THE EFFECTIVENESS OF REFLECTIVE TARPAULINS IN PROTECTING TREE SEEDLINGS AGAINST HEAT STRESS

by

ERNST INGVAR STJERNBERG

Registered Professional Forester, British Columbia

B.Sc.F., The University of Toronto, 1979

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Department of Forest Sciences

The University of British Columbia
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Abstract

Reflective tarpaulins are used extensively in western Canada to protect tree seedlings against solar radiation during on-site storage and transportation. This project determined heat transfer characteristics of new and used reflective tarpaulins, and a FIST (Fiberglass Insulated Seedling Transporter) canopy. Containerized white spruce (*Picea glauca* (Moench) Voss) seedlings in shipping boxes were stored under tarpaulins and in the FIST canopy for up to six days in June, 1992. An unprotected seedling box was used as the control. Average seedling box temperatures per each 15-minute period were recorded for 144 h. Solar irradiance and wind speed at the test site were recorded simultaneously with box temperatures. The electrical conductivity of needles was measured after storage. Seedlings were withdrawn daily for six days and outplanted in a nursery plot in a split-plot randomized complete block design with repeated measurements within each experimental unit. Root collar diameter, total height and survival were recorded for three growing seasons. Samples of the tarpaulins were also tested under controlled conditions for ability to resist heat transfer.

Seedling box temperatures ranged up to 35°C. Significant differences in 144-hour heat sums were found between: new and used tarpaulins; various used tarpaulins; tarpaulins and control; three new tarpaulins and the FIST canopy. Relative conductivities indicated no storage-induced damage to cell membranes. Survival after three years was 99.8%. Significant differences in growth were found but were more likely related to differences in soil nutrient differences within the plot. Storage temperature and length had no effect on growth and survival.

A laboratory experiment tested for storage-induced pre-conditioning effects. Controls and seedlings pre-conditioned 4 and 8 days for 3 h at 30°C, were heat stressed for 8 h or 48 h at 30, 35, and 40°C. Electrical conductivities of needles were measured. Seedlings were outplanted in two plots with a completely randomized design. Root collar diameter, total height and survival were recorded for two growing seasons.

No cell membrane damage was observed. Survival after two years was 99.5%. Evidence for a pre-conditioning effect is inconclusive. The 8-day pre-conditioning may have resulted in higher growth in seedlings heat stressed for 48 h.

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This thesis is dedicated to my family for their enduring patience and understanding during my studies.

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Introduction

There are between 200 and 300 million coniferous tree seedlings grown and planted in British Columbia every year (Canadian Council of Forest Ministers 1991). The vast majority of these seedlings are grown in containers, mostly of the Styroblock® type. Seedlings slated for spring planting are lifted in late fall, sorted and put into bundles which then are packed in polyethylene lined bags in cardboard boxes for overwinter freezer storage. In the spring these boxes are removed from the freezer and the seedlings allowed to thaw for a short period. They are then shipped, in temperature controlled semi-trailers or trucks, to a central cache at or near the planting area. Seedling boxes may be stored in the cache for up to a week depending on the progress of the planting programme. As needed, the seedling boxes are distributed from the central cache to smaller ones where individual planters or a group of planters get their supply of seedlings.

The principal factors that diminish seedling performance potential include root exposure, handling, storage conditions and length, shipping, temporary storage in the field, and planting (particularly micro-site selection) (Rietveld 1989). High temperatures can occur in the absence of other environmental stresses, although it is more common for high temperatures to accompany both atmospheric and edaphic water stress (Hällgren et al. 1991). Considerable evidence from research shows that seedlings should be stored at temperatures below 10°C to avoid the damaging effects of high temperature and to reduce the evaporative demand. Some of this evidence is presented in the literature review.

Reflective tarpaulins are used extensively in all planting operations in British Columbia and Alberta. They are used to cover the boxes in the central cache, for individual caches and also during local transportation of seedlings. Tarpaulins protect seedling boxes from solar radiation thus keeping seedlings cool until planted. Different brands and qualities of tarpaulins are commercially available. All are white on one side and 'silver' coloured on the other side. The tarpaulins are used with the white side facing upwards/outwards. However, they are folded, stretched, scuffed and exposed to the elements in normal use. This causes considerable wear and tear which can result in delamination of the materials, decreased reflectance of the white surface, and general deterioration of the tarpaulin material. When this happens, the tarpaulin's ability to resist heat transfer may be reduced and its effectiveness in protecting the seedlings from heat during field transportation and storage possibly decreased.

The B.C. Ministry of Forests' Silviculture Contracts specify that only reflective tarpaulins, in good repair and approved by the Ministry Officer, can be used to cover seedling boxes. Presently, there are no forest industry standards for the quality of the reflective tarpaulins to be used by planting contractors. As a result, some very poor quality, worn out tarpaulins are in use. Subjective decisions are made whether a tarpaulin is still useable but very little is known about how effective the tarpaulins are after one or more years of use in the field. This project was formulated to provide unbiased information on various types of tarpaulins, and to determine if there are any measurable effects on the seedlings as a result of storage under various types of reflective tarpaulins. The project was extended with a laboratory experiment after preliminary analysis of the results from the first year's seedling growth.

Literature Review

Heat stress in seedlings

Elevated temperatures affect seedlings in different ways. These have been explored to various extent in many studies. There are reports dealing with direct and indirect seedling damage as a result of excessively high temperatures (Baker 1929, Binder and Fielder 1995, Levitt 1980, Seidel 1986). Other reports explore the creation of food reserves and their use during high temperature exposures (Glerum and Balatinecz 1980, Puttonen 1980, Ritchie 1982). The physiology and survival of tree seedlings exposed to high temperatures are the topics of many studies (Hallman et al. 1978, Kauppi 1984, Mattsson 1986). This review looks at the effects of high temperature on tree seedlings from these perspectives. It also includes a discussion of the effects of storage and storage temperatures on seedling performance, and explores the effectiveness of various types of covers used to protect seedlings from elevated temperature. Some of the principles of heat transfer, and how these apply to reflective tarpaulins, are briefly reviewed.

Interest in heat tolerance of tree seedlings was high in the first half of this century since many of the problems involved with forest regeneration were considered to be caused by excessively high temperatures. However, during the last few decades there has been little research done on this subject (Colombo and Timmer 1992). An early report details an investigation into the problem of heat relations in seedlings to determine the importance of this factor, especially in the coniferous stands of the Pacific Coast (Baker 1929). Baker (1929) dealt with the subject of heat relations under six distinct headings: i) the nature of direct heat injury in the open; ii) reaction of

protoplasm of different species to high temperatures; iii) reaction of protoplasm of seedlings of different ages; iv) relation of internal temperatures to external air and soil temperatures; v) protective devices in conifers; and vi) effect of morphology and age on extent of temperature-induced injury.

Individual soils have vastly different heat properties owing to different colours, specific heat, radiation and conductivity (Munch 1914). These factors interact to make dark soils heat up more than light, loose soils heat more than compact, and dry soils heat more than wet and damp (Childs et al. 1985). As a result, dark loose moor soil is said to be very dangerous to seedlings; loose dry sand is about equally bad, followed by sandy loams, while light-colored, compact clay soils are relatively very cool (Baker 1929). The seedling receives heat by radiation, direct or reflected, and by conduction from the surface soil. Fatal internal temperatures are reached by different species under different external heat conditions, the degree of injury varying with age and with species (Alexandrov 1964, Baker 1929).

Soil surface temperatures on boreal planting sites often exceed 50°C and have a direct influence on coniferous seedling mortality (Koppenaal and Colombo 1988). Baker (1929) found that western American conifer seedlings (1-3 months old) are quickly killed when a soil temperature of about 54°C is reached. However, they can withstand a temperature only a few degrees lower for some time. Fatal temperatures are reached in seedlings only at the base of the stem which apparently was first suggested by Heinrich Mayr in 1909 (Baker 1929). This has been confirmed by more recent work (Koppenaal et al. 1991; Smith and Silen 1963). Direct heat injury under

field conditions involving the whole seedling has not been reported in the literature. Rather, it is the surface of the soil that becomes hot enough to cause direct heat injuries to the stem at, or just above, ground level (Baker 1929, Helgerson et al. 1992). This is attributed to the combined effect of direct radiation coupled with reflected heat (Hartley 1918).

Efforts to prevent such injuries through shading have been reported by several investigators (Childs et al. 1985, Helgerson 1990a and 1990b, Keijzer and Hermann 1966, Maguire 1955, Seidel 1986). Self-shading is the main morphological means of resisting heat damage (Helgerson 1990b). Some studies also suggest that shade-intolerant species require less shade than species that are more shade-tolerant although this trend is not clear (Helgerson 1990b). Maximum solar and sky radiation always occur at solar noon, whereas, maximum surface temperatures on level surfaces lag from one to two hours for all months except April and October when the earth is at mean distance from the sun (Maguire 1955). Heat damage, in the northern hemisphere, mostly occurs within mid-latitudes on flat sites, or on south-, southeast- or southwest-facing slopes (Helgerson 1990b). Heat damage tends to be associated with soils with low heat capacity and conductivity, a dry top layer, and surfaces that are covered with organic matter, or are dark or burned (Helgerson 1990b). These characteristics tend to be interrelated (Helgerson 1990b).

Smith and Silen (1963) related injuries to external symptoms, rather than to actual temperatures, by studying the anatomy of heat damaged Douglas-fir (*Pseudotsuga menziesii* (Mirb) Franco) seedlings. It was observed that anatomical changes within the hypocotyl were correlated both with external symptoms of heat damage and survival, which in turn was dependent primarily on depth to which the permanent injury extended. Smith and Silen (1963) outlined different levels of damage as indicated by, for example, a water-soaked appearance of the hypocotyl, a white

spot or streak appearing on the irradiated side of the hypocotyl, or a shallow constriction of the hypocotyl. The seedling cannot survive, regardless of age, when the damage reaches into the vascular cambium region or deeper. Damage appears to be due to progressive transfer of heat from the surface inwards, regardless of maturity of tissues present. Death results when external heat is present at high temperatures for a sufficient period of time to allow damage to extend inward to the cambium region. The claim that older seedlings are more heat resistant was not verified in this study and the authors believe this may have a physiological rather than a anatomical basis (Smith and Silen 1963). This was also suggested in later studies (Gauslaa 1984). However, Keijzer and Hermann (1966) observed that the older seedlings are, the less direct heat injuries they sustain, presumably because the succulence of exposed tissues decreases with age. This was also noted by Baker (1929). Losses of conifer seedlings because of excessive temperatures are greatest early in the growing season (Smith and Silen 1963; Seidel 1986). This suggests that the tolerance to higher soil temperatures increases as the growing season progresses. Tolerance may depend more on mechanical resistance to heat damage associated with increased size and morphological changes than on physiological changes (Baker 1929; Levitt 1980; Helgerson 1990b). There is also evidence that the length of exposure is a factor in the amount of damage sustained by seedlings (Baker 1929; Colombo and Timmer 1992; Seidel 1986). This particular aspect was also studied by Kauppi (1984). He tried to develop a dynamic injury model based on the fact that injury develops over time. The study by Kauppi (1984) is reviewed later under the heading 'storage effects'.

Seidel (1986) investigated the tolerance of seedlings of ponderosa pine (*Pinus ponderosa* Dougl. ex Laws), Douglas-fir, grand fir (*Abies grandis* (Dougl. ex D. Don) Lindl.) and Engelmann

spruce (*Picea engelmannii* Parry ex Engelm.) to high temperatures. The study objective was to estimate and compare the relationship between seedling survival and time of exposure to high temperature. A negative exponential relationship was found between time and temperature. This indicates that a short exposure to a high temperature will result in the same survival rate as a longer exposure to a lower temperature. This conclusion was also reached by other researchers (Colombo and Timmer 1992; Maguire 1955; Ingram et al. 1990). Seidel (1986) found that increasing the exposure time from 0 to 15 minutes dramatically reduced the temperature that the seedlings could tolerate. At longer exposures, over 60 minutes, the effect of time was less important in seedling mortality. Ponderosa pine was able to withstand the highest temperatures at any exposure time, while Engelmann spruce was the least tolerant. The difference between these two species was found to be 2 to 4°C. The abilities of Douglas-fir and grand fir to resist heat fell between the previous two species. The results of this study suggest that seedlings of many species suffer mortality from high temperatures within the approximate range of 48 to 68°C, depending upon the length of exposure (Seidel 1986). Soil temperatures of this magnitude have been recorded in many locations (Baker 1929). The time-temperature relationship investigated by Seidel (1986) may not, however, be representative of these species in all locations since the results were obtained from a single natural stand seed source. Seedling response under the greenhouse conditions of this study may also be different from the response in field conditions. This was observed in a study by Keijzer and Hermann (1966). They found that the outside environment seemed to trigger physiological changes that led to greater tolerance for heat in Douglas-fir seedlings, while the greenhouse environment did not induce such changes in the seedlings. Their treatments consisted of exposing seedlings of various ages to temperatures ranging from 67.5°C for 5 minutes to 61.3°C for 60 minutes. While all treated

seedlings showed signs of injury, the final tally of mortality 16 weeks after treatment showed that 8 - 12-week-old seedlings grown outside were able to recover from heat treatments that proved lethal to seedlings raised in the greenhouse. Seedlings grown outside also seemed to increase their tolerance to heat with age (Keijzer and Hermann 1966).

Binder and Fielder (1995) examined the effect of elevated post-cold-storage temperatures on 1+0 container white spruce (*Picea glauca* (Moench) Voss) seedlings. Their main objective was to determine the extent to which heat stresses applied to seedlings resulted in loss of growth potential and survival. Another objective was to ascertain if certain tests, which detect physiological changes, could be used to detect heat stress in seedlings prior to planting. Seedlings that had been exposed for up to 96 h to pre-planting temperatures of 5, 10 and 20°C, or up to 48 h at 30°C, and up to 24 h at 40°C had mortality of less than 8%. Seedling mortality only exceeded 20% after 72 h exposure at 30°C and after 48 h at 40°C. However, they recorded 100% mortality 63 days after planting in seedlings treated for 72 h or longer at 40°C (Table 1).

They (Binder and Fielder 1995) also observed that seasonal growth measured as shoot extension, stem diameter, shoot dry weight, and root dry weight was unaffected by pre-planting temperature treatments of 5, 10 and 20°C for up to 96 h. Exposure to 30°C for 48 h significantly reduced final shoot extension. Stem diameter and shoot dry weight were affected after exposure for 72 h and 96 h respectively. After 28 days, the percentage of non-flushing terminal buds was less than, or equal to, 10% among plants exposed to 5, 10 and 20°C for up to 96 h, 30°C for up to 24 h, and 40°C for up to 12 h, but increased with duration of exposure at 30 and 40°C.

Table 1. Mortality (%) of white spruce seedlings at time of harvest in November. Seedlings in sealed boxes stored for 6 months at -2°C, and thawed for 7 days at 5°C, were then exposed before planting to the temperatures and times shown below. Treated seedlings planted in 2 plots of 25 seedlings each during the last week in April for treatments, 5, 10 and 40°C and the first week in May for treatments 20 and 30°C. (from Binder and Fielder 1995).

Temperature	*Duration of exposure (h)*					
(°C)	*0*	*12*	*24*	*48*	*72*	*96*
5	0	4	2	2	2	6
10	2	2	4	4	4	2
20	6	2	2	4	8	6
30	2	4	2	6	26	40
40	4	2	8	56	100	100

Seedling damage and the reduced growth and survival at the higher temperatures in this study were considered to be the result of primary indirect heat injury, as defined by Levitt (1980). Binder and Fielder (1995) concluded that external box temperatures of 30 and 40°C caused a reduction in the quality of planted stock, and that the degree of damage was dependent on the duration of the heat treatment. Binder and Fielder (1995) also found that temperatures up to 20°C did not have any measurable effect on survival, growth, or ability of buds to flush.

Levitt (1980) divides heat injury into primary and secondary types. Primary heat injury is either direct or indirect. Direct heat injury results from very brief exposure to temperatures in the range of 45-60°C. It may cause membrane leakages to appear within seconds to 30 min., and may also result in damage to unheated parts. Primary indirect heat injury, on the other hand, is observed at temperatures around 40°C or lower. It is gradual and may not be apparent for days or weeks following exposure. This type of injury may cause starvation and protein breakdown depending on the length of exposure. Secondary heat induced injury is due to desiccation (Levitt 1980).

A phenomenon, known as the heat shock response (Howarth and Ougham 1993, Levitt 1980, Lindquist 1986), refers to the increase in the temperature normally lethal to an organism as a result of non-lethal exposure to high temperatures. The heat shock response induction temperature varies but is correlated with the normal range of environmental exposure (Lindquist 1986). The heat shock response in tree species has been documented (Gauslaa 1984, Koppenaal et al. 1991) but how it relates to the ability of plants to withstand direct and indirect forms of injury is not well understood (Howarth and Ougham 1993).

Colombo and Timmer (1992) studied the limits of tolerance to high temperatures causing direct and indirect damage, as defined by Levitt (1980), to black spruce (*Picea mariana* (Mill.) B.S.P.). In the experiment, ten seedlings were exposed to each of 98 temperature x length of exposure treatments, ranging from 40 to 60°C and from 5 seconds to 180 minutes. Some of the 13-15 week old seedlings in the study by Colombo and Timmer (1992) were heat shock conditioned by six daily cycles of 38°C air temperatures for 3 h per day. To minimize the effects of diurnal fluctuations in heat tolerance, they conducted all tests, including the conditioning, before noon (Colombo and Timmer 1992). After exposing both conditioned and non-conditioned black spruce seedlings to 49°C for periods from 3 to 150 minutes, it was found that direct heat damage was evident within several minutes of exposure. Volatile emissions, as noted by smell, were also always present. They noted such emissions only occasionally from needles with indirect damage. The lowest temperature at which direct damage was detected was 46°C. At that temperature 73% of the seedlings were showing direct damage while an exposure for 180 minutes at 40°C failed to cause direct damage, but did cause indirect damage in 40% of the seedlings. Indirect damage occurred at lower temperatures and with shorter lengths of exposure than did direct damage (Table 2).

Table 2. Percent of black spruce seedlings suffering from indirect damage following exposure to a range of high temperatures for varying lengths of time (from Colombo and Timmer 1992).

Exposure	Exposure temperature (°C).												
duration	40	44	46	47	48	49	50	52	54	56	58	59	60
5 s								0	0	36	8	0	8
15 s				0		0		0	0	33	100	100	100
30 s				0		0		0	0	83	100	100	100
1 min.				0		4		0	50	100	100	100	100
2 min.				0		4		3	100	100	100	100	100
3 min.	0	0	0	0	0	7	21	77	100	100	100		
5 min.				0		22	73	100					
10 min.				0	17	100	100	100					
15 min.	0	0	8	51	36	100	100	100					
30 min.	0	0	67	96	100	100	100	100					
45 min.	0	0	92	100	100	100	100	100					
60 min.	0	46	100	100	100	100	100						
75 min.	0					100							
90 min.	0	83	100	100	100								
120 min.	0			100									
150 min.	0			100									
180 min.	40												

For any exposure duration causing indirect damage, temperatures 1.5 to 3°C higher were necessary to cause the same amount of direct damage. The processes causing indirect damage were found to be more dependent on exposure temperature and less dependent on length of exposure than the processes causing direct damage. The investigation by Colombo and Timmer (1992) concluded that different mechanisms appear responsible for the two types of damage. The seedlings that had been heat shock conditioned tolerated higher temperatures and longer exposures than non-conditioned seedlings. Heat shock conditioning increased tolerance to both direct and indirect damage.

Another study also involving black spruce investigated the clonal variation in heat tolerance and heat shock protein expression (Colombo et al. 1992). Rooted one-year old cuttings were exposed to temperatures of 47°C for 30 minutes. Damage varied widely between clones of the same family indicating a large component of single tree variability in the factors contributing to heat tolerance.

High and low temperature treatments were used to illustrate changes in plant gene expression in response to environmental stresses in a review by Howarth and Ougham (1993). Stressful conditions include, besides temperature, drought and flooding, nutrient deficiency and heavy metal stress. The combined effect on the plant, where two or more stresses occur together, may be much greater than the sum of the individual stresses. There is also an interaction between the level of stress and its duration (Howarth and Ougham 1993). The heat shock response may be induced by a 5-10°C rise above the optimal growth temperature (Howarth and Ougham 1993, Lindquist 1986, Vierling 1990). Synthesis of most normal proteins is repressed and new transcription and translation of a small set of heat shock proteins is initiated (Benedict and

Hatfield 1988, Hällgren et al. 1991, Vierling 1990). It is very rapidly induced and is detectable within minutes of the onset of high temperature conditions. However, it is a transient phenomenon and a prolonged exposure to high temperatures does not result in a continuation of the heat shock response. In plants, heat shock proteins have a half-life of days (Howarth and Ougham 1993).

Koppenaal et al. (1991) looked at the duration of increased thermotolerance in jack pine (*Pinus banksiana* Lamb.), black spruce and white spruce. They compared heat injury from high temperatures in non-hardened seedlings with that in seedlings hardened at 38°C for 180 minutes a day for either 1, 3 or 6 days. In all three species, the duration of acquired thermotolerance increased with the number of days of heat hardening. For jack pine and white spruce seedlings, hardened at 38°C for 6 days, increased thermotolerance persisted for at least 14 and 10 days, respectively, after the end of the hardening treatment. In contrast, the thermotolerance of black spruce seedlings hardened at 38°C for 6 days remained elevated for only 4 days.

Koppenaal et al. (1991) also found that the persistence of thermotolerance varied between species and is also related to the severity of the initial heat shock treatment. A more extreme treatment would result in the synthesis of heat shock proteins for a longer time period (Howarth and Ougham 1993, Welch 1993). Thermotolerance is either intrinsic or acquired. Acquired thermotolerance is the result of acclimation and is not heritable, but the intrinsic is heritable since it is the result of evolutionary thermal adaptation (Howarth and Ougham 1993). Whether the acquisition of thermotolerance is a result of the synthesis of heat shock proteins is still not clear.

It may be that the role of these proteins is: i) the repair of heat-induced damage, and ii) the stabilization of proteins and other cellular components (Howarth and Ougham 1993).

Reports on heat stress in tree seedlings indicate few instances where direct heat injury occurs in nature. Heat stress is more likely to result in indirect heat injury where length of exposure in relation to temperature is the critical factor. Different species have different abilities to tolerate heat stress, and to repair heat-induced damage. Thermotolerance may develop and reduce direct and indirect damage.

Food reserves

Since food reserves are important in the life processes of a tree, their creation, storage and use have been studied extensively (Chalupa and Fraser 1968, Glerum and Balatinecz 1980, Hallman et al. 1978, Krueger and Trappe 1967, Olofinboba and Kozlowski 1973, Puttonen 1980 and 1986, Rikala 1983, Ritchie 1982). Increased temperatures increase the rate at which the food reserves are used up. They may therefore play an important role in the survival and growth of seedlings exposed to elevated temperatures, before or after planting (Puttonen 1980, Rikala 1983). Carbohydrates are also depleted during storage (McCracken 1979, Ritchie 1982).

