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PRELIMINARY STUDIES IN INTEGRATED MANAGEMENT OF *PHYTOPHTHORA* CROWN AND ROOT ROTS

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Radial growth of *Phytophthora cactorum* and *P. citrophthora* was inhibited by *T. harzianum* and reduced by *P. expansum* and *A. radiobacter*. *B. cereus* did not influence the mycelial growth of both *Phytophthora* species.

Disease resistance on *P. cactorum* was induced by a local pre-inoculation with the non-pathogen *P. megasperma*, *P. capsici*, or *P. drechsleri*. Besides, it was shown that cross-protection is not systemic but is localized in the area of original inoculation.

The influence of heat and flood in the defense mechanisms of peach tree to *P. cactorum*, *P. megasperma*, *P. capsici*, and *P. drechleri* was also evaluated. Development of *P. cactorum* on plants was not influenced from any treatment. *P. megasperma* developed little necrosis only on stressed plants. In contrast, *P. capsici* and *P. drechsleri* had not the ability to infect heat and flood-treated plants.

Metalaxyl and dimethomorph inhibited the mycelial growth of both *Phytophthora* species at rates of 10 ppm but not the growth of antagonists. In contrast, fosetyl-Al inhibited radial growth of antagonists and both *Phytophthora* species at concentration as high as 500 ppm. Based on these results, metalaxyl and dimethomorph may be good candidates for integrated approach to manage *Phytophthora* diseases.

Keywords: Biocontrol; Cross-protection; Fungicides; Peach trees; *Phytophthora*

INTRODUCTION

Phytophthora cactorum and *P. citrophthora* have a wide host range and cause crown and root rots (Erwin and Ribeiro, 1996). Attempts to control *Phytophthora* disease by, for example, pre-plant soil fumigation

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(Grimm and Alexander, 1971; Harris, 1991), and by the use of resistant plants, have shown little success (McCarter *et al.*, 1978; Shokes and McCarter, 1977). Furthermore, the use of fungicides, besides being expensive and involving risks to the environment associated with the application of chemicals, is not totally effective and may lead to the appearance of new, resistant strains of pathogens (Gisi and Cohen, 1996; Grinberger *et al.*, 1995). Therefore, an integrated approach to manage *Phytophthora* crown and root rots in the orchard is recommended.

Interest in biological control of *Phytophthora* spp. that attack fruit trees has developed only recently and little information is available. *Trichoderma* spp., *Penicillium* spp. and *Bacillus* spp. have been widely used in biological control studies against *Phytophthora* spp. (Ahmed *et al.*, 1999; Fang and Tsao, 1995; He *et al.*, 1994; Milner *et al.*, 1995; Osburn *et al.*, 1995; Ownley and Benson, 1992). *Agrobacterium radiobacter* strain K84 is used widely to control diseases caused by *Agrobacterium tumefeciens* (Kerr, 1980). No attempt has been done to test the ability of A. *radiobacter* against *Phytophthora* crown and root rots.

The present study evaluates the biological control potential of *Trichoderma harzianum*, *Penicillium expansum*, *Bacillus cereus* and *Agrobacterium radiobacter* against *P. cactorum* and *P. citrophthora*. Besides, it determined if, by inoculation an actively growing plant (peach tree) with *P. megasperma*, *P. capsici*, and *P. drechsleri*, normally not pathogenic to this cultivar, it could prevent or reduce subsequent infection by a pathogenic species (*P. cactorum*). Also the present study was undertaken to determine the effect of heat and flood treatments on the defense of peach trees. Finally, the possibility of using of the fungicides metalaxyl, fosetyl-Al and dimethomorph in an integrated approach to manage *Phytophthora* crown and root rots is tested.

MATERIALS AND METHODS

Experiment 1

One isolate of each *Phytophthora cactorum*, *P. citrophthora*, *Trichoderma harzianum*, *Penicillium expansum*, *Bacillus cereus* and

Agrobacterium radiobacter were used in this experiment. All fungi were kept on potato dextrose agar (PDA) and bacteria on nutrient agar.

