Variation in Pathogenicity of Cephalosporium gregatum Isolates

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ABSTRACT

Variation in pathogenicity of isolates of *Cephalosporium gregatum* is of two types. Type I isolates caused wilt symptoms on soybean cultivar Clark-63, and Type II isolates caused vascular browning but not wilt symptoms. On the resistant cultivar, P.I. 84.946-2, Type I isolates also caused more extensive vascular discoloration than did Type II isolates. Phytopathology 61: 1410-1411.

Additional key words: brown stem rot.

Variation in pathogenicity of *Cephalosporium gregatum*, Allington & Chamberlain, the cause of brown stem rot of soybean (collected from within a given geographical area) has not been reported. However, isolates of *C. gregatum* collected from different areas are variable in pathogenicity (4). Hamilton & Boosalis (3) reported that there are sporulating and nonsporulating types. The purpose of this research was to determine any differences in pathogenicity among isolates of *C. gregatum* collected in central Illinois. During the summer of 1970, 35 isolates of *C. gregatum* were obtained from diseased soybean plants selected at random from three fields in central Illinois. These isolates and the American Type Culture Collection (ATCC) culture of *C. gregatum* (No. 11073), included as a pathogenic isolate and morphological standard, were compared for pathogenicity. The isolates were maintained on potato-dextrose agar slants and later transferred to soybean stem-extract agar (1). For preparation of inoculum, hyphal tip transfers were made from each isolate to three bottles of soybean broth (four soybeans/50 ml water, autoclaved 25 min). After incubating at 22 C for 3 weeks, the cultures were used for pathogenicity tests.

Soybean (*Glycine max* [L.] Merr. 'Clark-63') seedlings were grown in a greenhouse in 6-inch clay pots containing steam-treated soil (five plants/pot) at 30-C day and 26-C night temperatures and 1,000 ft-c of light for 15 hr. The pots were watered weekly with a nutrient solution.

At 4 weeks, the seedlings were inoculated with individual isolates by making a wound with a sterile needle in the taproot 1 cm below the soil line. A small amount of mycelium was placed in the wound, and the root was covered with pasteurized soil. Four weeks later, the plants were examined for wilt and defoliation symptoms. Stems split longitudinally were measured for the extent of vascular browning. The experiment was repeated 3 times with the same isolate collection.

Characteristically, two types of pathogenic isolates were present: Type I, which repeatedly caused defoliation and leaf symptoms similar to those reported by Allington & Chamberlain (1) in addition to vascular discoloration; and Type II, which did not produce foliar symptoms but caused vascular discoloration. Inoculations with the ATCC culture of *C. gregatum* produced infection characteristic of Type II. The average length of vascular browning for Type I isolates was 215 mm as compared to 107 mm for Type II isolates. The ATCC culture of *C. gregatum* produced 118 mm of vascular browning.

In field observations for five growing seasons, Chamberlain & Bernard (2) reported that soybean cultivar P.I. 84.946-2 had a higher proportion of disease-free plants (52%) than did cultivar Lincoln (2%). A greenhouse experiment was designed to compare the pathogenicity of 2 Type I isolates and the ATCC culture of *C. gregatum* on Clark-63 (susceptible) and P.I. 84.946-2 (resistant) soybean cultivars. Plants of each cultivar were grown in pots of steam-treated soil. After 4 weeks, 10 plants of each cultivar were inoculated with the individual isolates. Five wounded plants of each cultivar without inoculum served as controls.

Three weeks after inoculation, both Type I isolates produced wilt symptoms and defoliation on inoculated Clark-63 seedlings. The ATCC culture did not produce wilt symptoms and defoliation on Clark-63. The first Type I isolate produced 173 mm vascular browning; the second Type I isolate produced 188 mm vascular browning. The ATCC isolate produced 110 mm vascular browning.

Neither Type I isolate produced wilt symptoms on the resistant P.I. 84.946-2 plants, and the ATCC isolate did not produce any symptoms. The first Type I isolate produced 166 mm of vascular browning in the inoculated plants, and the second Type I isolate produced 178 mm of vascular browning. The ATCC isolate produced only 62 mm of vascular browning. There were no foliar symptoms or vascular discoloration in any of the controls.

The intensity of vascular browning in the inoculated P.I. 84.946-2 plants as compared to Clark-63 plants was less in all cases regardless of the fungus isolate; vascular discoloration was spotty, and generally confined to scattered nodes, whereas in Clark-63 plants, vascular discoloration generally extended continuously from the soil line to the maximum extent of vascular browning.

Three isolates from Type I and two isolates from Type II were selected and compared with the ATCC culture of *C. gregatum* for their growth characteristics at various temperatures. A 4-mm plug of each isolate was transferred from plates of stem-extract agar to triplicate plates of soybean agar (10 g soybean seed boiled 30 min, and liquid decanted and diluted to 1 liter with water plus 16 g agar, pH 5.5). The plates were then placed in incubators at 17, 21, 24, and 27

C for 5 days, and the colony diameters recorded.

The growth of all isolates was less at 17 and 27 C than at 21 and 25 C. The Type I isolates grew faster at 21 C (30-mm average) and 25 C (30-mm average) than did the Type II isolates at 21 C (23-mm average) and 25 C (25-mm average).

Two types of pathogenic isolates of *C. gregatum* were found in field-collected plants. They have been designated Type I and Type II; Type I isolate consistently caused wilt symptoms on inoculated Clark-63 soybeans, while Type II isolate caused vascular discoloration but no wilt symptoms. The Type I isolate also caused a more extensive vascular discoloration in the resistant soybean cultivar (P.I. 84.946-2) than did the Type II isolate. These results demonstrate that variation in pathogenicity of *C. gregatum* is present and could have an influence on the reaction of cultivars in a program of breeding for disease resistance to this fungus disease.

LITERATURE CITED

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