EVALUATION OF FUNGICIDES AND SOME ECOFRIENDLY TREATMENTS AGAINST *Bipolaris sorokiniana*

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By

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CERTIFICATE

This is to certify that thesis entitled "Evaluation of fungicides and some eco-friendly treatments against *Bipolaris sorokiniana*" 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 *bonafide* research work carried out by Rokshana Panna, Registration No. 00872/2000-2001 under my supervision and guidance. No part of the thesis has been submitted for any other degree or diploma.

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

Dated: 27.12.2007
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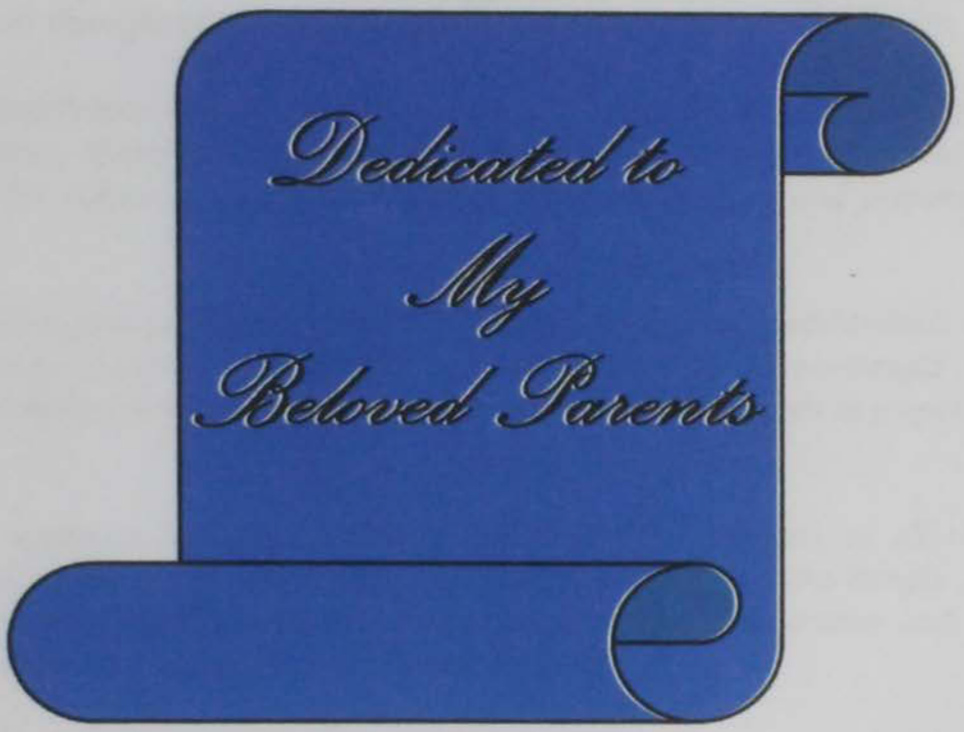
Dedicated to
My
Beloved Parents

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Evaluation of fungicides and some eco-friendly treatments against *Bipolaris sorokiniana*

By
Rokshana Panna

ABSTRACT

Experiments were conducted in the seed pathology laboratory, Department of Plant Pathology, Sher-e-Bangla Agricultural University to evaluate some ecofriendly treatments and fungicides on seed germination and incidence of *Bipolaris sorokiniana* of wheat. Seed treatments with fungicides reduced the incidence of *Bipolaris sorokiniana* over untreated control. The lowest incidence was counted (2.33%) in apparently healthy seed treated with Ridomil gold MZ 68 WP @ 0.2%. The highest seed germination (94.17%) was recorded in apparently healthy seed treated with brine solution @ 2% for 15 minutes followed by solarized apparently healthy seed for 5 hours (99.33%), polythene solarized apparently healthy seed for 5 hours (99.00%), apparently healthy seed treated with hot water at 52°C for 10 minutes (99.00%) and Burmuda grass extract (99.60%). Comparatively the lowest incidence of *Bipolaris sorokiniana* (1.50%) was observed in apparently healthy seeds treated with hot water at 52°C for 20 minutes followed by polythene solarized apparently healthy seeds for 15 hours (5.00%), apparently healthy seeds solarized for 15 hours (6.00%), apparently healthy seeds treated with brine solution @ 2% for 30 minutes (13.00%) and apparently healthy seeds treated with Neem extract (14.00%).

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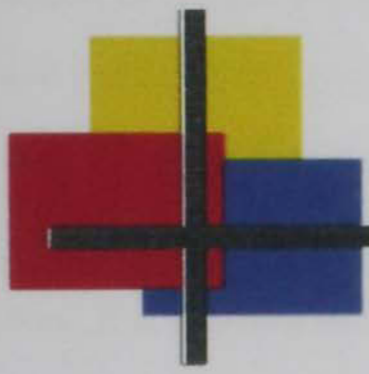
LIST OF ABBREVIATIONS

%	=	Percent
@	=	At the rate
°C	=	Degree Centigrade
Anon.	=	Anonymous
BARI	=	Bangladesh Agricultural Research Institute
BAU	=	Bangladesh Agricultural University
BBS	=	Bangladesh Bureau of Statistics
CIMMYT	=	International Maize and Wheat Improvement Center
cv.	=	Cultivar(s)
DMRT	=	Duncan's Multiple Range Test
e.g.	=	For example
et al.	=	And Others
etc.	=	Etcetera
FAO	=	Food and Agriculture Organization
g	=	Gram
hr	=	Hour(s)
i.e.	=	That is
IRRI	=	International Rice Research Institute

ISTA	=	International Seed Testing Agency
kg	=	Kilogram
LSD	=	Least Significant Difference
no.		Number
PDA	=	Potato Dextrose Agar
SAU	=	Sher-e-Bangla Agricultural University
T	=	Treatment
t/ha	=	Ton per Hectare
UNDP	=	United Nation Development Program
$^{w}/_{v}$	=	Weight per Volume
wt.	=	Weight



Chapter 1
Introduction

1. INTRODUCTION

Wheat (*Triticum aestivum* L.) is one of the most important cereal crops and main staple food all over the world. About two third of the World's population used wheat as staple food (Majumder, 1991). Dubin and Ginkel (1991) reported that the largest area of wheat cultivation in the warmer climates exists in the South-East Asia including Bangladesh, India and Nepal. It is the second most important grain crop in Bangladesh that plays a vital role in the national economy by reducing the volume of import of cereals (Razzaque *et al.*, 1992). Besides human nutrition, both wheat grain and straw are also used as animal feed. Wheat straw is used as fuel and as well as straw shade for the house of poor farmers of Bangladesh. Though the crop introduced in Bangladesh (former East Pakistan) in 1967 but its popularity increased after 1975. Now it is a raising crop over the country and well accepted by the farmers. Wheat cultivation has increased manifolds to meet up the food shortage in the country. In spite of its importance, the yield of the crop in our country is low in comparison to the other countries of the world, where average yield estimated 2.69 t/ha (FAO, 1997). Though the area, production and yield of wheat have been increasing dramatically during the last decade, but still it is too low (2.2 t/ha) in comparison to the developed countries like Japan, France, Germany and UK producing 3.76, 7.12, 7.28, and 5.00 t/ha, respectively (FAO, 2000). About 706.86 thousand hectares of land in Bangladesh is covered by wheat cultivation with the annual production of 1570 thousand tons (BBS, 2006).

