E-ISSN: 2789-3073
P-ISSN: 2789-3065
IJPPM 2022; 2(2): 33-37
Received: 19-05-2022
Accepted: 22-06-2022

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Effect of tricho-compost and tricho-leachate on germination, vigor, and seedling mortality of chili

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Abstract
The experiment was conducted to evaluate the effect of tricho-compost and its by-product tricho-leachate on germination, vigor, and suppression of seedling mortality of chili. Seven isolates of *Trichoderma harzianum* were screened by following the dual culture technique using *Sclerotium rolfsii* as a sample pathogen to find out the most effective isolates. Among all of the isolates, bioderma showed the highest inhibition of mycelial growth (71.41%), and the lowest mycelial growth inhibition was observed by Th3 (59.37%). That's why bioderma isolates were used in this experiment for the production of tricho-compost and tricho-leachate. There were 6 treatments generated in this study with different doses of tricho-compost and tricho-leachate namely T1 (100g tricho-compost + 25 ml tricho-leachate), T2 (50g tricho-compost + 25 ml tricho-leachate), T3 (100 g tricho-compost), T4 (25 ml tricho-leachate), T5 (100g cowdung), and T6 (control). The result revealed that the germination percentage of chili was significantly increased by T1 (96.43%) treatment over control (T6 = 43%). The root length and shoot length were sharply increased by T1 (10.84 cm and 9.45 cm, respectively) over control T6 (3.67 cm and 3.31 cm respectively). The highest vigor index was recorded in T1 (1957.0) and the lowest vigor index was recorded in T6 (300.29). Furthermore, the seedling mortality was significantly suppressed by T1 (2.33%) over control T6 (8.67%). The present studies suggested combination effect of tricho-compost and tricho-leachate in T1 treatment is suitable for the production of healthy and vigorous chili seedlings which impact significantly in improving germination, vigor, and suppression of mortality chili seedlings.

Keywords: Tricho-compost, Tricho-leachate, Germination, Vigor, and Mortality

1. Introduction
Chili is an important spice and valuable cash crop with excellent sources of vitamins, potassium, magnesium, and iron. In Bangladesh, about 102.25 thousand tons of chili were produced under the cultivation of 93.55 thousand hectares area in 2012-2013 which is very low compared to other chili-growing countries. (BBS, 2015) [3]. There are several factors behind that low yield of chili. Among them low organic matter content and disease incidence are prominent an ideal soil contains more than 3% organic matter. But in Bangladesh, most of the soil contains less than 1.5% organic matter which is gradually decreasing day by day (BARC, 1997) [2]. On the other hand, about 16% of annual crops are lost in Bangladesh due to plant diseases. (Fakir, 1983) [6]. Chili crops are also infected by more than 83 diseases such as *Pythium sp*., *Fusarium oxysporum*, *Sclerotium rolfsii*, *Phytophthora sp*, and *Rhizoctonia solani*, etc. (Singh, 1984) [17]. There are various methods to manage these diseases among them chemical method is found most effective which has a very hazardous effect on human health as well as the environment. That's why in the recent Integrated Disease Management system (IDM), biological control using antagonistic microbes has become more popular to minimize the use of chemical fungicide (Daamen *et al*., 1989) [5]. Bio-composts have the potential to provide biological control of disease and pathogens, through its microbial actions (Inbar *et al*., 1994) [9]. Tricho-compost and tricho-leachate is a bio-compost that is prepared by using *Trichoderma sp*. fungi in the composting process which has an antifungal agent and nutritive value for plant (Rahman and Birkey, 2015) [15]. It's also a good source of macro and micronutrients which can solve the lack of organic matter (Rahman, 2009) [13]. Besides Tricho-compost can be produced by utilizing large number of agricultural wastage with low capital involvement and higher volumetric productivity. Considering the facts mentioned above, the present investigation was undertaken to determine the effect of Tricho-compost and Tricho-leachate on germination, vigor, and suppression of seedling mortality of chili to achieve the

Following objective

- To select the effective isolate of *Trichoderma harzianum* using a soil-borne pathogens
- To find out the effect of tricho-compost and tricho-leachate on seed germination and seedling vigor.
- To find out the effect of tricho-compost and tricho-leachate on the suppression of seedling mortality of chili.