A. Antagonism of Trichoderma harzianum *and* Penicillium expansum *against* Phytophthora cactorum *and* P. citrophthora

The interactions were studied in petri plates (9 cm diameter) containing two different media: potato dextrose agar and V8 agar.

An agar plug (6 mm diameter) from the edge of an actively growing *Phytophthora cactorum* or *Phytophthora citrophthora* colony was transferred to each plate and a similar agar plug of *Trichoderma harzianum* or *Penicillium expansum*, cut in the same manner, was placed at a distance 3 mm. There were 40 plates for each medium. Twenty plates were used for each antagonist, 10 plates for each *Phytophthora* species. As control used plates inoculated only with *Phytophthora cactorum* or *Phytophthora citrophthora*. Plates were placed in an incubator at 23°C and any interactions were observed 6 days later.

B. Antagonism of Bacillus cereus *and* Agrobacterium radiobacter *strain K84 against* Phytophthora cactorum *and* P. citrophthora

Again the interactions was studied in petri plates. Both *Bacillus cereus* and *Agrobacterium radiobacter* were grown in nutrient broth for five days. One milliliter for each antagonist was transferred on potato dextrose agar and nutrient agar. An agar plug (6 mm diameter) from the edge of an actively growing *Phytophthora cactorum* or *P. citrophthora* colony was placed at a distance of 3 cm. Plates were incubated at 23°C in darkness. Any interactions were observed six days later.

There were 40 plates for each medium. Twenty plates were used for each antagonist, 10 plates for each *Phytophthora* species. As control used plates inoculated only with *Phytophthora cactorum* or *Phytophthora citrophthora*.

Experiment 2

One isolate for each *Phytophthora* species was used in all experiments. They were kept at the Benaki Phytopathological culture collection on cornmeal agar at 22°C.

One-year-old GF677 plants planted in 1000 cc pots containing sterile soil were used to determine the effect of heat and flood treatments on the plant defense to *P. capsici*, *P. drechsleri*, *P. megasperma*, and *P. cactorum*. Fifteen pots for each species were placed in oven for one hour at 45°C and 15 other plants were flooded for two days before inoculation. Fifteen non-treated plants were used as control. Plants were then inoculated 5 cm above soil surface with removing 6 mm trunk bark and placed agar plugs with mycelium of fungus directly on cambium. Wounds were covered with petroleum jelly and wrapped with adhesive tape to avoid desiccation. Incubation of plants were performed in glasshouse at 23–26°C for 10 days.

After incubation period, adhesive tape was removed and trunk bark around wound was scraped using a sharp knife. Results were collected by measuring the length of the resulting necrosis. Recovery of fungi was made using the selective medium developed by Jeffers and Martin (1986).

Experiment 3

Again, one isolate for each *Phytophthora* species was used in all experiments. GF677 plants were bought from a commercial tissue culture station (Vitro Hellas) and planted in the experimental field of Pomology Institute, Naoussa. Two-year-old plants were initially inoculated with *P. megasperma*, *P. capsici*, and *P. drechsleri* 10 cm above soil surface. Inoculations were made by removing a 6 mm periderm of bark using a flamed sharp knife and 6 mm agar plugs with mycelium of fungus were placed directly on cambium. To avoid desiccation, the wound was covered with petroleum jelly and wrapped with adhesive tape. Six hours, one and two days later, adhesive tape was removed and plants were re-inoculated with *P. cactorum*. Again wounds were wrapped with adhesive tape.

Ten days later, adhesive tape was removed and the trunk bark was scrapped until necrosis was exposed. Results were collected by measuring the length of necrosis.

There were 60 plants for each species, 20 for each re-inoculation time. Ten plants were re-inoculated in initial wound and other 10 plants were re-inoculated 5 cm above the initial inoculation. Also,

there were 10 plants for each species without any primary inoculation. Plants inoculated with *P. cactorum* were used as control.

Again, recovery of fungi was made using the selective medium described by Jeffers and Martin (1986).

Experiment 4

Stock solutions of fungicide were prepared by mixing metalaxyl, fosetyl-Al and dimethomorph solutions in potato dextrose agar (PDA).

