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Efficacy of bioagents against *Macrophomina phaseolina* causing root rot of soybean *in vitro*

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Abstract
Efficacy of seven bioagents antagonists *viz*., *Trichoderma viride*, *Trichoderma harzianum*, *Trichoderma asperellum*, *Trichoderma longibacter*, *Trichoderma koningii*, *Pseudomonas fluorescens* and *Bacillus subtilis* were evaluated for its antagonistic effect against *M.phaseolinain vitro* by dual culture technique. Among seven different antagonists tested against *M. phaseolina in vitro*, *Trichoderma asperellum* showed significantly maximum growth inhibition (66.66%) with lowest mycelial diameter (30.00 mm) of *M. phaseolina* which was at par with *T. harzianum* (66.29%) and *T. viride* (65.00%) *T. longibacter* (63.14%) and *T. koningii* (62.96%) with mycelial diameter of 30.33 mm, 31.50 mm, 33.16 mm and 33.33 mm, respectively (at $P = 0.01$).

Keywords: bioagents, *Macrophomina phaseolina*, soybean

Introduction
Macrophomina phaseolina is a necrotrphic phytopathogen with a wide host range including more than 500 cultivated and wild plant species belonging to more than 75 families (Khan, 2007; Salik, 2007). Sinclair and Shurlleffe (1975) [5, 9, 10] reported 100 pathogens known to affect soybean, out of which 35 are of economic importance. All parts of the soybean plant are susceptible to number of pathogens which reduces the quality and quantity of seed yield. 30-50 per cent yield losses due to *Macrophomina phaseolina* in soybean crop has been reported by Yang and Navi (2005) [13]. Losses of plant population up to 77 per cent has been reported by rhizoctonia bataticola (Muthusamy and Mariappan, 1991) [7]. Disease is more severe in the regions where climate is relatively dry and warm during growing season (Singh and Mehrotra, 1982) [11]. The disease can be managed to some extent by cultural, chemical and biological methods (Bristow and Wyllie, 1975; Gupta, 2004) [2, 4]. Biological control is becoming an important component of integrated plant disease management of root rot. There are several examples of fungi that are able to manage the plant pathogens, of these, *Trichoderma* spp. has provided one of the first biological control to manage root rot have been studied to the greater extent (Papavizas, 1985) [8]. It exerts biocontrol activity against fungal phytopathogens either indirectly by competing for nutients and space, which modify mechanisms and antibiosis, or directly by mechanisms such as mycoparasitism (Benitez *et al*., 2004) [1]. Considering the above facts, the present investigation was carried out.

Materials and Methods
Effect of different antagonists listed in Table 1 was studied by dual culture technique against *M. phaseolina* with following experimental details

a) Design: Completely Randomized Design (CRD)
b) Treatments: 8
c) Repetitions: 3
d) Method: Dual culture method (Dennis and Webster, 1971)

The antagonistic potential of each fungal antagonist was studied by dual culture method (Dennis and Webster, 1971). A 5 mm diameter disc of antagonist was placed individually at

one end of the Petri plate containing 20 ml PDA media and just opposite to that a 5 mm diameter disc of the pathogen (*M. phaseolina* Anand isolate MP4) was placed. For bacterial isolates *in vitro* antagonism was performed following the method of Toure *et al*. (2004). Two days old culture of bacterial isolates were streaked as a streak line in PDA plates and 5 mm mycelia disc of an actively growing culture of the pathogen was introduced opposite to the other edge of the Petri plate. Three repetitions were maintained for each antagonist. In control, the pathogen alone was inoculated at center. The cultures of bioagents were obtained from Department of Plant Pathology and Department of Agricultural Microbiology, BACA, AAU, Anand – 388 110. Department of Plant Pathology, NMCA, NAU, Navsari – 396 450, Department of Plant Pathology, Chaudhary Sarwan Kumar Himachal Pradesh Krishi Vishvavidhayalaya, Palampur, Himachal Pradesh – 176 062.

Table 1: List of bioagents tested against *M. phaseolina* by dual culture technique

Tr. No.	Antagonists
T_1	*Trichoderma viride*
T_2	*Trichoderma harzianum*
T_3	*Trichoderma asperellum*
T_4	*Trichoderma longibacter*
T_5	*Trichoderma koningii*
T_6	*Pseudomonas fluorescens*
T_7	*Bacillus subtilis*
T_8	Control (Test pathogen only)

Observations recorded

Observations on the radial growth (mm) was recorded from 24 h of incubation at 28±2°C till the complete growth of test pathogen in control plates. Per cent growth inhibition (PGI) over control was calculated by using following formula given by Vincent (1947).

$$PGI = \frac{C-T}{C} \times 100$$

Where,

PGI = Percent growth inhibition

C = Mean diameter of mycelial colony in control treatment (mm)

T = Mean diameter of mycelial colony in treated set (mm).

Results

The result pertaining to this study is presented in Table 2. The data revealed that all the antagonists tested against *M.phaseolina* had a significant antagonistic effect on the mycelial growth of *M.phaseolina* in dual cultures (P = 0.05 and P = 0.01), when compared to controls and the per cent growth inhibition ranged from 66.66% to 57.59%.