Carbohydrates in the form of sugars and starch are regarded as the principal classes of food reserves (Glerum and Balatinecz 1980, Krueger and Trappe 1967). These reserves fluctuate considerably by season and constitute a substantial percentage of plant dry weight. Krueger and

Trappe (1967) found little seasonal variation in crude fat or protein concentrations in Douglas-fir seedlings, and considered neither to be quantitatively important as a food reserve that accumulates to support future growth. Glerum and Balatinecz (1980) studied the formation and distribution of food reserves in jack pine. They divided the tree up into four tissue components: needles, bark, xylem, and roots. An increase in saponifiable lipid production, mainly in xylem, was observed over a period of about four weeks at the end of the growing season. Glerum and Balatinecz (1980) suggested that this buildup of lipids may play an important role in the process of frost hardiness.

Krueger and Trappe (1967) looked at seasonal growth of tops and roots of Douglas-fir seedlings and related usage of food reserves. In October, as diameter growth stops and root activity slows, sugar reserves begin to increase gradually. The rate of increase accelerates as autumn progresses, perhaps because cool temperatures reduce respiration. Maximum concentrations coincided with the coldest weather. In late winter, a decrease in sugar concentration coincided with a buildup of starch. Krueger and Trappe (1967) found that starch increased earlier in the roots than in the tops where the starch concentration reached a maximum in early April. It then declined rapidly with bud break and subsequent shoot growth, followed by only a limited increase in starch concentration during summer (Krueger and Trappe 1967). It was suggested that the late fall and winter increase in sugars is caused by the continued photosynthesis without concomitant use of the photosynthate (Olofinboba and Kozlowski 1973).

Late lifting not only minimizes storage duration, but also allows pre-lifting buildup of food reserves (Krueger and Trappe 1967). Such buildup should provide more respiratory substrate for

use during storage and more residual food reserves to support vigorous growth after outplanting (Krueger and Trappe 1967). However, evergreen species have a functioning photosynthetic system when shoot elongation begins, which supplies photosynthates for expansion of the new shoot (Olofinboba and Kozlowski 1973). This confounds the influences of the reserves (Olofinboba and Kozlowski 1973, Glerum and Balatinecz 1980) but appreciable shoot growth occurs even when currently produced carbohydrates are not available (Olofinboba and Kozlowski 1973). This indicates that reserve carbohydrates can be mobilized. There is considerable evidence that reserve carbohydrates are used in shoot growth (Olofinboba and Kozlowski 1973). In red pine (*Pinus resinosa* Ait.) these reserves were found to come from old needles as well as from twigs and stems (Olofinboba and Kozlowski 1973).

Chalupa and Fraser (1968) studied the effect of soil and air temperature on soluble sugars and growth of white spruce seedlings. They observed that soil temperatures had a significant effect on the root morphology and total growth of the seedlings. A small number of thick roots were produced at lower temperatures (18 - 21°C) while a finer, fibrous root system developed at higher temperatures (27 - 32°C) (Chalupa and Fraser 1968). The roots and needles of seedlings grown at the lowest soil temperature (11°C) had the greatest sugar content which decreased with increased soil temperatures. The increased sugar content was more obvious in the roots (Chalupa and Fraser 1968). The results indicate that if trees are exposed to low temperatures in the spring for an extended period during budbreak, sugar composition undergoes changes similar to those that occur in the fall under natural conditions (Chalupa and Fraser 1968). Under these conditions, low temperature affects enzyme activity and a higher level of soluble sugars is

attained. The sugar composition may be associated with developing frost hardiness though this relationship has not been definitely established (Chalupa and Fraser 1968).

Puttonen (1986) studied carbohydrate reserves in the needles of 2+1 bareroot Scots pine (*Pinus sylvestris* L.) seedlings with the aim of using it as an indicator of seedling quality. The depletion of carbohydrate reserves was measured directly, using an enzymatic method, and indirectly by calculating the dark respiration sum. Four dark storage treatments were applied to Scots pine seedlings in plastic bags: 1) cold storage at +2 to +4°C; 2) +20 to +25°C; 3) +24 to +27°C; and 4) outdoors under a spruce canopy where the temperature ranged from -4 to +20°C. The bags were closed in the cold storage treatment and left open in the others for ventilation. The cold storage treatment lasted 118 days, the other three storage times were 45 days. Each week for five weeks, 40 seedlings were outplanted in a nursery field and 20 seedlings in a regeneration site. Outplantings were done on the same day at both sites.

Concurrent with the weekly outplanting of seedlings, four seedlings from each treatment were randomly sampled for carbohydrate analysis. Puttonen (1986) assumed the depletion of carbohydrate reserves in the previous year's needles influenced the current shoot length by reducing the amount of reserves for growth initiation and the length of new needles. Only needles were analyzed as they were considered to contain the main storage reserves in young conifer plants (Puttonen 1986). It was also assumed that the rate of dark respiration represented the rate of maintenance respiration. The rate of dark respiration can be equated with the rate of loss of carbohydrate reserves in a non-photosynthesizing plant and thus, the accumulated dark respiration to the amount of carbohydrate reserves lost (Puttonen 1986).

Results indicated that the reduction in the concentration of total glucose in needles was relatively rapid during storage (Puttonen 1986). Both storage time and temperature had a highly significant effect on the total glucose concentration. Carbohydrate depletion was fastest in the two indoor treatments where the daily mean temperature was over +20°C (Puttonen 1986). Cold storage and outdoor storage resulted in similar reductions in carbohydrate reserves (Puttonen 1986). As the total glucose concentration decreased, current season shoot and needle lengths also decreased, while the mortality rate increased. However, the relationship was not linear. There appeared to be a threshold value of about 2% total glucose below which changes were very rapid (Puttonen 1986). Roots had a more intense dark respiration rate than did shoots. This reflects a higher content of metabolically active tissue in roots. It took three weeks of storage at 20°C to reach the carbohydrate reserve threshold of about 2% (Puttonen 1986). Obviously, the time to reach the threshold value depends on the initial concentration of carbohydrates. The rate of depletion is an exponential function of temperature. The depleted carbohydrate reserves affected the growth characteristics for at least two growing seasons after planting (Puttonen 1986). Over time, the rate of carbohydrate depletion slowed down. Indirectly, this indicates that the rate of dark respiration also decreased. Puttonen (1986) observed that spring photosynthesis makes only a small contribution to early shoot development which emphasizes the need for a build-up of adequate reserves through proper nursery practice during the previous season.

Puttonen (1980) also carried out a study with bareroot 2+1 Scots pine seedlings to determine the consumption of carbohydrates during temporary storage in the spring and its effect on the planting results. The seedlings in this study were kept for 16 days in different types of storage

conditions, i.e. external cellar, cellar below building, toolshed, against the outside wall of toolshed, the attic of a toolshed, a shaded spruce stand, the attic of a building, and a furnace room. Seedlings were in bundles of 25 with their roots protected by damp peat, and were watered as needed during storage. Another part of the study involved keeping seedlings in a dark incubator at +37°C for periods of time ranging from 0.5 h to 22 h. Both of these experiments were followed by outplanting in a nursery field. The dark respiration of different seedling parts, and the seedling carbohydrate reserves, were also monitored.

Seedlings grew taller the warmer the storage temperature (Puttonen 1980). Height growth in its initial stage is mainly determined by temperature (Hari et al. 1977). The leader shoot started growing during storage. The longer it was by the end of the storage period, the shorter its continued growth following outplanting in the nursery, and the shorter the total height growth (storage plus nursery) after the first growing season. Differences in total height observed after the first growing season were still evident at the end of the fourth growing season. Mortality caused by depletion of the carbohydrate reserves was only minimal at storage temperatures of less than 10°C. The mortality of seedlings stored at room temperature exceeded 50% about 12 days after planting. For those seedlings stored at 37°C, 50% mortality was attained within 5 days (Puttonen 1980). The mortality rate of the seedlings increased when the total glucose content fell below 2% of the dry-weight. Stem elongation showed a similar trend. Depletion of total glucose reserves does not alone appear to induce a high mortality rate or growth losses (Puttonen 1980). An increase in temperature, which increases the rate of utilization of reserves, is also associated with an increased transpiration rate (Puttonen 1980). These processes, together with possible

unfavorable conditions during the reforestation operations can considerably weaken seedlings which have just been outplanted.

Ritchie (1982) reported on carbohydrate concentrations in foliage, stems and roots of coastal Douglas-fir seedlings during a lifting season and following various periods of cold (+2°C) and freezer (-1°C) storage. He also examined the hypothesis that changes in root growth potential (RGP) are modulated in the plant by changes in carbohydrate reserves. Results showed that storage duration accounted for the greatest variance component in total nonstructural carbohydrate (TNC) (28%) and extractable sugars (27%) while lifting date accounted for the least (4 and 3%, respectively) (Ritchie 1982). Root growth potential varied appreciably with respect to date of lifting, beginning at about 150 cm per seedling in November, peaking at about 300 cm per seedling in January, then declining to about 100 cm per seedling in March (Ritchie 1982). This pattern closely paralleled the changes in foliar sugars through the lifting season. However, RGP was not obviously related to carbohydrate concentrations in either root or stem tissues (Ritchie 1982).

Total nonstructural carbohydrate (TNC) concentrations decreased rapidly in foliage, stem and roots during the +2 and -1°C storage (Ritchie 1982). The decline was most rapid in foliage and during the first 2 months. Stems and root tissues had lower initial TNC concentrations than foliage and declined at a slower rate. However, the rate of carbohydrate depletion, due to respiratory consumption, was higher at +2°C than at -1°C during the first 2 months in storage. As a result, the TNC concentrations were about 30% lower in cold-stored seedlings after 2 months. Total nonstructural carbohydrate decreased exponentially during storage so the rate of

respiration must have been declining. The decline may have been caused by a reduction in substrate concentration, available oxygen or both. Thus, by 6 months the difference in TNC between cold- and freezer-stored seedlings had disappeared. Ritchie (1982) found also that RGP was not significantly affected by storage temperature. Field survival was 95% or higher for all treatments except the February- and March-lifted seedlings which were stored 6 months. In those cases budbreak occurred within 2 weeks of planting in September and October and new growth was killed by a November frost. Of the seedlings stored 12 months, only 35% survived pot trials (Ritchie 1982).

Storage effects

During transportation and planting many types of physiological stresses occur. The development of water stress is commonly considered to be the most harmful (Hallman et al. 1978). The root tips of bareroot stock are destroyed to a great extent during, and as a result of, transportation (Hallman et al. 1978). In addition, the roots dry out during planting and often the fine rootlets fail to make good ground contact with the soil particles after planting. All these factors produce a post-planting, seedling water deficit as it is impossible for the seedling to take up water in sufficient amounts (Hallman et al. 1978). The water deficit will not cease until new root tips have grown and established soil contact. Stored carbohydrates are consumed during handling and transportation, where mechanical damage also can occur. This interference in the development of planted seedlings is called planting shock which has a harmful effect on the subsequent development of the plantation.

Hallman et al. (1978) examined studies of gas metabolism and height growth of plants in the field, especially those that concentrated on water deficit stresses. They then adapted the measurement and data analysis technique to planting shock studies with the objective of studying the self regulation of transpiration, photosynthesis and height increment of Scots pine seedlings. Differences in degree of self regulation have been assumed to accurately reflect planting shock (Hallman et al. 1978). The results indicated that planting can have pronounced and long lasting effects on the metabolism of a seedling (Hallman et al. 1978). Daily amounts of transpiration were reduced in a few days to 40% of potential transpiration, while daily photosynthesis decreased to 50% of its potential. The variation among seedlings was however, great in both transpiration and photosynthesis. Hallman et al. (1978) suggested one reason for this great variance may be differences in size and density of seedling root systems. A 20-min exposure of the uncovered roots to the sun caused, especially a few days after treatment, a more rapid decrease in the daily amounts of transpiration than did planting. Transpiration was then about 25% of its potential. The decrease in photosynthesis was of similar magnitude. They found planting shock effects lasted for a longer time than expected. Hallman et al. (1978) determined that recovery occurred at the same time as cessation of height increment and maximal growth of needles. They also found that the rapid growth of roots starts after the cessation of height increment. The recovery of the water balance is connected with the degree of root regeneration (Tranquillini 1973, Havranek 1975). The effect of environment on the self regulation was very clearly evident in photosynthesis which was as depressed, especially at elevated temperatures after planting, as it was with water deficit.

Lähde (1978) examined i) how long paper-pot and peat-pot planting stock could be temporarily stored at the planting site, and ii) what effect watering and covering 8-week old planting stock had on seedling development. Open styrofoam seedling boxes, containing the paper-pot and the peat-pot seedling containers, were placed directly on the soil of a large clearcut in one layer (Lähde 1978). For covering, the same kind of boxes were inverted on those containing the seedlings. It was found that paper-pot seedlings could be stored unwatered and uncovered for 1 month without significant reduction in survival during the following 2-4 years. Further, covering the paper-pot seedlings had a negative effect but it could be reduced by watering the seedlings before planting. The peat-pot seedlings were clearly affected by the storage time during the dry summer. A 2-week storage resulted in an almost complete failure of subsequent reforestation plantings. Survival increased when seedlings were watered before outplanting. By watering every second or fourth day and by covering the seedlings, the peat-pot seedlings could be stored for 2 weeks with good results, and with fair results for a month. The rate of survival was then about 70% (Lähde 1978). These results suggest that container type and watering are the determining factors. Paper-pot seedlings that were uncovered and unwatered survived after outplanting, while peat-pot seedlings under those conditions did not. Watering mitigates the negative effects of covering the paper-pots and improves survivability of seedlings in peat-pots. Covering the peat-pots may help retain moisture, thus improving survival rates compared to uncovered peat-pots.

Another study of planting site storage was made by Mattsson (1986). He studied the post-planting development of cold-stored and outdoor-stored Scots pine containerized seedlings for

several plantings in early May and June. His objective was to demonstrate possible differences in subsequent growth between seedlings planted without or after a period of field storage. Furthermore, he was interested in the importance of storage. The results indicated negative effects of field storage in cardboard boxes at the planting site. Without any other protection, they are more pronounced for outdoor-stored seedlings than for cold-stored seedlings (Mattsson 1986). Outdoor stored seedlings began shoot elongation during storage and were affected by desiccation. Differences in subsequent growth may presumably be explained by differences in respiration rates during field storage (Mattsson 1986). Negative effects of late planting dates are also somewhat more apparent for outdoor-stored seedlings. Their shoot elongation is in progress or has just been completed, while cold-stored seedlings are at the same phenological stage regardless of planting date (Mattsson 1986). Outdoor and cold-stored seedlings planted in early May without field storage showed similar development for all growth variables measured during the first three growing seasons following planting (Mattsson 1986).

Rikala (1983) carried out a theoretical analysis of the amount of heat released by 500 pine seedlings (5 g dry weight) through dark respiration in transport bags. He calculated that the rate of temperature increase was about 1°C per h. Thus, if the temperature to start with was +25°C, after 15 h it should be +40°C if the rate of increase was linear. Since respiration rate increases with higher temperatures the rise in temperature would accelerate it. However, as respiration depletes sugar reserves this will slow down the respiration rate as well as the rate of temperature increase (Rikala 1983). At +35°C, the heat released through respiration would cause a temperature increase of 1.8°C per h (Rikala 1983). According to Puttonen (1980) the peak of

the dark respiration curve is reached at about +45°C for Scots pine. Rikala (1983) suggested transport bags should be stacked so that air can circulate freely around the outside surface. This will help contain the temperature increase as a result of respiration, and minimize the probability of poor establishment.

Kauppi (1984) studied bareroot Scots pine transplants from the time they were lifted at the nursery to acclimation on the planting site. He wanted to develop concepts and methods for recognizing and analyzing the dynamic aspects of injury development in seedling transplants. Damage to plants is difficult to detect within the critical phases of transportation, storage and planting. Conifers, and broad-leaved plants in the leafless stage, are slow in displaying symptoms so damage often remains latent for several weeks (Kauppi 1984). Due to this delay, weak plants may be introduced into the expensive planting phase, as they appear vigorous. Moreover, also due to the delay of symptoms, it is difficult to keep track of where, when and why the damage was introduced. Thus, the mistake of planting moribund stock is likely to be repeated (Kauppi 1984).

There are two ways of looking at development of damage in seedlings (Kauppi 1984). One method relates the amount of damage observed in the seedling to the duration of the exposure which induced the damage. In this case, the environment has been assumed constant. The other method considers the duration of the critical period as a constant, and damage as a function of some quantitative variable characteristic of the plants' environment. Plants seldom experience sudden dosages of unfavorable conditions but more often such conditions prevail over a period of time, with their levels being sometimes weaker, sometimes stronger (Kauppi 1984).

Decreased survival and growth occurs only after a long period of time when, perhaps, the unfavorable conditions have already disappeared. Hence, the process is essentially dynamic in character (Kauppi 1984).

In developing his Dynamic Injury Model, Kauppi (1984) used the concepts of strain, stress and stress resistance as introduced by Levitt (Levitt 1972). Strain is used to describe the physiological pathway which precedes the injury. All plant properties, which in qualitative or quantitative terms differ from the respective properties of a vigorous tree, are grouped together under the concept of strain (Kauppi 1984). Stress has been defined as an environmental factor capable of inducing a potentially injurious strain on an organism (Levitt 1972). Stress resistance includes the plant factors affecting the process of injury (Levitt 1972). Stress resistance factors are genotypically fixed to vary within a certain range, but within this range there is phenotypic variability due to environmental growing conditions (Kauppi 1984). According to Kauppi (1984), strain results from stress and stress resistance, while injury results from strain and site environment.

Kauppi (1984) carried out an experiment to test whether the injury in outplanted bareroot Scots pine transplants depended only on the duration of stress conditions prior to planting. He studied the effects of exposing root systems to different temperature levels for various time periods. Water vapour deficit was also a variable. It was highest at the highest temperature and lowest at the lowest temperature. Exposure ranged from 1.5 to 11 h, 4 to 22 h, and 28 h to 216 h, at temperatures of 37°C, 24°C and 2°C, respectively. These conditions may be referred to as the 'high', 'moderate', and 'low' environments. The results showed that survival decreased to half in 5.4 h, 12 h and 50 h respectively. A Growth Index decreased to half in 4.0 h, 6.2 h and 37 h,

respectively (Kauppi 1984). This indicates that injury does not develop at a constant rate. It is not only dependent on the duration of the stress but also on the environmental conditions. A given decrease in survival was reached earlier in the 'high' environment than in the 'moderate' environment, and earlier in the 'moderate' environment than in the 'low' environment (Kauppi 1984).

In yet another experiment by Kauppi (1984), bareroot seedlings were protected from desiccation and excess heat during a 56 h exposure period. The solar irradiation for the 56 h period averaged 310 W/m^2 (Kauppi 1984). Exposure did not have any effect. In contrast to these results, all the plants died when exposed to solar radiation without any protection from heat or desiccation for just 5 h (Kauppi 1984). When seedlings were protected from the sun but not from desiccation, only 3 out of 20 seedlings survived an exposure of 22 h. The results provided a clear answer to the question of direct versus indirect stress due to solar irradiance. When desiccation and excess heat were removed, direct irradiance appeared to have no significance (Kauppi 1984). Unprotected plants were severely injured, with the injury being caused by indirect irradiance stress (Kauppi 1984). A similar conclusion was also reached by Nelson and Ray (1990) in a study of Sitka spruce (*Picea sitchensis* (Bong.) Carr.).

To test the significance of high temperatures as a stress factor in addition to evapotranspiration, Kauppi (1984) carried out two experiments with Scots pine. In one, seedlings were exposed to six temperature levels between 25 and 58°C, and in another to five temperature levels between 35 and 55°C. All seedlings were exposed to high temperatures for only 10 minutes. It was found that there was an abrupt increase in mortality between 45 and 55°C (Kauppi 1984). All reference plants, and plants exposed to 45°C or lower temperatures survived. Growth was also maintained

at the control level up to this critical temperature threshold. Damage would occur also at the lower temperatures if more time were given for the stress to build up (Kauppi 1984). Such slowly developing strains are likely due to gradual loss of carbohydrates (Puttonen 1980). With the experimental design used by Kauppi (1984), it was only possible to investigate abrupt strains such as dehydrolysation of plant proteins. It appeared that for such abrupt strain the threshold temperature is about 50°C.

A final experiment involved keeping the roots of seedlings protected from desiccation while the shoots of the transplants were unprotected at an air temperature of +22°C (Kauppi 1984). During the treatment, bags were watered daily to a constant weight. One group was kept in these conditions for six days and one group for nine days. Heat sums for the two treatments were 105 and 150 degree-days, respectively. Seedlings were planted in a cutover following the treatments. Storage at room temperature prior to planting appeared to result in substantial mortality only on one site. Obviously, the food reserves were not severely exhausted. This indicates that the plants must have been in good condition before the stress treatment (Kauppi 1984). These observations are in agreement with the view that conifers are rather flexible in their carbon metabolism (Glerum 1980). Injury due to the "room temperature stress", i.e. depletion of carbohydrates, seemed unimportant in terms of growth (Kauppi 1984). The variation in shoot growth of both stressed and non-stressed plants was clearly dominated by the site factor. Kauppi (1984) concluded the main focus should be the protection of bareroot seedling roots. The roots should be covered at all times. Additional measures of protection are needed because covering the roots does not prevent all desiccation. Seedlings should be stored in places where the level of evapotranspiration is low. Since high evapotranspiration often is associated with high temperatures, controlling these would serve as an indirect control of root desiccation (Kauppi

1984). The use of container stock may alleviate the problem of root desiccation to some extent because many roots are protected by the soil substrate which also holds water. However, in many container types, most roots grow on the outside of the soil plug and are therefore subject to desiccation if left unprotected after lifting.

Electrical conductivity

An increase in electrolyte leakage from cells is evidence of excessive stress to tissues, and is considered one of the first signs of direct injury to cell membranes (Levitt 1980, Tal and Shannon 1983, Ruter 1993). While it is often used to detect freezing stress (Burr et al. 1990, Colombo et al. 1984, Flint et al. 1967) it has also been used for other applications (McKay and Mason 1991, McKay 1992). Whitlow et al. (1992) proposed another method of examining ion leakage from plant tissue, the tissue ionic conductance (g_{Ti}), and compared it with electrical conductivity. They found g_{Ti} to be more reliable since it explicitly includes chemical driving force and tissue surface area.

Binder and Fielder (1995) used electrolyte leakage from stem and needle segments as an indicator of heat stress based on the assumption that a relationship exists between cellular membrane damage resulting from heat stress, and electrolyte efflux (Blum and Ebercon 1981, Burr et al. 1993, Ruter 1993, Tal and Shannon 1983). They expressed efflux of electrolytes due to membrane damage as a fraction of the total electrolytes in the tissue, using the term fractional release of electrolytes (FRE).

