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Full Length Research Paper

Biocontrol properties of indigenous *Trichoderma* isolates from North-east India against *Fusarium oxysporum* and *Rhizoctonia solani*

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***Trichoderma* is one of the best known and well described biocontrol fungi for its lytic activity and antagonistic properties against phytopathogens. In the present study, a total of 114 isolates of *Trichoderma* were isolated from the soil collected from diverse climatic conditions of Manipur on selective medium. Out of the total isolates, 80% shows high degree of antagonism against *Fusarium oxysporum* while 68% *Trichoderma* isolates gives strong activity against *Rhizoctonia solani*. Based on their antifungal activity in dual plate assay, 25 isolates were selected for further analysis. The interaction between the *Trichoderma* and fungal pathogens were examined microscopically. Several biocontrol mechanisms were studied and analysis data showed that the clearing zone diameter of protease activity of these indigenous *Trichoderma* isolates ranges from 10 to 60 mm. Among them, 84% gave high chitinase activity and their activity ranges between 10 to 85 mm. whereas, ß-1,3-glucanases activity showed a clearing zone diameter ranging from 10 to 70 mm. Based on their relative biocontrol potency, three indigenous *Trichoderma* isolates (T10, T17 and T83) were selected for pot culture experiment for testing their biocontrol efficacy against wilting and damping off diseases of common beans. Among all the treatments, T83 showed better biocontrol efficacy against the two test fungus as compared to the exotic *Trichoderma harzianum* (ITCC No. 6276) strain.**

Key words: Antagonistic, Chitinase, *Fusarium oxysporum,* protease, *Rhizoctonia solani, Trichoderma* sp.

INTRODUCTION

Plant diseases play a major role in the decrease of food production in agricultural scenario. Chemical compounds have been used to control plant diseases, but abuse in their employment has favored the development of pathogens resistant to fungicides.

Biological control using potential microorganisms having strong antifungal activity is coming up as an alternative strategy for disease management, which is also ecology-conscious and environment friendly. Among all the microbial biocontrol agents (BCAs), *Trichoderma* sp. is one of the most commonly used worldwide as safe BCAs. The success of *Trichoderma* strains as BCAs is due to their high reproductive capacity, ability to survive under very unfavourable conditions, efficiency in the utilization of nutrients, capacity to modify the rhizosphere, strong aggressiveness against phytopathogenic fungi and efficiency in promoting plant growth and defense mechanisms. So far, *Trichoderma* sp. is among the most studied fungal BCAs and commercially marketed as biopesticides (Harman, 2000).*Trichoderma* is a secondary opportunistic invader, a fast growing fungus, a strong spore producer, a source of cell wall degrading enzymes and important antibiotic producers (Francesco et al., 2008). The main biocontrol mechanisms exhibited by *Trichoderma* in direct confrontation with fungal patho-

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gens are mycoparasitism (Papavizas, 1985; Harman and Kubicek, 1998; Howell, 2003) and antibiosis (Howell, 1998; Sivasithamparam and Ghisalberti, 1998). As Manipur being in one of the rich Indo-Burma mega biodiversity hot-spot region of the world, is expected to occur diverse range of *Trichoderma* sp. having potential biocontrol activity.

Bean (*Phaseolus vulgaris* L.) is one of the most important food legumes for direct human consumption in the world. Several soilborne fungi cause wilt and damping-off diseases in beans. The main pathogens responsible for causing wilt and damping off incidence in beans are *Fusarium oxysporum* f.sp.*phaseoli* and *Rhizoctonia solani* (Kuhn) respectively (El-Mougy et al., 2007). Yield losses in severely infested areas may be as high as 50% (Estevez et al., 2001). Many researchers have demonstrated the potential of *Trichoderma* sp. in controlling wilt and damping-off diseases of crop plants caused by *Fusarium* sp. and *Rhizoctonia solani* (Dubey et al., 2007 and Rojo et al., 2007). Therefore, the present study aimed to study the biocontrol efficacy of the local strains of *Trichoderma* for the control of wilt and damping off disease in beans.

MATERIALS AND METHODS

Isolation of *Trichoderma* sp.

Soils from the agricultural fields of nine different districts of Manipur were collected and preserved on 4°C. *Trichoderma* strains were isolated using selective medium by following modified method given by Tate (1995). Then the colonies were transferred on potato dextrose agar (PDA) plates and incubated at 27°C for five to six days followed by morphological identification based on colony characteristics (Gams and Bisset, 1998) and microscopically according to the related literatures.

