AN INTEGRATED STUDY OF NURSERY STOCK CONDITIONING: OSCILLOSCOPE READINGS AND COLD HARDINESS

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Production of high quality conifer stock requires improved conditioning practices and development of better methods to evaluate stock quality. Efforts to evaluate quality of planting stock have included tests of cold hardiness (Timmis 1973, 1976; Sakai and Weiser 1973; van den Driessche 1969, 1973; Seidel 1974; Tanaka and Timmis 1974; Aronsson et al. 1976; Clerum 1976) and dormancy (Lavender and Cleary 1974, Ferguson et al. 1975, Cleary et al. 1978).

Dormancy and cold hardiness are phenomena that often occur in parallel (Levitt 1956). As seedlings are exposed to lower temperatures, hardiness increases until trees have reached their maximum cold hardiness. This progression often coincides with the dormant state reviewed by Cleary et al. (1978). If these phenomena are parallel, a method that measures the dormant state might also measure cold hardiness. In this study, the oscilloscope/square wave apparatus suggested for detecting dormancy (Ferguson et al. 1975) was investigated for its potential use in determining cold hardiness of Douglas-fir seedlings. Preliminary results are reported in this paper.

MATERIALS AND METHODS

Seedlings measured for square wave patterns and cold hardiness were samples from a comprehensive study on the combined effects of several nursery practices. Eighteen treatment combinations of two irrigation regimes, three root manipulation methods, and three seedbed densities were installed in three replications at the U.S. Forest Service's Wind River Nursery, Carson, Washington, and at the D. L. Phipps State Forest Nursery, Elkton, Oregon, in the fall of 1976. Seedlings were from separate seed sources, 1,500-foot (grown at Phipps) and 3,000-foot (grown at Wind River) origins in the Umpqua drainage in Oregon. A total of 60 2-0 seedlings per treatment per replication were lifted from Wind River in mid-September, mid-October, and mid-November of 1977, and at Phipps the week following each lifting at Wind River. These seedlings were

given several laboratory tests, including measurement of square wave pattern and cold hardiness. Trace observations for each lifting date were made on a subsample of 10 seedlings per treatment per replication immediately after lifting at the nursery. Seedlings were classified as slightly active, very active, dormant, or dead (Fig. 1).

A separate subsample of five seedlings per treatment per replication for each lifting date was potted in each of two pots. One pot of seedlings was subjected to a cold test at -4° C and the other at -7° C. A day or more after potting the seedlings were placed in a cooling chamber at an initial temperature of 1°C. Temperature was dropped by 1.4°C increments at half-hour intervals until the minimum temperature was reached. The seedlings were kept at the minimum temperature for 3 hours. The temperature was then increased in 1.4°C increments at halfhour intervals until the initial temperature was reached. The seedlings were then placed in a greenhouse under growing conditions, and extent of damage was observed after 6 weeks. Seedlings were classified dead or severely damaged if more than one-half of the foliage was brown and the terminal bud was injured. These observations were combined for analysis. Seedlings from both nurseries were tested simultaneously. The data were analyzed by linear regression methods, and coefficients of correlation were calculated. The observations for each temperature test were combined and each group of 10 seedlings for each treatment replication combination was used to obtain a data point.

Calculated correlations were compared visually with a theoretical model (Fig. 2). In the model, the general form of the negative correlation is indicated for three levels of damaging temperature. If we assume that dormancy and frost hardiness are parallel phenomena in Douglas-fir seedlings, then we would expect to observe a negative correlation between damage from freezing and percent of seedlings dormant. The percent of seedlings damaged or killed would probably also vary with the minimum temperature used in the test.

RESULTS

Looking at freeze damage and dormancy separately, I observed a seasonal trend in square wave form. Percent damage was higher in October than in September and November in both Wind River and Phipps stock (Tables 1 and 2).

The data for each nursery were analyzed separately for the September, October, and November lift dates. No correlation trends were detected. For seedlings from the Wind River Nursery, there was no correlation between the percentage of seedlings severely damaged or killed and the percentage of seedlings that were dormant as determined by the square wave trace (Fig. 3). In fact, the correlation coefficients decreased with each subsequent lifting. The data from the Phipps Nursery also indicated a lack of significant correlations for each lifting date (Fig. 4); however, the regression line for October sampling slightly resembled the model.







