

Persistence of *Pythium myriotylum* in Soils

Kenneth H. Garren

Plant Pathologist, Plant Science Research Division, ARS, USDA, Holland, Virginia 23391, and Adjunct Professor, Department of Plant Pathology and Plant Physiology, Virginia Polytechnic Institute and State University.

Cooperative investigations of Plant Science Research Division, ARS, USDA, and Research Division, Virginia Polytechnic Institute and State University.

Contribution No. 202, Department of Plant Pathology and Plant Physiology, Virginia Polytechnic Institute and State University, Blacksburg 24061.

ABSTRACT

Pythium myriotylum survived at lower inoculum:soil ratios, and survived longer in two soils to which it is not indigenous, than in a soil to which it is indigenous. A possible explanation is that there are no antagonists to *P. myriotylum* in the two soils that are present in the one where survival was reduced. *Phytopathology* 61:596-597.

Additional key words: competitive saprophytism, antagonisms, resurgence of fungi.

In a 1966 survey (5), *Pythium myriotylum* Drechs., a pathogen of pod breakdown of Virginia-type peanuts (*Arachis hypogaea* L.) (3), was not found in soil samples obtained from Virginia. The samples were taken from around fruits with pod breakdown, and *P. myriotylum* was isolated from many of these fruits. Still another pathogenic thermophilic *Pythium* sp. of southern coastal plain soils (6) was not found in the Virginia samples (5). Subsequently, I compared the persistence of artificial inoculum of *P. myriotylum* in soils at Cambridge, England and Blacksburg, Va. (to which this species seems not to be indigenous) with its persistence in the Virginia coastal plain soil. Results reported herein suggest that there are antagonists of *P. myriotylum* in the coastal plain soil that are not present in two soils in which *P. myriotylum* is not indigenous.

A modification of the "Cambridge method" for estimation of competitive saprophytic colonization (1, 4) was used. Flasks of sand-cornmeal medium were seeded with three different clones of *P. myriotylum*. After 14 days' growth, the inoculum was mixed with each soil in ratios of 1:1, 1:5, 1:39, 1:100, 1:200, 1:500, and 1:800. The raw soils were dried on a greenhouse bench and restored to 50% moisture-holding capacity before they were mixed with the inoculum or used in checks. Three soil types were used: Kettering loam, an English soil, with a pH of 5.8; Greendale, a silty clay loam of the mountains of Virginia, 5.3; and Woodstown loamy fine sand, a Virginia coastal plain soil in which *P. myriotylum* is indigenous, 5.8. There were three checks: soil alone, soil plus sand, and soil plus sterile sand-cornmeal medium. The sand and sterile medium were in the same ratios as in the inoculum:soil mixes. The soils and the inoculum:soil mixes were stored in closed

plastic bags in the laboratory. After 10 months at room temperature, the pH of the 1:5 inoculum:soil mixtures with the Virginia soils were 4.7 and 4.9, respectively.

Samples were taken from the storage jars after 5, 10, 15, 20, 25, 35, 45, 55, 65, 100, 200, and 300 days, and restored to their original moisture contents with distilled water. Ten 60-g lots of each sample were put in 90 × 25 mm petri dishes. To determine extent of fungal colonization, I partially buried germinated turnip seed (*Brassica rapa* L. 'Early Snowball') in five of the dishes, and immature Va. Bunch 46-2 peanut fruit in five dishes. Pieces of each lot of peanut fruit were surface-sterilized and plated on cornmeal-wheat-germ oil agar. No evidence of endocarpic (2) *Pythium* spp. was found, though half the plates were incubated for 2 days at 27 C and half at 39 C (6).

After 5 days' incubation at 27 C, the extent of colonization of turnip radicles or peanut fruit by *P. myriotylum* was determined by incubating pieces on cornmeal-wheat-germ oil agar at 27 and 39 C. Lysis or autolysis destroyed the introduced inoculum in all 1:1 ratios in 20-35 days. No thermophilic *Pythium* sp. other than *P. myriotylum* was found in any of these plates. The only other *Pythium* sp. found in any plates was *P. ultimum* Trow, found in several pieces of tissue taken from the three Greendale checks.

