Ecology and Epidemiology of Rhizoctonia solani in Field Soil

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ABSTRACT

The ecology of *Rhizoctonia solani* was studied for more than 2 years in a field plot at the University of Maryland Vegetable Research Farm, Salisbury, Maryland. The highest inoculum density of *R. solani* (expressed as percentage of colonization of tablebeet seed supplied as bait) was in August and September, after the bean tissue of the first crop had been plowed under, and the lowest from February to June. Bean root rot was more pronounced in the second planting than in the first. The percentage of colonization was positively correlated with bean root rot in the field and greenhouse ($r = .9858$). Colonization also was positively correlated with total inorganic N and NH_4-N, but not with NO_3-N. Inoculum in the field was confined to the upper 10 cm of soil. From a total of 500 isolations made from the plot, 13 morphologically distinct isolates were obtained from soil and two from plant debris. Eleven of the 13 isolates from soil and both isolates from debris belonged to AG-4 ('Praticola'-type). Two isolates from soil belonged to AG-3. The isolates ranged from nonpathogenic to highly pathogenic on bean, cotton, lettuce, radish, and sugarbeet. Isolates R-27 and R-29 from the plot survived better in soil at 10, 15, and 20 C than at −5, 5, 25, and 30 C.

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During the past 25 years considerable knowledge has become available on the ecology of *Rhizoctonia solani* Kühn (19) and the epidemiology of diseases caused by this pathogen (2). For example, the growth, saprophytic behavior, and survival of *R. solani* in soil have been studied extensively (1, 6, 11, 19, 21, 23), as have variation and variability, virulence, and physiology of parasitism (2, 4, 14). With a few exceptions, however, most of the ecological research on *R. solani* has been done in the laboratory and greenhouse. Although studies with models may sometimes be extrapolated to the field, such studies often do not reflect the natural conditions and stresses to which pathogens are subjected in the field. This paper presents data obtained during a 2-year field study on the ecology and epidemiology of *R. solani* in an experimental field cropped continuously to snapbeans (*Phaseolus vulgaris* L.) for more than 10 years.

MATERIALS AND METHODS.—*Field selection and soil sampling.*—The ecological and epidemiological study of *R. solani* was done at the University of Maryland Vegetable Research Farm, Salisbury, in cooperation with the Department of Botany and Plant Pathology of the University of Maryland. A rectangular plot, approximately 0.2 hectare (ha) (33 × 83 m) in area, was selected for the long-range study. This plot had two crops per year of snapbeans for more than 10 years, and was naturally infested with *R. solani, Fusarium solani* f. sp. *phaseoli, Pythium* spp., and *Thielaviopsis basicola*. Soil samples were collected from each of 28 standard sites in the field and brought to the laboratory once a month for various biological and chemical analyses. About 3.5 kg of soil was collected with a trowel from each site to a depth of 15 cm. Because of the great volume of soil involved in 0.2 ha and the relatively small samples collected, we believe that repeated sampling from the same sites did not affect or bias the data. Soil samples were also collected occasionally at various depths to 30 cm, from various sites of the plot. The pH of the soil was 6.1 when it was brought to the laboratory.

Disease severity in naturally infested soil.—Disease severity was determined on both beans grown in the Salisbury plot and beans planted in soil samples brought to the greenhouse from the 28 sites. Two plantings were made in the field, one in May and the second in July. A rye cover crop, the predominant cover crop in the Delmarva Peninsula, was plowed under in late March or early April. The bean plants of the first crop were disked into the soil in late July or early August, and the second planting followed in a few days or was done the same day. Before beans were planted in May and July, 8-16-16 fertilizer was broadcast in the field at 1,120 kg/ha. Also, trifluralin at 1.17 liters/ha was added to the soil before planting.

