo Ab, Umeå, Sweden) in planted Scots pine seedlings. Based on their results, arginine phosphate granules added to the planting hole of Scots pine seedlings at the time of planting increased the survival potential of seedlings in elevated planting positions (humus and mineral soil layers had been turned over the intact humus). However, arginine phosphate did not affect the survival of Scots pine seedlings in unprepared soil but increased growth of seedlings in some extent. They did not find any effects on growth in elevated planting positions. As far as we know, there is no published research into the effects of the addition of arginine phosphate to the planting hole at the time of planting of Norway spruce seedlings in Nordic boreal conditions.

Under field conditions, several factors affect seedlings' development. For example, in the study of Häggström et al. (2021), the survival of seedlings planted in elevated planting positions and unprepared soil was only 71%–77% and 58%, respectively. High risk of damage in unprepared soil complicates the growth comparison of different seedling treatments in forest conditions. Pine weevil feeding damage and abiotic damage causing agents both increase mortality and reduce the growth of damaged but still living seedlings (Luoranen et al. 2017, 2022). In addition, the digging up of the whole root system without cutting the long roots grown out from peat plugs in forest conditions is difficult. This is the case especially in unprepared soil with vegetation around a sampled seedling, when it is difficult to separate the roots of planted seedlings and other plants. The length of the longest roots can be 35 cm (Heiskanen et al. 2013) or even over 50 cm (J. Luoranen, personal observation) in seedlings planted in mounds after the first growing season. Precise measurements and comparisons of biomass in different treatments under more controlled conditions might therefore be preferable.

Root growth potential and the timing of the root growth of planted seedlings have long been studied in controlled indoor conditions, planting seedlings in cassettes (Mattsson 1991) or small pots (<1 l) (e.g. Luoranen et al. 2005, 2006, Luoranen 2018). In these small-pot tests, the volume of pots and soil layers has not been the same as in real conditions on regeneration sites. The question arose for us as to whether it would be possible to conduct these growth comparison tests on 'a mound scale' in more controlled conditions. The volume of big industrial scale plastic pots is about 40 l, about the same as the volume of the spot mound. Using such pots, a planting platform could be developed by adding forest soil to pots with the same layering structure as in soils without any MSP (soil layers in the same order as in regeneration sites before site preparation) or on spot mounds (double humus layer under the mineral soil cover; see Sikström et al. 2020). Planting seedlings in these pots and growing seedlings in these simulated planting places in more controlled growing conditions make treatment comparisons more accurate when there are no risks of damage disturbing growth. Using such a platform, the comparison of early growth, and especially root growth, in different seedling treatments in different soil layer structures mimicking MSP treatments could also be more economic, with fewer seedlings but sufficient statistical efficacy and generalizable results for regeneration site conditions.

Our study aims to test the suitability of the outlined simulated planting places with different soil layer structures in studying the early performance of newly planted seedlings, as well as to investigate the effects of arginine phosphate granules (later called ARG treatment) given to the planting hole at the time of planting on growth variables of Norway spruce and Scots pine seedlings. We tested this in simulated planting places with spot mound layers (later called spot mounds) and forest soil layers without any MSP treatment (later called No MSP treatment). The hypothesis was that (i) ARG treatment has no effects on growth parameters in mounds, but that it increases growth in simulated No MSP - treatment; and (ii) the effects on growth are similar in Norway spruce and Scots pine seedlings.

Material and methods

Seedling material

One-year-old Scots pine and 2-year-old Norway spruce container seedlings were grown in FinForelia Oy's Saarijärvi nursery in Central Finland. The seeds used to raise seedlings were from seed orchards (SV436 for Scots pine and SV235 for Norway spruce) and were appropriate for use in Central Finland's conditions. The seedlings were grown in hard-walled plastic containers (Plantek PL81F, 81 seedlings per container, cell volume: 85 cm^3, surface area of a cell: 18.3 cm^2, growing density: 546 cells m^{-2}; BCC Ab, Landskrona, Sweden), which were filled with base-fertilized and limed light sphagnum peat. All the seedlings were grown in raised pallets, and they were irrigated and fertilized regularly during nursery growing. On 18 May 2022, a seedling container from both tree species was transferred from Saarijärvi to Suonenjoki, where the seedlings were watered daily until planting in pots.

The planting experiment was established in a sandy field at the Suonenjoki Research Unit of Natural Resources Institute Finland (Luke) (62°39′N, 27°03′E, 142 m above sea level) in Central Finland. The experiment was established using a split-plot design with 10 blocks, one seedling within each tree species and treatment combinations within a block (Fig. 1). Tree species was used as the main plot within a block, site preparation (spot mound or No MSP) was used as the subplot, and seedling treatment (arginine phosphate application; with (ARG) or without (CO)) within the site preparation treatment was used as the sub-subplot. For the experiment, 40 seedlings per tree species were used, a total of 80 seedlings.

Establishment

The seedlings were planted in 40-l black plastic pots (diameter 50 cm; height 34 cm; holes in the bottom), which were filled with soil that mimicked the structure of soil layers in soil without any MSP or in a spot mound (Fig. 1a). To grow the Scots pine seedlings, the soil for the pots was taken from the regeneration site, which was formerly Scots-pine-dominated. The site type was a sub-xeric dryish heath (Tonteri et al. 1990; *Vaccinium* type in the Cajander (1949) forest site type classification), the soil was podzol soil with a medium-coarse soil texture type (sand), and with a 2–5 cm thick humus layer overlaying the mineral soil layer. The main tree species before cutting was Scots pine, and it was clearcut in the winter of 2020–2021. The soil for the Norway spruce pots was taken from a clearcut formerly Norway-spruce-dominated site (clearcut in the winter of 2022). The site type was mesic fresh heath (*Myrtillus* type). The site was quite wet, and the soil texture

