Genetic Divergence in Selected Populations of Loblolly pine*

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Introduction

A study of natural variation within a species of commercial interest is important because success in genetic manipulation depends on thorough understanding of both the amount and nature of such variation. This is especially important in species of wide natural distribution like loblolly pine whose range covers a large portion of the Southeastern U.S.A. Geographic variation in loblolly pine was studied and as part of a range-wide seed source study (Wells and Wakely 1966). Results from this study indicated strong clinal variation in growth and survival from the east coast westwards and from South to North on the eastern extremity of the species range. There were also indications of great geographic variation in fusiform rust (*Cronartium fusiforme* Hedc. and Hunt ex. Cumm.) resistance but this was not as clinal as the other two traits. Geographic variation in wood specific gravity was studied by Zobel (1961). In these studies, he found the relative proportions of geographic, sites within geographic areas, tree to tree and within tree variations to be 15%, 3%, 70% and 12% respectively. From these kinds of studies the breeder can determine where to place his efforts to the maximum benefit. For instance, large geographic variation suggests benefits from provenance testing and selection while large tree to tree variation suggest that intrapopulation selection program is a viable breeding strategy. Based on these kinds of information Zobel and Dorman (1973) have defined seven different provenances within the loblolly pine range that could be used in different areas as exotics.

Another objective for studying natural variation aims at determining the amount of genetic differentiation with a species range. This kind of information is very useful as a basis for planned preservation of gene pools which could be used, later on, to create new gene combinations. Wide crosses among such genetically differentiated races can play a key role in the creation of new gene combinations. The wide cross study, as has been detailed earlier offered some limited opportunities for studying the amount of genetic differentiation within the loblolly pine range. The specific objectives of this study were:

1. To estimate the additive genetic components for open-pollinated progeny from different geographic sources when grown in Halifax County, North Carolina.
2. To estimate the degree of genetic differentiation among the geographic sources for height growth, fusiform rust resistance, crown form, and stem straightness.

Historical Background

Most of the Southeastern U.S. was under water until recent geologic times. Wakely (1961) hypothesized that the present distribution of loblolly pine originated from two islandic relic sources; one in Florida and the other in Texas. He further hypothesized that the wide Mississippi drainage formed a barrier between the two relic sources. According to this hypothesis, population expansion has taken place from the Texas source eastwards and from the Florida source north and westwards.

Originally, loblolly pine occupied lowlands bordering or within swamps, pocosins and savannas (Wahlenberg 1962). Upon widespread agricultural failures of the colonial settlements of the 19th century, loblolly pine had ecological release into abandoned cotton old fields and most of the present natural stands are on such old-fields.

Genetic Memoranda

Geographic variation can be due to three possible causes: (1) differential selective pressures over the species range, (2) random genetic drift, (3) historical accidents that have contributed to the heterogeneity of the populations but not yet cancelled by diffusion (Cavalli-Sforza and Bodmer 1971). For loblolly pine, the clinal variation in growth and survival is evidence of differential selective pressures. Random drift could also have played a major role in genetic differentiation since the old field invasion started with small stands. Under such circumstances, large genetic drift effects could result in accordance with the founder *principle.* Possible crosses between Texas and Florida relics that have not reached equilibrium can be listed under historical accidents. Interspecific hybridization with other pine species with which loblolly readily hybridizes should be included in this category. Geographic variation can also be caused by the Baldwin effect at the species range extremeties whereby chance mutations or recombinations prove beneficial in these harsh environments and are subsequently fixed in isolate populations (Grant 1963). The above factors tend to cause geographic differentiation but can be counteracted by the forces of migration and gene flow. In continuous populations, effective population size is taken as the neighborhood size *i.e.,* the largest panmictic unit (Wright 1962). The formula $N = \pi\sigma\delta$ d where d = the number of trees per acre for hermaphroditic population having areally continuous range, $\sigma\delta$ = standard deviation of pollen dispersion distance was used by McElwee (1970) to determine the neighborhood sizes of from 1.5 acres (150 trees) to 5.75 ares (600 trees) in natural stands of loblolly pine. It has been shown by Wright (1962) that for a neighborhood size of N, sizeable differentiation (geographic races) can be expected within an area N times the neighborhood size. Using this rough estimation, genetic differentiation can be expected within 40 acres of continuous loblolly pine stand. The sources included in the present study were well beyond these limits and therefore, appreciable degree of genetic differentiation between them could be expected. It must, however, be remembered that these old field population have existed only for about five generations.

