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Evaluation of commercially available talc formulations of bioagents for management of sheath blight of rice and their effect on plant growth enhancement

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ABSTRACT

The effectiveness of commercially available talc formulations of Trichoderma harzianum and Pseudomonas fluorescens applications either alone or in combination on rice crop growth, sheath blight disease and grain yield was investigated in field experiments. Seed and soil application of T. harzianum and P. fluorescens resulted in significant reduction of sheath blight disease incidence caused by Rhizoctonia solani and was comparable to the treatment with a systemic fungicide carbendazim (seed and foliar application). Lowest sheath blight severity was recorded in the treatment comprising combined application of T. harzianum and P. fluorescens (9.50%) whereas, 10.6 per cent sheath blight severity was recorded in plot receiving seed and foliar application of carbendazim. Plots having seed and soil application of T. harzianum and P. fluorescens alone had 11.5 per cent and 11.8 per cent disease severity, respectively. Observations recorded at the time of disease incidence revealed that combined application of T. harzianum and P. fluorescens delayed disease incidence by 35 days which was maximum among all the treatments. Seed and foliar application of carbendazim also delayed disease incidence by 27 days which was significantly lower than combined application of T. harzianum and P. fluorescens. Observations on plant growth parameters revealed that plant growth was maximum in plots having combined application of T. harzianum and P. fluorescens, in this treatment higher plant height (112.0 cm), shoot dry weight (32.0 g), root dry weight (7.9 g) were recorded. No significant increase in plant growth was recorded in plots receiving seed and foliar application of carbendazim. Studies on different treatments on yield attributes also established combined application of T. harzianum and P. fluorescens as the most effective treatment and resulted in 17.56 per cent increase in plant yield followed by T. harzianum (16.75%) and P. fluorescens (14.86%) alone, only 9.45 per cent increase in plant yield was recorded in plots having seed and foliar application of carbendazim.

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INTRODUCTION

Rice is an important staple food crop for majority of the world. Many biotic stresses hamper rice production and specifically fungal diseases cause huge economic losses. Among different fungal diseases of rice, sheath blight (ShB) is an important one, responsible for losses in grain yield. Annual yield losses upto 40 per cent were reported with ShB under optimum conditions of disease development (Ou, 1985 and Wan et al., 2007). The fungus Rhizoctonia solani Kuhn., survives in the form of sclerotial bodies in the soil for several years in stubbles of the previous season crop and on weeds (Kozaka, 1961 and Kobayashi et al., 1997). The sclerotia of the fungus survive in soil and are disseminated by irrigation water (Premlatha and Dath, 1990). The disease is initially soil borne, subsequent spread is foliar. Effective management of sheath blight disease in rice is possible only when the pathogen is eliminated completely or the propagules are brought down below economic threshold limits at field level. Seed treatment (IRRI, 1980a), soil application (Chen and Chu, 1973) and foliar spray with systemic fungicides and antibiotics (Dev and Mary, 1986), have given effective control of the disease. However, these treatments are expensive and add pesticides to the environment. Use of beneficial micro-organisms as biofertilizers and biocontrol agents has become more important in recent years not only to improve plant growth and to manage plant diseases but also to avoid environmental pollution.

Among the fungal biocontrol agents *Trichoderma* spp. are the most promising in controlling foliar and root diseases in a wide range of agricultural crops (Elad, 2000 and Mathivanan *et al.*, 2004). At present several commercial formulations of Trichoderma are available worldwide for field application (Whipps and Lumsden, 2001). *Trichoderma* spp. have been shown to be effective for the control of diseae on rice (Tewari and Singh, 2005 and Naeimi *et al.*, 2010).

Among bacterial antagonists, fluorescent pseudomonads, particularly *Pseudomonas fluorescens* Migula is considered to be the prime candidate for

biological control. Fluorescent pseudomonads are known to inhibit the growth of plant pathogens by diverse mechanisms besides, they also act as plant growth promoting substances (Lifshitz *et al.*, 1987) and enhancing the crop yield (Dileep Kumar and Dube, 1992).

Use of multiorganisms as crop production and crop protection inputs is currently under practice in agriculture. Further combination of biocontrol agants was reported to offer an effective control of plant diseases (Duijiff *et al.*, 1999 and De Boer *et al.*, 2003). Present study was carried out to examine the influence of *P. fluorescens* and *T. harzianum* combination on the crop growth and yield in rice. In addition, the combined effect of these biocontrol agents on sheath blight of rice caused by *Rhizoctonia solani* was also evaluated.

