Advances in Biology & Earth Sciences
Vol.4, No.2, 2019, pp.93-106

EFFECT OF BIO-FORTIFIED COMPOST IN CONTROLLING SOIL-BORNE DISEASES OF LENTIL (*Lens culinaris* L.) AND ENHANCE THE CROP GROWTH AND YIELD

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Abstract. Poultry refuge mixed with bio-agent i.e *Trichoderma harzianum* isolate Pb-7 is compatible and has combined effect in controlling the pathogenic fungi viz. *Rhizoctonia solani* isolate Rs-3, *Fusarium oxysporum* f. sp. *lentis* isolate Fo-3 and *Sclerotium rolfsii* isolate Sr-3 which cause the foot and root rot, *Fusarium* wilt and collar rot disease of lentil variety BARI masur-6. Post-emergence mortality of plants due to damping off, foot and root rot, collar rot and *Fusarium* wilt disease were significantly reduced after the field treatment with *T. harzianum* isolate Pb-7 fortified compost. *T. harzianum* isolate Pb-7 spore suspension @ 8×10^8 ml^{-1} were used as seed treating biocontrol agents and application of poultry refuge in the soil as an organic source of nutrients. As a result shoot and root lengths, fresh and dry weight of shoot were increased significantly. The lowest disease incidence (DI) of foot and root rot, collar rot and *Fusarium* wilt were recorded in T_8 where field treated with *T. harzianum* isolate Pb-7 fortified compost. On the contrary, the highest disease incidence was observed in the control-2 where field treated with the test pathogens such as *R. solani* isolate Rs-3, *F. oxysporum* f. sp. *lentis* isolate Fo-3 and *S. rolfsii* isolate Sr-3 while minimum disease incidence was found in control-1 due to uninoculation. The highest percentage of foot and root rot, collar rot, *Fusarium* wilt disease over control-2 were recorded in the treatment T_8 followed by T_3, T_4 and T_6, respectively. The highest shoot length (33.87 cm), root length (13.60 cm), fresh weight (34.23 g), dry weight (21.57 g), 1000 seed weight (35.74 g) and yield (2.52 t ha^{-1}) were also recorded in the treatment T_8, where inoculated field treated with *T. harzianum* isolate Pb-7 fortified composted poultry refuge.

Keywords: *Bio-fortified compost, soil-borne fungal pathogens, yield of Lentil.*

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Received*: 28 May 2019;* ***Accepted****: 08 July 2019;* ***Published****: 10 August 2019.*

1. Introduction

Lentil (*Lentis culinaris* L.) is the 2nd major pulse crop of Bangladesh in terms of both area and production. In 2018, total production of lentil in Bangladesh is about 168837 metric tons (Anon., 2018). There are various reasons associated with low yield of lentil in Bangladesh, where diseases are considered as the major constraints. Various diseases may cause 30-40% yield loss in lentil (Begum & Bhuiyan, 2006). Among the various diseases, the soil-borne disease is very critical in realizing the yield potential of improved cultivars in several agricultural crops. Habitually, these diseases are very tough to manage because of their highly heterogeneous incidence and lack of

information about epidemiological aspects of soil-borne fungi. The legume crop such as lentil suffers from a number of diseases which are caused by many fungi, bacteria, viruses, nematodes and plant parasites as a result decline the grain yield abruptly. The foot and rot root disease caused by *Rhizoctonia solani*, collar rot caused by *Sclerotium rolfsii*, *Fusarium* wilt caused by *Fusarium oxysporum* f. sp. *lentis* and damping off or seedling mortality caused by all those of pathogens are the main limiting factors to successful cultivation. The biological management of soil-borne disease is increasingly gaining stature as a possible practical and safe approach. The potential antagonistic micro-organisms helps to reduce the intensity of crop damage by the soil-borne plant pathogens (Kashem *et al.*, 2011). In running era, there has been a worldwide swing to the use of eco-friendly and sustainable methods for protecting the crops from pests and diseases. *Trichoderma* spp. were found to be effective against different sclerotia forming fungi including *F. solani, R. solani, S. rolfsii* (Elad *et al.*, 1980). Control of the soil-borne pathogens *F. oxysporum, R. solani,* and *S. rolfsii* with chemicals is practically difficult. On the other hand, indiscriminate use of chemicals cause environmental pollution and health hazards (Gerhardson, 2002). Now a days, Integrated Disease Management (IDM) is very much popular for controlling plant diseases. There are several tactics within IDM, among them biological control being one of the most important tactics (Kwok *et al.*, 1987).

Trichoderma spp. are found in almost any tropical and temperate soil. The suppression of disease by *Trichoderma* is based on hyper-parasitism, antibiosis, induced resistance in the host plant and competition for nutrients and space (Harman *et al.*, 2004). The majority of *Trichoderma* spp. are antagonist of plant pathogenic fungi and have been broadly used as the most important biocontrol agent (Tjamos *et al*., 1992). A *Trichoderma* based compost fertilizer to be prepared by mixing a definite concentration of spore suspension of *T. harzianum* strain with measured amounts of processed raw materials such as poultry refuse, water hyacinth, tea waste etc. Nahar *et al.,* 2012 found that combined application of *Tricho*-compost and *Tricho*-leachate reduced the seedling mortalities by 40.9 to 64.5% in Gazipur and 53.3 to 62.1% in Bogra. *Trichoderma*-fortified compost was very effective in reducing different diseases of tomato and also significantly increased yield of tomato (Rahman, 2013).

