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EFFICACY OF PHOSPHO-VERMICOMPOST, BIOFERTILIZER, BIOPESTICIDES AND THEIR INTEGRATION AGAINST ROOT ROT OF LENTIL

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Abstract

Lentil is a valuable leguminous crop with high nutritional content, but it is susceptible to various soil-borne pathogens, including root rot, which can severely impact its yield and quality. In pursuit of sustainable and environmentally friendly agricultural practice, this study aimed to evaluate the efficacy of BINA phospho-vermicompost, biofertilizer BINA-LT-17 (*Rhizobium leguminosarum*), BINA biofungicide (*Trichoderma asperellum*) and their integration in managing root rot of lentil under field condition. Results from the study demonstrated that the application of BINA phospho-vermicompost (2 t kg^{-1}) during final land preparation, seed coating with biofertilizer BINA-LT-17 (peat soil-based *Rhizobium leguminosarum*) and soil application of chickpea bran based BINA biofungicide (*Trichoderma asperellum*) significantly reduced root rot incidence while enhancing plant vigor and yield. This integrated approach demonstrated a synergistic effect, suggesting that combining organic amendments with beneficial microorganisms, including both *Rhizobium* and *Trichoderma*, can provide a comprehensive strategy to control root rot in lentil.

Keywords: BINA phospho-vermicompsot, Biofertilizer BINA-LT-17, BINA biofungicide, Root rot, Lentil.

Introduction

Lentil (*Lens culinaris* fsp. *Culinaries* Mendikus) is an essential cool-season legume that play a significant role in both human and animal nutrition, as well as in maintaining soil fertility. It serves as an affordable source of dietary protein (22-35%), minerals, prebiotics and soluble fibers, which are crucial for alleviating malnutrition and micronutrient deficiencies, particularly in developing countries such as Bangladesh. Often referred to as "rural people's meat," lentil ranks fifth in global seed legume production (Lucas and Fuller, 2020). The total cultivated area for lentil is approximately 6.10 million hectares, yielding about 6.33 million tons annually, with a productivity rate of 1.04 MT/ha. The leading producers include Canada, India, the USA, Turkey, Australia, Kazakhstan, Nepal, the Russian Federation, Bangladesh and China (FAOSTAT, 2020). In Bangladesh, lentil holds the top position among legumes, being the most preferred and wisely consumed. It is cultivated over approximately 141, 296 hectares, with an average yield of 1.25 t/ha (BBS, 2020). However, current production level fall shorts of meeting the demand of the population.

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The low yield of lentil is primarily attributed to various biotic and abiotic stresses, with disease being a significant factor. A total of 15 pathogens causing 17 diseases have been identified in Bangladesh, with root rot, particularly caused by *Sclerotium rolfsii*, being the most devastating (Rashid *et al.*, 2007). This polyphagous soil-borne pathogen affects over 500 plant species globally and pose a substantial threat to lentil production, especially in regions with high soil moisture and temperature during the seedling stage. Root rot can lead to yield losses of up to 50% in affected fields (Asghar *et al.*, 2018). Symptoms include poor emergence, stunting, yellowing of leaves and decay of roots, often occurring in patches that can expand under favorable conditions. Controlling soil-borne pathogens, particularly *Sclerotium rolfsii*, using chemicals is challenging. Moreover, the use of inorganic pesticides has detrimental effects on soil fertility, beneficial microorganism and the natural resistance of crop, making them more susceptible to disease. This reliance on chemical control has raised concerns regarding human health and environmental issues, including pesticide residue in food and soil degradation (Rekha *et al.*, 2018). As a result, there is a growing trend towards ecofriendly methods for protection. Among these methods, *Trichoderma* species have emerged as effective biocontrol agents against fungal pathogens. They can suppress phytopathogens through competition for nutrients and space, environmental modification and enhancement of plant growth and defense mechanism (Hafez *et al.,* 2013). Additionally, rhizobia have been explored as biocontrol agent against legume root rot, as they occupy the same ecological niche as pathogenic fungi and can inhibit fungal growth (Volpiano *et al.*, 2019). Seed treatment with *Rhizobium leguminosarum* pv. *Viceae* has been shown to reduce damping-off and improve seedling growth and yield in legumes (Huang *et al.,* 2007). The combination of *Trichoderma* and bacterial biocontrol agents has gained attention for its synergistic effects in controlling agricultural pest and diseases. Co-inoculations of *Trichoderma* and bacteria have demonstrated significant reduction in fungal and oomycete pathogens (Izquierdo-García *et al.*, 2020; Firdu *et al.*, 2020, 2021). For instance, co-inoculating seed with *Trichoderma viride* and *Pseudomonas. fluorescens* have been effective in reducing seedling mortality caused by *Sclerotium rolfsii* through the production of hydrogen cyanide and volatile anti metabolites (Manjula *et al.*, 2004). Another promising alternatives is vermicompost, which not only enhance crop growth and yield but also suppresses diseases sustainably while protecting human health and the environment. Vermicompost enriches soil with nutrients, plant growth regulators and beneficial microbes, thereby improving crop resilience against soil-borne pathogens (Yatoo *et al.,* 2021). Research has shown that vermicompost can significantly suppress disease caused by pathogens such as *Pythium*, *Fusarium* and *Phytophthora* (Edwards and Arancon, 2004). Given the importance of lentil cultivation and the challenges posed by soil-borne diseases, the present study aims to evaluate the efficacy of BINA phospho-vermicompsot, biofertilizer BINA-LT-17 (*Rhizobium leguminosarum*) and BINA biofungicide (*Trichoderma asperellum*), either alone or in combination, in managing root rot disease of lentil under field conditions.

