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Enhancement of Biological Control of *Botrytis cinerea* on Cucumber by Foliar Sprays and Bed Potting Mixes of *Trichoderma harzianum* YC459 and Its Application on Tomato in the Greenhouse

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***Trichoderma harzianum* YC459 (Th 459), isolated from sawdust compost, was effective in controlling cucumber and tomato gray mold caused by *Botrytis cinerea* under controlled and plastic film tunnel conditions. A water suspension of the wettable powder formulation of Th 459 significantly ($P \leq 0.05$) reduced the severity of cucumber gray mold by foliar spraying at all tested concentrations from 10^5 to 10^8 colony forming unit (cfu)/ml in repeated experiments. The control efficacy was maintained at least seven days with the average control value of 70% in cucumber pot tests. Mixing one to eight grams of the granular formulation (10^8 cfu/g dry weight) of Th 459 into one liter nursery potting mix at seeding also significantly ($P \leq 0.05$) reduced the severity of cucumber gray mold by suppression of lesion formation three weeks after treatment. Application of mixing granular formulation at seeding in combination with foliar spraying during cultivation provided a more significant reduction ($P \leq 0.05$) of cucumber gray mold than granule mixing or leaf spray alone. The foliar spraying of the formulated wettable powder of Th 459 significantly ($P \leq 0.05$) reduced the infection of tomato fruits by *B. cinerea* as effective as the chemical fungicide, dichlofluanid, in three plastic film tunnel experiment trials. It is suggested that effective control of gray mold of cucumber and tomato can be provided by both treatment of Th 459 into potting mix and foliar spray through induction of systemic resistance and direct inhibition of the pathogen.**

Keywords : biological control, *Botrytis cinerea*, cucumer gray mold, induced resistance, *Trichoderma harzianum*

Gray mold caused by *Botrytis cinerea* is one of the major disease of vegetables, ornamentals and fruit crops produced in commercial greenhouses and fields all over the world. The pathogen attacks flowers, leaves and mature fruits in the pre- and post-harvest stages (Agrios, 1997). The effective method for control of gray mold has been chemical sprays with benzimidazole, dicarboximide and other fungicides in most greenhouses during the last two decades. However, *B. cinerea* has developed resistance to these fungicides by repeated applications of potent fungicides (Choi et al., 1995; Grindle et al., 1981). In addition, public concern about fungicide residues in edible products and environment has accelerated the search for alternative disease control strategies.

Biological control using antagonistic microorganisms is an alternative control method to the fungicide use and provides an ecologically based approach to integrated pest management in sustainable agriculture in crop production systems (Cook and Granados, 1991; Singh et al., 1999; Sutton and Peng, 1993). Antagonistic microorganisms such as *Trichoderma, Bacillus*, and *Pseudomonas* species have been often evaluated for the control of *B. cinerea* and developed for commercial products (De Meyer et al., 1998; Harman, 2000; Nelson and Powelson, 1988; Paulitz and Belanger, 2001; Redmond et al., 1987). A *Trichoderma*-based biofungicide, TRICHODEX (Makhteshim Chemical Works Ltd., Beer Sheva, Israel) is now commercially available for the control of *B. cinerea* (Elad, 2000a; 2000b). Multiple mechanisms of action including antibiosis, competition, mycoparasitism via production of chitinases and glucanases, solubilization of inorganic plant nutrients and inactivation of pathogen's enzymes involved in the infection process have been suggested for the biocontrol of *Trichoderma* species against *B. cinerea* (Altomare et al., 1999; Elad and Kapat, 1999; Hjeljord et al., 2001; Lorito et al., 1993). The mechanism of induced resistance for defense responses by *T. harzianum* has also been provided (De Meyer et al., 1998; Harman, 2000; Yedidia et al., 1999; 2000). Localized and systemic resistance responses are induced by *Trichoderma* species as avirulent symbionts of plants (Harman et al., 2004; Horst et al., 2005).

