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Effects of cold-storage and planting date on subsequent growth, starch and nitrogen content in Scots pine (*Pinus sylvestris*) and Norway spruce (*Picea abies*) seedlings

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Abstract

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The phenology of *Pinus sylvestris* (L.) and *Picea abies* (L.) Karst. seedlings, stored during winter in a cold-store (−5°C) and outdoors, respectively, was studied after planting-out on different dates from May to end of July. Seasonal changes in nitrogen and starch content were followed during the summer of planting.

In pine and spruce seedlings planted from mid-June to end of July, negative effects on dry weight, shoot and needle growth, bud diameter, and frost hardiness were observed. These effects were most pronounced for cold-stored seedlings and still evident after three growing seasons.

Needle starch levels showed equal variations for early plantings, independently of storage treatment, but differences increased successively for later planting dates. Cold-stored seedlings showed, on all planting dates, a rapid increase in needle starch concentration. Seedlings stored outdoors had high levels of starch on early, and low levels on late planting dates. High levels of starch were found in the root system for seedlings planted in May. The results of the starch determinations were similar for both species.

The nitrogen concentration in the old needles of the two species was low after planting for both storage treatments. Late planting dates resulted in low levels of nitrogen in current needles.

Key words: Pinus sylvestris, Picea abies, cold-storage, planting date, growth, starch, nitrogen.

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Introduction

Since Swedish forest sites are spread over a wide geographical area and the spring planting period is short, commonly only May and June, storage of coniferous seedlings in cold-stores during winter has made the handling and distribution of planting stock much easier for the Swedish nurseries.

Many investigations have been made on changes in seedling quality during the period of storage, as well as on survival and growth after planting (for references see e.g. Hocking & Nyland, 1971; Ritchie & Dunlap, 1980). To improve the results of planting, there is, however, still a need for information about the effects of cold-storage.

The effect of cold storage on carbohydrates has been studied in many conifer species (Hellmers, 1962; Winjum, 1963; Ronco, 1971, 1973; McCracken, 1979). These studies have, however, mainly concentrated on the period of storage, and do not take into account what happens afterwards until the planted trees become well established.

Many investigations show starch accumulation in conifer needles in spring, before budbreak (e.g. Rutter, 1957; Krueger & Trappe, 1967; Pomeroy et al., 1970; Senser et al., 1975; Ericsson, 1979). A spring increase in the starch content in the living part of the stem bark and in the roots of conifers has also been shown (e.g. Loach & Little, 1973; Ford & Deans, 1977; Ericsson, 1980). Thus, if the intention is to understand the plart played by starch reserves in connection with the planting of conifer seedlings, it is also necessary to investigate whether the seedlings build up these reserves during the period immediately after planting.

Another important factor for successful plant establishment is probably the nutrient status of the seedlings after planting, and it therefore seems necessary to investigate whether outplanting causes nutrient deficiency in the seedlings. A deficiency in e.g. nitrogen immediately after planting may result in growth reductions for both above and belowground parts of the seedling for several years.

The present work focusses on the phenological development of cold-stored and outdoor-stored Scots Pine (*Pinus sylvestris* L.) and Norway spruce (*Picea abies* (L.) Karst.) seedlings after planting on different dates from the beginning of May to the end of July. Seasonal changes in nitrogen and starch content were also followed during the summer of planting.

Material and methods

The study was carried out at a commercial nursery in Nässja (lat. 60°15′ N, long. 16°50′ E), situated 80 km NW of Uppsala in central Sweden. The plant material consisted of Scots pine (*Pinus sylvestris* L.) and Norway spruce (*Picea abies* (L.) Karst.) seedlings. The pine seeds (lat. 60°20′ N, alt. 0–100 m) were collected in a seed orchard, while the spruce seeds (lat. 60° 20′ N, alt. 0–100 m) were collected from a natural stand. Both the seed orchard and the stand were about 20 km from the nursery.

Pine and spruce seeds were sown on 6 April and 21 April, respectively. The containers (Lännen Tehtaat OY, Finland) used were Paperpot 508 for the pines and Paperpot 408 for the spruces and the substrate was peat (Hasselfors K 11, Sweden). The seeds were germinated and grown in a plastic greenhouse during the first part of the summer and thereafter moved outdoors.

The seedlings were irrigated and fertilized according to the standard routines of the nursery. The fertilizer was distributed dissolved in the irrigation water and consisted of a complete mineral nutrient solution (Wallco, Sweden). Fertilization started two weeks after sowing and continued for 12 weeks. Accumulated nitrogen supply during this period of time was equivalent to 35 g m^{-2}.

The pine seedlings were moved outdoors on 8 June and the spruces on 10 July. Late in autumn, some of the seedlings were placed in tightly closed cardboard boxes and stored in a cold store at −5°C and 80% RH during the winter of 1978–1979. The pines and spruces were placed in the cold-store on 21–24 October and 10–14 November, respectively. The remainder of the material was left outdoors during the winter.

Seedlings of approximately equal shoot length were selected on each planting occasion, to minimize the variation within the plant material (cf. Fig. 2). Cold-stored seedlings were taken from storage one week before planting and kept at 20°C in darkness in open boxes. On each planting occasion ca. 350 seedlings from each storage treatment and species were planted in randomized blocks containing 7 rows. The spacing between the seedlings was 0.2 m and the distance between the rows 0.5 m.

Seedlings from both storage treatments were planted out on a total of seven occasions during the spring and summer of 1979. The first were planted on 7 May and thereafter every two weeks until 31 July. The area chosen for planting was in the close vicinity of the nursery, and had previously been used for the production of bare-root conifer seedlings. The soil is a fine sand.

Dead and damaged seedlings were recorded in the autumn of 1979 and 1980. The seedlings used for this purpose were the same as those which were intended for shoot length measurements (n=50). Injuries caused by animals were recorded separately.

Shoot length measurements were made from the soil surface to the top of the seedling (±0.5 cm). Measurements were made on the dates of planting and in the autumns of 1979, 1980 and 1981. On the different planting occasions in 1979 the number of seedlings was 50. In the autumn of the same year and in 1980, the number of measured seedlings was lower, owing to the exclusion of dead seedlings from the original 50 seedlings. Since 10 seedlings from each planting occasion and storage treatment were used for dry weight determination in the autumn of 1980, the number of seedlings was even lower in the autumn of 1981 (See also below and Table 1).

In autumn of 1979, 25 seedlings were harvested from each planting group for *dry weight determination* of the aboveground part and the root system of the seedlings. The seedlings were divided into groups consisting of five seedlings, which were dried at 105°C for 20 h and weighed. Dry weight measurements in the autumn of 1980 were made on 10 seedlings taken from those which were used in 1979 for the length measurements. The determinations were restricted to the aboveground part, and each seedling was separately treated according to the method described above.

The dry matter content of the uppermost 2 cm of the top shoot was determined on seedlings on four occasions in the autumn of 1979 (18, 25 September; 2, 17 October). Cold-stored seedlings from each planting occasion were examined, but for those stored outdoors the study was restricted to three of the seven planting occasions. Samples were taken from 15 seedlings, and the samples were combined in subsamples, representing 5 seedlings each, before drying (105°C, 20 h) and weighing.

For *needle length measurements*, one needle from the middle of the current main shoot was sampled from 25 seedlings of each planting occasion and storage treatment in the autumn of 1979. In the autumn of 1980, one current needle from each of 10 seedlings was sampled. The seedlings were identical with those used for dry weight determination. During 1980, no samplings were made from the spruce seedlings, nor from the cold-stored pine seedlings planted out on 31 July. In the latter, current needles were missing as a result of damage.

