EFFECT OF DIFFERENT LEVELS OF BLACK POINTED SEED AND VERMICOMPOST ON LEAF BLIGHT *(BIPOLARIS SOROKINIANA)* SEVERITY, YIELD ATTRIBUTES AND SEED HEALTH STATUS OF WHEAT

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This is to certify that thesis entitled, ***"Effect of Different Levels of Black Pointed Seed and Vermicompost on Leaf Blight (Bipolaris sorokiniana) Severity, Yield Attributes and Seed Health Status of Wheat"*** *submitted to the Faculty of Agriculture, Sher-e-Bangla Agricultural University, Dhaka, in partial fulfillment of the requirements for the degree of* ***MASTER OF SCIENCE IN PLANT PATHOLOGY,*** *embodies the result of a piece of bona fide research work carried out by* ***Registration No. 08-03111*** *under my supervision and guidance. No part of the thesis has been submitted for any other degree or diploma in any institute.*

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

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DEDICATED TO

MY

BELOVED PARENTS

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ABSTRACT

An experiment was conducted during the period from November 2013 to July 2014 to find out the effect of different levels of black pointed seed and vermicompost on leaf blight (*Bipolaris sorokiniana)* severity, yield attributes and seed health status of wheat, cv. Shatabdi. Field experiment was carried out in RCBD design with 18 treatments. In the field condition, significant variations were found considering number of seedlings/m^2. The maximum (114.3 and 124.0) emergence of seedling was recorded in T_7 and the minimum (89.0 and 100.0) was in T_6 after 7 days and 15 days of sowing respectively. Leaf blight severity (0-5 scale) on flag leaf and penultimate leaf in flag leaf stage, panicle initiation stage, flowering stage, milking stage and hard dough stage were recorded minimum while applied treatment T_7 (Vermicompost @ 1.5 ton/ha and apparently healthy seed treated with Provax 200 @ 0.25%). The maximum leaf blight severity was recorded while applied treatment T_6 (without vermicompost and 25% black pointed seed in weight basis). The highest plant height (82.84 cm) and spike length (12.91cm) were recorded in T_7. The treatments showed significant variations on number and weight of grains/ear of different severity grades (0-5) of harvested seeds. Significantly higher 1000 seeds weight (42.70g), grain yield (3.66 t/ha) and straw yield (4.33 t/ha) were recorded in $T_{7.}$ The lowest 1000 seeds weight (37.42g), grain yield (2.33 t/ha) and straw yield (3.05 t/ha) were recorded in $T_{6.}$ In respect of seed germination, significant variations were observed among the treatments. Remarkable reduction of the seed germination was recorded in seeds with the maximum black pointed seed in the treatment T_6 (without vermicompost and 25% black pointed seed in weight basis). Germination percentage, normal seedling percentage and vigor index were the highest in T_7 and the lowest in $T_{6.}$ The shoot length, root length, seedling weight and vigor index (VI) were decreased with the increasing levels of black pointed seed. The shoot length, root length, seedling weight were the highest in T_7 and the lowest in $T_{6.}$

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LIST OF ABBREVIATED TERMS AND SYMBOLS

ABBREVIATION	FULL NAME
AEZ	Agro-Ecological Zone
BARI	Bangladesh Agricultural Research Institute
BAU	Bangladesh Agricultural University
BBS	Bangladesh Bureau of Statistics
BSMRAU	Bangladesh Sheikh Mujibur Rahman Agricultural University
CIMMYT	International Maize and Wheat Improvement Centre
cm	Centimeter
cv.	Cultiver (s)
DAS	Days After Sowing
e.g.	Example
et al.	*et* alibi (and others)
Etc.	Et cetera (and so on)
FAO	Food and Agriculture Organization
Fig	Figure
g	Gram
CRD	Complete Randomized design
hr	Hour
i.e.	Id est (that is)
ISTA	Inter National Seed Testing Agency
K	Potassium
Kg	Kilogram
Kg/ha	Kilogram per hectare
LSD	Least Significant Difference
m	Meter
m^2	Square meter
ml	Millimeter
ABBREVIATION	**FULL NAME**
mm	Millimeter
MP	Murate of Potash
N	Nitrogen
No.	Number
PDA	Potato Dextrose Agar
RCBD	Randomized Complete Block Design
S	Sulphur
SAU	Shere -e- Bangla Agricultural University
T	Treatment
t/ha	Ton per hectare
TSP	Triple Super Phosphate
Viz.	Videlict (namely)
@	At the rate
^0C	Degree Celcious
pH	Hydrogen ion potentiality
$\leq$	Less than or equal
$\geq$	More than or equal
%	Percent

CHAPTER I

INTRODUCTION

Wheat (*Triticum aestivum L.*) is the most important cereal crop in the world and is a major source of nourishment. It is one of the most important grain crops providing nearly 20% of the total world food requirement. In Bangladesh it is the second major cereal crop next to rice. Though the crop has been introduced in 1961 in the country, it has gained popularity after 1975. In Bangladesh 4,29,607 acres of land was under wheat cultivation and total production was about 3.033 metric tons per hectare (BBS, 2014). Wheat is counted the third among three big cereal crops in the world, and its yield has increased from 560 million tons in 2003 to 713 million tons in 2013 (FAO 2013).

About one third people of the world one billion as many as 43 countries and provides about 20% of total food calories. It contains carbohydrate (78.1%), Protein (14.7%), minerals (2.1%) and vitamins (Peterson, 1965).

Wheat is a winter crop. Its time for sowing of wheat in Bangladesh is between mid November and first week of December (Hossain and Alam, 1986), due to its own definite requirements for temperature and light for emergence, growth and flowering. The weather conditions particularly relative humidity is one of the most important congenial factors for the development of the disease.

Wheat suffers from about 26 seed borne pathogens causing 14 seed borne diseases (Fakir, 1999). Disease causes loss of yield approximately 15-20% equivalents to 20-30 million tons annually in developing countries (Hanson *et al.*, 1982). In Bangladesh about 10% yield reduction was reported due to diseases (Miah, 1985). Among the diseases, leaf blight caused by *Bipolaris sorokiniana* is the major and devastating disease of wheat in Bangladesh (Hossain and Azad, 1985). Leaf blight of wheat caused by *Bipolaris sorokiniana*, is one of the major cereal disease of global concern. The disease is one of the constraints for crops in warmer growing areas and cause significant yield losses (Aftabuddin *et al*., 1991). The abnormal weather conditions play a critical role in leaf blight problem including high moisture and temperature (Fischer, 1985; Hussain *et al*., 2010). The fungi, *B. sorokiniana* is generally considered highly seed transmitted pathogen expressed as black point on seed and possesses a potential threat to the subsequent crops. It produces seedling blight, leaf spot, leaf blotch and leaf blight on standing crop. It also causes black point, seed rot, shriveling of seed resulting poor seedling emergence, plant stand and low yield of subsequent crops (Aulakh *et al*., 1988).

Bipolaris leaf blight appears at the seedling stage (Alam *et al.,* 1994). *Bipolaris sorokiniana* is also a known cause of foliar blight, seedling blight, head blight and common root rot of wheat (Shahzad *et al.,* 2009). *Bipolaris sorokiniana* at flag leaf stage may cause of 7-100% reduction in formation of grains/ear (Hossain and Azad, 1994). The pathogen attacks wheat grains causing black point and the disease reduced yield up to 40% in field condition (Hossain *et al*. 1998), in severe cases it may cause of 57.6% and 64.5% yield reduction of wheat due to *Bipolaris* leaf blight in cvs shatabdi and sonalika, respectively (Rashid and Fakir 1998). The leaf blight causing pathogen is found most frequent in Bangladesh and currently becoming the major limitation for wheat cultivation in the country (Alam *et al*., 1994). The pathogen is named as a silent killer of the wheat crop as its losses are not well estimated in this part of the world compared to the rusts. *Bipolaris sorokiniana* is most frequently associated with poor germination and abnormal seedlings of wheat (Ammara *et al*., 2001). The head infection increase the unwanted black pointed, shriveled and infected seeds. In severe cases, the infected head may turn into blight.

The occurrence of black point disease of wheat seems to be increasing during the last year all over Bangladesh. The disease is reported to have deteriorating on quality of the seed. The discolored lustureless seeds having at least five types of visible symptom i, e. brown to black embryo tip, brown to blackish color around whole embryo, discoloration on embryo spreading to the part of endosperm and discoloration of whole embryo and most of the part of endosperm and whole seed discoloration and shrivelledness. Virtually, it is impossible to produce black point free seed; however, seed with a reduced level of black point can be produced that may ensure good germination, high seedling vigor (Aulakh *et al*.1988).

Seed health plays an important role for successful cultivation and yield exploitation of a crop species. Among various factors that affect seed health, the most important are the seed borne fungi that not only lower seed germination, but also reduce seed vigor resulting in low yield. Healthy seed plays an important role not only for unsuccessful cultivation but also for increasing yield of crop. Seed-borne pathogens of wheat are responsible to cause variation in plant morphology and also reducing yield up to 15 - 90 % if untreated seeds are grown in the field (Wiese, 1984).

Organic fertilizers are the key to improve the sustainability of agricultural farming system and soil productivity. Vermicompost helps in increasing the organic matter content of soil, which helps in increasing the natural productivity of soil. It is a viable technology for converting waste into organic fertilizers (Gutiérrez-Miceliet *et al.*, 2011) endowed with hormone- like substances. The chemical characteristics of vermicompost and its products may vary in a significant range, depending upon its initial substrate source (such as manure from cow, chicken, pig and sheep, or food waste):19.5 - 51.7 % of organic carbon, 1.3 - 5.8 % of total nitrogen, 0.4 - 4.5 % of total phosphorous, 0.4 - 9.2 % of total potassium, 5.3 to 8.0 pH and 0.12 to 4.5 dS/m electrical conductivity (Atiyeh *et al.,* 2000).

Besides the earthworm compost, its leachate drained during vermicomposting process is a remarkable natural plant growth promoter due to its biochemical properties, which includes humic substances.

The vermicomposting is bio-oxidation and stabilization of organic material involving the joint action of earthworm and microorganisms. Although, microbes are responsible for the biological degradation of the organic matter, earthworms are the important drivers of the process, conditioning the substrate and altering biological activity (Aira *et al.,* 2002).

Vermicompost is important in maintaining and enhancing the quality of environment and conserving resources for sustainable agriculture (Simanaviciene *et al.,* 2001). Use of vermicompost reduces the cost of production, increases plant’s health and resistance against biotic and abiotic causes and fertility and water holding capacity of the soil. Seed infection level had a significant effect on disease incidence and severity at different growth stages like flag leaf stage, panicle initiation stage, flowering stage, milk stage and hard dough stage (Chowdhury *et al.*, 2010). Soil amendment with vermicompost can be the eco-friendly approach to control the leaf blight disease.

Considering the above facts, the present research program has been designed to determine the effect of different levels of black pointed seed and vermicompost on leaf blight (*Bipolaris sorokiniana*) severity, yield attributes and seed health status of harvested seed of wheat.

The objectives of the present study were

i. To evaluate the effect of different levels of black pointed seeds and vermicompost on leaf blight severity of wheat.

ii. To determine the effect of different levels of black pointed seeds and vermicompost on plant growth parameters of wheat.

iii. To estimate the effect of different levels of black pointed seeds and vermicompost on yield and seed health status of harvested seed of wheat.

CHAPTER II

REVIEW OF LITERATURE

Impact of black pointed seeds on leaf blight severity by *Bipolaris sorokiniana* and yield attributes of wheat

Podder *et al.* (2012) reported that percent leaf area infection (%LAI) of flag leaf and penultimate leaf of wheat was increased with increasing the age of plant. The maximum and minimum Area Under Disease Progress Curve (AUDPC) of flag leaf and penultimate leaf were also recorded in highest and lowest level of black pointed seed, respectively.

Chowdhury *et al.* (2010) reported that the leaf blight severity of wheat was increased with the increasing level of seed infection. The different plant yield attributes like number of grains/ear, weight of grains/ear and weight of healthy grains/ear were increased with decreasing of seed infection level.

Duveiller and Sharma (2009) reported that among wheat diseases, spot blotch caused by *Bipolaris sorokiniana* (teleomorph: *Cochliobolus sativus*) singly or as foliar blight complex in combination with *Pyrenophora trictici-repentis* are the most economically important diseases. Foliar blight complex is also the most important disease in the non-traditional wheat-growing areas of the Indian subcontinent, which covers around 13 million ha land area adopting an intensive rice-wheat system in India, Bangladesh, and Nepal.

Reza *et al.* (2006) reported on the effect of different levels of seed and plant infection by *Bipolaris sorokiniana* on wheat determined under induced field condition. The maximum seed rot/seedling mortality (15.73%) followed by subsequent leaf blight severity (75.4%) was recorded as a result of sowing 30% infected seeds while the minimum (5%) infected seeds resulted in 3.1% and 57.53% of seed rot/seedling mortality and leaf blight severity, respectively. He also found that 65.36 percent disease severity interning the corresponding 17.42 percent seed infection.

Akbari *et al. (*2004) reported that Seed quality is very important to optimum growth and yield production in farm which influenced by many factors such as genetic characteristics, viability, germination percent, vigor, moisture content, storage conditions, survival ability and seed health, but their most important is germination percent and vigor. Generally, factors such as genetic structure, environment and parental nutrition, maturity stage in harvest time, mechanical damages, seed storages, age and aging and pathogens, affect seed germination and vigor.

Iram and Ahmed (2004) stated that the diseases varying levels of threats to the production of wheat in different agro-ecological zones of the country. Earlier studies on foliar blights inciting pathogens in the rice-wheat cropping system of revealed that *Bipolaris sorokiniana, Drechslera tetramera, Pyrenophora tritici-repentis, Alternari aalternata* and *Stemphylium* sp. are the major pathogens of wheat. In Southeast Asia the most frequently isolated fungus is *B.sorokiniana* and is associated with warm and humid climates. The spot blotch causing pathogen is found most frequent in Bangladesh and currently the major limitation for wheat cultivation in that country.

Mahto *et al*. (2002) observed that Spot blotch of wheat caused by *Bipolaris sorokiniana,* is one of the major cereal disease of global concern. The disease is one of the constraints for crops in warmer growing areas and cause significant yield loss. Epidemic has been observed in Indian Punjab with the dominant pathogen *B. sorokiniana* followed by *Fusarium* spp and the yield losses ranged from 27%-56.6% during 1998-99 in north eastern and north western plains of India due to the leaf blight caused by *B. sorokiniana.*

Mondal (2000) reported that the infected seeds and soils infested either with conditional suspension or colonized grains may serve as potential source for the survival of *Bipolaris sorokiniana* resulting germination failure, seedling mortality and leaf blight of development.

