

**FIFTY-SECOND**

**PROGRESS REPORT**

**OF THE**

**COOPERATIVE**

**FOREST TREE IMPROVEMENT**

**PROGRAM**

**By**

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## INTRODUCTION

The Western Gulf Forest Tree Improvement Program continues to change in ways that reflect the rapid restructuring of the forest industry. During the past year CellFor, Inc., a service company whose primary product is clonal varieties for reforestation, became the group's newest member. Boise was privatized and subsequently announced that their timberlands would be sold to a timber investment group. As the cooperative enters 2005, membership stands at fourteen. This includes five public state forestry agencies, a real estate investment trust, two timber investment management organizations, a regeneration service provider, and five traditionally integrated forest industries. Changing membership coupled with the rapid development of new technology has made it necessary for the cooperative to keep rethinking both its mission and its methods.

One substantial challenge has been defining our clients and hence, our breeding objectives in the face of declining numbers of the integrated forestry companies supporting tree improvement. Are the cooperative's clients the landowners who grow the trees or are they the manufacturers that process raw wood into a finished product? If our clients are the manufacturers that use wood as a raw material, then improvement in wood quality is an obvious selection criterion. A significant portion of the manufacturing sector, however, has divested itself of timberland ownership, in effect taking the position that there is an adequate supply of quality wood available at reasonable prices. By taking this step, they have forfeited any interest in influencing the direction of future breeding programs. If we, on the other hand, identify our clients as the landowners who grow timber for the commodity market, then the breeding objective becomes achieving the maximum volume production while maintaining only minimum quality standards. Under this scenario, the cooperative becomes unwilling to give up gains in growth rate for improvements in wood quality.

Most of our members have taken the position that wood is our ultimate product, and they desire simultaneous improvement in both wood quality and volume growth. For wood quality to be a viable selection criterion, however, it has to have a recognizable economic value reflected in procurement systems that explicitly pay a premium for better and more uniform wood. If not, landowners have no incentive to produce such material and the cooperative has no way of constructing rational selection indices. Despite the fact that the majority of current procurement systems are based on volume, the cooperative continues to develop strategies for incorporating wood quality into our breeding objectives.

Wood quality, as determined by stem straightness, wood specific gravity, and microfibril angle, has either neutral or slightly negative genetic correlations with volume growth in the loblolly pine progeny tests evaluated by the WGFTIP. Selection for volume growth alone can thus be expected to result in decreased wood quality over time. As a result, the WGFTIP must define the best possible set of selection criteria given that 1) no single set of breeding objectives can be considered optimal for the multiple products produced by our members and 2) improvement in wood quality has no readily recognized and commonly agreed upon economic importance

in the existing market. To meet this challenge, the WGFTIP has implemented different strategies for the mainline breeding population and the various deployment populations tailored to meet the needs of specific members. An elite wood quality population based on backwards selection of parents is also under development. The structure of this population and the creation of a selection population produced as either seedlings or clonal lines will be described below.

Each WGFTIP member tailors their independently managed deployment population to meet their unique needs. The choice of clones making up a new orchard can have considerable impact. For example, if clones for a South Arkansas orchard are selected for volume alone, they have an expected breeding value of nearly 38 percent for growth expressed as percent improvement in mean annual increment at age 20 compared to an unimproved source, a slight improvement in stem straightness, and lower specific gravity than the unimproved checklot. Selection of a twenty-clone orchard for the same deployment area using the pulp index method that gives specific gravity an economic weight seven times larger than volume growth would result in an expected breeding value of 15 percent for growth, a similar score for stem straightness and an improvement of 0.03 for specific gravity compared to the unimproved checklot. Actual orchard design is frequently a trade off between these two extremes as exemplified by those designed for two state agencies, which together grafted a total of 26 acres of new orchard in 2004. These orchards, developed to provide trees primarily to non-industrial private landowners, had a projected average improvement in volume growth of 35 percent in mean annual increment at age 20 while specific gravity was only slightly below the existing regional average.

The ability to tailor the makeup of individual member's deployment populations for multiple traits comes at a cost. More selections must be available than would be required to support improvement in a single trait. The cooperative's loblolly pine selection population now consists of 3,123 progeny tested first-generation parents, 248 progeny tested second-generation parents, and 1,339 additional second-generation selections that have been identified and are in the pipeline. Subsequent generations will not need to be this large as individuals that combine desired traits are selected for the mainline breeding population and as smaller more targeted elite populations are developed.

Field establishment of first-generation progeny tests was completed for all but the last remaining loblolly pine diallel during the 2003/04 planting season. One member established three duplicate plantings of five diallels containing 41 previously untested parents in East Texas. Only eight loblolly pine parents remain to be tested for which seed is already in hand. First-generation slash pine breeding has also been completed, but final progeny test establishment will be delayed until the summer of 2005. These accomplishments are notable milestones in the history of the WGFTIP and its members.

Members within breeding regions continue to work

<sup>1</sup>Lowe, W.J., T.D. Byram, and F.E. Bridgwater. 1999. Selecting loblolly pine parents for seed orchards to minimize the cost of producing pulp. For Sci. 45(2):213-216.

together to complete advanced-generation breeding and progeny test establishment. All available polymix seed is pooled and responsibility for test establishment is shared. This ensures that field tests are planted as rapidly as seed becomes available and has proven much more efficient than individual members attempting to run independent programs. In 2004, advanced-generation loblolly pine plantings were established in Arkansas and Texas containing 70 and 80 polymix families, respectively. Breeding is progressing at a sufficiently rapid pace to allow additional loblolly pine polymix tests to be sown in both of these regions during the summer of 2004. Advanced-generation slash pine polymix tests were also planted in 2003/04. These tests will provide field data on 45 second-generation parents for this species.

If the cooperative's first challenge from the continued realignment within the forestry industry was the need to identify our clients, the second challenge has been in defining our membership. Traditionally, only land-based organizations supporting large regeneration programs on corporate and/or non-industrial private land could justify the considerable investment needed to support a tree improvement program. For the most part, these organizations have been either state agencies promoting conservation and economic development or integrated forest industries with mills to supply. These organizations had a set of common goals, which included the benefits of ensuring an inexpensive and abundant raw material resource was available to the manufacturing sector. Furthermore, all organizations were very similar in that they had access to the same types of resources, could be expected to apply very similar methods, and could supply similar in-kind support to the collaborative effort. Recent changes in the business environment and advances in technology have made this description of the tree improvement community far too simplistic.

The first change the cooperative experienced was the emergence of the real estate investment trust and the timber investment management organization as major landowners. These groups have grown rapidly in both numbers and in size as traditionally integrated forest industries have sold significant amounts of forestland. As an example, nearly half of the commercial timberland owned by integrated forest industries in southeast Texas as recently as five years ago is now managed by timber investment organizations. Similar trends are apparent across the region. While many of these entities are too small to justify direct investments in tree improvement, even the larger organizations own few manufacturing facilities, produce timber primarily as a commodity to be sold to others, and many have been incorporated for a fixed period of time. They are, therefore, much more similar to non-industrial forest landowners than to our traditional corporate members. The second change was the emergence of forest biotechnology service corporations modeled closely after agricultural seed companies. These organizations typically own no land, and therefore have no internal use for planting material and no infrastructure to support traditional tree improvement programs.

The decline in the number of traditionally integrated forest industries and the simultaneous appearance of two novel business groups made it critical that the cooperative rethink its mission. As noted in previous annual reports, this has been

an ongoing process as the cooperative has dealt with issues such as data exchange, germplasm ownership, and intellectual property rights. An important step in this process was recognizing that the cooperative's primary mission was to improve the common base population from which each member draws to create their own individual products. A second important step was the realization that not all members need to support the program in the same way for their contribution to be equitable. The outcome of this discussion was the realization that the Western Gulf Forest Tree Improvement Program could appeal to any organization with a vested interest in improved genetic material, regardless of the uses to which it will be put. Furthermore, our mutual responsibility to each other is the willingness to support the development of the common base population from which all members draw. Explicitly restating these concepts made it possible to recognize that there were organizations outside our traditional group of land-based state agencies and integrated forest products industries that could and should support tree improvement. As a result, the cooperative accepted CellFor, Inc. as a full member in 2004. CellFor, Inc. has no land base and will not establish orchards. They have agreed to support the forward selection population by providing clonal propagules to allow the cooperative's elite populations to be clonally tested. As the cooperative continues to adapt to the changing realities of the business environment, we hope to identify other organizations with which mutually beneficial arrangements can be developed.

Another noteworthy development in 2004 was the establishment of the Forest Tree Molecular Cytogenetics Laboratory at Texas A&M University by the USDA Forest Service Southern Research Station. This facility will be part of the Southern Institute of Forest Genetics at Gulfport, MS, but will be located in the Forest Science Laboratory with the WGFTIP staff. This is the only facility in the nation whose primary focus will be on the molecular cytogenetics and physical genome mapping of forest trees. Dr. Nurul Faridi (Figure 1), the lead scientist for this effort, has already successfully karyotyped loblolly pine, slash pine, and American chestnut. Future projects will include the ordering of loblolly pine chromosome fragments preserved in the most comprehensive bacteria artificial chromosome (BAC) library to be created for this species. The outcome of this effort will be a better understanding of the genetic architecture of pines and it will be an essential part of any future loblolly pine genome project. Ultimately these studies will help answer fundamental questions about the evolutionary success of pines and will inform the design of the breeding population.

*Figure 1. Dr. Nurul Faridi, an employee of the Southern Institute of Forest Genetics, is the lead scientist in the USDA Forest Service Southern Research Station's Forest Tree Molecular Cytogenetics Laboratory located at the Forest Science Laboratory in College Station.*



# WESTERN GULF FOREST TREE IMPROVEMENT PROGRAM

## Highlights

- Two members grafted a total of 26 acres of advancing-front loblolly pine seed orchard. Families in these orchards have a predicted breeding value gain of 35 percent in mean annual increment at age 20 with a 10 percent improvement in height growth.
- The 2003 seed crop was outstanding. The members collected a total of 47,975 pounds of loblolly pine seed and 6,180 pounds of slash pine seed. Annual harvests over the last ten years have averaged 33,296 pounds for loblolly and 4,912 pounds for slash pine.
- First-generation progeny test establishment was completed for all but one loblolly pine diallel in 2003/04, leaving the cooperative with only eight parents left to evaluate out of a total of 3,123 that made up the first cycle.
- Polymix test series were planted in both the East Texas and Arkansas breeding regions in 2003/04 and a second round of progeny tests with a different set of parents were sown in each of these zones for planting in the fall of 2004.
- Site hazard maps for fusiform rust infection of loblolly pine were generated using the average infection levels for unimproved checklots. These maps showed hotspots in North Louisiana and South Mississippi.
- One standard deviation improvement in rust resistance was equal to a predicted breeding value decrease in rust infection levels of approximately 13.5 percent for both loblolly and slash pine on sites where the unimproved sources would incur 50 percent infection.
- The Forest Tree Molecular Cytogenetics Laboratory was established at Texas A&M University by the USDA Forest Service Southern Research Station.

