TOP GRAFTING LOBLOLLY PINE IN THE WESTERN GULF REGION

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Abstract:--Flowering data were collected from top grafts made in 1996 and 1997 at the Mississippi Forestry Commission's Craig Seed Orchard near Lumberton, MS. Scion material from twenty loblolly pine *(Pinus taeda)* second-generation selections was grafted onto five loblolly pine and five slash pine *(P. elliottii)* interstocks. All interstocks had a history of good cone production. Data collected included the number of female strobili per graft, scion growth, the number of branches above the graft union and the numbers of conelets per graft in the second year after grafting.

First-year measurements of 1996 grafts suggested that grafts on slash pine interstocks had an advantage over grafts on loblolly pine interstocks. Average survival of grafts on slash pine interstocks was 58%, of which 94% flowered with an average of 14.4 strobili per graft. Grafts on loblolly pine interstocks had an average survival of only 25%, of which 43% flowered with an average of 4.8 strobili per graft. Second-year measurements of the 1996 grafts showed much smaller differences between interstock species. Average graft survival declined slightly to 50% for slash pine interstocks and 21% for loblolly pine interstocks. Flowering increased to 84% on the surviving grafts with 12.7 strobili per graft on slash pine interstocks and 86% flowering with 10.5 strobili per graft on loblolly pine interstocks. The similar performance of interstock species in the second-year data suggested that the first-year differences were the result of a differential response to a hard spring freeze that occurred at the orchard in 1996.

Grafts made in 1997 had an average first-year survival of 91% on slash pine interstocks and 84% on loblolly pine interstocks. Of the surviving grafts, 56% flowered on slash pine interstocks producing an average 4.9 female strobili and 58% flowered on loblolly pine interstocks with an average of 5.2 female strobili. No significant differences were detected between the two interstock species in graft survival, average numbers of strobili, scion diameter or number of branches above the graft. Average scion length after one year's growth did differ between the slash pine (742 mm) and loblolly pine (586 mm) interstocks. Conelet survival after two years was not significantly different between loblolly pine interstocks (2.6 conelets per graft) and slash pine interstocks (2.2 conelets per graft). No significant interaction effect was observed between interstock species and scion clone for either the 1996 or 1997 grafts in the first or second year after grafting.

Top grafting has been shown to be an effective way to shorten the breeding cycle for loblolly pine improvement programs. The rapid production of strobili using young scion material can greatly accelerate breeding and shorten generation intervals. Members of the Western Gulf Forest Tree Improvement Program are already using top grafting as a part of their regular breeding programs. The results of this study suggest that flowering of loblolly pine scions can be promoted by top grafting on to either slash pine or loblolly pine interstocks.

Keywords: *Pinus taeda, P. elliottii,* tree breeding, seed orchard, grafting

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INTRODUCTION

Shortening the breeding cycle, the time between the selection of a genotype in one generation and the selection of a genotype in the following generation, is an important goal for tree breeders. One way to accelerate the breeding cycle is to shorten the generation interval, the time it takes to produce seed in large enough numbers to test and select genotypes. Forest tree species typically have a long generation interval with an extended juvenile stage of several years before reaching reproductive maturity. Techniques that result in earlier flowering shorten the generation interval and have important economic implications.

Top grafting is the grafting of young scion material into reproductively mature trees. This technique has been shown to promote early flowering of loblolly pine *(Pinus taeda* L.) in a Georgia seed orchard (Bramlett and Burris 1995). Early flowering in loblolly pine can also be induced by other methods, including hormonal and water stress treatments (Burris et al. 1991). These techniques can be effective, but are appropriate for an indoor, greenhouse-based accelerated breeding program. Top grafting is a relatively low-tech, low-cost technique that can be performed on trees outside in a scion bank or orchard using familiar skills and tools. The first objective of the study was to determine if top grafting effectively promotes early flowering in other parts of the range of loblolly pine, aside from the areas where it has been tested.