Hossain (2000) reported that seed germination and seedling emergence were significantly decreased with the increase in number of black pointed seed. The sample having 28% black pointed seed resulted maximum reduction in germination by 20.20% and 42.69% in the blotter and rolled paper towel method and emergence by 34.27% and 40.74% in the field and in the pot, respectively. The rate of reduction of growth was maximum by 28% black pointed seeds as recorded root length was 57.21% and for shoot length was 41.40%. Significantly the highest (1582.61) and the lowest (433.16) seedling Vigor Index (VI) were recorded while the best seed treated with Vitavax and in 28% black pointed seed and seed samples 28% having black pointed seeds, respectively. Significant decrease in plant stand in pot and field has been observed with the increase in number of black pointed seeds in seed samples. Higher the level of seed borne fungal infection, there will be higher primary inoculum level in the field resulted higher infection in the field and maximum infection severity was attained at hard dough stage. The higher level of black point infection in the seed sample incited more disease to the crop plants resulting formation of higher number of diseased seed in the field.

Ahmed (1999) reported that 40.0-51.17% yield loss due to leaf blight disease under the field condition. Foot and root rot disease also causes considerable yield loss in wheat. There is no absoluter resistant gene of BpLB and Foot and root rot diseases in Bangladesh. Various kinds of abiotic stresses like terminal heat, waters stress, weed infestation and imbalance fertilizers etc are known to influence the severity of the diseases in different degrees.

Mehta (1998) conducted that *Bipolaris sorokiniana* develops dark brown necrotic lesions on roots, crown, leaves and lower leaf sheaths. It develops oval to elongated light to dark brown blotches on leaf blades and sheaths, when it severely infect the roots and crown portions, the plants dry out without producing any seed. Similarly infected spikelets under favourable conditions produce shriveled grains. The conducive weather conditions i.e., continuous rain for 5-6 days followed by warmer temperatures (day average of 20–30oC), spot blotch epidemic can develop very rapidly.

Rahman and Islam (1998) reported that Black point has adverse effects on seed weight, germination and grain yield. The reduction in 1000-grain weight was as large as 41.96% when the grains were severely infected with the black point fungi .The germination of black point affected seeds was reduced to a great extent as compared to healthy grains.

Rahman and Islam (1998) reported that the effect of black point of seeds of wheat on its qualitative characters such as weight of 1000 grains total crude protein, total crude fiber, total ash dry matter and seed shoot and root growth into five different grades (Grade-0, I, II, III, IV) on the basis of level of black point infection .The decrease was more pronounced in grade III and IV infected seeds. Germination percentage decreased sharply with the increased severity of infection of the disease, while shoot and root growth also decreased as the grade of infection increased.

Reis *et al.* (1998) *observed that* the basic principle involved in the control of a seed-borne disease is reducing the population of primary inoculums of the pathogen. Infected seeds are one of the most important sources of primary inoculums that survive between wheat cropping seasons. Thus use of healthy seeds provides a means of reducing primary inoculums and in turn reduces foliage infection and seedling blight. The predominant black point fungus, *B. sorokiniana* is highly seed transmitted and more than 80% seed to plant transmission of this pathogen have been established in wheat.

Saari (1998) reported that Spot blotch disease of wheat caused by *Bipolaris sorokiniana* is a disease causing substantial damage to wheat (*Triticum aestivum L*.em.Thell) in south Asia. Some traits viz., erect leaf posture, leaf tip necrosis and stay green traits have been demonstrated to have positive effects on resistance to spot blotch.

Rashid *et al.* (1997) reported that a highly significant effect of seed borne infection by *Bipolaris sorokiniana* on the germination of seeds of wheat as well as pot experiment. At the maximum seed borne infection level (90%) both the cultivars yielded the minimum germination of 30.25 and 26.50%, respectively. Relationship between the levels of seed borne infection and present seed germination showed gradual reduction in germination of seed with the increase level of infection level. The maximum germination reductions were found as 71.5% and 68.00% in cv Sonalika and cv. Kanchan, respectively.

Williamson (1997) reported that Black point can result discoloration may cause the degradation of seed coat, which could be explained by increased levels of active enzymes and compounds in the seed coat triggered by humidity during the grain filling period. Peroxidase catalyses the oxidation of phenolic compounds. Black point reduced the germination rate, number of embryonic roots, and coleoptiles length and delayed seedling emergence and reduced seedling vigour.

Alam *et al*. (1995) estimated the yield loss of wheat cultivar of kanchon due to *Bipolaris* leaf blight at the farmers' fields of four different areas. The average yield losses of wheat due to *Bipolaris* leaf blight disease at Dinajpur, Jessore and Ishurdi were 13.9, 16.2, 14.8 and 14.5 percent, respectively. The average losses of grains were 14.9 percent over the location.

Alam *et al*. (1994) reported that in north eastern and north western plains of India due to the leaf blight caused by *B.sorokiniana*. Due to this pathogen severe losses were estimated up to15% on several farms over a number of years in Bangladesh, In Nepal the loss was reported up to 23.8%. This seed and soil borne pathogen affects the seed germination and seedling emergence significantly.

Rashid *et al. (*1994) reported that the seed quality deterioration is positively associated with the incidence of leaf blight caused by *Bipolaris sorokiniana* under field condition.

Mathur (1993) reported that infected ears may look normal but there may be elliptical, brown to dark brown lesions on the inner side of the glumes. Diseased kernels are discolored and appear withered, black pointed, or smudged. Discoloration generally occurs at the germ end of the caryopsis and in severe cases it extends along the crease and over the shoulders causing the shriveling of the whole seed. The dark brown discoloration at the embryo end is generally limited to the seed coat; the aleurone cells underneath are not damaged. However, the endosperm frequently turns slightly brownish grey. The discoloration from the scutellum may also spread some distance in the mesocarp on the lower and upper sides of the grooves of the grain.

Talukder and Fakir (1993) reported that under the favorable conditions of disease development, the airborne inocula of the causal fungi infect the spikes and ultimately lead to the point on the grains. However, the characteristic discoloration of the kernels usually appears during the soft dough to hard dough stages of grain development. The prevalence of different fungi associated with floret lemma and developing grains and to assess the incidence of black point disease under different exposure periods of spikes to airborne inocula of the causal fungi.

Hossain and Azad (1992) reported that controlling leaf blight disease; several approaches have been practiced, such as use of resistant variety, cultural control, chemical control, biological control and use of plant extract etc. Cultivation of resistant variety is the most acceptable method for controlling this disease. But none of the wheat varieties in the country is found resistant against this disease. The most acceptable method for controlling this disease is sowing of pathogen free seeds. Therefore along with routine seed health testing, seed treatment before sowing is necessary. Seed treatment of with seed-dressing fungicides was found to improve germination and decrease infection of seedling growth from the black pointed seeds.

Reis (1991) reported that the main sources of inoculums of *Bipolaris sorokiniana* cause of spot blotch wheat were infected seed, infected residues, soil having free dormant conidia and secondary hosts. These also represented the survival mechanisms of the pathogen. He also reported that the incidence of spot blotch caused by *Bipolaris sorokiniana* in early growth stages of wheat lower in the region where wheat was not previously grown. It also often observed that the infected seeds being the the sole source of inoculums and the transmission rate of the fungus from infected seed to coleoptiles was about 2:1.

Talukder and Fakir (1991) reported that the disease symptoms of leaf blight of wheat first appeared on the floret grains at the hard dough stage and it become pronounced at the dead ripe stage. The percentage of black point affected grain observed at the full and dead ripe stage 6.25 and 37.08%, respectively.

Aulakh *et al.* (1988) reported that to have deteriorating effect on quality of the seed. Black point of wheat is a disease of grain, discoloration is followed at the embryonal end and it severe cases the whole grain may be diseased and shriveled. *Bipolaris sorokiniana* causes seed rot poor stand reducing seedling emergence and yield of the subsequent crop.

Fakir (1988) observed that in Bangladesh no significant effect of sowing o.6 to 12 percent black point affects the seeds on the yield, incidence of seedling blight or leaf blight and development of black point in the harvested grains. However he showed that reduction in germination of black point affected seeds was directly related with the severity of infection. According to him *Drechslera sorokiniana* was responsible to cause more disease to the germinating seeds and seedling than other black point fungi.

Kaur (1988) reported that wheat seed germination and seedling survival were affected by the pathogen. The growth and survival of wheat seedlings to the attack of seed transmitted *B. sorokiniana* is lacking. Seedling infection is usually the primary phase of subsequent disease developments in plants by this seed transmitted pathogen .The relative effects of seed to plant transmission of the pathogen on different growth characters and survival wheat of seedlings.

Khanum *et al.* (1987) reported that the disease is characterized by brown to black discoloration usually restricted to the germ end of the seeds. Black point symptom may also develop on the endosperm, ventral crease or even on the brush end of the grain. In case of severe infection, particularly when *B. sorokiniana* is involved, the grain may be completely discolored and shriveled. Impaired seed germination and significant reduction in seedling vigor and grain yield to black point infection.

Vermicompost for controlling diseases

Singhai *et al.* (2011) reported that the potential of *Pseudomonad mosselii* strain R1 in promoting plant growth as well as inducing antimicrobial mechanisms systemically in the host plants facilitated by the organic amendment in the form of vermicompost.

Nesterenko *et al.* (2009) found that the lowest wheat disease incidence caused by *Bipolaris sorokiniana* in two crop plantings values were obtained from the second planting in soil like substrate (SLS) obtained by sequential wheat straw treatment with a fungus (*Pleurotus ostreatus*) and worms (*Eisenia foetida*). The values were 26% and 41% at the first and the second infection levels, respectively. For soil the values were 60% and 82%, respectively, and for sand they were 67% and 74%, respectively. SLS significantly suppressed the germination of *B. sorokiniana* conidia.

Chawla and Gangopadhyay (2009) reported that the biocontrol potentiality of *T. harzianum* and *P. fluorescence* was relatively better in presence of farm yard manure or mustard cake in suppressing cumin wilt caused by *Fusarium oxysporum* f. sp. *Cumini* while disease control due to seed treatment with *T. viride, T. harzianum or P. fluorescens* was similar in presence of vermicompost.

Importance of Vermicompost on yield of wheat

Jack (2010) reported that all composts are rich in beneficial soil microbes. Vermicompost is especially rich in microbial diversity. Earthworms further proliferates useful microbes in billions and trillions in soil. Earthworms can modify soil microbial community structure depending on the type of organic matter present in soil.

Pattnaik and Reddy (2009) reported that the nutrients- N, P, K, Ca, and Mg increased from vermicompost and compost while the organic carbon, C: N and C: P ratios decreased as the composting process progressed from 0 to 15, 30, 45 and 60 days. The nutrient status of vermicompost of all earthworm species produced from both the wastes was more than that of the compost and that of their respective substrates. Vermicompost, another type of compost which has attracted an increasing attention, is formed by earthworm activities from soil organic residues, mainly animal manures. Earthworms stabilize organic residues by producing earthworm casts which are soil conditioners that have a high nutrient bioavailability for plant growth.

Suthar (2006) reported that vermicompost may be potential sources of nutrients for field crops if applied in suitable ratios with synthetic fertilizers. The earlier workers have reported a positive effect of vermicompost application on growth and productivity of cereals and legumes.

Bhuiyan (2005) reported with the increasing doses of vermicompost and chemical fertilizers increased grain and straw yield of wheat. He also stated that vermicompost at 3 ton/ha or chemical fertilizers 120-80-60 kg/ha increased N, P, K and S concentrations in wheat plant significantly at different stages of plant growth and also enhanced N, P, K and S uptake at the ripening stage.

Chaoui *et al*. (2003) reported that Vermicompost has been shown to have high levels of total and available nitrogen, phosphorous, potassium and micronutrients ,microbial and enzyme activities and growth regulators and continuous and adequate use with proper management can increase soil organic carbon, soil water retention and transmission and improvement in other physical properties of soil like bulk density, penetration resistance and aggregation as well as beneficial effect on the growth of a variety of plants due to higher levels growth stimulating substances such as enzymes, antibiotics and growth hormones available in vermicompost.

Khandal and Nagendra (2002) studied the effect of chemical fertilizers (KPK fertilizers) and organic manure (vermicompost). Plant height, dry matter production and grain yield were higher at higher dose of vermicompost. Number of tillers and leaves per plant were very low at early stages of growth and suddenly increased after adding different concentrations of vermicompost and organic manure.

Orozco *et al.* (1996) reported that Vermicompost contains most nutrients in plant- available forms such as nitrates, phosphates, and exchangeable calcium and soluble potassium. There is accumulating scientific evidence that vermicomposts can influence the growth and productivity of plants significantly. Various greenhouse and field studies have examined the effects of a varity of vermicomposts on a wide range of crops including cereals and legumes, vegetable, ornamental and flowering plants and field crops. Annual application of adequate amounts of some organic residues (vermicompost) led to significant increase in soil enzyme activities such as urease, phosphomonoesterase, phosphodiesterase and arylsulphatase.

CHAPTER III

MATERIALS AND METHODS

The experiment was conducted in the Plant Pathology Laboratory and in the farm field allotted for the Department of Plant Pathology, Sher-e-Bangla Agricultural University, Dhaka to find out the effect of different levels of black pointed seed and vermicompost on leaf blight (*Bipolaris sorokiniana)* severity, yield attributes and seed health status of wheat.

3.1 Field experiment

The field experiment was conducted at the firm field allotted for the Department of Plant Pathology. The location of experimental site is sown in Fig 1. In previous season, the experimental field was occupied by rice.

3.1.1 Duration of the experiment

The experiment was conducted during the Rabi season from November 2013 to April 2014.

3.1.2 Planting material

The wheat variety (*Triticum aestivum*) Shatabdi, was used the most widely cultivated variety in the country.

3.1.3 Collection of seed sample

Wheat seed samples of variety Shatabdi was collected from a local farmer named Md. Ismail Hossain of Village- Gouri Gram, Thana-Santhia, and District-Pabna. After collection the seeds were kept in a plastic container with air tight lid and the container was stored in normal room temperature in Seed Pathology Laboratory, Sher-e-Bangla Agricultural University, Dhaka.

3.1.4 Treatment used in the experiment

Seeds were collected and the collected seed sample was physically sorted out to separate apparently healthy looking seeds with bold golden color and black pointed seeds. Then the black pointed seeds and apparently healthy looking seeds were mixed in different percentages of weight basis to prepare different treatments. The apparently healthy seed was treated with Provax 200@ 0.25% to prepare seed sample for a treatment. There were 18 treatments.

