Effects of Crop Rotation on Rhizoctonia Disease of White Potato

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ABSTRACT

Specht, L. P., and Leach, S. S. 1987. Effects of crop rotation on *Rhizoctonia* disease of white potato. Plant Disease 71: 433-437.

Sweet corn (Zea mays), Japanese millet (Echinocloa crusagalli), buckwheat (Fagopyrum esculentum), spring oat (Avena sativa), and annual ryegrass (Lolium multiflorum) were used as 1-yr rotation crops with potato (Solanum tuberosum) to evaluate their effectiveness in controlling the Rhizoctonia disease of potato (R. solani). The effects of incorporating the rotation crop residues as green, immature amendments versus mature, partially decomposed amendments were also examined. Populations of Rhizoctonia spp. were usually significantly higher in buckwheat-rotated soils, but the resulting disease severity level was not higher. Buckwheat may have selected for species or strains of Rhizoctonia that were nonpathogenic or less pathogenic on potato. Annual ryegrass gave the lowest overall disease severity, but this was only significantly lower than that for Japanese millet (P = 0.05). The effect of tillage regime on disease severity was not significant, except for Japanese millet; in this instance, the early-tilled soils had lower levels. Increased populations of bacteria and fungi, but not actinomycetes, were found in the early-tilled plots immediately after incorporation of the green, immature amendments. Except for the Japanese millet rotation, this increase in soil microorganism population was not associated with a suppression of the pathogen or a reduction in disease severity.

The Rhizoctonia disease of white potato is an important problem in Maine and other potato-growing areas (15,22). The major pathogen, Rhizoctonia solani Kühn, has a very wide host range (3). Other species of Rhizoctonia may also be involved (7). Within the species of R. solani, those strains that belong to anastomosis group 3 (AG-3) are consid-

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Accepted for publication 10 November 1986.

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ered most pathogenic on potato (2). Crop rotation has sometimes proven useful in controlling this disease (9,17) but not always (33). The nature of a crop residue can influence the activity of *R. solani* (8,12,13,27,28). Blair (8) found that high amounts of readily decomposable organic matter in amendments resulted in the greatest level of suppression. Papavizas and Davey (28) found that incorporation of green plant residues increased populations of bacteria, actinomycetes, and fungi antagonistic to *R. solani*.

Crop rotation may influence the activity of R. solani several ways. Direct effects may include the suitability of the crop and its residues for parasitic and saprobic colonization by R. solani. This pathogen can survive as a saprobe in the soil by competitively colonizing plant debris during its nonparasitic phases (27). Other direct effects include the influence of plant residue decomposition products on R. solani (24,25). One direct effect of crop rotation is antagonism toward R. solani resulting from qualitative and/or quantitative changes in the

composition of the soil microbiota (1,5,32,36,37,39). A reduction in populations of *R. solani* resulting from competition for available nutrients can arise from the increased activity of soil microorganisms (5). Increases in actinomycetes may suppress *R. solani* because of their antibiotic-producing ability (31). Increased populations of certain species of soil microorganisms may be important; for instance, high populations of *Trichoderma* spp. have been associated with a suppression of *R. solani* (10,18,26).

The objective of this study was to determine the influence of five rotation crops on populations of *Rhizoctonia* spp. and the resulting severity of the *Rhizoctonia* disease of white potato. Populations of bacteria, including actinomycetes, and fungi, including *Trichoderma* spp. were also determined, with these data being related to each other to determine possible mechanisms by which disease reduction may occur.

MATERIALS AND METHODS

Cultural practices. The plots used for this study had been in 2-yr rotations with potato for 5 yr. Two sets of similar plots were sampled and are referred to as experiments 1 and 2. The two experiments were staggered in their rotation schedule so that when one was planted to potato, the other was planted with rotation crops. Plots in experiment 1 were sampled over a 2-yr period from 1981 through 1982. Disease severity data on potato were taken in 1982; populations of Rhizoctonia spp. were determined in both 1981 and 1982. Plots in experiment 2 were only studied in 1982, when rotation crops were planted. Field plots were located at Newport, ME, in a Bangor silt loam. Individual plots were 4 × 8 m and were separated by 1-m grass strips. Each plot was planted with potato (cultivar Kennebec) and rotated with one of five rotation crops: sweet corn (Zea

mays L.), Japanese millet (Echinocloa crusagalli (Rozb.) Wright), buckwheat (Fagopyrum esculentum Moench), spring oat (Avena sativa L.), and annual ryegrass (Lolium multiflorum Lam.). Populations of microorganisms and disease severity were also determined in an adjacent set of nonrotated plots (i.e., planted to potatoes for five consecutive years).

