

**EVALUATION OF HEALTH STATUS AND QUALITY OF
VEGETABLES SEEDS COLLECTED FROM RANGPUR
DISTRICT OF BANGLADESH**

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**EVALUATION OF HEALTH STATUS AND QUALITY OF VEGETABLES
SEEDS COLLECTED FROM RANGPUR DISTRICT OF BANGLADESH**

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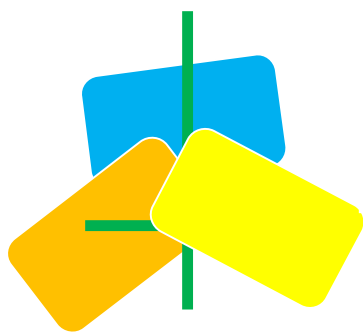
CERTIFICATE

*This is to certify that, the thesis entitled, "**EVALUATION OF HEALTH STATUS AND QUALITY OF VEGETABLES SEEDS COLLECTED FROM RANGPUR DISTRICT OF BANGLADESH**" submitted to the Department of Plant Pathology, Sher-e-Bangla Agricultural University, Dhaka, in partial fulfillment of the requirements for the degree of **MASTER OF SCIENCE (M.S.) IN PLANT PATHOLOGY** embodies the result of a piece of bona fide research work carried out by **RATAN CHANDRO MOHANTO**, Registration No.: **14-06310** under my supervision and guidance. No part of the thesis has been submitted for any other degree or diploma.*

I further certify that such help or source of information, as has been availed of during the course of this investigation has been duly been acknowledged by him.

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TO MY PARENTS

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EVALUATION OF HEALTH STATUS AND QUALITY OF VEGETABLES SEEDS COLLECTED FROM RANGPUR DISTRICT OF BANGLADES

ABSTRACT

A series set of test of this experiment was carried out to evaluate the seed health and quality status of ten selected vegetables viz. amaranth, Indian spinach, bottle gourd, sweet gourd, snake gourd, okra, bitter gourd, cucumber, brinjal and country bean during November 2015 to April 2016 at Seed Health Laboratory of Department of Plant Pathology, Sher-e-Bangla Agricultural University, Dhaka, Bangladesh. Untreated and unknown grower's bulk vegetable's loose seeds were collected from three different seed sources of Rangpur sadar viz. New Bangla Seeds; Islam Seeds and Rafiq Traders. Seed health study was conducted by following inspection of dry seed, blotter method, rolled paper towel method and growing on test. In inspection of dry seed method, pathogenic structures, mycelium (white and cottony) and acervuli were recorded. The observed physical abnormalities viz. deformed, shrinkage, swelling, spotted and undersized seeds were recorded. In most cases, the highest percentage of pure seed was recorded in seeds collected from Rafiq Traders followed by New Bangla Seeds. It was exposed that seeds of selected vegetables yielded six fungal species in blotter method. The identified fungi were *Aspergillus flavus*, *Aspergillus niger*, *Fusarium* spp., *Alternaria* spp., *Chaetomium* spp. and *Rhizopus* sp. Seed infection of an unidentified bacterium was also recorded. The highest total seed borne fungal infection was found in the seeds of New Bangla Seeds. In rolled paper towel methods, seed germination varied from 70-94%, normal seedlings were 56-84%, abnormal seedlings were 5-18%, rotten seeds were 4-15%, hard or ungerminated seeds were 2-18% and vigor index was 677.3 to 2747. In growing on test method, seed germination varied from 65-99%, normal seedlings were 51-90%, abnormal seedlings were 8-19%, rotten seed were 1-15%, hard seeds were 1-24% and vigor index was 666.6 to 2747. In both methods, the highest seed germination and seedling vigor was recorded in amaranth and country bean of New Bangla Seeds, respectively. The overall health status and quality of bulk vegetable's loose seeds were not up to the mark. Most of the seed-borne pathogens reduced the germination and produced diseased seedlings. Thus, seed treatment is suggested for the farmers/growers before sowing vegetable's loose seeds.

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LIST OF ABBREVIATIONS

Full word	Abbreviations
Bangladesh Agricultural Research Council	BARC
Bangladesh Agricultural University	BAU
Bangladesh Bureau of Statistics	BBS
Centimeter	cm
Coefficient of Variance	CV
Degree Celsius	°C
Etcetera	etc.
Food and Agricultural Organization	FAO
And others	<i>et al.</i>
Least Significant Difference	LSD
Percentage	%
Ton per hectare	t/ha
Sher-e-Bangla Agricultural University	SAU
Videlicet (namely)	viz.
World Health Organization	WHO

CHAPTER I

INTRODUCTION

Seed is the biological objected input in agriculture. Quality of seed has the pre-requisite of seed as a planting material for sowing in land in order to get disease free seedling and plant and finally to achieve satisfactory yield. Health of seeds can be affected by direct infection of pathogens or through contamination of seeds by pathogenic propagules as contamination in, on or with the seeds or as concomitant contamination (Rashid *et al.*, 2000). In Bangladesh, around 2.5 million tons of rice worth more than Tk. 12000 million was lost annually due to diseases caused by seed borne pathogens (Fakir *et al.*, 2003). In Bangladesh, agriculture accounts for 532,032 million taka of its gross domestic product (BBS, 2013). Vegetables constitute a potential and important group of crops in Bangladesh. They are important for their low production cost, short production time and high nutritive value. In 2010-2011 about 1115966 acres of land were under vegetable cultivation in Bangladesh and production was 3061840 metric tons (BBS, 2010). The contaminated seeds may fail to germinate, spread disease from seed to seedling and from seedling to growing plants. Seed borne pathogens affect seed quality. Islam *et al.* (2000) found that most lethal seed infection caused by *Fusarium moniliforme*, *Trichoconis padwickii* and *Curvularia* sp. The infected seeds may fail to germinate, transmit disease from seed to seedling and from seedling to growing plants (Farid *et al.*, 2002)

The major constraints to production in Bangladesh are; insects, weeds and diseases, and also seeds, environment, marketing, postharvest losses etc. Amongst the production constraints, pest and diseases are the major ones. Among them, infestation by fungi, bacteria, nematodes or viruses are major. Therefore, diseases of tomato act as the main limiting factors to its economic production. Over 200 diseases have been reported to affect the tomato plants in

the world (Watterson, 1986). Among them the seed borne pathogens mostly fungi plays a vital role in disease development. World-wide, there are numerous reports on seed borne fungi of tomato (Neegaard, 1979). Seed borne fungi are of considerable importance due to their influence on the over-all health, germination and final crop stand in the field. Farmers have to deal with significant losses due to infections by serious seed borne pathogens on their plants, which may start from germinating seed, seedling in the nursery, matured plants in the field and proceed till the products are harvested and fruits and seeds stored. Significant crop losses due to seed borne pathogens have been recorded by Pimentel and Perkins (1980).

Seeds are very important in crop production and high quality seed is an important pre-requisite for sustainable and profitable vegetable production. Seed health is an important factor in the control of diseases, since an infected seed is less viable, has low germination, reduced vigor and reduced yield. The control of seed borne pathogens is the first step in any agricultural crop production and protection programmed. Attempts have been made to reduce seed borne infection by chemical treatment of the seeds and some successes have been reported. Though, chemical controls of seed borne pathogens have been very successful. However, chemical pesticides have the additional potential disadvantages of accumulation in the ecosystem and induction of pesticide resistance in pathogens.

In Bangladesh, important vegetable are brinjal, Indian spinach, okra bottle gourd, sweet gourd, snake gourd, bitter gourd, country bean, yard long bean, cucumber and radish etc. Indian spinach under Amaranthaceae family. Leafy vegetables, flowering annual plant, height of up to 30 cm, flowers yellow-green, 3-4 mm diameter, small, hard, dry, lumpy fruit cluster (5-10mm), containing several seeds and source of vitamin A, vitamin-C, vitamin E and vitamin K etc. Spinach leafy vegetables supply nutrition all over Bangladesh in Rabi season.

Similarly Indian spinach is an important vegetable in Bangladesh. It grows all over Bangladesh in Kharif and Rabi season. Indian spinach (*Spinacia oleracea*) an edible flowering plant *Amaranthaceous*, an annual plant (rarely biennial) of central and south western Asia. Bottle gourd is important vegetable Plant of Cucurbitaceae family, edible vegetable, typically grows as an annual, Flower White or Yellow, fruit a berry, variable in size and shape. Similarly Sweet gourd is a plant of the family *Cucurbitaceae*, very popular, nutritious, fruit vegetables, seed is the main propagative material of this gourd. So the quality of the seed should be known to us for its proper use. Snake gourd and country bean is a common and important vegetable, which found in every season in Bangladesh Long yard bean is a vegetable of family *Fabeaceae*. It contain large amount of protein, carbohydrate and iron. It is a common vegetable in our country, to know the seed health status of loose vegetable seed. Seed- borne pathogens can affect the seed quality by damaging external or internal seed tissues and cause the important seed diseases like seed rot, seed necrosis, and seedling damage through the local or systemic infection (Bateman and Kwasna, 1999; Khanzada *et al.*, 2002)

There are different methods for determination of seed health test such as Inspection of dry seed, Incubation test viz. Blotter method, Agar plate method, Rolled paper towel method, Water agar plate method, Deep freezing blotter method and Seedling symptom test. Microscopic observation of suspension viz. washing test, Whole embryo count method, Indicator plant test, growing on test and Serological test etc. Here taking into consideration easier method such as inspection of dry seed and Incubation test like blotter method, rolled paper towel method and Seedling symptom test.

From the conclusion of this research work, we can understand about the health status of loose vegetable seeds and give suggestion to the farmers regarding seed treatment. The expected output may ensure the quality production of

vegetable in Bangladesh. So, to achieve these troubles which are created by using unknown quality loose seed, evaluation of seed health status is very effective.

Considering the above facts, the present research is undertaken -

- To assess the health status of untreated, bulk vegetable loose seeds collected from Rangpur district of Bangladesh; and
- To evaluated the seed and seedling quality of untreated, bulk vegetable loose seed collected from Rangpur district of Bangladesh.

CHAPTER II

REVIEW OF LITERATURE

Considerable numbers of research work have been conducted on the association of the seed borne fungal pathogens of vegetable crops both at home and abroad. Attempt has been made to review the relevant literature available in the subject.

Seed borne disease is a serious problem for successful crop production in Bangladesh. Among all the groups of seed borne pathogens, Fungi play significant role in causing seed borne diseases. Pathogenic fungi, bacteria and viruses are the most responsible as well as limiting factors in cultivation of the crop in many part of the world. Many researchers have been carried out research in relation to seed quality and health status of different vegetables. However, some of the recent and available literatures, relevant information on seed health and quality have been cited in this chapter

2.1 History of fungi in seeds

Knowledge concerning seed-borne fungi, which cause diseases in plants, is almost two and half centuries old. It was in France when showed that he could increase the number of smut-infected wheat plants by adding smut dust to wheat seeds before planting and reduce them if the seeds were treated with Copper Sulphate before planting (Agrios, 1997). However, it was only during the first half of the last century when seed borne diseases received greater attention, especially in Europe and North America.

2.2 Fungi associated with fruit vegetables seeds and their pathogenic effect

a) Cucurbitaceous vegetables

Aktar (2009) studied the health status of sweet gourd seeds collected from different villages of Madhupur Upazilla of Tangail District. After incubation of

sweet gourd seeds on blotter four different fungi such as *Aspergillus flavus*, *Aspergillus niger*, *Penicillium* sp., and *Botrytis cineria* were recorded.

Sultana (2009) conducted an experiment to study the germination and health status of Truthfully Labeled Seed (TLS) of bottle gourd, sweet gourd, snake gourd, Ridge gourd, cucumber, wax gourd and sponge gourd collected from BADC and other seed company. She observed eight fungi namely *Aspergillus* sp., *Botrytis* sp., *Curvularia* sp., *Colletotrichum* sp., *Fusarium* sp., *Penicillium* sp., *Phomopsis* sp. and *Rhizopus* sp., she found only *Aspergillus* sp. was highly prevalent in all the crop seeds ranging from 1.6-14%.

Sultana and Ghaffar (2009) studied that seed borne fungi of bottle gourd (*Lagenari siceraria*), a total of 22 genera and 45 species of fungi were isolated of which 35 have hitherto been recorded from seeds of bottle gourd in Pakistan. Both blotter and deep-freezing methods yielded quantitatively as well as qualitatively more fungi than agar plate method *Lasiodiplodia theobromae*, *Fusarium semitectum*, *Macrophomina phaseolina* and *Fusarium oxysporum* were most frequently isolated from 33, 91, 50 and 66% seed samples of bottle gourd, respectively.

Anonymous (2003) reported that *Fusarium* sp. fungus can be seed borne in case of squash and pumpkin. They suggested paying special attention in using disease free seed.

Alimova *et al.* (2002) obtained different seed borne fungi and bacteria in cucumber and tomato seeds. They conclude that *Aspergillus* sp, and *Penicillium* sp. were predominant in tomato and cucumber seeds.

Begum and Momin (2000) reported that the suitable detection techniques of associated seed borne fungi in the laboratory conditions. Fifty-four seed samples of three cucurbits, namely sweet gourd (*Cucurbita moschata*), white gourd (*Benincasa hispida*) and bitter gourd (*Momordica charantia*), were

collected from six different districts of Bangladesh, to find out a suitable dejection technique of associated seed borne fungi in laboratory conditions. Three different detection methods, namely dry inspection, blotter test seedling symptom test, were compared. Among the tested detection techniques, more infection rate was observed for *Aspergillus flavus* and *Penicillium* in all cucurbit seeds in blotter test method, whereas high infection caused by *Fusarium* sp. and *Rhizopus* sp. were recorded in test tube seedling symptom test. The higher germination percentage of cucurbit seed was observed in blotter test and it is less expensive, quick and useful for the detection of most infectious fungi

Kamble *et al.* (1999) found eight different fungi (*Aspergillus* sp., *Colletotrichum* spp., *Fusarium* sp., *Penicillium* sp., *Rhizopus* sp., *Rhizoctonia* sp., *Macrophomina phaseolina* and *Alternaria* sp.) from pumpkin (cv. Local), cucumber (cv. Pnekhire), water melon (cv. Sugar baby) and muskmelon (cv. Punjab hybrid).

Peregrine *et al.* (1984) and Peregrine and Ahmed (1983) reported that the fungi *Aspergillus*, *Rhizopus*, *Cladosporium*, *Helminthosporium*, *Corynespora* and *Penicillium* were associated as seed borne fungi with cucurbit crops.

b) Solanaceous and other vegetables

Hossain *et al.* (2015) reported that 6 company's okra seeds collected from in Mymensingh district and dominance of seed borne fungi was studied by blotter method. Six predominant fungal genera were identified namely *Fusarium oxysporum* (5.08%), *Aspergillus flavus* (4.50%), *Aspergillus niger* (6.50%), *Colletotrichum dematium* (4.67%), *Rhizopus stolonifer* (3.33%) and *Penicillium* sp. (3.00%). They also conclude germination percentage and fungal association varied from company to company.

Akter (2008) found *Colletotrichum dematium*, *Macrophomina phaseolina*, *Fusarium oxysporum*, *Fusarium moniliforme*, *Cercospora* sp., *Aspergillus niger*, *Aspergillus flavus* and *Penicillium* sp. as seed borne fungi on okra seeds.

Sarker *et al.* (2006) studied the effect of seed-borne fungal pathogen on the planting value of brinjal (*Solanum melongena*) seeds collected from different sources of Bangladesh during March 2002-March 2003. *Aspergillus flavus*, *Alternaria alternata*, *Colletotrichum dematium*, *Phomopsis* spp. *Curvularia lunata* (*Cochliobolus lunatas*), *Fusarium moniliforme* (*Gibberella moniliformis*), *F. oxysporum*, *Penicillium* sp. and *Rhizopus* sp. were commonly found prevalent in all the seeds.

Shome (2002) studied quality of brinjal, tomato and onion seeds. He recorded *Aspergillus* sp., *Curvularia* sp., *Fusarium* sp., *Penicillium* sp., and *Rhizopus* sp. where maximum prevalence of *Aspergillus* sp. was (12%) in farmer's seeds.

Thippeswamy *et al.* (2006) collected 145 brinjal seed samples were collected from different agro-climatic regions of Karnataka, India during 2001-03 and analysed for microflora. This crop is susceptible to phomopsis blight (*Phomopsis vexans*) and leaf spot (*Alternaria solani*) diseases. These diseases are seed borne fungal disease and reduce the yield up to 30-50 percent.

Alam (2004) studied the qualitative attributes of Brinjal (*Solanum melongena* L.) seeds obtained from different sources viz. BADC, BRAC, Local Seed Trader, Local Seed Company and Farmers. *Aspergillus* sp., *Penicillium* sp., *Curvularia* sp., *Rhizopus* sp. and *Phomopsis* sp. were identified in the seeds of BADC. Among the studied fungi, the highest (16%) prevalence of *Aspergillus* sp. was found jointly in the seeds of BADC and farmer followed by Local Seed Company (12%) and BRAC (8%), while it was absent in Local Seed Trader.

Alam (2002) studied the health of some vegetable seeds collected from different sources. He used the blotter method for detecting the seed-borne fungi. He found about six fungi in brinjal seeds such as *Alternaria sp.*, *Aspergillus sp.*, *Penicillium spp.*, *Curvularia sp.*, *Fusarium sp.* and *Rhizopus sp.* From the consequence of his experiment he found that the seed collected from Farmers and Local Seed Traders were infected by all most all fungi, which he detected. He also found that the higher prevalence, of all fungi was also in Farmer and Local Seed Trader seed.

Alam (2001) reported that by using blotter method, he obtained different pathogens from different vegetable seeds. He recorded *Alternaria sp.*, *Aspergillus sp.*, *Penicillium sp.*, *Curvularia sp.* and *Rhizopus sp.* from vegetables seeds. *Aspergillus sp.*, *Penicillium sp.*, *Curvularia sp.*, *Fusarium sp.*, *Phomopsis sp.* and *Rhizopus sp.* from brinjal seeds. *Aspergillus sp.*, *Penicillium sp.* and *Macrophomina sp.* from okra seeds.