During the study on development of biocontrol agents for control of gray mold on greenhouse crops, many bacterial and fungal antagonists were isolated at Jinju area in 1993 (Chung et al., 1993). Among fungal antagonists, Th 459

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isolated from sawdust compost produced abundant aerial spores and parasitized *Botrytis* mycelia rapidly by paired bioassay. The isolate was selected and its control efficacy was tested by foliar sprays in preliminary tests. Even drenching of spore suspension into rhizosphere soil was found to be effective in the control of gray mold by farmers in agricultural farms (unpublished information). The objectives of this work were to evaluate the efficacy of Th 459 for its ability to control gray mold of cucumber and tomato by foliar sprays and specifically to investigate the synergistic effect of foliar sprays and bed soil mixing.

Materials and Methods

Preparation of *T. harzianum* YC459 and pathogen inoculum. The formulations of Th 459 were developed in cooperation with JGreen Inc. (Changnyung, Gyeongnam province, Korea) in 2000 and concentrated conidia (10^{11} cfu/g dry weight), wettable powder (10^9 cfu/g dry weight) and granular (10^8 cfu/g dry weight) formulations of the commercial product (TORY®) were provided by the company. Conidial suspension of Th 459 was prepared by mixing concentrated conidia in water containing Tween 20 surfactant (250 μg/ml) and its population density was assessed by determining with the dilution plate method on a selective medium (Chung and Hoitink, 1990).

Pathogenic *B. cinerea* JM43 isolated from diseased tomato at Jinju area and stored at our laboratory was retrieved from the storage tube and cultured for 14 days at 20°C on potato dextrose agar (PDA, Difco). Plates were then flooded with sterilized distilled water and conidia were scraped with a rubber stick. Mycelial debris was removed by filtration through double layer cheesecloth (Choi et al., 1995). The conidial concentration was determined by using hemacytometer, and the concentration was adjusted to 10^5/ml with sterilized distilled water. Cucumber leaves of 2-4 leaf stage were then sprayed to runoff with this inoculum containing 0.1% (w/v) potato dextrose broth.

Plant growth conditions. For pot experiments, two seeds of cucumber (cv. Nambu chungjang, Seoul seed co., Korea) were planted into a commercial soilless, peat-based potting mix (Biomedia Co., Gyeongju, Korea) in 5-cm plastic pot containing 15% zeolite, 5% diatomaceous earth, 70% shredded coconut shell pieces, and 10% (vol/vol) Canadian sphagnum peat moss. The pots were placed in a plant growth chamber with a 12-h photoperiod and a temperature range of 23 to 25°C for three weeks and watered manually every day. Each pot served as a replicate and each treatment was replicated four times. For field experiments, tomato (cv. Seo-kwang, Heungnong seed Co., Korea) were cultivated in the plastic film tunnels (length 24 m × width 7.0 m × height 2.3 m) located at Sacheon, Cheongju, and Soonchun area. Each plot consisted of a single row 10-m long and 0.5m wide and plots were spaced 0.5m apart within the row. Other cultivation was followed by farmer's conventional methods. The experimental conditions recorded fluctuating diurnal temperatures 15-25°C and relative humidity 80-95% as common in winter.

Evaluation of biocontrol efficacy. In the laboratory tests, conidial suspension of Th 459 (10^5~10^8 cfu/ml) was sprayed to cucumber leaves at 3 to 4-leaf stages one day before inoculation of the *B. cinerea*. After inoculation of the pathogen (10^5 conidia/ml) to *Trichoderma* applied cucumber leaves, the plants were placed in a dew chamber at 20°C. Disease severity of the older two leaves was determined when symptoms developed at 3 to 5 days after inoculation on the basis of percentage of total leaf surface with necrotic lesions. To determine the residual control efficacy of foliar spraying of Th 459, conidial suspension (10^5 cfu/ml) was sprayed to 2-leaf stage cucumber leaves until runoff at 1, 3, 5, 7, and 9 days before inoculation of the *B. cinerea* and the severity was assessed as described above. For granules treatment, potting mixes were inoculated with the granule formulations of Th 459 (10^8 cfu/g dry weight) at a rate of one to eight grams/liter mix at seeding. Control plants mixes were not treated with Th 459. All plants were placed at 25°C after seeding and the disease severity of cucumber leaves was assessed three weeks after seeding.