Bud diameter was measured on 25 pine seedlings from each planting group at the end of the first summer after planting. The seedlings were those used for dry weight measurements. The determinations were made with vernier callipers, and were restricted to the pine seedlings, because of technical difficulties in obtaining an accurate value for the small spruce buds.

In the autumn of 1981, *stem diameter* was measured on the remaining seedlings intended for length measurements (n=30–40). Determinations on the pines were made on the middle of the internode extended in 1979. No measurements could be made on the cold-stored seedlings planted on 31 July, since the 1979 internode was missing from most of those seedlings. The stem diameter of the spruces was measured at a level of 5 cm from the soil surface.

Needles intended for *starch and nitrogen analysis* were sampled during the growing season of 1979. Samples were taken from the one-year-old needles at the time of planting, then weekly until the beginning of August. No needles were sampled on the seventh planting occasion from seedlings stored outdoors, since no living needles were available. The current needles were sampled every week throughout the season, as soon as they had reached a length of about 10 mm. On each sampling occasion a total of 100–120 needles was taken from each planting group of seedlings. The needle samplings were evenly distributed among the seedlings of the experimental group. The number of planted seedlings per storage treatment decreased successively from 120 on the first planting occasion to 50 on the last occasion. This experimental design was chosen to provide uniform disturbance of the seedlings, independently of the numer of needles sampled during the experimental period.

The root system of the seedlings was analysed for starch and nitrogen on the date of planting (n=50), 4 (n=25) and 6 (n=10) weeks after planting, and finally on 15 October (n=25). Except for the first planting occasion, no root samples were taken from seedlings stored outdoors at the time of planting. Analyses of starch content in the roots of the spruces were made only on seedlings planted on the first occasion. Nitrogen analysis of the root systems was restricted to the pine seedlings.

The extraction and analysis procedures for the starch determination were those described by Hansen & Møller (1975), as modified by Ericsson (1979). Two parallel determinations were made from each sample, to check the reproducibility of the analysis procedure.

The Kjeldahl-nitrogen determinations were made according to the routines of the Faculty of Forestry, Swedish University of Agricultural Sciences, Umeå. Samples of 150–200 mg dry weight were used for the analyses. Since a separate study of the method showed a precision (sensu Reeve & Crozier, 1980) of 4.4%, only one determination was made per sample.

The meteorological data given in Figure 1 are taken from the routine observations made in the nursery.

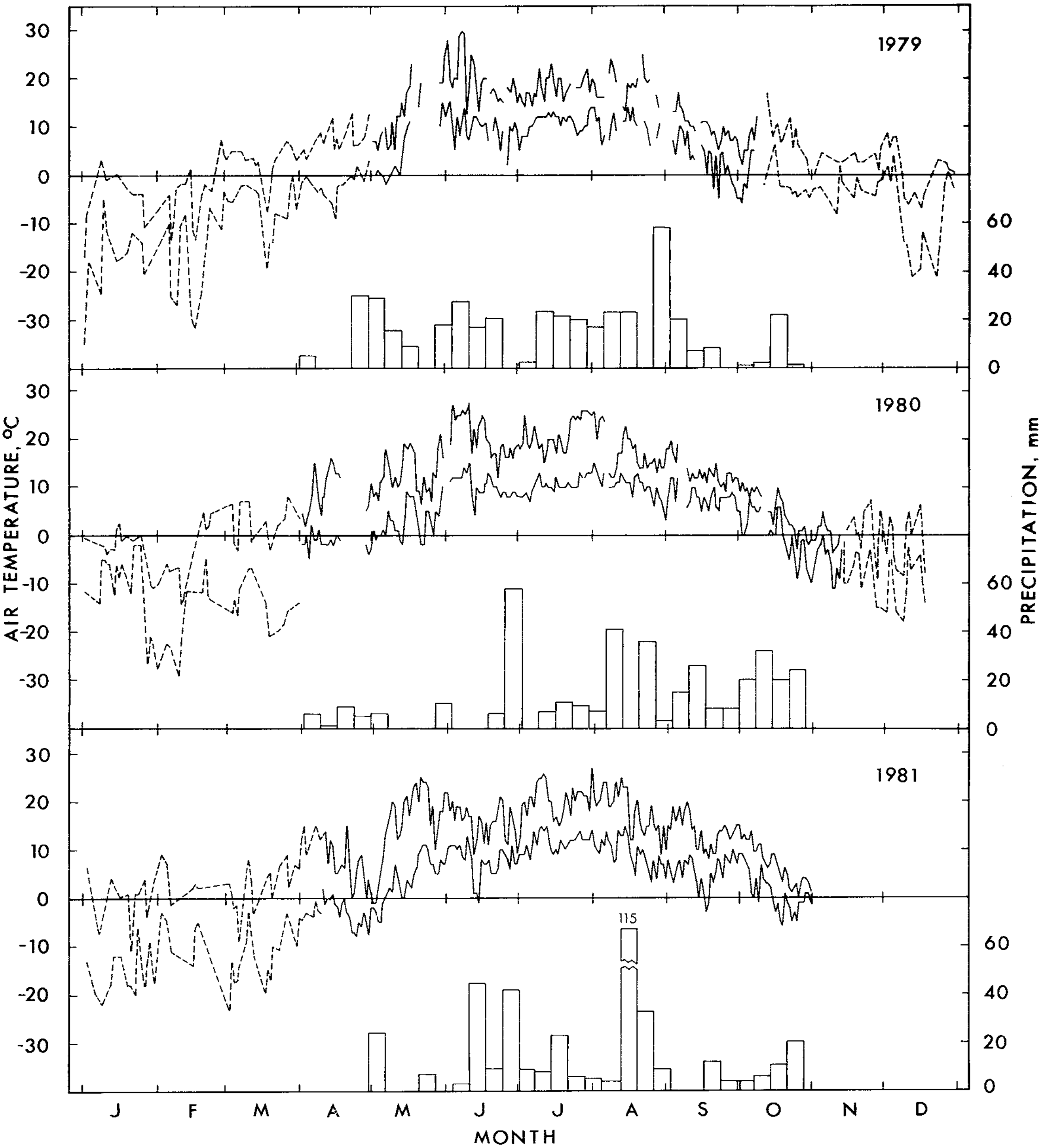

Fig. 1. Minimum and maximum air temperatures 1.5 m above ground and weekly rainfall (bars) during 1979–1981. The temperature observations were made irregularly during winter (– – – –), but every day during the summer months (——). The precipitation is shown for April–October only.

Results

Few Scots pine seedlings died during the summer of 1979 (Table 1). A few seedlings were damaged during the summer of 1979, but the damage was mainly caused by animals, and those seedlings are therefore excluded from the material when the effects on the different growth variables are discussed.

In August 1980, the number of dead pine seedlings was also low for all treatments (Tabel 1). It was found, however, that many of the pines had abnormal growth of the current shoot and other injuries or both. This was especially pronounced for the cold-stored seedlings, although some of the planting groups of the seedlings stored outdoors were also damaged.

Shoot length increment of the pine seedlings after planting was small during 1979 for all groups of seedlings (Fig. 2). Seedlings from the cold store and the two first-planted groups of seedlings stored outdoors grew about 5 cm. Seedlings stored outdoors which were planted late in the season had started their growth in the storage boxes, but ceased growth immediately after planting.

