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Study on the Different Modes of Action of Potential *Trichoderma* spp. from Banana Rhizosphere against *Fusarium oxysporum* f.sp. *cubense*

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A B S T R A C T

An attempt was made to study the different modes of action of the promising *Trichoderma* spp. from banana rhizosphere collected from different regions of Assam, Mizoram, Meghalaya and Nagaland. The results from the present investigation revealed that all the potential *Trichoderma* spp. produced IAA, NH_3, siderophore and HCN, though at different levels however, the promising *Trichoderma* spp. were not able to solubilize phosphate on solid medium containing insoluble inorganic phosphorus source. Considering the possibility of an improved potentiality of combined application, a study was also undertaken to check the effect of combined application of the *Trichoderma* spp. against *Fusarium oxysporum* f.sp. *cubense* (Foc), the causal organism of Fusarium wilt of banana. The per cent inhibition over control was calculated after 48, 72 and 96 hours after inoculation. The result revealed that the efficacy of all the treatments differed significantly with that of control at all the intervals. The per cent inhibition of radial growth of Foc *in vitro* was observed highest by the combination of the three *Trichoderma* spp. *viz. T. reesei* (RMF-25), *T. reesei* (RMF-13) and *T. harzianum* (RMF- 28) with 69.18 per cent followed by the combination of *T. reesei* (RMF-25), and *T. harzianum* (RMF-28) with 66.86 per cent and combination of *T. reesei* (RMF-13) and *T. harzianum* (RMF-28) with 68.60 per cent inhibition of the test pathogen after 96 hours of incubation.

Introduction

Plant diseases are the result of interactions among the components of disease triangle i.e. host, pathogen and environment. The use of biocontrol agents (BCAs) has been proved to be an environmental friendly disease management strategy in recent years (Xue *et al.*, 2015; Deltour *et al.*, 2017; Fu *et al.*, 2017). Biological control of soil borne diseases caused especially by *Fusarium oxysporum* is well documented (Marois *et al*., 1981; Sivan and Chet, 1986; Larkin and Fravel, 1998; Thangavelu *et al*., 2004). Several reports have previously demonstrated the successful use different species of *Trichoderma*, *Pseudomonas*, *Streptomyces*, non pathogenic *Fusarium* (np*Fo*) of both rhizospheric and endophytic in nature against Fusarium wilt disease under both glass house and field

conditions (Lemanceau and Alabouvette, 1991; Alabouvette *et al*., 1993; Larkin and Fravel, 1998; Weller *et al.,* 2002; Sivamani and Gnanamanickam, 1988; Thangavelu *et al.,* 2001; Rajappan *et al.,* 2002; Getha *et al.,* 2005).

Understanding how the bio control agents work can facilitate optimization of control as well as help to screen for more efficient strains of the agent (Junaid, 2013). Understanding the mechanisms of biological control of plant diseases through the interactions between biocontrol agent and pathogen may allow us to manipulate the soil environment to create conditions conducive for successful biocontrol or to improve bio control strategies (Chet, 1987).

The biocontrol activity is exerted either directly through antagonism of soil-borne pathogens or indirectly by eliciting a plant-mediated resistance response (Pozo and Azcón-Aguilar, 2007; Jamalizadeh *et al.,* 2011). Thus, envisaging the potential of rhizospheric microorganisms in plant disease management, the present work has been undertaken to isolate *Trichoderma* spp. from banana rhizosphere and to explore their biocontrol potential against *Fusarium oxysporum* f.sp. *cubense in vitro*.

Materials and Methods

Collection of samples

Rhizospheric soil samples were collected from healthy banana rhizosphere of different banana cultivars from Assam, Mizoram, Meghalaya and Nagaland. For collection of the soil samples, the area around healthy banana plants were dug upto a depth of about 5 -10 cm. The soils were collected close to the root of the banana plant and kept in polyethylene bags until it was brought to the lab for isolation.

Isolation of *Trichoderma* spp.

Microbial culture media *viz.* Potato Dextrose Agar (PDA) medium and *Trichoderma* Specific Medium (TSM) were used for the isolation of *Trichoderma* spp. The *Trichoderma* spp. were isolated following the protocol described by (Thangavelu and Gopi, 2015) where one gram of each of the rhizospheric soil collected from different cultivars of banana were transferred to 250 ml conical flasks containing 100 ml of sterile distilled water. The flasks were placed in rotary shaker for 10 min at 120 rpm to dissolve the soil thoroughly. From this, 1 ml of the supernatant were taken and serially diluted upto 10^{-5} dilutions. One ml of the dilution such as 10^{-3}, 10^{-4}, 10^{-5} was poured at the centre of sterilized Petri plates. Onto such plates specific media for the fungus were poured and rotated clockwise and anticlockwise. Finally the plates were incubated at 28^{0}C for 2 days and observed for emerging colonies. The fungal colonies were purified by single spore isolation technique and maintained in PDA slants.

Indole acetic acid (IAA) production

Assay for indole acetic acid (IAA) production was done following the protocol given by Noori and Saud (2012). Five discs of each of the rhizospheric microbes were transferred into respective universal bottles containing 10 mL of Potato Dextrose Broth (PDB) and incubated on the incubator shaker for 24 h. After 24 h of incubation, 1 mL of fungal inoculum was transferred into 250 mL conical flask containing 100 mL of sterile PDB with 5 mL of 0.2% (w/v) L-tryptophan and incubated at 28±2 °C for 72 h. Conical flask without rhizospheric microbes served as controls or blanks. A 1.5 mL of aliquot was sampled and centrifuged at 3,000 rpm for 30 min, 1 mL of the supernatant was then added with two drops of orthophosphoric acid and 4 mL of

salkowskis reagent (50 mL, 35% perchloric acid; 1 mL 0.5 M ferric chloride, $FeCl_3$). Appearance of red color indicates IAA production. To determine the amount of IAA produced from the isolates, the colour density (absorbance) was measured at 535 nm using spectrophotometer. The IAA produced was compared to the standard graph and expressed as $\mu g\ mL^{-1}$.