In plastic film tunnel experiments, the treatments were arranged in a completely randomized design with three replicate plots per treatment. Each plot was 20 m^2 and 10 plants were cultivated per plot. One rate of the formulated Th 459, 250 g (wettable powder) in 250 liter water containing surfactant (250 μg/ml, a.i. 60% alkylaryl polyethoxylate and sodium alkylsulfonated alkylate, Dongbu Hannong, Korea) was applied to tomato until runoff three times every seven days at the beginning of the gray mold development during March, 2002 (Soonchun and Cheongju) and February, 2003 (Sacheon). The fungicide dichlofluanid (a.i. 40% wettable powder, Dongbu Hannong) was used as a standard chemical fungicide. Disease incidence was determined by counting the number of naturally infected fruits by the gray mold fungus seven days after the final spray.

Statistical analysis. All experiments were arranged in a completely randomized design. Data were analyzed by analysis of variance (ANOVA) and means were compared using the Duncan's multiple range test. Statistical analysis was performed with SAS (version 9.0; SAS Institute).

Results

Evaluation of biocontrol efficacy. The biocontrol agent Th 459 sprayed to cucumber leaves followed 24 h later by inoculation of *B. cinerea* significantly (P≤0.05) reduced the percentage of diseased leaves by the mean of 76.6% compared with control at all tested concentrations (10^5, 10^6, 10^7, and 10^8 cfu/ml) in repeated experiments. At the concentrations of 10^5 and 10^8 cfu/ml, the severity of disease was 20% and 8%, respectively, compared with 98% of control (Fig. 1). The control efficacy of Th 459 lasted at least seven days when conidial suspension (10^5 cfu/ml) was sprayed to cucumber leaves 1, 3, 5, 7, 9 days before inoculation of the pathogen with disease severity of 22, 18, 20, 24, and 38%, respectively, at the growth chamber (Fig. 2).

Mixing one to eight grams of Th 459 (10^8 cfu/g dry wt) granules into one liter container medium at seeding resulted in a significant (P≤0.05) reduction in the severity of cucumber leaves gray mold relative to non-added control. Disease severity at the doses of 1, 2, 4, 8 g/liter potting mix was 20, 36, 28, 22%, respectively, and a typical dose-response relationship was not observed in repeated experiments (Fig. 3). When combination of granule mixing into potting mix and foliar spray of Th 459 was applied, a significant reduction in disease severity was achieved in six times repeated experiments (Fig. 4). The combination of both treatments was the most effective in reducing cucumber gray mold severity of 12%. Disease severity of cucumber treated with granule mixing or leaf spray alone was 32% and 22%, respectively, compared with the untreated control of 65% (Fig. 4).

In plastic film tunnel experiments, tomatoes sprayed with conidial suspension of Th 459 had significantly (P≤0.05) less disease than the untreated control plants (Fig. 5). The percentage of naturally infected tomatoes by *B. cinerea* in

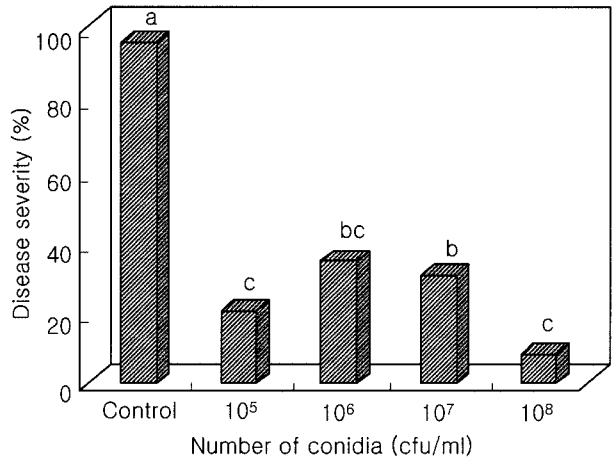

Fig. 1. Effects of *Trichoderma harzianum* YC459 at various spore concentrations on the severity of gray mold of cucumber leaves produced in a potting mix. Spore suspension of *T. harzianum* YC459 was sprayed to cucumber seedlings (3-4 leaf stage) one day before inoculation of the *B. cinerea*. Disease severity was determined at 3 to 5 days after inoculation on the basis of percentage of total leaf surface with necrotic lesions. Bars are the average of four replications per treatment. Bars with the same letter are not significantly different at P≤0.05.

