Root Electrolyte Leakage and Root Growth Potential as Indicators of Spruce and Larch Establishment

H. M. McKay

McKay, H.M. 1998. Root electrolyte leakage and root growth potential as indicators of spruce and larch establishment. Silva Fennica 32(3): 241–252.

The relationship between the condition of bare-rooted 2-year-old seedlings of Sitka spruce and larch at the time of planting and their survival and growth after 2 years was examined. Data were analysed for 2 experiments using seedlings lifted and stored at +1°C throughout the winter for planting in April and also for 2 experiments using seedlings planted directly on different dates without cold storage. Electrolyte leakage from the fine roots of spruce was closely correlated to survival following direct planting at different times from September to April and fine root leakage was a more accurate indicator of spruce performance than root growth potential. However the pattern of larch survival of directly planted stock was more closely related to root growth potential than to root leakage. When seedlings were cold-stored, root electrolyte leakage and root growth potential were modified during storage and following cold storage, the performance of both species was more closely related to root electrolyte leakage than root growth potential. These results are interpreted as meaning that successful establishment of bare-rooted seedlings requires a functional nursery root system that is capable of both supplying adequate water for a limited period immediately after transplanting and of producing roots to meet the seedling's increased water demand later in the growing season.

Keywords root growth potential, functional root

Author´s address Forest Commission Research Agency, Northern Research Station, Roslin, Midlothian, EH25 9SY, Scotland **E-mail** h.mckay@forestry.gov.uk

Accepted 30 June 1998

1 Introduction

There are many ways of assessing the potential of tree seedlings to survive and grow well after transplanting from nursery to forest. Ritchie (1984) organised these into material attributes, which can be measured directly, and performance attributes, which are measures of the performance of whole seedlings when subjected to specified test conditions. Material attributes include bud dormancy, water status, mineral nutrition, carbohydrate status, morphology and levels of growth regulators and enzymes while performance attributes include vigour, root growth, frost hardiness and seedling temperature. Since then methods of assessing plant quality have been described and evaluated, for example Duryea (1985) and Rose et al. (1990).

Root growth potential (RGP), or root growth capacity, is one of the most commonly used tests; it is defined as the seedlings' ability to grow roots when placed in a favourable environment. Ritchie and Dunlap (1980) found that 85 % of the 26 papers they reviewed showed a positive correlation between RGP and field performance and Ritchie and Tanaka (1990) found that, of 12 studies published in the intervening years, 75 % showed a positive relationship.

The ability of cell membranes to control the rate of ion movement in and out of cells is used as a test of damage to a great range of tissue samples from seeds (Sahlén and Gjelsvik 1993) and bulbs (Bonnier et al. 1994) to roots (McKay 1994), needles (Burr et al. 1990) and stem (Dean et al. 1995) following exposure to a wide range of environmental conditions including frost (Green and Warrington 1978, van den Driessche 1976), desiccation (Martin et al. 1987) and high temperatures (Binder and Fielder 1995). The rate of electrolyte leakage from needles or shoots has been used to determine frost hardiness (eg Colombo and Hickie 1987, Murray et al. 1989). More recently, Bigras and Calmé (1993) concluded that it was the best test to estimate damage to the root system a short time after simulated freezing of black spruce seedlings (*Picea mariana* (Mill.) B.S.P.). Electrolyte leakage from the fine roots of Sitka spruce (*Picea sitchensis* (Bong.) Carr.), Douglas fir (*Pseudotsuga menziesii* (Mirb.) Franco), and Japanese larch (*Larix leptolepis* (Sieb. and Zucc.) Gord.) was highly correlated to both survival and height growth after 2 growing seasons (McKay 1992); bare-root seedlings had been cold-stored at +1°C for various durations and seedlings were tested at the end of the cold storage period.

This paper describes two cold-storage experiments and two experiments using directly-planted stock. In all these experiments seedlings were lifted at intervals from Wykeham Research Nursery during the winter and cold-stored (1°C) or directly planted within 24 h on second-rotation sites on the North York Moors. Root electrolyte leakage and RGP, measured on a subsample of plants, were related statistically and graphically to survival and growth after two years. Although a range of conifers was included in these experiments, this paper is restricted to Sitka spruce and larch (Japanese larch and its hybrid with European larch (*L. eurolepis*)) to contrast an evergreen and deciduous conifer. The objective of this paper was to investigate the relationship between seedling performance and the physiological indicators REL and RGP measured at the time of planting contrasting a deciduous and evergreen conifer and cold-stored and directly-planted stock.

