Root Rot Reaction in Wheat: Resistance not Mediated by Rhizosphere or Laimosphere Antagonists

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ABSTRACT

Chromosome 5B is critical in differentiating the resistance to common root rot of the cultivars 'Apex' (A) and 'Cadet' (C) from the susceptibility of 'S-615' (S) and 'Rescue' (R). The validity and significance of a previously indicated association between resistance governed by chromosome 5B and the occurrence of bacteria in the rhizosphere antagonistic in vitro to the primary pathogen, *Cochliobolus sativus,* was evaluated.

Antagonists were more prevalent not only in the rhizosphere but also in the subcrown internode laimosphere of Apex and the resistant disomic chromosome substitution line S-615-Apex 5B (S-A5B) than they were in the rhizosphere or laimosphere of S-615. Seed bacterization with rhizosphere antagonists from Apex or S-A5B did not, however, increase the resistance of S-615 even though estimates based on plate-dilution frequency techniques showed that treatment with the S-A5B culture increased antagonists in the rhizosphere and that bacterization with either culture increased antagonists in the laimosphere to levels comparable to those in untreated Apex and S-A5B. Comparison of the parental cultivars, Apex, Cadet, S-615, and Rescue and their resistant (S-A5B, C-R5D) and susceptible (S-A5D, C-R5B) disomic chromosome substitution lines, showed no consistent relationship between resistance to common root rot and incidence of rhizosphere bacteria antagonistic in vitro to *C. sativus*. The cytogenetic evidence clearly showed that the incidence of antagonists is not controlled by the gene that determines root rot reaction in these lines.

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Soil microorganisms antibiotic to root pathogens have long attracted the attention of pathologists and microbiologists seeking biological control of various soil-borne diseases (7, 13, 19). Many reports deal with the effectiveness of selected bacteria, actinomycetes, or fungi in inhibiting the germination or growth of root pathogens in vitro or in controlling disease development following seed bacterization, soil modification, or both (3, 14, 17). Few convincingly documented examples point to the presence or absence of antibiosis as the critical disease-determining factor in normally cropped field soils. In a recent review Baker (1) indicated that much of the evidence that antibiosis is significant in control rests on correlations between suppression of disease and increase in antibiotic-producing organisms. He also suggested that there are few published examples of lack of correlation between antibiosis and biological control because few scientists publish negative data.

We have shown that chromosome 5B is critical in differentiating the resistance to common root rot of the spring wheat cultivar 'Apex,' from the susceptibility of the cultivar 'S-615' (8). Replacing chromosome 5B in S-615 with the corresponding chromosome from Apex makes the resulting chromosome substitution line, S-A5B, as resistant as Apex (8) and changes the characteristics of the rhizosphere microflora (11). The incidence of bacteria antibiotic in vitro to the primary causal agent of common root rot, *Cochliobolus sativus* (Ito and Kurib.) Drechs. ex Dastur, was the most obvious rhizosphere characteristic correlated with the disease reactions of the chromosome substitution line and its donor and recipient parents (11). Although none of the bacteria isolated from dilution plates of S-615 rhizosphere soil showed antibiosis to *C. sativus,* 20% of those similarly obtained from the rhizospheres of Apex and S-A5B were antagonistic. Despite the intriguing correlation that this genetically controlled system provided, we cautioned that it was not possible from such data "to associate conclusively resistance to root rot with the occurrence of these antagonistic microbes in the rhizosphere" (11). The present paper reports the results of further experiments we have carried out to determine the significance of this association. It also provides evidence for the existence of a "laimosphere" effect (9) about the subcrown internodes of wheat.

MATERIALS AND METHODS.—*Bacterization experiment.*—Seed was treated with rhizosphere bacteria that exhibited antibiosis to *C. sativus* in vitro to determine whether root rot ratings in naturally infested field soil would be reduced.

Three lines of spring wheat (*Triticum aestivum* L. emend. Thell.) were used: Apex, moderately resistant to common root rot; S-615, highly susceptible; and S-A5B, a disomic chromosome substitution line essentially the same as S-615 except for a pair of 5B chromosomes from Apex that produces root rot resistance equivalent to that of Apex (8). These lines were those used previously (11) and have been fully described elsewhere (8).

