

PROSPECTS FOR USING NATURAL LOBLOLLY x SHORTLEAF HYBRIDS
FOR RESISTANCE TO FUSIFORM RUST

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Abstract. --Natural hybridization of loblolly pine (*Pinus taeda* L.) and shortleaf pine (*Pinus echinata* Mill.) has been suggested as the probable cause of resistance to fusiform rust (*Cronartium fusiforme* Hedge and Hunt) in western provenances of loblolly pine. Phenology studies of loblolly and shortleaf pines in East Texas have indicated an overlap in pollen shed of the two species.

From a hybrid index based on five morphological characters, trees with characteristics intermediate to the two species have been detected.

Investigations are currently underway to determine what implications such introgression may have with respect to rust resistance and the possible uses of this source of natural resistance.

Additional keywords: phenology, hybrid index, numerical classification, sample sizes.

INTRODUCTION

The existence of phenotypes intermediate to both species has been observed within mixed natural populations of loblolly (*Pinus taeda* L.) and shortleaf (*P. echinata* Mill.) pines in East Texas (Zobel, 1953; Bilan, 1965). Should these intermediate types possess the acknowledged resistance to fusiform rust (*Cronartium fusiforme* Hedge and Hunt) exhibited by shortleaf pine (Snow and Kais, 1970) and the more desirable growth characteristics inherent in loblolly pine, identification and selection of these intermediates could represent a gene pool containing desirable recombinants.

Work by Dorman and Barber (1956) concluded that both loblolly and shortleaf were releasing pollen concurrently between March 20 and March 26 in 1951, in Nacogdoches County, Texas. Hicks, et al. (1972) confirmed this possibility in the same area in 1971. Twenty sexually mature trees of each species were monitored from February 12 to April 27. Results showed that 10 percent of the shortleaf pines under observation were producing pollen simultaneously with more than half of the loblolly pines. Since female strobili are normally receptive shortly following first pollen release, phenological opportunity exists for interspecific crosses.

This paper describes current research at the School of Forestry, Stephen F. Austin State University² designed to establish means for

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identifying loblolly x shortleaf hybrids with resistance to fusiform rust, which could be incorporated into a tree improvement program.

SELECTION OF TAXONOMIC TRAITS
AND WITHIN-TREE SAMPLE SIZES

Hicks (In press) tested 12 morphological characteristics among shortleaf, loblolly, and putative hybrids. His analysis was based upon a numerical taxonomic classification method proposed by Flake and Turner (1968). No attempt was made to evaluate environmental influence upon the traits. Table 1 shows the resulting estimates (using Stein's two-stage sample technique) of sample size required to measure each character within ± 5 percent of true mean (Steel and Torrie, 1960). Characters involving difficult determinations, or very large samples were considered least suitable for taxonomic determinations. Among the morphological characteristics examined, those which appeared best suited for segregating these taxa were needle length, fascicle sheath length, number of needles per fascicle, terminal bud width, and cone length.

Table 1. Sample size required to estimate within-tree means within $\pm 5\%$ of the true mean.

Character	Number of Samples Required	
	t = 90%	t = 95%
Needle length	17 needles	28 needles
Needle width	113 needles	192 needles
Fascicle sheath length	10 fascicles	18 fascicles
No. of needles per fascicle	31 fascicles	52 fascicles
No. of stomatal rows	67 needles	118 needles
No. of stomates per cm in rows	12 needles	20 needles
Terminal bud length	42 buds	82 buds
Terminal bud width	53 buds	102 buds
Axillary scale width	715 scales	1,210 scales
Cone length	26 cones	45 cones
Cone width	97 cones	169 cones
Seed weight		

*The within-tree sample size could not be calculated for seed weight due to the original sampling technique, i.e. a single within-tree sample.

Table 2 demonstrates the correlation coefficients (r) derived for the 12 traits. Hybrids of two different species should produce progeny which are intermediate for quantitative traits; thus characters which reflect genetic differences in the parental species should possess genetic correlations in an introgressed population. High (r) values could result from environmental effects; therefore should not be taken as an absolute measure of taxonomic worth. Low (r) values demonstrate little worth of a trait for taxonomic purposes.

Table 2. Matrix consisting of correlation coefficients (r) derived from all possible combinations of tree-mean values for morphological characters observed (after Hicks, 1971).