There are many constraints responsible for low yield of wheat in Bangladesh. Among the different factors that affect the production of wheat, use of unhealthy or diseased seeds is one of the major constraints. Government and semi government organizations supply only 22.8 % of the total wheat seed required during 1998-1999 (Motahar, 2000). These seeds are treated as quality seeds in Bangladesh. The rest 77.2 % of the seeds produced traditionally by the farmers with no or little care even for purity and germination and remain out of scope of certification. As a result, a huge crop loss is incurred every year in wheat due to seed diseases in the country.

Seed borne disease causes enormous losses to our crop (Fakir, 1982). Wheat plants at all growth stages are prone to the attack of numerous diseases. The crop is known to suffer from as many as 200 diseases of which the most important and damaging ones are seed borne (USDA, 1960). Seed borne infections of fungal pathogens are important not only due to the association with the seeds that cause germination failure, and/or causing disease to the newly emerged seedlings or growing plants, but also contaminate the soil by establishing its inocula permanently. Wheat suffers from as many as 26 seed borne pathogens causing 14 seed borne diseases. Among them leaf spot and black point caused by *Bipolaris sorokiniana* has become a serious concern in Bangladesh (Azhar *et al.*, 1972; Fakir, 1988).

The most expectable method for controlling the disease is cultivation of resistant variety, but not a single wheat cultivar in the country is found to be resistant (Hossain and Azad, 1992). The second most acceptable method for controlling of this disease is sowing of pathogen free seeds. Therefore along with routine seed health testing, seed treatment before sowing is necessary.

Treatment of seed with seed dressing fungicides was found to improve germination and decrease infection of seedling growth from the black pointed seeds. Many workers followed the use of different chemicals against the disease and Tilt-250 EC had been proved effective (Anonymous, 1989). Easy and economic way of controlling seed borne diseases may be the use of clean, healthy looking and washed seeds. It may play an important role to minimize disease incidence and reducing entry of pathogens to the field. In a preliminary study, it has been found that, cleaning and washing farmer's saved seed reduced seedling diseases up to 53.87% over uncleaned farmer's saved seed (Hossain and Asad-ud-Doullah, 1998). Seed cleaning and washing are easy and practically feasible method for our poor farmers (Hasan, 2000). This technique does not require much effort and it is free from chemical threat. It is an alternative method of avoiding seed treating chemicals and thus environment will be saving from pollution.

Though chemical treatment are most effective for controlling the disease, but continuous use of chemicals results in accumulation of harmful chemical residues in soil as well as in the plant products causing serious health hazard. Chemical fungicides also pollute the environment and develop tolerance of pathogen. The chemical fungicides are also costly that is sometimes burden for the poor farmers. In addition, their harmful effect is responsible for air, soil and water pollution.

In this aspect seed treatment with botanicals may be a nice option in controlling seed borne pathogens as well as seed transmitted disease in the field. Use of plant extracts in controlling pathogens is now successfully used against certain fungal pathogens (Assadi and Behroozin, 1987; Miah *et al*., 1990; Fakir and Khan, 1992; Suratuzzaman, 1995; Hossain *et al*. 1997). Hossain and Schlosser (1993) have been reported promising fungicidal effect of Neem (*Azadirachta indica*) extracts against *Bipolaris sorokiniana*.

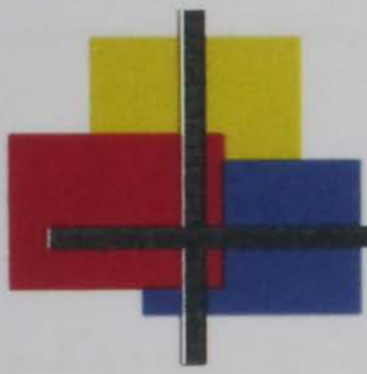
Physical seed treatments in controlling seed borne infection of *Bipolaris sorokiniana* and seed germination are found to be better in the investigation of different researchers. Reduction of incidence of seed borne (*Bipolaris sorokiniana)* was obtained by manually sorted healthy looking seed up to 66.15% over untreated control (Zobaer *et al., 2007*). They also found significant reduction of incidence of the same fungus achieved by treating with hot water and solar heat of 76.79% and 73.16% respectively.

Therefore, it is judicious to explore less expensive, less risky nonchemical components to treat seeds for freedom from the seed-borne pathogens. In this perspective, use of solar heat (Direct and polythene Solarization), seed treatment with hot water and Brine Solution and manual seed sorting to obtain apparently healthy seed would be good alternatives in controlling seed borne fungal pathogen.

Now a day's scientists are giving more emphasis on sustainable technology for agriculture. It would be possible when ecologically sound, economically viable, culturally appropriate and socially acceptable technology will be adopted.

Considering the above facts the present investigation was undertaken with the following objective:

- To evaluate the effect of some fungicides and eco-friendly treatments on seed germination and incidence of *Bipolaris sorokiniana* of wheat.



Chapter 2
Review of literature

2. REVIEW OF LITERATURE

Seed borne pathogens are reported to cause spots or discoloration on the seed. Seed treatment may reduce number of seed borne pathogens associated with seed and thus improves the quality of seed. Management of plant diseases with eco-friendly approaches, i.e. alternative to chemicals is now getting top priority. Use of eco friendly treatments instead of chemical fungicides in controlling disease is one of the recent approaches for plant disease control. Therefore, attempt has been taken to collect relevant literature on this aspect, which are given below:

2.1 Seed treatment with eco-friendly approaches

Bedi and Gill (1960) reported that a substantial loss in germination (11.0 to 37.3%) is associated with the spotted seeds. They found spotted seeds, which did not germinate was profusely covered with mycelium and the spore of *Helminthosporium oryzae*. Healthy seeds gave very high percentage germination as 99% and remained free from any seedling mortality. In the case of naturally infected seed there was a loss of about 5% among the seedlings. They also found appreciable loss in weight with spotted seeds. It was found to vary from 40.58% to 29%.

