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EVALUATION OF DIFFERENT CHEMICALS AND BOTANICAL EXTRACTS ON GERMINATION AND SEED-BORNE FUNGI OF COUNTRY BEAN

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Abstract

The experiment was carried out to estimate the prevalence of seed-borne fungi of country bean at laboratory condition. Before placed, the seeds were treated with six different treatments namely Autostin 50WP, Aimcozim 50WP, Ata leaf extract (1:2 w/v), Neem leaf extract (1:2 w/v), Biskatali leaf extract (1:2 w/v) and Mahogani leaf extract (1:2 w/v). In laboratory, different seed-borne fungi like *Aspergillus niger*, *Cercospora sesami*, *Rhizopus stolonifer*, *Alternaria* sp. and *Fusarium oxysporum* were detected from the collected seed sample by blotter method. After 7 days of incubation, germination percentage varied significantly and ranged from 33.3% to 83.3%. The highest germination was found in Autostin 50 WP (83.3%) which was statistically similar with Aimcozim 50 WP (77.8%) and among botanicals the highest germination was recorded from the treatment Biskatali leaf extracts. Autostin 50 WP was found as the best seed treating chemicals in controlling seed borne fungi *Aspergillus niger*, *Alternaria* sp., *Fusarium oxysporum, Rhizopus stolonifer* and *Cercospora sesame* compared to all other treatments.

***Keywords*:** Country bean, botanical, chemical, seed borne fungi.

Introduction

Country bean (*Lablab purpureus* L.) is one of the most important leguminous vegetables in Bangladesh. It is short lived creeping perennial but used as an annual legume. It is a vigorously trailing, twining herbaceous plant. It is normally grown during the rabi or winter season. The green pods of legume and developed unripe seeds serve as delicious vegetables. Immature green seeds are eaten boiled or used curries, ripe seeds are also used as pulse, "dhal" (Sultana, 2001). It improves the fertility status of soil through nitrogen fixation. High acidic soil and adverse climatic conditions like heavy rainfall, high humidity, seasonal flooding affect the vegetable production in Sylhet region. Several diseases and insect also attack crops and causes yield loss. Summer country bean during summer season in Sylhet region might be an excellent endeavor to the farmers to earn a lot from their existing farming system. Number of diseases of country bean is increasing day by day (Ahmed, 1986). Various approaches such as plant extracts, fungicides and use of resistant variety are used to control different diseases. Chemicals are very effective for controlling the diseases of crop plants in Bangladesh (Hoque *et al*., 2014). Though injudicious application of these chemicals pollute the environment and cause health hazard but other alternate approaches like use of plants extracts is very effective against the *Cercospora* sp. Botanical fungicides are unique because they can be produced easily (Roy *et al*., 2005). The use of plant extracts is a recent approach to plant disease management and it has drawn the special attention of the plant pathologist all over the world (Asharfuzzaman and Hossain, 1992; Suratuzzaman *et al.*, 1994). In consideration of the situation stated above, the present study was undertaken to evaluate the efficacy of different botanicals and chemicals on seed-borne fungi of country bean.

Materials and Methods

Collection and preparation of working sample

One high yielding variety BARI Seem-7 was used in this experiment. The variety was collected from Bangladesh Agricultural Research Institute (BARI), Gazipur. Fresh leaves of Neem (*Azadirachta indica*), Biskatali

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(*Polygonum hydropiper*), Mahogoni (*Swietenia macrophylla*) and Ata (*Anonasqua mosa*) were collected from Eco park area near of Sylhet Agricultural University, Sylhet. The extracts were prepared by following the method of Ashrafuzzaman and Hossain (1992). For preparation of extracts, fresh leaves were collected, weighted in an electronic balance and then washed in the water. After washing the large leaves were cut into small pieces. For getting extract, weighted plant parts were blended and added with distilled water. The pulverized plant tissue was squeezed through 3 folds of fine cotton cloth. When extract were used for field treatment as spraying purpose then its ratio was 1:4 (100 g plant parts materials crushed in 400 ml of distilled water).

