WIDE CROSSES IN THE SOUTHERN PINES

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Abstract.--Previous efforts to produce hybrids between members of the genus Pinus have resulted in a "set of rules" as to what can and cannot be crossed. Disregarding these rules, all possible crosses were attempted, especially those with the southern pines. The available seed were planted, and morphological characters prevalent from time of planting through one year's growth were measured.

The objective of this paper is to report on early performance of the putative hybrids compared to parental species. No claim of hybridity is made; however, intermediacy of the crosses compared to the parent species is suggested to be an indication of hybridity in some implausible crosses. Further assessment of hybridity will take place in the field plantings. Performance and putative hybridity is commented on by cross, followed by a brief summary.

Additional keywords: Interspecific,putative hybrids

The possibility of making hybrids amongst members of the genus Pinus has intrigued researchers for many years and has been explored more fully than in most genera of forest trees (Critchfield, 1975). The effort expended in this activity has resulted in a "set of rules" as to what can be crossed and what cannot. How these rules are applied depends on which of the several authorities on nomenclature one follows. The more common of these are listed in Table 1 (page 17) of Dorman (1976); others are listed in Duffield (1952) and Little and Critchfield (1969). An excellent listing of the species involved by group or section is given in Mirov (1967), starting on page 521. We are following Duffield's (1951) classification which divides the subgenus Diploxylon into groups. 2/

The general rule is that crossing among groups generally cannot be done; this is expressed by Critchfield (1975), "crossing is usually impossible among the 15 groups of species (subsections) currently recognized as making up the genus Pinus." There are some exceptions in which intergroup crosses were successful, as listed in Table 1 of Critchfield (1975); only two of these represent southern pines. These crosses are P. elliottii x P. clausa (Saylor and Koenig, 1967) and P. taeda x P. clausa (Critchfield, 1963). As a result of the rule

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2/ These are equivalent to subsections.

and the few successful crosses between groups, many crosses have not been tried and even scoffed at as "ridiculous to try" by some biologists.

In 1959 Hoerner-Waldorf started a pine arboretum in the Coastal Plain of North Carolina. Through the years, 80 species and 10 varieties were planted; of these, 50 survived. Many of these grew well enough to produce abundant flowers, some at an early age.

At that time the question arose as to the feasibility of making selective crosses among the flowering species. The decision was made to ignore the "rules," to throw away the book and try all possible crosses, especially those with any of the southern pines. Such crossing has been pursued over a number of years; in 1974 and 1975 the seed available were planted in the greenhouse, then moved into field plantings at Hoerner-Waldorf's Research Center at Tillery, North Carolina.

The objective of this paper is to report on early performance of the putative hybrids in the greenhouse, compared to parental species. No claim is made that the crosses are truly hybrids; final determination will be made after the trees planted in the field are older, based upon morphological, anatomical, flowering and isozyme analyses. The present paper will point out intermediacy of the crosses grown in the greenhouse as an indication of hybridity in some implausable crosses not expected to be successful based upon the rules.

MATERIALS AND METHODS

This study of parents and putative across-group pine hybrids was a joint effort of the Pine Cooperative at North Carolina State University and Hoerner-Waldorf Corporation of Roanoke Rapids, North Carolina. The crosses in the arboretum were made by Ray Brown at Tillery, North Carolina. The origin for each parent species is shown in Table 1. Species verification is not absolute and the nomenclature supplied by the seed supplier was accepted unless there was an obvious error or suspicion of mislabeling. As far as we know, each species was represented by a number of parents. Sixteen trees of each species were planted in a block in the arboretum. Most crosses made involved several female parents and a pollen mix from several parents, except when flowering was limited to one or two trees within the species block. A few of the seed lots were extracted at Raleigh, North Carolina, but most were extracted at Tillery and sent to North Carolina State University for planting in the greenhouse. When large enough, the trees were taken to Tillery for establishment in the hybrid plantation.