All plots to be planted with potatoes received in-row applications of a 12-15-15-1.8 commercial fertilizer at a rate of 1,500 kg/ha. Handcut seed pieces (43-60 g) were treated with PCNB + thiabendazole (50,000 + 1,500 ppm)respectively); this prevented the addition of outside inoculum in the form of tuberborne sclerotia. The seed pieces were hand-planted, with 22-cm spacings within rows and 0.9-m spacings between rows. Lime and pesticides were applied as needed following soil test results and recommended cultural practices. Plots planted with the five rotation crops received 560 kg/ha of 10-10-10-1.8 commercial fertilizer immediately before seeding. All rotation crops except sweet corn were broadcast-planted; sweet corn was planted in 0.9-m spaced rows with a hand planter. All rotation crops were mowed in mid-August, and ammonium nitrate (33.5% N) was broadcast at a 34 kg N/ha to promote maximum decomposition of crop residues. One-half of each plot was then tilled to incorporate the crop residue (to a depth of 15-20 cm) as a green, immature amendment; the crop residue and ammonium nitrate on the other half of the plot was left on the surface of the soil and incorporated in November as a mature, partially decomposed amendment.

The experimental design for experi-

ments 1 and 2 was a randomized complete block split plot, with tillage regime being used as the subplot treatment. Each main plot (rotation crop) was replicated four times for both experiments. For statistical analysis, the Bayes LSD value (K-ratio = 100, P =0.05) was used to make comparisons among the rotation crops. The subplot effect of tillage, averaged over all five rotation crops, was also tested, which left four degrees of freedom for testing the effect of tillage separately with each individual rotation crop. Four single degree of freedom orthogonal comparisons were therefore used to test the effect of tillage in the Japanese millet, buckwheat, oat, and annual ryegrass rotations. We chose not to test the effect of tillage in the sweet corn rotation because this would be an unlikely practice. Also, data collected from the nonrotated plots were not analyzed statistically because those plots were not included in the experimental design. However, the later data provided useful information and are presented for comparison.

Soil microbial assays. Soil samples were collected in both 1981 and 1982. In 1981, those plots planted with rotation crops were sampled on 31 August, 30 September, 9 October, and 21 November (experiment 1). Potatoes were planted in these plots the following year (1982), and soil samples were collected on 17 May, 15 June, 23 July, and 8 September. The rotation plots in experiment 2 were only sampled in 1982, when they were planted to rotation crops. Representative soil samples were collected from the plots by combining and thoroughly mixing 15-20 subsamples, collected to an average depth of 15 ± 2 cm with a 2.5-cm soil

probe. Soil samples collected from plots planted with potatoes were taken from the middle of the hill in the vicinity of plant roots. No attempts were made to distinguish rhizosphere from nonrhizosphere soil. The soil samples were sieved (6.4-mm openings) to remove stones and large pieces of plant debris, kept moist in waxed cardboard boxes, stored at 23 C. and assayed for microorganism populations within 24-48 hr.

In 1981, populations of Rhizoctonia spp. were determined by a soil debrisparticle isolation method (38). A 2% hydrogen peroxide solution was used to remove contaminating bacteria from debris (35). Organic debris, retained on a 0.2-mm sieve after wet-sieving a 10-g soil sample, was transferred to molten 1.5% water agar cooled to 47 C. Twentymilliliter aliquots were then immediately poured into petri plates and allowed to solidify. Plates were incubated at 23 C for 48-72 hr to allow fungi to grow out from organic matter and sclerotia. Any colonies that resembled a Rhizoctonia sp. were confirmed by examining their hvphal morphology (11). In 1982, populations of Rhizoctonia spp. were determined by the pellet soil samplerselective medium method described by Henis et al (19) with Ko and Hora's selective medium (23); this method allows a larger quantity of soil to be assayed (25 g of soil per sample) and produces results equal to the hydrogen peroxide method (6). Plates were incubated at 23 C for 36-48 hr and observed for colonies of Rhizoctonia spp. The multiple-colonization correction formula was used to account for multiple colonies emerging from single soil pellets

