DORMANCY, CHILLING REQUIREMENTS, AND STORABILITY OF CONTAINER-GROWN LOBLOLLY PINE SEEDLINGS

James N. Boyer and David B. South $^{
m l}/$

Abstract. -- The objective of this study was to learn whether storability coincides with satisfaction of the chilling requirement in several different seed sources of container-grown loblolly pine (Pinus taeda L.) seedlings. While a certain amount of chilling appeared to be necessary for storability, the ability to survive and grow following storage were not related to satisfaction of the chilling requirement for bud break for different seed sources.

INTRODUCTION

In late summer or fall, with proper conditioning, loblolly pine (Pinus taeda L.) seedlings cease height growth, set buds, and enter into a period of dormancy. The dormancy cycle is believed to progress through preliminary rest, mid-rest, and after-rest (Samish, 1954). While moisture stress may play a role in the induction of dormancy, decreasing daylength seems to be important in dormancy deepening, or entrance into mid-rest (Carlson, 1985). The mid-rest stage (deep dormancy) is defined as a failure to quickly resume normal growth even under favorable environmental conditions (Doorenbos, 1953; Romberger, 1963). Lifting western conifer seedlings in this stage seems to cause severe physiological disruption (Hermann et al., 1972). Dormancy state is usually measured by how rapidly a seedling breaks bud and resumes growth when placed in an environment which is ideal for growth.

The dormant period is commonly broken in nature only after exposure to near-freezing temperatures (Berry, 1965; Campbell and Sugano, 1975; van den Driessche, 1975; Lavender, 1981; Lyr et al., 1970; Nelson and Lavender, 1979; Nienstaedt, 1967; Steinhoff and Hoff, 1972; Wells, 1979). This "chilling requirement" provides an adaptive value to trees by preventing shoot growth during warm spells in fall or winter when the new growth would be damaged by subsequent low temperatures (Lavender, 1981; Nienstaedt, 1967). Before the chilling requirement is met, a seedling is in the true dormancy state or mid-rest. After the chilling requirement has been satisfied, the seedling is said to be in after-rest, which is equivalent to imposed dormancy or quiescence, and would quickly resume growth if placed in a greenhouse.

As early as 1920, it was recognized that plants would not resume normal growth in the spring unless they had been previously subjected to a period of chilling (Coville, 1920). By 1932 it was suggested that the number of hours the temperature was 8°C or lower be used as a measure of dormancy break in peach (Prunus persica Batsch) (Weinberger, 1956). Subfreezing temperatures are not effective in breaking dormancy (Lyr et al., 1970; Wareing, 1969).

^{1/}Respectively, Research Associate, School of Forestry, Alabama Agricultural Experiment Station, Auburn University, AL 36849; and Assistant Professor and Director, Auburn University Southern Forest Nursery Management Cooperative.

Although most of the early work on dormancy release was done on fruit trees, chilling requirements have now been reported for forest trees. The chilling requirement for breaking dormancy has been found to vary not only among tree species, but also among seed sources or families of the same species (Nelson and Lavender, 1979; Steinhoff and Hoff, 1972). Generally, provenances from the milder portions of a species' range have lower chilling requirements than provenances from more severe climates (Lavender, 1981).

Most bare-root loblolly pine seedlings are lifted from nursery beds from December through February, while transplanting in the field may continue past March. Lifting is generally completed before spring so as not to interfere with preparation of the seed beds for the coming season. Furthermore, survival of seedlings lifted while they are flushing is generally poor (Wakeley, 1954). Hence, many seedlings are kept in refrigerated storage until they can be transplanted in the field. When the planting season must be extended, cool-storage can be used to keep stock inactive longer than if left in the nursery beds (Hocking and Nyland, 1971). In cases where lifting operations are extended into less favorable periods to accommodate the longer planting season, especially earlier in the fall, reliable information is required on the relationships among lifting date, state of dormancy, duration of cool storage, and field survival and performance for different geographic seed sources (Garber and Mexal, 1980).