Fakir (2000) listed large number of seed borne fungi under 13 different types of vegetables. It was 15 in brinjal, 14 in okra and 13 in chilli seeds. The genera were *Alternaria sp.*, *Aspergillus sp.*, *Colletotricum sp.*, *Fusarium sp.*, *Penicillium sp.*, *Phomopsis sp.*, *Pythium sp.*, *Sclerotium sp.*, *Cercospora sp.*, *Macrophomina sp.*, *Peronospora sp.* and *Rhizopus sp.*

2.3 Fungal pathogens associated with leafy vegetables (amaranth, Indian spinach)

Begum (2012) collected different leafy vegetables viz. Laffa, Mustard, Indian spinach, Jute, Red amaranth, Swamp cabbage, Spinach, Cabbage and Amaranth seeds from different shops of Saidpur. Different seed- borne fungi such as *Alternaria sp.*, *Aspergillus flavus*, *Aspergillus niger*, *Curvularia sp.*, *Fusarium sp.*, *Phoma sp.*, *Penicillium sp.* and *Rhizopus sp.* were detected.

Khanom (2011) collected different leafy vegetables viz. Cabbage, Indian spinach, Indian cabbage, Spinach and Red amaranth seeds from different seed shops of Mymensingh. Different seed borne fungi such as *Alternaria* sp., *Aspergillus niger*, *Curvularia* sp., *Fusarium* sp., *Phoma* sp., *Penicillium* sp. and *Rhizopus* sp. were detected.

Koike and Correll (1993) reported foliar disease symptoms on commercially produced spinach in the saline valley in Monterey country, California, USA. The causal organism was located and identified as *Colletotrichum dematium* and its pathogenicity was confirmed.

Islam (2006) conducted a research to study the germination and health status of Indian spinach, red amaranth, and spinach collected from BADC, local seed company and farmer. He observed nine fungi in bottle gourd where seven fungi in Indian spinach. A fungus in red amaranth and six fungi in Indian spinach. He found that the prevalence of fungi and percent germination varied significantly depending on the seed categories and seed source. He also found, only *Fusarium moniliforme* was capable of transmitting disease to growing seedlings.

Richardson (1990) reported that *Alternaria amaranthi* that carried through the seeds of *Amaranthus* sp. He listed four pathogenic fungi namely *Fusarium oxysporum* f. sp. *lagenariae*, *Lasioidipodia theobromae*, *Macrophomina phaseolina* and *Rhizoctonia solani* in seeds of Bottle gourd. He also reported different fungal pathogens namely *Botrytis cinerea*, *Cladosporium variable*, *Colletotrichum dematium* f. sp. and *Verticillium* sp. were found to be transmitted through the seeds of spinach (*Spinacia oleracea*).

Wu *et al.* (2001) isolated *Colletotrichum dematium* from seeds of diseased amaranth. The amount of seed borne *C. dematium* was positively correlated with the amount of the abnormal seedlings and unmerging seeds of amaranth.

2.4 Fungal pathogen associated with pod vegetable (country bean)

Alves *et al.* (2009) isolated *Collectotrichum gossypii* var. *cephalosporioides*, *Colletotrichum truncatum* and *Colletotrichum lindemuthianum* from common bean (*Phaseolus vulgaris* L.) by the water restriction technique. Considering the seeds submitted to the blotter test, it was possible to identify *Fusarium* sp. on maize, *C. gossypii* var. *cephalosporioides*, *C. truncatum*, and *C. Lindemuthianum* were observed in the surface of inoculated seeds.

Yesuf and Sangchote (2007) made surveys of major seed-borne fungi associated with seeds of common bean (*Phaseolus vulgaris* L) in different bean-growing areas of Ethiopia. The major seed-borne fungi associated with common bean seeds were *Colletotrichum lindemuthianum*, *Phaeoisariopsis griseola* and *Ascochyta phaseolorum*. Bean anthracnose survived in infected seeds but not in the soil, and the primary source of bean anthracnose infection in the field was from infected seeds.

Domijan *et al.* (2003) identified seed-borne fungi on bean (*Phaseolus vulgaris* L.) crops grown in 13 countries of the Republic of Croatia. The most common fungi isolated were *Cladosporium* sp. (98%) *Alternaria* sp. (75%), *Aspergillus* sp. (73%), *Rhizopus* sp. (73%), *Penicillium* sp. (69%), *Fusarium* sp. (38%), and *Fusarium* sp. (38%).

Islam (1990) reported that farmer's seeds were heavily infected by seed-borne pathogens compared to BADC seed.

Fakir (1980) reported that at least 2-3% of the total seeds were rotted in storage per annum due to various fungi and bacteria in Bangladesh and lost to the tons of approximately Taka 430 million.

2.5 Bacteria associated with all vegetable seeds

Seed Pathology Centre (SPC), BAU, Mymensingh carried out an experiment to identify bacterial infection of 16 vegetable crops viz. Bean, Bitter gourd, Bottle gourd, Brinjal, Cabbage, Carrot, Cauliflower, Indian cabbage, Indian spinach, Radish, Red amaranth, Spinach, Sweet gourd, Turnip and Yard long bean collected from different shops of Mymensingh Sadar. *Pseudomonas* sp. were found in association with the seeds of Bean, Bottle gourd, Indian spinach, Radish, Red amaranth, Sweet gourd and Tomato. *Pseudomonas fluorescens* were found with the seeds of cabbage, carrot and Yard long bean. But bacteria have not been found with the seeds of Bitter gourd by Hossain and Purnima, 2011.

Miklas *et al.* (2011) reported Common bacterial blight (CBB; caused by *Xanthomonas oxonopodis* pv. *phaseoli*) a serious disease of dry and green beans (*Phaseolus vulgaris* L.) in warm humid climates. Crop loss up to 40% has been reported due to this disease, which is most prominent east of the continental divide in the United States.

Warriner *et al.* (2005) evaluated the efficacy of seed decontamination to enhance the safety of salad vegetables and herbs. Seeds (celery, coriander, lettuce, spinach and watercress) were inoculated (as a level of 3-5 log cfu g⁻¹) with either *Escherichia coli* P 36 or *Listeria monocytogenes* NCTC 7973 and decontaminated with ozone gas, acidified sodium chlorite (ASC) or quaternary ammonium salt preparation (QAS).

Tu (1980) assayed for seed-borne micro-organism in bean and isolated 10 species of bacteria. Of them 3 were pathogenic: *Xanthomonas campestris* pv. *Phaseoli* (present in 11% of seeds), *Pseudomonas syringae* (1%) and *Pseudomonas syringae* pv. *Pseudomonas syringae* pv. *phaseolicola* (1%).

CHAPTER III

MATERIALS AND METHODS

3.1. Experimental site

The experiments were conducted in Seed Health Laboratory of the Department of Plant Pathology of Shere-e-Bangla Agricultural University (SAU), Shere-e-Bangla Nagar, Dhaka-1207.

3.2. Experimental Period

The experiments were conducted during the period from November, 2015 to April, 2016.

3.3. Collection of seeds

Excluding tested, treated, procured, packed and imported seeds only growers bulk seed of ten vegetables were collected and preserved by the seed trader for the sac to the farmers were collected/taken from three seed stores namely New Bangla seed, Islam Seed and Rafiq Traders of Rangpur district in Bangladesh for conducting series of test for this experimentation. The samples were brought directly to Seed Health Laboratory of Department of Plant Pathology and kept in air tight until use for subsequent studies.

3.4. Vegetable species selected

Seeds of ten different categories of vegetables were studied for their health and quality study.

3.5. Test materials

The categories of vegetables were

I. Leafy vegetables viz. amaranth and Indian Spinach

II. Fruit vegetables viz. bottle gourd, sweet gourd, snake gourd, bitter gourd, cucumber, okra, and brinjal or eggplant.

III. Podded vegetables viz. country bean

Bengali or common name, English name, scientific name and family selected vegetable seeds are shown in Table 1.

Table 1. The list of different vegetable seeds

Common Name	English name	Scientific name	Family
Leafy vegetables			
Data	Amaranth	<i>Amaranthus spinosus</i>	Amaranthaceae
Pui Shakh	Indian spinach	<i>Basella alba</i>	Basellaceae
Fruit vegetables			
Lau	Bottle gourd	<i>Lagenaria siceraria</i>	Cucurbitaceae
Misti kumra	Sweet gourd	<i>Cucurbita moschata</i>	Cucurbitaceae
Korola	Bitter Gourd	<i>Momordica charantia</i>	Cucurbitaceae
Shosha	Cucumber	<i>Cucumis sativus</i>	Cucurbitaceae
Cicinga	Snake gourd	<i>Trichosanthes cucumerina</i>	Cucurbitaceae
Dherosh	Okra	<i>Abelmoschus esculentus</i>	Malvaceae
Begun	Brinjal	<i>Solanum melongena</i>	Solanaceae
Pod vegetables			
Shim	Country Bean	<i>Phaseolus vulgaris</i>	Fabaceae



A. Amaranth seeds



B. Indian spinach seeds



C. Bottle gourd seeds



D. Sweet gourds



E. Bitter gourd seeds



F. Cucumber seeds



G. Okra seeds



H. Brinjal seeds



I. Snake gourd seeds



J. Country bean

Plate 1 (A-J): Working samples of loose vegetable seeds collected from Rangpur Sadar

3.6. Laboratory experiment

A. Seed sampling: Three primary samples of each vegetable were collected from each source. The composite sample was formed by combination and mixing all the primary samples taken from the lot or containers or bags.

B. Working samples: Working sample was obtained by following the methods of Instrumental Rules of ISTA (1999).

3.6.1 Inspection of dry Seeds

The inspection of dry seeds is a method where the presence of fruiting structures of fungi and the effects of fungi on the physical appearance of seeds is examined. The method provides quick information on insect and mechanical damage to the seeds as well as on seed treatment pesticides so that the samples are handled with appropriate precautions. The fruiting structures of fungi can be in the form of pathogenical viz. acervuli, pycnidia, sclerotia on the seed surface or submerged in the seed coat. Physical abnormalities include shriveling of seed coat, reduction or increase in seed size, discoloration or spots in the seed coat. The inspection of dry seed is closely associated with the purity analysis as practiced at seed testing station.

Working procedure: For the inspection of dry seeds the working sample (2-80 g) various with their size and weight. It was advisable to use a sample of the same size as the one recommended for “purity analysis” for different crops by the International Rules for Seed Testing (1999). The seed sample were collected from New Bangla Seed, Islam Seeds and Rafiq Traders. This procedure maintain three times. For the purity analysis weight the three components (pure seeds, inert matter and seeds of other crops) separated and enter the result in the seed health report. Examine the pure seeds by naked eye and under a stereomicroscope, visual inspection helps to separate the abnormalities, shrinkage, swelling of seeds and fruiting structure observed under microscope. After three purity test, results added and divided by three and then get an average purity test result.

3.6.2 Blotter method

A working sample of 400 seeds was used for blotter test for detection of seed borne fungi following the International Rules for Seed Testing (ISTA, 2001). In this incubation method 9 cm diameter pyrex glass petridish and whatman no. 1 filter paper was used. The petridish was treated and incubated for seven days at 22⁰c under 12 hour alternating cycle of light and darkness. After incubation, fungi developed on each seed were examined under different magnifications of a stereomicroscope and identified. The identification of fungi was based on the way they grow on seeds “habit characters” and on the morphological characters of fruiting bodies, spores/conidia observed under a compound microscope.



Plate 2: Incubation of seed

3.6.3. Rolled paper towel method

Seedling infection and seedling vigor test was done by rolled paper towel method (Warham, 1990). In this method, 400 seeds were randomly selected from each sources and placed uniformly between a pair of moist paper towels. The towels were rolled and the two ends also closed with rubber band so that moist could not remove easily. Then the rolled papers containing seeds were

placed in an upright position for 7 days at room temperature under 12 hours photoperiod for 7 days. Length of shoot was measured from the base of the stem up to the growing point of the youngest leaf. Similarly, length of root was measured from starting point of the root to the largest available lateral root apex. Vigor of the seedling was determined by the following formula (Baki and Anderson, 1972).

Vigor index = (Mean of root length + Mean of shoot length) X % of seed germination

3.6.4. Growing on test

A sample of 400 seeds were drawn randomly from each of three replications in all the sources and sown manually in the tray of sand. The spacing maintain between the seeds was 2.0 cm and the raw was 15 cm. The adequate soil moisture was maintained. The emergence count was taken on 15th day of sowing. By this method, shoot length, root length, normal seedling, abnormal seedling, rotten seeds, hard seeds, germination and vigor index were counted.

3.7. Design of experiments: All the laboratory experiment were conducted following Complete Randomized Design (CRD) with three replications.

3.8. Analysis of data

The data obtained for different characters were statistically analyzed by “MSTAT-C” program to find out the significance of the difference levels of loose seeds collected from three seeds store. The mean values of all the characters were evaluated and analysis of variance was performing by the ‘F’ (variance ratio) test. The significance of the difference among the treatment combinations means was estimated by the Duncan’s Multiple Range Test (DMRT) at 5% level of probability (Gomez and Gomez, 1984).

CHAPTER IV

RESULTS AND DISCUSSION

4.1 Seed health status of leafy vegetables

Fungi detected in leafy vegetable viz. amaranth seeds that were collected from different seed shops like as New Bangla Seeds, Islam Seeds store and Rafiq Traders of Rangpur Sadar presented in different table and figures. Prevalence of seed-borne fungal infections varied depending on the seeds of different vegetables.

4.1.1 Seed health study of amaranth (*Amaranthus spinosus*)

4.1.1.1 Health status of amaranth seeds by inspection of dry seed method

A. Physical appearance of seeds: No physical abnormalities were observed in amaranth seeds by naked eye.

B. Presence of fruiting structures of fungi: No fungal fruiting structures were observed over the seed surface.

C. Purity Analysis

For purity analysis of three different store seeds two gm seeds of amaranth from each source were observed by naked eye. No inert matter and other crop seeds were found from any seed sources (Table 2).

4.1.1.2. Prevalence of seed-borne fungi of amaranth identified by blotter method

The fungal incidence varied significantly among the samples for all pathogens at 5 % level of significance.

A. Identified pathogen

Six fungal pathogens viz. *Aspergillus flavus*, *Aspergillus niger*, *Fusarium sp.*, *Alternaria sp.*, *Chaetomium spp.*, *Rhizopus sp.* and one unidentified bacteria was observed and recorded.

B. Incidence of seed borne pathogens

In case of fungal infection, the highest incidence was observed from the seed collected from New Bangla Seed store. However, the highest bacterial infection was recorded from another two sources. Incidence of *Aspergillus flavus*, *A. niger* were varied from 6.25 to 7.5% where, the highest was observed in the seeds of New Bangla Seed and the lowest was in Islam Seed. Incidence of *Fusarium sp.* was varied from 2.75 to 3.5%. Incidence of *Alternaria sp.* was varied from 2 – 4%, where the highest incidence was observed in Islam seed and lowest was in Islam seeds. *Chaetomium spp.* and *Rhizopus sp.* was varied from 5 – 6.5% and 3 – 4%, respectively (Table 3).

Table 2. Health status of amaranth seeds by inspection of dry seed method

Seed Sources	Total weight (g)	Pure seeds (g)	Pure seeds (%)	Inert matter (g)	Inert matter (%)	Other seeds (g)	Other seeds (%)	Fungal structures	Physical abnormalities of seed
New Bangla Seed	2	2	100	0	0	0	0	Absence	Absent
Islam Seeds	2	2	100	0	0	0	0	Absence	Absent
Rafiq Traders	2	2	100	0	0	0	0	Absence	Absent

Table 3. Prevalence of seed-borne fungi of amaranth identified by blotter method

Source Name	% Pathogen incidence							% Seed infection
	<i>Aspergillus flavus</i>	<i>Aspergillus niger</i>	<i>Fusarium</i> sp.	<i>Alternaria</i> sp.	<i>Chaetomium</i> spp.	<i>Rhizopus</i> sp.	Unidentified bacteria	
New Bangla Seeds	7.50 a	7.50 a	3.50 a	3.50 a	6.50 a	4.00 a	1.75 b	34.25
Islam Seeds	6.25 b	6.25 b	2.75 b	4.00 a	5.00 b	3.00 b	2.25 a	29.5
Rafiq Traders	7.25 a	7.25 a	3.50 a	2.00 b	6.50 a	3.00 b	2.25 a	31.75
LSD	0.997	0.843	0.596	0.565	0.843	0.653	0.460	
Level of significance	*	**	*	**	**	**	*	
CV %	8.91	7.53	11.47	11.16	8.78	12.25	13.86	

*= Significant at 5% level of significant as per DMRT, ** = Significant at 1% level of significant as per DMRT

4.1.2 Seed health study of Indian spinach (*Basella alba*)

4.1.2.1 Health status of Indian spinach seeds by inspection of dry seed method

A. Physical appearance of seeds: Shrinkage and deformed seeds were observed from the seeds collected from three stores for experiment (Table 4).

B. Presence of pathogenic structures of fungi: Only mycelium was observed over the seed surface.

C. Purity Analysis

In case of purity analysis of three different store seeds 60 gm seed of Indian spinach was seen in naked eye. Then determine by hand pure seed, inert matter and other crops seeds. Result of purity analysis given below Table 20 total seeds 60 gm, pure seeds 58 gm (96.7 %), inert matter 2 gm (3.3 %) and other crop seeds were not observed in case of New Bangla Seeds. But in Islam Seeds were observed pure seeds 59 gm (98.33 %), inert matter 1 gm (1.67 %) and other crop seeds were not observed. Pure seeds 57 gm (95 %), inert matter 3 gm (5 %) and no observation of other crops were found in Rafiq Traders seeds (Table 4).

4.1.2. 2. Prevalence of seed-borne fungi of Indian spinach identified by blotter method

The fungal incidence varied significantly among the samples for all pathogens at 5 % level of significance.

A. Identified pathogen

Aspergillus flavus, *Aspergillus niger*, *Fusarium* sp., *Alternaria* sp., *Chaetomium* spp., *Rhizopus* sp. and Unidentified bacteria were observed.

B. Incidence of seed borne pathogens

In case of fungal infection, the highest incidence was observed from the seed collected from Islam seeds. However, the highest bacterial infection was recorded in Islam seeds. Incidence of *Aspergillus flavus*, *A. niger* were varied from 5.75 to 8.0% where, the highest was observed in the seeds of New Bangla Seed and the lowest was in Islam Seed store. Incidence of *Fusarium* sp. was varied from 2.87 to 3.5%. Incidence of *Alternaria* sp. was varied from 2.25 – 2.50%, where the highest incidence was observed in New Bangla seed and lowest was in Islam seeds. *Chaetomium* spp. and *Rhizopus* sp. was varied from 5.50 – 6.0% and 5.25 – 6.0% respectively (Table 5).