Evaluation of antifungal activity

Fungal inhibition assay of the indigenous *Trichoderma* isolates were performed by dual plate assay against the test pathogens such as *Fusarium oxysporum* (C.O. of wilt) and *Rhizoctonia solani* (C.O. of damping off) isolated from the infected beans collected from the different crop fields of Manipur (Morton and Strouble, 1955) and plates were incubated at 28°C ± 2°C for five to six days on PDA plates. For each treatment, three triplicate plates were maintained. The treatment without antagonist served as control. In dual cultures, the *Trichoderma* isolates were categorized as effective biocontrol agents based on their ability to overgrow and inhibit the pathogens. The interaction between the *Trichoderma* and pathogens were examined under the microscope.

Determination of different cell wall degrading enzymes of the *Trichoderma* isolates

Protease activity

Protease activity of *Trichoderma* isolates was determined according to the modified method given by Berg et al. 2002. Skim milk agar medium (51.5/lt.) was used for detection of protease activity. Culture disc from 5-6 days old *Trichoderma* cultures were inoculated on skim milk agar medium and incubated at 28°C ± 2°C for three to four days. *Trichoderma* which shows proteases activity gave a clearance zone indicating the production of protease enzymes.

Chitinase activity

Chitinase activity of the *Trichoderma* isolates was determined according to the modified method given by Roberts and Selitrennikoff (1988) on chitin detection medium.

Preparation of colloidal chitin: 5.0 g of chitin was added to 60 ml of concentrated HCl (acid hydrolysis) by constant stirring using a magnetic stirrer at 4°C and kept in refrigerator overnight. The resulting slurry was then added to 2000 ml. of ice-cold 95% ethanol and kept at 26°C for overnight (ethanol neutralization). Then it was centrifuged at 3000 rpm for 20 min at 4°C. The pellet was washed with sterile distilled H_2O. The final colloidal chitin was stored at 4°C until further use.

Final chitinase detection medium: The final chitinase detection medium per litre comprises of 4.5 g colloidal chitin, 0.3 g magnesium sulphate, 3.0 g ammonium sulphate, 2.0 g potassium dihydrogen phosphate, 1.0 g citric acid monohydrate, 15 g agar, 0.15 g bromocresol purple and 200 µl of tween-80. The pH of the media was maintained at 4.7 and autoclaved at 121°C for 15 min. The fresh culture plugs of *Trichoderma* isolates to be tested for chitinase activity were inoculated into the sterile plates containing chitinase detection medium and incubated at 28±2°C for two to three days and observed for the colored zone formation. Chitinase activity was identified due to the formation of purple colored zone. Color intensity and diameter of the purple colored zone were taken as the criteria to determine the chitinase activity after three days of incubation.

ß-1,3-Glucanases activity

For plate screening of ß-1,3-glucanases activity, carboxy methyl cellulose agar (CMC agar) medium amended with laminarin was used according to the modified method given by El-Ketatny et al. (2001). A 6 mm culture disc was placed at the center of the plate. Plates were incubated at 25°C for three days. ß-1,3-glucanases activity on the plates were observed by dipping in 0.1% congored dye for 15 to 20 min followed by distaining with 1 N NaCl and then with 1 N NaOH for 15 min. The destaining was repeated twice. ß-1, 3-glucanases activity was recorded with the clearance zone formation.

Biocontrol potential of the indigenous *Trichoderma* isolates in green house trial

Based on the relative biocontrol potency such as antifungal activity and the production of enzymatic activities, only three *Trichoderma* isolates (T10, T17, and T83) were selected for pot culture experiment. A pot experiment was conducted during 2009 to 2010 in the greenhouse of Institute of Bioresources and Sustainable (IBSD), Takyelpat, Imphal using a completely randomized block design with different treatments. The disease control efficacy of the

three local isolates was compared with a reference *Trichoderma harzianum* strain (ITCC No. 6276) against wilt (C.O. *F. oxysporum*) and damping off (C.O. *R. solani*) of beans. The surface sterilized bean seeds were planted in the plastic pot, which was filled with 3 kg of sterilized soil. Two days ahead of sowing the bean seeds, each pot was pretreated with seven to eight days-old fungal pathogens (Dubey et al., 2007). Bean seeds were treated with eight days old *Trichoderma* spores concentration of 10^{-9} along with 1% CMC and charcoal powder to ensure uniform distribution of the biocontrol agent on the seed surface and seeds were shade dried. The pots without pathogen application were taken as controls. Different treatments taken for pot experiments were named as: (i) control, (ii) F. O, (iii) F.O + *T. harzianum*, (iv) F. O. + T10, (v). F. O. + T17, (vi) F. O. + T83 (vii) R. S. (viii)R. S. + *T. harzianum*, (ix) R. S. + T10, (x). R. S. + T17 and (xi) R. S. + T83. The plants were observed for the occurrence of wilt and damping off incidence after 30 days of sowing and compared with the control pots.