Several researchers have noted that for best results with storage, fall-lifting should be delayed to ensure the proper state of dormancy (Garber and Mexal, 1980; Hermann et al., 1972; Hocking and Nyland, 1971; Lavender and Wareing, 1972; Nyland, 1974). Garber and Mexal (1980) suggested that the survival potential of seedlings when planted immediately may be relatively constant over a long period of time, whereas the storage potential may change a great deal.

A number of investigators have attempted to correlate seedlings' ability to withstand storage, or "storability," with date of lifting (Austin, 1961; Garber and Mexal, 1980; Hermann, 1967; Hermann et al., 1972; Hocking and Ward, 1972; Lavender, 1964; Nyland, 1974; Oldenkamp and Elk, 1966; Stone and Schubert, 1959a; Stone and Schubert, 1959b; Winjum, 1963; Winjum, 1961). However, many now recognize that optimum lifting dates may vary from year to year depending upon the weather (Garber and Mexal, 1980; Hocking and Nyland, 1971; Ursic et al., 1966). These optima may vary by location and may also depend upon the seed source or family being raised. The need for a more standardized measure of storability became quite evident during the planting season of 1982-83. The fall of 1982 was exceptionally mild, with fewer chilling hours than usual. For example, in 1980 and 1983, approximately 500 chilling hours accumulated at Auburn by 31 December. In 1982, just over 300 hours had accumulated by that date. The U. S. Forest Service estimated losses at \$220,000 that year due to deterioration of seedlings in storage (Oak, 1983). Improper physiology or state of dormancy was suggested to be the cause of the deterioration.

Garber and Mexal (1980) found that fulfillment of storage potential and satisfaction of the chilling requirement for bud break occurred at approximately the same time in two separate studies using seed sources from a similar latitude. The objective of this study was to learn more about timing

of storability for loblolly pine and determine whether storability coincides with satisfaction of the chilling requirement for bud dormancy.

MATERIALS AND METHODS

During the fall and winter of 1983-1984, two studies investigated the relationships among chilling (cold temperature accumulation), dormancy state, and ability to withstand cool storage. In one study, loblolly pine seedlings from four different seed sources (Table 1) were grown outside and and received natural photoperiod and chilling. In the other study, seedlings from the northernmost (southeast Virginia) and southernmost (southeast Georgia) sources were raised in the greenhouse under a constant photoperiod and received artificial chilling in a refrigerated room. Seedlings were grown in 164-cm³ Leach-cell containers (Ray Leach "Cone-tainer" Nursery, Canby, Oregon) in a medium of peat moss, vermiculite, and perlite (2:2:1, by volume) with a time-release fertilizer added to the mix.

Outside, seedlings were sampled after various amounts of chilling, from 98 to 407 chilling hours. A chilling hour was one hour of accumulated time between 0° and 8°C (32-46°F). At each sampling time, seedlings were divided into three groups: for bud break or dormancy release; for immediate outplanting; and for cool storage followed by outplanting. In the bud break study, seedlings were moved into the greenhouse under a 14-hour photoperiod, and number of days for bud break to occur was monitored. For cool storage, seedlings were placed in plastic bags at approximately 2°C (36°F) for 11 weeks. Seedling roots were kept in the containers and potting media for storage, except for duplicate samples of one source (southwest Arkansas), which had roots bared for storage and planting. Survival and height growth one year after outplanting were used to compare storability of seedlings.

Seedlings raised in the greenhouse were grown under a 16-hour photoperiod. In mid-September, each replication of each seed source was divided into three daylengths: 10, 12, or 14 hours. These photoperiod treatments were continued for seven weeks. Following the photoperiod treatments, all seedlings were moved to a refrigerated room with a 10-hour photoperiod. Temperature inside the room was kept at approximately 2°C (36°F). Seedling samples were taken after approximately the same numbers of chilling hours as in the outside study (101-485 hours). Seedlings were divided into three groups: for bud break or dormancy release; for immediate vigor testing (stress testing); and cool storage followed by vigor testing. In the bud break study, seedlings were moved into the greenhouse under a 14-hour photoperiod, and number of days for bud break to occur was monitored. Cool storage took place in plastic bags, with roots bared, at 2°C (36°F) for 10 weeks. Vigor testing consisted of exposing bare-root seedlings to 32°C (90°F) and 30 percent relative humidity for 20 minutes, followed by potting in sand. Survival six weeks after transplanting was used as a measure of seedling vigor or ability to withstand stress.