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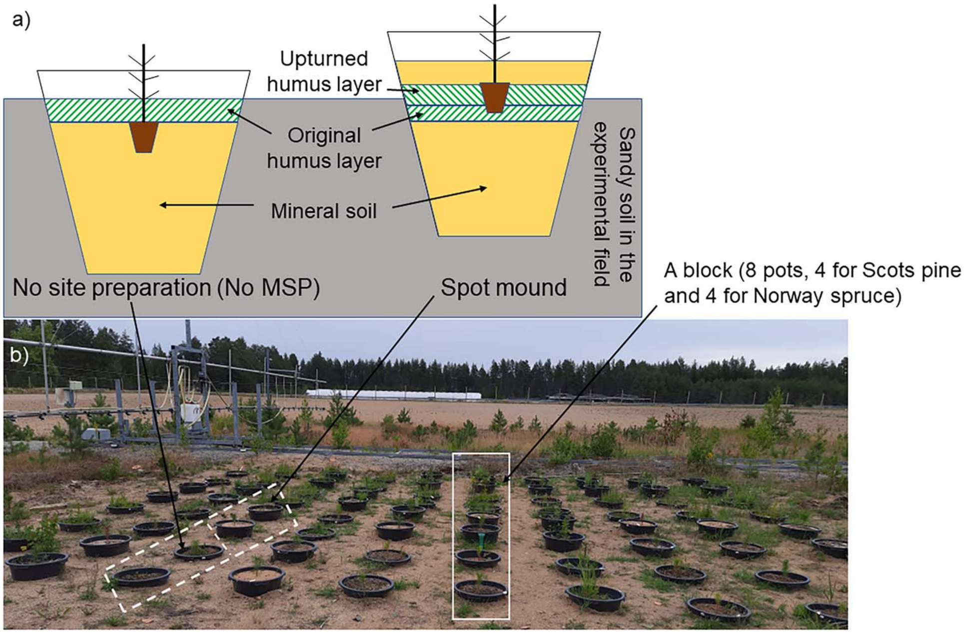

Figure 1. a) Schematic illustration of soil layers in the 40 l pots and b) the experimental design for the experiment with two simulated site preparation treatments, spots mound, and no MSP; on the photo, a block consisting of eight pots, four pots for Norway spruce and four for Scots pine, have been drawn with white square; the squares with broken lines indicate sub-plots with two pots for simulated mounds and two pots that had been filled with normal forest soil layers (no site preparation); seedling treatments, either arginine phosphate or control, were randomized within these subplots; photo: Jaana Luoranen.

type was fine (fine sand, partly silt sand). An organic layer about 10 cm thick was on the mineral soil composed partly of organic material, which indicated that the site had perhaps previously been arable land. On the bottom of the pots, there was root barrier fabric (Cello, polypropylate 220 g m^{-2}; produced for Kesko Oy, Finland) and on this, a 5 cm layer of light expanded clay aggregate granules (Leca©; 4–10 mm round particles; loose bulk density 275 kg m^{-2} ± 15%; Saint-Gobain Finland Oy, Helsinki, Finland). On these layers, the simulated structures of the No MSP treatment and spot mounds were built up by taking different soil layers from clearcut sites. The simulated spot mounds were made in 40 pots (20 pots with soil layers taken from a Scots pine site, 20 pots from a Norway spruce site) by putting mineral soil in the pots, then a humus layer including also field layer (called later simply humus layer) in the same position as in the forest, a humus layer upside-down on the first humus layer, and finally, on the turned humus layer, a 6 cm thick layer of mineral soil (Fig. 1a). Before adding the upper mineral soil layer, the lower layers were packed compactly, mimicking the effect of a mounding device. Simulated planting places in No MSP-treatment were also made in 40 pots (20 from the soil taken from the Scots pine site, 20 from the soil taken from the Norway spruce site) by putting the fabric and Leca-granules in the mounds and then adding mineral soil and humus layers in the same position as they were in the forest. The pots were not fully filled, with about 10 cm from the surface of the soil to the top of the pot. Pots with No MSP treatment were then placed in the soil in Suonenjoki so that the soil level in pots was at the same level as the soil surface in the surrounding soil (Figs 1 and 2). Pots with mounds were placed so that the soil level in the pots was 10 cm above the soil surface around the pot (simulating the height of the mounds in the regeneration site conditions).

When the pots were in the soil, the seedlings were planted to a depth of 5 cm in the center of pots, one seedling per pot, using a planting tube (Pottiputki, BCC Ab, Landskrona, Sweden) on 6 June 2022. At the time of planting, one dose (0.4 g) of granular arginine phosphate (arGrow© Granulat, Arevo Ab, Umeå, Sweden) was added with dosing feeder to the bottom of the planting hole of one seedling within a site preparation treatment, a total of 10 seedlings in each seedling treatment and tree species. The control (CO) seedlings were planted without arginine phosphate granules.

The soil's water content in the pots was measured with a portable TDR (Trime-FM, Imko Micromodultechnik, Germany, accuracy 0.1 %) from two pots per site preparation treatment and tree species once or twice a week. Between 29 June and 8 August, soil water contents in Scots pine pots were on average 22 ± 3 (standard deviation) and 21 ± 3 for No MSP and mounds, and, respectively, in Norway spruce 21 ± 5 and 26 ± 3 vol-%. Seedling pots were watered with 20 mm of water with an irrigation boom twice a week, excluding times when precipitation was >20 mm between irrigations according to the rain gauge placed between pots in the study field, and the soil's water content in the pots was >10 vol-%.

Larvae of the web-spinning sawfly (*Acantholyda hieroglyphica*) were found in 14 Scots pine seedlings. The larvae were removed as soon as they were found.

Weather conditions

Weather data were collected from the Suonenjoki Research Nursery's weather station. Monthly mean temperature in July 2022 was near the long-term average, but June, August, and October were warmer, and September cooler, and the precipitation sums in all months was lower than Suonenjoki's long-term (2000–2022) values (Table 1).

