Ten-year growth comparison between rooted cuttings and seedlings of loblolly pine

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Abstract: Rooted cuttings and seedlings of loblolly pine (*Pinus taeda* L.) were established in a central Alabama field trial. Five, full-sib families, with an average number of six clones per family, were evaluated. Mean cutting/seedling height ratios revealed that despite initial differences in size, relative growth rates of both propagule types stabilized and were equal by age 7 years. Through age 10 years, results show virtually no difference in height, diameter at breast height, volume, or stem taper between the rooted cuttings and seedlings.

Résumé : Des boutures racinées et des semis de pin à encens (*Pinus taeda* L.) ont été Ctablis dans une plantation dans le centre de l'Alabama. Cinq descendances biparentales, avec un nombre moyen de six clones par descendance, ont été évaluées. Les ratios moyens de la hauteur des boutures sur celles des semis montrent que, malgré les différences initiales de dimension, les taux de croissance relative des deux types de propagules se sont stabilisés et Ctaient Cgaux après 7 ans. Jusqu'à 10 ans, les résultats ne comportent pratiquement aucune différence dans la hauteur, le diamètre à hauteur de poitrine, le volume ou le défilement de la tige entre les boutures racinées et les semis.
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Introduction

Worldwide interest in cloning forest trees has given rise to many programs to develop and apply either rooted cutting or tissue culture technology (Ritchie 1992; Lambeth et al. 1994). Recent breakthroughs in rooted cutting technology of the traditionally difficult-to-root species, such as loblolly pine (*Pinus taeda* L.) (Foster and Shaw 1987; Hughes 1987), have sparked widespread interest in this form of plant propagation throughout the commercial forest region of the southeastern United States. A prerequisite to the use of rooted cuttings in either research studies or commercial deployment, however, is the assurance that the growth and development of clonal propagules is comparable to that of seedlings.

Field trials comparing the growth of *Pinus* sp. rooted cuttings and seedlings have yielded mixed results. Fielding (1970) reported that in three studies ranging in age from 4 to 27 years, Monterey pine (*Pinus radiata* D. Don) rooted cuttings were slightly taller than seedlings. Pawsey (197 1) and Sweet and Wells (1974), however, found that the survival and growth of Monterey pine rooted cuttings were poorer than seedlings through ages 11 and 5 years, respectively. Rooted cuttings of eastern white pine (*Pinus strobus* L.) performed as well or better than seedlings for survival, height, and diameter at breast height after 40 years in the field (Struve et al. 1984). But, in a more recent study, Struve and McKeand (1990) reported that eastern white pine rooted cuttings were significantly shorter than seedlings after age 4 years, with differences between stock types increasing in magnitude through year 8. Foster et al. (1987) found that loblolly pine rooted cuttings performed better than seedlings through age 4 years, whereas in a companion study the reverse was true through age 6 years. In a third study with loblolly pine, Foster (1988) found no significant differences in growth or crown morphology between the two propagule types through age 3 years.

These conflicting results have been attributed to the age of the donor trees, and the unsuitable physiological condition and size of the rooted cuttings at the time they were planted. Maturation effects on rooting ability and subsequent growth of rooted cuttings are well known for a number of forest tree species (Sweet 1973; Greenwood 1984; Foster et al. 1987; Struve and McKeand 1990). In a study with slash pine (*Pinus elliottii* Engelm.) rooted cuttings, Franklin (1969) found a negative correlation between ortet (donor plant) age and height or diameter growth of the ramets (rooted cuttings). With loblolly pine, Foster et al. (1987) observed noticeable reductions in height and diameter growth of ramets from ortets as young as 4 or 5 years. Inherent differences in rooting ability led to variability in the growth and morphology of root systems (Greenwood and Nussbaum 198 1; Foster et al. 1984). Studies with eastern cottonwood (*Populus deltoides* Bartr. ex Marsh.), eastern white pine, and western hemlock (*Tsuga heterophyllu* (Raf.) Sarg.) have shown a positive relationship between root system quality and subsequent growth (Ying and Bagley 1974; Struve et al. 1984; Foster et al. 1985; Paul et al. 1993).