## Seed Orchards

The annual report has traditionally been divided into three broad sections discussing the deployment population, the breeding population, and supporting activities of the WGFTIP members and staff. During the entire history of the cooperative, the segment on the deployment population has centered on the use of seed. Initially this discussion included the selection of appropriate geographic seed sources and the management of local seed production areas. Early annual reports considered seedling seed orchards. However, as the program matured, this discussion rapidly changed to emphasize the use of grafted clonal orchards. Now new technolo-

gies are providing operationally viable alternatives to seed. In recent years, several members have planted large trials of vegetatively propagated loblolly and slash pine. Indications are that clones will soon be available in operationally meaningful quantities from a number of different providers and southern pine clonal forestry will be a reality. While this technology bypasses mass production of seed, clones are copies of individuals that originated from the traditional breeding program and are just as much a product of the WGFTIP as orchard seed. Clones will, therefore, have to be included in any future discussion of the deployment population in order for the annual report to reflect the total extent and value of the reforestation effort conducted with WGFTIP material. For the short-term, however, discussion of the deployment population will continue to be exclusively devoted to seed orchards (Figure 2).



Figure 2. French Wynne in Potlatch Corporation's Prescott Seed Orchard at age five.

## Orchard Establishment and Roguing

During the 2004 grafting season, the Mississippi Forestry Commission and the Oklahoma Department of Agriculture, Food and Forestry grafted a total of 26 acres of advancing-front loblolly pine seed orchard. These two state agencies selected clones for their new orchards that will maximize value for the non-industrial landowners growing timber for the open market. Therefore, they emphasized adaptability, growth rate, and stem straightness. Efforts were made to maintain specific gravity at regional averages although this was clearly a secondary priority. The two orchards had an average predicted gain in breeding value for volume of 35 percent for mean annual increment at age 20 and an improvement in height growth of 10 percent. Because of the large number of individuals from which selections could be made, these gains were accomplished with the negligible loss of 10 kg/cubic meter in predicted wood density compared to the regional averages. Genetic gain captured in this year's orchard establishment effort is down

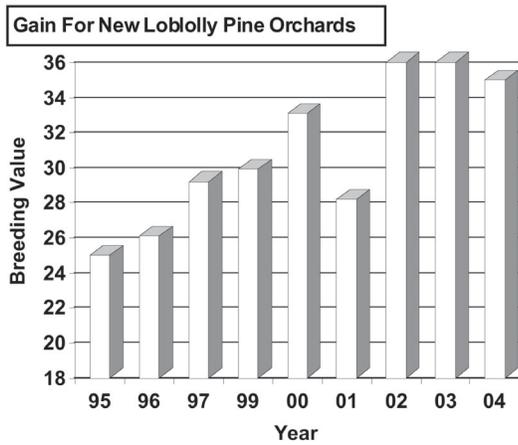


Figure 3. Gain in breeding value for volume growth averaged over new loblolly pine seed orchards by year of establishment.

slightly from the gain of 36 percent achieved in each of the last two years because the orchards represented in previous years were designed for different deployment zones (Figure 3).

The two loblolly pine orchards grafted in 2004, on opposite ends of the east-west transect of the cooperative, draw in part from the same procurement zone. The Mississippi Forestry Commission orchard (Figure 4), designed to support reforestation programs in North Mississippi, draws from a procurement zone that includes Mississippi and South Arkansas. The Oklahoma orchard also draws from trees selected and tested along the same latitude, specifically trees that originated from the Arkansas breeding population. This coincidence serves to illustrate two important aspects of collaborative tree improvement programs. The first is that genetic gain, in part, is a matter of selection differential. Therefore, breeding populations from overlapping procurement zones may be mutually supportive, sometimes in unanticipated ways. The second point is that as tree improvement matures, adequate progeny testing programs will be ever more important in making deployment decisions. Seed movement guidelines that were good for wild populations will be superseded by recommendations based on progeny test performance making access to field data crucial.



Figure 4. Robert Stewart of the Mississippi Forestry Commission examining grafts in their newest orchard expansion for the North Mississippi deployment zone.

The Oklahoma Department of Agriculture, Food and Forestry also grafted a shortleaf pine seed orchard with a predicted gain in phenotypic superiority of 25 percent for volume growth. The new orchard, designed with progeny tested clones, replaces two first-generation orchards originally established with Mountain and Lower Shortleaf provenances from within the state. The clonal composition of this new orchard reflects the fact that no performance differences were ever demonstrated for these two seed sources and once again emphasizes the value of progeny testing. Demand for this species has also declined as plantation management continues to rely primarily on loblolly pine even beyond the western and northern extent of its natural range. Reducing the number of shortleaf pine orchards from two to one and adjusting the orchard acreage to be more compatible with actual seed demand will make it possible for the Oklahoma Department of Agriculture, Food and Forestry to more efficiently use the limited area available at their Idabel, OK complex.

The members of the cooperative now manage a total of 2,341 acres of orchard (Figure 5). Of this total, 1,169 acres are heavily rogued first-generation orchards and 1,172 acres are advancing-front orchards established with the best available material regardless of breeding cycle. The cooperative has discontinued use of untested material for orchard establishment even though they may have very high predicted breeding values based on parental performance. Mid-parent values have proven accurate on average but are very unreliable for predicting performances of specific selections.

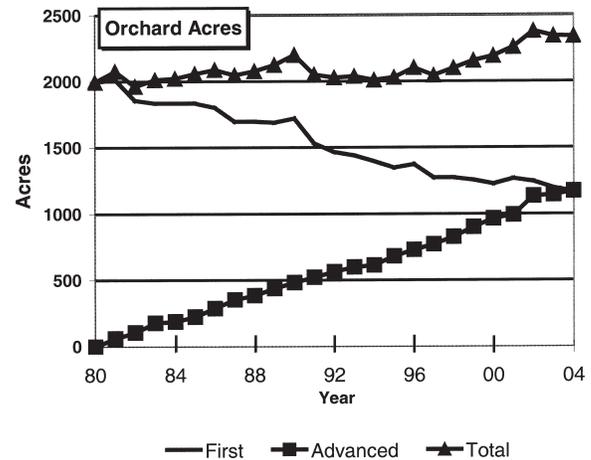


Figure 5. Seed orchard acres managed by the cooperative.

Total orchard acres somewhat belie the importance of the younger orchards within the cooperative. In most years, only seed from the very best clones that are predominately in the younger, newer orchard blocks are collected. Older orchards are maintained for insurance and for the few excellent parents they sometimes contain.

Three organizations prepared sites in 2004 for future orchard expansions. Boise planted rootstock in a previously unused area to further their loblolly pine orchard ex-

pansion program in 2005. Deltic Timber Corporation cleared eight acres of older orchard in 2004 to begin recycling more of their original first-generation orchard site into advancing-front orchard blocks. The Mississippi Forestry Commission also prepared ground in 2004 for rootstock establishment in 2005 and grafting in 2006. This expansion will include both a new slash pine orchard and a South Mississippi advancing-front loblolly pine seed orchard.

### Orchard Yields

The 2003 seed crop was outstanding. The members collected a total of 47,975 pounds of loblolly pine seed and 6,180 pounds of slash pine seed (Figure 6). Annual harvests over the last ten years have averaged 33,296 pounds for loblolly pine and 4,912 for slash pine. This exceeds the annual demand of 30,000 pounds of loblolly pine seed required by the cooperative and has allowed seed inventories to be maintained. The 2003 seed harvest was excellent across the whole WGFTIP region and accurately reflected individual member's orchard acres. Twenty percent of the loblolly pine seed crop was collected by a single member with a total collection of 4.5 tons from five different orchard complexes. The remainder of the crop was evenly divided among members with three organizations collecting more than 2.5 tons each and six others each collecting more than one ton. Yields averaged 1.19 pound of seed per bushel, which is moderate by historical standards. However, given the uncertain insect control that we have been able to achieve with the dwindling suite of cone and seed insect control techniques currently available, this figure was quite gratifying.

Figure 6. ETT, L.P.'s Slayden Creek Orchard managed by Molpus Timberland Management at age 6 showing the outstanding 2004 flower crop.



The first harvest from ETT, L.P.'s five-year old orchard (managed by Molpus Timberlands Management) was 3.6 pounds of seed per acre. This orchard is on a soil type the cooperative is favoring for new orchard establishment based on its early productive capacity. There were 65 bushels of cones collected from this same 24 acres at age six and if the 2004 flower crop is any indication, this orchard will break all records for early seed production in 2005 when it will be seven years old (Figure 7).

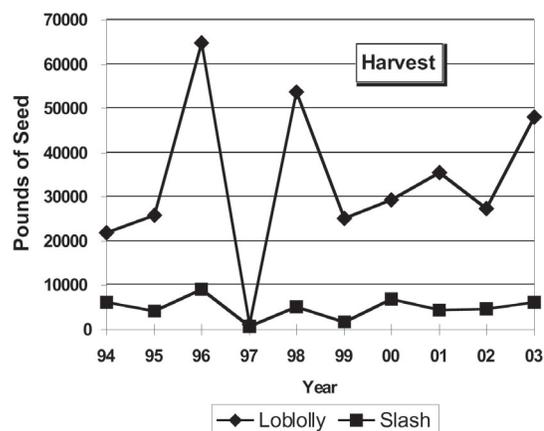


Figure 7. Pounds of seed harvested by the cooperative from 1994 to 2003

The cone crop in 2004 was well below average and unevenly distributed across the region. Several cooperators, primarily in southeast Texas, made the decision to drastically reduce cone collection efforts and to draw down seed inventories replenished the previous year. The combined harvest for all of the cooperative members totaled only 13,495 bushels of loblolly pine cones and 1,765 bushels of slash pine cones. If the 2004 flower crop is a reliable indicator, the 2005 cone crop should break all previous production records. In addition to heavy flower production in orchards, progeny test trees only four years from planting were also observed to be producing copious quantities of flowers.