Measurements

Each seedling's height (accuracy 1 mm) and stem base diameter (accuracy 0.1 mm) were measured before and after planting in June and again at the end of the experiment in October. Heights were measured from the surface of a peat plug (before planting and after harvesting) or from the surface of the mineral soil in

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Figure 2. Pots filled with soil layers mimicking a, c) no site preparation treatments and b, d) mounds and to which a,b) Norway spruce or c, d) Scots pine seedlings had been planted; photos: Luke/Sirpa Kolehmainen.

Table 1. Monthly mean and minimum (in brackets) air temperatures (°C; at 2 m) and precipitation sum (mm) from June to October 2022, as well as the long-term average values (22-year average from 2000–2022) at Suonenjoki Research Nursery in Finland.

Month	**Air temperature, °C**		**Precipitation sum, mm**	
	2022	**Long-term average**	**2022**	**Long-term average**
June	16.0 (4.4)	14.4	48	64
July	17.2 (9.6)	17.3	66	78
August	17.1 (6.6)	15.2	41	70
September	7.8 (−1.1)	9.8	30	56
October	5.1 (−5.3)	3.8	21	50

the pots (after planting or before harvesting). The diameters were measured 2 cm above the peat plug or soil surface in pots. Each seedling's exact planting depth was calculated by the difference in height before and after planting.

On 18 October 2022, the seedlings were lifted from the pots. Uplifted seedlings were divided based on the current and previous years' needles and stems. The separation of the first- and second-year part of Scots pine seedlings were difficult in some cases due to the growth form of young seedlings. The roots grown out from peat plugs were cut and washed (current-year roots). The roots grown inside the peat plugs were called old roots and considered to be roots that had grown before planting. The peat around the roots was washed off. All plant parts were dried in an oven (48 h at 60°C) and weighed to an accuracy of 0.001 g. After weighing, the current-year needles of seedlings in three (or four) blocks were combined in three samples per treatment combination for both tree species. Needles of several seedlings had to be pooled to get enough biomass to nutrient analysis. From the samples, the nitrogen (N) concentration was determined with a CHN analyzer (modified ISO 13878 standard; LecoCN Trumac, LECO Corp., St. Joseph, MI, USA) in Luke's laboratory in Jokioinen, Finland. The nutrient concentrations of phosphorous (P), potassium (K), calcium (Ca), magnesium (Mg), manganese (Mn), boron (B), iron (Fe), zinc (Zn), aluminum (Al), and copper (Cu) were determined using an ICP-OES (Thermo Scientific iCAP 6500 Duo) in Luke's laboratory in Viikki.

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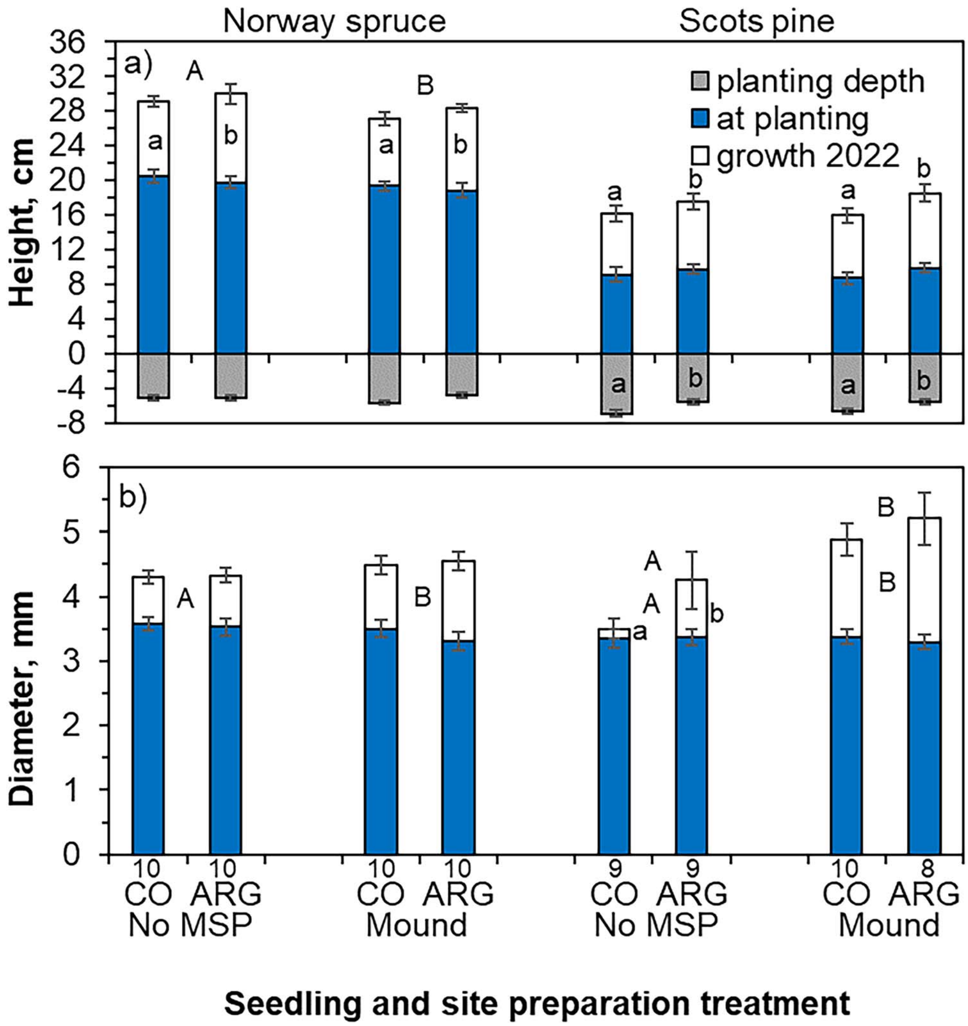

Figure 3. a) Height and planting depth (length of the stem below ground) and b) stem base diameter at the time of planting (measured after planting at soil surface) of Norway spruce and Scots pine seedlings in the rooting experiment; the figure also includes the height and diameter growth during the growing season; seedlings were planted in pots mimicking soil layers in No MSP treatment or spot mounds; at the time of planting, arginine phosphate granules were added to half the seedlings (ARG), and the other half were grown without it as a control (CO); the numbers below the bars indicate the number of healthy seedlings in each treatment combination included in the analysis; the lowercase letters indicate statistically significant differences between seedling treatments within a site preparation treatment and tree species, and the uppercase letters between site preparation treatments within a tree species; the letters above bars are for total heights or diameters, and the letters inside or between bars are for growth variables.