NH_3 production

Bakker and Schipper's (1987) protocol was followed to detect the production of NH_3 by the three most effective rhizospheric microbes. Freshly grown rhizospheric microbes were inoculated in culture tubes containing 8-10 ml peptone water broth and incubated at 25-26°C for 48 hours. Nesseler's reagent (1 ml) was added in each tube. The development of colour from yellow to brownish orange was a positive test for ammonia (Bakker and Schipper, 1987).

Hydrogen cyanide (HCN) production

HCN production of the three effective rhizospheric microbes was tested qualitatively following the method of Bakker and Schipper (1987). The rhizospheric microbes were inoculated on petriplates containing Tryptic Soya Agar (TSA) supplemented with 4.4 g L^{-1} of glycine. A Whatman filter paper soaked in alkaline picric acid solution (2.5 g of picric acid; 12.5 g of Na2CO3; 1000 ml of distilled water) was placed in the upper lid of each plate. The plates were incubated at 25±2°C for 7 days. A change in colour of the filter paper from yellow to light brown, brown or reddish brown was recorded as indication of HCN production (Meera and Balabaskar, 2012).

Siderophore production

Chrome Azurol S (CAS) agar method (Schwyn and Neiland, 1987) with a few modification was used to detect the mobilization of iron by the three effective rhizospheric microbes. The rhizospheric microbes were first cultured on PDA plates after which 5mm fungal mats from each isolate were transferred to CAS agar plates and incubated at 25±2 ^oC for seven days. Fe-CAS indicator gave a medium a blue colour. When the iron ligand complex was formed the release of the free dye was accompanied with a color change. Iron mobilization was done via the production of complex acids or siderophores. The Fe (III) gave the agar a rich blue color and concentration of siderophores excreted by iron starved organisms gave a color change to orange. The orange hallow surrounding the colony indicated the excretion of siderophore and its dimension evaluated the amount of siderophore excreted.

Phosphate solubilizing activity

The three best performing rhizospheric microbes were screened qualitatively for inorganic phosphate solubilization as per methodology described by Gupta *et al*., (1994). A 5mm mycelia disc of each isolates were placed on the centre of Pikovskaya agar with insoluble tricalcium phosphate (TCA) and incubated at 25±2 ^oC for 7 days. The experiment was performed on CRD with five replications each. After incubation, the colonies with clear halo zones (solubilizing zone) around colony indicated positive solubilization of mineral phosphate (Noori and Saud, 2012).

In vitro testing of promising *Trichoderma* spp. for their compatibility

The Compatibility studies were carried out to observe whether the selected antagonists were compatible with each other against Foc. Dual culture method described by Dennis and Webster (1971) was employed to observe for the zone of inhibition. The test was carried *in*

vitro with all possible permutations and combinations to study their compatibility with each other.

Effect of promising *Trichoderma* spp. against Foc individually and in combination

Efficacy of the promising antagonists was studied individually and in combinations against Foc *in vitro* based on the compatibility test following Zegeye *et al.,* (2011) with slight modification. The design of the experiment followed was completely randomized design (CRD) with five replications for each treatment (individually or in combination).

Results and Discussion

Identification of *Trichoderma* spp.

All the rhizospheric microbes isolated during the present investigation were tested for their antagonistic activity against Foc by dual culture plate technique. Identification of *Trichoderma* spp. was carried out only for the three best performing rhizospheric microbes by sequencing of 18S rRNA and the results revealed that the first (RMF-25) and the second best (RMF-13) promising rhizospheric microbes were *Trichoderma* reesei while the third best promising rhizospheric microbe (RMF-28) was *T. harzianum*. These three potential *Trichoderma* spp. were then used for testing their different modes of action like production of IAA, NH_3, HCN, Siderophore and Phosphate solubilisation activity.

Indole acetic acid (IAA) production

The results for the production of IAA have been presented in Table 1 and depicted in Plate 2. In the present investigation, it was observed that all *Trichoderma* spp. elucidated positive results for IAA production. Maximum IAA production was observed in *T. reesei* (RMF-25) with 13.38 µg mL^{-1} of IAA followed by *T. harzianum* (RMF-28) and *T. reesei* (RMF-13) with 9.34 6.32 µg mL^{-1} IAA production respectively. IAA has been implicated in virtually every aspect of plant growth and development, as well as defense responses. The result of the present investigation is also supported by the findings of Mohiddin *et al.,* (2017) who isolated *Trichoderma* species from chilli rhizosphere. Their studies revealed that the amount of IAA produced by *Trichoderma* spp. ranged from 1.538 µg mL^{-1} to 6.605 µg mL^{-1}. Similar findings were recorded several workers (Badawi *et al.,* 2011; Aarti and Meenu, 2015) who also reported the amount of IAA produced by *Trichoderma* spp. as in the range obtained in present investigation.