### Breeding and Progeny Testing

The progeny testing program proceeded apace in 2004 as first-generation progeny test establishment was completed for all but one loblolly pine diallel and the last remaining first-generation breeding for the slash pine program was finished. In the second-generation loblolly pine breeding program, polymix test series were planted in both the East Texas and Arkansas breeding regions in 2003/04 and a second round of progeny tests for these regions using a different set of parents were sown in the greenhouse in 2004.

A common element in the breeding and progeny testing program is the increasing complexity as the cooperative makes the transition from the first-generation to advanced-generation breeding. This is a result of an increasing number of populations, complementary mating schemes in the advanced-generation breeding strategies, and the fact that a single individual may belong to multiple

populations and contribute to different breeding cycles. The difficulties that arise from managing a complex breeding program manifest themselves primarily as bookkeeping problems. A second difficulty can occur in failing to recognize that a cross made for one population may also support another. The outcome of managing this complexity effectively is a broadening of the selection population for all parts of the program. Mutually supportive breeding programs and faster turn-around times achieved with accelerated-breeding techniques such as topgrafting are contributing significantly to converting tree improvement from what was little better than a mass selection scheme into a modern plant breeding program (Figure 8). Innovations in marker-assisted breeding and clonal progeny testing can only accelerate this process.



Figure 8. Becky Maxwell of Plum Creek Timber Company tending topgrafts intended to support accelerated breeding efforts.

### Elite Breeding Programs

The cooperative manages two elite breeding programs directed primarily to support the deployment population. The first of these is the Super-Breeding Groups and the second is the Wood Quality Elite Population. The underlying strategy for both programs is identical although the selection criteria and organization of the crossing patterns are different. The basic concept is to use backward selection to identify outstanding individuals that were not previously crossed together in the mainline breeding program, make these crosses and plant block plots from which outstanding individuals can be identified and selected. These selections will then be topgrafted, crossed with polymix pollen, and evaluated in the same polymix progeny tests that support the mainline advanced-generation testing program. Individuals with proven performance will then be available for use in orchards or for additional breeding.

### Super-Breeding Groups

Individuals in super-breeding groups are selected on the same basis as those selected for the mainline breeding program, although with a higher selection differential. The

primary selection criterion is, therefore, growth potential with secondary emphasis placed on disease resistance, bole straightness, and wood quality. The crossing pattern consists of mating the top three selections from each of two paired breeding groups both managed by the same member. Because the only intention is to create a selection population from parents with known breeding values, the precise crossing scheme is not important. Some crosses cut across breeding groups and will be appropriate for use in orchards or for further breeding only within elite breeding populations. Other crosses, however, are between parents in the same breeding group and can be used in the mainline breeding program as well. By maintaining structure in the breeding population through the mating of parents from paired breeding groups, no matter the degree of inbreeding incurred within super-breeding groups, there will always be unrelated individuals from which a deployment population can be formed.

To date, five members of the cooperative have established a total of 93 crosses from nine different super-breeding groups. Twenty-four super-breeding group selections have been identified by Potlatch Corporation and will be incorporated into their testing program. Eleven of these selections are from crosses where both parents originated from the same breeding group. Should they prove of sufficient quality when evaluated in polymix progeny tests, they will be incorporated into the next cycle of breeding in the mainline population.

### Wood Quality Elite Breeding Population

The wood quality elite breeding population is selected with a pulp index score that combines performance data for both growth and specific gravity. The economic weight for specific gravity is approximately seven times larger than that for volume production. These values were chosen to maximize the profit from a hypothetical kraft paper mill where the quantity of dry matter per cubic volume, and therefore the quantity of cellulose that can be loaded into a digester, are important factors in reducing the final cost per ton of pulp. It is also anticipated that this population will yield a higher percentage of stress graded dimensional lumber because of the positive correlation between specific gravity and strength.

The wood quality elite breeding population will ultimately consist of the thirty individuals with the highest pulp indices selected from each of four breeding zones. Development of this program has been slower than anticipated for two reasons. First, even with our large base population, identifying individuals with the desired combination of specific gravity and volume traits has been difficult (Table 1). Secondly, breeding has been slower than anticipated because most of these individuals are only found in a limited number of scion banks where they may be older, taller trees with difficult to reach crowns. While individuals selected for the wood quality elite population must have acceptable growth rates, they are not, as a rule, in the upper echelon of fast growers and have not been widely used in orchards. When they are located in scion banks, they tend to have few flowers and are difficult to pollinate due to their size and

**Table 1. Average performance of the loblolly pine parents in the elite breeding populations for improvement in volume growth and wood specific gravity.**

Zone	Parent (number)	Volume <sup>1</sup> (%)	Specific Gravity <sup>2</sup>	Economic Index (Kraft Pulp) <sup>3</sup>
Arkansas	11	19.3	0.023	\$13.72
North Louisiana	5	25.4	0.018	\$13.57
Texas	18	18.9	0.031	\$16.34
South MS/South LA	4	22.4	0.030	\$17.40

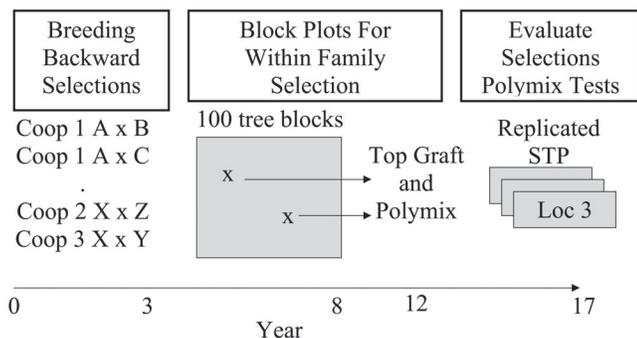
<sup>1</sup> Breeding value expressed as change in mean annual increment at age 20 compared to an unimproved checklot performance.

<sup>2</sup> Absolute change in specific gravity compared to an unimproved checklot.

<sup>3</sup> Reflects expected savings per ton of kraft pulp produced from a land base of fixed size for wood with improvements in both volume and specific gravity.

closer spacing. Scion is being shared and topgrafted into younger scion banks or into orchards to expedite the breeding program.

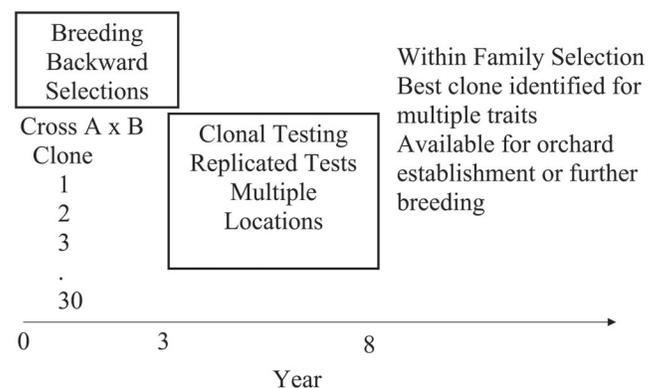
The original plan for generating a selection population in this breeding program was to establish block plots of control-pollinated seedlings. Multiple selections in each cross would be identified at age 5, topgrafted, crossed with a polymix pollen, and the resultant seedlings evaluated in replicated field trials (Figure 9). The entire cycle from identifying the parents through to the point of having progeny tested selections for use in orchards or for breeding stock would take approximately 17 years. This process is also expected to be relatively inefficient in identifying individuals with simultaneous improvements in low heritability traits such as volume growth and negatively correlated wood quality characteristics.



*Figure 9. Schematic showing the timeline for using block plots for within-family selection in the elite breeding program. The steps include breeding backward selections, establishing block plots with seedlings, identifying within-family selections, and polymix breeding and test establishment for breeding value estimation.*

To overcome these difficulties, the cooperative will use clonal testing to evaluate the portion of this population that is amenable to vegetative propagation. Rather than test 100 seedlings per cross in a block plot where within-family selection is based strictly on phenotype, the cooperative will evaluate approximately 30 clonal lines per full-sib cross in replicated plantings (Figure 10). This scheme has a number of advantages. Clonally replicated progeny tests should

improve overall selection efficiency and may also drastically shorten the breeding cycle. Crosses in this population will not be inbred and to the degree that genetic variation in loblolly pine is additive, clonal performance should reflect breeding value. With this assumption, the period from identifying parents to having evaluated material for an additional round of breeding is shortened to 8 years. Multiple generations of breeding and selection coupled with improved selection efficiency should dramatically increase gain per year. Clones identified in these tests will be available to all members for use in seed orchards or for further breeding.



*Figure 10. Schematic showing time line for using clonal replicates for evaluating within-family selections in the elite breeding program. Steps include breeding backward selections, producing clonal replicates, establishing replicated plantings of clonal lines in field tests, and identifying within-family selections for additional breeding or seed production.*

Clonal testing for improved forward selection is not tied to a single propagation method. Because the selection effort is strictly limited to identifying the best clonal lines within each full-sib family, it may also be possible to include multiple propagation methods within a single test series by treating families as main plots and lines as subplots. It should be emphasized that only the proportion of the wood quality elite breeding population that is easily propagated vegetatively will be clonally tested. Seedlings will continue to be used to create the selection population for crosses that produce few seed and those that are recalcitrant to clonal propagation. Periodically, selections will have to be evaluated in polymix seedling progeny tests to ensure that clonal performance is in actuality correlated with breeding value. Polymix progeny tests will certainly be required to verify the breeding value for any selection used in seed orchards. By topgrafting, polymix crossing can be conducted simultaneously with orchard establishment and data can be available before seed production commences.

Highly selected small breeding programs provide the ideal population in which to evaluate new breeding and progeny testing techniques. Their small size facilitates the rapid turnover of generations without impacting the base population on which the cooperative's long-term genetic gain depends.