MATERIAL AND METHODS

Biocontrol agents: Biocontrol formulations of *Trichoderma harzianum* (2x10⁸ cfu) and *Pseudomonas fluorescens* (2 x 10⁹ cfu) were procured from G.B. Pant University of Ag. and Tech., Pantnagar.

Seed treatment:

About 1 kg of rice seeds (variety PS-5) were uniformly treated. 10 g formulation of *T. harzianum* and *P. fluorescens* separately were used to treat rice seeds, whereas for combined application of biocontrol agents 5 g of *T. harzianum* and 5 g of *P. fluorescens* were used as mixture for treating 1 kg seeds, using rice gruel as sticker and seeds were shade dried for 30 minutes prior to sowing. Similar method was adopted for seed treatment by carbendazim @ 2.5 g/kg seed.

Soil application:

Soil treatment by *T. harzianum* and *P. fluorescens* formulations was done @ 5 kg/ha separately and above formulations were used @ 2.5 kg each for treating soil of one hectare, when used in combination. For soil treatment FYM was used @ 100 kg FYM/ha. Biocontrol formulations were mixed with FYM and this mixture was kept in shade for 10 days, intermittent mixing was done

to promote uniform distribution of biocontrola agents in FYM.

Foliar spray of carbendazim @ 2g/lit was done at 15 days after transplanting.

Field experiments were conducted during *Kharif* 2010, 2011, 2012 in crop cafeteria of Krishi Vigyan Kendra, Rampur. Specific plot having history of significant sheath blight incidence was selected for conducting trials. The experiment was laid out in Randomized Block Design (RBD) with three replications by maintaining a plot size of 5.0× 4.0 m². Sharbati, the local rice variety was used for the study. Seeds were sown as per treatments and nursery was raised. Rice seedlings were removed after 20 days and transplanted in the main field. All the recommended agronomic practices were followed for rice cultivation.

Shoot length and root lengths of plants were recorded at ninety days after transplanting. Plant height, shoot and root dry weight and days of 50 per cent flowering were recorded at flowering stage. Twenty hills from each treatment per replication were selected at random and tagged to observe the sheath blight incidence. The desease score was recorded at flowering stage according to standard evaluation system of International Rice Research Institute (IRRI, 1980b). The healthy and infected tillers were observed and percentage of sheath blight infected tillers was calculated. The crop was harvested at maturity after threshing grain yield in each plot was recorded and expressed in qt/ha at 14 per cent moisture level.

Data of three year trials were compiled and their mean was considered for further analysis. The experimental data were analyzed statistically by one way ANOVA, using MINITAB software. The least significant difference (LSD) was used to separate group mean values when ANOVA was significant at P=0.05.

RESULTS AND DISCUSSION

Seed treatments and soil treatment of *P. fluorescens* and *T. harzianum* significantly increased the shoot and root lengths of rice plants when compared with fungicide treated and untreated control. In combined application of *P. fluorescens* and *T. harzianum* this effect was more pronounced then other treatments. The shoot and root lengths were 31.3 cm and 10.5 cm, but no significant difference was observed as compare to sole application by *P. fluorescens* or *T. harzianum* which resulted in 29.2 cm and 30.5 cm shoot length and 9.6 cm and 9.8 cm root length, respectively as against 25.5 and 7.5 cm, respectively in control (Table 1).

Growth attributes such as plant height, shoot and root dry weights were recorded at flowering stage in treated and control plots. Application of P. fluorescens and T. harzianum either alone or in combination and cabendazim significantly increased the growth parameters in rice over control. Maximum increase in all the growth parameters was recorded in the plots with combination of P. fluorescens and T. harzianum and the minimum was in the carbendazim treated plots. The growth promotion in rice was high following the combined application of P. fluorescens and T. harzianum indicating that synergism existed between the two beneficial micro-organisms as shown for the combination of other micro-organisms (Camprubi et al., 1995). However, there was no significantly significant difference among biocontrol agents and fungicide treated plots for plant height (Table 1). Fluorescent pseudomonads and Trichoderma have been reported to improve overall plant growth and suppress the disease incidence in different crops (Windham et al., 1986; Kloepper et al., 1988; Mathivanan et al., 2000 and Thirup et al., 2003).

