Effect of Interaction Between Fusarium, Pythium, and Rhizoctonia on Severity of Bean Root Rot

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ABSTRACT

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The effect of *Fusarium solani* f. sp. *phaseoli*, *Pythium ultimum*, and *Rhizoctonia solani* singly and in combination on beans was studied in artificially-infested, pasteurized, bean-field soil in a controlled environment. Root rot was more severe in soil infested with both *P. ultimum* and *F. solani* f. sp. *phaseoli*. Data suggest that a synergistic relationship exists between these pathogens. Root-rot ratings of plants growing in soil infested with *P. ultimum*, *F. solani* f. sp. *phaseoli*, or a combination of the two organisms was 3.2, 0.8, and 5.1, respectively. Similarly, reduction in total dry weight per plant was 52, 12, and 74%, respectively. No interaction was observed between *R. solani* and *F. solani* f. sp. *phaseoli*. However, *R. solani* significantly reduced the severity of root rot incited by *P. ultimum* when the two were combined, suggesting an antagonistic relationship. Observations in recent years suggest that the synergistic interaction between *P. ultimum* and *F. solani* f. sp. *phaseoli* occurs under field conditions in New York, particularly during cool, wet periods.

Additional key words: *Phaseolus vulgaris*.

Bean root rot is a limiting factor in the production of snap beans. It is particularly severe under the cool, wet, soil conditions that often prevail during the spring throughout the bean-growing areas of New York. The disease can be incited by *Fusarium solani* (Mart.) Appel & Wr. f. sp. *Phaseoli* (Burk.) Snyd. & Hans., *Pythium ultimum* Trow, *Rhizoctonia solani* Kuehn, and *Thielaviopsis basicola* (Berk. & Br.) Ferr. (22). The lesion nematode, *Pratylenchus penetrans* (Cobb) Filip. & Schuurm.-Stekh., also is associated consistently with bean roots and soils in New York (11) and has caused considerable root damage and dry weight loss under greenhouse conditions (Abawi, *unpublished*). Depending on soil and environmental conditions, these pathogens may act independently or as a complex in any possible combination on beans.

Interactions between soilborne plant pathogens have been demonstrated to greatly influence disease incidence and severity on many crops (8, 19), but most reports on bean root rot are concerned primarily with damage caused by individual pathogens (22). However, Hutton et al. (10) reported that the incidence of root rot of dry beans caused by *F. solani* f. sp. *phaseoli* was increased by combination with *P. penetrans* or *Meloidogyne* spp., but severity of the disease was not affected. In similar studies, Burkholder (4) reported that *T. basicola* and *F. solani* f. sp. *phaseoli* together reduced the yield of dry beans more than either fungus alone. Maloy (17) studied all possible combinations of the four pathogens on Red Kidney and reported that no combination resulted in more severe root rot than that incited by *F. solani* f. sp. *phaseoli* alone.

Our research results, field observations, and greenhouse tests with selective fungicides for the control of bean root rot suggest that there are interactive effects between the bean root-rot pathogens. Therefore, this study was done to determine the effect of *P. ultimum*, *R. solani*, and *F. solani* f. sp. *phaseoli*, singly and in combination, on the severity of root rot of snap beans.

MATERIALS AND METHODS

Isolates of *P. ultimum* and *R. solani* used in this study were recovered from field-infected snap beans in New York. The isolate of *F. solani* f. sp. *phaseoli* was isolated from beans in Nebraska (J. R. Steadman, Department of Plant Pathology, University of Nebraska, Lincoln, NE 68503). Cultures of these fungi were maintained by periodic transfers on potato-dextrose agar (PDA) or cornmeal agar plates incubated at 21-25 C. Field soil (sandy loam, pH 6.8, and 2.8% organic matter) with a history of severe bean root rot was used in all tests. The soil was pasteurized by aerated steam (60 C for 30 min) and stored in loosely covered cans for 3-4 wk before infestation with the three pathogens, singly and in all combinations. Conidial suspensions of *F. solani* f. sp. *phaseoli* were prepared from 2- to 4-wk-old cultures growing on PDA plates at 25 C under 12 hr of fluorescent light (5,000 lux) per day. The concentration of the conidial suspension was estimated by counting with a microscope the number of spores in four 0.01-ml aliquots. Conidia (mostly macroconidia) were thoroughly mixed into moist soil (20% moisture) at a rate of 500 conidia/g oven-dry soil. Inoculum of *P. ultimum* was prepared and