The total shoot length after two growing season, i. e. in the autumn of 1980, was approximately the

Table 1. *Per cent of dead and damaged* P. sylvestris *and* P. abies *seedlings in the autumn of 1979 and 1980 in relation to date of planting and storage treatment.*
A = dead seedlings, B = damaged seedlings. Each value is based on 50 seedlings. Values within parentheses represent damage caused by animals

Species ...	*P. sylvestris*								*P. abies*							
Storage treat ...	Outdoors				Coldstore				Outdoors				Coldstore			
Year ...	1979		1980		1979		1980		1979		1980		1979		1980	
Planting date 1979	A	B	A	B	A	B	A	B	A	B	A	B	A	B	A	B
7 May	0	0	0	0	2	4	2	14 (4)	0	0	0	12	0	0	0	8
21 May	0	4 (4)	0	26 (6)	0	0	0	10	0	2 (2)	0	20 (6)	0	0	0	18
5 June	0	0	0	8	2	0	2	12	0	0	0	12 (2)	0	2	0	8 (2)
18 June	0	0	0	10 (4)	0	0	0	14	0	0	0	16	2	2	2	24 (4)
2 July	0	0	0	0	0	0	0	12 (2)	0	4 (2)	0	8 (2)	0	0	2	24 (2)
16 July	0	0	0	0	0	0	0	14 (2)	4	0	6	10	0	10 (2)	4	60 (2)
30 July	0	8 (8)	2	12 (10)	0	2 (2)	0	20 (2)	0	6 (6)	8	78 (6)	4	2	36	64

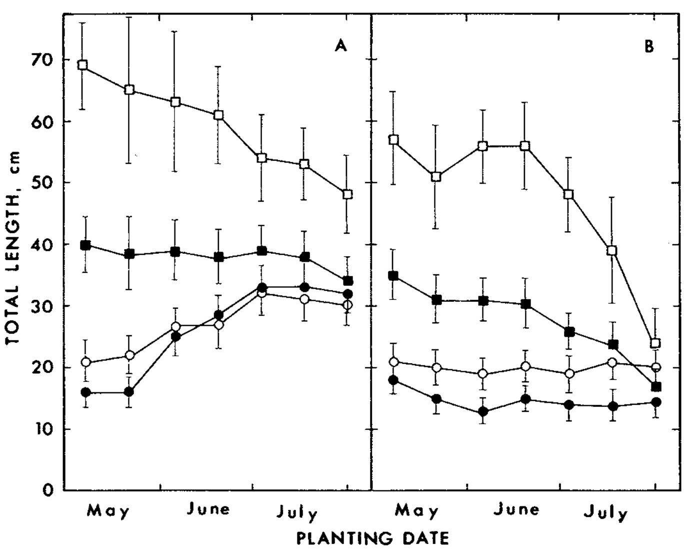

Fig. 2. The effect of planting date on the total shoot length of outdoor (A) and cold-stored (B) *P. sylvestris* seedlings. ● = length at planting in 1979 (n = 50), ○ = autumn 1979 (n = 47–50), ■ = autumn 1980 (n = 46–50), □ = autumn 1981 (n = 36–40). Vertical lines show ± standard deviation.

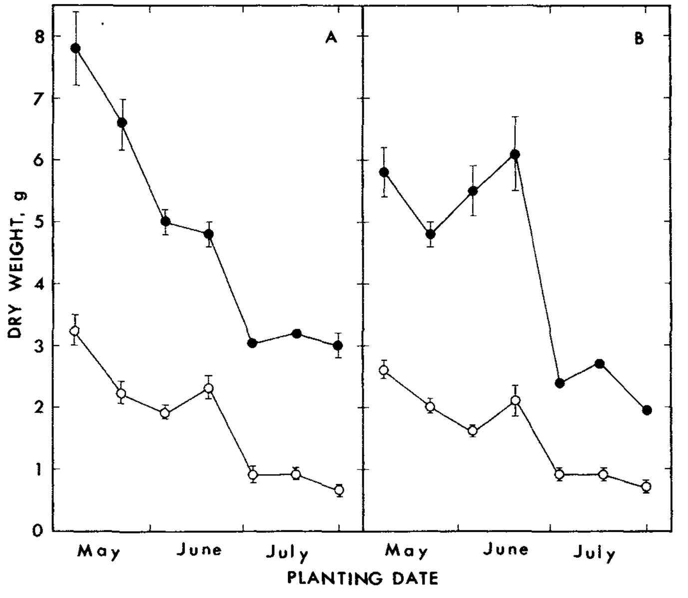

Fig. 3. The effect of planting date on the dry weight for outdoor (A) and cold-stored (B) *P. sylvestris* seedlings in autumn 1979. Planting was carried out in the summer of 1979. ● = aboveground part of the seedlings, ○ = root system. Total number of seedlings = 25, n = 5. Vertical lines show ± standard error.

same for all pine seedlings from the material stored outdoors, while shoot length of the cold-stored seedlings decreased the later they were planted (Fig. 2). The growth of the seedlings stored outdoors decreased successively from about 20 cm for the first planting occasions to about 2.5 cm for the latest one. Corresponding values for the cold-stored seedlings were 15 cm and no increase at all. There was thus a tendency that in the second summer cold-stored seedlings grew in general less than seedlings stored outdoors.

The measurements after three growing seasons showed that the pine seedlings stored outdoors were taller on all planting occasions than the corresponding cold-stored seedlings (Fig. 2). It was also evident that the negative effects of the late plantings were still severe after three growing seasons, for both types of storage treatment.

In the autumn of 1979 *dry weights of Scots pine seedlings* planted in early summer were greater than those of seedlings planted later in the summer (Fig. 3). This was true for both outdoor and cold-stored seedlings. The weights for the root system, as well as of the aboveground parts, were lower on nearly all occasions for the cold-stored seedlings than for the outdoor-stored ones.

The results of the dry weight measurements of the aboveground parts of the pine seedlings in 1980 showed the same tendencies as in the previous year (Fig. 4A), i. e. decreasing dry weights when planted late and the lowest values for the cold-stored seedlings.

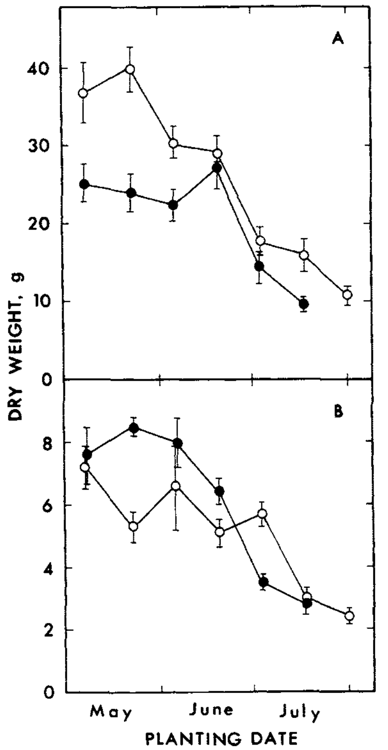

Fig. 4. The effect of planting date on the dry weight of the aboveground part of *P. sylvestris* (A) and *P. abies* (B) seedlings. Results from autumn 1980 after planting in 1979. ○ = outdoor seedlings, ● = cold-stored seedlings. Vertical lines show ± standard error, (n = 10).