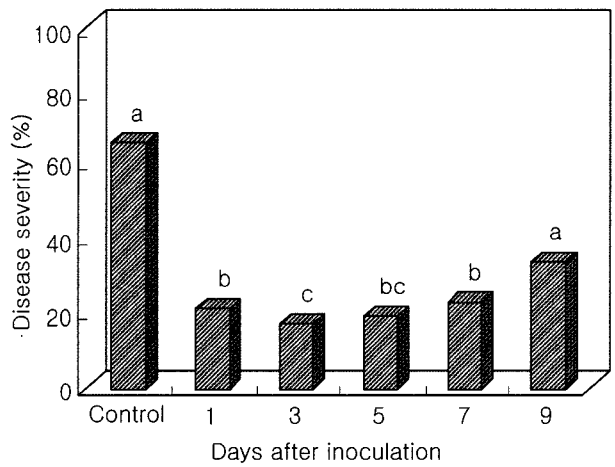

Fig. 2. Residual efficacy of *Trichoderma harzianum* YC459 for control of gray mold of cucumber leaves produced in a potting mix. Spore suspension (10^5 cfu/ml) of *T. harzianum* YC459 was sprayed to 2-leaf stage cucumber leaves until runoff at indicated days before inoculation of the *B. cinerea* (10^5 conidia/ml) and the severity was assessed at 3 to 5 days after inoculation on the basis of percentage of total leaf surface with necrotic lesions. Bars are the average of four replications per treatment. Bars with the same letter are not significantly different at P≤0.05.

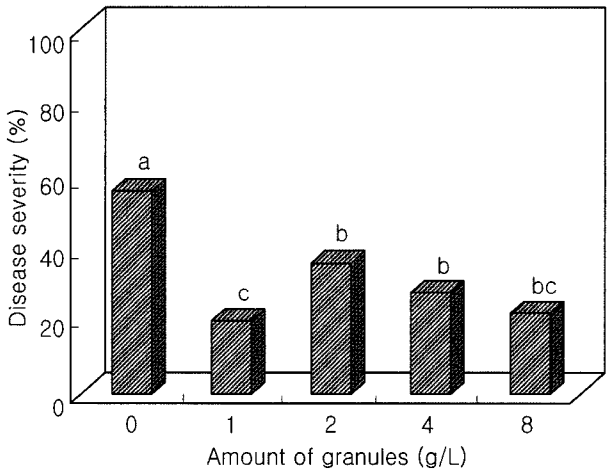

Fig. 3. Effects of granule mixing of *Trichoderma harzianum* YC459 in potting mix on the severity of gray mold of cucumber leaves. Potting mixes were inoculated with the granule formulations of *T. harzianum* YC459 (10^8 cfu/g dry weight) at seeding and the disease severity of cucumber leaves was assessed three weeks after seeding on the basis of percentage of total leaf surface with necrotic lesions. Bars are the average of four replications per treatment. Bars with the same letter are not significantly different at P≤0.05.

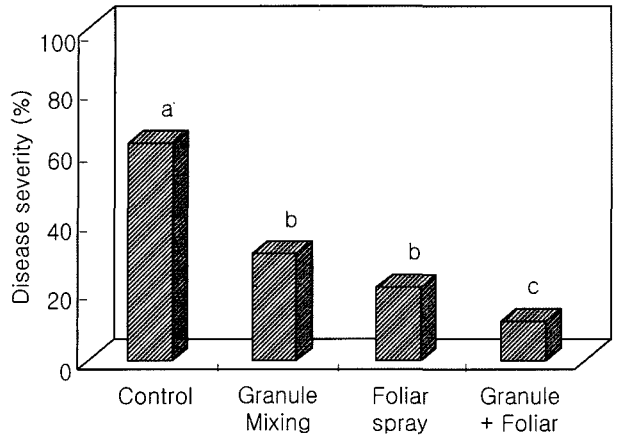

Fig. 4. Effects of different application methods of *Trichoderma harzianum* YC459 on the severity of gray mold of cucumber leaves. Granule mixing: one litter potting mix was inoculated with the one gram granule of *T. harzianum* YC459 (10^8 cfu/g dry weight) at seeding. Foliar spray: spore suspension (10^5 cfu/ml) of *T. harzianum* YC459 was sprayed to 3-week old cucumber leaves one day before inoculation of the *B. cinerea* (10^5 conidia/ml). Granule+Foliar: both granule mixing and foliar spray were applied. Disease severity was assessed at 3 to 5 days after inoculation of the pathogen on the basis of percentage of total leaf surface with necrotic lesions. Bars are the average of four replications per treatment. Bars with the same letter are not significantly different at $P \leq 0.05$.