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added to soil as described previously (18) at the rate of 500 sporangia/g of oven-dry soil. Inoculum of *R. solani* was prepared by the method of Ko and Hora (14) and 5.5 g were added per kilogram of soil. The inoculum densities used in this study were based on results from preliminary tests to determine populations that consistently caused moderate amounts of disease.

To further document the observed interactions between *F. solani* f. sp. *phaseoli* and *P. ultimum*, pasteurized soil infested with each fungus alone or in combination was treated with Dexon (*p*-dimethylamino-benzenediazo sodium sulfonate) which is active only against pythiaceous fungi (16). Dexon (35 WP) in water was incorporated into moist soil at the rate of 230 mg a.i./kg of oven-dry soil. In other tests, nonpasteurized soil from two bean fields with a history of root rot were treated similarly with Dexon. Portions of these same soils not treated with the fungicide served as controls.

Eight-day-old seedlings of the cultivar, Early Gallatin, grown in sterilized sand at 22-23 C, were used throughout this study. The bean seedlings were transplanted as described previously (18) into 10-cm diameter plastic pots filled with the various treated soils. Four plants were grown per pot, each treatment was replicated four times, and all tests were repeated once. The experiments were conducted in a growth chamber at 21 ± 1 C, 75% relative humidity, and with 14 hr of cool-white fluorescent light (11,000 lux) per day. Water was added to the surface of each pot daily to maintain the soil in a "visibly" moist condition at all times.

The experiments were terminated 24 or 28 days after transplanting. Plant height was recorded and the roots were washed free of soil and rated for root-rot severity. The hypocotyls and roots were rated separately on a scale of 0 to 6, wherein 0 indicates no apparent symptoms and 6 refers to the most severe disease development (dead plant). Isolations from hypocotyl and root tissues from all treatments were made on water agar and a selective medium for *Pythium* spp. (20). Total dry weight was determined by weighing plants that had dried for 72 hr at 95 C. Data in the tables from representative experiments were analyzed by the Waller-Duncan's Bayesian K-ratio (LSD) rule (21).

RESULTS

In pasteurized soil, *F. solani* f. sp. *phaseoli* alone incited severe hypocotyl decay, little or no root rot, and slight stunting; whereas *P. ultimum* caused root necrosis, limited hypocotyl decay, stunting, and considerable loss in dry weight (Table 1, Fig. 1). Root-rot severity and stunting were greater for plants grown in soil infested with a combination of *P. ultimum* and *F. solani* f. sp. *phaseoli* (Table 1, Fig. 1). The interaction between the two pathogens apparently was synergistic since root-rot severity from the combination was greater than the sum of their individual effect. For example, *P. ultimum*, *F. solani* f. sp. *phaseoli*, or both together caused 52, 12, and 74% reduction, respectively, in dry weight per plant and the root-rot severities were 3.2, 0.8, and 5.1, respectively (Table 1). Reduction of plant heights followed a similar pattern (Table 1, Fig. 1).

Infection by *R. solani* alone resulted in restricted hypocotyl and root lesions, but there was no evidence of an interaction between *R. solani* and *F. solani* f. sp. *phaseoli*. Dry weight of plants grown in soil infested with *R. solani*, *F. solani* f. sp. *phaseoli*, or both together was reduced 14, 12, and 27%, respectively. Similarly, hypocotyl and root-rot severity was not increased significantly in soil infested with a combination of the two pathogens. Root-rot severity ratings of plants grown in soil infested with *R. solani*, *P. ultimum*, or both together was 1.0, 3.2, and 2.4, respectively, which indicates that the two were antagonistic. The same treatments caused 14, 52, and 42% reduction, respectively, in dry weight (Table 1, Fig. 1).

At the termination of each experiment, the pathogen(s) added to soil always was recovered from hypocotyl or root tissues of the infected plants. However, none of these pathogens was isolated from plants growing in noninfested soil.

