

## **Comparative efficacy of *Trichoderma viride*, *T. harzianum* and carbendazim against damping-off disease of cauliflower caused by *Rhizoctonia solani* Kuehn**

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### **ABSTRACT**

*Trichoderma viride* and *Trichoderma harzianum* were evaluated along with fungicide (carbendazim 50 WP) as a seed treatment and soil drench against damping-off disease in cauliflower caused by *Rhizoctonia solani*. Under *in-vitro* conditions the bio-control agents significantly inhibited the growth of *Rhizoctonia solani* and under field conditions they caused significant reduction in damping-off incidence, increased the seed germination and improved plant growth vigor as compared to carbendazim and control. The findings of the present study concluded that seed treatment with *Trichoderma harzianum* and *T. viride* along with farm yard manure offered better performance against damping-off disease and seedling growth of cauliflower. Seed treatment and seedbed treatment with *Trichoderma* spp., with combination farm yard manure might be suggested to control soil-borne diseases and plant growth vigor.

**Key words:** Cauliflower, carbendazim, damping off, *Rhizoctonia*, *Trichoderma* spp.

### **INTRODUCTION**

Many soil borne fungi play a major role in causing several diseases such as damping-off, root-rot, seed decay, collar rot, crown rot and wilt, etc. Cabbage, an important vegetable crop is attacked by several diseases, mostly caused by fungi and bacteria leading to severe crop losses. Among the fungal diseases, the damping off incited by *Rhizoctonia solani* Kuehn is a major constraint in the production of cabbage seedlings. *Rhizoctonia solani* is essentially soil-borne pathogen which inflicts heavy losses under favourable condition (Seema and Devaki, 2010). The management of this disease is difficult owing to long saprophytic survival ability of pathogen in soil (Dey, 2005). Reduction or elimination of soil borne inoculum is the only effective solution to overcome the problem and this may be achieved through use of effective fungal antagonists. Harman *et al.* (2004) reported biological and cultural control measures as two alternatives feasible options to synthetic pesticides in an integrated diseases management programme. Control of the plant diseases by chemicals can be spectacular but this is relatively a short term and

moreover, the accumulation of the harmful chemical residues sometimes causes serious ecological problem. In recent years, the increasing use of potentially hazardous pesticides and fungicides in agriculture has been the result of growing concern of both environmentalists and public health authorities. Biological methods, on the other hand can be economical, long lasting and free from residual side effects. The main purpose of the biological control of the plant disease is to suppress the inoculum load of the target pathogen to a level, which would not cause potential economic loss in a crop. Therefore, the present study has been carried out with following objectives, to isolate *Rhizoctonia solani* from infected seedlings of cauliflower and to study the compatibility of biocontrol agents (*T. viride* and *T. harzianum*) and a chemical (Carbendazim) against selected fungal pathogens under laboratory conditions. To evaluate *T. viride* and *T. harzianum* against damping-off disease of cauliflower.

## MATERIALS AND METHODS

*Rhizoctonia solani* was isolated from the infected cauliflower seedlings as per standard method and identified on the basis of cultural and morphological characters (Mehrota and Aggarwal, 2003). *Trichoderma viride* and *T. harzianum* were obtained from department of Microbiology and Microbial Technology, AAIDU, Allahabad. The efficacy of antagonists against *R. solani* was initially evaluated on potato dextrose agar (PDA) medium by dual culture technique (Dennis and Webster, 1971). Also, three concentrations of carbendazim 50 WP *viz.*, 10, 50 and 100 ppm were screened against the pathogen on PDA by poison food technique (Nene and Thapliyal, 1993). Five replications of each treatment along with control, maintained in completely randomized design, were incubated at 27°C. The radial growth of antagonist and pathogen was measured at 24 hrs intervals till sixth days.