2 Methods

2.1 Nursery Phase

Plants were raised at Wykeham Research Nursery, NE England (0°32'W, 54°16'N). The nursery is 215 m above sea level with an annual precipitation of 760 mm. Sitka spruce of Queen Charlotte Islands origin and Japanese larch of unknown seed origin were used for a cold-storage experiment in 1989–1990, both a cold-storage and a direct-planting experiment in 1990–91, and a direct-planting experiment in 1991–92. The second direct-planting experiment also included hybrid larch from seed collected from a seed orchard with equal numbers of Japanese and European larch clones. Two-year-old seedlings were raised in an open nursery using a regime of precision sowing, undercutting and wrenching (Mason 1994). In both years, seedlings were first undercut in late June or early July, side cut 2 weeks later and then wrenched at two-weekly intervals until early October.

In 1990, plants were lifted and planted within 2 days on 8 dates at two-week intervals from 4 September to 11 December. In the following year, there were 13 lifting and planting dates from 1 October to early April. Plants for cold-storage were lifted on 9 dates between 1 October 1989 and 28 February 1990 and on 8 dates between 13 November 1990 and 12 March 1991. In both cold-storage experiments, seedlings were removed from cold storage and planted in early

April along with seedlings freshly lifted in early April. On each lifting date, plants were carefully removed from 4 randomly located positions along each nursery bed. Plants were sealed in co-extruded black and white polythene bags. Directly-planted seedlings were planted within 24 h of lifting. Cold-storage plants were placed in the dark at 1°C. Plants for testing were sent overnight to the Northern Research Station.

Meteorological data (total monthly precipitation, mean monthly air temperature, maximum and minimum air temperature to occur in the month, and mean monthly soil temperature recorded at 9.00 am) were obtained from a nearby station (High Mowthorpe, 00°38'W, 56°06'N, 175 m altitude). Data are relevant to seedlings in both the nursery and in the planting sites.

2.2 Outplanting Experiments

2.2.1 Sites

The site used in 1990 (0°31'W, 54°16'N) had been clear felled in 1988–89 and ploughed with a double mould board plough to a depth of 45 cm and disturbed to a further 15 cm with a tine (D45 T60) in January 1990. The site was flat, sloping slightly to the south, with an elevation of 155 m and annual precipitation of 788 mm. An iron pan soil had developed over lower calcareous grit of jurassic age. The vegetation regrowth, mainly brambles (*Rubus fruticosa* agg L.), birch (*Betula pendula* Roth.) and oak (*Quercus robur* L.), was controlled by glyphosate and handweeding. Unground rock phosphate was added at 450 kg ha^{-1} in July 1991. The planting site used in 1991–92 had been clearfelled in 1989 following gale damage and ploughed (D45 T60) in August 1991. There was a 7° slope to the west with an elevation of 190 m. Iron pan soils had developed over a soft limestone lithology. Cultivation gave good vegetation control and no further control was necessary; no fertiliser was added.

2.2.2 Assessments

Height was measured at planting and after one and two growing seasons. Survival was assessed after one and two growing seasons in November to February. Spruce was considered alive if any green needles were present even if the leading shoot had died back. Larches were considered alive if the buds were well formed and the stem, when scratched, was green. Because of this assessment method, it is possible that trees classified as not surviving the first growing season may regrow in the second year and also that first year growth may be negative.

2.3 Physiological Tests

2.3.1 Root Growth Potential

The height and root collar diameter of each seedling were measured and the root system was sandwiched between moist peat contained in a long, 3-sided growth box and a clear acrylic panel which formed the fourth side following the method of Tabbush (1988). Each box was stacked at a slight angle against another box to exclude light and encourage root growth against the clear viewing panel. Seedlings were grown for 2 weeks at a temperature of 20°C, a relative humidity of 75 % and a photon flux density of 350 μmol m^{-2} s^{-1} for a 16 h day. The total number of new roots longer than 1 cm was counted for each seedling. At lifting, 20 replicates were used but, because of limited growth room space, only 16 replicates were used for seedlings coming out of cold-store.

2.3.2 Root Electrolyte Leakage

Electrolyte leakage from fine roots (ie < 2 mm diameter) was determined using the relative conductivity method of Wilner (1955, 1960) as modified in McKay (1992). Root material was washed in cold tap water to remove soil and rinsed in deionized water to remove surface ions. The central root mass of each plant was sampled, in this case by removing a 2 cm wide band of roots at a distance of approximately 8–10 cm from the root collar. Samples were taken at random from the band of roots. Sample fresh weight ranged from 100 to 500 mg. Individual samples were added to 28-mL universal glass bottles containing 16 mL distilled water of a known conductivity. The bottles were capped, shaken, and left at room temperature for 24 h. The bottles were shaken again, and the conductivity of the bathing solution was measured using a conductivity probe with in-built temperature compensation. Samples were killed by autoclaving at 110°C for 10 mins. When the samples had cooled to room temperature the total conductivity of each sample was measured. The 24 h conductivity was expressed as a percentage of the value after autoclaving, having first subtracted the distilled water conductivity value from both. At lifting, 15 replicates were used but after storage only 10 replicates were used because of the quantity of material to be tested.