Materials and Methods

Site description

The experiment was conducted at the research farm of Bangladesh Institute of Nuclear Agriculture (BINA) Sub-station, Ishurdi during November 2019 to March 2020 and November 2020 to March 2021. The land was medium high having sandy loam textured soil with soil pH 7.65 (Annonymous, 2012). Growing period was suitable and weather was optimally dry and humid, suitable for lentil production.

Experimental details

The experiment was carried out in a randomized complete block design with three replications. The seeds of susceptible lentil variety Binamasur-6 were collected from Plant Breeding Division, BINA, Mymensingh. The seeds were sown in rows on second weeks of November in two consecutive growing seasons (2019-20 and 2020-21). Size of the plots was 2.0 m × 2.0 m and plant spacing was 30 cm with continuous sowing. The fertilizers were applied as per the Fertilizer Recommendation Guide (BARC, 2018) and normal cultural practices were followed. Eight treatments were T_1= Soil amendment with BINA phospho-vermicompost @ 2 tkg^{-1}, T_2=Seed treatment with biofertilizer BINA-LT-17 (*Rhizobium leguminosarum*) @ 50 g/kg of seed weight, T_3= Seed treatment with BINA biofungicide (*Trichoderma asperellum*) @ 3% of seed weight, T_4= Soil amendment with BINA biofungicide (*Trichoderma asperellum*) @ 150 $kgha^{-1}$, T_5= Soil amendment with BINA phospho-vermicompost + Seed treatment with biofertilizer BINA-LT-17, T_6= Soil amendment with BINA phospho-vermicompost + Seed treatment with BINA biofungicide, T_7= Soil amendment with BINA phospho-vermicompost+ Seed treatment with biofertilizer BINA-LT-17 + Soil amendment with BINA biofungicide and T_8= Control (no treatment).

Biofertilizer BINA-LT-17 was collected from the Soil Microbiology Laboratory, Soil Science Division, BINA. The composition of the biofertilizer was peat soil and *Rhizobium leguminosarum*, which was isolated from the nodules of legume plants. For preparing peat based biofetilizer, at first 500 g peat soil was poured in a polythene bag then sterilized followed by inoculation with a previously prepared 5 ml broth culture of *R. leguminosarum* (10^{11} CFU/ml) and, then mixed thoroughly for proper distribution. Then the materials were incubated for 7 days at 25 ± 2^{o} C. After 7 days of incubation, it was ready for use. Before sowing, required number of seeds were taken in a beaker and a few drops of molasses were added for moistening the seed surface uniformly to allow maximum adherence of biofertilizer on the whole surface of seeds. Then the seeds were thoroughly mixed with biofertilizer (@ 50 g/kg of seed weight) where the biofertilzer contained 10^8 cells/g carrier materials.

Phospho-vermicompost was collected from the Soil Science Division, BINA. The composition of BINA developed phospho-vermicompost was 50% Cowdung + 25% Water hyacinth + 25% Rice straw + 4% Rock phosphate. It has 15.2% Organic Carbon, 1.42% Nitrogen, 1.45% Phosphorus, 1.52% Potassium and 0.35 Sulfur and the C: N ratio was 10:7.

Besides, total counts of bacteria were 7 x 10^{12} per gram (Haque *el al.* 2020). Phospho-vermicompost was added to the soil during final land preparation @ 2 t/ha. Phospho-vermicompost was P enriched through rock phosphate, so it was contained more nutrients than other only residues or cow dung based vermicompost.

BINA biofungicide (*Trichoderma* based) was collected from Plant Pathology Division, BINA. The antagonistic "*Trichoderma* (*Trichoderma asperellum*)" is the active ingredient of this biofungicide which was developed through gamma radiation and then formulated with pulse bran (chickpea bran). It contains 10^6 CFU g^{-1} of culture medium. The application rate was 150 kg ha^{-1}. During application 50 kg was applied at 7 days before seed sowing and the rest 100 kg was applied in two splits at 20 days interval. For seed treatment, the biofungicide was applied @ 3% of seed weight.

Pathogen mass culture and inoculation

Fresh diseased plant samples were collected from lentil field. Roots of diseased plant showing symptoms were washed thoroughly with running tap water followed by cutting into small pieces with the help of sterilized blade. Then surface sterilization was done with 3% Sodium Hypochlorite and 70% ethanol followed by three washings with sterilized distilled water to remove traces of sodium hypochlorite and ethanol. Then the pieces were transferred aseptically to Petri plates containing sterilized PDA and incubated at 25 ± 2 ^{o}C for three to five days and examined at frequent intervals to see the growth of the fungus developing from different pieces. As and when fungal colony appears they were transferred to PDA slant for purification of culture. Purified *S. rolfsii* was mass multiplied in lentil grain media for inoculation. Then multiplied culture of *S. rolfsii* were inoculated in collar zone of lentil plant within 30 days after sowing in both growing season (Koshariya *el al.*, 2020).

Data collection and Statistical Analysis

Data on different parameters i.e. germination, plant stand, plant height, no. of branches, no. of nodule, weight of nodule $plant^{-1}$, dry shoot weight, root length, dry root weight, no. of pods and grain yield were collected from five randomly selected plants from each plot. The incidence of root rot of lentil was recorded at 10 days interval. The incidence of root rot disease of lentil was calculated by the following formula:

$$\text{Incidence of root rot (\%)} = \frac{\text{Number of infected plants}}{\text{Total number of plants}} \times 100$$

The data were analyzed using statistical software Statistix10 (Version 10.0) and Microsoft Excel program.