Seed treatment with plant extracts

For the purpose of seed treatment 1:2 (w/v) ratio was prepared by crushing 100 g plant parts material in 200 ml of distilled water. A total of 105 seed samples of country bean were treated for 21 plots in the experiment. Treated seeds were tested for fungal growth by the Standard Blotter Method (ISTA, 2006). In case of control, seeds were treated only with distilled water. After proper covering the seed coat with extracts, seeds were used for studying the efficacy of the applied plant extract.

Seed treatment with fungicides

Two fungicides namely Autostin 50 WP and Aimcozim 50 WP (Group of carbendazim) collected from local market in Sylhet were used as seed treatment. For treating seeds, 250 mg of each fungicide along with 100 g seeds was taken separately in 250 ml Erlenmeyer flasks. The flasks were then shaken manually for 10-15 minutes for proper coating of the fungicides. The treated seeds were kept overnight as it is in the flasks and then tested for the presence of fungal detection.

Blotter incubation test

The fungal pathogens associated with country bean seed samples were detected by blotter method (ISTA, 2006). In this method, three pieces of 9 cm filter papers (Whatman No.1) were soaked in distilled water and placed at the bottom of the Petridish (9 cm diameter). Forty two seeds were taken from sample and then placed on the moist filter paper in seven petridishes. The petridishes with seeds were then incubated at room temperature (25±3)°C in incubation chamber of the laboratory for seven days. After seven days of incubation, each seeds was observed under stereo-binocular microscope at 10X magnification in order to record the presence of seed borne fungal pathogens and identified by observing their growth characters on the incubated seeds. Number of germinated seed was recorded along with the seed borne infection of fungal pathogens incubation. The results were expressed in percentage. Incidence of fungi was calculated according to following formula (Ahmed, 1986).

$$\text{Incidence of seed yielding fungi (\%)} = \frac{Number\ of\ infected\ seed}{Total\ number\ of\ seed} \times 100$$

Experimental design and statistical analysis

The experiment was laid out in laboratory conditions using Completely Randomized Design (CRD) with three replications. The data obtained for different parameters were statistically analyzed to find out the significant difference among the treatment. The analysis of variance was performed by using R program. The difference among the treatment means was estimated by LSD (Least Significance Difference) at 5% level of probability.

Results and Discussion

Detection and identification of seed-borne fungi on blotter

A total of five seed-borne fungi like *Aspergillus niger, Alternaria* sp., *Fusarium oxysporum, Rhizopus stolonifer,* and *Cercospora sesame* were detected by standard blotter method (Plate 1). The incidence of *Aspergillus niger, Alternaria* sp., *Fusariumo xysporum, Rhizopus stolonifer* and *Cercospora sesame* were ranged between 33.3-88.9%, 22.2-88.9%, 38.9-88.9%, 27.8-83.3% and 27.8-88.9%, respectively.

