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Ecofriendly Disease Management of Lentil (*Lens culinaris*) Seedlings

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Abstract

To determine the effect of Biofertilizer and Integrated Pest Management (IPM) biopesticide for controlling foot and root rot diseases of lentil it is very important for conducting experiment in the field. It was marked that both Biofertilizer and IPM Biopesticide found significantly lower disease incidence of Bangladesh Institute of Nuclear Agriculture (BINA) released popular lentil variety Binamasur-1 and Binamasur-2 seedlings compared with the control treatment. Soil treatment with Biofertilizer as Bangladesh Agricultural Research Institute (BARI) released Bari-Biofertilizer performed the lowest disease incidence of lentil variety Binamasur-1 and Binamasur-2 at 20 days after sowing (DAS) showed a reduction of disease incidence up to 68.80% and 71.70% over the control. While after 28 DAS, it was found up to 91.27% and 91.34% reduction of disease incidence over control. Furthermore, after 35 DAS, it exhibited up to 69.37% and 69.28% reduction of disease incidence over control. Bari-Biofertilizer significantly increased the fresh weight and the number of nodules per plant.

Keywords

Ecofriendly, Disease, Biofertilizer, Biopesticide, Foot Rot

1. Introduction

Lentil (*Lens culinaris*) is a direct source of protein for human beings and also for animals in the country [1]. It is one of the principal legume crops and is considered one of the staple protein sources for the people of Bangladesh. Lentil is especially honored as a cheap protein source in comparison with high-cost animal protein. It is also considered as rural people's meat [2]. Lentil contains about

25% when uncooked, decreasing to about 10% when cooked, due to the extra water absorbed [3]. They are also a very good source of folate and B vitamins. The price of animal protein is increasing and availability is decreasing day by day, therefore the protein shortage in the diet of the people in this country can be met up through improvement of lentil varieties for increasing the production. Moreover, legumes have the remarkable quality of fixing the symbiotic nitrogen having a relationship with *Rhizobia* and improving soil fertility. Lentil is cultivated in many countries in the world, including Canada, much of Asia, Australia and Southern Europe; Canada is the biggest exporter of lentil [3]. Lentil is an important pulse crop of Asia-Pacific region (which covers about 53% and produces 49% of world's lentil [3]. Among the legumes, lentil possesses the first position for its wide uses in this country. Lentil is cultivated all over the country of about 141,296 ha with an average yield of 1.25 t/ha [4]. The main genetic and cultural factors contributing to the low productivity of lentil in Bangladesh include low yield potential of local cultivars, Local cultivars' susceptibility to major diseases.

There are various causes associated with a lower yield of pulses in the country, where diseases play one of the most important factors. Many phytopathogenic soil-borne and seed-borne fungi are responsible for disease development which damage plants during seedling to reproductive stages. Especially fungi are more destructive at the seedling stage. Foot and root rot (caused by *Fusarium oxysporum* and *Sclerotium rolfsii*) is considered the most common and destructive disease of pulse crops in Bangladesh and also in almost all legume growing countries of the world [5] [6].

Chemical control of soil-borne *F. oxysporum* and *S. rolfsii* is costly, time consuming and impractical. A hundred percent lentil mortality occurs in Bangladesh due to foot and root rot and wilt-infected plants [6] [7]. Indiscriminate uses of chemicals also cause environmental pollution and health hazards. The use of antagonistic bacteria as biological control means may provide an alternative for plant pathologists. *Trichoderma harzianum* is an effective biocontrol agent of soil and seed-borne plant pathogenic fungi [8] [9] [10]. Seed treatment with *Rhizobium leguminosarum* pv. *viceae* was effective to reduce damping-off and increasing seedling height, root nodule mass, root biomass, shoot biomass, and seed yield of pea and lentil [11]. *T. harzianum* has been reported as an effective biocontrol agent of soil and seed-borne plant pathogenic fungi [8] [9] [10]. Practically development of disease resistant variety requires seven to eight years and it is possible to reduce economic loss by applying the bio-control means. The present study was undertaken to observe the effect of Bari-Biofertilizer and IPM Biopesticide on foot and root rot disease of lentil under field condition.