2. Materials and Method

The efficacy of Tricho-compost and Tricho-leachate on germination, seedling vigor, and suppression of seedling mortality of chili was investigated in the Plant Disease Clinic (PDC) and Plant Pathology Laboratory of the Department of Plant Pathology, Patuakhali Science and Technology University (PSTU).

2.1 Screening of Trichoderma herzianum as a biocontrol agent using Sclerotium rolfsii

Six isolates of *Trichoderma harzianum* were collected from the different agricultural land of Dumki, Patuakhali through the dilution plate method, and one *Trichoderma* formulation named Bioderma was collected from the supermarket. Their antagonistic effect was determined through the duel culture technique using *Sclerotium rolfsii* as a tested pathogen (Tuite, 1996) [19]. The dual culture was developed on Potato Dextrose Agar (PDA) media. 5mm diameter mycelial disc of *T. herzianum* and *S. rolfsii* was placed simultaneously on the edge of each PDA petridish in opposite direction. After 7 days, the inhibition percentage of mycelia growth of *S. rolfsii* was calculated by following the formula as suggested by Sunder *et al*. (1995) [20].

$$\% \text{ inhibition } = \frac{X - Y}{X} \times 100 .$$

Where

- X= Mycelial growth (mm) of the pathogen in absence of antagonists.
- Y= Mycelial growth of the pathogen in presence of antagonists.

2.2 Preparation of Tricho-compost and Trichho-leachate

Tricho-compost and Tricho-leachate were produced by using the procedure of housing system which was measured 10 ft. in length x 5 ft. in width x 4.5 ft. in height (Rahman & Birkey, 2015) [15]. Raw materials of tricho-compost, such as cow dung (45 kg), poultry refuse (45 kg), water hyacinth (45 kg), sawdust (2 kg), maize bran (1 kg), neem leaf (1 kg), ash (1 kg), and molasses (500g) were collected from nearby areas. All the raw materials were chopped into small pieces and mixed together. The *Trichoderma* inoculum suspension was prepared by mixing with 500 g of molasses and 2 liters of water. The inoculum suspension was drenched onto the compost mixture in a layering method and kept for 45 days maintaining the moisture content around 60% until fully decomposed. Tricho-leachate was collected during the decomposition of Tricho-compost materials.

2.3 Experimental design

The experiment was conducted by following a Completely Randomized Design (CRD) with three replications in the tray. There were 18 unit plots and the size of each plot was 0.052 m^2. Following five different proportions of tricho-compost and tricho-leachate were used as a treatment in this experiment. They were

- T1= 100g Tricho-compost + 25 ml Tricho-leachate per tray
- T2=50g Tricho-compost + 25 ml Tricho-leachate per tray
- T3= 100 g Tricho-compost per tray
- T4 = 25 ml tricho-leachate per tray
- T5 = 100g cow dung per tray
- T6= control

2.4 Germination and Mortality Test

After 7 days of sowing, the number of germinated seedlings, pre-emergence, and post-emergence mortality data was recorded. Dead seedlings were counted every alternate day and counted for 20 days after sowing. The dead seed and seedlings were uprooted gently to determine the reason for the death. The germination percentage was calculated using the following formula:

% Germination = (Number of seeds germinated/Number of seeds sown) × 100

2.5 Vigor Test

For determination of seedling vigor index, 6 seedlings were randomly selected from each tray and their individual shoot and root length were measured. The root length was scaled from the collar region to the tip of the root. The shoot length was measured from the collar region to the point of attachment of cotyledons to the tip of the shoot. The vigor of the seedlings was determined by following the formula of Abdul-Baki and Anderson (1972) [1]:

Vigor index = [mean of root length (cm) + mean of shoot length (cm)] × percentage of seed germinations

2.6 Analysis of Data

The data obtained from different parameters were statistically analyzed through Analysis of variance (ANOVA) by using the MSTAT-C program and means were compared by LSD test (Least Significance Difference) at the 5% level of probability (Gomez and Gomez, 1984) [8]

3. Results Discussion

3.1 Screening of Trichoderma harzianum Isolate

The antimicrobial effect of seven *Trichoderma herzianum* isolates was screened out using *Sclerotium rolfsii* pathogen on PDA media (Table 1). The result showed that each *T. herzianum isolate* was able to inhibit the mycelial growth of *S. rolfsii* which was more than 50% over control. The inhibition percentage ranged from 59.37% to 71.41%. The highest mycelial inhabitation percentage observed in Bioderma (71.41%) followed by Th5 (68%), Th4 (67.43%), Th6 (64.43%) and Th1 (63.43%) isolates. The lowest mycelial inhabitation percentage was observed in Th3 (59.37%). Th4 isolates were statistically similar to Th5 isolates and Th1 isolates were statically similar to Th6 isolates. Among all isolates, bioderma showed the best result to inhibit the mycelial growth of *Sclerotium rolfsii* which was used for producing tricho-compost and tricho-leachate.

Table 1: Inhibition of mycelial growth of *Sclerotium rolfsii* on PDA medium culture by selected seven isolates of *Trichoderma harzianum*

Trichoderma harzianum isolates	% inhibition of mycelial growth *Sclerotium rolfsii*
Th1	63.43 c
Th2	62.04 d
Th3	59.37 e
Th4	67.43 b
Th5	68.00 b
Th6	64.43 c
Bioderma	71.41 a
LSD (P = 0.05)	1.35

Values within a column with a different letter(s) differ significantly (P=0.05) by LSD

3.2 Effect of Tricho-compost and Tricho-leachate on Germination of Chili Seed

The seed germination percentage of chili was significantly (P=0.05) increased by the application of different doses of Tricho-compost and Tricho-leachate. The germination percentage ranged from 43 to 96.43 (Table 2). The maximum seed germination was recorded in the T1 treatment (100g Tricho-compost + 25 ml Tricho-leachate; 96.43%) which was significantly different from others. Minimum germination percentage was recorded in T6 treatment (Control; 43%). Germination percentage of T2 (50 g Tricho-compost + 25 ml Tricho-leachate; 90.89%) is statically similar to T3 (100 g Tricho-compost; 91.61%). From the results, it was revealed that Tricho-compost was able to increase the seed germination of chili. Some researchers also found the efficacy of Tricho-compost on the germination of various plants. Rahman *et al.* (2010) [14] reported that Tricho-compost enhanced the germination percent of chili. Nahar *et al.* (2012) [12] investigated that the seed germination percentage was significantly increased due to the application of Tricho-compost in soil. Sarker *et al.* (2017) [16] showed that Tricho-compost performed the best compared to botanical treatment in terms of germination.

Table 2: Effect of Tricho-compost and tricho-leachate on germination of chili seed

Treatment	Germination percentage
T1	96.43 a
T2	90.89 b
T3	91.61 b
T4	66.67 c
T5	51.33 d
T6	43.00 e
LSD (P ≥ 0.05)	3.96

Values within a column with a different letter(s) differ significantly (P=0.05) by LSD

