## Comparative Pathogenicity of Pythium aphanidermatum and Pythium myriotylum to Twelve Plant Species and Intraspecific Variation in Virulence

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## ABSTRACT

Tomato, bean, cucumber, rye, oats, wheat, peanut, sorghum, soybean, cotton, corn, and tobacco were inoculated with 14 isolates each of *Pythium aphanidermatum* and *Pythium myriotylum*. The crops differed greatly in susceptibility to the two organisms, tomato, bean, and rye being most susceptible,

cotton and corn most resistant. The two *Pythium* spp. often differed in pathogenicity on a given plant species, and individual isolates of each organism varied markedly in virulence on susceptible crops. Phytopathology 60:264-268.

Pythium aphanidermatum (Edson) Fitz. and Pythium myriotylum Drechs, attack a large number of economically important crops grown in the southeastern United States. Middleton (9) listed 81 hosts for P. aphanidermatum; a recent host list for P. myriotylum included 15 species (7). We are concerned with these organisms as causes of root and stem rots of fall-seeded small grains, particularly rye and spring-seeded tomato used for transplant production. Both Pythium spp. appear to be common inhabitants of soils in the Southeast, are favored by similar environmental conditions, are commonly associated with the same hosts, and produce similar symptoms. Because of these similarities, it is probable that these organisms have been confused in some previous work. Little has been reported on their comparative pathogenicity on major southeastern crops, so the present work appeared needed.

Limited information is available concerning differences in virulence among isolates of each organism. Freeman et al. (3) inoculated small grains with three isolates of *P. aphanidermatum* from different hosts and found neither a difference in virulence nor evidence of host specificity. However, Morgan & Hartwig (10) reported that isolates of *P. aphanidermatum* from soybean (*Glycine max*) were more virulent to that crop than were isolates from soil. McCarter & Littrell (7) found that one isolate of *P. myriotylum* was less virulent to several plant species than three other isolates tested.

The present studies were made to compare the pathogenicity of *P. aphanidermatum* and *P. myriotylum* on 12 plant species, and to determine the amount of intraspecific variation in virulence. A preliminary report has been published (8).

MATERIALS AND METHODS.—Fourteen isolates each of *P. aphanidermatum* and *P. myriotylum* were used to inoculate tomato (*Lycopersicon esculentum Mill.* 'Campbell 17'), bean (*Phaseolus vulgaris L.* 'Tendercrop'), cucumber (*Cucumis sativus L.* 'Boston Pickling'), rye (*Secale cereale L.* 'Wrens Abruzzi'), oats (*Avena sativa L.* 'Florida 500'), wheat (*Triticum aestivum L.* 'Georgia 1123'), peanut (*Arachis hypogaea L.* 'Early Runner'), sorghum (*Sorghum bicolor* [L.]

Moench 'Lindsey 77F'), soybean 'Hampton', cotton (Gossypium hirsutum L. 'Atlas 66'), corn (Zea mays L. 'Dixie 18'), and tobacco (Nicotiana tabacum L. 'Hicks'). Pre- and postemergence inoculations were made on all crops except tobacco, which was inoculated only after transplanting. In the pre-emergence tests, seeds were planted in 10-cm pots filled with soil infested separately with the various fungal isolates. Seeding rates in seeds per pot were as follows: tomato, rye, wheat, oats-30; bean, cucumber, soybean, grain sorghum-10; and peanut, corn, cotton-6. In the postemergence tests, the plants were grown 7- to 14-cm tall before inoculation. The same numbers of plants per pot were used as in the pre-emergence tests, except that bean, cucumber, soybean, and sorghum were reduced to six plants, and tomato to 10 plants. One tobacco plant was established in each pot. Potting soil used in all tests was an autoclaved (2 hr at 15 psi) Goldsboro loamy sand mixed (3:1, v/v) with vermiculite, and fertilized with a complete mixture.