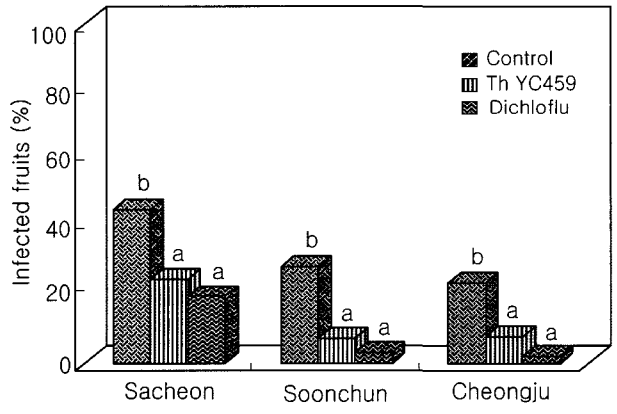

Fig. 5. Effects of foliar sprays with *Trichoderma harzianum* YC459 and dichlofluanid (Dichloflu, a.i. 40% wettable powder) on the infection of tomato fruits under plastic film tunnels. One rate of the formulated *T. harzianum* YC459, 250g (wettable powder) in 250 liter water containing surfactant was applied to tomato until runoff three times every seven days at the beginning of the gray mold development during March, 2000 (Soonchun and Cheongju) and February, 2003 (Sacheon). The number of infected fruits by the gray mold fungus was counted seven days after the final spray. Bars are the average of three replications per treatment. Bars with the same letter are not significantly different at $P \leq 0.05$.

treated plots was 7.6% and 8.0% at Soonchun and Cheongju, whereas that of the untreated plants was 28.5% and 23.7%, respectively. In the experiment at Sacheon area, the infection rate of treated tomatoes was 24.9%, but that of untreated plants was 45.3%, significantly higher than control plants. All treatments with Th 459 did not differ significantly from the fungicide spray of dichlofluanid in reducing gray mold. Especially, senescent petal falls were frequently observed on many tomato fruits sprayed with Th 459 within three to five days after treatments.

Discussion

Strain Th 459 reduced gray mold of cucumber leaves significantly at the ranges from 10^5 to 10^8 cfu/ml without dosage-response relationship. It appears that the conidial concentration of Th 459, 10^5 cfu/ml, is enough for suppression of *B. cinerea* in this experiment. The average control value of this strain was 77%, which is higher than that of the other commercially developed *Trichoderma* strains T-39 (Trichodex), 35-44% under similar conditions of disease development (Elad, 2000; Freeman et al., 2004). Foliar sprays of *Trichoderma* conidial suspension containing 10^7 to 10^8 cfu/ml are considered to be necessary to suppress *B. cinerea* in many other experiments (Gullino, 1992; Hjeljord and Tronsmo, 2003; Hong et al., 1998). However, Th 459 was not in the case probably due to its higher effectiveness on the suppression of the pathogen and colonization activity of old plant tissues than other strains at the low concentration (Kim and Chung, 2004). The residual control activity of Th 459 lasted seven days after spray on cucumber leaves, which showed that this strain would survive well on cucumber leaves until one week after spray in growth chamber. The same trend was observed in other test using T-39 (Freeman et al., 2004). Th 459 is likely to be effective in reducing gray mold of many crops on a seven-day spray schedule in commercial greenhouses during winter.

Spores in wettable powder or granule formulations of Th 459 effectively controlled gray mold of cucumber, but there was no significant difference in the efficacy with increasing doses of spore suspension or granule of Th 459. Lewis and Papavizas (1985) also found that no correlation existed between cfu of *Gliocladium* and *Trichoderma* species in soil and extent of survival and saprophytic growth of *R. solani*. Therefore, high titer in formulations may not be critical for Th 459, if the minimal amount of propagules which can reduce inoculum potential of the pathogen is applied for biocontrol and its mechanism is systemic induced resistance (Honeycutt and Banson, 2001). Control of gray mold of cucumber leaves by inoculation of Th 459 into potting mix confirms earlier reports on systemic control of *B. cinerea* by *T. harzianum* T-39 and *T. hamatum* T382 (De Meyer et al., 1998; Horst et al., 2005). A combination of the application of granular formulation into potting mix at seeding and foliar sprays during cultivation could

provide significantly more effective reduction of cucumber gray mold than granule mixing or leaf spray alone. It seems that the synergism between resistance induction and direct inhibition may occur in the control of *B. cinerea* (Elad and Kapat, 1999; Elad, 2000b; Hjeljord et al., 2001; Horst et al., 2005).