2. Materials and Methods

Lentil seeds of Binamasur-1 and Binamasur-2 were collected from Bangladesh Institute of Nuclear Agriculture, Mymensingh. IPM-Biopesticide (based on *Tri-*

choderma sp.) and Bari-Biofertilizer were collected from IPM Laboratory, Department of Plant Pathology, BAU, Mymensingh and Bangladesh Agricultural Research Institute (BARI), Joydevpur, Gazipur, respectively.

This experiment was conducted in the field of Department of Plant Pathology, Bangladesh Agricultural University (BAU), Mymensingh from November 2019 to April 2020. The land was medium-high with loamy soil texture having pH range of 5.5 - 6.8, low organic matter content and medium K (Potassium) minerals. Soil texture of the experimental plots was sandy loam to Sonatola soil series under Old Brahmaputra Flood Plain AEZ (Agro-ecological Zone). The experiment was laid out in a Randomized Complete Block Design with four replications with a unit plot size of 6 m × 2 m. The space for line to line was 30 cm and plant to plant was 12 cm. The treatments were: 1) T_1 = Control, 2) T_2 = IPM Biopesticide as soil treatment, 3) T_3 = IPM Biopesticide in lines at sowing time, 4) T_4 = IPM Biopesticide as seed treatment, 5) T_5 = Bari-Biofertilizer as seed treatment and 6) T_6 = Bari-Biofertilizer in lines at sowing time.

The Biopesticide was mixed with top soil @ 6.4 g/m^2 at seven days before sowing. At the time of sowing Biopesticide was given @ 1 g/line. For seed treatment required amount of seeds were taken in a beaker and a few drops of water was added for moistening the seed surface uniformly to allow maximum adherence of the IPM Biopesticide on the whole surface of seeds. Seeds were then treated with IPM Biopesticide @ 4% of seed weight until the whole surfaces of seeds were coated with the IPM Biopesticide. Similarly, seeds of lentil were treated with Biofertilizer @ 4% of seed weight and the required amount of Biofertilizer (@2 kg/ha) was given in lines at the time of sowing. Seeds were sown in lines at about 5 cm depth @ 25 kg/ha. Three times weeding was done at 15, 30 and 45 DAS (Days after sowing). No irrigation was done in the field.

Data of disease incidence (%) were recorded at 20 DAS, 28 DAS and 35 DAS. The weight of each plant (g), no. of nodules per plant and weight of nodule per plant (g) were recorded at 60 DAS and 85 DAS. The incidence of foot and root rot disease in the experimental plots was recorded by the following formula:

% plant infection = (Number of infected plants/Total number of plants) × 100

The data were analyzed following a statistical package (MSTATC) program. Treatment means were compared with Duncan's Multiple Range Test (DMRT) [12].

3. Results

Effect of Biofertilizer and Biopesticide on Disease Incidence of Foot and Root Rot of Binamasur-1

Disease incidence was reduced in plots treated with IPM Biopesticide and Bari-Biofertilizer. The lowest disease incidence was recorded by treating soil at sowing time with Bari-Biofertilizer, while the highest disease incidence was recorded in control plots (**Table 1**). The lowest disease incidence (%) was 4.7%,

Table 1. Effect of IPM Biopesticide and Bari-Biofertilizer on disease incidence (%) of foot and root rot of lentil cv. Binamasur-1 and Binamasur-2.

Treatments	Disease incidence (%)					
	Binamasur-1			Binamasur-2		
	20 DAS	**28 DAS**	**35 DAS**	**20 DAS**	**28 DAS**	**35 DAS**
Control	15.1[a]	21.5[a]	20.3[a]	18.4[a]	22.7[a]	21.7[a]
IPM Biopesticide as soil treatment (7 days before sowing)	5.2[b] (65.5%)	3.1[b] (85.5%)	6.6[b] (67.4%)	5.6[b] (69.3%)	3.0[b] (86.7%)	6.9[b] (68.2%)
IPM Biopesticide in lines at sowing time	6.8[b]	6.3[b]	7.3[b]	6.8[b]	3.3[bc]	8.6[b]
IPM Biopesticide as seed treatment	6.8[b]	4.6[bc]	8.4[b]	8.0[b]	6.8[b]	8.3[b]
Bari-Biofertilizer as seed treatment	5.5[b]	3.9[bc]	7.5[b]	6.3[b]	3.5[bc]	6.6[b]
Bari-Biofertilizer in lines at sowing time	4.7[b] (68.8%)	1.8[c] (91.6%)	6.2[b] (69.4%)	5.2[b] (71.7%)	1.9[c] (91.6%)	6.6[b] (69.5%)
LSD ($p \geq 0.05$)	3.17	2.58	3.4	3.07	4.39	3.80

Data presents the mean of four replications. In a column, data with similar letter do not differ.