The seed from each lot were subjected to a 30-day stratification. They were then treated with arasan and planted in small peat pots in the greenhouse at the Genetics Nursery at North Carolina State University. A soil mixture of two parts humus soil, one part sand, and one part vermiculite was used as the planting medium. A light layer of fine sand was spread over the pots and a periodic mist begun at time of seeding. The temperature in the greenhouse was kept in the 70° - 80° (F.) range.

A small number of seedlings was lost to damping off at an early age. Several applications of 50 percent Captan in water solution were made to control the disease which was prevalent because of excess moisture. There was

Table 1.--Origin of the parents used to produce the putative hybrids

Species	Common name	Group[a/]	Origin
P. echinata	shortleaf pine	Australes	Widespread in southeastern U. S.
P. pungens	table-mountain pine	Australes	Mountains of southeastern U. S.
P. rigida	pitch pine	Australes	Eastern U. S. highlands in its southern range
P. serotina	pond pine	Australes	Southeast U. S., coastal sites
P. taeda	loblolly pine	Australes	Widespread in southeastern U. S.
P. banksiana	jack pine	Contortae	Northeast U. S. & Canada
P. clausa	sand pine	Contortae	Florida and southern Alabama
P. virginiana	Virginia pine	Contortae	Southeastern U. S., primarily in uplands in its southern range
P. pinaster	maritime pine	Sylvestres	Southwest Europe and northwest Africa
P. thunbergii[b/]	Japanese black pine	Sylvestres	Japan and Korea
P. densiflora	Japanese red pine	Sylvestres	Japan

[a/] Group and subsection are used interchangeably.
[b/] This species has recently been renamed P. thunbergiana.

some wilting such as noted by Franklin (1969) in which the cotyledons wilted and then the seedling died two to three weeks after germination. This may have been caused by the conditions in the greenhouse or possibly by some physiological breakdown which prevented water uptake and further development. 3/

One trend noted in putative hybrids of both plantings was the tendency for the seed coats to adhere to the cotyledons. When not shed naturally, the seedlings often died. When the seed coats were removed manually, the cotyledons were twisted and misshapen but usually recovered and appeared to start normal growth within a few days.

The morphological characteristics examined were those that are evident from the time the seeds are planted until the end of the first growing season. Germination speed and characteristics were recorded daily, as was secondary needle formation later in the year.

3/ Personal communication, Dr. R. C. Kellison, N. C. State University.

The number of cotyledons and length of the hypocotyl were recorded periodically to determine number, rate of development and other characteristics. Height measurements were taken for the mean for each parent or putative cross after one growing season (approximately six months). Height was accurately measured from the soil surface to the tip of the apical bud and appears to be a most useful indicator of hybridity. Hypocotyl color differences were great and always observed on the same number of days after germination for parents and putative crosses; this was a subjective measurement, however, and its **use**[f]ulness as an indicator of hybridity could sometimes be questioned.

Many crosses were attempted with each parent species in the arboretum. The 29 putative hybrids with sufficient seed to plant are listed in Table 2 with the groups they represent. As there are seven intragroup crosses and 22 intergroup crosses; only P. taeda x P. clausa (Critchfield, 1963) has been reported as being successful. The only other successful intergroup cross reported is P. elliottii x P. clausa (Saylor and Koenig, 1967).

RESULTS AND CONCLUSIONS

No claim of hybridity can be made solely on greenhouse tests, In most cases, however, the putative cross exhibits morphological characteristics of the seedlings that are distinctly different from seedlings of either parent species. These differences, which are often intermediate, can be seen from Table 2 by comparing each putative cross with its respective parents. Outstanding examples of differences or intermediacy are commented upon in the following paragraphs with comments about some unusual features observed. Space does not enable discussion of every cross.

The strongest characteristics denoting hybridity in the intergroup cross P. thunbergii x P. pungens was the reduced average height of the cross after one growing season. The lesser height may indicate physiological incompatibility between P. pungens (group Australes) and P. thunbergii (group Sylvestres). The rate of secondary needle formation was similar to the female parent P. thunbergii.