NH_3 production

The results for NH_3 production has been presented in Table 1. All the *Trichoderma* spp. showed positive result for ammonia production by turning initial peptone water broth from yellow to brownish orange (Plate 2). It had also been observed that *T. reesei* (RMF-13) produced more amount of NH_3 while *T. reesei* (RMF-25) and *T. harzianum* (RMF-28) produced mediocre amount of NH_3. Ammonia production by the *Trichoderma* isolates may influence plant growth indirectly which is directly or indirectly useful for plants (Ahemad and Kibret, 2014). The ACC (1-aminocyclopropane-1- carboxylic acid) synthesized in plant tissues by ACC synthase is thought to be exuded from plant roots and be taken up by neighboring micro-organisms. *Trichodrema* may hydrolyze ACC to ammonia (Ahemad and Kibret, 2014). The result of the present investigation is in agreement with reports of several workers (Aarti and Meenu, 2015; Chadha *et al.,* 2015) who reported the production of ammonia by *Trichoderma* spp. Similar findings was also reported by Mohiddin *et al.,* (2017) who reported that out of 20 *Trichoderma* spp., isolated from chilli

rhizosphere, 13 isolates were found to produce ammonia.

Hydrogen cyanide (HCN) production

The results of HCN production (Table 1) revealed that *T. reesei* (RMF-13) and *T. harzianum* (RMF-28) were able to produce HCN as there was a change in colour of filter paper from yellow to reddish brown (Plate 3). It was also observed that *T. harzianum* (RMF-28) produced more amount of HCN as compared to *T. reesei* (RMF-13) which produced mediocre amount. However, HCN production was not observed in *T. reesei* (RMF-25). HCN production is an important trait found in various soil micro-organisms as it indirectly promotes plant growth by controlling some soil borne diseases (Kremer and Souissi, 2001; Siddiqui *et al.,* 2006). This is mainly due to cyanide production by microbes which can acts as a general metabolic inhibitor to avoid predation or competition without harming the host plants (Noori and Saud, 2012.). The result of the present investigation is also supported by Aarti and Meenu (2015), Ng *et al.,* (2015) and Mohiddin *et al.,* (2017) who reported the positive production of HCN by *Trichoderma* spp.

Siderophore production

The results (Table 1 and Plate 4) revealed that *T. reesei* (RMF25) and *T. reesei* (RMF-13) were able to secrete siderophore by the production of yellow halo surrounding the growing *Trichoderma* spp. The observations revealed that *T. reesei* (RMF-25) secretes more amount of HCN as compared to *T. reesei* (RMF-13) which produced mediocre amount however secretion of siderophore production was not observed by *T. harzianum* (RMF 28). Siderophores are low molecular iron chelating compounds produced by fungi and bacteria under iron stress condition (Ghosh *et al.,* 2017**)**. Siderophores are produced for scavenging iron from the environment and have an high affinity for iron (III) (Hider and Kong, 2010). Fe^{3+}-chelating molecules can be beneficial to plants because they can solubilise the iron which is otherwise unavailable and can suppress the growth of pathogenic microorganisms by depriving the pathogens of this necessary micronutrient (Leong, 1986). However siderophore production can vary considerably depending on the strain of *Trichoderma* spp (Anke *et al.,* 1991). This is in conformity with the result of the present finding as secretion of siderophore was not observed in *T. harzianum* (RMF 28). Gosh *et al.,* (2017) and Vinale *et al.,* (2013) also revealed that antagonistic spp. of *Trichoderma* namely *T. viride, T. harzianum, T. longibrachiatum* and *T. asperellum* produced considerable amount of siderophore

Phosphate solubilizing activity

The result of the qualitative estimation of phosphate solubilisation for all the *Trichoderma* spp. did not show any clear zone on Pikovskaya's Agar after incubation at room temperature for 0-7 days (Table 1 and Plate 5). The finding of the present investigation was in contrast with El-Katatny (2004), who reported that *Trichoderma* isolates are relatively good in P-solubilization. Phosphate solubilization of *Trichoderma* species is one of the mechanisms of these fungi as the plant growth promoting fungi. However, the ability of *Trichoderma* species depends on the kind and strain of *Trichoderma* and source of phosphate (Kapri and Tewari, 2010; Promwee, 2011). Our finding was also supported by many workers (Rawat and Tewari, 2011; Promwee *et al.,* 2014; Ng *et al.,* 2015) who reported that even though *Trichoderma* species revealed good mycelia growth, there was no formation of halo-zone on the solid medium containing insoluble inorganic phosphorus source. In addition, Nautiyal (1999) reported that the

criterion for isolation of phosphate solubilizers based on the formation of a visible halo-zone on Pikovskaya's agar is not a reliable technique because many isolates of Phosphate Solubilizing Microorganisms (PSM), which did not show any clear zone on agar plates, could be able to solubilize insoluble inorganic phosphates in liquid medium.

In vitro testing of effective rhizospheric microbes for their compatibility

Considering the possibility of an improved potentiality of combined application of the three best performing rhizospheric microbes, a study was undertaken to record the combined effect of the rhizospheric microbes in comparison to single application. The experiment was carried out in all permutations and combination amongst the rhizospheric microbes. The result of the experiment revealed that all the *Trichoderma* spp. were found to be compatible with each other in all combinations without inhibiting each other (Plate 6). Such reports of positive compatibility amongst the rhizospheric microbes have been reported by many researchers (Dandurand and Knudsen 1993; Duffy *et al.*, 1996; Raupach and Kloepper, 1998). Further, since they are of one fungus their compatibility is justified (Thangavelu and Gopi, 2015a; Baruah *et al.*, 2018).