Materials and Methods

There were 32 open-pollinated seed identified by mother tree from 10 widely distributed sources in the natural range of loblolly pine. All the families were represented in the planting in Halifax County, North Carolina. Randomized

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complete block design was used with ten-tree row plots planted at 9' by 9' spacing.

Statistical Models and Data Analysis

The analysis for open-pollinated progeny at a single location was based on the following model:

$$\bar{Y}_{ij} = mu + r_i + f_j + (rf)_{ij}$$

where

$\bar{Y}_{ij}$ = the mean measurement of the jth family in the ith replicate
mu = general mean of all families over all replications
r_i = added effect of the ith replication ($i = 1,2,\ldots,h$)
f_j = added effect of the jth family ($j = 1,2,\ldots,m$)
$(rf)_{ij}$ = effect of the interaction of the jth family with the ith replicate

The expected mean square of the appropriate analysis of variance are shown in *table 1.*

Table 1. — Form of analysis of variance on plot mean basis of open-pollinated progenies tested in one location.

Source of variation	DF	Expected mean square[1])
Replications	n — 1	$(\sigma^2_w/_k + \sigma^2_{rf}) + m\sigma^2_r$
Families	m — 1	$(\sigma^2_w/_k + \sigma^2_{rf}) + n\sigma^2_f$
Reps × families	(m — 1) (n — 1)	$(\sigma^2_w/_k + \sigma^2_{rf})$

[1]) σ^2_w = within plot variance
σ^2_{rf} = variance du to rep × families interaction
σ^2_r = variance due to replication differences
σ^2_f = variance due to family differences
k = harmonic mean of the number of trees/plot

A similar analysis for the open-pollinated families nested within their areas of origin was based on the following model

$$\bar{Y}_{ijk} = mu + g_i + f_{j(i)} + r_k + e_{ijk}$$

$\bar{Y}_{ijk}$ = the mean of the kth replicated of the jth family nested within the ith region or origin
mu = mean of all observations
g_i = added effect of the ith region
$f_{j(i)}$ = added effect of the jth family within ith region
r_k = effect of the kth replicate
e_{ijk} = residual

The expected mean squares in the corresponding analysis of variance are shown in *table 3.*

Results and Discussion

The estimates of additive genetic variance presented in *table 2* are calculated according to the relationship $\sigma^2_f = 1/4\,\sigma^2_A$. The assumptions necessary for such a relationship to hold have been detailed by STONECYPHER (1966). As he has pointed out, the most questionable of the assumptions is that the open-pollinated progenies are half-sibs. This can only be a realistic assumption if each seed tree has an equal chance of receiving pollen from any tree in the reference population. Even in uniformly stocked stands, differences in flowering time can cause deviations from random pollination. Of greater disturbance, in this particular study, is the fact that the selected trees were widely separated within a geographic source. Thus within a source, selected trees were separated by distances of the order of 10 miles (WOESSNER 1968). The assumption that these constitute half-sibs from a given source should therefore be highly suspect.

Table 2. — Estimates of additive genetic variance for four characters based on open-pollinated progeny performance in Halifax County, N.C.[1])

Family groups	Characters			
	Height growth	Rust Score	Crown Score	Strightness Score
Champion	0	0	0	0
Louisiana	2.83	0	.717	.014
Bowaters	9.58	.324	0	0
Continental Can	0	0	0	0
Kimberly-Clark	.525	0	2.528	2.475
Texas	0	0	.52	0
Weyerhaeuser	2.56	.036	.20	0
Hoerner-Waldorf	0	0	1.777	.84
Westvaco	.233	.027	.827	2.49
	1.747	.043	.729	.757

[1]) Additive genetic variance of zero represent the cases where the estimate of σ^2_f was either zero or negative.

Table 3. — Form of analysis of variance for open-pollinated progenies based on plot means with families nested within areas of origin.[1])

Source of variation	DF	Expected mean square[1])
Replications	8	—
Regions	9	[2]) $(\sigma^2_w/_k + \sigma^2_{rf}) + k_2\,\sigma^2_f + k_3\sigma^2_g$
Families (Regions)	27	$(\sigma^2_w/_k + \sigma^2_{rf}) + k_1\,\sigma^2_f$
Error	278	$(\sigma^2_w/_k + \sigma^2_r)$

[1]) Only the performance in Halifax County, N.C. was analysed.
[2]) $k_1 = k_2$ = number of replications per half-sib family.
k_3 = nm where n is the number of replications and n is the harmonic mean of the number of families per region.
σ^2_g = variance due to differences between regions.