Statistical analysis

The differences in the morphological variables and nutrient concentration of needles between site preparation and seedling treatments within tree species were analyzed with linear mixed model analysis (MIXED) and normal variance analysis (ANOVA) respectively, and the probability of insect observation in Scots pine seedlings with a generalized linear mixed model (GLMM) in IBM SPSS Statistics Version 28.0.1.0. The fixed effects in the models were site preparation and seedling treatment and their interaction. In MIXED and GLMM analysis random effects were block, site preparation within a block, and seedling treatment within a site preparation and block. Normal distribution was tested with the Kolmogorov–Smirnov test, and homogeneity of variance with Levene's test. Where necessary, the values were log-transformed to achieve normal distributions and homogenous variance of residuals. Multiple comparisons were based on the Bonferroni method. A difference with a *P*-value of <0.05 was considered significant.

Results

During the experiment, one Scots pine seedling died, probably because of drought. In Scots pine seedlings no differences in damage caused by the larvae of web-spinning sawfly between treatments were found. Although attempts were made to remove larvae as soon as they were found, they ate needles from three pine seedlings (one control seedling in No MSP and two seedlings in ARG treatment planted in mounds), and these seedlings were removed from further analyses.

The mean height of planted Norway spruce seedlings was 20 ± 0.4 cm, and the stem base diameter was 3.5 ± 0.06 mm (Fig. 3). Seedlings were planted to a depth of 5 cm. At the end of the experiment, Norway spruce seedlings planted in No MSP treatment were 2 cm taller, but their diameter growth was poorer than in the seedlings planted in mounds. In both site preparation treatments, ARG treatment increased the height growth of seedlings compared with control seedlings without a statistically significant effect on the diameter growth or dry mass of seedlings

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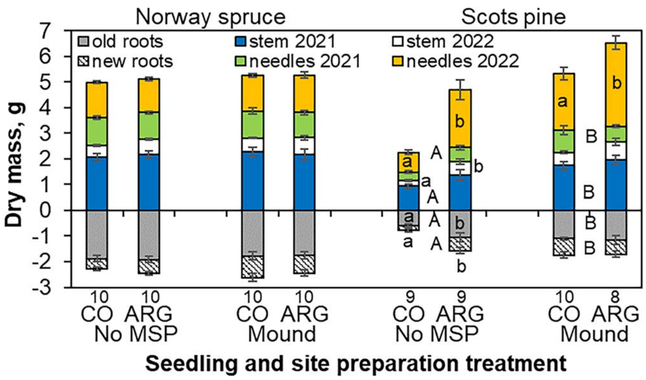

Figure 4. Dry mass of roots grown within root plugs (old roots) or grown out from plugs (new roots) and dry masses of aboveground parts (stems and needles) grown before planting (2021) and after planting (2022) in Norway spruce and Scots pine seedlings; the seedlings were grown in pots simulating No MSP or spot mounds; at the time of planting, arginine phosphate granules were added to half the seedlings (ARG), and the other half of the seedlings were grown without it as a control (CO); the numbers below the bars indicate the number of healthy seedlings in each treatment combination that were included in the analysis; the lowercase letters indicate statistically significant differences between seedling treatments within a site preparation treatment and tree species, and the uppercase letters between site preparation treatments within a tree species.

(Fig. 4; Table 2). There were no differences in any of the dry mass components between site preparation treatments. New roots to new shoots ratios did not differ statistically significantly between seedling and site preparation treatments.

The mean height of Scots pine seedlings was 15.5 ± 0.3 cm, and the stem base diameter was 3.5 ± 0.06 mm at the time of planting. Control seedlings were planted 1 cm deeper and were 2 cm shorter at the end of the first growing season than seedlings to which ARG granules were added (Fig. 3; Table 2). In No MSP treatment, the diameter growth of Scots pine seedlings was poorer, and seedlings were thinner than the seedlings planted in simulated mounds. ARG application affected seedlings' diameter during the experiment statistically significantly only in No MSP treatment in which ARG treated seedlings were thicker than control seedlings. Dry masses of old and new roots, as well as previous years' stems and needles and total masses of roots and shoots, were greater in mounds than in No MSP treatment (Fig. 4). Total dry masses of shoots and roots were also greater in ARG treatment than in control. Neither the seedling nor site preparation treatments affected the ratio of roots and shoots (needles + stem) grown after the planting.

Nutrient concentrations

The N and P concentrations of Norway spruce needles were higher in seedlings grown in mounds than in No MSP (Table 3). In Scots pine needles, Mn concentrations were statistically significantly higher in mounds than in No MSP treatment. As a response to the ARG treatment, the differences between seedling treatments were statistically significant only in B concentrations in Norway spruce needles. The ARG treatment also decreased K concentrations in Norway spruce needles grown in No MSP treatment but not in mounds.

Discussion

In the simulated planting places embedded in large volume pots the differences between mounds and No MSP treatment, as well as the effects of arginine phosphate granules on seedlings' growth, were clearly visible. The effects of soil layer structures mimicking MSP methods and seedling treatments on the growth of seedlings were clearer in Scots pine than in Norway spruce seedlings. Unfortunately, in the Norway spruce seedlings, the soil texture on the site from which the soil in pots was taken was less well-structured podzol soil than on the Scots pine site. The Norway spruce site was probably former arable land, and clear layers of mineral soil and humus for simulated mounds were less easy to create than for the Scots pine pots. When comparing site preparation methods, the results for Norway spruce may therefore be less generalizable as those for Scots pine.