In the fall of each year, the bean plants were plowed under and 1-2 days later a rye cover crop was planted. Dates of disking and planting for the spring and summer of 1971-73 were: 1971—bean plant remnants of the first planting disked into soil 2 August and beans planted 4 August; 1972—rye cover crop plowed under 11 April and beans planted 22 May; bean plant remnants disked into soil 27 July and beans planted the same day; 1973—rye cover crop plowed under 26 March and beans planted 4 May; bean plant remnants disked into soil 26 August and beans planted the same day. Bean field appearance and disease severity index (DSI) values on field-grown plants were usually obtained 3-4 weeks after planting. To obtain bean seed emergence and DSI values in the greenhouse, we planted 'Topcrop' bean seed each month in soil samples (1,500 g in No. 6 bulb pots) from the 28 sites in a 24 ± 3 C greenhouse.

Determination of saprophytic activity and survival.—Saprophytic activity of *R. solani* was assayed by a modification of a method previously described (20). One gram of autoclaved tablebeet (*Beta vulgaris* L.) seed, suggested for use to us by D. J. De Zeeuw of Michigan

State University, was mixed with 100 g soil in 9-cm diameter petri dishes. After 2 days of incubation in soil, the seed were recovered on a 1.41-mm screen, washed for 20 minutes in running tap water, and transferred to petri dishes (7-8 seeds per dish) containing 15 ml water agar (2% agar). Chlorotetracycline hydrochloride and streptomycin sulfate (50 mg/liter of each antibiotic) were added to the agar after it had been autoclaved and cooled to 48-50 C. Seed were examined with a compound microscope for *R. solani* colonization after the dishes were incubated at 25 C for 24 hours.

To test the effect of temperature on survival of *R. solani,* batches of Salisbury soil free of the pathogen were infested separately with isolates R-27 and R-29, originally obtained from the Salisbury plot. The two isolates were grown on sand-corn-meal for 3 weeks, and inoculum was added to moist soil at the rate of 2% (w/w). The infested soil batches were kept in the laboratory at approximately 24 C for 1 week and then subdivided into 0.8-kg portions. These were placed in 1-kg beakers and covered with polyethylene film to prevent loss of moisture. The infested soils, kept at −18 bars moisture (approximately 50% of moisture holding capacity), were incubated at −5, 5, 10, 15, 20, 25, and 30 C for 40 weeks. Periodically, 100 g soil were withdrawn, and the population surviving was determined by the tablebeet seed colonization method.

Isolation of Rhizoctonia solani.—Hyphal-tip isolations of the fungus growing from colonized seed on the antibiotic medium were made directly to potato-dextrose agar (PDA) slants. Comparative cultural and microscopic examinations of the transfers were made on PDA and Czapek-solution agar (CSA) at 25 C, 12 and 24 days after transferring. To obtain *R. solani* from debris particles, we fractionated soil samples by the method of Boosalis and Scharen (8). Fifty debris particles picked at random from soils from selected sites were transferred to water agar dishes (eight particles/dish). The particles were examined microscopically after 24 and 48 hours of incubation, and isolations were made as described before.

Anastomosis and heterokaryon formation among field isolates.—Anastomosis tests were performed by a method described by Parmeter et al. (22). The isolates of *R. solani* obtained from the Salisbury plot were opposed on sterile cellophane resting on water agar (2% agar) with the tester strains of Parmeter et al. (22). Heterokaryon formation among field isolates of *R. solani* was studied by methods recently described (7, 10).

Relative pathogenicity of isolates.—Thirteen isolates from the Salisbury plot, selected on the basis of morphological differences on PDA and CSA, were tested individually for pathogenicity on five different host plants: snapbean (cultivar Topcrop), cotton (*Gossypium hirsutum* L. 'Rowden'), lettuce (*Lactuca sativa* L. 'Boston'), radish (*Raphanus sativus* L. 'Comet'), and sugarbeet (cultivar US H2). Sand-corn-meal inoculum of *R. solani* was added at the rate of 0.5% (w/w) to soil treated with aerated steam at 83 C for 2 hours. Control soil was supplemented with noninoculated sand-corn-meal medium. After incubation of the inoculum in moist soil for 7 days, the infested soils were apportioned among the No. 5 plastic pots and planted to surface-disinfected seed of the six host (10 seeds per pot). Readings of cotton, lettuce, radish, and sugarbeet seedling emergence were taken 1, 2, and 4 weeks after planting, and the emergence counts made at 4 weeks were used to calculate total damping-off. Root rot of bean seedlings was assessed 4 weeks after planting. To calculate the DSI, we rated each seedling on an arbitrary scale from 0 (no visible infection)