1.8% and 6.2% at 20 DAS, 28 DAS and 35 DAS, respectively while the highest disease incidence was recorded in control which was 15.1%, 21.5% and 20.3% at 20 DAS, 28 DAS and 35 DAS, respectively. The second-lowest disease incidence was recorded by treating with IPM Biopesticide as soil treatment (7 days before sowing) which were 5.2%, 3.1% and 6.6% at 20 DAS, 28 DAS and 35 DAS, respectively. Treating of soil with Bari-Biofertilizer resulted in up to 68.8%, 91.6% and 69.4% reduction of foot and root rot at 20 DAS, 28 DAS and 35 DAS, respectively over the control and also treating with IPM Biopesticides as soil treatment (7 days before sowing) resulted up to 65.5%, 85.5% and 67.4% reduction of foot and root rot at 20 DAS, 28 DAS and 35 DAS, respectively over the control. Seed treatments with Bari-Biofertilizer and IPM Biopesticides also resulted in significant effect on disease incidence of lentil.

Effect of Biofertilizer and Biopesticide on Disease Incidence of Foot and Root Rot of Binamasur-2

Disease incidence (%) was significantly reduced in treated plots of lentil compared to control (Table 1). The lowest disease incidence (%) was recorded by treating soil at sowing time with Bari-Biofertilizer which were 5.2%, 1.9% and 6.6% at 20 DAS, 28 DAS and 35 DAS, respectively while the highest disease incidence was recorded in control which was 18.4%, 22.7% and 21.7% at 20 DAS, 28 DAS and 35 DAS, respectively. Treating of soil with Bari-Biofertilizer resulted up to 71.7%, 91.6% and 69.5% reduction of foot and root rot at 20 DAS, 28 DAS and 35 DAS, respectively over the control. IPM Biopesticide reduced almost 69.3%, 86.7% and 68.2% foot and root rot at 20 DAS, 28 DAS and 35 DAS, respectively over the control. Seed treatments with IPM Biopesticide also resulted in significant effect on disease incidence of lentil.

Effect of Biofertilizer and Biopesticide on fresh weight of plant

Binamasur-1 found the maximum plant fresh weight (4.9 g) at 60 DAS was recorded in case of treating the soil with Bari-Biofertilizer while the minimum fresh weight (2.8 g) was recorded in control (Table 2). The fresh weight of lentil seedlings was found to be increased up to 75.0% and 35.7% by treating the soil with Bari-Biofertilizer and IPM Biopesticide, respectively. In Binamasur-2 the maximum fresh weight of plant (5.3 g) at 60 DAS was recorded in case of treating seeds with Bari-Biofertilizer, while the minimum fresh weight of plant (1.6 g) was recorded in control (Table 2). The fresh weight of lentil seedlings was found to be increased by treating the soil with Bari-Biofertilizer up to 187.5%.

Effect of Biofertilizer and Biopesticide on Number of Nodules

IPM Biopesticide and Bari-Biofertilizer applied in different methods showed an effect on the number of nodules/plant at 60 DAS. In Binamasur-1 maximum number of nodule/plant at 60 DAS (8.4) was found in case of treating soil with Bari-Biofertilizer in lines at sowing time. The second highest nodules/plant (7.5) was recorded in case of treating seeds with Bari-Biofertilizer. On the other hand, the lowest number of nodules (3.7) has been observed under control (Table 2). The number of nodules/plant of lentil seedlings was found to be increased by treating the soil with Bari-Biofertilizer and IPM Biopesticide up to 127.0% and 75.6% respectively, over the control. In Binamasur-2 the maximum number of nodule/plant at 60 DAS was found in case of treating soil with Bari-Biofertilizer (Table 2). The second highest nodules/plant was recorded in case of treating seeds with Bari-Biofertilizer. Furthermore, the lowest number of nodules/plant was observed under control condition. The number of nodules/plant of lentil seedlings was found to be increased by treating the soil with Bari-Biofertilizer up to 121.2% over the control.

Table 2. Effect of IPM Biopesticide and Bari-Biofertilizer on fresh weight, number of nodules and weight of nodules of Binamasur-1 and Binamasur-2.