In the study, variations in other factors than studied treatments were tried to minimize to test how well the simulation of soil texture layers in pots works. For that, there was only one seedling material from one nursery per tree species, controllable growing conditions without disturbing damage causing agents and the same weather conditions to all seedlings. In this study environment, small seedling numbers (10) in each treatment were sufficient for observing possible statistically significant differences. At the same time, all these factors, as well as the fact that the study was made only in 1 year and in one soil texture type per tree species, impair the generalizability of the results. By this technique, however, it was possible to observe the previously well-known results that the growth of seedlings in soil structure with double humus layer (simulated mounds) improved growth in the comparison with structure with humus layer above mineral soil. This fact would suggest that the effects of arginine phosphate on the growth of seedlings in different soil layer structures mimicking different planting places were real.

The minor effect of site preparation treatment on the height growth of seedlings in the first growing season corroborates previous results (Johansson et al. 2005; Nilsson et al. 2019). At least a part of the current-year height growth in Scots pine (Thompson 1976) and Norway spruce (von Wühlisch and Muhs 1986) seedlings is predetermined in the second and third growing seasons. In predetermined growth, the previous year's growing conditions during bud development affects the stem units within a bud, and the season's height growth is therefore determined in both the previous year's conditions and in the growing season's conditions. The slightly increasing effect of arginine phosphate treatment on height growth is therefore probably caused by these current-year conditions. The effects of different soil layer structures on the height growth can be seen better in the later years. For example, in the study of Häggström et al. (2021), arginine phosphate increased the length of the leader shoot in Scots pine seedlings in the second season planted in unprepared soil, but the effect was minimal in elevated planting places. The reduced height growth of conifer seedlings in unprepared soil in the later years has been observed in several previous studies (e.g. Raulo and Rikala 1981; Johansson et al. 2005; Nilsson et al. 2019; Luoranen et al. 2022).

It was already possible to observe the effects of soil layer structures on other variables than height in the first growing season. The diameter growth of all seedlings and the dry mass of different plant parts in Scots pine seedlings planted in the treatment without MSP were smaller than those of the seedlings planted in mounds. The biomass results correspond to previous results: For example, Heiskanen and Rikala (2006) and Heiskanen et al. (2013) observed that the biomass of Norway spruce seedlings planted in unprepared soil was lower than in the seedlings planted in different kinds of mounds. The minor effect of site preparation on the growth of Norway spruce seedlings was probably caused

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Table 2. Statistical significances (*P*-values) given by the linear mixed models for the morphological variables measured at planting (2021) and at the end of the experiment (2022) in Scots pine and Norway spruce seedlings; significant *P*-values have been bolded.

Variable	Effect			
Norway spruce	Intercept	Site preparation (SP)	Seedling treatment (ST)	SP x ST
Planting depth	<0.001	0.549	0.122	0.091
Height at planting	<0.001	0.172	0.404	0.908
Height at the end of the experiment	<0.001	**0.028**	0.110	0.968
Height growth	<0.001	0.204	**0.001**	0.834
Diameter at planting	<0.001	0.246	0.342	0.577
Diameter at the end of the experiment	<0.001	0.378	0.930	0.995
Diameter growth	<0.001	**0.030**	0.292	0.551
Dry mass of stem before planting	<0.001	0.530	0.986	0.453
Dry mass of stem after planting (new)	<0.001	0.096	**0.007**	0.798
Dry mass of needles before planting	<0.001	0.519	0.336	0.969
Dry mass of needles after planting (new)	<0.001	0.428	0.951	0.291
Dry mass of roots before planting	<0.001	0.349	0.967	0.828
Dry mass of roots grown after planting (new)	<0.001	**0.008**	0.946	0.183
Total dry mass of shoots (needles+stem)	<0.001	0.475	0.822	0.861
Total dry mass of needles	<0.001	0.816	0.607	0.559
Total dry mass of roots	<0.001	0.408	0.993	0.400
New roots to new shoots ratio	<0.001	0.736	0.973	0.344
Scots pine				
Planting depth	<0.001	0.657	**<0.001**	0.253
Height at planting	<0.001	0.827	0.190	0.595
Height at the end of the experiment	<0.001	0.258	**0.027**	0.852
Height growth	<0.001	0.470	0.180	0.646
Diameter at planting	<0.001	0.656	0.665	0.844
Diameter at the end of the experiment	<0.001	**0.002**	0.083	0.479
Diameter growth	<0.001	**0.001**	**0.024**	0.416
Dry mass of stem before planting	<0.001	**<0.001**	0.072	0.552
Dry mass of stem after planting	<0.001	**0.008**	**0.009**	0.616
Dry mass of needles before planting	<0.001	**0.005**	0.849	**0.018**
Dry mass of needles after planting	<0.001	**0.003**	**<0.001**	0.341
Dry mass of roots before planting	<0.001	**0.028**	0.064	0.146
Dry mass of roots grown after planting	<0.001	**0.046**	0.301	0.073
Total dry mass of shoots (needles+stem)	<0.001	**0.002**	**0.007**	0.093
Total dry mass of needles	<0.001	**0.005**	**0.003**	**0.033**
Total dry mass of roots	0.090	**0.010**	0.103	0.051
New roots to new shoots ratio	<0.001	0.883	0.553	0.136

by the thick organic material layer in the soil and the fact that the mineral soil and organic material layers were less clear layers than they should have been and as they were in the Scots pine pots.