Prabhu and Prasada (1970) controlled Alternaria leaf blight of wheat caused by *Alternaria triticiana* by soaking seed in water at 52 to 54°C for 10 min. Lowest dead seeds (9.9%) recorded at temperature 51-52°C, which was lower than dead seeds (12.8%) at control condition. Increase in temperature above 51- 52°C, there observed a continuous significant increase in dead seeds indicating the negative effect of heat against viability of seeds. The highest dead seeds (53.6%) recorded at temperature 58-59°C.

Singh and Sharma (1978) compared the antifungal activity of 34 crude extracts of Indian flowering plants which strongly inhibited *Helminthosporium sativum*, *Colletotrichum falcatum* and *Fusarium oxysporum*.

Couture and Sutton (1980) observed that in dry heat treatments of wheat seeds naturally infected with *Bipolaris sorokiniana*, as the temperature and the time of exposure to treatment increased survival of both fungus and seeds decreased but not at the same rate. A severe reduction in viability of wheat was produced under conditions required to eliminate the fungus from the seeds. The best seed survival associated with a treatment giving complete elimination of the fungus was only 42% (60 h at 90°C).

According to IRRI (1983) *Bipolaris oryzae* caused brown spot of rice as a seed transmitted fungus effectively controlled by the hot water seed treatment at 53-54°C for 10-12 minutes. This treatment controlled primary infection at the seedling stages. Presoaking the seed in cold water for 8 hours increased effectivity of the treatment.

Alice and Rao (1987) obtained garlic extract have significant effect in controlling seed-borne *Drechslera oryzae*. Evaluation of plant extracts against seed-borne infection of fungus and increased the germination ability of the treated seeds (Alice and Rao, 1987).

Ashrafuzzaman and Hossain (1992) evaluated Pudina (*Mentha viridis*) extract against *Bipolaris sorokiniana* and observed that the extract inhibited mycelial growth and spore germination. In the same work they found that extract of castor (*Ricinus communis*) and Dantha Kalash (*Leucas aspera*) were inhibitory against mycelial growth and spore germination of *Bipolaris sorokiniana*..

Khan and Kumar (1992) observed the antifungal activity of leaves extract of Neem (*Azadirachta indica*) with different dilutions of wheat seeds Mycoflora. They recorded a marked reduction in seed Mycoflora and enhance seed germination of wheat seeds.

Rovesti *et al.* (1992) also found aqueous Neem extract effective against some important diseases of different herbaceous crops.

Hossain and Schlosser (1993) found Neem seed extracts and Neem oil cake effective against *Bipolaris sorokiniana*. The extract inhibited the growth of the fungus *in vitro* and also reduced its pathogenecity on wheat leaves. Germination rate of wheat seeds increased after treatment with extracts of Neem seed and cake.

Hossain *et al.* (1993) evaluated that extracts of *Lawsonia alba, Ipomoea fistulosa, Allium sativum* and *Leucas aspera* against *Rhizoctonia solani* and *Bipolaris sorokiniana*. Among the test extract, *A. sativum* completely inhibited the mycelial growth at dilution ratio of 1:4 (w/v).

Khan and Hossain (1993) observed that extracts of *Allium cepa , A. sativum , Datura stramonium, D. plumeiri, Lawsonia alba, Ricinus communis, Leomurus sibiricus* and *Metha viridis* completely inhibited spore germination of *B. sorokiniana* at 1:3 (w/v) dilution ratio.

Winter *et al.* (1994) compared hot water treatment of certified wheat seeds dipped in water at 15°C for 2hrs, then air-dried at 40°C for 5hrs with chemical seed treatment with fenpiclonil and carboxin at 400 ml/kg seed for control of seed-borne fungi of wheat. According to them hot water treatment was equally or more effective than the fungicides in controlling *Gerlachia nivalis* and *Seploria nodorum*.

Bisht and Khulbe (1995) studied the efficacy leaf of extract of *Allium sativum* in controlling the growth of *Drechslera oryzae*. The fungitoxic properties of *Allium sativum* have been observed and significant reduction of the mycelial growth compared with the control was obtained.

Hossain *et al.* (1995) reported that extract of Mehedi (*Lawsonia alba*) was found to be effective against *Bipolaris sorokiniana*

Jahan (1996) demonstrated that solar heat treatment effectively inhibited seed-borne fungi of jute.

Khaleduzzaman (1996) studied hot water treatment of wheat seeds at 49°C, 52°C, 55°C and 61°C, respectively for 5 and 10 min in controlling seed borne infection. Hot water treatment at 52°C-55°C for 10 minutes gave highest control of *Alternaria tenuis*, *Aspergillus flavus, Aspergillus roger, Bipolaris sorokiniana, Curvularia luanata, Fusarium spp.* and *Penecillium spp.* and increased seed germination.

Winter *et al.* (1996) found that hot water treatment of barley seeds at 52° for 5 or 10 minutes was partially effective against seed borne *Drechslera teres* and *Helminthosporium sativum*. However, hot water treatment at 52°C for 10 min sometimes reduced germination and field emergence but the effect was less with 5 min treatment.

Khaleduzzaman (1996) reported the effect of plant extracts viz Bishkatali (*Polygonum hydropiper*), Garlic (*Allium sativum*), Ginger (*Zingiber officinale*) and Neem (*Azadirachta indica*) and a seed dressing chemical Vitavax-200 (Carboxin) on seed-borne infection of wheat following blotter method of seed health testing. Vitavax -200 was found best in reducing seed borne infection and increasing germination of seeds. All the four plant extracts were found effective against seed-borne fungi of wheat resulting statistically similar effect like Vitavax-200. However, Garlic was turned up as superior among the extract followed by Ginger and Neem.

Hossain *et al.* (1997) demonstrated that the extract of *Allium sativum* and *Lawsonia alba* showed marked effect in controlling the spore germination and mycelial growth of *Bipolaris sorokiniana* and pathogenecity to wheat seeds and *Nigella sativa* showed positive antifugal activity in reducing the pathogenicity of *Bipolaris sorokiniana* of wheat seeds.

Fakir and Jahan (1998) carried out an experiment to control seed-borne, fungal pathogens of jute by seed treatment with solar heat. Solar heal treatment effectively reduced 91.3% seed-borne infections and increased 9.0% seed germination.

Hossain and Asad-ud-Doullah (1998) reported that cleaning and washing of farmer's rice seeds reduced the seedling diseases up to 53.87% over the uncleand farmer's seed. Use of healthy seedlings resulted 14.77% increasing in grain yield over the unbiased use of normal seedlings from seedbed.