The treatments are:

T_1 = (Apparently healthy seed treated with Provax 200 @ 0.25%)

T_2 = (5 % black pointed seed in weight basis)

T_3 = (10 % black pointed seed in weight basis)

T_4 = (15 % black pointed seed in weight basis)

T_5 = (20 % black pointed seed in weight basis)

T_6 = (25 % black pointed seed in weight basis)

T_7 = (Vermicompost @ 1.5 ton/ha + T_1)

T_8 = (Vermicompost @ 1.5 ton/ha + T_2)

T_9 = (Vermicompost @ 1.5 ton/ha + T_3)

T_{10} = (Vermicompost @ 1.5 ton/ha + T_4)

T_{11} = (Vermicompost @ 1.5 ton/ha + T_5)

T_{12} = (Vermicompost @ 1.5 ton/ha + T_6)

T_{13} = (Vermicompost @ 3.0 ton/ha + T_1)

T_{14} = (Vermicompost @ 3.0 ton/ha + T_2)

T_{15} = (Vermicompost @ 3.0 ton/ha + T_3)

T_{16} = (Vermicompost @ 3.0 ton/ha + T_4)

T_{17} = (Vermicompost @ 3.0 ton/ha + T_5)

T_{18} = (Vermicompost @ 3.0 ton/ha + T_6)

3.1.5 Vermicompost preparation

Kitchen waste especially vegetable wastes and cow dung were used as vermin bed substrates due to their availability in large quantities in Bangladesh. Earthworms are chosen for the resistance to extreme conditions and feeding and reproductive rate. *Eisenia fetida* is the most efficient in waste processing in Bangladesh. Vermicompost was produced by using vermicomposting "chari" method. In this method, 75% cow dung and 25% kitchen wastes were subjected to produce vermicompost by the decomposition of organic wastes facilitated through earthworm *Eisenia fetida.*

3.1.6 The Experimental Design and Layout

The experiment was laid out in RCBD design with three replications. The experiment was divided into three blocks. Blocks represented the replication. So the total number of unit plots in the entire experiment was 3x18=54. Size of each unit plot was 1.5m x1m = 1.5m^2.The distance between sub plot was 0.5m and block was 1m.

3.1.7 Application of vermicompost

Vermicompost was applied as per requirement of the treatments.

3.1.8 Climate of experimental site

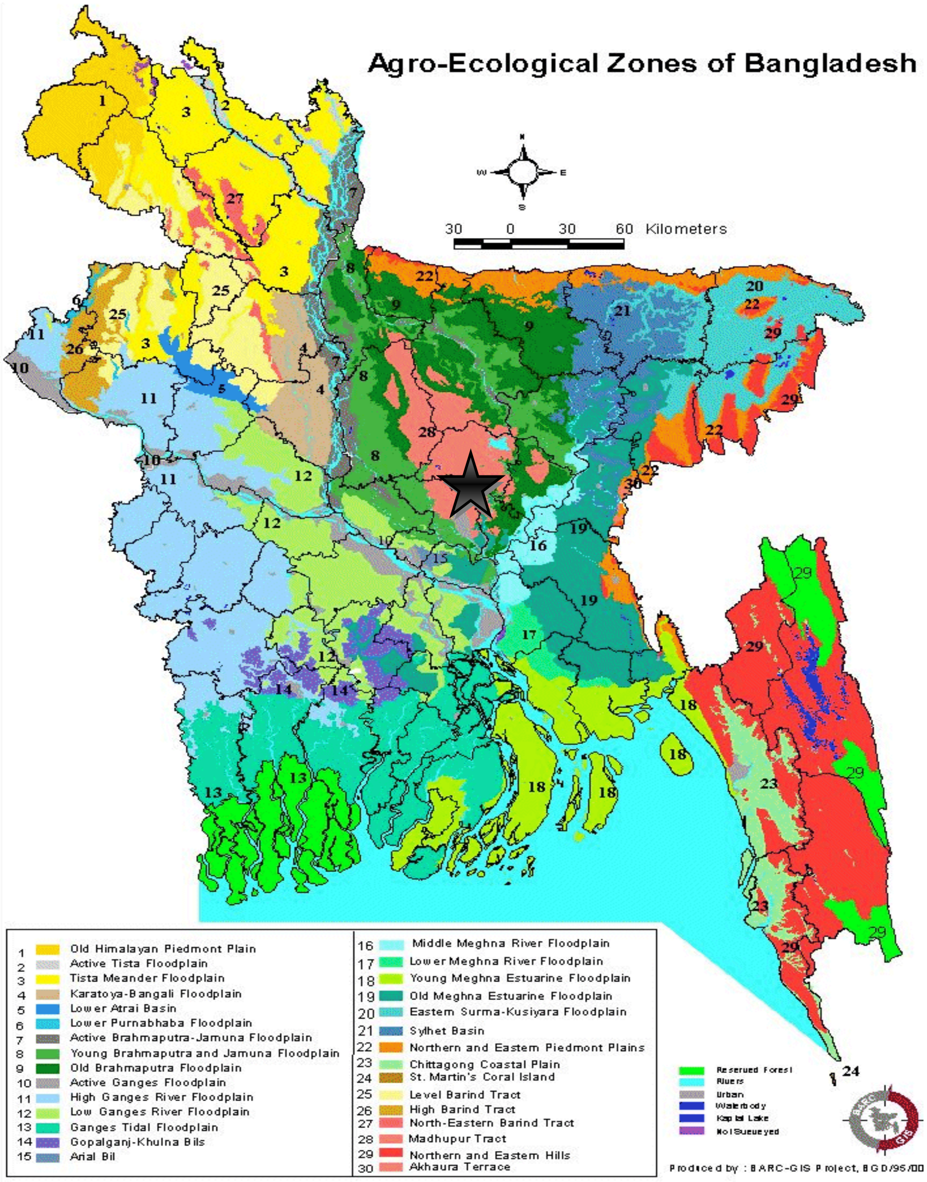
The experimental area was under the subtropical climate which is characterized with the comparatively high rainfall, high humidity, high temperature, relatively long day during April to September and scanty, low humidity, low temperature and short day period during October to March. The later period (October to March) is favorable for wheat cultivation. Monthly air temperature, relatively humidity (%) and total rainfall (mm) and average sunshine hour of site during the experimental period have shown in appendix-1.

3.1.9 Soil

The soil of the experimental soil belongs to the agro –ecological region of Madhpur Tract, AEZ-28. It was red brown terrace soil and belongs to ‘’ Nodda’’ cultivated soil series. The soil was silty clay in texture. Organic matter content was very low (0.78%) and soil pH was 5.6. The physical and chemical characteristics of soil have been shown appendix –II.

3.1.10 Land preparation

The experimental field was thoroughly ploughed and cleaned prior to seed sowing and application of fertilizers and manure was done in the field. The experimental field was prepared by thorough ploughing followed by laddering to have a good tilth. Finally the land was properly leveled before seed sowing.



★ Indicate experimental site (AEZ-28)

Source: www.fao.org

Fig 1: Location of experimental field

3.1.11 Applications of fertilizers

The field was fertilized at the rate of 220 Kg Urea, 180 Kg TSP, 50 Kg MP, 120 Kg Gypsum and 10 tons Cow dung per hectare (Krishi Projukti Hatbooi, 2005). Two third of Urea, full dose of TSP, MP, Gypsum and Cow dung was applied at the time of final land preparation. Remaining one third of Urea was applied at 21 days after seed sowing.

3.1.12 Sowing of seeds

Wheat seeds were sown in the field at the rate of 120 kg/ha. The seeds were placed continuously in lines properly at a depth of 5cm and were covered by soil with the help of land. The distance between the lines was 25 cm.

3.1.13 Intercultural operations

Weeds were controlled thoroughly. The weeds were controlled by nirani (hand hoe). After sowing, light irrigation was given for proper germination. Then flood irrigation was given at crown root initiation (20 DAS), heading (50 DAS) and grain filling (70 DAS) stages. During irrigation, care was taken so that water can not flow from one plot to another or overflow the boundary of the plots.

3.1.14 Plant protection activities

Special care was taken for 12 days after sowing to protect the crop from birds especially at sowing and germination stages and at the ripening stage of the crop.

3.1.15 Tagging and data collection

Randomly 10 plants were selected and tagged for rating and mean values were determined to get rating score of each treatment.

3.1.16 Isolation and identification of pathogen

The diseased leaves were collected and were taken to the laboratory. The leaves were then cut into small pieces (about 0.5 cm) with diseased and healthy portion and surface sterilized with $HgCl_2$ solution (0.01%) for 30 second. The cut pieces were then washed in water at three times and were placed onto PDA media in petridish. The plates were then incubated at 25±1°C for 7 days. Later the pathogen was purified using hyphal tip culture method and grown on PDA media at 25±1°C for two weeks and identified as *Bipolaris sorokiniana* with the help of relevant literature.

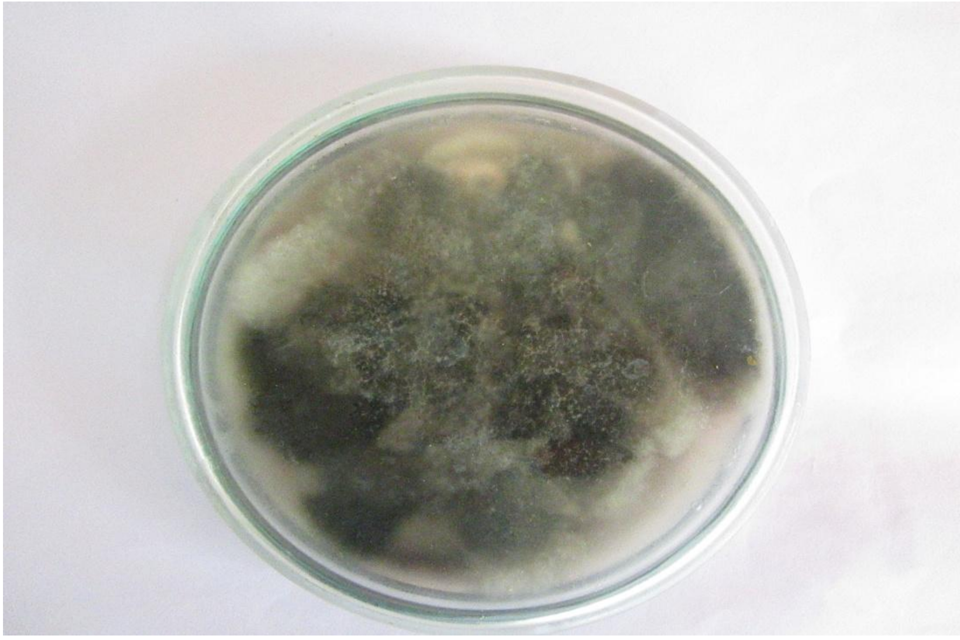


Figure 2: Pure culture of *Bipolaris sorokiniana*

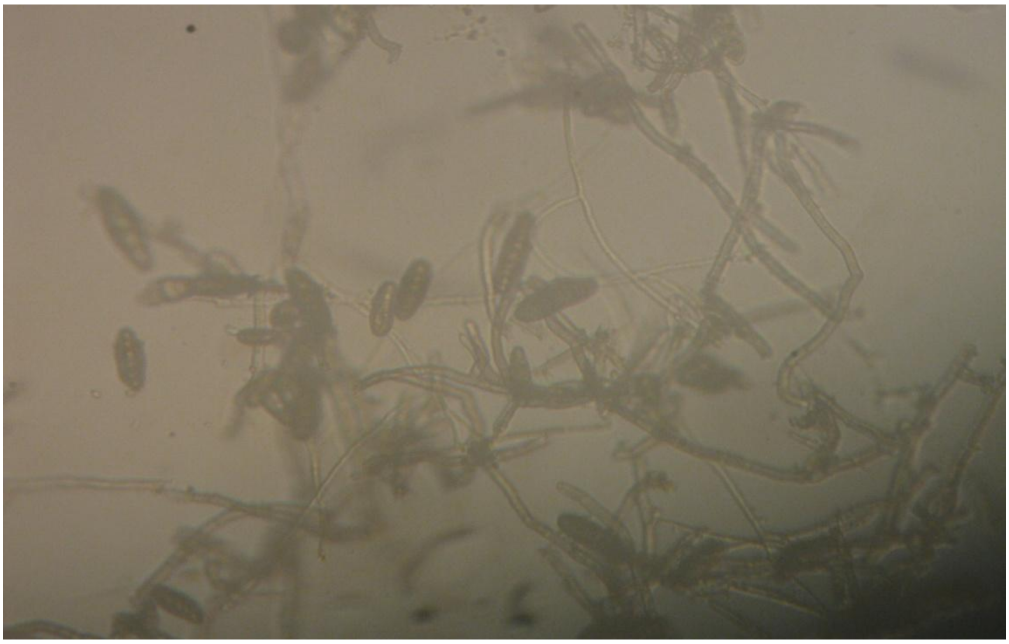


Figure 3: Mycelia and conidia of *Bipolaris sorokiniana* under compound Microscope (X 100)

3.1.17 Evaluation of leaf blight severity

Disease severity of Flag leaf and Penultimate was measured by using the following "0-5" scale (Horsfall and Barratt, 1945) at five growth stages of plant viz. flag leaf stage, panicle initiation stage, flowering stage, milking stage and hard dough stage. The Leaf blight severity of the disease was recorded following 0-5 grade. The grades are given below:

%LAD	Scale/Grade
0	0
0.1-5	1
5.1-12	2
12.1-25	3
25.1-50	4
>50	5

Percent Disease Index (PDI) of Flag leaf and Flag leaf -1 was measured by using the following formula (Horsfall and Barratt, 1945) at five growth stages of plant viz. flag leaf stage, panicle initiation stage, flowering stage, milking stage and hard dough stage.

$$\text{PDI} = \frac{\text{Sum of total ratings/grading}}{\text{Total No. of observation} \times \text{Highest Grade in the scale}} \times 100$$

3.1.18 Collection of data on yield and contributing characters

Data of plant growth and yield contributing characters was recorded from the randomly selected 10 tagged plants of each unit plot on the following parameters.

i) Number of Seedlings/m^2 were counted after 7 and 15 days of sowing .

ii) Plant height (cm)

Iii) Spike length (cm)

iv) Number of different Grade of seeds (0-5 scale)

v) Weight of different Grade of seeds (0-5 scale)

vi) 1000 grain weight (g)

vii) Straw yield (t/ha)

viii) Grain yield (t/ha)

3.1.19 Grading of seeds

The disease severity grade of harvested seeds was done following the 0-5 rating scale (Gilchrist, 1985). The rating scale is as follows:

0 = Free from infection

1 = Only embryo blackish

2 = Embryo and its adjacent area slightly infected

3 = Embryo and less than ¼ of grains are discolored

4 = Embryo and ½ of grain are infected

5 = Grains are shriveled, almost completely discolored or more than ½ of grains were discolored.