Despite the slightly unclear soil layers in Norway spruce pots, the N concentrations of Norway spruce needles were significantly lower in seedlings planted in the No MSP pots than in mounds. The N concentrations of Norway spruce needles grown in No MSP pots were near the deficiency and significantly lower than observed in previous studies. For example, in Finnish regeneration site conditions, the N concentrations of needles in 1- and 2-year-old Norway spruce seedlings were >20 g kg^{-1}, both in unprepared soil and mounds (Heiskanen and Rikala 2006). In mounds, peat plugs of planted seedlings were in the double layer of organic material while the bottom of peat plugs in No MSP treatment were in fine textured mineral soil. N may bind to the fine soil particles and may have decreased its availability to seedlings in No MSP treatment. In pots of MSP treatment, there were also more competing field vegetation, especially in Norway spruce pots (not measured, but visible in the pots; see Fig. 2). The effect of vegetation on planted seedlings is competition from water, as water content measurements in this study also showed, but also competition for nutrients (Örlander et al. 1996). Furthermore, Norway spruce seedlings probably took more N in mounds since net N mineralization can be higher in mounds than in unprepared soil treatment during planting summer (Smolander and Heiskanen 2007). Soil for Scots pine pots were taken from a poorer site type with thinner humus layer and N mineralization was probably lower and no differences in needles' N concentrations were found.

The Mn concentrations of control seedlings grown in No MSP treatment were also low, but the values can vary greatly, as the variation from 380 to 1010 mg kg^{-1} in different site preparation treatments and years in the study of Heiskanen and Rikala (2006) show. Compared with the values presented by Heiskanen and Rikala (2006), the concentrations of other nutrients in Norway spruce seedlings were at the same levels.

The N concentrations of needles in Scots pine pots, both unprepared soil and mounds, were about the same as they were in Norway spruce planted in mounds. Based on the results of Gruffman et al. (2014) and Lim et al. (2021), Scots pine seedlings can take up more organic N than inorganic N, and an organic source of N (like arginine) can markedly increase the nitrogen concentration

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Table 3. Nutrient concentrations (mean SE) of needles in Norway spruce and Scots pine seedlings planted in soil texture layers simulating No MSP or mounds and added arginine phosphate (ARG) or not (CO) at the time of planting. Nutrient concentrations were determined from three samples (pooled needles from three to four seedlings) in each treatment; statistical significances (*P*-values) given by ANOVA for site preparation (SP), seedling treatment (ST), and their interactions are also presented separately for tree species; the different letters indicate statistically significant differences between seedling treatments within a site preparation treatment (based on multiple comparison of Bonferroni method in SP x ST interactions; $P \leq 0.05$): lowercase for unprepared soil and uppercase for mounds; the italics in mound values indicate statistically significant differences between site preparation treatments; the bolded values are for statistically significant *P*-values.

Tree species	Treatment	N	P	K	Ca	Mg	B	Mn
		g kg^{-1}					mg kg^{-1}	
Norway spruce	Unp. CO	10.9 (1.5)	2.0 (0.2)	8.1 (0.3)a	3.4 (0.3)	1.3 (0.05)	11 (0.4)a	374 (34)
	Unp. ARG	11.2 (0.8)	2.0 (0.2)	7.2 (0.3)b	5.6 (0.4)	1.3 (0.1)	11 (0.4)a	511 (34)
	Mound CO	*22.5 (0.5)*	*2.8 (0.2)*	7.5 (0.3)A	6.7 (0.9)	1.1 (0.1)	12 (0.9)A	567 (53)
	Mound ARG	*19.6 (1.8)*	*2.6 (0.2)*	8.2 (0.1)A	5.2 (0.3)	1.3 (0.2)	9 (0.7)B	489 (46)
P-values	*SP*	***<0.001***	***0.005***	*0.422*	*0.065*	*0.481*	*0.410*	*0.079*
	ST	*0.353*	*0.645*	*0.782*	*0.853*	*0.438*	***0.016***	*0.510*
	SP x ST	*0.253*	*0.521*	***0.018***	***0.020***	*0.747*	*0.084*	***0.035***
Scots pine	Unp. CO	22.0 (1.7)	2.3 (0.1)	6.8 (0.5)	2.9 (0.2)	1.2 (0.06)	15 (1.7)	340 (50)
	Unp. ARG	20.9 (0.9)	2.2 (0.1)	7.2 (0.4)	2.6 (0.2)	1.3 (0.05)	11 (1.8)	365 (14)
	Mound CO	25.1 (0.5)	2.3 (0.03)	6.7 (0.3)	2.4 (0.2)	1.3 (0.04)	15 (1.5)	*534 (43)*
	Mound ARG	22.2 (1.5)	2.1 (0.1)	6.0 (0.2)	2.3 (0.2)	1.4 (0.1)	13 (1.2)	*503 (86)*
P-values	*SP*	*0.109*	*0.774*	*0.121*	*0.088*	*0.557*	*0.696*	***0.016***
	ST	*0.134*	*0.196*	*0.746*	*0.351*	*0.078*	*0.119*	*0.960*
	SP x ST	*0.493*	*0.642*	*0.183*	*0.779*	*0.859*	*0.652*	*0.624*

in Scots pine seedlings. However, no differences in N between seedling treatments in Scots pine in our study were found.

In both tree species, some growth-increasing effects of arginine phosphate were found, but the effects were stronger in Scots pine seedlings and in No MSP treatment. Both tree species can uptake arginine (Öhlund and Näsholm 2001), and the differences in responses probably have other causes. Arginine phosphate significantly increased the biomass growth in the current-year parts, i.e. roots grown out from peat plugs, current-year needles, and the stem of Scots pine seedlings planted in No MSP treatment, and the biomass growth was almost comparable with the growth of seedlings in mounds. Arginine phosphate has been observed to stimulate the root growth over the shoot growth in the nursery (Gruffman et al. 2012), but we observed no differences in the ratio of new roots to new shoots after planting.