Treatments	Binamasur-1			Binamasur-2		
	Fresh weight/ plant (g)	Number of nodules/plant	Fresh weight of nodules/ plant (g)	Fresh weight/ plant (g)	Number of nodules/plant	Fresh weight of nodules/ plant (g)
Control	2.8^{c}	3.7^{d}	0.01^{b}	1.6^{c}	4.7^{d}	0.01^{b}
IPM Biopesticide as soil treatment (7 days before sowing)	3.8^{c} (35.7%)	6.5^{abc} (75.6%)	0.01^{b}	3.1^{b} (93.7%)	7.4^{bc} (57.4%)	0.01^{b}
IPM Biopesticide in lines at sowing time	3.5^{bc}	5.1^{cd}	0.01^{b}	2.9^{b}	5.8^{cd}	0.01^{b}
IPM Biopesticide as seed treatment	3.9^{abc}	5.4^{bcd}	0.01^{b}	2.5^{b}	5.0^{d}	0.01^{b}
Bari-Biofertilizer as seed treatment	4.4^{ab}	7.5^{ab}	0.02^{a} (100.0%)	5.3^{a}	8.1^{b}	0.02^{a}
Bari-Biofertilizer in lines at sowing time	4.9^{a} (75.0%)	8.4^{a} (127.0%)	0.02^{a} (100.0%)	4.6^{a} (187.5%)	10.4^{a} (121.2%)	0.02^{a} (100.0%)
LSD ($p \geq 0.05$)	1.09	2.04	0.006	0.76	1.78	0.006

Data presents the mean of four replications. In a column, data with similar letter do not differ.

Effect of Biofertilizer and Biopesticide on Fresh Weight of Nodules

In Binamasur-1 the maximum fresh weight of nodules/plant was observed in case of treating the soil with Bari-Biofertilizer (Table 2). Soil and seed treatment with Bari-Biofertilizer also showed higher fresh weight of nodules/plant (0.02 g). The fresh weight of nodules of lentil seedlings was found to be increased by treating soil and seed with Bari-Biofertilizer up to 100.0% over the control. In Binamasur-2 the maximum fresh weight of nodules/plant (0.02 g) was observed in case of treating the soil with Bari-Biofertilizer. Seed treatment with Bari-Biofertilizer also showed higher fresh weight of nodules/plant (0.02 g) (Table 2). The fresh weight of nodules of lentil seedlings was found to be increased by treating the soil with Bari-Biofertilizer up to 100.0% over the control.

In Figure 1, field view of the experimental field (healthy and diseased) has been shown. From the two photographs, it was found that Bari-Biofertilizer treated plot showed lower diseases incidence and the control plot showed higher diseases incidence. In Figure 2, one photograph showed the foot and root rot infected lentil seedlings in the laboratory and another photograph showed healthy seedlings in the field. In Figure 3, isolated nodules from lentil seedlings have been shown.

(A) (B)

Figure 1. Field view: (A) Bari-Biofertilizer treated plot showing lower diseases incidence, (B) Bari-Biofertilizer untreated plot (control) showing higher diseases incidence.

(A) (B)

Figure 2. The lentil seedling: (A) Foot and root rot infected, (B) Healthy.

Figure 3. Isolated nodules from lentil seedling.

4. Discussion

In case of both Binamasur-1 and Binamasur-2, disease incidence was significantly reduced in plots treated with Biofertilizer and biopesticide which was supported by the mentioned findings. *Rhizobial* inoculations showed antagonistic effect against *Fusarium oxysporium* and *Sclerotium rolfsii in vitro* and showed up to 62.5% and 73.3% reduction of *Fusarium* and *Sclerotium* foot and root rot, respectively in pot experiment and obtained up to 87.43% reduction of foot and rot disease of lentil by using *Rhizobial* inoculants treated seeds [13]. Biofertilizer treated seed resulted in up to 84.03% reduction of foot and root rot [14]. Seed treatment with Bari-Biofertilizer (*Rhizobial* inoculants) and BAU-Biofungicide (*Trichoderma harzianum*) resulted in up to 26.47% higher field emergence, post-emergence deaths of plants due to foot rot disease were also successfully reduced [15]. Soil treatment with Bari-Biofertilizer resulted in the lowest disease incidence of a pulse crop chickpea var. Hyprosola, Binasola-2, Binasola-3 and Binasola-4 at 20 DAS (Days after sowing) that displayed reduction of disease incidence up to 83.77%, 54.48%, 70.76% and 71.45% respectively over control which was also supported the result [16]. *T. harzianum* and *T. viride* as the most efficient in inhibiting the growth of *Fusarium oxysporum* f. sp. *ciceri* by 60% and 58%, respectively [10]. *Trichoderma lignorum*, *T. viride* and *T. virens* inhibited the growth of *S. rolfsii* by 68% and 67%, while the growth of *R. bataticola* was inhibited to the extent of 76% by *T. viride* and *T. harzianum* [10]. *Trichoderma harzianum* resulted in foot and root rot disease in legumes control ranging from 66.67% to 100% [17]. The lowest foot and root rot incidence (6.9%), highest seed germination (82.08%), maximum plant stand (93.12%) and the highest seed yield (3726.67 kg·ha^{-1}) were recorded in plots where *T. harzianum* (TG-2 isolate) was applied [18]. *Trichoderma viride* was found most effective in suppressing the mycelial growth (51.11%) and sclerotia production (95.90%) of the target pathogen [19].