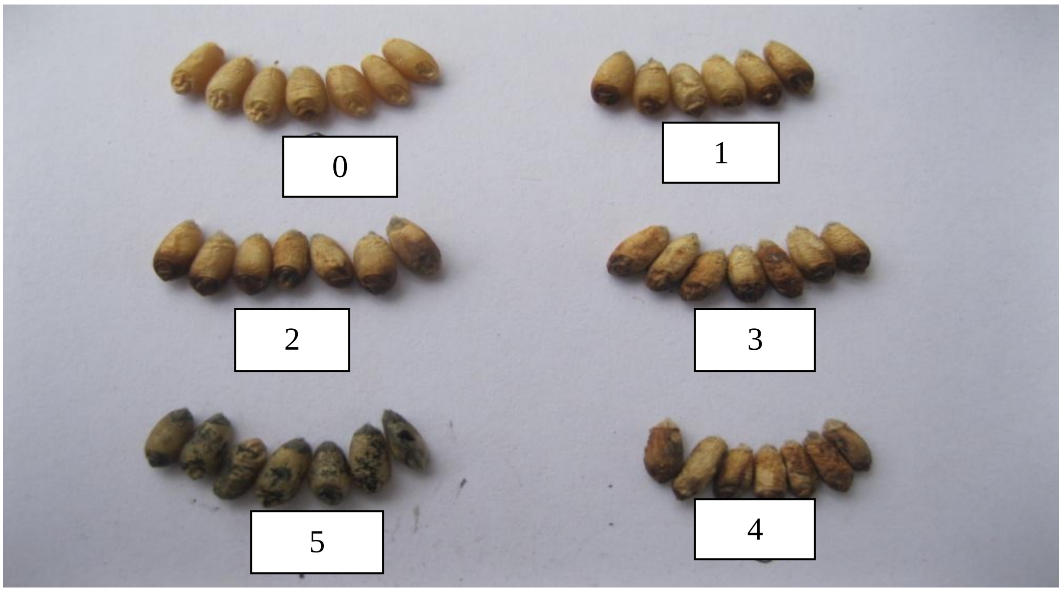


Figure 4: Wheat seed showing different (0-5) grades

3.1.20 Harvesting of crop

The crop was harvested at full ripening stage on 30 March, 2014.

3.2 Laboratory experiment (after harvesting)

The laboratory experiment was conducted in the Molecular Plant Pathology Laboratory of Department of Plant Pathology. Sher-e-Bangla Agricultural University, Dhaka during the period of April 2014 to July 2014.

3.2.1 Seed health study on Laboratory experiment

Laboratory experiment was performed in the Molecular Plant Pathology Laboratory of Department of Plant Pathology, Sher-e-Bangla Agricultural University, Dhaka. For studying the seed health different methods were used. These are as follows

3.2.2 Design of experiment

CRD with three replications

3.2.3 Blotter test

To determine the seed health status, the blotter method (ISTA, 1996) was used. 400 seed were tested for each treatment. In this method, three layers of blotter paper were soaked in sterilized water and placed at the bottom of each sterilized glass petridish. Then, twenty five seeds were plated on the blotter paper in a petridish maintaining equal distance and covered with lid. The seeds on petridishes were incubated in an air cooled room at about 20°C temperature under 12/12 hr light and darkness cycle for 7 days. Sterilized water was added time to time to maintain the moisture. After 7 days of incubation, the seeds were observed for the presence of seed-borne *Bipolaris sorokiniana* under stereo binocular microscope. Germination of the seeds was also being recorded.

3.2.4 Rolled Paper Towel Method

Seedling infection and seedling vigor test was done by following the Rolled Paper Towel Method (Warham, 1990). In this method, one hundred and fifty seeds were randomly taken from each treatment. 50 seeds were placed uniformly between a pair of moist paper towels. The towels were rolled and the two ends were closed with rubber bands. Then the rolled papers containing seeds were placed in an upright position for 7 days at room temperature under normal 12/12 light and darkness cycle. After incubation period, observations pertaining to germination, number of normal seedlings, number of abnormal seedlings and number of dead seeds were recorded. The normal and abnormal seedlings were categorized according to ISTA rules (1996). The normal seedlings were categorized by following points:

i) Intact seedling with all essential structures, well developed, complete in proportion of all structures and healthy.

ii) Seedlings with slight defects of their essential structures and otherwise satisfactory and balanced development comparable to that of intact seedling in the same test.

iii) Seedlings with secondary infection that would have fallen into categories i or ii but found infection by fungi or bacteria from sources other than the parent seed.

During recording the abnormalities of germinating seeds and seedlings, the following points were considered:

i) Seminal roots missing/stunted or broken and decayed due to primary infection.

ii) Coleoptile missing/split and deformed or bent over.

iii) Shoot system (the mesocotyl if developed) broken/decayed.

iv) Leaf missing/extending less than half-way up the coleoptile, shredded or deformed.

v) Seedling as a whole deformed spindly, discolored or decayed as a result of primary infection.

vi) Blackened dead or decayed seed

Twenty seedlings were randomly selected from each paper and their individual shoot and root length will be measured. The shoot and root portions were blotted dry with fine tissue paper and fresh weight will be taken. Vigor of the seedling was determined by the following formula (Baki and Anderson, 1972):

Vigor Index = (Mean of root length + Mean of shoot length) × % Seed germination

3.3 Analysis of Data

The collected data for different parameters were compiled and tabulated in proper form. Appropriate statistical analysis was made by MSTAT Computer Package program. The treatment means were compared by Duncan's Multiple Range Test (DMRT).

CHAPTER IV

RESULTS

4.1. Field Experiment

4.1.1. Effect of different levels of black pointed seed and vermicompost on disease incidence at 7 DAS of wheat in the field

Considering number of seedlings/m^2, the treatments differed significantly (Table 1 and plate 1). The values ranged from 89.0 to 114.3, where the highest number of seedlings /m^2 was found in T_7 followed by T_{13} (112.00) and T_8 (109.0) and the lowest seedlings /m^2 was recorded in T_6 preceded by T_5 (93.0) and T_4 (94.33).

4.1.2. Effect of different levels of black pointed seed and vermicompost on disease incidence at 15 DAS of wheat in the field

Considering number of seedlings/m^2, the treatments differed significantly (Table 1). The values ranged from 100.7 to 124.0, where the highest number of seedlings /m^2 was found in T_7 followed by T_8 (121.0) and T_{13} (120.7) and the lowest number of seedlings /m^2 was recorded in T_6 preceded by T_5 (102.0) and T_4 (104.3).

Table 1. Effect of different levels of black pointed seed and vermicompost on disease incidence at 7 DAS and 15 DAS

Treatment	Number of seedlings m^{-2}	
	7 DAS	15 DAS
T_1	102.3 c-f	110.7 b-h
T_2	100.7 d-h	109.7 c-h
T_3	96.00 f-i	107.0 e-h
T_4	94.33 g-i	104.3 f-h
T_5	93.00 hi	102.0 gh
T_6	89.00 i	100.7 h
T_7	114.3 a	124.0 a
T_8	109.0 a-c	121.0 ab
T_9	106.7 a-e	118.3 a-d
T_{10}	103.0 c-f	117.7 a-e
T_{11}	102.3 c-f	114.0 a-f
T_{12}	101.3 c-g	111.7 b-h
T_{13}	112.0 ab	120.7 a-c
T_{14}	108.3 a-d	117.3 a-e
T_{15}	105.3 b-e	115.3 a-f
T_{16}	104.0 c-e	112.0 b-g
T_{17}	101.0 d-g	110.7 b-h
T_{18}	100.3 e-h	107.7 d-h
LSD $_{(0.05)}$	7.77	11.04
CV (%)	4.57	5.92

T_1 = (Apparently healthy seed treated with Provax 200 @ 0.25%); T_2 = (5 % black pointed seed in weight basis); T_3 = (10 % black pointed seed); T_4 = (15 % black pointed seed); T_5 = (20 % black pointed seed) ; T_6 = (25 % black pointed seed); T_7 = (Vermicompost @ 1.5 ton/ha + T_1); T_8 = (Vermicompost @ 1.5 ton/ha + T_2); T_9 = (Vermicompost @ 1.5 ton/ha + T_3); T_{10} = (Vermicompost @ 1.5 ton/ha + T_4); T_{11} = (Vermicompost @ 1.5 ton/ha + T_5); T_{12} = (Vermicompost @ 1.5 ton/ha + T_6); T_{13} = (Vermicompost @ 3.0 ton/ha + T_1); T_{14} = (Vermicompost @ 3.0 ton/ha + T_2); T_{15} = (Vermicompost @ 3.0 ton/ha + T_3); T_{16} = (Vermicompost @ 3.0 ton/ha + T_4); T_{17} = (Vermicompost @ 3.0 ton/ha + T_5); T_{18} = (Vermicompost @ 3.0 ton/ha + T_6)

Plate 1: A view of experimental field showing Wheat seedlings at 7 DAS.

4.1.3. Effect of different levels of black pointed seed and vermicompost on leaf blight severity (0-5) scale on Flag leaf of wheat at flag leaf stage

Significant variations were found in leaf blight severity on Flag leaf (Table 2). In case of flag leaf, the disease severity ranged from 0.00 to 5.33 %. The maximum (5.33 %) value was found in T_6 (5.33 %). On the contrary, T_7 and T_{13} showed minimum (0.00 %) disease severity.

4.1.4. Effect of different levels of black pointed seed and vermicompost on leaf blight severity (0-5) scale on Flag leaf of wheat at panicle initiation stage

Significant variations were found in leaf blight severity in panicle initiation stage on Flag leaf (Table 2). In case of flag leaf, the disease severity ranged from 1.00 to 10.00 %. The maximum (10.00 %) value was found in T_6. On the contrary, T_7 showed minimum (1.00 %) disease severity. Treatments T_7 (1.00 %) and T_{13} (1.67 %) showed statistically similar results.

4.1.5. Effect of different levels of black pointed seed and vermicompost on leaf blight severity (0-5) scale on Flag leaf of wheat at flowering stage

Significant variations were found in leaf blight severity in flowering stage on Flag leaf (Table 2). In case of flag leaf, the disease severity ranged from 2.67 to 14.00 %. The maximum (14.00 %) value was found in $T_{6.}$ On the other hand, the minimum (2.67 %) disease severity was found in Treatment T_7 which was statistically similar with T_{13} (3.33 %).

4.1.6. Effect of different levels of black pointed seed and vermicompost on leaf blight severity (0-5) scale on Flag leaf of wheat at milking stage

Significant variations were found in leaf blight severity in milking stage on Flag leaf (Table 2). In case of flag leaf, the disease severity ranged from 20.00 to 40.67 %. The maximum (40.67 %) value was found in T_6 (40.67 %) and which was statistically similar with T_5 (37.33 %) and T_4 (36.00 %). On the other hand, the minimum (20.00 %) disease severity was found in Treatment T_7 which was statistically similar with T_8 (24.67 %) and T_{13} (23.00 %).

4.1.7. Effect of different levels of black pointed seed and vermicompost on leaf blight severity (0-5) scale on Flag leaf of wheat at hard dough stage

Significant variations were found in leaf blight severity in hard dough stage on Flag leaf (Table 2). In case of flag leaf, the disease severity ranged from 32.67 to 58.67 %. The maximum (58.67 %) value was found in T_6 (58.67 %) which was statistically similar with T_5 (54.00 %). On the other hand, the minimum (32.67 %) disease severity was observed in Treatment T_7 which was statistically similar with T_8 (35.67 %) and T_{13} (36.00 %).

4.1.8. Effect of different levels of black pointed seed and vermicompost on leaf blight severity (0-5) scale on Penultimate leaf of wheat at flag leaf stage

Significant variations were found in leaf blight severity in flag leaf stage on Penultimate leaf (Table 3). In case of Penultimate leaf, the disease severity ranged from 2.00 to 7.33 %. The maximum (7.33 %) disease severity was observed in T_6 followed by T_{18} (6.00 %) On the contrary, T_7 showed the minimum (2.00 %) disease severity. T_8 (2.67 %) and T_9 (2.67 %) showed statistically similar results.

4.1.9. Effect of different levels of black pointed seed and vermicompost on leaf blight severity (0-5) scale on Penultimate leaf of wheat at Panicle initiation stage

Significant variations were found in leaf blight severity in panicle initiation stage on Penultimate leaf (Table 3).In case of Penultimate leaf, the disease severity ranged from 2.67 to 12.00 %. The maximum (12.00 %) disease severity was observed in Treatment T_6, T_{18} (10.00 %) and T_5 (9.67 %) were statistically similar. On the other hand, the minimum (2.67 %) disease severity was observed in T_7 statistically similar with by T_8 (3.00 %) and T_{13} (3.33 %).

4.1.10. Effect of different levels of black pointed seed and vermicompost on leaf blight severity (0-5) scale on Penultimate leaf of wheat at flowering stage

Significant variations were found in leaf blight severity at flowering stage on Penultimate leaf (Table 3). In case of Penultimate leaf, the disease severity ranged from 4.66 to 15.67 %. The maximum disease severity was (15.67 %) observed in Treatment T_6 followed by T_{18} (14.00 %) and T_5 (13.33 %). On the other hand, the minimum disease severity (4.66 %) was observed in T_7 which was statistically similar with T_8 (5.33 %) and T_{13} (5.33 %).

4.1.11. Effect of different levels of black pointed seed and vermicompost on leaf blight severity (0-5) scale on Penultimate leaf of wheat at milking stage

Significant variations were found in leaf blight severity at milking stage on Penultimate leaf (Table 3). In case of Penultimate leaf, the disease severity ranged from 25.67 to 44.67 %. The maximum disease severity was (44.67 %) observed in T_6 statistically similar with T_5 (41.33 %) and T_4 (40.00 %). On the other hand, the minimum disease severity (25.67 %) was observed in T_7 which was statistically similar with T_8 (28.33 %) and T_{13} (28.33 %).

4.1.12. Effect of different levels of black pointed seed and vermicompost on leaf blight severity (0-5) scale on Penultimate leaf of wheat at hard dough stage

Significant variations were found in leaf blight severity in hard dough stage on Penultimate leaf (Table 3). In case of Penultimate leaf, the disease severity ranged from 41.33 to 65.33 %. The maximum disease severity was (65.33 %) observed in T_6 which was statistically similar with T_5 (62.67 %). On the other hand, the minimum disease severity (41.33 %) was observed in T_7 which was statistically similar with T_8 (44.00 %).