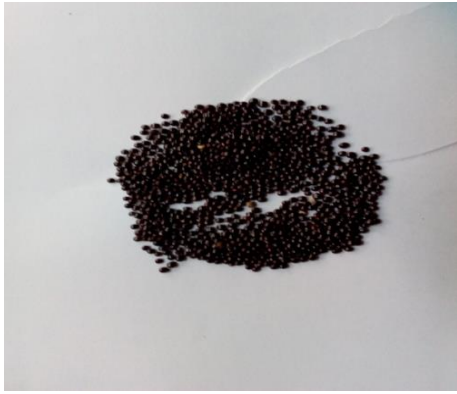
Table 4. Health status of Indian spinach seeds by inspection of dry seed method

Seed Sources	Total weight (g)	Pure seeds (g)	Pure seeds (%)	Inert matters (g)	Inert matters (%)	Other seeds (g)	Other seeds (%)	Fungal structure	Physical abnormalities of seed
New Bangla Seeds	60	57	95	3	5	0	0	Mycelium	Shrinkage, Deformed
Islam Seeds	60	58	96.7	2	3.3	0	0	Mycelium	Shrinkage, Deformed
Rafiq Traders	60	59	98.33	1	1.67	0	0	Mycelium	Shrinkage, Deformed

Table 5. Prevalence of seed-borne fungi of Indian spinach identified by blotter method

Source Name	% Pathogen incidence							% Seed infection
	<i>Aspergillus flavus</i>	<i>Aspergillus niger</i>	<i>Fusarium</i> sp.	<i>Alternaria</i> sp.	<i>Chaetomium</i> spp.	<i>Rhizopus</i> sp.	Unidentified bacteria	
New Bangla Seeds	8.00 a	5.75 b	3.50 a	2.50 a	6.00 ab	6.00 a	2.25 b	34
Islam Seeds	6.50 b	8.00 a	2.87a	2.75a	6.75 a	5.25 a	3.50 a	35.62
Rafiq Traders	8.00 a	7.25 a	3.25 a	2.25 a	5.50 b	5.25 a	2.75 b	34.22
LSD	1.25	1.06	0.741	0.532	0.884	1.13	0.532	
Level of significance	*	**	Ns	ns	*	ns	**	
CV %	10.42	9.52	14.46	13.33	9.09	12.86	11.76	

*= Significant at 5% level of significant as per DMRT, ** = Significant at 1% level of significant as per DMRT



A. Pure seeds of amaranth

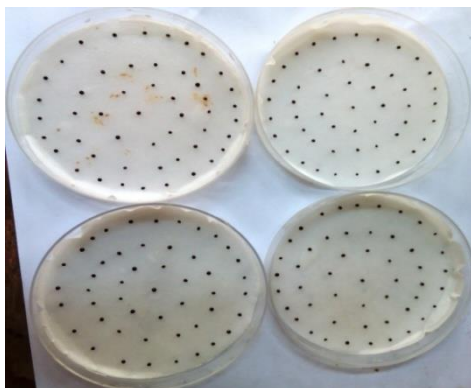


B. Pure of Indian spinach



B. Inert matter of Indian spinach

Plate 3: Pure seeds and inert matter observed by dry inspection method



A. Seed health of amaranth by blotter method



B. Seed health of Indian spinach by blotter method

Plate 4 (A-B): Seed health of amaranth and Indian spinach by blotter method

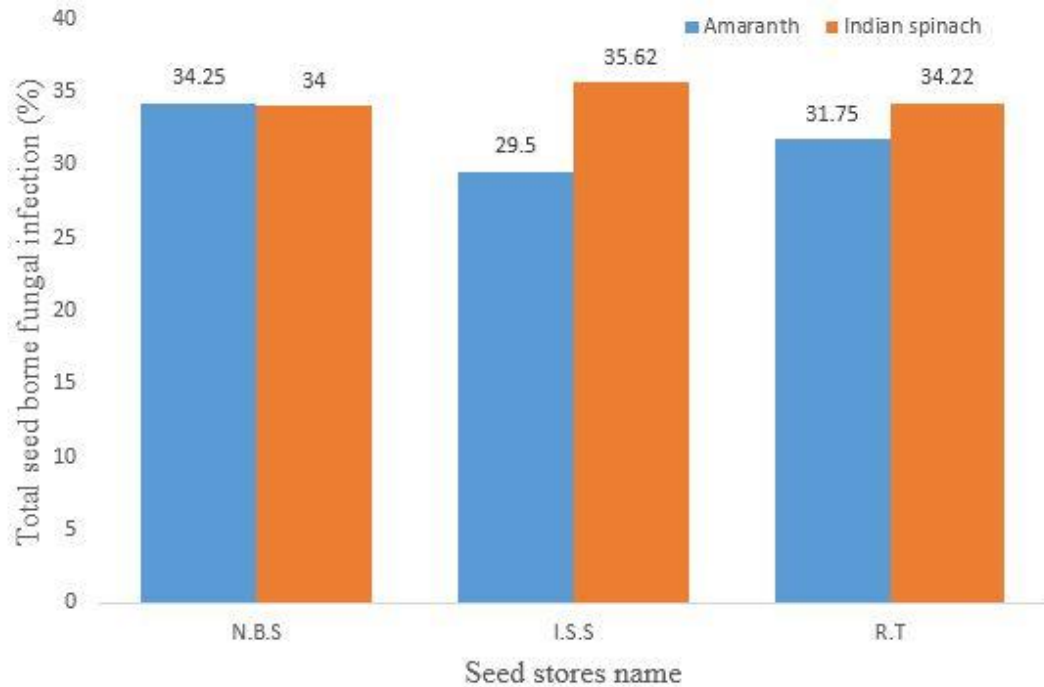


Figure 1: Total seed borne fungal infection (%) of leafy vegetables

N.B.S. - New Bangla seeds, I.S.S- Islam seeds, R.T. - Rafiq traders

The present study indicate that in all seeds store showed the highest value seed borne fungal infection for Indian spinach (34%, 35.62%, 34.22%). Whereas seeds of amaranth showed the lowest value seed borne fungal infection for all seeds stores.

4.2 Seed health status of fruit vegetables

Fungi detected in fruit vegetable seeds namely bottle gourd, brinjal, cucumber, bitter gourd, sweet gourd, snake gourd and okra seeds that were collected from different seed shops like as New Bangla Seeds, Islam Seeds store and Rafiq Traders of Rangpur Sadar presented in table and figures. Prevalence of seed-borne fungal infections varied depending on the seeds of different vegetable seed sources.

4.2.1 Seed health study of bottle gourd (*Lagenaria siceraria*)

4.2.1.1 Health status of bottle gourd seeds by inspection of dry seed

A. Physical appearance of seeds

Deformed seeds and discoloration of seeds were observed in the seeds collected from New Bangla seeds, whereas shrinkage and deformed of seeds were observed in the seeds that collected from Islam Seeds and deformed seeds were observed in the seeds collected from Rafiq traders (Table 6).

B. Presence of pathogenic structures of fungi: Acervuli and mycelium were observed in all seeds that collected from three seeds stores (Table 6).

C. Purity analysis

In case of purity analysis of three store seeds 80 gm seeds of bottle gourd were seen in naked eye. Then determine by hand; pure seeds, inert matter and other crops seeds. Total seed 80 gm, pure seeds 78.3 gm (97.8 %), inert matter 0.5 gm (0.625 %) and other crop seeds 1.12 gm (1.4 %) were observed in case of New Bangla seeds. But in Islam seeds seeds were observed pure seeds 79.05 gm (98.8 %), inert matter 0.92 gm (1.15 %) and other crop seeds no observed. Moreover, from Rafiq traders seeds, the pure seeds 78.04 gm (97.55 %), inert matter 0.83 gm (1.03 %) and other crop seeds 1.12 gm (1.5 %) were observed (Table 6).

4.2.1.2. Prevalence of seed-borne fungi of bottle gourd identified by blotter method

A. Identified pathogen: *Aspergillus flavus*, *Aspergillus niger*, *Fusarium* sp., *Alternaria* sp., *Chaetomium* spp., *Rhizopus* sp. and unidentified bacteria were observed.

B. Incidence of seed borne pathogens

The highest incidence was observed from the seed collected from New Bangla Seed in case of fungal infection. However, the highest bacterial infection was recorded from New Bangla seeds. Incidence of *Aspergillus flavus*, were varied from 6.0 to 8.0 % where, the highest was observed in the seeds of Rafiq traders and the lowest was in New Bangla Seeds. Incidence of *A. niger* varied from 5.0 to 6.25%. Highest infection was observed in Islam seeds (6.25%). Incidence of *Fusarium* sp. was varied from 2.75 to 4.0 %. Incidence of *Alternaria* sp. was varied from 2.75 – 4.25 %, where the highest incidence was observed in New Bangla Seed and lowest was in Rafiq traders. *Chaetomium* spp. and *Rhizopus* sp. were varied from– 4.5-5.75% and 5 – 6.25%, respectively (Table 7).

Table 6. Health status of bottle gourd seeds by inspection of dry seed method

Seed Sources	Total weight (g)	Pure seeds (g)	Pure seeds (%)	Inert matters (g)	Inert matters (%)	Other seeds (g)	Other seeds (%)	Fungal structure	Physical abnormalities of seed
New Bangla Seeds	80	78.3	97.8	0.5	0.625	1.2	1.5	Acervuli	Deformed
Islam Seeds	80	79.05	98.8	0.92	1.15	0	0	Acervuli	Shrinkage, Reduced
Rafiq Traders	80	78.04	97.55	0.83	1.03	1.12	1.4	Acervuli	Deformed

Table 7. Prevalence of seed-borne fungi of bottle gourd identified by blotter method

Source Name	% Pathogen incidence							% Seed infection
	<i>Aspergillus flavus</i>	<i>Aspergillus niger</i>	<i>Fusarium</i> sp.	<i>Alternaria</i> sp.	<i>Chaetomium</i> spp.	<i>Rhizopus</i> sp.	Unidentified bacteria	
New Bangla Seeds	6.5 b	5.75 ab	4.00a	4.25 a	5.25 ab	5.50 ab	4.50 a	35.75
Islam Seeds	6.0 b	6.25 a	2.75b	3.50 b	5.75 a	6.25 a	2.50 b	33
Rafiq Traders	8.0 a	5.0 b	3.25b	2.75 c	4.50 b	5.00 b	1.50 c	30
LSD	1.193	0.997	0.532	0.653	0.753	0.799	0.653	
Level of significance	**	**	**	**	**	**	**	
CV %	10.91	11.0	10.0	11.66	9.12	8.96	10.50	

** = Significant at 1% level of significant as per DMRT



A. Pure seed of bottle gourd



B. Inert matter of bottle gourd



C. Other seeds

Plate 5 (A-C): Seed health of bottle gourd by dry inspection method



Plate 6: Seed health of bottle gourd by blotter method

4.2.2 Seed health study of sweet gourd (*Cucurbita moschata*)

4.2.2.1 Health status of sweet gourd seeds by inspection of dry seed method

A. Physical appearance of seeds

Shrinkage seeds were observed in the seeds collected from New Bangla seeds and Islam Seeds, whereas shrinkage and deformed of seeds was observed in the seeds that collected from Rafiq Traders (Table 8).

B. Presence of pathogenic structures of fungi: Acervuli and mycelium were observed over the seed surface of all seeds that collected from three seeds store (Table 8).

C. Purity analysis

In case of purity analysis of three store seeds 80 gm seeds of sweet gourd were seen in naked eye. Then determine in hand pure seeds, inert matter and other crops seeds. Total seed 80 gm, pure seeds 77 gm (96.25 %), inert matter 1.8 gm (2.25 % and other crop seeds 1.2 gm (1.5%) were observed in case of New Bangla Seeds. But in Islam Seeds seeds were observed pure seeds 78 gm (97.5 %), inert matter 1.3 gm (1.625 %) and other crop seeds were observed 0.7 gm (0.87%). Moreover, from Rafiq traders seeds, the pure seeds 79 gm (98.75 %), inert matter 0.3 gm (0.375 %) and other crop seeds 0.7 gm (0.87 %) were observed (Table 8).

4.2.2.2 Prevalence of seed-borne fungi of sweet gourd identified by blotter method

A. Identified pathogen

Aspergillus flavus, *Aspergillus niger*, *Fusarium* sp., *Altenaria* sp., *Chaetomium* spp., *Rhizopus* sp. and unidentified bacteria were observed.

B. Incidence of seed borne pathogens

In case of fungal infection, the highest incidence was observed from the seed collected from New Bangla Seeds. However, the highest bacterial infection was recorded from all seeds store. Incidence of *Aspergillus flavus*, *A. niger* were varied from 8.50 to 7.75 % and 8.25 to 9.0 % where, the highest was observed in the seeds of Islam seeds and the lowest was in Rafiq traders. Incidence of *Fusarium* sp. was varied from 4.25 to 5.0 %. Incidence of *Alternaria* sp. was varied from 4.25 – 4.75 %. But in case of *Chaetomium* spp. and *Rhizopus* sp. were varied from 8 – 9 % and 7.5 – 8.75%, respectively (Table 9).

Table 8. Health status of sweet gourd seeds by inspection of dry seed method

Seed Sources	Total weight (g)	Pure seeds (g)	Pure seeds (%)	Inert matters (g)	Inert matters (%)	Other seeds (g)	Other seeds (%)	Pathogenic structures	Physical abnormalities of seed
New Bangla Seeds	80	77	96.25	1.8	2.25	1.2	1.5	Acervuli	Shrinkage
Islam Seeds	80	78	97.5	1.3	1.625	0.7	0.87	Acervuli	Shrinkage
Rafiq Traders	80	79	98.75	0.3	0.375	0.7	0.87	Acervuli	Shrinkage, Deformed

Table 9. Prevalence of seed-borne fungi of sweet gourd identified by blotter method

Source Name	% Pathogen incidence							% Seed infection
	<i>Aspergillus flavus</i>	<i>Aspergillus niger</i>	<i>Fusarium</i> sp.	<i>Alternaria</i> sp.	<i>Chaetomium</i> spp.	<i>Rhizopus</i> sp.	Unidentified bacteria	
New Bangla Seeds	8.25a	9.00a	4.50 a	4.25 a	9.00 a	8.75 a	3.75 a	47.5
Islam Seeds	8.50a	8.25b	4.25 a	4.50 a	8.25 a	7.50 a	4.00 a	45.25
Rafiq Traders	7.75a	9.00 a	5.00 a	4.75 a	8.00 a	7.75 a	4.25 a	46.5
LSD	1.25	0.704	0.884	0.923	1.16	1.25	0.843	
Level of significance	ns	**	ns	ns	ns	**	**	
CV %	9.57	5.04	10.06	8.83	8.63	9.77	11.18	

*= Significant at 5% level of significant as per DMRT, ** = Significant at 1% level of significant as per DMRT



A. Pure seed of bottle gourd



B. Inert matter of bottle gourd



C. Other seed

Plate 7 (A-C): Seed health of sweet gourd by dry inspection method



Plate 8: Seed health of sweet gourd by blotter method

4.2.3 Seed health study of bitter gourd (*Momordica charantia*)

4.2.3.1 Health status of bitter gourd seeds by inspection of dry seed

A. Physical appearance of seeds

Deformed and black spot seeds were observed in the seeds collected from New Bangla seeds whereas shrinkage and deformed of seeds was observed in the seeds that collected from Islam Seeds. But swelling and deformed seeds was observed which seeds that collected from Rafiq Traders (Table 10).

B. Presence of pathogenic structures of fungi: Mycelium was observed over all seeds surface that collected from three seeds store (Table 10).

C. Purity Analysis

In case of purity analysis of three store seeds 80 gm seeds of bitter gourd were seen in naked eye. Then determine in hand pure seeds, inert matter and other crops seeds. Total seed 80 gm, pure seeds 79.46 gm (99.32 %), inert matter 0 gm (0 %) and other crop seeds 0.54 gm (0.67%) were observed in case of New Bangla Seeds. But in Islam Seeds seeds were observed pure seeds 79.02 gm (98.77 %), inert matter 0.3 gm (0.37 %) and other crop seeds were observed 0.6 gm (0.75%). Moreover, from Rafiq Traders seeds, the pure seeds 80 gm (100 %), inert matter and other crop seeds were observed 0 gm (Table 10).

4.2.3.2 Prevalence of seed-borne fungi of bitter gourd identified by blotter method

A. Identified pathogen

Aspergillus flavus, *Aspergillus niger*, *Fusarium* sp., *Alternaria* sp., *Chaetomium* spp., *Rhizopus* sp. and unidentified bacteria were observed.

B. Incidence of seed borne pathogens

The highest incidence was observed from the seed collected from Rafiq Traders in case of fungal infection. However, the highest bacterial infection was also recorded from Rafiq Traders. Incidence of *Aspergillus flavus*, were varied from 7.25 to 8.0 % where, the highest was observed in the seeds of Rafiq Traders and the lowest was in New Bangla Seed. Incidence of *A. niger* varied from 5.75 to 7 %. Highest infection was observed in Rafiq Trader's (7 %). Incidence of *Fusarium* sp. was varied 4.75 % 6.75 to 9 %, where the highest incidence was observed in Rafiq Trasers. In case of *Rhizopus* sp. was varied from 4 – 6.5% respectively. The highest incidence was recorded in Rafiq Traders and lowest in New Bangla Seeds (Table 11).

Table 10. Health status of bitter gourd seeds by inspection of dry seed method

Seed Sources	Total weight (g)	Pure seeds (g)	Pure seeds (%)	Inert matters (g)	Inert matters (%)	Other seeds (g)	Other seeds (%)	Fruiting structures	Physical abnormalities of seed
New Bangla Seeds	80	79.46	99.32	0	0	0.54	0.67	Mycelium	Deformed, Black spot
Islam Seeds	80	79.02	98.77	0.3	0.37	0.6	0.75	Mycelium	Deformed, Shrinkage
Rafiq Traders	80	80	100	0	0	0	0	Mycelium	Swelling, Deformed

Table 11. Prevalence of seed-borne fungi of bitter gourd identified by blotter method

Source Name	% Pathogen incidence							% Seed infection
	<i>Aspergillus flavus</i>	<i>Aspergillus niger</i>	<i>Fusarium</i> sp.	<i>Alternaria</i> sp.	<i>Chaetomium</i> spp.	<i>Rhizopus</i> sp.	Unidentified bacteria	
New Bangla Seeds	7.25 a	6.75a	3.25ab	3.75b	7.50b	4.00 c	3.50 b	36
Islam Seeds	7.50 a	5.75 b	3.00 b	3.75 b	6.75 b	5.25 b	3.50 b	35.5
Rafiq Traders	8.00 a	7.00 a	3.75 a	4.75 a	9.00 a	6.50 a	4.25 a	43.25
LSD	0.884	0.843	0.532	0.596	1.16	0.799	0.704	
Level of significance	ns	**	**	**	**	**	**	
CV %	7.29	8.11	10.00	9.13	9.37	9.52	11.76	

ns = Non significant, ** = Significant at 1% level of significant as per DMRT



A. Pure seeds



B. Inert matter

Plate 9 (A-C): Seed health of bitter gourd by dry inspection method



Plate 10: Seed health of bitter gourd by blotter method

4.2.4 Seed health study of cucumber (*Cucumis sativus*)

4.2.4.1 Health status of cucumber seeds by inspection of dry seed

A. Physical appearance of seeds

Deformed seeds was observed in the seeds which collected from New Bangla seeds whereas shrinkage and deformed of seeds was observed in the seeds that collected from Islam seeds and Rafiq Traders (Table 12).