2.4 Statistical Analysis

The relationship between performance (survival and height increment in the first and second years) and the physiological measurements (REL and RGP) was investigated separately for each experiment and species by fitting linear and (if necessary) quadratic functions. When both were necessary, the linear and quadratic functions were fitted in orthogonal form. The quadratic term then indicates the amount and direction of curvature in the relationships, while the coefficient of the linear term gives the amount and direction of the linear trend combined with the curvature.

3 Results

Climate prior to lifting is expected to affect the seedlings' cold hardiness and water and carbohydrate status when they are lifted for direct planting or cold storage. All three winters at a nearby meteorological station had a decline in mean air temperature from 12–13°C in September to 4°C in December with at least 20 mm rainfall per month (Table 1) so conditions were conducive to hardening. The lowest air temperatures were around –9°C which were unlikely to cause shoot damage to Sitka spruce (McKay and Mason 1991) or larch (McKay and Morgan in prep). Soil temperatures followed air temperatures and temperatures in the rooting zone were unlikely to cause damage (McKay 1994). However January and February 1990 were mild and soil temperatures were probably high enough to allow active root growth. Climate after planting is expected to affect the stress experienced by the seedling and may influence the relationship between indicators and performance within and between years. Low temperature per se was un-

Table 1. Total monthly precipitation (mm), mean monthly air temperature at 1 m in a Stevenson screen and soil (30 cm depth) temperature (°C), and the maximum and minimum temperature to occur in the month at High Mowthorpe. (NA = not available)

Month	1989–90					1990–91					1991–92				
	Temperature				Precipitation	Temperature				Precipitation	Temperature				Precipitation
	Mean Air	Max Air	Min Air	Mean Soil		Mean Air	Max Air	Min Air	Mean Soil		Mean Air	Max Air	Min Air	Mean Soil	
September	13.6	24.1	6.1	13.6	20	11.9	23.0	3.7	12.9	35	13.5	22.8	3.5	14.4	30
October	10.4	16.9	2.4	10.8	57	10.9	19.8	2.8	10.8	74	9.1	16.8	0.3	10.0	64
November	5.8	12.6	–1.6	7.0	39	5.9	14.3	–0.8	6.8	65	6.1	15.1	–3.4	NA	86
December	3.7	10.6	–3.6	4.6	75	3.6	10.8	–0.7	4.0	102	3.8	12.3	–9.1	4.4	32
January	5.1	13.3	–0.5	4.8	75	1.9	10.9	–3.2	2.1	49	3.0	11.4	–4.6	3.7	34
February	5.5	14.4	–1.6	4.8	68	0.8	11.9	–8.7	NA	87	4.5	11.9	–5.8	3.6	33
March	6.9	17.8	–4.6	5.9	17	7.1	17.9	–2.4	5.9	51	5.6	13.4	–1.2	5.6	85
April	6.9	20.4	–4.5	7.2	17	6.4	16.6	–1.8	7.0	34	7.4	16.6	–0.7	7.3	63
May	10.7	21.7	1.1	11.8	33	9.1	17.6	1.7	9.6	12	11.3	23.7	2.0	11.4	12
June	12.4	21.0	5.3	13.4	127	10.7	18.7	1.2	11.4	70	14.7	25.6	7.1	15.0	48
July	14.8	27.1	4.3	15.6	27	16.3	26.3	8.5	15.9	31	14.9	23.3	7.1	15.7	95
August	16.9	33.2	7.2	16.5	33	16.3	25.4	6.5	16.2	9	14.5	22.3	6.1	15.0	74

likely to cause damage (McKay and Mason 1991, McKay 1994, and McKay and Morgan in prep). However, in April 1990, the combination of –4.5°C and low rainfall may have lead to excessive water loss especially if they coincided with strong winds; this would apply to all cold storage treatments since they were planted out within a few days of one another. Conditions seemed favourable for all directly planted seedlings in September to December 1990 but it is possible that, during the following winter, seedlings planted in December, January and February may have been exposed to a stressful combination of low temperatures and low rainfall leading to excessive water loss.

3.1 Direct Planting

3.1.1 Sitka Spruce and Japanese Larch Planted between Early September and Mid- December, 1990

Sitka spruce survival was closely and negatively related to REL; non-linear regressions between REL and first and second year survival explained 98 and 84 % of the variation with P < 0.001 and 0.004 respectively (Table 2). Survival was not significantly related to RGP over this period. Height growth in general was not closely related to either indicator.

Japanese larch survival and height growth were closely related to REL, especially when non-linear relationships were used; REL accounted for 87 % (P = 0.002) and 69 % (P = 0.024) of the variation in first and second year survival respectively (Table 2). REL was negatively related to survival and first year height growth but was positively related to growth in the second year and over the 2 years after planting. Survival and first-year growth were not related to RGP over this period though second-year and total height growth were positively related to RGP, which explained 75 % of the variation in total height growth. Although there was no significant statistical relationship between larch survival and RGP, there were some similarities in their patterns (see Fig. 1).