Effect of promising rhizospheric microbes against Foc individually and in combination

The effect of the three promising *Trichoderma* spp. were further studied to observe their efficacy in reducing the growth of Foc individually as well as in combinations. The result revealed that the efficacy of all the treatments differed significantly with that of control at all the intervals. The per cent inhibition over control was calculated after 48, 72 and 96 hours after inoculation. The results for the combined effect of *Trichoderma* spp. against Foc have been presented in Table 2 and depicted in Plate 7. After 96 hours of incubation, the per cent inhibition of radial growth of Foc *in vitro* was observed highest by the combination of the three *Trichoderma* spp. *viz*. *T. reesei* (RMF-25), *T. reesei* (RMF-13) and *T. harzianum* (RMF- 28) with 69.18 per cent followed by the combination of *T. reesei* (RMF-25), and *T. harzianum* (RMF 28) with 66.86 per cent and combination of *T. reesei* (RMF-13) and *T. harzianum* (RMF 28) with 68.60 per cent inhibition of the test pathogen.

Table.1 Production of IAA, NH_3, HCN, Siderophore and Phosphate solubilisation activity by isolated *Trichoderma* spp

Sl. No.	*Trichoderma* spp.	IAA Production (µg mL^{-1})	NH3 Production	HCN Production	Siderophore Production	Phosphate Solubilization
1.	*T. reesei* (RMF-25)	13.38	+	-	++	-
2.	*T. reesei* (RMF-13)	6.32	++	+	+	-
3.	*T. harzianum* (RMF-28)	9.34	+	++	-	-
+	indicates mediocre amount of production					
++	indicates more amount of production					
-	indicates no production					

Table.2 Effect of *Trichoderma* spp. individually and in combination on the growth and per cent inhibition of Foc

Sl. No.	Combinations	Growth of Foc (cm)	Per cent inhibition of Foc	Growth of Foc (cm)	Per cent inhibition of Foc	Growth of Foc (cm)	Per cent inhibition of Foc
		48 hrs		72 hrs		96hrs	
1.	*T. reesei* (RMF25)	1.12	46.66	1.14	55.81	1.16	66.27
2.	*T. reesei* (RMF13)	1.14	45.71	1.18	54.26	1.2	65.12
3.	*T. harzianum* (RMF 28)	1.16	44.76	1.16	55.03	1.18	65.69
4.	*T. reesei* (RMF25) + *T. reesei* (RMF13)	1.06	49.52	1.08	58.13	1.10	68.02
5.	*T. reesei* (RMF25) + *T. harzianum* (RMF 28)	1.1	47.62	1.12	56.58	1.14	66.86
6.	*T. reesei* (RMF-13) + *T. harzianum* (RMF 28)	1.04	50.47	1.06	58.91	1.08	68.60
7.	*T. reesei* (RMF25) + *T. reesei* (RMF13)+ *T. harzianum* (RMF 28)	1.02	51.42	1.04	59.68	1.06	69.18
8.	Control	2.1	0.00	2.58	0.00	3.44	0.00
	SEd±	0.06		0.04		0.03	
	CD (p=0.05)	0.12		0.09		0.07	

Plate.1 Indole Acetic Acid (IAA) production test by promising *Trichoderma* spp.

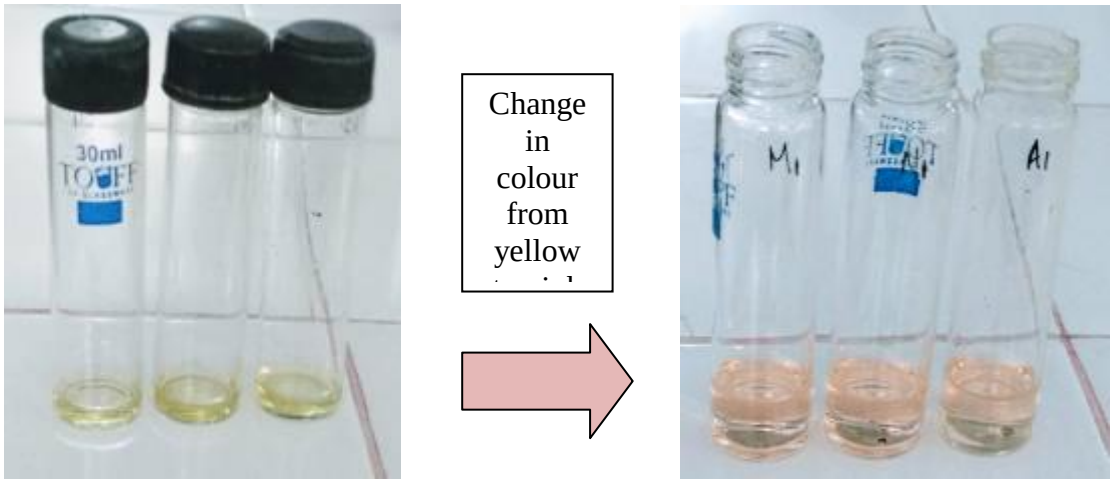


Plate.2 NH_3 production test by promising *Trichoderma* spp.