Effects of the *Trichoderma* spp. in promoting seedling establishment, enhancement of plant growth and elicitation of plant defense reaction in some vegetable crops have been examined by a number of researchers (Rabeerdran *et al.*, 2000; Hoyos- Carvajal *et al.*, 2009). However, due to lack of suitable carriers for large scale production, several organic substances were tested and poultry refuse was found to be a suitable medium for growing *T. harzianum*. Additionally, combined application of poultry refuse and *T. harzianum* was found to be highly effective to control soil-borne diseases in seedbed nurseries. A few works have been done on the management of plant disease with bio control microorganism in Bangladesh. Yet information on biological control of foot and root rot, collar rot and *Fusarium* wilt of lentil is inadequate under Bangladesh condition. Therefore, the present study was undertaken to control major soil-borne fungal diseases of lentil by using *T. harzianum* fortified compost and increase the yield of lentil.

2. Materials and Methods

Collection, isolation and preservation of test pathogens

Each of the three isolates of *F. oxysporum* f. sp. *lentis* designated as Fo-1, Fo-2 and Fo-3, *R. solani* designated as Rs-1, Rs-2 and Rs-3 and *S. rolfsii* designated as Sr-1, Sr -2 and Sr-3 were isolated from the rhizosphere and rhizoplane of lentil (*Lens culinaris* L.), potato (*Solanumtuberosum*), chilli (*Capsicum frutesence*), Bush bean (*Phaseolus vulgaris* L.) and Soybean (*Glycine max* L.). The specimens which had typical symptoms of *Fusarium* wilt, foot and root rot and collar rot were selected from infected fields. The fungal isolates were isolated following standard method (Mian, 1995). The fungal colonies were grown on PDA and identified by following standard key (Barnet & Hunter, 1998). The pure culture of *F. oxysporum* f. sp.*lentis*, *R. solani* and *S. rolfsii*were preserved separately by using PDA slants at 10°C in refrigerator as stock culture for further use.

Inoculum preparation of test pathogens

Inocula of the test pathogens were prepared individually with autoclaved moist wheat grains by following standard method (Rubayet & Bhuiyan, 2016). After preparation, the inoculum was stored at 4°C for future use.

Pathogenicity test

The pathogenicity of *F. oxysporum* f. sp. *lentis*, *R. solani* and *S. rolfsii* isolates were done separately by following standard soil infestation method (Rubayet *et al*., 2017) in pot culture under the shade condition. The number of treatment and replication for each pathogen was three and design of experiment was Completely Randomized Design (CRD). Five seeds were sown in each individual pot. Disease development was observed regularly and were recorded at 15 to 30 days after sowing to pre- and post-emergence seedling mortality. The causal agents of pre-emergence seedling mortality were confirmed after re-isolation of the pathogen from ungerminated seeds.

Collection, isolation and preservation of T. harzianum isolates

A total of 20 isolates of *T. harzianum* were isolated from the rhizosphere and rhizoplane of lentil, rice, jute potato, brinjal, tomato, carrot, bush bean, chilli, maize and pea field following the soil dilution plate technique (Mian, 1995). All the isolated *Trichoderma* spp. were identified as *T. harzianum* based on the different characteristics like colony color, hyphal growth and spore formation (Shaiesta*et al.*, 2012.). The pure culture of *T. harzianum* was preserved by using PDA slants at 10°C in refrigerator as stock culture for future use.

Screening of T. harzianum isolates against virulent isolate of test pathogens

The isolates Fo-3, Sr-3 and Rs-3 were found as the highly virulent isolates of *F. oxysporum* f. sp. *lentis*, *S. rolfsii* and *R. solani*, respectively against lentil. Screening was conducted to evaluate the antagonistic effect of 20 isolates of *T. harzianum* against *F. oxysporum* f. sp. *lentis* isolate Fo-3, *R. solani* isolate Rs-3and *S. rolfsii* isolate Sr-3 on potato dextrose agar (PDA) medium by dual culture technique (Dhingra & Sinclair, 1985). One discs of *T. harzianum* was placed at the edge of each PDA plate and one disc of *F. oxysporum* f. sp. *lentis*, *R. solani* and *S. rolfsii w*ere placed at another edge separately. All plates were incubated in the dark for 25℃ until the mycelium of *F.*

oxysporum f. sp. *lentis, R. solani* and *S. rolfsii* covered the control plate fully. The isolates of *T. harzianum* after come in contact with the test pathogen overgrew and lysed the *F. oxysporum* f. sp. *lentis, R. solani* and *S. rolfsii* mycelium. A distinct inhibition zone was observed of which arms and height were measured. After 7 days of incubation inhibition percentage of the radial growth of *F. oxysporum* f. sp. *lentis, R. solani* and *S. rolfsii* were calculated by following the formula (Sundar *et al.*, 1995).

$$\% \text{ Inhibition of growth} = \frac{X - Y}{X} \times 100$$

where,

X = Mycelial growth of pathogen in absence of *T. harzianum*

Y= Mycelial growth of pathogen in presence of *T. harzianum*

Preparation of wheat grain colonized T. harzianum isolate Pb-7

The highly antagonist isolate of *T. harzianum* Pb-7 was selected on the basis of screening test. Then, inoculum of the *T. harzianum* isolate Pb-7 was prepared with autoclaved moist wheat grains by following standard method (Rubayet & Bhuiyan, 2016). After preparation, the inoculum was stored at 4°C for future use.