TABLE 1. Correlation coefficients obtained by computer analysis among selected variables studied for 2 years in a bean field infested with *Rhizoctonia solani*

Variable	No. of times assayed	Correlation coefficient		
		Rhizoctonia colonization	*Rhizoctonia* DSI[a]	
			Greenhouse	Field
Rainfall	23	.2248	.3297	−.1164
Maximum temperature	23	.4644*[b]	.5571**	−.1325
Minimum temperature	23	.4741*	.5794**	−.3860
Rhizoctonia colonization	22	1.0000	.8537**	.9858*
Greenhouse *Rhizoctonia* DSI	21	.8537**	1.0000	.8940
Greenhouse bean seed emergence	21	−.2343	−.0256	−.9024
Greenhouse *Pythium* blight	21	.1418	.2538	.2176
P. aphanidermatum population	19	.1674	.0121	−1.0000
P. ultimum population	19	.0778	−.1788	−1.0000
Ammonium N	16	.7115**	.7442**	.6184
Nitrate N	16	.3752	.3682	−.5470
Total inorganic N	16	.5163*	.5225*	−.2208
Soil conductivity	9	−.2818	.5158	1.0000
Soil color	8	.1214	.2296	−.7892
Field *Rhizoctonia* DSI	6	.9858*	.8940	1.0000
Field plant stand	6	−.3465	.1948	.4272
Field plant weight	5	−.1714	−.5378	1.0000
Organic matter (%)	5	−.3437	−.5420	1.0000
Field appearance	4	.2411	.7961	1.0000
Total field DSI	3	−.9750	1.0000	0.0000
Field *Pythium* blight	3	1.0000	1.0000	.9244

[a]Disease severity index: 0 = healthy, 5 = plants dead.
[b]* = significant correlation at $P = 0.05$; ** = highly significant correlation at $P = 0.01$; − = negative correlation.

TABLE 2. Field and greenhouse evaluations of soil nitrogen content, substrate colonization by *Rhizoctonia solani* and root rot of beans (primary pathogen, *R. solani*) in soil obtained at various intervals from 28 locations in a single field

Month and year of sampling	Tablebeet seed colonization (%)	DSI[a]		Nitrogen content (μg/g)		
		Greenhouse	Field	Total inorganic	Ammonium	Nitrate
August 1971	44.3 A[b]	2.2 A	3.3 A	40.5 A	9.6 B	30.9 A
September 1971	43.6 A	2.2 A		14.6 CD	13.1 A	1.5 F
October 1971	39.2 A	2.1 A	3.2 A	11.6 DE	7.2 CD	4.3 DE
August 1972	25.1 B	1.3 B	1.7 B	10.6 EF	8.0 C	2.6 EF
September 1972	19.5 B	1.5 B		31.1 B	11.9 A	19.1 B
October 1972	13.3 C	0.7 C		11.7 DE	7.0 CD	4.7 D
February 1972	8.2 CD	0.7 C		8.0 G	5.1 E	2.9 EF
July 1972	3.7 DE	0.6 C	0.3 C	7.9 G	5.1 E	2.6 EF
February 1973	3.4 DE	0.6 C		13.5 D	6.4 DE	7.1 C
April 1972	1.4 E	0.4 CD		9.9 EFG	6.3 DE	3.7 DE
June 1972	1.4 E	0.3 CD	0.2 C	10.8 EF	6.3 DE	4.3 DE
June 1973	0.4 E	0.1 D	0.4 C	9.2 FG	6.1 DE	3.0 DEF

[a]Disease severity index: 0 = healthy, 5 = plants dead.
[b]Numbers followed by the same letter do not differ significantly ($P = 0.05$) by Duncan's multiple range test.