We obtained cultures from several hosts or substrata, and locations to study differences in virulence among isolates of each Pythium. The isolates of P. aphanidermatum originated as follows: six from tomato in Georgia; one each from Citrus sp., Chrysanthemum sp., rye, and soil in Florida; one from unknown hosts each in California and North Carolina; and two from unknown sources. The isolates of P. myriotylum originated as follows: three from peanut in Virginia; two each from peanut, tomato, and bean in Georgia; one each from rve and soil in Georgia; one each from tobacco and rye in Florida; and one from azalea (Rhododendron sp.) in Virginia. All isolates were maintained on V-8 juice agar at 18 C. Cultures for inoculum production were grown in petri dishes on V-8 agar for 2 days at 30 C. Details on methods given below for infesting soil in pre-emergence tests and for inoculating plants in postemergence tests were given earlier (7). In the pre-emergence tests, contents of two petri dish cultures were fragmented in 500 ml of water in a Waring Blendor, and the resulting suspension was mixed with 9 kg (air dry basis) of soil. In the postemergence tests, two petri dish cultures were blended in 250 ml of water,

and 30 ml were injected with a syringe and cannula at six locations in each pot.

The 28 isolates of the two Pythium spp. were tested simultaneously on a group of crops (usually six), and tests with each group were made twice. Four replications of each treatment combination and controls were used in each test. All tests were made in the greenhouse from March through July 1968, the postemergence tests in March to May, and the pre-emergence in June and July. Soil temperatures during this period ranged from 21 to 39 C, but were usually maintained at 30-32 C (near optimum for both organisms) by heating or cooling the greenhouse as needed. Thermograph records were maintained to verify that temperatures during the tests were optimum for disease development. Soil moisture was maintained at field capacity or higher after inoculation. In the pre-emergence tests, surviving plants were counted 14 days after seeding. Data on disease severity in the postemergence study were taken 14 days after inoculation. Roots of surviving plants were washed, and the plants were rated for disease severity on a previously described (7) 0 to 6 disease index where 0 = no discoloration or rot of roots or stems, 1 to 5 = increasing degrees of root and stem rot,and 6 = plant dead.

RESULTS.—The various crops differed greatly in susceptibility to the two *Pythium* spp. in both pre- and postemergence tests (Fig. 1, 2). Stands of rye, oats, wheat, tomato, bean, and soybean were markedly reduced by several isolates of both *P. aphanidermatum* and *P. myriotylum* when seeds were planted in infested soil. Rye was the most susceptible of all the species

tested. Peanut and sorghum were less susceptible than the above crops, although one or more isolates of each organism caused moderate stand reductions. Cotton and corn germinated and survived well in soil infested with either organism. Typical damage caused by both organisms on susceptible crops was a decay of the seed or seedling prior to emergence, or a rot of the stem soon after emergence. In the postemergence tests, tomato, bean, and tobacco were damaged moderately to severely by the two organisms. Cotton and corn were highly resistant to both organisms. The other crops varied in susceptibility depending on the organism and isolate used. Damage to the various crops caused by the two organisms ranged from a minor amount of root decay, as occurred on cotton and corn, to complete kill of plants as occurred when bean and tomato were inoculated with some isolates. The typical symptom following attack by both Pythium spp. was the appearance near the soil level of dark brown lesions, which often advanced several cm up the stem.

The two *Pythium* spp. often differed in pathogenicity on a given plant species (Fig. 1, 2). For example, in pre-emergence tests, stands of tomato, cucumber, and soybean were reduced more by *P. aphanidermatum* than by *P. myriotylum*, whereas the reverse occurred on bean. In the postemergence tests *P. myriotylum* caused considerable damage to rye, oats, wheat, peanut, and sorghum, but *P. aphanidermatum* usually caused only slight damage to these crops. Bean was highly susceptible to *P. myriotylum* and moderately susceptible to *P. aphanidermatum*.