**Table 2. Progeny tests established during the 2003/04 planting season.**

<b>Cooperator</b>	<b>Number of Tests</b>	<b>Diallel by Location Combinations</b>
<b><u>First-Generation Loblolly Pine Tests</u></b>		
Temple-Inland Forest	3	15
<b>First-Generation Loblolly Pine Total:</b>	<b>3</b>	<b>15</b>
<b><u>Advanced-Generation Loblolly Pine Polymix Tests</u></b>		
	Number of Tests	Number of Families
Arkansas Forestry Commission	1	68
Deltic Timber Corporation	1	69
International Paper Company	3	148
Texas Forest Service	1	81
<b><u>Advanced-Generation Slash Pine Polymix Tests</u></b>		
Boise	1	45
Louisiana Department of Ag and Forestry	1	45
Texas Forest Service	1	45

**First-Generation Breeding and Progeny Test Establishment**

This will likely be the last annual report that discusses first-generation breeding and progeny test establishment as a separate topic. During the 2003/04 planting season, Temple-Inland Forest established five loblolly pine diallels of first-generation crosses representing 39 parents (Table 2). This test series represented the last first-generation progeny tests required to complete evaluation for this species in East Texas. First-generation breeding has been completed and the last diallel for loblolly pine and three diallels for slash pine will be established in the field in 2005/06.

**Slash Pine**

There were no first-generation progeny tests established in 2003/04. However, breeding was completed for the last three remaining diallels. Seed is in hand and the final first-generation test series will be sown in 2005.

The slash pine program started with approximately 1,000 first-generation selections. This number was culled to around 500 individuals based on screening for fusiform rust resistance at the USDA Forest Service’s Resistance Screening Center (RSC) at Asheville, NC prior to diallel breeding. This step delayed pedigree breeding by three or more years as the seed submitted to the RSC was created with a polymix of susceptible males. This was done to ensure that resistance inherited from the paternal side of the cross would not mask susceptibility in the female parent. When the final progeny tests are planted in 2005, we will have established approximately 400 parents that passed the screening at the

RSC in control-pollinated progeny tests for growth and form evaluation in the field. There are an additional 64 parents in this species for which we have open-pollinated progeny test data that predates the Asheville screening.

Gain in rust resistance as gauged by the number of rust-free trees at age five has increased dramatically because of the screening at the RSC. The WGFTIP evaluates rust resistance as the number of standard deviations less rust expressed by the select tree family compared to the unimproved checklot. The actual response is curvilinear and varies according to the inherent risk level at each site. On sites at the extremes of the range, those with zero and 100 percent rust infection, one would expect that the improved sources would vary little from the performance of the susceptible checklot. Maximum differences should be expressed in stands where the unimproved checklot suffers 50 percent infection. This was shown to be true when rust resistance data was analyzed in 1989<sup>2</sup>. Very similar curves were created when all 107 currently available slash pine progeny tests were evaluated. Curves were fitted to predict the magnitude of one standard deviation in rust resistance for improved material based on average plantation infection levels for all plantings with more than 20 percent infection. The phenotypic performance was then reduced to account for a heritability of 0.75 for resistance (Figure 11). A one standard deviation improvement in rust resistance equates to approximately a 13.5 percentage point reduction in rust infection in a stand where the unimproved checklot incurs 50 percent infection. In other words a family with a one standard deviation improvement in rust

<sup>2</sup>Byram, T. D., T. A. Greene, W. J. Lowe, C. R. McKinley, J. F. Robinson, and J. P. van Buijtenen. 1989. 37th Progress Report of the Cooperative Forest Tree Improvement Program. Tex. For. Ser. Cir. 283, 27 p.

resistance would have an expected infection level of 36.5 percent. This relationship was relatively strong with  $R^2=0.95$ . The best 20 first-generation parents available for orchard establishment average a reduction in rust resistance exceeding 2.7 standard deviations.

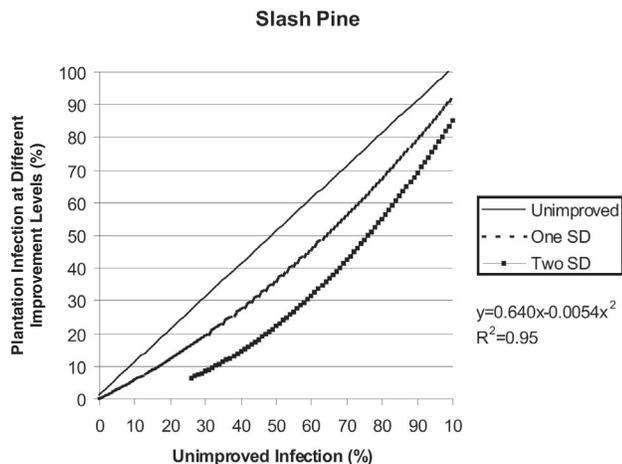


Figure 11. Predicted rust infection levels for slash pine families with one and two standard deviations of improvement in rust resistance over a range of sites with different infection levels for the unimproved checklot. Phenotypic values need to be adjusted by the heritability to obtain breeding values.

This method of reporting rust resistance in progeny tests treats disease infection as a quantitative trait and ignores the much more complicated reality of gene-for-gene interactions expressed by real world organisms. This method will give an accurate performance prediction when the select trees are exposed to the same pathogen population to which they were exposed when they were progeny tested. However, even the most rust resistant family will fail if exposed to a new and virulent pathogen population. Deploying a mosaic of families with different resistance genes can mitigate this type of risk.

### Loblolly Pine

During the 2003/04 planting season Temple-Inland Forest established five diallels of first-generation crosses representing 39 parents in East Texas. This series of progeny tests represent the last first-generation progeny tests required by Temple-Inland Forest to complete evaluation for this species.

The cooperative has now established 2,606 first-generation loblolly pine parents in balanced control-pollinated progeny tests and an additional 517 parents in open-pollinated or polymix progeny tests (Figure 12). The database will eventually include performance data on 3,123 different individuals for this species. The plantings established during the 2003/04 planting season will evaluate the performance of an additional 41 parents, all of which are new to the program (Figure 13).

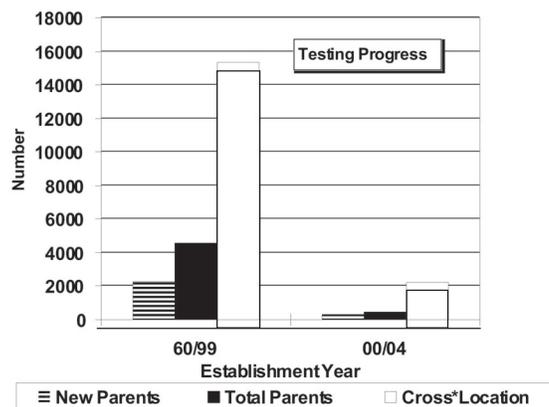


Figure 12. The number of loblolly pine crosses (total number of cross by location combinations), the total number of parents, and the number of parents established in tests for the first time from 1960/99 and from 2000/04.

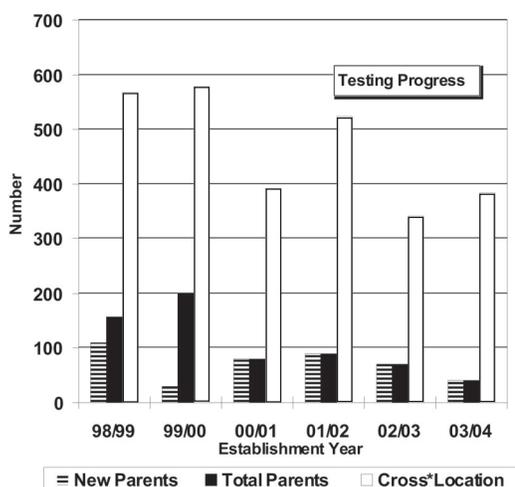


Figure 13. The number of loblolly pine crosses (total number of cross by location combinations), the total number of parents, and the number of parents established in tests for the first time in each of the last six years. The 1998/99 establishment year represents progeny tests that were evaluated in 2003/04.

### Virginia Pine

The Texas Forest Service and the Oklahoma Department of Agriculture, Food and Forestry established a third series of polymix tests to evaluate Virginia pine parents for Christmas tree production in 2003/04. Fifty-three parents have now been established in 13 progeny tests with the collaboration of Christmas tree growers in the states of Oklahoma, Texas, Louisiana, and Mississippi. Trees in these tests are being evaluated for survival and height growth at one, two and three years of age. Christmas tree quality will be evaluated at age four. In 2006 the Texas Forest Service will be the first to use this data by grafting the initial blocks of an orchard with improved qualities for Christmas tree production. This clonal orchard will replace an existing seedling seed orchard and should provide seed with a significant improvement in both genetic quality and uniformity over that previously available.

## Test Measurement and Second-Generation Selection Activity

During the 2003/04 measurement season, the cooperative evaluated 97 progeny tests. Of these, 66 were loblolly, 17 were slash pine, ten were Virginia pine, and three were shortleaf pine. First-year survival was assessed in ten tests. Forty-seven of the measured tests were age ten while 18 tests were age five. The five-year-old tests provided an evaluation of 444 families, 207 for the first time (Figure 13). The cooperative continues to evaluate all plantings at ages 5 and ten. Ten-year measurements have proven valuable in validating judgments made at age five and allowing additional second-generation selections to be identified. A subset of tests is measured through age 20 (Figure 14). This has been invaluable in continuing the effort to improve age-age correlations and to develop breeding values for mature traits such as improvement in site index.



Figure 14. An eleven-year-old loblolly pine test being thinned by Boise in order to maintain it through age 20.

The Arkansas Forestry Commission requested that two loblolly tests be added to the 2003/04 schedule. These tests were measured and screened for selections at age nine. This early evaluation was done to allow the entire test site, which contained multiple plantings, to be row-thinned in an effort to reduce future roosting by egrets. Droppings from the sheer number of birds in the roosts caused considerable mortality in several plantings resulting in the need to clearcut entire replications.

Cooperators made a total of 80 second-generation selections in 2004 with slash pine predominating the effort with 44 new selections. Temple-Inland Forest added 14 slash pine second-generations selections this season, followed by the Louisiana Department of Agriculture and Forestry, who added eight and the Mississippi Forestry Commission who

added seven. The greatest number of new loblolly pine selections was made by Plum Creek Timber Company with 15, with the majority of those coming from south Arkansas. Boise added 14 loblolly pine selections to the cooperative's advanced-generation population (Figure 15). The cooperative has now identified a total of 1,587 loblolly pine and 224 slash pine second-generation selections (Figure 16).



Figure 15. An eleven-year-old loblolly pine second-generation selection preserved in a row designated for thinning in a Boise progeny test.