Fifteen milliliters of medium containing 0, 10, 100, 500 ppm of metalaxyl, dimethomorph and fosetyl-Al were added to each of 60 petri plates per concentration, 10 for each of *Trichoderma harzianum*, *Penicillium expansum*, *Bacillus cereus*, *Agrobacterium radiobacter*, *Phytophthora cactorum* and *Phytophthora citrophthora*. One colonized agar plug of each fungus or 0.5 ml of each bacterial culture was placed in the center of each plate. Plates were incubated at 23°C and fungus or bacterial growth was recorded 4 days later

Analysis of results The experimental design used throughout experiments was completely randomized. Data were analyzed by one-way analyses of variance. All experiment were conducted three times. To combine experiments, the Bartlett's test of homogeneity of variance was used and treatment means were separated by Duncan's multiple range test ($p = 0.05$).

RESULTS

Experiment 1

A. Antagonism of Trichoderma harzianum *and* Penicillium expansum *against* Phytophthora cactorum *and* P. citrophthora

No difference was observed between potato dextrose agar and V8 agar. *Trichoderma harzianum* showed a rapid colonization of the medium which grew over the *Phytophthora cactorum* and *Phytophthora citrophthora*. *T. harzianum* hyphae coiled around of *Phytophthora cactorum* and *Phytophthora citrophthora* mycelium.

Finally, *T. harzianum* inhibited the growth of both *Phytophthora* species.

Penicillium expansum reduced the growth of both *Phytophthora* species. Mycelium of *Phytophthora* was grown only in the opposite from antagonism side. However, *Penicillium expansum* hyphae did not coil around those of *Phytophthora cactorum* or *P. citrophthora*. Growth of *Penicillium expansum* was not influenced by the presence of *Phytophthora*.

B. Antagonism of Bacillus cereus *and* Agrobacterium radiobacter *(K84) against* Phytophthora cactorum *and* P. citrophthora

Growth of both *Phytophthora cactorum* and *P. citrophthora* was not influenced by the presence of *Bacillus cereus*. In contrast, *Agrobacterium radiobacter* reduced the growth of both *Phytophthora* species. The border between *A. radiobacter* and *Phytophthora* colony was clear.

Experiment 2

None of the non-treated plant inoculated with *P. capsici*, *P. drechsleri* or *P. megasperma* caused any obvious damage to the trunk (Table I). After heating and flooding treatments, plants inoculated with *P. megasperma* were colonized by fungus. Development canker had the typical red-brown discoloration and necrosis was extended both upward and downward. Some of the plants showed the symptom of gummosis around the wound point. Significant less necrosis was observed on plants inoculated with *P. megasperma* compared with those inoculated

TABLE I Effect of heat and flood treatments on resistance of GF677 peach plants to *P. megasperma*, *P. cactorum*, *P. drechsleri*, and *P. capsici*

Pathogen	*Treatments*		
	Non-treated	*Heat-treated*	*Flood-treated*
P. cactorum	4,6[x] a[y]	4,4 a	4,65 a
P. megasperma	0 b	2,2 b	2,5 b
P. capsici	0 b	0 c	0 c
P. drechsleri	0 b	0 c	0 c

[x]Each value represents the mean of two experiments, each with 15 replicates.
[y]Values followed by different letters are significantly different ($p = 0.05$) according to Duncan's multiple range test.

with *P. cactorum*. Growth of *P. cactorum* was similar on treated and non-treated plants. In contrast, *P. capsici* and *P. drechsleri* had not the ability to infect stressed plants.

Only *P. cactorum* was recovered from non-treated plants while, *P. cactorum* and *P. megasperma* was re-isolated from at least one heat and flood–treated plant.

Experiment 3

In the cross-protection trials, observations were made 10 days after the challenge inoculations with *P. cactorum*. None of *P. megasperma*, *P. capsici*, and *P. drechsleri* showed evidence of pathogenic activity on plants (Table II). In contrast, colonization of plants inoculated with *P. cactorum* was extended and plants showed the typical crown rot symptoms.