RESULTS AND DISCUSSION

For the two parallel studies, results were obtained from only the storability part of the outside study and only the bud break portion of the greenhouse study. Regardless of chilling, trees grown outside did not flush

when brought into the greenhouse. This is thought to be explained by drying of roots which had grown out of the container. Roots of trees grown in the greenhouse did not occupy the entire container and had not emerged from the container bottom. Their roots did not dry out and bud break progressed normally. Storability is not reported for greenhouse-grown trees since survival was very poor following storage, even for trees which received large amounts of chilling prior to storage. Nonstored trees survived the vigor test at or near 100 percent regardless of chilling. The morphology of the greenhouse-grown seedlings (relatively slender and nonlignified stems with very sparse root systems) may have been a factor in their lack of storability. Furthermore, many of the stored trees had badly molded foliage. For the outside study, stored trees survived as well as nonstored trees if they received adequate chilling prior to storage.

In the bud break study, chilling increased speed of bud break for both seed sources (Table 2). Longer photoperiods prior to chilling delayed the breaking of dormancy. For trees under short photoperiods prior to chilling, the southeast Georgia source clearly reached its maximum speed of bud break by 341 hours, while the southeast Virginia source continued to increase up to 485 hours, showing it had a greater chilling requirement (Figure 1). Differences in speed of bud break between seed sources were not so clear for trees under longer photoperiods prior to chilling.

In the storability study, the only sampling time where survival of stored trees was significantly poorer than for freshly planted seedlings was 98 chilling hours (Table 3). By 223 chilling hours, all seed sources in the outside study had received enough chilling for maximum storability. This pattern was observed for all seed sources. Height growth responded similarly, with stored trees from th first sampling time consistently shorter than all other trees (Table 4). Seedlings which had their roots bared survived and grew similarly to those from the same seed source which were kept in the potting media, except for seedlings stored after just 98 chilling hours. For this treatment, bare-rooted trees survived and grew less than those kept in the containers.

Because of possible differences in effectiveness of chilling, comparisons of chilling hours received outside with those received in the cold room may not be valid. Therefore, it is not possible to definitely say whether storability of outside-grown trees coincided with satisfaction of their chilling requirement for bud break. However, the data throw doubt on that hypothesis. While the greenhouse study showed that the seed sources varied in chilling requirement for bud break, all seed sources responded similarly to chilling for survival and height growth following long-term storage. While at least some chilling was necessary for storing loblolly pine seedlings, 223 chilling hours appeared to be sufficient for all seed sources to adequately survive more than 2 months of cool storage. This amount of chilling was less than was required by even the southernmost seed source for maximum speed of bud break.

Garber and Mexal (1980) found the chilling requirement for their Arkansas seed source to be satisfied by 19 December, which corresponded to about 400 chilling hours at their location for that year. This same source, in our study, was storable after just 223 hours, which corresponded to 6 December, 1983 at Auburn. Bare-root seedlings from a different seed source were lifted

on 6 December, 1983 in South Carolina and were able to withstand 8 weeks of cool storage (personal communication, John Conn, Champion International Corporation).

Hermann et al. (1972) reported that lifting western conifer seedlings which are in deep dormancy (defined as a failure to commence growth under favorable conditions) can be very detrimental. Our findings also suggest that lifting seedlings while in a deep dormancy state (i. e. mid-November in this study) is ill-advised, if the seedlings are to be stored. We also agree with Garber and Mexal (1980) that timing of lifting may not be so important if the seedlings will be planted immediately.

In this study, containerized seedlings from different provenances grown outdoors achieved storability at the same time. Two of these seed sources were shown to have different chilling requirements for bud break under controlled conditions. For both of these sources, the amount of chilling (at constant temperature) required for maximum speed of bud break was greater than was required (outdoors) for storability. A difference in the effectiveness of chilling outside versus under controlled conditions may have influenced the results. Furthermore, photoperiod, in addition to chilling, may play a significant role in the achievement of storability. More work in this area will be needed in order to elucidate the individual effects of and interactions between chilling and photoperiod on seedling storability. It may turn out that bud dormancy status has little to do with storability, but rather other changes in the seedling which take place at the same time the bud is progressing into post-dormancy control tolerance of cool storage. These possibilites need to be investigated so nursery managers can make informed decisions concerning when they may safely lift and store seedlings.