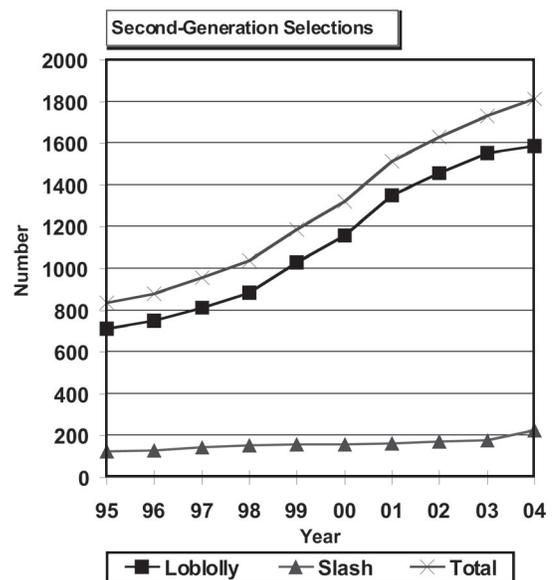


Figure 16. Cumulative number of WGFTIP second-generation selections.

## Slash Pine Demonstration Plantings

Ten-year data were collected from seven slash pine demonstration plantings designed to exhibit differences in fusiform rust resistance among unimproved commercial checklots from South Mississippi and Georgia, and a group of improved WGFTIP families. The Georgia source was similar to the unimproved material widely planted in the Western Gulf Region before orchard seed from local selections became available. The South Mississippi checklot was a standard comparison in all WGFTIP slash pine progeny tests. The improved source consisted of ten families selected by the WGFTIP for high rust resistance in previous field trials.

Plantings were established in 1993 and 1994 and maintained by Boise, Temple-Inland Forest, and the Louisiana Department of Agriculture and Forestry. Four locations were planted each year; seven of the eight sites are still active. Each planting consists of two replications with large block plots (80-100 trees) from each source. Boise has two locations and the Louisiana Department of Agriculture and Forestry has a single location in Beauregard Parish, LA. Temple-Inland Forest has one location in Hardin County, TX, and two in Vernon Parish, LA, all of which also include an additional improved loblolly pine seed source. The Louisiana Department of Agriculture and Forestry also has a test location in East Feliciana Parish, LA, which also includes an improved loblolly pine seed source. Five-years results from these demonstration plantings were reported in the 46th Progress Report of the Cooperative Forest Tree Improvement Program.

Although individual plantings were established strictly for demonstration, it was possible to do a statistical analysis by combining information across sites. Survival, rust infection, and planted tree volume were evaluated after 10 years in the field. The only trait for which differences among sources were statistically significant at age ten was volume. Improved slash clearly produced more volume than either of the unimproved slash sources (Figure 17). Differences among sources for rust infection levels, while not statistically different were very nearly so (Prob. > F = 0.12) and were in the expected direction (Figure 18). A change in relative response to higher rust pressures in the second year of the study resulted in a significant rust infection by year interaction. In the 1993 plantings, when average rust infection was 24.9 percent, rust infection levels in the improved sources were approximately 20 percentage points lower than the unimproved sources (19.7% compared to 31.1%). In the 1994 plantings when average rust infection levels increased to 36.4 percent, the improved sources were more than 25 percentage points lower (26.0% compared to 52.3%). In other words, when the rust infection pressure was higher, so was the benefit of planting the improved stock.

The ten families included in the improved source averaged approximately two standard deviations below the checklot for rust infection in previous field trials. Based on this performance, the expected level of rust infection for the improved sources in these plantings were predicted to be 9 and 24 percent in the stands where the unimproved sources had 31 and 52.3 percent infection, respectively (see Figure 11). Actual infection levels for the improved sources were

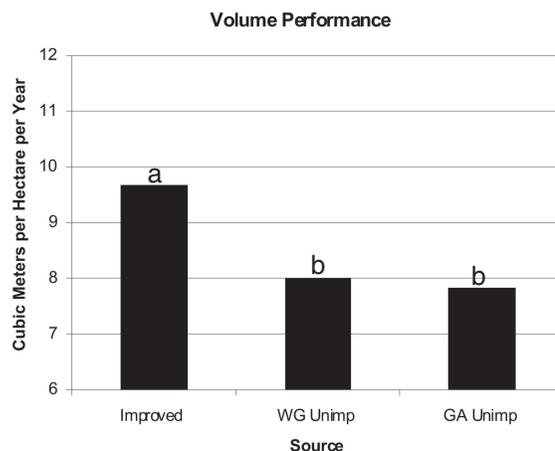


Figure 17. Average volume production for the improved and unimproved sources in the slash pine demonstration plantings. Bars with different letters represent values that are statistically different at the 10 percent level of confidence.

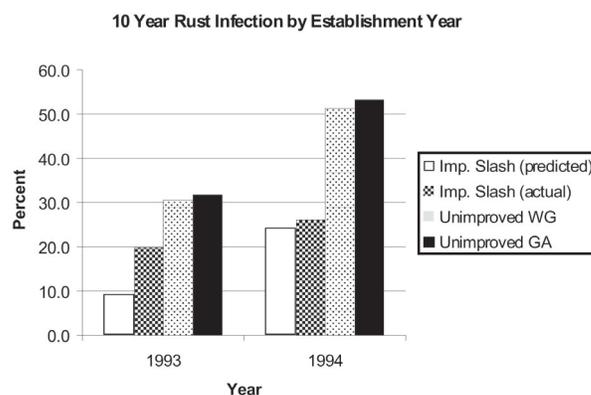


Figure 18. Rust infection levels for the improved and unimproved sources in the slash pine demonstration plantings by year of establishment. The left most bars represent the predicted rust infection level of the improved sources given performance in previous progeny tests.

19.7 and 26.0 percent indicating that our predictions may be more reliable at moderate to high infection levels.

In the combined analysis of all plantings that included a loblolly pine seed source, the loblolly pine had less rust than the improved slash pine, but no more volume. At individual sites, loblolly pine produced more volume than slash pine in the Hardin County, TX test, while the improved slash pine outperformed loblolly at the East Feliciana Parish, LA location.

These results demonstrate that substantial gains have been made for volume and rust resistance in the slash pine program. Slash pine is an exotic in much of the Western Gulf region, but does have a place in reforestation, particularly where it is used in its native range such as in the Florida Parishes of Louisiana. It is also the preferred species for planting on flatwoods sites if bedding and phosphorous fertilization are not used. Slash pine may also have a higher value in management systems that favor pole production.

## Fusiform Rust Resistance in Loblolly Pine

Loblolly pine resistance to fusiform rust infection has not been a major concern for the WGFTIP breeding program. Rust infection levels within the region have been historically low both because the western seed sources are inherently more resistant and the weather conditions over much of the region are not conducive to the spread of fusiform rust spores. Despite the fact that fusiform rust seldom caused economic losses in the past, this situation could change for any number of reasons. Plantation management is likely to be more conducive to disease outbreaks than natural stand management. Furthermore, lack of exposure to infection in progeny tests makes it difficult to eliminate susceptible parents from the program making it more likely that these parents will be included in the production population. There was some evidence that this is happening in the Lower Gulf Elite Breeding Population as the eastern Atlantic coastal sources, from a generally recognized susceptible provenance, had less rust than did the western sources.

The cooperative has taken two initial steps to be proactive about this potential problem. The first step was the development a rust hazard map based on fusiform rust infection levels in the unimproved loblolly pine checklot. The second step was to use all of the loblolly pine progeny tests with fusiform rust to prepare curves of expected infection levels for one and two standard deviations of improvement in rust resistance. These curves were constructed using the same techniques as the slash pine resistance curves first prepared in 1983.

The rust hazard map for fusiform rust infection depicts only counties/parishes that contain WGFTIP progeny tests in which rust was evaluated. In general, this map (Figure

19) conforms to similar maps published by the USDA Forest Service. Rust infection levels are generally low in Arkansas, Oklahoma, and East Texas. Rust infection levels are much higher in Mississippi and hot spots exist in both southeast Mississippi and North Louisiana. Moderate levels of rust infection levels in the unimproved checklot of between 15 and 40 percent prevail in Mississippi (where infection levels are historically higher) and in scattered counties and parishes throughout the region. Superficially, the counties and parishes with moderate infection levels have a number of things in common. They have hardwood bottoms in which the alternate host red oak species flourish and plantation management is widely practiced.

Genetic differences in rust resistance can only be evaluated when the disease incidence is sufficiently severe to insure that resistant trees are truly separated from escapes. The WGFTIP considers that this condition has been met in plantings where either the average infection level in the plantation or the infection level in the unimproved checklots exceed 30 percent and there are statistically detectable differences among families. For the purposes of developing these curves, however, all 631 loblolly pine progeny tests for which rust infection had been scored were used. The curves reflect the expected value of one and two standard deviations improvement in rust resistance given the infection level of the unimproved sources (Figure 20). These curves were very similar to the curves generated for slash pine (see Figure 11). Statistical fit was excellent ( $R^2=0.94$ ) but it is expected that the predictions in the middle portion of the range will be most accurate as mean separations are expected to be most precise at infection levels of approximately 50 percent. When the unimproved sources have 50 percent infection, families with one standard deviation improvement in rust resistance should exhibit 36.5 percent infection (a reduction of 13.5 percentage points).

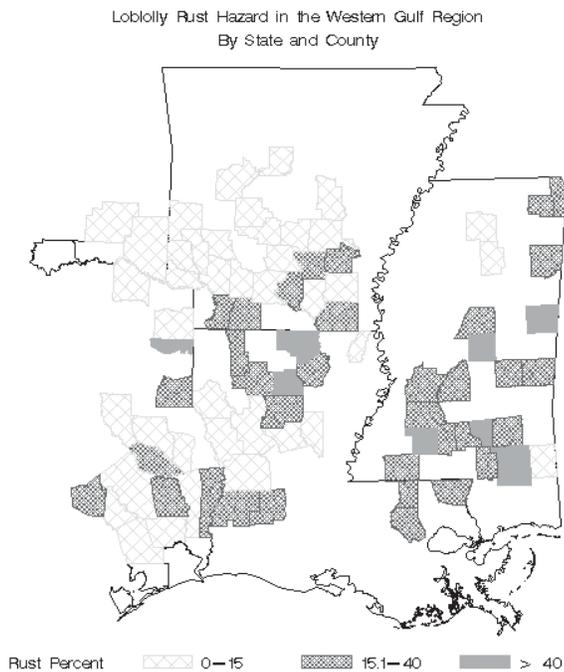


Figure 19. Rust hazard map for loblolly pine based on the performance of the unimproved checklot. Only counties/parishes with progeny tests evaluated for rust are shown.