The pathogen inocula were prepared in potato dextrose broth contained in 250 mL flasks and incubated at 27°C for ten days. Each mycelial growth on PDB scrapped and thoroughly shaken in 250 mL conical flasks containing 50 mL sterilized distill water on an electric shaker for 15 minutes. The mycelia were discarded and liquid sclerotia suspension collected separately. The suspension was centrifuged at 300 rpm for one minute. The pathogen inoculum (*Rhizoctonia solani*) with sclerotia load  $4 \times 10^6 \text{ mL}^{-1}$  was applied to nursery seedbeds @  $250 \text{ mL m}^{-2}$ ) four days before seed sowing of seeds as per the treatment (Shabir and Rubina, 2010; Chakraborty and Prasanta, 2001). The FYM was applied @  $12 \text{ t ha}^{-1}$  as per treatment a week before any other treatment. The cabbage cv. 'Bahar' seeds were sown in infested nursery beds. Before sowing, the seeds as per treatment were treated with biocontrol agents (*T. viride* and *T. harzianum*) or carbendazim (@  $2.5 \text{ gm kg}^{-1}$ . in case of soil drench, *T. harzianum* and *T. viride* was applied @  $2.5 \text{ kg ha}^{-1}$  (mixed in 500l water). Carbendazim 50 WP was applied (@ 0.25%). One untreated control was also maintained. The field experiment was laid in a randomized block design with each treatment replicated three times. The data with

respect to seed germination, damping-off incidence and plant growth vigor i.e. plant height, number of leaves and fresh weight were recorded at 45 DAS. The data was statistically analyzed with Randomized Block Design.

## RESULTS AND DISCUSSION

Under *in-vitro* conditions both the antagonists i.e. *Trichoderma harzianum* and *T. viride* inhibited the mycelial growth of *R. solani* ranged between 85.5 and 83.0 % as compared to control. Carbendazim 50 WP at 10, 50 and 100 ppm inhibited mycelial growth of pathogen 75, 78 and 81%, respectively significantly, as compared to control. The inhibition of *R. solani* by *Trichoderma* species could probably be due to the secretion of extracellular cell degrading enzymes such as chitinase B-1, 3- glucanase, cellulose and lectin , which help mycoparasites in the colonization of their host. The inhibition of pathogen may be also be attributed to the production of secondary metabolites by antagonists such as glioviridin, viridin and gliotoxin. (Shabir and Rubina, 2010; KamLesh and Gurjar, 2002; Muhammad and Amusa, 2003).

Both the species of *Trichoderma* under field condition improved seed germination and reduced the damping-off disease incidence as compared to untreated control. Overall 90.2 and 89.1% seed germination was recorded in soil drench with *T. viride* + FYM application treatment and seed treatment with *T. harzianum*, respectively. These are followed by seed treatment with *T. viride* and soil drench with *T. harzianum* + FYM application and seed germination, respectively, as compared to control. All treatments are effective in improving seed germination. Seed treatment with *T. viride* and *T. harzianum* had minimum damping-off incidence followed by seedbed treatment with *T. harzianum* + FYM as compared to control. Seedbed drenching with *T. harzianum* and *T. viride* + FYM reduced the damping-off incidence as compared to seedbed treatments with *T. harzianum* and *T. viride*, seedbed treatment with FYM and seed treatment with carbendazim

**Table 1.** Effect of *Trichoderma viride* and *T. harzianum* on growth of cauliflower and damping off disease incidence

Treatments	Seed germination 25 DAS	Damping-off incidence At 30DAS	Seedling height (cm) At 45 DAS	No. of leaves/seedling At 45 DAS	Fresh weight (g) At 45 DAS
Control	78.2	22.1	11.6	6.6	1.9
(FYM)	85.0	13.6	14.0	9.3	2.7
Seed + <i>Trichoderma harzianum</i>	89.1	8.0	16.4	11.6	4.8
Seed + <i>Trichoderma viride</i>	87.7	6.0	16.1	12.3	4.9
Seed + Carbendazim	86.2	13.6	14.2	9.6	3.3
Seedbed + <i>Trichoderma harzianum</i>	86.5	10.0	14.8	8.3	3.1
Seedbed + <i>Trichoderma viride</i>	82.8	11.0	14.7	8.6	3.5
Seedbed + both <i>Trichoderma</i> spp.	85.6	13.0	14.4	8.6	3.5
Seedbed + both <i>Trichoderma</i> spp. + FYM	86.8	11.0	15.8	9.6	3.8
Seedbed + <i>Trichoderma viride</i> + FYM	90.2	13.3	15.8	11.6	4.0
Seedbed + <i>Trichoderma harzianum</i> + FYM	87.1	13.6	15.9	11.3	4.2