Results show FRE values for needles of less than 0.2 for the 30 and 40°C treatments of up to 48 h. For the 72 h and 96 h treatments at those temperatures, the FRE values for needles increased sharply. Binder and Fielder (1995) found good correlation between FRE and needle damage after 14 days. They found that fractional release of electrolytes from stem segments at the 40°C treatment increased after 24 h while for needles it increased only after 48 h. This difference at equivalent treatment combinations may suggest a higher thermotolerance of needles (Binder and Fielder 1995).

Operational storage temperature recommendations

Many stock handling guidelines specify a maximum storage temperature. If refrigerated local storage is available, DeYoe (1986) suggested that seedlings shipped for spring planting be kept as close to 33°F (+0.5°C) as possible. If no such storage space is available, box temperature should be kept below 40°F (4.4°C).

Binder and Fielder (1995) studied the effect of elevated post-cold-storage temperatures on physiology and survival of boxed white spruce seedlings. Seedlings showed considerable tolerance to "heat stress" above 10°C over short durations. However, they recommended, as a precaution, that seedlings should not be exposed to temperatures above 5°C after cold storage, but prior to planting.

The Alberta Contract Seedling Supply Manual (Alberta Forest Service n.d.) does not specify how seedlings are to be kept cool. However, for overwintered stock the Manual specifies that trees should be planted before temperatures, in the boxes, reach 12°C. Further, the Manual says if temperatures reach 12°C, seedlings should be placed upright and watered.

Tests of seedling covers

Seedling bags were tested for their ability to protect bareroot seedlings under severe, but realistic, field conditions (Kauppi 1984). Five different bags were compared: A= white polythene bag; B= double-walled polythene bag with a white exterior and a black inner wall; C= black polythene bag; D= double-walled polythene bag with metallic silver exterior and black inner wall; E= paper bag with three layers of paper having air-spaces between the layers. The wall materials were tested in detail. Measurements were taken of short wave radiation transmittance, short wave radiation reflectance, spectral distribution of the reflected short wave radiation and spectral distribution of transmitted thermal radiation (Kauppi 1984). He described the environmental conditions inside the bags. Thus, temperature and potential evaporation inside the bags were monitored under field conditions.

Each bag was filled with 150 bareroot 2+1 Scots pine transplants and then tightly closed. Temperature was measured at 100 second intervals with thermocouples that remained in the bags for 42 days, from May 11, 1978. A planting experiment with storage as a treatment was also conducted in conjunction with this study (Kauppi 1984). For this experiment, twenty fresh transplants were placed horizontally on top of the original transplants inside the bags and another twenty beneath them, on June 14. The test plants were left in the bags for a week and then planted in an open nursery field . Possible damage to the plants was described 12 weeks after

planting when shoot and needle growth for that season had ceased. The transplants were then graded into four vigor classes. Dead transplants, i.e. transplants with no obvious living buds or needles, were grouped into Class 0. The remaining three classes were reserved for living transplants with increasing vigor from Class 1 to Class 3 (Räsänen et al. 1970).

The results of the bag wall material tests by Kauppi (1984) are shown below.

Bag type	Relative proportion (%) Transparency (T)	Reflectance (R)	Absorbance (100 - T - R)
A = white	50 [+/-10]	62 [+/-10]	-
B = white / black	3.0	67	30
C = black	0.1	9.5	90
D = silver / black	0.01	28	72
E = paper	2.0 (27)[1]	47 (47)	51 (26)

Measurements of internal bag temperatures showed that at night, bag internal temperatures approached ambient air temperature. In sunshine, however, the internal temperature ranged from 10 to 25°C higher than the external temperature. The rate of temperature increase was about 2°C per minute when the sun came out after a cloudy period at midday, with a similar rate of decrease when it disappeared. The highest instantaneous temperature reached was 50°C in Bag A for a period of about two minutes.

The potential evaporation rate inside the bags also appeared to follow the course of the incident solar radiation. In some cases, the potential evaporation rate doubled or even tripled within five minutes (Kauppi 1984).

[1] Number in brackets are for a single layer of Bag E

Most transplants that were exposed to field conditions for one week, except those in Bag A, survived the one-week storage treatment in relatively good shape (Kauppi 1984). Twelve weeks after planting, they were classified into either of two classes: good (Class 2) or very good (Class 3). However, transplants located on top of the pile in Bag A appeared to have suffered from the treatment (Kauppi 1984).

Conclusions drawn from this part of the study were that temperature varied more strongly inside the bag compared to the variation outside the bag. Bag A had several properties (high reflectance; bluish in colour; transmittance of thermal long wave radiation) which should have served to maintain low temperatures and evaporation rates. But, these positive factors could not compensate for the effect of transparency, in increasing temperature and evaporation. The transplants stored in Bag A, were less vigorous than those from other bags. The results indicate a high transparency to short wave radiation will, in certain field conditions, generate particularly severe conditions inside the bag.

DeYoe et al. (1986) conducted a study to evaluate materials used by growers and foresters to protect seedlings from overheating when packed in Kraft® paper bags. The objective was to determine whether certain materials were capable of keeping seedlings cool during exposures to solar radiation similar to that encountered under operational conditions. The study, in May 1982, evaluated seven protective treatments plus control: #1- dark green canvas; #2- canvas painted off-white on both sides; #3- a white cloth sheet; #4- a silver (outer surface) and blue (inner surface) crinkled-foil thermal wrap; #5- Mylar® with a white outer surface and a silver inner surface; and, #6- (the reverse) Mylar® with a silver outer surface and a white inner surface.

Another Poly-Kraft® bag of seedlings was placed in heavy shade with good air circulation. The bags, containing 2+0 Douglas-fir seedlings, were carried to the site each day and set out in a quarter-acre area. Dial thermometers were inserted into each bag with the temperature sensor close to the center of the packed seedlings. Temperatures were read at 30-minute intervals. Temperatures taken during one day were averaged to produce a mean daily temperature for each treatment. Results were significantly different among treatments (DeYoe et al. 1986). The temperature in the control bag (without cover) reached 31.6°C in 420 min. while the temperature in the dark-green canvas (#1) bag reached 40°C after the same time period. The white-painted canvas (#2) had a maximum temperature of 26.7°C, nine degrees less than control. The white sheet (#3) and the crinkled-foil wrap (#4) were close to control and differed only by 1.6°C. Mylar® with the silver surface towards the sun (#6) kept the bag temperature to 21.7°C after 420 minutes while the Mylar® with the white surface facing the sun (#5) kept the bag temperature essentially the same as in deep shade, i.e. 15°C after 420 min. It was concluded that the Mylar® tarp makes an excellent tool for seedling protection during all phases of handling (DeYoe et al. 1986).

Some heat transfer principles as they apply to reflective type tarpaulins

Heat transfer can take place by three modes: conduction, convection and radiation (Erhardt 1977; Thomas 1980). Transfer is always from a warmer to a colder region. Conduction and convection require a medium while radiation can take place across a perfect vacuum. Radiation only needs two regions of differing temperatures that "see" each other. Radiant energy travels in a straight line through space at the "speed of light": it is the main means of energy transfer

throughout the known universe (Gates 1980). There are two important bands of electromagnetic waves: the solar spectrum and the longwave spectrum. Sunlight consists of short wavelengths, about 200 to 2600 nm (Fairey 1986). The visible portion of the solar spectrum has wavelengths of about 400 to 700 nm while the 700 to 2600 nm part of the spectrum is invisible, near-infrared radiation. There is both near-infrared and far-infrared radiation. On earth, regions of different temperatures that "see" each other exchange energy via far-infrared radiation in the 4000 to 40000 nm wavelength band (Fairey 1986). This latter is sometimes called "thermal" or long-wave radiation. The effect of both is heat (Gates 1980).

A reflective tarpaulin acts as a barrier to stop far-infrared radiation from increasing seedling box temperature. Radiant barriers are materials that restrict the transfer of far-infrared radiation across an airspace (Fairey 1986). They do this by reflecting most of the radiation that strikes them and, at the same time, by not absorbing and re-radiating much of the radiant energy. A material that has this capability is said to have a very low emissitivity. The lower the emissitivity the better the radiant barrier (Fairey 1986).

Emissitivity values range from 0 to 1 (Thomas 1980). Emissitivity plus transmissitivity plus reflectivity for any material must always equal one. A material with an opaque surface has a transmissitivity of zero, thus its emissitivity equals one minus its reflectivity. Aluminum foil (polished) is an excellent radiant barrier (Fairey 1986). It has a low emissitivity (0.05) and therefore it eliminates 95% of the radiant transfer potential (Thomas 1980). Aluminum foil is also a good thermal conductor. Consequently it has an extremely low R-value (very little

resistance - c.f. aluminum wiring). However, if it is placed between materials that are attempting to transfer thermal energy by radiation (rather than by conduction), and if it is separated from these materials by an air layer, the foil effectively eliminates the normal radiant energy exchange across the airspace. This is the operating principle of a radiant barrier (Fairey 1986).

A reflective type tarpaulin is made from a lamination of metalized mylar, nylon or polyethylene scrim and an outer surface of white vinyl or polyethylene that reflects most of the solar radiation (Stjernberg 1991). Reflective type tarpaulins are opaque and radiation that is not reflected by the white surface is absorbed and heats the white outside material. Some of that heat is then transferred through conduction to the cooler inner surface of metalized mylar. This material has very low emissitivity and therefore only a small amount of the heat received from the white outside layer is released inwards through radiation to the airspace around the seedling boxes (Stjernberg 1991).

Objectives

1. To determine the effectiveness of new and used reflective tarpaulins of various qualities in resisting heat transfer.

2. To determine if there are immediately measurable detrimental biological responses in seedlings stored under these tarpaulins for different lengths of time and at different temperatures.

3. To determine if there are significant effects on growth and survival of seedlings stored under various types of reflective type tarpaulins for different lengths of time and at different temperatures.

4. To determine if pre-conditioning significantly affects growth and survival of heat-stressed seedlings (Objective added after the first year).

Experiments

Field Experiment

Materials and methods

The materials selected for testing are shown in Table 3.

Table 3. List of treatments and test materials in the field experiment.

Treatment No.	Test Material	Type[2]	Status
1	Lightweight Silvicool®[3] 3	8 x 8	New
2	Standard Silvicool® 3	12 x 10	New
3	Original Silvicool® type[4]	18 oz.	New
4	Econocool	8 x 8	New
5	Silvicool® 2	12 x 12.	2-years old
6	Original Silvicool® type	18 oz.	7-years old
7	Original Silvicool® type	10 oz.	1-year old
8	Unprotected box	N/A	New
9	FIST[5] Canopy	N/A	New
10	Silvicool® 2 shelter[6]	12 x 12	3-years old

Species and stocktype

Container grown white spruce (*Picea glauca* (Moench) Voss) seedlings for the tests were provided by the Northwood Reforestation Centre (Northwood Pulp & Timber Ltd.) in Prince

[2] Reflective tarpaulins are distinguished in two way; i.e. by the weight, in ounces per square foot (e.g. 18 oz), and in the case of woven materials, by the number of strands per inch in two directions (e.g. 8 x 8).

[3] Silvicool® is a registered trademark of Bushpro Supplies Ltd., Anmore, B.C.

[4] The 'Original Silvicool®' reflective tarpaulin was a custom made 12 oz. vinyl material.

[5] The Fiberglass Insulated Seedling Transporter (FIST) Canopy consists of two fiberglass shells with 4 cm of solid foam insulation in between. It is manufactured by the Horizon Fiberglass Products Ltd., Delta, B.C.

[6] Treatment No. 10 consisted of a purpose-built shelter made of Silvicool® 2 tarpaulin material, and manufactured commercially by Bushpro Supplies Inc., Anmore, B.C. The dimensions of this shelter was somewhat larger than for those set up for this trial (W = 90 cm; L = 120 cm; H = 50 cm)

George. The white spruce seeds, from B.C.M.O.F. seedlot #2240 collected at 640 m elevation in the McGregor area in 1973, were sown on March 7, 1991, in PSB 313A (Beaver Plastic, Edmonton, AB) containers (62 ml). Seedlings were lifted November 15, 1991, packaged in waxed seedling boxes with plastic lined Kraft bags and cold stored until June 1, 1992 at -2°C. The seedlings were then removed from the freezer and thawed at an ambient temperature of about +16°C until June 4 when they were returned to cool storage (+2°C). On June 5 the seedlings were rebundled and labeled for the experiment. This was carried out inside the cold storage facility in order to keep the seedlings cool. The seedling boxes were transported to the Red Rock Research Station by pick-up truck on June 8 and stored during the remainder of that day in their cool storage facility.

In order to maintain the same physical environment in the boxes after seedlings had been withdrawn for planting, the vacated space was filled by replacement seedlings provided by the Faculty of Forestry at the University of British Columbia. Replacement seedlings were stored in the FIST until used.

Seedling shelters

Seedling shelter frames were constructed of 1"x2" wood and measured 120 x 90 cm with a height of 50 cm. The reflective tarpaulins chosen for testing were cut down to fit these shelters and extended to the ground on all sides. Tarpaulins were secured with rope and tent pegs (Figure 1). The shelters and the FIST canopy were set up in three rows on a grassy area beside the Red Rock Research Station, 15 km south of Prince George, British Columbia.

Figure 1. Reflective tarpaulin shelters set up outside the Red Rock Research Station, Prince George, in June 1992. The Fiberglass Insulated Seedling Transporter (FIST) canopy is seen in the far left corner of the picture. The shelter to the left of the canopy is Treatment 10 - a purpose-built shelter manufactured commercially. The ambient temperature was recorded inside the weather station shown on the right.

Seedling box temperatures

The temperatures inside the seedling boxes were measured on a continuous basis. One thermocouple, type T (copper-constantan) was placed in the physical center of each seedling box, i.e. in amongst the foliage of the seedlings which were kept upright. All thermocouples were connected to a data logger (model CR10, Campbell Scientific, Inc., Edmonton, Alberta) equipped with a multiplexer. All thermocouples were scanned every 10 seconds and the average value for each 15-minute period was recorded by the data logger. Recordings started at 21:00 hours on June 8, 1992, and continued uninterrupted for 144 h.

Environmental measurements

The ambient temperature was measured with a thermocouple placed inside the Stevenson Screen at the weather station at Red Rock Research Station. Wind speed was measured with an anemometer (Met-One, Model 014, Campbell Scientific, Inc., Edmonton, Alberta), centrally located amongst the shelters. A LI-200SA Pyranometer Sensor (Campbell Scientific, Inc., Edmonton, Alberta) located on top of the weather station's thermometer enclosure measured the incoming solar radiation in the 400-1100 nm band. Ambient air temperature, wind speed and solar irradiance were measured at the same intervals as the temperatures in the seedling boxes, using the same CR10 datalogger.

Electrical conductivity measurements

Needle samples for conductivity tests were collected from the seedlings to be planted each day for six days (Times 1-6). Six needles were removed with tweezers from each of the 24 (four seedlings * six blocks) seedlings per repeated treatment, and put into 10 mL plastic vials with screw tops. The 24 (six needles * four seedlings) needles constituted one sample resulting in one conductivity sample per block per repeated treatment. Thus, a total of 120 conductivity samples were collected each day for six days. When all samples for one day had been collected, 6 mL of distilled water was added to each vial. Samples were then left for approximately 21 h at room temperature in order for the electrolyte to diffuse out. Conductivity measurements were made with a Digital Conductivity Meter (Cole-Parmer Model 1481-60, Cole-Parmer Instrument Company, Chicago, Illinois). Following measurement, samples were "killed" by placing them in an oven at 90°C for 2 h. Measurements of the conductivity in the "killed" samples were taken

after the electrolyte in the needles had diffused for at least 21 h. The procedure was adapted from the methodology used by the Research Laboratory of the British Columbia Ministry of Forests and others (Colombo et al. 1984, Oleinikova 1965).

Altogether, there were 720 conductivity samples. Control conductivity measurements were obtained at Time 1, i.e. before the seedling boxes were put into the shelters. Two blanks, i.e. vials with distilled water only, were included with the other samples for each day. Relative conductivity was calculated according to the following formula:

RC = (EC1 - B1) * 100 / (EC2 - B2)

where: RC = relative conductivity; EC1 = electrical conductivity of leachate before "killing"; EC2 = electrical conductivity of distillate after "killing"; B1 = average electrical conductivity of the two blanks before "killing"; B2 = average electrical conductivity of the two blanks after "killing" (Colombo et al. 1984).

Experimental design

The experimental design is a split-plot randomized complete block design with repeated measurements within the experimental unit. Each of the six blocks was divided into six time zones; each time zone was then divided into ten treatment parts and each part was planted with two sets of four seedlings, in an alternate fashion. The layout is shown in Figure 2 and Tables A-1 and A-2 in Appendix I.

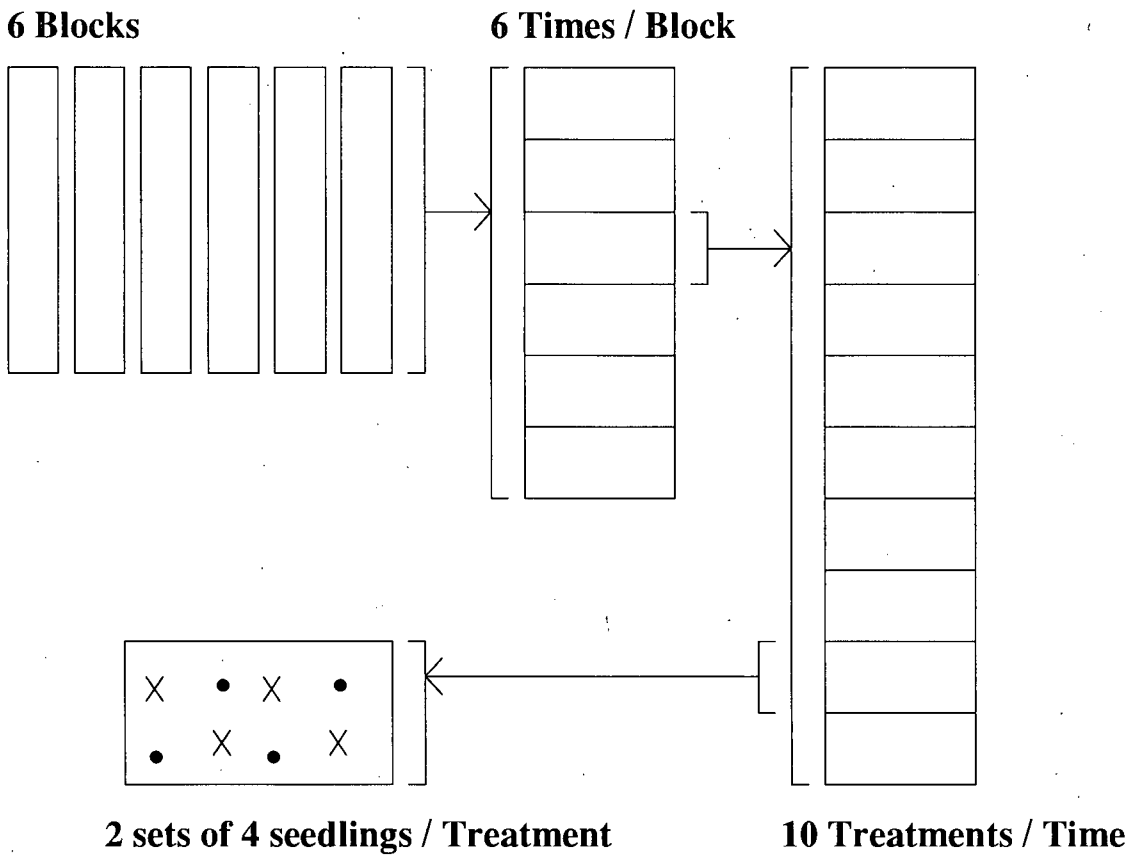

Figure 2. Schematic layout of sample plot with a split-plot randomized complete block design, with repeated measurements of the experimental units. Each of the six blocks was divided into six time zones; each time zone was divided into ten treatment parts and each part was planted with two sets of four seedlings, in an alternate fashion, x = set 1 and • = set 2.

Conductivity data were statistically analyzed using analysis of variance (ANOVA) in the STAT module of SAS® for Windows (Version 6.08). A simpler version of the model shown in Table A-3 Appendix II, that excluded the Block factor was used.

Outplantings

Seedlings were outplanted in prepared nursery beds at the Red Rock Research station. The seedling spacing in the beds was 25 x 25 cm. Each seedling sample consisted of two sets of 24 seedlings from each of the ten replicated treatments. The 480 seedlings treated each day were planted in ten randomly selected locations in the designated time zone within each of the six blocks. Four seedlings from each of the two sets were planted alternately in the location selected for that treatment (Figure 2). The first outplanting, designated as Time 1 and used as Control for the experiment, were removed from seedling boxes and planted on June 8 before the boxes were put into the shelters. Because the following day, June 9, was rainy and cold, no planting was done on that day. Samples of seedlings were then withdrawn from the stored seedling boxes daily for another five days , i.e. Times 2 (= June 10, etc.), 3, 4, 5 and 6, and planted according to the layout (Figure 2).

The experimental unit consisted of 8 seedlings per treatment. A total of 2880 seedlings were planted from the ten replicated treatments. Root collar diameter and total height were measured and recorded immediately after planting. These measurements were repeated after the first, second and third growing seasons. In addition, survival surveys were done each spring (see Appendix III for a time-line).

Statistical analysis

Statistical analysis (covariance) of the growth data was carried out with the STAT module in SAS® for Windows (Version 6.08)[7] for personal computers, and with the SAS program on

[7] SAS® for Windows (Version 6.08), SAS Institute Inc., Cary, NC., 1993.

Unixg at the University of British Columbia. The General Linear Model (GLM) was used in both programs, to allow for missing data from dead seedlings. The statistical model is shown in Table A-3, Appendix II. Because of the unequal number of observations per experimental unit, averages are calculated as LSMEAN (Least Square Mean), i.e. they are determined from the data in such a way as to minimize the sum of squares of the deviations from the means (Hicks 1982).

Soil and foliage analysis

To determine what may have induced growth differences in the various parts of the plot, soil and foliage samples were collected and commercially analyzed[8]. Foliage samples were collected from seedlings adjacent to the soil sample locations in the fall of 1994.

Controlled Conditions Experiment

Materials and methods

A set of tests were carried out to determine the ability of the selected tarpaulin materials to resist heat transfer under controlled conditions. Tarpaulins used in the field testing were divided into 40 cm squares and three squares per tarpaulin were randomly selected and cut out. Since each tarpaulin was replicated in the field, a total of six samples per tarpaulin type were tested.

[8] Pacific Soil Analysis Incorporated, Richmond, B.C.

For the tests, a box was constructed of 12 mm thick plywood, with inside dimensions of 30 cm x 30 cm x 30 cm, and insulated with 15 cm of styrofoam on all sides. A removable lid, with a 30 cm square opening, allowed tarpaulin material to be placed as a cover for the inside top of the box. Thermocouples (type T, copper-constantan) were placed 2.5 cm below the surface of the tarpaulin material inside the box, one in each corner and one in the centre. Another thermocouple was placed 2.5 cm above the surface of the tarpaulin, in the centre of the opening. Two photo floodlights[9] (General Electric, 250 Watt, type BBA, 3400°K) were mounted 45 cm above the surface of the tarpaulin in such a way that the center of their beams struck the tarpaulin at an angle of about 30°. These floodlights provided the heat during the tests.