Enough *P. myriotylum* inoculum persisted in 65-day-old mixes with the Kettering soil to invade turnip radicles from all inoculum:soil ratios except those of 1:1, 1:500, and 1:800. Contact with wounds where fruit were detached from plants seemed necessary for invasion of fruit by *P. myriotylum* in inoculum:soil ratios of 1:100 and 1:200 held for 65 days. When 1-cm incisions were made in fruits, enough *P. myriotylum* inoculum was present in the 1:500 inoculum:soil ratio after 65 days to cause some rot around the incisions.

Similar results were obtained in Virginia in 1969-70 with the Greendale soil, but not with the Woodstown soil. In these studies, fruit were wounded and the above inoculum:soil ratios were used. After 15 days, there was no invasion of fruit or radicles from Woodstown soil at the 1:200 ratio; after 65 days, the only invasion of radicles and rot of fruit (Fig. 1) in Woodstown was at the 1:5 ratio. In the Greendale soil there was invasion of radicles and rot of fruit at the 1:500 ratio and some invasion of fruit at 1:800 (Fig. 1).

The indigenous *P. myriotylum* in the Woodstown checks was not noticeably active on radicles when the soil was held for 65 days, but it did cause some rot at incisions on a few fruit. When inoculum was introduced into Woodstown soil which was pasteurized rather than sterilized, enough inoculum persisted for up to 35 days to invade radicles. When Woodstown soil sterilized in an autoclave was not handled aseptically, *Trichoderma viride* Pers. ex Fr. grew rapidly in it, and there was no evidence of invasion of radicles or fruit by introduced *P. myriotylum* inoculum after 15 days.

The introduced *P. myriotylum* inoculum did not always die off in the Woodstown soil. It resurged and