B. Presence of fruiting structures of fungi: Pathogenic structure was not observed (Table 12).

C. Purity analysis

In case of purity analysis of three store seeds 80 gm seeds of cucumber were seen in naked eye. Then determine by hand pure seeds, inert matter and other crops seeds. Total seed 80 gm, pure seeds 79 gm (98.75%), inert matter 1 gm (1.25 %) and other crop seeds 0 gm (0 %) were observed in case of New Bangla seeds. But in Islam seeds seeds were observed pure seeds 79.59 gm (99.37 %), inert matter 0.6 gm (0.75 %) and other crop seeds were not observed. Moreover, from Rafiq Traders seeds, the pure seeds 79.4 gm (99.25 %), inert matter 0.6 gm (0.75%) but other crop seeds were observed 0 gm (Table 12).

4.2.4.2 Prevalence of seed-borne fungi of cucumber identified by blotter method

A. Identified pathogen

Aspergillus flavus, *Aspergillus niger*, *Fusarium* sp., *Alternaria* sp., *Chaetomium* spp., *Rhizopus* sp. and unidentified bacteria were recorded.

B. Incidence of seed borne pathogens

In case of fungal infection, the highest incidence was observed from the seed collected from Rafiq Traders. However, the highest bacterial infection was recorded from Rafiq Traders. Incidence of *Aspergillus flavus* was varied from 5.53 to 6.5 % where, the highest was observed in the seeds of New Bangla seeds store and the lowest was in Islam Seeds. Incidence of *A. niger* varied from 6 to 6.5 %. Highest infection was observed in New Bangla Seeds. Incidence of *Fusarium* sp. was varied 3.25 to 4 %, where the highest incidence was observed in New Bangla seeds. For *Alternaria* sp., ranged varied from 3 to 3.75%. In case of *Chaetomium* spp., was varied from 5.75 to 6.25 %. In case of *Rhizopus* sp. was varied from 4.75 to 6.75% respectively. The highest incidence was recorded in Rafiq Traders and lowest in New Bangla seeds (Table 13).

Table 12. Health status of cucumber seeds by inspection of dry seed method

Seed Sources	Total weight (g)	Pure seeds (g)	Pure seeds (%)	Inert matters (g)	Inert matters (%)	Other seeds (g)	Other seeds (%)	Pathogenic structure	Physical abnormalities of seed
New Bangla Seeds	80	79	98.75	1	1.25	0	0	No observed	Deformed
Islam Seeds	80	79.5	99.37	0.6	0.75	0.6	0.75	No observed	Deformed, Shrinkage
Rafiq Traders	80	79.4	99.25	0.6	0.75	0	0	No observed	Swelling, Deformed

Table 13. Prevalence of seed-borne fungi of cucumber identified by blotter method

Source Name	% Pathogen incidence							% Seed infection
	<i>Aspergillus flavus</i>	<i>Aspergillus niger</i>	<i>Fusarium</i> sp.	<i>Alternaria</i> sp.	<i>Chaetomium</i> spp.	<i>Rhizopus</i> sp.	Unidentified bacteria	
New Bangla Seeds	6.50 a	6.50a	4.00 a	3.25ab	5.75 a	4.75 b	3.00 b	33.75
Islam Seeds	5.53 b	6.00a	3.25 b	3.00 b	6.00 a	6.25 a	3.75 ab	33.78
Rafiq Traders	5.75 b	6.25a	3.75ab	3.75 a	6.25 a	6.75 a	4.25 a	36.75
LSD	0.706	0.596	0.653	0.653	0.843	0.884	0.753	
Level of significance	*	ns	*	*	ns	**	**	
CV %	7.46	5.96	11.13	12.25	8.78	9.34	10.86	

ns = non-significant, *= Significant at 5% level of significant as per DMRT, ** = Significant at 1% level of significant as per DMRT



A. Pure seeds



B. Inert matter

Plate 11: Seed health of cucumber by dry inspection method



Plate 12: Seed health of cucumber by blotter method

4.2.5 Seed health study of snake gourd (*Trichosanthes cucumerina*)

4.2.5.1 Health status of snake gourd seeds by inspection of dry seed

A. Physical appearance of seeds

Deformed seeds was observed in the seeds collected from New Bangla seeds whereas shrinkage and deformed of seeds was observed in the seeds that collected from Islam seeds and Rafiq Traders (Table 14).

B. Presence of pathogenic structures of fungi: White mycelium was observed seeds that collected from New Bangla seeds. But white cottony mycelium and mycelium were observed in both case of two seeds store (Table 14).

C. Purity analysis

In case of purity analysis of three store seeds 80 gm seeds of snake gourd were seen in naked eye. Then determine by hand pure seeds, inert matter and other crops seeds. Total seed 80 gm, pure seeds 78 gm (97.5%), inert matter 1.4 gm (1.75 %) and other crop seeds 0.6 gm (0.75 %) were observed in case of New Bangla seeds store. But in Islam Seeds seeds were observed pure seeds 79 gm (98.75 %), inert matter 0.4 gm (0.5 %) and other crop seeds 0.6 (0.75 %) were observed. Moreover, from Rafiq Traders seeds, the pure seeds 80 gm (100 %), but inert matter and other crop seeds were not observed (Table 14).

4.2.5.2 Prevalence of seed-borne fungi of snake gourd identified by blotter method

A. Identified pathogen

Aspergillus flavus, *Aspergillus niger*, *Fusarium* sp., *Alternaria* sp., *Chaetomium* spp., *Rhizopus* sp. and unidentified bacteria were recorded.

B. Incidence of seed borne pathogens

In case of fungal infection, the highest incidence was observed from the seed collected from Rafiq Traders. Whereas, the highest bacterial infection was recorded from New Bangla seeds. Incidence of *Aspergillus flavus* and *A. niger* were varied from 7.25 to 8.5 % and 6 to 6.5 %. Incidence of *Fusarium* sp. was varied 2.5 to 4 %, where the highest incidence was observed in Islam seeds store. For *Alternaria* sp., ranged varied from 3 to 3.5%. In case of *Chaetomium* spp., was varied from 6.5 to 8.5 %. The highest incidence was recorded in Rafiq traders and lowest in Islam seeds. In case of *Rhizopus* sp. was varied from 5.75 to 6.75%, respectively (Table 15).

Table 14. Health status of snake gourd seeds by inspection of dry seed method

Seed Sources	Total weight (g)	Pure seeds (g)	Pure seeds (%)	Inert matters (g)	Inert matters (%)	Other seeds (g)	Other seeds (%)	Fruiting structures	Physical abnormalities of seed
New Bangla Seeds	80	78	97.5	1.4	1.75	0.6	0.75	White mycelium	Deformed
Islam Seeds	80	79	98.75	0.4	0.5	0.4	0.75	Cottony mycelium	Deformed, Shrinkage
Rafiq Traders	80	80	100	0	0	0	0	Mycelium	Swelling, Deformed

Table 15. Prevalence of seed-borne fungi of snake gourd identified by blotter method

Source Name	% Pathogen incidence							% Seed infection
	<i>Aspergillus flavus</i>	<i>Aspergillus niger</i>	<i>Fusarium</i> sp.	<i>Alternaria</i> sp.	<i>Chaetomium</i> spp.	<i>Rhizopus</i> sp.	Unidentified bacteria	
New Bangla Seeds	7.25a	6.00a	2.50b	3.00 a	7.25 b	5.75b	2.75a	34.5
Islam Seeds	8.50a	6.50a	4.00a	3.00 a	6.50 b	6.25ab	2.25 b	37
Rafiq Traders	8.50 a	6.00 a	3.75a	3.50 a	8.50 a	6.75 a	2.25 b	39.25
LSD	1.27	0.753	0.704	0.653	0.884	0.799	0.460	
Level of significance	ns	ns	**	*	**	*	*	
CV %	9.89	7.64	12.91	12.89	7.45	8.00	11.95	

ns= non-significant, *= Significant at 5% level of significant as per DMRT,** = Significant at 1% level of significant as per DMRT



A. Pure seeds



B. Other seeds

Plate 13: Seed health of snake gourd by dry inspection method



Plate 14: Seed health of snake gourd by blotter method

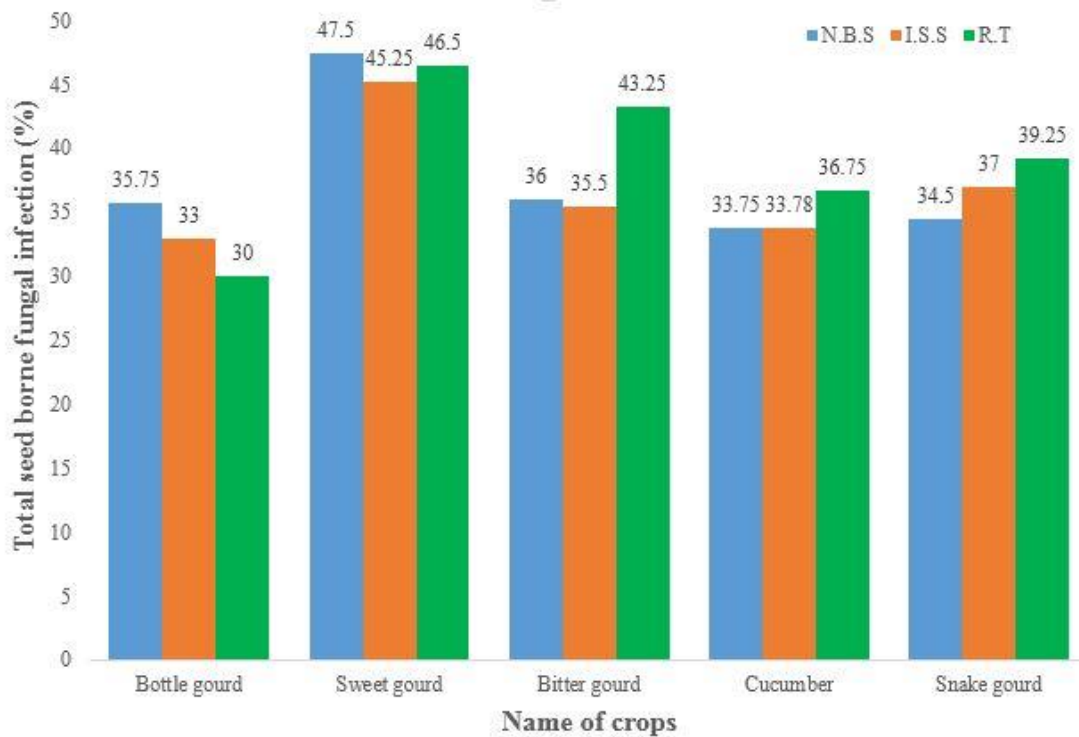


Figure 2: Total seed borne fungal infection (%) of Cucurbitaceae vegetables seeds of different stores

N.B.S. - New Bangla seeds, I.S.S- Islam Seeds, R.T. - Rafiq Traders

The figure 2 indicate that New Bangla seeds showed highest seed borne fungal infection for bottle gourd (35.75 %), sweet gourd (47.5%) and snake gourd (39.25%). Whereas, bitter gourd and cucumber seeds that collected from Rafiq traders showed highest fungal infection. But Islam seeds showed moderate value of seed borne fungal infection for most of the seeds.

4.2.6 Seed health study of okra (*Abelmoschus esculentus*)

4.2.6.1 Health status of okra seeds by inspection of dry seed

A. Physical appearance of seeds

Black spot seeds were observed in the seeds collected from New Bangla seeds whereas shrinkage, deformed and black spot of seeds was observed in the seeds that collected from Islam seeds and Rafiq traders (Table 16).

B. Presence of pathogenic structures of fungi: *Aspergillus* and mycelium was observed seeds that collected from New Bangla seeds. But white cottony mycelium and mycelium were observed in both case of two seeds store (Table 16).

C. Purity analysis

In case of purity analysis of three store seeds 20 gm seeds of okra were seen in naked eye. Then determine by hand pure seeds, inert matter and other crops seeds. Total seed 20 gm, pure seeds 18.5 gm (92.5%), inert matter 1.3gm (6.5 %) and other crop seeds 0.2 gm (1 %) were observed in case of New Bangla seeds. But in Islam Seeds seeds were observed pure seeds 18.99 gm (94.95 %), inert matter 0.2 gm (1 %) and other crop seeds 1.02 (5.1 %) were observed. Moreover, from Rafiq Traders seeds, the pure seeds 18 gm (90 %), but inert matter 1.2 gm (6%) and other crop seeds were observed 0.8 gm (4 %) (Table 16).

4.2.6.2 Prevalence of seed-borne fungi of okra identified by blotter method

A. Identified pathogen

Aspergillus flavus, *Aspergillus niger*, *Fusarium* sp., *Alternaria* sp., *Chaetomium* spp., *Rhizopus* sp. and unidentified bacteria were observed.

B. Incidence of seed borne pathogens

In case of fungal infection, the highest incidence was observed from the seed collected from New Bangla seeds. However, the highest bacterial infection was recorded in New Bangla Seeds and Islam Seeds. Incidence of *Aspergillus flavus* was varied from 7.5 to 9%. The Highest was observed in New Bangla seeds. In case of *A. niger*, ranged varied from 7.5 to 8.75 %. Incidence of *Fusarium* sp. was varied 3.75 to 4 %. For *Alternaria* sp., ranged varied from 3.5 to 4.5%. Highest was observe in New Bangla seeds but lowest in other two stores seeds. In case of *Chaetomium* spp. was varied from 3.75 to 6.25 %. The highest incidence was recorded in Rafiq Traders and lowest in Islam Seeds. In case of *Rhizopus* sp. was varied from 4 to 5.25%, respectively (Table 17).

Table 16. Health status of okra seeds by inspection of dry seed method

Seed Sources	Total weight (g)	Pure seeds (g)	Pure seeds (%)	Inert matters (g)	Inert matters (%)	Other seeds (g)	Other seeds (%)	Pathogenic structure	Physical abnormalities of seed
New Bangla Seeds	20	18.5	92.5	1.3	6.5	0.2	1	Mycelium	Block Spot
Islam Seeds	20	18.99	94.95	0.2	1	1.02	5.1	Mycelium	Deformed, Shrinkage
Rafiq Traders	20	18	90	1.2	6	0.8	4	Cottony mycelium	Deformed, Shrinkage

Table 17. Prevalence of seed-borne fungi of okra identified by blotter method

Source Name	% Pathogen incidence							% Seed infection
	<i>Aspergillus flavus</i>	<i>Aspergillus niger</i>	<i>Fusarium</i> sp.	<i>Alternaria</i> sp.	<i>Chaetomium spp.</i>	<i>Rhizopus</i> sp.	Unidentified bacteria	
New Bangla Seeds	9.00 a	8.75 a	4.00 a	4.50 a	5.50 a	4.75 ab	3.75 a	40.25
Islam Seeds	8.00 ab	8.00 ab	4.00 a	3.50 b	6.25 a	5.25 a	3.50 a	38.5
Rafiq Traders	7.50 b	7.50 b	3.75 a	3.50 b	3.75 b	4.00 b	2.75 b	32.75
LSD	1.35	1.03	0.596	0.843	0.843	0.843	0.532	
Level of significance	*	*	ns	*	**	*	**	
CV %	10.41	7.99	9.52	13.75	10.20	11.29	10.00	

ns = non-significant, *= Significant at 5% level of significant as per DMRT, ** = Significant at 1% level of significant as per DMRT



A. Pure seeds



B. Inert matter

Plate 15: Seed health of okra by dry inspection method



Plate 16: Seed health of okra by blotter method

4.2.7 Seed health study of brinjal (*Solanum melongena*)

4.2.7.1 Health status of brinjal seeds by inspection of dry seed

A. Physical appearance of seeds

Black spot, swelling and deformed seeds were observed in the seeds collected from New Bangla seeds whereas shrinkage, deformed seeds were observed in the seeds that collected from Islam Seeds and Rafiq Traders (Table 18).

B. Presence of fruiting structures of fungi:

Mycelium was observed over the seeds that collected from New Bangla seeds, Islam seeds and Rafiq Traders (Table 18).

C. Purity analysis

In case of purity analysis of three store seeds 2 gm seeds of brinjal were seen in naked eye. Then determine by hand pure seeds, inert matter and other crops seeds. Total seeds 2 gm, pure seeds 2 gm (100 %), inert matter and other crop seeds 0 gm (0 %) were observed in case of New Bangla seeds and Islam Seeds. But in Rafiq Traders seeds were observed pure seeds 1.99 gm (99.5 %), inert matter 0.02 gm (0.5 %) and other crop seeds 0 (0 %) were observed (Table 18).

4.2.7.2 Prevalence of seed-borne fungi of brinjal identified by blotter method

A. Identified pathogen

Aspergillus flavus, *Aspergillus niger*, *Fusarium* sp., *Alternaria* sp., *Chaetomium* spp., *Rhizopus* sp. and unidentified bacteria were observed.

B. Incidence of seed borne pathogens

In case of fungal infection, the highest incidence was observed that the seeds which collected from Rafiq Traders. However, the highest bacterial infection was recorded in Rafiq Trader. Incidence of *Aspergillus flavus* was varied from 5.5 to 6.75 %.The Highest was observed in Rafiq Traders. In case of *A. niger*, ranged varied from 5.25 to 6.5 %.The Highest was observed in New Bangla seeds store but lowest in Islam Seeds. Incidence of *Fusarium* sp. was varied 4 to 4.25 %. For *Alternaria* sp., ranged varied from 3.5 to 4 %. In case of *Chaetomium* spp., was varied from 6.25 to 4.5 %. The highest incidence was recorded in Rafiq Traders and lowest in Islam Seeds. In case of *Rhizopus* sp. was varied from 3.75 to 5 %, respectively (Table 19).