Table 2. Effect of different levels of black pointed seed and vermicompost on leaf blight severity (0-5) scale on Flag leaf of wheat at flag leaf stage, panicle initiation stage, flowering stage, milking stage and hard dough stage

Treatment	Disease severity in different growth stages at Flag leaf (%)				
	Flag leaf stage	Panicle initiation stage	Flowering stage	Milking stage	Hard dough stage
T_1	0.67 i	2.33 hi	4.00 ij	28.67 e-h	41.33 e-g
T_2	1.33 h	3.67 g	6.33 fg	30.67 c-g	44.00 d-f
T_3	2.67 f	6.00 de	8.00 e	34.00 b-d	48.00 b-d
T_4	2.67 f	7.33 c	10.00 d	36.00 ab	50.67 bc
T_5	4.00 c	8.67 b	11.67 bc	37.33 ab	54.00 ab
T_6	5.33 a	10.00 a	14.00 a	40.67 a	58.67 a
T_7	0.00 j	1.00 j	2.67 k	20.00 j	32.67 h
T_8	0.67 i	1.67 ij	4.67 hi	24.67 h-j	35.67 gh
T_9	1.33 h	2.00 hi	5.33 gh	27.33 g-i	43.33 d-f
T_{10}	2.00 g	4.67 f	6.67 f	30.67c-g	41.33 e-g
T_{11}	2.00 g	5.33 ef	8.33 e	33.33 b-e	44.00 d-f
T_{12}	3.00 e	6.33 d	10.67 cd	34.67 bc	46.00 c-e
T_{13}	0.00 j	1.67 ij	3.33 jk	23.00 ij	36.00 gh
T_{14}	2.00 g	2.67 h	5.33 gh	27.67 f-i	39.33 fg
T_{15}	1.33 h	4.67 f	7.33 ef	29.33d-h	43.33 d-f
T_{16}	2.67 f	5.67 de	9.67 d	32.67b-f	46.67 c-e
T_{17}	3.33 d	6.00 de	10.00 d	35.33bc	48.67b-d
T_{18}	4.67 b	8.33 b	12.00 b	36.00ab	50.67bc
$LSD_{(0.05)}$	0.32	0.84	1.27	5.01	6.24
CV (%)	8.71	10.3	9.81	9.67	8.41

T_1 = (Apparently healthy seed treated with Provax 200 @ 0.25%); T_2 = (5 % black pointed seed in weight basis); T_3 = (10 % black pointed seed); T_4 = (15 % black pointed seed); T_5 = (20 % black pointed seed) ; T_6 = (25 % black pointed seed); T_7 = (Vermicompost @ 1.5 ton/ha + T_1); T_8 = (Vermicompost @ 1.5 ton/ha + T_2); T_9 = (Vermicompost @ 1.5 ton/ha + T_3); T_{10} = (Vermicompost @ 1.5 ton/ha + T_4); T_{11} = (Vermicompost @ 1.5 ton/ha + T_5); T_{12} = (Vermicompost @ 1.5 ton/ha + T_6); T_{13} = (Vermicompost @ 3.0 ton/ha + T_1); T_{14} = (Vermicompost @ 3.0 ton/ha + T_2); T_{15} = (Vermicompost @ 3.0 ton/ha + T_3); T_{16} = (Vermicompost @ 3.0 ton/ha + T_4); T_{17} = (Vermicompost @ 3.0 ton/ha + T_5); T_{18} = (Vermicompost @ 3.0 ton/ha + T_6)

Table 3. Effect of different levels of black pointed seed and vermicompost on leaf blight severity (0-5) scale on Penultimate leaf of wheat at flag leaf stage, panicle initiation stage, flowering stage, milking stage and hard dough stage

Treatment	Disease severity in different growth stages at Penultimate Leaf (%)				
	Flag leaf stage	Panicle initiation stage	Flowering stage	Milking stage	Hard dough stage
T_1	2.00 h	4.67 g	6.67 hi	32.67 d-g	52.67 c-f
T_2	4.00 e	6.33 f	8.00 g	34.67b-g	55.00 b-e
T_3	4.00 e	7.67 de	9.33 ef	36.00 b-f	58.00 a-d
T_4	4.00 e	8.00 cd	11.33 d	40.00 a-c	60.67 a-c
T_5	4.67 d	9.67 b	13.33 bc	41.33 ab	62.67 ab
T_6	7.33 a	12.00 a	15.67 a	44.67 a	65.33 a
T_7	2.00 h	2.67 h	4.66 k	25.67 h	41.33 g
T_8	2.67 g	3.00 h	5.33 jk	28.33 gh	44.00 fg
T_9	2.67 g	4.33 g	6.00 ij	32.00 e-h	46.67 e-g
T_{10}	3.33 f	6.33 f	7.33 gh	34.00 c-g	48.00 e-g
T_{11}	4.67 d	7.00 ef	9.67 e	36.00 b-f	50.67 d-g
T_{12}	5.33 c	8.67 c	11.33 d	37.33b-e	52.00 c-f
T_{13}	2.00 h	3.33 h	5.33 jk	28.33 gh	47.33 e-g
T_{14}	2.00 h	4.67 g	6.33 h-j	30.33 f-h	47.67 e-g
T_{15}	3.33 f	6.67 f	8.33 fg	33.67 c-g	49.67 d-g
T_{16}	4.67 d	7.67 de	10.00 e	35.67 b-f	52.33 c-f
T_{17}	5.33 c	8.67 c	12.67 c	37.00 b-f	54.00 b-e
T_{18}	6.00 b	10.00 b	14.00 b	39.33 a-d	58.00 a-d
LSD $_{(0.05)}$	0.52	0.99	1.13	6.83	9.62
CV (%)	8.15	8.87	7.44	11.82	11.03

T_1 = (Apparently healthy seed treated with Provax 200 @ 0.25%); T_2 = (5 % black pointed seed in weight basis); T_3 = (10 % black pointed seed); T_4 = (15 % black pointed seed); T_5 = (20 % black pointed seed) ; T_6 = (25 % black pointed seed); T_7 = (Vermicompost @ 1.5 ton/ha + T_1); T_8 = (Vermicompost @ 1.5 ton/ha + T_2); T_9 = (Vermicompost @ 1.5 ton/ha + T_3); T_{10} = (Vermicompost @ 1.5 ton/ha + T_4); T_{11} = (Vermicompost @ 1.5 ton/ha + T_5); T_{12} = (Vermicompost @ 1.5 ton/ha + T_6); T_{13} = (Vermicompost @ 3.0 ton/ha + T_1); T_{14} = (Vermicompost @ 3.0 ton/ha + T_2); T_{15} = (Vermicompost @ 3.0 ton/ha + T_3); T_{16} = (Vermicompost @ 3.0 ton/ha + T_4); T_{17} = (Vermicompost @ 3.0 ton/ha + T_5); T_{18} = (Vermicompost @ 3.0 ton/ha + T_6)

Plate 2: A view of experimental field at panicle initiation stage

Plate 3: A view of experimental field at hard dough stage

4.1.13. Effect of different levels of black pointed seed and vermicompost on Plant height and Spike length

The treatments were found to differ significantly in respect of plant height and spike length (Table 4).

The plant height ranged from 75.67 to 82.84 cm, where the highest plant height (82.84) was found in T_7 which was statistically similar with T_8 (82.55). The lowest plant height (75.67 cm) was found in T_6 which was statistically indifferent with T_5 (76.75)

In respect of spike length, the treatments showed significant variation. The spike length, ranged from 10.98 to 12.91 cm, where the highest spike length (12.91) was found in T_7 which was statistically similar with T_8 (12.72). The lowest spike length (10.98 cm) was found in T_6 which was indifferent with T_5 (11.54).

Table 4. Effect of different levels of black pointed seed and vermicompost on Plant height and Spike length

Treatment	Plant height (cm)	Spike length (cm)
T_1	79.72 a-c	12.28 a-c
T_2	78.48 a-c	12.24 a-c
T_3	77.99 a-c	11.85 a-d
T_4	77.82 a-c	11.72 b-d
T_5	76.75 bc	11.54 cd
T_6	75.67 c	10.98 d
T_7	82.84 a	12.91 a
T_8	82.55 a	12.72 ab
T_9	81.58 ab	12.54 a-c
T_{10}	81.44 ab	12.42 a-c
T_{11}	80.92 a-c	12.36 a-c
T_{12}	79.72 a-c	12.24 a-c
T_{13}	82.30 ab	12.61 a-c
T_{14}	81.53 ab	12.43 a-c
T_{15}	80.84 a-c	12.31 a-c
T_{16}	80.24 a-c	12.24 a-c
T_{17}	79.45 a-c	12.00 a-d
T_{18}	79.28 a-c	11.83 a-d
$LSD_{(0.05)}$	5.73	1.09
CV (%)	4.32	5.39

T_1 = (Apparently healthy seed treated with Provax 200 @ 0.25%); T_2 = (5 % black pointed seed in weight basis); T_3 = (10 % black pointed seed); T_4 = (15 % black pointed seed); T_5 = (20 % black pointed seed) ; T_6 = (25 % black pointed seed); T_7 = (Vermicompost @ 1.5 ton/ha + T_1); T_8 = (Vermicompost @ 1.5 ton/ha + T_2); T_9 = (Vermicompost @ 1.5 ton/ha + T_3); T_{10} = (Vermicompost @ 1.5 ton/ha + T_4); T_{11} = (Vermicompost @ 1.5 ton/ha + T_5); T_{12} = (Vermicompost @ 1.5 ton/ha + T_6); T_{13} = (Vermicompost @ 3.0 ton/ha + T_1); T_{14} = (Vermicompost @ 3.0 ton/ha + T_2); T_{15} = (Vermicompost @ 3.0 ton/ha + T_3); T_{16} = (Vermicompost @ 3.0 ton/ha + T_4); T_{17} = (Vermicompost @ 3.0 ton/ha + T_5); T_{18} = (Vermicompost @ 3.0 ton/ha + T_6)

4.1.14. Effect of different levels of black pointed seed and vermicompost on number of grains/ear of different severity grades (0-5) of harvested seeds of wheat

Grading of seeds was done in 0-5 rating scale where ‘o’ indicates apparently healthy seed ’5’ indicate seed with the maximum diseases symptom .Significant effects of the treatments were found on the formation of seeds under Grade -0 (Free from infection i.e. apparently healthy seeds), Grade-1 (only embryo blackish), Grade-2 (Embryo and its adjacent area slightly infected), Grade-3 (Embryo and less than ¼ of grains are discolored), Grade-4 (Embryo and ½ of grain are infected), Grade-5 (Grains are shriveled, almost completely discolored or more than ½ of grains were discolored).

The number of grains under grade-0 was found in a range of 34.27 to 45.50. The maximum (45.50) count of healthy seeds was observed in T_7 which was statistically similar with T_8 (44.13) and T_{13} (43.03). The minimum (34.27) count of healthy seeds was recorded in T_6 preceded by T_5 (34.83) and T_4 (35.00). The number of grains under grade-1 was found in a range of 3.23 to 3.73. The highest (3.73) number of seeds was observed in T_6. The minimum (3.23) number of seeds was recorded in T_7 preceded by T_8 (3.27).

The number of grains under grade-2 was found in a range of 0.12 to 0.45. The highest (0.45) number of seeds was observed in T_6 followed by T_{18} (0.42). The minimum (0.12) number of seeds was recorded in T_7 preceded by T_{13} (0.15). The number of grains under grade-3 was found in a range of 0.23 to 0.55. The highest (0.55) number of seeds was observed in T_6 and T_{18}. The minimum (0.23) number of seeds was recorded in T_7 and T_{13} (0.23) preceded by T_8 (0.30). The number of grains under grade-4 was found in a range of 0.45 to 0.73.The highest (0.73) number of seeds was observed in T_6 which was statistically similar with T_{18} (0.73) and T_5 (0.73). The lowest (0.45) number of seeds was recorded in T_7 which was statistically similar with T_{13} (0.47) and T_8 (0.50). The number of grains under grade-5 was found in a range of 0.73 to 1.13.The highest (1.13) number of seeds was observed in T_6 and T_5 (1.13) which was statistically similar with T_4 (1.07). The lowest (0.73) number of seeds was recorded in T_7 statistically similar with T_{13} (0.76).

Table 5. Effect of different levels of black pointed seed and vermicompost on number of grains/ear of different severity grades (0-5) of harvested seeds of wheat

Number of different Grade seeds						
Treatment	0	1	2	3	4	5
T_1	41.13 cd	3.40 c-f	0.19 fg	0.32 gh	0.53 gh	0.80 h-k
T_2	39.67 de	3.43 b-f	0.24 ef	0.36 fg	0.53 gh	0.87 f-i
T_3	37.00 f-h	3.57 a-c	0.23 ef	0.40 ef	0.60 ef	0.96 c-e
T_4	35.00 hi	3.57 a-c	0.37 b-d	0.46 cd	0.65 c-e	1.07 ab
T_5	34.83 hi	3.63 ab	0.40 a-c	0.50 a-c	0.73 a	1.13 a
T_6	34.27 i	3.73 a	0.45 a	0.55 a	0.73 a	1.13 a
T_7	45.50 a	3.23 f	0.12 h	0.23 i	0.45 i	0.73 k
T_8	44.13 ab	3.27 ef	0.19 fg	0.30 h	0.50 hi	0.80 i-k
T_9	42.33 bc	3.30 d-f	0.25 e	0.35 f-h	0.55 fg	0.83 g-j
T_{10}	41.50 cd	3.40 c-f	0.33 d	0.41 de	0.60 ef	0.88 e-i
T_{11}	39.17 d-f	3.49 b-e	0.36 cd	0.46 bc	0.66 cd	0.95 c-f
T_{12}	38.50 e-g	3.55 a-c	0.40 a-c	0.50 a-c	0.70 abc	0.98 b-d
T_{13}	43.03 a-c	3.30 d-f	0.15 gh	0.23 i	0.47 i	0.76 jk
T_{14}	41.13 cd	3.43 b-f	0.21 ef	0.32 gh	0.53 gh	0.82 h-k
T_{15}	39.62 de	3.46 b-f	0.26 e	0.39 ef	0.55 fg	0.89 e-h
T_{16}	37.26 e-h	3.51 a-d	0.33 d	0.48 bc	0.62 de	0.92 d-g
T_{17}	36.50 g-i	3.63 ab	0.38 b-d	0.51 ab	0.68 bc	0.97 c-e
T_{18}	35.29 hi	3.66 ab	0.42 ab	0.55 a	0.73 ab	1.03 bc
LSD $_{(0.05)}$	2.50	0.23	0.05	0.05	0.05	0.09
CV (%)	3.85	3.95	6.24	5.28	4.73	6.38

T_1 = (Apparently healthy seed treated with Provax 200 @ 0.25%); T_2 = (5 % black pointed seed in weight basis); T_3 = (10 % black pointed seed); T_4 = (15 % black pointed seed); T_5 = (20 % black pointed seed) ; T_6 = (25 % black pointed seed); T_7 = (Vermicompost @ 1.5 ton/ha + T_1); T_8 = (Vermicompost @ 1.5 ton/ha + T_2); T_9 = (Vermicompost @ 1.5 ton/ha + T_3); T_{10} = (Vermicompost @ 1.5 ton/ha + T_4); T_{11} = (Vermicompost @ 1.5 ton/ha + T_5); T_{12} = (Vermicompost @ 1.5 ton/ha + T_6); T_{13} = (Vermicompost @ 3.0 ton/ha + T_1); T_{14} = (Vermicompost @ 3.0 ton/ha + T_2); T_{15} = (Vermicompost @ 3.0 ton/ha + T_3); T_{16} = (Vermicompost @ 3.0 ton/ha + T_4); T_{17} = (Vermicompost @ 3.0 ton/ha + T_5); T_{18} = (Vermicompost @ 3.0 ton/ha + T_6)

4.1.15. Effect of different levels of black pointed seed and vermicompost on weight of grains/ear of different severity grades (0-5) of harvested seeds of wheat

The weight of grains under grade-0 was found in a range of 1.71 to 2.28.The highest (2.28) weight of seeds was observed in T_7 which was statistically similar with T_8 (2.21). The minimum (3.23) weight of seeds was recorded in T_6 which was statistically similar with T_5 (1.75).