In 1979, *the length of current needles in the Scots pines* was affected by the date of planting (Fig. 5A), i.e. late planting dates reduced elongation of the needles. Compared to seedlings stored outdoors, the cold-stored seedlings were found to have shorter needles on all planting occasions, with the exception of two occasions in June. Length of current needles in the autumn of 1980 was of the same order, independently of planting date.

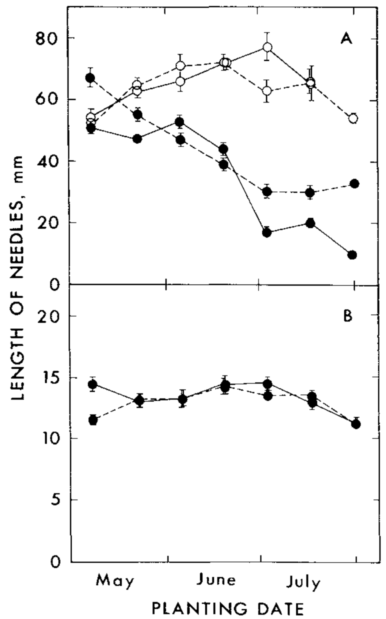

Fig. 5. The effect of planting date on the needle length of outdoor (– – –) and cold-stored (——) *P. sylvestris* (A) and *P. abies* (B) seedlings. The length measurements were made on current needles of 1979 (●, n = 25) and 1980 (○, n = 10), repectively. Planting was carried out in 1979. Vertical lines show ± standard error.

The number of dead and damaged Norway spruce seedlings in 1979 was approximately the same as that for the corresponding pines (Table 1); i.e. a few seedlings were dead or damaged, and no general trends in the pattern of damage could be observed. Many spruce seedlings were dead in August 1980, probably due to frost damage in the previous winter. Most dead and damaged seedlings belong to the last planting occasion, and the cold-stored seedlings were most affected.

The results of the *shoot length measurements on spruce* in 1979 were approximately the same as for the Scots pine (Fig. 6). Growth was very slight during the summer of planting for all cold-stored seedlings, and for the two first-planted groups of seedlings stored outdoors. Seedlings stored outdoors, which were planted during June and July, had already started shoot elongation at the time of planting, but no further growth could be observed for these after planting.

In August 1980, the total shoot lengths of the spruces stored outdoors were approximately the same for all seedlings. As a result of frost damage during winter, the total length of the cold-stored seedlings decreased when they were planted late in the previous summer.

The shoot growth of the spruce seedlings during 1981 was lowest for the last planting occasions, but the differences between planting dates seemed not to be as pronounced as that found for the pines.

As was the case with the pine seedlings, *the dry weights of the spruce* in the autumn of 1979 were low for the late planting dates (Fig. 7). The cold-stored seedlings of the first four plantings weighed more than the corresponding outdoor-stored seedlings,

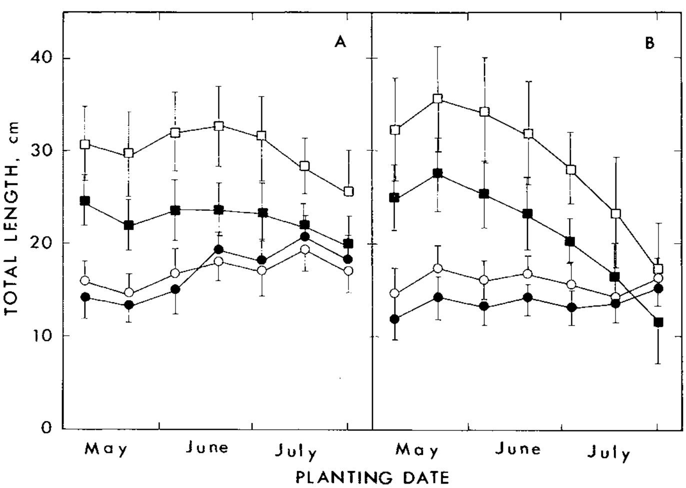

Fig. 6. The effect of planting date on the total shoot length of outdoor (A) and cold-stored (B) *P. abies* seedlings. ● = length at planting in 1979 (n = 50), ○ = autumn 1979 (n = 47–50), ■ = autumn 1980 (n = 31–50), □ = autumn 1981 (n = 31–40). Vertical lines show ± standard deviation.

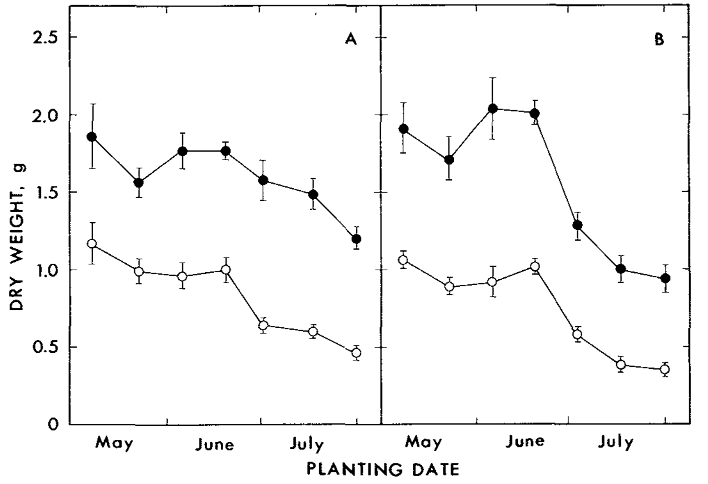

Fig. 7. The effect of planting date on the dry weight of outdoor (A) and cold-stored (B) *P. abies* seedlings in autumn 1979. Planting was carried out in the summer of 1979. ● = aboveground part of the seedlings, ○ = root system. Total number of seedlings = 25, n = 5. Vertical lines show ± standard error.

while the opposite result was found for the three latest plantings. This results was confirmed in the autumn of 1980 (Fig. 4B).

No pronounced effects of the storage treatments or of the planting dates were observed in 1979 on the *length of the current needles of the spruce* seedlings (Fig. 5B), and no further measurements were made in 1980.

The examination of *bud growth* was restricted to the pine seedlings, since the buds of the spruces were very small and thus difficult to measure accurately. Diameter measurments on the 1979 year's main bud showed that its development was affected by both the storage method during the previous winter and the date of planting (Fig. 8). No investigations of bud growth were made during 1980.

Stem diameter measurements were made in the autumn of 1981, and demonstrated that late planting dates in 1979 resulted in thin stems, even though three growing seasons had passed (Fig. 9). This result was found for both the pine and spruce seedlings.

The dry matter content of the two uppermost centimetres of the leading shoot demonstrated, that all

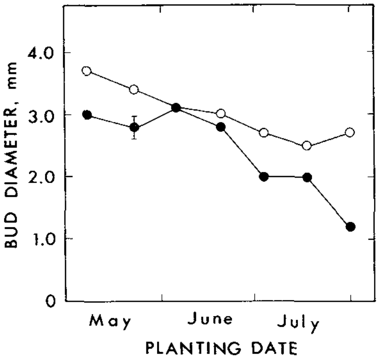

Fig. 8. The effect of planting date on the bud diameter of outdoor (○) and cold-stored (●) *P. sylvestris* seedlings. The measurements were made in autumn 1979. Planting was carried out in 1979. The vertical lines show ± standard errors >0.1 mm (n = 25).