Increase in root and shoot lengths were recorded after 45 days of growth. The percentage reduction of roots lengths and shoot lengths were evaluated and compared with the control pots.

Disease incidence (%) was calculated using the formula n/N × 100%, where, n = number of plants affected by disease, N = Number of plants assessed. Disease reduction (%) was also calculated using the formula:

Disease reduction (%) = [(Growth in control – Growth in treatment) / Growth in control] × 100

The ANOVA approach was used to evaluate the biocontrol efficiency of *Trichoderma* biocontrol agent both *in vitro* and *in vivo* experiments. Comparison among treatment are means of appropriate control treatment which were made at P=0.05. In case of the green house experiment, pool data of the pot experiments were subjected at ANOVA.

RESULTS

Isolation and characterization of *Trichoderma* strains

A total of 114 *Trichoderma* isolates obtained from the different soil samples collected from diverse agricultural fields of Manipur were morphologically and microscopically characterized. The distinctive morphology of *Trichoderma* includes rapid growth, bright green or white conidial pigments and a repetitively branched, but some of them have poorly defined conidiophore structure. Cultures showed very fast growth rate between 25 to 30°C. Microscopically they were identified based on the shape of conidiophores, conidia and philiades using 100X in Olympus BX61 microscope. Some of the conidiophores were highly branched and difficult to define or measure.

And others were loosely or compactly tufted, often formed in distinct concentric rings and borne along the scant aerial hyphae. Main branches of the conidiophores produce lateral side branches and often phialides arising directly from the main axis near the tip. Typically the conidiophore terminates in one or a few phialides. Phialides are typically enlarged in the middle, cylindrical or nearly subglobose (Figure 1).

Antifungal activity

The antifungal activities of 114 *Trichoderma* isolates against *F. oxysporum* and *R. solani* were determined by dual plate assay on PDA plates. Out of the total 114 isolates, 80% of the *Trichoderma* isolates showed high degree of antifungal activities against F.O while 68% of *Trichoderma* isolates gives strong antagonistic activity against *R. solani* (Table 1). Based on their adaptive focused acoustics (AFA) on dual plate assay 25 test isolates were selected for further analysis of different biocontrol mechanisms.

Determination for the production of different cell wall degrading enzymes

Each of the 25 selected biocontrol *Trichoderma* isolates were evaluated for the production of different enzymatic activities such as proteases, chitinases and β-1,3-glucanases *in vitro* condition.

Proteases activity

The production of protease, a fungal cell wall degrading enzymes which is an important mechanism of fungal growth inhibition was detected for all selected isolates. Protease activity was evaluated from the clearance zone given on the skim milk agar plates and the degree of protease activities were measured based on the diameter of clearance zone which ranges from 10 to 60 mm. Out of the total 25 *Trichoderma* isolates, 72% of them gave maximum protease activity. Among them, T54 and T69 were the two isolates which showed highest protease activity with more than 50 mm zone width (Figure 2).

Chitinases activity

Chitinase activity, an important enzymatic activity to degrade the chitin which is a major structural polysaccharide of fungal pathogens was evaluated by using the chitin detection medium. The formation of purple colour zone on the chitin detection medium indicated the presence of chitinase activity. Chitinase activity of the 25 *Trichoderma* isolates was determined according to the diameter and intensity of the colour zone. The diameter of the colour zone ranged from 10 to 85 mm approximately and 84% out of the 25 *Trichoderma* showed maximum chitinase activity. T17, T69, T80 and T83 showed highest chitinase activity which were above 60 mm in zone diameter (Figure 3).

