J. Microbiol. Biotechnol. (2012), **22**(7), 883–888
http://dx.doi.org/10.4014/jmb.1201.01008
First published online April 18, 2012
pISSN 1017-7825 eISSN 1738-8872

JOURNAL OF MICROBIOLOGY AND BIOTECHNOLOGY

Integrated Management of Foot Rot of Lentil Using Biocontrol Agents under Field Condition

Hannan, M. A.[1], M. M. Hasan[2], I. Hossain[1], S. M. E. Rahman[3,4], and Deog-Hwan Oh[4]*

[1]*Department of Plant Pathology, Bangladesh Agricultural University, Mymensingh-2202, Bangladesh*
[2]*Department of Agronomy and Agricultural Extension, Faculty of Agriculture, University of Rajshahi, Rajshahi-6205, Bangladesh*
[3]*Department of Animal Science, Bangladesh Agricultural University, Mymensingh-2202, Bangladesh*
[4]*Department of Food Science and Biotechnology and Institute of Bioscience and Biotechnology, Kangwon National University, Chuncheon, Gangwon 200-701, Korea*

Received: January 5, 2012 / Revised: March 8, 2012 / Accepted: March 12, 2012

The efficacy of cowdung, Bangladesh Institute of Nuclear Agriculture (BINA)-biofertilizer, and Bangladesh Agricultural University (BAU)-biofungicide, alone or in combination, was evaluated for controlling foot rot disease of lentil. The results exhibited that BINA-biofertilizer and BAU-biofungicide (peat soil-based *Rhizobium leguminosarum* and black gram bran-based *Trichoderma harzianum*) are compatible and have combined effects in controlling the pathogenic fungi *Fusarium oxysporum* and *Sclerotium rolfsii*, which cause the root rot of lentil. Cowdung mixing with soil (at 5 t/ha) during final land preparation and seed coating with BINA-biofertilizer and BAU-biofungicide (at 2.5% of seed weight) before sowing recorded 81.50% field emergence of lentil, which showed up to 19.85% higher field emergence over the control. Post-emergence deaths of plants due to foot rot disease were significantly reduced after combined seed treatment with BINA-biofertilizer and BAU-biofungicide. Among the treatments used, only BAU-biofungicide as the seed treating agent resulted in higher plant stand (84.82%). Use of BINA-biofertilizer and BAU-biofungicide as seed treating biocontrol agents and application of cowdung in the soil as an organic source of nutrient resulted in higher shoot and root lengths, and dry shoot and root weights of lentil. BINA-biofertilizer significantly increased the number of nodules per plant and nodules weight of lentil. Seeds treating with BAU-biofungicide and BINA-biofertilizer and soil amendment with cowdung increased the biomass production of lentil up to 75.56% over the control.

Keywords: Integrated management, foot rot, lentil, biocontrol agents, field condition

**Corresponding author*
Phone: +82-33-250-6457; Fax: +82-33-241-0508;
E-mail: deoghwa@kangwon.ac.kr

Different phytopathogenic soilborne as well as seedborne fungi are responsible for disease development of pulses, which attack plants during seedling to maturity stages and are more destructive at the seedling stage [8]. Foot rot (causal agents *F. oxysporum* and *S. rolfsii*) is considered as an important and destructive disease of pulses in almost all legume-growing countries of the world [1].

Control of the soilborne pathogens *F. oxysporum* and *S. rolfsii* with chemicals is practically difficult. On the other hand, indiscriminate use of chemicals causes environmental pollution and health hazards [9]. Nowadays, 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 [18]. *Trichoderma* may be used as an ecofriendly biocontrol agent in this regard. The biocontrol agent *Trichoderma* has the potential to protect seedlings against several plant pathogenic fungi. *Trichoderma* spp. have been widely used as antagonistic fungal agents against several pests as well as plant growth enhancers [28]. *Trichoderma harzianum* has been reported to be effective in controlling seed- and soilborne diseases of different crops, namely legumes and vegetables [2, 6, 12, 20, 24, 26]. The use of antagonistic bacteria as a biological control means may provide a great alternative for plant pathologists [11]. Hossain *et al.* [11] also reported prominent antagonistic effect of *Rhizobium* against foot and root rot pathogens (*F. oxysporum* and *S. rolfsii*) of pulses. This antagonist also increased the percentage of seedling emergence, plant height, fresh weight, and vigour index [12, 22]. It has been reported that rhizobial strains have significant effect in reducing the severity of foot and root of chickpea [16]. *Rhizobium* spp. and *Trichoderma* sp. are compatible and have combined effects in controlling the fungi *F. oxysporum* and *S. rolfsii*, which caused the root rot of lentil [21]. Application of cowdung and manures in the soil is aimed to supply nutrients to the crops, creating a positive environment of inducing disease resistance to the plant. As a result, plants may recover from the disease or be resistant to disease, or overcome the disease epidemic [14, 17].