The intergroup cross P. taeda x P. pinaster showed some characteristics different from the parents. The average height of the cross was less than either parent, and the time required for germination and secondary needle formation was much longer than for both parent species. The reciprocal cross P. pinaster x P. taeda showed similar tendencies as P. taeda x P. pinaster but seedlings exhibited a strong reduction in chlorophyll content, having nearly white needles and light pink hypocotyls. This was retained after outplanting.

P. clausa x P. virginiana was intermediate in most characteristics measured. Most outstanding was the intermediate height obtained from a considerable number of seedlings of both parents and the cross. No abnormal seedlings were noted. The reciprocal cross P. virginiana x P. clausa was intermediate in some measurements but heights were similar and hypocotyl color was intermediate. Based on greenhouse observations, both of these within-group crosses appear to be good candidates for true hybrids.

Table 2.--Comparison of physical characteristics of parents and crosses[2/]

Cross and parents	Number seed planted	Number seed germ.	Number cotyledons	Color of hypocotyl[c/]	Length of hypocotyl (cm)	Avg. Ht. (1 growing season) (cm)	Secondary needle formation (days)	Groups crossed d/
				1974 Planting				
P. clausa	50	34	5 - 8	lt. red	1.6 - 3.2	14.4	69	
P. clausa x P. virginiana	50	24	5 - 7	lt. red	1.2 - 4.2	8.2	92	C x C
P. virginiana	50	37	5 - 7	lt. red	2.0 - 3.0	6.1	97	
P. pinaster	50	30	6 - 9	lt. purple	2.0 - 2.8	15.2	62	
P. pinaster x P. echinata	50	35	7 - 10	lt. red-purple	1.3 - 3.0	12.9	75	S x A
P. echinata	50	35	5 - 7	red	0.7 - 2.0	7.9	83	
P. pinaster x P. pungens	50	37	7 - 10	deep purple	2.0 - 3.3	17.2	64	S x A
P. pungens	50	13	7 - 8	lt. red	2.0 - 2.8	6.8	83	
P. pinaster x P. serotina	50	21	6 - 9	deep purple	2.0 - 3.3	13.3	82	S x A
P. serotina	50	42	5 - 8	pink	0.5 - 2.1	14.4	76	
P. taeda	50	33	6 - 8	red	2.5 - 4.3	15.8	70	
P. taeda x P. pinaster	50	6	7	red	3.0 - 4.8	11.8	85	A x S
P. taeda x P. thunbergii	50	1	8	red	3.8	9.0	68	A x S
P. thunbergii	50	27	6 - 8	pale red	1.5 - 2.5	5.7	65	
P. taeda x P. virginiana	50	19	6 - 8	red	2.5 - 4.1	12.2	89	A x C

Cross and parents	Number seed planted	Number seed germ.	Number cotyledons	Color of hypocotyl [c/]	Length of hypocotyl (cm)	Avg. Ht. (1 growing season) (cm)	Secondary needle formation (days)	Groups crossed [d/]
				1974 Planting				
P. thunbergii x P. clausa	50	22	5 - 8	lt. green	1.7 - 3.0	8.8	60	S x C
P. thunbergii x P. pungens	50	30	7 - 9	lt. green	2.0 - 3.0	4.9	65	S x A
P. thunbergii x P. serotina	50	27	5 - 8	pink	1.3 - 2.8	7.2	68	S x A
P. thunbergii x P. taeda	50	12	6 - 8	green	1.5 - 2.7	5.0	76	S x A
P. taeda x P. clausa	50	0						A x C
				1975 Planting				
P. pinaster x P. taeda	27	4	7	red	[a/]	10.8	128+ [b/]	S x A
P. rigida	75	58	5 - 7	lt. red	2.5 - 3.2			
P. rigida x P. clausa	17	8	5 - 7	lt. red	1.1 - 2.5	8.8	99	A x C
P. taeda x P. banksiana	36	4	4 - 6	[a/]	0.2 - 1.0	3.2	128+ [b/]	A x C
P. taeda x P. clausa	49	3	6 - 8	red	1.7 - 3.8	7.8	128+ [b/]	A x C
P. taeda x P. serotina	100	85	4 - 7	pale red	0.5 - 2.3	10.4	128	A x A
P. taeda x P. virginiana	100	78	5 - 8	purple	1.8 - 5.0	14.3	109	A x C
P. thunbergii x P. rigida	33	5	6 - 8	pink	2.3 - 3.0	[a/]	91	S x A