3.1.2 Sitka Spruce, Japanese Larch and Hybrid Larch Planted between Late September 1991 and Early April 1992

Growth of Sitka spruce was not related to either REL or RGP but survival was related to REL especially when non-linear regressions were used and to RGP when non-linear regression were

Table 2. The significance and correlation coefficients of the relationship between physiological indicators (REL and RGP) measured at the time of planting and the performance of stock planted without storage during 1990. The significance and correlation coefficients of quadratic relationships are given only where they were more highly significant than linear regressions.

	Sitka spruce				Japanese larch			
	REL		RGP		REL		RGP	
	Linear	Quadratic	Linear	Quadratic	Linear	Quadratic	Linear	Quadratic
Year 1								
Survival	.078, 33.5	<.001, 98.0	ns		.031, 49.7	.002, 87.3	ns	
Height increment	ns		ns		.030, 50.2	.006, 81.6	ns	
Height increment %	ns		ns		.031, 49.6	.005, 82.9	ns	
Year 2								
Survival	ns	.004, 84.0	ns		ns	.024, 68.7	ns	
Height increment	ns		ns		.014, 60.5	.013, 75.5	ns	.020, 68.5
Height increment %	.019, 56.3		ns		<.001, 85.3	<.001, 90.0	ns	
Total height increment	ns		ns		ns		.011, 63.8	.010, 74.9
Total height increment %	ns		ns	.040, 60.7	.044, 44.3	.024, 68.7	.041, 45.4	.030, 64.2

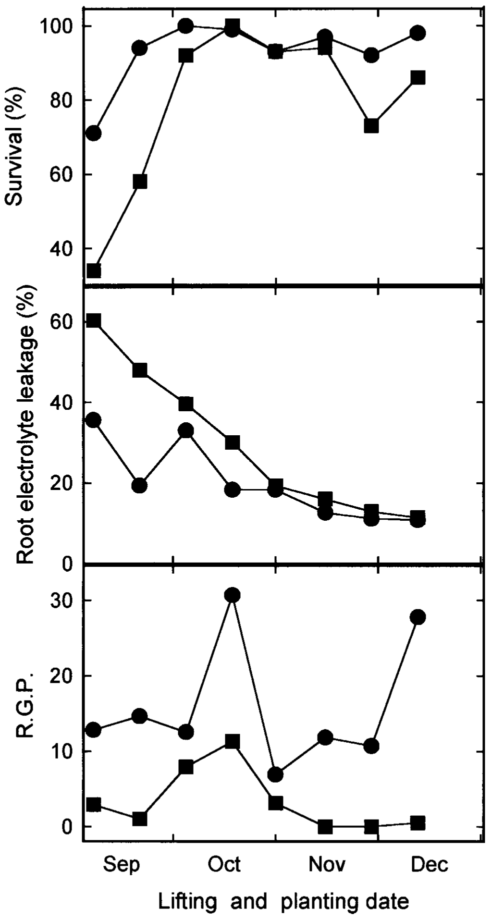

Fig. 1. The mean survival after 2 growing seasons, root electrolyte leakage and root growth potential measured at the time of planting of Sitka spruce (●) and Japanese larch (■) planted without storage in 1990.

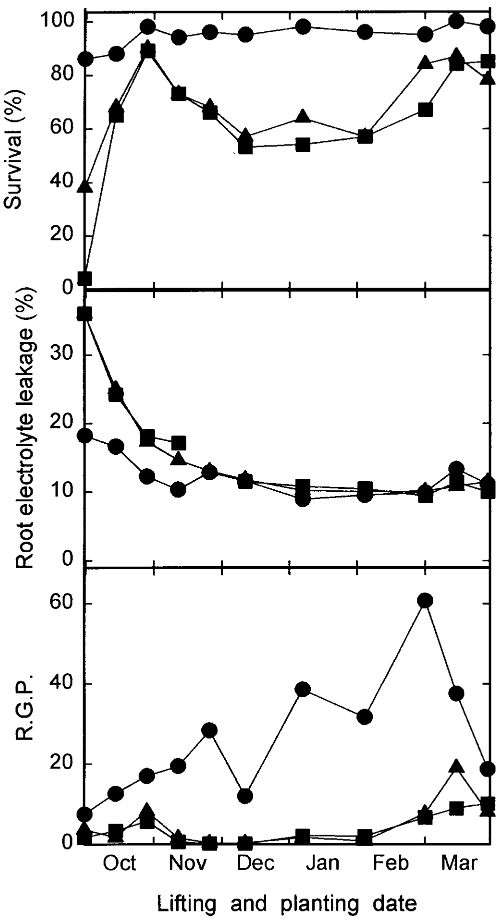

Fig. 2. The mean survival after 2 growing seasons, root electrolyte leakage and root growth potential measured at the time of planting of Sitka spruce (●), Japanese larch (■), and hybrid larch (▲) planted without storage in 1991–92.

used (Table 3). Over the whole planting season REL explained more of the variation in second year survival than RGP (67 vs 57 % respectively) and the pattern of REL resembled the survival pattern more closely than RGP (Fig. 2).