Table 18. Health status of brinjal seeds by inspection of dry seed method

Seed Sources	Total weight (g)	Pure seeds (g)	Pure seeds (%)	Inert matters (g)	Inert matters (%)	Other seeds (g)	Other seeds (%)	Fruiting structures	Physical abnormalities of seed
New Bangla Seeds	2	2	100	0	0	0	0	Mycelium	Block Spot, Deformed, Swelling
Islam Seeds	2	2	100	0	0	0	0	Mycelium	Deformed, Shrinkage
Rafiq Traders	2	1.99	99.5	0.02	0.5	0	0	Cottony mycelium	Deformed, Reduced size

Table 19. Prevalence of seed-borne fungi of brinjal identified by blotter method

Source Name	% Pathogen incidence							% Seed infection
	<i>Aspergillus flavus</i>	<i>Aspergillus niger</i>	<i>Fusarium</i> sp.	<i>Alternaria</i> sp.	<i>Chaetomium</i> spp.	<i>Rhizopus</i> sp.	Unidentified bacteria	
New Bangla Seeds	5.75 b	6.50 a	4.25 a	3.50 a	5.50 a	4.75 a	2.75 b	33
Islam Seeds	6.75 a	5.25 b	4.01 a	3.50 a	4.50 b	3.75 b	2.75 b	30.6
Rafiq Traders	5.50 b	6.00ab	4.00 a	4.00 a	6.25 a	5.00 a	3.25 a	34
LSD	0.843	0.799	0.715	0.653	0.961	0.753	0.460	
Level of significance	**	**	ns	ns	**	**	*	
CV %	8.78	8.45	10.95	11.13	11.09	10.48	9.90	

ns= non-significant, * = Significant at 5% level of significant as per DMRT, ** = Significant at 1% level of significant as per DMRT



A. Pure seeds

Plate 17: Seed health of brinjal by dry inspection method

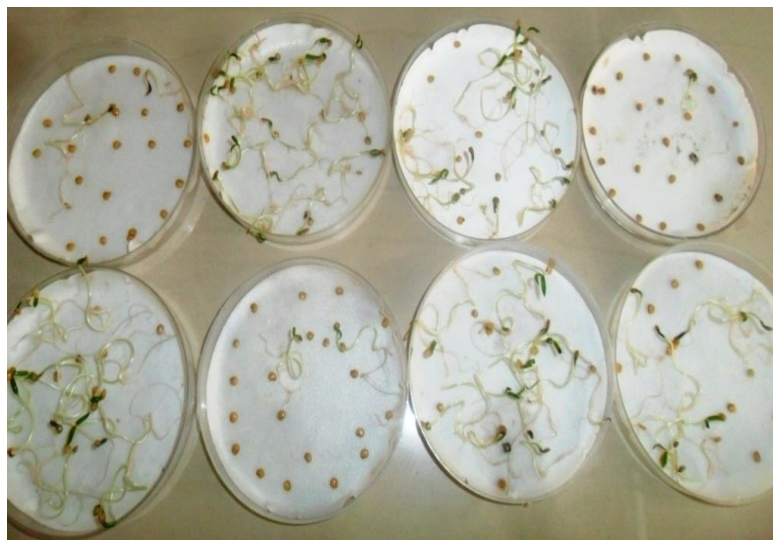


Plate 18: Seed health of brinjal by blotter method

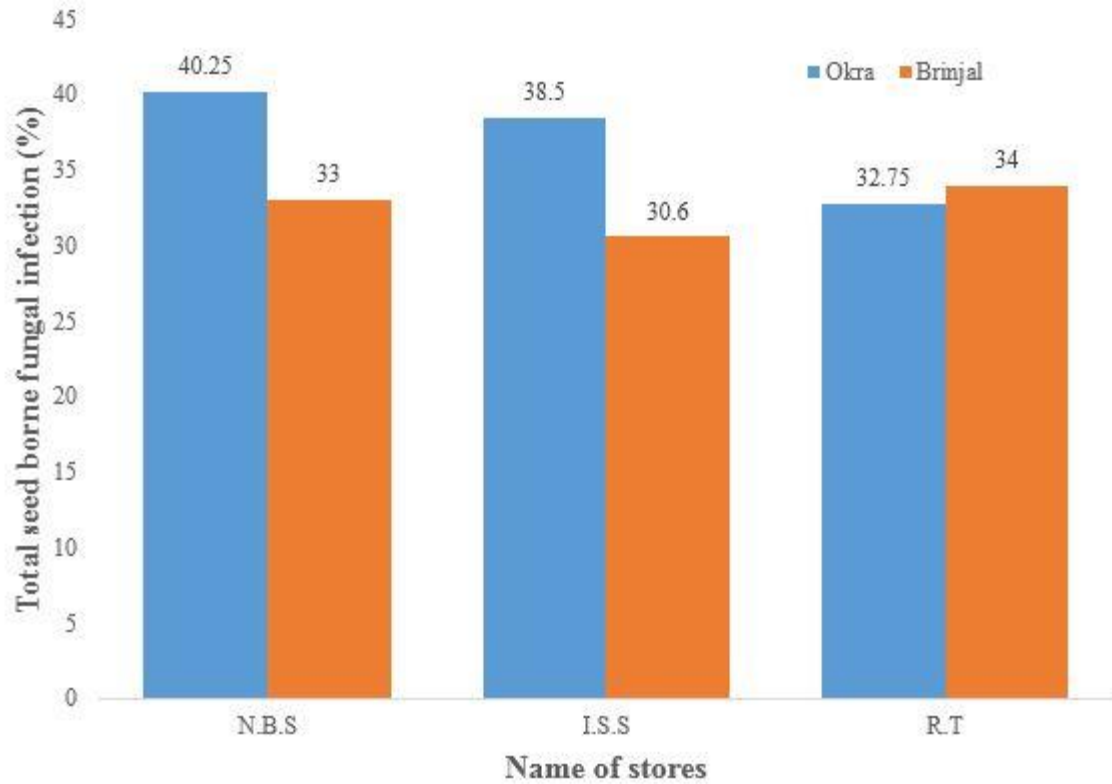


Figure 3: Total seed borne fungal infection (%) of Solanaceous and other vegetables seeds of different stores

N.B.S. - New Bangla seeds, I.S.S- Islam Seeds, R.T. - Rafiq Traders

The present study indicate that okra seeds showed highest value of seed borne fungal infection for New Bangla seeds and Islam Seeds from where seeds were collected. But brinjal seeds showed lowest value of seed borne fungal infection.

4.3 Seed health status of pod vegetable

Fungi detected in pod vegetable like as country bean seeds that were collected from different seed stores like as New Bangla Seed, Islam Seeds and Rafiq Traders of Rangpur sadar presented in table and figures. Prevalence of seed-borne fungal infections varied depending on the seeds of different vegetables.

4.3.1 Seed health study of country bean (*Phaseolus vulgaris*)

4.1.3.1 Health status of country bean seeds by inspection of dry seed

A. Physical appearance of seeds:

Black spot, swelling and deformed seeds were observed in the seeds collected from New Bangla seeds whereas swelling, deformed seeds were observed in the seeds that collected from Islam seeds and Rafiq Traders (Table 20).

B. Presence of fruiting structures of fungi: Mycelium was observed over the seed surface.

C. Purity analysis

In case of purity analysis of different store seeds 80 gm seed of country bean was seen in naked eye. Then determine by hand pure seeds, inert matter and other crops seeds. Result of purity analysis given below table 18. Total seed 80 gm, pure seeds 80 gm (100%), inert matter and other crop seeds was not observed in case of New Bangla seeds. But in Islam Seeds, pure seeds 78.09 gm (97.6 %), inert matter 0.8 gm (1 %) and other crop seeds 1.11 (1.4 %) were observed and 79.02 gm (98.77) pure seeds, 0.35 gm (0.43 %) inert matter and no observation of other crops were found in Rafiq Traders (Table 20).

4.3.1.2. Prevalence of seed-borne fungi of country bean identified by blotter method

The fungal incidence varied significantly among the samples for all pathogens at 5 % level of significance.

A. Identified pathogen

Aspergillus flavus, *Aspergillus niger*, *Fusarium* sp., *Alternaria* sp., *Chaetomium* spp., *Rhizopus* sp. and unidentified bacteria were observed.

B. Incidence of seed borne pathogens

In case of fungal infection, the highest incidence was observed that the seeds which collected from Rafiq Traders. However, the highest bacterial infection was also recorded in Rafiq Traders and New Bangla seeds. Incidence of *Aspergillus flavus* was varied from 7 to 8 %. In case of *A. niger*, ranged varied from 6 to 8.5 %. Highest was observed in Rafiq Traders and Islam Seeds but lowest in New Bangla seeds. Incidence of *Fusarium* sp. was varied 4.25 to 4.75 %. For *Alternaria* sp., ranged varied from 4.75 to 3.75 %. Where the highest incidence was observed in Rafiq Traders but lowest in New Bangla Seeds. In case of *Chaetomium* spp., was varied from 7.25 to 8.25 %. In case of *Rhizopus* sp. was varied from 6.25 to 7.5 % respectively. The highest incidence was recorded in Islam seeds and lowest in New Bangle Seeds (Table 21).

Table 20. Health status of country bean seeds by inspection of dry seed method

Seed Sources	Total weight (g)	Pure seeds (g)	Pure seeds (%)	Inert materials (g)	Inert materials (%)	Other seeds (g)	Other seeds (%)	Phatogenic structures	Physical abnormalities of seed
New Bangla Seeds	80	80	100	0	0	0	0	Mycelium	Block Spot, Deformed,
Islam Seeds	80	78.09	100	0	0	0	0	Mycelium	Deformed, Swelling
Rafiq Traders	80	79.02	98.77	0.35	0.43	0	0	Mycelium	Deformed, Swelling

Table 21. Prevalence of seed-borne fungi of country bean identified by blotter method

Source Name	% Pathogen incidence							% Seed infection
	<i>Aspergillus flavus</i>	<i>Aspergillus niger</i>	<i>Fusarium</i> sp.	<i>Alternaria</i> sp.	<i>Chaetomium</i> spp.	<i>Rhizopus</i> sp.	Unidentified bacteria	
New Bangla Seeds	8.00 a	6.00 b	4.75 a	3.75 b	8.25 a	6.25 b	3.87 a	40.87
Islam Seeds	7.00 a	8.50 a	4.75 a	4.00 b	7.25 a	7.50 a	2.50 b	41.5
Rafiq Traders	7.75 a	8.50 a	4.25 a	4.75 a	7.75 a	7.00 ab	3.75 a	43.75
LSD	1.38	0.753	0.799	0.653	0.961	1.09	0.639	
Level of significance	ns	**	ns	**	ns	**	**	
CV %	11.42	6.15	10.91	9.80	7.75	9.94	11.84	

ns= non-significant,* = Significant at 5% level of significant as per DMRT, ** = Significant at 1% level of significant as per DMRT



A. Pure seeds



B. Other seed



C. Inert matters

Plate 19: Seed health of country bean by dry inspection method

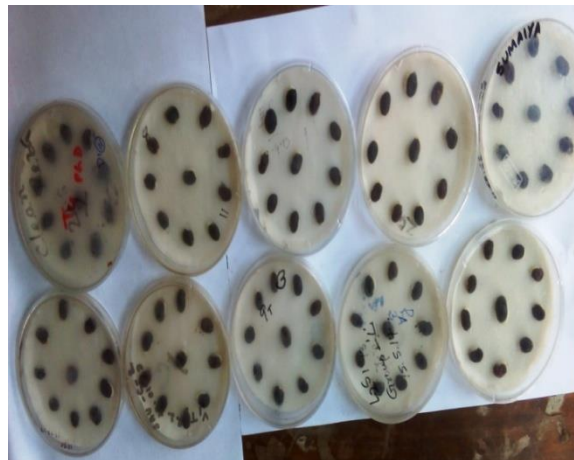


Plate 20: Seed health of country bean by blotter method

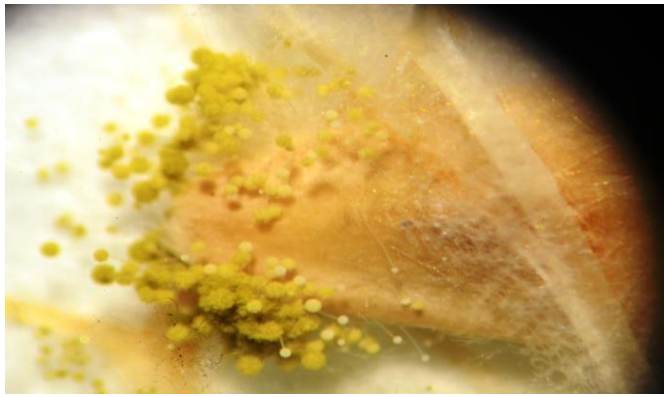


Plate 21: Stereomicroscopic view of *Aspergillus flavus*

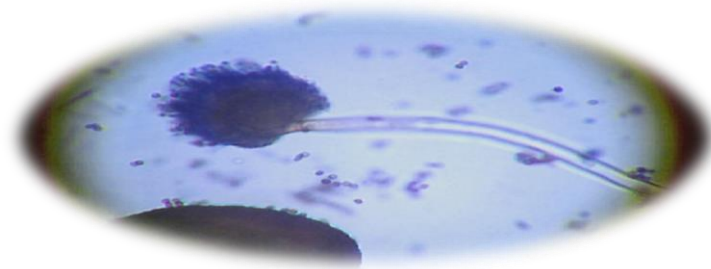


Plate 22: Compound microscopic view of *Aspergillus flavus*

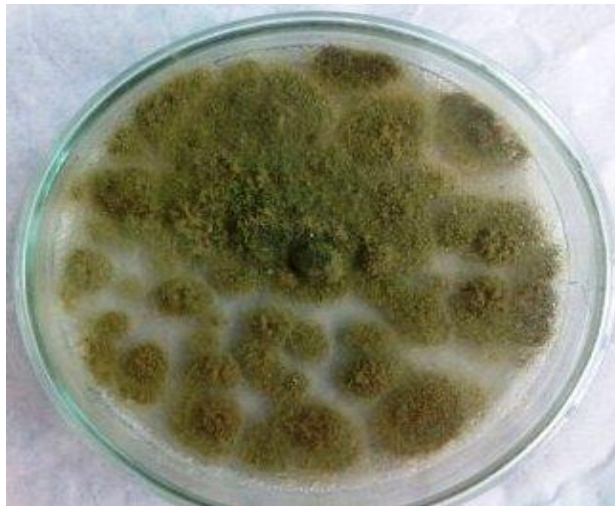


Plate 23: Pure culture of *Aspergillus flavus* on PDA medium



Plate 24: Stereomicroscopic view of *Aspergillus niger*

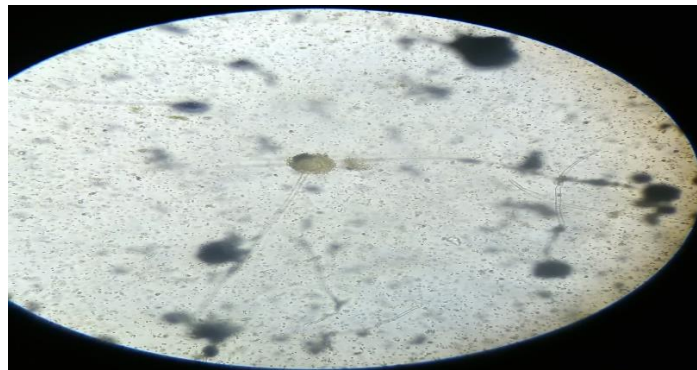


Plate 25: Compound microscopic view of *Aspergillus niger* (40X)



Plate 26: Pure culture of *Aspergillus niger* on PDA medium

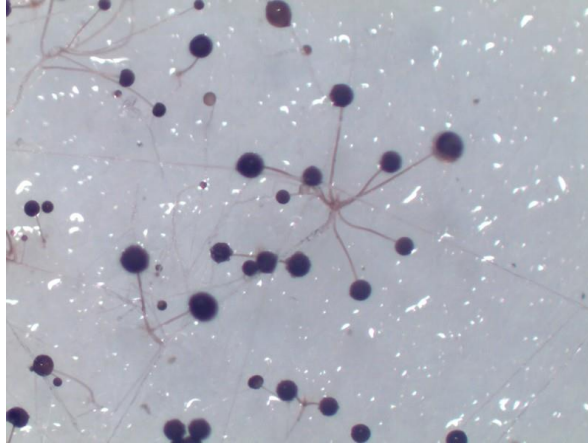


Plate 27: Stereomicroscopic view of *Rizopus* sp. on okra seed



Plate 28: Compound microscopic view of *Rizopus* sp. on okra seed (40 X)



Plate 29: Pure culture of *Rhizopus* sp. on PDA medium

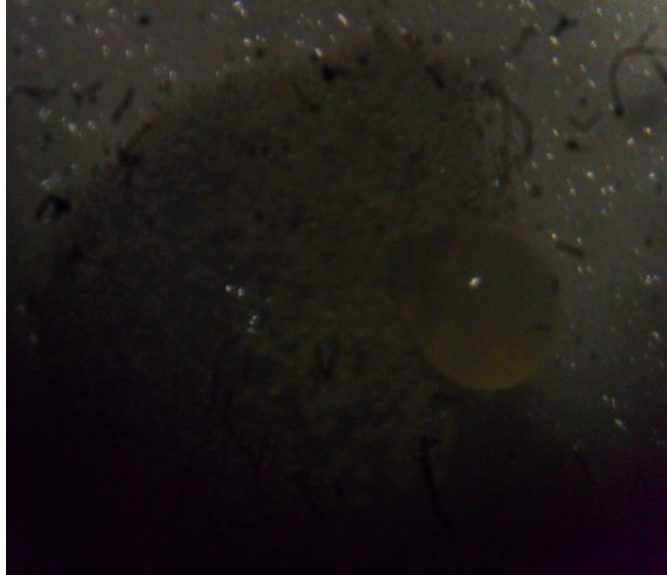


Plate 30: Bacterial ooze on okra seed



Plate 31: Bacterial ooze on sweet gourd seed

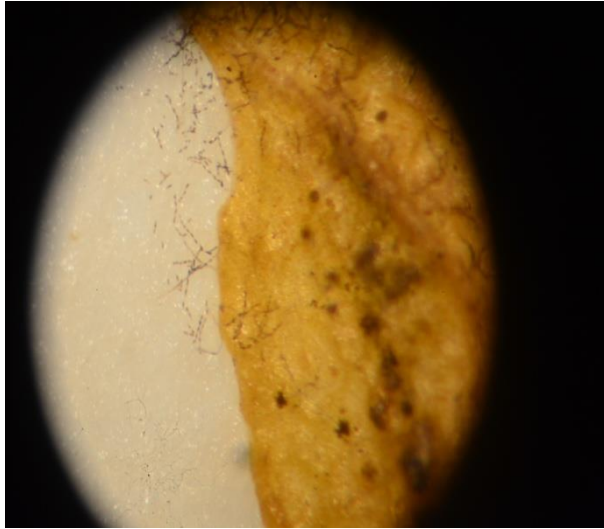


Plate 32: Stereomicroscopic view of *Alternaria* sp. on sweet gourd seed

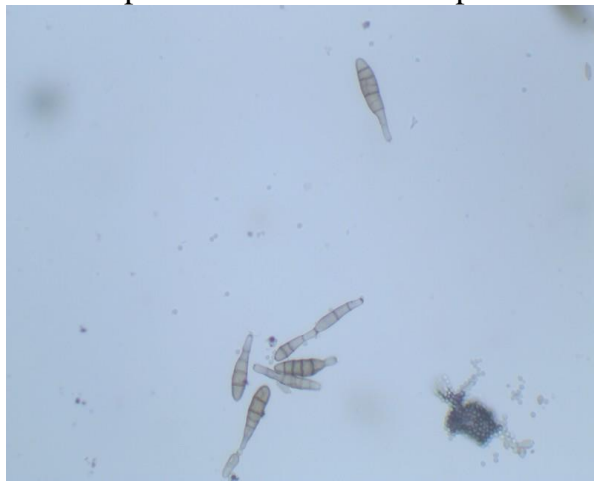


Plate 33: Compound microscopic view of *Alternaria* sp. on sweet gourd seed (40X)