Cross and parents	Number seed planted	Number seed germ.	Number cotyledons	Color of hypocotyl[c/]	Length of hypocotyl (cm)	Avg. Ht. (1 growing season)(cm)	Secondary needle formation	Groups crossed [d/]
				1975 Planting				
P. virginiana x *P. banksiana*	100	58	5 - 7	red-orange	2.0 - 3.5	5.2	128+[b/]	C x C
P. virginiana x *P. clausa*	100	73	4 - 7	pale red	1.0 - 2.7	5.7	109	C x C
P. virginiana x *P. echinata*	36	4	5 - 7	pale red	1.5 - 3.0	4.5	128+[b/]	C x A
P. virginiana x *P. pinaster*	32	5	5 - 8	red	2.0 - 3.0	7.4	128	C x S

The following crosses did not produce any seedlings:

	Number of seed	Groups crossed
P. echinata x *P. banksiana*	31	A x C
P. echinata x *P. pinaster*	45	A x S
P. densiflora x *P. rigida*	43	S x A
P. densiflora x *P. taeda*	30	S x A

[a/] Value not obtained

[b/] Approximate value

[c/] This was a subjective measurement.

[d/] Group designations: A = Australes, C = Contortae, S = Sylvestres.

The unexpected intergroup cross P. thunbergii x P. taeda shows many of the characteristics of the parent P. thunbergii with the exception of the time required for secondary needle formation, which was a little longer than for either parent. Because of the resemblance to the female parent, the hybridity of this cross is rather doubtful. However, the reciprocal cross P. taeda x P. thunbergii, of which only one of 50 seeds germinated, showed intermediacy in most measurements. One seedling is insufficient to make any statement of hybridity even if it has intermediate characteristics.

The intergroup cross P. thunbergii x P. serotina was intermediate in average height and somewhat so for time required for secondary needle formation. One dwarf seedling was noted that had a thickened hypocotyl and never developed primary needles; it was outplanted when approximately five centimeters in height. Characteristics of seedlings and fairly good seed germination show definite indications of hybridity.

P. pinaster x P. pungens, an intergroup cross, appeared to have characteristics different from the parents; it had much the same form as P. pinaster. The average height of the cross is somewhat misleading because some of the seedlings of the cross were as much as fifteen centimeters taller than the parent P. pinaster. These tall seedlings produced primary needles along approximately three-fourths of the stem, had good straight form, and were vastly superior to the P. pungens seedlings. This could well be a hybrid. Percent germination was much better than both. parents.

The intergroup cross P. thunbergii x P. clausa was intermediate in height between the parents and the color of the hypocotyl was very different from the parents; secondary needle formation was faster than for either parent. We feel this is a good cross.

The intergroup cross P. taeda x P. virginiana was established in both years' plantings. It exhibited some intermediate and some characteristics of each parent in both tests. Hybridity at this stage is uncertain but there are enough characteristics like the pollen parent that hybridity may be present. There were four abnormal seedlings which had short, hard, thickened hypocotyls and did not appear to produce any new growth after the primary needles were formed. If confirmed, this putative hybrid will be of special interest since the P. taeda x P. virginiana cross has been tried by several persons without apparent success.