Spore suspension preparation of T. harzianum isolate Pb-7

Mycelial disc 5 mm diameter of *T. harzianum* isolate Pb-7 strains were obtained from 7 days-old culture and transferred to 50 ml PDA in a 250 ml conical flask separately incubated at 28ºC for 7 days. At the end of the incubation period 30 ml of sterile distilled water was added to each culture flask and the flasks were shaken at 50 rpm for 30 min in an orbital shaker. Then the content of each conical flask was filtrate through sterile muslin cloth. The filtrate with the spores was collected and a concentration of spore suspension was adjusted to $8\times10^{8}ml^{-1}$ by use of a haemocymeter under a light microscope.

Preparation of bio-fortified compost

The poultry refuge extract was prepared from 5 months old according to the method described by Brinton (1995). Compost and tap water were mixed in the ratio of 1:5 (Khalequzzaman, 2016) in polyethylene non-degradable containers with covers. The mixtures were supplied with aeration using aquarium pumps and were stirred once daily manually using a wooden rod and left at ambient temperature. Subsequently, the mixture was filtered through double-layered cheese cloth after the desired duration of extraction. The filtered extracts were kept at 20-25°C out of direct sunlight. The extracts produced were then fortified with *T. harzianum* isolate Pb-7 inoculum (Yasmeen, 2008). Lentil seed soaked with extraction of *T. harzianum* isolate Pb-7 fortified composted poultry refuge at least 5-6 h before sowing.

Treatments of the experiment

T_1=Uninoculated field (Control-1)

T_2=Inoculated with test pathogen field (Control-2)

T_3=T_2 + Seed treated with spore suspension of *T. harzianum* isolate Pb-7

T_4=T_2 + Seed treated with extracted composted poultry refuge

T_5=T_2 + Seed treated with *T. harzianum* isolate Pb-7 fortified composted poultry refuge

T_6=T_2 + Field treated with wheat grain colonized *T. harzianum* isolate Pb-7

T_7=T_2 + Field treated with composted poultry refuge

T_8=T_2 + Field treated with *T. harzianum* isolate Pb-7 fortified composted poultry refuge

Application of bio-fortified compost

Three separate field experiment for three test pathogens at same season was conducted to assess *T. harzianum* isolate Pb-7 fortified different composting substrate @ 2.5 Kg plot^{-1} (4.17 t ha^{-1}) in each plot except in the control-1 and control-2 of different experiments. On the contrary, wheat grain colonized inoculum of selected three test pathogens were applied @100g plot^{-1} (0.167t ha^{-1}) of different experiments in all treatment except T_1.

Cultivation of lentil in the field soil

Land was prepared by using a tractor driven disc plough, rotavator and harrow. The experiment was designed in the Randomized Complete Block Design (RCBD) with 3 replications. The unit plot size of the experiment was 3 m × 2 m where row to row distance 30 cm. Distance between block to block was 1.0m. Drains were made surrounding the each unit plots and the excavated soil was used for raising plots 15 cm high from the general soil surface. Seeds were sown in rows uniformly at a depth of 2.0 centimeter. Weeding, mulching and irrigation were done in the experimental field whenever necessary.

Data recording

Data were recorded on germination, pre and post-emergence mortality, number of healthy plants and number of infected plant from vegetative and up to harvesting. Determination of seedling mortality and also percent mortality reduction over control-2 was measured by following equation.

$$\% \text{ Mortality reduction} = \frac{X - Y}{X} \text{ x } 100$$

where, X=Total mortality of control-2

Y=Total mortality of individual treatment

Plant with soil-borne diseases symptoms were observed during the growing to harvesting. Ten plants in each plot were randomly selected and uprooted carefully, washed with tap water, checked exclusively and disease severity was rated on 0-4 scale in which 0 = no symptoms, 1=1-25%, 2=26-50%, 3=51-75% and 4=76-100% of lentil organ covered with lesions (Sen, 2010; Morid *et al*., 2012). The disease incidence and disease severity were measured by the following formula (Rahman *et al.*, 2013).

$$\text{Disease Incidence Percentage (\% DI)} = \frac{\text{Number of infected plants}}{\text{Total number of plants assessed}} \text{ x } 100$$

$$\text{Percent Disease Index} = \frac{\text{Summation of all ratings}}{\text{Total number of rating x Maximum disease grade (4)}} \text{ x } 100$$

The percent disease incidence reduction over control-2 was measured by following formula.

$$\% \text{ Disease incidence reduction} = \frac{X - Y}{X} \text{ x } 100$$

where, X=Disease incidence of control-2
Y=Disease incidence of individual treatment

Shoot and root length

At the reproductive phase, five plants were randomly selected from each individual treatment and separated into shoots and roots. The distances from crown to leaf tip and root tip were measured as shoot length and root length, respectively. Then, the mean values of the two parameters were computed.

