Influence of Soil Water Potential and Temperature on Severity of Pythium Root Rot of Snap Beans

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ABSTRACT

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Soil water and temperature had significant effects on the severity of root rot of snap beans caused by *Pythium ultimum*. The disease was studied in pasteurized soil infested with the pathogen and maintained at fluctuating water potentials of 0 to −1, 0 to −5, and 0 to −12 bars, each at 15, 21, and 27 C. At any one temperature, root-rot severity increased as soil water potential increased. At any one water potential, root-rot severity decreased as soil temperature increased. Pythium root rot was most severe and caused the greatest plant dry weight loss in soil at 15 C and soil water potential of 0 to −1 bar. When plants grown in infested soil for 15 days at 15 C and at soil water potential of 0 to −1 bar were shifted to 27 C and/or soil water potential 0 to −5 bars for another 15 days, they had greater dry weight and less root rot than did plants maintained in soil at 15 C and 0 to −1 bar for 30 days. Damage from *Pythium* also occurred, although to a lesser extent, when plants were grown at 27 C and soil water potential of 0 to −12 bars for 15 days and then shifted to a 15 C and/or soil water potential of 0 to −1 bar. Soil populations of *P. ultimum* increased at all temperatures and soil water potentials tested.

Additional key words: *Phaseolus vulgaris*, epidemiology, soil moisture.

In New York State, root rot of snap beans (*Phaseolus vulgaris* L.) causes economic losses annually. Furthermore, the incidence and severity of root rot has increased steadily over the last decade and now is considered to be a limiting factor in the production of snap beans, particularly under cool, wet conditions. *Pythium ultimum* Trow, the principal seed-decay and damping-off pathogen of beans (5), also has been shown to play a major role in the bean root-rot complex in New York (17, 18). Moreover, *P. ultimum* and *Fusarium solani* (Mart.) Appel & Wr. f. sp. *phaseoli* (Burk.) Snyd. & Hans. may interact synergistically to cause root rot of snap beans (17). However, there are only few studies available that quantitatively describe environmental conditions that influence disease expression on beans (9). Considerable yearly variations in root-rot severity have been observed within and between bean fields with a history of severe root rot in New York State. Field observations suggest that soil water status and temperature greatly influence the incidence and severity of root rot. These environmental factors may cause the annual variations in the severity of root rot.

This paper reports the influence of fluctuating soil water potentials and constant soil temperatures on the severity of Pythium root rot of snap beans. A preliminary account of this work has been published (16).

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MATERIALS AND METHODS

A sporangium isolate of *P. ultimum*, from roots of snap bean from a field in New York (18), was used. The culture was maintained by periodic transfers on plates of cornmeal agar incubated at 21 C. For all experiments, inoculum was prepared by the methods described previously (18) and incorporated into pasteurized soil (sandy loam, pH 6.8, and 2.8% organic matter) at the rate of 500 sporangia/g oven-dry soil. The soil was collected from a bean field with a long history of severe root rot. The water content of the soil at water potential values of 0, −0.01, and −0.04 bar were 0.571, 0.474, and 0.334 g water per gram oven-dry soil, respectively.

The effect of fluctuating soil water potential and constant or fluctuating soil temperature on root rot was studied in controlled temperature tanks (6). The tanks were located in a growth chamber maintained at 21 C, 75-80% relative humidity, and 11,000 lux of cool-white fluorescent light for 14 hr/day.