Considering the above facts the present study was undertaken to find out the effect of cowdung, BINA-biofertilizer (peat-based *R. leguminosarum*), and BAU-biofungicide (organic substrate-based *T. harzianum*), either alone or in combination on foot rot disease of lentil under field conditions.

Materials and Methods

Collection of BINA-Biofertilizer

Peat-based biofertilizer was collected from the Soil Microbiology Laboratory of the Bangladesh Institute of Nuclear Agriculture (BINA), Mymensingh, Bangladesh. The composition of the biofertilizer was peat soil and *Rhizobium leguminosarum*. The *R. leguminosarum* was firstly collected from the nodules of legume plants. After collection, *Rhizobium* broth was prepared. Sterilized 500 g peat soil was poured in a polythene bag and inoculated with a previously prepared 5 ml broth of *R. leguminosarum* (10^{11} CFU/ml) and mixed thoroughly for proper distribution. Then the materials were incubated for 7 days at $25 \pm 2^{o}C$. After 7 days of incubation, it was ready for use. The material can be stored up to 6 months at $22 \pm 1^{o}C$ for future use.

Collection of BAU-Biofungicide

BAU-biofungicide was collected from the Disease Resistance Laboratory, Department of Plant Pathology, BAU, Mymensingh, Bangladesh. BAU-biofungicide was invented from a naturally occurring fungus, *Trichoderma harzianum* growing on an organic substrate (black gram bran). The *T. harzianum* was firstly collected from the rhizosphere of a lentil field. The isolated fungi were cultured on potato dextrose agar (PDA) at $28 \pm 2^{o}C$. Then 50 g of black gram bran was moistened with 1.5 ml of water and sterilized in an autoclave. After cooling, the sterilized substrate was inoculated with previously prepared 7-day-old culture of *T. harzianum* (four to five 1 cm block of *T. harzianum*). After 7 days of incubation, it was ready for use. The material can be stored up to 6 months at $22 \pm 1^{o}C$ for future use.

Collection of Seeds of Pulse

Seed samples of lentil variety BINA Masur-1 were collected from the Bangladesh Institute of Nuclear Agriculture (BINA), Mymensingh, Bangladesh. The collected seeds were kept in a paper bag and stored in the refrigerator at $5-7^{o}C$ for one month for subsequent studies.

Seed Treatment

Required amounts of seeds were taken in a beaker and a few drops of water were added for moistening the seed surface uniformly to allow maximum adherence of BAU-biofungicide on the whole surface of seeds. Seeds were treated with BAU-biofungicide at 2.5% weight of seeds until the whole surface of the seeds were coated, where 2.5% biofungicide contained 10^6 conidia [19]. For seed coating with biofertilizer, seeds were initially moistened with water. Then the seeds were thoroughly mixed with biofertilizer (at 2.5% of seed weight) where the biofertilzer contained 10^8 *Rhizobium* cells/mg formulations. The inoculant-coated seeds were placed in a cool and dry place under shade for drying. The treated seeds were kept in paper bags and stored in the refrigerator at $5-7^{o}C$ for one month for subsequent studies.

In the present study, the following treatments were used:

Control
Cowdung
BINA-biofertilizer
BAU-biofungicide
Cowdung + BINA-biofertilizer
Cowdung + BAU-biofungicide
BINA-biofertilizer + BAU-biofungicide
Cowdung + BINA-biofertilizer + BAU-biofungicide

Cowdung (5 t/ha) was mixed with the soil during the final land preparation as per treatment specification [15].

