## Pathogenicity, Host Range, and Distribution of Colletotrichum graminicola on Corn

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## ABSTRACT

All isolates of *C. graminicola* obtained from corn (*Zea mays* L.) during 1968-1971 (a total of 30 isolates from six different states) readily attacked nonwounded corn seedlings. Isolates from nine other grass genera failed to attack corn. Corn isolates attacked all members of the genus *Sorghum* tested, but failed to produce symptoms on oats, barley, wheat, tall fescue, or millet. Low light intensity and prolonged periods of high

humidity appear to be important environmental factors which increase disease severity. The ability of corn isolates to attack both corn and sorghum, including sorghum cultivars resistant to isolates of *C. graminicola* from sorghum, indicates that a race of this fungus potentially capable of causing severe damage to both plants is widely distributed in the United States.

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Anthracnose, caused by Colletotrichum graminicola (Ces.) Wils., is a common disease of cereals and grasses. In North America, the fungus has caused serious damage to rye, wheat, barley, and many other grasses (3); but on corn, anthracnose usually appears as a leaf-spotting disease restricted to the lower leaves of the plant and has been of little economic importance (2). In other countries, anthracnose has been reported to be severely damaging both as a leaf and as a stalk disease of corn (1, 4).

Isolates of *C. graminicola* from corn appear to vary greatly in both pathogenicity and host range. In 1963, Williams and Willis (10) reported that isolates associated with a local outbreak of stalk rot in Ohio caused symptoms on corn seedlings sprayed with spore suspensions, but only if the leaves were wounded before inoculation. These isolates caused no symptoms on wounded leaves of oats, barley, or wheat. In the same year, Dale (2) reported that nonwounded corn seedlings were readily attacked when sprayed with a spore suspension of a corn isolate from Arkansas. He also reported that the same corn isolate would not attack sorghum and that an isolate from sorghum would not attack corn. These results agreed with those obtained in France (4), but were in contrast with results from India

where a corn isolate readily attacked sorghum and nine other grasses (1).

During the past two yr, unusually severe outbreaks of anthracnose on dent corn have been reported from Kentucky (9) and Maryland (5). Crop failure in sweet corn caused by anthracnose has been reported from Indiana (7). In laboratory tests, an isolate of *C. graminicola* obtained from corn in Kentucky in 1969 was highly pathogenic when nonwounded corn seedlings were sprayed with spore suspensions (8). Greenhouse tests of 21 commercial hybrids and 28 inbred lines of corn showed that most were susceptible or highly susceptible, some were moderately resistant; none was highly resistant or immune (6). This report will be concerned with the pathogenic capabilities and host ranges of isolates of *C. graminicola* from corn and other members of the grass family. Some of the results have been published in abstracts (8, 9).

MATERIALS AND METHODS.— Isolates of C. graminicola were maintained on oatmeal agar slants. Inocula were prepared by flooding 30-day-old cultures with sterile water and scraping the surface of the culture with a wire loop to dislodge spores. The resulting spore suspensions were filtered through a small wad of absorbent cotton

to remove debris, and then washed three times by centrifugation and resuspension in water. Spore concentrations were determined with a hemocytometer, and one drop of Tween 40 was added to each 50 ml of spore suspension used as inoculum. Controls were sprayed with sterile distilled water containing the same amount of Tween 40.

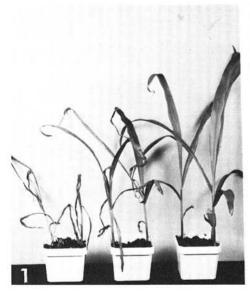
Plants for laboratory tests were grown in a controlled-environment chamber at 25 C and a 14-h light 10760 lx (1,000 ft-c) period. Unless otherwise noted, nonwounded 10- to 12-day-old seedlings at the three-leaf stage were sprayed with spore suspensions to the point of runoff. After inoculation, plants were placed in moist chambers (plastic bell jars partially lined with wet paper towels) and held under continuous low light ca. 1,076 lx (100 ft-c) for 24-72 h before they were returned to the growth chamber. Plants grown in the greenhouse were inoculated at 8-10 days of age and placed in moist chambers constructed of clear plastic for 24-72 h.

RESULTS.—In preliminary laboratory tests, 20 cultivars, which included sweet and dent corn hybrids, and dent inbred lines, were sprayed with a spore suspension (2×10<sup>5</sup> spores/ml) from an isolate of *C. graminicola* obtained from corn in 1969 (8). After inoculation, the plants were held for 72 h in moist chambers, and then returned to the growth cabinet. The range of reactions seen 5 days after inoculation is shown in Fig. 1. A sweet corn hybrid, 'Illini Chief Super Sweet', was the most susceptible and the inbred 33-16 the most resistant of the group tested. These two, rated highly susceptible and moderately resistant (respectively), plus the dent hybrid, Funk's G-5757, rated susceptible (Fig. 1) were selected for further tests.