Soil water potential was measured using soil thermocouple psychrometers and an HR-33 dewpoint microvoltmeter (Wescor Inc., Logan, UT 84321). Prior to each experiment the psychrometers were cleaned and calibrated at the different temperatures used. One psychrometer was placed in the center of each 12.5-cm-diameter plastic pot 5 cm below the soil surface. A perforated, polyvinyl chloride watering tube (1-cm diameter) was placed down the center of each pot to facilitate more uniform distribution of water throughout the soil mass. Four 8-day-old bean seedlings of the cultivar Early Gallatin were transplanted into each pot as described previously (18). Immediately after transplanting, all pots were watered uniformly. The soil water potential for each pot was determined daily. Depending on the soil temperature, decreases in soil water potential were usually detected 3-4 days after each experiment was initiated. The appropriate volume of water was added to each pot to readjust the water potential to near 0 bar whenever the lower limit of each preselected water potential value was reached. Three soil water potentials (ψ) of 0 to −1, 0 to −5, and 0 to −12 bars were maintained and henceforth will be referred to as wet, intermediately dry, and dry soil treatments, respectively. Each pot received 0.5 g of a complete fertilizer (20-20-20) at 1 and 15 days after transplanting.

In the first series of experiments, bean seedlings were transplanted into wet, intermediately dry, and dry soils, each maintained at a constant temperature of either 15, 21, or 27 C for 30 days. In a second series of experiments, plants were grown at 15 C and either in wet or intermediately dry soils. After 15 days, half of the plants grown in each of the wet and intermediately dry soils were shifted to 27 C. The other half was maintained at 15 C, but the soil water status was reversed from wet to intermediately dry (ψ = 0 to −1 to ψ = 0 to −5 bars) and vice versa. In this same experiment, plants also were started at 27 C and shifted to 15 C after 15 days. The soil water status was similarly altered from wet to intermediately dry and vice versa.

Thirty days after transplanting, the plants were removed from the pots, and the roots were washed and visually rated for root-rot severity. Roots and hypocotyls were rated separately using a scale of 0 to 6 for which 0 represents the absence of disease symptoms, and 6 refers to a dead root or hypocotyl. A rating of one represents light discoloration of the tissues without visible necrotic lesions. Ratings of 2, 3, 4, and 5 indicate that up to 25, 26 to 50, 51 to 75, and over 75% of the roots or hypocotyl tissues exhibit dark discoloration and necrosis. Root tissues from all treatments were plated on water agar to confirm the presence of *Pythium*. The soil population of *P. ultimum* was determined for the treatments where soil moisture and temperature remained constant for 30 days. All samples were passed through a 1.68-mm-mesh screen and mixed thoroughly. Soil dilution series of 1:10, 1:50, and 1:100 (w/v) were made in 0.03% sterile water agar. The surface-soil-dilution plate technique was used as described previously (18) except the selective medium of Tsao and Ocana (22) was used. The plates were incubated in the dark at 20 C, and colony counts were made 24 and 48 hr later and recorded as the number of propagules/g oven-dry soil. The plants were dried for 48-72 hr at 80 C and weighed.

All experiments were repeated once and each treatment was replicated three times. Data were analyzed by the Waller-Duncan's Bayesian K-ratio (LSD) rule (23). Data in tables are those of representative experiments.

RESULTS

The severity of root rot and plant stunting caused by *P. ultimum* decreased as available soil water was decreased and temperature was increased (Table 1, Fig. 1). The most severe symptoms were extensive stunting, pruning, and discoloration of the roots (Fig. 1-A, B, C, D). There were very few symptoms on the hypocotyls. At 15 C, the root-rot severity rating was 5.0, 3.8, and 2.8; and the dry weight

TABLE 1. Influence of fluctuating soil water potential (ψ) and constant temperature on the population of *Pythium ultimum* and severity of bean root rot

