INBREEDING AS A MEANS OF GENETIC IMPROVEMENT OF LOBLOLLY PINE

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Traditionally, there has been much speculation and little basic scientific understanding about the potential uses of inbreeding as a means of improving forest trees. Many of us at some time have indulged in a starry-eyed vision of a four-way, double-cross, super-hybrid loblolly pine (or substitute your own favorite species). Recent data from a study in loblolly pine indicate that inbreeding has rather limited potential as a direct means of tree improvement for this species. The study involved comparisons of 75 self- and 75 cross-pollinated families in two replicated greenhouse experiments (Franklin 1968). Additional data were provided from subsequent field plantings, established and maintained in cooperation with North Carolina State University and Albemarle Paper Company. 1

EFFECTS OF INBREEDING

Lower Yields of Seedlings

Seedling yields per cone harvested from self- and cross-pollinations on the same trees have been compared (table 1). Number of 6-month-old seedlings per cone harvested was only one-seventh as great from selfs as from crosses. Thus, to obtain equivalent seedling yields, seven times more selfed canes as crossed cones are required. When the seedlings are outplanted and grow older, there will continue to be more mortality among selfs than crosses, particularly in cases of environmental stress. Comparative costs of producing equivalent numbers of inbred and crossbred seedlings must be a major consideration in evaluating inbreeding as a method of tree improvement.

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Table 1.--Comparison of seed and seedling yields after self- and cross-pollination

	Self	Cross
Total seed per cone	32.7	36.7
Minus empty seed per cone	-28.0	-9.5
Filled seed per cone	4.7	27.2
Minus nongerminating seed	-1.2	-4.0
Germinable seed per cone	3.5	23.2
Minus mortality at six weeks	-0.2	-0.5
Number of living seedlings per cone at six weeks of age	3.3	22.7
Cones required for equivalent seedling yields at six weeks of age	7	1

Pregerminative Selection

The large number of empty seeds following selfing (table 1) indicates that self-fertilized embryos failed to mature in the majority of ovules. Self-pollination yielded only 11 percent viable seed compared to 63 percent following cross-pollination. The stronger natural selection pressure against self-fertilized zygotes eliminates a substantial proportion of the selfbred population before the breeder even collects seed. Natural selection is operative on a trait of no economic importance; namely, pregerminative survival capacity under selfing. The relationships of this trait to economically important traits are not known. Therefore, with selfing the breeder is forced to accept a highly selected population, without knowing how the preselection has biased his chances for genetic gain through further selection.

Imbalance of Experimental Designs

Performance was consistently poorer after selfing than after crossing (table 2). Selfed families had lower germination and lower survival than comparable crossed families. Furthermore, variation in germination and mortality was strongly associated with individual parent tree responses to selfing. Regarding experimental design, this meant that often all the trees from several family plots were lost, instead of a few trees from many different family plots being lost. Consequently, missing plots were much more numerous among families after selfing than after crossing. In one field planting, 675 selfbred and 675 crossbred 1-0 seedlings were planted in three-tree row plots. One year later 143, crossbred seedlings had died, resulting in one missing plot; but, 232 selfbred seedlings had died, resulting in 17 missing plots. Analytical problems caused by missing plots increase the costs and decrease the efficiency of statistical analyses.

Table 2.--Family means, coefficients of variation and correlation coefficients after self- and cross-pollination, for seed and seedling traits

Trait	Means		Coefficients of variation		Correlation[a] coefficients
	Self	Cross	Self	Cross	Self x Cross
Filled seed (percent)	21.7	76.5	60.8	20.2	0.02
Seed weight (mg)	22.7	23.3	27.4	27.9	.69
Germination (percent)	73.1	84.9	2.3	0.8	.29
Germination rate (days to germinate)	6.53	6.31	16.1	16.7	.57
Hypocotyl height (mm)	37.7	40.7	15.8	9.5	.63
6-month height (cm)	23.8	27.3	12.7	7.2	.84
Survival at 6 weeks (percent)	97.1	98.6	5.0	2.4	.15
2-year height[b] (feet)	1.07	1.23	23.3	21.7	.48
Survival at 2 years[b] (percent)	65.6	78.8	42.5	31.3	.27

[a]First seven correlations estimated with 51 degrees of freedom; the last two estimated with 44 degrees of freedom.

[b]One year after outplanting as 1-0 stock.