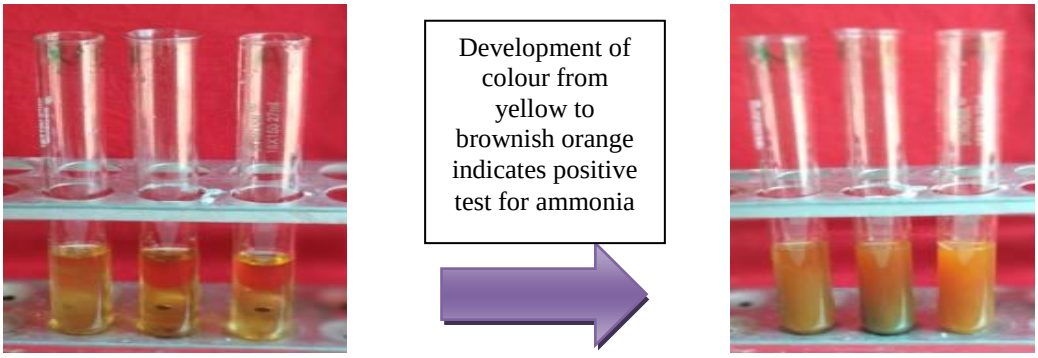


Plate.3 HCN production test by promising *Trichoderma* spp.
A) *T. reesei* (RMF-25), B) *T. reesei* (RMF-13), C) *T. harzianum* (RMF-28)

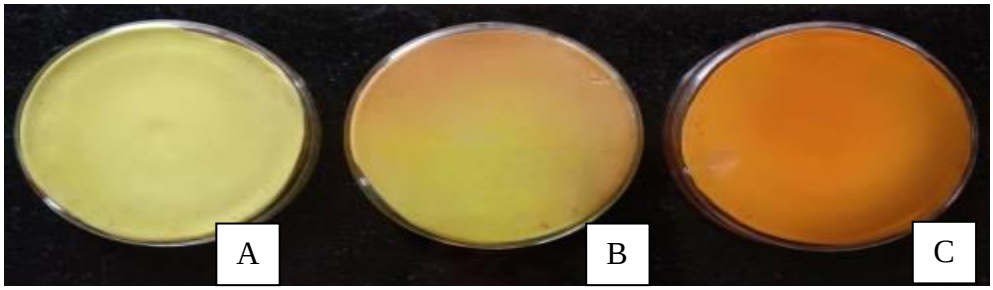
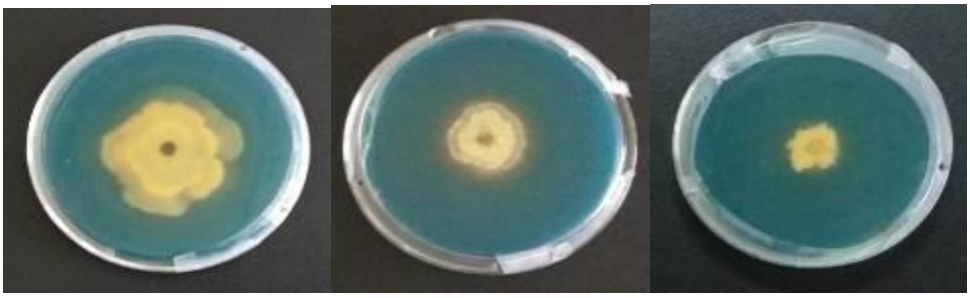
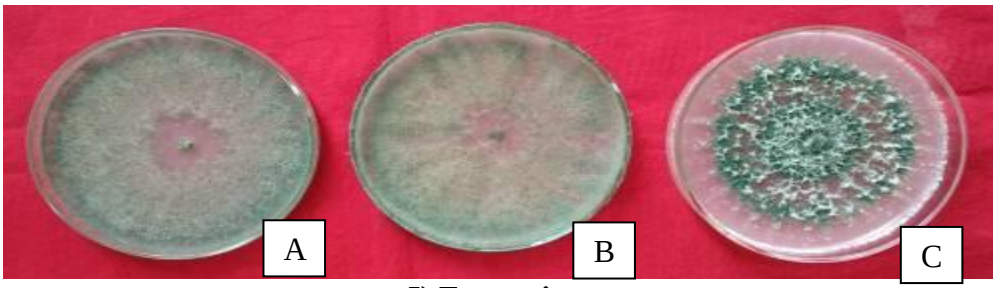


Plate.4 Siderophore production test by promising *Trichoderma* spp.
A) *T. reesei* (RMF-25), B) *T. reesei* (RMF-13), C) *T. harzianum* (RMF-28)



A) **B)** **C)**

Plate.5 Phosphate solubilisation test by promising *Trichoderma* spp.
A) *T. reesei* (RMF-25), B) *T. reesei* (RMF-13), C) *T. harzianum* (RMF-28)



I) Front view

Plate.6 In vitro testing of promising *Trichoderma* spp. for their compatibility A) *T. harzianum* (RMF-28) + *T. reesei* (RMF-13) + *T. reesei* (RMF-25) B) *T. reesei* (RMF25) + *T. reesei* (RMF13) C: *T. reesei* (RMF13) + *T. harzianum* (RMF 28) D: *T. reesei* (RMF25) + *T. harzianum* (RMF 28)