Field Experiment

Seed sowing. The field experiment was conducted in a randomized block design with 3 replications. The size of the individual plot was 2 m × 1 m, and the spaces between the plots and blocks were 1 m and 1 m, respectively. Treated seeds were sown (35 kg/ha) in lines about 2.0 cm in depth and the seeds were immediately covered with soil. Two times of weeding were preformed, one after 25 days and another 40 days after sowing. No plant protecting chemicals (insecticides or fungicides) were applied in the field.

Determination of foot rot disease. The experimental plots were inspected routinely to observe the foot rot disease on plant. In case of complexity to identify the disease, symptoms-bearing plants were collected from the field using polythene bag and brought to the Disease Resistance Laboratory, Department of Plant Pathology, BAU, Mymensingh. From the infected plants, the fungi were isolated following tissue planting methods [4]. After incubation, the fungi that grew over potato dextrose agar (PDA) were purified by the hyphal tip culture method. The isolated fungi were identified as *F. oxysporum* and *S. rolfsii* according to reference mycology books and manuals [3, 5, 25]. The pure cultures of the fungi were preserved in PDA slants at 4°C in the refrigerator as stock culture for future use.

Data collection and analysis. Data on different parameters (*viz*., germination, post-emergence death of plants, plant stand, shoot length, root length, dry shoot weight, dry root weight, number of nodule/plant, weight of nodule/plant, and biomass) were taken. Five plants were randomly selected and uprooted carefully from each plot for recording data. Data were expressed as the means ± standard errors. The results were analyzed using the SPSS statistical package (ver. 16, SPSS Inc., Chicago, IL, USA) and Microsoft excel program. An analysis of variance (ANOVA) was used to evaluate the treatment. Duncan's Multiple Range Test was used to determine the significant difference at $p \leq 0.05$.

Results

Effect of Cowdung, BINA-Biofertilizer, and BAU-Biofungicide Either Alone or in Combination on Germination, Post-Emergence Death of Plants, and Plant Stand of Lentil

The effects of cowdung, BINA-biofertilizer, and BAU-biofungicide either alone or in combination on germination of lentil (var. BINA Musur-1) is shown in Table 1. The

Table 1. Effect of cowdung, BINA-biofertilizer, and BAU-biofungicide either alone or in combination on germination, post-emergence death, and plant stand of lentil.

Treatment	Germination (%)	Post-emergence death of plants (%)	Plant stand (%)
Control	68.0 ± 2.116b	36.26 ± 1.0a	63.07 ± 2.36d
Cowdung	78.17 ± 0.09a	24.56 ± 1.59b	75.43 ± 2.06bc
BINA-biofertilizer	78.83 ± 0.67a	20.07 ± 1.13c	79.05 ± 0.59b
BAU-biofungicide	81.33 ± 0.87a	15.79 ± 1.03d	84.82 ± 2.11a
Cowdung + BINA-biofertilizer	79.67 ± 0.19a	20.18 ± 0.53c	70.56 ± 1.28c
Cowdung + BAU-biofungicide	80.67 ± 1.92a	17.37 ± 0.21d	84.08 ± 2.28a
BINA-biofertilizer + BAU-biofungicide	80.0 ± 2.88a	15.11 ± 0.06d	82.96 ± 1.73a
Cowdung + BINA-biofertilizer + BAU-biofungicide	81.50 ± 1.76a	16.80 ± 0.16d	84.65 ± 1.74a
Significance level (P)	**	***	***

***, P<0.001; **, P<0.01.
The data are mean ± standard error from a representative experiment. Data represent the mean of three replications.
The experiment was repeated twice. Mean values with same letters in a column are not significantly different: Duncan's Multiple Range Test; P<0.05.

treatments resulted in significant effects on the germination of seeds of lentil. The highest germination (81.50%) was recorded by treating seeds with BAU-biofungicide and BINA-biofertilizer and application of cowdung in the soil as an organic source of nutrient, and the result was statistically identical with cowdung, BINA-biofertilizer, BAU-biofungicide, cowdung + BINA-biofertilizer, cowdung + BAU-biofungicide, and BINA-biofertilizer + BAU-biofungicide, whereas the lowest germination (68.0%) was recorded in the control. The post-emergence death of BINA-Masur-1 plants due to foot rot was found to be caused by *F. oxysporum* and *S. rolfsii*. The minimum post-emergence death of plants (15.11%) was obtained by treatment of seeds with BAU-biofungicide and BINA-biofertilizer, which was followed by BAU-biofungicide, cowdung + BAU-biofungicide, and cowdung + BINA-biofertilizer + BAU-biofungicide, respectively. The maximum plant stand (84.82%) was obtained by seed treatment with BAU-biofungicide, which was statistically identical with the combinations of seed treatment with BAU-biofungicide and application of cowdung in the soil (84.08%), seed treatment with BINA-biofertilizer + BAU-biofungicide (82.96%) and applying cowdung in the soil, as well as seed treatment with BINA-biofertilizer + BAU-biofungicide (84.65%), respectively. The lowest plant stand (63.06%) was recorded in the control.