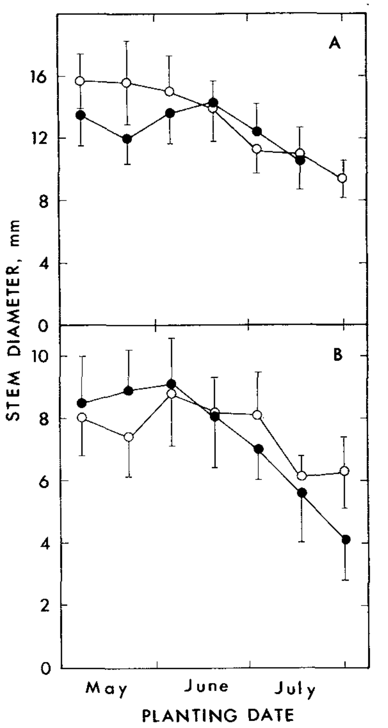

Fig. 9. The effect of planting date on the stem diameter of outdoor (○) and cold-stored (●) *P. sylvestris* (A) and *P. abies* (B) seedlings. The measurements were made in the autumn of 1981. Planting was carried out in 1979. Vertical lines show ± standard deviation (n = 31–40).

investigated seedlings stored outdoors seemed to be well frost-hardened in the middle of September, according to dry matter limit values found by Rosvall-Åhnebrink (pers. commun.) for one-year-old seedlings of pine (34%) and spruce (36%), respectively (Fig. 10). The cold-stored seedlings showed low dry matter values on both sampling occasions in September, and the last 3–4 planting groups never reached the recommended values during the period of investigation. The assumption that the dry matter content was a good indicator of the frost-hardiness level of the seedlings was partly supported by the results shown in Table 1.

Seasonal variation of starch concentration in Scots pine. Starch content during 1979 in the one-year-old (primary) needles of the different planting groups of Scots pine is shown in Figure 11. The levels on the first planting occasion were about 5 and 20% of dry weight for the cold-stored and outdoor seedlings, respectively. After approximately one month, the shapes of the curves for the two treatments were almost indentical. Results were similar for the second planting as well as for the third, except that in the last case the amount of starch never reached the same levels as for the previous two.

When the sampling occasions for the fourth and fifth planting groups were compared, the seasonal changes in starch content differed increasingly between the two types of storage treatment. On the last two planting dates this trend would probably have continued, but since almost no green primary needles were available on the outdoor seedlings, few values can be used for comparison.

In summary, the results show that the starch content of the needles increased from about 5% of dry weight to a maximum value of about 24% in the cold-stored pine seedlings, independently of the date of planting. The outdoor seedlings were found to have relatively high levels on the planting occasions in the beginning of the season, and low values at late planting dates. The variations in starch content during the first growing season showed the same general pattern for the early plantings, independently of storage treatment, but differed according to how late in the season planting was carried out.

The analysis of the starch content in the root system was restricted to a few occasions only, because of the destructive sampling method and limited amounts of material. The results showed that at the first sampling for the first planting occasion, the root system of the outdoor pines had a starch level of about 18% of dry weight (Fig. 12). After one month the level was still 18%, but decreased thereafter during the next two weeks to 10%. The cold-stored seedlings were found to have a starch concentration of about 6% on the first planting date. The content inreased during the next month to 12% and decreased therafter again to 6%.

Values are missing for the outdoor seedlings for the time of planting for the rest of the planting occasions, but the results after one month demonstrated that the roots for these seedlings never reached starch levels of the same order as those found on the first planting occasion.

The starch content was approximately 6% at the date of planting, independently of the storage time, for seedlings taken from cold-storage, and increased thereafter during the first four weeks. With the exception of the fourth and fifth planting occasions, the amount decreased for all groups during the following two weeks.

At the sampling in October, starch reserves in the

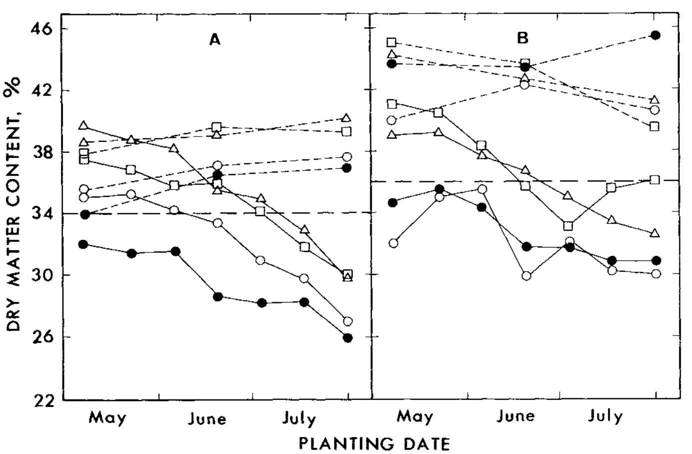

Fig. 10. The effect of planting date on the dry matter content of the uppermost 2 cm of the leading shoot of outdoor (---) and cold-stored (——) *P. sylvestris* (A) and *P. abies* (B) seedlings. Planting was carried out in 1979. ● = 790918, ○ = 790925, △ = 791002, □ = 791017. Each point represents the mean of 15 seedlings divided into 3 groups at the determinations. Limiting values for cold-hardiness according to Rosvall-Åhnebrink (pers. comm.) indicated by dashed line in the figure.

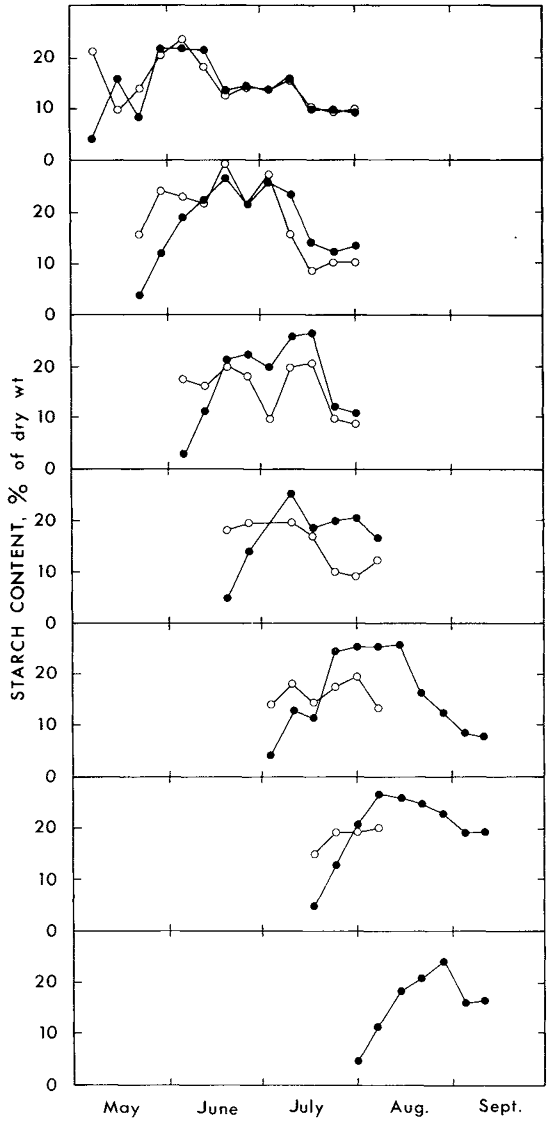

Fig. 11. Seasonal changes in starch content of 1-year-old (primary) needles of outdoor (○) and cold-stored (●) *P. sylvestris* seedlings during 1979. The first points on each curve represent the planting date. The values are mean of two determinations, which seldom differed by more than 1% of dry weight.