Shoot dry weight

The shoots of plants in each replication were separately packed and dried at 70ºC until a constant weight was reached. Then, shoot dry weight was measured by digital weighing machine.

1000 seeds weight

One thousand seeds were selected randomly from individual treatment and weighted by digital weighing machine.

Pod yield weight in each plot was recorded and pod yield per hectare was calculated by following formula (Razaq *et al.*, 2015).

$$\text{Total pod yield } (\text{tha}^{-1}) = \frac{\text{Yield per plot (kg)}}{\text{Area of plot } (\text{m}^2) \text{ x } 1000 \text{ (Kg)}} \text{ x } 10000 \text{ m}^2$$

Data analysis

Statistically data were analyzed by using the MSTAT-C program. The treatment means were compared with following Duncan's Multiple Range Test (Gomez & Gomez, 1984).

3. Results and Discussion

Isolation of test pathogens

Each of the three isolates of *F. oxysporum* f. sp. *lentis* designated as Fo-1, Fo-2 and Fo-3, *R. solani* designated as Rs-1, Rs-2 and Rs-3 and *S. rolfsii* designated as Sr-1, Sr -2 and Sr-3 were isolated from the rhizosphere and rhizoplane of different field crops.

Pathogenicity test for the selection of virulent isolates of the test pathogens

The pathogenicity tests of the three selected isolates of *F. oxysporum* f. sp. *Lentis* isolate Fo-3, *R. solani* isolate Rs-3 and *S. rolfsii* isolate Sr-3 were tested in the pot culture. All the isolates of the tested pathogens were virulent but variable in causing total seedling mortality of lentil ranged from 5.00-94.61% except the *F. oxysporum* (Fo-2) isolate collected from chickpea caused only 6.00% seedling mortality significantly differ to the control pot (Table 1). The isolates Fo-3, Sr-3 and Rs-3 were found as the highly virulent isolates of *F. oxysporum* f. sp. *lentis*,*S. rolfsii* and *R. solani*, respectively. These virulent isolates of the tested pathogens were selected for the field inoculation. The pre-emergence and post emergence mortality of different vegetable crops including lentil caused by the tested pathogens are also reported by several investigators (Rojo *et*

al., 2007; Dubey *et al*., 2007; Kashem, 2011; Naziha, 2012; Montaser, 2013; Rubayet & Bhuiyan, 2016).

Table 1. Pathogenicity test of *F. oxysporum* f. sp. *lentis*, *R. solani* and *S. rolfsii* isolates on lentil variety BARI masur- 6

Isolates	% mortality		Total mortality
	Pre-emergence	Post-emergence	
	F. oxysporum f. sp. *lentis*		
Fo-1	35.00	26.92	61.92
Fo-2	2.00	4.00	6.00
Fo-3	40.00	39.58	79.58
	S. rolfsii		
Sr-1	50.00	30.00	80.00
Sr-2	10.00	20.00	30.00
Sr-3	52.5	42.11	94.61
	R. solani		
Rs-1	45.00	34.09	79.09
Rs-2	10.00	12.00	22.00
Rs-3	38.75	48.72	87.47

Screening of T. harzianum isolates against virulent isolates

The antagonistic ability of *T. harzianum* against three isolates of *R. solani* isolate Rs-3, *S. rolfsii* isolate Sr-3, *F. oxysporum* f. sp. *lentis* isolate Fo-3 on PDA medium was recorded and presented in the Table 2 and Plate I. All the isolates of *T. harzianum* have antagonistic effect against *S. rolfsii*, *R. solani* and *F. oxysporum* f. sp. *lentis* with different degrees of inhibition. Twenty isolates of *T. harzianum* performed more than 50% inhibition of radial growth of the tested pathogens as compared with the control. The tested isolates Pb-7 showed the highest 89.81, 86.30 and 88.33% inhibition of the radial growth in case of *F. oxysporum* f. sp. *lentis*, *R. solani* and *S. rolfsii*, respectively. Among the tested 20 isolates of *T. harzianum*, Pb-7 isolate showed growth inhibition at Bell's scale R_1 against *F. oxysporum* f. sp. *lentis*, *R. solani* and *S. rolfsii*. Rest of the isolates of *T. harzianum* showed Bell's scale R_2 antagonism against the tested pathogens except the isolate Pb-10 and Pb-11 which showed scale R_3 growth inhibition in case of *F. oxysporum* f. sp. *lentis*, *R. solani* and *S. rolfsii*. The performance of the isolate Pb-7 of *T. harzianum* was better than all other isolates in inhibiting the radial growth of the tested pathogens. Therefore, the isolate Pb-7 can be used as a bio control agent for the environment friendly management of foot and root rot disease, collar rot disease and *Fusarium* wilt disease incidence in order to increase lentil yield in Bangladesh. The study of the screening of *T. harzianum* isolates against the major soil-borne pathogen including *F. oxysporum* f. sp. *lentis*, *R. solani* and *S. rolfsii* by dual culture technique are observed by several investigators throughout the world including Bangladesh (Said *et al.* 2013; Rubayet & Bhuiyan, 2016; Talukdar *et al*., 2017). Based on screening the highly antagonist *T. harzianum* isolate Pb-7 was selected to prepare the *Trichoderma* spore suspension and preserved for further use in the PDA slant at 10°C.