Effects of inoculum concentration, duration of high humidity, and plant age on disease severity.—Twelve 10day-old plants of each of the three lines selected (Fig. 1) were sprayed with suspensions which contained  $3 \times 10^6$ .  $1\times10^6$ ,  $2\times10^5$ ,  $5\times10^4$ , or  $1\times10^4$  spores/ml and held in moist chambers for 72 h. Disease severity increased with increased inoculum concentration. At the highest concentration, all plants were killed, whereas at the lowest concentration only a few lesions developed on the two susceptible cultivars. A similar test in which the plants were sprayed with 2×10<sup>5</sup> spores/ml and then held in moist chambers for 24, 48, or 72 h, showed that disease severity increased with increases in exposure to high humidity following inoculation. In an attempt to simulate dew conditions which might occur in nature, plants were sprayed with  $2 \times 10^5$ spores/ml, held for 24 h in moist chambers, and then returned to the growth cabinet during daytime and replaced under moist chambers at night for the following two days. With the three lines tested, this method gave results entirely similar to those shown in Fig. 1 and unless otherwise noted was adopted as a standardized inoculation procedure in all further experiments.

The same 20 cultivars used in preliminary tests were inoculated by the standard procedure when the plants were 21-23 days old. In contrast to results with 10- to 12-day-old plants, none of these older plants was killed. Highly susceptible cultivars were severely damaged whereas moderately resistant plants had only a few lesions on the lower leaves (Fig. 2).

Pathogenicity to corn of C. graminicola isolates from various hosts.— All isolates of C. graminicola obtained from corn during the 1971 season, a total of 26 from six different states (Delaware, Georgia, Kansas, Kentucky, Mississippi, and North Carolina), readily attacked non-wounded corn seedlings inoculated by the standard procedure. Similar results were obtained with four additional cultures isolated from corn in North Carolina in 1968. In



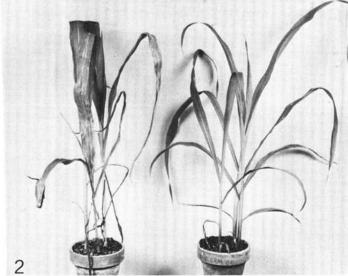


Fig. 1-2. Reactions of corn seedlings sprayed with spore suspensions of an isolate of *Colletotrichum graminicola* from corn. 1) Reactions of seedlings inoculated at 10 days of age. From the left the cultivars are 'Illini Chief, Super Sweet' (highly susceptible), 'Funk's G-5757' (susceptible), and inbred 33-16 (moderately resistant). 2) Reactions of seedlings inoculated at 23 days of age. On the left, Illini Chief Super Sweet and on the right, inbred 33-16.

TABLE 1. Host range of two isolates of Colletotrichum graminicola from corn

		Reaction to isolate from b	
Host plant	Cultivars tested a	Kentucky	Kansas
Oats (Avena sativa L.)	Aristogold, Compact, Park Dubois,		
	Victograin 48-93	0	0
Barley (Hordeum vulgare L.)	Barsoy	0	0
Wheat (Triticum aestivum L.)	Knox 62	0 -	0
Fescue (Festuca arundinacea Schreb)	Kenwell, Alta, Fawn, Ky 31, NC-1, NC-6,		
	NC-11, NC-17	0	0
Pearl Millet (Pennisetum typhoides S. & H.)	DeKalb PM604	0	0
Sorghum (S. bicolor L. Moench)	Pioneer Brand 878 c	2	1
	Pioneer Brand XB913 <sup>c</sup>	2	i
	Pioneer Brand XB935 <sup>c</sup>	3	2
	Pioneer Brand 944d	3	3
	DeKalb 931 d	3	3
	Taylor Evans Milkmaker <sup>d</sup>	3	3
	Asgrow Duet <sup>d</sup>	3	3
Sudangrass [S. sudanense (Piper) Stapf]	Monarch, Piper	3	3
Johnsongrass [S. halepense (L.) Pers.]	Seed collected near Lexington, Kentucky.	2	4

<sup>&</sup>lt;sup>a</sup>Each cultivar was tested at least twice with six to eight plants inoculated by the standard procedure in each test.