Results and Discussion

Effect of phospho-vermicompost, biofertilizer BINA-LT-17 and BINA biofungicide on germination, root rot incidence and plant stand of lentil

The Table 1 shows the results of the effects of various treatments on germination, root rot incidence and plant stand of lentil over two growing seasons (2019-2020 and 2020-2021). The results indicated that the treatments T_4 (soil amendment with BINA biofungicide) and T_7 (soil amendment with BINA phospho-vermicompost + seed treatment with biofertilizer BINA-LT-17 + soil amendment with BINA biofungicide) had the highest germination rates, which ranged from 80.16% to 88.37% in both seasons. The treatment T_8 (control) had the highest root rot incidence, reaching 43.33% in 2019-2020 and 35.09% in 2020-2021. The treatment T_4 (soil amendment with BINA biofungicide) and T_7 (soil amendment with BINA phospho-vermicompost + seed treatment with biofertilizer BINA-LT-17 + soil amendment with BINA biofungicide) had the lowest root rot incidence, with values below 18%. The treatments T_4 and T_7 also had the highest plant stands, with values exceeding 80% in both seasons. The treatment T_8 (control) had the lowest plant stand, ranging from 53.64% to 63.34%. The soil application of BINA biofungicide (*Trichoderma asperellum*) significantly reduced the incidence of root rot compared to other treatments, as demonstrated by the results. The result revealed that, the combination of soil amendment with phospho-vermicompost, seed treatment with biofertilizer BINA-LT-17 and soil amendment with BINA biofungicide (T_7) was the most effective in improving seed germination, reducing root rot incidence and increasing plant stand of lentil compared than the control treatment.

Table 1. Effect of BINA phospho-vermicompost, biofertilizer BINA-LT-17 and BINA biofungicide on germination, root rot incidence and plant stand of lentil

Treatments	Seed germination (%)		Root rot incidence (%)		Plant stand (%)	
	2019-2020	2020-2021	2019-2020	2020-2021	2019-2020	2020-2021
T_1	79.97a	80.33abc	28.80bcd	17.94c	70.73abc	81.25c
T_2	79.00ab	81.67ab	37.52ab	20.39c	61.41cd	79.05c
T_3	76.20ab	74.67cd	34.00abc	19.25c	63.23bcd	80.18c
T_4	80.44a	80.67abc	18.02d	10.87e	80.16a	88.37a
T_5	74.54ab	75.67bc	22.02cd	14.63d	75.67ab	84.90b
T_6	72.92b	78.33abc	39.85ab	28.80b	60.03cd	70.44d
T_7	80.73a	82.33a	16.43d	11.50e	83.03a	88.24a
T_8	64.81c	69.00d	43.33a	35.09a	53.64d	63.34e
LSD (0.05)	6.84	6.82	12.74	2.90	13.00	2.90
CV %	3.12	3.04	14.75	5.09	6.56	1.27

T_1= Soil amendment with phospho-vermicompost, T_2=Seed treatment with biofertilizer BINA-LT-17 (*Rhizobium leguminosarum*), T_3= Seed treatment with BINA biofungicide (*Trichoderma asperellum*), T_4= Soil amendment with BINA biofungicide (*Trichoderma asperellum*), T_5= Soil amendment with BINA phospho-vermicompost vermicompost + Seed treatment with Biofertilizer BINA-LT-17, T_6= Soil amendment with BINA phospho-vermicompost + Seed treatment with BINA biofungicide, T_7= Soil amendment with BINA phospho-vermicompost + Seed treatment with biofertilizer BINA-LT-17 + Soil amendment with BINA biofungicide and T_8= Control (no treatment).

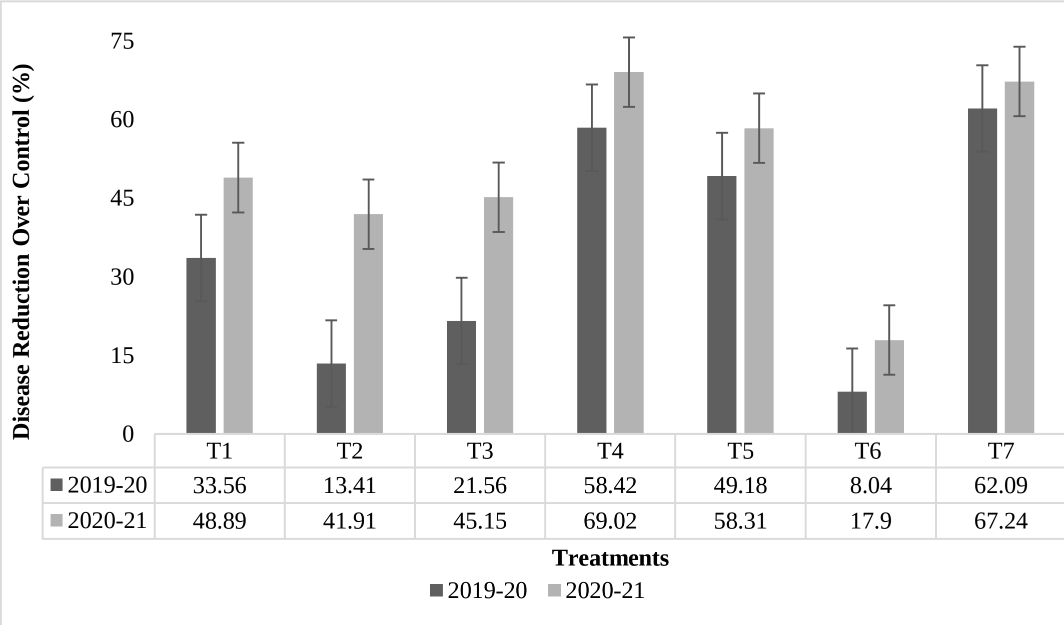

Fig. 1. Root rot disease reduction over control (%) in lentil using various treatments in 2019-20 and 2020-21.