The tarpaulin squares were placed in the box in random order and each was exposed to the heat from the floodlights for 30 minutes. The thermocouples were connected to a datalogger (model CR10, Campbell Scientific, Inc., Edmonton, Alberta) which scanned their temperature every five seconds, averaged it for each 1-minute period, and recorded it.

For the data analysis, the heat sums in degree-hours below and above the tarpaulin were calculated using the ambient temperature as the base temperature. Tested materials could be compared using the ratio of these two heat sums, based on 30 minutes of recordings, and rated according to their ability to resist heat transfer. The ratio was used in analysis of variance (ANOVA) and Bonferroni's multiple range tests, both available in the STAT module of SAS® for Windows (Version 6.08).

[9] These are tungsten filament lamps with a maximum radiant power of about 200 microwatts per 100 Å per Lumen at a wavelength of 0.8 microns.

Laboratory Experiment

Materials and methods

Materials

Seedlings from the same seedlot as used in the main experiment were provided by Northwood Reforestation Centre, Prince George. The seedlings, transported over-night by courier from Prince George to the Faculty of Forestry at University of British Columbia, were shipped frozen directly from the cold storage. The seedlings were shipped as they were stored, i.e. inside Kraft-paper bags in waxed cardboard boxes. Seedlings were nearly thawed by the time they arrived 21 h later, and were then placed in cool storage at +5°C. The first pre-conditioning treatment started six days later, on June 14, 1993.

Treatments

The treatments listed below were applied to a total of 960 seedlings used for this experiment. The laboratory at the Department of Forest Sciences of the University of British Columbia was utilized for this experiment.

Pre-conditioning treatments:

Pre-conditioning 0 days (controls): 320 seedlings, left in cool storage at +5°C.

Pre-conditioning 4 days: 320 seedlings removed from cool storage, placed in Kraft-paper bag, left at room temperature for 2 h, put into an incubator at 30°C for 3 h, left at room temperature for 2 h, and then returned to cool storage at +5°C.

Pre-conditioning 8 days: 320 seedlings removed from cool storage, placed in Kraft-paper bag, left at room temperature for 2 h, put into an incubator at 30°C for 3 h, left at room temperature for 2 h, and then returned to cool storage at +5°C.

Heat stress treatments:

80 seedlings from each of the 3 pre-conditioning treatments = no heat stress treatments (controls), left in cool storage at +5°C.

80 seedlings from each of the 3 pre-conditioning treatments = placed in Kraft-paper bag, put into an incubator at 30°C for 8 h (40 seedlings), and 48 h (40 seedlings).

80 seedlings from each of the 3 pre-conditioning treatments = placed in Kraft-paper bag, put into an incubator at 35°C for 8 h (40 seedlings), and 48 h (40 seedlings).

80 seedlings from each of the 3 pre-conditioning treatments = placed in Kraft-paper bag, put into an incubator at 40°C for 8 h (40 seedlings), and 48 h (40 seedlings).

Electrical conductivity measurements

Needle samples for conductivity tests were collected after the heat stress treatments were applied and before the seedlings were returned to Prince George for outplanting. Six needles were removed with tweezers from each seedling. The 24 needles (6 needles * four seedlings)

were put into a 10 mL plastic vial with screw top to constitute one conductivity sample. Five samples plus one blank sample were taken per replicated treatment. Thus, a total of 240 conductivity samples and 48 blank samples were collected. Measurements and relative conductivity calculations were done following the same procedure as used for the field experiment.

Outplanting:

Following the heat treatments, the seedlings were shipped by over-night courier to the Red Rock Research Station. Four shipments on consecutive days were made. Seedlings were outplanted in two plots (Plot I and Plot NI) with a completely randomized design. Plot I could be irrigated, while Plot NI could not. All treatments were equally represented in both plots.

Results

Field Experiment

Environmental conditions

Solar irradiance was recorded continuously for the period from 21:00 hours on June 8 to 21:00 hours on June 14, 1992 (Figure 3). The first day (June 9) was cloudy and rain fell most of the day. The weather then improved for the remainder of the test, and days 3 to 5 were sunny with cloudy periods.

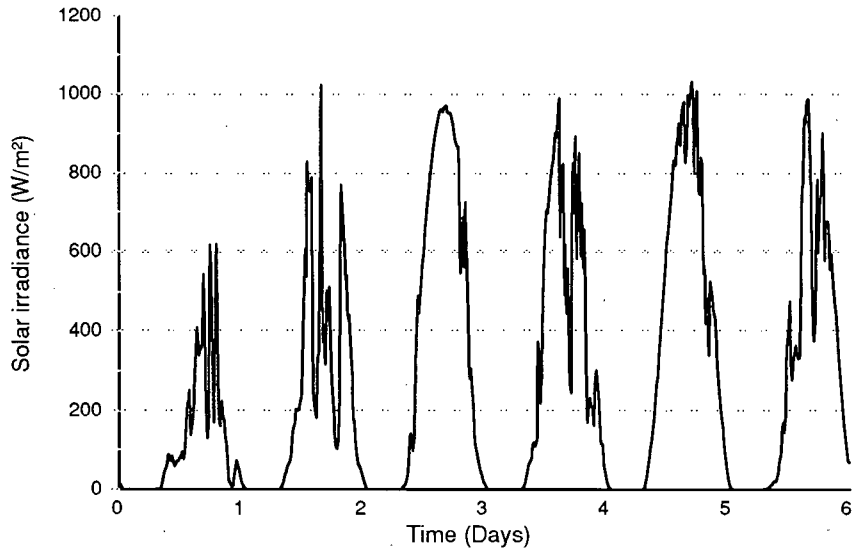

Figure 3. Solar irrradiance recorded on site during the field experiment period. Data recording stared at 21:00 h on June 8, 1992 and continued for 144 h.

Wind speed recording started only on the second day because of a technical problem (Figure 4). The wind increased as the experiment progressed and reached a maximum of about 5 $m \cdot s^{-1}$ during the fifth day.

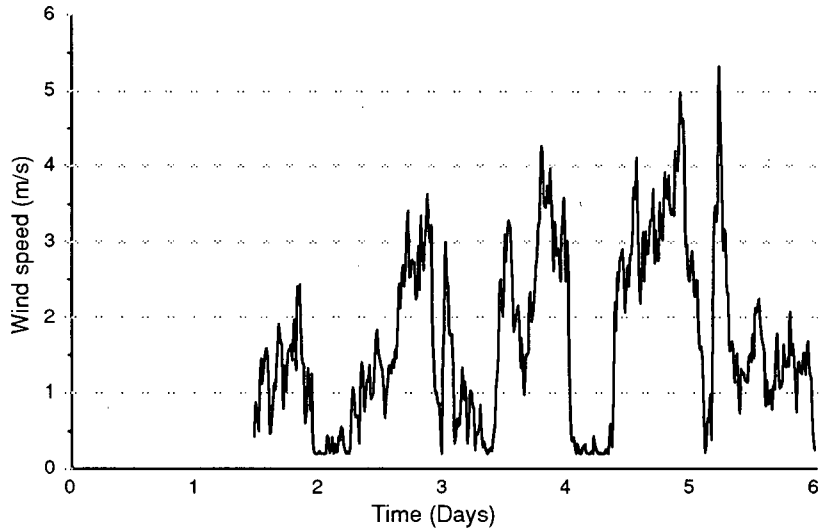

Figure 4. Wind speed recorded on site during the field experiment period. Data recording started on June 10, 1992. Zero on the X - axis represents 21:00 h on June 8, 1992.

Seedling box temperatures

Temperatures in the seedling boxes were also monitored continuously from 21:00 h on June 8 to 21:00 h on June 14, 1992, i.e. for 144 h. The minimum temperatures were generally reached between 06:00 and 07:00 hours in the morning (Figure 5) while the maximum temperatures occurred approximately between 16:00 and 17:00 h (Figure 6). For most treatments, the two shelters of the same material reached the minimum and maximum temperatures at the same time, or within 30 min. of each other. The daily minimum box temperatures for each treatment followed the same pattern. It can also be seen that the first two full days (June 9 and 10) were

substantially cooler than the following days. Minimum and maximum temperatures for June 8 are not included, since monitoring only started at 21:00 hours on that day.

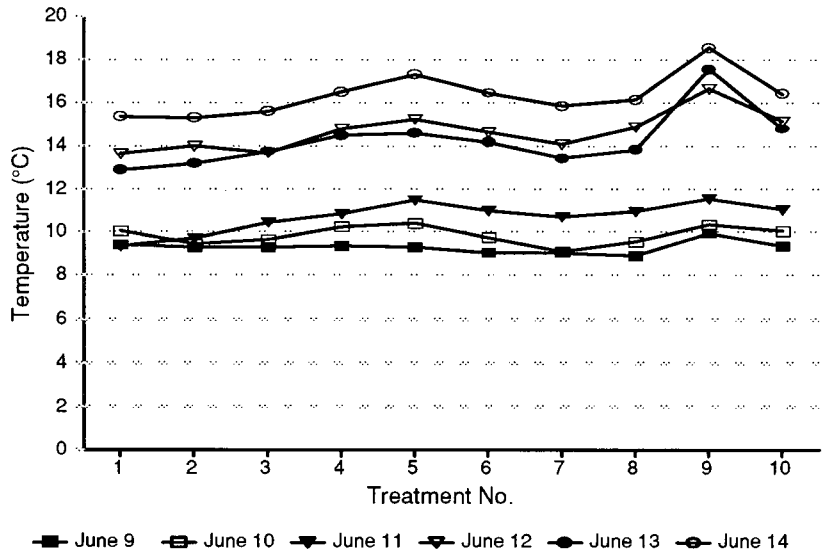

Figure 5. Daily minimum seedling box temperatures for the 10 treatments in the field experiment.

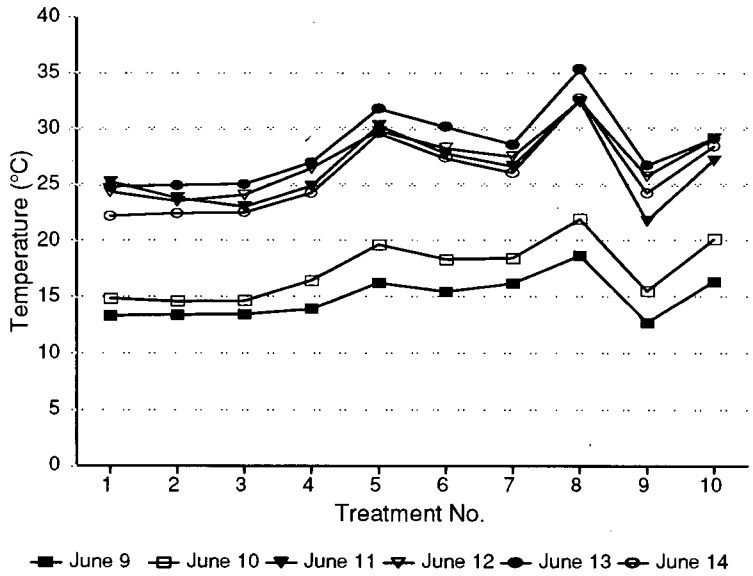

Figure 6. Daily maximum seedling box temperatures for the 10 treatments in the field experiment.

The daily maximum box temperatures for the ten treatments exhibited the same general pattern as did the minimum temperatures; the exceptions being that treatments 8 and 9 are reversed. Treatment 8 is the unprotected seedling box which predictably had the highest daily maximum temperature, while Treatment 9 is the FIST canopy which usually had the lowest daily maximum. This can be expected since the FIST canopy is insulated and the inside temperature increases at a slower rate. On the other hand, it also decreases slower at night which is reflected in the highest minimum temperatures (Figure 5).

There is no apparent reason for the unexpectedly high maximums for treatments 1 and 2 on June 11. A rather abrupt increase in the temperatures occurred between 16:30 and 16:45 when the average (of 2 shelters) maximums for treatments 1 and 2 increased by 3.1°C and 1.6°C, respectively. These steep increases do not follow the general pattern which tends to be a gradual increase. The sample trees were removed from the boxes and planted in the morning of that day and the boxes were not disturbed after that.

The heat sum in degree-hours for each treatment was calculated using a base temperature of 10°C[10]. Thus, Heat sum = (T-10°C)*No. of h at the temperature T. For example, if the temperature in a seedling box was 15°C for four hours the heat sum would be 20 degree-hours. The heat sums are shown in Table 4. Bonferroni's multiple range test was applied to determine which treatments were significantly different. The outplanted seedlings were exposed to less heat than these figures show since the samples were removed gradually over a period of six days. An analysis of variance of heat sums indicated significant differences among the treatments.

[10] The base temperature of 10°C was chosen because it is also the maximum temperature recommended by the British Columbia Ministry of Forests for seedlings in storage.

Table 4. Heat sums after 144 h for Treatments #1-10. Heat sums with the same letter are not significantly different, $\alpha = 0.05$, using Bonferroni's multiple range test.

Treatment No.	Test Material	Status	Heat sum (°-h)
1	Lightweight Silvicool® 3	New	844.9^{f}
2	Standard Silvicool® 3	New	840.51^{f}
3	Original Silvicool® type	New	832.22^{f}
4	Econocool	New	985.11^{def}
5	Silvicool® 2	2-years old	1262.25^{ab}
6	Original Silvicool® type	7-years old	1116.84^{bcd}
7	Original Silvicool® type	1-year old	1045.55^{cde}
8	Unprotected box	New	1373.03^{a}
9	FIST Canopy	New	1025.23^{cde}
10	Silvicool® 2	3-years old	1193.29^{bc}
cf.	Ambient air temperature in Stevenson Screen		912.68^{ef}

Treatments 1, 2 and 3, all new tarpaulins, have the lowest heat sums. The tarpaulin in treatment 4 was also new but was not as effective in resisting heat transfer. The heat sum for the unprotected box (Treatment 8) is, as expected, the highest. Silvicool® 2 in Treatments 5 and 10, incorporated a layer of black plastic between the metalized mylar surface and the scrim. The heat sums of these two treatments are only slightly better than the unprotected box. The relative differences between the heat sums are shown in Table 5 as the ratio between the heat sum for a treatment and the unprotected seedling box heat sum. The figures show the best treatments, three new tarpaulins, allow a heat accumulation that is less than 75% of the heat accumulated in an unprotected box.

Heat sums in degree-hours for seedlings that were planted in the sample plot are shown in Table 6, and graphically in Figure 7. These heat sums are calculated from the time the seedlings in each treatment were put into storage (21:00 h on June 8 for all) until the time they were withdrawn

on the day they were planted. Since the seedlings from the various treatments were planted in a random order each day, the times of removal from the shelters varied. Time 1 represents the time before the seedlings were put into the shelters.

Table 5. Relative difference in heat sums between treatments after 144 h. Ratio based on unprotected seedling box heat sum = 1.000.

Treatment No.	Treatment	Status	Ratio	Rank
1	Lightweight Silvicool® 3	New	0.748	3
2	Standard Silvicool® 3	New	0.745	2
3	Original Silvicool® type	New	0.742	1
4	Econocool	New	0.815	4
5	Silvicool® 2	2-years old	0.948	9
6	Original Silvicool® type	7-years old	0.877	7
7	Original Silvicool® type	1-year old	0.843	6
8	Unprotected box	New	1.000	10
9	FIST Canopy	New	0.835	5
10	Silvicool® 2	3-years old	0.915	8
cf.	Ambient air temperature in Stevenson Screen		0.773	

Table 6. Heat sums by treatment for seedlings planted in sample plot. Heat sum accumulations started at 21:00 h on June 8 and finished when seedlings from each treatment were removed from the shelter on each planting day. Time 1 has a heat sum of zero for all treatments. See also Figure 7.

Treatment No.	Time 2	Time 3	Time 4	Time 5	Time 6
1	41	94	332	511	725
2	36	87	318	486	751
3	52	101	278	487	712
4	47	130	334	570	892
5	73	189	465	761	1114
6	57	158	462	659	960
7	51	192	359	628	924
8	101	202	531	850	1156
9	54	117	314	585	929
10	117	231	417	744	1025

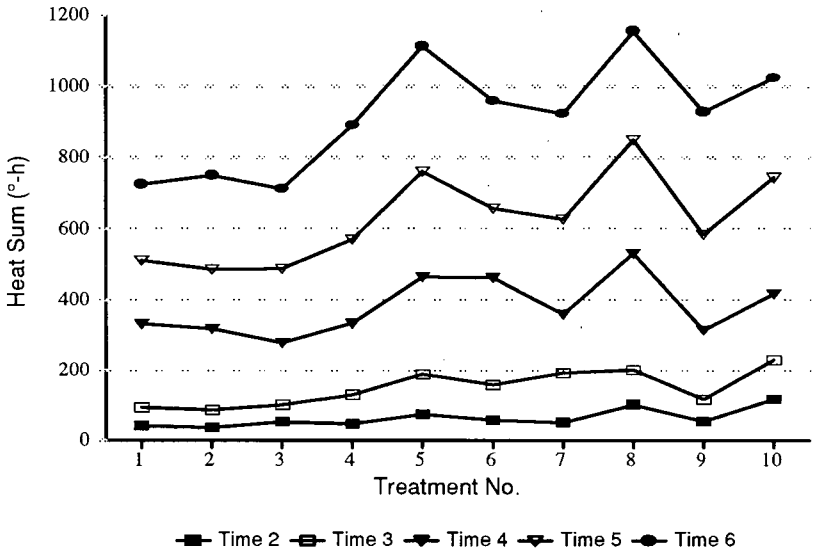

Figure 7. Heat sums for seedlings planted in sample plot, by treatment. Heat sum accumulations started at 21:00 hours on June 8 and finished when seedlings from each treatment were removed from the shelter on each planting day. Time 1 has a heat sum of zero for all treatments.

Electrical conductivity

Relative conductivities (RC) for all 720 samples ranged from 5.68% to 12.35%, with an average of 8.39% (std. dev. = 1.17). The analysis of variance for the variable Relative Conductivity (RC) is shown in Table 7. Interaction between Time and Treatment is significant at the $\alpha = 0.05$ level, and is presented in Figure 8.

There is no pattern to the interaction between Times and Treatments shown in Figure 8. Treatments were therefore analyzed individually for the time factor by treatment. The relative conductivities by time and treatment are shown in Table 8, and in Figures 9 and 10.

Table 7. The analysis of variance (ANOVA) of the calculated Relative Conductivity of needles in the field experiment.

Source	DF	Mean Squares	F Value	Pr > F
Block (B)	5	1.66	1.30	0.2540
Time (T)	5	4.73	3.70	0.0028
T*B	25	0.61	0.47	0.9865
Treatment (TR)	9	5.38	4.21	0.0001
T*TR	45	3.46	2.71	0.0001
B*TR	45	1.16	0.91	0.6398
T*B*TR	225	1.01	0.79	0.9715
Sampl. Error	360	1.28		
Total	719			

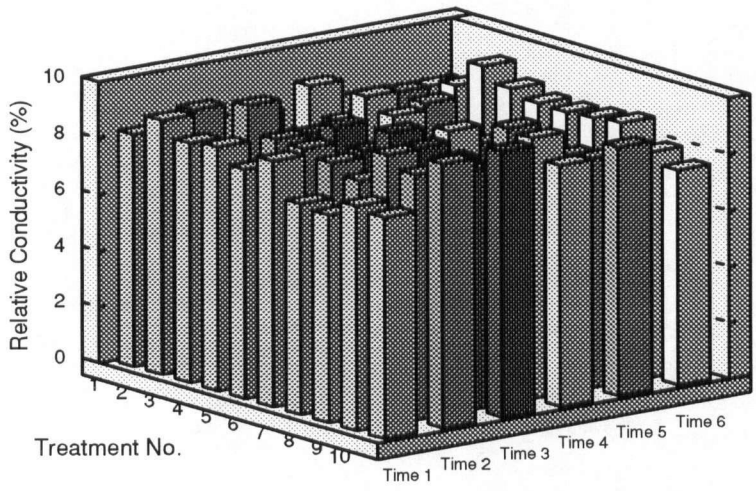

Figure 8. The percent relative conductivity for all treatments and times. The interaction Treatment*Time is significant (p = 0.0001).

Table 8. Calculated Relative Conductivities (RC) (%) of needles for Time 1 to 6, and rank of tarpaulin efficiencies. Treatments were analyzed individually. The means without any letter BY ROW are not significantly different. The means with the same letter BY ROW are not significantly different.

Treatment No.	Time 1	Time 2	Time 3	Time 4	Time 5	Time 6	cf. Tarp. Rank
1	8.256	8.757	8.253	8.850	8.133	8.089	3
2	9.048[a]	7.409[b]	7.670[b]	7.889[ab]	8.540[ab]	8.390[ab]	2
3	8.514	9.482	8.092	9.044	8.432	9.364	1
4	8.669[a]	8.555[a]	8.622[a]	8.630[a]	7.369[b]	8.864[a]	4
5	8.159	8.519	8.073	7.426	7.566	8.612	9
6	8.752	8.296	8.996	8.667	8.387	8.586	7
7	7.508[ab]	7.960[ab]	8.733[a]	7.244[b]	7.523[ab]	8.659[ab]	6
8	7.390[b]	9.142[a]	8.573[ab]	8.552[ab]	7.989[ab]	8.781[ab]	10
9	8.002	8.741	8.504	9.266	8.168	8.011	5
10	7.878[bc]	9.413[a]	9.460[a]	8.585[abc]	8.846[ab]	7.645[c]	8

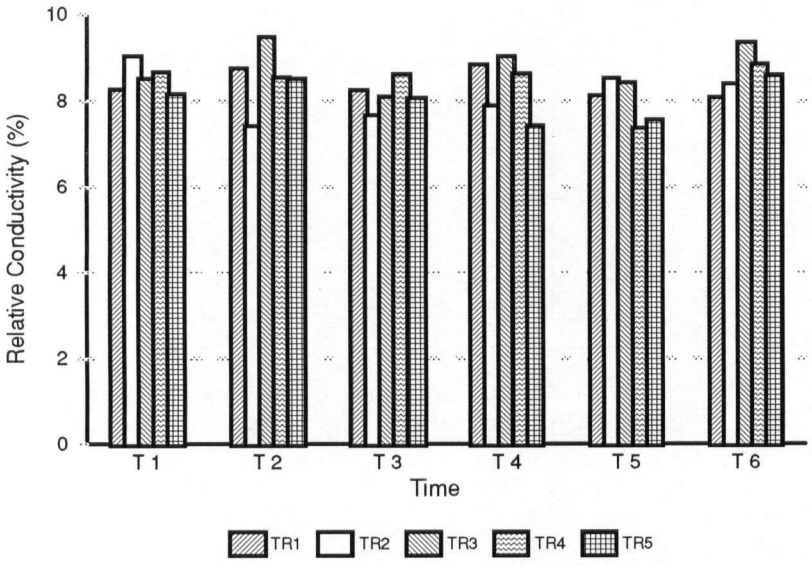

Figure 9. Relative Conductivities (%) of needles for Treatments 1 - 5 in the field experiment, when treatments are analyzed individually [T1...T6 = Time 1...Time 6].