All species gave good degree of protection against infection by challenge fungus. Measuring the length of necrosis collected the results. This local cross-protection occurred when at least 6–48 h had elapsed between inoculations for *P. megasperma*. Greatest reduction of growth of *P. cactorum* was achieved when the elapse time between inoculation with *P. megasperma* and *P. cactorum* was 2 days. Cross-protection was in evidence when at least 24–48 h had elapsed between inoculation for *P. capsici* and *P. drechsleri*. This indicated a slower activity of the defense mechanism of plants. Growth of *P. cactorum*

TABLE II Effect of prior inoculation of GF677 peach plants with nonpathogenic species of *Phytophthora* on subsequent suceptibility of these plants to *P. cactorum*

	Vertical distance of canker development					
	P. megasperma		*P. capsici*		*P. drechsleri*	
	within 5 cm	*above 5 cm*	*within 5 cm*	*above 5 cm*	*within 5 cm*	*above 5 cm*
P. cactorum	6,25[x]a[y]	6,25 a	5,2 a	5,20 a	5,28 a	5,28 a
6 h	3,64 b	6,30 a	3,8 ab	5,25 a	5,30 a	5,14 a
1 day	3,36 b	5,98 a	2,8 b	5,13 a	3,08 b	5,21 a
2 days	1,55 c	6,13 a	2,5 b	5,18 a	2,4 b	5,16 a
Nonpathogen	0 d	0 b	0 c	0 b	0 c	0 b

[x]Values are the means of two experiments, each with ten replicates for each inoculation time.

[y]Values followed by different letters are significantly different ($p = 0.05$) according to Duncan's multiple range test.

TABLE III Effect of various concentration of metalaxyl, fosetyl-Al and dimethomorph on growth of *Phytophthora cactorum*, *P. citrophthora*, *Trichoderma harzianum*, *Penicillium expansum*, *Bacillus cereus* and *Agrobacterium radiobacter*

Fungicides	*A*[x]	*B*	*C*	*D*	*E*	*F*
Metalaxyl						
Control	4,88[y] a[z]	2,1 a	2,5 a	3,1 a	2,55 a	2,1 a
10 ppm	5,2 a	2,2 a	0 b	0 b	2,5 a	2,25 a
100 ppm	3,1 b	1,4 b	0 b	0 b	0 b	0 b
500 ppm	0 c	0 c	0 b	0 b	0 c	0 c
Dimethomorph						
Control	5,1 a	1,9 a	2,3 a	3,05 a	2,4 a	2,5 a
10 ppm	5,6 a	1,95 a	0 b	0 b	2,2 a	2,3 a
100 ppm	1,2 b	1,8 b	0 b	0 b	0,9 b	1,7 b
500 ppm	0 c	0 c	0 b	0 b	0 c	0 c
Fosetyl-Al						
Control	4,9 a	2,3 a	2,5 a	2,9 a	2,5 a	2,4 a
10 ppm	5 a	2,15 a	2,6 a	3,1 a	2,45 a	2,25 a
100 ppm	5,3 a	2 a	2,55 a	2,85 a	2,25 a	2,2 a
500 ppm	0 b	0 b	0 b	0 b	0 b	0

[x] A = *Trichoderma harzianum*, B = *Penicillium expansum*, C = *Phytophthora cactorum*, D = *Phytophthora citrophthora*, E = *Bacillus cereus*, F = *Agrobacterium radiobacter*.
[y] Each value is the mean of three experiments, each with ten replicates.
[z] Values followed by different letters are significant different ($p = 0.05$) according to Duncan's multiple range test.

was not influenced when the inoculation was made at 5 cm above initial inoculation for at least 6–48 h.

P. cactorum was recovered from at least one plant in any treatment.

Experiment 4

Growth of *Phytophthora cactorum* and *P. citrophthora* was inhibited *in vitro* by metalaxyl and dimethomorph at rates of 10 ppm. In contrast, development of *Trichoderma harzianum*, *Penicillium expansum*, *Bacillus cereus* and *Agrobacterium radiobacter* was not influenced by metalaxyl and dimethomorph at concentration as low as 10 ppm. Dimethomorph reduced significant the growth of antagonists at rates of 100 ppm. Metalaxyl reduced also the development of *T. harzianum* and *P. expansum* and inhibited the growth of *Bacillus cereus* and *Agrobacterium radiobacter* at rates of 100 pm. Both metalaxyl and dimethomorph inhibited the growth of antagonists at concentration as high as 500 ppm (Table III).