Each value indicates mean value of three replications

and respectively. The result are in agreement with Islam and Faruq (2008), Manoranjitham *et al.* (1999) and Bunker and Mathur (2001); Roy *et al.* (1998); Faruk *et al.* (2002) and Champawat and Shama (2003) who employed biocontrol agents for the disease control and revealed the inhibitory effect was probably due to hyperparasitism/mycoparasitism, competition for space and nutritional source and antagonistic chemicals produced and released into the environment. *Trichoderma* sp., have been reported to produce antibiotic compounds (Trichodermin) extracellular enzymes (chitinase, cellulose) unsaturated monobasic acids (Dermadine) and peptides that either damage plant pathogen or enhance their population in biota.

All treatments significantly increased seedling height as compared to control. Seed treatments with *T. viride* and *T. harzianum*, seedbed drenching with treatments with *T. harzianum* and *T. viride* + FYM, followed by seedbed drenching with *T. harzianum*, *T. viride*, seedbed drenching with both BCA and seed treated with carbendazim showed significantly better seedling height as compared to control. The seed treated with *T. viride* and *T. harzianum*, seedbed treatments with BCA seed treatment with carbendazim, seedbed treatment with FYM and seedbed drenched with *T. harzianum* and *T. viride* significantly increases the number of leaves of cauliflower seedlings respectively as compared to control. The highest

seedling weight was recorded in seed treatment *T. viride* as compared to control. Seedbed treatment with FYM shows lowest fresh weight as compared to other treatment. Similar findings have been reported by Inbar *et al.* (1994); Manoranjitham *et al.* (2001); Champawat and Sharma (2003); Stephen *et al.*, (2003); Srivastava (2004); Sathar (2008); Shabir and Rubina (2010); M-uddin *et al.* (2011) and they suggested several possible mechanisms to explain this phenomenon including control of minor pathogens, production of plant hormones, production of vitamins, conversion of non utilizable materials into a form that can be utilized by the plant and increased uptake and translocation of minerals, increases the efficiency of nutrient uptake solubilizing certain insoluble nutrient elements like rock phosphate. This is actually one of the reason due to which growth rate of vegetables is increased in *Trichoderma* treated nursery beds.

It can be stated from the present study, that combination of bio control agent with Farm yard manure has significant effect on control of soil-borne pathogens. Simultaneously, it also enhanced the growth of vegetable with respect to control. The use of each combinations in the present study is easily producible, biodegradable, less expensive and cause no environment hazards to human health. These are ecologically safe and culturally more acceptable among the farmers.