Total populations of bacteria, including

Table 1. Populations^a of *Rhizoctonia* spp. as affected by rotation crop

Sample date	Potatob	Corn	Millet	Buckwheat	Oat	Ryegrass	Bayes LSD (P = 0.05)	
Experiment 1 ^c							· · · · · · · · · · · · · · · · · · ·	
31 August 1981	•••	1.8	1.5	4.3	3.0	2.5	NS^d	
30 September 1981	•••	3.4	4.5	13.6	10.6	3.4	NS	
9 October 1981	•••	2.3	1.6	10.6	5.1	3.5	6.0	
21 November 1981	•••	2.8	0.9	12.0	5.9	1.1	6.0	
17 May 1982	1.3	0.8	1.3	7.8	4.4	3.4	NS	
15 June 1982	0.5	0.6	0.6	5.9	1.7	1.7	5.3	
23 July 1982	2.8	1.7	1.8	7.5	3.2	3.1	NS	
8 September 1982	2.6	2.0	1.7	7.1	3.5	2.0	4.7	
Experiment 2 ^e								
10 August 1982	•••	1.0	1.0	7.1	1.5	2.2	4.0	
24 August 1982		2.4	6.1	13.7	3.3	5.4	3.6	
7 October 1982	•••	0.9	2.6	7.7	2.8	2.6	5.0	

^a Population counts of *Rhizoctonia* spp. given as propagules per 10 g of dry soil as determined by either the soil debris particle method (in 1981) or the soil pellet sampler-selective medium method (in 1982).

^bData from the nonrotated continuous potato plots not part of experimental design or analysis. Data from continuous potato plots only taken if the plots being sampled were planted with potatoes.

Experiment 1: rotation crops present in 1981, with four sample dates (31 August, 30 September, 9 October, and 21 November) after early tillage carried out on 20 August; late tillage carried out on 15 November. Four sample dates (17 May, 15 June, 23 July, and 8 September) the year after rotation while potatoes were grown.

Not significant.

Experiment 2: rotation crops present in 1982, with data from one sample (10 August) prior to any tillage treatment. Early tillage carried out on 17 August; late tillage carried out in mid-October after the last sample date.

actinomycetes, and fungi, including *Trichoderma* spp., were determined using the soil dilution-plate count technique. Bacteria, actinomycetes, and fungi were enumerated on soil extract agar, sodium caseinate agar, and rose bengal agar, respectively (29). Populations of these other microorganisms were determined only in 1982.

Identification of isolates of R. solani. To distinguish between Rhizoctonia-like fungi and multinucleate R. solani, nuclei in vegetative cells (<4 days old) from cultures of Rhizoctonia grown on potato-dextrose agar (PDA) were stained with 0.5% aniline blue in lactophenol with an acidified wetting agent (Tween 20) (20). Multinucleate isolates were tested to determine their anastomosis groupings by pairing each isolate with a tester isolate from each of AG-1, AG-2, AG-3, AG-4, and AG-5 following accepted procedures (21,30).

Assessment of disease severity. Disease severity was evaluated by the system described by Frank et al (16). The rating system evaluated a total of five aspects of the disease: percent nonemergence, number of stems with lesions and degree of severity, percent stolons pruned, percent stolons with lesions, and percent unusable weight of harvested tubers. Tubers were rated unusable if they had 10 or more sclerotia or were malformed. Plants were evaluated for R. solani damage 6 and 10 wk after planting. An overall rating was determined on the basis of the five aspects of the disease evaluated. A total of five plants was rated per plot.

RESULTS

Rhizoctonia spp. assay. Populations of Rhizoctonia spp. were always high in

the buckwheat-rotated soils, with significant differences (P = 0.05) on seven of the 11 sample dates (Table 1). Population levels of Rhizoctonia spp. supported by the other four rotation crops (corn, Japanese millet, oat, and ryegrass) did not differ significantly, except on sample date 24 August 1982, when the population level of Rhizoctonia spp. with the millet rotation was significantly greater than with the corn rotation. Population levels of R. solani in the continuous potato plots were similar to those supported by the last four rotation crops. The early- and late-tillage regimes did not produce significantly different populations of Rhizoctonia spp.

Disease severity. When the main plot effect of rotation crop was examined, there were no significant differences in percent nonemergence, percent stolons pruned, percent stolons with lesions, or percent unusable yield (Table 2). However, the stem lesion severity ratings did differ significantly between the plots rotated with Japanese millet versus buckwheat (28.7 vs. 6.6, respectively). Also, the overall final disease rating following Japanese millet (2.60) was significantly higher than that following annual ryegrass, which had on overall final rating of 1.58 (Table 2). The overall final rating in the nonrotated potato plots was 2.50, but this value was not analyzed statistically.