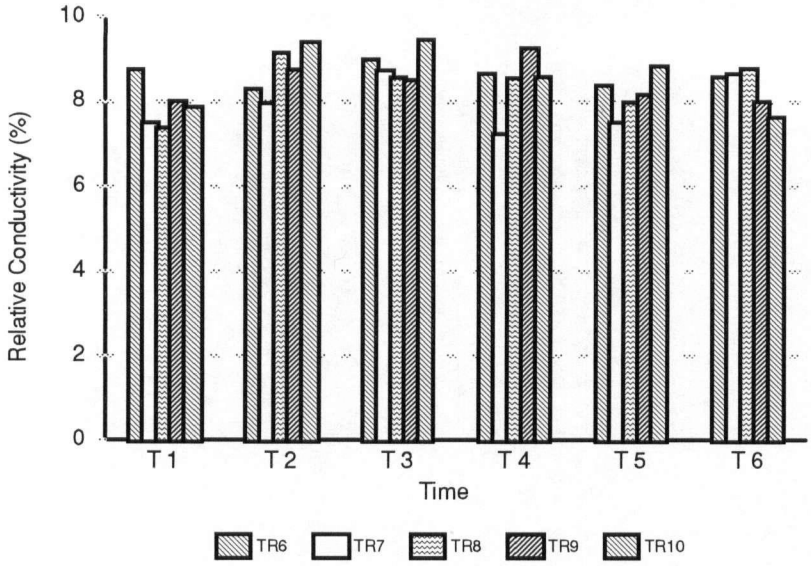

Figure 10. Relative Conductivities of needles for Treatments 6 - 10 in the field experiment, when treatments are analyzed individually [T1...T6 = Time 1...Time 6].

There are no significant differences in relative conductivity (RC) between Times in Treatments 1, 3, 5, 6, and 9. In Treatment 2, the RC at Time 2 and 3 are significantly lower than the RC at Time 1 only. As shown in Figure 9, the RC decreased sharply at Time 2 and then slowly recovered. A similar pattern can be seen in Treatments 4 and 5 although the decrease happened in Times 5 and 4, respectively, and the differences in RC were not significant in Treatment 5. In Treatment 4 the RC at Time 5 was significantly lower than the other RC's. In Treatment 7, the RC at Time 4 was the only one significantly lower than the RC at Time 3. In Treatment 8, only the RC in Time 1 was significantly lower than the RC in Time 2. Finally, in Treatment 10 the RC was significantly higher at Times 2 and 3 than at Times 1 and 6. The RC at Time 6 was also significantly lower than the RC at Time 5. In short, there is no pattern to the RC changes by treatment.

Seedling survival and growth

The survival of the seedlings was very high. Of the 2880 seedlings in the field experiment, only seven had died after three growing seasons. There was no discernible pattern in terms of Times, Blocks and Treatments for the dead seedlings.

Terminology

Seedling stem volume (V) was estimated by the formula:

$V = diameter*diameter*height*0.2618.$

where diameter = root collar diameter in mm, and height = total height in mm.

This formula assumes that seedling stems are cone-shaped.

All volumes shown are LSMEAN (Least Square Mean) as calculated by the SAS/STAT[11] program, and are expressed in mm^3.

V1 = seedling volume at time of planting. V1 is used as covariate in the statistical analysis to remove the influence different seedling sizes may have had on the volume growth of the seedlings.

V2 = seedling volume after the first growing season;

V3 = seedling volume after the second growing season;

V4 = seedling volume after the third growing season;

[11] SAS® for Windows (Version 6.08), SAS Institute Inc., Cary, NC., 1993.

G1 = V2 - V1; growth in the first growing season only

G2 = V3 - V2; growth in the second growing season only;

G3 = V3 - V1; growth in first and second growing seasons;

G4 = V4 - V3; growth in the third growing season only;

G5 = V4 - V1; growth in the first, second and third growing seasons;

Time = length of storage in days;

Time 1 = no storage, --- seedlings used as Controls;

Time 2, 3, 4, 5, 6 = seedlings stored for 2, 3, 4, 5, and 6 days

Block = position in sample plot (B1, B2...B6);

Blocks consisted of nursery beds with four seedlings planted abreast;

Treatment = protection method (Treatment 1,2...10);

NOTE: Each Block (B 1,2...6) was randomly divided into six Time zones (T 1, T2, ...T6). Each Time zone was randomly divided into ten Treatment parts (TR 1, TR2,...TR10). Each Block consisted of 480 seedlings; each Time zone consisted of 80 seedlings, and each Treatment consisted of eight seedlings (four seedlings in each of two shelter setup using the same tarpaulin material) per Time zone per Block.

The General Linear Method (GLM) method was used to statistically analyze differences in seedling volume (V) and growth (G) between treatments. The GLM procedure uses the method

of least squares to fit general linear models, and must be used for analysis of variance and covariance when data is unbalanced (e.g., from dead seedlings).

The **first step** in the analysis was carried out for growth in the second growing season (G2), and for total volume after the second year (V3). The analyses were based on the statistical model shown in Table A-3, Appendix II. This is the most extensive model which includes all factors that can be analyzed.

The results showed significant interactions between Time, Block, and Treatment in both V3 and G2. To trace the source of the interactions, the V3 LSMEAN volumes for the these factors were plotted as shown in Figure 11 and Figure 12.

Figure 11 shows the Blocks in the order they were established in the plot, with Block 6 the most southerly. Time 1 seedlings were planted without having been stored, and are used as Controls. All other seedlings were therefore compared to the Time 1 seedlings. In Figure 11, *shading* is used to show those Treatments and Times for which the V3 LSMEAN volumes were larger than those of the Controls. In other words, if the seedlings grew better after having been field stored for some length of time, that Treatment and Time in Figure 11 is shaded. The numbers in Figures 11 and 12 show the percentage larger (*shaded*), or smaller (*unshaded*) that the V3 LSMEAN seedling volumes were, compared to the Time 1 seedlings (Control). In Figure 11, the ten Treatments are plotted in sequence for each Time, while in Figure 12 the five Times are plotted in sequence for each Treatment.

TIME Comp.	TR #	B6 V3	B2 V3	B4 V3	B5 V3	B3 V3	B1 V3
T1 - T2	1	5	68	6	29	13	4
T1 - T2	2	9	66	3	57	10	45
T1 - T2	3	14	56	12	0	10	3
T1 - T2	4	54	25	3	68	7	12
T1 - T2	5	20	62	13	91	2	11
T1 - T2	6	9	44	2	18	28	49
T1 - T2	7	8	27	22	77	14	7
T1 - T2	8	14	58	32	68	2	9
T1 - T2	9	18	65	16	34	5	5
T1 - T2	10	10	53	8	63	13	5
T1 - T3	1	42	39	18	4	8	4
T1 - T3	2	48	30	21	8	21	23
T1 - T3	3	16	33	34	14	2	37
T1 - T3	4	8	44	12	16	19	1
T1 - T3	5	60	22	24	23	35	1
T1 - T3	6	20	12	9	5	24	89
T1 - T3	7	29	47	46	15	18	13
T1 - T3	8	59	13	39	27	4	7
T1 - T3	9	42	12	48	21	30	33
T1 - T3	10	33	6	25	18	23	11
T1 - T4	1	50	64	10	49	38	7
T1 - T4	2	41	56	7	45	32	23
T1 - T4	3	57	9	1	20	4	69
T1 - T4	4	44	19	19	47	20	5
T1 - T4	5	62	50	8	56	12	29
T1 - T4	6	51	20	8	16	70	62
T1 - T4	7	63	9	23	58	5	44
T1 - T4	8	58	40	19	39	25	17
T1 - T4	9	64	31	39	31	21	3
T1 - T4	10	60	38	41	70	82	30
T1 - T5	1	57	23	16	14	37	3
T1 - T5	2	61	20	44	4	32	2
T1 - T5	3	66	10	16	14	41	6
T1 - T5	4	26	51	21	27	42	2
T1 - T5	5	61	29	1	48	4	21
T1 - T5	6	53	8	20	14	27	82
T1 - T5	7	55	50	8	18	3	22
T1 - T5	8	74	23	10	9	24	0
T1 - T5	9	51	6	37	20	9	34
T1 - T5	10	60	4	20	67	2	21
T1 - T6	1	10	55	32	17	31	6
T1 - T6	2	36	65	16	21	3	2
T1 - T6	3	15	55	14	8	6	34
T1 - T6	4	25	50	5	26	29	13
T1 - T6	5	4	69	33	9	100	15
T1 - T6	6	46	52	17	6	48	62
T1 - T6	7	3	46	46	6	51	15
T1 - T6	8	11	32	3	15	16	11
T1 - T6	9	19	66	48	4	38	3
T1 - T6	10	14	51	13	12	86	4

TIME Comp.	TR #	B6 V3	B2 V3	B4 V3	B5 V3	B3 V3	B1 V3
T1 - T2	1	5	68	6	29	13	4
T1 - T3	1	42	39	18	4	8	4
T1 - T4	1	50	64	10	49	38	7
T1 - T5	1	57	23	16	14	37	3
T1 - T6	1	10	55	32	17	31	6
T1 - T2	2	9	66	3	57	10	45
T1 - T3	2	48	30	21	8	21	23
T1 - T4	2	41	56	7	45	32	23
T1 - T5	2	61	20	44	4	32	2
T1 - T6	2	36	65	16	21	3	2
T1 - T2	3	14	56	12	0	10	3
T1 - T3	3	16	33	34	14	2	37
T1 - T4	3	57	9	1	20	4	69
T1 - T5	3	66	10	16	14	41	6
T1 - T6	3	15	55	14	8	6	34
T1 - T2	4	54	25	3	68	7	12
T1 - T3	4	8	44	12	16	19	1
T1 - T4	4	44	19	19	47	20	5
T1 - T5	4	26	51	21	27	42	2
T1 - T6	4	25	50	5	26	29	13
T1 - T2	5	20	62	13	91	2	11
T1 - T3	5	60	22	24	23	35	1
T1 - T4	5	62	50	8	56	12	29
T1 - T5	5	61	29	1	48	4	21
T1 - T6	5	4	69	33	9	100	15
T1 - T2	6	9	44	2	18	28	49
T1 - T3	6	20	12	9	5	24	89
T1 - T4	6	51	20	8	16	70	62
T1 - T5	6	53	8	20	14	27	82
T1 - T6	6	46	52	17	6	49	62
T1 - T2	7	8	27	22	77	14	7
T1 - T3	7	29	47	46	15	18	13
T1 - T4	7	63	9	23	58	5	44
T1 - T5	7	55	50	8	18	3	22
T1 - T6	7	3	46	46	6	51	15
T1 - T2	8	14	58	32	68	2	9
T1 - T3	8	59	13	39	27	4	7
T1 - T4	8	58	40	19	39	25	17
T1 - T5	8	74	23	10	9	24	0
T1 - T6	8	11	32	3	15	16	11
T1 - T2	9	18	65	16	34	6	5
T1 - T3	9	42	12	48	21	30	33
T1 - T4	9	64	31	39	31	21	3
T1 - T5	9	51	6	37	20	9	34
T1 - T6	9	19	66	48	4	38	3
T1 - T2	10	10	53	8	63	13	5
T1 - T3	10	33	6	25	18	23	11
T1 - T4	10	60	38	41	70	82	30
T1 - T5	10	60	4	20	67	2	21
T1 - T6	10	14	51	13	12	86	4

Figure 11 (Left of center line). Percent difference between V3 volumes of T1 (Control) and T2, T3...T6, with Times plotted side-by-side (see text for explanation).

Figure 12 (Right of center line). Percent difference between V3 volumes of T1 (Control) and T2, T3...T6, with Treatments plotted side-by-side (see text for explanation).

There is a distinct difference, as shown in Figure 11, between the three Blocks 6, 2, and 4, and Blocks 5, 3, and 1. Seedlings in most treatments planted in Blocks 5, 3, and 1 grew better than the controls. The opposite was true for Blocks 6,2, and 4. This block-effect was unexpected and partly explains the Block*Time*Treatment interaction found in the statistical analysis.

The shading pattern in Figure 12 suggests that seedlings given Treatments 4 to 10, for all storage Times in Blocks 5, 3, and 1 generally appear to have grown better than the others.

When the volumes for the second growing season (G2) are plotted, in the sequence each treatment was located in a block, a very definite gradient from one end of the sample plot to the other is found (Figure 13). The regression equation, the coefficients of determination (r^2) and the standard error of the Y-estimate, are given in Table 9. Note that the sequence of treatments in each block was allocated randomly before the experiment.

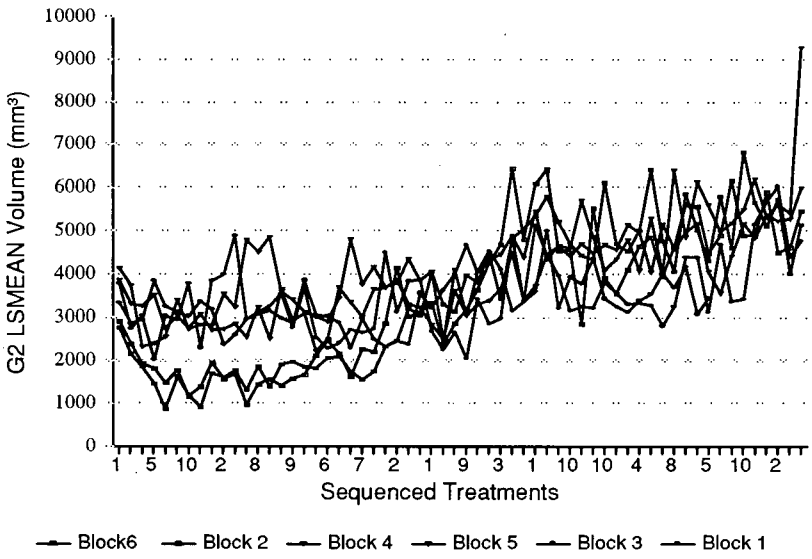

Figure 13. The mean seedling volume in the second growing season (G2) for all the treatments. The ten treatments are plotted in the same random sequence used to allocate their location in each of the six time zones in each block. See Table A-2, Appendix I.

Table 9. Data for regression equation $y = b_0 + b_1 * X_n$ based on the data of Figure 13.

Block No.	X_n	b_0 (Constant)	b_1 (Coefficient)	r^2	S_{y*x}
1	1-60	2665.121	19.141	0.271	553.626
2	1-60	1134.822	75.596	0.699	873.149
3	1-60	2761.098	23.584	0.212	801.366
4	1-60	2730.274	51.266	0.559	802.142
5	1-60	2490.419	42.688	0.654	546.707
6	1-60	917.049	84.298	0.779	790.811

Blocks 6 and 2 in particular show a very strong increasing gradient (b1) from east to west in the sample plot and also have the highest coefficient of determination (r^2) values. The estimated increase in seedling growth for these two blocks is almost six times from one end of the block to the other. This variance within the blocks was also unexpected and unpredictable, and together with the between-block variance explains the Time*Block*Treatment interaction found in the statistical analysis. The standard error of the Y-estimate (S_{y*x}), which indicates the spread of the observations around the regression line, is lowest for Block No.'s 1 and 5. The other four blocks have a standard error that is almost 50% higher. A three-dimensional representation of growth in the second growing season (G2) is shown in Figure 14. The volume data from each block are arranged in same order that the treatments were located in the plot. The increases in volume towards the west end of the blocks and the northern blocks at the eastern end are very distinct.

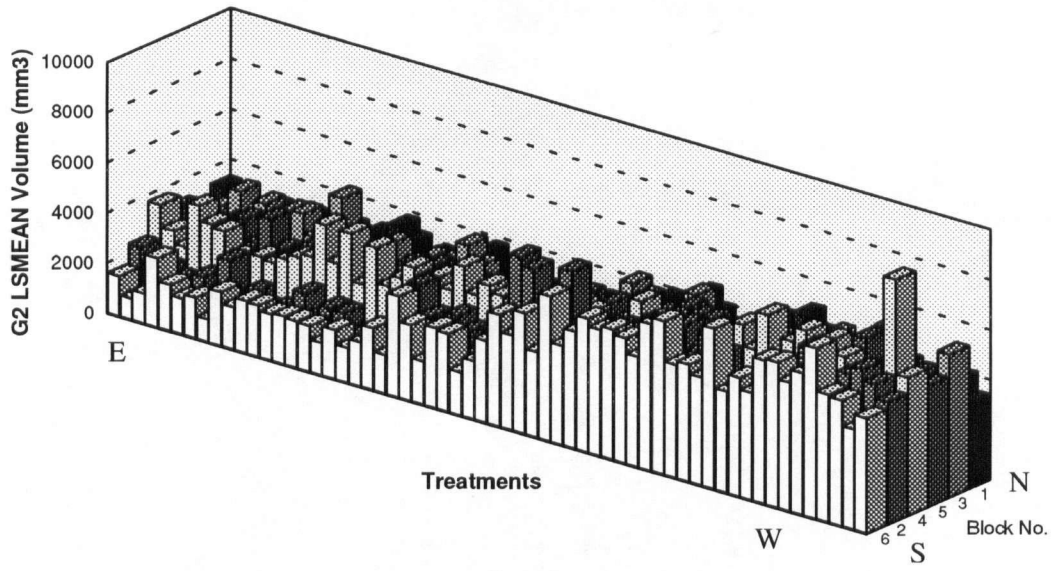

Figure 14. The mean growth in the second growing season (G2), plotted in the sequence that treatments were located in each block. Blocks are in the same order as in the plot.

The **second step** in the analysis was to determine if there were any Time by Treatment, and Time by Block effects for variable V3. This was done by including the Time*Block*Treatment interaction in the Experimental Error. The result of the covariance analysis is shown in Table A-4, Appendix II. Significant Time*Treatment, and Time*Block interactions are indicated in Table A-4. The V3 seedling volume data for Time by Treatment are shown in Figure 15. Though the Time 1 (Control) volumes are highest for some treatments, there are no significant differences between the Times for any of the treatments. The interaction stems from the variability between treatments for the different times.

The Time*Block interaction is significant. This was also suggested by Figures 11 and 12. Figure 16 shows the volumes after the two growing seasons (V3) plotted in the actual Time zone order, by block.

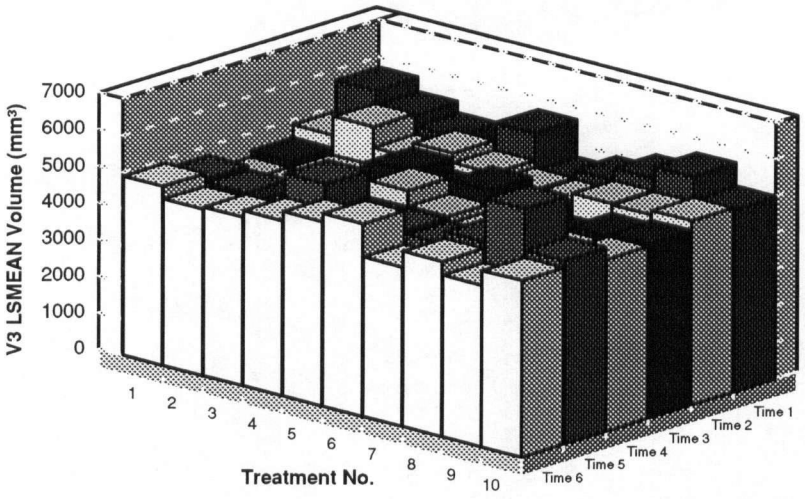

Figure 15. The mean seedling volumes after the second growing season (V3), by Time and Treatment.

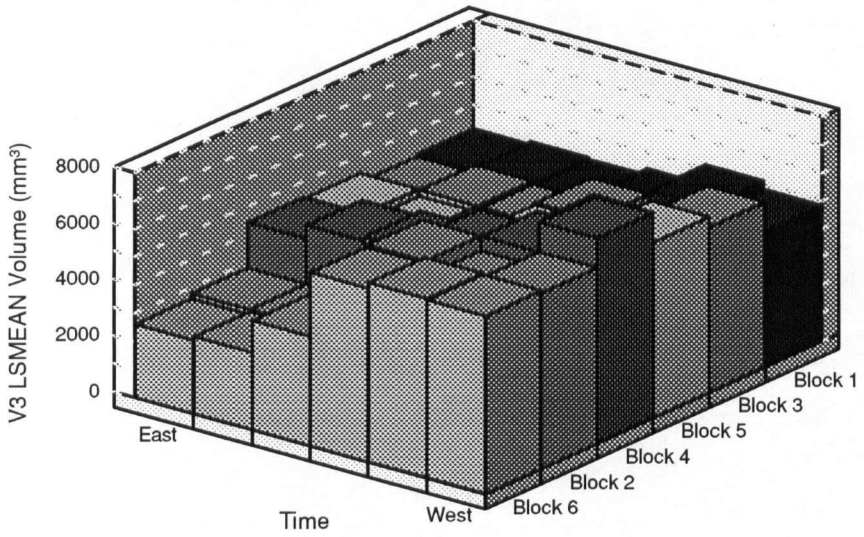

Figure 16. The mean seedling volumes after the second growing season (V3) by Time, plotted in the same sequence as they occurred in each block.

The data shown in Figure 16 indicate an increase in seedling volumes towards the west in the sample plot. The depressed volumes in the east end of Blocks 6 and 2 are noticeable. This accounts for the significant Time*Block interaction.

The **third step** in the statistical analysis used data obtained after three growing seasons. Each block was analyzed separately since each contained complete sets of Time and Treatment combinations. The statistical model for the covariance analysis of Block 1 is shown in Table A-5, Appendix II. The same model was used for all the blocks. The combined volume growth in the first two growing seasons (G3), the volume growth in the third growing season only (G4), and the combined volume growth in the first three growing seasons (G5) were analyzed. The statistical model is shown in Table A-6, Appendix II, which also shows the results of the probability of significance for the various factors (α=0.05) from the GLM covariance analysis.

Significant Time*Treatment interactions were found in all blocks, except for G4 and G5 in Block 6, G4 in Block 2, and G3 in Block 5. Only Times were significantly different in these blocks (Table A-6, Appendix II). A Bonferroni T test on the variable G4 in Block 6 showed Time 1 and Time 2 had significantly higher volume than Time 3, Time 4 and Time 5. Also, Time 6 had significantly higher volume than Time 5. The volumes decreased from Time 1 to Time 5 but Time 6 had the third highest volume in this block. However, when Times are plotted in the order of block layout, as shown in Figure 17, the increase in volume follows the same pattern as was shown in Figure 14 for growth in the second year. The volume growth in the third growing season (G4) in Block 2 was very similar to Block 6 (data not shown).