Effect of Cowdung, BINA-Biofertilizer and BAU-Biofungicide Either Alone or in Combination on Shoot and Root Lengths of Lentil (Variety BINA Masur-1)

The effects of cowdung, BINA-biofertilizer, and BAU-biofungicide either alone in combination were evaluated

Table 2. Effect of cowdung, BINA-biofertilizer, and BAU-biofungicide either alone or in combination on length of shoot and root of lentil.

Treatment	Shoot length (cm)			Root length (cm)		
	60 DAS	80 DAS	110 DAS	60 DAS	80 DAS	110 DAS
Control	15.13 ± 0.50b	37.13 ± 0.45c	38.0 ± 1.73c	5.66 ± 0.66c	5.76 ± 0.53c	6.45 ± 0.25c
Cowdung	15.13 ± 0.13b	39.27 ± 1.88c	39.60 ± 1.02bc	6.0 ± 0.28bc	6.40 ± 0.23bc	7.0 ± 0.57bc
BINA-biofertilizer	16.0 ± 1.04b	40.13 ± 1.22c	44.77 ± 2.30b	7.13 ± 0.13ab	7.33 ± 0.33b	7.60 ± 0.44abc
BAU-biofungicide	24.0 ± 1.52a	58.53 ± 1.73a	59.0 ± 1.73a	7.0 ± 0.14ab	7.46 ± 0.26b	7.97 ± 0.56abc
Cowdung + BINA-biofertilizer	14.98 ± 0.88b	44.93 ± 2.09b	45.20 ± 1.29b	6.76 ± 0.46abc	7.53 ± 0.30b	7.88 ± 0.30abc
Cowdung + BAU-biofungicide	24.10 ± 0.55a	57.0 ± 1.15a	58.0 ± 1.65a	6.02 ± 0.02bc	7.53 ± 0.41b	8.33 ± 0.76ab
BINA-biofertilizer + BAU-biofungicide	23.73 ± 0.73a	57.0 ± 1.73a	58.70 ± 1.40a	6.44 ± 0.43bc	7.20 ± 0.20b	7.77 ± 0.44abc
Cowdung + BINA-biofertilizer + BAU-biofungicide	25.0 ± 0.57a	58.67 ± 0.88a	60.0 ± 2.85a	7.85 ± 0.49a	9.13 ± 0.65a	9.33 ± 0.70a
Significance level (P)	***	***	***	***	***	***

***, P<0.001.
The data are mean ± standard error from a representative experiment. Data represent the mean of three replications.
The experiment was repeated twice. Mean values with same letters in a column are not significantly different: Duncan's Multiple Range Test; P<0.05.

regarding shoot length of plants (Table 2). The effects of different treatments on shoot length of BINA masur-1 at 60 DAS, 80 DAS, and 110 DAS showed marked variation. In the case of 60, 80, and 100 DAS, maximum shoot lengths of 25.0 cm, 58.67 cm, and 60.0 cm, respectively, were found after application of cowdung in the soil and treating seeds with BAU-biofungicide and BINA-biofertilizer, whereas the minimum shoot lengths at 60, 80, and 100 DAS were 15.13 cm, 37.13 cm, and 38.07 cm, respectively, in the control. It has been observed that the length of roots at 60, 80, and 110 DAS varied significantly under different treatments. The highest root lengths at 60 DAS, 80 DAS, and 110 DAS were 7.85 cm, 9.13 cm, and 9.33 cm, respectively, when the seeds were treated with BAU-biofungicide and BINA-biofertilizer and soil was incorporated with cowdung. On the other hand, the lowest root lengths of 5.66 cm, 5.76 cm, and 6.45 cm were recorded at 60 DAS, 80 DAS, and 110 DAS under the control. It was evident that all the treatments increased the root lengths of plant over the control.