Growth of Japanese larch was not significantly related to either REL or RGP. Survival was closely related to REL especially the non-linear relationship which explained 65 % of the variation in survival after 2 years. Hybrid larch survival or growth were not significantly related to REL. RGP explained some of the variation in height growth in the first year and in second year survival. The pattern of larch survival reassembled the pattern of RGP. Both had peaks in autumn and spring with lower values during midwinter.

Table 3. The significance and correlation coefficients of the relationship between physiological indicators (REL and RGP) measured at the time of planting and the performance of stock planted without storage during 1991–92. The significance and correlation coefficients of quadratic relationships are given only where they were more highly significant than linear regressions.

	Sitka spruce				Japanese larch				Hybrid larch			
	REL		RGP		REL		RGP		REL		RGP	
	Linear	Quadratic	Linear	Quadratic	Linear	Quadratic	Linear	Quadratic	Linear	Quadratic	Linear	Quadratic
Year 1												
Survival	.013, 45.9	.003, 71.5	ns	.004, 68.4	.038, 33.0	.008, 62.2	ns		ns		ns	
Height increment	ns		ns		ns		ns		.033, 34.8		.49, 29.4	.034, 46.2
Height increment %	ns		ns		ns		ns		ns		.047, 30.1	.025, 50.4
Year 2												
Survival	.010, 49.2	.005, 67.0	ns	.013, 57.4	.049, 29.3	.006, 64.6	ns		ns		.038, 33.0	
Height increment	ns		ns		ns		ns		ns		ns	
Height increment %	ns		ns		ns		ns		ns		ns	
Total height increment	ns		ns		ns		.038, 33.0		.038, 33.0		ns	
Total height increment %	ns		ns		ns		ns		ns		ns	

3.2 Cold Storage of Sitka Spruce and Japanese Larch

3.2.1 Storage at +1°C on Different Dates from Early October 1989 to Early March 1990 with Planting in Early April

Sitka spruce survival and height growth were significantly, negatively related to REL at the end of cold storage (Table 4 and Fig. 3). The relationship between survival and REL was particularly close. A non-linear relationship explained a higher proportion of the variation in survival and height increment than linear relationships. Survival and first-year height growth were not significantly related to RGP and RGP explained a slightly smaller proportion of the variation in total increment than REL. For these reasons, REL was a better indicator of Sitka spruce performance in this experiment.

The survival and first-year height growth of larch were significantly negatively related to REL at the end of storage (Table 4). Non-linear relationships explained more of the variation than linear regressions. Height growth in the second year and total 2-year increment when expressed as proportions of initial height were not generally influenced by REL. RGP was not related to survival or first-year growth. Although RGP was significantly related to second-year and total height increment it explained < 45 % of the variation. Consequently, REL was generally a better indication of Japanese larch performance after storage than RGP.

3.2.2 Storage at +1°C on Different Dates from Mid-November 1990 to Mid-March 1991 with Planting in Early April

Both species established well over these storage dates, for example the range in survival of Sitka spruce and Japanese larch was 88–100 % and 89–97 % respectively while the range in their total height increment was 51–64 cm and 62–87 cm respectively, and neither REL or RGP at planting was related to performance.

3.2.3 Changes in REL and RGP during Cold Storage

In 1989 the REL rates of both species lifted in October for cold storage increased during storage indicating deterioration (Fig. 3). The deterioration decreased with later storage dates and by storage dates in mid-winter there was no significant difference (P < 0.001) between REL before

Table 4. The significant and correlation coefficients of the relationship between physiological indicators (REL and RGP) measured at the time of planting and the performance of stock after storage at +1°C and planting in April 1990. The significance and correlation coefficients of quadratic relationships are given only where they were more highly significant.

	Sitka spruce				Japanese larch			
	REL		RGP		REL		RGP	
	Linear	Quadratic	Linear	Quadratic	Linear	Quadratic	Linear	Quadratic
Year 1								
Survival	<.001, 97.3	<.001, 98.3	ns		.001, 75.3	<.001, 89.4	ns	
Height increment	.002, 73.6		ns		.019, 50.9	.013, 68.8	ns	
Height increment %	.003, 70.0		ns		.015, 5.6	.009, 71.9	ns	
Year 2								
Survival	<.001, 97.3	<.001, 98.1	ns		.005, 65.8	<.001, 90.4	ns	
Height increment	ns		.036, 41.7		ns		.029, 44.8	
Height increment %	ns		ns		ns		ns	
Total height incremen	.028, 45.1	.014, 67.8	.039, 40.5		.028, 45.5	.018, 65.4	.041, 39.8	
Total height increment %	.037, 41.0	.021, 63.0	.037, 41.3		ns		.033, 43.1	