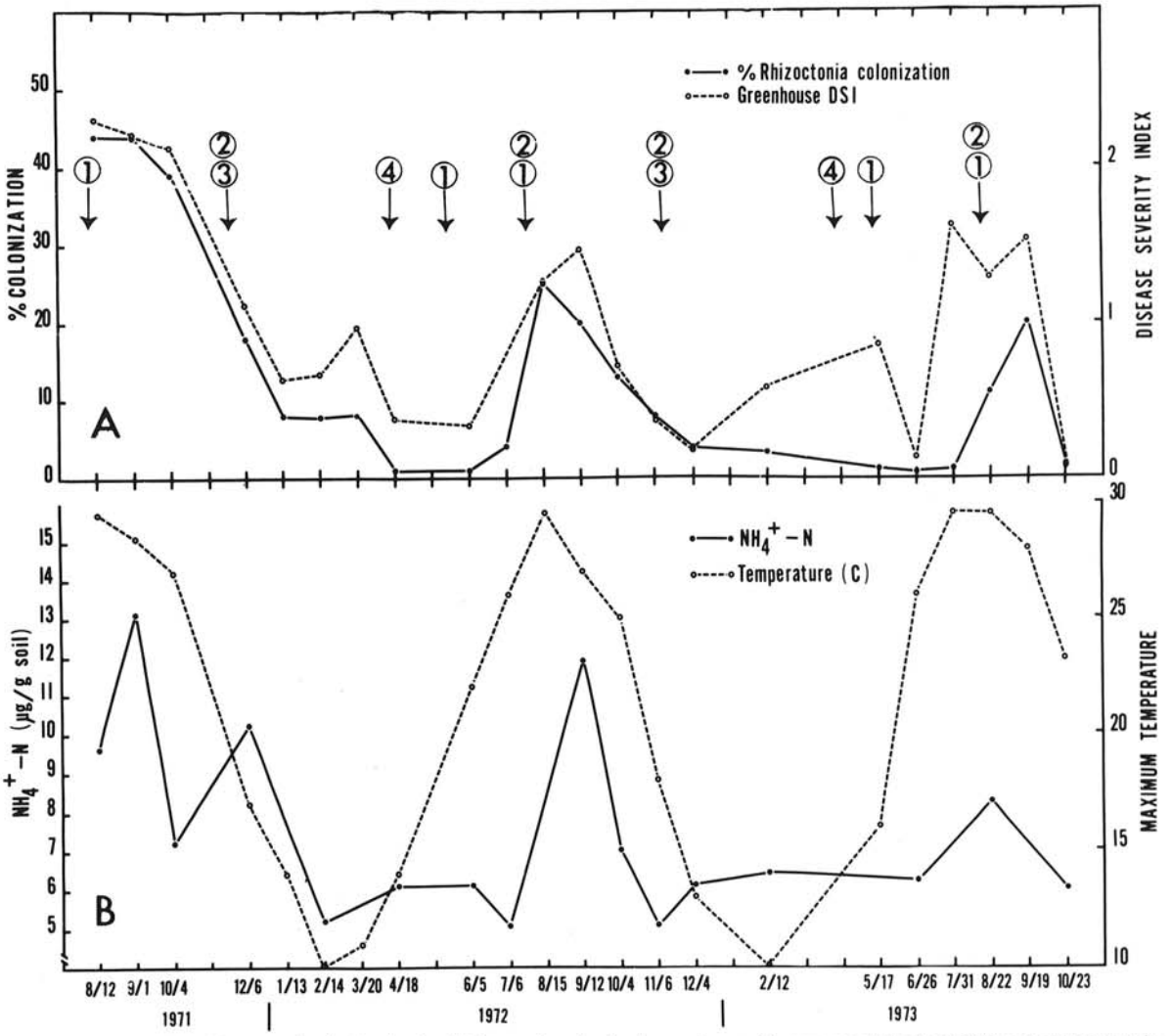

Fig. 1-(A, B). A) Tablebeet seed colonization by *Rhizoctonia solani* and greenhouse disease severity index (DSI) on beans grown in soil from 28 locations of a field collected at monthly intervals in 2 years. The two parameters were significantly correlated at $P = 0.01$. **B)** Ammonium-N content of soil and maximum temperature. The two parameters were significantly correlated at $P = 0.01$. Encircled numbers indicate the following: 1, beans planted; 2, bean plants disked into soil; 3, rye planted; 4, rye cover crop disked into soil.

to 5 (hypocotyls completely girdled).

Soil physical and chemical characteristics.—The physical and chemical characteristics of the soils were determined by standard methods (13). These included textural analysis, field capacity, pH, organic matter content by ignition oxidation, and salt content (conductivity) by electrical conductance of soil solutions. Exchangeable NH_4-N and NO_3-N were determined in 2-g portions of sieved soil with steam distillation by the magnesium oxide-Devarda alloy method (9). Total inorganic N by this method included the sum of NH_4-N and NO_3-N. Available P in the form of phosphate and available S in the form of sulfate were determined colorimetrically in 5-g portions of soil by the extraction methods of Nelson et al. (16) and Bardsley and Lancaster (3), respectively.

To evaluate soil color, we diluted soil 1/16 from each site and used 6 ml of each dilution to group the samples by color from lightest to darkest.

Climatological data.—Daily minimum and maximum temperatures and precipitation throughout the experiment were recorded by the University of Maryland Vegetable Research Farm.