No significant variation was observed among the treatments in respect of grade-1, grade-2, grade-3, grade-4 and grade-5 seeds.

Table 6. Effect of different levels of black pointed seed and vermicompost on weight of grains/ear of different severity grades (0-5) of harvested seeds of wheat

Weight of different Grade seeds						
Treatment	0	1	2	3	4	5
T_1	2.06 b-d	0.17	0.01	0.02	0.03	0.04
T_2	1.97 c-f	0.17	0.01	0.02	0.03	0.04
T_3	1.86 e-i	0.18	0.01	0.02	0.03	0.05
T_4	1.76 g-i	0.18	0.02	0.02	0.03	0.05
T_5	1.75 hi	0.18	0.02	0.03	0.04	0.06
T_6	1.71 i	0.19	0.02	0.03	0.04	0.06
T_7	2.28 a	0.16	0.01	0.01	0.02	0.04
T_8	2.21 ab	0.16	0.01	0.02	0.02	0.04
T_9	2.12 a-d	0.17	0.01	0.02	0.03	0.04
T_{10}	2.10 a-d	0.17	0.01	0.02	0.03	0.04
T_{11}	1.96 c-g	0.17	0.02	0.02	0.03	0.05
T_{12}	1.92 d-h	0.18	0.02	0.02	0.04	0.05
T_{13}	2.15 a-c	0.17	0.01	0.01	0.02	0.04
T_{14}	2.06 b-e	0.17	0.01	0.02	0.03	0.04
T_{15}	1.99 c-f	0.17	0.01	0.02	0.03	0.04
T_{16}	1.86 e-i	0.18	0.02	0.02	0.03	0.05
T_{17}	1.83 f-i	0.18	0.01	0.03	0.03	0.05
T_{18}	1.76 g-i	0.18	0.02	0.03	0.04	0.05
LSD $_{(0.05)}$	0.2032	NS	NS	NS	NS	NS
CV (%)	6.16	5.76	5.38	2.53	7.34	3.57

NS=Not Significant

T_1 = (Apparently healthy seed treated with Provax 200 @ 0.25%); T_2 = (5 % black pointed seed in weight basis); T_3 = (10 % black pointed seed); T_4 = (15 % black pointed seed); T_5 = (20 % black pointed seed) ; T_6 = (25 % black pointed seed); T_7 = (Vermicompost @ 1.5 ton/ha + T_1); T_8 = (Vermicompost @ 1.5 ton/ha + T_2); T_9 = (Vermicompost @ 1.5 ton/ha + T_3); T_{10} = (Vermicompost @ 1.5 ton/ha + T_4); T_{11} = (Vermicompost @ 1.5 ton/ha + T_5); T_{12} = (Vermicompost @ 1.5 ton/ha + T_6); T_{13} = (Vermicompost @ 3.0 ton/ha + T_1); T_{14} = (Vermicompost @ 3.0 ton/ha + T_2); T_{15} = (Vermicompost @ 3.0 ton/ha + T_3); T_{16} = (Vermicompost @ 3.0 ton/ha + T_4); T_{17} = (Vermicompost @ 3.0 ton/ha + T_5); T_{18} = (Vermicompost @ 3.0 ton/ha + T_6)

4.1.16. Effect of different levels of black pointed seed and vermicompost on 1000 seeds weight and yield of wheat

Significant variations were observed among the treatments in respect of 1000 seeds weight, grain yield and straw yield of wheat.

The 1000 seeds weight (g) of wheat ranged from 37.42 to 42.70g. The highest (42.70) weight of 1000 seeds was found in T_7 which was statistically similar with T_{13} which provides 41.91g weight of 1000 seeds. The lowest (37.42) weight of 1000 seeds was found in T_6 which is statistically similar with T_5 which provide 38.02 g weight of 1000 seeds.

Considering grain yield (t/ha) ranged from 2.33 to 3.66.The highest (3.66) grain yield (t/ha) was found in T_7 statistically similar with $T_{8,}$ which provide 3.54. The lowest (2.33) grain yield (t/ha) was found in T_6 which was statistically similar with T_5 which provide 2.48 (t/ha).

Considering straw yield (t/ha) ranged from 3.05 to 4.33.The highest (4.33) straw yield (t/ha) was found in T_7 statistically similar with T_8 and T_{13} which provide 4.21 and 4.13 (t/ha). The lowest (3.05) straw yield (t/ha) was found in T_6 statistically similar with T_5 which provide 3.22 (t/ha).

Table 7. Effect of different levels of black pointed seed and vermicompost on 1000 seeds weight and yield of wheat

Treatment	1000 seed weight (g)	Grain yield (t ha^{-1})	Straw yield (t ha^{-1})
T_1	41.13 a-d	3.20 c-e	3.93 a-d
T_2	40.34 b-e	3.03 d-f	3.83 b-e
T_3	39.85 b-f	2.94 e-g	3.68 c-e
T_4	38.98 d-g	2.68 gh	3.45 e-g
T_5	38.02 fg	2.48 hi	3.22 fg
T_6	37.42 g	2.33 i	3.05 g
T_7	42.70 a	3.66 a	4.33 a
T_8	41.81 a-c	3.54 ab	4.21 ab
T_9	41.09 a-d	3.32 b-d	4.07 a-c
T_{10}	40.71 a-e	3.21 c-e	3.96 a-d
T_{11}	40.05 b-f	3.17 de	3.84 b-e
T_{12}	39.56 c-g	2.90 e-g	3.63 c-f
T_{13}	41.92 ab	3.50 a-c	4.13 ab
T_{14}	41.22 a-d	3.28 b-d	4.02 a-d
T_{15}	40.62 a-e	3.14 de	3.91 a-d
T_{16}	39.89 b-f	3.02 d-f	3.79 b-e
T_{17}	39.04 d-g	2.93 e-g	3.60 d-f
T_{18}	38.65 e-g	2.74 f-h	3.46 e-g
$LSD_{(0.05)}$	2.28	0.32	0.45
CV (%)	3.43	6.24	7.14

T_1 = (Apparently healthy seed treated with Provax 200 @ 0.25%); T_2 = (5 % black pointed seed in weight basis); T_3 = (10 % black pointed seed); T_4 = (15 % black pointed seed); T_5 = (20 % black pointed seed) ; T_6 = (25 % black pointed seed); T_7 = (Vermicompost @ 1.5 ton/ha + T_1); T_8 = (Vermicompost @ 1.5 ton/ha + T_2); T_9 = (Vermicompost @ 1.5 ton/ha + T_3); T_{10} = (Vermicompost @ 1.5 ton/ha + T_4); T_{11} = (Vermicompost @ 1.5 ton/ha + T_5); T_{12} = (Vermicompost @ 1.5 ton/ha + T_6); T_{13} = (Vermicompost @ 3.0 ton/ha + T_1); T_{14} = (Vermicompost @ 3.0 ton/ha + T_2); T_{15} = (Vermicompost @ 3.0 ton/ha + T_3); T_{16} = (Vermicompost @ 3.0 ton/ha + T_4); T_{17} = (Vermicompost @ 3.0 ton/ha + T_5); T_{18} = (Vermicompost @ 3.0 ton/ha + T_6)

4.2. Laboratory Experiment

4.2.1. Effect of different levels of black pointed seed and vermicompost on germination and incidence of *Bipolaris sorokiniana* of harvested seeds of wheat after harvesting (Blotter method)

In blotter method, the effect of the treatments was significantly different regarding germination percentage and incidence of *Bipolaris sorokiniana* of wheat. The maximum seed germination (93.33%) was found in T_7 statistically similar with T_{13} (91.33%). On the other hand, the lowest seed germination (65.33%) was recorded in T_6 statistically similar with T_{18} (70.33 %).

Bipolaris sorokiniana ranged from 8.667 to 53.67 % where the treatment T_7 (8.667%) was found to have lowest infection preceded by T_{13} (11.33%) and T_1 (13.33%). On the other hand, the highest incidence (53.67%) was recorded in T_6 statistically similar with T_{18} (50.33%).

Table 8. Effect of different levels of black pointed seed and vermicompost on germination and incidence of *Bipolaris sorokiniana* of harvested seeds of wheat after harvesting (Blotter method)

Treatment	Blotter Method	
	% Germination	% *Bipolaris sorokiniana*
T_1	88.33 a-d	13.33 m-o
T_2	84.67 a-e	18. 67 kl
T_3	80.33 c-g	25.33 ij
T_4	75.00 e-h	35.00 fg
T_5	71.67 f-h	43.33 cd
T_6	65.33 h	53.67 a
T_7	93.33 a	8.667 o
T_8	90.67 a-c	14.67 l-n
T_9	86.00 a-d	19.00 kl
T_{10}	81.00 b-g	28.33 hi
T_{11}	77.67 d-g	37.67 ef
T_{12}	72.33 f-h	46.33 bc
T_{13}	91.33 ab	11.33 no
T_{14}	87.67 a-d	16.67 lm
T_{15}	82.33 b-f	22.67 jk
T_{16}	78.67 d-g	31.00 gh
T_{17}	73.00 f-h	40.33 de
T_{18}	70.33 gh	50.33 ab
LSD $_{(0.01)}$	10.99	5.094
CV (%)	6.14	8

T_1 = (Apparently healthy seed treated with Provax 200 @ 0.25%); T_2 = (5 % black pointed seed in weight basis); T_3 = (10 % black pointed seed); T_4 = (15 % black pointed seed); T_5 = (20 % black pointed seed) ; T_6 = (25 % black pointed seed); T_7 = (Vermicompost @ 1.5 ton/ha + T_1); T_8 = (Vermicompost @ 1.5 ton/ha + T_2); T_9 = (Vermicompost @ 1.5 ton/ha + T_3); T_{10} = (Vermicompost @ 1.5 ton/ha + T_4); T_{11} = (Vermicompost @ 1.5 ton/ha + T_5); T_{12} = (Vermicompost @ 1.5 ton/ha + T_6); T_{13} = (Vermicompost @ 3.0 ton/ha + T_1); T_{14} = (Vermicompost @ 3.0 ton/ha + T_2); T_{15} = (Vermicompost @ 3.0 ton/ha + T_3); T_{16} = (Vermicompost @ 3.0 ton/ha + T_4); T_{17} = (Vermicompost @ 3.0 ton/ha + T_5); T_{18} = (Vermicompost @ 3.0 ton/ha + T_6)

Plate 4: Healthy seeds on blotter method

Plate 5: Infected seeds on blotter method

4.2.2. Effect of different levels of black pointed seed and vermicompost on germination %, normal and abnormal seedling%, dead seed % of wheat after harvesting (Rolled Paper towel method)

Wheat seeds samples having different levels of black pointed seed and vermicompost had significant effect on seed germination, normal and abnormal seedling production and dead seed percentage of wheat in Rolled Paper towel method (Table 9 and plate 7, 8). The germination varied from 64.67 to 90.67 % .The highest germination (90.67%) was found in T_7 statistically similar with T_{13} (89.33%) and T_8 (88.33%) and. The lowest (64.67%) was found in T_6 statistically similar with T_{18} (68.67%) and T_5 (69.33%).

The treatment showed significant difference from one another regarding percent normal seedling and the results for all the treatments ranged from 82.00 to 37.33% ,where the maximum counts (82.00%) were found in T_7 statistically similar with T_8 (77.67%) and T_{13} (76.67%). The lowest (37.33%) was found in T_6 statistically similar with T_{18} (44.00%) and T_5 (45.00%).

The treatments showed significant differences from one another regarding percent abnormal seedlings .The number of abnormal seedlings was found to be increased with the increase of black pointed seeds. In case of abnormal seedlings, they ranged from 8.667 to 27.33% where the highest (27.33%) percent was observed in T_6 followed by T_{18} (24.67%). The lowest (8.667%) was found in T_7 statistically similar with T_8 (10.67%).

The treatments showed significant differences from one another regarding percent dead seed. The result varied from 35.33 to 9.33 %. Percent dead seed was found the minimum 9.33 in T_7 which was statistically similar with T_{13} (10.67%). The highest (35.33%) dead seed was found in T_6 followed by T_{18} (31.33%) and T_5 (30.67%)

Table 9. Effect of different levels of black pointed seed and vermicompost on germination %, normal and abnormal seedling%, dead seed %of wheat after harvesting (Rolled Paper towel method)

Treatment	Paper towel method			
	% Germination	% Normal seedling	% Abnormal seedling	% Dead seed
T_1	86.67 a-c	72.33 bc	14.33 g-i	13.33 jk
T_2	83.33 a-e	67.00 c-e	16.33 fg	16.67 gh
T_3	77.00 c-g	57.33 f-h	19.67 de	23.00 de
T_4	74.00 d-h	52.00 g-i	22.00 cd	24.67 d
T_5	69.33 gh	45.00 ij	24.33 bc	30.67 b
T_6	64.67 h	37.33 j	27.33 a	35.33 a
T_7	90.67 a	82.00 a	8.667 k	9.33 l
T_8	88.33 ab	77.67 ab	10.67 jk	11.67 j-l
T_9	83.67 a-d	71.00 b-d	13.33 hi	16.33 hi
T_{10}	79.00 b-g	63.33 d-f	15.67 gh	21.00 ef
T_{11}	76.33 c-g	58.00 fg	18.33 ef	24.67 d
T_{12}	70.33 f-h	50.00 g-i	20.33 de	29.67 bc
T_{13}	89.33 ab	76.67 ab	12.67 ij	10.67 kl
T_{14}	86.33 a-c	71.00 b-d	15.33 gh	13.67 ij
T_{15}	80.67 a-f	62.00 ef	18.67 ef	19.33 fg
T_{16}	76.00 c-g	56.00 f-h	20.00 de	24.00 d
T_{17}	72.33 e-h	49.00 hi	23.67 bc	27.67 c
T_{18}	68.67 gh	44.00 ij	24.67 b	31.33 b
$LSD_{(0.01)}$	11.26	8.52	2.417	2.914
CV (%)	6.44	6.33	6.01	6.17

