An Epiphytotic of Diaporthe Stem Canker of Soybean in South Carolina

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ABSTRACT

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Stem canker disease of soybean, caused by Diaporthe phaseolorum var. caulivora, has been confirmed for the first time in South Carolina following an epiphytotic. The causal organism isolated from infected soybean stems was identified by in vivo and in vitro morphological studies and a pathogenicity test.

Stem canker of soybean, caused by Diaporthe phaseolorum (Cke. & Ell.) Sacc. var. caulivora Athow & Caldwell, has been a damaging disease in the north central United States for many years (3,5). More recently, the disease has spread to soybean-producing areas of the southern Mississippi Valley and Gulf Coast states (2,4). Before 1982, stem canker disease of soybean had not been

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confirmed in South Carolina.

In September 1982, soybeans in numerous fields in widely scattered areas of South Carolina began to die prematurely. These soybeans showed symptoms characteristic of stem canker disease (5). Plants in varying stages of pod-fill died rapidly. Necrotic, dried leaves remained attached to the stem. On the middle to lower portions of the stems of diseased plants, reddish brown lesions were observed, usually progressing from a leaf scar or node (Fig. 1). Green, nonnecrotic tissue remained above and below most lesions, even at the time of plant death. Advanced lesions consisted of slightly sunken, reddish brown cankers, often 6-10 cm long. Frequently, cankers girdled the stem and killed the

Statewide soybean losses caused by the disorder were not high except in scattered areas where certain cultivars, especially Hutton, Bragg, and Coker 237, sustained losses ranging as high as about 80%. This investigation was undertaken to verify if this disease was the stem canker disease of soybean.

MATERIALS AND METHODS

To isolate the pathogen, small pieces of stem tissue from the margins of stem cankers were surface-disinfected in 2% sodium hypochlorite for 10 min, then rinsed in sterile water and plated on acidified potato-dextrose agar (PDA). After 3 days of incubation at 20 C, mycelium from the edge of advancing colonies was transferred to PDA in petri plates for maintenance.

Pathogenicity of the isolates was tested by a modified toothpick inoculation method for soybean seedlings as reported by Keeling (4). Flat toothpicks were boiled for 30 min in each of three changes of tap water, then dried and placed in glass vials $(2.5 \times 5.5 \text{ cm})$. Potato-dextrose broth was added to the vials to a depth of 1 cm. The vials were capped loosely and autoclaved for 15 min at 120 C. The cooled broth was inoculated with mycelium from an actively growing culture on PDA and incubated at 21 C for 15 days before use.

Soybean seedlings of cultivar Coker 338 were grown in a steam-disinfested Varina sandy loam-sand mixture (2:1) in 10-cm-diameter clay pots. Eleven plants were inoculated 15 days after planting. A small hole was made with a dissecting needle in the stem 1 cm below the cotyledon and a toothpick tip overgrown with mycelium was inserted in the hole. Eleven other seedlings that served as uninoculated checks were pierced with toothpicks soaked for 15 days in potatodextrose broth that had not been inoculated with the fungus. Plants were placed in a moist chamber at 100% RH and 23 C for 4 days before they were removed to a greenhouse bench at 23 ± 3 C for six more days, then the symptoms were evaluated. Reisolation from inoculated stems showing canker symptoms was made using the method described previously.

Microscopic examination was made of fungal fruiting bodies on stem cankers from naturally infected field-grown soybeans and from PDA cultures of the isolated fungus.

RESULTS AND DISCUSSION

Occasionally, within cankers on naturally infected soybean stems, black, globose perithecia, predominantly in caespitose groups within a stroma, were embedded within the cortical tissue of the stem and had protruding beaks. The thinwalled, eight-spored asci were elongate-clavate and sessile. Paraphyses were absent, and no pycnidia were found within the cankers examined.

Isolations from cankers on infected soybean stems consistently yielded a fungus with white mycelium, initially



Fig. 1. Characteristic stem canker on soybean stem caused by *Diaporthe phaseolorum* var. caulivora. Note the nonnecrotic tissue above and below the canker that appears to emanate from a leaf node. The interveinal necrosis on the leaf is a nonspecific symptom of the disease.

appressed but later becoming flocculent. The mycelium remained white despite the age of the culture. On PDA maintained in alternating light and darkness, black perithecia with protruding beaks developed abundantly in groups within mycelial tufts. No pycnidia formed. Asci were similar to those described from the perithecia found in cankers on infected soybean stems. Ascospores were hyaline, two-celled, elongate-ellipsoid, slightly constricted at the septum, and biguttulate in each cell.

All soybean plants inoculated with the suspected pathogen developed cankers averaging 1.9 cm long within 10 days of

inoculation. These plants showed overall symptoms ranging from slight wilting to plant death. The control plants, pierced with uninoculated toothpicks, showed no signs of canker formation or disease symptoms. The fungus isolated from the inoculated soybean seedlings was similar to the fungus initially isolated from cankers on the naturally infected soybean plants.

Symptoms of this disease are characteristic of those of stem canker disease of soybean caused by D. phaseolorum var. caulivora (5). The fungus observed on and consistently isolated from cankers on naturally infected soybean plants was also similar to D. phaseolorum var. caulivora as described by Athow and Caldwell (1). The fungus isolated was pathogenic on inoculated soybean seedlings and produced stem canker symptoms as reported by Keeling (4). We conclude that this disease is truly stem canker disease of soybeans caused by D. phaseolorum var. caulivora. This is the first confirmed report of this disease in South Carolina.

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