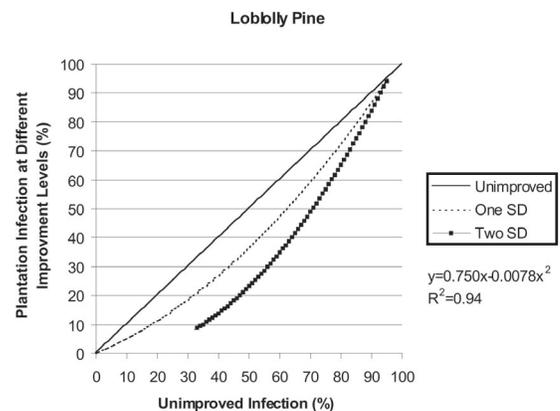


Figure 20. Predicted rust infection levels for loblolly pine families with one and two standard deviations of improvement in rust resistance over a range of sites with different infection levels for the unimproved checklot. Phenotypic values need to be adjusted by the heritability to obtain breeding values.

As mentioned above in the discussion of slash pine, this method of reporting rust resistance is a gross simplification of the complex gene-for-gene action that is certainly a major component of this disease complex. It is,

however, predictive when material is deployed in areas where it is exposed to the same conditions under which it was tested. Even the most resistant families will prove susceptible when they are challenged by a novel and virulent pathogen. This risk can be mitigated by 1) testing families against the pathogen population that they are expected to encounter and 2) by deploying a mosaic of families with different resistance genes.

### Shortleaf Pine

Shortleaf pine is a minor species in the WGFTIP as only two members maintain seed orchards and only one member has an active progeny testing program. Most of the older progeny tests were established with open-pollinated seed and have now been measured periodically for 20 years. Dr. David Gwaze, now a Resource Scientist with the Missouri Department of Conservation, recently examined genetic components in three of these progeny tests to 1) validate that there is a sufficient genetic variation to conduct a tree improvement program for this species and 2) ascertain the value of early selection programs in this species. This last aspect is particularly of interest as this species tends to be grown on a much longer rotation interval than either slash or loblolly pine.

The test layout was the same for each of the three plantings and consisted of a randomized complete block design replicated twelve times with six-tree row plots for each family. Initial spacing was eight feet by eight feet and all tests were evaluated at 5, 10, 15 and 20 years of age for height and diameter. Volume of living trees was calculated using the formula for a cone. All three plantings were located in Cleburne County, Arkansas and included 24, 25 and 27 half-sib families. Summary statistics for the three plantings at various ages are included in Table 3. Data was not pooled across the three tests because they had very few families in common.

**Table 3. Overall means (± standard deviation) and number of trees for height, diameter and volume in three shortleaf pine tests at four ages.**

Age	Test	HT (m)	DBH (cm)	VOL(dm <sup>3</sup> )	Measured trees
5	AFC448	2.21 ± 0.40	2.11 ± 0.80	0.324 ± 0.255	1571
	AFC449	1.92 ± 0.44	1.55 ± 0.72	0.174 ± 0.183	1557
	AFC450	2.29 ± 0.37	2.32 ± 0.85	0.400 ± 0.310	1489
10	AFC448	6.91 ± 0.97	10.31 ± 1.66	20.23 ± 7.36	1559
	AFC449	6.15 ± 0.93	9.76 ± 1.67	16.39 ± 6.54	1534
	AFC450	6.55 ± 0.80	10.62 ± 1.66	20.31 ± 7.07	1467
15	AFC448	10.23 ± 1.24	14.89 ± 2.38	62.24 ± 21.34	1476
	AFC449	9.43 ± 1.23	14.84 ± 2.40	57.22 ± 20.68	1471
	AFC450	9.93 ± 1.15	15.66 ± 2.49	66.85 ± 23.48	1436
20	AFC448	13.67 ± 1.35	17.86 ± 2.84	119.49 ± 40.63	1283
	AFC449	12.94 ± 1.22	17.78 ± 2.89	112.49 ± 40.04	1379
	AFC450	10.15 ± 2.05	17.87 ± 2.98	89.50 ± 38.18	1376

Genetic parameters for shortleaf pine did not appear to differ substantially from those reported for other southern pine species (Table 4). Heritabilities for volume production

ranged from 0.11 to 0.44 and tended to increase with age. Heritabilities for height and dbh were very similar. Genetic correlations for height between ages 5 and 20 were moderate, ranging from 0.66 to 0.71. This indicated that early selections are likely to be efficient.

**Table 4. Heritabilities (standard error) for height, diameter, and live tree volume for three shortleaf pine tests assessed at four ages.**

Age	Test	HT	Heritability	
			DBH	Volume
5	AFC448	0.171(0.071)	0.162(0.069)	0.114(0.055)
	AFC449	0.136(0.060)	0.157(0.069)	0.134(0.063)
	AFC450	0.176(0.075)	0.254(0.097)	0.220(0.088)
10	AFC448	0.244(0.091)	0.104(0.052)	0.152(0.066)
	AFC449	0.145(0.063)	0.136(0.060)	0.176(0.071)
	AFC450	0.219(0.087)	0.328(0.116)	0.357(0.124)
15	AFC448	0.152(0.067)	0.103(0.052)	0.155(0.067)
	AFC449	0.122(0.057)	0.146(0.064)	0.156(0.067)
	AFC450	0.306(0.111)	0.381(0.131)	0.435(0.144)
20	AFC448	0.314(0.114)	0.145(0.068)	0.215(0.087)
	AFC449	0.125(0.060)	0.140(0.064)	0.171(0.072)
	AFC450	0.319(0.116)	0.408(0.138)	0.438(0.120)

The Oklahoma Department of Agriculture, Food and Forestry and the Arkansas Forestry Commission manage small shortleaf pine programs. Both of these organizations support regeneration programs outside the natural range of loblolly pine. Shortleaf pine is favored for ecosystem restoration by both the USDA Forest Service and some non-industrial private landowners in these areas.

### Second-Generation Breeding and Testing

The real strength of the cooperative was once again illustrated by the level of collaboration demonstrated in the advanced-generation breeding and progeny testing program. Polymix progeny testing for parental breeding value estimation is organized on a regional basis. All available polymix seed lots from within a breeding region are grouped together and the responsibility for growing and establishing the progeny tests are divided among members. By working together, polymix seed lots are established in progeny tests as soon as sufficient numbers become available for a test series. This has proven much more efficient than waiting for an individual member to have a sufficient amount of seed to plant stand-alone progeny test series.

This collaborative system made it possible to establish loblolly pine advanced-generation progeny test series in both the East Texas and the Arkansas breeding regions in each of the last two years. In 2003/04, there were 81 families planted in an East Texas polymix progeny test series and 69 families in an Arkansas/Oklahoma polymix progeny test series. This was followed by sowing another two test series in the summer of 2004 with 77 East Texas advanced-generation parents and 56 Arkansas/Oklahoma advanced-generation parents, respectively. Such rapid

conversion of the breeding effort into field tests is essential to continue improvement in the production population.

During the 2003/04 planting year, Boise, the Louisiana Department of Agriculture and Forestry, and the Texas Forest Service collaborated to establish three locations of a slash pine polymix test series containing 45 families. Deltic Timber Corporation, the Arkansas Forestry Commission and International Paper Company each established one location of an Arkansas/Oklahoma loblolly polymix test containing 69 families. International Paper Company established two locations and the Texas Forest Service planted the third to complete an East Texas loblolly polymix test containing 81 families (Table 2 and Figure 21). Finally, the Texas Forest Service established two locations of a Virginia pine polymix test containing 17 families. The Oklahoma Department of Agriculture, Food and Forestry established the third location.



*Figure 21. Gerald Lively and Adam Crain on the site of the 2003/04 Texas Forest Service planting of the East Texas advanced-generation polymix progeny test at the Arthur Temple Research Area. This site was reused after a previous progeny test was harvested.*

Advanced-generation breeding includes both the very best first-generation parents and second-generation selections. Of the approximately 1,900 loblolly pine parents identified for polymix testing, 845 are established in two or more locations. Of this number, 248 have reached sufficient age to have performance data. In the slash pine program there are 224 true second-generation selections of which approximately 80 are established in field tests. The first test series was measured when it reached age five, but the data was not considered meaningful because there was no rust pressure on this initial test series. Volume performance for these parents will be incorporated into the database when these plantings reach age 10.

The second part of the advanced-generation breeding program is the establishment of pedigree crosses to form the population for the next cycle of selection. These crosses are being planted in unrepliated block plots containing either 100 trees at one location or 49 trees at two locations. Seven cooperators have established selection plots for 367 control-pollinated families of both loblolly and slash pine families.

## Additional Activities

### Contact Representatives' Meeting

The annual Contact Representatives' Meeting was held May 11-12, 2004 in Lomax Hall on the campus of Louisiana Tech University in Ruston, LA. Dr. John Adams, Director of Forestry at Louisiana Tech University, made it possible for the WGFTIP to have access to these excellent facilities and helped host the meeting. The theme for the first day of presentations was the potential impact of clonal forestry. Members heard about the operations and future plans of one clonal forestry provider in a presentation given by John Pait, Senior Vice-President of Business Development for Cellfor, Inc. Dr. Tom Byram followed up with a talk detailing how clones could be used to improve advanced-generation selection procedures. Drs. Terry Clason and Michael Blazier set the stage for the afternoon tour of the Hill Farm Research Station near Homer, LA. They highlighted past, present, and future forest management research efforts on the 350 acres of pine plantation contained within the borders of the Station. Jackie and Glenda Robbins graciously sponsored the evening social at their Irrigation Mart warehouse.

Presentations on the second day centered on cone and seed insect control with reports given by Drs. Alex Mangini, John Taylor (USDA Forest Service) and Don Grosman (Texas Forest Service). Members were also given an overview of new herbicide combinations in a report by Dr. Jimmy Yeiser (Stephen F. Austin State University), as well as a report on chip bud grafting in hardwoods presented by Robert Stewart of the Mississippi Forestry Commission. Attendees received 7.0 CEU's from the SAF.