Two separate cultures of rhizosphere bacteria, one from Apex, the other from S-A5B were used to treat the seeds. Each culture was a mixture of 20 isolates shown to be antagonistic to *C. sativus* in vitro (11). The isolates composing each culture were gram-positive rods with similar characteristics, but between the two cultures there were slight differences in the morphological and physiological characteristics of the bacteria and in the solubility properties of the antibiotic(s) they produced. In each instance, the bacteria were highly specific in their antagonism for *C. sativus* (J. L. Neal, *unpublished*). The bacterial isolates were maintained separately in sterile soil at 28 C and increased in nutrient broth (28 C) for the bacterization experiments. The individual nutrient broth cultures were centrifuged after 96 hr and the bacteria were washed and resuspended in sterile distilled water. They

were then mixed to form the inoculating suspension. Greenhouse-grown seeds of Apex, S-615, and S-A5B were added in individual lots of 300 each to separate 100-ml aliquots of a suspension of each culture and stirred continuously for 30 min. Three hundred seeds of each line were similarly treated in sterile distilled water. After treatment, the seeds were transferred to sterile filter paper in petri dishes and allowed to drain for 1 hr at 37 C.

The nine treatments (three wheats × three seed treatments) were laid out in a randomized block design with six replicates, each replicate contained in a metal flat. Because of space limitations, two growth cabinets were used, one with four replicates, the other with two. Both cabinets were programmed to provide a diurnal cycle with a 16-hr photoperiod at 18 C and an 8-hr dark period at 13 C. Within each replicate the plots were arranged in a three × three format. Each plot consisted of six rows 5 cm apart, each containing seven seeds spaced 2.5 cm apart. The seed was placed with clean forceps on 7.6 cm of well-mixed field soil naturally infested with *C. sativus*. The soil had been treated with 0.1% Lytron soil conditioner (Monsanto Canada Ltd., Rexdale, Ontario). The seed was covered with 6.3 cm of the same soil. The seed was planted at this depth so that the subcrown internodes would be long enough to rate for root rot reaction and to provide an adequate soil sample for estimating the microbial populations of the laimosphere. To avoid improper use of the term rhizosphere, "laimosphere" has recently been proposed to designate the zone of influence of belowground stems or leaves on soil microbes (9).

Residual seed from each treatment held at room temperature was assayed for the presence of viable antagonists of *C. sativus* after 2 and 5 days. In each test, ten seeds from each of the nine treatments were placed, five seeds per petri dish, on potato-dextrose agar made up in nutrient broth instead of water, then incubated for 96 hr at 28 C. The presence or absence of bacterial growth about each seed was noted. Conidia of *C. sativus* were then dusted over the plates. The occurrence of clear zones about the seeds, indicating inhibition of *C. sativus,* was noted after a further 96 hr of incubation.

Seven weeks after the bacterization experiment was seeded, the middle five plants from each of the center four rows of each plot were lifted. Roots from all twenty plants of each sample and subcrown internodes from ten of them were used to estimate the total populations of bacteria and of antagonists as outlined below. The other subcrown internodes were rated immediately for root rot reaction (2). Forty-eight hr later, the subcrown internodes of those plants used to provide the laimosphere samples were rated for root rot. These results were included with the earlier ratings. The percentage data were transformed to arc sines and analysis of variance applied.

The plants sampled for microbial population estimates were carefully shaken to dislodge adhering nonrhizosphere soil, and then separated into seminal root and subcrown internode portions. Separate soil suspensions were obtained from each structure as previously described (11). Successive fourfold dilutions of the initial soil suspensions were made and suitable aliquots were plated on soil-extract agar to permit the total bacterial populations and the numbers of antagonists of *C. sativus* on the same plates to be estimated by plate-dilution frequency techniques (6, 10). A 100-g composite sample of nonrhizosphere soil from each replicate was subjected to the same analyses.

The data of each replicate were independently adjusted to a basis of 1 g of oven-dry soil. Analysis of variance was carried out on the data transformed to $\log_{10}$. Differences among means were compared using Duncan's multiple range test.