Fosetyl-Al did not influence the development of all antagonists and *Phytophthora* species at rates of 100 ppm. In contrast, this fungicide inhibited their growth at concentration as high as 500 ppm.

DISCUSSION

In this work, *Trichoderma harzianum*, *Penicillium expansum* and *Agrobacterium radiocacter* produced good results in controlling *P. cactorum* and *P. citrophthora*. *T. harzianum* inhibited the radial growth of *Phytophthora* species apparently by direct antagonism with minor inhibition by antibiosis. It has been found that *T. harzianum* was promising as biological control agent of *Phytophthora* (Ahmed *et al.*, 1999; Cruz and Cisterna, 1998; Roiger and Jeffers, 1995; Smith *et al.*, 1990). Lederer *et al.* (1992), found that *Trichoderma* spp. produced antibiotics leading to inhibition of the mycelial growth of *Phytophthora cactorum*. *Penicillium expansum* could reduce the radial growth of both *Phytophthora cactorum* and *P. citrophthora* apparently by antibiosis. Fang and Tsao (1995) reported that *Penicillium funiculosum* reduced *Phytophthora* root rot of azalea and sweet orange. Also, *Penicillium janthinellum* reduced the incidence of *Phytophthora* root rot of azalea (Ownley and Benson, 1992). *Bacillus cereus* could not influence the radial growth of both *Phytophthora* species. In contrast, it has been reported that *Bacillus cereus* may have potential as a biocontrol agent against *Phytophthora* diseases (Handelsman *et al.*, 1990; Osburn *et al.*, 1995; Stabb *et al.*, 1994). This bacterium produces antibiotics that inhibit the growth of plant pathogen *Phytophthora medicaginis* (He *et al.*, 1994; Milner *et al.*, 1995). Radial growth of both *Phytophthora* species seemed to be reduced from *Agrobacterium radiobacter*. The intensity of inhibition did not differ according to the medium used. In contrast, Ahmed *et al.* (1999), found that *T. harzianum* reduced the growth of *P. cactorum* more on Czapek than V8c and Water agar media.

In the cross-protection tests with *P. megasperma*, *P. drechsleri*, and *P. capsici*, the reduction of growth of *P. cactorum* on peach trees was found. This was lasting protection since it was effective for at least 2 days. Similarly Paxton and Chamberlain (1967), protected soybean plants against a virulent race of *P. sojoe* by prior inoculation with an

avirulent race. Also, Stromberg (1995), induced resistance in potato cultivars to late blight by a local pre-inoculation with the non-pathogen *P. cryptogea*. Agrios (1988), reported that when a pathogen comes in contact with a host cell an early event takes place that triggers a fairly rapid response in each organism that either allows or impedes further growth of the pathogen and development of disease. It may be one of many biochemical substances, structures, and pathways. Many researchers have reported phytoalexins as the cause for inhibiting fungal development (Chamberlain and Gerdemann, 1966; Egea *et al.*, 1996a,b; Jones *et al.*, 1974; Klarman and Gerdemann, 1963; Miller *et al.*, 1984; Musell and Staples, 1971; Olah *et al.*, 1985; Paxton and Chamberlain, 1967; Shih and Kuc, 1973).

The required time for activating of the plant defense is also possible to depend on *Phytophthora* species. For this reason the elapsed time for reducing of growth of challenging fungus was shorter for *P. megasperma* compared with other tested species. Martyn (1991), reported that pathogens closely related to the challenge isolate are better inducers of resistance than those that are non-pathogens or pathogens of unrelated hosts.

Cross-protection was not systemic because only those cells found around the initial inoculation point were capable of restricting the growth of subsequently introduced pathogens. The results agree with those obtained by Paxton and Chamberlain (1967).

As only plant inoculated with *P. megasperma* developed necrosis when they were heat and flood-treated, this showed that resistance to non-pathogens is not governed by a single mechanism. Similar results were obtained by Chamberlain (1972). A partial suppression of biochemical reactions that take place in the cells and tissues is an answer. It is possible that the concentration of phytoalexin was not reduced enough so that *P. drechsleri* and *P. capsici* could grow on plants and also different concentration of phytoalexin is possible inhibitory for each *Phytophthora* species.