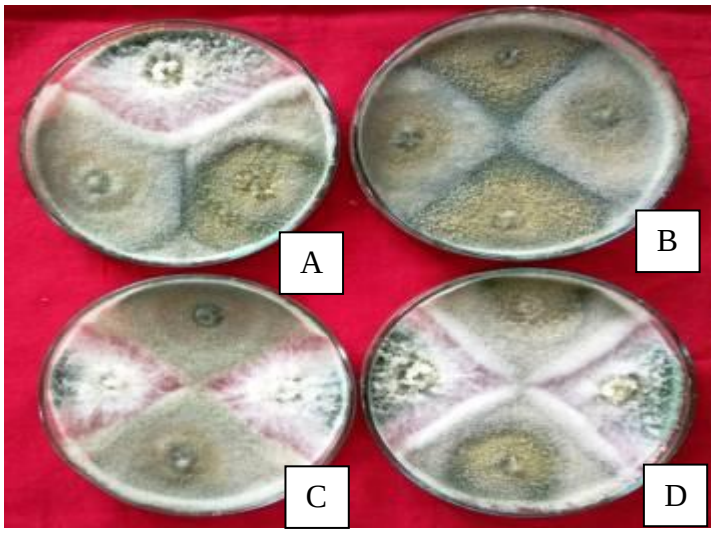
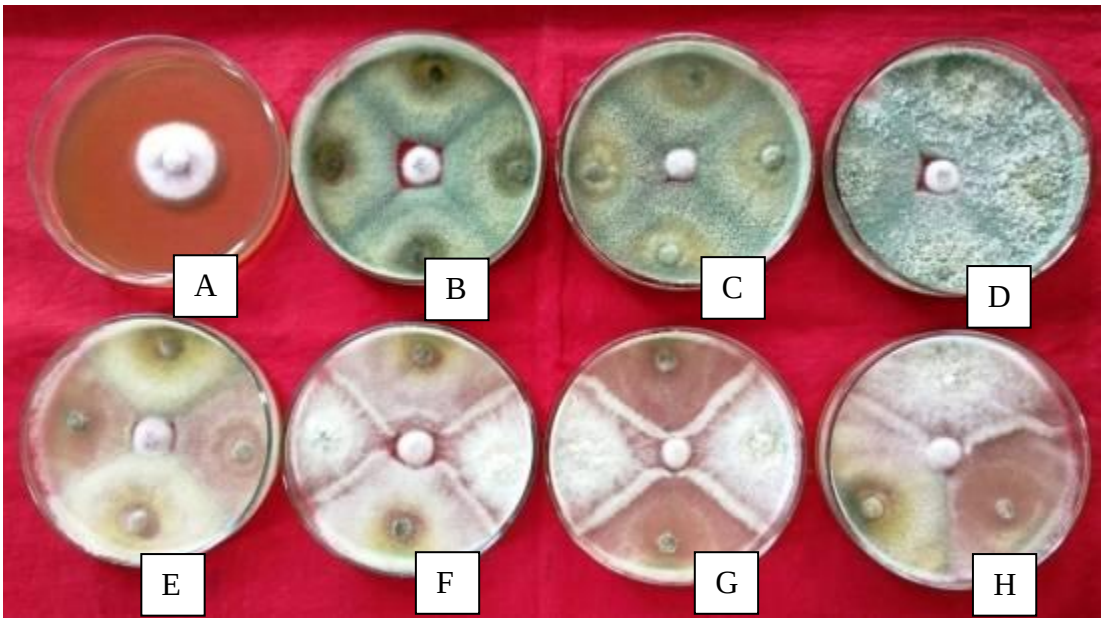


Plate.7 Effect of promising *Trichoderma* spp. individually and in combination against Foc A) Control B) *T. reesei* (RMF-25) alone C) *T. reesei* (RMF-13) alone D) *T. harzianum* (RMF-28) alone E) *T. reesei* (RMF-25) + *T. reesei* (RMF-13) F) *T. reesei* (RMF-25)+ *T. harzianum* (RMF-28), G) *T. reesei* (RMF-13) + *T. harzianum* (RMF-28) H)*T. reesei* (RMF-25) + *T. harzianum* (RMF-28) + *T. reesei* (RMF-13)



The percent inhibition recorded by the rest of the rhizospheric microbes either singly or in combination ranged from 65.12 per cent in case of *T. reesei* (RMF-13) alone to 68.02 per cent in case of combination of *T. reesei* (RMF-25) and *T. reesei* (RMF13).

It had been reported that combined application of biocontrol agents is more effective over a single biocontrol agent in the management of several plant diseases (Crump, 1998; Pierson and Weller, 1994). Similar finding was reported by Akrami *et al.*, (2011) who reported that *T. harzianum* and *T. asperellum* isolates and their combination reduced Fusarium rot disease severity from 20 to 44 per cent and increased the dry weight from 23 to 52 per cent in lentil under glasshouse conditions. Thangavelu and Gopi (2015a) reported that the rhizospheric and endophytic *Trichoderma* isolates, which recorded effective control against Foc pathogen were compatible with each other under *in vitro* condition. Otadoh Sobre *et al.*, (2011) also evaluated Isolates of *Trichoderma* from Embu soils for their ability to control *Fusarium oxysporum* f. sp. *phaseoli., in vitro* They found that *Trichoderma* solates significantly reduced the mycelial growth of the pathogen where combination of *T. reesel and T. koningii* were most effective. Since the data obtained from the present investigation also indicates significant reduction in the growth of Foc, thus it corroborates with the findings of the earlier workers.

References

Aarti, T., and Meenu, S. 2015. Role of volatile metabolites from *Trichoderma citrinoviride* in biocontrol of phytopathogens. *Int. J Res Chem Environ*. 5: 86-95.

Ahemad, M., and Kibret, M. 2014. Mechanisms and Applications of Plant Growth Promoting Rhizobacteria: Current Perspective. *Journal of King Saud University-Science.* 26: 1-20.

Akrami, M., Golzary, H., and Ahmadzadeh, M. 2011. Evaluation of different combinations of *Trichoderma* species for controlling Fusarium rot of lentil. *African. J. Biotechnol*. 10: 2653-2658.

Alabouvette, C., Lemanceau, P., and Steinberg, C. 1993. Recent advances in the biological control of *Fusarium* wilts. *Pesticides Science*. 37: 365–373.

Anke, H., KinnKarl-Erik, J., and Sterner, B. O. 1991. Production of siderophores by strains of the genus *Trichoderma.* Biol. Metals. 4:176-180.

Bakker, A.W., and Schippers, B. 1987. Microbial cyanide production in the rhizosphere in relation to potato yield reduction and Pseudomonas spp.-mediated plant growth-stimulation. *Soil Biology and Biochemistry*. 19: 451–457.