Table 2. Screening of *T. harzianum* isolate against *F. oxysporum* f. sp. *lentis* isolate Fo-3, *R. solani* isolate Rs-3 and *S. rolfsii* isolate Sr-3 by dual culture technique

Isolates of *T. harzianum*	*F. oxysporum* f. sp. *lentis*		*R. solani*		*S. rolfsii*	
	% inhibition	Bell's scale	% inhibition	Bell's scale	% inhibition	Bell's scale*
Pb-1	73.33	R_2	70.74	R_2	72.96	R_2
Pb-2	66.67	R_3	67.41	R_3	75.19	R_2
Pb-3	71.85	R_2	64.44	R_2	74.44	R_2
Pb-4	64.07	R_2	74.07	R_2	73.33	R_2
Pb-5	70.74	R_2	72.22	R_2	70.00	R_2
Pb-6	68.89	R_2	65.19	R_2	64.44	R_2
Pb-7	89.81	R_1	86.30	R_1	88.33	R_1
Pb-8	67.78	R_2	72.96	R_2	72.96	R_2
Pb-9	70.74	R_2	64.07	R_3	70.37	R_2
Pb-10	52.22	R_3	53.33	R_3	57.04	R_3
Pb-11	65.56	R_3	66.67	R_3	69.63	R_3
Pb-12	75.19	R_2	68.15	R_3	68.89	R_3
Pb-13	71.11	R_2	75.56	R_2	69.26	R_2
Pb-14	68.52	R_2	61.48	R_2	72.22	R_2
Pb-15	63.70	R_2	63.70	R_2	64.07	R_2
Pb-16	69.26	R_2	64.07	R_2	65.93	R_2
Pb-17	59.63	R_3	69.63	R_2	69.26	R_2
Pb-18	71.11	R_2	67.78	R_2	70.74	R_2
Pb-19	72.59	R_2	60.74	R_2	70.74	R_2
Pb-20	62.22	R_2	71.11	R_2	68.89	R_2
Control	0.00		0.00		0.00	

* Bell's scale: R_1= 100% Overgrowth of *T. harzianum*, R_2= 75% Overgrowth of *T. harzianum*, R_3= 50% Overgrowth of *T. harzianum*, R_4= Block at point of contact and R_5= Pathogen overgrowth against antagonist.

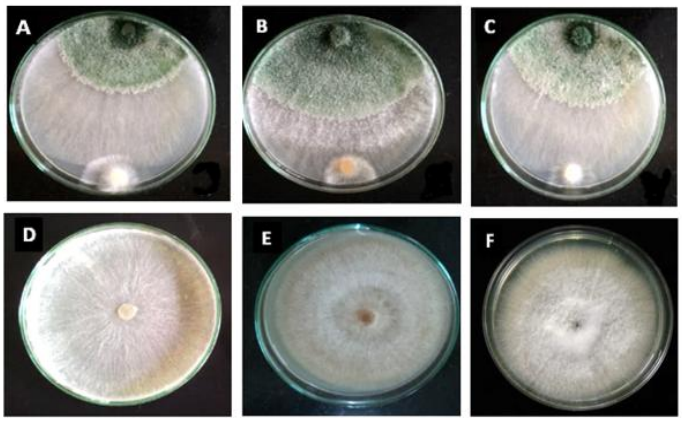

Plate I. Growth inhibition of different pathogens by *T. harzianum* isolate Pb-7 in dual culture PDA plate **A**. *T. harzianum* and *S. rolfsii* isolate Sr-3 **B.** *T. harzianum* and *R. solani* isolate Rs-3 **C.** *T. harzianum* and *F. oxysporum* f. sp. *lentis* isolate Fo-3 **D.** *S. rolfsii* isolate Sr-3 **E.** *R. solani* isolate Rs-3 **F.** *F. oxysporum* f. sp. *lentis* isolate Fo-3.

Effect of T. harzianum isolate Pb-7 on seedling mortality of lentil

Immediately after sowing of lentil seed, pre-emergence and post-emergence seedling mortality caused by *F. oxysporum* f. sp. *lentis* isolate Fo-3, *R. solani* isolate Rs-3 and *S. rolfsii* isolate Sr-3 were recorded up to three weeks of the plant growth. The highest percentage of total mortality was recorded in treatment T_2 (89.00%) followed by

the treatment T_1 (43.75%). In treatment T_8 the lowest (18.75%) seedling mortality was observed where *T. harzianum* fortified composted poultry refuge was applied. On the other hand, the similar result in total mortality percentage with T_8 was also observed in treatment T_6 and T_3, respectively (Table 3). In respect of all treatments, the highest 78.93% reduction of total mortality over control -2 was also found in treatment T_8 followed by T_6 (76.12%) and T_3 (69.10%), respectively (Fig. 1). The control-1 treatment was uninoculated field. As result, the inoculum pressure was poor and disease infestation was also minimum compared to control-2. The emergence of seedling mortality of different crops including lentil caused by soil-borne pathogens are also reported by many investigators (Dubey *et al*., 2007; Montaser, 2013; Rubayet & Bhuiyan, 2016).