Needle Length	Needle Width	No. of Stomates Per cm in Rows	Fascicle Sheath Length	No. of Needles Per Fascicle	Terminal Bud Length	No. of Stomatal Rows	Terminal Bud Width	Axillary Scale Width	Cone Length	Cone Width	Seed Weight	
Needle Length		.821**	.018ns	.858**	.856**	.039ns	.215ns	.722**	.894**	.822**	.719**	.791**
Needle Width			.350*	.788**	.821**	.641**	.597**	.814**	.895**	.728**	.631**	.696**
No. of Stomates Per Cm In Rows				.172ns	.139ns	.159ns	.793**	.233ns	.170ns	.113ns	.070ns	.073ns
Fascicle Sheath Length					.842**	.493**	.312*	.829**	.857**	.709**	.654**	.689**
No. of Needles Per Fascicle						.501**	.265ns	.752**	.861**	.728**	.620**	.661**
Terminal Bud Length							.294*	.712**	.689**	.314*	.310*	.369*
No. of Stomatal Rows								.395**	.337*	.288*	.217ns	.245ns
Terminal Bud Width									.849**	.634**	.576**	.575**
Axillary Scale Width										.774**	.680**	.762**
Cone Length											.908**	.883**
Cone Width												.797**
Seed Weight												

** From the estimated value of r, it can be concluded that the population correlation coefficient is significantly different from 0 at the 0.1 level.

* Significant at the 0.05 level.

ns Not significant at the 0.05 level.

FREQUENCY OF INTERSPECIFIC HYBRIDS
WITHIN NATURAL STANDS IN EAST TEXAS

In the fall of 1971, samples were collected from 164 trees in 16 stands in East Texas for the purpose of determining the frequency of natural hybridization between loblolly and shortleaf pines (Cotton, 1972). Three basic mathematical techniques were applied to determine how intermediate types might be differentiated within a natural population based upon correlated morphological traits.

Samples of cones, buds, and needles were measured for five morphological traits. These were cone length, terminal bud width, sheath length, needle length, and the number of needles per fascicle.

Following derivation of within-tree means for each character, tree-mean values of the trees exhibiting the minimum and maximum raw measurements for each trait were assigned scores of 0.0 and 5.0, respectively. The mean values for other individuals were transformed to a linear scale of 0.0 to 5.0 by interpolation. These scores were then used for analysis by hybrid index (Anderson, 1949), cluster analysis (Veldman, 1967), and factor analysis (Lawley and Maxwell, 1963).

Figure 1 shows the frequency distribution of individuals by hybrid index scores with three cluster analysis groups given letter designations (Hicks, et al., 1973). The distribution indicates two distinct, normally distributed populations representing loblolly and shortleaf pine. Morphologically intermediate trees were present but appeared to be normal deviates of their respective parental populations.

The fact that three cluster analysis groups exist does not necessarily imply that the intermediates are natural hybrids. Due to the nature of the technique, any specified number of cluster groups will be formed at some level.

Principal component analysis (Figure 2) developed two component factors, one of which accounted for almost 99% of the total variation in the five morphological traits.

The bulk of the evidence at this point indicates that there is little justification for claiming widespread introgression among loblolly and shortleaf pines. Isoenzyme analysis of seed proteins of trees with a range of morphological types conceivably should be beneficial in establishing whether hybridity is the source of some morphological variability. The latter observation has prompted the beginning of isoenzyme studies by electrophoresis in the near future.

TESTING FOR RUST RESISTANCE

Preparation for further testing began in the fall of 1972 when needle, bud, and mature cone samples were collected from 52 trees at four locations within a sixty-mile radius of Nacogdoches (Florence, 1973). Trees sampled were selected on the basis of their position on the hybrid index scale employed by Cotton (1972). Trees ranging from



Figure 1. Frequency distribution of the sample population based on hybrid index values. Letter designations indicate cluster analysis groups. (Plotting by Hicks, from data by Cotton, 1972).