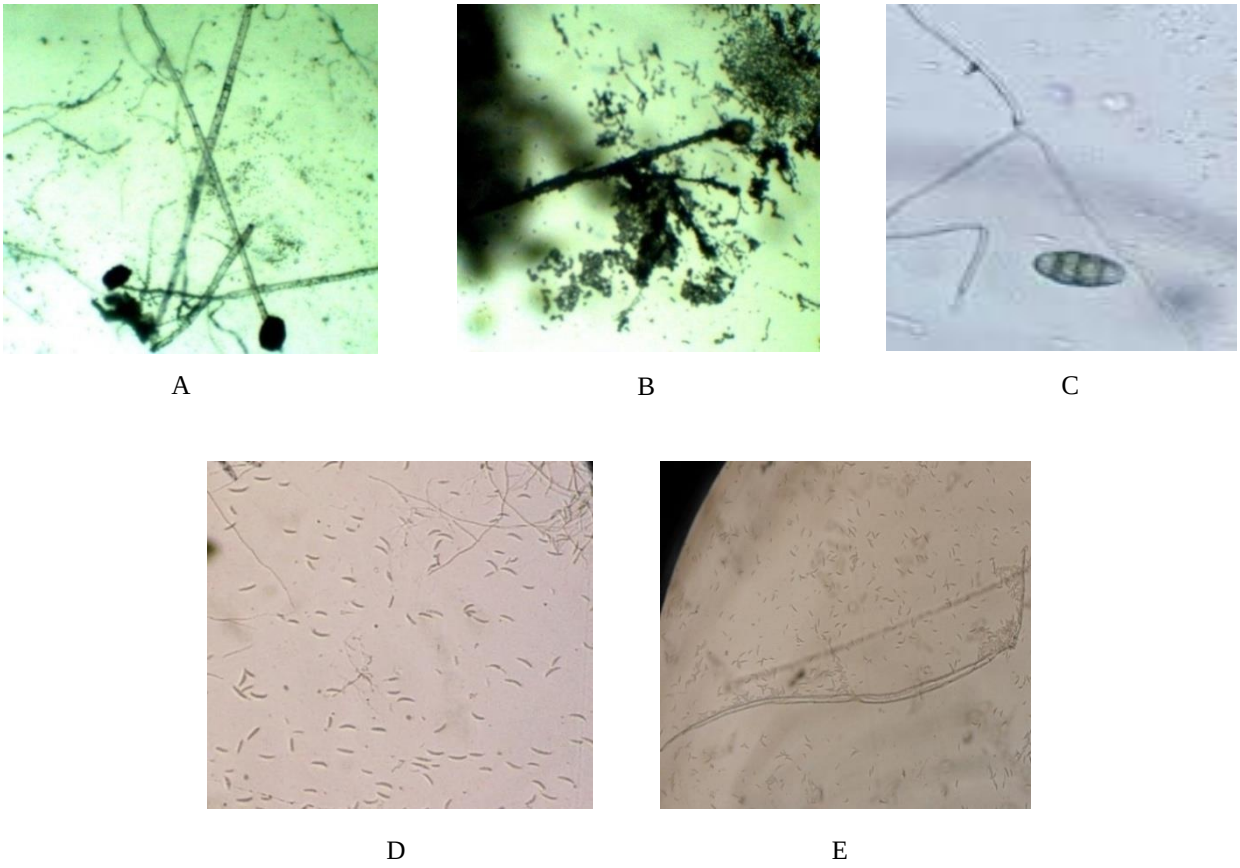


Plate 1. Conidia of seed borne fungi under compound microscope (10 X). (A) *Rhizopus stolonifer*, (B) *Aspergillus niger*, (C) *Alternaria* sp., (D) *Fusarium oxysporum*, (E) *Cercospora sesami*

Efficacy of different chemicals and botanicals on seed germination

Result of different treatments on germination percentage of country bean seeds are presented in Fig 1. After 7 days of incubation, percent seed germination varied significantly among different treatments. The germination percentage was ranged from 33.3 to 83.3%. The highest germination (83.3%) was found in seed treatment by Autostin 50 WP; which was statistically similar with Aimcozim 50 WP (77.8%). The lowest germination was recorded in control treatment (33.3%). Effect of different treatments on germination has been shown in Plate 2.