Effects of Cowdung, BINA-Biofertilizer, and BAU-Biofungicide Either Alone or in Combination on Dry Shoot and Root Weight, Number of Nodule/Plant, and Weight of Nodule/Plant of Lentil

Except for the cowdung, most of the treatments (*viz*., BINA-biofertilizer, BAU-biofungicide, cowdung + BINA-biofertilizer, cowdung + BAU-biofungicide, BINA-biofertilizer + BAU-biofungicide, and cowdung + BINA-biofertilizer + BAU-biofungicide) significantly increased the dry shoot weight of lentil over the control at 80 DAS and 110 DAS, respectively (Table 3). Cowdung, BINA-biofertilizer, and BAU-biofungicide in combination showed increase in dry root weight of the lentil variety. Maximum dry root weights of 1.92 g and 0.84 g at 80 DAS and 110 DAS were recorded when seeds were treated with BAU-biofungicide and BINA-biofertilizer and soil amendment with cowdung. Minimum dry root weight (0.65 g) at 110 DAS was recorded in the control. Cowdung, BINA-biofertilizer, and BAU-biofungicide either alone or in combination showed significant effects on the number of nodule/plant. The maximum number of nodule (13.80) at 60 DAS was obtained by treating seeds with BINA-biofertilizer, which was followed by seed treatment with BINA-biofertilizer and BAU-biofungicide and applying cowdung in the soil. On the other hand, the minimum number of nodules (8.8) was observed under control treatment. It was observed that treating seeds with BINA-biofertilizer increased the number of nodule/plant up to 56.81% over the control. Maximum weight of nodules (0.113 and 0.114 g) at 60 DAS was recorded when the seeds were treated with BINA-biofertilizer alone or in combination with BAU-biofungicide and soil application of cowdung, whereas the lowest weight of nodules (0.053 g) was observed in the control.

Cowdung, BINA-biofertilizer, and BAU-biofungicide either alone or in combination showed significant effects on biomass production of the lentil variety (Fig. 1). The maximum biomass production (6.25 t/ha) was recorded by applying cowdung in soil and treating seeds with BINA-biofertilizer and BAU-biofungicide. On the other hand, the

Table 3. Effects of cowdung, BINA-biofertilizer, and BAU-biofungicide either alone or in combination on dry shoot weight and dry root weight of lentil.

Treatment	Dry shoot weight (g)		Dry root weight (g)		Number of nodule/ plant	Weight of nodule/ plant (g)
	80 DAS	110 DAS	80 DAS	110 DAS	60 DAS	60 DAS
Control	4.95 ± 0.25b	4.80 ± 0.46c	1.20 ± 0.05d	0.65 ± 0.02c	8.80 ± 0.46e	0.053 ± 0.00e
Cowdung	6.0 ± 0.57b	5.62 ± 0.35c	1.20 ± 0.05d	0.75 ± 0.02abc	9.37 ± 0.21cd	0.084 ± 0.002d
BINA-biofertilizer	7.85 ± 0.49a	6.74 ± 0.12b	1.50 ± 0.14c	0.76 ± 0.03abc	13.80 ± 0.11a	0.113 ± 0.001a
BAU-biofungicide	8.85 ± 0.43a	7.81 ± 0.46a	1.90 ± 0.05ab	0.82 ± 0.01ab	10.67 ± 0.38c	0.094 ± 0.002c
Cowdung + BINA-biofertilizer	7.70 ± 0.46a	6.84 ± 0.19b	1.65 ± 0.12bc	0.70 ± 0.05bc	12.92 ± 0.04ab	0.109 ± 0.00a
Cowdung + BAU-biofungicide	8.80 ± 0.23a	7.82 ± 0.23a	1.89 ± 0.05ab	0.80 ± 0.02ab	12.73 ± 0.13b	0.101 ± 0.001b
BINA-biofertilizer + BAU-biofungicide	8.84 ± 0.25a	7.80 ± 0.11a	1. 90 ± 0.15ab	0.83 ± 0.01a	12.67 ± 0.38b	0.110 ± 0.002a
Cowdung + BINA-biofertilizer + BAU-biofungicide	8.88 ± 0.39a	7.87 ± 0.07a	1.92 ± 0.02a	0.84 ± 0.05a	13.53 ± 0.30a	0.114 ± 0.001a
Significance level (P)	***	***	***	**	***	***

***, P<0.001; **, P<0.01. The data are mean ± standard error from a representative experiment. Data represent the mean of three replications. The experiment was repeated twice. Mean values with the same letters in a column are not significantly different: Duncan's Multiple Range Test; P<0.05.