From the Fig. 1, the treatments involved BINA biofungicide, either alone (T_4) or in combination with other treatments (T_7), provided the highest disease reduction rates, particularly in the second year, highlighting the effectiveness of the applied treatments in managing disease incidence in lentils.

Soil amendment with both BINA phospho-vermicompost and BINA biofungicide, along with seed treatment using biofertilizer BINA-LT-17, resulted in seed germination rates of 80.73% and 82.33% during 2019-20 and 2020-21, respectively, representing a 24.56% and 19.31% increase over the control. Hannan *et al.* (2012) reported that the application of cow dung in soil, combined with seed treatment using BAU-biofungicide or BINA-biofertilizer, exhibited 81.33% seed germination. During the 2019-20 Rabi season, the lowest root rot incidence (43.33%) was recorded in plots treated with both BINA phospho-vermicompost and BINA biofungicide, combined with seed treatment using biofertilizer BINA-LT-17, which also resulted in the highest plant stand (83.03%). Soil amendment with BINA biofungicide (*Trichoderma asperellum*) was the second most effective treatment for root rot control, followed by soil amendment with BINA phospho-vermicompost along with seed treatment using biofertilizer BINA-LT-17. In the 2020-21 Rabi season, the minimum root rot incidence (10.87%) was recorded in plots treated with BINA biofungicide as a soil amendment, which resulted in the maximum plant stand (88.37%). In this season, the combination of phospho-vermicompost in soil, seed treatment with biofertilizer BINA-LT-17 and BINA biofungicide as a soil amendment ranked second. Soil amendment with only BINA phospho-vermicompost was the third most effective treatment, while seed treatment with BINA biofungicide ranked fourth. Ahmed *et al.* (2021) observed that the combined application of *Trichoderma harzianum* and *Rhizobium leguminosarum* pv. *viceae* reduced foot and root rot disease incidence in Binamasur-1 and Binamasur-2 while increasing plant fresh weight, the number of nodules per plant and nodule fresh weight. Arya *et al.* (2021) found that seeds treated with *T. harzianum* and *Pseudomonas fluorescens* resulted in the lowest disease incidence of collar rot (*Sclerotium rolfsii*) (6.66%) and maximum yield in lentil. A greenhouse experiment conducted by Kushwaha *et al.* (2018) showed that the combined application of *T. harzianum* and *T. asperellum* was more effective in controlling Fusarium rot of lentil (*Fusarium oxysporum*), reducing disease severity by 20-44% and increasing dry weight by 23-52%. Additionally, Kashem *et al.* (2011) reported that in lentils, the lowest incidence of foot and root rot (6.9%), highest seed germination (82.08%), maximum plant stand (93.12%) and highest seed yield (3,726.67 kg ha^{-1}) were achieved in plots treated with the isolate *T. harzianum* against *F. oxysporum.* The mechanisms of antagonism employed by *Trichoderma* spp. include nutrient and niche competition, antibiosis through the production of volatile and non-volatile compounds inhibitory to a range of soil-borne fungi and parasitism. Synergism between different modes of action also enhances the biocontrol efficacy of fungal pathogens under natural conditions. Environmental factors, including abiotic (e.g., soil type, temperature, pH and water potential) and biotic factors (e.g., plant species, variety and microbial activity in the soil), as well as application methods and timing, significantly influence the biological control efficacy of *Trichoderma* isolates (Akrami *et al.*, 2011). Nagamani *et al.* (2017) screened twenty *Trichoderma* isolates for efficacy against soil-borne plant pathogens (*R. bataticola*, *F. oxysporum ciceri* and *S. rolfsii*) in chickpea. The isolates *T. asperellum* (ATPU 1), *T. harzianum* (ATPP 6), *T. asperellum* (KNO 2) and *T. asperellum* (KNPG 3) were highly efficient in producing volatile and non-volatile compounds.

The study's findings underscore the importance of integrated soil amendment and seed treatment strategies in enhancing lentil crop productivity. The combined use of BINA phospho-vermicompost, BINA biofungicide and biofertilizer BINA-LT-17 not only improve seed germination and plant health but also provides effective control of root rot disease, contributing to higher yields.