Baruah, N., Bhattacharyya, A., Thangavelu, R., and Puzari, K. C. 2018. *In vitro* Screening of Native Banana Rhizospheric Microbes and Endophytes of Assam against *Fusarium oxysporum* f. sp. *cubense*. *Int. J. Curr. Microbiol. App. Sci.* 7(6):1575-1583.

Chadha, N., Prasad, R., and Varma, A. 2015. Plant promoting activities of fungal endophytes associated with tomato roots from central Himalya, India and their interaction with *Piriformospora indica*. *International Journal of Pharma and Bio Sciences*. 6: 333-343.

Chet, I. 1987. *Trichoderrna* - application, mode of action, and potential as a biocontrol agent of soilborne plant pathogens. In Innovative Approaches to Plant Disease Control. Pp. 137-160. Edited by I. Chet. New York: John Wiley.

Dandurand, L. M. and Knudsen. 1993. Influence of *Pseudomonas fluorescence* on hyphal growth and biocontrol activity of *Trichoderma harzianum* in the spermosphere and rhizosphere of pea. *Phytopathol*. 83: 265-270.

Deltour, P., França, S. C., Pereira, O. L., Cardoso, I., Höfte, M. 2017. Disease suppressiveness to Fusarium wilt of banana in an agroforestry system: Influence of soil characteristics and

plant community. *Agric Ecosyst Environ.* 239: 173-181.

Dennis, C., and Webster, J. 1971. Antagonistic properties of species groups of *Trichoderma*. II. Production of volatile antibiotics. *Transact. Brit. Mycol. Soc.* 57: 41-48.

Duffy, B. K., Simon, A., and Weller, D. M. 1996. Combinations of *Trichoderma koningii* and fluorescent pseudomonad for control of take-all of wheat. *Phytopathol.* 86: 188-194.

El-Katatny, M. S. 2004. Inorganic phosphate solubilisation by free or immobilized *Trichoderma harzianum* cells in comparison with some other soil fungi. *Egyptian J. Biotechnol*. 17: 1338-1353.

Fu, L., Penton, C. Y., Ruan, Y. Z., Shen, Z. Z., and Shen, Q. R. 2017. Inducing the rhizosphere microbiome by biofertilizer application to suppress banana Fusarium wilt disease. *Soil Biol Biochem*. 104: 39-48.

Getha, K., Vikineswary, S., Wong, W., Seki, T., Ward, A. and Goodfellow, M. 2005. Evaluation of *Streptomyces* sp. strain G10 for suppression of Fusarium wilt and rhizosphere colonization in pot grown banana plantlets. *Journal of Industrial Microbiology and Biotechnology*. 32: 24-32.

Ghosh, S. K., Banerjee, S., and Sengupta, C. 2017. Bioassay, characterization and estimation of siderophores from some important antagonistic fungi. *JBiopest*. 10(2): 105-112.

Gupta, R. R., Singal, R., Shanker, A., Kuhad, R. C., Saxena, R. K. 1994. A modified plate assay for screening phosphate solubilizing microorganisms. *J. Gen. Appl. Microbiol*. 40: 255–260.

Jamalizadeh, M., Etebarian, H. R., Aminian, H., and Alizadeh, A. 2011. A review of mechanisms of action of biological control organisms against post-harvest fruit spoilage. *EPPO Bulletin*. https://doi.org/10.1111 /j.1365-2338.2011.02438.x

Junaid, J. M., Dar, N. A., Bhat, T. A., Bhat, A. H., and Bhat, M. A. 2013. Commercial Biocontrol Agents and Their Mechanism of Action in the Management of Plant Pathogens. *International Journal of Modern Plant & Animal Sciences.* 1(2): 39-57.

Kapri, A., and Tewari, L. 2010. Phosphate solubilization potential and phosphatase activity of rhizospheric *Trichoderma spp. Brazilian Journal of Microbiology.* 41(3): 787-795.

Kremer, R. J., and Souissi, T. 2001 Cyanide production by rhizobacteria and potential for suppression of weed seedling growth. *Current Microbiol*. 43: 182-186.

Larkin, R. and Fravel, D. 1998. Efficacy of various fungal and bacterial biocontrol organisms for the control of Fusarium wilt of tomato. *Plant Disease*. 82: 1022-1028.

Larkin, R. and Fravel, D. 1998. Efficacy of various fungal and bacterial biocontrol organisms for the control of Fusarium wilt of tomato. *Plant Disease*. 82: 1022-1028.

Lemanceau, P., and Alabouvette, C. 1991. Biological control of *Fusarium* diseases by fluorescent *Pseudomonas* and non-pathogenic *Fusarium*. *Crop Protection*. 10: 279-286.

Marois, J. J., Mitchel, D. J., and Somada, R. M. 1981. Biological control of Fusarium crown and root rot of tomato under field condition. *Phytopathology*. 12: 1257-1260.

Meera, T., and Balabaskar, P. 2012. Isolation and characterization of *Pseudomonas fluorescens* from rice fields. *International Journal of Food, Agriculture and Veterinary Sciences.* 2 (1):113-120.

Mohiddin, F. A., Bashir, I., Padder, S. A., and

Hamid, B. 2017. Evaluation of different substrates for mass multiplication of *Trichoderma* species. *Journal of Pharmacognosy and Phytochemistry.* 6(6): 563-569.