Storage and manipulation of data.—Data were collected for the following 36 variables: rainfall and maximum and minimum temperatures; greenhouse and field *R. solani* DSI and total field DSI; tablebeet seed colonization by *R. solani;* greenhouse bean seedling emergence; field bean crop appearance, plant stand, yield, and plant weight; seed depth at planting; greenhouse and field *Pythium* lesions and blight; populations of *P. ultimum, P. aphanidermatum, T. basicola,* and *Fusarium* spp. in the field; organic matter and C content; total inorganic and total N; NO_3-N, NH_4-N, and soil C:N ratio; soil pH, conductivity, and color; available S and P content; sand, silt, and clay content; and field capacity. Variables assayed more than once are shown in Table 1. The data for each sampling date were analyzed by computer for possible correlations. The correlation of each biological variable with each of the other variables was determined individually. Duncan's multiple-range test was used for each variable, with either dates or field sites as replications. In addition, correlation coefficients were determined between variables for each date with field sites as replications.

TABLE 3. Substrate colonization by *Rhizoctonia solani* in soil samples collected from various depths of the Salisbury, Maryland, plot

Depth (cm)	Colonization in soil from indicated field areas (%)				
	A-6[a]	B-5	C-6	D-5	E-5
0-5	12	18	18	20	46
5-10	2	2	4	8	4
10-30[b]	0	0	0	0	0

[a]Site designations.
[b]Sampled at 5-cm increments in depth.

RESULTS.—*Inoculum density, disease severity, and physiochemical variables.*—The highest inoculum density of *R. solani* (expressed as percentage of colonization) was observed in August and September each year and the lowest from February to June, Table 2. Saprophytic activity peaked after the bean plants of the first crop had been disked into the soil, Fig. 1-A. Saprophytic activity also differed from year to year. For instance, in 1971, colonization maximized at about 45%, but in 1973, only at about 20%.