TABLE 1. Effect of bean root-rot pathogens singly and in combination on root-rot development and growth response of snap beans grown in pasteurized soil in a controlled environment

Treatment[w]	Disease rating[x] Hypocotyl	Disease rating[x] Root	Dry wt/ plant (g)	Dry wt loss (% control)	Plant ht (cm)[y]
Control	0.0 e[z]	0.0 f	1.45 a	...	13.1 a
Pythium	1.6 d	3.2 b	0.69 e	52	9.7 e
Fusarium	3.6 c	0.8 e	1.28 b	12	11.9 bc
Rhizoctonia	1.3 d	1.0 d	1.25 b	14	12.3 ab
Phythium + Fusarium	5.0 a	5.1 a	0.38 g	74	5.2 g
Pythium + Rhizoctonia	1.2 d	2.4 c	0.84 d	42	10.8 d
Fusarium + Rhizoctonia	3.4 c	1.0 d	1.06 c	27	11.0 cd
Pythium + Fusarium + Rhizoctonia	4.5 b	4.9 a	0.46 f	68	7.2 f

[w]*Pythium ultimum* and *F. solani* f. sp. *phaseoli* were incorporated into the soil at the rate of 500 propagules/g oven-dry soil. *Rhizoctonia solani* inoculum was mixed into soil at the rate of 5.5 g/kg oven-dry soil.

[x]Disease-severity rating was based on a scale of 0 to 6 with 0 = no symptoms and 6 = most severe symptoms (dead plant).

[y]Plant height was measured from the soil surface to the base of the petiole of the uppermost leaf on each plant.

[z]Means in a column followed by the same letter do not differ significantly ($P = 0.05$) by the Waller-Duncan's Bayesian K-ratio (LSD) rule.