Another disadvantage of selfed families was that they generally had higher coefficients of variation than crossed families (table 2). Compared to crossbreds, inbreds seemed to be hypersensitive to environmental stresses. This is also an important consideration in experimental design, because greater variation among experimental units necessitates larger experiments to achieve comparable precision for estimates, other factors being equal.

The use of inbred material presents some difficult and potentially expensive problems in seedling production, artificial selection, and experimental design. The question is whether the benefits derived from inbred material justify its use in preference to crossbred material despite the disadvantages discussed above.

USES OF INBRED MATERIAL

Production of Base Populations for Selection

The success of corn breeders using the inbreeding-outcrossing hybrid method has always impressed tree breeders. As early as 1929, Kolesnikov suggested that pure line breeding would be a good approach for forest tree improvement. Even today, as the corn breeders are turning more and more to outcrossing, recurrent selection methods (Duclos and Crane 1968), some tree breeders still advocate inbreeding programs for tree improvement (Righter 1960; Dieckert 1964b; Pawsey 1964; Keiding 1968). "On the basis of present information, hybridization programs were initiated in maize before there was genetic need for such programs. . . . In the 1920's, when the current hybridization programs were initiated, selection was thought to be ineffective. Current information indicates that selection, properly performed, is still an efficient tool. The results with maize, therefore, do not constitute an adequate justification for a hybridization program with other crops" (Sprague 1966, page 336).

The biological and economic inferences most strongly supported by the results with controlled inbreeding in loblolly pine indicate that crossbreeding should be favored over inbreeding in production of base populations for selection. The same conclusion has been reported for slash pine (Snyder 1968). Costs in time and capital outlay would necessarily be greater and gains would be more slowly achieved with inbreeding than with crossbreeding programs. Sacrifices in long-term genetic flexibility are inherent in an inbreeding-outcrossing hybrid method because, once the hybrid is obtained, the breeder must start over again each time to produce a new variety for a newly discovered need. This lack of flexibility is particularly disadvantageous with a plant such as loblolly pine because generation intervals are so long. Crossbreeding programs, such as reciprocal recurrent selection, offer long term flexibility in selection of several strains simultaneously and show promise of exceeding the gains possible with inbreeding (Penny 1968). To really profit from the experience of corn breeders, outbreeding methods should be used in loblolly pine. By so doing, we may be able to avoid the tree breeders' version of--

The Shattered Dream of a Corn Breeder

Selfed a hundred corn plants,
Put each in a cross;
Selfing without testing,
Means a heavy loss.

Looked around the country,
Found a fertile field,
Used a ten-ten lattice
To find out how they'd yield.

Analyzed the variance,
Wanted just the best;
Planted only thirty,
Threw away the rest

Thirty, good in hybrids,
That would be a plenty;
Heavy rains, and lodging;
Then there were twenty.

Still had twenty inbreds
Looking mighty keen;
Hot, humid weather;
Smut left thirteen.

Lucky thirteen inbreds,
Glad to be alive;
Wilt, blight, and aphids;
Then there were five.

So passed the summer,
Full of sweat and tears;
Came then the harvest--
Four had rotted ears.

One sturdy inbred,
All, all alone;
It has no sex appeal
Can't find a home.

Frederick D. Richey, Knoxville, Tenn.

Briggs, F. M. , and Knowles, P. L. Introduction to plant breeding. Pp. 237-238. New York: Reinhold Publishing Corp. 1967.

Progeny Testing

In loblolly pine there are moderately high correlations between self-combining ability and cross- (1. e. , general) combining ability in most traits, particularly height growth (table 2). Similar correlations for several traits have been reported in other species (Dieckert 1964a; Fowler 1965; Bingham 1966; Nikles 1966; Snyder 1968). These results have led to the suggestion that selfing might be used as a method of progeny testing. With this method, candidate trees would be ranked according to the performance of their offspring resulting from self-fertilization. The assumption is that ranking based on selfing would be similar to the ranking based on some other form of progeny testing, such as using a pollen mix or a tester system.

In progeny testing, the ideal is to have the progeny performances in dicate perfectly the breeding value of the tested parent trees. A disadvann tage of crossbreeding systems is that the breeder must arbitrarily choose a limited number of tester trees or sources of pollen for mixes. He must then assume that the sample will reliably represent the average performance of parent trees under seed orchard or other seed production conditions. Progeny testing using selfbred progeny would eliminate the need of an arbitrary choice of test pollen and would thereby standardize the testing procedure to some extent. An experimental comparison of crossing and selfing as progeny testing methods was obtained from height data from 51 selfbred and crossbred families (Franklin 1968). There was a fairly strong overall correlation between the ranked array of families produced by crossing with a five-tree pollen mix, and the corresponding families produced by selfing (figure 1). Nevertheless, the best 10 parent trees, chosen on the basis of self-combining ability, included only six of the parent trees indicated to be best on the basis of cross-combining ability.