The radial growth of both *Phytophthora* species was inhibited by metalaxyl and dimethomorph at rates of 10 ppm. In contrast, development of antagonists did not influence at the same concentration. Their mycelial growth was inhibited at concentration as high as 500 ppm. Based on these results, metalaxyl and dimethomorph may be good candidates for integrated approach to manage *Phytophthora* diseases. The

results also showed that fosetyl-Al could not use in integrated management of *Phytophthora* diseases because it inhibited both antagonists and *Phytophthora* at the same concentration.

References

Ahmed, A.S., Perez-Sanchez, C., Egea, C. and Candela, M.E. (1999). Evaluation of *Trichoderma harzianum* for controlling root rot caused by *Phytophthora capsici* in pepper plants. *Plant Pathology*, **48**, 58–65.

Agrios, N.G. (1988). Plant Pathology. Third Edition, Academic Press, 97.

Chamberlain, D.W. (1972). Heat-induced susceptibility to nonpathogens and cross protection against *Phytophthora megasperma* var. *sojoe* in soybean. *Phytopathology*, **62**, 645–646.

Chamberlain, W.D. and Gerdemann, J.W. (1966). Heat-induced susceptibility of soybeans to *Phytophthora megasperma* var. *sojoe*, *Phytophthora cactorum* and *Helminthosporium sativum*. *Phytopathology*, **56**, 70–73.

Cruz, M.A. and Cisterna, V.O. (1998). Integrated control of *Phytophthora capsici* in pepper. I. Effect of antagonist fungi on plant growth. *Agricultura Tecnica*, **58**, 81–92.

Egea, C., Alcazar, D.M. and Candela E.M. (1996a). Capsoidiol: its role in the resistance of *Capsicum annum* to *Phytophthora capsici*. *Physiologia Plantarum*, **98**, 737–742.

Egea, C., Perez, M.D.M. and Candela, E.M. (1996b). Capsidiol accumulation in *Capsicum annuum* stems during the hypersensitive reaction to *Phytophthora capsici*. *J. Plant Physiol.*, **149**, 762–764.

Erwin, D. and Ribeiro, O. (1996). *Phytophthora* Diseases Worldwide, 245, 288 pp.

Fang, J.G. and Tsao, P.H. (1995). Efficacy of *Penicillium funiculosum* as a biological control agent against *Phytophthora* root rots of azalea and citrus. *Phytopathology*, **85**, 871–878.

Gisi, U. and Cohen, Y. (1996). Resistance to phenylamide fungicides: A case study with *Phytophthora infestans* involving mating type and race stractur. *E. Ann. Rev. Phytopathol.*, **34**, 549–572.

Grimm, R.G. and Alexander, F.A. (1971). Fumigation of *Phytophthora* in sandy soil by surface application of methyl bromide and methyl bromide-chloropicrin. *Plant Disease*, **55**, 929–932.

Grinberger, M., Kadish, D. and Cohen, Y. (1995). Infectivity of metalaxyl-sensitive and -resistant isolates of *Phytophthora infestans* to whole potato tubers as affected by tuber aging and storage. *Phytoparasitica*, **23**, 165–175.

Handelsman, J., Raffel, S., Wunderlich M.L. and Grau, C.R. (1990). Biological control of damping-off of alfalfa seedlings with *Bacillus cereus* UW85. *Applied and Environmental Microbiology*, **3**, 713–718.

Harris, C.D. (1991). A comparison of dazomet, chloropicrin and methyl bromide as soil disinfestants for strawberries. *Journal of Horticulture Science*, **66**, 51–58.

He, H., Silo-Suh, L.A., Handelsman, J. and Clardy, J. (1994). Zwittermicin A, an antifungal and plant protection agent from *Bacillus cereus*. *Tetrahedron Letters*, **35**, 2499–2502.

Jeffers, S.N. and Martin, S.B. (1986). Comparison of two media selective for *Phytophthora* and *Pythium* species. *Plant Diseases*, **70**, 1038–1043.