The induced root system can also be affected by environmental preconditioning of the individual donor plants, especially when the donor plants are growing in different locations. This effect has frequently been referred to as a common, or "C," effect and can result in an artificial inflation of clonal variance (Libby and Jund 1962; Wilcox and Farmer 1968; Foster et al. 1984; Cannel1 et al. 1988; Paul et al. 1993). In response to this problem, Libby and Jund (1962) developed a two-stage rooting procedure that has minimized, and in some cases eliminated, C-effects among rooted cuttings after a few

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Table 1. Average performance of rooted cuttings (RC) and seedlings (S) from five, full-sib families for a loblolly pine propagule comparison trial in St. Clair Co., Ala.

Trait	Propagule type	Family A × B	A × C	D × E	F × G	H x l	Family average
HT1 (m)	RC	0.42	0.41	0.40	0.41	0.45	0.42
	S	0.53	0.57	0.48	0.39	0.28	0.45
HT2 (m)	RC	1.38	1.24	1.15	1.14	1.21	1.22
	S	1.36	1.40	1.04	1.33	0.67	1.16
HT3 (m)	RC	2.68	2.62	2.36	2.29	2.43	2.48
	S	2.49	2.73	2.26	2.77	1.77	2.40
HT7 (m)	RC	9.18	8.65	8.95	8.77	8.80	8.87
	S	9.45	9.07	8.61	9.14	8.69	8.99
HT10 (m)	RC	12.74	11.64	12.23	12.26	11.80	12.13
	S	12.50	12.04	11.58	12.50	11.58	12.04
DBH3 (cm)	RC	3.82	3.40	3.31	2.76	2.93	3.24
	S	3.30	3.18	2.67	3.56	2.02	2.90
DBH7 (cm)	RC	14.64	13.97	14.07	13.45	13.90	14.01
	S	15.49	11.43	13.72	16.38	11.18	13.64
DBH10 (cm)	RC	19.54	18.01	18.62	19.13	18.63	18.79
	S	21.34	14.35	18.54	21.72	14.73	18.14
VOL7 (m^3)	RC	0.058	0.050	0.052	0.047	0.050	0.054
	S	0.066	0.035	0.047	0.071	0.032	0.049
VOL10 (m^3)	RC	0.142	0.115	0.124	0.133	0.120	0.131
	S	0.166	0.077	0.114	0.170	0.073	0.120
TAPER3 (%)	RC	58.12	54.90	55.59	47.21	48.87	52.94
	S	49.06	45.50	44.18	52.99	35.00	45.35
TAPER7 (%)	RC	82.02	78.98	79.08	81.43	82.66	80.83
	S	78.58	64.40	69.70	77.71	77.19	73.52
TAPER1 0 (%)	RC	78.20	78.45	79.51	81.27	82.26	79.94
	S	74.49	64.46	69.64	88.46	69.05	72.82

Note: Trait codes explained in text.

years in the field (Cannel1 et al. 1988; Farmer et al. 1988; Paul et al. 1993).

The calculation of relative growth rate values, and the correction of these values for covariance on tree size, enables one to evaluate growth rates independently of planting size (Sweet and Wells 1974). A simpler method of comparing relative growth rates between rooted cuttings and seedlings established at different sizes, however, is to plot the cutting/seedling ratio for a given trait over time (Frampton 1986; Amerson et al. 1988; Frampton and Isik 1987). If rooted cuttings and seedlings are growing at the same rate this ratio will remain constant over time. An increase or decrease of this ratio over time indicates a respective increase or decrease of the relative growth rate of the rooted cuttings compared with seedlings.

The objective of this study was to compare the growth of rooted cuttings and seedlings from the same full-sib families of loblolly pine.

Material and methods

Seeds were obtained from five full-sib families of loblolly pine of coastal North Carolina origin. The parents were randomly chosen from a population of first-generation selections in an industrial tree improvement program operating in the region. Nine unrelated parents yielded five full-sib families in which only two families shared a common parent; otherwise, the families arose from single-pair matings.

A two-stage cloning procedure similar to Libby and Jund (1962) was used to produce the cutting material. Seedlings from each family were grown during the winter and spring of 1983, and cuttings were collected and set for rooting in June 1983. Once rooted, the cuttings (primary ramets) were planted in pots and grown in a greenhouse during the winter and spring of 1984. In June 1984, cuttings were collected from the primary ramets and set for rooting. At this point, the trees were 18 months old from seed. Following rooting, the rooted cuttings (secondary ramets) were grown in a greenhouse over the winter of 1985.