ψ Soil (bars)	Soil temp (C)	Disease severity rating[t] Hypocotyl	Root	Total dry weight per plant Control (g)	Infected (g)	Dry weight[u] (% Control)	*P. ultimum* propagules/g dry soil[vwx] (no.)
0 to −1	15	1.2 a[y]	5.0 a	2.05 b[z]	0.55 k	27	1200 g
	21	1.2 a	2.9 c	2.21 a	0.76 gh	34	2080 ef
	27	1.2 a	0.7 d	1.69 c	1.32 d	78	4640 b
0 to −5	15	1.5 a	3.8 b	1.00 e	0.73 ghi	73	3960 bc
	21	0.2 b	0.3 de	0.84 fg	0.90 ef	107	7040 a
	27	0.1 b	0.0 e	0.78 fgh	0.80 fgh	102	2560 de
0 to −12	15	1.8 a	2.8 c	0.76 gh	0.79 fgh	104	3320 cd
	21	0.0 b	0.2 e	0.70 hij	0.62 ijk	89	2800 de
	27	0.0 b	0.0 e	0.68 hij	0.58 jk	85	1320 fg

[t]Hypocotyls and roots were rated 30 days after transplanting using a scale of 0 to 6; 0 = no apparent disease symptoms and 6 = most severe symptoms. Disease ratings for all controls were 0.

[u]Total dry weight of infected plants presented as percent of the dry weight of the control.

[v]Initial population in infested soil was 440 propagules/g dry soil.

[w]Each calue is the mean of five replicate plates per soil treatment.

[x]*Pythium ultimum* was not detected in soil of the controls.

[y]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.

[z]Comparisons for significance are applicable within and between columns for Control and Infected.