	September		October		November	
$reatment \frac{1}{2}$	Trees dormant 2/	Trees 3/ damaged or dead	Trees dormant	Trees damaged or dead	Trees dormant	Trees damaged or dead
				Percent		
1	0	0	53	13	57	3
2	0	0	30	3	73	0
3	0	10	30	10	63	0
4	3	7	47	0	70	0
5	0	0	43	7	40	0
6	0	7	60	7	67	0
7	10	0	40	0	70	0
8	7	3	53	0	47	0
9	7	0	33	0	50	0
10	7	3	47	3	50	0
11	7	0	40	0	40	0
12	0	0	40	3	45	0
13	20	0	40	10	63	0
14	17	10	53	7	90	0
15	0	7	27	3	80	0
16	0	0	37	0	53	0
17	0	0	33	0	63	0
18	3	0	57	0	63	0

Table 1. Percent of Douglas-fir seedlings damaged or killed by freezing tests and percent of trees dormant, from Wind River Nursery, Carson, Washington

1/ Three replications combined, totaling 30 trees except in November test; treatments 9, 12, 14, and 15 were two replications combined, totaling 20 trees. Treatments were combinations of two irrigation regimes, three root manipulation methods, and three seedbed densities.

2/ Determined by square wave trace form.

 $\overline{3}$ / More than one-half of seedling damaged.

$Treatment \frac{1}{2}$	September		October		November	
	Trees dormant 2/	Trees 3/ damaged or dead	Trees dormant	Trees damaged or dead	Trees dormant	Trees damaged or dead
			P	ercent		
1	7	3	23	13	100	0
2	7	3	47	7	93	7
3	17	0	27	13	97	0
4	3	3	53	13	93	0
5	0	17	17	13	97	3
6	20	20	33	27	93	20
7	27	17	40	37	100	13
8	3	7	50	13	97	10
9	27	27	63	20	90	3
10	17	7	40	10		7
11	7	20	17	27		7
12	30	17	27	10		3
13	17	0	45	15		10
14	17	20	30	20		
15	23	23	47	23		10
16	23	13	27	33		20
17	15	15	30	20		0
18	10	10	30	17		7

Table 2.	Percent of Dougla	s-fir seedlings	damaged or killed b	by freezing tests and
percent	of trees dormant,	from D. L. Phip	ops Nursery, Elkton,	Oregon

1/ Three replications combined, totaling 30 trees except September when treatment 17 was 2 replications combined, totaling 20 trees; in October treatments 13 and 17 were 2 replications combined, totaling 20 trees; in November treatment 8 was 2 replications combined, totaling 20 trees. Treatments were combinations of two irrigation regimes, three root manipulation methods, and three seedbed densities.

- 2/ Determined by square wave trace form.
 3/ More than one-half of seedling damaged.
- 4/ No data available



FIGURE 2. THEORETICAL RELATIONSHIP BETWEEN PERCENT OF DOUGLAS-FIR SEEDLINGS DAMAGED OR KILLED BY FREEZING TESTS AND THE PERCENT OF TREES DORMANT, EACH LINE REPRESENTS A

DAMAGING TEMPERATURE.



FIGURE 3. CORRELATION BETWEEN DOUGLAS-FIR SEEDLINGS FROM WIND RIVER NURSERY DAMAGED OR KILLED BY FREEZING

TEST AND TREES DORMANT AS DETERMINED BY SQUARE WAVE TRACE.



FIGURE 4. CORRELATION BETWEEN DOIUULAS FIR SEEDLINGS FROM D. L. PHIPPS NURSERY DAMAGED OR KILLED BY FREEZING TEST AND TREES DORMANT AS DETERMINED BY SQUARE WAVE TRACE

The data were also analyzed for effects of individual treatments. No correlative trends between trace form and damage were detected for seedlings of each irrigation treatment, root manipulation treatment, or seedbed density. Treatment interactions were not analyzed.

DISCUSSION

Observations were carefully made to minimize factors that might influence the test results. Factors that might have had some effect were (1) Square wave observations were taken after lifting rather than in situ; (2) trace and freezing damage observations were made on different seedlings; and (3) time intervals between lifting seedlings, potting them for testing, and testing them may have subtly influenced the seedlings' capability to withstand freezing.

A major purpose for determining dormancy is to decide when seedlings are well enough conditioned to withstand storage or outplanting; i.e., seedlings are cold hardy. Visual observation of trace form such as was done in this study would be the likely technique used by nursery personnel. Data from this study indicate that such observations do not correlate with hardiness determined by freezing tests. Therefore, I question the use of the oscilloscope square wave technique as a tool for measuring cold hardiness.

In her evaluation of the oscilloscope technique for detection of dormancy and survival potential of Douglas-fir seedlings, Askren (1978) classified trace forms using voltage measurements at three constant points on the trace. Ratios of these measurements were used to typify the trace form. Seasonal changes in trace form were then compared with seasonal changes in bud growth responses. She found that although the trace form demonstrated a seasonal trend, the transition from dormancy deepening to the dormant state was not reflected in any significant change of trace form. She did however, observe that the trace form changed greatly during the period in which cambial growth ceased (late October to early November). Van den Driessche, (1969) using a ratio of tissue impedance at low frequency to tissue impedance at high frequency, also observed changes in his measurements coinciding with cessation of cambial activity. He further noted that cambial activity ceased at the same time cold hardiness began to develop.

We intend to further evaluate the use of the oscilloscope/square wave technique during the 2d year of a continuing study of seedling conditioning.

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