



ANTAGONISTIC EVALUATION OF SOME BIO AGENTS AND THEIR EFFECT AS SEED TREATMENT ON GERMINATION GROWTH PARAMETERS AND WILT INCIDENCE IN LENTIL

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ABSTRACT

Antagonistic activity of four bioagents viz. *Trichoderma viride*, *T. harzianum*, *Pseudomonas fluorescens* and *Gliocladium virens* against *Fusarium oxysporum* f. sp. *lentis* inciting wilt of lentil was found that *T. viride* was found most effective than others, representative maximum 72.85 per cent inhibition on mycelia growth of test pathogen. Effect of seed treatment with bioagents have also revealed that the increased seed germination by 54.28 to 72.85 per cent and decreased wilt incidence from 11.0 to 1.12 per cent. Among the bioagents, *T. viride* was found more effective giving maximum seed germination with 85.0 per cent, increased root and shoot length 9.5 and 4.5 cm and minimizing wilt incidence by 1.12 per cent which was followed by *T. harzianum* in paper towel method. Similar observation has also recorded in case of glass house experiment.

Key Words : bio agents, lentil wilt and germination.

Lentil (*Lens esculenta* Medik.) is grown as major rabi pulses crop in India with annual production of 994.2 million tones from 1476 million hectare of land (Anonymous, 2006). The major lentil growing states in India is Uttar Pradesh, Madhya Pradesh, Bihar, West Bengal, and Rajasthan. *Fusarium* wilt or true wilt caused by *Fusarium oxysporum* f. sp. *lentis* is economically most devastating disease causing yield loss up to 50 per cent in India and 12 per cent North West Syria. The disease has been reported from 17 countries of the world. In India, wilt of lentil is serious problems in Uttar Pradesh (especially Bundelkhand region), Madhya Pradesh, Bihar, West Bengal, Rajasthan. The pathogen being seed and soil borne in nature and is

very difficult to control. However chemicals are somehow better than other methods for managing of disease but continuous use of chemical is being discouraged now a days for the reason that the fungicides are not ecofriendly for being hazard our to mammalian group and responsible for creating the environmental pollution in air, soil and water. Therefore, there is a search of an ecofriendly method for management of disease.

The different strains of bioagent when incorporate in the soil and applied to seed were reported to more effective against diseases of many crop (Chaudhary and Prajapati, 2004, Singh and Mukhopadhyay, 2000 and Singh et al., 2004). Use of bioagent is reported to be

quite effective; in expensive and ecofriendly keeping in view the present investigation was under taken to evaluate different bioagent in different way like seed treatment, laboratory, bioassay and paper towel method and in pot culture under glass house condition.

MATERIALS AND METHODS

Antagonistic evaluation of bioagents against *Fusarium oxysporum* f. sp. *lentis* (in-vitro) :

Four bioagent viz. *T. viride*, *T. harzianum*, *Pseudomonas fluorescens* and *Gliocladium virens* were assessed for their efficacy against *Fusarium oxysporum* f. sp. *lentis* by using dual culture technique . The culture of test fungus and antagonists was multiplied on Potato Dextrose Agar medium (PDA). Five mm disc of test fungus and the antagonist cut from the edge of seven days old culture plate and were placed at opposite direction to each other at a distance of 5 mm from the periphery of the Petri-plates. A disc of test fungus was placed alone only one side on P.D.A. plates serve as control. Three replications were kept for each treatment. Then all the petriplates were incubated at 25 ± 1°C. The data was recorded after seven days of inoculation when the inhibition zone was formed and calculates per cent inhibition by using following formula :

$$\text{Per cent inhibition} = \frac{\text{Colony diameter (mm) in check} - \text{colony diameter (mm) in treatment}}{\text{Colony diameter (mm) in check}} \times 100$$

Effect of seed treatments with bioagent by paper towel method :

The experiment was conducted by paper towel method by using lentil variety Sehare-74-3 to find out the effect of seed treatment with bioagents on seed germination, root length, shoot length, vigour index and wilt incidence in lentil. Seeds were treated with spore and mycelial suspension of bioagents collected from seven days old culture liquid broth which was mixed with 1 per cent carboxyl methyl cellulose for proper coating of spore on the seed surface. Four replications each of 100 seeds were taken for each treatment with suitable control. The data were recorded on seed

germination after 7 days of sowing .The root length, shoot length and wilt incidence were taken 21 days after sowing (DAS). The seedling vigour index of all treatment was also calculated by the following formula :

$$\text{Vigour index} = \text{Seed germination} \div \text{x} [\text{Root length (cm)}] \times [\text{Shoot length (cm)}]$$

Effect of by seed treatment with bioagents in pots under glass house condition :

The seeds of variety Sehare-74-3 were treated with bioagents viz. *T. viride*, *T. harzianum*, *P. fluorescens* and *G. virens* (3-4 g/kg seed). The treated 25 seed were sown in pots which were previously filled by soil containing *F. oxysporum*. f. sp. *lentis* @ 2 per cent by weight per plot. The experiment was conducted in C.R.D. with four replications under glass house condition. The observation on seed germination, seedling emergence and wilt incidence was recorded. The experiment was carried out for two consecutive years 2005-06 and 2006-07.