β-1,3-Glucanases activity

β-1,3-glucanases are enzymes which hydrolyzes the O-

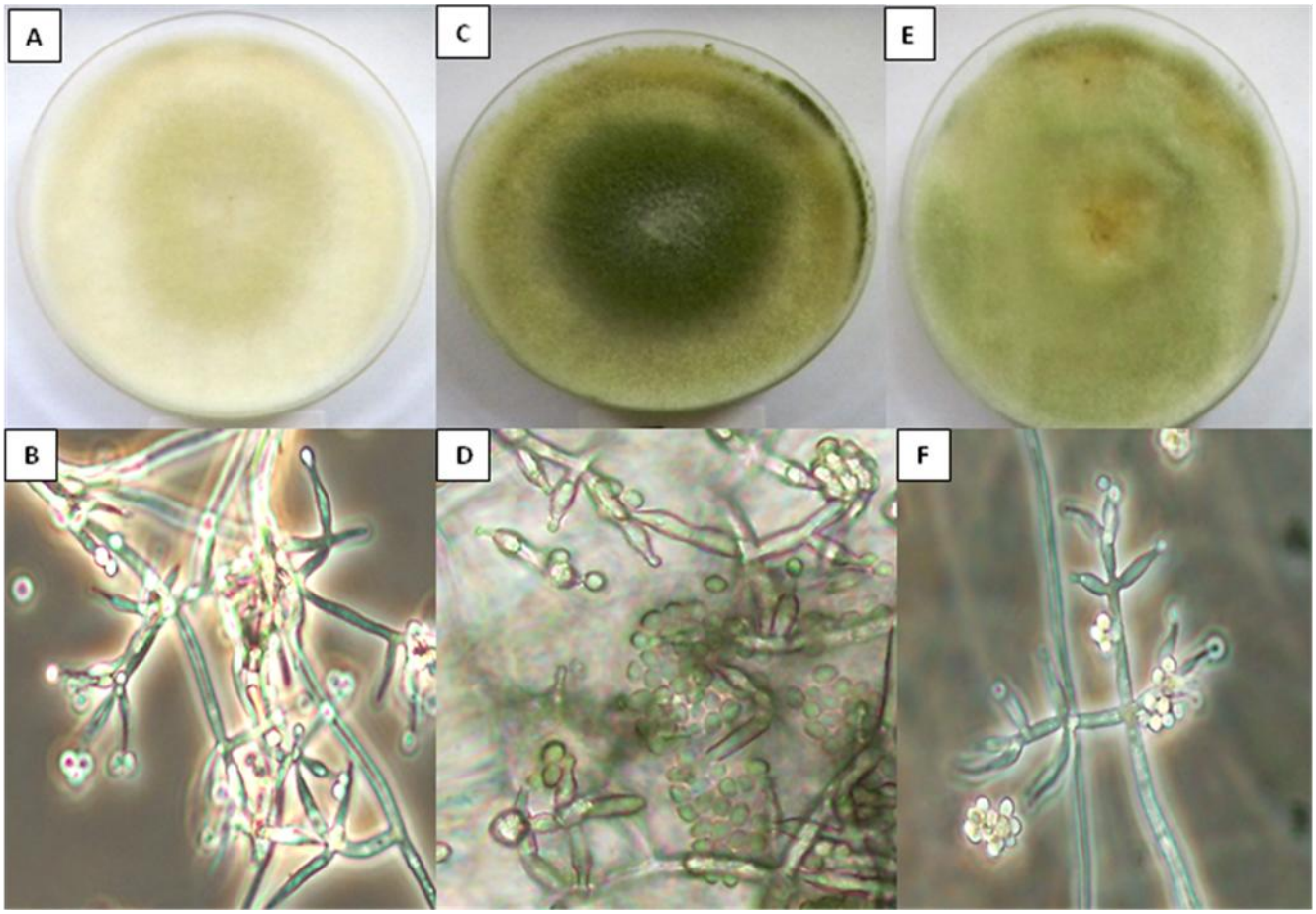

Figure 1. A, Colony morphology of T10 isolates on PDA plate; B, microscopic view of T10 isolate; C, colony morphology of T17 isolates on PDA plate; D, microscopic view of T17 isolate; E, colony morphology of T83 isolate on PDA plate; F, microscopic view of T83 isolate. PDA, Potato dextrose agar.

glycosidic linkages of β-glucan chains and are among the plant defense responses to pathogen attack. The production of β-1,3-glucanases activity by the 25 *Trichoderma* isolates were determined by using the CMC agar medium. The production of β-1,3-glucanases was detected according to the clearance zone formation. The diameter of the clearance zone ranged from 10 to 70 mm. Among the selected isolates, 60% showed maximum activity. T8, T10, T17, T72 and T83 were among the *Trichoderma* isolates which showed maximum activity with more than 50 mm in diameter (Figure 4)

Biocontrol activity in green house trial

Three *Trichoderma* isolates with high antagonistic activity against *F. oxysporum* and *R. solani* were selected for the evaluation of their biocontrol efficacy in a green house trial and compared their activity with the control untreated pot. In this test, no *Trichoderma* could completely protect the plants against *F. oxysporum* and *R. solani*, although all isolates significantly increased shoot length, root length and fresh weight as compared to the infested control plants (Table 2). Plant height was recorded after 45 days of growth in the trial pots. In the *F. oxysporum* treated pots, the plants treated with T83 consistently provided significant increase in both root and shoot length, with a highest root and shoot length reduction percentage of 6.88 and 8.88, respectively. Whereas in case of *R. solani* treated plant, root length was found to increase when treated with the T10 but increase in shoot length was recorded in the plant treated with T83. Whereas in the pot of *F. oxysporum* treated with reference strain *T. harzianum* (ITCC No. 6276), the root length reduction value was found to be 12.05% and shoot length reduction value of 17.75% and in case of *R. solani* and *T. harzianum (*ITCC No. 6276) treated plants, the percentage reduction of root length and shoot length were 32.75 and 50.41, respectively. The least significant difference (LSD) (P=0.05) value for root length was 0.014

Table 1. Antifungal activity (% inhibition) against *F. oxysporum* and *R. solani* by the selected indigenous *Trichoderma* isolates using dual culture methods.