Effect of BINA phospho-vermicompost, biofertilizer BINA-LT-17 and BINA biofungicide on plant height, no. of branches, no. of nodule plant^{-1} and weight of nodule plant^{-1} of lentil

The effects of various treatments on plant height, number of branches, number of nodules per plant and weight of nodules per plant in lentil were evaluated for the 2019-2020 and 2020-2021 growing seasons (Table 2). Significant differences were observed in plant height, number of nodules per plant and weight of nodules per plant across treatments, while the number of branches showed no significant variation. Plant height ranged from 37.34 cm to 42.50 cm in 2019-2020 and from 43.11 cm to 46.22 cm in 2020-2021. The tallest plants were observed with the T_5 (Soil amendment with BINA phospho-vermicompost + Seed treatment with biofertilizer BINA-LT-17) treatment in both seasons. T_7 (BINA phospho-vermicompost + BINA-LT-17 + BINA biofungicide) also showed significant highest results, reached 40.10 cm and 44.89 cm in the respective years. The control (T_8) had the shortest plants, measuring 37.34 cm in 2019-2020 and 43.11 cm in 2020-2021. The number of branches varied between 2.89 and 4.00 in 2019-2020 and between 2.89 and 3.78 in 2020-2021, with no statistically significant differences among treatments.

Table 2. Effect of BINA phospho-vermicompost, biofertilizer BINA-LT-17 and BINA biofungicide on plant height, no. of branches, no. of nodule and weight of nodule plant^{-1} of lentil

Treatments	Plant height (cm)		No. of branches		No. of nodule		Weight of nodule/ plant (g)	
	100 DAS		100 DAS		65 DAS		65 DAS	
	2019-20	2020-21	2019-20	2020-21	2019-20	2020-21	2019-20	2020-21
T_1	40.31ab	46.22a	3.33	2.89	11.67 cd	13.56cd	0.098d	0.111cd
T_2	39.78ab	42.12b	3.44	3.78	16.67ab	17.55ab	0.124b	0.135abc
T_3	40.00ab	45.22ab	3.44	3.00	12.07bcd	13.11cd	0.101cd	0.107cd
T_4	38.56ab	43.00ab	3.55	3.11	15.67bc	13.67cd	0.122bc	0.112bcd
T_5	42.50a	44.22ab	3.80	3.55	15.33bcd	17.89ab	0.120bc	0.140ab
T_6	41.37ab	43.11ab	3.44	3.11	11.33cd	15.67bc	0.094d	0.128abc
T_7	40.10ab	44.89ab	4.00	3.22	20.67a	18.85a	0.173a	0.154a
T_8 (Control)	37.34b	43.11ab	3.21	3.00	10.67d	11.22d	0.084d	0.091d
LSD (0.05)	4.03	3.34	1.27	2.04	4.67	2.67	0.02	0.03
CV %	3.50	2.64	12.42	22.05	11.38	6.09	6.49	8.15

The table shows the mean values of plant height, number of branches, number of nodules and weight of nodules per plant of lentil across two consecutive growing seasons, 2019-2020 and 2020-2021. T_1= Soil amendment with phospho-vermicompost, T_2=Seed treatment with biofertilizer BINA-LT-17 (*Rhizobium leguminosarum*), T_3= Seed treatment with BINA biofungicide (*Trichoderma asperellum*), T_4= Soil amendment with BINA biofungicide (*Trichoderma asperellum*), T_5= Soil amendment with phospho-vermicompost + Seed treatment with biofertilizer BINA-LT-17, T_6= Soil amendment with phospho-vermicompost + Seed treatment with BINA biofungicide, T_7= Soil amendment with phospho-vermicompost + Seed treatment with biofertilizer BINA-LT-17+ Soil amendment with BINA biofungicide and T_8= Control (no treatment). DAS: Days After Sowing.

The number of nodules per plant showed significant variation across treatments. The highest average number of nodules was observed in T_7 (soil amendment with BINA phospho-vermicompost + seed treatment with biofertilizer BINA-LT-17 + soil amendment with BINA biofungicide), with an average of 19.76 nodules over the two years. Treatments T_2 (seed treatment with biofertilizer BINA-LT-17) and T_5 (soil amendment with BINA phospho-vermicompost + seed treatment with biofertilizer BINA-LT-17) also resulted in significant nodule formation, averaging 17.11 and 16.61 nodules, respectively. In contrast, the control treatment (T_8) exhibited the lowest average nodule formation, with only 10.945

nodules. The weight of nodules per plant was significantly higher in T_7, with an average of 0.1635g across both seasons, followed by T_5 (0.130g) and T_2 (0.1295g). These treatments demonstrated substantial nodule weight compared to the control, which had the lowest average nodule weight of 0.0875g. The application of phospho-vermicompost, biofertilizer BINA-LT-17 and BINA biofungicide, either alone or in combination, significantly enhanced the growth of lentils (Fig. 2). Specifically, the treatment T_7 (combined treatment) consistently outperformed than other treatments, leading to increased plant height, number of branches, nodule formation and nodule weight, thereby indicating their potentiality for improving lentil productivity.