LITERATURE CITED

- Austin, R. C. 1961. p. 4 <u>In</u> Date of Lifting Symposium. Minutes by R. C. Austin. Crown Zellerbach Corp. 5-p. mimeo. November 2, 1961.
- Berry, C. R. 1965. Breaking dormancy in eastern white pine by cold and light. USDA For. Serv. Res. Note SE-43, 3 p.
- Campbell, R. K. and A. I. Sugano. 1975. Phenology of bud burst in Douglas-fir related to provenance, photoperiod, chilling, and flushing temperature. Bot. Gaz. 136: 290-298.
- Coville, F. V. 1920. The influence of cold in stimulating the growth of plants. J. Agric. Res. 20: 151-160.
- Doorenbos, J. 1953. Review of the literature on dormancy in buds of woody plants. Meded. Landbhoogesch. Wageningen, 53: 1-24.
- Garber, M. P. and J. G. Mexal. 1980. Lift and storage practices: their impact on successful establishment of southern pine plantations. N. Z. J. For. Sci. 10: 72-82.

- Hermann, R. K. 1967. Seasonal variation in sensitivity of Douglas-fir seedlings to exposure of roots. For. Sci. 13: 140-149.
- Hermann, R. K., D. P. Lavender and J. B. Zaerr. 1972. Lifting and storing western conifer seedlings. For. Res. Lab., Oregon State Univ., Corvallis. Res. Pap. 17, 8 p.
- Hocker, H. W. 1955. Climatological summaries for selected stations in and near the southern pine region, 1921-1950. USDA For. Serv., S. E. For. Exp. Sta. Pap. 56, 11 p.
- Hocking, D. 1972. Nursery practices in cold storage of conifer seedlings in Canada and the United States: a survey. Tree Planters' Notes 23(2): 26-29.
- Hocking, D. and R. D. Nyland. 1971. Cold storage of coniferous seedlings a review. Appl. For. Res. Inst., State Univ. of New York, Syracuse. Res. Pap. 6, 70 p.
- Hocking, D. and B. Ward. 1972. Late lifting and freezing in plastic bags improve white spruce survival after storage. Tree Planters' Notes 23(3): 24-26.
- Lavender, D. P. 1981. Environment and shoot growth of woody plants. For. Res. Lab., Oregon State Univ., Corvallis. Res. Pap. 45, 47 p.
- Lavender, D. P. 1964. Date of lifting for survival of Douglas-fir seedlings. For. Res. Lab., Oregon State Univ., Corvallis. Res. Note 49, 20 p.
- Lavender, D. P. and P. F. Wareing. 1972. Effects of daylength and chilling on the responses of Douglas-fir (<u>Pseudotsuga menziesii</u> (Mirb.) Franco) seedlings to root damage and storage. New Phytol. 71: 1055-1067.
- Lyr, H., G. Hoffman and R. Richter. 1970. On the chilling requirement of dormant buds of <u>Tilia platyphyllos</u>. Biochem. Physiol. Pflanzen 161: 133-141.
- Nelson, E. A. and D. P. Lavender. 1979. The chilling requirement of western hemlock seedlings. For. Sci. 25: 485-490.
- Nienstaedt, H. 1967. Chilling requirements in seven <u>Picea</u> species. Silvae Genet. 16: 65-68.
- Nyland, R. D. 1974. Fall lifting for over-winter cold storage of conifers.
 Appl. For. Res. Inst., State Univ. of New York, Syracuse. Res. Pap. 22, 5
 P.
- Oak, S. W. 1983. Evaluations of a loblolly pine seedling survival and storage problem on the Sumter, Chattahoochee, and Oconee National Forests, 1983. USDA For. Serv. Rep. #83-1-22. Asheville, North Carolina. 15 p.