Another potential source of error is the sample size. For each source, the number of open-pollinated progeny ranged from 4 to 8; from past experience with sampling problems in tree breeding experiments, these samples sizes are well below the minimum required for any meaningful genetic interpretation. A combined analyses for all the open-pollinated progeny resulted in estimates of 1.214, .008, .862 and .911 for additive genetic variance for height growth, fusiform rust score, crown score and stem straightness score respectively. In most cases, the sampling errors were larger than the estimates themselves. The great variation in *table 2* can therefore be attributed mainly to the very small samples used.

The data from these open-pollinated progenies was used in analyses aimed at inferences about genetic differentiation within the loblolly pine natural range. Such analyses have been attempted elsewhere by STERN (1961) who also enumerated the necessary assumptions for genetic interpretations from the analyses. The assumptions are similar to those discussed by STONECYPHER (1966) with the added assumption that the degree of inbreeding is the same for all the geographic subpopulations (regions). As has been pointed out earlier, intra-specific genetic differentiation in a species of wide natural range like loblolly pine is likely to be due differences in allelic frequencies. The expected allelic disequilibrium $D = \sum_{i=1}^{n} [(p_i - q_i)^2/4p_iq_i]$ (BURROWS 1971) where n is the number of loci involved and p and q are the frequencies of the favorable alleles in population 1 and 2 respectively.

On the assumption of uniform degree of inbreeding, the additive genetic variance is related to allelic disequilibrium as $\sigma^2_A = 2 \sum_{i=1}^{n} p_i q_i (1 - D_i) \alpha^2_i$ where α is the average effect of gene substitution. Using components as derived in *table 3*, Stern (1961) has shown that

$$E(\sigma^2_g) = 2\sigma^2_A \text{ and } E(\sigma^2_r) = \frac{(1\text{-}D)}{4} \sigma^2_A.$$

where E stands for expected value. Estimates of D for four characters are shown in *Table 4*. High values of D indicate great genetic differentiation. It is evident from these results that there is considerable genetic differentiation for fusiform rust resistance which concurs with results or other studies like the south wide seed source study (Wakely 1961) where it was reported that the Texas source were uniformly resistant to fusiform rust attack.

Table 4. — Degree of genetic divergence (D) for height growth, rust resistance, crown form and stem straightness.

Character	σ^2_g	σ^2_f	σ^2_A	$\hat{D}$	$\frac{D}{\sigma^2_A} \times 100$
Height growth	.2234	.2539	3.9523	.0283	.71
Rust resistance	.0047	.006	.0936	.0256	27
Crown form	.0470	.0199	.2944	.0798	27
Stem straightness	.0519	.0524	.8123	.0319	2.7

σ^2_g = variance due to differences between regions.

1) σ^2_f = variance due to differences between families within regions.

σ^2_A = additive genetic variance.

$\hat{D}$ = measure of genetic divergence.

A comparatively little genetic differentiation was observed for height growth and only moderate amount for stem straightness. Since the effectiveness of wide crosses as a means of broadening genetic bases of breeding populations depend on the amount of genetic differentiation in the species range, results of the present study indicate that wide cross approach is justifiable for some traits like fusiform rust resistance and crown form, but not for others like height growth and stem straightness.

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Summary

Genetic differentiation of upto 27% of the additive genetic variance was found in the loblolly pine range sampled Differences in the degree of genetic differentiation were observed for different traits; for example, genetic differentiation of under 1% was found for height growth whereas a value of 27% was calculated for resistance to fusiform rust. This suggests that when a breeder is looking for new gene combinations through wide crossing, he is likely to be more successful with such traits as rust resistance and crown form but not stem straightness and height growth.

Key words: *Pinus taeda*, open pollinated progenies, gene frequency differences.

Zusammenfassung

32 frei abgeblühte Nachkommenschaften aus zehn weit voneinander entfernten Beständen aus dem natürlichen Verbreitungsgebiet von *Pinus taeda* wurden in einem Versuch mit vollständigen Blöcken auf Kronenform, Stammform, Höhe und Resistenz gegen *Cronartum fusiforme* untersucht. Das allelische Ungleichgewicht (Burrows 1971) wird als Maß für die intraspezifische genetische Differenzierung aus den Varianzkomponenten für Bestände und Familien in Beständen geschätzt. Es ergibt sich geringe Differenzierung für die Höhe und Stammform, dagegen große Differenzierung für Kronenform und Resistenz, was bedeutet, daß für diese letzteren Merkmale die genetische Basis von Zuchtpopulationen durch Kreuzung weit voneinander entfernter Eltern wünschenswert verbreitert werden kann.

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