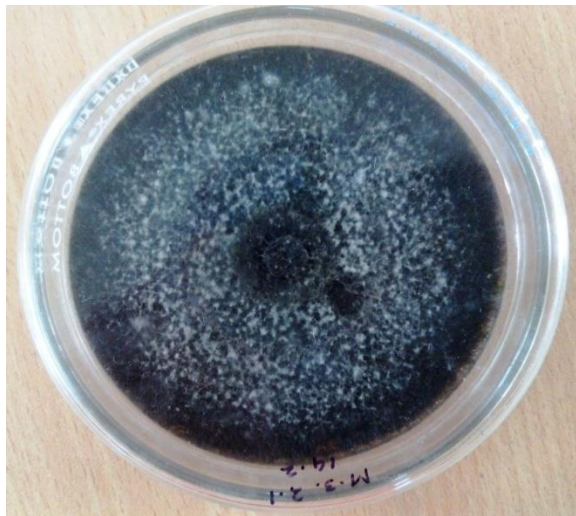


Plate 34: Pure culture of *Alternaria* sp. on PDA medium

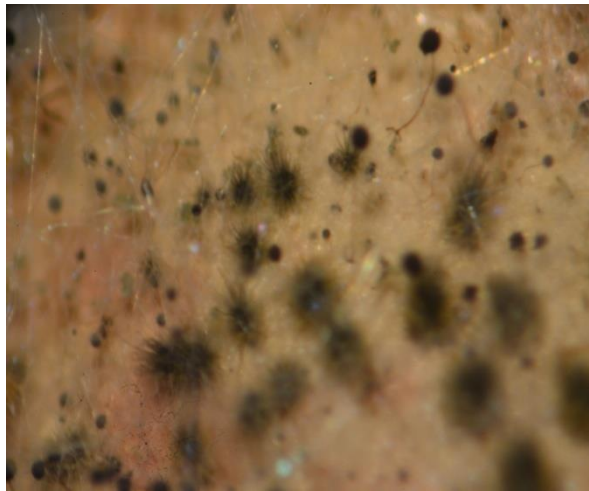


Plate 35: Stereomicroscopic view of *Chaetomium* spp. on sweet gourd seed

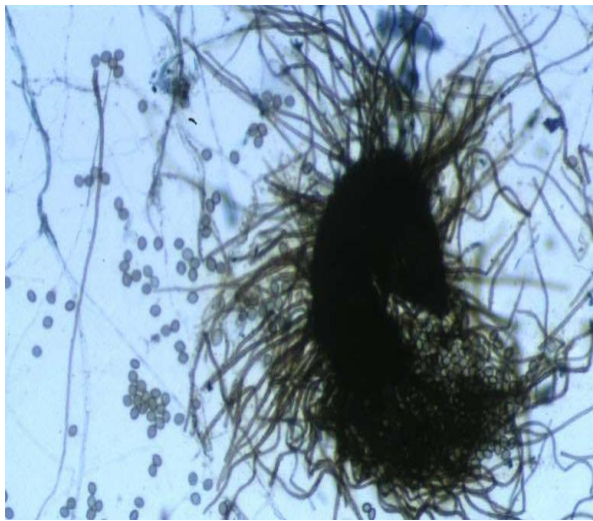


Plate 36: Compound microscopic view of *Chaetomium* spp. on sweet gourd seed (40X)

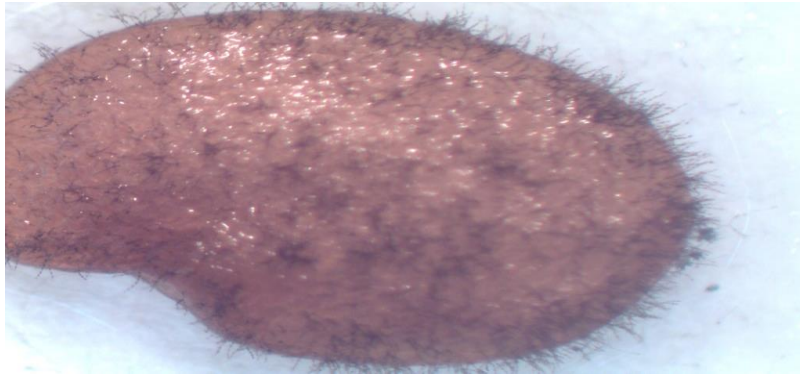


Plate 37: Stereomicroscopic view of *Alternaria* sp. on brinjal seed



Plate 38: Compound microscopic view of *Alternaria* sp. on brinjal seed (40X)

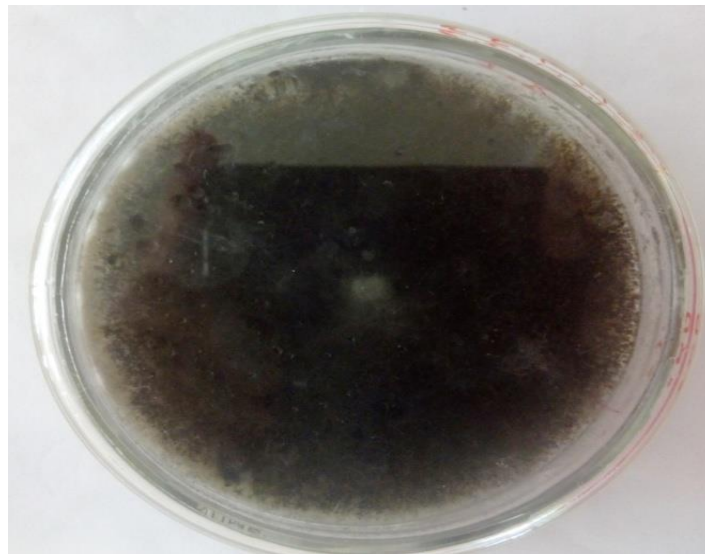


Plate 39: Pure culture of *Alternaria* sp. on PDA medium

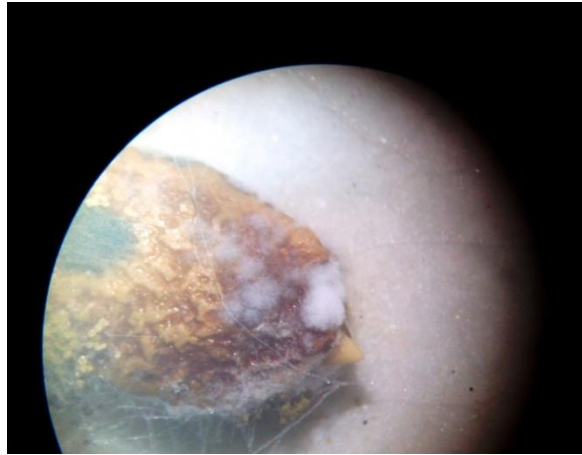


Plate 40: Stereomicroscopic view of *Fusarium* sp. on bitter gourd seed



Plate 41: Compound microscopic view of *Fusarium* sp. on bitter gourd seed (40X)



Plate 42: Pure culture of *Fusarium* sp. on PDA medium



Plate 43: Stereomicroscopic view of *Fusarium* sp. on bottle gourd seed

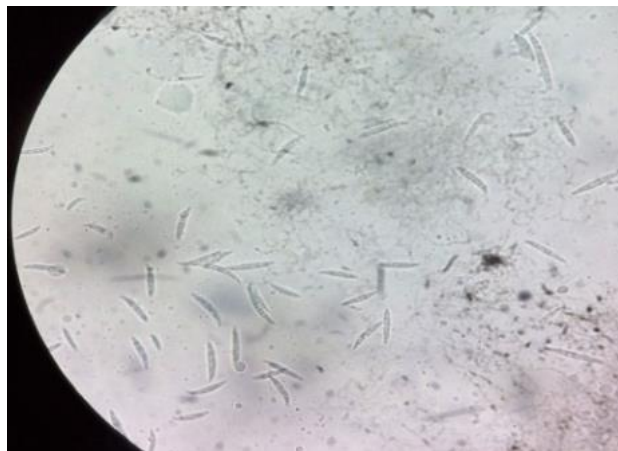


Plate 44: Compound microscopic view of *Fusarium* sp. on bottle gourd seed (40X)

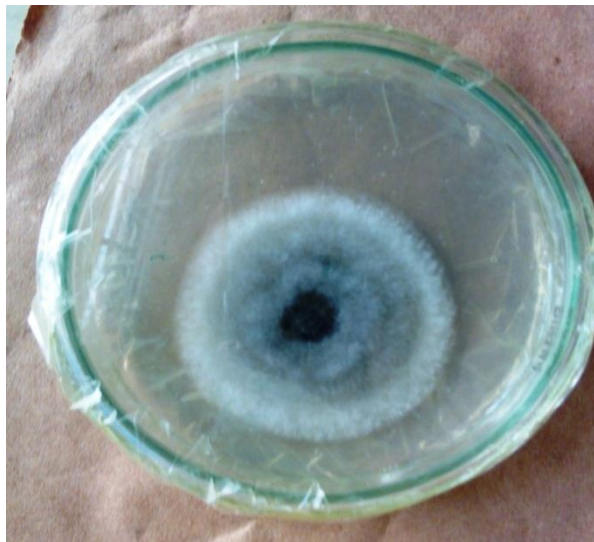


Plate 45: Pure culture of *Fusarium* sp. on PDA medium

4.4 Determination of vigor index by rolled paper towel method

4.4.1 Percentage seed germination

Germination records of different types of vegetable seeds that collected from three stores at Rangpur sadar investigated through rolled paper towel method. Percent germination differs significantly from one to other types vegetables that ranged 94.0 % - 70.0%. The highest germination was found leafy vegetable seeds namely amaranth (94.0 %) that collects from New Bangla seeds which was statistically similar with all other vegetables seeds but differ at numerical value. The lowest germination was recorded in root vegetable seeds like Indian spinach (70 %) that present in table 22.

4.4.2 Shoot and root length of all types of vegetable seeds by rolled paper towel method

All types of vegetable seeds shoot and root length were determine by rolled paper towel method. Shoot length ranged was 18.95 cm to 4.17 cm. The highest shoot length was found in country bean vegetable (18.95 cm) comparatively to other vegetable crops seeds as they are statically similar and dissimilar within the types of vegetable and the store, collection of seeds place. The lowest shoot length was recorded amaranth seeds (4.17 cm) compare with other vegetables which statistically similar between three shops but similar and dissimilar from others.

In case of root length, ranged was 15.40 cm to 3.12 cm. The highest root length was found in country bean vegetable (15.40 cm) comparatively to other vegetable crops seeds as they are statically dissimilar within the store; similar and dissimilar the types of vegetable. The lowest root length was recorded amaranth seeds (3.12 cm) which statistically similar between them but compare with other vegetables statistically similar and dissimilar from others types vegetable.

4.4.3 Percentage of normal and abnormal seedling of all types of vegetable seeds by rolled paper towel method

Normal and abnormal seedling percentage was recorded of different types vegetables that collect from three stores at Rangpur Sadar investigated through rolled paper towel method. Percent normal seedling differs significantly from one to other types vegetables that ranged 84.0 % - 56.0 %. The highest normal seedling was found leafy vegetable seeds namely amaranth (84.0 %) that collects from New Bangla seeds which was statistically similar with two stores seeds and all other vegetables seeds but differ at numerical value. The lowest germination was recorded in root vegetable seeds like Indian spinach (56.0 %) that present in table 22

4.4.4 Non germinated (hard and rotten seed) of different types vegetable seeds be rolled paper towel method

Non germinated seed includes hard seed and rotten seed. Percentage of hard seedling ranged from 18 % to 2 %. The highest percentage of hard seedling was recorded on Indian spinach (18%) seeds that collected from New Bangla seed which was statically different from other two stores seeds and also statically similar and dissimilar from other vegetable crop seeds. The minimum percentage of hard seedling was recorded amaranth (2 %) seeds which were statically identical brinjal, country bean, and bitter gourd seeds (table 22).

The rotten percentage of seed ranged from 4 % to 15 %. The highest percentage of rotten seedling was recorded on bottle gourd (15%) which was collected from Islam Seeds, statically from similar other seeds crop but also different from the two seeds stores and another types crop seed. The lowest percentage of rotten seedling was found in leafy vegetable like amaranth (4%) seed that statically different from two stores and other type crops.

4.4.5 Seedling vigor index

Results of seedling vigor index of ten different vegetable crop seeds by rolled paper towel method were presented in table 22. Vigor index ranged from 677.3 to 2747. Highest vigor index was observed in country bean (2747) seeds that collected from New Bangla seed store which statically similar to other two stores seeds and also the other vegetable varieties. But the lowest seedling vigor index was found in amaranth seeds.

Table 22. Evaluation of seed and seedling quality of leafy and podded vegetables seeds by rolled paper towel method

Crop Name	Treatments	Germination (%)	Shoot Length (cm)	Root Length (cm)	Normal Seedling (%)	Abnormal Seedling (%)	Rot Seed (%)	Hard Seed (%)	Vigor Index (%)
Leafy Veg.	1	94 a	4.17 a	3.12a	84 a	10 b	4 c	2 b	686.1 a
Amaranth	2	91 a	4.62 a	3.20 a	77 a	14 a	7 b	2 b	716.6 a
	3	89 a	4.45 a	3.17 a	81 a	8 c	8 a	5 a	677.3 a
LSD		7.54	0.604	0.195	11.18	1.30	0.653	0.460	74.60
CV%		5.16	8.56	3.87	8.67	7.65	6.45	9.62	6.73
Indian Spinach	1	70 a	6.47 a	4.15 a	56 a	14 ab	11 a	18 a	744.3 a
	2	73 a	6.52 a	3.77 b	60 a	13 b	10 a	17 a	750.8 a
	3	72 a	6.75 a	3.97ab	57 a	15 a	11 a	17 a	770.9 a
LSD		10.39	0.570	0.311	8.49	1.306	1.508	1.68	143.9
CV%		9.07	5.42	4.94	9.21	5.83	8.84	6.08	11.91
Pod Veg.	1	79 a	18.42b	14.20c	70 a	9 b	10 b	11 a	2578 a
Country Bean	2	80 a	18.95a	15.4a	71 a	9 b	12 a	8 b	2747 a
	3	76 a	17.5 c	14.88b	67 a	11 a	11 ab	11 a	2469 a
LSD		6.30	0.281	0.503	8.26	0.923	1.306	0.923	324.2
CV %		5.03	0.96	2.12	7.45	5.97	7.42	5.77	7.80

N.B.: 1 = New Bangla seeds, 2 = Islam Seeds and 3 = Rafiq Traders

Table 23. Evaluation of seed and seedling quality of fruit vegetables seeds by rolled paper towel method (Continued)

Crop Name	Treatments	Germination (%)	Shoot Length (cm)	Root Length (cm)	Normal Seedling (%)	Abnormal Seedling (%)	Rot Seed (%)	Hard Seed (%)	Vigor Index (%)
Bottle gourd	1	81 a	14.13b	12.73b	68a	13b	6c	13a	2176a
	2	76a	16a	14.05a	64a	12b	15a	9b	2285a
	3	76.5a	14.65b	13.15b	64a	15a	11b	10 b	2197a
LSD		9.52	0.718	0.637	9.65	1.306	1.19	1.13	365.9
CV%		7.65	3.01	2.99	9.24	6.12	6.99	6.63	10.31
Cucumber	1	72b	9.05 a	7.15 a	59a	15b	12a	14a	1191a
	2	77.5a	7.52 b	6.42 ab	61a	16b	11a	12b	1075b
	3	79a	6.55 c	6.20 b	61a	18a	12a	11b	1005b
LSD		2.96	0.924	0.772	10.2	1.30	1.36	1.306	76.89
CV %		2.44	7.50	7.32	110	5.0	7.0	6.62	4.41
Bitter gourd	1	72a	18.40a	14.25a	58a	14b	12ab	15a	2351a
	2	74a	17.45b	13.65b	57a	17a	13a	13b	2302a
	3	73a	17.05b	13.48b	60a	13b	11b	16a	2230a
LSD		7.90	0.563	0.350	5.54	1.306	1.36	1.306	318.3
CV %		6.77	2.0	1.58	5.94	5.57	6.80	5.57	8.67
Sweet gourd	1	84a	14.10a	13.20a	72a	12b	10a	5 a	2294a
	2	90a	13.45a	2.98 ab	78a	12b	5b	5a	2375a
	3	85.a	13.52a	12.65b	71a	14a	10a	4b	2223a
LSD		9.947	0.8358	0.4878	10.7	1.306	1.11	0.6537	233.4
CV %		7.20	3.82	2.36	8.63	6.45	8.49	8.75	6.35

Continued									
Snake gourd	1	78a	16.60a	15.23 a	68a	12.5a	8a	12b	2479a
	2	73a	15.82b	14.95 a	61a	12ab	11a	16a	2247a
	3	75a	15.95b	15.18 a	64a	11b	11a	13b	2337a
LSD		10.98	0.635	0.331	12.3	1.19	5.84	1.50	410.8
CV%		9.11	2.46	1.37	12.6	6.30	36.1	6.90	10.91
	1	74a	6.57a	3.32 b	57a	17a	10 a	14a	731b
Okra	2	76a	6.17 a	3.87 a	58a	21.5a	11 a	14a	753b
	3	80a	6.62 a	4.12 a	62a	18 a	11 a	9a	859a
LSD		8.79	0.505	0.510	9.71	8.90	5.43	8.72	98.54
CV%		7.17	4.90	8.48	109	29.56	31.7	44.25	7.88
Brinjal	1	81a	5.12 c	3.65 a	63a	18a	9c	10b	708 b
	2	77a	6.62 a	4.07 a	62a	5b	12a	11b	827 a
	3	75a	5.85 b	3.87 a	61a	14b	10b	15a	724b
LSD		6.07	0.570	0.540	6.31	1.508	0.93	1.30	101.8
CV%		4.89	6.08	8.73	6.36	6.02	5.59	6.80	8.45

N.B.: 1 = New Bangla seeds, 2 = Islam Seeds and 3 = Rafiq Traders



A. Germination of snake gourd by rolled paper towel method



B. Normal seedling of snake gourd



C. Abnormal seedling of snake gourd



D. Dead seeds of snake gourd

Plate 46 (A-D): Determination of seeds and seedling quality of vegetables seeds by rolled paper towel method

4.5 Determination of seed health status by growing on test

4.5.1 Percentage seed germination

Germination records of different type vegetables seeds that collect from three stores at Rangpur sadar investigated through growing on test method. Percent germination differs significantly from one to other types vegetables that ranged (99 – 65) %. The highest germination was found leafy vegetable seeds namely amaranth (99%) that collects from New Bangla seeds which was statistically similar with all other vegetables seeds but differ at numerical value and statistically differ from other two stores seeds of same seeds,. The lowest germination was recorded in root vegetable seeds like okra (65 %) (Table 23).