RESULTS AND DISCUSSION

The Results obtained from the present investigation has been discussed below :

In vitro evaluation of bioagents against pathogens by dual culture method:

The results are interpreted in the Table 1 showed that all the bio agents are able to inhibit the mycelial growth of *F. oxysporum*. f. sp. *lentis* ranging from 54.28 to 72.85 per cent . However, the maximum colony growth inhibition of *F. oxysporum*. f. sp. *lentis* was recorded by *T. viride* with 72.85 per cent, followed by 70 per cent 66.71 per cent and 54.28 per cent in *T. harzianum*, *P. fluorescens* and *G. virens*, respectively. Chaudhary and Prajapati (2004), also reported that six bioagents collected from different places of India were tested against *Fusarium udum* and found that among the isolates *T. harzianum* (Pantnagar), *T. viride* (Kanpur), *Penicillium citrinum* (Lucknow) were found more effective than *G. virens* (Pantnagar) and *T. viride* (Coimbatore) isolates.

Table 1: Antagonistic effect of bioagents against *Fusarium oxysporum* f. sp. *lentis* in vitro

Sr. No.	Treatments	Radial growth (mm)	Per cent inhibition
1.	<i>Trichoderma viride</i>	19	72.85
2.	<i>Trichoderma harzianum</i>	21	70.00
3.	<i>Pseudomonas fluorescens</i>	24	65.71
4.	<i>Gliocladium virens</i>	32	54.28
5.	Control	70	–
	S.E. ±	1.17	–
	C.D. (P=0.05)	3.25	–

Table 2: Effect of seed treatment with bioagents on seed germination, root length, shoot length, vigour index and wilt incidence by paper towel method

Sr. No.	Bio-agents	Dosages g/kg	Seed germination (%)	Root length (cm)	Shoot length (cm)	Vigour index	Wilt incidence (%)
1.	<i>Trichoderma viride</i>	6	85* (67.30)	9.5	4.5	1190.0	1.12 (6.0)
2.	<i>Trichoderma harzianum</i>	6	83 (65.76)	9.1	4.4	1120.5	1.14 (6.05)
3.	<i>Pseudomonas fluorescens</i>	6	78 (62.09)	8.7	3.8	975.0	2.75 (9.50)
4.	<i>Gliocladium virens</i>	6	72 (58.13)	8.2	3.4	835.2	4.02 (11.52)
5.	Control	-	7 (56.84)	7.0	3.0	700.0	11.00 (11.30)
	S.E. ±	-	1.68	0.27	0.22	18.12	0.68
	C.D. (P=0.05)	-	4.66	0.76	0.60	50.19	1.91

* Figure in parentheses indicate transformed values

Table 3 : Evaluation of seed treatment with bioagents on seed germination seedling emergence and wilt incidence in lentil under glass house condition

Sr. No.	Bioagents	Doses g/ka seed	Seed germination (%)			Seedling emergence (%)			Wilt incidence (%)		
			2005-06	2006-07	Average	2005-06	2006-07	Average	2005-06	2006-07	Average
1.	<i>Trichoderma viride</i>	6	90 (71.85)*	85 (67.35)		85 (67.35)	80 (63.52)		10 (18.25)	15 (22.57)	
2.	<i>Trichoderma harzianum</i>	6	90 (71.66)	85 (67.35)		85 (67.35)	80 (63.56)		15 (22.76)	15 (22.78)	
3.	<i>Pseudomonas fluorescens</i>	6	83 (67.21)	80 (63.43)		80 (63.43)	75 (60.00)		25 (30.00)	20 (26.56)	
4.	<i>Gliocladium virens</i>	6	75 (60.00)	75 (60.00)		70 (56.78)	70 (56.78)		20 (26.22)	25 (30.00)	
5.	Control	-	65 (53.75)	60 (50.77)		60 (50.70)	60 (50.78)		40 (39.22)	45 (42.03)	
	SEm ±	-	1.28	1.23			1.35		0.91	0.99	
	C.D. (P=0.05)	-	3.56	3.41			3.66		2.51	2.76	

Effect of seed treatments with bioagents by paper towel method:

The experiment was conducted *in vitro* condition to determine the effect of seed treatment with bioagent on seed germination, root length, shoot length, vigour index and wilt incidence in lentil. It is revealed that bioagents significantly reduced wilt incidence and improved the seed germination, root length, shoot length and vigour of the seedlings (Table 2). Among the treatment *T. viride* was found most effective in giving maximum seed germination with 85 per cent, increased root length (9.5 cm), shoot length (4.5 cm) and minimizing wilt incidence (1.22 %), followed by *T. harzianum* representing 83 per cent seed germination, 9.1 cm root length, 44 cm shoot length and 1.1 per cent wilt incidence. Papavizar (1985), also reported that *T. harzianum*, *T. viride* and *G. virens* inhibited the pathogen by competition, mycoparasitism and antibiosis.

Evaluation of seed treatment with bioagents in pots under glass house condition :

For further confirmation the experiment was conducted in pots under glass house condition. The result in the table 3 showed that *Trichoderma viride* and *T. harzianum* resulted in better seed germination seedling emergence and minimizing wilt incidence which was followed by *P. fluorescens* and *G. virens*. The different strains of bioagent when incorporate in the soil and applied to seed were reported to more effective against diseases of many crop (Kumawat, et al. 2010, Chaudhary and Prajapati, 2004, Singh and Mukhopadhyay, 2000 and Singh et al., 2004). Singh *et al.* (2004) also reported that antagonist *viz.* *T. viride*, *T. harzianum*, *G. virens* and *Aspergillus nidulans* as

seed, soil and combined seed and soil treatments for the control of tomato wilt in green house. According to them application of the *T. harzianum*, *T. viride* and *Gliocladium virens* as seed treatment 10 g/kg seed were effective in controlling seedling mortality upto 85 per cent and were at par with carbendazim. They also found that soil treatment with antagonist was less effective than seed treatment.

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