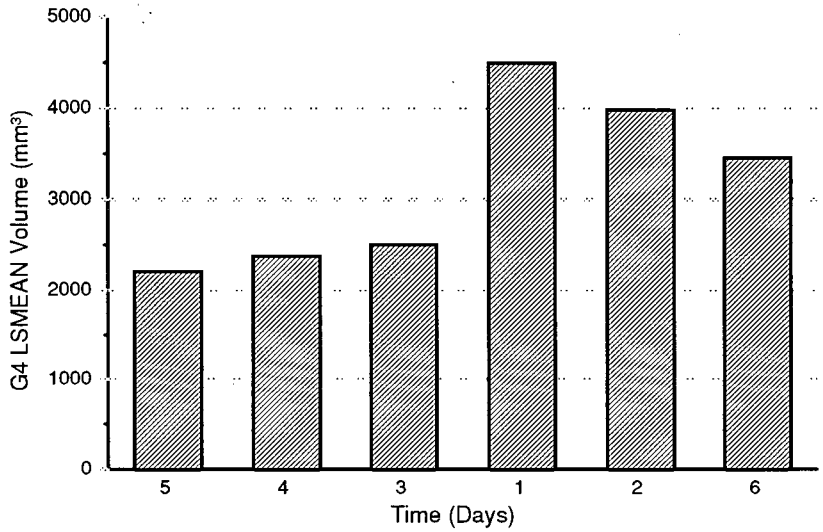

Figure 17. Growth in the third growing season (G4) for Block 6. The significant volume differences by Time are plotted by their order in the block from East to West.

The type of Time*Treatment interaction indicated in Table A-6, Appendix II, is graphically illustrated in Figure 18 for G4 in Block 1. The Times are displayed in the graph in the same random order in which they were planted in the block. There are no particular patterns to the volumes which indicate that treatments gave different results for different storage times and location in the block.

Blocks 3, 4, and 5 also had significant Time*Treatment interactions in G3, G4 and G5 (except G3 in Block 5) (Table A-6, Appendix II; Figures 19 -21). No particular pattern can be established which could explain the treatment differences for different lengths of storage and the noted interaction.

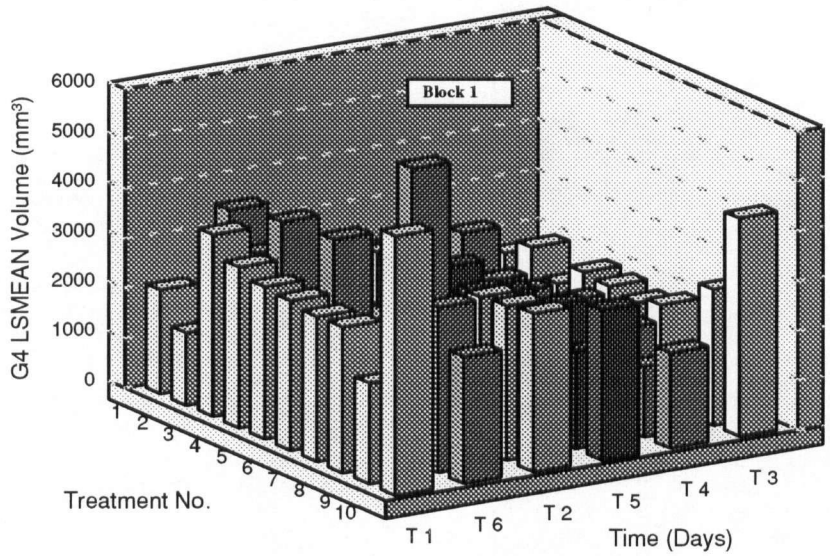

Figure 18. Seedling volume growth in the third growing season (G4) for Block 1. The treatment differences by Time are plotted by their order in the block, from East to West.

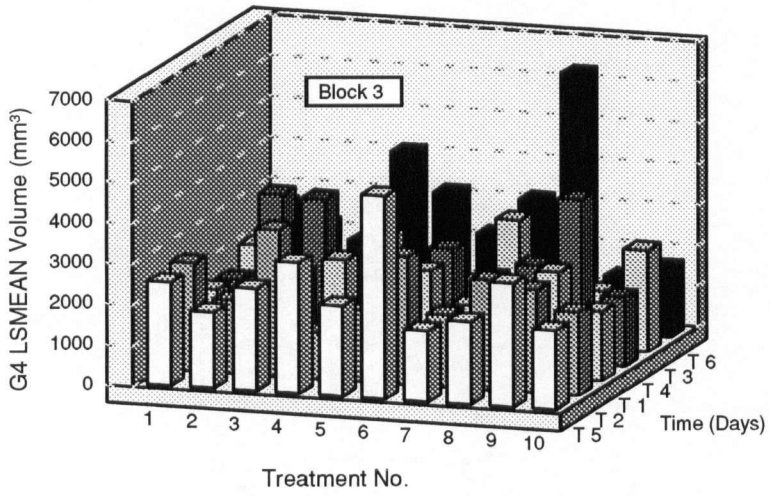

Figure 19. Seedling volume growth in the third growing season (G4) for Block 3. The treatment differences by Time are plotted by their order in the block, from East to West.

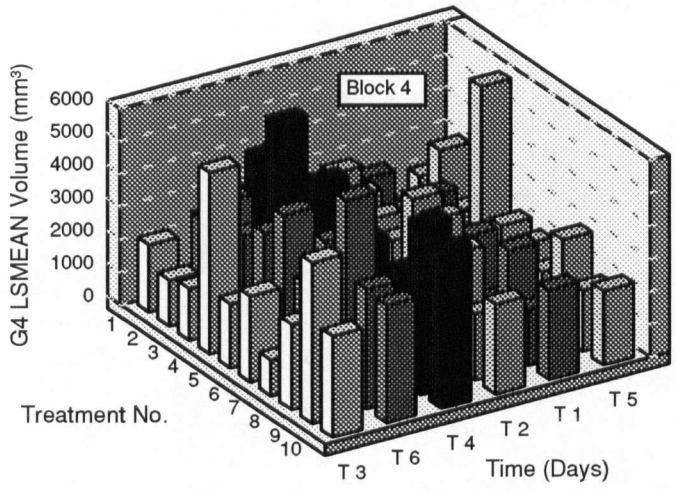

Figure 20. Seedling volume growth in the third growing season (G4) for Block 4. The treatment differences by Time are plotted by their order in the block, from East to West.

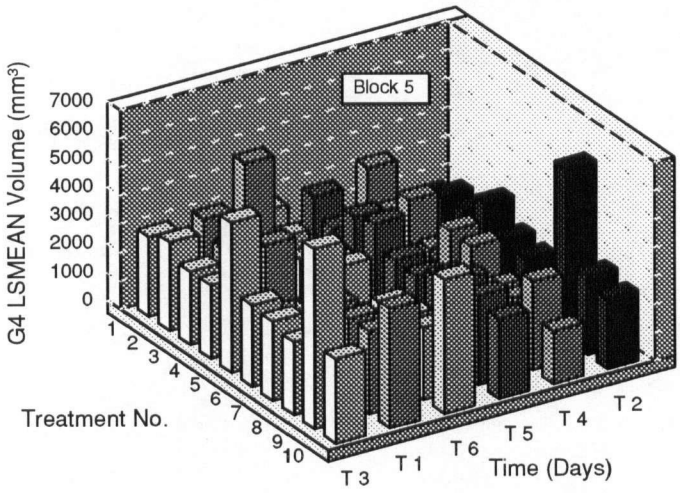

Figure 21. Seedling volume growth in the third growing season (G4) for Block 5. The treatment differences by Time are plotted by their order in the block, from East to West.

Soil and foliage analysis

Some of the results from the soil and foliage analysis are presented in Appendix IV. Tables A-10 and A-11 show the data from the soil nutrient and the foliage nutrient sampling.

Controlled Conditions Experiment

The test of the tarpaulins under controlled conditions determined the relative heat transfer characteristics between samples of seven of the tarpaulins used in the field experiment. A ratio of the heat sum above to below the tarpaulin sample was calculated and statistically analyzed (Table A-7, Appendix II). The average ratios for the samples from each tarpaulin type, obtained from the Bonferroni T test, were then expressed relative to Treatment 1. Treatment 1 had the lowest heat transfer of all the samples tested. The relative heat sum ratios are shown in Figure 22.

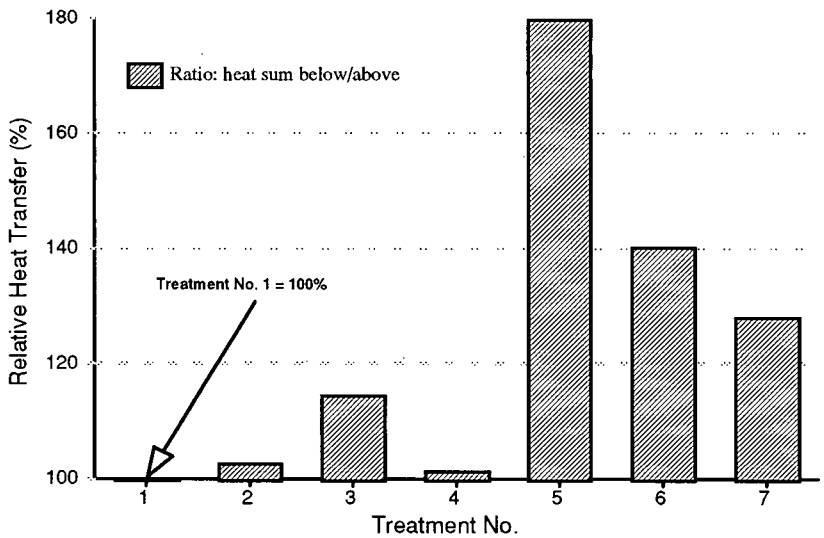

Figure 22. Ratios of relative heat transfer for pieces of reflective tarpaulins taken from Treatments 1-7 used to compare the reflective tarpaulins in the various treatments to Treatment 1 which was set to 100%. This test was carried out under controlled conditions, using floodlights as a heat source.

The four new tarpaulins used for Treatments 1-4 were found to have the lowest relative heat sums, while the tarpaulin in Treatment 5 had the highest at 80% more than Treatment 1. The results agree with those obtained in the field experiment, though Treatments 3 and 4 are reversed in this controlled condition experiment. The used tarpaulins in Treatments 5, 6, and 7 transfer significantly more heat than the new tarpaulin materials in Treatments 1, 2, 3, and 4. Treatments 3 and 7 were not significantly different, nor Treatments 6 and 7. Treatment 5 was significantly different from all the others.

Laboratory Experiment

Electrical Conductivity

The absolute values for relative conductivity were almost double those found in the field experiment, i.e. 16% vs. 8%. Statistical analysis of the data showed significant two-way and three-way interactions between Pre-conditioning, Heat, and Time (Table 10, and Figures 23 and 24).

Table 10. The analysis of variance (ANOVA) of the relative conductivity data in the laboratory experiment.

Source	DF	Mean Square	F Value	PR > F
Pre-con.	2	2.92	0.31	0.7356
Heat	3	18.41	1.94	0.1240
Time	1	91.55	9.65	0.0021
Pre-con.*Time	2	97.70	10.30	0.0001
Pre-con.*Heat	6	35.47	3.74	0.0015
Heat*Time	3	64.22	6.77	0.0002
Pre-con.*Heat*Time	6	24.36	2.57	0.0201
Model	23	39.12	4.12	0.0001
Error	216	9.49		
Corrected Total	239			

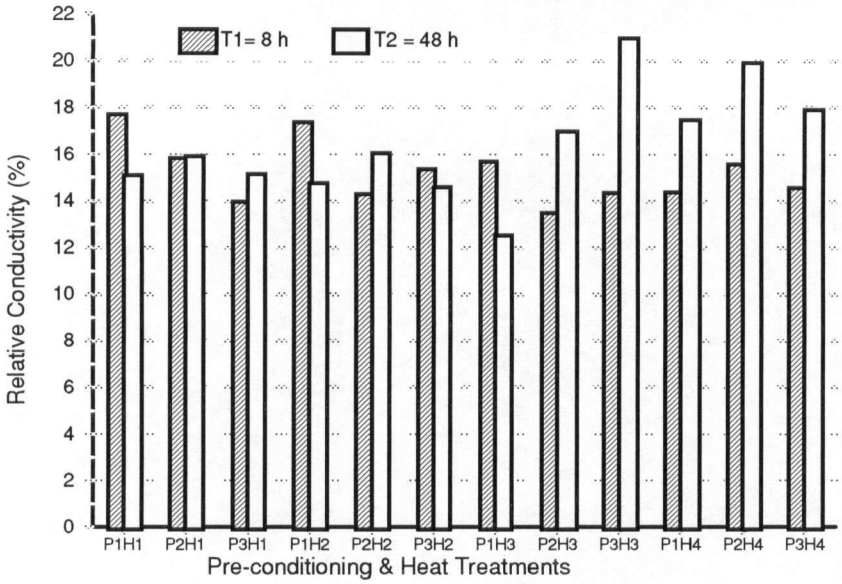

Figure 23. Relative conductivities for seedlings given heat stress treatments in the laboratory experiment. Statistically significant interaction for Pre-conditioning*Heat for two Times is shown (p=0.0201). [P1: pre-conditioning = 0 days; P2: pre-conditioning = 4 days; P3: pre-conditioning = 8 days; T1: heat stress duration = 8 h; T2: heat stress duration = 48 h; H1: heat stress temperature = 5°C; H2: heat stress temperature = 30°C; H3: heat stress temperature = 35°C; H4: heat stress temperature =40°C].

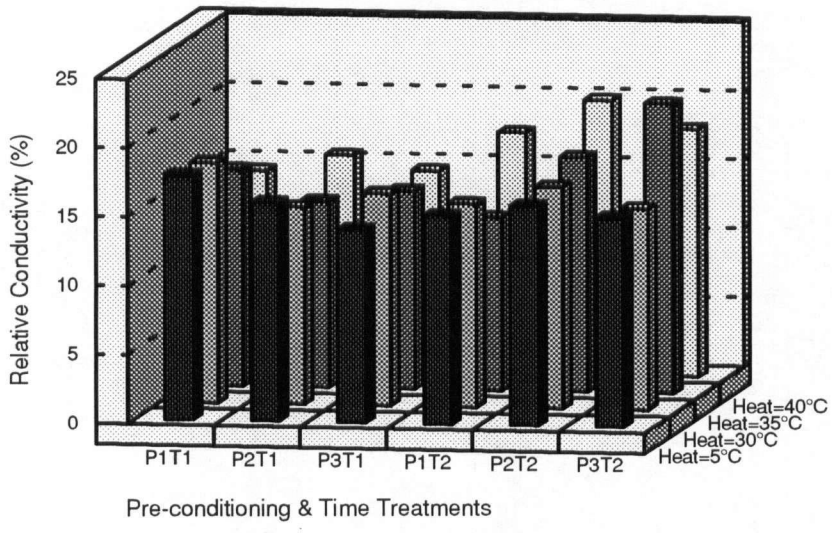

Figure 24. Relative conductivities for seedlings given heat stress treatments in the laboratory experiment. Statistically significant interaction for Pre-conditioning*Time, for four Heat treatments is shown (p=0.0201). [P1: pre-conditioning = 0 days; P2: pre-conditioning = 4 days; P3: pre-conditioning = 8 days; T1: heat stress duration = 8 h; T2: heat stress duration = 48 h]

Though there are significant interactions between the different Pre-conditioning, Heat and Time treatments, there does not appear to be a pattern of treatment effects. However, the relative conductivity did increase in the 48-hour treatments for seedlings pre-conditioned 4 and 8 days at 35°C, and for all pre-conditioned seedlings at 40°C (Figures 23 and 24).

Seedling survival and growth

One plot (Plot I) was located adjacent to the field experiment plot, and the other (Plot NI) was established about 150 meters west of the field experiment plot. All were in the same nursery field. Irrigation was to be applied to Plot I while Plot NI was not to be irrigated. Because there was sufficient rain during the summers of 1993 and 1994, no irrigation took place and therefore the two plots were exposed to approximately the same summer weather, and similar growing conditions. However, Plot NI was in a part of the nursery field where winter winds blew away the snow and as a consequence, exposed the seedlings to the weather. As a result, about 21% of the seedlings in this plot suffered winter damage in form of dead terminals during the winter of 1993/94, i.e. after the first growing season. Only about 3% of the seedlings in the Plot I had dead terminals. A data filter applied during the statistical analysis excluded seedlings coded as winter damaged.

Survival of the seedlings was very high. After the first growing season, one (1) seedling had died. There were five dead seedlings after the second growing season (less than one percent): two in Plot I, and three in Plot NI where seedlings were exposed to desiccating winds.

Growth in the first (G1) and second (G2) growing seasons for Plots I and NI were analyzed with original seedling volume (V1) as covariate. Plot I and Plot NI were analyzed together (Table A-8, Appendix II). The significant factors in the mean growth in the first year (G1) were interactions Plots*Pre-conditioning*Time and Plots*Time (Figures 25 and 26, respectively). Growth, in both plots, was higher for pre-conditioned seedlings exposed to the 48-hour heat stress treatment. In Plot I, even seedlings which had not been pre-conditioned had higher mean growth in the 48-hour heat stress treatment as compared to those exposed to the 8-hour treatment (Figure 25).

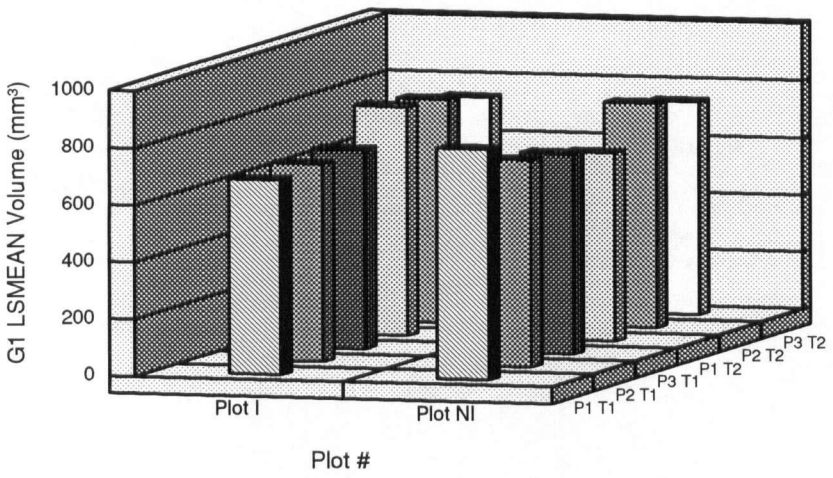

Figure 25. The mean volume growth in the first year (G1) for Plots I and NI analyzed together. The interaction Plots*Pre-conditioning*Time is significant (p=0.0462) (Table A-8, Appendix II). [P1: pre-conditioning = 0 days; P2: pre-conditioning = 4 days; P3: pre-conditioning = 8 days; T1: heat stress duration = 8 h; T2: heat stress duration = 48 h] (n=73-80).

Based on the same data as the previous graph but excluding the pre-conditioning effect, the significant Plots*Time interaction is plotted in Figure 26. As before, the mean growth was different in the two plots for the two exposure times and the growth in Plot I increased for the

48-hour heat stress treatment compared to the 8-hour treatment, while in Plot NI it decreased slightly.

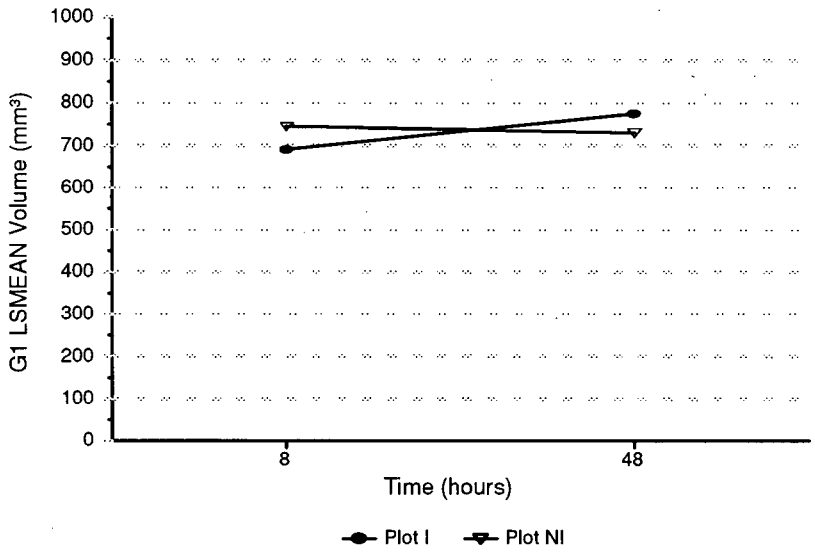

Figure 26. The mean volume growth in the first year (G1) for Plots I and NI analyzed together. The interaction Plots*Time is significant (p=0.0401) (Table A-8, Appendix II). [T1= heat stress duration = 8 h; T2: heat stress duration = 48 h] (n=231-238).

Analysis of mean growth in the second year (G2) revealed significant Pre-conditioning*Heat interaction (Figure 27). The volume of pre-conditioned seedlings decreased at 30°C compared to their volume at the control temperature, 5°C. The volume for the 4-day pre-conditioning treatment decreased further at the 35°C treatment level, but then increased sharply at the 40°C level. This was a different reaction to seedlings that had no pre-conditioning and those pre-conditioned for 8 days. The seedlings without pre-conditioning increased in mean volume at the 30 and 35°C levels but then decreased at the highest level, 40°C. Seedlings pre-conditioned for 8 days decreased at the 30°C level, increased at the 35°C and then decreased again at the 40°C level of treatment.

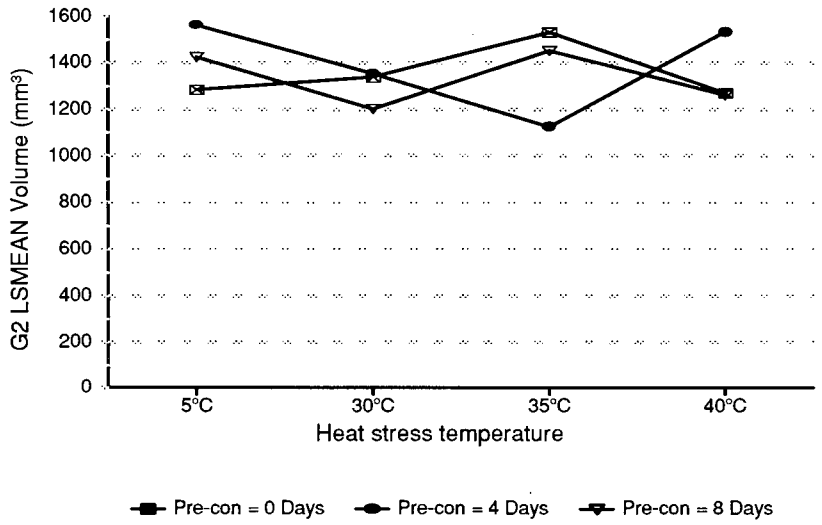

Figure 27. The mean volume growth in the second year (G2) for Plots I and NI analyzed together. The interaction Pre-conditioning*Heat is significant (p=0.0057), (Table A-9, Appendix II) (n=64-71).

