Infection of Shortleaf × Loblolly Pine Hybrids Inoculated with *Cronartium quercuum* f. sp. *echinatae* and *C. quercuum* f. sp. *fusiforme*

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ABSTRACT

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In this artificial inoculation study interspecific hybrids of shortleaf pine (*Pinus echinata*) × loblolly pine (*P. taeda*) were no more resistant to infection by the form of fusiform rust fungus (*Cronartium quercuum* f. sp. *echinatae*) that infects shortleaf pine than were seed orchard shortleaf pine. As expected, however, they were much more resistant to infection by the form of the rust fungus (*C. quercuum* f. sp. *fusiforme*) that commonly attacks loblolly pine.

Hybrids of the interspecific cross of shortleaf pine (*Pinus echinata* Mill.) × loblolly pine (*Pinus taeda* L.) generally are more resistant to the form of the fusiform rust fungus that attacks loblolly pine (*Cronartium quercuum* (Berk.) Miyabe ex Shirai f. sp. *fusiforme*, hereafter abbreviated to *Cqf*) than the highly susceptible loblolly pine parent (6). Backcrosses of hybrid F_2s to the loblolly pine parent have generally shown excellent growth with high levels of rust resistance (6). Interest is increasing in the use of loblolly pine strains with fusiform rust resistance transferred from shortleaf pine for planting where incidence of this disease is high, but many questions remain to be answered.

One question concerns the level of resistance of the hybrid to infection by *C. quercuum* (Berk.) Miyabe ex Shirai f. sp. *echinatae* (hereafter abbreviated to *Cqe*) the form of the rust that commonly attacks shortleaf pine (1). Although this question will not be answered until the hybrid can be field tested where *Cqe* is prevalent, results described here indicate that the hybrid will be no more resistant to *Cqe* than shortleaf pine from seed orchards of trees selected for good form and growth rate. Results also reaffirm the relatively high resistance of the hybrid to infection by *Cqf*.

MATERIALS AND METHODS

The objective of this study was to test the hypothesis that there is no difference in resistance of shortleaf pine, loblolly pine, and different types of shortleaf × loblolly pine hybrids to infection by the two *formae speciales* of *C. quercuum*. Since work with this interspecific hybrid is still in its early stages we were most interested in using test material with theoretically different levels of hybridization (backcrosses are 75% loblolly, F_1s and F_3s are 50% loblolly) involving as many different parents as possible, and the hybrid generations were considered to be at the species level for comparisons.

A total of 21 seedlots were used in the study: six backcrosses (loblolly pine × F_2 hybrids), six F_1 hybrids (loblolly pine × shortleaf pine), six F_3 hybrids (wind-pollinated seed from F_2), two loblolly pine check lots, and one shortleaf pine check lot.

One of the loblolly pine check lots was a wind-pollinated seed orchard collection from Georgia; the other was a wind-pollinated collection from trees known to be susceptible to *Cqf*. The shortleaf pine check lot was a wind-pollinated collection from a Georgia seed orchard of clones selected by the Tennessee Valley Authority (TVA).

After stratification for 2 mo, seeds were germinated and transplanted into flats containing 20 seedlings each. At 4 wk of age the seedlings were inoculated by passing the flats under an aqueous spray of basidiospores (5.0×10^4 spores per milliliter in 8-ml aliquots) as specified for the concentrated basidiospore spray system of Matthews and Rowan (9). Six flats of 20 seedlings each for each of the 21 seedlots were exposed to basidiospores of *Cqe* derived from a mixture of aeciospores from 10 galls on shortleaf pine in Stone County, MS. Equal numbers of seedlings were exposed to *Cqf* basidiospores derived from a 10-gall mixture of aeciospores from loblolly pine in Clarke County, GA. Flats were inoculated in random order. Immediately after inoculation, the seedlings were placed in a mist chamber and held at 21 C for 24 hr,

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grown in the greenhouse for 9 mo, and those with visible galls were counted. Rust infection data were transformed to the arc sin square root percentage of rustfree seedlings, and a variety of standard statistical analyses were made.

RESULTS

After inoculation with *Cqe*, 86% of the loblolly pine controls and 81% of the shortleaf pine controls remained rustfree. Average percentages of rustfree seedlings were somewhat lower for the hybrids and hybrid backcrosses, but only the average for the F_3 hybrids was significantly lower than those of the controls (Table 1).

An average of 28% of the loblolly pine controls and 93% of the shortleaf pine controls remained rustfree after inoculation with *Cqf*. The backcrosses had about as much infection as the loblolly pine controls. Percentages of rustfree F_1 and F_3 hybrids ranged widely (41–99%), but these hybrids were significantly more resistant than the loblolly pine controls (Table 1). In general, galls produced by *Cqe* were globose while those produced by *Cqf* were fusoid (Fig. 1).