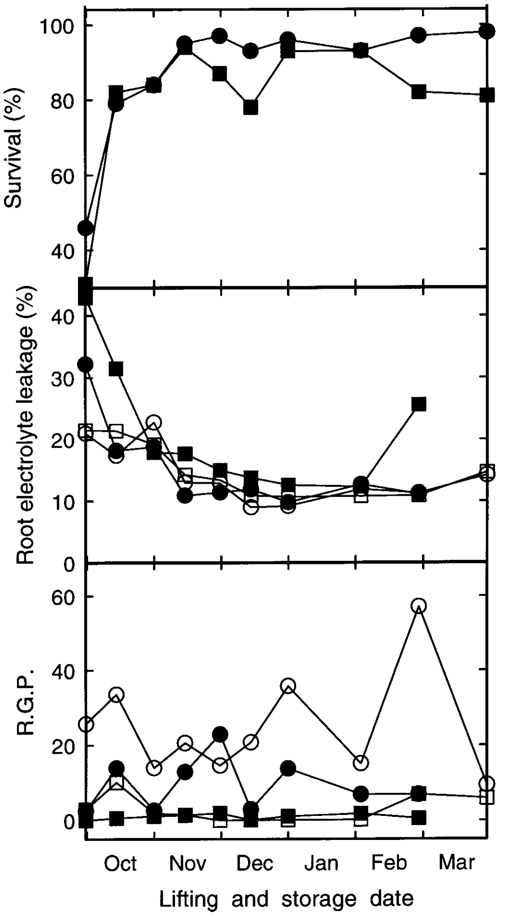

Fig. 3. The mean survival after 2 growing seasons and root electrolyte leakage and root growth potential measured after cold storage of Sitka spruce (●) and Japanese larch (■) planted in April 1990 after storage at +1°C on different dates in 1989–90. Mean root electrolyte leakage and root growth potential at lifting of Sitka spruce (○) and Japanese larch are also shown (□). Values before and after storage can be compared using the 95 % confidence intervals of 3.1 for REL and 3.9 for RGP.

and after storage in either 1989–90 or 1990–91. In fact, in some cases the leakage levels after storage were slightly less than those at lifting.

The RGP of spruce was lower after storage for almost all storage dates than at lifting (Fig. 3) but the patterns before and after storage were generally similar. In contrast, the RGP pattern of larch altered during storage. The peaks evident in stock lifted in autumn and spring declined while the low mid-winter values of directly lifted stock increased slightly. Consequently the double peak patterns of freshly lifted larch was not evident after cold-storage.

4 Discussion

The survival and in a few cases height growth of Sitka spruce was closely related to REL measured at planting in stock that was both directly planted and cold stored. This has been observed in other experiments with evergreen conifers which differed in the quality of their fine roots because of cold storage (McKay and Mason 1991, McKay 1993), and desiccation (McKay and White 1996). In one of the present experiments, survival was also significantly related to RGP but even in this instance, non-linear regressions of RGP explained a smaller proportion of the variation in second-year survival than REL (57 vs 67 %).

The present finding that survival of both larch and spruce after storage was more closely related to REL than RGP may be explained as follows. The primary cause of transplanting shock is water stress (Blake and Sutton 1987, Grossnickle 1988) and it is frequently stated that water stress after transplanting is overcome by root growth (Burdett 1988). Soil temperatures during the planting season are normally sub-optimal for root growth and may even be lower than the minimum for root growth which ranges from 2 to 5°C depending on species (Tabbush 1986, Coutts and Philipson 1987). Tabbush (1986) has shown that Sitka spruce at 8°C produced only 2 roots longer than 1 cm in 18 days and less than 20 roots in total after 10 days at 8°C. Therefore, in the period immediately after planting, the seedling must rely on water uptake via its existing root system. Electrolyte leakage is an index of the semi-permeable properties of cell membranes (Palta et al. 1977); if fine roots are capable of maintaining a low level of electrolyte leakage it is probable that they will also be capable of water uptake. As water demand increases during spring because of the increase in needle area and evapotranspiration, the original root system may not supply sufficient water; the transplanted seedling then relies on root growth. Root growth enhances water absorption by i) increasing the surface area of unsuberized roots ii) providing access to a larger soil volume and iii) improving root-soil contact (Burdett 1988). Both Sitka spruce (Philipson 1988) and larch, as demonstrated in RGP tests, are capable of producing some roots from stored carbohydrate so root growth is not entirely dependent on maintaining sufficient water levels to allow photosynthesis to proceed. Nevertheless, root growth is faster when photosynthesis is possible (Philipson 1988). The present cold storage results suggest firstly that, immediately after planting, the condition of the existing root system is of greater importance than the plant's potential to produce roots and secondly, provided root condition is adequate to support vital processes during the early stages of establishment, root growth in time influences survival and growth.