Another important consideration in top grafting is the effect of interstock, the section of the mature tree between the original rootstock and the newly top grafted scion (reviewed in Jayawickrama et al. 1991). Both loblolly and slash pine are commonly used as rootstock for loblolly pine grafts. The effect of rootstock on grafted conifers is an important consideration for orchard managers, even if the mechanisms of graft success and incompatibility are not fully understood. As there are mature seed orchards of both loblolly pine and slash pine *(Pinus elliottii* Engelm.) suitable for top grafting in the Western Gulf region, a secondary objective of the study was to determine if choice of interstock species would affect survival and flower production in loblolly pine top grafts.

METHODS

The Mississippi Forestry Commission (MFC) selected ten clones for use as interstocks from the Craig seed orchard complex located near Baxterville, MS (Byram and Lowe 1996). Five interstock clones were loblolly pine and five were slash pine. The interstock clones all had a history of good cone production and did not show signs of incompatibility, needle cast or any other disease.

Scions were collected in January of 1996 from one- to six-year old grafts from twenty selections in the Craig seed orchard second-generation scion bank. The ramets in the scion bank were originally grafted from five-year old (19 selections) or ten-year old (one selection) ortets in progeny tests. Only selections that had not previously flowered in the scion bank were chosen for scion collection. Approximately twelve pieces of scion were collected from each selection and stored until grafting.

One graft of each scion clone was made on each interstock clone so that each scion-interstock combination was represented once in the experiment, for a total of 200 grafts. In most cases, one interstock ramet did not have enough branches to accommodate all twenty grafts and a second interstock ramet was used. Cleft grafts were made in the upper crown using the typical wax grafting method (White et al. 1983). All grafts on a particular interstock clone were made at the same time, and all grafting was completed within a few days of scion collection. The twenty scion clones were grafted at random on each interstock clone, and each graft was tagged with a unique number.

The experiment was repeated in 1997 using the same scion clones and nine of the ten original interstock clones, although different ramets of the interstock clones were used. One of the loblolly pine interstock clones used in the 1996 grafting experiment started to die out in the orchard in 1997, so it was replaced with another interstock clone. The replacement interstock clone had a similar history of good cone production as the clone it replaced and showed no sign of incompatibility or disease. Grafting followed the same procedures as in 1996.

After the first growing season, the following data were collected: graft survival, number of female strobili, presence or absence of male strobili, diameter of the scion 5 cm above the graft union, length of the scion from the graft union to the terminal, and the number of first-order branches on the scion. These data were collected again for the 1996 grafts two years after grafting. Grafts made in 1997 were measured only after the first year, but graft survival was tallied and conelets on these grafts were counted after the second growing season. Data was analyzed using the SAS (SAS Institute, Cary, NC) GLM procedure. Mean values for scion length, scion diameter, number of branches, number of female strobili and number of conelets were estimated using the least squares means procedure to account for imbalance in the data due to mortality. Graft survival and the proportion of living grafts with male and female strobili were calculated directly from the data. All tests of significance were performed at the 10% level of probability.

RESULTS AND DISCUSSION

Measurements of 1996 grafts after the first year of the study indicated an advantage to grafting on slash pine interstock as compared to grafting on loblolly pine interstock (Byram et al. 1997). Although top grafting promoted flower production in both interstock species, graft survival, growth and strobilus production was much higher on slash pine interstocks than loblolly pine interstocks (Table 1).

Table 1. First-year growth and flowering of loblolly pine grafted on two interstock species in 1996.

Interstock Species	Survival (%)	Scion length (mm)	Scion diameter (mm)	Number of branches	Living grafts with female strobili (%)	Number of female strobili/ living graft	Living grafts with male strobili (%)
Loblolly Pine	25	377.7 a'	8.5 a	3.6 a	43	4.8 a	4
Slash Pine	58	505.3 b	10.8 b	4.6 a	94	14.4 b	12

Least squares means followed by the same letter are not significantly different at the 10 percent confidence level by Student's t test.

Survival and flowering percentages of grafts on loblolly pine interstocks were less than half that of grafts on slash pine interstocks. The average number of female strobili produced per surviving graft was significantly different between interstock species, approximately three times higher for grafts on slash pine interstocks than those on loblolly pine interstocks. Scion length, diameter and number of branches were all higher for grafts on slash pine interstocks than grafts on loblolly pine interstocks. Differences

between interstock species in scion lengths and scion diameters were statistically significant, but the difference between interstocks in the number of branches per graft was not significant.