There was a highly significant interaction of seedlot types × inoculum sources caused primarily by the higher resistance of the loblolly pine and backcross seedlots to infection by *Cqe* ($P = 0.01$). There was also a highly significant interaction among seedlots of the six backcrosses ($P = 0.01$), indicating that backcrosses resistant to *Cqf* are likely to be susceptible to *Cqe*, and vice versa.

There was no significant difference between the F_1 and the F_3 hybrid types in infection by either *Cqe* or *Cqf*. Approximately 70% of the seedlings of both hybrid types remained free of infection from each spore type (Table 1). Among individual F_1 seedlots, there were significant differences in susceptibility to both spore types ($P = 0.01$), but no significant interaction was found. Among individual F_3 seedlots, however, there was a significant interaction ($P = 0.01$), indicating that F_3 seedlots resistant to *Cqf* may be susceptible to *Cqe*.

The F_3s were significantly ($P = 0.01$) more susceptible than the loblolly pine controls to infection by *Cqe*. These F_3s were not made by control pollination, and the F_2 stand is in an area exposed to pollen from surrounding loblolly and shortleaf pine. Since the flowering time of the F_2 hybrid more nearly coincides with shortleaf pine flowering, the higher susceptibility of these seedlots could be due to backcrossing to wild shortleaf pine.

The F_3s were significantly more susceptible to *Cqe* than were seedlings from the shortleaf pine seedlot. This difference was not expected, but could be due to the shortleaf pine seedlot having come from trees selected by TVA for use in seed orchards. Presumably one criterion for selection was absence of galls caused by *Cqe*. The relatively low resistance of the backcrosses to *Cqf* is in contrast to previous results (7,8). Additional studies of the resistance of this backcross type are needed.

DISCUSSION

Overall, the resistance of the shortleaf pine hybrids to infection by *Cqe* is encouraging. Only two of the hybrids were significantly less resistant ($P = 0.05$) than an open-pollinated collection from a shortleaf pine seed orchard. If the hybrids are faster growing in field tests than pure shortleaf pine they have potential for use on some shortleaf pine sites which are presently being planted to loblolly pine.

The significant interaction in the F_3s is probably a reflection of the wide range of flowering times in the F_2 stand. As mentioned earlier, the flowering time of most F_2s coincides with shortleaf pine, but some F_2 trees are probably backcrossing with loblolly pine.