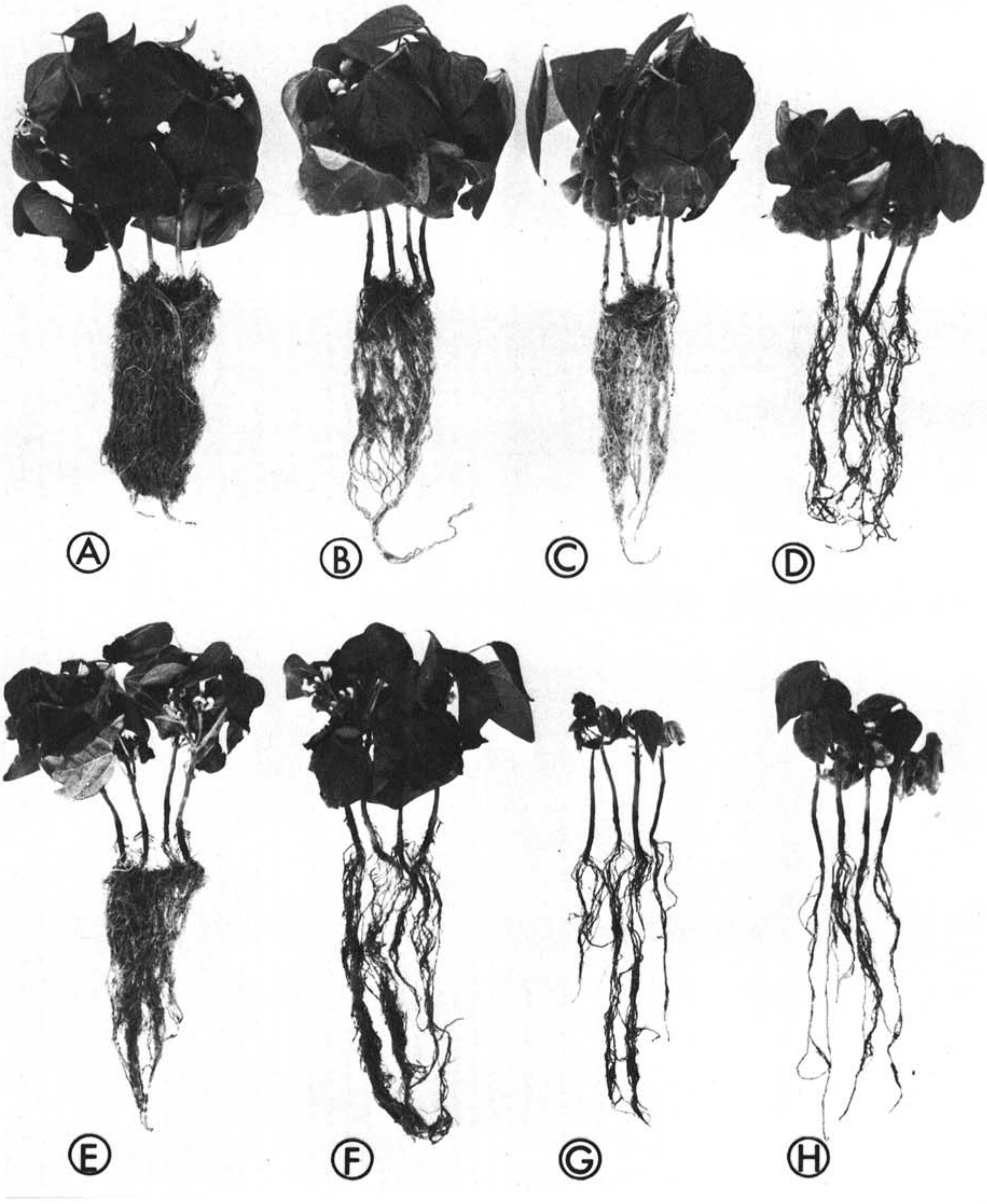

Fig. 1-(A to H). Root-rot symptoms and stunting on 28-day-old snap beans grown in **A)** pasteurized, noninfested soil and in pasteurized soil infested with **B)** *Fusarium solani* f. sp. *phaseoli*; **C)** *Rhizoctonia solani*; **D)** *Pythium ultimum*; **E)** *F. solani* f. sp. *phaseoli* and *R. solani*; **F)** *P. ultimum* and *R. solani*; **G)** *P. ultimum* and *F. solani* f. sp. *phaseoli*; **H)** *P. ultimum*, *R. solani*, and *F. solani* f. sp. *phaseoli*. Note differences in hypocotyl and root necrosis and plant stunting.

The synergistic interaction between *P. ultimum* and *F. solani* f. sp. *phaseoli* also was observed in pasteurized soil without the addition of Dexon (Table 2, Fig. 2). Soil application of Dexon did not reduce damage caused by *F. solani* f. sp. *phaseoli* alone, but did prevent hypocotyl and root necrosis incited by *P. ultimum* alone, although the fungus was occasionally reisolated from the roots. Disease severities and loss in dry weight were reduced greatly by application of Dexon to soil infested with both pathogens. Furthermore, only symptoms typical of those incited by *F. solani* f. sp. *phaseoli* were evident on plants grown in Dexon-treated soil. Plants grown in

TABLE 2. Effect of soil application of Dexon on severity of bean root rot caused by *Pythium ultimum* and *Fusarium solani* f. sp. *phaseoli* in pasteurized bean-field soil

	Disease rating[y]					
	Hypocotyl		Root		Dry wt/plant (g)	
Treatment[x]	No Dexon	With Dexon	No Dexon	With Dexon	No Dexon	With Dexon
Control	0.0 d[z]	0.0 d	0.0 e	0.0 e	0.67 a	0.57 bc
Pythium	1.0 c	0.0 d	2.9 b	0.0 e	0.41 e	0.57 bc
Fusarium	3.4 b	3.6 b	0.8 d	1.8 c	0.62 ab	0.47 d
Pythium + *Fusarium*	5.1 a	3.5 b	5.1 a	1.9 c	0.30 f	0.54 c

[x] *Pythium ultimum* and *F. solani* f. sp. *phaseoli* were incorporated into the soil at the rate of 500 propagules/g of oven-dry soil.
[y] Disease-severity rating was based on a scale of 0 to 6 with 0 = no symptoms and 6 = most severe symptoms (dead plant).
[z] Means followed by the same letter within and between the no-Dexon and with-Dexon columns, under each individual heading (hypocotyl, root, or dry wt) only, do not differ significantly ($P = 0.05$) by the Waller-Duncan's Bayesian K-ratio (LSD) rule.