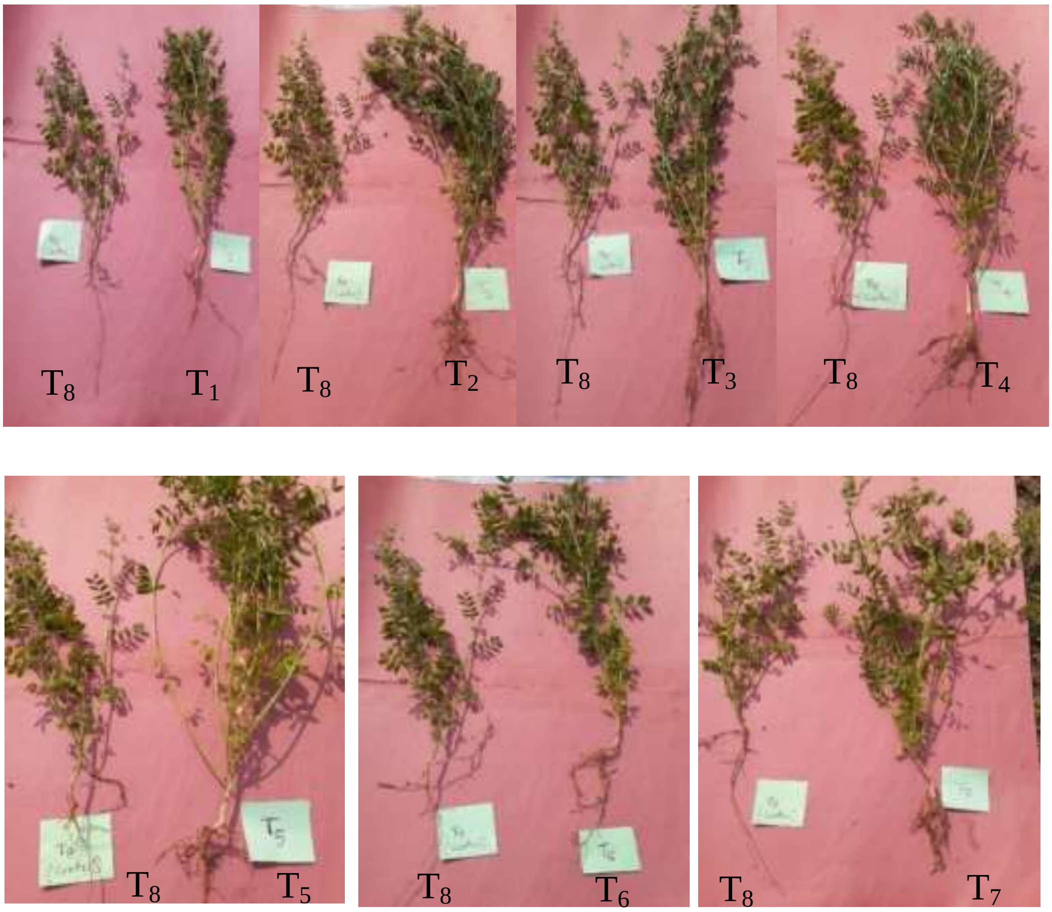

Fig. 2. Effect of BINA phospho-vermicompost, biofertilizer BINA-LT-17 and BINA biofungicide on growth and development of lentil plants.

Effect of BINA phospho-vermicompost, biofertilizer BINA-LT-17 and BINA biofungicide on dry shoot weight, root length, dry root weight, no. of pods and grain yield of lentil

Table-3 summarizes the effects of various treatments on dry shoot weight, root length, dry root weight, number of pods and grain yield of lentil over two growing seasons (2019-20 and 2020-21). This table presents the effects of various treatments, including BINA phospho-vermicompost, biofertilizer BINA-LT-17 and BINA biofungicide, applied either individually or in combination, on several growth parameters of lentils.

Table 3: Effect of BINA phospho-vermicompost, biofertilizer BINA-LT-17 and BINA biofungicide on dry shoot weight, root length, dry root weight, no. of pods and grain yield of lentil

Treatments	Dry shoot weight (g)		Root length (cm)		Dry root weight (g)		No. of Pods		Grain Yield (kg ha^{-1})	
	100 DAS		100 DAS		100 DAS		100 DAS		100 DAS	
	2019-20	2020-21	2019-20	2020-21	2019-20	2020-21	2019-20	2020-21	2019-20	2020-21
T_1	5.65cd	5.58ab	9.08ab	10.11ab	1.01ab	2.00ab	65.00bc	72.00bc	824.00cd	803.33c
T_2	5.37de	5.80ab	8.52abc	10.11ab	0.92b	2.05a	86.33ab	51.93e	931.20bc	720.83d
T_3	5.52cd	4.62b	8.55abc	10.52a	1.07ab	1.80bc	68.44bc	78.17ab	791.40de	800.83c
T_4	6.87ab	5.63ab	9.39a	9.33bc	1.53a	1.52d	85.11ab	80.63ab	903.80bcd	895.00b
T_5	5.93bcd	5.35ab	6.98c	9.28bc	0.95b	1.21e	99.67a	65.70cd	1087.50a	785.67c
T_6	6.43bc	4.48b	9.91a	9.28bc	1.00ab	1.20e	99.44a	56.10de	990.00ab	751.67d
T_7	7.95a	6.12a	7.57bc	9.44bc	1.09ab	1.66cd	85.44ab	84.65a	946.10b	990.00a
T_8 (Control)	4.26e	3.10c	9.04ab	8.44 c	0.94b	1.05e	56.60c	47.33e	691.30e	665.83e
LSD (0.05)	1.09	1.38	1.58	1.06	0.54	0.24	21.75	11.648	14.93	31.955
CV %	6.31	9.39	6.35	3.84	17.43	5.35	9.35	6.03	4.46	1.38

The Table shows the mean values of dry shoot weight, root length, dry root weight, no. of pods and grain yield of lentil across two consecutive growing seasons, 2019-2020 and 2020-2021. T_1= Soil amendment with phospho-vermicompost, T_2= Seed treatment with biofertilizer BINA-LT-17 (*Rhizobium leguminosarum*), T_3= Seed treatment with BINA biofungicide (*Trichoderma asperellum*), T_4= Soil amendment with BINA biofungicide (*Trichoderma asperellum*), T_5= Soil amendment with phospho-vermicompost + Seed treatment with biofertilizer BINA-LT-17, T_6= Soil amendment with phospho-vermicompost + Seed treatment with BINA biofungicide, T_7= Soil amendment with phospho-vermicompost+ Seed treatment with biofertilizer BINA-LT-17+ Soil amendment with BINA biofungicide and T_8= Control (no treatment).DAS: Days After Sowing.

