## Increase in Virulence of Cronartium fusiforme on Resistant Slash Pine

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#### ABSTRACT

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Cronartium fusiforme from field-infected members of a highly resistant slash pine family infected more seedlings of the same parent than did samples of *C. fusiforme* inocula

collected from the general rust population. Greater virulence of inocula from resistant trees is evidence of pathogenic specialization in *C. fusiforme*.

Additional key words: pathogenic variation, Pinus elliottii var. elliottii, Quercus nigra, disease resistance, epidemiology.

Breeding pines with genetic resistance to fusiform rust is a promising way to control the disease, but the prospect that Cronartium fusiforme Hedge. & Hunt ex Cumm, will specialize to overcome that resistance has long been recognized (1, 3, 4, 7, 8). Existing plantations of resistant loblolly (Pinus taeda L.) and slash (P. elliottii var. elliottii Engelm.) pines are small and scattered; resistant materials are just now becoming available for extensive use. The time is near, however, when C. fusiforme will be exposed to selection pressure imposed by large acreages of resistant trees and the stability of that resistance will be continually tested. Pathogenic variability in the rust, a prerequisite for specialization, has been demonstrated by differential responses of slash pines to inocula from throughout the southeastern USA (9). The potential therefore exists for shifts in virulence of the rust population.

The rate at which such shifts will occur, and whether or not they will be important in future plantations of resistant trees is unknown. This research was done to determine if inocula derived from the few field-infected trees of a highly resistant slash pine family were more virulent on seedlings from the same parent tree than were inocula representing the general rust population in that locality. Higher virulence of inocula from resistant trees would be evidence that the frequency of virulent strains in the rust population will increase when the pathogen encounters large numbers of resistant trees.

### MATERIALS AND METHODS

Aeciospores were collected in 1974 from 10 of the few available sporulating galls on open-pollinated progeny of one slash pine parent tree (Institute of Forest Genetics, Code No. 8-7). The field-infected progeny are located in two plantations: one established in 1963 near Gulfport, Mississippi, and the other established in 1964 near Bogalusa, Louisiana. Rust incidence was high both within and in the vicinity of both plantations (5). In the Mississippi plantation, only 3% of the IFG 8-7 progeny were infected as opposed to an overall mean of 45%. Comparable data for the Louisiana plantation were 5%

and 42%, respectively. To sample the general rust population (hereafter referred to as wild-type inocula), aeciospores were collected from nine galls on slash pines growing outside, but within 1.6 km of, the two field plantings. Spore collections from each gall were processed separately and used to establish 18 rust cultures on water oak (Quercus nigra L.) seedlings (Table 1). Seventeen cultures were derived from individual galls. Culture 5, however, originated from two galls on the same tree because spores were in short supply from both galls. Equal samples of aeciospores from three galls (those used individually to establish cultures 1, 2, and 6) were combined and applied to oak seedlings to produce a composite inoculum because such composites are commonly used in routine screening tests. To minimize the possibility of selection by the oak host, each rust culture was propagated on water oak seedlings from 10

TABLE 1. Gall development on seedlings of resistant slash pine parent IFG 8-7<sup>a</sup> 6 months after inoculation with *Cronartium fusiforme* from field-infected IFG 8-7 trees and with wild-type inocula

Inocula from resistant 8-7 trees		Inocula from field-run trees	
Culture no.b	Percent of plants with active galls <sup>c</sup>	Culture no.b	Percent of plants with active galls <sup>c</sup>
1	94.4	10	1.4
2	63.5	11	15.3
3	84.3	12	38.8
4	90.3	13	54.1
5	88.4	14	0.0
6	73.6	15	1.4
7	76.1	16	16.7
8	72.7	17	29.1
9	51.8	18	0.0
Mean	77.2	Mean	17.4

<sup>a</sup>Institute of Forest Genetics tree code designation.

<sup>b</sup>Rust cultures were from individual galls except culture 5, which was a composite of spores from two galls on one tree. Cultures 1 and 10 were from the Mississippi plantation; other cultures were from the Louisiana plantation.

Averages of three replications of 23-24 seedlings each.

parent trees. Inoculum for pines was mixed from leaves of five of these oak seedlings.