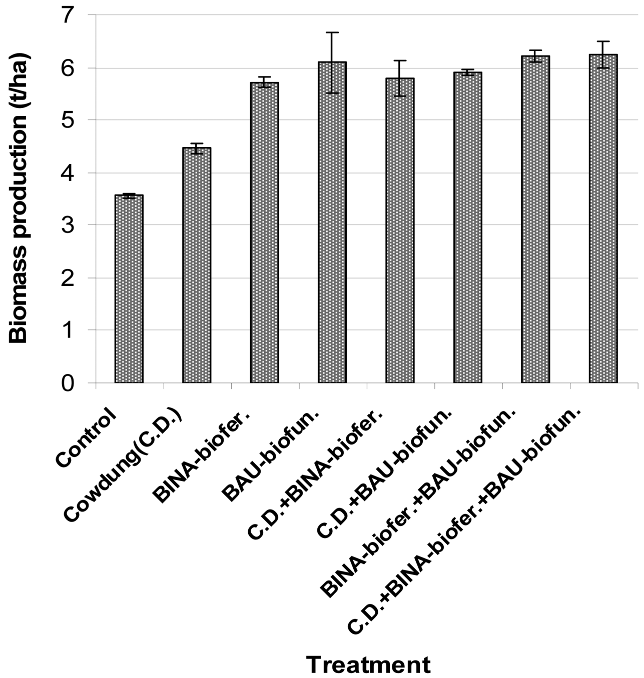

Fig. 1. Effects of cowdung, BINA-biofertilizer, and BAU-biofungicide either alone or in combination on biomass production of lentil at harvest.

Treatments = Control, Cowdung (C.D.), BINA-biofertilizer, BAU-biofungicide, C.D. + BINA-biofertilizer, C.D. + BAU-biofungicide, BINA-biofertilizer + BAU-biofungicide, and C.D. + BINA-biofertilizer + BAU-biofungicide. Data represent the mean ± SE of three replications per treatment. The experiment was repeated twice.

minimum biomass production (3.56 t/ha) was recorded in the control. It has been observed that applying cowdung in the soil and seed treated with BINA-biofertilizer and BAU-biofungicide increased biomass production up to 75.56% over the control.

DISCUSSION

The field experiments were carried out to find out the effects of cowdung, BINA-biofertilizer, and BAU-biofungicide either alone or in combination in controlling foot rot disease of lentil. It has been observed that application of cowdung in the soil and seed treatment with BINA-biofertilizer and BAU-biofungicide either alone or in combination have a great effect on germination of seeds, post-emergence death of plants, plant stand, shoot and root length, dry weight of shoot and root, number of nodule/plant, weight of nodule/plant, and biomass production of lentil. Application of cowdung in the soil and seed treatment with BAU-biofungicide either alone or in combination with BINA-biofertilizer exhibited 81.33% seed germination, which indicated a 19.85% increased seed germination over the control. Hossain and Fakir [10] used antagonist *Trichoderma* as the seed-treating agent and obtained increased number of germination of okra, bottle gourd, sweet gourd, white gourd, sanke gourd, cucumber, and tomato. In another study, increased germination of sweet gourd seed up to 13% over control with the same antagonist has also been observed [13, 22].