The treatments were evaluated based on dry shoot weight (g), root length (cm), dry root weight (g), number of pods and grain yield (kg ha^{-1}) at 100 days after sowing (DAS) during the 2019-2020 and 2020-2021 growing seasons. T_1 (Soil amendment with BINA phospho-vermicompost) treatment resulted in moderate dry shoot weight and root length, with the grain yield being slightly lower than the top treatments. T_2 (Seed treatment with biofertilizer BINA-LT-17) treatment produced similar root length and dry shoot weight as T_1 but had a higher grain yield in 2019-2020. T_3 (Seed treatment with BINA biofungicide) showed a reduction in dry shoot weight and grain yield compared to T_1 and T_2. T_4 (Soil amendment with BINA biofungicide) treatment increased dry shoot weight and grain yield significantly, especially in 2020-2021. T_5 (Soil amendment with BINA phospho-vermicompost + Seed treatment with biofertilizer BINA-LT-17) resulted in the highest grain yield in 2019-2020, although the results were lower in 2020-2021. T_6 (Soil amendment with BINA phospho-vermicompost + Seed treatment with BINA biofungicide) showed improved root length (Fig. 3) and grain yield, though not as high as T_5 or T_7. T_7 (Combination of all treatments) had the highest dry shoot weight and grain yield in 2020-2021, making it the most effective combination. The control group (T_8), which received no treatment, showed the lowest results across all parameters, highlighting the efficacy of the applied treatments. The findings suggest that integrated treatments involving both soil amendments and biofertilizers (T_5 and T_7) consistently provided superior results compared to individual treatments or control.

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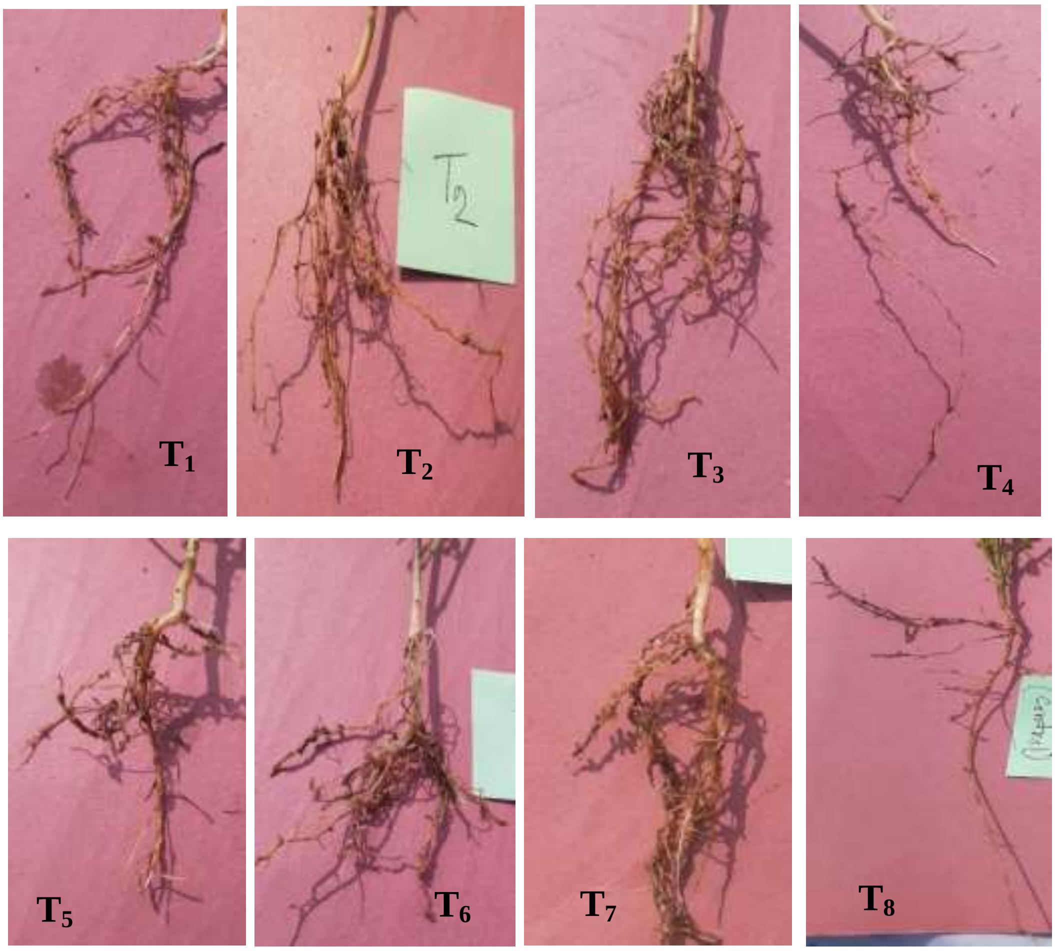

Fig. 3. Effect of BINA phospho-vermicompost, biofertilizer BINA-LT-17 and BINA biofungicide on root growth of lentil plants.