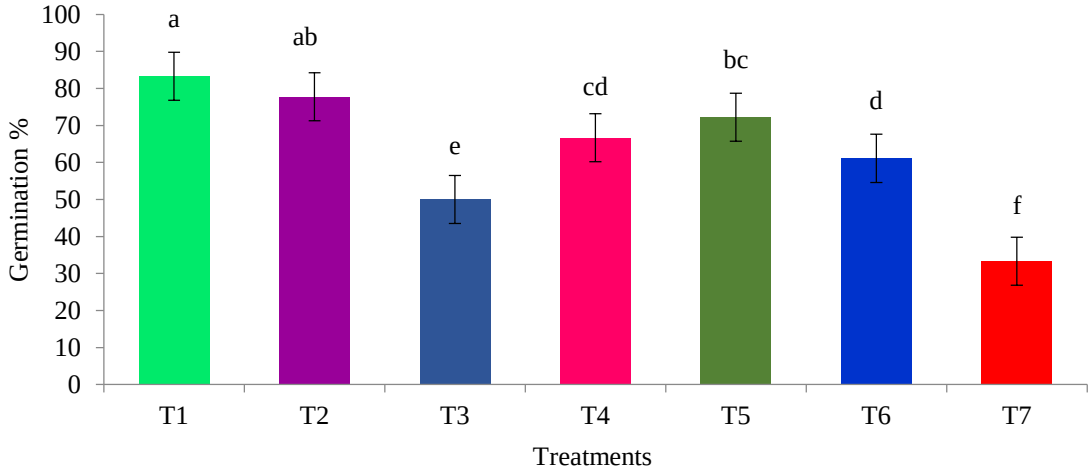
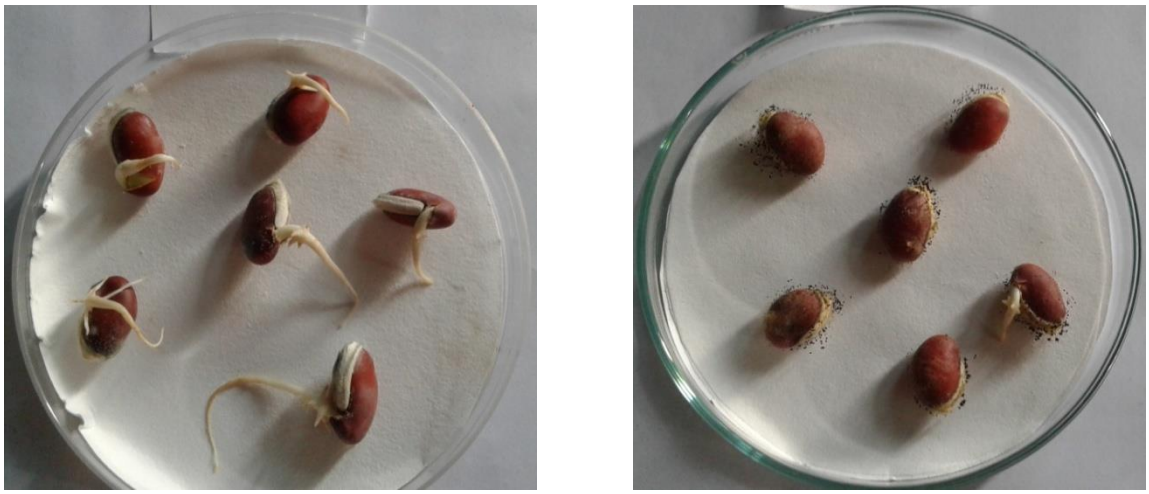


Fig. 1. Effect of treatments on percent seed germination of country bean

(Chemicals = Autostin 50 WP, Aimcozim 50 WP), (botanicals = Ata leaf extracts, Neem leafextracts, Biskatali leaf extracts, Mahogani leaf extracts, Control)



Autostin 50 WP treated seeds Untreated seeds (Control)

Plate 2. Effect of Autostin 50 WP treatment on seed germination of country bean

In the present study, Autostin 50 WP treated seeds performed best result. Hossain (2014) reported that seed treated with Bavistin at 0.2% increased germination of 39% over control. Biskatali leaf extracts and neem leaf extracts performed better than control. Similar results were also obtained by Haque *et al.* (2009) and Rahman *et al.* (1999) and they reported that plant extracts like Garlic, Neem, Biskatali also improved germination of various seeds such as jute, rice, chilli, cotton and soybean.

Performance of different treatments against *Rhizopus stolonifer*

In suppressing the growth of *Rhizopus stolonifer*, all the treating chemicals and plant extracts inhibited the incidence of *Rhizopus stolonifer* compared to untreated control (Table 1). The highest incidence was found in control treatment (83.3%) and the minimum incidence was found in Autostin 50 WP (27.8%) treated seeds which were statistically similar with Aimcozim 50 WP (33.3%) and Biskatali leaf extract (38.9%) treated seeds. It was revealed that Autostin 50 WP treated seeds came out as the best treatment in controlling seed yielding *R. stolonifer*. Salam (2004) reported that vitavax-200 treated seeds showed better performance to control seed borne micro flora than hot water treatment. All treated seeds showed better performance compared to untreated seeds.