4.5.2 Shoot and root length of all types of vegetable seeds by growing on test

All types of vegetable seeds shoot and root length were determine by growing on test method. Shoot length ranged was 25.5 cm to 4.10 cm. The highest shoot length was found in country bean vegetable (25.5 cm) comparatively to other vegetable crops seeds as they are statically similar and dissimilar within the types of vegetable and the shops, collection of seeds place. The lowest shoot length was recorded amaranth seeds (4.10 cm) compare with other vegetables which statistically similar between three shops but similar and dissimilar from others.

In case of root length, ranged was 23.25 cm to 3.30 cm. The highest root length was found in country bean vegetable (23.25 cm) comparatively to other vegetable crops seeds as they are statically dissimilar within the store; similar and dissimilar the types of vegetable. The lowest root length was recorded amaranth seeds (3.30 cm) which statistically similar between them but compare with other vegetables statistically similar and dissimilar from others types of vegetable seeds.

4.5.3 Percentage of normal and abnormal seedling of all types of vegetable growing on test method

Normal and abnormal seedling percentage was recorded of different type vegetables seeds that collect from three stores at Rangpur Sadar investigated through rolled paper towel method. Percent normal seedling differs significantly from one to other types vegetables that ranged 90 % - 51.0 %. The highest normal seedling was found leafy vegetable seeds namely amaranth (90.0 %) that collects from New Bangla seeds which was statistically similar with two store seeds and all other vegetables seeds but differ at numerical value. The lowest germination was recorded in root vegetable seeds like okra (51.0 %), collected from third seed store that present in table 23.

In case of abnormal seedling, range was 8 % to 19 %. The highest normal seedling was found root vegetable seeds namely Indian spinach (19 %) that collects from islam seeds store which was statistically similar with third store seeds and all other vegetables seeds but differ at numerical value. The lowest germination was recorded in leafy vegetable seeds like amaranth (8 %) which was statically similar to snake gourd seeds.

4.5.4 Non germinated (hard and rotten seed) of different types vegetable seeds by growing on test method

Non germinated seed includes hard seed and rotten seed. Percentage of rotten seedling ranged from 15% to 1%. The highest percentage of rotten seedling was recorded on Indian spinach, okra and bottle gourd (15%) seeds that collected from Islam seeds store and Rafiq Traders which was statically different from other two store seeds and also statically similar and dissimilar from other vegetable crop

seeds. The minimum percentage of rotten seedling was recorded amaranth (1%) seeds which was statically different from other vegetable seeds (Table 23).

The hard seedling percentage of different vegetables seed ranged from 1 % to 24 %. The highest percentage of hard seedling was recorded on okra (24%) which was collected from Islam Seeds, statically from similar other seeds crop but also different from the two seeds store and another types crop seed. The lowest percentage of rotten seed was found in leafy vegetable like amaranth (1 %) seed that statically different from two stores and other type crops.

4.5.5 Seedling vigor index

Results of seedling vigor index of ten different vegetable crop seeds by rolled paper towel method were presented in table 23. Vigor index ranged from 666.6 to 2747. Highest vigor index was observed in country bean (2747) seeds that collect from New Bangla Seed which statically similar to other two store seeds and also the other vegetable varieties. But the lowest seedling vigor index was found in amaranth seeds (666.6) which was statically different from other vegetable see

Table 24. Evaluation of seed and seedling quality of leafy and podded vegetables by growing on test method

Crop Name	Treatments	Germination (%)	Shoot Length (cm)	Root Length (cm)	Normal Seedling (%)	Abnormal Seedling (%)	Rot Seed (%)	Hard Seed (%)	Vigor Index (%)
Leafy Veg.	1	99 a	4.25 ab	3.60 a	90a	9 b	1c	1b	778a
Amaranth	2	90 b	4.77 a	3.45 a	82b	8 c	2b	5.25a	741.7b
	3	90 b	4.10 b	3.3 a	80b	10 a	6a	5.0 a	666.6 b
LSD		6.993	0.598	0.410	7.23	0.653	0.42	0.601	82.04
CV %		5.16	8.56	3.87	8.67	7.65	6.45	9.62	7.04
Indian Spinach	1	80.5a	6.83 a	2.73 b	63a	17 b	9.0 c	11 a	765.2a
	2	78a	6.35 a	3.1 ab	59b	19 a	13 b	8 c	739.9a
	3	75a	6.25 a	3.4 a	56b	19 a	15 a	9 b	724.9a
LSD		7.21	0.556	0.4 4	3.45	1.30	1.31	0.923	93.84
CV %		5.79	5.37	8.20	3.64	4.45	6.62	6.19	7.89
Pod Veg.	1	79a	18.42b	14.2c	70a	9b	10b	11 a	2578a
Country Bean	2	80a	18.95 a	15.4a	71a	9 b	12a	8b	2747 a
	3	76a	17.5 c	14.8b	67a	11 a	11ab	11a	2469 a
LSD		6.30	0.281	0.503	8.26	0.923	1.30	0.923	324.2
CV %		5.03	0.96	2.12	7.45	5.97	7.42	5.77	7.80

N.B.: 1 = New Bangla seeds, 2 = Islam Seeds and 3 = Rafiq Traders

Table 25. Evaluation of seed and seedling quality of fruit vegetables by growing on test method (Continued)

Crop Name	Treatments	Germination (%)	Shoot Length (cm)	Root Length (cm)	Normal Seedling (%)	Abnormal Seedling (%)	Rot Seed (%)	Hard Seed (%)	Vigor Index (%)
Fruit Veg.	1	70.5a	7.10 b	6.17 ab	54b	15a	9c	18 b	942.7 a
Okra	2	74a	6.60c	5.77b	59a	15a	15 a	11c	942.4 a
	3	65b	7.92 a	6.40 a	51b	13.7a	13 b	24a	930.4 a
LSD		5.41	0.435	0.463	3.016	1.38	1.13	1.30	87.26
CV %		7.17	4.90	8.48	10.29	9.56	11.87	14.5	7.88
Brinjal	1	81a	5.12 c	3.65a	63a	18a	9c	10b	799.9 a
	2	77a	6.62 a	4.07a	62a	15b	12a	11b	792.9 a
	3	75a	5.85 b	3.87a	61a	14b	10b	15a	775.6 a
LSD		6.07	0.570	0.540	6.30	1.50	0.923	1.30	102.1
CV %		4.89	6.08	8.73	6.36	6.02	5.59	6.80	8.08
Bottle gourd	1	81a	14.13b	12.73 b	68a	13b	6c	13a	2176 a
	2	76a	16.00 a	14.05 a	64a	12b	15a	9b	2285 a
	3	76.5a	14.65 b	13.15b	64a	15a	11b	10b	2197 a
LSD		9.52	0.718	0.637	9.65	1.30	1.19	1.13	365.9
CV %		7.65	3.01	2.99	9.24	6.12	6.99	6.63	10.31
Cucumber	1	72b	9.05a	7.15a	59a	15b	12a	14a	1191 a
	2	77.5a	7.52b	6.42ab	61a	16b	11 a	12b	1075 b
	3	79a	6.55 c	6.20b	61a	18a	12 a	11b	1005 b
LSD		2.96	0.924	0.772	10.72	1.30	1.30	1.30	76.89
CV %		2.44	7.50	7.32	11.10	5.0	7	6.62	4.41

Continued									
Bitter gourd	1	72a	18.40 a	14.25 a	58a	14b	12ab	15 a	2491a
	2	74a	17.45 b	13.65 b	57a	17a	13a	13b	2522a
	3	73a	17.05 b	13.48 b	60a	13b	11b	16a	2147 b
LSD		7.90	0.563	0.350	5.54	1.30	1.30	1.30	290
CV %		6.77	2.00	1.58	5.94	5.57	6.80	5.57	7.60
Sweet gourd	1	84a	14.10 a	13.20 a	72a	12b	10a	5a	2388a
	2	90a	13.45 a	12.98ab	78a	12b	5b	5a	2177 b
	3	85a	13.52 a	12.65 b	71a	14a	10a	4 b	2403a
LSD		9.94	0.835	0.487	10.17	1.30	1.13	0.65	175.3
CV %		7.20	3.82	2.36	8.63	6.45	8.49	8.75	4.72
Snake gourd	1	89a	15.98b	14.02 a	81a	8c	3 c	7c	2670 a
	2	74b	16.88 a	13.9a	59.7b	13b	12b	17a	2275 b
	3	75b	16.38ab	14a	59b	16a	12a	13a	2279 b
LSD		7.54	0.684	0.589	4.96	1.13	1.13	1.13	217.4
CV %		5.94	2.60	2.64	4.66	5.73	9.22	5.73	5.64

N.B.: 1 = New Bangla seeds, 2 = Islam Seeds and 3 = Rafiq Traders



A. Germination test of sweet gourd seeds by growing on test method



B. Normal seedling



C. Abnormal seedling



D. Dead seeds

Plate 47 (A-D): Determination of seeds health status by growing on test method

CHAPTER V

DISCUSSION

Two leafy vegetables (amaranth and Indian spinach), seven fruit vegetables (bottle gourd, sweet gourd, cucumber, bitter gourd, snake gourd, okra and brinjal) and one pod vegetable (country bean) were evaluated to determine the health status of loose vegetables seeds that collected from Rangpur Sadar. A considerable amount of seed borne pathogenic fungi and unidentified bacteria were observed by inspection of dry method, blotter method, rolled paper towel and growing on test method.

Altogether five genera of seed borne fungal pathogens viz. *Alternaria*, *Aspergillus*, *Chaetomium*, *Fusarium* and *Rhizopus* were recorded in ten vegetable seeds viz. amaranth, Indian spinach, okra, cucumber, bottle gourd, bitter gourd, sweet gourd, snake gourd, country bean and brinjal collected from three shops of Rangpur Sadar.

In Dry inspection method, white and cottony mycelium, acervuli were identified. In blotter method, five seed borne pathogenic genera were identified namely *Aspergillus*, *Fusarium*, *Alternaria*, *Chaetomium*, *Rhizopus* and unidentified bacterium.

In dry inspection of amaranth, highest pure seeds (100 %) were found all shops from seeds collected. Six pathogens were observed on seeds collected from three shops. And the pathogens that were come a crossed i.e. *Aspergillus flavus*, *Aspergillus niger*, *Fusarium* sp., *Alternaria* sp., *Chaetomium* spp., *Rhizopus* sp. The highest total seed borne pathogen infections (34.24%) were New Bangla seeds. To the view of my knowledge the overall health status of loose seeds of amaranth was not up to the mark. This is because, saprophytic pathogenic genera were observed with the seed that may deteriorate the health status and quality of seed and seedling.

The present investigation revealed that altogether six different fungi viz. *Alternaria* sp., *Aspergillus flavus*, *Aspergillus niger*, *Chaetomium* spp., *Fusarium* sp. and *Rhizopus* sp. were found to be associated with Amaranth seeds. Bebum, 2012 reported by a considerable number of seed borne fungal pathogens belonging to the genera *Aspergillus*, *Curvularia*, *Fusarium* and *Penicillium* had been detected in Amaranth seeds. The present findings clearly showed that *Alternaria* sp., *Aspergillus flavus*, *Aspergillus niger*, *Chaetomium* spp., *Fusarium* sp. and *Rhizopus* sp. were associated with the tested seed samples of amaranth significantly reduced percent germination. Similar result was reported by earlier worker (Islam, 2005)

In dry inspection of Indian spinach, the highest pure seed were found (98.33 %) in Rafiq Traders and lowest pure seeds were found New Bangla seeds (95%). Whereas, shrinkage and deformed seeds were observed on seeds collected from three seeds sources. In case of Indian spinach seeds five different fungi viz. *Alternaria*, *Aspergillus*, *Chaetomium*, *Fusarium* and *Rhizopus* were found to be associated. The highest (34 %) total seed borne pathogen infestation was Islam seeds. From my view, the overall health status of loose seeds of Indian spinach was not up to the mark. Because of, few saprophytic pathogens were found with the seeds that may deteriorate the health status and quality of seed and seedling.

A considerable number of seed borne fungal pathogens belonging to the genera *Alternaria*, *Curvularia*, *Fusarium* and *Penicillium* had been detected in Spinach seeds by many researchers (Begum, 2012; Khanom, 2011). The present findings clearly showed that *Alternaria*, *Aspergillus*, *Chaetomium* and *Fusarium* were associated with the tested seed samples significantly reduced percent germination. Similar result was reported by earlier worker Islam (2005).

In dry inspection of bottle gourd, the highest pure seeds were found (79.05 %) in Islam Seeds store and lowest pure seed were found Rafiq Traders (78.04 %).

Whereas, shrinkage, reduced in size and deformed seeds were observed on seeds collected from three seed sources. In case of blotter method five different pathogens were observed namely *Alternaria* sp., *Aspergillus flavus*, *Aspergillus niger*, *Chaetomium* spp., *Fusarium* sp. and *Rhizopus* sp. The highest (35.75 %) total seed borne pathogen infestation was New Bangla seeds and lowest infestation were recorded (30) the seeds source namely Rafiq Traders. From my view, the overall health status of loose seeds of bottle gourd was not up to the mark. Because of, few saprophytic pathogens were found with the seeds that may deteriorate the health status and quality of seed and seedling.

However, the seed borne fungi of bottle gourd, a total of 22 genera and 45 species of fungi were isolated where 35 have hitherto been recorded from seeds of bottle gourd in Pakistan reported by Sultana and Ghaffar (2009).

In dry inspection of sweet gourd, highest pure seeds were found (98.75 %) in Rafiq Traders and lowest pure seeds were found New Bangla seeds (96.25 %). Whereas, shrinkage and deformed seeds were observed on seeds collected from three seeds sources. In case of sweet gourd seeds five different fungi viz. *Alternaria*, *Aspergillus*, *Chaetomium*, *Fusarium* and *Rhizopus* were found to be associated. The highest (47.5 %) total seed borne pathogen infestation was New Bangla seeds. Whereas, lowest infestation (45.25 %) were observed Islam seeds store. From my view, the overall health status of loose seeds of Indian spinach was not up to the mark. Because of, few saprophytic pathogens were found with the seeds that may deteriorate the health status and quality of seed and seedling.

In case dry inspection of bitter gourd, the highest pure seeds were found (100 %) in Rafiq Traders and the lowest pure seeds were found Islam seeds (98.77%). Whereas, shrinkage and deformed seeds were observed on seeds collected from three seed sources. The present investigation revealed that altogether six different fungi viz. *Alternaria* sp., *Aspergillus flavus*, *Aspergillus*

niger, *Chaetomium* spp., *Fusarium* sp. and *Rhizopus* sp. were found to be associated with bitter gourd seeds. The highest (43.25 %) total seed borne pathogen infestation was Rafiq Traders. Where, lowest infestation (35.5 %) was observed Islam seeds. From my view, the overall health status of loose seeds of Indian spinach was not up to the mark. Because of, few saprophytic pathogens were found with the seeds that may deteriorate the health status and quality of seed and seedling.

In dry Inspection of cucumber, highest pure seed were found (99.37%) in Islam seeds and lowest pure seeds were found New Bangla seeds (98.75 %). Whereas, shrinkage and deformed seeds were observed on seeds collected from three seed sources. In case of cucumber seeds five different fungi viz. *Alternaria*, *Aspergillus*, *Chaetomium*, *Fusarium* and *Rhizopus* were found to be associated. The highest (36.75 %) total seed borne pathogen infestation was Rafiq Traders. Where, lowest infestation (33.75 %) was observed New Bangla seeds and Islam Seeds. From my view, the overall health status of loose seeds of Indian spinach was not up to the mark. Because of, few saprophytic pathogens were found with the seeds that may deteriorate the health status and quality of seed and seedling.

In case of cucumber seeds four different fungi viz. *Alternaria* sp., *Curvularia* sp., *Fusarium* sp. and *Penicillium* sp. were recorded. A considerable number of seed borne fungal pathogens belonging to the genera *Alternaria*, *Curvularia*, *Fusarium* and *Penicillium* had been detected in cucumber seeds by many researchers (Chowdhury *et al.* 2005; Alimova *et al.* 2002; Nasreen and Sultana, 2000; Puspa *et al.* 1999). The present findings clearly showed that *Alternaria* sp., *Curvularia* sp., *Fusarium* sp. and *Penicillium* sp. were associated with the tested seed samples significantly reduced percent germination. Similar result was reported by some earlier workers (Begum and Momin, 2000; Alimova *et al.* 2002; Islam, 2005).