Progeny from the original IFG 8-7 pine parent were grown from open-pollinated seed collected in 1973. Twenty-four seedlings were inoculated with each rust culture in each of three replications; one complete replication was done each week. Pine seedlings were inoculated individually with a forced-air device (10), and spore density was maintained at 12-18 basidio-spores/mm² on a plane surface. The sequence of inoculation for inoculum source (IFG 8-7 progeny or other trees) and culture within source was determined randomly for each replication. Since 3 weeks elapsed between the first and third replication, pines were planted in sequence so that seedlings were 5-6 weeks old for all inoculations. Oaks were inoculated in a similar sequence, and 3-week-old telia were used for all pine inoculations.

Inoculated pine seedlings were grown in 10-cm diameter pots in the greenhouse and were examined for purple lesions 2 months after inoculation. The presence of galls, gall length, and seedling height were recorded after 6 months. The 6-month data were summarized as percentage of seedlings with active galls (Table 1). The distinction between active and inactive galls is necessary because resistance to *C. fusiforme* is often expressed by the formation of small galls that become inactive a few months after inoculation (9). Analysis of variance for a randomized block was made with the 6-month gall data. A single-degree-of-freedom comparison was made for the two sources of inocula; variability among inocula within sources was evaluated in separate analyses of variance.

#### RESULTS AND DISCUSSION

Only one culture representing the general rust population approached those from offspring of the resistant parent in virulence (Table 1). Two wild-type cultures (14 and 18) were avirulent, and two others (10 and 15) caused active galls on only one seedling each. Averaged over all wild-type cultures, the percentage of seedlings with active galls was only 17.4%. In contrast, active galls formed on an average of 77.2% of the seedlings inoculated with cultures from the resistant family. With a significance level of P = 0.05, this difference in virulence was determined to be at least fourfold, indicating that the frequency of specialized forms, such as culture 13, increased in response to planting resistant trees. In a related experiment, three of eight individual gall cultures collected randomly in Mississippi infected more than 30% of other IFG 8-7 progeny (9), indicating that forms of the pathogen virulent to IFG 8-7 progeny are present in other areas.

Differences among individual cultures from offspring of the IFG 8-7 parent were not significant, but substantial variability was apparent (Table 1). The range of variation approached that observed among wild-type cultures. Virulence of culture 5, a culture derived from two galls on the same tree, was similar to that of other cultures from the resistant family. The performance of the composite culture was 76.4%, which approximated the average, 77.4%, observed for the three cultures, 1, 2, and 6, from which it was derived. This finding suggests similar mixtures of inocula may be useful in future work.

These results support our previous conclusion that an

increase in frequency of virulent strains of C. fusiforme will occur if pines with one or few forms of resistance are planted extensively (6, 9). Several factors, however, should moderate the rate of increase in number of such strains. (i) Most damaging pine infections occur within the first 5 to 6 years after trees are planted and the rate at which an epidemic would progress in a particular stand would diminish when trees are over 6 years old. (ii) The pathogen is not naturally transmitted from pine to pine; 2-3 years are required to complete the cycle from pine to oak and back to pine. (iii) The uredial stage, which is capable of propagating the rust vegetatively, is formed only on oak, and more than one generation of uredia annually is rare in nature. (iv) Pines are infected by basidiospores produced following an assumed meiotic division of diploid nuclei in teliospores; thus, the genetic constitution of the organism is subject to change after each generation on pine. (v) Both slash and loblolly pines are highly heterozygous in comparison to agricultural crops, and therefore less likely to succumb to a single strain of pathogen. Because of the time factors involved for reinfection of pines and the genetic factors inherent in the pathogen and host, we do not think that epidemics will result from virulent strains of C. fusiforme. However, we can visualize an erosion of resistance in improved stock if the capacity of this organism to respond to selection pressure is not recognized. Efforts should be made to identify different types of resistance, especially types more stable than that possessed by the IFG 8-7 progeny. A better understanding of genetic interaction between host and pathogen is essential to the maintenance and utilization of genetic diversity of southern pines (2) so that excessive losses resulting from specialization in C. fusiforme can be avoided.

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# April 1976]

SNOW ET AL.: CRONARTIUM VIRULENCE INCREASE

513

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