Control efficacy of the strain Th 459 was comparable to chemical sprays with the fungicide, dichlofluanid for control of tomato gray mold under plastic film tunnel conditions. Th 459 was similar or superior to other commercial biocontrol agents, *T. harzianum* T-39 and T-22 in reducing gray mold incidence of tomato fruits (Elad, 2000a, b; Harman, 2000). In many preliminary experiments of commercial farms, Th 459 was found to be effective against *B. cinerea* on greenhouse tomatoes, strawberry, cucumber, zucchini squash, wild sesame and hot pepper. In addition, Th 459 could remove old flower petals which can be a possible source of the pathogen inoculum by active colonization of these tissues (Kim and Chung, 2004). The application of the strain Th 459 may allow growers an opportunity to limit the use of chemical fungicides and rely more on biological measures for control of gray mold. This strain was developed as a biofertilizer in 2000 and marketed in Korea and the process is undergoing for registration of Th 459 as a biofungicide.

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References

Agrios, G. N. 1997. Plant Pathology. Academic Press, New York, NY, USA. 606 pp.

Altomare, C., Norvell, W. A., Bjokman, T. and Harman, G. E. 1999. Solubilization of phosphates and micronutrients by the plant-growth promoting and biocontrol fungus *Trichoderma harzianum* Rifai 1295-22. *Appl. Environ. Microbiol.* 65:2926-2933.

Choi, I. S., Chung, Y. R. and Cho, K. Y. 1995. Variations in phenotypic characteristics, pathogenicity and fungicide resistance of *Botrytis cinerea*, gray mold rot fungus, isolated from various host plants. *Kor. J. Mycol.* 23:246-256.

Chung, Y. R. and Hoitink, H. A. J. 1990. Interactions between thermophilic fungi and *Trichoderma hamatum* in suppression of *Rhizoctonia* damping-off in a bar compost-amended container medium. *Phytopathology* 80:73-77.

Chung, Y. R., Shin, W. G. and Kang, S. W. 1993. Development of the microbial pesticide for controlling gray mold rot of greenhouse crops. Research Report. Rural Development Administration of Gyeongnam Province, Jinju, Korea. 24 pp.

Cook, R. J. and Granados, R. R. 1991. Biological control: making it work. In: *Agricultural Biotechnology at the cross roads*, ed. M. J. F. MacDonald, pp. 213-227. National Agricultural Biotechnology Council, Ithaca.

De Meyer, G., Bigirimana, J., Elad, Y. and Höfte, M. 1998. Induced systemic resistance in *Trichoderma harzianum* T39 and biocontrol of *Botrytis cinerea*. *Eur. J. Plant Pathol.* 104:279-286.

Elad, Y. and Kapat, A. 1999. The role of *Trichoderma harzianum* protease in the biocontrol of *Botrytis cinerea*. *Eur. J. Plant Pathol.* 105:177-189.

Elad, Y. 2000a. *Trichoderma harzianum* T39 preparation for biocontrol of plant diseases-control of *Botrytis cinerea, Sclerotinia sclerotiorum* and *Cladosporium fulvum*. *Biocontrol Sci. and Tech.* 10:499-507.

Elad, Y. 2000b. Biological control of foliar pathogens by means of *Trichoderma harzianum* and potential modes of action. *Crop Protect.* 19:709-714.

Freeman, S., Minz, D., Kolesnik, I., Barbul, O., Zveibil, A., Maymon, M., Nitzani, Y., Kirshner, B., Rav-David, D., Bilu, A., Dag, A., Shafir, S. and Elad, Y. 2004. *Trichoderma* biocontrol of *Colletotrichum acutatum* and *Botrytis cinerea* and survival in strawberry. *Eur. J. Plant Pathol.* 110:361-370.