Govindachari *et al.* (1998) studied that the activity of Neem oil (*Azadirachta. indica*) against *Drechslera oryzae* (*Cochliobolus miyabeanus*), *Fusarium oxysporum* and *Alternaria tenuis* (*Alternaria. alternata*). They observed that the active fractions of those plant extracts contained major compound such as 6-deacetylnimbin, azadiraduione, nimbin, salannin and epoxyazadiradione. Pure azadradione, nimbin, salannin and epoxyazadiradione did not show antifungal activity. However, when terpenoids (Methanol extraction) were mixed and bioassayed, they showed antifungal activity, suggesting possible additives; synergistic effects

Rahman *et al.* (2000) carried out an experiment to improve seed quality by seed cleaning (manual sorting and flotation in water) in four seed samples of rice cv. BR11. The seed borne fungi associated with the treated and untreated seeds were *Bipolaris oryzae, Trichoconis padwickii, Curvularia lunata, Nigrospora oryzae, Alternaria tenuis, Aspergillus spp.* and *Penicillium spp.* All the three seed treatment methods reduced all seed borne fungal infections. The best method was treatment with Vitavax-200, followed by manual seed sorting and flotation. Germination test following paper towel method showed that chemical treatment was the best in producing normal seedlings, followed by manual sorting and flotation method. Vitavax-200 treated seeds and manually sorted seed produced the highest number of tillers/hill, percentage of healthy seeds and 1000 seed weight. Grain yield was increased in manually sorted, flotation and Vitavax-200 treated seeds/ plots by 30.5, 13.5, and 27.3%, respectively.

Muniz (2001) stated that the dry heat treatment on the control of seed transmitted pathogens and its effects on the viability of tomato seeds treated at 70°C for 12 days eradicated fungi associated with tomato seeds. But in hot water treatment at 50 C for 30 minutes under laboratory research the associated fungi in tomato seeds were eradicated.

Fallik *et al.* (2002) studied the effectiveness of a short pre-storage hot water rinsing and brushing on resistance to decay development and chilling injury on pink tomato cv. 189 fruit that were kept for 15 days at 5 or 12° C plus three days at 22°C. He suggested the alternative method of a very short (15 S) HWRB (Hot Water Rinsing & Brushing) at 52°C for desirable tomatoes. This treatment extended storability well over three weeks at 5°C by minimizing CI (Chilling Injury) and enhancing resistance against pathogen during Fallik *et al.* (2002) studied the effectiveness of a short pre-storage hot water rinsing and brushing on resistance to decay development and chilling injury on pink tomato cv. 189 fruit that were kept for 15 days at 5 or 12° C plus three days at 22°C. He suggested the alternative method of a very short (15 sec) HWRB (Hot Water Rinsing & Brushing) at 52°C for desirable tomatoes. This treatment extended storability well over three weeks at 5°C by minimizing CI (Chilling Injury) and enhancing resistance against pathogen during storage.

Hossien (2002) reported that farmer's clean seed, washed farmer's seed, washed clean seed and seed treated with Vitavax-200 increased 16.62%, 16.45%, 23.39% and 26.6% grain yield, respectively over farmer's saved seeds of rice (BR11).

Jiskani (2002) reported that the brown spot or blight of rice is a much more wide spread and a common disease in almost all rice growing area of the world. He prescribed that brown spot or blight of rice caused by *Helminthosporium oryzae* effectively controlled by hot water seed treatment at 54° C for 10 minutes.

Siddique *et al.* (2002) reported that reaction of five wheat varieties to *Bipolaris sorokiniana* and *Fusarium moniliforme* was assessed in terms of prevalence of the pathogen on the seeds, symptom severity on the growing plants and effects of the pathogens on the yields. Prevalence of *B. sorokiniana* was 4-4.25% in kanchan and Akbar and of *Fusarium monilifome* was 2.25-2.75% in Gourab and Kanchan. Disease severity was the highest in Barkat both in laboratory and field conditions. However, *Bipolaris sorokiniana* infection was more severe than that of *Fusarium moniliforme*. Percent diseased grains were high in Barkat, while grain yield was low in barkat and sonalika due to inoculation of *B. sorokiniana* and *F. moniliforme*. Thus, there was a significant variation in reaction among the wheat varieties to the pathogens.

Chowdhury (2005) observed that highly infected / contaminated seed samples with seed borne fungi of rice, wheat, cosmos, Zinnia, sunflower and radish were subjected to seed treatment with 1:0,1:1, 1:5,1:10 and 1:20 dilution of crude/ nascent extract of garlic, datura and turmeric; 1:1, 1:5, 1:10 and 1:20 dilution of commercially available oil extracts of neem, mahogany and koromcha; hot water treatment for 15 minutes at 50°C, 52°C, 54°C, 56°C and 58°C temperatures and chemical seed treatment with

Vitavax-200 a 0.1%, 0.2% and 0.3% of the seed weight. Botanicals at all concentrations reduced the occurrence of mycoflora on the seed significantly and thereby increased seed germination. Some fungi were totally removed at 1:10 dilution of commertially available plant oil extract.

Hossain *et al.* (2005) tested different plants viz. Bishkatali, Vatpata, Garlic, Gagra, Bitter Guard and Neem against fungi associated with wheat seed. After dipping in the crude extract and alcoholic extract both in undiluted and diluted form for 24 hours, wheat seed were invested and five different fungi viz. *Bipolaris sorokiniana, Alternaria tenuis, Curvularia lunata, Fusarium spp* and *Aspergillus spp* were found to reduced significantly by seed treatment with plant extracts over control and increased seed germination. Crude extracts were superior to the dilution extract. Out of 6 plant species, Neem extracts was turned up as superior among the selected extracts followed by Garlic, Bishkatali and Vatapta.

Ozer (2005) reported that the kernels of the bread wheat (*Triticum aestivum* L.) cultivars Flamura 85, Katia, Pehlivan and Sana from farmer fields of Tekirdag province in 2000 and 2001 were analyzed to identify the fungi associated with the black point disease. Fungi in the embryos, endosperms and seed coats of the kernels (healthy and black pointed) separately, were determined by plating them on agar medium. The same cultivars were used to test the effect of black point on emergence at day five and ten, and seedling vigour (length of shoot, dry weight of roots and

shoots) in plate experiments. *Bipolaris sorokiniana* was the dominant fungus in black pointed kernels for both years and isolated from the endosperm and seed coat especially, but present at low level. None of the fungi from black pointed kernels were isolated from healthy kernels. Black point delayed seedling emergence from the kernels of 2000. However the reduction in seedling vigour was significantly higher in the affected kernels of 2001.