A highly positive correlation was found between inoculum density of *R. solani* (expressed as percentage of colonization of tablebeet seed supplied as bait) and the DSI obtained from infected beans grown in soil, of which samples were brought to the greenhouse each month, Table 1. Correlation between inoculum density and DSI in the field was also statistically significant at $P=0.05$. *R. solani* inoculum was also positively correlated with NH_4-N, total inorganic N, and mean maximum and minimum temperature, Fig. 1-B. No correlation could be found in

TABLE 4. Pathogenicity and anastomosis grouping of morphologically distinct isolates of *Rhizoctonia solani* from the Salisbury, Maryland, plot

Isolate	DSI[a] on bean	Damping-off of indicated plants in infested soil[b]				Anastomosis group
		Cotton	Lettuce	Radish	Sugarbeet	
R-10	4.7 (HP)[c]	100	71	67	100	AG-4
R-29	4.3 (HP)	30	50	26	29	AG-4
R-5	4.2 (HP)	100	75	90	100	AG-4
R-27	4.0 (HP)	64	33	0	92	AG-4
R-32	3.3 (P)	75	42	22	62	AG-4
R-28	3.1 (P)	86	40	17	61	AG-4
R-30	3.0 (P)	8	64	15	42	AG-4
R-33	2.4 (MP)	15	16	0	67	AG-4
R-31	2.2 (MP)	19	33	3	36	AG-4
R-7	0.5 (NP)	46	28	12	0	AG-4
R-8	0.3 (NP)	18	37	17	44	AG-3
R-9	0.2 (NP)	14	28	10	6	AG-3
R-26	0.2 (NP)	18	61	15	67	AG-4

[a]Disease severity index: 0 = healthy, 5 = plants dead.
[b]Damping-off was recorded 4 weeks after planting, and is expressed as percentage of the control (controls planted in noninfested soil were considered as having 0% damping-off).
[c]HP = highly pathogenic; P = pathogenic; MP = mildly pathogenic; NP = nonpathogenic.

this field between inoculum density and NO_3-N, rainfall, conductivity, and soil color.

It has been observed in Maryland, Delaware, and New Jersey that bean root rot caused by *R. solani* is more severe during the second bean crop (planted in July) than the first (planted in May). Results shown in Fig. 1-A indicate that the inoculum density had been at its lowest point before the winter cover crop was plowed under and beans were planted. In both years of research, an increase in *R. solani* activity was observed immediately after the bean residue from the first crop was disked into the upper few centimeters of soil. Each year, the inoculum was at its highest level between August 10 and the end of September (when the second bean crop was disked). The inoculum level then declined gradually to its lowest level the next spring.

Soil samples were collected at various depths to 30 cm from various sites of the Salisbury plot and analyzed for *R. solani* inoculum density. The pathogen was confined almost entirely to the upper 5 cm of soil, Table 3. Very little activity was observed between 5 and 10 cm, and no activity below 10 cm.

Isolations from soil and debris and characteristics of isolates.—From more than 500 transfers made from the original water-agar cultures of tablebeet seed buried in soils from the different sites, 13 morphologically distinct isolates were obtained on PDA and CSA from 1971-73. Besides the 13 isolates, two more morphologically distinct isolates were obtained from debris particles. Data collected during the transfers indicated that 90% of the transfers made directly from the colonized beet seed were pure cultures of *R. solani*.

For detection of heterokaryon formation, all field isolates listed in Table 4 and isolates R-40 and R-41 obtained from debris were paired on PDA containing 1% charcoal (PDCA) in all possible combinations. Four pairings only (R-7 with R-41, R-26 with R-41, R-30 with R-41, R-40 with R-41) developed tufts of mycelia where the cultures met on PDCA, indicating heterokaryon formation (7, 10). Isolate R-41 from debris was involved in all pairings observed. The rest of the pairings on PDCA developed no tufts and were considered incompatible.

Eleven of the 13 isolates from soil and the two debris isolates were of the 'Praticola'-type, AG-4 (Table 4). Two isolates, R-8 and R-9, were the AG-3 type. No isolate was found that belonged in groups AG-1 or AG-2.

