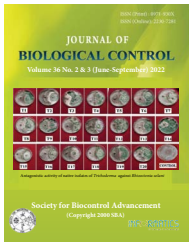

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Research Article

Antagonistic potential and growth promoting activities of novel indigenous strains of *Trichoderma*

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ABSTRACT: Rice (*Oryza sativa* L.), as a cereal grain, is the most widely consumed staple food for a large part of the world's human population, especially in Asia and Africa. Rice production is often subjected to several biotic and abiotic stresses. Rice sheath blight is one of the most destructive diseases causing economic losses in rice yields and affecting quality worldwide. Twenty soil samples were collected from the rhizosphere of rice crop from different regions of Northern Karnataka. *Trichoderma* spp. were isolated from the rhizospheric soil samples. The antagonistic potential of *Trichoderma* spp. was studied using dual culture technique. Among twenty strains of *Trichoderma* the highest inhibition (>50%) was recorded in nine strains which ranged from 54.20 to 65.10 per cent. *Trichoderma harzianum* recorded highest seed germination (100%), root length (13.73 cm), shoot length (8.64 cm) and seedling vigour index. In pot culture experiment, the *Trichoderma* strains significantly improved the growth parameters such as root length, shoot length and number of tillers per plant. Among the different treatments, seed treatment + root dipping + foliar spray with *T. harzianum* strain was highly effective in increasing the shoot length, root length and seedling vigour index at 30, 60 and 90 days after sowing. This strain was found highly effective in inhibiting the pathogen and promoting the growth of rice plants.

KEYWORDS: Plant growth promotion, rice, sheath blight, *Trichoderma* strains

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INTRODUCTION

Rice (*Oryza sativa* L.), as a cereal grain, is the most widely consumed staple food for a large part of the world's human population, especially in Asia and Africa.

Sheath blight is one of the major devastating diseases of rice. In India, sheath blight is known to occur in almost all the rice growing states of the country causing up to 58 per cent loss in yield. The losses in grain yield varied between 10-36 per cent depending upon the stage of crop at the time of occurrence of disease (Roy, 1979). Sustainable agriculture is vital in today's world as it offers the potential to meet our agricultural needs, something that conventional agriculture fails to do. Microbial populations are responsible for instrumental to fundamental processes that drive stability and productivity of agro-ecosystems. Many chemical control methods are available to combat the disease and often sheath blight outbreaks are common. Effective management of sheath blight in rice is possible only when the pathogen is eliminated completely or the propagules are brought down below economic threshold levels at field level. Present management strategies mostly involve use of chemical fungicides. The adverse effects of chemical fungicides on environment and beneficial microflora are well known. Moreover, the soil borne inoculum is difficult to control so, an economic and viable alternative for Sheath blight management is essential.

Biological control is an effective alternative to chemicals and the promising bio-control agents have been successfully exploited for the management of soil borne plant diseases.

Trichoderma is one of the potential antagonistic fungus which was exploited maximum for the management of plant diseases. *Trichoderma* produces extra cellular enzymes, antibiotics and promote plant growth and induce resistance. They have the potential to proliferate abundantly in various soils. Because of their many advantages *Trichoderma* spp. are used in controlling plant pathogens. The present investigation was carried out to assess the bio efficacy of indigenous *Trichoderma* strains against sheath blight of rice.

MATERIALS AND METHODS

Isolation of Pathogen

Sheath blight diseased samples were collected during *Kharif* 2020 were used for the isolation of pathogen. The samples were washed thoroughly with tap water. Small portion of infected parts containing healthy as well as diseased tissues were cut in to 0.5 cm pieces with the help of sterilized scalpel blade. These pieces were then surface sterilized with 1 percent sodium hypochlorite solution for 1 minute with 3 subsequent changes in sterilized water to remove traces of the chemical. The pieces were then transferred aseptically to Petri dishes containing sterilized Potato Dextrose Agar (PDA) and incubated at 28±2 ºC under BOD incubator. The Petri dishes were examined at regular time intervals for fungal growth radiating from the infected pieces. The pure culture of the pathogen was obtained from hyphal tip culture and such pure culture was used for the further studies. Later, the pathogen culture was subjected to Koch's postulates.