S/N	Name of *Trichoderma*	Antifungal activity	
		Fusarium oxysporum	***Rhizoctonia solani***
1.	T1	74.33 ± 2.33^{b}	79.00 ± 3.78^{b}
2.	T2	71.67 ± 2.03^{b}	64.67 ± 2.60^{c}
3.	T4	72.33 ± 1.45^{b}	70.00 ± 3.60^{b}
4.	T7	29.00 ± 2.08^{e}	19.67 ± 1.67^{g}
5.	T8	75.33 ± 2.03^{b}	75.67 ± 4.70^{b}
6.	T10	72.00 ± 3.51^{b}	62.33 ± 1.45^{c}
7.	T15	65.33 ± 2.60^{c}	70.67 ± 0.67^{b}
8.	T17	62.33 ± 1.45^{c}	75.00 ± 2.89^{b}
9.	T20	25.67 ± 3.48^{e}	60.67 ± 0.67^{c}
10.	T22	21.67 ± 6.00^{e}	15.00 ± 2.89^{g}
11.	T34	69.00 ± 2.08^{c}	41.67 ± 1.67^{d}
12.	T36	72.33 ± 1.45^{b}	72.33 ± 4.33^{b}
13.	T38	62.33 ± 1.45^{c}	21.67 ± 1.67^{f}
14.	T40	69.67 ± 3.93^{c}	76.67 ± 4.41^{b}
15.	T47	64.67 ± 3.71^{c}	78.33 ± 4.41^{b}
16.	T51	79.00 ± 3.78^{b}	61.67 ± 4.41^{c}
17.	T54	45.00 ± 2.89^{d}	69.00 ± 2.08^{c}
18.	T62	82.33 ± 1.45^{a}	45.00 ± 2.89^{d}
19.	T69	79.33 ± 2.33^{b}	41.67 ± 1.67^{d}
20.	T72	79.00 ± 3.78^{b}	71.67 ± 4.41^{b}
21.	T75	79.33 ± 2.33^{b}	75.00 ± 2.89^{b}
22.	T80	14.00 ± 3.05^{f}	31.67 ± 0.67^{e}
23.	T83	82.33 ± 1.45^{a}	81.67 ± 1.67^{a}
24.	T112	77.33 ± 3.93^{b}	69.00 ± 2.01^{c}
25.	T114	79.33 ± 1.76^{b}	42.33 ± 1.45^{d}

Values are average of three replicates; ± SEM; values with the same letter do not differ significantly ($P<0.05$).

while for the shoot length, it was found to be 0.034 thereby indicating a significant increase in root and shoot length (Table 2).

It is evident from Table 2 that the plant treated with *F. oxysporum* and T83 gave an effective disease control of wilt diseases of beans with a record of only 9% disease incidence as compared to control plants, and the highest disease reduction percentage of wilt diseases of bean was also recorded from T83 treated plant. Whereas the disease incidence of *F. oxysporum* with *T. harzianum* was 33.33% and the disease reduction percentage of this treatment was 51.5%. In case of the plant infested with *R. solani*, the treatment plant of *R. solani* and T83 showed the lowest disease incidence as well as highest disease reduction percentage of damping off disease of bean with a reading of 11 and 76.6% respectively. Whereas, the disease incidence percentage of R. S. + *T. harzianum* was 66.67% and their disease reduction percentage was 14.1%. This result shows that the indigenous *Trichoderma* T83 isolates gives better disease control activity in both the wilt and damping off diseases of bean as compared to the reference *T. harzianum*.