- Oldenkamp, L. and B. C. M. van Elk. 1966. (The storage period for forest plants.) Jaarb. Proefsta. Boomkwek., Boskoop 1966, 143-144. (Cited in For. Abstr. 28: 80. 1968.)
- Romberger, J. A. 1963. Meristems, growth, and development in woody plants. USDA Tech. Bull. 1293. 214 p.
- Samish, R. M. 1954. Dormancy in woody plants. Ann. Rev. Plant Phys. 5: 183-204.
- Steinhoff, R. J. and R. J. Hoff. 1972. Chilling requirements for breaking dormancy of western white pine seedlings. USDA For. Serv. Res. Note INT-153, 6 p.
- Stone, E. C. and G. H. Schubert. 1959a. Ponderosa pine planting stock. Calif. Agric. 13(3): 11-12.
- Stone, E. C. and G. H. Schubert. 1959b. The physiological condition of ponderosa pine (<u>Pinus ponderosa</u> Laws.) planting stock as it affects survival after cold storage. J. For. 57: 837-841.
- Ursic, S. J., H. L. Williston and R. M. Burns. 1966. Late planting improves loblolly survival. USDA For. Serv. Res. Pap. S0-24, 12 p.
- van den Driessche, R. 1975. Flushing response of Douglas-fir buds to chilling and to different air temperatures after chilling. British Columbia For. Serv. Res. Note 71, 22 p.
- Wakeley, P. C. 1954. Planting the southern pines. USDA Monogr. 18. 233 p.
- Wareing, P. F. 1969. The control of bud dormancy in seed plants, p. 241-262.

 <u>In</u> Woolhouse, H. W. (ed.). Dormancy and survival. Symp. Soc. Exp. Biol.,
 No. 23. Academic Press, New York.
- Weinberger, J. H. 1956. Prolonged dormancy trouble in peaches in the Southeast in relation to winter temperatures. Proc. Amer. Soc. Hort. Sci. 67: 107-112.
- Wells, S. P. 1979. Chilling requirements for optimal growth of Rocky Mountain Douglas-fir seedlings. USDA FOR. Serv. Res. Note INT-254, 9 p.
- Winjum, J. K. 1963. Effects of lifting date and storage on 2+0 Douglas-fir and noble fir. J. For. 61: 648-654.
- Winjum, J. K. 1961. p. 2 <u>In</u> Date of Lifting Symposium. Minutes by R. C. Austin. Crown Zellerbach Corp. 5-p. mimeo. November 2, 1961.

Table 1. Geographical and climatological data for seed sources (climatological data from Hocker, 1955).

Seed source	Approximate latitude	Approximate longitude	Approximate elevation	Average length of frost-free season	
			-(m)-	-(days)-	
Southeast Virginia (VA)	37 ° N	76° w	5	228	
Southwest Arkansas (AR)	34° N	94° W	120	229	
North Georgia (NG)	34° N	84° W	240	215	
Southeast Georgia (SG)	32° N	81° W	5	278	

Table 2. Mean days to reach 50 percent bud break for controlled environment study.

	Photoperiod ²	Time to 50% bud breakl			
Chilling		SG	VA		
-(h)-	-(h)-	-(days)-		
101	10	36.0 abc	37.0 ab		
	12	34.5 bcd	37.0 ab		
	14	36.0 abc	43.5 a		
245	10	21.0 fghi	27.0 defg		
	12	30.0 bcde	28.0 cdef		
	14	24.0 efgh	35.0 bcd		
341	10	16.0 hi	23.0 efgh		
	12	23.0 efgh	24.0 efgh		
	14	21.5 fghi	35.0 bcd		
413	10	15.5 hi	20.0 fghi		
	12	20.0 fghi	19.0 ghi		
	14		28.0 cdef		
485	10	17.0 hi	14.0 i		
	12	18.0 hi	17.0 hi		
	14	17.0 hi	27.5 def		

Means of 3 replications; means followed by the same letter are not significantly different at the 0.05 level of probability as compared by Duncan's Multiple Range Test.

² Photoperiod regime prior to chilling (hours of daylight).