Isolation and maintenance of *Trichoderma* strains

Trichoderma strains were isolated from rice rhizosphere by serial dilution on *Trichoderma* specific medium. Ten grams of soil sample was taken and suspended in 90 ml of sterilized distilled water and stirred well to get 1:10 dilution (10^{-1}). One ml from this was transferred to test tube containing 9 ml of sterilized distilled water to get 1:100 (10^{-2}) dilution. Likewise, the dilution of the sample was prepared up to 1:100000 (10^{-5}). One ml of a final dilution of each sample was pipetted out into each sterile Petri plate separately to which a quantity of 15-20 ml of sterilized and molten medium was poured and gently rotated for uniform mixing and the plates were incubated at 28 ± 1°C for about 6-10 days. The Petri plates were kept under observation daily for the appearance of *Trichoderma* colonies. From the isolated plates, among the different colonies, an actively growing colony of *Trichoderma* strain was selected and plated on PDA (Potato Dextrose Agar) medium and plates were incubated at 28 ± 1°C for about 4 days. Likewise, twenty strains of *Trichoderma* were obtained from soil samples collected from different regions of North Karnataka. For the maintenance, the cultures of *Trichoderma* strains were sub cultured on PDA slants, allowed to grow at 28 °C and such slants were preserved in refrigerator at 4 °C and sub cultured once in 30 days.

Antagonistic activity of native *Trichoderma* strains against *R. solani*

Efficacy of *Trichoderma* strains was tested by dual culture method described by Broadbent *et al.,* (1971). Twenty ml of PDA was poured in each of the sterilized Petri plate of 90 mm diameter. On solidification dual inoculation was done in each plate using 5 mm disc of sheath blight pathogen *Rhizoctonia solani* at one side and that of test bio-agent (*Trichoderma* strain) at other side. Petri plates were then incubated at 25±1 ˚C. The observations on growth of *Trichoderma* over the pathogen were recorded after five days of incubation with the help of plastic scale. The mean of three replications was calculated and expressed in mm. The per cent inhibition over the control was calculated by using the formula

$$I = \frac{(C - T)}{C} \text{ X } 100$$

Where;

I = Per cent inhibition

C = Radial growth of fungus in control

T = Radial growth of fungus in treatment

Molecular identification of efficient strains of *Trichoderma*

The *Trichoderma* strains which were efficient in inhibiting the growth of *R. solani* (> 50%) in dual culture assay were identified by amplifying the Internal Transcribed Spacer (ITS) region. The total genomic DNA of fungi was extracted by using the Cetyl Trimethy lammonium Bromide (CTAB) method. ITS regions were amplified from fungal genomic DNA using fungal universal primers; ITS1-F (CTTGGTCAT-TTAGAGGAAGTAA) and ITS4-R (TCCTCCGCT-TATTGATATGC). Primer sequences were synthesized at commercial facilities (Eurofins, Bangalore, India). Sequencing was carried out by Sanger's dideoxy chain-termination method and aligned by using BLAST analysis (http://www.ncbi.nlm.nih.gov/BLAST).

Elucidation of plant growth promoting activity of efficient indigenous *Trichoderma* strains

Roll Towel Method

Four *Trichoderma* strains which were highly efficient in inhibiting the growth of *R. solani* in dual culture assay were further tested for elucidation of plant growth promoting activity. The efficient strains were assessed for plant growth promotion based on the seedling vigour index using the standard roll towel method (ISTA, 1993). Ten rice seeds which were treated with the each isolate of bio-agent were be kept over the pre-soaked germination paper. The seeds were held in position by placing another pre-soaked germination paper strip and gently pressed. The polythene sheet along with seeds were then rolled and incubated in growth chamber for 14 days. Three replications were maintained for each treatment. Seeds soaked in sterile water served as control. Root length and shoot length of individual seedlings were measured and the germination percentage of seeds was calculated. Later, the seedling vigour index was calculated by

using the formula (Baki and Anderson, 1973).

Vigour index (VI) = Seedling length (Mean root length + Mean shoot length) × Germination in percentage

Per cent germination of rice seeds

The observations on number of *Trichoderma* treated rice seeds germinated and total number of seeds kept for incubation were recorded after 14 days of incubation under growth chamber. Later, percent germination was calculated.