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**REFERENCES**

- Bunker, R. N. and Mathur, K. 2001. Antagonism of local biocontrol agents to *Rhizoctonia solani* inciting dry root-rot of chilli. *Journal of Mycology Plant Pathology*, **31**(1): 50-53.
- Chakraborty, A. and Prasanta, K. S. G. 2001. Some biochemical changes in susceptible pigeonpea seedlings in response inoculation with non-pathogenic fusaria and its significance in induction of resistance against *Fusarium* wilt. *Journal of Mycology Plant Pathology*, **31**(1): 42-45.
- Cook, R. J. 1985. Biological control of plant pathogens: theory to application. *Pathopathology*, **12**: 75- 80.
- Champawat, R. S. and Sharma, R. S. 2003. Integrated management of nursery disease in brinjal, chilli, cabbage and onion, *Journal of Mycology Plant Pathology*, **33**(2): 290-291.
- Dennis, C. and Webster, J. 1971. Antagonist properties of species group of *Trichoderma*. III hyphal interaction. *Trans British Mycology Society*, **57**:363.
- Dey T. K. 2005. Effect of soil solarization in controlling damping-off disease of true potato seedlings. *Bangladesh Journal of Plant pathology*, **21**(1&2): 93.
- Faruk, M. I., Rahman, M. L. and Bari, M. A. 2002. Management of seedling disease of cabbage through *Trichoderma harzianum* amendment in seedbed. *Bangladesh Journal of Plant Pathology*, **18**(1-2): 49-53.
- Harman, G. E., Charles, R. H., Ada Viterbo, Chet, I. and Matteo, L. 2004. *Trichoderma* – opportunistic, avirulent plant symbionts. *Nature reviews Microbiology*, **2**: 43-56.
- Inbar, J., Abramsky, M., Cohen, D. and Chet I. 1994. Plant growth enhancement and disease control by *Trichoderma harzianum* in vegetable seedlings grown under commercial conditions. *European Journal of Plant Pathology*, **100**: 337-346.
- Islam, M. T. and Faruq, A. N 2008. Effect of selected soil amendments on seed germination, seedling growth and control of damping-off of chilli seedlings. *Journal of Sher-e-Bangla Agricultural University*. **2**(2): 12-16.
- Kamlesh, M. and Gujar, R. S. 2002. Evaluation of different fungal antagonistic, plant extracts and oil cakes against *Rhizoctonia solani* causing stem rot of chilli seedlings. *Annual Plant Protection Sciences*, **10**(2): 319-322.
- Manoranjitham, S. K., Prakassam, V. and Rajappan, K. 1999. Effect of antagonists on *Pythium aphanidermatum* (Edson) Fitz and the growth of chilli seedlings. *Journal of Biological Control*, **13**(1-2): 101-106.
- Manoranjitham, S. K., Prakasam, V. and Rajappan, K. 2001. Biological control of damping-off disease using talc based formulations of antagonists. *Annual Plant Protection Sciences*, **8**(2): 159-162.
- M-Uddin, M., Akther, N., Islam, M. T. and Farug, N. A. 2011. Effect of *Trichoderma harzianum* and some soil amendment against damping-off disease complex of potato and chilli. *The Agriculturists*, **9**(1&2): 106-116.
- Mehrotra, R. S. and Aggarwal, A. 2003. In: *Plant Pathology*. Tata McGraw-Hill Publishing Company limited. 493 P.
- Muhammad, S. and Amusa, N. A. 2003. *In-vitro* inhibition of growth of some seedling blight inducing pathogens by compost-inhabiting microbes. *African Journal of Biotechnology*, **2**(6): 161-164.
- Nene, Y. I. and Thapiyal, P. N. 1993. In: *Principles of pesticides*. Oxford and IBH Publishing. New Delhi. 3<sup>rd</sup> edition. 691 P.
- Roy, S. K., Das, B. C. and Bora, L. C. 1998. Non-pesticidal management of damping off of cabbage and cauliflower caused by *Rhizoctonia solani* Kuehn. *Journal of Agricultural Sciences Society*, **11**(2): 127-130.
- Shabir, R. and Rubina, L. 2010. Biological control of damping-off disease of cabbage caused by *Rhizoctonia solani* Kuehn. *Applied Biological Research*, **12**: 38-41.
- Sathar, S. 2008. Bioconversion of post harvest crop residues and cattle shed manure into

- value added products using earthworms *Eudrilus eugeniae* (Kinberg). *Ecological Engineering*, **32**: 206-214.
- Seema, M. and Devaki, N. S. 2010. Effect of some essential oils on *Rhizoctonia solani* Kuhn infecting flue-cured Virginia tobacco. *Journal of Biopesticides*, **3**(3): 563-566.
- Srivastava, V. K. 2004. *Trichoderma* spp. - a boon for better crop health. *Pestology*, **28**(8): 40-45.
- Stephan, Z. A., Jbara, I. M. and Al-Rawi F. A. 2003 Effect of soil moisture content and thermal treatment on the activity of bioagent fungi *Trichoderma harzianum* Rifai and *Paecilomyces lilacinus* (thom) Samson on some tomato plant growth parameters. *Arabian Journal of Plant Protection*, **21**(1): 1-5.

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