Post-emergence death of plants of lentil due to foot rot (*F. oxysporum* and *S. rolfsii*) was found to be reduced by treating seeds with BAU-biofungicide and BINA-biofertilizer either alone or in combination. The mycoparasitism of both *T. harzianum* and *T. longibrachitum* on *Fusarium oxysporum* f. sp. *phaseoli* causing wilt disease in beans has been studied. The pathogen was completely engulfed by the hyphae of *Trichoderma* and treated seeds reduced the post-emergence death of seedling [29]. Another study observed that seeds of chickpea treated with rhizobial inoculants resulted in significant effect in reducing the severity of foot and root rot up to 70.16% [16]. The plant stand of the lentil has been found to increase owing to the use of cowdung, BINA-biofertilizer, and BAU-biofungicide either alone or in combination. Among the treatments, BAU-biofungicide increased the plant stand of lentil up to 34.48% over the control, which was followed by seed treatment with BAU-biofungicide and BINA-biofertilizer and soil application of cowdung. Application of cowdung in the soil and use of BINA-biofertilizer and BAU-biofungicide as seed treating biocontrol agent displayed higher shoot length, root length, dry shoot weight and root weight, number of nodule/plant, and weight of nodule/plant over the control. This study is in accordance with the findings of Shoresh *et al.* [24] who recorded mycoparasitism of *Trichoderma* against the chickpea wilt caused by *F. oxysporum* and found that the plant becomes healthy and the growth of chickpea roots, shoots, and leaves was enhanced. In another experiment, it has been found that Mung (*Vinga radiata* L.) seed inoculation with rhizobium increased nodulation (in terms of their number and dry weight), over the control [23]. Application of *Trichoderma* as a seed-treating agent resulted in higher shoot length, root length, and shoot weight of vegetable seedlings [12, 13, 21]. Biomass production of lentil has been found to be increased owing to use of cowdung, BINA-biofertilizer, and BAU-biofungicide either alone or in combination with one another. Among the treatments used in the present study, combined use of cowdung, BINA-biofertilizer, and BAU-biofungicide resulted in higheist biomass production of lentil by 75.56% in comparison with the control. This finding is also supported by many researchers [7, 11, 23, 27].

From the present study, we observed that combined use of cowdung, BINA-biofertilizer, and BAU-biofungicide showed a profound effect in reducing root rot disease and in increasing plant growth parameters of lentil under field conditions. It has been clearly pointed out that BAU-biofungicide in combination with cowdung and biofertilizer can successfully be used for cultivation of lentil.