### Other Activities

There were several noteworthy meetings and program reviews held in 2004 in which members of the cooperative played pivotal roles both as individuals and as representatives of the larger tree breeding community. The first of these was a satellite meeting held in conjunction with the Information Exchange Group conference in June at Jekyll Island, GA. This meeting was organized by Dr. Dave Neale, a USDA Forest Service scientist, with the intent of developing a community wide position on the benefits and possible organization of a loblolly pine genome project. Several WGFTIP members attended and provided input from the perspective of organizations involved in applied breeding programs. As a part of this effort, an industry subgroup was formed and a position paper outlining potential industry participation and expectations was prepared. The final report for this and other subgroups can be viewed at <http://dendrome.ucdavis.edu/lpgp>.

Membership interests were also represented by the cooperative staff and member participation in Technical Advisory Visits (TAVs) for the Southern Institute of Forest Genetics, the Project on Insect and Diseases in Southern Pine Forests and the Southern Hardwood Forestry Research Group. Held every five years, the format of the TAVs allows the

USDA Forest Service research scientists to provide updates of current research and to receive feedback on future research plans. Participation in the TAVs provided an opportunity to stress to the USFS management that these units do research that is of critical importance to our members and that we are interested in finding ways to increase collaboration on problems of mutual concern.

### Formal Reviews

Five formal reviews, including one review delayed from 2003, were scheduled and completed in 2004. The rapidly changing organizational environments in which our members operate almost always ensure that the formal reviews are timely for a variety of reasons. These comprehensive program assessments are conducted every three years and are intended to provide the participants an overview of the accomplishments of their respective programs. They also provide an opportunity to reassess the strategic goals of the individual members and to insure that the WGFTIP is positioned to support these goals. Formal Reviews were conducted in 2004 for the Arkansas Forestry Commission, International Paper Company, the Oklahoma Department of Agriculture, Food and Forestry, Potlatch Corporation and the Texas Forest Service.

### USDA Forest Service - Forest Tree Molecular Cytogenetics Laboratory<sup>3</sup>

The Southern Institute of Forest Genetics (SIFG, USDA Forest Service Southern Research Station, Saucier, MS) has re-initiated its work in molecular cytogenetics. Dr. Nurul Faridi has been hired to lead this effort and is co-located with the Western Gulf Forest Tree Improvement Program in Texas A&M University's Forest Science Laboratory. In addition Dr. Faridi has adjunct appointments with the Departments of Forest Science and Soil and Crop Science.

The main focus of the Forest Tree Molecular Cytogenetics Laboratory (FTMCL) is to study the genomic organization of forest trees and in particular the genus *Pinus*. Along with Drs. Dana Nelson (Project Leader, SIFG) and Tom Kubisiak (Research Geneticist, SIFG), Dr. Faridi has developed standard loblolly pine and slash pine fluorescent in-situ hybridization (FISH) -based karyotypes using 18S-28S rDNA, 5S rDNA, *Arabidopsis*-type telomere probes and AT-rich banding. This work utilized statistical analyses of chromosomal measurements collected from several trees and cells within trees to make possible the definitive identification of each of the 12 chromosomes of both loblolly and slash pine (manuscript in preparation).

The FTMCL is presently working on the following projects: (1) Developing shortleaf pine and longleaf pine FISH-based karyotypes and comparing these to loblolly and slash pines, and (2) Locating the physical position of fusiform rust resistant gene (Fr1) in loblolly pine. In addition Dr. Faridi has been awarded funds from National Science Foundation (NSF) and Department Of Energy (DOE) to provide

molecular cytogenetic analyses of loblolly pine and hybrid poplar through the following projects: NSF -- Accelerating pine genomics through development and utilization of molecular and cytogenetics resources, with Drs. Daniel Peterson, Mississippi State University and Dana Nelson; and DOE-- Assembling the poplar genome sequence into chromosomal scaffolds and examining chromosome structure using FISH, with Dr. Stephen DiFazio, Oak Ridge National Laboratory.

The FTMCL has recently identified a new retrotransposable element in pine. In the project to locate Fr1 in loblolly pine, Drs. Faridi, Nelson, and Kubisiak teamed up with two other groups (Drs. John Davis and Alison Morse, University of Florida and Kathy Smith SIFG/UFL, and Dr. Henry Amerson, North Carolina State University). Dr. Davis' group cloned three DNA fragments linked to the Fr1 locus. The total length of these three clones, 8 Kb, is presumably adequate for cytological location using FISH. Two of these clones (B8 and J4) contained retrotransposable elements, which hybridized to the whole *Pinus* genome, including loblolly, slash, and longleaf pines. Interestingly this retroelement has not invaded the centromeric and 18S-28S rDNA sites in pine (Figure 22). A database search found no such retroelement sequence in any gymnosperm species, but did find a significant match in an angiosperm species. This retroelement will be studied in various species of *Pinus* and other conifers including some distantly related gymnosperms.



Figure 22. Longleaf pine chromosomes showing the binding sites for a previously unknown retroelement.

<sup>3</sup> Contributed by Dr. Nurul Faridi, USDA Forest Service.

# HARDWOOD TREE IMPROVEMENT PROGRAM

## Highlights

- The Western Gulf Forest Tree Improvement Program – Hardwood has completed first-generation progeny testing for green ash, sweetgum, sycamore, cherrybark oak, water/willow oak, and yellow poplar.
- All of the Nuttall oak progeny test series have been measured once at age 5 and the oldest planting has been measured at both ages 5 and 10.
- Lack of agreement between ages 5 and 10 in the first series of Nuttall oak progeny tests argues that early evaluations for designing orchards will not be successful for this species.
- Twenty-year measurements on Potlatch Corporation’s natural regeneration study Project 1000 confirm that any type of mechanical site preparation encourages desirable species. Shearing seems to favor oaks while chopping favors sweetgum. The oak component increased relative to sweetgum between ages 15 and 20.

## Tree Improvement

### Progeny Testing

The Western Gulf Forest Tree Improvement Program – Hardwood continues to close progeny tests after they are evaluated at age 20. During the 2003/04 measurement year, two cherrybark oak tests were measured for the final time and a water/willow oak test was dropped prior to measurement due to its poor condition. The cooperative has now completed first-generation evaluations for all of the species for which orchards have been established. This includes green ash, sweetgum, sycamore, cherrybark oak, water/willow oak, and yellow poplar. Active progeny tests are currently maintained to evaluate first-generation parents for Nuttall oak, and to evaluate advanced-generation selections from sweetgum and sycamore (Table 5).

**Table 5. Active progeny tests in the Hardwood Tree Improvement Program.**

Species	Number of Tests
Nuttall oak	22
Sweetgum	2
Sycamore	1

In addition to the two twenty-year-old progeny tests measured during the 2003/04 season, three five-year-old and three ten-year-old Nuttall oak tests were also measured. The ten-year measurements represented the second evaluation for

this series of tests and were used to assess the potential for early selection in this species.

### Cherrybark Oak

The two cherrybark tests evaluated at age 20 were located in Tyler County, TX and Washington Parish, Louisiana (Table 6). These plantings contained open-pollinated families from two different sets of parents and evaluation of genotype by environment interaction was not possible. Growth rates across these two locations illustrate both the excellent growth potential for this species and the degree of site selectivity and management required if this species is to be managed in plantations. Both sites were selected because they appeared to be appropriate for this species, which prefers a well-drained bottomland soil. It was possible to mow one site regularly, while the other site was visited infrequently because of difficulties with access. As a result, small site differences compounded by different levels of management inputs made large differences in growth rate. The average tree in the Tyler County test was 3.5 m taller and 5.7 cm larger in diameter than the average tree in Washington Parish. The Tyler County test has been thinned by Temple-Inland Forest for conversion into a seed production area (Figure 23).

**Table 6. Twenty-year results from the cherrybark oak progeny tests measured in 2003/04 measurement season. Family differences were significant at the 10 percent level or less for all traits except as noted (ns).**

Location (Co./Par., State)	Plantation Average			
	Survival (%)	Height (m)	Diameter (cm)	Volume (dm <sup>3</sup> )
Tyler, TX	80.61	8.21	8.21	44.1
Washington, LA	84.2(ns)	12.8	12.6	57.5



*Figure 23. Jim Tule in a cherrybark oak progeny test converted by Temple-Inland Forest into a seed production area after its 20-year evaluation.*

## Nuttall Oak

The cooperators of the Western Gulf Forest Tree Improvement Program – Hardwood established 22 Nuttall oak progeny tests in five planting series between 1994 and 1999 to evaluate the performance of 216 parents. During 2003/04, the members measured three Nuttall oak progeny tests, representing the last series of tests for this species (Table 7). Survival among all three plantings was very similar. The Lonoke County, AR test, which lacked significance for all the traits evaluated, had suffered repeated attacks of red oak borer. This insect bores into the trunk and kills the stem above the point of attack. Trees with multiple attacks frequently die; however, the main effect observed in this test was loss of height growth and forking.

**Table 7. Five-year results from the Nuttall oak progeny tests measured in 2003/04 measurement season. Family differences were significant at the 10 percent level or less for all traits except as noted (ns).**

Location (Co./Par., State)	Plantation Average			
	Survival (%)	Height (m)	Diameter (cm)	Volume (dm <sup>3</sup> )
Angelina, TX	85.5	2.71	2.18	0.52
Sharkey, MS	94.1	3.73	4.57	2.14
Lonoke, AR	81.5(ns)	3.18(ns)	2.77(ns)	0.72 (ns)

Three plantings in the first series of tests established with Nuttall oak were measured at age 10 (Table 8). These plantings had received excellent site-preparation prior to establishment and regular maintenance during their life. The three locations were analyzed separately and then combined to evaluate differences among locations and genotype by environment interactions. Finally, age ten evaluations were compared to previous evaluations at age five.

**Table 8. Ten-year results from the Nuttall oak progeny tests measured in 2003/04 measurement season. Family differences were significant at the 10 percent level or less for all traits except as noted (ns).**

Location (Co./Par., State)	Plantation Average			
	Survival (%)	Height (m)	Diameter (cm)	Volume (dm <sup>3</sup> )
Desha, AR	73.4	5.5	6.6	5.9
Sharkey, MS	80.5	5.7	7.2	7.7
Lonoke, AR	96.0	8.1	9.8	21.6

In the combined-location analysis, the only trait for which there were no discernable differences among families was survival. This species has proven to be very tenacious, even when planted on less than optimal sites, and good survival seems to be a trait possessed by the majority of families. There were differences among families and genotype by environment interactions for height, diameter, and volume growth. The genotype by environment

interactions were legitimate changes in ranks among parents at different locations indicating that selecting families that do well across a range of sites may be challenging (Table 9). These sites were located along a northwest to southeast transect. The two sites that were the most distant were no more different from each other than they were from the middle site indicating that these differences are not occurring in response to clinal variation in the environment.