Table 3. Effect of *T. harzianum* isolate Pb-7 against seedling mortality of lentil in field

Treatments	% mortality		Total mortality
	Pre-emergence	Post-emergence	
T_1	12.50	31.25	43.75 [b]
T_2	47.00	42.00	89.00 [a]
T_3	13.75	13.75	27.50 [e]
T_4	23.75	15.00	38.75 [c*]
T_5	16.25	15.00	31.25 [d]
T_6	12.50	8.75	21.25 [f]
T_7	18.50	16.25	35.00 [c]
T_8	11.25	7.50	18.75 [f]

*Means within a column having a common letter(s) do not differ significantly (P = 0.05) by DMRT.

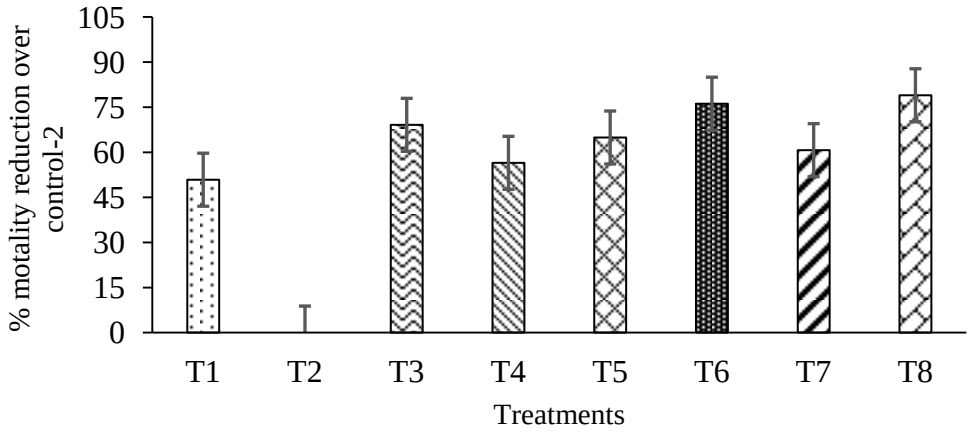

Fig. 1. Percent mortality reduction over control-2

where, T_1 = Uninoculated field (Control-1), T_2= Inoculated with test pathogen field (Control-2), T_3= T_2 + Seed treated with spore suspension of *T. harzianum* isolate Pb-7, T_4= T_2 + Seed treated with extracted composted poultry refuge, T_5= T_2 + Seed treated with *T. harzianum* isolate Pb-7 fortified composted poultry refuge, T_6= T_2 + Field treated with wheat grain colonized *T. harzianum* isolate Pb-7, T_7= T_2 + Field treated with composted poultry refuge, T_8= T_2 + Field treated with *T. harzianum* isolate Pb-7 fortified composted poultry refuge.

Assessment of disease incidence and disease management of lentil in field soil

The incidence of foot and root rot disease ranged from 5.00-23.33%. Among the eight treatments of lentil, the highest 23.33% incidence of foot and root rot disease caused by *R. solani* was observed in the treatment T_2 where only pathogens are added without any other amendments. The lowest incidence of foot and root rot disease 5.00% was observed in the treatment T_8 where *T. harzianum* isolate Pb-7 fortified compost was applied that statistically different from other treatments (Table 4 and Fig. 2). The results are in agreement with Prasad *et al.* (2002).The highest 72.02% reduction of the collar rot disease was observed with the treatment T_8 followed by T_6 where wheat grain colonized *Trichoderma* isolate Pb-7 was applied. Collar rot disease reduction in the treatment T_8 and T_6 were identical but superior to the other treatments. Significantly reduction of collar rot disease caused by *S. rolfsii* were also observed in the treatment T_3, T_4, T_5 and T_7 in comparison to the control-2 where only pathogens were inoculated. But the treatments were significantly differ from each other in controlling the incidence of collar rot disease. The incidence of *Fusarium* wilt disease ranged from 4.17-29.17%. Among the eight treatments of lentil, the highest 29.17% incidence of *Fusarium* wilt disease was observed in the treatment control-2. The lowest incidence of *Fusarium* wilt disease 4.17% was observed in T_8 which was identical with T_3 and T_6. Based on the *Fusarium* wilt disease, the highest reduction 85.71% in T_8 where the field treated with *T. harzianum* isolate Pb-7 fortified composted poultry refuge was followed by T_3 and T_6 (82.86%), where the seed treated with spore suspension of *T. harzianum* isolate Pb-7 and field treated with pathogens and wheat grain colonized bio-agent was applied. Inoculation with *T. harzianum* isolates reduced the incidence of root rot and increased seed germination in lentil (Vyas & Mathur, 2002). *Tricho*-compost was found most effective in controlling soil-borne diseases of cabbage (Naher *et al*., 2012). Rahman (2013) reported that *Trichoderma*-fortified composted poultry refuge was very effective in controlling several soil-borne pathogens of tomato and increased yield significantly in comparison to the untreated control in case of tomato. The results of the current study are in agreement with above mentioned investigators.