The rooting protocol followed the one described by Hughes (1987). Briefly, the cuttings were dipped in water followed by sticking 1 cm of the base into a talc-based rooting compound (Hare 1979). Each treated cutting was then set to a depth of 1 cm in a 97-cm^3 polyethylene pot containing a 3: 1 (v/v) mixture of peat and perlite. Air temperature in the propagation greenhouse was maintained at 2 1 °C, and relative humidity was maintained at 100% by generating a 4- min fog every 8 min. An average CO, concentration of 1500 ppm was supplied by compressed gas cylinders. High urea foliar feed fertilizer (27: 15: 12 N–P–K) was applied daily at 33 ppm N through an overhead traveling boom irrigator. An 18-h photoperiod was maintained throughout the rooting phase via incandescent and fluorescent lights. Three weeks after the cuttings were set, the rooting medium was leached on a weekly basis to remove any excess soluble salts that accumulated from the foliar fertilization. The cuttings were kept in this propagation phase for 12 to 16 weeks, after which time humidity levels were gradually decreased and the rooted cuttings were allowed to acclimate to normal greenhouse conditions.

Additional seedlings from the original family seed lots were produced in an adjoining greenhouse during the winter and spring of 1985. They were grown in the same containers and medium as the rooted cuttings and under similar environmental conditions minus the

Table 2. Analysis of variance probability values and family-mean correlations (r_{CS}) between rooted cuttings (C) and seedlings (S) associated with several growth traits for a loblolly pine propagule comparison trial in St. Clair Co., Ala.

Trait	Source of variation”			R^2	r_{CS}
	P	F	P × F		
HT1	0.5918	0.3096	0.1000	0.28	–0.74*
HT2	0.6249	0.0034	0.0140	0.46	0.33
HT3	0.7730	0.0036	0.0409	0.47	0.41
HT7	0.4656	0.2840	0.8965	0.52	0.35
HT10	0.9614	0.5502	0.8863	0.49	0.56
DBH3	0.3486	0.0999	0.4007	0.31	0.41
DBH7	0.7397	0.0004	0.0011	0.54	0.48
DBH10	0.773 1	0.0005	0.0134	0.49	0.86*
VOL7	0.8946	0.0009	0.0066	0.50	0.19
VOL10	0.7152	0.0003	0.0949	0.48	0.92*
TAPER3	0.0811	0.2959	0.3535	0.33	0.22
TAPER7	0.1251	0.0001	0.0181	0.72	0.92
TAPER10	0.1514	0.000 l	0.0014	0.63	0.38

Note: Trait codes explained in text
*Significant at α = 0.05.
[a]P, propagule type; F, family.

high relative humidity levels supplied by the intermittent fog. These seedlings along with the rooted cuttings (secondary ramets) from each family were planted in St. Clair County, Alabama (AL), on April 29–30, 1985. Care was taken to coordinate the growth of the seedlings in the greenhouse so that they were of similar size to the rooted cuttings.

A completely randomized field design was used. Two to four seedlings represented each family in a noncontiguous plot fashion. In addition, one to four ramets from each of five to eight cloned individuals per family were planted. As with the seedlings, ramets of each clone were planted in a noncontiguous arrangement, intermingled with the seedlings. The field test consisted of a total of 15 seedlings and 132 rooted cuttings. Trees were spaced at 2.44 x 2.75 m.

The following traits were measured over a lo-year period in the field: (i) total height (m) at age 1 (HT1), 2 (HT2), 3 (HT3), 7 (HT7), and 10 (HT10) years; (ii) stem diameter (cm) at 1.3 m above groundline at age 3 (DBH3), 7 (DBH7), and 10 (DBH10) years; (iii) stem diameter (cm) at 15 cm above groundline at age 3 (DGL3), 7 (DGL7), and 10 (DGL 10) years.

From the measured traits, the following traits were derived: (iv) stem taper, at age 3 (TAPER3), 7 (TAPER7), and 10 (TAPERIO) years as calculated by

$$[1] \quad \text{TAPER} = (\text{DBH}/\text{DGL}) \times 100$$

(v) stem volume (m^3), at age 7 (VOL7) and 10 (VOL10) years, as calculated by Smalley and Bower (1968):

$$[2] \quad \text{VOL} = (0.011\,82 + (0.001\,998\,94(\text{DBH}^2) \times \text{HT})) \times 0.028\,32$$

The cutting/seedling ratios for mean height and mean diameter (1.3 m above groundline) of each family were plotted over time to compare the relative growth rates among the propagule types. The seedlings were considered the standard of comparison, and their mean size was used as the denominator of the ratio.