Plant growth promotion activity under glass house conditions

One highly efficient strain of *Trichoderma* which was found highly superior in the *in vitro* evaluation test (roll towel method) was further selected to carry out the studies on plant growth promoting activities under glass house conditions. The strain was inoculated into pots through different delivery systems. Each treatment was replicated three times and un inoculated plants were kept as control.

Methods of application of *Trichoderma* strain

Seed treatment

Rice seeds were surface sterilized with 2 % Sodium hypochlorite solution and soaked in suspension of *Trichoderma* ($9x10^8$cfu ml^{-1}). The seeds were allowed to sprout for 24 h before sowing in pots (Vidhyasekaran *et al.,* 1997).

Root dipping

Rice seedling in bundles were dipped in 250 ml of superior *Trichoderema* strain suspension ($9x10^8$cfu ml^{-1}) for 2 h, ensuring that roots alone were immersed in the inoculum and planted in the pots. For this, ten plants were maintained per pot.

Foliar application

25 ml of superior *Trichoderma* strain suspension (9 × 108 cfu ml^{-1}) was sprayed to 30 days old seedlings (Vidhyasekaran *et al.,* 1997).

The following treatment combinations were formulated for pot experiment to know the plant growth promoting activity of superior *Trichoderma* strain under *in vitro* (Table 1).

Observations on growth parameters

Shoot and root length

The shoot and root length of different treatments were measured at 30, 60 and 90 days with standard scaling method. Observations were taken for all the ten plants in the pot and mean was calculated and then expressed in centimeters.

Table 1. Treatment details for pot culture experiment

Sl. No.	Treatment details
1	Seed treatment with *T. harzianum*
2	Seed treatment+ seedling dip with *T. harzianum*
3	Seed treatment + seedling dip + spraying *T. harzianum*
4	Control

Total number of tillers $hill^{-1}$

In each pot, the number of tillers were counted for each plant at 30, 60 and 90 DAS and expressed as total number of tillers $hill^{-1}$.

Statistical analysis

Statistical analysis was carried out as per the procedures given by Panse and Sukhatme (1985). Actual data in per cent were converted to angular transformed values before analysis.

RESULTS

The pathogen *R. solani*

The samples of affected sheath showing typical blight symptom collected from different geographic regions were used for isolation of pathogen by standard tissue isolation method. The fungus was obtained in pure form by hyphal tip isolation as explained in material and methods and maintained in pure form on PDA.

Upon inoculation of the *R. solani* on medium, it produced shade of brown hypha, constriction at the point of branchingand right angle branching in matured hyphae. The isolate shared typical characteristics of *R. solani* branching at right angle near the distal septum of the cell, formation of a septum in the branch near the point of origin, constriction of the branch at origin. Further, it also produced sclerotia which were undifferentiated aggregations of thick-walled cells, small (1-3 mm diameter) irregular-shaped, brown to black structures. The koch's postulates were proved successfully to know the identity of the pathogen and thus the isolated culture was confirmed as *R. solani.*

Isolation and maintenance of indigenous *Trichoderma* strains

Twenty rhizospheric soil samples of rice obtained from different regions of Northern Karnataka were used for the isolation of *Trichoderma* strains.

Twenty indigenous *Trichoderma* strains were successfully isolated by serial dilution technique on *Trichoderma* specific medium and all the twenty isolates showed typical characters of greenish fungal colony and produced typical characteristics of *Trichoderma* spp. when observed under microscope.

Antagonistic potential of *Trichoderma* strains against *R. solani*

Twenty strains of *Trichoderma* spp. isolated from healthy rhizospheric soil of rice were screened against *R. solani*for mycelial inhibition by dual culture technique. Zone of inhibition of mycelium (in mm) was recorded and the per cent inhibition was calculated. The results (Table 2) indicated that per inhibition of mycelial growth of pathogen varied greatly among the twenty strains. The highest inhibition (> 50%) was recorded in nine strains which ranged from 54.20 to 65.10 per cent and these nine strains were highly significant in inhibiting the growth of pathogen when compared to other strains and control. Maximum per cent inhibition of 65.10 per cent was observed in TD-14 followed by TD-9 (58.28 %) and TD-3 (56.46 %), while least inhibition of 41.24 per cent was observed in TD-2 and TD-7 strains (Table 2).