Although it may be possible to use inbred offspring for progeny testing, the overwhelming evidence is that alternative methods will be more efficient. For example, some candidate trees could never be progeny tested using selfed offspring because they would never yield viable seed when selfed. In addition to requiring seven times more cones for equivalent yields of 6-month-old seedlings (table 1), larger coefficients of variation (table 2) will necessitate larger experiments to achieve equivalent precision on estimates. Added to these problems is the apparent hypersensitivity of selfs to environmental stress which increases mortality.

Progeny tests based on self-pollinated families will give essentially the same information as tests with corresponding cross-pollinated families, i. e., general combining abilities. Therefore, use of selfs for progeny testing has no important advantage, but does have some significant disadvantages, and should not be considered as a general method of progeny testing loblolly pine.

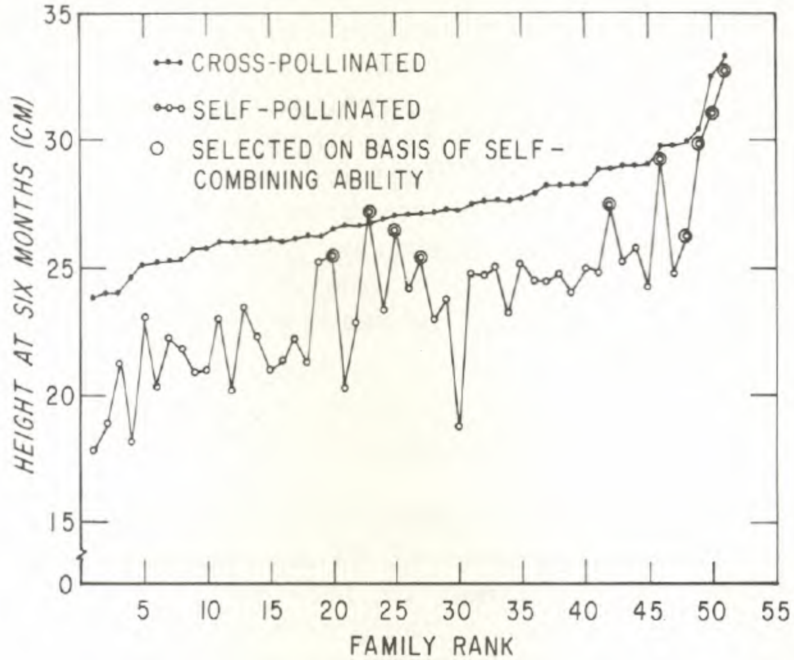

Figure 1.--Correlation between family mean heights based on self- and cross-pollinations of 51 parent trees (cross pollen was a five-tree mix).

Estimation of Genetic Components of Variance

Selection indices will be powerful tools for tree improvement in the near future. To obtain efficient indices, precise estimates of genetic components of variance and covariance are needed. Estimation of these components requires large amounts of control-pollinated material suitable for use in relatively refined statistical designs. Results with controlled inbreeding in loblolly pine prove it to be an inefficient method of obtaining large quantities of such material. Adequate mating designs based entirely on outcrossing are available for variance component estimation (Stonecypher 1966). Therefore, it is neither necessary nor efficient to try to use inbreeding for estimation of genetic components of variance in loblolly pine, except in the rare instance when the desired information can be obtained in no other way.

SUMMARY

The utility of inbreeding as a means of tree improvement has long been a topic for debate. Inbreeding has been suggested as a method of progeny testing, as a method of producing base populations from which to make se-

lections, and as a means of estimating components of genetic variance. Recent results for loblolly pine showed that seedling yields per cone harvested after selfing were only one-seventh as great as those after crossing. Seedling mortality also increased significantly as a result of selfing. Increased mortality after selfing poses difficult problems in experimental design and interpretation under the rather severe environmental conditions of forest testing. Consequently, inbred seedlings are more difficult and more expensive to produce and to use than are crossbred seedlings. With respect to tree improvement methods and procedures, information and materials obtained by inbreeding can usually be obtained at a much lower cost by crossbreeding. Therefore, the utility of inbreeding for genetic improvement of loblolly pine is quite limited.

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