Rhizosphere analyses of additional chromosome substitution lines with differential root rot reactions.—To determine whether the occurrence of antagonists in the rhizosphere was related to disease reaction, the incidence of bacteria antibiotic to *C. sativus* in vitro was assessed in the wheats previously tested (11) and in similar untested lines differential for root rot reaction. The cultivars 'Cadet', moderately resistant to root rot, and 'Rescue,' a highly susceptible hybrid selection of Apex and S-615, and their disomic chromosome substitution lines C-R5B and C-R5D were used. Because it carries the critical 5B chromosome from Rescue, the line C-R5B, though genetically like Cadet in most respects, is susceptible to root rot; C-R5D exhibits the same level of resistance as Cadet (8). In addition to S-615, Apex, and S-A5B, the related disomic chromosome substitution line, S-A5D, was included. Unlike S-A5B, S-A5D is as susceptible to root rot as S-615 (8). The S-615-Apex and Cadet-Rescue chromosome substitution series are conversely related in that in S-615-Apex the recipient and donor parents are root rot-susceptible and -resistant, respectively, whereas in Cadet-Rescue, the relationship is reversed (8).

These wheats, sown 2.5 cm deep in "disease-free" Lytron-treated field soil, were grown in the greenhouse as in our previous investigation (11) and replicated three times. Seven weeks after seeding, nonrhizosphere and rhizosphere soil samples were obtained and used to estimate the total bacterial populations and bacteria antagonistic in vitro to *C. sativus*. The procedures were as described above except that tenfold, rather than fourfold, dilutions were made (6).

RESULTS.—*Bacterization experiment.*—Bacterization did not alter the root rot reactions characteristic of the three wheat lines (Table 1). Analysis of variance showed no significant differences in root rot ratings between bacterization treatments or between replicates, and no interaction between lines and treatments. The highly significant difference between lines was accounted for by the inherent difference between S-615 and the other two wheats.

Failure of seed bacterization to reduce the root rot susceptibility of S-615 or to increase the resistance of Apex and S-A5B could be attributed to failure of the antagonists to become established in the rhizosphere of the test plants. The seed bioassay showed, however, that the bacterized seeds were carrying viable antagonists of *C. sativus* 2 and 5 days after treatment. Only the seeds that had been treated with either of the cultures of rhizosphere antagonists were consistently surrounded by an obvious clear zone indicating inhibition of *C. sativus*.

The rhizosphere data for the untreated lines in disease-producing field soil confirm our previous findings for "disease-free" soil (11): that the root rot-susceptible recipient parent, S-615, had significantly larger numbers

TABLE 1. Percent root rot ratings of wheat lines grown from untreated seed and from seed treated with cultures of rhizosphere bacteria antibiotic in vitro to *Cochliobolus sativus*

Wheat line[a]	Untreated	Source of bacterial culture		Mean[b]
		Apex	S-A5B	
S-615	95.0	96.7	93.0	94.9
Apex	19.3	20.3	20.4	20.0
S-A5B	21.0	19.8	20.0	20.3
Mean[c]	45.1	45.6	44.5	

[a] S-A5B is a chromosome substitution line of S-615 in which chromosome 5B has been replaced by the corresponding chromosome from Apex.
[b] Line means, F = 729.97 ($P < .01$).
[c] Treatment means, F = .919 ($P > .99$).
Line × treatment interaction, F = 1.121 ($P > .99$).

of bacteria in the rhizosphere but significantly fewer antagonists, both in numbers and percentage, than the root rot-resistant donor parent, Apex (Table 2). As before, the rhizosphere populations of Apex and the chromosome substitution line, S-A5B, were similar. Bacterization with antagonists from Apex did not alter these relative relationships. Treatment with the S-A5B culture, however, increased the numbers but not the percentage of antagonists in the rhizosphere of S-615 to a level comparable with that of Apex and S-A5B. Bacterization with the S-A5B culture produced a significant percentage increase in antagonists in the rhizosphere of Apex but a contrasting decrease in those of S-A5B. Apex and S-A5B showed increased total bacterial populations when bacterized with their own antagonists, but no such trend was evident when they were treated with the bacteria isolated from the opposite line.