Fresh weight of plant was significantly increased in both Binamasur-1 and Binamasur-2 in plots treated with Biofertilizer and biopesticide which was sup-

ported by the following findings. Seed treatment with *T. harzianum* resulted in increasing the fresh shoot weight up to 263.33% and fresh root weight 157.14% over control [20]. Lentil cv. *utfala* seed was treated with *Rhizobium* inoculants showed 11.6% increased dry weight of plants over control [21]. Soil treatment with Bari-Biofertilizer resulted in the maximum fresh weight of chickpea var. Hyprosola, Binasola-2, Binasola-3 and Binasola-4 at 60 DAS (Days after sowing) and the fresh weight of seedlings was found to be increased up to 70.39%, 142.14%, 167.19% and 98.75% respectively over control which was also supported the result [16].

Bari-Biofertilizer and IPM Biopesticide applied in different methods showed significant increase on the number of nodules/plant at 60 DAS in both Binamasur-1 and Binamasur-2 which was supported by the following findings. The use of Bari-Biofertilizer in different methods resulted in higher number of nodules per plant in chickpea. Mungbean (*Vigna radiata* L.) seed treated with *Rhizobium* had the number of nodulation, biomass and grain yield increased [22]. Biofertilizer as seed treatment resulted in 53.9% higher nodules/plant over control [23]. Biofertilizer and *Rhizobium* strains, viz. BINA-LTL4, BINA-LT-634 and BINA-LT-640, increased number of nodules/plant in 58.4% in another study [14]. Soil treatment with Bari-Biofertilizer resulted in the maximum number of nodules/plant of a pulse crop chickpea var. Hyprosola, Binasola-2, Binasola-3 and Binasola-4 at 60 DAS (Days after sowing) and the number of nodule/plant was found to be increased up to 293.48%, 109.03%, 124.68% and 183.33% respectively over control which also supported the result [16].

The fresh weight of nodules increased was significantly increased in both Binamasur-1 and Binamasur-2 in plots treated with Biofertilizer and biopesticide which was supported by the mentioned findings. The fresh weight of nodules increased when the soil was treated with Bari-Biofertilizer in case of chickpea variety. In chickpea, they found 35% increase of weight of nodules [24]. Also found 61.39% increase of weight of nodules in chickpea [25]. Soil treatment with Bari-Biofertilizer resulted in the maximum fresh weight of nodules of seedlings of a pulse crop chickpea var. Hyprosola, Binasola-2, Binasola-3 and Binasola-4 at 60 DAS (Days after sowing) and the fresh weight of nodules of seedlings was found to be increased up to 172.58%, 164.15%, 226.83% and 98.75% respectively over control [16].

5. Conclusion

In this study, it was observed that both Bari-Biofertilizer and IPM Biopesticide resulted in lower disease incidence of Binamasur-1 and Binamasur-2 over the control. Therefore, control of pulse diseases by biocontrol means could be an important point of consideration. Bari-Biofertilizer and IPM Biopesticide also increased the fresh weight of the plant, the number of nodules per plant and fresh weight of nodules per plant. The findings of the present study have pointed out that Bari-Biofertilizer and IPM Biopesticide in soil or seed treatment may

use for the overcome the diseases of lentil in the farmer's field.

Conflicts of Interest

Authors have declared that no competing interests exist.

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