The difference between interstock species for 1996 grafts was sharply reduced in the second year after grafting (Byram et al. 1998). Most of the grafts that survived the first year were still alive after two years, so although there was still a large difference in graft survival between slash pine and loblolly interstocks, the change in survival during the second year was small and similar for both interstock species. Flowering success on surviving grafts two years after grafting was approximately equal for slash pine and loblolly pine interstocks (Table 2). Interstock species did not differ significantly in the number of strobili produced or the number of branches above the graft union. Interstock species did differ significantly in average scion length and diameter.

Table 2. Second-year growth and flowering of loblolly pine grafted on two interstock species in 1996.

Interstock Species	Survival (%)	Scion length (mm)	Scion diameter (mm)	Number of branches	Living grafts with female strobili (%)	Number of female strobili/ living graft	Living grafts with male strobili (%)
Loblolly Pine	21	939.3 a[1]	21.0 a	9.1 a	86	10.5 a	0
Slash Pine	50	1295.3 b	25.0 b	10.6 a	84	12.7 a	40

Least squares means followed by the same letter are not significantly different at the 10 percent confidence level by Student's t test.

Grafts made in 1997 had high average survival after one year. First-year measurements of grafts made in 1997 showed much smaller differences between loblolly pine interstocks and slash pine interstocks in graft survival and flower production (Table 3). Grafts on slash pine and loblolly pine interstocks were significantly different in scion length, but not in number of female strobili, scion diameter, or number of branches per graft.

Survival of grafts made in 1997 dropped slightly in the second year to 90% for grafts on slash pine interstocks and remained at 84% for grafts on loblolly pine interstocks. After two years, there were 2.6 conelets surviving per graft on loblolly pine interstock and 2.2 conelets surviving per graft on slash pine interstocks. Differences between interstock species in survival and number of conelets were not statistically significant at the 10% level.

The strong difference between interstock species in the first-year measurements of the 1996 grafts was most likely due to a hard spring freeze. Graft success for both species was very poor relative to that of the grafts made the following year. The grafts on the slash pine interstock appeared to resist freezing damage better than grafts on loblolly pine interstock, based on the survival, flowering and growth data. This difference was not seen in the second-year measurements of the 1996 grafts or the first-year measurements of the 1997 grafts.

Table 3. First-year growth and flowering of loblolly pine grafted on two interstock species in 1997.

Interstock Species	Survival (%)	Length (mm)	Diameter (mm)	Number of branches	Grafts with female strobili (%)	Number of female strobili/ living graft	Grafts with male strobili (%)
Loblolly Pine	84	585.7 a[1]	11.9 a	4.7 a	58	5.2 a	0
Slash Pine	91	741.6 b	12.7 a	5.0 a	56	4.9 a	3

[1] Least squares means followed by the same letter are not significantly different at the 10 percent confidence level by Student's t test.

Significant differences in graft survival, scion growth and flowering success were observed among scion clones for both the 1996 and 1997 grafts at one and two years after grafting. This was expected, because there are usually strong differences in graft take, growth rate, and quantity and timing of flowering among clones in the same orchard. Although clonal differences were strong, no interaction effects between scion clone and interstock species were detected for any of the traits analyzed in this study.

There was a slight difference between 1996 grafts and 1997 grafts in the proportion of grafts that produced male strobili. Only three grafts on one slash pine interstock produced male strobili in the 1997 grafts. In contrast, 12% of the grafts on slash pine interstock and 4% of the grafts on loblolly pine interstock produced male strobili in the 1996 grafts after one year. After two years, 40% of the grafts on slash pine interstocks produced male strobili and grafts on loblolly pine interstocks produced no male strobili. This substantial change may be due to the hard spring freeze in 1996. The damage to the newly grafted tissue may have induced early production of male strobili as an 'emergency response'. The more typical weather conditions in the spring of 1997 did not present a survival challenge to the new grafts and male flower production was very low, which was expected for such young scion material grafted into the upper crown. Higher production of male strobili would be expected for grafts made into the lower crown of mature trees, but not for grafts in upper crowns as in this experiment.