The laimosphere data of the subcrown internodes provide interesting similarities to, and contrasts with, those of the corresponding rhizospheres (Table 2). In contrast with the rhizosphere, the laimosphere of the untreated lines show that the soil about the subcrown internode of S-615 harbors no larger bacterial populations than either Apex or S-A5B. Nevertheless, as in the rhizosphere, significantly fewer bacteria antibiotic to *C. sativus* were found in the laimosphere of S-615 than in either of the root rot-resistant lines. Bacterization with either of the antibiotic-producing cultures increased the percentage of antagonists in the laimosphere of S-615 to levels comparable with those of the root rot-resistant lines. Despite this, there was no decrease in the severity of disease symptoms on the subcrown internode of S-615 (Table 1). The only other statistically significant change produced in the laimosphere by bacterization was a decrease in the percentage of antagonists when S-A5B was inoculated with the Apex culture. The reverse relationship did not hold. Because little soil adhered to the subcrown internodes, our data may be more representative of the laimoplane than of the laimosphere. Special methods for collecting laimosphere soil around subcrown internodes are needed to obtain larger samples.

Rhizosphere analyses of additional chromosome substitution lines with differential root rot reactions.—The root rot-susceptible lines showed a

TABLE 2. Population estimates of total bacteria and of bacteria showing antibiosis in vitro to *Cochliobolus sativus* in the rhizospheres and laimospheres of wheat lines grown from untreated seed and from seed treated with rhizosphere bacteria antibiotic to *C. sativus*[u]

Wheat line or sample source[v]	Reaction to common root rot[w]	Source of bacterial culture	Rhizosphere			Laimosphere	
			Total bacteria (× 10^6)	Antagonists (× 10^6)	Antagonists (%)[x]	Total bacteria (× 10^6)	Antagonists[y] (%)[x]
S-615	S	Untreated	332 a[z]	14 c	4.2 e	114 a	1.6 c
Apex	R	Untreated	161 c	25 ab	15.7 b	109 a	11.2 a
S-A5B	R	Untreated	165 bc	25 ab	15.3 b	112 a	10.4 a
S-615	S	Apex	324 a	15 c	4.5 de	122 a	12.7 a
Apex	R	Apex	204 b	33 a	15.8 b	112 a	9.7 a
S-A5B	R	Apex	166 bc	28 ab	16.8 ab	115 a	3.7 bc
S-615	S	S-A5B	339 a	21 b	6.3 d	123 a	11.0 a
Apex	R	S-A5B	160 c	29 ab	18.4 a	118 a	8.6 ab
S-A5B	R	S-A5B	191 bc	25 ab	13.2 c	111 a	10.1 a
Nonrhizosphere, nonlaimosphere soil			32 d	1 d	3.7 e	32 b	3.7 bc

[u] Per gram of soil, oven-dry basis. Each value is the geometric mean of six replicates.
[v] S-A5B is a chromosome substitution line of S-615 in which chromosome 5B has been replaced by the corresponding chromosome from Apex.
[w] Relative reaction types: S = susceptible, R = resistant. See Table 1 for percentage ratings.
[x] Percent data are arithmetic means of six replicates.
[y] Laimosphere antagonists are presented only as percentages because there were no significant differences between treatments in total bacteria counts.
[z] Means in a column followed by the same letter do not differ statistically at $P = 0.01$.

TABLE 3. Population estimates of total bacteria and bacteria showing antibiosis in vitro to *Cochliobolus sativus* in the rhizospheres of wheat lines differing in susceptibility to root rot[W]

Wheat line or sample source[X]	Reaction to common root rot[Y]	Total bacteria ($\times 10^6$)	Antagonists ($\times 10^6$)	Antagonists (%)
S-615	S	183 a[Z]	11 d	5.8 b
Apex	R	94 b	27 bc	28.4 a
S-A5B	R	83 b	26 bc	31.6 a
S-A5D	S	190 a	36 b	18.9 a
Cadet	R	88 b	32 bc	36.2 a
Rescue	S	187 a	64 a	34.0 a
C-R5B	S	170 a	46 ab	28.9 a
C-R5D	R	66 c	20 c	29.7 a
Nonrhizosphere soil		33 d	2 e	5.9 b

[W]Per gram of soil, oven-dry basis. Each value represents the geometric mean of three replicates.

[X]S-A5B and S-A5D are chromosome substitution lines of S-615 in which chromosome 5B and 5D, respectively, have been replaced by the corresponding chromosome from Apex. Similarly, C-R5B and C-R5D are lines of Cadet in which chromosomes 5B and 5D, respectively, have been replaced by the corresponding chromosome from Rescue.