Akhter *et al.* (2006) reported that inhibition of conidial germination of *Bipolaris sorokiniana* was tested using eight ethanolic plant extracts, ten aqueous plant extracts in combination with cow dung and five aqueous plant extracts in combination with cow urine. Hundred percent of conidial germination was inhibited with the application of ethanolic extracts of *Adhatoda vasica* (leaf) and *Zingiber officinale* (rhizome) at 2.5% concentration. After the treatment with *Vinca rosea*, Piper betle and *Azadirachta indica* extracts in combination with cow dung suspension showed 100% inhibition of conidial germination; where the lowest was noted in case of *Rauwolfea serpentina* (30%) extract at same concentration. At 2.5% concentration of *Calotropis procera* extracts in combination with cow urine, 91% inhibition of conidial germination was observed. In most cases, *Ocimum sanctum* extract exhibited less inhibitory effect against *Bipolaris sorokiniana*.

Islam (2006) tested different plant extracts with three concentrations viz 1:1, 1:2, 1:4 ($^{w}/_{v}$) to control seed borne *Bipolaris sorokiniana*. All the plant extracts except mehedi increased seed germination in the laboratory in compared to untreated control. The highest incidence of seed borne *Bipolaris sorokiniana* was recorded under control. A remarkable reduction of the incidence of fungi was achieved by treating seeds with botanicals.

Rahman *et al.* (2006) tested 33 plant extracts in vitro against *Bipolaris sorokiniana*. The extracts of kalijira, turmeric, ginger, garlic, onion, neem, allamanda, nayantara, mandar, naglingam and duranta showed good effect in controlling *Bipolaris sorokiniana*. The extracts of durba, neem, allamanda, turmeric, ginger, garlic, onion, kalijira, katamehedi, tulsi, mandar, eucalyptus, mutha, naglingam, bel have profound effect in increasing seed germination and controlling seed borne *Bipolaris sorokiniana*. Among the tested plant species durba, allamanda, turmeric, garlic, onion, katamehedi, tulsi, mutha, bel, lemon treated seeds produced maximum number of healthy seedlings of wheat in compared to untreated control.

2.2 Seed treatment with chemicals

Peltonen and Karjalainen (1992) observed that application of Tilt increased grain yield. They also found that Tilt 250 EC significantly increased nitrogen uptake, grain weight and protein quality in a good growing season but in a cold and wet weather Tilt 250 EC did not increase yield or quality of cultivars.

Fakir and Khan (1992) reported that garlic bulb extract was effective in controlling seed-borne fungal pathogen of jute such as *Macrophomina phaseolina* and *Fusarium spp.* by seed treatment.

Black point of wheat caused, by *Bipolaris sorokiniana* is the major disease constraint of wheat production. An integrated approach is required to control the disease as regards to reduce the fungal inoculums by using seed with low level of infection. Seed treatment, spraying appropriate fungicide and other agronomical practices are the important approaches. (Duveiller and Gilchrist, 1994)

Meisner *et al.* (1994) found that seed dressing with Vitavax-200 decreased soil borne pathogen population and seedling infection caused by *Bipolaris sorokiniana.*

Mondal *et al.* (1994) evaluated four commercial fungicides to evaluate their efficacy in controlling Black point of wheat under natural epiphytotic conditions during 1991-1992 and 1992-1993. Among them, Tilt-250 EC (0.05%) was the most effective and

profitable one, which controlled the disease significantly. The disease severity was also reduced by Dithane M-45 (0.2%) and Pencozeb (0.15%) and gave profitable yield while application of Rovral (0.2%) was round uneconomic offering the lowest gross margin.

Khaleduzzaman (1996) evaluated the effect of plant extracts viz bishkatali (*Polygonum hydropiper*), garlic (*Allium sativum*), ginger (*Zingiber officinale*), neem (*Azadirachta indica*) and a seed dressing chemical Vitavax-200 (Carboxin) on incidence of seed-borne fungi of wheat following blotter method of seed health testing. Vitavax–200 was found best in reducing seed–borne infection and increasing germination of seeds. All the four plant extracts were found effective against seed-borne fungi of wheat resulting statistically similar effect like Vitavax–200. However, garlic was turned up as superior among the extract followed by ginger and neem.

Mahfuzul (1997) evaluated some plant extract viz. garlic (*Allium sativum*), ginger (*Zingiber officinale*), nisinda (*Vitex negundo*), dolkalmi (*Ipomoea fistulosa*) and marigold (*Tagetes erecta*) against major seed–borne fungal pathogens of chilli. Among the plant extracts garlic was found to be most effective followed by neem leaf. The garlic and neem leaf extracts at the dilution ratio of 1:1 were almost equally effective.

Rahman *et al.* (1999) found that bishkatali (*Polygonum hydropiper*), garlic (*Allium sativum*), ginger (*Zingiber officinale*) and neem (*Azadirachta indica*) extracts were effective against seed borne infections by *Alternaria tenuis, Bipolaris sorokiniana, Curvularia lunata, Fusarium* spp. of wheat. However, garlic was found superior to ginger and neem.

Rahman *et al.* (1999) reported the effect of seed treatment with Vitavax-200 (Carbendazim + Thiram) at 4 g/kg against black point of wheat caused by *Bipolaris sorokiniana* (*Cochliobolus sativus*). They observed a significant effect incase of reduction of leaf blight severity and increased yield were observed with seed treatment with Vitavax combining with foliar spray of Tilt-250 EC.

Rashid *et al.* (2001) reported that Tilt-250 EC (Propiconazole) is a good fungicide in controlling leaf blight of wheat

Rahman *et al.* (2001) found that black point (caused by *Bipolaris sorokiniana*) of wheat can be controlled by using Vitavax-200 (0.4%) and Tilt 250EC (0.1%) but their combined effect of (Vitavax-200 + Tilt-250 EC) showed better control than the single one.

Patil *et al.* (2002) reported that the efficiency of 0.1% Propiconazole (Tilt 250EC & 25% EC), 0.1% Haxaconazole 25% EC, 0.05% Tridemorph 200 WC. 0.1% Carbendazim 50 WP (Bavistin), 0.1% Triadimefon, 0.0025% Mancozeb (Pencozeb), 0.020% Chlorothalonil, 0.3% Copper oxychloride 50 WP and 0.03% Nimbicidin in controlling black point of wheat. The incidence of the disease was not observed in plant sprayed with 0.1% Propiconazole (Tilt) and increased the yield and biomass.

Siddique (2003) reported that the seeds remained on sieve showed better performance in respect of seed germination, seedling vigour and seed health in different wheat varieties (Kanchan, Akbar and Sourov). Seeds were cleaned by bamboo sieve having mesh size 2.25mm. Seeds were taken on bamboo sieve and manually shaken for different duration (two, four, six and eight minute). The seeds remained on sieve were considered as healthy seeds and those were dropped the sieve were consider as unhealthy seeds. Germination and seedling vigour increased with increasing duration of sieving. Seed cleaning by bamboo sieve reduced prevalence of pathogens. Infection percentage gradually decreased with increasing duration of sieving.