Isolates of the two organisms varied greatly in their

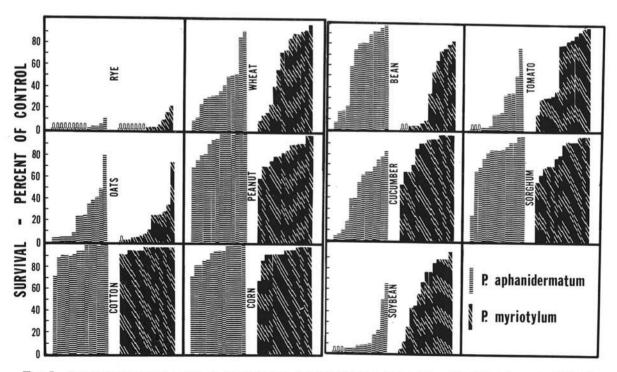


Fig. 1. Survival of 11 plant species 14 days after seeding in soil infested separately with 14 isolates each of Pythium aphanidermatum and Pythium myriotylum. Each bar represents the results with an individual isolate. 0 = no survival.

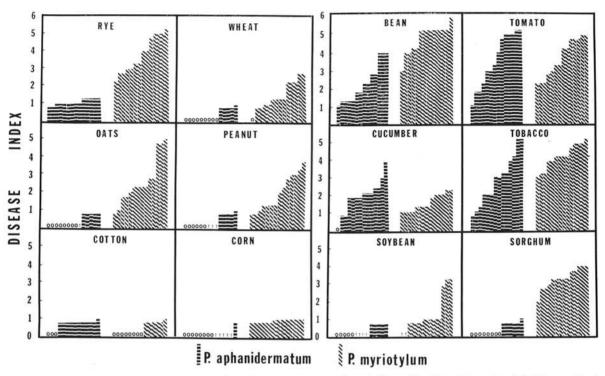


Fig. 2. Disease indices on 12 plant species after postemergence inoculation with 14 isolates each of *Pythium aphanidermatum* and *Pythium myriotylum*. Each bar represents the results with an individual isolate. 0 = no disease, T = trace or isolated lesions, 1-5 = increasing degrees of root and stem rot, and 6 = plants dead.

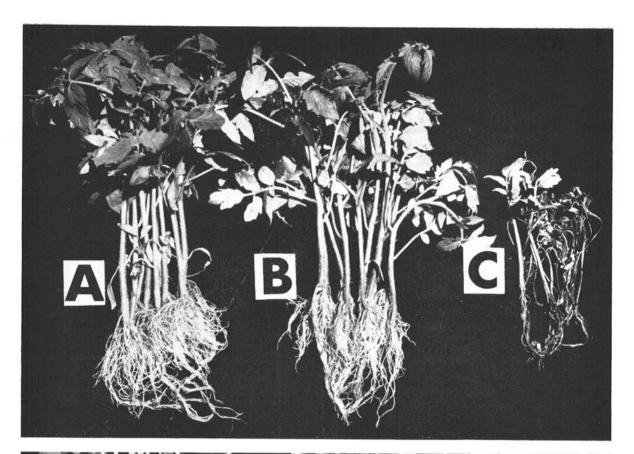
capacity to reduce plant stands when seeds were planted in infested soil (Fig. 1), and to cause damage to established plants (Fig. 2). In the postemergence studies, isolates of P. aphanidermatum ranged in virulence from low to high on tomato (Fig. 3, above) and tobacco, and from low to moderate on bean and cucumber. Isolates of P. myriotylum varied in virulence from low to high on tomato, rye, and oats; from low to moderate on wheat, sorghum, and peanut (Fig. 3, below); and from moderate to high on bean and tobacco. Three isolates of P. myriotylum (all from peanut in Virginia) were moderately damaging to soybean, whereas none of the other 11 isolates caused significant damage. Some isolates of the two organisms rated consistently low in virulence to all crops; other isolates were highly virulent to all susceptible crops; and still other isolates varied in virulence among crops.