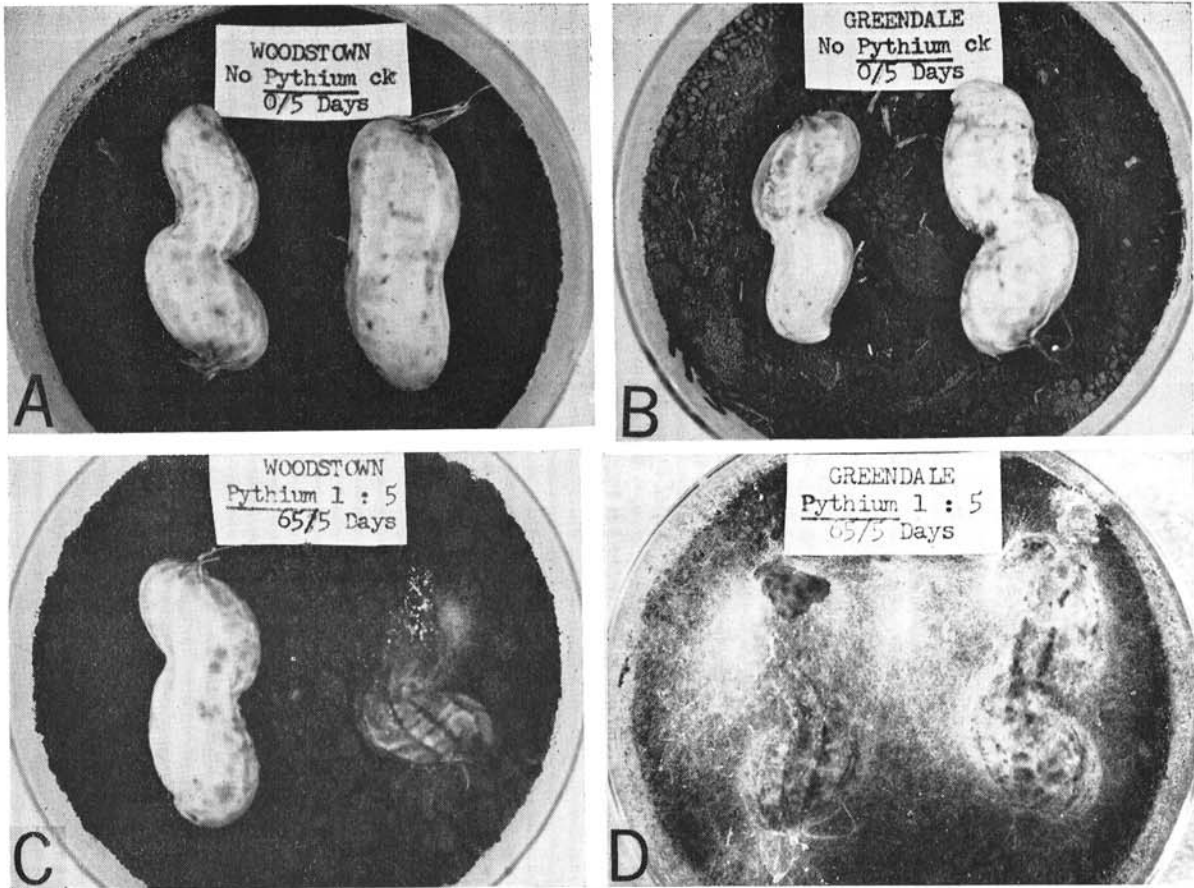


Fig. 1. Reactions of wounded, immature Va. Bunch 46-2 peanut fruits to 5 days' incubation at 27 C on Woodstown loamy fine sand and Greendale silty clay loam with or without inoculum of *Pythium myriotylum*. A and B had no inoculum. C and D had one part inoculum to five parts soil. D is enlarged slightly.

invaded radicles and fruit at the 1:5 ratio at 65 days when the mixture was moistened with calcium or potassium sulfate solutions; however, there was no such resurgence when the mixtures were moistened with distilled water. Thus, either a Ca or K cation and radicle plus fruit wound exudates seemed necessary to the activity of the *P. myriotylum* propagules from the artificial inoculum in Woodstown soil. There are at least two possible explanations for all this. Some chemical or physical characteristic of the Woodstown soil could have promoted more autolysis of inoculum in it than in the two other soils. However, it could be simply that Woodstown soil has antagonists of *P. myriotylum* which Kettering and Greendale soils do not have. Soil dilution plates gave no evidence that soil types had different effects on propagule formation; and almost no sporangia or oospores could be found by examining drops of the soil dilutions with a microscope. Nevertheless, there must have been more propagules of the introduced inoculum of *P. myriotylum* close to radicles and fruit wounds in the mixtures with Kettering and Greendale soils than in the mixtures with Woodstown soil. Since the artificial inoculum persisted

longer in pasteurized Woodstown than in sterilized Woodstown, the explanation based on activity of organisms other than *P. myriotylum* seems more plausible.

LITERATURE CITED

1. BUTLER, F. C. 1953. Saprophytic behaviour of some cereal root-rot fungi. I. Saprophytic colonization of wheat straw. II. Factors influencing saprophytic colonization of wheat straw. *Ann. Appl. Biol.* 40: 284-311.
2. GARREN, K. H. 1966. Peanut (groundnut) mycofloras and pathogenesis in peanut pod rot. *Phytopathol. Z.* 55:359-367.
3. GARREN, K. H. 1967. Relation of several pathogenic organisms and the competition of *Trichoderma viride* to peanut pod breakdown. *Plant Dis. Repr.* 51:601-605.
4. GARRETT, S. D. 1963. *Soil fungi and soil fertility*. Pergamon Press, Oxford. 165 p.
5. HENDRIX, F. F., JR., & W. A. CAMPBELL. 1970. Distribution of *Phytophthora* and *Pythium* species in soils in the Continental United States. *Can. J. Bot.* 48:377-384.
6. LITRELL, R. H., & S. M. McCARTER. 1970. Effect of soil temperature on virulence of *Pythium aphanidermatum* and *Pythium myriotylum* to rye and tomato. *Phytopathology* 60:704-707.