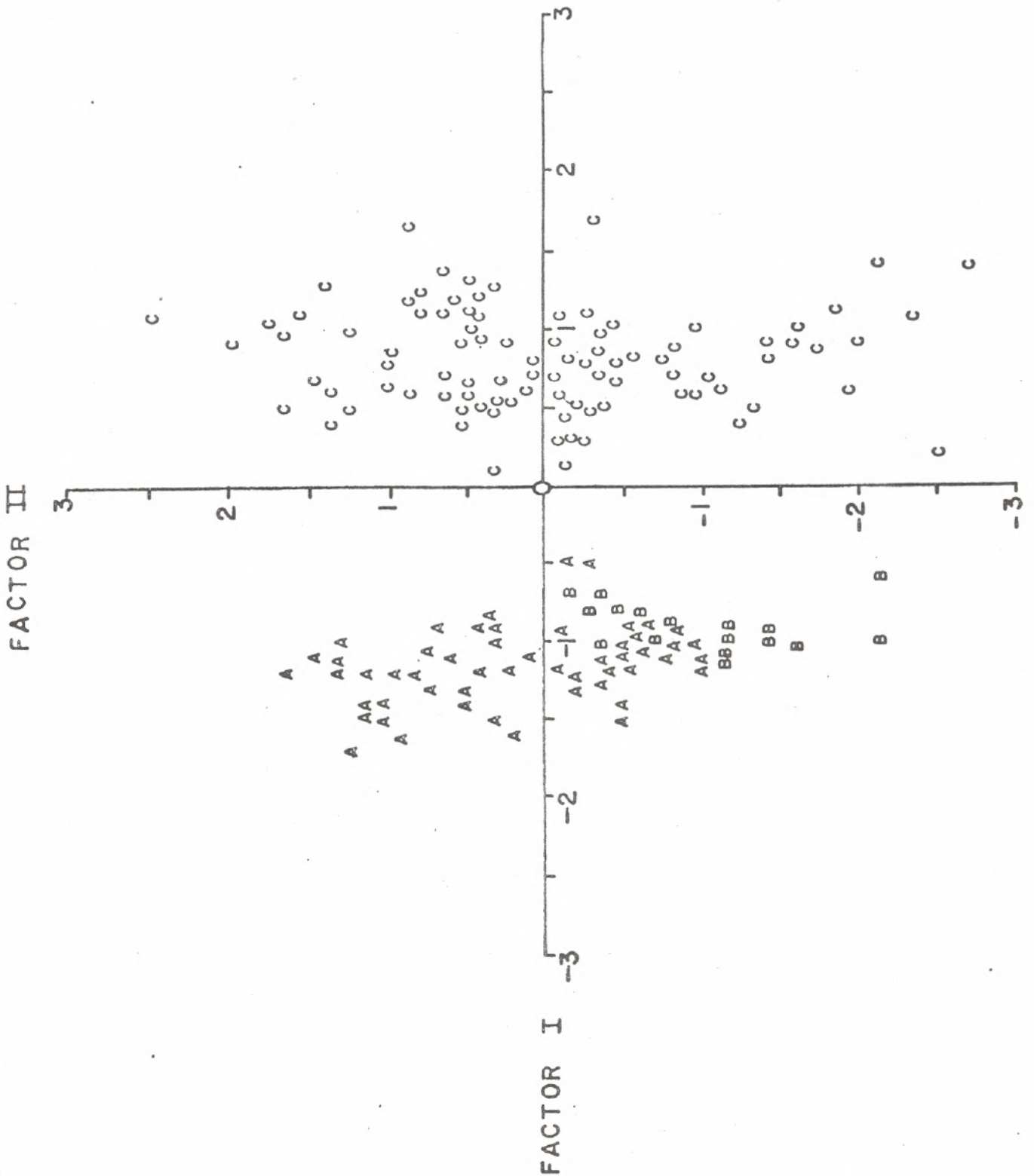


Figure 2. Plotting of the sample population by principal axis factor scores from principal components analysis. Letter designations indicate cluster groupings of individuals into three cluster groups. (Plotting by Hicks, from data by Cotton, 1972).

shortleaf types to loblolly types, including intermediates, were sampled.

Seed from the 1972 samples were extracted, cleaned, and stratified during the past winter. Fifteen half-sib family lots of each subjective group (45 lots total) have been submitted for testing in this year's fusiform rust trials conducted at the U. S. Forest Service's Forest Pest Management facilities at Asheville, North Carolina. Herein lies the prime objective of the current study.

At this point in our research at Stephen F. Austin State University we wish to culminate what has been learned of the East Texas pine population into a manageable framework of information. Hopefully, such a framework will permit us to select for loblolly types with greater surety of a resistant individual's genotype.

Subsequent testing is planned to reinforce this goal through progeny tests conducted in controlled greenhouse studies and electrophoretic protein separations. Each of these studies will include use of these same 45 families soon to be tested for their resistance to fusiform rust.

SUMMARY

Sequential research of shortleaf and loblolly pine in East Texas at Stephen F. Austin State University during the past three years has shown their phenology to permit natural crossing.

Based upon five correlated morphological characteristics, it has been concluded that natural hybrids are infrequent in the natural population if morphological characters are good indicators of such hybrids.

Current research is devoted to determining to what extent intermediates may express resistance to fusiform rust as compared to parental types. Concurrently, studies are in progress to more fully develop the progeny morphology and biochemical relationships to rust resistance as an addendum to practical field selection from the natural parental population.

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