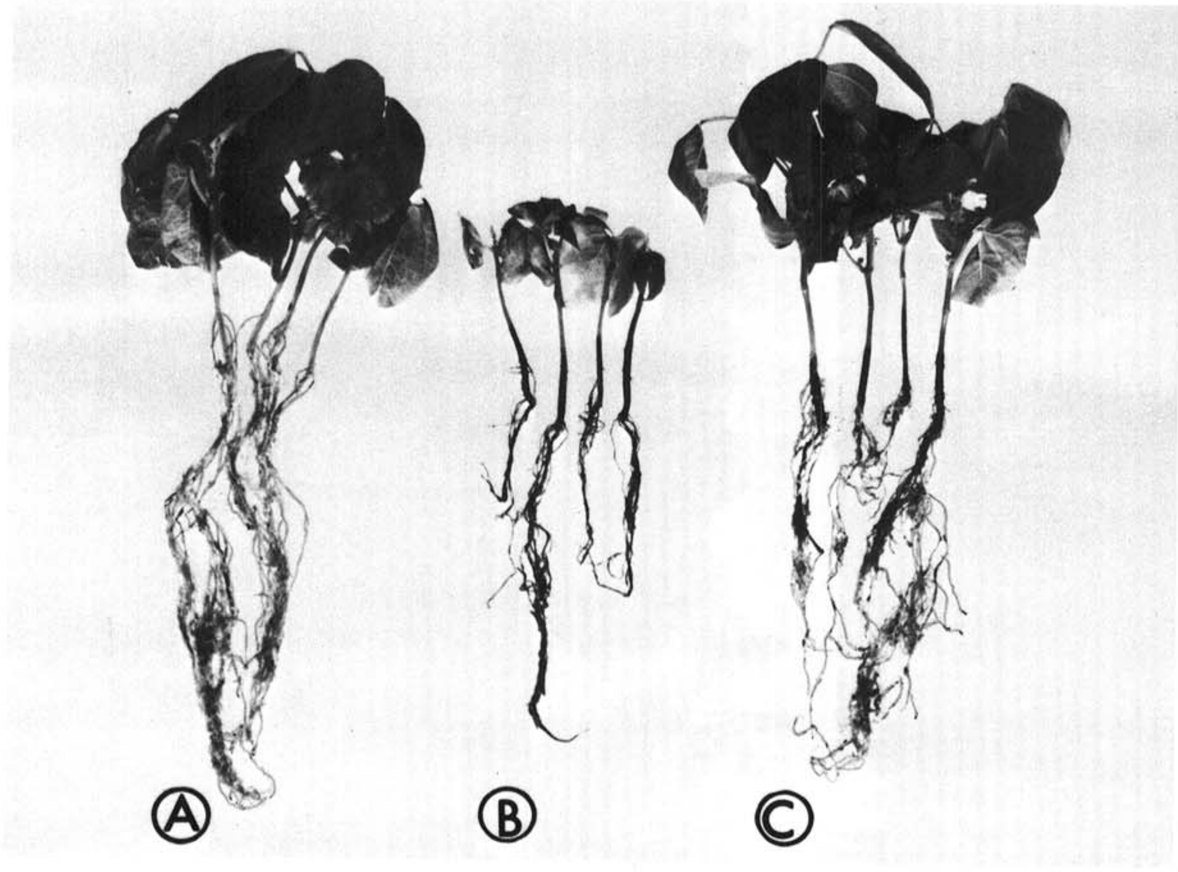

Fig. 2-(A to C). Effect of soil application of Dexon on severity of bean root rot in pasteurized soil infested with *Pythium ultimum* and *Fusarium solani* f. sp. *phaseoli*. **A)** Beans grown in pasteurized, noninfested soil treated with Dexon. **B)** Plants grown in infested soil without Dexon. **C)** Plants grown in infested soil with Dexon.

pasteurized, noninfested soil treated with Dexon (control) were slightly reduced in dry weight as compared with those grown in pasteurized soil without Dexon, suggesting that Dexon was slightly phytotoxic.