Jones, D.R., Graham, W.G. and Ward, W.E.B. (1974). Ultrastructural changes in pepper cells in a compatible interaction with *Phytophthora capsici*. *Phytopathology*, **64**, 1084–1090.

Kerr, A. (1980). Biological control of crown gall through production of Agrocin 84. *Plant Disease*, **64**, 25–33.

Klarman, W.L. and Gerdemann, J.W. (1963). Resistance of soybeans to three *Phytophthora* species due to the production of Phytoalexin. *Phytopathology*, **53**, 1317–1320.

Lederer, W., Lorens, K.H. and Seemuller, E. (1992). Studies on antagonistic effects of *Trichoderma* isolates against *Phytophthora cactorum*. *J. Phytopathology*, **136**, 154–164.

Martyn, J.D. (1991). Induced resistance to *Fusarium* wilt of watermelon under simulated field conditions. *Plant Diseases*, **75**, 847–877.

McCarter, S.M., Jaworski, C.A. and Johnson, A.W. (1978). Effect of continous plant culture and soil fumigation on soilborne plant pathogens and on growth of tomato transplants. *Phytopathology*, **68**, 1475–1481.

Miller, S.A., Davidse, L.C. and Maxwell, P.D. (1984). Expression of genetic susceptibility, host resistance, and nonhost resistance in alfalfa callus tissue inoculated with *Phytophthora megasperma*. *Phytopathology*, **74**, 345–348.

Milner, J.L., Raffel, S.J., Lethbridge, B.J. and Handelsman, J. (1995). Culture conditions that influence accumulation of zwittermicin A by *Bacillus cereus* UW85. *Appl. Microbiol. Biotechnol.*, **43**, 685–691.

Mussell, W.H. and Staples, R.C. (1971). Phytoalexin compounds apparently involved in strawberry resistance to *Phytophthora fragariae*. Phytoalexin, **61**, 515–517.

Osburn R.M., Milner, J.L., Oplinger, E.S., Smith, R.S. and Handelsman, J. (1995). Effect of *Bacillus cereus* UW85 on the yield of soybean at two field sites in Wisconsin. *Plant Diseases*, **79**, 551–556.

Olah, A.F., Schmitthenner, A.F. and Walker, A.K. (1985). Glyceollin accumulation in soybean lines tolerant to *Phytophthora megasperma* f. sp. *glycinea*. *Phytopathology*, **75**, 542–546.

Ownley, B.H. and Benson, D.M. (1992). Evaluation of *Penicillium janthinellum* as a biological control of *Phytophthora* root rot of azalea. *J. Amer. Soc. Hort. Sci.*, **117**, 407–410.

Paxton, J.D. and Chamberlain, D.W. (1967). Acquired local resistance of soybean plants to *Phytophthora* sp. *Phytopathology*, **12**, 352–353.

Roiger, D.J. and Jeffers, S.N. (1991). Evaluation of *Trichoderma* spp. for biological control of *Phytophthora* crown and root rot of apple seedlings. *Phytopathology*, **81**, 910–917.

Shih, M. and Kuc, J. (1973). Incorporation of ^{14}C from acetate and mevalonate into rishitin and steroid glycoalkaloids by potato tuber slices inoculated with *Phytophthora infestans*. *Phytopathology*, **63**, 826–829.

Shokes, F.M. and McCarter, S.M. (1977). Occurrence of plant pathogens in irrigation ponds in southern Georgia. *Proc. Am. Phytopathol. Soc.*, **3**, 342 (abstr.).

Smith, V.L., Wilcox, W.F. and Harman, G.E. (1990). Potential for biological control of *Phytophthora* root and crown rots of apple by *Trichoderma* and *Gliocladium* spp. *Phytopathology*, **80**, 880–885.

Stabb, E.V., Jacobson, M.L. and Handelsman, J. (1994). Zwittermicin A-Producing strains of *Bacillus cereus* from diverse soils. *Applied and Environmental Microbiology*, **60**, 4404–4412.

Stromberg, A. (1995). Systemically induced resistance in potato cultivars with different degree of resistance to late blight caused by *Phytophthora infestans* (Mont.) de Bary. *J. Phytopathology*, **143**, 27–31.