Early-tilled plots had a significantly lower stem lesion severity rating than the late-tilled plots (10.6 vs. 19.2) but a significantly higher percentage of stolons with lesions (13.5 vs. 8.5). The overall final disease rating did not differ significantly between the early- versus late-tilled plots. Of the four rotation crops tested individually for an early-

versus late-tillage effect (Japanese millet, buckwheat, oat, and annual ryegrass). only the Japanese millet rotation produced a significant difference. The earlytilled Japanese millet plots produced potato plants with a significantly (P =0.05) lower level of disease compared with the late-tilled Japanese millet plots. The overall disease ratings for these two regimes were 1.85 and 3.34, respectively (not shown in tables). The data of experiment 1 (sample dates 17 May, 15 June, 23 July, and 8 September 1982) were reanalyzed as a randomized complete block split-split plot, using the four sample dates as the additional split plot. Populations of Rhizoctonia spp. did not differ significantly among these four dates. Figure 1 shows the observed relationship between populations of Rhizoctonia spp. (averaged over the four sample dates) and the overall final disease rating for the different rotations, including the nonrotated, continuous potato plots. The buckwheat rotation resulted in a high population of *Rhizoctonia* spp., but this was not associated with a higher final disease rating.

The high *Rhizoctonia* spp. populations found in the buckwheat plots were associated with populations of binucleate *Rhizoctonia*-like fungi that were three times those found in plots of other crops. The populations of *R. solani* AG-3 were about equal in all plots, thus the similar disease ratings observed. The remaining anastomosis groups were about equal in all plots.

Populations of bacteria, actinomycetes, and fungi. No differences among the five rotation crops were consistently observed with respect to population levels of total bacteria, actinomycetes, and fungi, or Trichoderma spp. In experiment 1

Table 2. Rhizoctonia solani disease ratingsa on Kennebec potato following the previous year's rotation crop and tillage regime (experiment 1, 1982)

	Main plot effect of crop									
	Potatob	Corn	Millet	Buckwheat	Oat	Ryegrass	Bayes LSD (P = 0.05)			
Percent nonemergence	17.9	19.5	22.0	27.7	23.3	19.2	NS°			
Stem lesion severity	20.4	16.5	28.7	6.6	14.1	8.7	17.0			
Percent pruned stolons	22.8	9.5	18.1	7.1	5.9	6.7	NS			
Percent stolons with lesions	4.4	9.1	9.0	11.3	13.6	12.4	NS			
Percent unusable yield	10.2	17.7	11.9	10.3	15.0	5.9	NS			
Overall final rating	2.50	2.02	2.60	1.95	2.02	1.58	1.02			
	Subplot effect of tillage ^d									
		E	arly		Late					
Percent nonemergence		21.4			23.3					
Stem lesion severity		10.6*°			19.2					
Percent pruned stolons	7.6				11.3					
Percent stolons with lesions	13.5**				8.5					
Percent unusable yield	10.9				13.4					
Overall final rating			2.21							

^a Rhizoctonia disease rating system as described by Frank et al (16).

^bData from potato rotation not part of experimental design or analysis.

^c Not significant.

^d Previous year's rotation crops were incorporated in mid-August for early-tillage regime and in mid-November for late tillage regime. Only the overall effect of tillage regimes averaged over the five rotation crops is presented.

e Probability of a significant difference between tillage regimes: observed differences significant at * = P = 0.05 and ** = P = 0.01.

(1982), the early- and late-tillage regimes of the previous rotation crop did not produce significantly different populations of these microorganisms. However, in experiment 2 (1982), significantly higher populations of both total bacteria and fungi were observed with the early-tillage regime (8/24) immediately after incorporation of the green amendment. The early-tillage regime did not affect the population of actinomycetes. In contrast,

actinomycete populations were higher in the late-tilled soils on 7 October (Fig. 2). This sample date was prior to incorporating the mature, partially decomposed residues lying on the surface of the soils. The overall effect of tillage regime on populations of microorganisms was consistent with the effects observed for each individual crop. Total populations of *Trichoderma* spp., which accounted for 2-3% of the total fungal plate counts,

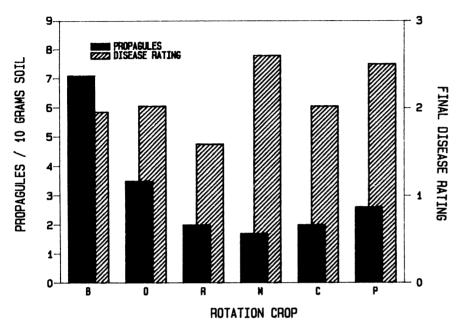


Fig. 1. Populations of *Rhizoctonia* spp. (average of sample dates 17 May, 15 June, 23 July, and 8 September) and final disease ratings in potato soils following the previous year's crop. Alternate crops were buckwheat (B), oat (O), ryegrass (R), millet (M), corn (C), and potato (P). Experiment 1, 1982. Bayes LSD (P = 0.05): propagules = 5.6, disease rating = 1.0. Data from potato (nonrotated) not part of experimental design or analysis.

