# Anthracnose of Alfalfa Observed in Minnesota

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#### ABSTRACT

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Anthracnose of alfalfa was observed for the first time in Minnesota near Stanton in July 1978 and again near Rosemount in August 1979. Isolates obtained from plants growing at both locations were identified as race 1 of Colletotrichum trifolii.

Additional key words: Medicago sativa

Anthracnose, caused by Colletotrichum trifolii Bain, is a major disease of alfalfa (Medicago sativa L.) in the Middle

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Atlantic and southeastern states, where it is considered the principal cause of "summer decline" (1,2). Sporadic epidemics have been reported from New York and Washington (3,8). We have observed C. trifolii in some states of the Midwest, including Iowa and Nebraska and as far north as southern Wisconsin, sometimes causing severe damage: The fungus, however, has not previously been reported on alfalfa in Minnesota.

Two races of *C. trifolii* occur on alfalfa, based on the differential reactions of cultivars Saranac, Arc, and Saranac-AR (6,7,9). Saranac is susceptible to both races, Arc is resistant to race 1 and susceptible to race 2, and Saranac-AR is resistant to both races. Race 1 is most common, whereas race 2, which was first found in 1977–1978, occurs locally in North Carolina (9) and Maryland (6).

In August 1978, anthracnose symptoms were observed on a susceptible strain of alfalfa in research plots near Stanton, MN, and C. trifolii was isolated from typical stem and crown lesions. Anthracnose was also observed in August 1979, and C. trifolii was isolated from alfalfa in plots on the Agricultural Research Station near Rosemount, MN.

The first report of *C. trifolii* on alfalfa in Minnesota is given in this paper. The pathogenicity of an isolate (MN 1-4) from Stanton and three (Ct-1, Ct-2, and Ct-4) from Rosemount are compared with isolates of race 1 (PA) and race 2 (NC-4) at Raleigh, NC.

### **MATERIALS AND METHODS**

Isolates of *C. trifolii* were grown on potato-dextrose agar and stored at 3-4 C until used for the preparation of inoculum. Petri dishes of lima bean agar were flood-inoculated with conidia from potato-dextrose agar slants of each isolate and incubated for 7 days at 24-26 C. Conidia were washed from the agar surface with distilled water, and the suspensions were adjusted to 10<sup>6</sup> conidia per milliliter in a final volume of 1 L of distilled water supplemented with two drops of Tween 20 and 30 ml of freshly squeezed and filtered orange juice (4).

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The race 1 isolate of *C. trifolii* was originally obtained from the U.S. Regional Pasture Laboratory, University Park, PA, and has been the standard isolate used to test for resistance in North Carolina germ plasm. The race 2 isolate was originally obtained in Rowan County, NC, in 1977 (9).

Before planting, seeds of Arc and Saranac-AR cultivars of alfalfa were scarified and inoculated with an appropriate strain of Rhizobium meliloti Dang. (The Nitragin Co., 3101 W. Custer Ave., Milwaukee, WI 53209). Seeds were planted in a mixture of Metromix-200 (Florist Products, Inc., 780 W. Oakton St., Des Plaines, IL 60018) and fumigated sand (1:1, v/v). Plants were watered and fertilized as needed to maintain vigorous growth. Seedlings were grown in pressed board trays (14 ×  $19 \times 7$  cm) and thinned to 25 per row. When the plants were 3 wk old, the trays were enclosed in unvented plastic freezer bags and sealed.

The plants were inoculated by being sprayed with the spore suspension through a slit in the bag until dripping wet. After inoculation, the bags were removed and the trays were put into a mist chamber at 20–25 C for 3 days; intermittent misting kept the foliage wet. The seedlings were returned to a bench in a greenhouse at 18–24 C for 3–4 wk of incubation. Check plants were sprayed with Tween 20 and orange juice in water without adding conidia of C. trifolii. Check-1 was sprayed before and check-2 after the seedlings had been inoculated with the test isolates.

The experiment was arranged in a randomized complete block design with two replications and repeated once. Twenty-five seedlings of each cultivarisolate combination and uninoculated control were included in each replication.

Seedlings were scored on a five-point scale with 1 = no lesions or only hypersensitive flecking; 2 = small lesions, without sporulation; 3 = typical diamond-shaped lesions not girdling the stem, with sporulation and setae in the acervuli; 4 = stem-girdling lesions with sporulation, but with new shoots originating from lower axillary buds; and 5 = a dead plant. Plants scored 1 and 2 were considered resistant.

## RESULTS AND DISCUSSION

All isolates of C. trifolii were pathogenic and caused typical anthracnose symptoms in susceptible alfalfa seedlings. Resistant reactions (a score of 1 or 2) were observed in 95-99% of the uninoculated seedlings. Not all control plants survived, but isolations from the dead plants indicated that the anthracnose fungus was not the cause of death. The Arc cultivar had 66-80% resistant seedlings when inoculated with the Minnesota isolates, 77% when inoculated with PA (race 1), and 9% when inoculated with NC-4 (race 2). Saranac-AR had 54-66% resistant seedlings when inoculated with the Minnesota isolates, 75% when inoculated with PA, and 56% when inoculated with NC-4.

An additional 13 single-spore cultures from isolates of *C. trifolii* obtained from diseased plants at Stanton were tested on Saranac-AR and Arc by the seedling box method (5). Saranac-AR was highly susceptible to all isolates and Arc was highly resistant.

Because Arc was resistant to all Minnesota isolates, we concluded that the anthracnose fungus at Rosemount and Stanton was race 1 of *C. trifolii*. A

herbarium specimen (RHM 101) of *C. trifolii* from the Stanton location was deposited in the Plant Pathology Mycological Herbarium of the University of Minnesota (MPPD).

In the past, plant pathologists have assumed that the climate in the northern states of the Midwest was not conducive to the development of anthracnose in alfalfa. The recent occurrence of this disease at two locations in Minnesota, however, and our observations of significant damage in alfalfa stands in southern Wisconsin and Iowa suggest that anthracnose should be considered a potential factor in stand losses, especially during hot and wet seasons, in the northern areas of the Midwest.

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