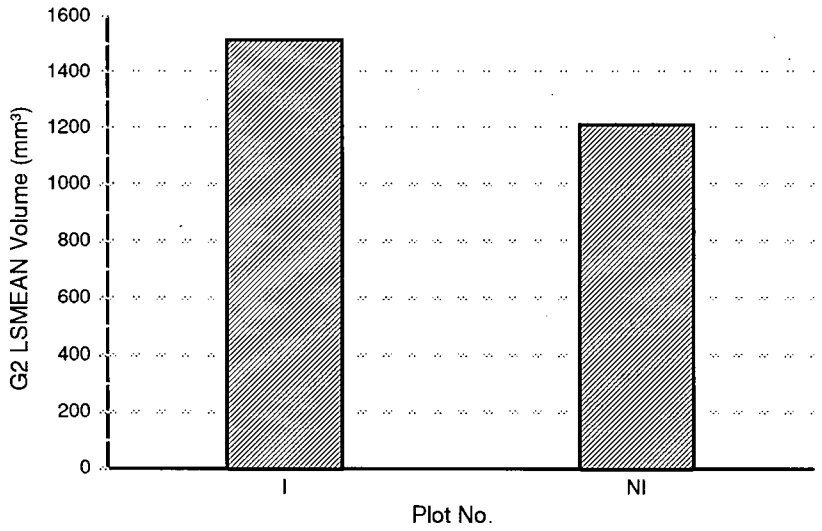

Figure 28. The mean volume growth in the second year (G2) for Plots I and NI analyzed together. The difference between Plots is significant (p=0.0001) (Table A-9, Appendix II) (n=450 for I, n=372 for NI). [Plot No. I = 'irrigated' plot; Plot No. NI = 'non- irrigated' plot; NOTE: no irrigation took place].

The mean growth in the second year (G2) was significantly different for the two plots (Figure 28). Seedlings in Plot NI grew significantly less than those in Plot I (Table A-9, Appendix II).

The data for the plots were also analyzed separately. It was found that for Plot I, only Time was significant (p= 0.0161) for growth in the first growing season. Seedlings exposed to the heat stress for 48 h had higher mean volume (data not shown). No significant differences were present for the growth in the second growing season.

In Plot NI, the interactions Pre-conditioning*Time and Pre-conditioning*Heat were significant in the first growing season. The pre-conditioned seedlings had higher growth when exposed to 48 h of heat stress compared to 8 h of exposure (Figure 29).

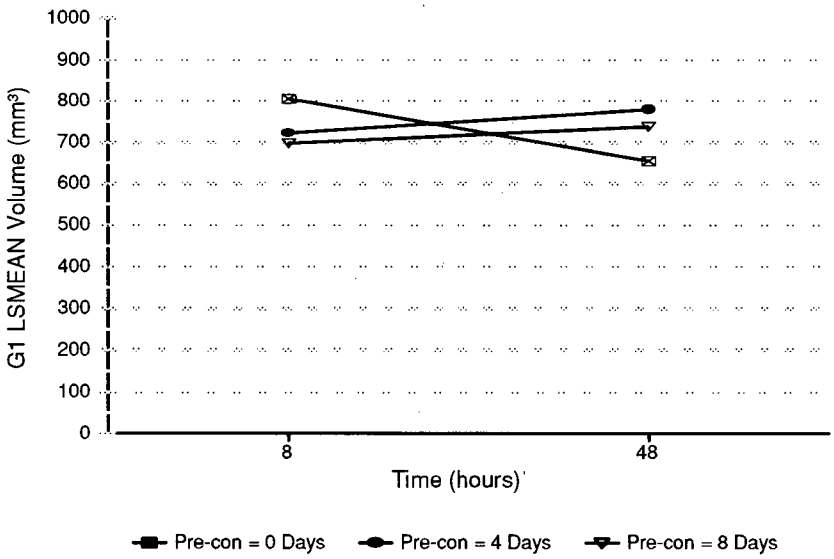

Figure 29. The interaction Pre-conditioning*Time for G1 in Plot NI. The interaction is significant (p= 0.0095). [T1: heat stress duration = 8 h; T2: heat stress duration = 48 h; G1 = growth in first growing season].

The Pre-conditioning*Heat interaction is plotted in Figure 30. The 8-day pre-conditioning is shown to have increased growth with increased heat stress temperature. The 4-day pre-conditioning effect on growth in the first growing season is not as clear. It decreases for the first two stress temperatures and increases for the highest. The 0-day pre-conditioning, i.e. control, had increased volume for the first two stress temperatures and decreased volume for the highest. Analysis of the Pre-conditioning*Heat interaction for growth in the second growing season (G2) shows results very similar to the those from the first growing season. Only the 8-day pre-conditioning treatment changed slightly as illustrated in Figure 31.

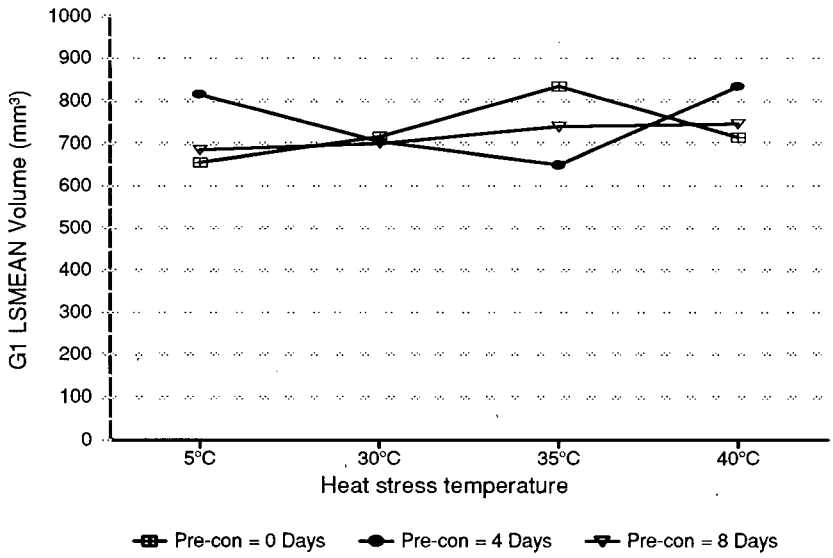

Figure 30. The interaction Pre-conditioning*Heat for G1 in Plot NI. The interaction is significant (p= 0.0438). [G1 = growth in first growing season].

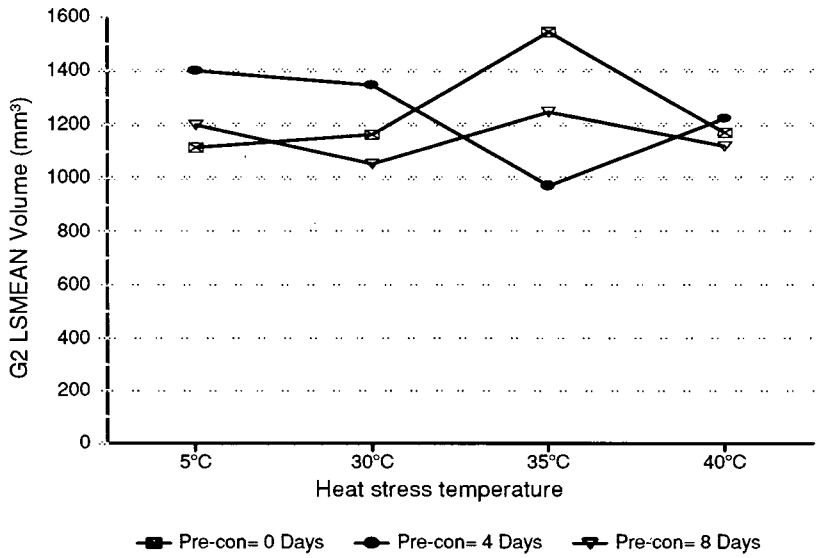

Figure 31. The interaction Pre-conditioning*Heat for G2 in Plot NI. The interaction is significant (p= 0.0367) [G2 = growth in the second growing season].

Discussion

Field Experiment

Environmental conditions

The environmental conditions recorded during the field experiment are typical of conditions in late spring, early summer at planting sites in the central interior of British Columbia. The June 9 and 10 temperatures were low compared to the following four days and below the 30-year mean daily temperature in June at Prince George Airport - 12.9°C (Environment Canada n.d.). Planting was postponed on June 9 since it rained most of the day, and the shelters with stored seedlings had been exposed to very limited solar radiation. The highest levels of solar radiation occurred during the afternoon hours while the wind speed typically peaked in the early evening hours each day (Figure 3). Some wind was present during the early part of the night (Figure 4). While solar radiation is the source of any heat buildup in the seedling boxes, the presence of a wind will affect the rate of the buildup and its subsequent decrease during the night.

Reflective tarpaulins and seedling box temperatures

Minimum seedling box temperatures varied little among treatments during the first 72 h (Figure 5), mainly because of cool day temperatures and rain on the first day seedlings were in the shelters. There was also little variation in minimum temperatures among treatments for the fourth and fifth days. The FIST (Fiberglass Insulated Seedling Transporter) had the highest minimum temperature each day of the experiment. This unit has four centimeters of rigid foam

insulation between the outside and inside fiberglass shells. The effect of this insulation can be seen in the daily maximum seedling box temperatures (Figure 6) which show the FIST had some of the lowest maximums. Because heat transfer always proceeds from a warmer to a colder region (Gates 1980) heat is transferred into the FIST canopy during the day, and out during the night, if outside temperature is lower than the inside temperature. However, because of insulation in the walls and roof of the FIST canopy, heat transfer is slower in both directions compared to heat transfer in the tarpaulin shelters.

Heat sums were calculated to quantify the amount of heat that seedlings were exposed to over time (Table 4). The heat sums for Treatments 1 - 4, all new tarpaulins, are significantly different (α=0.05) from the rest and are the lowest heat sums. Though the heat sum for the new tarpaulin in Treatment 4 is higher by about 17% than the heat sums for the other three, the difference is not statistically significant. The heat sum in Treatment 5 was not significantly different from the unprotected box which recorded the highest heat sum. The reflective tarpaulin in Treatment 5 had a layer of black plastic between the white outer surface and the scrim (Stjernberg 1991). This layer made the tarpaulin completely opaque even in a worn condition, which was the purpose of having it there, but the black plastic acted as a heat sink and retained heat conducted from the white outer layer. This resulted in a heat sum for Treatment 5 about 50% greater than for the best three new tarpaulins. Treatment 10 was a purpose-built shelter manufactured commercially and was somewhat larger in size than the other shelters. The tarpaulin material was the same as used for Treatment 5 although it appeared to be in a better condition. However, there was no statistical difference between heat sums for the two treatments.

The heat sum for the FIST (Treatment 9) was about 25% higher than for the three best tarpaulins but it is not significantly different from Treatments 6 and 7 (Table 5). The higher heat

sum for the FIST is a direct result of the *rate* of heat transfer through the canopy insulation. However, in a typical planting operation the FIST would normally be loaded with many more boxes of seedlings than was done in this experiment. The amount of heat required to increase the temperature in a fully loaded canopy would then be much higher, and as consequence the buildup of heat during the day would not be as high as was the case here. The period during the night when heat is transferred to the outside may also be reduced or not occur if the inside temperature remains below the outside. The FIST canopy can operationally be expected to have a much lower heat sum than was reported in this experiment. Von der Gönna (1990) found temperatures inside the FIST remained well below ambient air temperature when the canopy was carrying a full load of seedling boxes. In this experiment (Table 5), the FIST attained greater heat sums than ambient air temperature.

The tarpaulins used in Treatments 6 and 7 were both of the Original Silvicool® *type*, but of different ages. The difference in their heat sums was, however, not significant. There were significant differences between new and used tarpaulins but the differences within each group were small, and not significant in the case of new tarpaulins. It is shown that tarpaulin usage caused increased heat transfer and thus decreased the efficiency of the tarpaulins. However, age by itself is not necessarily a good indicator of tarpaulin efficiency. It is the quality of the material, and the abuse it has undergone which will determine the efficiency of the tarpaulin.

Heat sums for outplanted seedlings were different because seedlings were withdrawn daily, and at different times during the day for each treatment as a result of the randomization process (Figure 7). The first two days of storage did not add much to the heat sums but by Time 4, the differentiation between treatments was clearly established. The heat sum increments after Time 4 are very similar in size for all treatments. The heat sum differences between Treatment 8

(unprotected box, i.e. control) and Treatments 5 and 10 are small. If high heat sums, to a base temperature of 10°C, were to induce physiological perturbations in the seedlings one would expect this to be displayed in relative conductivities, and perhaps in survival and growth in Treatments 8, 10 and 5.

Electrical conductivity

The relative conductivity test determined if cell membranes were damaged by the heat buildup (heat sums) in the seedling boxes. Damaged cell membranes would allow the cell contents to leak out. This would be indicated by an increase in the electrical conductivity of the leachate. This technique is commonly used to determine frost hardiness in seedlings (Colombo et al. 1984, Hawkins and Binder 1990) and has been used by Binder and Fielder (1995) to detect seedling heat damage. Preliminary heat exposure tests done before the field experiment indicated that a relative conductivity of above 30% should be expected when significant damage to cell membranes had occurred.

The results from the electrical conductivity measurements in the field experiment gave a relative conductivity in the range of 6% to 12% (Figure 8). While there was a statistically significant interaction between Time and Treatment, the relative conductivities were too low to signify any cell membrane damage (Table 7, Figure 8). Natural variations among the seedlings could account for the small differences in relative conductivity. Analysis of the relative conductivities for Time in individual treatments do not show a pattern that can be correlated with the heat stress received over time (Table 8, Figures 9 and 10).

Treatments 8, 10 and 5 which had the greatest heat sums by the last planting (Table 6) did not have the highest RC at Time 6 (Table 8). In fact, Treatment 3 which had the lowest heat sum had the greatest RC. This further suggests the present RC data was indicative of natural variations among the seedlings rather than physiological damage induced by the heat sums. In short, it appears the level of heat sums observed in this experiment did not result in any cellular membrane damage for seedlings of this spruce seedlot.

Seedling survival and growth

All but seven seedlings were alive after three growing seasons - a survival rate of 99.8%. No pattern relating the dead seedlings to any Time or Treatment is discernible. Three year survival rates of this order would suggest the heat treatments had induced no physiological perturbation in the stock. Mortality of 1 to 2 percent is usually expected in this common garden at Red Rock Research Station in the first season (C. Hawkins, pers. comm., May 1994). High survival as observed could result if the seedlot had a high degree of heat tolerance, if it was of exceedingly high physiological quality at the time of planting, or if the heat treatments were of no physiological significance. The best or any hypothesis cannot be selected at this time.

Analyzing the growth of the seedlings presented a statistical challenge. The experiment was designed as a split-plot randomized complete block with repeated measurements within the experimental units. The statistical model includes all main effects (Block, Time, Treatment) but also two-way and three-way interactions between these effects. All significant interactions must

be explained. The total stem volume after two growing seasons (V3) and the stem growth in the second growing season (G2) were chosen to start analysis.

Unfortunately, there were significant three-way interactions between Block, Time and Treatment both in G2 and in V3. Analysis showed there were distinct differences between the blocks from south to north in the plot (Figures 11 to 14). The plot was established in a bareroot forest seedling nursery field where soils have been regularly cultivated, and where there were no visual differences among blocks. However, when volumes for the treatments are compared to the volume of the control there are obvious differences between the blocks. In an examination of the growth in the second growing season there was no pattern (Figures 11 and 12) until the treatments were plotted in the same sequence that was randomly selected for the outplanting in the blocks (Figures 13 and 14). The data clearly indicate the increased growth toward the west end of the blocks. This was confirmed by a regression analysis (Table 9). An obvious outlier in Figure 13 represents data for a number of seedlings that grew very well. The data indicate that there were more effects from micro-sites than from any heat treatment or storage time.

The analysis with the three-way interaction effect included in the error term still produced significant Time and Treatment interaction, and Time and Block interaction for the total volume after two growing seasons (V3). However, when the least-square-mean volumes are plotted in the same order as the treatments were planted, the variations within the blocks are reduced (Figures 15 and 16). This indicates that there is an effect dependent on where in the plot the treatment was located, rather than what the treatment - time combination was. Again, microsite overrode all else in the analysis of the data.

When more data were collected in year three, the growth in the third growing season (G4) was analyzed block by block. Also included in this analysis was growth in the first two growing seasons (G3) and growth in the first three growing seasons (G5). Time and Treatment interactions were present in most blocks (Figures 17 to 21). However, there were no significant interactions in Block 6 (Figure 17), and the growth in the third growing season confirms the previous trend of increasing growth towards the west part of the plot. Not unexpectedly, the analysis of G4 and G5 further confirmed the observation of the first two analysis: planting location had a far greater effect than did Treatment and storage duration. Therefore, it was decided to do soil and foliar analysis on the plots to see if any site related growth differences were related to possible nutrient deficits / excesses.

Soil and foliage analysis

Changes in the soil and foliage samples are shown in Appendix IV, going from east to west and from south to north. Seedling growth generally increased as one moved from east to west within a block and from south to north as one moved from block to block (Figure 14). Most of the nutrients increase towards the west and north which is the same trend as the demonstrated with the seedling growth. This could also help explain microsite growth differences. A foliage sample taken at the south-east corner of the plot showed anomalous readings of iron (408 ppm) and aluminum (468 ppm). These are very high compared to the readings from seven other nearby samples that averaged 134 ppm for iron and 205 ppm for aluminum. While references to aluminum levels are not readily available, the level of foliage iron was at toxic level and is a likely cause for the depressed growth (van den Driessche 1989). The soil sample taken from that spot also showed readings of copper, zinc, iron, and manganese that were lower than the

average from 15 other samples. Total nitrogen, sulfur and organic matter were also lower in this corner than the averages from samples in other parts of the plot. This may explain why seedling growth in this corner was much lower than in the rest of the plot as seen in Figure 16. The soil sample values are not easily interpreted because of the off-setting or compensating effects different nutrients may have at different levels (R. van den Driessche, pers. comm. March 1995). These data indicate the importance of 'knowing' your site prior to planting. Ideally, soil sampling should have been done before block layout and the area mapped based on fertility. That would have allowed uniformity among the blocks and minimized micro-site differences.

Controlled Condition Experiment

The test of the tarpaulin samples resulted in the same ranking of the tarpaulins as in the field experiment (Figure 22), though two tarpaulins were switched (Treatments 3 and 4). The light spectrum for the floodlights contains more infrared light than solar radiation (Michael Norris, General Electric, pers. comm., April 1995) and this is most likely the cause for differences in the two experiments.

If deemed crucial by the silviculture industry, tarpaulins could be tested for their efficiency before decisions are made to retain or dispose of them. However, based on the present results obtained under extreme operational durations, there is no biological evidence to suggest tarpaulin quality was an issue in this experiment with a single spruce seedlot.

Laboratory Experiment

Electrical conductivity

Based on tests carried out prior to the field experiment, relative conductivities above 30% would be indicative of cell membrane damage. Because RC were low, survival high and there were no obvious treatment or storage duration effects found for the initial planting in 1992, it was postulated that the diurnal fluctuation of temperatures was pre-conditioning the seedlings prior to planting. This possibility was alluded to earlier when it was suggested that the seedlings had a high degree of heat tolerance, the heat treatments were not physiologically significant or the seedlot was of exceedingly high physiological quality. A laboratory pre-conditioning experiment was conducted to assess the possibility of a pre-conditioning phenomenon.

Relative conductivities observed were in the 15% to 20% range which indicates that there was very little, if any, damage to the cell membranes as a result of the treatments (Figures 23 and 24). These values were about 7 to 10 relative units greater than for the field trial (Figures 8 to 10). This indicates a seasonality in RC even when it is the same seedlot grown in the same nursery with only year differing. The observed relative conductivities are similar to those reported by Binder and Fielder (1995) for the similar treatment combinations. As with relative conductivities in the field experiment, natural variations among seedlings could explain the small differences that did occur and which resulted in the interactions shown in the data (Table 10). There was no demonstrated effect from the pre-conditioning treatments or from the 8-hour treatment at any temperature. A couple of readings for the 48-hour treatment appear to stand out in Figures 23 and 24, but the standard deviations were large (i.e. 19.9%± 4.1 and 21.0%±7.1). It is therefore reasonable to conclude that the heat stress treatments did not cause

significant cell membrane damage. This conclusion is supported by the results of other studies (Alexandrov 1964, Burr et al. 1993). Alexandrov (19964) found that the temperature at which heat irreversibly damaged cell components ranged from 48.5 to 56.4°C in eight different species. Burr et al. (1986) recorded much higher electrolyte leakage in cell membranes when the temperature increased from 48 to 52°C. Binder and Fielder (1988) reported that conductivities increased in needle segments in 30 and 40°C treatments after 72 h. Had the present experiment continued past 48 h increased relative conductivities could therefore have been expected.

Seedling survival and growth

The seedling survival was 99.5 % after two growing seasons which must be considered high, given the heat stress treatments. It would indicate that at least, this particular seed source of white spruce is very tolerant to heat stress. Different results were obtained in an experiment by Binder and Fielder (1995). They (Binder and Fielder 1995) recorded eight percent mortality in white spruce seedlings after one growing season when they exposed the seedlings to a 24-hour 40°C heat treatment. Their mortality increased to 56% when the duration was extended to 48 h, and 100% when heat treatment duration was 72 h and 96 h. In this experiment the maximum duration was 48 h. The length of exposure to the heat stress is important as has been confirmed in other studies (Baker 1929, Binder and Fielder 1988, Colombo and Timmer 1992, Ingram 1990).

The seedlings in this experiment were exposed to two transportation segments of 800 km each - one before the treatments and one after the various pre-conditioning and heat stress treatments. Seedlings were shipped by courier between Prince George and Vancouver in non-temperature

controlled vehicles. Transportation distances for seedlings in British Columbia and Alberta average less than 400 km, most of it in temperature controlled environment, based on a recent stock handling study. Very few seedlings are transported in excess of 1000 km in total (Stjernberg 1995). Thus, the seedlings in this experiment were exposed to extreme shipping and handling conditions, although they are assumed to be equal across treatments.

When the field experiment data for the first growing season were analyzed, the data suggested a pre-conditioning effect for seedlings at temperature exposures of up to 35°C. There are reports indicating that seedlings can withstand temperatures that normally would be fatal, if they have been pre-conditioned by higher-than-normal temperatures (Burke 1990, Colombo et al. 1992, Howarth and Ougham 1993, Koppenaal et al. 1991).