Combined application of *P. fluorescens* and *T. harzianum* induced early flowering of rice when 50 per

Treatments	Shoot length (cm)	Root length (cm)	Plant height (cm)	Shoot dry weight (g)	Root dry weight (g)	Days of 50% flowering
P. fluorescens	29.2 ^b	9.6ª	106.3 ^b	28.3 ^b	6.3ª	95.0 ^b
T. harzianum	30.0^{a}	9.8ª	108.5 ^a	29.8 ^a	6.5 ^a	95.0^{b}
Pf + Th	31.3^{a}	10.5 ^a	112.0 ^a	32.0^{a}	7.9^{a}	93.0^{a}
Carbendazim	26.5°	8.2ª	105.0 ^b	25.0°	5.5 ^b	96.0°
Control	25.5 ^a	7.5 ^b	101.0°	23.5^{d}	4.3°	96.0°
C.D. (P=0.05)	1.72	2.56	4.33	2.16	1.89	1.05

In column mean value of three experiments, followed by same letter are not significantly different

cent flowering was observed at 93 days after transplanting as against 96 days in control plots. In plots receiving sole application of *P. fluorescens* and *T. harzianum* 50 per cent flowering occurred at 95 DAT, whereas, in plots having carbendazim treatment 50 per cent flowering occurred at 96 DAT which was similar to control plot. These growth promotion activities could be due to secretion of plant growth promoting substances and plant growth hormones by the rhizosphere microorganisms as demonstrated by Ureta *et al.* (1995).

The sheath blight severity in rice was significantly decreased by P. fluorescens and T. harzianum either alone or in combination and in carbendazim treated plots when compared with control. Minimum disease severity of 9.5 per cent was recorded in plots having combined application of P. fluorescens and T. harzianum as against 21.5 per cent in control. The disease reduction ranged from 46.04 per cent to 55.81 per cent for biocontrol agents and fungicide treated plots compared with control. Disease reduction was at par and maximum in combined application by P. fluorescens and T. harzianum and carbendazim treated plots, whereas, disease reduction was significantly less in plots having sole application of P. fluorescens (46.51%) and T. harzianum (46.04%). Pseudomanas fluorescens inhibited the growth of sheath blight pathogen, R. solani by the production of antibiotics (Gaffney et al., 1994) and siderophores (Savitry and Gnanamanickam, 1987) whereas, T. harzianum degraded the chitin polymers from the cell wall of R. solani by secreting chitinase (Krishnamurthy et al., 1999). Many reports indicate that combining fluorescent pseudomonads with other biocontrol agents resulted in effective control of plant diseases (Raupach and Kloepper, 1998; Duijiff et al., 1999 and De Boer et al., 2003).

Disease incidence was significantly delayed in biocontrol agents treatment either alone or in combination

and carbendazim treated plots. Maximum delay of 35 days was recorded in plots having combined application of *P. fluorescens* and *T. harzianum* (Table 2). This delay in disease incidence could be attributed to rhizospheric competition for space and food between biocontrol agents and pathogenic fungi thus, disrupted population build up of *R. solani*.

Plots given combined application of *P. fluorescens* and T. harzianum yielded a maximum grain yield of 43.50 qt/ha compared to the control (37.0 qt/ha). In general, the grain yields ranged from 40.0 qt/ha to 43.5 qt/ha. All three treatments by biological control agents were at par with each other, whereas, yield in plots receiving seed treatment and foliar spray by carbendazim was significantly lower (40.0 qt/ha) as compare to biological control agents treatments. Promotion of grain yield in biocontrol treated plots is correlated to the vigorous early crop growth due to plat growth promoting activity of the used organisms. In addition, the cumulative effect of nutrient uptake, hormone secretion and disease suppression accelerated by P. fluorescens and T. harzianum might have ultimately resulted in enhanced rice yield as demonstrated by Kloepper et al. (1988) and Sakthivel et al. (1986).

Our results were similar to results for biocontrol agents and fungicide in reduction of rice sheath blight disease and enhancement of crop growth. The results of present investigation suggest that there was no negative effect when the two biocontrol agents were applied together. Synergism between *P. fluorescens* and *T. harzianum* is quite evident in our findings. Hence, the talc formulations of biocontrol agents in combination can be recommended to the farmers as one of the crop protection strategies for management of sheath blight of rice because combined treatment appeared to be promosing, If one organism failed, the other organism(s) possibly will take care of the biological control of the

Treatments	% disease incidence	% decrease in disease incidence	Time of disease appearance (DAT)	Delay in disease (Days)	Yield (qt/ha)	% increase in yield
P. fluorescens	11.5 ^b	46.51	51 ^b	31	42.5 ^a	14.86
T. harzianum	11.8 ^b	46.04	52ª	32	43.2ª	16.75
Pf + Th	9.5 ^a	55.81	55 ^a	35	45.0^{a}	21.62
Carbendazim	10.6 ^a	50.69	47 ^b	27	40.0^{b}	8.10
Control	21.5°	-	$20^{\rm c}$	-	37.0°	-
C.D. (P=0.05)	1.06	-	3.85	-	1.38	-

In column mean value of three experiments, followed by same letter are not significantly different

target pathogen.

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