Grindle, M. 1981. Variations among field isolates of *Botrytis cinerea* in their sensitivity to antifungal compounds. *Pest Sci.* 12:305-312.

Gullino, M. L. 1992. Control of Botrytis rot of grapes and vegetables with *Trichoderma* spp. In: *Biological control of plant diseases*, ed. by E. C. Tjamos, G. C. Papavizas, and R. J. Cook, pp. 125-132, Plenum Press, New York.

Harman, G. E. 2000. Myths and dogmas of biocontrol changes in perceptions derived from research on *Trichoderma harzianum* T-22. *Plant Dis.* 84:377-393.

Harman, G. E., Howell, C. R., Viterbo, A., Chet, I. and Lorito, M. 2004. *Trichoderma* species-Opportunistic, avirulent plant symbionts. *Nature Rev.* 2:1-14.

Hjeljord, L. G., Stensvand, A. and Tronsmo, A. 2001. Antagonism of nutrient-activated conidia of *Trichoderma harzianum* (*atroviride*) P1 against *Botrytis cinerea*. *Phytopathology* 91:1172-1180.

Hjeljord, L. G. and Tronsmo, A. 2003. Effect of germination initiation on competitive capacity of *Trichoderma atroviride* P1 conidia. *Phytopathology* 93:1593-1598.

Honeycutt, E. W. and Banson, D. M. 2001. Formulation of binucleate *Rhizoctonia* spp. and biocontrol of *Rhizoctonia solani* on impatiens. *Plant Dis.* 85:1241-1248.

Hong, C. X., Michailides, T. J. and Holtz, B. A. 1998. Effects of wounding, inoculum density, and biological control agents on post-harvest brown rot of stone fruits. *Plant Dis.* 82:1210-1216.

Horst, L. E., Locke, J., Krause, C. R., McMahon, R. W., Madden,

L. V. and Hoitink, H. A. J. 2005. Suppression of Botrytis blight of begonia by *Trichoderma hamatum* 382 in peat and compost-amended potting mixes. *Plant Dis.* 89:1195-1200.

Kim, G. G. and Chung, Y. R. 2004. Colonization and degradation of senescent flowers of zucchini squash by *Trichoderma harzianum* YC459, a biocontrol agent of gray mold, *Botrytis cinerea*. *J. Zhejiang Univ.* 30:402 (Abstr.).

Lewis, J. A. and Papavizas, G. C. 1985. Effect of mycelial preparations of *Trichoderma* and *Gliocladium* on populations of Rhizoctonia solani and the incidence of damping-off. *Phytopathology* 75:812-817.

Lorito, M., Harman, G. E., Hayes, C. K., Broadway, R. M., Tronsmo, A., Woo, S. L. and Di Pietro, A. 1993. Chitinolytic enzymes produced by *Trichoderma harzianum*: Antifungal activity of purified endochitinase and chitobiosidase. *Phytopathology* 83:302-307.

Nelson, M. E. and Powelson, M. L. 1988. Biological control of gray mold of snap beans by *Trichoderma hamatum*. *Plant Dis.* 72:727-729.

Paulitz, T. C. and Belanger, R. R. 2001. Biological control in greenhouse systems. *Ann. Rev. Phytopahtol.* 39:103-133.

Redmond, J. C., Marois, J. J. and Macdonald, J. D. 1987. Biological control of *Botrytis cinerea* on roses with epiphytic microorganisms. *Plant Dis.* 71:799-802.

Singh, P. P., Shin, Y. C., Park, C. S. and Chung, Y. R. 1999. Biological control of *Fusarium* wilt of cucumber by chitinolytic bacteria. *Phytopathology* 89:92-99.

Sutton, J. C. and Peng, G. 1993. Biocontrol of *Botrytis cinerea* in strawberry leaves. *Phytopathology* 83:615-621.

Yedidia, I., Benhamou, N. and Chet, I. 1999. Induction of defense responses in cucumber plants (*Cucumis sativus* L.) by the biocontrol agent *Trichoderma harzianum*. *Appl. Environ. Microbiol.* 65:1061-1070.

Yedidia, I., Benhamou, N., Kapulnik, Y. and Chet, I. 2000. Induction and accumulation of PR proteins activity during early stages of root colonization by the mycoparasite *Trichoderma harzianum* strain T-203. *Plant Physiol. Biochem.* 38:863-873.