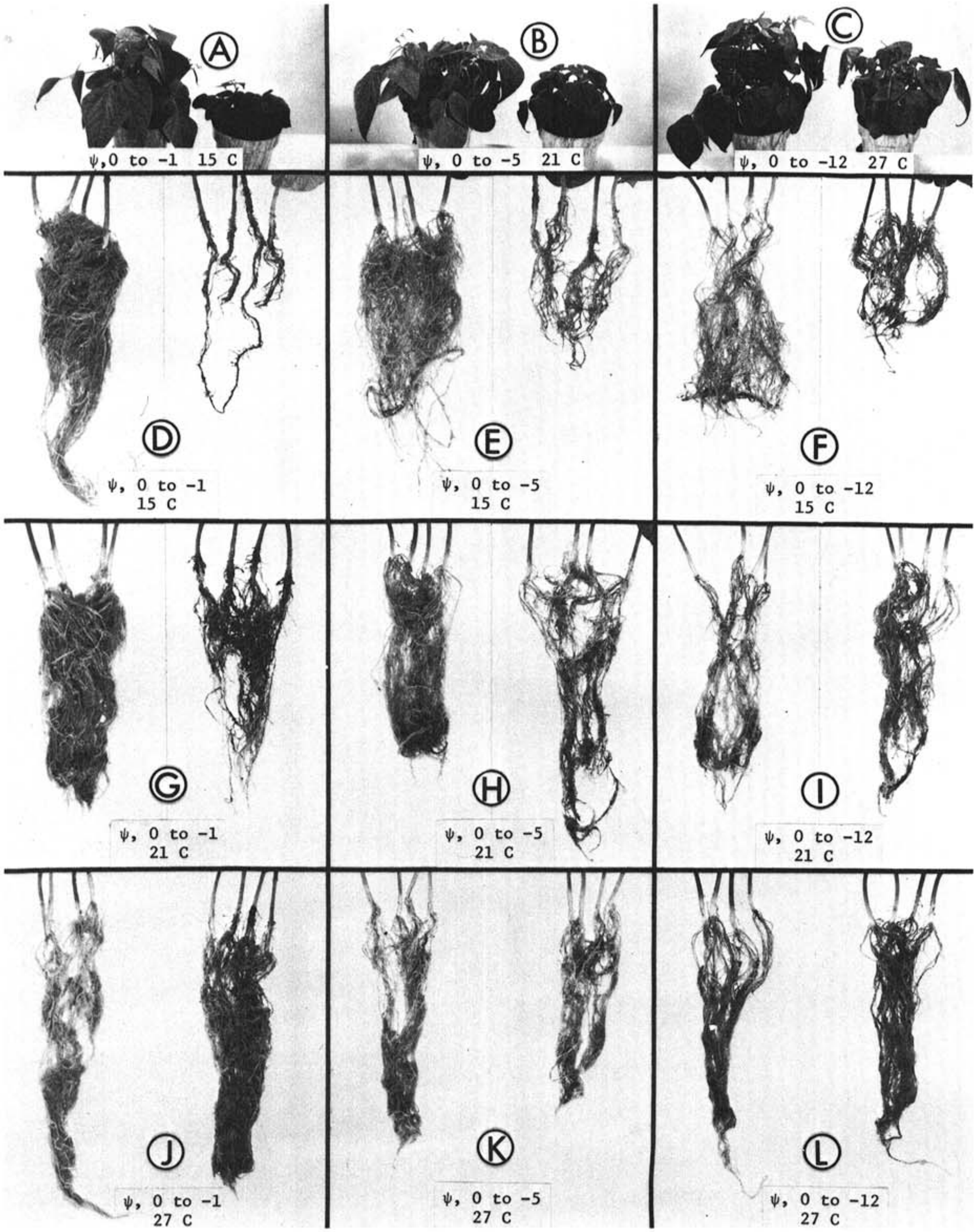

Fig. 1-(A to L). Bean plants (A-C) and roots of plants (D-L) grown in Pythium-infested pasteurized soil for 30 days at different fluctuating soil water potentials (ψ) and constant temperatures. Plants or roots on the left in each set were grown in noninfested pasteurized soil. Soil treatments were: **A)** $\psi = 0$ to -1 bar at 15 C; **B)** $\psi = 0$ to -5 bars at 21 C; **C)** $\psi = 0$ to -12 bars at 27 C; **D-F)** $\psi = 0$ to -1, 0 to -5, and 0 to -12 bars, respectively, at 15 C; **G-I)** $\psi = 0$ to -1, 0 to -5, and 0 to -12 bars, respectively, at 21 C; and **J-L)** $\psi = 0$ to -1, 0 to -5, and 0 to -12 bars, respectively, at 27 C.

of infected plants was 27, 73, and 104% of the controls in the wet ($\psi = 0$ to -1 bar), intermediately dry ($\psi = 0$ to -5 bars), and dry ($\psi = 0$ to -12 bars) soil treatments, respectively. Water status was limiting for plant growth in the dry soil treatment ($\psi = 0$ to -12 bars) which resulted in no significant difference in dry weights between the control and infected plants (100% vs. 104%) in spite of a disease rating of 2.8. However, a rating of 2.9 reduced plant dry weight to 34% of that of the control in the wet soil treatment at 21 C. At 21 C, only limited disease developed in the intermediately dry and the dry soil treatments, and plant dry weight was not significantly different from the controls. At 27 C, only limited plant stunting and root necrosis and discoloration were evident, even in the wet soil treatment; however, there was a significant loss in dry weight of plants in this treatment. No root necrosis was apparent in the intermediately dry and dry soil treatments at 27 C, and the root systems were comparable to those of the control treatments (Fig. 1-G, H, I).

Factorial analysis of dry weights showed that temperature and moisture both influenced disease development significantly ($P = 0.05$); however, there was no significant interaction between temperature and moisture.

Pythium ultimum was reisolated from the roots and hypocotyls of plants grown in infested soils in all treatments. The population of *P. ultimum* in soil at the termination of each experiment was 3 to 16 times greater than the initial population of 440 propagules/g (Table 1). In the wet soil treatment, the population increased with increasing temperature. However, in the intermediately dry and dry soil treatments, the population tended to decrease with increasing temperature.