T_1 = (Apparently healthy seed treated with Provax 200 @ 0.25%); T_2 = (5 % black pointed seed in weight basis); T_3 = (10 % black pointed seed); T_4 = (15 % black pointed seed); T_5 = (20 % black pointed seed) ; T_6 = (25 % black pointed seed); T_7 = (Vermicompost @ 1.5 ton/ha + T_1); T_8 = (Vermicompost @ 1.5 ton/ha + T_2); T_9 = (Vermicompost @ 1.5 ton/ha + T_3); T_{10} = (Vermicompost @ 1.5 ton/ha + T_4); T_{11} = (Vermicompost @ 1.5 ton/ha + T_5); T_{12} = (Vermicompost @ 1.5 ton/ha + T_6); T_{13} = (Vermicompost @ 3.0 ton/ha + T_1); T_{14} = (Vermicompost @ 3.0 ton/ha + T_2); T_{15} = (Vermicompost @ 3.0 ton/ha + T_3); T_{16} = (Vermicompost @ 3.0 ton/ha + T_4); T_{17} = (Vermicompost @ 3.0 ton/ha + T_5); T_{18} = (Vermicompost @ 3.0 ton/ha + T_6)

Plate 6: Rolled Paper towel method in laboratory

Plate 7: Seedlings of T_7 in rolled paper towel method

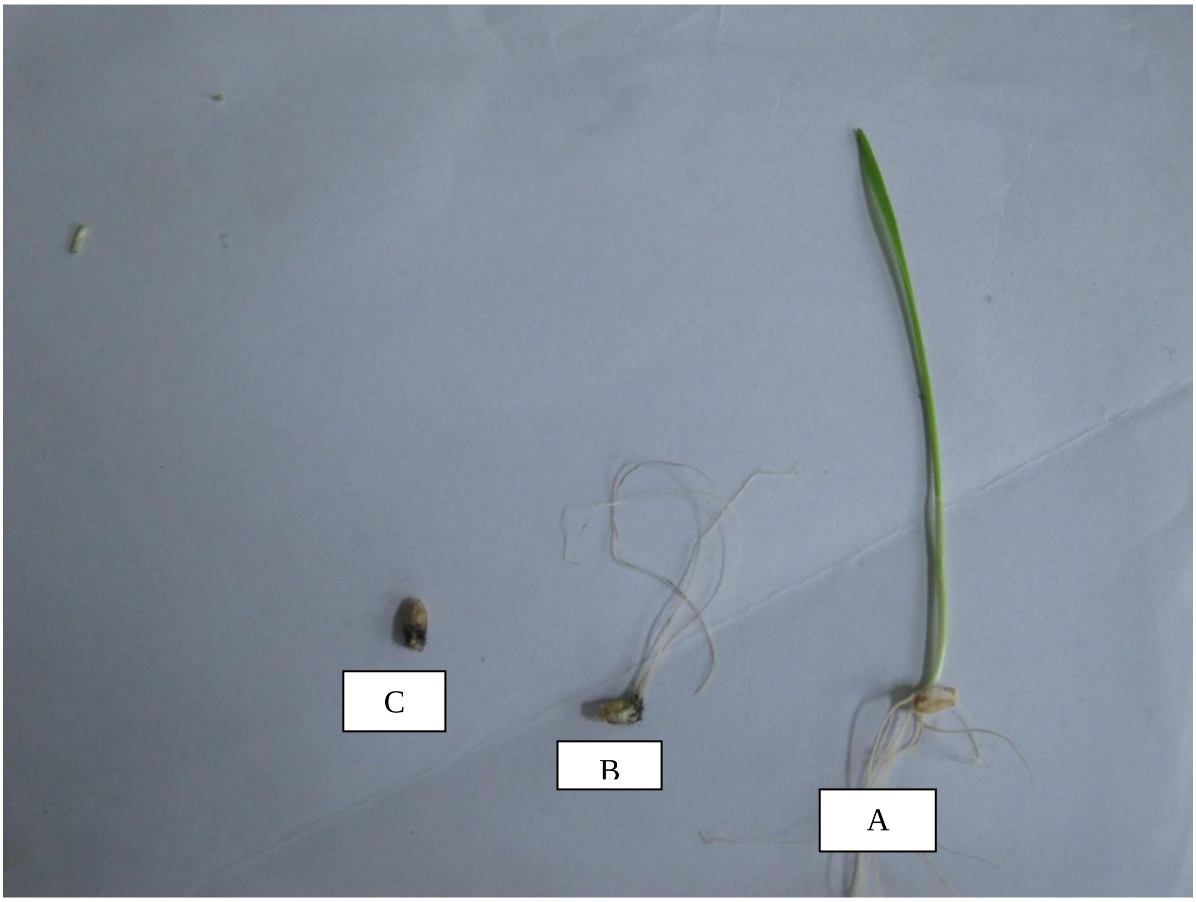


Plate 8: Normal (A), Abnormal (B) and dead seedling (C) in paper towel

4.2.3. Effect of different levels of black pointed seed and vermicompost on root length (cm), shoot length (cm), seedling weight and vigor index in the laboratory (Rolled Paper towel method)

The result showed that the treatments significantly influenced shoot length (cm) root length (cm), seedling weight and vigor index of 7 days old seedlings (Table 10). The root length varied from 11.13 to 13.77 cm, where the highest (13.77 cm) root length was found in T_7 statistically similar with T_{13} (13.53 cm). The minimum (11.13 cm) root length was found in T_6 which was statistically similar with T_5 (11.47 cm) as well as $T_{18.}$

In case of shoot length, The shoot length varied from 7.87 to 9.83 cm, where the highest (9.83 cm) shoot length was found in T_7 which was statistically similar with T_{13} (9.57 cm). The minimum (7.87 cm) shoot length was found in T_6 which was statistically similar with $T_{5.}$

Considering seedling weight, the values ranged from 3.43 to 4.30 g. The highest (4.30 g) seedling weight was found in T_7 statistically similar with T_8 (4.17). The minimum (3.43g) seedling weight was found in T_6 which was statistically similar with T_5 (3.53 g) as well as preceded by T_{18} (3.53) and T_{12} (3.57).

Vigor index (VI) for all the treatments differed significantly with a range of 1226 to 2157. The maximum vigor index (2157) was recorded in seedlings under T_7 which was statistically similar with T_{13} (2064). The minimum vigor index (1226) was counted in T_6 which was statistically similar with T_{18} (1346) and T_5 (1353).

Table 10. Effect of different levels of black pointed seed and vermicompost on root length (cm), shoot length (cm), seedling weight and vigor index in the laboratory (Rolled Paper towel method)

Treatment	Paper towel method			
	Root length (cm)	Shoot Length (cm)	Seedling weight (g)	Vigor index
T_1	13.10 a-d	9.23 a-d	4.03 a-d	1935 ab
T_2	12.73 a-f	8.97 b-e	3.92 a-e	1806 bc
T_3	12.37 c-g	8.73 c-g	3.80 b-f	1626 c-f
T_4	11.87 e-h	8.33 e-h	3.67 d-f	1494 e-g
T_5	11.47 gh	8.07 gh	3.53 ef	1353 gh
T_6	11.13 h	7.87 h	3.43 f	1226 h
T_7	13.77 a	9.83 a	4.30 a	2157 a
T_8	13.03 a-d	9.43 a-c	4.17 ab	1984 ab
T_9	12.83 a-f	9.03 b-e	4.03 a-d	1829 bc
T_{10}	12.53 b-g	8.83 b-f	3.93 a-e	1687 c-e
T_{11}	12.13 d-h	8.50 d-h	3.73 c-f	1574 d-g
T_{12}	11.77 f-h	8.40 e-h	3.57 ef	1418 f-h
T_{13}	13.53 ab	9.57 ab	4.13 a-c	2064 a
T_{14}	13.23 a-c	9.23 a-d	4.03 a-d	1940 ab
T_{15}	12.93 a-e	8.90 b-e	3.93 a-e	1761 b-d
T_{16}	12.53 b-g	8.73 c-g	3.83 b-f	1616 c-f
T_{17}	11.93 e-h	8.33 e-h	3.67 d-f	1463 e-g
T_{18}	11.47 gh	8.13 f-h	3.53 ef	1346 gh
$LSD_{(0.01)}$	1.10	0.75	0.41	231.10
CV (%)	3.97	3.85	4.77	6.19

T_1 = (Apparently healthy seed treated with Provax 200 @ 0.25%); T_2 = (5 % black pointed seed in weight basis); T_3 = (10 % black pointed seed); T_4 = (15 % black pointed seed); T_5 = (20 % black pointed seed) ; T_6 = (25 % black pointed seed); T_7 = (Vermicompost @ 1.5 ton/ha + T_1); T_8 = (Vermicompost @ 1.5 ton/ha + T_2); T_9 = (Vermicompost @ 1.5 ton/ha + T_3); T_{10} = (Vermicompost @ 1.5 ton/ha + T_4); T_{11} = (Vermicompost @ 1.5 ton/ha + T_5); T_{12} = (Vermicompost @ 1.5 ton/ha + T_6); T_{13} = (Vermicompost @ 3.0 ton/ha + T_1); T_{14} = (Vermicompost @ 3.0 ton/ha + T_2); T_{15} = (Vermicompost @ 3.0 ton/ha + T_3); T_{16} = (Vermicompost @ 3.0 ton/ha + T_4); T_{17} = (Vermicompost @ 3.0 ton/ha + T_5); T_{18} = (Vermicompost @ 3.0 ton/ha + T_6)

CHAPTER V

DISCUSSION

5.1. Field Experiment

Under in vivo condition, it was observed that the number of seedling/m^2 after 7 days and 15 days of sowing were significantly varied among the treatments in both observations. The number of seedling/m^2 after 7 days of sowing, the maximum number of seedlings/m^2 (114.3) was found in apparently healthy seeds (T_7) while the minimum number of seedlings/m^2 (89.00) was recorded in T_6. After 15 days of sowing, the maximum number of seedlings/m^2 (124.0) was found in apparently healthy seeds (T_7) while the minimum number of seedlings/m^2 (100) was recorded in T_6.

The present result is supported by Hossain (2000) who reported that black point infection greatly affected seed germination and seedling emergence of wheat and percent reduction in germination become higher with the increase level of black pointed seed. These findings were also supported by other researchers (Machacek and Grane, 1938). Machacek and Graney (1938) who reported that seed infected with *Helminthosporium sativum* produced only 24.8 % plant stand and resulted 80.6% seedling infection.

Considering main effect of different levels of black pointed seed, it has been found that leaf blight severity (0-5 scale) on flag leaf and penultimate leaf in different growth stage viz. flag leaf stage, panicle initiation stage, flowering stage, milking stage and hard dough stage were gradually increased with increasing levels of seed infection. The minimum leaf blight severity was recorded in the plots of T_7 (Vermicompost @ 1.5 ton/ha and apparently healthy seed treated with Provax 200 @ 0.25%) in every growth stages. The maximum leaf blight severity was recorded in the plots of T_6 (without vermicompost and 25 % black pointed seed in weight basis) in every growth stages. The disease severity was found to increase with the age of plant and the maximum disease severity was observed in hard dough stages in all treatments than the other stages. Leaf blight development is a usual consequence of the seed to plant to seed transmission of the pathogen *Bipolaris sorokiniana* under field condition (Rashid, 1996; Rashid and Fakir, 1998). Moreover, higher the level of seed borne fungal infection, there should be higher primary inoculums level in the field. So higher level of black pointed seed resulted with higher infection in the field in the present experiment.

The present findings were well supported by previous researchers (Nema and Joshi, 1974; Hossain and Azad, 1992; Malakar, 2003; Hossain, 2000; Reza, *et al.* 2006). Nema and Joshi (1974) reported that age was one of the important factor influencing disease intensity and susceptibility of wheat plant to *H. sativum*. Hossain and Azad (1992) reported that age of crop plant resulted higher incidence of leaf spot caused by *B. sorokiniana*. Malakar (2003) found that different grade of black point affected seeds caused significant variation in leaf blight development in adult plant. Reza *et al.* (2001) also reported that blight severity in adult plants of wheat increased with the increase in level of *Bipolaris sorokiniana* infected seeds. Temperature is also is an important factor for leaf blight incidence. Hossain (2000) reported that temperature 25-28^{0}C normally prevails in March when wheat plant turns to soft dough to hard dough stage. Therefore, maximum leaf blight disease incidence occurs at that time. The result of the present investigation is also supported by Hossain (2000) who found that significant leaf severity of wheat at flowering and milk ripening stages under field condition when different black point infected seeds were sown.

The effect of different levels of black pointed seed was highly significant in respect of plant height and spike length. The highest plant height (82.84 cm) and spike length (12.91cm) were recorded in T_7 (Vermicompost @ 1.5 ton/ha and apparently healthy seed treated with Provax 200 @ 0.25%). The minimum plant height (75.67 cm) and spike length (10.98cm) were observed in T_6 (without vermicompost and 25 % black pointed seed in weight basis).

The effect of plant population density on plant growth was reported by Das (2002) who mentioned that with the density treatments, a progressive increase in plant height was noted. Partially similar opinion was given by Roy and Biswas (1991); Mozumder (2001) and Pandey *et al*., (2004). They reported that there was no significant different among the seed rate in respect of plant height. Roy and Biswas (1991) and Fazil *et al., (*2004) reported that the spikelet formation i.e. number of spikelets/ear, number of healthy spikelets/ear and number of diseased spikelets / ear. The maximum black pointed seed attained the lowest number of spikelets / ear and healthy spikelets / ear but the highest number of diseased spikelets / ear was increased.

The present result was not supported by Nalli (1986). He found that *Bipolaris sorokinina* inoculated seeds produced tiller of lower height with induced number of seeds. On the other hand Hossain (2000) found that plant height and spike length did not varied significantly when different levels of black pointed seed were used for sowing.

Edwards and Burrows (1988) reported that vermicompost consistently improved seed germination, enhanced seedling growth and development, and increased plant productivity significantly. Application of vermicompost in potted and field crops displayed excellent growth performances in terms of height of plants, colour and texture of leaves, appearance of fruiting structures etc.as compared to chemical fertilizers.

Considering number of grains/ear of different severity grades (0-5) of harvested seeds, the treatments showed significant variations Treatment T_7 (Vermicompost @ 1.5 ton/ha and apparently healthy seed treated with Provax 200 @ 0.25%) attained the highest amount of seeds under Grade - 0 (Free from infection i.e. apparently healthy seeds), Grade - 1(only embryo blackish), Grade- 2(Embryo and its adjacent area slightly infected), Grade- 3 (Embryo and less than ¼ of grains are discolored), Grade- 4 (Embryo and ½ of grain are infected), Grade- 5 (Grains are shriveled, almost completely discolored or more than ½ of grains were discolored). The findings revealed that with the higher levels of black pointed seed resulted the higher number of harvested seeds with black point.

The treatments showed significant variations weight of grains/ear of different severity grades (0-5) of harvested seeds, in respect of Treatment T_7 (Vermicompost @ 1.5 ton/ha and apparently healthy seed treated with Provax 200 @ 0.25%) attained the highest weight of seeds under Grade - 0 (Free from infection i.e. apparently healthy seeds), No significant effect of the treatments were found on Grade - 1 seeds (only embryo blackish), Grade- 2 seeds (Embryo and its adjacent area slightly infected), Grade- 3 seeds (Embryo and less than ¼ of grains are discolored), Grade- 4 seeds (Embryo and ½ of grain are infected) and Grade- 5 seeds (Grains are shriveled, almost completely discolored or more than ½ of grains were discolored).