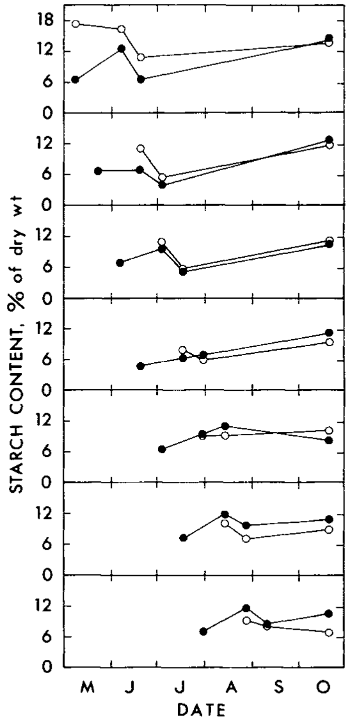

Fig. 12. Seasonal changes in starch content of the roots of outdoor (○) and cold-stored (●) *P. sylvestris* seedlings during 1979. The first points on each subdiagram represent the planting dates. Except for the first planting occasion, no determinations were made for outdoor seedlings at the time of planting. The values are means of two determinations, which seldom differed by more than 1% of dry weight.

root system showed decreasing concentrations the later the seedlings were planted in the summer. This result occurred for both storage treatments. The values were found to be 15 and 7% for the first and the last planting group of outdoor seedlings, respectively. Corresponding values for cold-stored seedlings were ca. 15 and 10%.

Seasonal variation of starch content in Norway spruce. The variation in the starch level of the needles of the spruces was only analysed for the first, third and fifth planting occasions (Fig. 13). The starch concentration was the same for all three groups of cold-stored seedlings on the different planting dates, i.e. about 5% of dry weight. In all three cases the level increased within a month to more than 30%. The outdoor seedlings showed high values on all three planting dates: 20, 32 and 33% of dry weight, respectively. After one month, the amount of starch was roughly at the same level for both types of storage treatment, independently of the date of planting, a result which differed from that obtained for the Scots pine seedlings (cf. Fig. 11).

The analysis of the root system of the spruce seedlings was restricted to the first planting occasion (Fig. 14). The starch contents were 6 and 19% of dry weight for the two experimental groups, with the lower value for the cold-stored seedlings. The amount increased for the cold-stored seedlings during the first month to approximately the same level as was recorded for the spruces stored outdoors. On the last two sampling occasions, the levels were the same for both storage treatments.

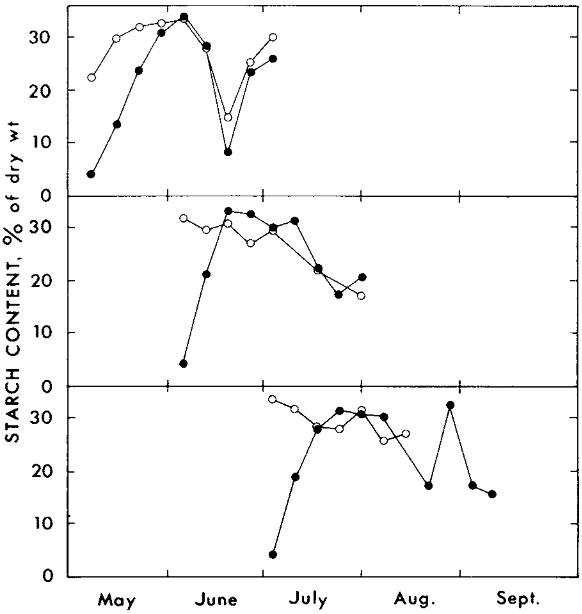

Fig. 13. Seasonal changes in starch content of 1-year-old needles of outdoor (○) and cold-stored (●) *P. abies* seedlings during 1979. The first points on each curve represent the planting dates. The values are means of two determinations, which seldom differed by more than 1% of dry weight.

Seasonal variation of nitrogen concentration. Fluctuations in the nitrogen level of the pine needles during 1979 were approximately the same for the first three planting occasions, regardless of storage treatment (Fig. 15). The concentrations in the one-year-old needles of these seedlings were found to be 1.0–1.3% of dry weight at the beginning of the experimental period, but decreased thereafter to about 0.75% at the end of June. In the latter part of July, the level increased again to ca. 1.25% of dry weight.

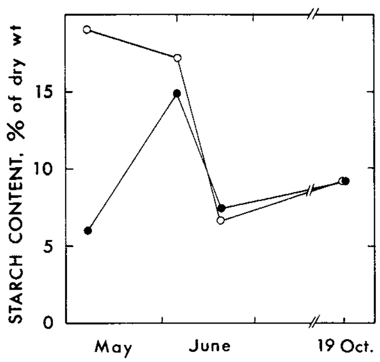

Fig. 14. Seasonal changes in starch content of the roots of outdoor (○) and cold-stored (●). *P. abies* seedlings during 1979. The first points on each curve represent the planting date. The values are means of two determinations, which seldom differed by more than 1% of dry weight.

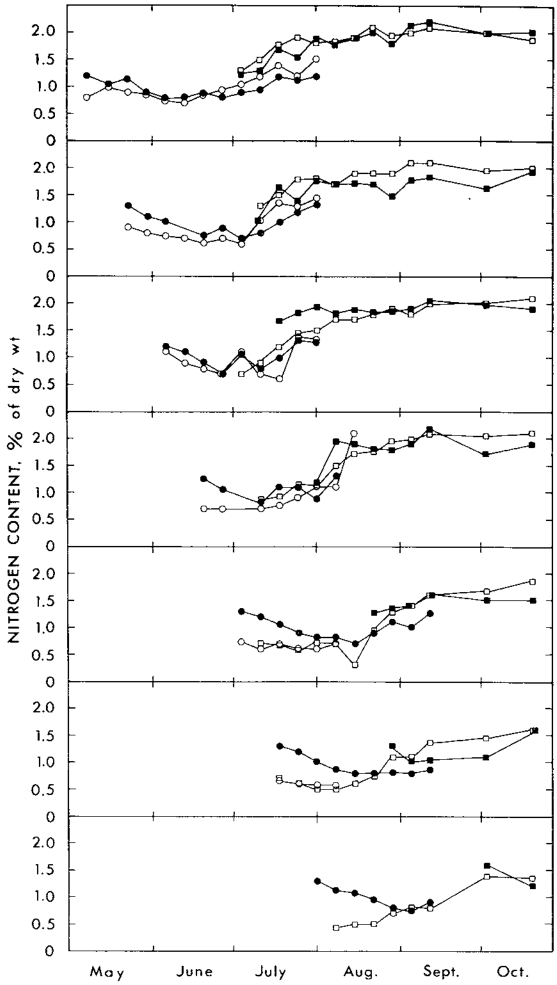

Fig. 15. The seasonal changes in total nitrogen content of 1-year-old (primary) and current (secondary) needles of outdoor (○, □) and cold-stored (●, ■) *P. sylvestris* seedlings during 1979. The first points on each subdiagram represent the planting dates, ○, ● = 1-year-old needles; □, ■ = current needles. Each point represents one determination.

The one-year-old needles of the outdoor pine seedlings of the fourth, fifth and sixth planting occasions were found to have values of 0.50–0.75% of dry weight on their planting dates. Except for the fourth planting occasion, when values reached about 1.1%, no further increase in nitrogen content could be detected during the period of sampling. No measurements were made on the one-year-old needles of the outdoor seedlings of the last planting date, for the same reason as in the case of the corresponding starch analysis.