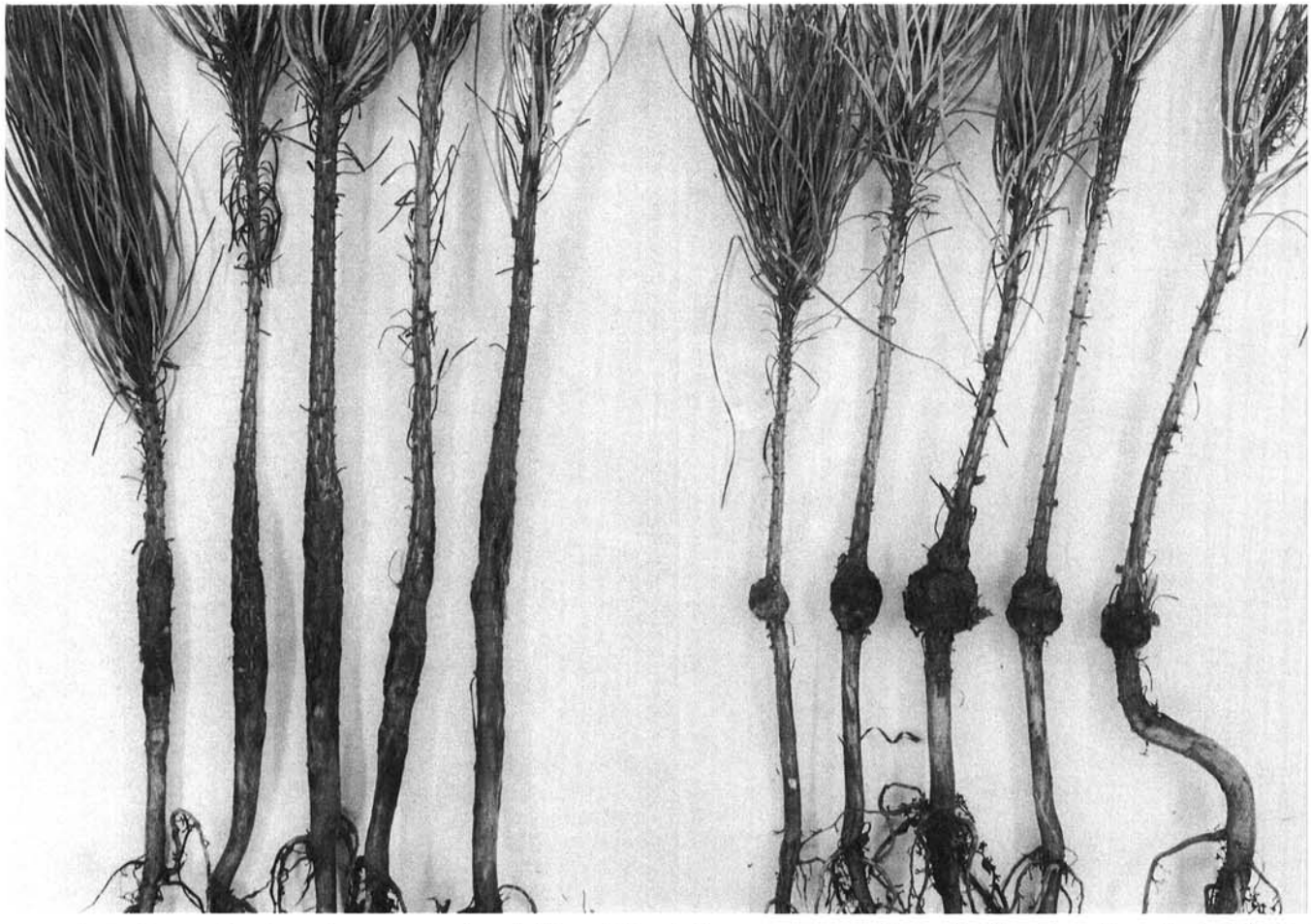

Fig. 1. Typical galls caused by *Cronartium quercuum* f. sp. *fusiforme* (left) and *C. quercuum* f. sp. *echinatae* (right) on F_3 seedlings of a single family of shortleaf × loblolly pine hybrids.

TABLE 1. Percent rustfree pine seedlings 9 mo after artificial inoculation with *Cronartium quercuum* f. sp. *echinatae* and *C. quercuum* f. sp. *fusiforme*

Seedlots	Inoculum	
	f. sp. *echinatae*	f. sp. *fusiforme*
Backcrosses		
1	98.7	21.6
2	76.3	41.0
3	65.4	33.7
4	62.9	21.0
5	62.7	48.8
6	···	32.1
Means	73.2 abc[z]	33.0 c
F_1s		
7	96.2	99.2
8	84.9	83.6
9	79.0	98.4
10	64.5	69.1
11	61.1	49.6
12	50.0	55.9
Means	72.6 abc	76.0 ab
F_3s		
13	84.0	41.2
14	77.8	70.8
15	67.1	88.1
16	63.8	82.7
17	59.8	58.2
18	56.7	92.6
Means	68.2 c	72.3 b
Loblolly controls		
19	85.7	26.0
20	85.3	30.5
Means	85.5 a	28.2 c
Shortleaf control		
21	80.8 ab	92.8 a

[z]In a column, means not sharing a common letter are significantly different $P = 0.05$, according to a one-way ANOVA.

At the same time it is obvious that the susceptibility of the hybrids to both *formae speciales* could function as a factor in the development of races of *Cqf* with potential for increased virulence to shortleaf pine. Since natural hybridization between the two pines already occurs in varying frequency throughout their common range, the development of such virulent races has probably taken place in the past, and presumably will continue. The possibility that such races may develop is being studied in a separate test using spores collected from infected artificial pine hybrids to inoculate a wide range of shortleaf pine seed sources.

With the exception of the backcrosses, the relative resistance of shortleaf × loblolly pine hybrids to *Cqf* in this study substantiates results of previous studies (2–5,8,10,11). The backcrosses in this study used loblolly pine as the female parent while those of La Farge and Kraus (7,8) were made using loblolly pine as the male parent. These results suggest that the direction of backcrossing may be an important factor in resistance to *Cqf*. Tests of reciprocal crosses between specific trees are currently under way to clarify the situation. The F_1 and F_3 hybrids in this study had over twice as many rustfree seedlings at 9 mo as the loblolly pine check lots.

Data on hand at Macon also indicate that the hybrids may be more resistant in the field than the artificial inoculation tests indicate. Fourteen seedlots which averaged 52% rustfree following greenhouse inoculation in 1974 are averaging 71% rustfree in a 5-yr-old field planting in which the loblolly check lots are only 8% rustfree (T. La Farge and J. F. Kraus, *unpublished*).

A long-term effort by tree breeders will be required to produce substantial amounts of a strain of loblolly pine with rust resistance from shortleaf pine. All indications are that the effort will be worthwhile, and some organizations have already established seed orchards for the production of hybrid seed.

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