The 13 selected isolates tested for pathogenicity on five kinds of plants ranged from nonpathogenic to highly pathogenic, Table 4. Although all highly pathogenic isolates belonged in the AG-4 group, not all AG-4 group isolates were highly pathogenic. With a few exceptions, isolates that were responsible for high DSI values on beans also caused a high percentage of damping-off. Some of the exceptions, however, were quite interesting. For instance, isolate R-29 was highly pathogenic on bean, but did not cause appreciable damping-off. Isolate R-27, highly pathogenic on bean, cotton, and sugarbeet, was nonpathogenic on radish and moderately pathogenic on lettuce.

Effect of temperature on survival.—Since inoculum density of *R. solani* in the field had a high correlation with mean maximum and minimum temperature (Table 1, Fig. 1) experiments were performed in the laboratory to assess

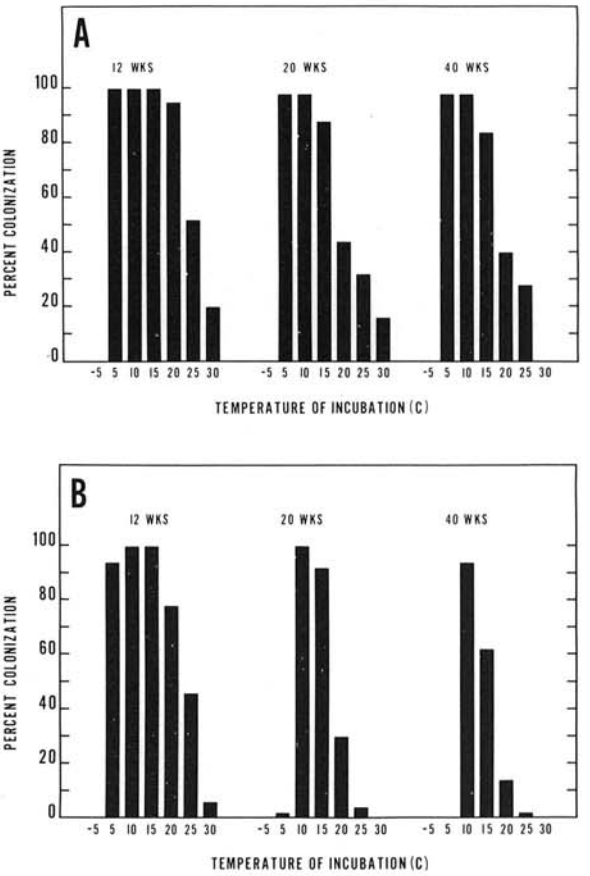

Fig. 2-(A, B). Effect of various temperatures on the survival of *Rhizoctonia solani* in soil determined by the colonization of tablebeet seed. **A)** Isolate R-27. **B)** Isolate R-29.

the importance of temperature on survival of *R. solani*. After 12 weeks of incubation, *R. solani* had lost its activity at −5 C (Fig. 2). At 5 C, isolate R-27 was active after 40 weeks, Fig. 2-A. Isolate R-29, however, did not survive at 5 C after 20 weeks of incubation, Fig. 2-B. Both isolates gradually lost their survivability at 25 and 30 C. The loss was more pronounced at 30 C, especially with R-29. Soil temperatures of 5, 10, and 15 C were the most conducive to survival of isolate R-27, and 5 and 10 C to that of R-29.

DISCUSSION.—Results obtained with saprophytic colonization techniques and pathogenicity tests established that saprophytic activity (and therefore inoculum density) of *R. solani* was at its peak in the upper 10 cm of soil in August and September, after beans from the first crop were disked. Saprophytic activity in the field was at its lowest before the first bean planting in May. It gradually increased and peaked after the first bean harvest. In the same sequence, bean root rot was extremely light in the first bean planting and heavy in the second. The gradual decline of *R. solani* activity during winter and spring may be due to a gradual reduction of food bases in soil, because of decomposition and food exhaustion and the adverse effect of low temperatures

during winter. The present data and previous research (6, 8, 17, 19, 21, 23) suggest that *R. solani* is highly dependent on plant tissue and almost disappears when the latter is exhausted. The pathogen grows rapidly from a suitable energy base, but survives poorly in competition once a selective substrate is gone. The laboratory experiment in Fig. 2 suggested that *R. solani* may not survive in soil at −5 C or even at 5 C, and that high temperatures (25 and 30 C) were not conducive to prolonged survival. These observations are in complete agreement with those of Benson and Baker (5) who showed that survival of *R. solani* in soil was three times better at 15 than at 26 C. Soil temperatures of −5 to 5 C at depths of 5-10 cm are not very uncommon in Salisbury after November.