Molecular identification of efficient strains of *Trichoderma*

Four highly efficient strains of *Trichoderma* such as TD-1, TD-14, TD-3 and TD-9 were sent for identification as

Table 2. Antagonistic activity of native isolates of *Trichoderma* strains against *R. solani*

Sl. No	Trichoderma strains	Mycelial growth (mm)*	Mycelial inhibition (%)*
1	TD-1	40.33	54.15 (47.71)
2	TD-2	51.67	41.27 (40.29)
3	TD-3	38.33	56.42 (48.69)
4	TD-4	45.67	48.09 (43.97)
5	TD-5	47.67	45.86 (42.28)
6	TD-6	39.67	54.91 (48.15)
7	TD-7	51.67	41.27 (40.30)
8	TD-8	49.33	43.93 (41.50)
9	TD-9	36.67	58.31 (50.48)
10	TD-10	40.33	54.15 (47.71)
11	TD-11	48.33	45.06 (42.48)
12	TD-12	38.67	56.04 (49.48)
13	TD-13	49.33	43.93 (41.49)
14	TD-14	30.67	65.13 (53.85)
15	TD-15	53.33	39.38 (37.43)
16	TD-16	49.33	43.93 (41.50)
17	TD-17	47.00	46.58 (43.70)
18	TD-18	45.00	48.85 (44.83)
19	TD-19	41.33	53.01 (45.96)
20	TD-20	38.67	56.04 (49.92)
21	Control	88.00	0.00 (0.00)
S.Em.±		-	**0.56**
C. D. at 1 %		-	**2.15**

*Mean of three replications Figures in parenthesis are arc sine transformed.

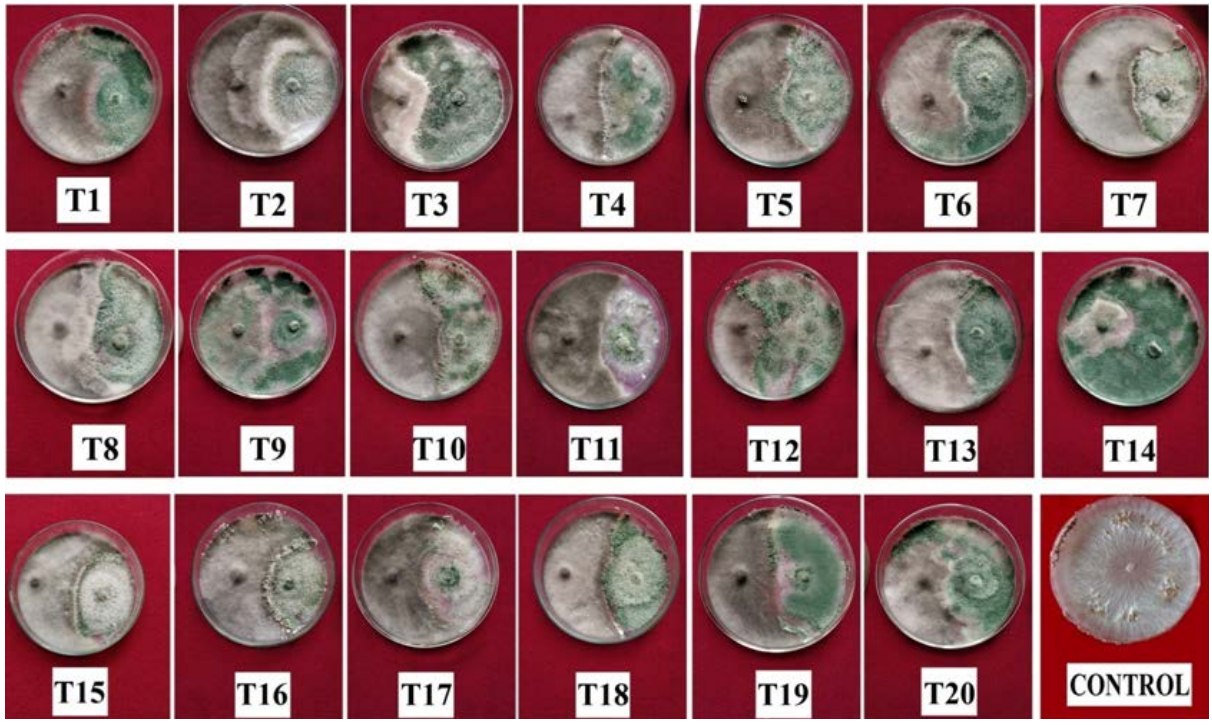

Figure 1. Antagonistic activity of native isolates of *Trichoderma* against *R. solani.*