DISCUSSION

Among the fungal biocontrol agents, *Trichoderma* is one of the most commonly used organisms for the control of soil borne fungal pathogens (Harman, 2000). The aim of the current study was isolation, screening and selection of the potential indigenous *Trichoderma* from Manipur to be utilized as a potential antagonist against the *F. oxysporum* and *R. solani* associated with wilt and damping off diseases in beans. Isolation of local strains of *Trichoderma* is essential for successful identification of potential biocontrol agents (Williams and Asher, 1996). Field soil used in this study was collected from different agricultural fields of Manipur having pH ranging from 6.3 to 6.8 on selective medium (Elad et al., 1981). A total of 114 *Trichoderma* isolates were evaluated for their

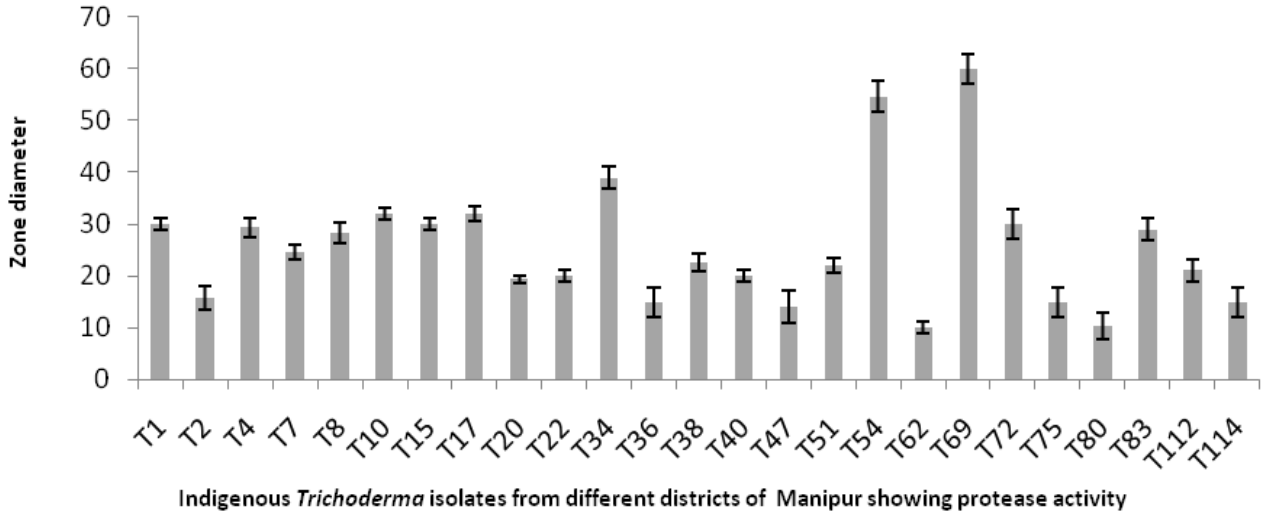

Figure 2. Specific activity of protease from different selected *Trichoderma* isolates. Each bar represents the average of three independent measurements.

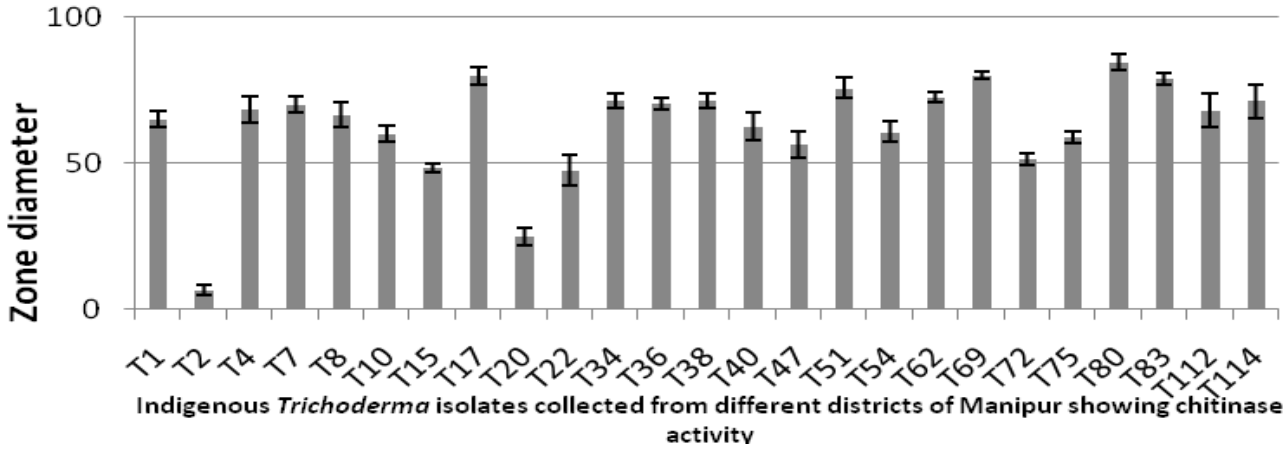

Figure 3. Specific activity of chitinase from different selected *Trichoderma* isolates. Each bar represents the average of three independent measurements.

antifungal activity against *F. oxysporum* and *R. solani.* The characterization of all the *Trichoderma* isolates into species aggregates are made on the basis of cultural and morphological characters according to Rifai (1969). In a similar study, Sariah et al. (2005) morphologically identified *T. harzianum*, *Trichoderma virens, Trichoderma koningii* and *Trichoderma longibrachiatum*. Zakaria (1989) identified five species of *Trichoderma* from the rhizosphere of rubber in Malaysia. Similar studies by Lim and Tech (1990) isolated three species of *Trichoderma* in Malaysian soil, *T. hamatum* (Bon.) from cocoa grown on ex-rubber areas, *T. koningii* (Rifai) from rubber and cocoa and *T. harzianum* (Rifai) from rubber and oil palm.