Plants grown in nonpasteurized bean soil treated with Dexon had significantly lower hypocotyl- and root-rot disease ratings and greater dry weights than plants grown in soil without the fungicide (Table 3). Only symptoms typical of those caused by *F. solani* f. sp. *phaseoli* and occasionally *T. basicola* were observed on plants grown in Dexon-treated nonpasteurized soil. Both fungi were isolated from surface-sterilized tissues of such plants on PDA. However, *Pythium* spp. were isolated from root segments of some and all plants grown in the Dexon-treated and nontreated soils, respectively.

Similar results were obtained when these experiments were repeated.

DISCUSSION

Pythium ultimum is considered to be the principal pathogen causing seed decay and preemergence damping-off of snap beans in New York State (5) and it also is an important root pathogen on older bean plants (18). The synergistic interaction between *P. ultimum* and *F. solani* f. sp. *phaseoli* demonstrated in this study may not be operative at times when soil conditions are not favorable for *P. ultimum* activity (18).

According to growers and processors, only about 60% of the snap bean acreage was harvested and yield was reduced considerably during the growing season of 1976 when weather conditions in New York were cool and wet. Root rot was the major factor contributing to these losses; root and aboveground symptoms on infected plants were similar to those reproduced under our controlled conditions with soil infested with a combination of *P. ultimum* and *F. solani* f. sp. *phaseoli*. Typical symptoms were severe necrosis of root and hypocotyl, stunting of plants, yellowing of foliage, death of lower leaves, and eventual death of plants. Both *P. ultimum* and *F. solani* f. sp. *phaseoli* were readily isolated from these plants.

Only limited information is available concerning the importance of *Pythium* species in the bean root-rot complex (5, 9, 15, 22). However, several interactions involving one or more *Pythium* species are known to occur on other hosts (8). Escobar et al. (6) reported that the combination of *Pythium* sp. and *F. solani* f. sp. *pisi* resulted in more severe root rot of peas than that caused by either pathogen alone. Kerr (12) reported that interaction between *F. oxysporum* f. sp. *pisi* and *P. ultimum* on peas resulted in more disease than either pathogen produced alone. He suggested that infection of roots by *Pythium* spp. favors infection by *F. oxysporum* which otherwise does not readily infect healthy roots. As shown in this study, *F. solani* f. sp. *phaseoli* is primarily a bean hypocotyl pathogen and causes only limited damage of roots. Burke (1, 2, 3) has shown that, whereas *F. solani* f. sp. *phaseoli* does not readily attack healthy bean roots, infection is more severe when roots are wounded or their growth is restricted by other causes. Therefore, damage to healthy roots by *P. ultimum* may predispose beans to infection by *F. solani* f. sp. *phaseoli*, resulting in more damage than produced by either pathogen alone.

Root-rot severity and stunting caused by *P. ultimum* were significantly greater than in soil infested with a combination of *P. ultimum* and *R. solani*. Klein (13) reported a similar reduction in damage to soybeans in soil infested with a combination of *Phytophthora* sp. and *Rhizoctonia* sp. as compared with soil infested with *Phytophthora* sp. alone. Similarly, Flowers and Littrell (7) reported that certain isolates of *R. solani* were antagonistic to *P. aphanidermatum* in sterilized and nonsterilized soil. The *R. solani* isolate used in our study also may be antagonistic to *P. ultimum*, but more work is needed to determine the nature of this interaction.