mentioned in material methods and the accession numbers were obtained. The sequence of strains, TD-1, TD-14,TD-3and TD-9 were identified as *T. asperellum* (Acc. No.: MW063489), *T. harzianum* (ON514140), *T. asperellum* (ON514143) and *T. asperelloides* (ON513885), respectively anddeposited in NCBI genebank. Further, these four identified strains of were used for plant growth promotion studies.

Plant growth promoting activity of *Trichoderma* strains

Germination percentage

The results revealed that germination per centof rice seeds treated with four highly efficient strains of *Trichoderma* varied from 97.67 to 100 per cent. Among them, *T. harzianum* showed the highest germination percentage (100%) followed by *T. asperellum* (TD-3) and *T. asperelloides* with the germination percentage of 97.67. Least germination percentage was recorded in *T. asperellum* (TD-1) (Table 3).

Root length

The results showed that root length ranged from 9.14 to 13.73 cm. The highest root length of 13.73 cm was shown by *T. harzianum* followed by *T. asperelloides* (11.42 cm) and *T. asperellum* (TD-1) (11.41 cm), while *T. asperellum* (TD-3) recorded least mean root length of 9.14 cm compared to 6.44 cm root length in control treatment (Table 3).

Shoot length

Mean shoot length ranged from 8.49 to 8.64 cm. Among the species, *T. harzianum* treated seeds showed highest mean shoot length of 8.64 cm followed by *T. asperellum* (TD-1) which showed 8.59 cm. The lowest mean shoot length was shown by *T. asperelloides*with mean shoot length of 8.49 cm, where control showed 4.47 cm (Table 3).

Seedling Vigor Index (SVI)

The results (Table 3) indicated that seedling vigor index of rice seeds treated with species of *Trichoderma* ranged from1727.78 to 2237.0. Highest seedling vigor index was recorded in isolate *T. harzianum* (2237) followed by *T.*

Table 3. Plant growth promoting activity of efficient *Trichoderma* strains in rice under roll towel method

Sl. No.	Trichoderma strain	Seed germination (%)*	Mean length (cm)*		SVI
			Root	Shoot	
1	*Trichoderma asperellum* (TD-1)	89.67 (71.99)	11.41	8.59	1792.50
2	*Trichoderma harzianum* (TD-14)	100.00 (89.99)	13.73	8.64	2237.00
3	*Trichoderma asperellum* (TD-3)	97.67 (89.82)	9.14	8.55	1727.78
4	*Trichoderma asperelloides* (TD-9)	97.67 (89.82)	11.42	8.49	1944.61
5	Control	70.00 (56.78)	6.44	4.47	826.70
S. Em.±		**0.04**	**0.14**	**0.11**	
C. D. at 1 %		**0.18**	**0.57**	**0.42**	

*Mean of three replications Figure in the parenthesis are arc sine transformed values SVI- Seedling Vigour Index

asperelloides (1944.61) and *T. asperellum* (TD-1) (1792.5). The least SVI was shown by *T. asperellum* (TD-3) (1727.78) whereas control showed SVI of 826.7.

SVI- Seedling Vigour Index

Elucidation of plant growth-promoting activity by efficient *Trichoderma harzianum* strain in rice under pot culture

The observations were taken for the parameters like shoot length, root length and number of tillers at 30, 60 and 90 DAS. The results are presented (Table 4).