When the interaction Plots*Pre-conditioning*Time data for growth in the first growing season (G1) are plotted pre-conditioned seedlings that were heat stressed for 48 h appear to have grown better than seedlings that were not pre-conditioned (Figure 25). However, even the seedlings not pre-conditioned grew better in one plot. There were no significant differences in growth between pre-conditioned and non-pre-conditioned seedlings exposed to the 8-hour stress treatment. This would indicate that the differences lie only in the length of exposure. A significant Plots*Time interaction suggests that seedlings in the "irrigated" Plot I grew better than in the other plot, even though there was no actual irrigation (Figure 26). The distinct differences in soil nutrients within the adjacent field experiment plot allows for the possibility that there could be soil nutrient differences between the Plots I and NI also, though this was not investigated. However, the non-irrigated plot was relatively more exposed to desiccating winds, particularly in the late fall and early spring when the soil was frozen and there was no snow to offer protection. This is evident from the measured over winter foliar injuries observed in the NI

plot. Unfortunately, neither scenario can be assigned responsibility for growth differences. The length of stress exposure is an important factor as noted above. In the present experiment, seedlings with the longer 48-hour heat stress exposure had better growth than did those with the 8 h exposure.

The Pre-conditioning*Heat interaction for the second growing season growth (G2) was significant but there was no consistency between the various combinations (Figure 27). It is difficult to draw any conclusions based on this information but at temperatures at and below 30°C there was no effect of pre-conditioning. However, at 35 and 40°C there were an indication of a pre-conditioning effect, albeit without pattern. The analysis does show a significant difference between G2 in Plot I and G2 in Plot NI (Figure 28). As discussed above, the most likely cause is related to the winter injury acquired in plot NI. Plot NI had considerably more winter injury than any other of the 1993 plants at Red Rock Research Station (C. Hawkins, pers. comm., March 1995).

To eliminate the confounding differences between plots, the data for each plot were analyzed separately. This is valid because there was no irrigation done and the plots were therefore equally treated with respect to irrigation. Interpretation of results from Plot I are straightforward. Only a time effect is significant and it confirms the previous conclusion that the longer heat stress duration caused a higher growth. However, it does not explain why. It was reported by Binder and Fielder (1995) that heat stress exposure for 24 h at 30°C and 12 h at 40°C appeared to mildly stimulate growth. They speculated that short term metabolic changes, or increased thermotolerance (Burr et al. 1993) due to synthesis of stable heat shock proteins (Koppenaal et al. 1991) are involved.

The clearly indicated pre-conditioning effect in Plot NI for the growth in the first growing season (G1) (Figure 29) is not present in the second growing season (G2). This could result if winter injury in 1993 / 94 was so severe it became the major factor regulating seedling growth. The pre-conditioning effect is also difficult to interpret when it is interacting with the levels of heat stress (Figure 30). The interaction is very similar for both growing seasons (Figure 31). In the first year, the 8-day pre-conditioning appears to increase the volume growth with increasing temperature, but some of this effect is lost in the second year. Again, this may in part be attributable to winter injury in plot NI.

The overall interpretation of the data suggests that the 48-hour stress duration increased growth in the first year. This is similar to what other Binder and Fielder (1995) found for shorter (12 h - 40°C and 24 h - 30°C) exposure durations, but contrary to what was intuitively expected in this experiment. The reasons are elusive. Seedlings that underwent the 8-day pre-conditioning and 48-hour heat stress treatment were observed as having buds on the lower laterals starting to flush, needles on upper laterals going yellow, no white roots or a limited number, and very brittle needles. Many of the other seedlings from the shorter pre-conditioning and heat stress treatments had white roots growing after the treatments were finished. Light is obviously not required to initiate root growth in white spruce. Binder et al. (1995) report the same to be true for Sitka spruce.

Temperatures in the incubators were stable and varied only within a few tenths of a degree from the settings. However, the temperatures dropped somewhat when the bags of seedlings were put in, and there were some lag before the incubator temperatures again reached target stress temperatures. No formal measurements of the bag temperatures were recorded. This decrease in

temperature is difficult to avoid and has been reported in other similar experiments (Binder and Fielder 1988).

Results from these experiments suggest that pre-conditioned seedlings acquired thermotolerance which prepared them for the sublethal heat stress treatment of 40°C for up to 48 h. The biological basis for this is not obvious, but I suggest that these treatments stimulated the seedlings to become more efficient and better able to survive and grow when outplanted. This is a reasonable suggestion considering evidence presented here and in other studies of similar nature.

Conclusions

Field experiment and controlled conditions experiment

Results of temperature monitoring in the field test and heat transfer testing under controlled conditions show that using tarpaulins made from the Silvicool® 2 material to protect seedlings would not be significantly different from leaving the seedling boxes unprotected. However, no related detrimental effects on seedling survival or growth were observed.

New tarpaulins tested showed little difference in performance with respect to heat transfer. The determining factors for purchase should be quality of materials.

The insulated FIST canopy had a higher accumulated heat sum than did the new tarpaulins, mainly because the heat dissipated slower once it had accumulated inside. A FIST canopy loaded with seedling boxes directly from cool storage can be expected to maintain low box temperatures longer than boxes protected by tarpaulins, because of the slow rate of heat transfer through the insulation.

There is a distinct increase in heat transfer capacity after a tarpaulin has been used for a period of years, though differences between different types of used tarpaulins are not great.

Relative conductivity analysis indicated no cell membrane damage in needles of seedlings as a result of exposure to temperatures of up to 35°C, and storage times of up to six days.

Results of the statistical growth analysis are interpreted as showing no significant differences between treatments in total seedling volume after two growing seasons, or in the growth in each of the first, second or third growing seasons. Furthermore, interpretation of the data indicate no

significant differences in seedling volumes after three growing seasons regardless of length of seedling storage.

Storage of white spruce seedlings at temperatures ranging up to 35°C for up to six days did not cause mortality.

Laboratory experiment

The results of this experiment indicate containerized white spruce seedlings are very heat tolerant. Temperatures of up to 40°C for 48 h did not cause any appreciable cell membrane damage, did not kill the seedlings and did not reduce their growth. Such temperatures actually enhanced growth. Pre-conditioning of seedlings may have been beneficial in the short term for seedlings exposed to 48-hour heat stress. However, the seedlings were outplanted in nursery bed conditions, and the results may have been different had the seedlings been outplanted on a harvested site. The results suggest further studies should be made to confirm the findings of this study with respect to heat tolerance and pre-conditioning. Heat tolerance limits for containerized white spruce and other major species used for reforestation in western Canada should be established for seedlings outplanted in field conditions.

Implications for the Forest Industry

Some implications for the forest industry from this research project are:

1. The Silvicool® 2 type of reflective tarpaulins should not be used if heat exposure is of concern.

2. The effectiveness of reflective tarpaulins deteriorate significantly with usage though it is not a linear relationship. Better quality tarpaulins are more effective than lower quality ones.

3. The FIST canopy is effective in protecting seedlings against solar radiation. However, it should not be used for longer-term storage since heat slowly builds up and then does not dissipate as quickly as it would under a tarpaulin. The more seedling boxes in the FIST, the longer they will stay cool if seedlings are cool when loaded.

4. A standard practice is to dispose of stored seedlings that have been heated to 20-30°C for more than a day as a result of a malfunctioning refrigeration unit on the semi-trailer. Since a load of seedlings may represent a value of $35,000 to $40,000 (in 1994), this is a costly practice that should be re-evaluated in light of the heat tolerance demonstrated for containerized white spruce seedlings in this study.

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Appendix I

Sample plot layout in field experiment

Table A-1. Plot layout showing Time zones within each of the six blocks in the field experiment. Blocks are shown in the order of the plot. See also Figure 2.

	BLOCK 6	BLOCK 2	BLOCK 4	BLOCK 5	BLOCK 3	BLOCK 1
West	Time 6	Time 5	Time 5	Time 2	Time 6	Time 3
	Time 2	Time 3	Time 1	Time 4	Time 3	Time 4
	Time 1	Time 1	Time 2	Time 5	Time 4	Time 5
	Time 3	Time 4	Time 4	Time 6	Time 1	Time 2
	Time 4	Time 2	Time 6	Time 1	Time 2	Time 6
East	Time 5	Time 6	Time 3	Time 3	Time 5	Time 1
	South					**North**

Table A-2. Treatments in the order of planting in the blocks. West is top of the block. T = Time zone; TR = Treatment.

BLOCK 6	BLOCK 2	BLOCK 4	BLOCK 5	BLOCK 3	BLOCK 1
T6 - TR4	T5 - TR9	T5 - TR9	T2 - TR5	T6 - TR9	T3 - TR5
9	5	8	2	7	1
2	6	1	7	5	9
7	8	7	8	10	8
5	10	2	10	1	6
6	1	10	4	4	7
10	4	6	1	3	4
3	2	3	3	2	2
1	3	5	6	8	3
8	7	4	9	6	10
T2 - TR3	T3 - TR4	T1 - TR8	T4 - TR3	T3 - TR9	T4 - TR8
9	10	9	4	4	4
10	2	6	1	8	10
8	7	5	6	6	9
1	5	2	2	1	7
5	9	4	9	10	1
6	1	10	10	5	6
4	8	3	7	3	2
7	6	1	8	7	3
2	3	7	5	2	5

continued from previous page					
T1 - TR7	T1 - TR6	T2 - TR3	T5 - TR10	T4 - TR4	T5 - TR10
10	3	4	9	9	9
5	1	1	5	6	6
8	2	9	1	10	8
9	9	2	8	8	1
3	5	10	3	5	4
4	8	5	4	7	3
1	10	7	2	2	2
2	4	6	7	3	7
6	7	8	6	1	5
T3 - TR7	T4 - TR7	T4 - TR3	T6 - TR3	T1 - TR8	T2 - TR7
10	10	10	2	5	3
3	6	8	10	4	5
4	9	2	5	1	6
6	4	7	6	3	10
1	3	6	1	9	2
9	5	4	4	2	9
5	2	1	9	10	4
2	8	9	7	6	1
8	1	5	8	7	8
T4 - TR1	T2 - TR6	T6 -TR5	T1 - TR9	T2 - TR4	T6 - TR4
2	4	7	7	7	7
6	7	1	10	5	2
3	3	9	1	6	1
8	10	3	8	8	6
4	1	10	3	9	10
9	2	4	5	3	8
5	8	8	6	1	3
7	9	6	4	10	5
10	5	2	2	2	9
T5 - TR1	T6 - TR4	T3 - TR5	T3 - TR7	T5 - TR4	T1 - TR7
10	6	9	2	9	1
8	5	2	4	1	8
3	9	10	9	6	4
7	2	6	3	7	5
2	7	7	10	10	3
6	3	8	6	3	6
5	10	3	1	2	2
8	1	1	5	5	9
4	8	4	8	8	10

Appendix II

Statistical Models

Table A-3. Statistical model for a split-plot randomized complete block design, to analyze differences in seedling volume and growth between treatments.

Source	DF
Block	6-1= 5
Time	6-1= 5
Block * Time (Error 1)	(6-1)(6-1)= 25
Treatment	(10-1)= 9
Treatment * Time	(10-1)(6-1)= 45
Treatment * Block	(10-1)(6-1)= 45
Block * Time * Treatment	(6-1)(6-1)(10-1)= 225
Replication (Error 2)	10*6*6(2-1)= 360
Sampling error	10*6*6*2(4-1)= 2160
Total	(10*6*6*2*4)-1= 2879

*Table A-4. Covariance analysis of V3 with an experimental error term that includes the Time*Treatment*Block factor.*

Source	DF	Sum of Squares	F Value	Pr > F
Block	5	394832843	26.16	0.0001
Time	5	107408005	0.16	0.9732
Time*Block	25	3257992101	43.7	0.0001
Treatment	9	47973030	1.77	0.0716
Time*Treatment	45	256504745	1.89	0.0006
Exper. Error	630	1901905616	1.07	0.1511
V1	1	1544353678	545.90	0.0001
Sampl. Error	2154	6093681194		
Corr. Total	2874	13956820060		

Table A-5. Covariance analysis of G3 for Block 1. The other blocks were analyzed with the same model.

Source	DF	Sum of Squares	F Value	Pr > F
Time (T)	5	69936006.5	7.71	0.0001
Treatment (TR)	9	24296670.0	1.49	0.1503
TI*TR	45	130557592.2	1.60	0.0112
Replication (TI*TR)	60	107491690.2	0.99	0.5056
V1	1	187619071.3	103.47	0.0001
Sampling Error	354	641900132.1		
Corrected Total	474	1189275560.5		

Table A-6. The Probability of significance from the GLM covariance analysis of: G3 (Growth in the first two growing seasons); G4 (Growth in the third growing season), and G5 (Growth in the first three growing seasons), analyzed by individual blocks. Note 1: Only the decimal values are shown. Numbers below 0500 indicate statistical significance at α=0.05 level. Note 2: Blocks are shown in the order of the plot.

Source	Block 6			Block 2			Block 4			Block 5			Block 3			Block 1		
	G3	**G4**	**G5**	**G3**	**G4**	**G5**	**G3**	**G4**	**G5**	**G3**	**G4**	**G5**	**G3**	**G4**	**G5**	**G3**	**G4**	**G5**
Time(T)	0001	0001	0001	0001	0001	0001	0001	0001	0001	0001	0094	0001	0001	0190	0019	0001	0001	0001
Treatm. (TR)	3205	1272	2852	7129	7288	7949	0182	0004	0010	5839	0201	0455	0269	3016	6383	1503	0137	0054
TI*TR	0107	3187	4445	0001	4416	0033	0011	0006	0001	7904	0001	0260	0006	0014	0380	0112	0256	0391
Rep(TI*TR)	1000	9476	9993	5077	8794	7596	8068	6373	6377	9398	8271	8806	9544	7436	9515	5056	6886	5992
V1	0001	1523	0001	0001	8432	0007	0001	0001	0001	0001	0001	0001	0001	0008	0001	0001	0001	0001
Exp. Error	0001	0222	0001	0001	0022	0001	0001	0001	0001	0001	0003	0001	0001	0037	0009	0001	0003	0001
R^2 (0.xxx)	631	335	485	658	348	586	521	393	488	431	360	413	454	340	353	460	358	433

Table A-7. The analysis of variance (ANOVA) data for Ratio, in the Controlled Conditions Experiment.

Source	DF	Mean Square	F Value	Prob > F
Treatment	6	0.01790482	72.29	0.0001
Tarpaulin	1	0.00012274	0.50	0.4873
Treatment*Tarpaulin	6	0.00006540	0.26	0.9490
Error	28	0.00024769		
Corrected Total	41			

Table A-8. The statistical model (GLM) and values for G1 in Laboratory Experiment.

Source	DF	Mean Square	F Value	Pr > F
Plots (B)	1	5712.2	0.04	0.8369
Pre-conditioning (P)	2	47917.0	0.36	0.7007
Heat stress (S)	3	196498.9	1.46	0.2243
Time (T)	1	280107.1	2.08	0.1496
B*P	2	13953.1	0.10	0.9016
B*S	3	36041.7	0.27	0.8488
B*T	1	569401.7	4.23	0.0401
P*S	6	237996.0	1.77	0.1029
P*T	2	161356.5	1.20	0.3023
S*T	3	37043.2	0.28	0.8434
B*P*S	6	253918.0	1.89	0.0805
B*P*T	2	415472.8	3.08	0.0462
B*S*T	3	23472.6	0.17	0.9138
P*S*T	6	120564.5	0.90	0.4977
B*P*S*T	6	111326.4	0.83	0.5494
V1	1	4951602.5	36.76	0.0001
Error	884	134691.7		
Corrected Total	932			

Table A-9. The statistical model (GLM) and values for G2 in Laboratory Experiment.

Source	DF	Mean Square	F Value	Pr > F
Plots (B)	1	18606029.6	27.51	0.0001
Pre-conditioning (P)	2	235243.5	0.35	0.7064
Heat stress (S)	3	543881.5	0.80	0.4918
Time (T)	1	20956.6	0.03	0.8603
B*P	2	327930.9	0.48	0.6160
B*S	3	333415.4	0.49	0.6873
B*T	1	451071.6	0.67	0.4144
P*S	6	2073028.5	3.06	0.0057
P*T	2	674378.2	1.00	0.3695
S*T	3	345146.8	0.51	0.6753
B*P*S	6	596516.1	0.88	0.5076
B*P*T	2	1246338.4	1.84	0.1591
B*S*T	3	124504.5	0.18	0.9073
P*S*T	6	867920.8	1.28	0.2625
B*P*S*T	6	592695.7	0.88	0.5118
V1	1	1382375.3	2.04	0.1533
Error	773	676444.6		
Corrected Total	821			

Appendix III

Research Program Timeline

May 1992	Preliminary tarpaulin tests at FERIC office
June 1992	Test of reflective tarpaulins at Red Rock Research Station
	i) Electrical conductivity measurements
	ii) Outplanting and measurements of seedlings
September 1992	Measurement of root collar and total height of seedlings
January 1993	Test of reflective tarpaulins under controlled conditions at FERIC
April 1993	Survival assessment of seedlings
June 1993	Laboratory experiment with pre-conditioning of seedlings
	i) pre-conditioning of seedlings and heat stressing
	ii) electrical conductivity measurements of needles
	iii) outplanting and measurement of seedlings
September 1993	Measurement of root collar and total height of all seedlings
April 1994	Survival assessment of all seedlings
October 1994	Measurement of root collar and total height of all seedlings

Appendix IV

Soil and foliage analysis

The following changes were found in two samples along the northern part of the plot, going from East to West:

Soil - increasing:	pH, total sulfur, organic matter, K, Mg, Cu, Zn, Fe, Mn;
Foliage - increasing:	P, Ca, Mg, K, Zn, Mn, B;
Soil - decreasing:	salts, P;
Foliage - decreasing:	N, S, Cu, Fe, Al;
Soil - no change:	total N, Ca;
Foliage - no change:	N/A

The following changes were found in three samples along the southern part of the plot going from East to West:

Soil - increasing:	total sulfur, organic matter, total nitrogen, P, Cu, Zn, Fe, Mn;
Foliage - increasing:	K, Mn, B;
Soil - decreasing:	pH;
Foliage - decreasing:	N, S, Cu,
Soil - variable and flat:	salts, K, Ca, Mg, B;
Foliage - variable and flat:	P, Ca, Mg, Zn, Fe, Al.

The following changes were found in two samples along the middle part of the plot going from South to North (includes one sample from each of the two previous sections):

Soil - increasing:	total sulfur, organic matter, total nitrogen, P, Cu, Zn, Fe, Mn;
Foliage - increasing:	N, Ca, S, Cu, Zn, Fe, Mn, B, Al;
Soil - decreasing:	pH, salts, K, Mg;
Foliage - decreasing:	P, Mg, K;
Soil - no change:	Ca;
Foliage - no change:	N/A

The following changes were found in two samples along the Western part of the plot going from South to North (includes one sample from each of the two first sections)::

Soil - increasing:	total sulfur, organic matter, Cu, Zn, Fe, Mn;
Foliage - increasing:	N, Ca, Mg, S, Zn, Mn,
Soil - decreasing:	pH, salts, total nitrogen, P, K, Mg,
Foliage - decreasing:	P, K, Fe, Al;
Soil - no change:	Ca, B;
Foliage - no change:	Cu;

Table A-10. Results from soil nutrient sampling in field plot.

Sample #	pH	S	E.C	O.M.	N	P	K	Ca	Mg	Cu	Zn	Fe	Mn	B
1	6.2	0.006	0.44	1.0	0.03	200	90	600	105	0.8	0.3	55	15	0.3
2	5.1	0.005	0.42	1.9	0.05	382	70	500	80	1.1	1.1	85	34	0.2
3	5.4	0.008	0.40	1.8	0.05	355	80	450	105	1.0	1.0	75	26	0.3
4	5.6	0.009	0.32	1.5	0.04	259	75	500	110	1.0	0.8	65	20	0.2
5	5.2	0.012	0.30	2.5	0.05	341	85	500	100	1.7	1.5	120	45	0.2
6	5.5	0.010	0.32	2.1	0.06	355	90	500	110	1.1	1.2	95	32	0.2
7	5.1	0.014	0.38	2.0	0.05	382	85	450	75	1.2	1.2	110	42	0.2
8	5.5	0.013	0.34	2.7	0.08	382	80	550	115	1.3	1.5	100	47	0.2
9	5.1	0.008	0.44	1.8	0.05	409	95	500	95	1.2	1.4	145	48	0.1
10	5.9	0.004	0.28	0.7	0.02	159	80	600	135	0.8	0.6	78	19	0.1
11	6.2	0.008	0.32	1.5	0.04	241	90	700	145	0.8	0.6	75	19	0.2
12	6.6	0.007	0.28	1.3	0.03	223	90	650	140	0.8	0.8	75	21	0.2
13	6.6	0.007	0.32	1.4	0.04	232	75	700	150	0.8	0.9	55	22	0.1
14	6.4	0.006	0.28	1.1	0.03	177	70	650	120	0.9	0.6	45	17	0.1
15	6.4	0.004	0.26	1.1	0.03	200	85	800	155	0.8	0.9	60	21	0.2
16	5.9	0.008	0.34	2.3	0.05	282	95	750	100	0.7	2.1	90	31	0.2

LEGEND: S = Total sulphur (%) E.C. = Salts (mmhos/cm) O.M. = Organic matter (%) N = Total nitrogen (%)
P = Phosphorous (ppm) K = Potassium (ppm) Ca = Calcium (ppm) Mg = Magnesium (ppm)
Cu = Copper (ppm) Zn = Zinc (ppm) Fe = Iron (ppm) Mn = Manganese (ppm)
B = Boron (ppm)

Sample # : 1, 4, 6 - along southern part of plot going from East to West.
Sample # : 2, 5 - along northern part of plot going from East to West.
Sample # : 3 - between samples 2 and 4 in the middle of plot.
Sample # : 7...16 - from locations outside of plot.

Table A-11. Results from foliage nutrient sampling in field plot. Foliage sample numbers corresponds to the soil sample numbers since needles were collected from seedlings adjacent to the soil sample spot.

Sample #	N	P	Ca	Mg	K	S	Cu	Zn	Fe	Mn	B	Al
1	0.71	0.23	0.50	0.12	0.59	0.100	7	51	408	184	20	468
2	0.62	0.21	0.47	0.10	0.52	0.114	5	52	129	322	23	210
3	0.64	0.28	0.43	0.11	0.68	0.102	4	47	99	338	24	179
4	0.60	0.25	0.39	0.11	0.60	0.092	4	44	114	271	22	186
5	0.60	0.22	0.49	0.11	0.58	0.088	3	64	93	365	24	171
6	0.58	0.23	0.47	0.10	0.62	0.086	3	55	173	283	25	248
7	0.58	0.24	0.43	0.11	0.56	0.074	3	47	213	263	19	232
8	0.64	0.26	0.56	0.12	0.58	0.092	4	59	119	263	26	209

LEGEND: N = Nitrogen (%) P = Phosphorous (%) Ca = Calcium (%) Mg = Magnesium (%)
K = Potassium (%) S = Sulphur (%) Cu = Copper (ppm) Zn = Zinc (ppm)
Fe = Iron (ppm) Mn = Manganese (ppm) B = Boron (ppm) Al = Aluminum (ppm)