Since cloned individuals were represented by multiple ramets but seedlings were not, clone means for the rooted cuttings and individual values for the seedlings were used in the analysis of variance. Propagule type was treated as a fixed effect, and all other sources were considered to be random effects. A least squares analysis was used to calculate the type Ill sums of squares and appropriate F-statistics (SAS Institute Inc. 1989). To test for association of the growth performance between the propagule types, family-mean correlations were calculated between propagule types for each trait.

Results and discussion

The rooted cuttings have survived (92%) and grown quite well (HT10 = 12.13 m; Table 1). Unfortunately, few seedlings from each family were available for planting. Furthermore, survival of the seedlings was only 67% at age 3 years because of mechanical damage that inadvertently killed 20%. The remaining seedlings, however, have grown similarly to the rooted cuttings (HT10 = 12.04 m).

Despite the imbalance of numbers of seedlings (n = 10) versus numbers of rooted cuttings (n = 121), the use of clone means greatly reduced the disparity, since there were, on average, six clones per family (34 clones total). When seedlings were compared with clonal averages, rooted cuttings and seedlings displayed virtually identical heights, stem diameters, and stem volumes (Table 1) with no significant difference for any trait (Table 2). Even in the case of stem taper, rooted cuttings did not significantly differ from seedlings.

Large differences for the mean height cutting/seedling ratio existed, however, among the families at age 1 year (Fig. 1). Measurements were not obtained at the time of outplanting, but the height of all transplants was relatively uniform. These differences in first-year cutting/seedling height ratios are likely due to variation in the quality of cutting root systems among families and consequent differences in early establishment. Early propagule differences, both within and among families, dramatically diminished within 7 years. By age 7, all the trees within a family were virtually the same size, regardless of propagule type. The observation that differences between propagule types diminished over time supports the speculation that family variation for earlier ratios resulted from differences in root system quality rather than permanent maturity effects.

Family genotype had a strong effect on the early relative growth rates for height between propagule types. Rooted cuttings were shorter than seedlings in three of the five families, and the relative growth rate of the rooted cuttings increased over time compared with seedling growth. In the fourth family, the rooted cuttings lagged behind their respective seedlings, but by age 7 years regained most of the ground they lost. The cutting/seedling ratio in the fifth family was most likely inflated because both of the seedlings were very small.

Relative growth rate comparisons for tree diameter were less conclusive (Fig. 2). The cutting/seedling ratios calculated at age 3 years show greater family differences than mean height cutting/seedling ratios. For three of the five families, some convergence to 1 .O was observed as time progressed. The ratio for two families, however, remained at 1.2, indicating that the rooted cutting propagules were growing faster than the seedlings.

Rooted cuttings are notoriously difficult to match in initial size and maturation level with seedlings (Sweet 1973; Foster et al. 1987; Frampton and Foster 1993). The initial size difference subsequently influences the growth rate (Sweet 1973). In addition, rooted cuttings may be in a different phase of maturation than seedlings, which leads to differences in growth and morphology (Sweet 1973; Greenwood 1984; Foster et al. 1987). Even though the propagule types in the present study

Fig. 1. Comparison of relative growth rates between loblolly pine rooted cuttings and seedlings by plotting over time the mean cutting/seedling height ratios for each of the five full-sib families denoted in the insert. Initial height data at time of outplanting were not collected.