In comparison to plants maintained at a constant 15 C for 30 days, root-rot severity was less and dry weight was more when plants were grown in Pythium-infested soil for only 15 days at 15 C and the other 15 days at 27 C, regardless of whether the warmer temperature was used during the first or last 15-day period (Table 2, Fig. 2). This was also true regardless of whether plants were grown in the wet soil treatment for the entire 30 days; in the wet and intermediately dry soil treatments for the first and last 15-day period, respectively; or the reverse of the latter treatment. However, plant dry weight was greater in treatments that combined both the dry soil treatments and high temperature. Furthermore, disease ratings were lower and dry weights greater for plants grown in infested

TABLE 2. Influence of fluctuating soil water potential (ψ) and temperature on severity of Pythium root rot of snap beans

Incubation period	Soil (bars)[u] (ψ)	Soil temp[v] (C)	Disease severity rating[w]		Total dry weight per plant[y]		Dry weight[z] (% Control)
			Hypocotyl	Root	Control (g)	Infected (g)	
1st 15 days 2nd 15 days	0 to −1 0 to −1	15 15	2.0 abc[x]	5.0 a	1.40 b	0.43 jk	31
1st 15 days 2nd 15 days	0 to −1 0 to −1	15 27	1.8 bcd	3.3 b	1.28 c	0.53 hi	41
1st 15 days 2nd 15 days	0 to −1 0 to −1	27 15	1.8 bcd	3.2 bc	1.55 a	0.76 f	49
1st 15 days 2nd 15 days	0 to −1 0 to −5	15 15	2.4 ab	5.0 a	0.87 e	0.32 l	37
1st 15 days 2nd 15 days	0 to −1 0 to −5	15 27	1.8 bcd	2.7 d	0.84 e	0.48 ij	57
1st 15 days 2nd 15 days	0 to −1 0 to −5	27 15	1.2 d	1.3 e	0.96 d	0.66 g	69
1st 15 days 2nd 15 days	0 to −5 0 to −1	15 15	1.6 cd	4.8 a	0.76 f	0.38 kl	50
1st 15 days 2nd 15 days	0 to −5 0 to −1	15 27	2.6 a	3.3 b	0.81 ef	0.42 jk	52
1st 15 days 2nd 15 days	0 to −5 0 to −1	27 15	1.5 cd	2.8 cd	0.83 ef	0.60 gh	72

[u]Refers to the soil ψ values (bars) that were maintained during the first and second 15 days of plant growth.

[v]Refers to the soil temperatures that were maintained during the first and second 15 days of plant growth.

[w]Hypocotyls and roots were rated at 30 days after transplanting using a scale of 0 to 6; 0 = no apparent disease symptoms, 6 = most severe symptoms. Disease ratings for all controls were 0.

[x]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.

[y]Comparisons for significance are applicable within and between columns for Control and Infected.

[z]Total dry weight of infected plants is presented as percent of the dry weight of the controls.

soil for 15 days first at 27 C and then for 15 days at 15 C as compared to plants started at 15 C and then grown at 27 C regardless of the soil water treatments. Increasing soil temperature to 27 C resulted in the growth of fibrous and adventitious roots that were white and without apparent disease symptoms.