Howlader (2003) observed that seed treatment with *Allamanda* leaf extract (1:1) effectively increased germination of egg plant seeds and tremendously decreased nursery diseases.

Kabir (2003) reported that the germination test of farmer's saved seed treated with Vitavax-200 resulted highest percentage of germination, followed by soaked washed clean seed. Seedling emergence in seedbed was significantly higher (21.35%) in soaked washed cleaned seed than all other treatment (untreated farmer's seed, cleaned seed, washed farmer's seed soaked washed farmer's seed, washed clean seed, and chemical treatment of farmer's saved seed with Vitavax-200). The disease severity of brown spot, narrow brown spot, blast, sheath blight and sheath rot was the lowest in plots under farmer's saved seed treated with Vitavax-200, followed by soaked washed cleaned seed. The Vitavax-200 treated seed significantly resulted highest plant height,

panicle length and yield (5.88 tons/ha), which was 18.07% higher over the use of untreated farmer's seed; while soaked washed cleaned seed and washed cleaned seed gave 16.47%, 14.86% increased seed yield, respectively. Maximum numbers of apparently healthy seeds were obtained by using farmer's seed treated with Vitavax-200 (76.24%), followed by soaked washed cleaned seed (70.47%) over untreated farmer's seed (57.69%).

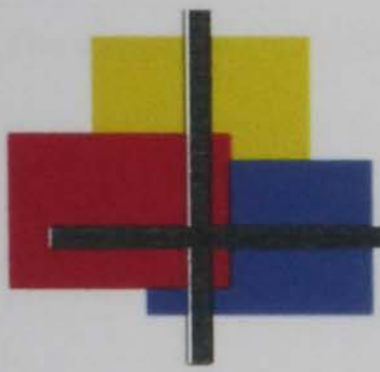
Uddin (2005) reported that seed borne pathogens were significantly reduced by treating seeds with chemical (vitavax-200) followed by garlic extract, brine solution, hot water and physically sorted seeds in Lentil. The highest reduction of seed borne fungal flora were observed in case of chemical treatment followed by garlic extract, brine solution, hot water and physically sorted seeds. In the field condition, germination percentage was higher in physically sorted seeds.

Hossain *et al.* (2005) reported that extract of different plant; viz. bishkatali, vatpata, garlic, gagra, bitter guard and neem were effective against fungi associated with wheat seed. Out of six plant species, neem extract was turned up as superior among the selected extracts followed by garlic, bishkatali and vatapta.

Islam (2006) evaluated eight plant extracts including Vitavax-200 against leaf spot (*Bipolaris sorokiniana*) of wheat. Among eight plant extracts, onion, garlic, kalijira, ginger, bishkatali and neem extract showed statistically similar grain yield as of seed treatment with Vitavax-200. Seed treatment with bishkatali extract increased 29.74% grain yield over untreated control.

Kabir (2006) tested some physical and chemical seed treatments to control seed borne *Bipolaris sorokiniana*. The highest germination was found in case of manually sorted apparently healthy looking seed treated with vitavax-200. The highest grain yield was also obtained from apparently healthy seed treated with Vitavax-200, over farmer's saved seed.

Zobaer *et al.* (2007) reported the effect of seed sorting, seed solarization and seed treatment with hot water on black point (*Bipolaris sorokiniana*) and seed yield of wheat. Highest reduction of incidence of seed borne *Bipolaris sorokiniana* was obtained by manually sorted healthy looking seeds treated with Vitavax 200. Significant reduction of incidence of same fungus was achieved by treating seeds with hot water and solar heat. Seed treatment of apparently healthy seed with vitavax 200 (0.4%) was found best in reducing the incidence, of *Bipolaris sorokiniana* and increased seed germination and seed yield over un-cleaned farmer's saved seeds. Only manually sorted seed reduced 66.15% incidence of *Bipolaris sorokiniana* over untreated control. But sun-dried sorted seed and hot water treated sorted seed reduced 76.79% and 73.16% incidence of *Bipolaris sorokiniana* over untreated control respectively.



Chapter 3
Materials and Methods

3. MATERIALS AND METHODS

The experiment was conducted at the Seed Pathology Laboratory, Department of Plant Pathology, Sher-e-Bangla Agricultural University; Dhaka, Bangladesh.

3.1. Experimental period

The experiment was carried out during the period from April, 2007 to December, 2007.

3.2. Design of experiment

The experiment was laid out in Completely Randomized Design (CRD). 400 seeds were tested for each treatment. The method used for seed health testing was Blotter method of ISTA, 1999. Data was recorded on germination percentage and % seed yielding *Bipolaris sorokiniana*.

3.3. Treatments

Six different sets of experiments were carried out during this investigation.

3.3.1. Evaluation of fungicides against *Bipolaris sorokiniana*

The treatments were:

T_1 = Untreated (Control)
T_2 = Cupravit 50 WP (0.7%)
T_3 = Ridomil Gold MZ 68 WP (0.2%)
T_4 = Tilt 250 EC (0.2%)
T_5 = Rovral 50WP (0.2%)
T_6 = Bavistin 50WP (0.2%)

3.3.2. Evaluation of brine solution against *Bipolaris sorokiniana*

Seed treatment with brine solution (2%) for:

T_1 = Untreated (Control)
T_2 = 5 minutes
T_3 = 15 minutes
T_4 = 30 minutes
T_5 = 60 minutes

3.3.3. Evaluation of seed solarization against *Bipolaris sorokiniana*

Seed solarization for:

T_1 = Untreated (Control)
T_2 = 5 hours
T_3 = 10 hours
T_4 = 15 hours
T_5 = 20 hours

3.3.4. Evaluation of polythene solarization of seed against *Bipolaris sorokiniana*

Polythene solarization of seed for:

T_1 = Untreated (Control)
T_2 = 5 hours
T_3 = 10 hours
T_4 = 15 hours
T_5 = 20 hours

3.3.5. Evaluation of hot water (52°C) against *Bipolaris sorokiniana*

Seed treatment with hot water (52°C) for:

T_1 = Untreated (Control)
T_2 = 5 minutes
T_3 = 10 minutes
T_4 = 15 minutes
T_5 = 20 minutes

3.3.6. Evaluation of plant extracts against *Bipolaris sorokiniana*

Five plant extracts were evaluated:

		Treatments	Concentration (w/v)
Allamanda:	T_1 =	Untreated (Control)	
	T_2 =	Allamanda extract	1:2
	T_3 =	,,	1:4
	T_4 =	,,	1:6
	T_5 =	,,	1:8
Neem:	T_1 =	Untreated (Control)	
	T_2 =	Neem extract	1:2
	T_3 =	,,	1:4
	T_4 =	,,	1:6
	T_5 =	,,	1:8
Nut Sedge:	T_1 =	Untreated (Control)	
	T_2 =	Nut Sedge (Mutha) extract	1:2
	T_3 =	,,	1:4
	T_4 =	,,	1:6
	T_5 =	,,	1:8

Barmuda Grass:	T_1 =	Untreated (Control)	
	T_2 =	Barmuda Grass extract	1:2
	T_3 =	,,	1:4
	T_4 =	,,	1:6
	T_5 =	,,	1:8
Black Cumin:	T_1 =	Untreated (Control)	
	T_2 =	Black Cumin seed extract	1:2
	T_3 =	,,	1:4
	T_4 =	,,	1:6
	T_5 =	,,	1:8

3.4. Collection of seeds

Wheat (*Triticum aestivum* L.) cv. Kanchan was collected from a farmer of Gazipur district and used in this study. About 6 kg seed sample of wheat cv. Kanchan was collected. After collection, the seeds were kept in at air tight plastic container and stored at normal room temperature in MS laboratory of the Department of Plant Pathology, Sher-e-Bangla Agricultural University.