Nautiyal, C. S. 1999. An efficient microbiological growth medium for screening phosphate solubilising microorganisms. *FEMS Microbiology Letters,* 170: 265-270. http://dx.doi.org/ 10.1111/j.1574-6968.1999.tb13383.x

Ng, L. C., Ngadin, A., Azhari, M., and Zahari, N. A. 2015. Potential of *Trichoderma* spp. as Biological Control Agents Against Bakanae Pathogen (*Fusarium fujikuroi*) in Rice. *Asian Journal of Plant Pathology.* 9: 46-58.

Noori, M. S. S., and Saud, H. M. 2012. Potential plant growth-promoting activity of *Pseudomonas* sp. isolated from paddy soil in Malaysia as biocontrol agent. *J. Plant Pathol. Microbiol*. 3: 10.4172/2157-7471.1000120

Otadoh, J. A., Sheila, A., Okoth, Ochanda, J., James, P., and Kahindi. 2011. Assessment of *Trichoderma* isolates for virulence efficacy on *Fusarium oxysporum* f. sp. *phaseoli*. *Trop. Subtrop. Agroecosys*. 13: 99-107.

Pozo. M. J., and Azcón-Aguilar, C. 2007. Unraveling mycorrhiza-induced resistance. *Curr Opin Plant Biol.* 10: 393-398.

Promwee, A. 2011. Role of *Trichoderma* spp. as phosphate solubilizing microorganism. *Thai Journal of Soils and Fertilizers.* 33(1): 17-30.

Promwee, A., Issarakraisila, M., Intana, W., Chamswarng, C., and Yenjit, P. 2014. Phosphate Solubilization and Growth Promotion of Rubber Tree (*Hevea brasiliensis* Muell. Arg.) by *Trichoderma* Strains.

Rajappan, K., Vidhyasekaran, P., Sethuraman, K., and Baskaran, T. L. 2002. Development of powder and capsule formulations of *Pseudomonas fluorescens* strain Pf-1 for the control of banana wilt. *Zeitschrift für Pflanzenkrankheiten und Pflanzenschutz.* 109: 80–87.

Raupach, G. S., and Kloepper, J. W. 1998. Mixtures of plant growth-promoting rhizobacteria enhance biological control of multiple cucumber pathogens. *Phytopathol.* 88: 1158-1164.

Rawat, R., and Tewari, L. 2011. Effect of abiotic stress on phosphate solubilization by biocontrol fungus *Trichoderma* sp. *Current Microbiology*. 62(5): 1521-1526. http://dx.doi.org/10. 1007/s00284-011-9888-2.

Schwyn, B., and Neilands, J. B. 1987. Universal chemical assay for the detection and determination of siderophores. *Analytical biochemistry*. 160: 47-56.

Siddiqui, I. A., Shaukat, S. S., Sheikh, I. H., and Khan, A. 2006. Role of cyanide production by *Pseudomonas fluorescens* CHAO in the suppression of root-knot nematode, M*eloidogyne javanica* in tomato. *World J Microbiol Biotechnol*, 22: 641-650.

Sivamani, E., and Gnanamanickam, S. S. 1988. Biological control of *Fusarium oxysporum* f.sp. *cubense* in banana by inoculation with *Pseudomonas fluorescens*. *Plant Soil*. 107: 3 9.

Sivan, A., and Chet, I. 1986. Biological control of *Fusarium* spp. in cotton, wheat and muskmelon by *Trichoderma harzianum*. *J. Phytopathol*. 116: 39–47.

Thangavelu, R., and Gopi, M. 2015. Combined application of native *Trichoderma* isolates possessing multiple functions for the control of Fusarium wilt disease in banana cv. Grand Naine, *Biocontrol Science andTechnology*. 25(10): 1147-1164.

Thangavelu, R., and Gopi, M. 2015a. Field

suppression of Fusarium wilt disease in banana by the combined application of native endophytic and rhizospheric bacterial isolates possessing multiple functions. *Phytopathol. Medit*. 54: 241-252.

Thangavelu, R., Palaniswami, A., and Velazhahan, R. 2004. Mass production of *Trichoderma harzianum* for managing *Fusarium* wilt of banana. *Agriculture, Ecosytems and Environment.* 103: 259–263.

Thangavelu, R., Palaniswami, A., Ramakrishnan, G., Sabitha, D., Muthukrishnan, S., and Velazhahan, R. 2001. Involvement of Fusaric acid detoxification by *Pseudomonas fluorescens* strain Pf10 in the biological control of Fusarium wilt of banana caused by *Fusarium oxysporum* f.sp. *cubense*. *Journal of Plant Disease and Protection*. 108: 433-445.

Vinale, F., Nigro, M., Sivasithamparam, K., Flematti, G., Ghisalberti, E. L., Ruocco, M., Varlese, R., Marra, R., Lanzuise, S., Eid, A., Woo, S. L,. and Lorito, M. 2013. Harzianic acid: a novel siderophore from *Trichoderma harzianum*. *FEMS Microbiol Lett*. 347: 123–129.

Weller, D. M., Raaijmakers, J. M., McSpadden Gardener, B. B., and Thomashow, L. S. 2002. Microbial populations responsible for specific soil suppressiveness to plant pathogens. *Annual Review of Phytopathology*. 40: 309–48.

Xue, C., Penton, C. R., Shen, Z., Zhang, R., Huang, Q., Li, R., Ruan, Y., Shen, Q.R. 2015. Manipulating the banana rhizosphere microbiome for biological control of Panama disease. *Sci Reports*. 5: 11124.

Zegeye, E. D., Santhanam, A., Gorfu, D., Tessera, M., and Kassa, B. 2011. Biocontrol activity of *Trichoderma viride* and *Pseudomonas fluorescens* against *Phytophthora infestans* under greenhouse conditions. *J. Agril. Technol.* 7: 1589-1602.

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