Antagonistic effect based on the dual culture experiments showed that *Trichoderma* isolates significantly inhibited the mycelial growth of *F. oxysporum* and *R. solani.* 80% of *Trichoderma* isolates showed high degree of antagonism against *F. oxysporum* while 68% of *Trichoderma* isolates gave strong antagonistic activity against *R. solani*. Chung and Choi (1990) reported effective inhibition of *F. oxysporum* f. sp. sesame by *T. viride*. Perello et al. (2003) reported that *Trichoderma* sp. significantly inhibited *Drechslera tricirepentis* colony growth between 50 and 74% utilizing dual culture techniques on PDA. Hermosa et al (2000) deduced that *T. harzianum* had a potential biocontrol activity in a dual culture studies against the phytopathogenic fungi of *Phoma betae, Rosellinia necatrix, Botrytis cinera* and *F.*

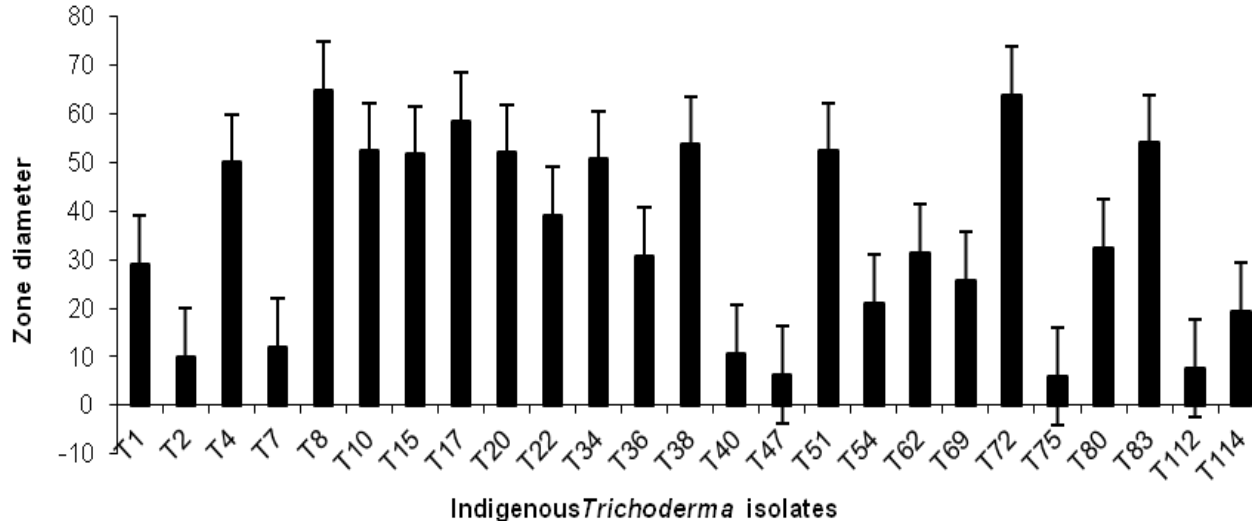

Figure 4. Specific activity of β-1,3 glucanases from different selected *Trichoderma* isolates. Each bar represents the average of three independent measurements.

Table 2. Biological efficacy of different *Trichoderma* isolates against the *F. oxysporum* and *R. solani* in beans.