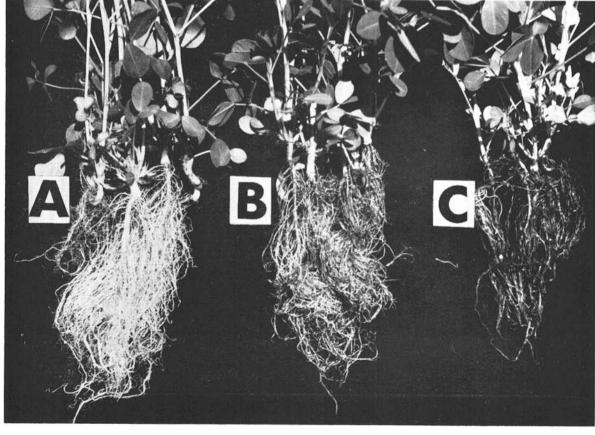
DISCUSSION.—We have shown that (i) the crops tested vary greatly in susceptibility to *P. aphanidermatum* and *P. myriotylum*; (ii) there are basic differences in pathogenicity of these organisms on several of the crops; and (iii) there is considerable intraspecific variability in virulence.

All of the twelve crops except peanut were previously reported as hosts of *P. aphanidermatum* (3, 9, 10),

and all except sorghum, soybean, cotton, and corn as hosts of P. myriotylum (2, 4, 5, 7, 9). We found cotton and corn highly resistant to both Pythium spp. Freeman et al. (3) also found corn to be resistant to P. aphanidermatum. Established tobacco and bean plants were more susceptible to P. myriotylum than to P. aphanidermatum, whereas the two organisms were about equally destructive to tomato. P. myriotylum has been implicated in a pod rot of peanut (4), and Bell (1) found that it attacked peanut seedlings grown in test tube culture. Our results demonstrate that some isolates of P. myriotylum also cause a root and stem rot of established peanut plants, whereas P. aphanidermatum does not cause appreciable damage to peanut. Morgan & Hartwig (10) found that P. aphanidermatum caused both pre- and postemergence killing of soybean. Our results differed somewhat, in that the organism did not cause significant damage in postemergence inoculations. This difference probably can be explained on the basis of the different inoculation procedures used. They made wound inoculations on the stem, but we injected the inoculum into the soil. Our report appears to be the first that establishes sorghum and soybean as being susceptible to P. myriotylum.

Fig. 3. (Above) Tomato plants 14 days after postemergence inoculation with the least virulent (B) and most virulent (C) of 14 isolates of *Pythium aphanidermatum*. A, control. (Below) Peanut plants 14 days after postemergence inoculation with the least virulent (B) and most virulent (C) of 14 isolates of *Pythium myriotylum*. A, control.





Plant age apparently influences the reaction of several crops to the two *Pythium* spp. For example, stands of rye, oats, and wheat were greatly reduced when planted in soil infested with the two organisms. *P. aphanidermatum*, however, did not cause significant damage to these crops when plants 13-15 cm tall were inoculated. *P. myriotylum* caused rather severe damage to established plants.

Morgan & Hartwig (10) reported that isolates of P. aphanidermatum from soybean were more virulent to that crop than were isolates from soil. We did not observe such specificity among our isolates. For example, some isolates from soil were as virulent to tomato as were isolates from tomato. Our results do agree with the findings of Morgan & Hartwig (10) that isolates of P. aphanidermatum differ in virulence. The extreme variability in virulence of the 14 isolates of P. aphanidermatum and P. myriotylum in our tests indicates the importance of testing several isolates before drawing a conclusion on the susceptibility of a test plant. The findings of Kilpatrick (6) that varieties of certain small grains differ in susceptibility to several Pythium spp., including P. aphanidermatum, may also suggest both the advisability of using several varieties in inoculation tests and the possibility of breeding for resistance to these diseases.

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