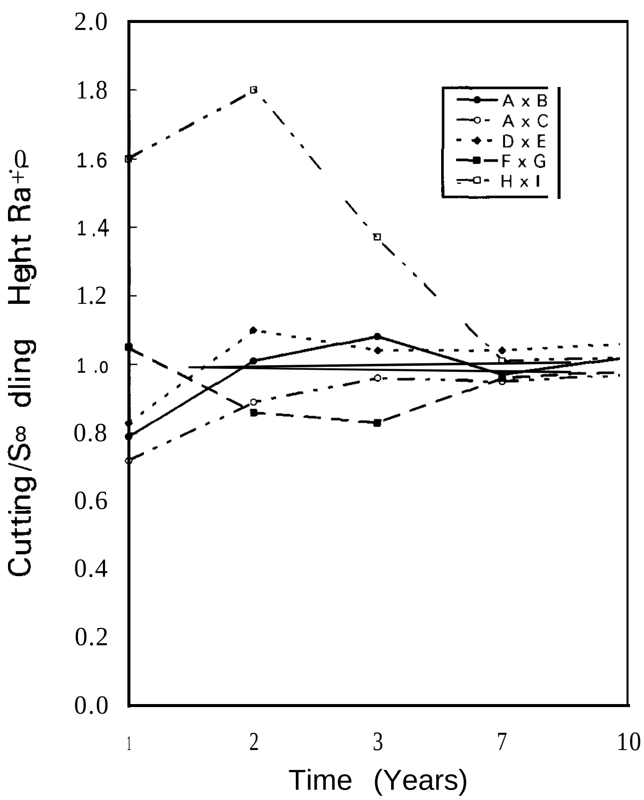
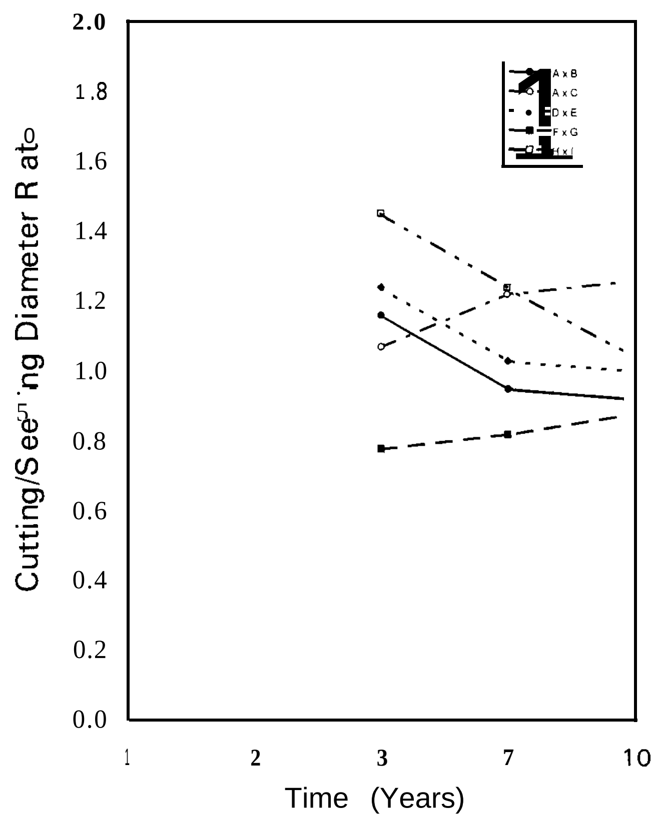


Fig. 2. Comparison of relative growth rates between loblolly pine rooted cuttings and seedlings by plotting over time the mean cutting/seedling diameter ratios for each of the five full-sib families denoted in the insert. All diameter data were collected 1.3 m above groundline.



were somewhat mismatched, the juvenile nature of the rooted cuttings (18 months from seed) encouraged growth and development that mimicked that of a seedling.

The family effect was significant for all traits except HT1, HT7, HT10, DBH3, and TAPER3 (Table 2). The small sample sizes for seedlings or clones within families inflated error variances and caused significant family effects to be detected only when among-family variances exceeded about 25% of the total variance.

The propagule type by family interaction effect was significant for HT2, HT3, DBH7, DBH10, VOL7, TAPER7, and TAPER10 (Table 2). This finding may indicate the differential progression of maturation by family and trait, or it may be a function of lack of test precision due to small sample sizes, especially for the seedlings. Foster et al. (1987) found little evidence for significant propagule type x family interaction in their study of loblolly pine. Therefore, the current result is suspected to be an artifact of sample size, especially given the convergence toward 1.0 for the overall cutting/seedling ratio for height and diameter in Figs. 1 and 2, respectively.

The family-mean correlation between propagule types for HT1 displayed a significant and strong negative correlation (Table 2). This is most likely the result of initial size and vigor differences between propagule types (Burdon and Sweet 1976). As the trees grew, the correlations for HT_ became more positive. At age 10 years, both DBHlO and VOL10 displayed a strong and significant positive correlation. These correlations, together with the pattern of relative growth rates for height (Fig. 1) and diameter (Fig. 2), indicate a converging agreement between propagule types.

Although comparisons between the growth of rooted cuttings and seedlings for several forest trees species have accumulated in the literature for a number of years, results for loblolly pine are just beginning to appear. Ten-year results from this study indicate that rooted cuttings from juvenile donor plants grow in a very similar fashion to seedlings and that they can be used for either reforestation or research. Comparison of relative growth rates, however, suggests early-age propagule differences exist and caution should be exercised in short-term research studies.

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