Table 4. Effect of bio-fortified compost on disease incidence of soil-borne diseases of lentil in the field condition

Treatments	% disease incidence (DI)		
	Foot and root rot (*R. solani*)	Collar rot (*S. rolfsii*)	*Fusarium* wilt (*F. oxysporum* f. sp. *lentis*)
T_1	12.50 [b]	15.00 [b]	10.00 [c]
T_2	23.33 [a]	20.83 [a]	29.17 [a]
T_3	8.33 [d]	9.00 [c]	5.00 [d]
T_4	7.50 [d]	10.00 [c]	10.83 [c]
T_5	10.00 [c]	6.67 [d]	13.33 [b]
T_6	9.00 [c]	6.00 [e]	5.00 [d]
T_7	10.00 [c]	13.33 [b]	12.00 [bc*]
T_8	5.00 [e]	5.83 [e]	4.17 [d]

*Means within same column followed by common letter(s) are not significantly different (P=0.05) by DMRT.

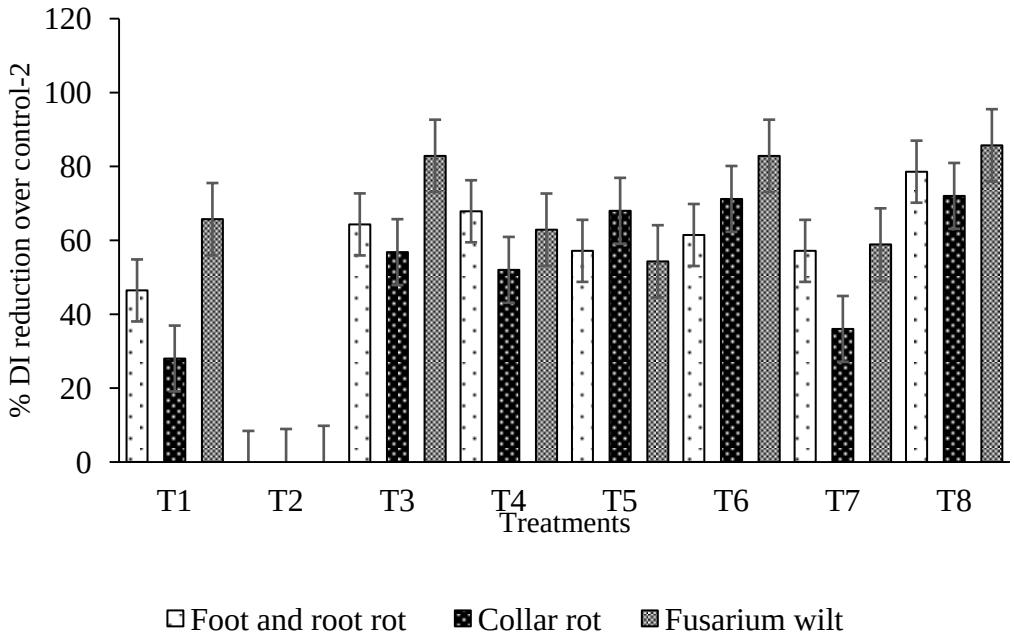

Fig. 2. Percent disease incidence reduction over control-2
where, T_1 = Uninoculated field (Control-1), T_2= Inoculated with test pathogen field (Control-2), T_3= T_2 + Seed treated with spore suspension of *T. harzianum* isolate Pb-7, T_4= T_2 + Seed treated with extracted composted poultry refuge, T_5= T_2 + Seed treated with *T. harzianum* isolate Pb-7 fortified composted poultry refuge, T_6= T_2 + Field treated with wheat grain colonized *T. harzianum* isolate Pb-7, T_7= T_2 + Field treated with composted poultry refuge, T_8= T_2 + Field treated with *T. harzianum* isolate Pb-7 fortified composted poultry refuge.

Effect of bio-fortified compost on growth promotion of lentil

All the *T. harzianum* isolate Pb-7 fortified composted poultry refuge treatments significantly increased the growth promotion traits compared to control-2 plot. In case of shoot length in treatment T_8 was identical (33.87 cm) with the treatment T_6 (31.53 cm) where wheat grain colonized *T. harzianum* isolate Pb-7 was applied. The other treatments T_3, T_4, T_5 and T_7 where seed treated with spore suspension of *T. harzianum* isolate Pb-7, seed treated with poultry refuge extract, seed treated with extract of *T. harzianum* isolate Pb-7 fortified composted poultry refuge and field treated with only poultry refuges, respectively were also significantly increased shoot length. Similar result were also observed in case of root length, fresh shoot weight, dry shoot weight and 1000 seeds weight (Table 5). The same parameter of different crops increasing growth promoting traits were observed by other investigator with the application of *Trichoderma*-fortified composted poultry refuges (Rahman, 2013; Talukdar *et al.* 2017).