Gruffman et al. (2013) studied the uptake of different sources of nitrogen (N), observing that in conditions in which the carbohydrate supply was restricted by environmental conditions, the Scots pine cuttings benefited from using organic N like arginine. In unprepared soil, the soil temperature and other environmental conditions are more challenging than in prepared soil and especially mounds, and seedlings may therefore utilize more organic arginine phosphate granules in unprepared soil. Norway spruce seedlings were planted in probable previous arable soil with thick organic layer and fine textured mineral soil making it difficult to generalize those results. In Scots pine, the arginine phosphate increased the growth of seedlings more in No MSP treatment than in mounds. In typical boreal forest soils, amino acids like arginine dominate in the diffusive flux of N compounds, and plant roots and mycorrhizal fungi are exposed to them (Inselsbacher and Näsholm 2012). In addition, as discussed above, the net mineralization of N from the organic material of the humus layer in mounds is high in the first growing season after site preparation (Smolander and Heiskanen 2007). There is thus probably enough organic N in the soil, and the roots of planted Scots pine seedlings have no need of the added arginine phosphate granules in mounds, and no effects were therefore found.

B levels in current-year needles of Scots pine seedlings were higher than they were in Norway spruce seedlings, but even in Norway spruce, the concentrations in all treatments were above the B deficiency limit of 3–8 mg kg^{-1}, which can cause deficiency symptoms (Brække 1983; Möttönen et al. 2001). In mounds, B concentrations in the current-year needles of Norway spruce seedlings treated with arginine phosphate were only 9 mg kg^{-1}. Based on the results of Möttönen et al. (2001), the low B concentrations may have affected the seedlings' root growth. The soil taken from Norway spruce sites contained much more organic material than soil taken from Scots pine sites. B can be bound to organic material (Lehto 1995), and this probably explains the lower B concentrations in Norway spruce seedlings than in the Scots pines in our study.

Conclusions

In conclusion, the simulation of soil layer structures mimicking different site preparation treatments in large-volume pots and growing seedlings in more controlled conditions were a promising and effective method for testing early responses of seedling treatments in different site preparation treatments without the disturbing biotic or abiotic damage usually observed in regeneration site tests. However, care must be taken when selecting soils for the experiment since in our case soil in Norway spruce pots were probably from former arable land and those results may not be generalizable to typical forest site types in boreal forest. Therefore, the comparison of responses between tree species is questionable but it seems that arginine phosphate affects the growth of seedlings more in poorer growth conditions as was the case in Scots pine seedlings planted in soil mimicking situation without MSP. Arginine phosphate increased the biomass growth

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of Scots pine seedlings planted in soil without MSP and to some extent in mounds. In soil without structure of any MSP, the diameter growth of all seedlings and the biomass growth of Scots pine seedlings were lower compared with seedlings planted in mounds. Further studies are needed to ensure that the better growth of treated Scots pine seedlings in soils without structures of any MSP continues in the following years, when the competition of field vegetation increases. It would be furthermore interesting to examine whether there are any effects on Norway spruce seedlings in real forest podzol soil conditions suitable for spruce.

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Data Availability

The data underlying this article will be shared on reasonable request to the corresponding author.

References

Brække FH. Micronutrients-prophylactic use and cure of forest growth disturbances. *Commun Inst For Fenn* 1983;**116**:20–6. http://urn.fi/URN:ISBN:951-40-0623-2.

Burdett AN. Physiological processes in plantation establishment and the development of specifications for forest planting stock. *Can J For Res* 1990;**20**:415–27. https://doi.org/10.1139/x90-059.

Cajander AK. Forest types and their significance. *Acta For Fenn* 1949;**56**:article id 7396. https://doi.org/10.14214/aff.7396.

Grossnickle SC. Importance of root growth in overcoming planting stress. *New For* 2005;**30**:273–94. https://doi.org/10.1007/s11056-004-8303-2.

Gruffman L, Ishida T, Nordin A. *et al.* Cultivation of Norway spruce and Scots pine on organic nitrogen improves seedling morphology and field performance. *For Ecol Manage* 2012;**276**:118–24. https://doi.org/10.1016/j.foreco.2012.03.030.

Gruffman L, Palmroth S, Näsholm T. Organic nitrogen uptake of Scots pine seedlings is independent of current carbohydrate supply. *Tree Phys* 2013;**33**:590–600. https://doi.org/10.1093/treephys/tpt041.

Gruffman L, Jämtgård S, Näsholm T. Plant nitrogen status and co-occurrence of organic and inorganic nitrogen sources influence root uptake by Scots pine seedlings. *Tree Phys* 2014;**34**:205–13 10.1093/treephys/tpt121.

Häggström B, Domevscik M, Öhlund J. *et al.* Survival and growth of Scots pine (*Pinus sylvestris*) seedlings in North Sweden: effects of planting position and arginine phosphate addition. *Scand J For Res* 2021;**36**:423–33. https://doi.org/10.1080/02827581.2021.1957999.

Hansson LJ, Ring E, Franko MA. *et al.* Soil temperature and water content dynamics after disc trenching a sub-xeric Scots pine clearcut in Central Sweden. *Geoderma* 2018;**327**:85–96. https://doi.org/10.1016/j.geoderma.2018.04.023.

Heiskanen J, Rikala R. Root growth and nutrient uptake of Norway spruce container seedlings planted in mounded boreal forest soil. *For Ecol Manage* 2006;**222**:410–7. https://doi.org/10.1016/j.foreco.2005.10.047.

Heiskanen J, Saksa T, Luoranen J. Soil preparation method affects outplanting success of Norway spruce container seedlings on till soils susceptible to frost heave. *Silva Fennica* 2013;**47**:article id 893. https://doi.org/10.14214/sf.893.

Inselsbacher E, Näsholm T. The below-ground perspective of forest plants: soil provides mainly organic nitrogen for plants and mycorrhizal fungi. *New Phytol* 2012;**195**:329–34. https://doi.org/10.1111/j.1469-8137.2012.04169.x.

Johansson K, Söderbergh I, Nilsson U. *et al.* Effects of scarification and mulch on establishment and growth of six different clones of *Picea abies*. *Scand J For Res* 2005;**20**:421–30. https://doi.org/10.1080/02827580500292121.

Johansson K, Nilsson U, Allen HL. Interactions between soil scarification and Norway spruce seedling types. *New For* 2007;**33**:13–27. https://doi.org/10.1007/s11056-006-9010-y.