The survival and growth of Sitka spruce planted throughout the winter was also more closely related to REL than RGP. Clear seasonal patterns in REL were evident in the present experiments; leakage rates were high in early winter and decreased to a minimum for some period between mid-December and mid-January with a very gradual increase in spring. This general trend has been found in a range of evergreen conifers over 7 years (see McKay and Mason 1991, McKay 1993, McKay 1994 and unpublished data) and appears to reflect an intrinsic response of plants to the overwinter environment in the same way as RGP periodicity has been related to season (Ritchie and Dunlap 1980). The pattern is so consistent that it is unlikely to result from differences in damage in the same way as high leakage values of plants stored early or for long periods reflect cell membrane damage. I suggest instead that seasonal changes in the REL at lifting are due to some aspect of the molecular changes that plasma membranes undergo during cold acclimation. In most plants there are qualitative changes in membrane proteins (Yoshida and Uemura 1990) and, since proteins constitute the specific carriers and channels in the plasma membrane which mediate movement in and out of cells (Larsson et al. 1990), any such changes are likely to influence the leakage rate of undamaged cells. There are also many changes in lipid composition associated with freezing tolerance in particular an increase in the relative content of phospholipids (Yoshida and Uemura 1990) which may also affect membrane permeability. It is not clear how such protein and lipid changes might influence survival; it seems more probable that REL and survival are linked indirectly rather than casually related. It is possible that seedlings with high RELs in autumn have still active cell division and expansion and are more easily damaged than seedlings later in the year when RELs have decreased.

There were marked similarities in the pattern of survival and of RGP for freshly lifted larch. In mid-winter survival was apparently reduced by low RGP even though the existing root system was in good condition and should have been capable of taking up limited amounts of water. The present similarity between larch RGP and survival suggests that, in mid winter when root growth would be negligible, the nursery root system of larch was insufficient to supply water to meet the needs of larch in exposed, windy and comparatively dry sites. Water loss through the periderm depends on its structure and the density and permeability of the lenticels and in summer is only about 1 % of the potential evapotranspiration (Larcher 1995). However no figures are available for young larch in winter.

In mid winter, larch did not, even in favourable growth room conditions, produce roots even though the low REL indicated that the cell membranes had not been ruptured and were able to control ion movement. It seems unlikely that the double peak pattern could be caused by fluctuations in either current or stored carbohydrate. These points suggest that, in mid winter, larch roots are undamaged and probably have sufficient starch reserves but are not stimulated to produce roots. Initiation of new root primordia follows a stimulus originating in the shoot which is transported in the phloem (Ritchie and Dunlap 1980). There is some evidence the stimulus for root initiation in Douglas-fir (*Pseudotsuga menziesii* (Mirb.) Franco.) and red pine (*Pinus resinosa* Ait.) come mainly from the needles (see Ritchie and Dunlap 1980) but there is no information available for larch.

In conclusion, differences between larch and spruce suggest that the establishment of bare-rooted seedlings has 2 phases and that successful establishment requires a nursery root system that is both capable of supplying adequate water immediately after transplanting for a limited period and producing roots to meet the increased water demand later in the growing season.

Acknowledgements

The research foresters at Wykeham Field Station helped to establish and assess the experiment. Sandwich students Steven Moore, Ben Wilkes and Kirsty Venner helped with the laboratory assessments. Two anonymous reviewers made helpful comments on the manuscript.

References

Bigras, F.J. & Calmé, S. 1994. Viability tests for estimating root cold tolerance of black spruce seedlings. Can. J. For. Res. 24: 1039–1048.

Binder, W.D. & Fielder, P. 1995. Heat damage in boxed white spruce (Picea glauca [Moench.] Voss.) seedlings: its pre-planting detection and effect on field performance. New Forests 9: 237–259.

Blake, T.J. & Sutton, R.F. 1987. Variation in water relations of black spruce stock types planted in Ontario. Tree Physiology 3: 331–343.

Bonnier, F.J.M., Keurentjis, J. & van Tuyl, J.M. 1994. Ion leakage as a criterion for viability of lily bulb scales after storage at –2°C for 0.5, 1.5, and 2.5 years. HortScience 29: 1332–1334.

Burdett, A.N. 1990. Physiological processes in plantation establishment and the development of specification for forest planting stock. Can. J. For. Res. 20: 415–427.

Burr, K.E., Tinus, R.W., Wallner, S.J. & King, R.M. 1990. Comparison of three cold hardiness tests for conifer seedlings. Tree Physiology 6: 351–369.

Colombo, S.J. & Hickie, D.F. 1987. A one-day test for determining frost hardiness using the electrical conductivity technique. Ontario Tree Improvement and Forest Biomass Institute, Ontario Ministry of Natural Resources, Maple. Ont. For. Res. Note 45.

Coutts, M.P. & Philipson, J.J. 1987. Structure and physiology of Sitka spruce roots. Proceedings of the Royal Society of Edinburgh 93B: 131–144.