Because most of the second-generation selection clones used in this study had still not flowered in the scion bank after the first year of this experiment, some of the strobili produced by the 1996 grafts were used for the MFC's controlled pollination program. The installation and removal of pollination bags was easier on the slash pine interstock than the loblolly pine interstock. This was due to the faster growth of the scions on slash pine interstocks, resulting in sturdier limbs to support the pollination bags.

Strobili on five top grafts on one of the slash pine interstock clones were control pollinated with polymix pollen in the spring of 1997. Normally, polymix breeding is done on scion material several years after it is grafted into a scion bank. Breeding is not normally performed on scion material as young as that used in the top grafting experiment. Flower retention for these top graft crosses was 79%, compared to 68% for the operational loblolly pine polymix breeding completed the same year. This difference was attributed to the difference in sample size (49 flowers debagged on top grafts compared to 400 flowers in the operational sample). Also, it is likely that the top grafts were treated more carefully during installation and removal of pollination bags. When strobili were counted in December 1997, 46 of the

original 49 top graft flowers remained. Cones were collected in the fall of 1998, including 39 sound cones from the top grafts. The seed yield from the top graft cones (60.3 sound seed per cone) compared very favorably to the 61.9 sound seed per cone from the operational polymix crosses.

CONCLUSION

Top grafting effectively reduced the generation interval in loblolly pine under the conditions in this study. While we cannot rule out the possibility that serial grafting may increase flowering more than first-time grafting, the results of this study suggest that loblolly pine scion material can be top grafted on to either loblolly pine or slash pine interstocks successfully. The results of the first-year measurements of the 1996 grafts did show a difference in graft take and flowering success between interstock species, but this could be attributed to differential damage from a hard spring freeze in 1996. No strong interstock species differences were apparent in the 1996 grafts after two years or in the 1997 grafts after one year.

The lack of interstock species differences suggests that orchard managers may choose to use either loblolly pine or slash pine for interstocks without a penalty in graft take or strobilus production. However, anecdotal evidence suggested that faster growth of grafts on slash pine branches made bagging slightly easier and faster. The preliminary seed yield data suggested that control-pollinated strobili on top grafts suffer no reduction in seed yield compared to control-pollinated strobili on older scion material. This study suggests that with top grafting, advanced generation selections can be made and grafted in one spring, produce strobili and be pollinated the next spring, and produce cones with a good yield of sound seed in the fall of the following year, shortening the breeding cycle by five to six years. This reduction in generation interval can increase the genetic gain captured per year by significantly shortening the generation interval.

Based on the results of this study, most members of the Western Gulf Forest Tree Improvement Program have incorporated top grafting into their standard breeding strategies. While all of the loblolly pine second-generation selections identified in 1998 were grafted into traditional scion banks for preservation, many were also top grafted for accelerated flower production.

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LITERATURE CITED

Bramlett, D. L. and L. C. Burris. 1995. Topworking young scions into reproductively-mature loblolly pine. In: Proc. 23rd SFTIC Conf. Asheville, NC. pp. 234-241.

Burris, L. C., C. G. Williams, and S. D. Douglass. 1991. Flowering response of juvenile selections in loblolly pine. In: Proc. 21st SFTIC Conf. Knoxville, TN. pp. 110-119.

Byram, T. D. and W. J. Lowe. 1996. 44th Progress report of the cooperative forest tree improvement program. Tex. For. Ser. Circular 296. 25 pp.

Byram, T. D., F. E. Bridgwater, G. D. Gooding and W. J. Lowe. 1997. 45th Progress report of the cooperative forest tree improvement program. Tex. For. Ser. Circular 297. 30 pp.

Byram, T. D., F. E. Bridgwater, G. D. Gooding, and W. J. Lowe. 1998. 46 th Progress report of the cooperative forest tree improvement program. Tex. For. Ser. Circular 301. 23 pp.

Jayawickrama, K. J. S., J. B. Jett, and S. E. McKeand. 1991. Rootstock effects in grafted conifers: A review. New Forests 5: 157-173.

White, G., W. J. Lowe, and J. Wright. 1983. Paraffin grafting techniques for loblolly pine. Southern Journal of Applied Forestry 7(3): 116-118.