3.3 Effect of Tricho-compost and Tricho-leachate on Seedling Vigor

The root length, shoot length, and vigor index was significantly influenced by different concentrations of tricho-compost and tricho-leachate (Table3). The highest root length was recorded in the T1 treatment (100g Tricho-compost + 25 ml Tricho-leachate; 10.84) and the lowest root length was recorded in the T6 treatment (Control; 3.67). Maximum shoot length was measured in the T1 treatment (100g Tricho-compost + 25 ml Tricho-leachate; 9.45) which is significantly similar to T3 Treatment (100 g Tricho-compost; 9.14). Minimum shoot length was measured in T6 Treatment (Control: 3.31). The vigor index of chili ranges from 300.39 to 1957. The highest vigor index was recorded in the T1 treatment (100g Tricho-compost + 25 ml Tricho-leachate; 1957.0) which is significantly different from others. The lowest vigor index was recorded in the T6 treatment (Control; 300.29). Treatment T2 (50g Tricho-compost + 25 ml Tricho-leachate; 1520.9) is statistically similar to T3 (100 g Tricho-compost; 1675.2). The result revealed that the combined effect of Tricho-compost and Tricho-leachate increased the vigor index of chili seedlings. Begum *et al.* (2010) and Mukhtar observed the highest vigor index when chili and okra seeds were treated with *T. harzianum* respectively. Lo and Lin (2002) noted that *Trichoderma* strains significantly increased seedling height and root exploration of bitter gourd, loo fah, and cucumber.

Table 3: Effect of Tricho-compost and Tricho-leachate on Seedling Vigor

Treatment	Mean Root Length	Mean Shoot Length	Vigor Index
T1	10.84 a	9.45 a	1957.0 a
T2	9.32 b	7.40 b	1520.9 b
T3	9.67 b	9.14 a	1675.2 b
T4	7.76 c	6.17 c	940.64 c
T5	5.54 d	5.28 c	556.03 d
T6	3.67 e	3.31 d	300.29 e
LSD (P ≥ 0.05)	1.1517	0.9970	183.43

Values within a column with different letter(s) differ significantly (P=0.05) by LSD

3.4 Effect of Tricho-compost and Tricho-leachate on Seedling Mortality

Seedling mortality was significantly reduced due to the application of different doses of Tricho-compost and

Tricho-leachate (Table 4). The lowest seedling mortality was recorded in T1 treatment (100g Tricho-compost + 25 ml Tricho-leachate; 2.33%) which was statistically similar to T2 treatment (50g Tricho-compost + 25 ml Tricho-leachate; 3.33%). The highest seedling mortality was observed in T6 treatment (Control; 8.67%). T2 treatment (3.33%) is statistically similar to T3 treatment (100 g Tricho-compost; 3.67%) and T4 (25 ml tricho-leachate; 5.67%) is statistically similar to T5 treatment (100g cow dung; 6.33%). Similarly, Nahar *et al*. (2012) [12] thesis observed that the application of tricho-compost and tricho-leachate reduced 98% of the seedling mortalities of cabbage caused by *Sclerotium rolfsii*. Sarker *et al*. (2017) [16] found that Tricho-compost significantly reduced seedling mortality of Indian spinach. Faruk (2019) [7] reported that seedling mortality of barley was sharply reduced due to the application of the soil amendment with Tricho-composts.

Table 4: Effect of tricho-compost and tricho-leachate on seedling mortality

Treatment	Mortality Rate (%)
T1	2.33 d
T2	3.33 cd
T3	3.67 c
T4	5.67 b
T5	6.33 b
T6	8.67 a
LSD (P ≥ 0.05)	1.2579

Values within a column with a different letter(s) differ significantly (P=0.05) by LSD

4. Conclusion

From the result of the present study, it can be concluded that the combined effect of Tricho-compost and Tricho-leachate provided the maximum germination, highest index vigor, and reduced seedling mortality of chili. As tricho-compost and Tricho-leachate has an antimicrobial effect and high nutrient values, which can be effectively used as fertilizer as well as soil amended. It can play a vital role in the depletion of chemical fertilizers and fungicides. Besides, it utilizes our agriculture wastage and minimizes production costs. So application of Tricho-compost and Tricho-leachate can be an excellent combination to produce healthy seedlings and improve crop production.

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