Shoot length

The results (Table4) revealed that *T. harzianum* enhanced the shoot length in all the treatments compared to the untreated control. At 30 DAS, the treatment combination containing seed treatment, seedling dip and foliar spray with *T. harzianum* showed significantly highest mean shoot length (28.12 cm) followed by seed treatment, and seedling dip (26.45 cm). The lowest mean shoot length of 22.50 cm was observed in the treatment where only seed treatment was done with *T. harzianum*. The control treatment recorded least mean shoot length of 15.50 cm (Table 4). The treatment combination seed treatment, seedling dip and foliar spray with *T. harzianum* showed significantly highest mean shoot length (43.00 cm) followed by seed treatment and seedling dip with *T. harzianum* (37.50 cm) at 60 DAS. Lowest mean shoot length of 32.70 cm was observed in the treatment where only seed treatment was done with *T. harzianum* when compared to the control treatment, which recorded least mean shoot length of 24.50 cm (Table 4). Similarly, at 90 DAS, the same treatment combination, seed treatment, seedling dip and foliar spray with *T. harzianum* showed significantly highest mean shoot length (51.40 cm) followed by seed treatment, seedling dip and foliar spray with *T. harzianum* (46.30 cm) and Lowest mean shoot length of 43.00 cm was observed in the treatment where only seed treatment was done with *T. harzianum*. The control treatment recorded least mean shoot length of 31.00 cm (Table 4).

Root length

The results (Table 4) revealed that *T. harzianum* enhanced the root length in all the treatments compared to the untreated control. At 30 DAS, seed treatment, seedling dip and foliar spray with *T. harzianum* showed significantly highest mean root length (13.00 cm) followed by seed treatment and seedling dip with *T. harzianum* only (12.48 cm). The lowest mean root length of 12.00 cm was observed in the treatment where only seed treatment was done with *T. harzianum*. The control treatment recorded least mean root length of 9.50 cm (Table 3).At 60 DAS, seed treatment, seedling dip and foliar spray with *T. harzianum* showed significantly highest mean root length (18.50 cm) followed by seed treatment, seedling dip and foliar spray with *T. harzianum* (15.50 cm). Lowest mean root length of 13.50 cm was observed in the treatment where only seed treatment was done with *T. harzianum*. The control treatment recorded least mean root length of 11.00 cm (Table 4). At 90 DAS, the treatment combination containing seed treatment, seedling dip and foliar spray with *T. harzianum* showed significantly highest mean root length (21.20 cm) followed by seed treatment and seedling dip with *T. harzianum* (19.47 cm). Lowest mean root length of 16.50 cm was observed in the treatment where only seed treatment was done with *T. harzianum*. The control treatment recorded least mean root length of 14.00 cm (Table 4).

Number of tillers per plant

The results (Table 4) revealed that *T. harzianum* enhanced the number of tillers in all the treatments compared to the untreated control. At 60 DAS, the treatment combination, seed treatment, seedling dip and foliar spray with *T. harzianum* showed significantly highest number of tillers per plant (13.30) followed by seed treatment and

Table 4. Plant growth promoting activity of efficient *Trichoderma* spp. in rice under pot culture

Sl. No.	Treatment	Mean shoot length (cm)*			Mean root length (cm)*			Number of tillers plant^{-1}		
		Days After Sowing (DAS)								
		30	60	90	30	60	90	30	60	90
1	Seed treatment with *T. harzianum*	22.50	32.70	43.00	13.00	13.50	16.50	-	11.30	13.27
2	Seed treatment+ seedling dip with *T. harzianum*	26.45	37.50	46.30	12.48	15.50	19.47	-	11.30	14.00
3	Seed treatment + seedling dip + spraying *T. harzianum*	28.12	43.00	51.40	13.00	18.50	21.20	-	13.30	16.00
4	Control	15.50	24.50	31.00	9.50	11.00	14.00	-	8.30	11.00
S. Em. ±		**0.67**	**0.58**	**0.68**	**0.61**	**0.76**	**0.65**	**-**	**0.89**	**0.76**
C. D. at 1 %		**2.75**	**2.38**	**2.80**	**2.52**	**3.13**	**2.68**	**-**	**3.66**	**3.14**

*Mean of three replications

seedling dip with *T. harzianum* (11.30). Lowest number of tillers per plant of 10.53 was observed in the treatment where only seed treatment was done with *T. harzianum*. The control treatment recorded least number of tillers per plant of 8.30 (Table 4). At 90 DAS, seed treatment, seedling dip and foliar spray with *T. harzianum* showed significantly highest number of tillers per plant (16.00) followed by seed treatment and seedling dip with *T. harzianum* (14.00). Lowest number of tillers per plant of 13.27 was observed in the treatment where only seed treatment was done with *T. harzianum*. The control treatment recorded least number of tillers per plant of 11.00 (Table 4).