Lehto T. Boron retention in limed forest mor. *For Ecol Manage* 1995;**78**: 11–20. https://doi.org/10.1016/0378-1127(95)03599-7.

Lim H, Jämtgård S, Oren R. *et al.* Organic nitrogen enhances nitrogen nutrition and early growth of *Pinus sylvestris* seedlings. *Tree Phys* 2021;**42**:513–22. https://doi.org/10.1093/treephys/tpab127.

Luoranen J. Autumn versus spring planting: the initiation of root growth and subsequent field performance of Scots pine and Norway spruce seedlings. *Silva Fennica* 2018;**52**:article id 7813.15. 10.14214/sf.7813.

Luoranen J, Rikala R, Konttinen K. *et al.* Extending the planting period of dormant and growing Norway spruce container seedlings to early summer. *Silva Fennica* 2005;**39**:article id 361. 10.14214/sf.361.

Luoranen J, Rikala R, Konttinen K. *et al.* Summer planting of *Picea abies* container-grown seedlings: effects of planting date on survival, height growth and root egress. *For Ecol Manage* 2006;**237**:534–44. https://doi.org/10.1016/j.foreco.2006.09.073.

Luoranen J, Viiri H, Sianoja M. *et al.* Predicting pine weevil risk: effects of site, planting spot and seedling level factors on weevil feeding and mortality of Norway spruce seedlings. *For Ecol Manage* 2017;**389**:260–71. https://doi.org/10.1016/j.forece.2017.01.006.

Luoranen J, Laine T, Saksa T. Field performance of sand-coated (Conniflex®) Norway spruce seedlings planted in mounds made by continuously advancing mounder and in undisturbed soil. *For Ecol Manage* 2022;**517**:120259. https://doi.org/10.1016/j.foreco.2022.120259.

Mattsson A. Root growth capacity and field performance of *Pinus sylvestris* and *Picea abies* seedlings. *Scand. J. For. Res.* 1991;**6**:105–12. https://doi.org/10.1080/02827589109382653.

Möttönen M, Lehto T, Aphalo PJ. Growth dynamics and mycorrhizas of Norway spruce (*Picea abies*) seedlings in relation to boron supply. *Trees* 2001;**15**:319–26. https://doi.org/10.1007/s004680100106.

Nilsson O, Hjelm K, Nilsson U. Early growth of planted Norway spruce and Scots pine after site preparation in Sweden. *Scand J For Res* 2019;**34**:678–88. https://doi.org/10.1080/02827581.2019.1659398.

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Nilsson U, Örlander G. Effects if regeneration methods on drought damage to newly planted Norway spruce seedlings. *Can J For Res* 1995;**25**:790–802. https://doi.org/10.1139/x95-086.

Nilsson U, Örlander G. Vegetation management on grass-dominated clearcuts planted with Norway spruce in southern Sweden. *Can J For Res* 1999;**29**:1015–26. https://doi.org/10.1139/x99-071.

Öhlund J, Näsholm T. Growth of conifer seedlings on organic and inorganic nitrogen sources. *Tree Phys* 2001;**21**:1319–26. https://doi.org/10.1093/treephys/21.18.1319.

Örlander G, Nilsson U, Hällgren J-E. Competition for water and nutrients between ground vegetation and planted *Picea abies*. *New Zeal J For* 1996;**26**:99–117 https://www.scionresearch.com/__data/assets/pdf_file/0004/59557/NZJFS261-21996ORLANDER99-117.pdf.

OSF: Natural Resources Institute Finland. Finnish Statistical Yearbook of Forestry. *Silviculture Soil preparation, 2001–2020*. https://www.luke.fi/en/statistics/about-statistics/statistical-publications/finnish-statistical-yearbook-of-forestry (7 February 2023, date last accessed).

Raulo J, Rikala R. Initial development of Scots pine, Norway spruce and silver birch seedlings planted on a forestation site prepared in different ways. *Folia For* 1981;**462**:1–13. (In Finnish with English summary). http://urn.fi/URN:ISBN:951-40-0507-4.

Siddappa S, Marathe GK. What we know about plant arginases? *Plant Physiol Biochem* 2020;**156**:600–10. https://doi.org/10.1016/j.plaphy.2020.10.002.

Sikström U, Hjelm K, Hanssen KH. *et al.* Influence of mechanical site preparation on regeneration success of planted conifers in clearcuts in Fennoscandia – a review. *Silva Fennica* 2020;**54**:article id 10172.35. 10.14214/sf.10172.

Smolander A, Heiskanen J. Soil N and C transformations in two forest clear-cuts during three years after mounding and inverting. *Can J Soil Sci* 2007;**87**:251–8. https://doi.org/10.4141/S06-028.

Solvin, T., Fløistad, I.S. and Fjellstad, K. 2021 *Statistics: Forest Seeds and Plants in the Nordic Region*. Nordic Council of Ministers, The Nordic Genetic Resource Centre (Nord Gen), Alnarp, Sweden. 14. http://urn.kb.se/resolve?urn=urn:nbn:se:norden:org:diva-12242 (3 February 2023, date last accessed)

Thompson S. Some observations on the shoot growth of pine seedlings. *Can J For Res* 1976;**6**:341–7. https://doi.org/10.1139/x76-045.

Tonteri T, Hotanen J-P, Kuusipalo J. The Finnish forest site type approach: ordination and classification studies of mesic forest sites in southern Finland. *Vegetatio* 1990;**87**:85–98. https://doi.org/10.1007/BF00045658.

Wallertz K, Björklund N, Hjelm K. *et al.* Comparison of different site preparation techniques: quality of planting spots, seedlings growth and pine weevil feeding damage. *New For* 2018;**49**:705–22. https://doi.org/10.1007/s11056-018-9634-8.

von Wühlisch G, Muhs HJ. Influence of age on sylleptic and proleptic free growth of Norway spruce seedlings. *Silvae Genetica* 1986;**35**:42–8.

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