3.5. Preparation of seeds for different treatments:

3.5.1. Seed sorting

Apparently healthy looking seeds were separated from farmer's saved seed by manual seed sorting, eliminating inert matter, varietal mixture, other crop seeds, weed seeds, crop residues and black pointed seeds (Plate-1).

A B

Plate 1: Seeds used for the experiment
A. Apparently healthy seed
B. Farmer's saved seed

3.5.2. Seed treatment with fungicides

Five fungicides were used for seed treatment by dipping method. The fungicides were Ridomil Gold MZ 68 WP (0.2%), Bavistin 50WP (0.2%), Tilt 250 EC (0.2%), Cupravit 50 WP (0.7%) and Rovral 50WP (0.2%). Seeds were dipped in the fungicidal suspension (plate-2) of desired concentration for 15 minutes. After seed treatment the suspension was removed. The treated seeds were then kept on blotting paper to remove excess moisture from seed surface prior to placing in the petridishs.

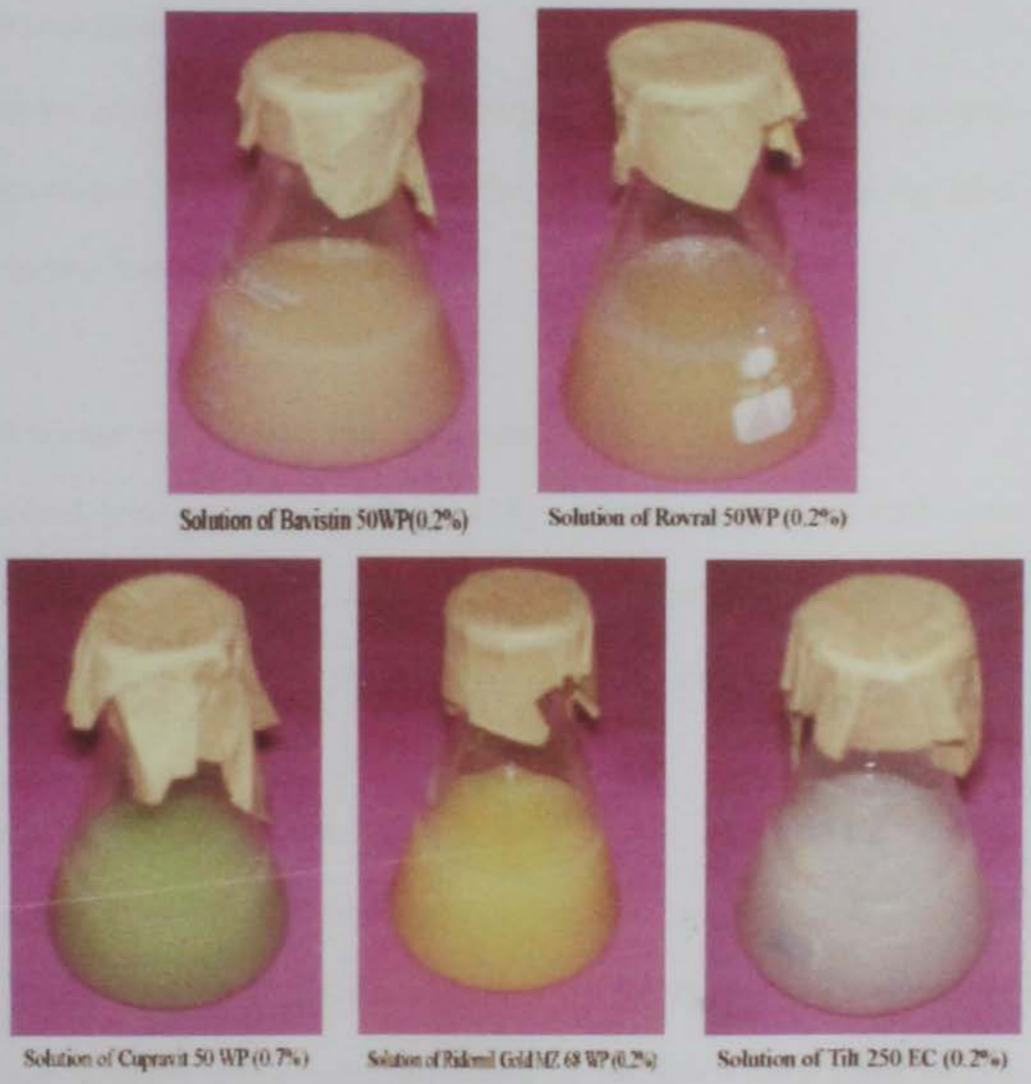


Plate 2: Different fungicidal suspension prepared for the experiment

3.5.3. Seed treatment with brine solution

At first 2% brine solution was prepared by mixing 100 ml tap water with 2g edible salt (NaCl). Therefore farmers saved seed and apparently healthy seeds were soaked in the solution separately for 5, 15, 30, 60 minutes respectively. After treating seeds the excess water was removed and the seeds were air dried in the laboratory prior to placing in the Petridishs.

3.5.4. Seed treatment by sundrying

Farmer's saved seed and apparently healthy seeds were sundried separately for 5, 10, 15, 20 hours respectively. After treating the seeds were brought in the laboratory prior to placing in the Petridishs.

3.5.5. Seed treatment by polythene solarization

Farmer's saved seed and apparently healthy seeds were packed with clear polythene and sun dried separately for 5, 10, 15, 20 hours respectively. After treating the seeds were brought in the laboratory prior to placing in the Petridishs.

3.5.6. Seed treatment with hot water

Apparently healthy and farmer's saved seed were treated with hot water at 52°C separately for 5, 15, 30, 60 minutes respectively. After treating seeds the excess water was removed and the seeds were air dried in the laboratory prior to placing in the Petridishs.

