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RESEARCH ARTICLE

Management of root rot disease in French beans (*Phaseolus vulgaris*) by using microbial consortium

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SUMMARY

Root rot disease of beans is caused by *Rhizoctonia solani*. Biocontrol agents were tested for their antagonistic activity against root rot pathogen *Rhizoctonia solani.* Among the tested isolates, *Trichoderma viride* recorded the maximum (86.04 %) inhibition on the mycelial growth of pathogen followed by *Pseudomonas fluorescence* which recorded 79.12 per cent inhibition on the mycelial growth. Treatment with (SA+ST with *P. fluorescens* + *T. viride*) + *Neem* cake was recorded maximum (78.55%) disease reduction in the field condition with the yield of 2654 kg/acre. Combined application of biocontrol agents having ability to reduce root rot disease in French bean and increase the yield siginificantly.

Key Words : Beans root rot, *Rhizoctonia solani*, *Trichoderma* sp., *Pseudomonas* sp., Biological control

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French beans (*Phaseolus vulgaris*) are suitable for cultivation in the high, middle and lower elevations of the Nilgiris and other similar hilly tracts. It is majorly grown in the temperate and tropical regions of India and shows higher productivity around temperature of 21°C. It is a very rich source of proteins, vitamins, β – carotene, neoxanthin, lutein, violaxanthin, Zea-Xanthin and also contains antioxidant capacity. Root rot is caused by *Rhizoctonia solani* which is an important fungal disease of French bean causing considerable damage to the crop. *Rhizoctonia solani* is a necrotropic soil borne pathogen with high competitive saprophytic activity. Soil-borne diseases can be devastating in beans production areas, especially if they are grown repeatedly in the same place. Cultural practices serve an important role in prevention and management of plant diseases.

Management of the diseases through chemicals and the use of resistant varieties are possible to some extent. But the hazardous impact of agrochemicals on the environment, development of resistant mutants, increasing cost of pesticides and frequent breakdown of resistant varieties strongly demand a sustainable and an alternative management approach for the disease. However due to residual effect of synthetic fungicides, there is demand for more eco friendly substances like bio pesticides. Such exploration of natural resources for

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their antifungal potential against the pathogen is quite inevitable for a sustainable and eco friendly management of the pathogen.

Biocontrol is an important component of integrated disease management (IDM) that provides disease control while being relatively harmless to humans, non-polluting and bio-degradable, selective in mode of action, difficult for pathogens to develop resistance, unlikely to harm other beneficial micro-organisms and generally improves soil health and sustainability of agriculture (Sheo Raj *et al.*, 2004). Palmieri *et al.* (2017) revealed that combined application of microbial biocontrol agents highly suppress the disease compared with the individual isolates.

Beneficial effects of PGPR that have been documented by many scientists, which include direct plant growth promotion, biological disease management and inducing host resistance. Knowledge on the molecular mechanisms of the PGPR enables the scientists, to bring the effective technique for managing the diseases. The introduction of antimicrobial compounds *viz*., phenazines and phloroglucinals do contribute significantly in the biological control of diseases (Kloepper and Beauchamp, 1992).

Biological control of plant diseases with antagonistic micro-organisms is now considered as a promising alternative to the use of fungicides. To manage certain crop diseases no single approach has been proved to be satisfactory. Hence, it is necessary to integrate microbial biocontrol agents for effective management of diseases. In the present investigation, the combined application of biocontrol agents were tested against root rot disease of beans caused by *Rhizoctonia solani*.

MATERIAL AND METHODS

Isolation of pathogen:

The pathogen was isolated from the diseased tissues of beans root by tissue segment method (Rangaswami, 1958). The infected portions of diseased plants were cut into small pieces using sterilized scalpel and these were surface sterilized with 0.1 per cent mercuric chloride for one minute and washed in three changes of sterile distilled water and then placed on previously poured and solidified Petri dish containing potato dextrose agar (PDA) medium. These plates were incubated at room temperature (28 ± 2^{o}C) for five days and observed for the growth of the fungus. The hyphal tips of fungi grown from the pieces were transferred aseptically to PDA slants for maintenance of the culture.

Screening of the antagonists against *Rhizoctonia solani* under *in vitro* condition:

Trichoderma viride and *Pseudomonas fluorescens* were screened against *Rhizoctonia solani* by dual culture method (Dennis and Webster, 1971). A nine mm mycelial disc of *Rhizoctonia solani* and *Trichoderma viride* were placed opposite to each other near the periphery of the Petri plate and incubated at room temperature (28±2^{0}C). After four days of incubation, mycelial growth of the pathogen and inhibition zone was measured as well as in control plates. Per cent inhibition (PI) of mycelial growth was calculated using the formula suggested by Pandey *et al.* (2000).

Efficacy of biocontrol agents against root rot of beans in pot culture:

To study the biocontrol potential of *Trichoderma viride* and *Pseudomonas fluorescens* pot culture experiment was conducted in glass house. The talc based formulation of *Trichoderma viride* and *Pseudomonas fluorescens* were delivered as per the treatment. The pathogen was mass multiplied on sand maize medium and incorporated in the pots at 5 per cent (w/w) under glass house condition. The disease incidence was recorded at frequent intervals.

Efficacy of biocontrol agents against root rot of beans in the field condition:

Field experiment with the following treatments was laid out at Nanjanad, Nilgiris district during 2016-2017 where the crop is grown every year. Randomized Block Design (RBD) was used in the experiments with plot size 5 x 4 m^{2}. The experiment was conducted with seven treatments and each treatment replicated thrice. Individual application of bioagents, organic amendments and its different combinations were applied as soil application. All normal agronomical practices were followed at regular intervals. Disease incidence and yield was recorded.