[Y]Relative reaction types: S = susceptible, R = resistant. See reference (8) for percentage ratings.

[Z]Means in a column followed by the same letter do not differ statistically at $P = 0.01$.

greater rhizosphere effect than did the resistant lines (Table 3). On the other hand, there was no consistent relationship between the disease reaction of the lines tested and the relative abundance of antagonistic bacteria in their rhizospheres. The populations of antagonists in the rhizospheres of Apex, S-615, and S-A5B (Table 3) showed the same relative relationships as we found before [Table 2 and (11)], but the root rot-susceptible chromosome substitution line, S-A5D, tested here for the first time, yielded as many antagonists as the resistant lines S-A5B and Apex. Similarly, in the conversely related Cadet-Rescue chromosome substitution series, more antagonists were enumerated in the rhizosphere of the root rot-susceptible donor parent, Rescue, and as many from the susceptible substitution line, C-R5B, as were enumerated in the root rot-resistant recipient parent, Cadet. The resistant line C-R5D had the fewest. When the numbers of antagonists are expressed as percentages of the total rhizosphere bacterial populations, the root rot-susceptible and -resistant lines exhibit no significant differences with the exception of S-615, which yielded fewer antagonists.

DISCUSSION.—Our failure to verify any cause-and-effect relationship between the incidence of bacteria antibiotic to *C. sativus* in vitro and resistance to common root rot, emphasizes the importance of rigorously testing such correlations before attributing any significance to them (1, 11). It also reinforces what others have repeatedly cautioned: that antibiosis in culture does not necessarily indicate antibiosis in soil (1, 16). That such must be true for the genetically related lines used in these investigations is evident not only from the results of the bacterization experiment but also from the failure of the rhizosphere analyses of additional lines to support the original correlation (11).

In the bacterization experiment, the failure of augmented laimosphere populations of antagonists to reduce the susceptibility of S-615 could be attributed to the failure of bacteria, originally isolated from Apex and S-A5B, to function antibiotically in the S-615 environment. The antibiotic potential of these microbes is probably not realized even in the rhizospheres and laimospheres in which they normally flourish. This is suggested by the failure of the resistant reaction of S-A5B to be affected by the unexpected decrease in laimosphere antagonists that occurred when the line was bacterized with the Apex culture. We offer no explanation for the decrease, which was evident in all replicates, but we do not doubt its validity: the total bacterial population estimate, which was based on the same plate dilution series as those used for enumerating the antagonists, showed no similar decrease. Use of the same dilution series for estimating both the total population and numbers of antagonists makes possible the valid calculation of the percentage of antagonists (10).

Cytogenetic evidence, implicit in the results of the rhizosphere analyses, indicates that the incidence of antagonists is not controlled by the gene we have shown to determine reaction to root rot (8). For example, the root rot-susceptible chromosome substitution line, S-A5D, has significantly more antagonists than the equally susceptible S-615 parent, indicating that the low number of antagonists characteristic of the S-615 rhizosphere is controlled by chromosome 5D as well as by 5B, even though chromosome 5D does not affect root rot reaction in this system. Furthermore, the large numbers of antagonists characteristic of the susceptible line Rescue is significant because Rescue is a selection from the hybrid of S-615 and Apex. Apparently, chromosome 5B of Rescue carries, as the result of a cross-over, the S-615 allele for root rot susceptibility and at least one Apex allele at another locus promoting high numbers of antagonists, or Rescue chromosome 5D carries a similarly effective gene inherited from Apex, or both.

Whether rhizosphere or laimosphere microbial populations play any role in determining the differential root rot reactions of these genetically related wheats is not known. Definitive studies involving the monoxenic culture of the host and pathogen are required to resolve this critical point, but the results of comparable investigations (4, 5) and our own observations indicate that rhizosphere or laimosphere populations, or both, are probably involved. We have found, for example, that rhizosphere soil from susceptible lines harbors greater numbers of cellulolytic, pectinolytic, and amylolytic bacteria than does rhizosphere soil from resistant lines (12). Enzymatic breakdown of cell walls interacting with toxin produced by the pathogen has been implicated in the development of common root rot (15).