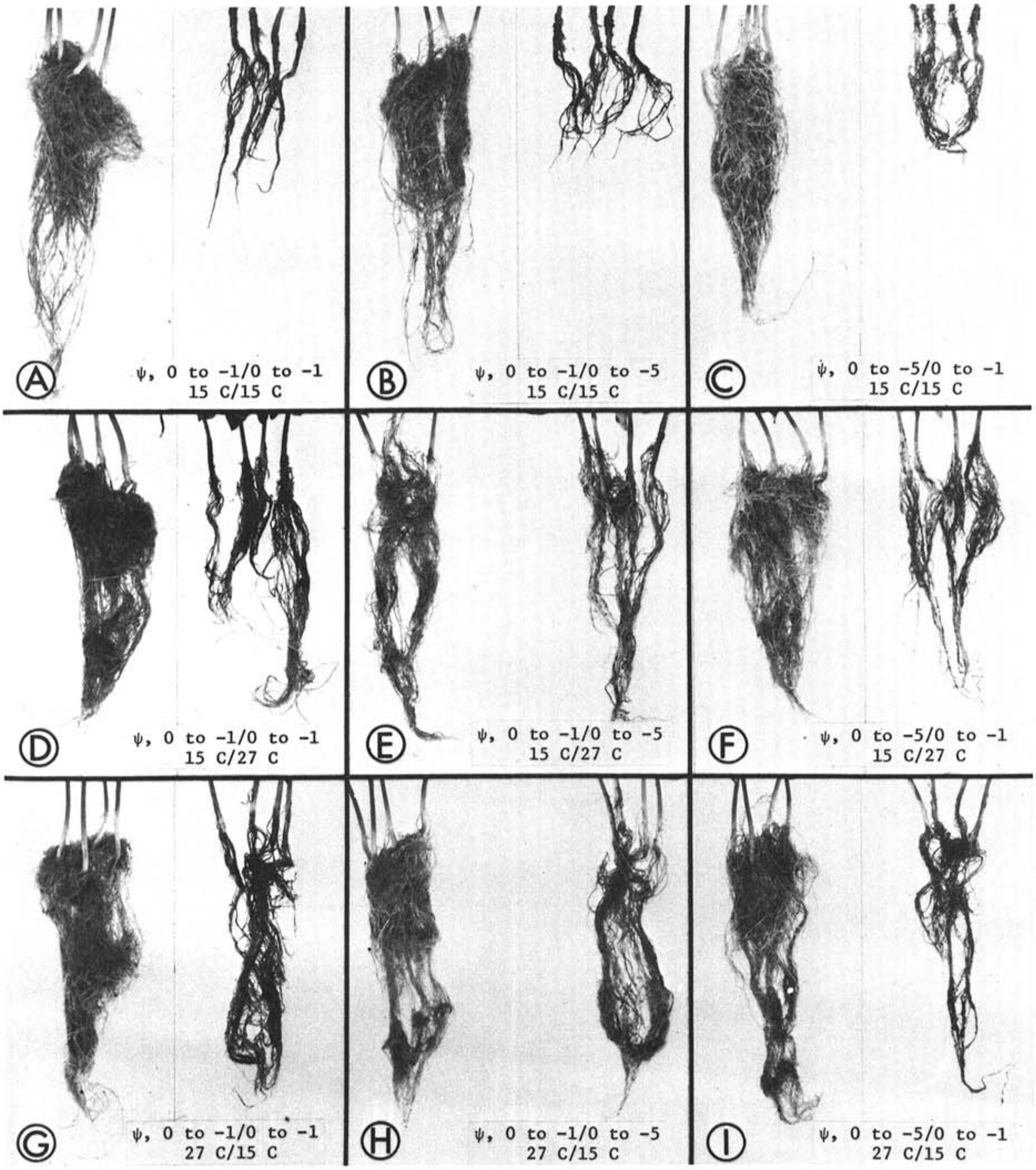

Fig. 2-(A to I). Influence of fluctuating soil water potential (ψ) and temperature on severity of Pythium root rot of beans when the initial soil ψ and/or temperature was changed 15 days after transplanting and was maintained for and additional 15 days. Roots on the left in each set were from plants grown in noninfested pasteurized soil. Soil treatment combinations for the first and second 15-day periods (15 days/15 days) were: **A)** $\psi = 0$ to $-1/0$ to -1 bar, **B)** $\psi = 0$ to $-1/0$ to -5 bars, and **C)** $\psi = 0$ to $-5/0$ to -1 bars, at 15 C; **D)** $\psi = 0$ to -1 bar/$\psi = 0$ to -1 bar, **E)** $\psi = 0$ to -1 bar/$\psi =$ to -5 bars, and **F)** $\psi = 0$ to -5 bars/$\psi = 0$ to -1 bar at 15 C/27 C, respectively; **G)** $\psi = 0$ to -1 bar/ψ, 0 to -1 bar, **H)** $\psi = 0$ to -1 bar/$\psi = 0$ to -5 bars and **I)** $\psi = 0$ to -5 bars/$\psi = 0$ to -1 bar at 27 C/15 C, respectively.