The one-year-old needles of the cold-stored pine seedlings contained, independently of storage time, approximately the same amount of nitrogen (1.25%)

when they were planted. The concentration decreased to 0.75% within 6 weeks after planting and increased thereafter to ca. 1.25% for all groups, except for the last two planting occasions. The latter result is probably a consequence of too early termination of sampling.

The current needles of the different planting groups of pine showed low values in October when planting was late in the season (Fig. 15). Seedlings planted in May and June were found to have a concentration of about 2%, while seedling planted at the end of July showed a level of only 1.25% in the autumn. It was also evident that when seedlings were planted late in the season the nitrogen content of the current needles was very low during July, August and September.

Table 2. *The seasonal mean (±SD) nitrogen content in roots of outdoors and coldstored* P. sylvestris *seedlings during 1979 in relation to planting date. The determinations were made on the same seedlings as shown in Figure 12*

Planting date 1979	Outdoors N % of d.w.	n	Coldstore N % of d.w.	n
7 May	0.92±0.08	4	0.98±0.12	4
21 May	0.95±0.17	3	0.99±0.08	4
5 June	0.99±0.16	3	1.03±0.08	4
19 June	1.07±0.11	3	1.10±0.08	4
3 July	0.93±0.14	3	1.01±0.12	4
17 July	0.87±0.05	3	1.03±0.09	4
31 July	0.73±0.06	3	0.90±0.11	4

The nitrogen concentration of the root system of the pine seedlings was investigated on the same seedlings as were used for the corresponding starch analysis (cf. Fig. 12). The determinations showed very small fluctuations in nitrogen content during the experimental period, as well as small differences between the two storage treatments (Table 2).

The variation in nitrogen content of the one-year-old needles of the spruce seedlings showed roughly the same general pattern as for the corresponding pines (Fig. 16).

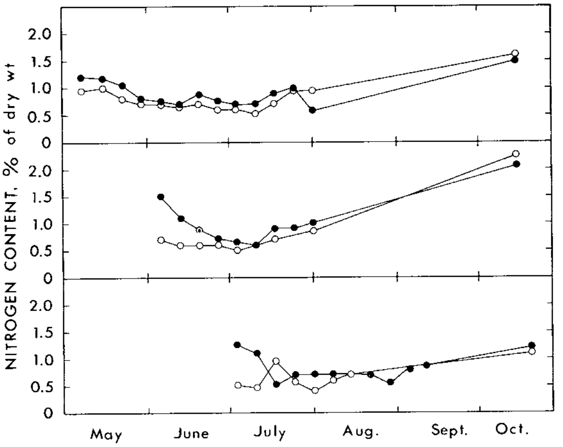

Fig. 16. The seasonal changes in total nitrogen content of 1-year-old needles of outdoor (○) and cold-stored (●) *P. abies* seedlings during 1979. The first points on each curve represent the planting date. Each point represents one determination.

Discussion

The general pattern of variations in the growth variables measured was, with a few exceptions, the same for the two species investigated. The same was found for the variations in starch and nitrogen content. The discussion will therefore concentrate primarily on the Scots pine seedlings, which were the main species in this study, and some differences in the reaction pattern of the two species will be indicated.

Both planting date and storage method affected the shoot growth of the pine seedlings. Growth was depressed for at least three years after planting (Fig. 2), when seedlings were planted from mid-June to the end of July. Cold-storage aggravated the negative effect of late planting.

Since shoot elongation is strongly dependent on the water potential of the plant (e.g. Kaufmann 1968), the slight growth during the first summer after planting is probably a result of disturbance in the water uptake, caused by damage to the root system at planting. The results show that when the seedlings were disturbed before the period of shoot elongation, final shoot lengths were approximately the same, independently of planting date. This is illustrated by the values obtained for the outdoor pine seedlings planted before budbreak (Fig. 2A), and for all planted cold-stored seedlings (Fig. 2B).

The shoot lengths did not correlate well with the total biomass. This is exemplified by the shoot lengths and dry weights of the outdoor seedlings (Figs. 2A, 4A). Most of the negative correlation found is probably explained by the development of the needle biomass after planting. The needle length was shorter, the later in the season the seedlings were planted (Fig. 7A). This must be of decisive importance to the total dry weight of the shoot, since, as a result of the predetermined growth pattern of Scots pine (Kozlowski, 1971, passim), the number of needles should be the same for all seedlings.

The decreased needle elongation of the outdoor pines planted in the middle of the summer, is probably a result of water stress (cf. Kaufmann, 1968; Garrett & Zahner, 1971), in combination with decreased day length, the latter deciding the duration of needle growth (Ekberg et al., 1979). Thus, early spring planting of outdoor pine seedlings results in decreased shoot elongation, caused primarily by water stress, while needle development will be almost undisturbed. On the other hand, when seedlings are planted in the middle of the summer, the shoot will grow undisturbed in the storage boxes, but needle development will be depressed by water stress.

The effect of water stress on needle and shoot growth will be somewhat different for the cold-stored seedlings. For early planting dates the situation will be similar for both kinds of storage treatment, while late planting of cold-stored seedlings will affect both shoot and needle elongation rate (Figs. 2B, 7A), since these seedlings are in the same post bud-break development stage on all planting occasions.

Late planting dates and cold-storage were both negative factors for bud development (Fig. 8). The depressed bud growth of the outdoor pine seedling planted late in the season is probably a result of a general disturbance in the activity of the apical meristem, caused primarily by water and nutrient stresses (Clements, 1970; Sucoff, 1971; Pollard & Logan, 1979), at a time when bud growth is most intense.

Since the cold-stored pine seedlings were in the same phenological stage on all planting occasions, the disturbance of bud growth for these may not be of the same nature as for the corresponding outdoor seedlings. In addition to water and nutrient stress, the duration of the growing season, in combination with shortened day length must be of importance. This means that the most severe effects of late planting on bud development would occur in cold-stored seedlings, a conclusion which is also supported by the results of the present investigation (Fig. 8).

The determination of the dry matter content of the uppermost 2 cm of the shoots demonstrated that cold-stored pine seedlings were probably less cold-hardened than the corresponding outdoor seedlings (Fig. 10A). This was supported to some extent by the number of damaged seedlings found in the following summer (Table 1). The low dry weight values may be a result of the reduced length of the growing season, leading to a limited ability of the seedlings to mature before autumn and winter start. In contrast to the cold-stored seedlings, the outdoor seedlings will have normal daylengths during the summer. Thus any disturbance in the process of cold-hardening of the outdoor seedlings is primarily a result of factors other than a shortened summer.

While carbohydrates may be fundamental to the re-establishment of the physiological functions disturbed by planting, the most severe effect of planting on growth is probably caused by water stress. Such stress will also affect carbohydrate production, through decreased photosynthesis (Hallman et al., 1978). Determinations indicated, however, that carbohydrate availability in the aboveground parts of the seedlings was not limiting during the period immediately after planting (Fig. 11).

This finding was especially pronounced for the cold-stored seedlings, which showed, independently of planting date, a rapid increase in starch concentration of the needles during the first weeks after planting. It therefore seems that these seedlings, in this respect at least, reacted exactly as they should have been expected to do during a normal spring (Rutter, 1957; Krueger & Trappe, 1967; Little, 1970*a*, *b*, 1974; Ericsson, 1979).