Among all the seven antagonists, *Trichoderma asperellum* showed significantly maximum growth inhibition (66.66%) with lowest mycelial diameter (30.00 mm) of *M.phaseolina* which was at par with *Trichoderma harzianum* (66.29%) and *Trichoderma viride* (65.00%) with mycelial diameter of 30.33 mm and 31.50 mm, respectively.

The next better antagonists were *Trichoderma longibacter* (63.14%), *Trichoderma koningii* (62.96%) and *Pseudomonas fluorescens* (62.22%) with mycelial diameter of33.16 mm, 33.33 mm and 34.00 mm, respectively and all the three were at par. Least inhibition was recorded by *Bacillus subtilis* with 57.59% and mycelial diameter of 38.16 mm.

The similar results were observed at CD (P = 0.01) significance level and along with *T. harzianum* and *T. viride*, *T. longibacter* and *T. koningii* were also found at par with *T. asperellum* thus observed difference was statistically highly significant.

Table 2: *In-vitro* antagonism of different bio-agents against *M. phaseolina*

Tr. No.	Bio-agents	*M. phaseolina*	
		Mycelial growth (mm)	Growth Inhibition (%)
T_1	*Trichoderma viride*	31.50	65.00
T_2	*Trichoderma harzianum*	30.33	66.29
T_3	*Trichoderma asperellum*	30.00	66.66
T_4	*Trichoderma longibacter*	33.16	63.14
T_5	*Trichoderma koningii*	33.33	62.96
T_6	*Pseudomonas fluorescens*	34.00	62.22
T_7	*Bacillus subtilis*	38.16	57.59
T_8	Control (Test pathogen only)	90.00	0.00
S. Em. ±		0.86	-
C.D. at 5 %		2.60	-
C.D. at 1 %		3.57	-
C.V. %		3.75	-

Discussion

The present study is in harmony with earlier workers. Doley and Jite (2012) [3] found that *T. viride* showed significant antifungal activity by inhibiting the mycelial radial growth of *M. phaseolina* by 71.42%. Kumar *et al.* (2013) [6] observed that maximum inhibition of the mycelial growth of *M. phaseolina* isolates was done by *T. harzianum* that varied from 61.1 to 70.1%. Ashwini *et al*. (2014) reported that *Pseudomonas fluorescens* showed best antagonistic activity against *M. phaseolina* (62.41%).

This antagonistic nature might be due to antibiosis, nutrient competition, and/or cell wall degrading enzymes (Kumar, 2013) [6]. A large variety of volatile secondary metabolites could be produced by *Trichoderma* spp. such as ethylene, hydrogen cyanide, aldehydes, and ketones, which play an important role in controlling various plant pathogens (Vey *et al*., 2001) [12].

References

1. Benitez T, Rincon AM, Limon MC, Condon AC. Biocontrol mechanisms of *Trichoderma* strains. International Microbiology. 2004; 7(4):249-260.
2. Bristow PR, Wyllie TD. The effect of crop rotation and date of planting on charcoal rot of soybean (Abstract). Proceedings of the American Phyto pathological Society. 1975; 2:83.
3. Doley K, Jite PK. *In vitro* efficacy of *Trichoderma viride* against *Sclerotium rolfsii* and *Macrophomina phaseolina*. Notulae Scientia Biologicae. 2012; 4(4):39-44.
4. Gupta GK. Soybean diseases and their management. In: Singh, N. B., Chauhan, G. S., Vyas, A. K. & Joshi, O. P. (eds) Soybean Production and Improvement in India. Indore, India, NRC for Soybean, 2004, 145-168.
5. Khan SN. *Macrophomina phaseolina* as causal agent for charcoal rot of sunflower. Mycopathologia. 2007; 5:111-118.
6. Kumar M, Gaur VK, Kant K, Sharma A. Host range and pathogenic variation in isolates of *Macrophomina phaseolina* incident of dry root rot in Moth bean. Annals of Plant Protection Sciences. 2013; 21(2):333-336.
7. Muthusamy S, Mariappan V. Disintegration of sclerotia of *Macrophomina Phaseolina (soybean isolate)* by oil cake extracts. Indian Phytopathology. 1991; 44:147-150.

8. Papavizas GC. *Trichoderma* & *Gliocladium* biology, ecology and potential for bio control. Annual Review of Phytopathology. 1985; 23:23-54.
9. Salik NK. *Macrophomina phaseolina* as causal agent for charcoal rot of sunflower. Mycopathologia. 2007; 5(2):111-118.
10. Sinclair JB, Shurlleff MC. Compendium of soybean diseases. American Phytopathological Society, Inc Minnesota, USA, 1975, 5p.
11. Singh P, Mehrotra RS. Influence of soil and temperature on *Rhizoctonia bataticola* infection of gram. Indian Phytopathology. 1982; 35:327-329.
12. Vey A, Hoagland RE, Butt TM. Toxic metabolites of fungal biocontrol agents, p: 311-346. In: "*Fungi as biocontrol agents*: Progress, Problems and Potential" (T. M. Butt, C. Jackson, N. Magan, eds.). CAB International, Bristol, UK, 2001, 390p.
13. Yang XB, Navi SS. First report of charcoal rot epidemics caused by *Macrophomina phaseolina* in soybean, Iowa. Plant Disease. 2005; 89(5):62.