Except for cotyledon number the intergroup cross P. pinaster x P. echinata has intermediate characteristics in color of the hypocotyl, average height and time required for secondary needle formation. This appears to be most positive of the many putative hybrids grown; even percent seed germination was good. No seed germinated from the reciprocal P. echinata x P. pinaster. The significance of this is mentioned later.

P. pinaster x P. serotina, another intergroup cross, appears to be a hybrid; at least its characteristics were sometimes much different from P. pinaster. The average height was less than for either parent, and the time required for secondary needle formation was considerably longer. No abnormal seedlings were noted but percent of germination was quite good.

The cross P. virginiana x P. banksiana showed a marked contrast from either parent. Intermediacy was evident in every measurement except secondary needle formation. The time for secondary needle formation was much longer than for either parent. This is a good candidate for a hybrid as an intragroup cross.

P. taeda x P. serotina looks very much like the male parent. All measurements are similar to the P. serotina, indicating hybridity. This hybrid would be expected but the direction toward the male, rather than intermediacy, was a surprise.

The intergroup cross P. virginiana x P. echinata has some characteristics of P. echinata but has depressed height growth. The lack of clear-cut differences or intermediacy makes it difficult to assess the hybridity of this cross.

The cross P. taeda x P. banksiana responded as crosses between groups often do. Only four seed germinated from the 100 that were planted; secondary needle formation was very slow on the survivors, and the length of the hypocotyl and the average height were less than for either parent. The small number of seedlings does not allow a statement of hybridity but it appears from the characteristics of the four seedlings that this is a hybrid.

An historically unsuccessful intergroup pine cross, P. taeda x P. clausa, has produced only seedlings that died soon after germination (Critchfield, 1963). The first planting of this cross also produced no seedlings, but the second planting produced three seedlings from 49 seed, including one albino. There appears to be considerable intermediacy and deviation from either parent. These three seedlings were field planted; the albino seedling acquired some green pigmentation in the field planting.

P. virginiana x P. pinaster may be a successful intergroup cross. The strongest characteristic denoting hybridity was its intermediate height; also it varied greatly in time required for secondary needle formation. Some of the traits of P. pinaster are seen in the putative cross. The number of cotyledons and much longer time required to produce secondary needles reinforce the suspicion of hybridity.

The intergroup crosses P. rigida x P. clausa and P. thunbergii x P. rigida appear to be successful hybrids; however, there was insufficient information obtained to come to a conclusion. The color of the hypocotyl was intermediate in both crosses, but values for other characteristics are not very informative.

There were five intergroup crosses for which seed did not germinate. P. echinata x P. banksiana, P. echinata x P. pinaster, P. densiflora x P. taeda, and P. densiflora x P. rigida yielded no seedlings. P. taeda x P. clausa yielded no seedlings in the first planting but three seeds germinated in the second planting. This does not prove success or failure but nongermination has been unusual in many crosses between different groups.

In his new book, Dorman (1976) has a section outlining hybridity among taxonomic groups of pines, with emphasis on the southern pines. A cross of P. silvestris x P. palustris has been reported but is considered to be doubtful. Similarly, Dorman states that a P. taeda x P. densiflora cross in 1950

is suspect. Crosses of P. contorta x P. pungens and P. densiflora x P. **rigida** have been made but hybridity is as yet unproved. The hybrid of P. **rigida x** P. radiata has been made and reported numerous times in Korea.

Of the intragroup crosses made, most appeared to be hybrids; of the 20 intergroup crosses, 12 appear to be good candidates, seven are doubtful but differ from the parents, and one appears to be a noncross. It will be very interesting to confirm or reject these findings after the trees have been grown about five years in the field and a multidiscipline attack to hybridity has been made.

SUMMARY

Many seemingly impossible crosses were made on pines in the Hoerner-Waldorf arboretum at Tillery, North Carolina. Some of these crosses produced seed. These seed were planted in the greenhouse where many measurements and observations were made. No claim of hybridity is made, based on these greenhouse tests; however, several intergroup crosses are offered as putative hybrids based on physical measurements and observations.

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