DISCUSSION

The *R. solani* isolate shared typical characteristics of branching at right angle near the distal septum of thecell, formation of a septum in the branch near thepoint of origin, constriction of the branch at origin. The results are in agreement with Sunder *et al.,* (2003), who reported the production of light brown, brown and dark brown mycelium on the PDA media. The discolorations of the growth media is mainly attributed to the production of pigments by the pathogen. Branching of the mycelium was found near the distal septum of a cell in young and advanced hyphae. In older hyphae, branching may occurred at any place along the cell.

Twenty indigenous *Trichoderma* strains were successfully isolated by serial dilution technique on *Trichoderma* specific medium. Sharma and Singh (2014) also isolated thirty isolates of the *Trichoderma* spp. from the rhizosphere soils from districts of Uttarakhand (India) on *Trichoderma* selective medium using serial dilution technique. These *Trichoderma* isolates were maintained in slants as pure cultures and used for further studies.

In the present study, the results indicated that per cent inhibition of my celial growth of *R. solani* varied greatly among the twenty strains. Similarly, Seema and Devaki (2012) evaluated the efficacy of fungal bio agents *viz*., *T. viride* and *T. harzianum* under *in vitro* conditions against *R.solani* and reported that the percentage inhibition of growth by *T. viride, T. harzianum*on *R. solani* was 70 per cent and 67 per cent, respectively.Similar findings were also made by Kumari *et al*. (2016) who also tested the several isolates of *Trichoderma* against *R. solani.* Result indicated that the antagonistic potential of 26 isolates of *Trichoderma spp.* against *R. solani* were varied which inhibited *R. solani* ranges 33-54%. Among isolates of *Trichoderma*, seven isolates showed strong antagonistic potential which inhibited >50% my celial growth of *R. solani*, *viz*., RCT1 (53.71%) followed by RCT22 (52.6%), RCT3 (51.85%), RCT7 (51.11%), RCT10 (50.37%), RCT 8 (50%) and RCT14 (50%). Moreover, seventeen (17) isolates were also showed inhibitory with <50% of the my celial growth while two isolates (RCT12 and RCT17) showed <40% my celial growth. These potential isolates of *Trichoderma* may be further exploited as bio control agent against *R. solani* as well as other soil borne phytopathogenic fungi.

The study showed that species of *Trichoderma* have shown plant growth promoting activity in both roll towel method and pot culture in rice. These results are in support of the findings of Zheng and Shetty (2000) who reported that *Trichoderma* spp. induced phenolic compound production in the plant during seed germination and led to enhancement of seed vigour. The ability of *Trichoderma* spp. in increasing the percent seed germination and plant growth in rice was also reported by Das *et al.,* (1995) and Ravi *et al.,* (1999).Among the fungal antagonists, *Trichoderma* spp. and *Gliocladium* spp. are widely used in the management of rice Sheath blight disease. These fungal antagonists are either applied to rice seed, soil, root dip and foliar spray for managing the disease. In pot culture studies, seed treatment of the bio agent *T. viride* resulted in Sheath blight disease reduction. Further, the efficacy of *T. viride* was comparatively more than the bacterial bio agent *B. subtilis* (Das *et al.,* 1998).

The method of delivery of *Trichoderma* strains also plays an important role in the promotion of plant growth. Highest shoot length, root length and more number of tillers were obtained in that treatment where, it received all the three delivery systems *viz.*, seed treatment, root dipping and foliar application. The results of our present investigation are in accordance with Patil *et al.,* (2003) who concluded that biological control agents were more effective and economical when applied using combination of delivery systems.

CONCLUSION

All twenty *Trichoderma* strains inhibited the my celial growth of *R. Solani* and among them nine strains recorded more than 50% inhibition.The superior strainswereidentified as *T. asperellum*, *T. harzianum, T. asperellum* and *T. asperelloides*. Among these, *T. harzianum* was highly superior in promoting plant growth by recording highest seed germination, shoot length, root length and seedling vigour index. The treatment combination, seed treatment + root dipping + foliar spray with *T. harzianum* suspension was highly effective by recording highest root length, shoot length and number of tillers per plant in pot culture experiment.

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