In an earlier investigation on 20-year-old Scots pine trees, Ericsson (1980) found that the pattern of fluctuations in starch content during the summer months was roughly the same for the above and belowground parts of the tree. This synchronized fluctuation in the starch levels for needles and roots was not found in the present investigation, especially not for the cold-stored seedlings. The maximum levels of starch in the roots were also lower throughout than was found for the different diameter classes of the root system of the 20-year-old pines (Ericsson & Persson, 1980).

Low starch levels in the roots of the outdoor seedlings are perhaps not surprising, since, with the exception of the first two planting dates, the sink strength of the root system, as well as of the elongating shoot, must be high during June and July: i. e. currently produced photosynthates will be immediately consumed for growth and respiration, instead of being transformed into starch reserves.

The growth rate of the cold-stored pines was minimal on all planting dates, and it therefore seems reasonable to conclude that both the root system and the needles should build up starch reserves. The results do, however, demonstrate that high levels of starch could be detected only in the needles. One reason for this may be that the soil temperature was so high on the different planting occasions (although this was not measured), that the growth and respiration of the roots immediately after planting reached levels which consumed all currently translocated carbohydrates. If this explanation is correct, it must be concluded that the amount of translocated carbohydrates is a limiting factor for the building-up of the starch reserves in the roots and may also be limiting for the growth of the partly destroyed root system.

An alternative explanation may be, that when the seedlings are taken from cold-storage, the phloem functions poorly or not at all. Such a situation, combined with the sudden start of shoot growth at high temperatures, will strongly limit the accumulation of carbohydrates in the roots. This implies, that it is necessary for the pine seedlings to have a spring period with low temperatures, yet about 0°C, for the restoration of the translocation system and for the building-up of starch reserves in the root system.

It must, however, be emphasized that the present investigation does not provide sufficient evidence that carbohydrate availability really is limiting for the growth and survival of the seedlings on late planting dates. The number of samples taken from the root system was small, and the results from the starch determinations of the needles showed that no carbohydrate limitation existed on a whole-seedling basis. However, the results indicate that carbohydrate dynamics must be better understood, and possibly taken into consideration, when conifer seedlings are planted out.

The cold-stored seedlings were found to have a nitrogen concentration in the one-year-old primary needles of about 1.3% of the dry weight on all planting occasions (Fig. 15). This value is probably also equal to the concentration that would have been found in the needles when the seedlings were put in the cold-store. Since a nitrogen level in the range of 1.7%–2.5% seems to be optimal for Scots pine (Aronsson & Elowson, 1980), one must conclude that the seedlings were already suffering from nitrogen deficiency in the previous autumn. Nitrogen deficiency probably also occurred in the outdoor seedlings, since the cold-storage treatment started so late in the autumn that no additional nutrient uptake of importance was to be expected.

In comparison with the cold-stored pine seedlings, the outdoor seedlings showed in general the lowest nitrogen concentrations on the different planting occasions (Fig. 15). This is most probably a result of the remobilization of nitrogen reserves used for growth processes (cf. Kreuger, 1967; Smith et al., 1970; Meyer & Splittstoesser, 1971), which had started before planting.

Approximately one month after planting, the nitrogen content of the primary needles decreased to the same low levels (0.5%–0.75%) for both kinds of storage treatment. This low nitrogen concentration seems to be the absolute minimum level before needle death occurs. According to the results illustrated in Figure 14, it must therefore be concluded, that on the four latest planting dates at least, the nitrogen reserves of the outdoor seedlings had already been used up.

The nitrogen content of the current needles of the two storage treatments showed the same patterns of fluctuations for seedlings planted on the same date (Fig. 15). When seedlings were planted during May and June, the concentrations increased within some weeks to about 2.0% of dry weight. There was, however, a tendency that the later was the date of planting, the later was this nitrogen level reached. The current needles of the seedlings planted on the three latest occasions never reached a concentration of the same order as was found for the previous four planting dates. Thus, it seems reasonable to conclude that seedlings planted in the middle of the summer will suffer from nitrogen deficiency during the second part of their first growing season.

The results discussed above show that the nitrogen level of the seedlings during the summer of outplanting varied strongly with planting date. It is therefore not surprising that the phenological development differed for the planting groups, although it must be borne in mind that the nitrogen status is only one of many factors important to growth. The nitrogen deficiency of the seedlings may thus be a result of a poorly established root system, which perhaps depends primarily on water stress and limited carbohydrate availability.

The most severe effects on the following years' growth were found for seedlings in which the development of the new needles and buds had been disturbed. The growth of the current needles and buds is most intense during July and August, respectively (Ericsson, 1979). Therefore, nitrogen availability during these months of the first summer after planting will probably be a factor which significantly contributes to decreased growth in subsequent years

(Clements, 1970; Pollard & Logan, 1979). Nitrogen deficiency during these two months should be most serious for cold-stored seedlings planted late in the season, since, as has already been mentioned, the decreased length of the summer also limits normal development of the seedlings.

It is also evident that in the autumn, the levels of starch in the root-system and of nitrogen in the needles, were highest for seedlings planted at the beginning of summer. This may also be important for rapid growth during subsequent years (Glerum & Balatinecz, 1980).

As has been mentioned, the results obtained for Norway spruce seedlings were mainly the same as those for the Scots pines. The overall impression is, however, that the negative effects of cold-storage on some of the growth variables were less evident for spruce than for pine.

One pronounced difference between the two species was the effect of late planting dates and cold-storage on needle length (Fig. 7). In contrast to the results for pine, all spruce seedlings had approximately the same needle length in the autumn after the first summer. This is probably explained by the differences in the needle growth strategy of the two conifers. The needle growth of spruce is primarily a result of cell elongation, in contrast to the active growth of the basal needle meristem of pine (Mirov 1967). This means that the final length of the spruce needles is dependent on daylength, in contrast to the situation in pine (Ekberg et al., 1979). This may contribute to the equal needle length for spruce, independently of the date of planting or of storage treatment.

Another difference between the two species in needle growth is that the needles of spruce elongate immediately after bud-break, i. e. during the elongation of the shoot axis (Cannell et al., 1976). This growth pattern may increase the possibilities for normal needle lenght growth of the spruce seedlings, since the period of needle elongation will be longer than that for the pine seedlings.

The determinations of dry matter content of the uppermost 2 cm of the shoot showed approximately the same trends for the two species investigated (Figure 10), indicating decreased cold-hardiness of cold-stored seedlings the later they were planted out. These findings were partly supported by the distribution of damaged and dead seedlings in the following summer (Table 1). The usefulness of measurements of dry matter content for estimating the cold-hardiness stage of the seedlings (Rosvall-Åhnebrink, 1977) is, however, limited, since e. g. outdoor-stored spruces planted on the last occasion showed both high dry weight values in autumn and much damage in the following summer. The results of the present investigation do, however, indicate that the development of cold-hardiness in spruce was more disturbed by planting than was that of the corresponding pine seedlings. This disturbance was most pronounced for late planting dates, and seemed also to be more severe for cold-stored seedlings.

The discussion above has mainly concentrated on different planting dates and the effect on seedling development in relation to the first summer of establishment. It is, however, necessary to point out that, in the case of cold-storage, the duration of the storage period as such may also contribute to decreased vigour of the planting stock (Ritchie & Dunlap, 1980). Finally, it must be emphasized that the results of the present study do not prove that cold-storage in general decreases the quality of seedlings as compared with outdoor seedlings. It is possible, for instance, that the winter of storage was extremely good for the outdoor seedlings, and that the summer of planting also favoured such seedlings.

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