Activity of *R. solani* was confined to the upper 10 cm of soil in the field plot, Table 3, even during August and September when activity reached its peak. Very little inoculum could be found at any time below a depth of 5-10 cm. This observation may be of considerable practical value if deep plowing of bean plant residue to a depth of 20-25 cm, rather than disking, were employed. In this manner, pathogen inoculum contained in plant roots and hypocotyls, and bean plant residue from the first planting would be buried deeply in soil. This practice may reduce inoculum potential and survival because *R. solani* may not be able to colonize extensively plant residue at depths of 20-25 cm. Although this is only a hypothesis, it may be expected to occur as a result of CO_2 accumulation released during plant residue decomposition. Previous reports summarized elsewhere (19) indicated the adverse effects of CO_2 on the saprophytic and pathogenic phases of *R. solani*.

The growers could also reduce inoculum of *R. solani* by planting beans once a year in May. The second bean crop could be replaced by a late crop of corn, sorghum, or any other suitable crop. That *R. solani* is at its lowest point before the first planting in May, may also be exploited for control. Perhaps the pathogen can be suppressed then with a minimal amount of fungicide that would not add excessively to the cost of production.

Computer analyses of the data established that competitive saprophytic activity of *R. solani* had a positive correlation with the magnitude of bean root rot in the greenhouse, and to a lesser degree, in the field. Such correlation enhances Papavizas' view (17) that competitive saprophytic ability in the sense of Garrett (12), measured by colonization of a suitable substrate, can be considered a highly reliable criterion of survivability of *R. solani* in soil. The correlation is also an indirect, but statistically accurate, measure of its inoculum density in soil. Similar conclusions were drawn by Sneh et al. (23), who used a different statistical approach to this problem, and by Martinson (15), who used a different colonization method.

Our results also showed a positive correlation between saprophytic activity of *R. solani* and total inorganic N and NH_4-N, but not NO_3-N. The correlation between activity and total inorganic N was expected, since survival of *R. solani* and the root rot it causes are known to be enhanced by total N (17, 19, 21). The positive correlation between activity and NH_4-N, however, was unexpected, since NH_4-N previously had appeared to decrease, rather than increase, saprophytic activity and survival (18). In those greenhouse studies, NO_3-N increased survival and saprophytic activity. The discrepancy may be due to several factors in the field that may be different from those in the greenhouse (previous cropping, soil condition, time of application) or to the NH_4-N:NO_3-N ratio, N-application rate and stability, or residual N in the field from previous fertilizations.

R. solani is known to be composed of an indefinite number of races distinguished by asexual morphological and physiological characteristics (14). More recent studies (22) showed that in *Thanatephorus cucumeris,* the perfect state of *R. solani,* at least four anastomosis groups can be found. Our results lead us to conclude that the population of *R. solani,* even in a small field, is composed of a number of distinct isolates with various saprophytic and parasitic potentialities. As might be expected, the majority of isolates from soil and debris in this field belonged to the 'Praticola'-type (AG-4), with very few belonging only to AG-3 (potato isolate). In these limited studies, isolates within an anastomosis group varied in pathogenicity from highly pathogenic to nonpathogenic. For instance, several isolates in AG-4 were very pathogenic on beans, whereas other isolates in the same group were mildly pathogenic or nonpathogenic. More studies are needed to determine whether distinct parasitic patterns are unique to the different anastomosis groups known today.

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