The demonstration of a laimosphere effect about the subcrown internode of wheat raises the question of the relative contributions made by rhizosphere and laimosphere microbial populations in determining reaction to common root rot. The disease ratings in our

work were based on extent of lesioning on subcrown internodes but antibiotics or other metabolites elaborated in the rhizosphere could be translocated to, and be effective in, the subcoronal tissue (18). If microorganisms in the laimosphere are important in determining reaction to *C. sativus,* it may be the quality rather than the quantity of exudates from the subcrown internode that are critical since there was no difference in magnitude of the laimosphere effect between root rot-susceptible and -resistant lines.

LITERATURE CITED

1. BAKER, R. 1968. Mechanisms of biological control of soil-borne pathogens. Annu. Rev. Phytopathol. 6:263-294.
2. BURRAGE, R. H., and R. D. TINLINE. 1960. Common root rot and plant development following treatments of wheat seed with aldrin, gamma BHC, and heptachlor, with and without mercury fungicides. Can. J. Plant Sci. 40:672-679.
3. CHANG, I-PIN, and T. KOMMEDAHL. 1968. Biological control of seedling blight of corn by coating kernels with antagonistic microorganisms. Phytopathology 58:1395-1401.
4. COHEN, E., S. B. HELGASON, and W. C. MC DONALD. 1969. A study of factors influencing the genetics of reaction of barley to root rot caused by Helminthosporium sativum. Can. J. Bot. 47:429-443.
5. HARDING, H. 1971. Effect of Bipolaris sorokiniana on germination and seedling survival of varieties or lines of 14 Triticum species. Can. J. Bot. 49:281-287.
6. HARRIS, R. F., and L. E. SOMMERS. 1968. Plate-dilution frequency technique for assay of microbial ecology. Appl. Microbiol. 16:330-334.
7. JACKSON, R. M. 1965. Antibiosis and fungistasis of soil microorganisms. p. 363-369. *In* K. F. Baker, and W. C. Snyder [ed.]. Ecology of soil-borne plant pathogens. Univ. California Press, Berkeley.
8. LARSON, R. I., and T. G. ATKINSON. 1970. A cytogenetic analysis of reaction to common root rot in some hard red spring wheats. Can. J. Bot. 48:2059-2067.
9. MAGYAROSY, A., and J. G. HANCOCK. 1972. Microbial population of the laimosphere of squash (Cucurbita maxima). Plant Soil 37:187-190.
10. NEAL, J. L., JR. 1971. A simple method for enumeration of antibiotic-producing microorganisms in the rhizosphere. Can. J. Microbiol. 17:1143-1145.
11. NEAL, J. L., JR., T. G. ATKINSON, and R. I. LARSON. 1970. Changes in the rhizosphere microflora of spring wheat induced by disomic substitution of a chromosome. Can. J. Microbiol. 16:153-158.
12. NEAL, J. L., JR., R. I. LARSON, and T. G. ATKINSON. 1973. Changes in rhizosphere populations of selected physiological groups of bacteria related to substitution of specific pairs of chromosomes in spring wheat. Plant Soil 39:209-212.
13. SANFORD, G. B. 1959. Root-disease fungi as affected by other soil organisms. p. 367-376. *In* C. S. Holton et al. [ed.]. Plant pathology; problems and progress 1908-1958. Univ. Wisconsin Press, Madison.
14. STEVENSON, I. L. 1956. Antibiotic activity of actinomycetes in soil and their controlling effects on root-rot of wheat. J. Gen. Microbiol. 14:440-448.
15. SUBRAMANIAN, C. V. 1965. Host-pathogen interaction in root rot of wheat. Tagungsber. Dtsch. Akad. Landwirtschaftwiss. 74:133-148.
16. THAYSEN, A. C. 1950. Antibiotics in soil. Nature (Lond.) 166:93-94.
17. WEINDLING, R. 1934. Studies on a lethal principle effective in the parasitic action of Trichoderma lignorum on Rhizoctonia solani and other soil fungi. Phytopathology 24:1153-1179.
18. WINTER, A. G., H. PEUSS, and F. SCHONBECK. 1960. The influence of biotic factors on the development of soil fungi. p. 76-83. *In* D. Parkinson and J. S. Waid [ed.]. The ecology of soil fungi. Liverpool Univ. Press, Liverpool, England.
19. WOOD, R. K. S., and M. TVIET. 1955. Control of plant diseases by use of antagonistic organisms. Bot. Rev. 21:441-492.