LITERATURE CITED

1. BURKE, D. W. 1965. The near immobility of Fusarium solani f. phaseoli in natural soils. Phytopathology 55:1188-1190.
2. BURKE, D. W. 1966. Predisposition of bean plants to Fusarium root rot. Phytopathology 56:872 (Abstr.).
3. BURKE, D. W. 1968. Root growth obstructions and Fusarium root rot of beans. Phytopathology 58:1575-1576.
4. BURKHOLDER, W. H. 1919. The dry root-rot of the bean. Cornell Univ. Agric. Exp. Stn. Mem. 26:1003-1033.
5. DICKSON, M. H., and G. S. ABAWI. 1974. Resistance to Pythium ultimum in white-seeded beans (Phaseolus vulgaris). Plant Dis. Rep. 58:774-776.
6. ESCOBAR, C., M. K. BEUTE, and J. L. LOCKWOOD. 1967. Possible importance of Pythium in root rot of peas. Phytopathology 57:1149-1151.
7. FLOWERS, R. A., and R. H. LITTRELL. 1973. Influence of Rhizoctonia solani on population densities of Pythium aphanidermatum in soil. Abstract 1058. 2nd Int. Congr. Plant Pathol., 5-12 September 1973, Minneapolis, Minnesota (unpaged).
8. HENDRIX, F. F., JR., and W. A. CAMPBELL. 1973.

TABLE 3. Effect of soil application of Dexon on severity of bean root rot in nonpasteurized bean-field soil with a history of root rot

	Disease rating[y]				Dry wt plant (g)	
	Hypocotyl		Root			
Treatment	No Dexon	With Dexon	No Dexon	With Dexon	No Dexon	With Dexon
Field 1	2.8 a	2.5 a	3.1 a	1.3 b	0.52 b	0.59 a
Field 2	2.8 a	0.9 b	3.9 a	2.6 b	0.47 a	0.57 a

[y]Disease-severity rating was based on a scale of 0 to 6 with 0 = no symptoms and 6 = most severe symptoms (dead plant).

[z]Means followed by the same letter between the no-Dexon and with-Dexon columns, under each individual heading (hypocotyl, root, or dry wt) for each field, do not differ significantly ($P = 0.05$) by the Waller-Duncan's Bayesian K-ratio (LSD) rule.

Pythiums as plant pathogens. Annu. Rev. Phytopathol. 11:77-98.
9. HOCH, H. C., D. J. HAGEDORN, D. L. PINNOW, and J. E. MITCHELL. 1975. Role of Pythium spp. as incitants of bean root and hypocotyl rot in Wisconsin. Plant Dis. Rep. 59:443-447.
10. HUTTON, D. G., R. E. WILKINSON, and W. F. MAI. 1973. Effect of two plant-parasitic nematodes on Fusarium dry root rot of beans. Phytopathology 63:749-751.
11. KEPLINGER, J. A., and G. S. ABAWI. 1976. Plant-parasitic nematodes associated with snap bean roots and soils in New York State. Proc. Am. Phytopathol. Soc. 3:307 (Abstr.).
12. KERR, A. 1963. The root rot-Fusarium wilt complex of peas. Austr. J. Biol. Sci. 16:55-69.
13. KLEIN, H. H. 1959. Etiology of the Phytophthora disease of soybeans. Phytopathology 49:380-383.
14. KO, W., and F. K. HORA. 1971. A selective medium for the quantitative determination of Rhizoctonia solani in soil. Phytopathology 61:707-710.
15. KRAFT, J. M., and D. W. BURKE. 1971. Phythium ultimum as a root pathogen of beans and peas in Washington. Plant Dis. Rep. 55:1056-1060.
16. KREUTZER, W. A. 1963 Selective toxicity of chemicals to soil microorganisms. Annu. Rev. Phytopathol. 1:101-126.
17. MALOY, O. C. 1959. Microbial associations in the Fusarium root rot of beans. Plant Dis. Rep. 43:929-933.
18. PIECZARKA, D. P., and G. S. ABAWI. 1978. Populations and biology of Pythium species associated with snap bean roots and soils in New York. Phytopathology 68:409-416.
19. POWELL, N. T. 1971. Interactions between nematodes and fungi in disease complexes. Annu. Rev. Phytopathol. 9:253-274.
20. TSAO, P. H., and G. OCANA. 1969. Selective isolation of species of Phytophthora from natural soils on an improved antibiotic medium. Nature (Lond.) 223:636-638.
21. WALLER, R. A., and D. B. DUNCAN. 1969. A Bayes rule for the symmetric multiple comparisons problem. J. Am. Stat. Assn. 64:1484-1503.
22. ZAUMEYER, W. J., and H. R. THOMAS. 1957. A monographic study of bean diseases and methods for their control. U.S. Dep. Agric. Tech. Bull. 868. 255 p.