Effect of bio-fortified compost on increasing the yield of lentil

The highest 2.52t ha^{-1} lentil seed was produced in the treatment T_8 where *T. harzianum* isolate Pb-7 fortified composted poultry refuge was applied which was significantly higher in comparison to all other treatments. The second highest yield (2.27 t ha^{-1}) was produced in the treatment T_6 significantly higher than the treatment T_3, T_4, T_5 and T_7, respectively but superior to the control-2 treatments (Table 5 and Fig. 3). The results are also in agreement with the other investigators whose were also observed

the disease reduction and increased of yield of other different crops by the application of *Trichoderma*-fortified compost (Prasad *et al*., 2002; Rahman, 2013; Akter, 2016).

Table 5. Effect of bio-fortified compost on growth promotion and yield of lentil

Treatments	Shoot length (cm)	Root length (cm)	Fresh shoot weight (g)	Dry shoot weight (g)	1000 seed weight (g)	Yield (gm^{-2})	Yield (tha^{-1})
T_1	24.13 ^{c}	9.40 ^{d}	24.00 ^{e}	11.77 ^{e}	23.47 ^{e}	100.67 ^{ef}	1.01 ^{ef*}
T_2	21.53 ^{d}	8.93 ^{d}	22.00 ^{ef}	11.73 ^{e}	21.43 ^{f}	88.33 ^{f}	0.88 ^{f}
T_3	25.73 ^{b}	11.50 ^{b}	27.40 ^{b}	18.57 ^{c}	28.00 ^{bc}	191.67 ^{c}	1.92 ^{c}
T_4	26.27 ^{b}	10.60 ^{c}	26.43 ^{cd}	14.13 ^{d}	27.60 ^{c}	125.33 ^{e}	1.25 ^{e}
T_5	28.53 ^{b}	11.1 ^{b}	25.00 ^{d}	20.60 ^{b}	30.13 ^{b}	181.67 ^{d}	1.81 ^{d}
T_6	31.53 ^{a}	12.20 ^{a}	32.00 ^{a}	20.97 ^{a}	32.43 ^{ab}	227.00 ^{b}	2.27 ^{b}
T_7	24.93 ^{bc}	10.60 ^{c}	25.50 ^{de}	15.05 ^{d}	27.53 ^{c}	159.00 ^{d}	1.59 ^{d}
T_8	33.87 ^{a}	13.60 ^{a}	34.23 ^{a}	21.57 ^{a}	35.74 ^{a}	251.85 ^{a}	2.52 ^{a}

*Means within same column followed by common letter(s) are not significantly different (P=0.05) by DMRT.

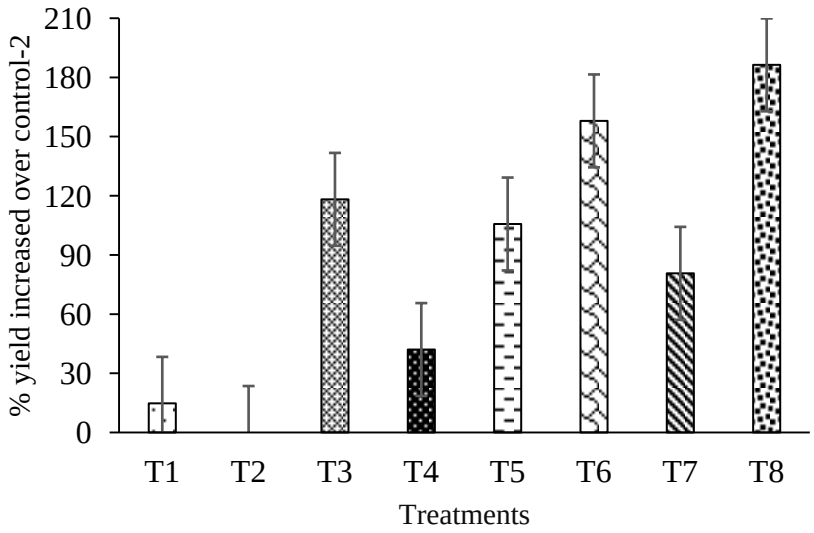

Fig. 3. Percent yield increased over control-2

where, T_1 = Uninoculated field (Control-1), T_2= Inoculated with test pathogen field (Control-2), T_3= T_2 + Seed treated with spore suspension of *T. harzianum* isolate Pb-7, T_4= T_2 + Seed treated with extracted composted poultry refuge, T_5= T_2 + Seed treated with *T. harzianum* isolate Pb-7 fortified composted poultry refuge, T_6= T_2 + Field treated with wheat grain colonized *T. harzianum* isolate Pb-7, T_7= T_2 + Field treated with composted poultry refuge, T_8= T_2 + Field treated with *T. harzianum* isolate Pb-7 fortified composted poultry refuge.

4. Conclusion

Based on the present study it may be concluded that application of *T. harzianum* isolate Pb-7 fortified composted poultry refuge is one of the excellent substrate in controlling soil-borne diseases of lentil. The composted may also enhance the efficacy of bio-agent against the soil-borne pathogens. In the meantime, it provides solubilized nutrients to the plant, improving soil health status and increase the yield of lentil.

Acknowledgement

The authors have been thankful to the Ministry of Science and Technology, Dhaka, Bangladesh and Higher Education Quality Enhancement Project (HEQEP), CP-2075, Bangabandhu Sheikh Mujibur Rahman Agricultural University, Gazipur-1706, Bangladesh for providing financial support.

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