DISCUSSION

It is well known that soil moisture and temperature have a significant influence on the incidence and severity of root rot caused by *Pythium* species on several hosts (2, 3, 4, 9, 14, 15). In this study, root rot caused by *P. ultimum* was most damaging to beans in the wet soil treatment (ψ = 0 to −1 bar) at 15 C. Generally, root-rot severity decreased and dry weight of plants increased with increasing temperature (21 or 27 C) and decreasing soil moisture (ψ = 0 to −5 or 0 to −12 bars).

However, these results contradict those of Hoch et al. (9) who reported that damage to beans caused by *P. ultimum* was similar at 16 and 28 C. They also reported that bean root rot was more severe at a constant −10 mbars than at −200 mbars soil water potential, the lowest tested. A naturally infested soil was used in their study from which *P. aphanidermatum* (Edson) Fitz, *P. splendens* Braun, *P. ultimum, F. solani,* and *Rhizoctonia solani* Kuehn were isolated. Research dealing with a wide range of constant and variable soil water potentials is needed to further elucidate the role of soil water on the incidence and severity of root rot of beans caused by *P. ultimum* and other species.

Data presented in this paper showed that beans can produce new roots in a relatively short time and at least partially escape severe damage from *Pythium* if soil conditions are changed from those favorable (e.g., ψ = 0 to −1 bar at 15 C) to unfavorable (e.g., ψ = 0 to −12 bars at 27 C) to the pathogen. Furthermore, when plants were grown first in soil with conditions unfavorable for Pythium root rot and then soil conditions became favorable for the disease, root rot still developed, but not to the same extent observed with constant root-rot-favorable conditions. The latter confirms the finding that *P. ultimum* can be an important pathogen on roots of older bean plants (18).

High soil moisture has been reported to be necessary for survival, spore germination, and saprophytic activities of *Pythium* species (1, 13, 20). It also has been suggested that wet soils provide an ecological advantage for *Pythium* species which are, otherwise, poor competitors (7, 9, 20). High soil moisture (decreased oxygen) conditions also may affect host physiology as it increases root exudations which stimulate spore germination and ultimately increase disease incidence and severity (12, 20). The above factors, in addition to low temperature (8), contribute to the success of *P. ultimum* as a root-rot pathogen of beans in cool, wet soils.

The data suggesting that beans grown in infested soil with the wet treatment at 27 C may be stunted and have lower dry weight yet show no apparent root necrosis is of particular interest. *Pythium ultimum* and other species have been reported to reduce plant growth of a number of crops, even when damage to roots and/or disease symptoms cannot be directly established (10, 24). This suggests that *P. ultimum* may infect plant roots under a range of environmental conditions, but cause extensive damage only under cool, wet soil conditions that reduce the vigor of the host.

The population of *Pythium* spp. is known to increase in soils planted to susceptible hosts (11, 21). In this study, the population of *P. ultimum* increased markedly over a 30-day period in soil planted to beans. Significant increases over the initial population occurred regardless of soil moisture or temperature. Since *Pythium* spp. are not active in dry soils (13), population increases detected in the drier soil treatments probably occurred during the period of wetness immediately following each watering. Soil populations of *Pythium* spp. in natural bean field soils in New York State appear to remain relatively constant throughout the growing season (18). Possibly, the lack of a specific antagonistic microflora in the pasteurized field soil (60 C for 30 min) allowed the population of *P. ultimum* to increase more rapidly than normal.

Results of this study and those in the literature suggest that, for beans, proper selection of fields with coarse-textured, well-drained soils that warm up rapidly in the spring may reduce root rot in early season plantings. Soil moisture regulation by the use of raised beds or ridges (19) also may reduce bean root-rot severity under field conditions.

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