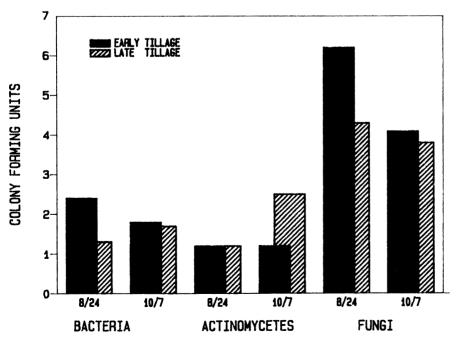


Fig. 2. Populations of microorganisms as affected by tillage treatment (overall effect). Early tillage carried out on 17 August; late tillage was not carried out until after last sample date. * = Tillage treatments significantly different on given sample date (P = 0.05). Counts given as colony-forming units multiplied by 10^8 (bacteria), 10^7 (actinomycetes), and 10^5 (fungi).

were not altered by either rotation crop or tillage regime.

DISCUSSION

Results obtained with either the soil debris particle isolation method or the pellet soil sampler-selective medium method were similar. The pellet soil sampler-selective medium method was used the second year of this study because it permitted the assay of larger quantities of soil. Slow-growing strains of Rhizoctonia spp. were detected as easily as fast-growing strains, provided that the incubation period was at least 48 hr. Results indicated that there was an increase in the population of Rhizoctonia spp, when buckwheat was used as a rotation crop with white potato. The major portion of the observed increase was because of high populations of binucleate Rhizoctonia-like fungi, not multinucleate R. solani strains. Frank and Murphy (17) also found high populations of Rhizoctonia spp. in some rotated potato soils. In their study, the observed populations did not correlate with the resulting severity of disease on potatoes. It is very possible that what they observed was similar to what was found in this study until we tested for nuclei number and anastomosis groups. This suggests that some crops may select for species or strains of Rhizoctonia-like fungi that are nonpathogenic or less pathogenic on potato. Such a selection, if it occurred, may have been a result of a proliferation of only those species or strains that possess a high competitive saprobic ability. Populations of those fungi possessing greater competitive saprobic abilities would be favored when large quantities of crop residue were added to soil. Other species of Rhizoctonia are known to infect some of the rotation crops examined in this study. For instance, Sumner and Bell (34) reported the isolation of both R. solani and R. zeae from the roots of diseased corn plants. Davis and McDole (14) found AG-4 of R. solani to be the most common in soils involved in a potato-grain rotation and not the highly pathogenic (to potato) AG-3 strain. A study conducted by Bandy et al (7) indicates that binucleate strains of Rhizoctonia spp., as well as anastomosis groups of R. solani other than AG-3, are commonly present in Maine potato fields.

The nonrotated, continuous potato plots were found to have plants that had a high percentage of pruned stolons and a low percentage of stolons with just lesions. One possible explanation for this is that the nonrotated potato soils selected for highly pathogenic (to potato) strains of *Rhizoctonia* spp., assuming that a high percentage of pruned stolons indicates a high level of pathogenicity. Further field studies would have to be conducted to test these hypotheses and also to determine what species and strains of *Rhizoctonia* are being selected

for by various rotation crops. Alternatively, the buckwheat rotation may have also promoted an increase in the population of antagonistic microorganisms, which suppressed the parasitic activity of R. solani. However, our results do not support this idea, because populations of total bacteria, actinomycetes, fungi, and Trichoderma spp. were not found to differ among the five rotation crops. However, certain species of antagonistic microorganisms may have been altered by the rotation crops.

Tillage was found to alter the resulting incidence of disease only with the Japanese millet rotation, where early tillage resulted in less disease. Populations of *Rhizoctonia* spp. were not affected in any instance by tillage regime, except for some sample dates with the buckwheat rotation, where higher populations of *Rhizoctonia* spp. were not observed in early-tilled soils. Observed increases in populations of bacteria and fungi with the early-tillage regime were not associated with any reduction in disease severity, except for Japanese millet.

The results of this study indicate that in general, a 1-yr rotation of potatoes in Maine with corn, oats, buckwheat, annual ryegrass, or Japanese millet has little effect on the *Rhizoctonia* disease complex of potatoes or the soil population of *R. solani* AG-3. The lower disease ratings observed after annual ryegrass, though not significant, and the significant difference when immature versus mature Japanese millet was incorporated into the soil is evidence that continued work is needed in this area.

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