References

1. Ahmed, H. U. 1985. Disease problems of pulse and oil seed crops in Bangladesh. A paper presented in the 1st National Phytopathology. *Conf. BARI Gazipur*.
2. Anonymous. 2003. *Use of BAU-Biofungicide to Control Seed Borne Diseases of Vegetables and Pulses*. Agricultural technologies developed by Bangladesh Agricultural University Research System (BAURES), Mymensingh.
3. Barnett, H. L. and B. B. Hunter. 1972. *Illustrated Genera of Imperfect Fungi*. 3rd Ed. Burges Co., Minneapolis, USA.
4. Baxter, A. P., I. H. Rong, C. Roux, and E. J. Van der Linde. 1999. *Collecting and Preserving Fungi-A Manual for Mycology.* Plant Protection Research Institute. Private Bag X134, Pretoria, 0001 South Africa.
5. Booth, C. 1971. *The Genus Fusarium*. Commonwealth Mycology Institute Kew, Survey, England.
6. Chowdhury, M. S. M., I. Hossain, G. A. Fakir, F. M. Aminuzzaman, and M. R. Islam. 2000. Tolerance of *Trichoderma harzianum* and *Gliocladium viride* to agrochemicals and their antagonistic effect on seed borne mycoflora of pigeon pea. *Bangladesh J. Seed Sci. Tech*. **4:** 83–86.
7. Devi, H. J., T. K. Maity, and N. C. Paria. 2003. Effect of different sources of nitrogen on yield and economics of cabbage. *Environ. Ecol*. **21:** 878–880.
8. Fakir, G. A. 1983. Status of research on pulse disease at the BAU, Department of Plant Pathology BAU, Mymensingh.
9. Gerhardson, B. 2002. Biological substitutes for pesticides. *Trends Biotechnol*. **20:** 338–343.
10. Hossain, I. and G. A. Fakir. 2001. *Biological Control of Seed Borne Pathogens of Some Crops*, 1st Ed. Seed Pathology Laboratory (SPL), Bangladesh Agricultural University (BAU), Mymensingh, Bangladesh.
11. Hossain, I., M. A. I. Khan, and A. K. Podder. 1999. Seed treatment with *Rhizobium* in controlling *Fusarium oxysporum* and *Sclerotium rolfsii* for biomass and seed production of lentil (*Lens cuilinaris* M.). *Bangladesh J. Environ. Sci*. **5:** 61–64.
12. Hossain, I. and M. H. A. Naznin. 2005. BAU biofungicide in controlling seedling disease of some summer vegetables. *BAU Res. Progr*. **15:** 32–35.
13. Hossain, I. and S. M. Shamsuzzaman. 2003. Developing *Trichoderma* based bio-fungicide using agro-waste. *BAU Res. Prog*. **14:** 49–50.
14. Hossain, I., G. Yahia, and M. Jahiruddin. 1995. Effect of copper boron and molybdenum on leaf spot disease and grain set of wheat. *Bangladesh J. Training Dev*. **8:** 77–81.
15. Islam, S. M. 1995. *Crop Nutrition and Nutrient Deficiency Symptom*. Division of Soil Science, Bangladesh Agricultural Research Institutes, Joydebpur, Gazipur.
16. Kibria, M. G. and I. Hossain. 2004. Seed treatment with biofertilizer and rhizobial strains in controlling foot rot of chickpea. *Bangladesh J. Seed Sci. Tech*. **8:** 49–52.
17. Kundu, S., I. Hossain, and M. Jahiruddin. 1996. Effect of boron on leaf blotch (*Bipolaris sorokiniana*) and grain yield of wheat cv. Kanchan. *Progress. Agric*. **7:** 171–175.
18. Kwok, O. C. H., P. C. Fahy, H. A. J. Hoitink, and G. A. Kuter. 1987. Interaction between bacteria and *Trichoderma hamature* in suppression of *Rhizoctonia* damping-off in bark compost media. *Phytopathology* **77:** 1206–1212.
19. Naznin, H. A. and I. Hossain. 2004. Effect of BAU-biofungicide on germination and seedling vigour of some summer vegetables. *Bangladesh J. Seed Sci. Tech*. **8:** 85–90.
20. Ozbay, N. and S. E. Newman. 2004. Biological control with *Trichoderma* spp. with emphasis on *T. harzianum. Pakistan J. Biol. Sci*. **7:** 478–484.
21. Shaban, W. I. and M. A. El-Bramawy. 2011. Impact of dual inoculation with *Rhizobium* and *Trichoderma* on damping off, root rot diseases and plant growth parameters of some legumes field crop under greenhouse conditions. *Int. Res. J. Agric. Sci. Soil Sci*. **1:** 98–108.
22. Shamsuzzaman, S. M., A. Islam, and I. Hossain. 2003. Production of *Trichoderma* conidia in agro-wastes. *Bangladesh J. Environ. Sci*. **9:** 146–150.
23. Sharma, L. C., A. K. Bhargava, and C. Jagdesh. 1995. Seed dressing fungicides in combination with *Rhizobium in* Moog (*Vigna radiate* L.) Wilezek and their effect on nodulation and yield. *Indian J. Mycol. Plant Pathol*. **25:** 42.
24. Shoresh, M., I. Yedidia, and I. Chet. 2005. Involvement of Jasmonic acid/ethylene signaling pathway in the systemic resistance induced in cucumber by *Trichoderma asperellum* T_2O_3. *Phytopathology* **95:** 76–84.
25. Singh, R. S. 1982. *Plant Pathogens*. *The Fungi*. Oxford and IBM Publishing Co., New Delhi.
26. Sultana, N., M. S. M. Chowdhury, and I. Hossain. 2001. Growth and storability of *Trichoderma harzianum* and its effect on germination of tomato seeds. *Bangladesh J. Seed Sci. Tech*. **5:** 117–121.
27. Sultana, N. and I. Hossain. 1999. Biological control of foot and root rot of lentil with *Trichoderma harzianum*. *Bangladesh J. Seed Sci. Tech*. **3:** 107–111.
28. Verma, M., K. Satinder, R. D. Brar, R. Y. Tyagi, and J. R. Valero. 2007. Antagonistic fungi, *Trichoderma* spp.: Panoply of biological control. *Biochem. Eng. J*. **37:** 1–20.
29. Younis, N. A. 2005. Mycoparasitism of *Trichoderma harzianum* and *Trichoderma longibrachiatum* on *Fusarium oxysporum* f.sp. *phaseoli* the causal of bean wilt disease. *Bull. Faculty Agric. Cairo Univ*. **56:** 201–219.