Table 3. Seedling survival for natural environment study (one year following outplanting).

Sampling			Survivall					
Date	Chilling	Storage	VA	AR	NG	SG	BR ²	
-1983-	-(h)-	-(weeks)-			(%) -			
15 Nov.	98	0	84 a	86 a	89 a	72 ab	84 a	
		11	55 ъ	61 ь	28 Ь	12 c	32 b	
6 Dec.	223	0	85 a	95 a	88 a	89 ab	94 a	
		11	92 a	86 a	89 a	91 ab	89 a	
15 Dec.	314	0	75 ab	92 a	82 a	71 ab	86 a	
		11	85 a	96 a	89 a	95 a	90 a	
21 Dec.	407	0	81 a	100 a	89 a	68 Ъ	89 a	
		11	94 a	86 a	79 a	90 ab	95 a	

¹ Means of 5 replications; means followed by the same letter (within a seed source) are not significantly different at the 0.05 level of probability as compared by Duncan's Multiple Range Test.

² Southwest Arkansas seed source stored and planted as bare-root stock.

Table 4. First-year height of surviving outplanted seedlings for natural environment study.

Sampling			Height ^l				
Date	Chilling	Storage	VA	AR	NG	SG	BR ²
-1983-	-(h)-	-(weeks)-			- (cm) -		
15 Nov.	98	0	33.2 abc	29.8 ab	33.5 ab	32.0 bc	27.2 b
		11	26.5 d	25.8 ь	24.2 c	26.7 c	20.5 c
6 Dec.	223	0	35.1 ab	34.0 a	36.6 a	36.9 ab	27.3 b
		11	32.0 abc	30.8 a	33.3 ab	32.5 b	29.3 ab
15 Dec.	314	0	31.3 bcd	31.7 a	28.7 bc	33.4 ab	26.1 b
		11	35.0 ab	32.8 a	33.8 ab	37.6 ab	31.2 a
21 Dec.	407	0	28.8 cd	30.7 a	32.7 ab	31.9 bc	26.8 b
		11	36.6 a	34.4 a	34.8 a	38.3 a	28.9 ab

Means of 5 replications; means followed by the same letter (within a seed source) are not significantly different at the 0.05 level of probability as compared by Duncan's Multiple Range Test.

 $^{^2}$ Southwest Arkansas seed source stored and planted as bare-root stock.

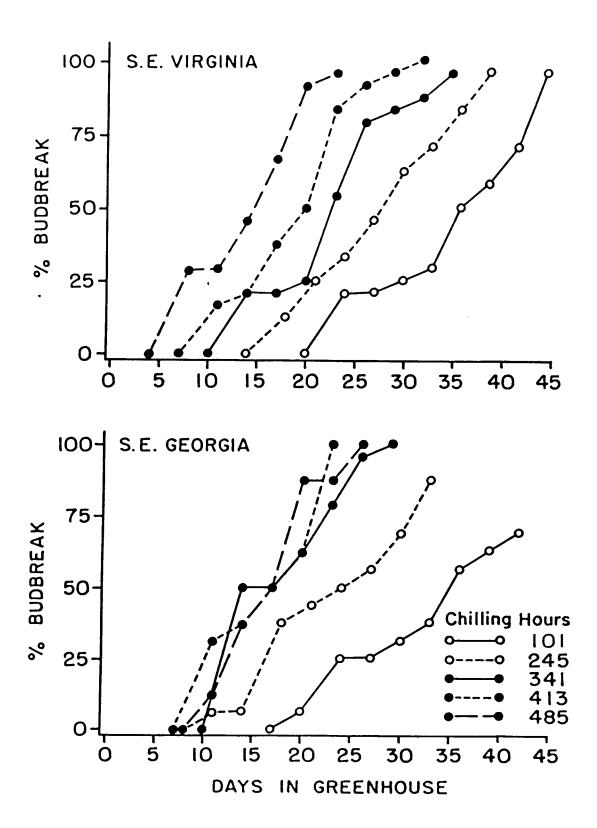


Figure 1. Bud break progression for the two seed sources. Seedlings received a 10-hour photoperiod for 7 weeks prior to chilling.