RESULTS AND DISCUSSION

Biocontrol agents were tested for their antagonistic activity against root rot pathogen *Rhizoctonia solani.* Among the tested isolates, *T.viride* recorded the maximum (86.04 %) inhibition on the mycelial growth of pathogen followed by *Pseudomonas* *fluorescens* which recorded 79.12 per cent inhibition on the mycelial growth under *invitro* condition (Table 1). *T. harzianum* and *T. viride* (Pers.) were antagonistic to *F.oxysporum* in onion under *in vitro* and *in vivo* conditions (Rod, 1984). Fungal antagonists *T. viride*, *T. harzianum*, *T. hamatum*, *T. koningii*, *T. pseudokoningii* were effective against *Fusarium oxysporum* f.sp.*cepae* infecting onion under *in vitro* conditions (Rajendran and Ranganathan, 1996).

A pot culture experiment was conducted to evaluate the biocontrol agents against root rot disease of beans. In this experiment, seven treatments were tested, among the treatments T_3 (ST+ SA with Pf + Tv + *Neem* cake) treatment was significantly recorded maximum disease reduction (75.82 %) (Table 2).

In the present study, biocontrol agents were tested against root rot disease of beans in field condition. In this experiment, seven treatments were tested, among the treatments T_3 (ST+ SA with Pf + Tv+ *Neem* cake) treatment was recorded maximum (78.55) per cent disease reduction with yield of 2654 kg/acre followed by T_2 and T_1 treatments which accounted 74.39 and 73.37 per cent reduction of the disease (Table 3). The chemical check carbendazim was recorded 79.71 per cent disease reduction with higher yield 2712 kg/acre.

Pseudomonas have been found to produce broad spectrum antibiotics *viz*., Phenazine, Pyrrolnitrin, Pyoverdine, 2,4-diacetylphloroglucinol (Gardener *et al*., 2000), lytic enzymes such as chitinases and b-1,3-glucanases which degrade fungal chitin (Velazhahan *et*

Table 1: Screening of biocontrol agents against mycelial growth of *Rhizoctonia solani* under *in vitro* condition

Sr. No.	Treatments	Mycelial growth (cm)*	Per cent reduction over control
1.	*Trichoderma viride*	1.25	86.04
2.	*Pseudomonas fluorescens*	1.87	79.12
3.	*Bacillus subtilis*	2.52	71.87
4.	Control	8.96	-
	C.D. (P=0.05)	0.45	-

*Mean of three replications

Table 2: Efficacy of biocontrol agents against root rot disease of beans in pot culture

Sr. No.	Treatments	PDI*	Perc ent reduction over control
1.	T_1- ST+ SA with *P. flourescens* (Pf)	18.78	70.89
2.	T_2- ST+ SA with *T.viride* (Tv)	18.46	71.35
3.	T_3 – (ST+ SA with Pf + Tv) + *Neem* cake	15.60	75.82
4.	T_4- *Panchaghavya* 3%	42.35	34.36
5.	T_5- Carbendazim (0.1%)	15.28	76.31
6.	T_6- *Copper oxychloride* (0.1%)	25.44	60.57
7.	T_7- Untreated control	64.52	-
	C.D. (P=0.05)	2.74	-

*Mean of three replications, ST- Seed treatment, SA- Soil application

Table 3 : Efficacy of biocontrol agents against root rot disease of beans in field condition

Sr. No.	Treatments	PDI*	Per cent reduction over control	Yield kg/acre
1.	T_1- ST+ SA with *P. fluorescens*	13.97	73.37	2421
2.	T_2- ST+ SA with *T.viride*	13.43	74.39	2449
3.	T_3 - (ST+ SA with Pf + Tv) + *Neem* cake	11.25	78.55	2654
4.	T_4- *Panchaghavya* 3%	40.35	23.08	1253
5.	T_5- Carbendazim (0.1%)	10.64	79.71	2712
6.	T_6- *Copper oxychloride* (0.1%)	20.52	60.88	1745
7.	T_7- Untreated control	52.46	-	953
	C.D. (P=0.05)	1.47	-	-

*Mean of three replications, Seed treatment (ST)- 4g/kg of seed (*T.viride*), 10g/kg of seed (*P. fluorescens*)
Soil Application (SA) - @ 2.5 kg/ha (*T.viride* / *P. fluorescens) Neem* cake- @ 150 kg/ha

al., 1999), production of siderophore (Loper, 1988), production of HCN (Ahl *et al*., 1986) and induced systemic resistance (Van Peer *et al*., 1991).

Marimuthu *et al*. (2002) reported that there is synergistic effect of combined application of *Azospirillum* and *Pseudomonas fluorescens* Pf 1 on reduction of root rot incidence and enhanced plant growth and yield of cotton under field conditions. The yield increase under field conditions might be due to the growth-promoting compounds such as gibberellin, cytokinins, auxin from tryptophan which produced by different biocontrol agents which increased the growth rate and yield of groundnut under field condition (Pal *et al*., 2000).

Soil application of *P. fluorescens* increased the rhizosphere population of *P. fluorescens*. Fluorescent Pseudomonads, which constitute 20 per cent of the total bacterial population, are expected to give better protection against soil-borne pathogens. They had high affinity for amino acid exudates and probably this might have contributed to their high rhizosphere competence (Bakker and Chet, 1982).

One of the emerging strategies for managing plant diseases is the use of microbial biocontrol agents, with the aim of reducing pesticide usage, providing non-polluted produce and eventually to safeguard human health and environment. The present study strengthen that well-defined goal, giving abundant evidence to prove that field application of bioformulations in beans to manage economically important diseases. In conclusion, the biocontrol agents effectively reduce the root rot incidence and enhanced the plant growth parameters and there by increased yield in beans.

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