In the present study, the maximum number and weight of nodules were observed during two consecutive seasons (2019-20 and 2020-21) in plots where BINA phospho-vermicompost and BINA biofungicide were applied as soil amendments and seeds were treated with biofertilizer BINA-LT-17. A higher number of nodules was consistently found in plots where biofertilizer BINA-LT-17 was applied as a seed treatment compared to other treatments. A similar trend was observed for dry shoot weight, root length, dry root weight, number of pods and grain yield. Soil amendment with BINA phospho-vermicompost and seed treatment with biofertilizer BINA-LT-17 also played a vital role in increasing the number of pods and grain yield during one of the two growing seasons. Soil amendment with only BINA biofungicide (*Trichoderma asperellum*) also positively influenced dry shoot weight, root length, dry root weight, number of pods and grain yield. Saber *et al.* (2009) reported a synergistic effect of *Trichoderma* and *Rhizobium* in both the biocontrol of chocolate spot disease and improving the productivity of faba bean. They observed that dual inoculation of seeds with a mixture of *Rhizobium leguminosarum* and *T. viride* tag4, followed by foliar spraying of the plants with the spore suspension of *T. viride* tag4 at 35 and 55 days after sowing, reduced chocolate spot disease while enhancing nodulation, nitrogenase activity and nitrogen-fixing bacterial populations in the rhizosphere. This treatment also improved physiological activities, including photosynthetic pigments, total phenol content and polyphenol oxidase activity, resulting in enhanced plant growth and yield. On average, this method led to a 57% reduction in chocolate spot disease and a 23% increase in faba bean yield compared to untreated control plants. Khalequzzaman *et al.* (2010) found that BARI Masur *5*, when inoculated with BARI Biofertilizer Rlc 104 (*Rhizobium* sp.), had the minimum foot and root rot incidence (11.2%), which led to maximum plant survival (88.8%), the highest number of nodules per plant (18.1) and the highest grain yield (1952.9 kg ha^{-1}). In another study, Khalequzzaman (2015) observed that the use of BARI *Rhizobium* Biofertilizers in BARI Chickpea resulted in the highest plant survival rate (90.22%) and maximum grain yield (2040 kg ha^{-1}). Mohammad and Hossain (2003) reported that biofertilizers reduced foot and root rot disease incidence by 77.79% in Binamoog-3 and by 76.78% in Binamoog-4. Additionally, biofertilizers increased seed yield by up to 20.83% in Binamoog-3 and by 12.79% in Binamoog-4 compared to untreated control plots. Hannan *et al.* (2012) observed that mixing cow dung into the soil at a rate of 5 t/ha during final land preparation, combined with seed coating using BINA-biofertilizer and BAU-biofungicide (2.5% of seed weight) before sowing, resulted in 81.50% field emergence of lentil, which was 19.85% higher than the control. They also found that using BAU-biofungicide as a seed treatment alone resulted in the highest plant stand (84.82%). When BINA-biofertilizer and BAU-biofungicide were used as seed treatments alongside cow dung application as an organic nutrient source, the shoot and root lengths and dry shoot and root weights of lentil increased significantly. Furthermore, BINA-biofertilizer notably enhanced the number and weight of nodules per plant. The combined use of BAU-biofungicide, BINA-biofertilizer and cow dung as soil amendments increased the biomass production of lentil by up to 75.56% over the control.

The role of vermicomposts in disease suppression is also well-documented. Vermicomposts have been shown to effectively reduce the incidence of soil-borne diseases, such as those caused by *Pythium*, *Phytophthora*, *Fusarium* and *Rhizoctonia* (Simsek-Ersahin, 2011; Szczech *et al.,* 2002). The integration of vermicompost with nonconventional chemicals and bioagents has been shown to enhance plant defense mechanisms, against to systemic acquired resistance (Sahni *et al.,* 2008). Additionally, vermicompost amendments have been reported to improve plant growth and yield by increasing microbial diversity and activity, which contribute to pathogen suppression through both specific and general mechanisms (Edwards & Arancon, 2004; Wang *et al.*, 2021). The fortification of vermicompost with biocontrol agents, such as *Trichoderma harzianum*, *Bacillus subtilis* and *Pseudomonas fluorescens*, has been particularly effective in reducing disease incidence and promoting plant growth. Studies have shown that biofortified vermicompost not only suppresses pathogens but also enhances microbial colonization, further improving its biocontrol efficacy (Keswani *et al.*, 2014; Basco *et al.*, 2017). Recent research of You *et al.* (2019) and others has confirmed the potential of vermicompost in combination with biocontrol agents to reduce disease incidence in various crops, including cucumber and tomato (Peerzada *et al.*, 2020; Zhang *et al.*, 2020). The result revealed that, the integration of phospho-vermicompost and biofertilizers, particularly BINA-LT-17, enhanced nodulation, growth parameters and disease resistance in lentil. Findings of the present study have the tremendous importance of adopting sustainable agricultural practices that could incorporate organic amendments and biocontrol agents to optimize crop productivity and soil health.

Conclusion

Soil amendment with BINA biofungicide (*Trichoderma asperellum*) both alone or in combination with BINA phospho-vermicompost and biofertilizer BINA-LT-17, effectively reduced the root rot disease and improved the growth of lentil. However, further research could be conducted to explore the potential cost-benefit ratio of using *Trichoderma asperellum* along with BINA phospho-vermicompost and biofertilizer BINA-LT-17 under field condition for the biological control of root rot caused by *Sclerotium rolfsii* in lentil which could be eventually lead to the commercialization of this technology.

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