In dry inspection of snake gourd, the highest pure seeds were found (100 %) in Rafiq Traders and the lowest pure seeds were found New Bangla seeds (97.75 %). Where, shrinkage, swelling and deformed seeds were observed on seeds collected from three seed sources. In case of snake gourd seeds five different fungi viz. *Alternaria*, *Aspergillus*, *Chaetomium*, *Fusarium* and *Rhizopus* were found to be associated. The highest (34.25%) total seed borne pathogen infestation was Rafiq Traders. Where, lowest infestation (39.25%) was observed New Bangla seeds. From my view, the overall health status of loose seeds of Indian spinach was not up to the mark. Because of, few saprophytic pathogens were found with the seeds that may deteriorate the health status and quality of seed and seedling.

In dry inspection of okra, highest pure seeds were found (94.95 %) in Islam seeds store and lowest pure seeds were found Rafiq Traders (90%). Where, shrinkage, block spot and deformed seeds were observed on seeds collected from three seed sources. In case of okra seeds five different fungi viz. *Alternaria*, *Aspergillus*, *Chaetomium*, *Fusarium* and *Rhizopus* were found to be associated. The highest (40.25%) total seed borne pathogen infestation was Rafiq traders. Where, the lowest infestation (32.75%) was observed New Bangla seed. From my view, the overall health status of loose seeds of Indian spinach was not up to the mark. Because of, few saprophytic pathogens were found with the seeds that may deteriorate the health status and quality of seed and seedling.

The present investigation revealed that altogether six different fungi viz. *Aspergillus flavus*, *Aspergillus niger*, *Fusarium* sp., *Penicillium* sp., *Colletotrichum dematium* and *Macrophomina phaseolina* were found to be associated with okra seeds. A considerable number of seed borne fungal pathogens belonging to the genera *Aspergillus*, *Fusarium*, *penicillium*, *Colletotrichum* and *Macrophomina* had been detected in okra seeds by many

researchers (Sultana, 2009; Akter, 2008; Alam, 2001; Jamadar *et al.* 2001; Fakir, 2000). The present findings clearly showed that *Aspergillus flavus*, *Aspergillus niger*, *Fusarium* sp., *Penicillium* sp., *Colletotrichum dematium* and *Macrophomina phaseolina* were associated with the tested seed samples significantly reduced percent germination. Similar result was reported by some earlier workers (Jamadar *et al.* 2001; Gupta *et al.* 1989; Adisa and Aborisade, 1987; Neergaard, 1979).

In dry inspection of brinjal, highest pure seeds were found (100 %) in New Bangla seeds and Islam seeds. The lowest pure seed were found Rafiq Traders (99.5 %). Where, swelling, shrinkage and deformed seeds were observed on seeds collected from three seed sources. In case of brinjal seeds five different fungi viz. *Alternaria*, *Aspergillus*, *Chaetomium*, *Fusarium* and *Rhizopus* were found to be associated. The highest (33%) total seed borne pathogen infestation was Rafiq Traders. Where, lowest infestation (30.51%) was observed Islam Seeds. From my view, the overall health status of loose seeds of Indian spinach was not up to the mark. Because of, few saprophytic pathogens were found with the seeds that may deteriorate the health status and quality of seed and seedling.

The present investigation revealed that altogether five different fungi viz. *Alternaria*, *Aspergillus*, *Chaetomium*, *Fusarium* and *Rhizopus* were found to be associated with Brinjal seeds. *Aspergillus flavus*, *Aspergillus niger*, *Curvularia* sp., and *Fusarium* sp. had been detected in brinjal seeds by many researchers (Habib *et al.* 2007; Sarker *et al.* 2006; Basak *et al.* 1989). The present findings clearly showed that *Aspergillus flavus*, *Aspergillus niger*, and *Fusarium* sp. were associated with the tested seed samples significantly reduced percent germination. Similar result was reported by earlier worker (Habib *et al.* 2007).

In dry inspection of country bean highest pure seeds were found (100 %) in New Bangla seeds and Islam seeds. The lowest pure seeds were found Rafiq

Traders (98.77%). Where, swelling, shrinkage and deformed seeds were observed on seeds collected from three seed sources. In case of country bean seeds five different fungi viz. *Alternaria*, *Aspergillus*, *Chaetomium*, *Fusarium* and *Rhizopus* were found to be associated. The highest (43.75 %) total seed borne pathogen infestation was Rafiq Traders. Where, the lowest infestation (40.87 %) was observed New Bangle seeds. From my view, the overall health status of loose seeds of Indian spinach was not up to the mark. Because of, few saprophytic pathogens were found with the seeds that may deteriorate the health status and quality of seed and seedling.

Domijan *et al.* (2003) reported that seed borne on bean (*Phaseolus vulgaris*) and most common fungi were isolated viz. *Alternaria* sp. (75 %), *Aspergillus* (73 %), *Rhizopus* (73 %), *Cladosporium* sp. (98 %), *Penicillium* sp. (69%), *Fusarium* sp. (38 %).

The fungi isolated from seeds of cauliflower where *Alternaria* (14.8%) was the most common genus, other common genera prevalence were *Helminthosporium* (12.8%), *Aspergillus falvus* (8.2%), *Rhizopus* (5.6%), *Curvularia* (4.8%), *A. niger* (4.6%), *Cercospora* (3.8%), *Fusarium* (3.4%) and *Chaetomium* (3.2%) reported by Iqbal *et al.* 2012.

Germination of vegetable seeds recorded by blotter method varied significantly. Germination was highest in amaranth (94 %) and lowest in Indian spinach (72 %). There was a highly positive relationship between germination failure and prevalence of seed borne fungal infection. In the present study, germination categories of seeds of 10 vegetables were recorded. Normal seedlings, abnormal seedlings and non-germinated seeds were the categories. Normal seedlings ranged from 56 to 84 % in 10 vegetables. Normal seedlings were found high in amaranth (84 %), whereas abnormal seedlings ranged from (5-21.5%). Non germinated seeds like (hard and rotten) were found to be ranged from (2-18%

and 4- 15%) in seeds of 10 vegetables. The notorious fungi, *Fusarium* sp. cause seedlings infection or foot and root rot disease. *Fusarium* sp. is a well-known seed borne pathogen able of causing germination failure/seed rot, damping-off, root rot and wilts etc. in many crops (Mathur *et al.*1975; Richardson, 1990). The cause of non-germination was pathogenic infection and sometimes it was physiological cause. Among the 10 vegetable seeds the vigour index was ranged from 677.3 to 2747.

The above study exposed that very good connection between seed borne infections and germination failure of vegetable seeds existed. Therefore, further studies with more representative seed samples from different agro-ecological zones of the country should be undertaken in order to reveal the exact picture regarding the prevalence of seed borne fungi and the role they do play on seed germination.

Here also another test was done for the germination of seeds namely growing on test method. Germination was highest in amaranth (99 %) and lowest in okra (65 %). There was a highly positive relationship between germination failure and incidence of seed borne fungal infection. In the present study, germination categories of seeds of 10 vegetables were recorded. Normal seedlings, abnormal seedlings and non-germinated seeds were the categories. Normal seedlings ranged from 51 to 90 % in 10 vegetables. Normal seedlings were found high in amaranth (90 %), whereas abnormal seedlings ranged from (8-19%). Non germinated seeds like (hard and rotten) were found to be ranged from (1-24 % and 1- 15 %) in seeds of 10 vegetables.

Naznin and Hossain (2004) studied the effect of BAU Bio-fungicide on germination of some vegetables and showed that in sweet gourd, BAU-Bio-fungicide increased germination up to 5.22% in pot and 46.25% in field over the untreated control, while in cowpea, cucumber and okra the germination

increased up to 46.245, 7.92% and 50.80%, respectively in pot experiment and 102.95, 28.8% and 47.8% in field trial. Sultana *et al.* (2009) used BAU Bio-fungicide for seed treatment to control *Bipolaris sorokiniana*, leaf spot of wheat. Kanchan. And observed (94%) germination which 12.7% higher over control. They reported that *Trichoderma* based preparation increased germination of seeds.

CHAPTER VI

SUMMARY AND CONCLUSION

Considering the importance of vegetable in Bangladesh perspective, seed health status examined of bulk untreated loose seed. For this experiment seed collected from three seeds shops from Rangpur Sadar. And determination of seed borne infection and germination, seedling vigor of ten vegetable seeds viz. amaranth, Indian spinach, okra, cucumber, sweet gourd, bottle gourd, bitter gourd, snake gourd, country bean and brinjal were tested in the Seed Pathology Laboratory, Department of Plant Pathology, Sher-e-Bangla Agricultural University, Dhaka-1207.

Four seed health testing methods viz. inspection of dry seed, blotter method, and roller paper tower method and growing on test were followed. By inspection of dry seed method farmers normally get primary idea about seed value of loose vegetable by observing visually. In blotter method most of the seed borne pathogen can be easily identified. That's why this method is the most acceptable method of seed health testing and using worldwide. On the other hand, another two methods, farmer can get idea about seeds germination and vigorous capability.

Occurrence of seed borne pathogen of ten vegetables recorded in both Inspection of dry seed and blotter method varied significantly depending on seed categories and seed sources. Among three seeds sources, each source mention that their purity percentage of seeds and total seed borne fungal infections. Through inspection of dry seed, present research work mention that in most case seeds collected from New Bangla Seeds showed lowest purity percentage of seed except amaranth

country bean and brinjal. Where highest purity percentage of seed were observed seed collected from Rafiq Traders. New Bangla seeds gave an average result for all loose vegetable seeds. On inspection of dry seed, the highest pure seed percentage was found in snake gourd, brinjal and country bean (100 %).

In blotter method, out of the tested seeds of ten vegetable crops, six fungi were detected. The fungi recorded were *Aspergillus flavus*, *Aspergillus niger*, *Fusarium* sp., *Alternaria* sp., *Chaetomium* spp. and *Rhizopus* sp. Out of these fungi, six fungi were detected on amaranth, Indian spinach, bottle gourd, sweet gourd, snake gourd, okra and brinjal seeds; in order of prevalence of average pathogen infestation were observed in New Bangla seeds and lowest were Islam seeds. The infestation of seed borne fungi was highest in sweet gourd. Based on the present research study, it was said that the pure seed percentage and infestation of seed borne fungi were varied significantly depending on the seeds of vegetables and the seed source.

In rolled paper towel method and seedling symptom test, the maximum number of seed germination (94 % and 99 %) in amaranth and highest seedling vigor (2747) in country bean seed were recorded that collected from New Bangla seeds. The maximum numbers of non-germinated seeds like rotten and hard seeds were observed in okra (15 %) and bottle gourd (15 %) seed that collected from Islam Seeds store and New Bangla seeds store. In case of hard seeds, highest percentage was observed in okra (24 %) and Indian spinach (18%) seeds in New Bangla seeds from where seeds collected.

High quality seed is not only important for increasing crop production but also proper establishment of sound seed industry in the country. Seed is a common carrier of plant pathogens. Pathogen free seed is the important input material in

agriculture. The present experiment showed that a lot of seed borne pathogens were associated with loose vegetable seeds. Seed borne fungi appeared may be due to improper management of vegetable seeds in storage. Considering the over-all findings it was exposed that the seed health status of loose vegetable seeds is not a satisfactory level. Farmers are therefore advised to collect the seeds from reliable source and check their seed health status before sowing in the main field.

CHAPTER VII

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CHAPTER VIII

APPENDICES

Appendix I: Analysis of variance of data on prevalence of seed borne fungi of amaranth identified by blotter method

Amaranth	df	Seed borne infection (%)						
		<i>Aspergillus flavus</i>	<i>Aspergillus niger</i>	<i>Fusarium</i> sp.	<i>Alternaria</i> sp.	<i>Chaetomium</i> spp.	<i>Rhizopus</i> sp.	Unidentified bacteria
Between	2	1.75*	1.75**	0.750*	4.33**	3.00**	1.33**	0.333*
Within	9	0.389 ^{ns}	0.278 ^{ns}	0.139 ^{ns}	0.125 ^{ns}	0.278 ^{ns}	0.167 ^{ns}	0.083 ^{ns}
Total	11							

Appendix II: Analysis of variance of data on prevalence of seed borne fungi of indian spinach identified by blotter method

Indian spinach	df	Seed borne infection (%)						
		<i>Aspergillus flavus</i>	<i>Aspergillus niger</i>	<i>Fusarium</i> sp.	<i>Alternaria</i> sp.	<i>Chaetomium</i> spp.	<i>Rhizopus</i> sp.	Unidentified bacteria
Between	2	3.00*	5.25**	0.396 ^{ns}	0.250 ^{ns}	1.58*	0.750 ^{ns}	1.58**
Within	9	0.611 ^{ns}	0.444 ^{ns}	0.215 ^{ns}	0.111 ^{ns}	0.306 ^{ns}	0.50 ^{ns}	0.111
Total	11							

Appendix III: Analysis of variance of data on prevalence of seed borne fungi of bottle gourd identified by blotter method

Bottle gourd	df	Seed borne infection (%)						
		<i>Aspergillus flavus</i>	<i>Aspergillus niger</i>	<i>Fusarium</i> sp.	<i>Alternaria</i> sp.	<i>Chaetomium</i> spp.	<i>Rhizopus</i> sp.	Unidentified bacteria
Between	2	4.33**	1.58*	1.57**	2.25**	1.54**	1.53**	20.33**
Within	9	0.556 ^{ns}	0.389 ^{ns}	0.111 ^{ns}	0.167 ^{ns}	0.222 ^{ns}	0.250 ^{ns}	0.167 ^{ns}
Total	11							

Appendix IV: Analysis of variance of data on prevalence of seed borne fungi of brinjal identified by blotter method

Brinjal	df	Seed borne infection (%)						
		<i>Aspergillus flavus</i>	<i>Aspergillus niger</i>	<i>Fusarium</i> sp.	<i>Alternaria</i> sp.	<i>Chaetomium</i> spp.	<i>Rhizopus</i> sp.	Unidentified bacteria
Between	2	1.75**	1.53**	0.079 ^{ns}	0.333 ^{ns}	3.08**	1.75**	0.333*
Within	9	0.278 ^{ns}	0.250 ^{ns}	0.200 ^{ns}	0.167 ^{ns}	0.361 ^{ns}	0.222 ^{ns}	0.083 ^{ns}
Total	11							

Appendix V: Analysis of variance of data on prevalence of seed borne fungi of cucumber identified by blotter method

Cucumber	df	Seed borne infection (%)						
		<i>Aspergillus flavus</i>	<i>Aspergillus niger</i>	<i>Fusarium</i> sp.	<i>Alternaria</i> sp.	<i>Chaetomium</i> spp.	<i>Rhizopus</i> sp.	Unidentified bacteria
Between	2	1.04*	0.25 ^{ns}	0.583*	0.573*	0.25 ^{ns}	4.33**	1.53**
Within	9	0.195 ^{ns}	0.139 ^{ns}	0.167 ^{ns}	0.163 ^{ns}	0.278 ^{ns}	0.306 ^{ns}	0.222 ^{ns}
Total	11							

Appendix VI: Analysis of variance of data on prevalence of seed borne fungi of bitter gourd identified by blotter method

Bitter gourd	df	Seed borne infection (%)						
		<i>Aspergillus flavus</i>	<i>Aspergillus niger</i>	<i>Fusarium</i> sp.	<i>Alternaria</i> sp.	<i>Chaetomium</i> spp.	<i>Rhizopus</i> sp.	Unidentified bacteria
Between	2	0.583 ^{ns}	1.75**	0.563*	1.33**	5.25**	6.25**	0.750 ^{ns}
Within	9	0.306 ^{ns}	0.278 ^{ns}	0.111 ^{ns}	0.139 ^{ns}	0.528 ^{ns}	0.250 ^{ns}	0.194 ^{ns}
Total	11							

Appendix VII: Analysis of variance of data on prevalence of seed borne fungi of sweet gourd identified by blotter method

Sweet gourd	df	Seed borne infection (%)						
		<i>Aspergillus flavus</i>	<i>Aspergillus niger</i>	<i>Fusarium</i> sp.	<i>Alternaria</i> sp.	<i>Chaetomium</i> spp.	<i>Rhizopus</i> sp.	Unidentified bacteria
Between	2	0.583 ^{ns}	0.750*	0.583 ^{ns}	0.250 ^{ns}	1.83 ^{ns}	1.75 ^{ns}	0.250*
Within	9	0.611 ^{ns}	0.194 ^{ns}	0.306 ^{ns}	0.333 ^{ns}	0.528 ^{ns}	0.611 ^{ns}	0.278 ^{ns}
Total	11							

Appendix VIII: Analysis of variance of data on prevalence of seed borne fungi of snake gourd identified by blotter method

Snake gourd	df	Seed borne infection (%)						
		<i>Aspergillus flavus</i>	<i>Aspergillus niger</i>	<i>Fusarium</i> sp.	<i>Alternaria</i> sp.	<i>Chaetomium</i> spp.	<i>Rhizopus</i> sp.	Unidentified bacteria
Between	2	2.08 ^{ns}	0.333 ^{ns}	2.58**	0.333*	4.08**	1.00*	0.333*
Within	9	0.639 ^{ns}	0.222 ^{ns}	0.194	0.167 ^{ns}	0.306 ^{ns}	0.250 ^{ns}	0.083 ^{ns}
Total	11							

Appendix IX: Analysis of variance of data on prevalence of seed borne fungi of okra identified by blotter method

Okra	df	Seed borne infection (%)						
		<i>Aspergillus flavus</i>	<i>Aspergillus niger</i>	<i>Fusarium</i> sp.	<i>Alternaria</i> sp.	<i>Chaetomium</i> spp.	<i>Rhizopus</i> sp.	Unidentified bacteria
Between	2	2.33*	1.51*	0.083 ^{ns}	1.33*	6.58**	1.58*	1.08**
Within	9	0.722 ^{ns}	0.417 ^{ns}	0.139 ^{ns}	0.278 ^{ns}	0.278 ^{ns}	0.278 ^{ns}	0.111 ^{ns}
Total	11							

Appendix X: Analysis of variance of data on prevalence of seed borne fungi of country bean identified by blotter method

Country Bean	df	Seed borne infection (%)						
		<i>Aspergillus flavus</i>	<i>Aspergillus niger</i>	<i>Fusarium</i> sp.	<i>Alternaria</i> sp.	<i>Chaetomium</i> spp.	<i>Rhizopus</i> sp.	Unidentified bacteria
Between	2	1.03 ^{ns}	8.33**	0.333 ^{ns}	1.08**	1.00 ^{ns}	1.58*	2.31**
Within	9	0.750 ^{ns}	0.222 ^{ns}	0.250 ^{ns}	0.167 ^{ns}	0.361 ^{ns}	0.472 ^{ns}	0.160 ^{ns}
Total	11							