Deans, J.D., Billington, H.L. & Harvey, F.J. 1995. Assessment of frost damage to leafless stem tissues of Quercus petraea: A reappraisal of the method of relative conductivity. Forestry 68: 25–34.

Duryea, M.L. (ed.) 1985. Evaluating seedling quality: principles, procedures, and predictive abilities of major tests. Proceedings of the workshop held October 16–18, 1984. Forest Research Laboratory, Oregon State University, Corvallis.

Green, L.M. & Warrington, I.J. 1978. Assessment of frost damage in radiata pine seedlings using the diffusate conductivity technique. N.Z. J. For. Sci. 8: 344–350.

Grossnickle, S.C. 1988. Planting stress in newly planted jack pine and white spruce, 2. Changes in tissue water potential components. Tree Physiology 4: 85–97.

Larsson, C., Møler, I.M. & Widell, S. 1990. Introduction to the plant plasma membrane – its molecular composition and organization. In: C. Larsson, and I.M. Møler (eds): The Plant Plasma Membrane. Springer-Verlag, Berlin, Heidelberg.

Martin, V., Pallardy, S.G. & Bahari, Z.A. 1987. Dehydration tolerance of leaf tissues in six woody angiosperm species. Physiol. Plant. 69: 182–186.

McKay, H.M. 1992. Electrolyte leakage from fine roots of conifer seedlings: a rapid index of plant vitality following cold storage. Can. J. For. Res. 22: 1371–1377.

— 1993. Tolerance of conifer fine roots to cold storage. Can. J. For. Res. 23: 337–342.

— 1994. Frost hardiness and cold storage tolerance of the root system of Picea sitchensis, Pseudotsuga menziesii, Larix kaempferi and Pinus sylvestris bare-root seedlings. Scand. J. For. Res. 9: 203–213.

— & Mason, W.L. 1991. Physiological indicators of tolerance to cold storage in Sitka spruce and Douglas-fir seedlings. Can. J. For. Res. 21: 890–901.

Murray, M.B., Cape, J.M. & Fowler, D. 1989. Quantification of frost damage in plant tissues by rates of electrolyte leakage. New Phytol. 113: 307–311.

Palta, J.P., Levitt, J. & Stadelmann, E.J. 1977. Freezing injury in onion bulb cells. I. Evaluation of the conductivity method and analysis of ion and sugar efflux from injured cells. Plant Physol. 60: 393–397.

Philipson, J. 1988. Root growth in Sitka spruce and Douglas-fir transplants: dependence on the shoot and stored carbohydrates. Tree Physiol. 4: 101–108.

Ritchie, G.A. 1984. Assessing seedling quality. In: Forestry nursery manual: Production of bare-root seedlings. Eds. M.L. Duryea and T.D. Landis. Martinus Nijhoff/Dr W. Junk Publishers. The Hague/Boston/Lancaster. Forest Research Laboratory, Oregon State University, Corvallis.

Ritchie, G.A. & Dunlap, J.R. 1980. Root growth potential: its development and expression in forest tree seedlings. N.Z. J. For. Sci. 10: 218–248.

— & Tanaka, Y. 1990. Root growth potential and the target seedling. In: Target Seedling Symposium. Proceedings, Combined Meeting of the Western Nursery Associations. August 13–17, 1990. Roseburg, Oregon. Eds. R. Rose, S.J. Campbell and D. Landis. USDA Forest Service. General Technical Report RM-200.

Rose, R., Campbell, S.J. & Landis, T.D. (eds). 1990. Target seedling symposium: Proceedings, Combined Meeting of the Western Nursery Association. August 13–17, 1990. Roseburg, Oregon. USDA Forest Service. General Technical Report RM-200.

Sahlén, K. & Gjelsvik, S. 1993. Determination of Pinus sylvestris seed maturity using leachate conductivity measurements. Can. J. For. Res. 23: 864–870.

Tabbush, P.M. 1986. Rough handling, soil temperature, and root development in outplanted Sitka spruce and Douglas fir. Can. J. For. Res. 16: 1385–1388.

— 1988. Silvicultural systems for upland restocking. For. Comm. Bull., 76: Her Majesty's Stationery Office, London.

van den Driessche, R. 1976. Prediction of cold hardiness in Douglas-fir seedlings by index of injury and conductivity methods. Can. J. For. Res. 6: 511–515.

Wilner, J. 1955. Results of laboratory tests for winter hardiness of woody plants by electrolyte methods. Proc. Am. Hortic. Sci. 66: 93–99.

— 1960. Relative and absolute electrolyte conductance tests for frost hardiness of apple varieties. Can. J. Plant. Sci. 40: 630–637.

Yoshida, S. & Uemura, M. 1990. Responses of the plasma membrane to cold acclimation and freezing stress. In: C. Larsson, and I.M. Møller (eds): The Plant Plasma Membrane. Springer-Verlag, Berlin, Heidelberg.

Total of 32 references