**Table 9. Pearson rank correlations among families represented in the ten-year old Nuttall oak progeny tests measured in 2003/04 measurement season.**

Location (Co./Par., State)	Lonoke, AR	Desha, AR	Sharkey, MS
Lonoke, AR	1	0.25	0.28
Desha, AR		1	0.26
Sharkey, MS			1

Growth rates and uniformity have been excellent at all three of these locations raising the hope that five-year data might be predictive of 10-year performance. If this were true, then early evaluations could be used to begin orchard establishment for this species as all series have been evaluated at age five. Unfortunately, there was poor agreement between the age five evaluations and the age 10 evaluations (Table 10). It will, therefore, be necessary to wait until there are a sufficient number of parents with ten-year evaluations to begin designing orchards.

**Table 10. Pearson rank correlations among families between ages five and ten.**

Location (Co./Par., State)	Trait		
	Height	Diameter	Volume
Lonoke, AR	0.70	0.79	0.63
Desha, AR	0.71	0.58	0.64
Sharkey, MS	0.51	0.42	0.55
<i>Average</i>	<i>0.64</i>	<i>0.60</i>	<i>0.61</i>

## Seed Orchards

Louisiana Forest Seed Company, the hardwood cooperative's newest member started an ambitious seed orchard establishment programs for green ash, Chinkapin oak, willow oak, and cherrybark oak. During the first year, both chip budding for all species and rooted cuttings for the willow oak were attempted. The green ash grafting was successful and orchard establishment will begin with transplanted grafts in 2004/05. Attempts to graft the oaks were less successful, due in part to poor weather conditions. At the time the chip buds should have been knitting, the region experienced three weeks of daily rainfall. Many of the chip bud unions, which were held together by grafting wax rather than the paraffin used in normal cleft grafts, simply mildewed. Rooting of willow oaks was somewhat more successful and

orchard establishment with these propagules will begin in 2005/06. More rooting for orchard establishment will be attempted in the future.

The Louisiana Department of Agriculture and Forestry continues to develop numerous hardwood orchards at their Monroe Nursery. The largest of these is a bald cypress orchard. Eventually this site will also contain orchards for bald cypress, cherrybark oak, and Nuttall oak. The Mississippi Forestry Commission continued to establish Nuttall oak scion banks at their Winona Nursery. They also began establishment of separate orchards for both water and willow oaks in 2003/04. Both the Mississippi Forestry Commission and Louisiana Forest Seed Company are establishing pure willow oak orchards. The water/willow oak program initially depended on progeny tests of acorns collected from ortets in the wild. Because of the nature of these older stands, the fact that these two species frequently hybridize, and the similarity of the seed, it was not always possible to reliably distinguish between these two species. Therefore, progeny testing and original orchard establishment was done for the water/willow oak complex. Now that the cooperative has had an opportunity to observe these trees both as grafts and as seedlings in replicated progeny tests, a number of selections have been identified as pure willow oak which will be established as separate orchards.

### Natural Regeneration – Project 1000

Potlatch Corporation established a hardwood natural regeneration study in 1983 with four site preparation treatments following an operational clear cut. The treatments included clearcut only (operational control), high shear, chop, and chemical injection of residuals. The study consisted of ten-acre treatment blocks with three replications. Pre-harvest conditions were recorded and subsequent species composition and growth of dominant trees were measured periodically. At age 20, 16 permanent measurement plots in each treatment block were revisited and all trees greater than four inches in diameter on a 0.1-acre plot were tallied. Height growth was measured on the two dominant trees of each species.

At age 20, 40 percent of the total 50.3 square feet of basal area across all treatments was in sweetgum, 30 percent in oaks and 23 percent in miscellaneous desirables (mostly pine). Undesirable species, which had been a significant component of the stand at earlier ages, began to decline in importance (Figure 24). Only the shear treatment resulted in more basal area in oaks than the untreated control. This treatment also resulted in the establishment of naturally seeded loblolly pine and may lead to a mixed pine/hardwood stand in what had previously been a hardwood bottom. The oak proportion of the total number of stems (rather than the basal area) increased in all treatments between ages 15 and 20. However, the increase was largest in the inject treatment which had previously trailed the shear treatment (Figure 25).

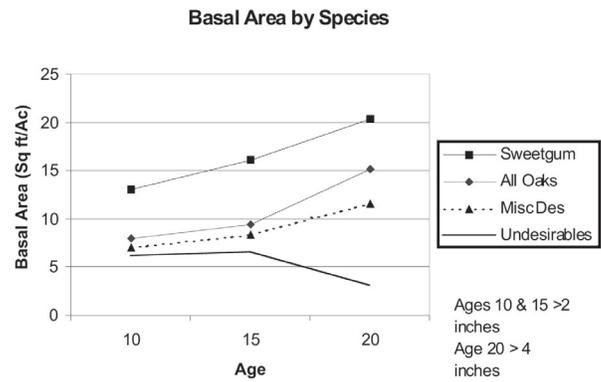


Figure 24. Basal area at 10, 15, and 20 years for different species classes averaged across all treatments in Potlatch Corporation's natural regeneration study.

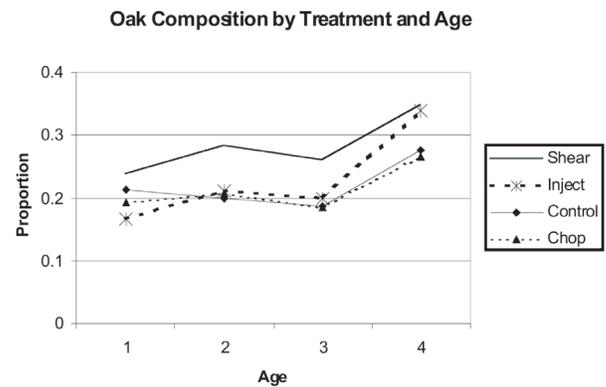


Figure 25. The proportion of the total number of stems represented by oaks over different ages for the different treatments in Potlatch Corporation's natural regeneration study.

Conclusions drawn from this study to date are that any mechanical site preparation will have the effect of reducing the undesirables and encouraging sweetgum and oak regeneration. Shearing appears to favor the oaks while chopping favors sweetgum. Finally, early evaluations are meaningless in predicting stand composition. It has taken 20 years for the relative dominance of the sweetgum to begin to give way to the slower growing oaks that are predicted to ultimately dominate the stand.

## PERSONNEL

This year there were no personnel changes to report in the cooperative staff or the Texas Forest Service tree improvement programs. The WGFTIP is pleased to welcome Dr. Nurul Faridi, whose office and laboratory will be in the Forest Science Laboratory building with the WGFTIP. Dr. Faridi is a research scientist employed by the Southern Institute of Forest Genetics, USDA Forest Service. His research project will be the only one in the country completely dedicated to the cytology of forest trees.

### The staff now includes the following people:

T. D. Byram..... WGFTIP Geneticist  
L. G. Miller.....WGFTIP Assistant Geneticist  
E. M. (Fred) Raley.....WGFTIP Assistant Geneticist  
P. V. Sieling.....Staff Assistant  
J. G. Hernandez..... Research Specialist  
G. R. Lively..... Research Specialist  
I. N. Brown.....Research Specialist  
D. M. Travis, Jr.....Research Specialist  
G. F. Fountain.....Aide to Specialist  
A. R. Crain..... Aide to Specialist

## PUBLICATIONS

- Bridgwater, F. E., T. Kubisiak, T. D. Byram, and S. E. McKeand. Risk management with current deployment strategies for genetically improved loblolly and slash pines. Information Exchange Group Meeting. Wrightsville Beach, NC. September 17-18, 2003. (Accepted by the Southern Journal of Applied Forestry).
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- Roberds, J. H., Strom, B. L., Hain, F. P., Gwaze, D. P., McKeand, S. E. and Lott, L. H. Estimates of genetic parameters for oleoresin and growth traits in juvenile loblolly pine. Canadian Journal of Forest Research (Accepted).
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- White, T. L. and T. D. Byram. 2004. Slash pine tree improvement. In: E.D. Dickens, J.P. Barnett, W.G. Hubbard and E.J. Jokela (Editors). Slash pine: still growing and growing! Proc. of the slash pine symposium. Gen. Tech. Rep. SRS-76. Asheville, NC: U.S. Department of Agriculture, Forest Service, Southern Research Station. pp.7-19.

## COOPERATIVE TREE IMPROVEMENT PROGRAM MEMBERS

### Western Gulf Forest Tree Improvement Program Membership

#### Pine Program

Full members of the Western Gulf Forest Tree Improvement Pine Program in 2004 include the Arkansas Forestry Commission, Boise, CellFor, Inc., Deltic Timber Corporation, ETT, L.P. (represented by Molpus Timberlands Management), International Paper Company, Louisiana Department of Agriculture and Forestry, Mississippi Forestry Commission, Oklahoma Department of Agriculture, Food and Forestry, Plum Creek Timber Company, Potlatch Corporation, Temple-Inland Forest, Texas Forest Service, Weyerhaeuser Company.

Associate members include International Forest Seed Company, Louisiana Forest Seed Company, and Robbins Association.

### Hardwood Program

The WGFTIP Hardwood Program includes the Arkansas Forestry Commission, Louisiana Department of Agriculture and Forestry, Louisiana Forest Seed Company, Mississippi Forestry Commission, Potlatch Corporation, Temple-Inland Forest, and the Texas Forest Service.

### Urban Tree Improvement Program

Membership in the Urban Tree Improvement Program includes the following municipalities and nurseries: Aldridge Nurseries (Von Ormy), Altex Nurseries (Alvin), Baytown, Burleson, Carrollton, Dallas, Dallas Nurseries (Lewisville), Fort Worth, Garland, Houston, LMS Landscape (Dallas), Plano, Rennerwood (Tennessee Colony), Richardson, Robertson's Tree Farm (Whitehouse), and Superior Tree Foliage (Tomball).

## FINANCIAL SUPPORT

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Agricultural Experiment Station, the Texas Forest Service, the Texas Christmas Tree Growers Association, and the USDA Forest Service.