These findings were well supported by Rashid, (1996); Rashid and Fakir, (1998); who reported that development of black point infection in the field was due to seed to plant to seed transmission of black point pathogen. Hossain (1998) observed that leaf infection at flowering and milking stages has direct effect on the reduction of formation of healthy grains with the increase in number of black pointed as well as discolored grains. Hossain (2000) also reported significant relationship of leaf blight severity with grain infection. Reza *et al.*, (2006) reported that 65.36 % percent disease severity the corresponding 17.42 percent seed infection.

Significant variations in respect of 1000 seeds weight and both grain and straw yield indicated the significant effect of different levels of seed infection. The highest 1000 seeds weight was found in the plots under Treatment T_7 (Vermicompost @ 1.5 ton/ha and apparently healthy seed treated with Provax 200 @ 0.25%) used and the lowest in the plots under Treatment T_6 (without vermicompost and 25 % black pointed seed in weight basis). Grain yield as well as straw yield were found in maximum in the Treatment T_7 and the minimum yields were obtained in T_6 having maximum black pointed seed in weight basis.

Malakar (2003) reported that there was a decreasing trend in yield and yield contributing characters with increasing in severity of black point infected seeds. Yield reduction due to sowing of black pointed seeds has also been reported by the other workers (Parashar and Chohan 1967); Zwatz, 1975 and Nalli, 1986. Ahmed and Hossain (2005) found the highest 1000 grain weight and the highest yield (3.63 t/ha) in wheat variety kanchan when sprayed with fungicide Tilt-250 EC. Ferdous *et al.*, (2003) found grain yield of wheat was 2880-4000 kg/ha by using different micronutrients in the field. The results are in accordance with Oppitz and Hoesser (1979) who reported that seed borne pathogens of wheat not only reduced the germination but also affected seedling vigor that resulting in low yield.

Baker and Barrett (1994) found that the earthworms (*Aporrectodea trapezoids*) increased growth of wheat crops by 39% and grain yield by 35%. It also reducedcrop diseases as compared to the control. Palanisamy (1996) also reported that earth worms and its vermicompost improve the growth and yield of wheat by more than 40%. Bhatia *et al*., (2000) also studied impact of vermicompost on potted wheat crops and came with very encouraging results.

5.2. Laboratory Experiment on seed health status

Significant variations were observed among the treatments in respect of seed germination in blotter method and rolled paper towel method test. It was evident that the maximum seed germination was found in seed sample which had lowest black pointed seed in the Treatment $T_{7.}$ With the increase of black pointed seed, remarkable reduction of the seed germination was recorded and the lowest germination was found in seeds with the maximum black pointed seed in Treatment T_6. The minimum germination respectively was 65.33 % and 64.67 % in blotter method and rolled paper towel method found in the Treatment T_6.

The present results are well supported by the other researchers (Hanson and Christensen, 1953; Choudhary *et al*., 1984; Khanum *et al*., 1987; Hossain, 2000). Similar trend of variation in germination of *Helminthosporium sativum* infected wheat seeds were reported by Hanson and Christensen (1953). They reported 66% and 62% seed germination having seed infection 81% and 74%, respectively with *Helminthosporium sativum.* Choudhary *et., al*. (1984) reported that germination of the infected (black pointed) seeds both in blotter and pot soil was found to decrease by 11.6% and 16.0%, respectively. Khanum *et al., (*1987) found 55-96% and 34.5-71% germination for healthy grains and diseased grains, respectively. Hossain (2000) reported that maximum reduction of germination was found by 20.20 and 42.69% in blotter and rolled paper towel method, respectively in 28% black pointed seeds. Reduction in germination of wheat seeds due to black point infection was also recorded by other workers (Parashar and Chohan, 1967; Rana and Gupta, 1982; Sinha and Thapliyal 1984, Zhang *et al*., 1990). Chowdhury *et al.* (2010) reported that germination of wheat seeds having different levels of black pointed seed ranged 69.00-97.00 and 72.67-96.67 respectively in blotter method and rolled paper towel method depending on level of seed infection in wheat seeds.

Malakar (2003) found abnormal seedlings and dead seeds increased with the increase inoculums of *B. sorokiniana*. Siddique (2003) reported that the highest normal seedlings and lowest abnormal seedlings were found in clean seeds. The prevalence of higher number of black pointed seeds might be responsible for causing seed rot.

From the present study, it was revealed that different levels of black pointed seed by *Bipolaris sorokiniana* had significant relationship with seedling infection as well as seedling health. Seedling infection increased with the increasing level of black pointed seed in rolled paper towel method. The shoot length, root length, seedling weight and also vigor index (VI) were decreased with the increasing levels of black pointed seed.

The findings of the present study are supported by the earlier reports (Rana and Gupta, 1982; Rahman and Islam, 1998; Rashid and Fakir, 1998; Hossain, 2000). Rana and Gupta (1982) found that black point infection greatly affected root and shoot growth of the seedlings, the effect being very prominent on root growth. Rahman and Islam (1998) observed significant reduction in seedling vigor in respect of germination, shoot and root length with the increase of black point infection. Rashid and Fakir (1998) reported that percent reduction in shoot and root length increased with the increase of infection grade of seed transmitted *B.sorokiniana* and the overall reductions were highest for root length. He also mentioned that the seedlings that developed from such seed were usually poor vigorous. Hossain (2000) found that the rate of reduction of growth was the maximum by 28% black pointed seeds as recorded root length was 57.21 cm and for shoot length was 41.40 cm. He also mentioned that the vigor index was found with maximum reduction (72.63%) resulted by the seedlings of 28% black pointed seeds. Chowdhury *et al.* (2010) found that different levels of black pointed seed greatly affected root and shoot growth of wheat seedlings where as vigor index of seedling was 1851.81, 1392.02, 1203.55, 971.16, 841.62 and 669.36.

CHAPTER VI

SUMMARY AND CONCLUSION

Wheat (*Triticum aestivum* L.) is the most important cereal crop of Bangladesh andgrown during winter from November-December to March-April. In Bangladesh, wheat is considered as the second most important cereal crop. The present study was conducted in the Molecular Plant Pathology Laboratory and in the field allotted for the Department of Plant Pathology, Sher-e-Bangla Agricultural University, Dhaka to find out the effect of different levels of black pointed seed and vermicompost on leaf blight (*Bipolaris sorokiniana)* severity, yield attributes and seed health status of wheat during the period of November 2013 to July 2014.

Under field condition, experiment was carried out in RCBD design and there were 18 treatments. Here, significant variations were found considering number of seedlings/m^2. The maximum (114.3-124.0) emergence of seedling was recorded in T_7 (Vermicompost @ 1.5 ton/ha and apparently healthy seed treated with Provax 200 @ 0.25%) and the minimum (89.0-100.0) was in T_6 (without vermicompost and 25 % black pointed seed in weight basis) after 7 days and 15 days of sowing indicating general tendency of decreasing seedling emergence with the maximum use of black pointed seed.

In case of main effect of different levels of black pointed seed it has been found that leaf blight severity on flag leaf and penultimate leaf in flag leaf stage, panicle initiation stage, flowering stage, milking stage and hard dough stage was gradually increased with increasing levels of black pointed seed. The minimum leaf blight severity was recorded in the plots of T_7 (Vermicompost @ 1.5 ton/ha and apparently healthy seed treated with Provax 200 @ 0.25%) in every growth stages recorded. The maximum leaf blight severity was recorded in the plots of T_6 (without vermicompost and 25 % black pointed seed in weight basis) in every growth stages.

The effect of different level of black pointed seed was highly significant in respect of plant height and spike length. The highest plant height (82.84 cm) and spike length (12.91cm) were recorded in T_7 (Vermicompost @ 1.5 ton/ha and apparently healthy seed treated with Provax 200 @ 0.25%). The minimum plant height (75.67) and spike length (10.98cm) were observed in T_6 (without vermicompost and 25 % black pointed seed in weight basis).

It was found that the treatments had significant effect on grain number of different severity grades (0-5) of harvested seeds. The minimum number of grains/ear was found in T_6 (without vermicompost and 25 % black pointed seed in weight basis) and the maximum was found in T_7 (Vermicompost @ 1.5 ton/ha and apparently healthy seed treated with Provax 200 @ 0.25%).

The minimum weight of grains/ear was found in T_6 and the maximum found in $T_{7.}$ Considering weight of grains/ear of different severity grades (0-5) of harvested seeds, the treatments showed significant variations Treatment T_7 (Vermicompost @ 1.5 ton/ha and apparently healthy seed treated with Provax 200 @ 0.25%) attained the highest weight of seeds under Grade -0 (Free from infection i.e. apparently healthy seeds). No significant effect of the treatments were found on Grade-1 seeds (only embryo blackish), Grade-2 seeds (Embryo and its adjacent area slightly infected), Grade-3 seeds (Embryo and less than ¼ of grains are discolored), Grade-4 seeds (Embryo and ½ of grain are infected), Grade-5 seeds (Grains are shriveled, almost completely discolored or more than ½ of grains were discolored).

Significant variations were also observed among the treatments in respect of 1000 seeds weight, grain yield and straw yield of wheat. The highest (42.70) weight of 1000 seeds was found in T_7 and the lowest (37.42) weight of 1000 seeds was found in $T_{6.}$ Considering grain yield (t/ha) ranged from 2.33 to 3.66.The highest (3.66) grain yield (t/ha) was found in T_7 (vermicompost @ 1.5 ton/ha and apparently healthy seed treated with Provax 200 @ 0.25%) and the lowest (2.33) grain yield (t/ha) was found in T_6 (without vermicompost and 25 % black pointed seed in weight basis). The highest (4.33) straw yield (t/ha) was found in T_7 (Vermicompost @ 1.5 ton/ha and apparently healthy seed treated with Provax 200 @ 0.25%) and the lowest (3.05) straw yield (t/ha) was found in T_6 (without vermicompost and 25 % black pointed seed in weight basis).

In laboratory condition under blotter method, the result showed that black pointed seed by *Bipolaris sorokiniana* had injurious effect on seed germination. The effect of the treatments was significantly different regarding germination percentage and incidence of *Bipolaris sorokiniana* of wheat. The maximum seed germination (93.33%) was found in T_7 (Vermicompost @ 1.5 ton/ha and apparently healthy seed treated with Provax 200 @ 0.25%). On the other hand, the lowest seed germination (65.33%) was recorded in T_6 (without vermicompost and 25 % black pointed seed in weight basis).

Seed health test also indicated that sample with increased percentage of black pointed seed usually had higher percentage of *Bipolaris sorokiniana*. In case of the incidence of *Bipolaris sorokiniana* of wheat, it was observed the highest (53.67%) in T_6 and the lowest (8.667%) inT_7 were observed.

Wheat seeds samples having different levels of black pointed seed had significant effect on seed germination, normal and abnormal seedling production and dead seed percentage of wheat in Rolled Paper towel method. The highest germination percentage (90.67%) was found in T_7 and the lowest (64.67%) was found in $T_{6.}$ The maximum count of normal seedling (82.00%) was found in T_7 and the lowest (37.33%) was found in $T_{6.}$ In case of abnormal seedling, the highest abnormal (27.33%) percent was observed in T_6 and the lowest (8.667%) was found in $T_{7.}$ Percent of dead seed was found the minimum 9.33 in T_7 and the highest (35.33%) dead seed was found in $T_{6.}$

The result showed that the treatments significantly influenced shoot length (cm) root length (cm), seedling weight and vigor index of 7 days old seedlings. The highest (13.77 cm) root length was found in T_7 and the minimum (11.13 cm) root length was found in $T_{6.}$ In case of shoot length, the highest (9.83 cm) shoot length was found in T_7 and the minimum (7.87 cm) shoot length was found in T_6. Considering seedling weight, the highest (4.30 g) seedling weight was found in T_7 and the minimum (3.43g) seedling weight was found in T_6.

Vigor Index (VI) was observed the maximum (2157) was recorded in T_7 and the minimum vigor index (1226) was counted in T_6.

In the view of above finding, it has been found that minimum level of black pointed seeds resulted minimum disease incidence and subsequent disease development in the field as well as for healthy seed production.

Considering the findings of the present studies it may be conducted that the seed sample treated with treatment T_7 (vermicompost @ 1.5 ton/ha and apparently healthy seed treated with Provax 200 @ 0.25%) showed promising results in respect of emergence of seedling, disease severity and seed production. However, more investigations are needed to be persued in different Agro-ecological Zones to fix up a suitable seed health standard against leaf blight of wheat (*Bipolaris sorokiniana)* for healthy seed production.

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APPENDICES

Apendix I. Monthly average humidity, maximum and minimum temperature, rainfall and sunshine hour of the experimental period (November 2013 to April 2014)

Year	Month	Air temperature ^{0}C		Average RH (%)	Total rainfall (mm)	Sunshine (hr)
		Maximum	Minimum			
2013	November	29.6	19.2	77	34.4	5.7
	December	26.4	14.1	69	12.8	5.5
2014	January	25.4	12.7	68	7.7	5.6
	February	28.1	15.5	68	28.9	5.5
	March	32.5	20.4	64	65.8	5.2
	April	33.7	23.6	69	165.3	4.9

Source: Bangladesh Meterological Department (Climate division), Agargaon, Dhaka-1207.

Apendix II. Physical and Chemical characteristics of initial soil in the experimental field

Charracteristics	Value
% Sand	27
% Silt	43
Clay	30
Textural class%	Silty-clay
pH	5.6
Organic carbon (%)	0.45
Organic matter(%)	0.78
Total N (%)	0.03
Available P (ppm)	20.00
Exchaneable K (meq/100g soil)	0.10
Available S (ppm)	45

Source: Soil resource development institute (SRDI), 2012.Khamarbari, Farmgate, Dhaka

Appendix: III- Layout of the experiment field.

Line to line distance – 25cm; Plot size- 1.5m×1 m; Block to block distance- 1 m; Plot to plot distance- 0.5 m

20 m

N

W **E**

15 m

T_1	T_4	T_{12}	T_5	T_3	T_{13}
T_{11}	T_7	T_{15}	T_2	T_{10}	T_6
T_{14}	T_{18}	T_8	T_{16}	T_9	T_{17}

T_{15}	T_{11}	T_3	T_8	T_{12}	T_{14}
T_4	T_2	T_{10}	T_1	T_5	T_7
T_{17}	T_6	T_{16}	T_{18}	T_9	T_{13}

T_{12}	T_{16}	T_2	T_8	T_3	T_{13}
T_{18}	T_6	T_4	T_{11}	T_{10}	T_1
T_{14}	T_{17}	T_7	T_{15}	T_9	T_5

S