S/N	Treatment	Root length	% Reduction of root length	Shoot length	% reduction of shoot length	Disease incidence (%)
1.	Control	82.67 ± 1.2[a]	0	83.33 ± 0.89[a]	0	0
2.	*F. oxysporum*	30 ± 1.15[f]	65.91	30.67 ± 0.67[f]	93.71	89
3.	*F. oxysporum* +*T. harzianum*	68.33 ± 2.02[c]	12.05	69.00 ± 0.67[c]	17.75	33.33
4.	*F. oxysporum* + T10	73.33 ± 0.89[b]	18.93	73.33 ± 1.45[b]	11.30	11
5.	*F. oxysporum* + T17	74.33 ± 0.89[b]	10.35	74.00 ± 1.15[b]	10.09	11
6.	*F. oxysporum* + T83	75.33 ± 1.45[b]	6.88	74.67 ± 1.45[b]	8.88	9
7.	*R. solani*	10.00 ± 1.15[g]	65.49	10.00 ± 1.15[g]	87.90	89
8.	*R. solani* + *T. harzianum*	42.00 ± 0.57[e]	32.75	41.00 ± 0.57[e]	50.41	66.67
9.	*R. solani* + T10	51.33 ± 0.89[d]	1.71	51.00 ± 1.73[d]	37.91	44.33
10.	*R. solani* + T17	57.33 ± 0.89[d]	10.35	37.00 ± 1.15[f]	54.43	22.33
11.	*R. solani* + T83	57.33 ± 1.45[d]	13.76	67.00 ± 1.15[c]	18.14	11
	LSD (P=0.05)	0.014		0.034		

Values are average of three replicates; ± SEM; values with the same letter do not differ significantly (P<0.05).

oxysporum f, sp. *dianthea* in the three different media. *Whipps* (1987) stated that *T. harzianum* appears to be a promising organism, particularly for use against *R. solani*. *Trichoderma* sp. are known to act through several mechanisms such as hyperparasitism, inhibition and antibiosis (Weindling, 1932; Hadar et al., 1979).

Different enzymatic activities such as proteases, chitinases and β-1,3-glucanases of the selected *Trichoderma* isolates were determined *in vitro* condition. Proteases activity of the 25 *Trichoderma* isolates ranges from 10 to 60 mm while for the chitinase activity, it ranges from 10 to 85 mm approximately. Out of the total 25 *Trichoderma* isolates, 60% of them showed maximum β-1,3-glucanases activity and ranges from 10 to 70 mm in diameter.

Benitez et al., (2004) demonstrated that *Trichoderma* strains that overproduce chitinases have been shown to be effective biocontrol agents against pathogens such as *B. cinera* and *Rhizoctonia meloni*. Howell (2003) reported that biocontrol of *B. cinerea* by *T. harzianum* has been attributed in plant to the action of proteases produced by the BCA that inactivate hydrolytic enzymes produced by this pathogen on bean leaves. Benitez et al. (1998) demonstrated that proteases from *T. harzianum* play an important role in biological control. Szekeres et al. (2004) reported the role of proteases in mycoparasitism and has

been reinforced with the isolation of new proteases-overproducing strains of *T. harzianum*. Howell (2003) tested the role of chitinases in mycoparasitism and believed that chitinases is a key enzyme in this process. Harman et al. (2004) also described the types of chtinases detected from *T. harzianum*, *T. atroviride* and *T. virens*. Simmoms, 1994 described that glucanases are among the plant defense responses to pathogen attack. Production of four β-1,3-glucanases by *T. harzianum* has been described by de la Cruz et al. (1995); Kitamoto et al. (1987); Lorito et al. (1994).The application of the indigenous *Trichoderma* isolates in our study against *F. oxysporum* and *R. solani* in beans shows good result and satisfactory reduction in the disease symptoms. Similarly, Singh and Singh (2004) screened the two strains, viz, *T. harzianum* (IMI. No. 359869) and *T. virens* (ITCC No. 106695) having the potential to control the collar rot disease of *Mentha* species caused by *Sclerotium rolfsii*. Larralde-Corona et al. (2008) reported the biocontrol potential of novel native *Trichoderma* strains against *Macrophomina phaseolina* isolated from sorghum and common bean.

In the present study, the effect of treatment was found to be significant (LSD (P=0.05) of root length = 0.014 and LSD of shoot length = 0.034) against wilt and damping off incidence respectively. The maximum wilt and damping off disease reduction were observed in pots treated with F. O. + T83 and R. S. + T83, respectively (Table 2). This could be due to a high degree of mycoparasitism and production of some lytic enzymes by this *Trichoderma* isolate. *Trichoderma* sp. have been shown to decrease wilt incidence in chickpea plants (Dubey et al., 2007) and increase root development in numerous other plants (Harman, 2006).From the current study, it can be postulated that there is maximum disease reduction in the application of indigenous *Trichoderma* isolate (T83) against the wilt and damping off diseases of bean as compared to the reference *T. harzianum* (ITCC No.6276) strain. This may be due to a number of reasons, including pathogenecity of isolates, climatic adaptability, influence of the pathogen origin and even the influence of local bean cultivars use in this region (Harman, 2006; Sharon et al., 2007). Therefore, T83 isolate native to Manipur could be an excellent candidate for providing long-term biocontrol agent against *F. oxysporum* and *R. solani* in beans with the aim of reducing the use of chemical pesticides in this region.

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