Performance of different treatments against *Fusarium oxysporum*

All the treatments appeared to be effective to control *Fusarium oxysporum* in comparison with the untreated control (Table 1). The maximum incidence was found in control (88.9%) and the lowest was recorded in Autostin 50 WP (38.9%) which was statistically similar with Aimcozim 50 WP (44.4%). The present study showed that Autostin 50 WP performed the best compared to all other treatment sin controlling seed borne *F. oxysporum*. Similar results were also supported by Sinha *et al*. (2004) and they reported that Dithane M-45 was effective against *Alternaria alternata, Curvularia lunata, Rhizopus stolonifer, Aspergillus flavus, A. niger* and *Penicillium citrinum.*

Performance of different treatments against *Alternaria* sp

It was found that all treating chemicals and plant extracts inhibited *Alternaria* sp. compared to untreated control (Table 1). Maximum incidence was recorded in control (66.7%). The minimum was recorded in Autostin 50 WP (16.7%) which was statistically similar with Aimcozim 50 WP (22.2%) and Biskatali leaf extract (27.8%) treated seeds. The results showed that Autostin 50 WP was the best seed-treating chemicals. Neem leaf extracts and Biskatali leaf extracts was also effective against the pathogens. Similar results were also supported by Uddin *et al.* (2013) and Khan and Kumar (1992) and they reported that extracts of Garlic, Ginger, and Biskatali leaf extracts

were effective against seed borne infection of *Fusarium* sp., *Alternaria tenuis, A. alternata, Bipolaris sorokiniana* and *Curvularia lunata* isolated from wheat seeds.

Performance of different treatments against *Cercospora sesami*

The highest incidence by *Cercospora sesami* was recorded in control (61.1%) and minimum incidence in Autostin 50 WP (16.7 %) treated seeds which were statistically similar with Aimcozim 50 WP (22.2%) and Biskatali leaf extract (27.8%) treated seeds (Table 1). The results showed that Autostin 50 WP was the best seed-treating agent among the chemicals used to control *C. sesami*. Similar results were also supported by Rahman *et al*. (1999) and reported that Biskatali leaf extracts, extracts of Garlic, Ginger were effective against seed borne infection of *Fusarium* sp., *Alternaria tenuis, Bipolaris sorokiniana* and *Curvularia lunata* isolated from wheat seeds.

Performance of different treatment against *Aspergillus niger*

The performance of different treatments against *Aspergillus niger* is presented in Table1. Autostin 50 WP showed the highest effectiveness against *Aspergillus niger*. The highest percentage of incidence was recorded from control (88.9%) and the lowest in Autostin 50 WP (33.3%) treated seeds which were statistically similar with Aimcozim 50 WP (38.8%) and Biskatali leaf extract (44.4%). It was observed that Autostin 50 WP was highly effective against *A. niger*. Similar results were supported by Haque *et al.* (2009) and they reported that Vitavax 200 was found most effective against seed borne pathogens of chilli.

Table 1. Efficacy of chemicals and botanical extracts on seed yielding fungi of country bean

Treatments	Incidence of seed borne fungi (%)				
	Aspergillus niger	*Rhizopus stolonifer*	*Fusarium oxysoporum*	*Alternaria* sp.	*Cercospora sesami*
Autostin 50 WP	33.33 c	27.77 c	38.90 d	16.7 e	16.7 e
Aimcozim 50 WP	38.8 c	33.30 c	44.43 d	22.2 e	22.2 e
Ata leaf extract	77.7 ab	72.23 ab	77.77 ab	44.4 bc	44.4 bc
Neem leaf extract	66.70 b	61.13 b	66.70 bc	38.9 cd	38.9 cd
Biskatali leaf extract	44.43 c	38.87 c	50.00 cd	27.8 de	27.8 de
Mahagoni leaf extract	72.23 ab	66.70 b	77.77 ab	55.6 ab	55.6 ab
Control	88.87 a	83.30 a	88.87 a	66.7 a	61.1 a
LSD	18.00	12.72	17.99	14.22	12.72
CV	17.04	13.27	16.19	15.27	14.53

Conclusion

From the above study, it may be concluded that among all treatments, application of Autostin 50 WP provided as the best seed treating chemical in controlling seed borne fungi of country bean. In depth study could be also conducted to evaluate the efficacy of plant extracts as these are very available and environmentally sound.

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