TABLE 2. Reaction of corn seedlings, grown and inoculated under various light conditions, to Colletotrichum graminicola

Cultivars	Growth chamber plants inoculated in <sup>a</sup>		Greenhouse plants inoculated in		
	Laboratory	Greenhouse	Laboratory	Greenhouse	Greenhouse (shaded)
DeKalb F880 Illini Chief	2.2	2.2	2.7	1.2	2.5
Super Sweet Average of five cultivars	4.9 3.2	2.5 2.2	4.9 4.0	2.0 1.7	2.9 2.8

<sup>&</sup>lt;sup>a</sup>Reactions ratings are averages of eight plants of each cultivar and are based on a scale from 0 (= immune) to 5 (= all plants killed) (6).

contrast, isolates of *C. graminicola* from other genera of host plants (*Avena*, *Medicago*, *Hordeum*, *Bromus*, *Triticum*, *Calamagrostis*, *Festuca*, *Sorghum*, and *Danthonia*) were nonpathogenic on corn. Two isolates of the closely related species, *C. falcatum*, obtained from sugarcane in Louisiana, also failed to attack corn. Two isolates from corn, one from Kentucky and one from Kansas, which were slightly more pathogenic to corn than the other 28 tested, were selected for host range studies.

Host range of corn isolates of C. graminicola.—Of the grasses tested, only members of the genus Sorghum were attacked by the two corn isolates (Table 1). Both Sudangrass cultivars were equally susceptible to both isolates. In contrast, three of seven S. bicolor (L.) Moench cultivars were somewhat more susceptible to the Kentucky than to the Kansas isolate, and johnsongrass was markedly more susceptible to the Kansas than to the Kentucky isolate.

Johnsongrass was inoculated with an isolate of *C. graminicola* obtained from sorghum in Texas. In contrast to results with corn isolates (Table 1), this isolate produced only minute red spots which never developed into sporulating lesions. When inoculated with the Texas sorghum isolate, the sorghum cultivars Pioneer Brand 878 and 944

gave 4-type reactions, XB913 was rated 3, and XB935 rated 1. These results were consistent with reactions of these cultivars to anthracnose observed in the field (Vance York, personal communication). Reactions of these sorghum cultivars to corn isolates were clearly different from field reactions and those obtained with the sorghum isolate. In particular, XB935 which is highly resistant to the sorghum isolate and to anthracnose in the field, was more severely damaged by corn isolates than was the highly susceptible cultivar 878 (Table 1).

Effect of light intensity on disease severity.— Poneleit et al. (6), when screening corn varieties for resistance to anthracnose, observed that plants grown and inoculated in the greenhouse were less severely damaged than those grown in growth chambers and inoculated in the laboratory. Since neither temperature nor light intensity could be accurately controlled in the greenhouse, the effects of these two factors on disease severity were investigated. Tests with plants grown in growth chambers indicated that temperature fluctuations over the range which occurred in the greenhouse (22-30C) had little, if any, effect on disease severity. It therefore appeared likely that light intensity, either before or during the inoculation process, might be

<sup>&</sup>lt;sup>b</sup> Reaction ratings based on a scale from 0 (=immune) to 5 (= all plants killed) (6).

<sup>&</sup>lt;sup>c</sup> Grain sorghum.

d Forage sorghum.

an important factor. To test this possibility, separate lots of greenhouse- and growth chamber-grown plants were inoculated under both greenhouse (high light) and laboratory (low light) conditions. Another lot of greenhouse-grown plants was shaded with several layers of cheesecloth after inoculation in the greenhouse. Five cultivars, which ranged from moderately resistant (DeKalb F880) to highly susceptible (Illini Chief Super Sweet) were inoculated with the Kentucky isolate by the standard procedure. Except for DeKalb F880 plants grown in growth chambers, low light intensity following inoculation resulted in increased disease severity. (Table 2).

DISCUSSION.— Several lines of evidence indicate that a new race or races of *C. graminicola* may be developing on corn. The isolates used in this study differ from those previously found in Ohio (10) in that they readily attack nonwounded plants and from those reported from Arkansas (2) in that they attack sorghum. Corn anthracnose was reported for the first time from Maryland in 1971 (5) and caused severe damage for the first time in Indiana in 1972 (7). The ability of corn isolates to attack sudangrass and sorghum, including sorghum cultivars highly resistant to sorghum isolates of *C. graminicola*, suggests that additional problems with anthracnose may arise on these crops. Johnsongrass, which is severely attacked by some corn isolates, is a common weed which may serve as a reservoir host for the fungus.

Increased disease severity when plants were inoculated under conditions of low light intensity and prolonged exposure to high humidity may account, at least in part, for the fact that corn anthracnose is usually restricted to the lower three to four leaves of the plant. Since the disease occasionally progresses to upper leaves, stalks and ears (7),

these organs are not physiologically immune. Severe damage from anthracnose may be expected after prolonged periods of heavily-overcast, rainy weather.

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