3. 5.7. Seed treatment with plant extracts

Botanicals were collected from different places (Plate 3-7). Seeds of black cumin were collected from the Agargaon market, Tejgaon, Dhaka. Barmuda grass (Durba), leaves of Neem and Allamanda were collected from Sher-e-Bangla Agricultural University campus. And Rhizomes of nut sedge (Mutha) were collected from village-Sharpati, P.S.-Chowddagram, District-Comilla. The extracts were prepared by using the method of Hossain *et al.* (2005). For preparation of extracts, collected leaves, seeds and rhizomes were weighted in an electric balance and then washed in water. After washing the big leaves and rhizomes were cut into small pieces. For getting extract, weighted plant parts were blended in an electric blender and then distilled water was added into the jug of the blender.

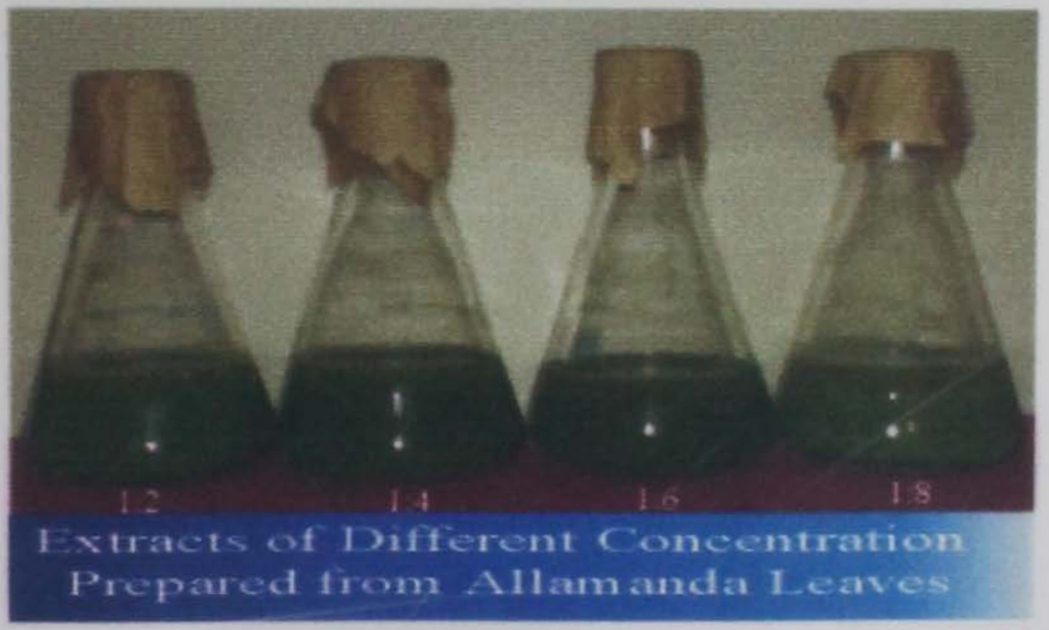
For getting 1:2 (w/v) ratios, 200 ml of distilled water was added with 100 g plant parts. Similarly to get 1:4 (w/v), 1:6 (w/v) ratios and 1:8 (w/v) ratios, 400, 600 and 800 ml distilled water was added in 100g plant parts respectively (Plate 3-7).

The particulars of the botanicals used for the experiment are cited below:

Common name	Botanical name	Plant parts Used	Concentration
Allamanda	*Allamanda cathartica*	Leaf	1:2, 1:4, 1:6, 1:8 (W/V)
Neem	*Azadirachta indica*	Leaf	1:2, 1:4, 1:6, 1:8 (W/V)
Kalizira	*Nigella sativa*	Seed	1:2, 1:4, 1:6, 1:8 (W/V)
Burmuda grass (Durba)	*Cynodon dectylon*	Whole plant	1:2, 1:4, 1:6, 1:8 (W/V)
Nut sedge (Mutha)	*Cyperus rotundus*	Rhizome	1:2, 1:4, 1:6, 1:8 (W/V)

Seeds were treated by following dipping method separately in different extracts. Different plant extracts with different concentrations were taken in different sterilized beaker as per requirement. Then 400 seeds were dipped in the solution for 1hour. The treated seeds were then taken out of the extract and kept in blotting paper to remove excess moisture from seed surface prior to placing in the Petridishs.

A



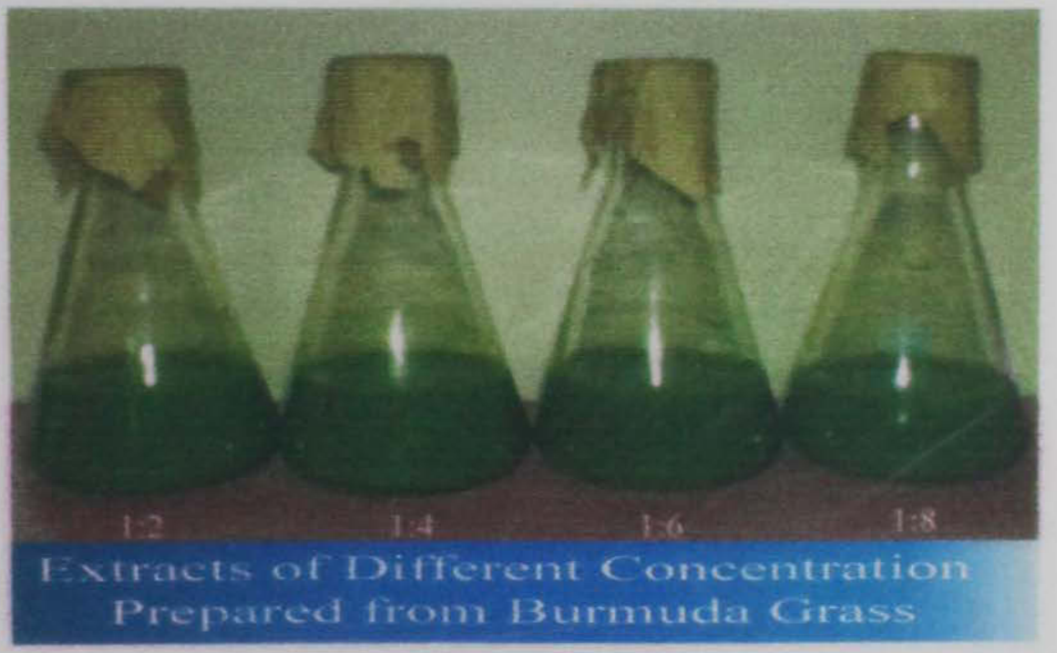
B

Plate 3.

A. Allamanda leaves

B. Extracts prepared from Allamanda leaves

A



B

Plate 4.

A. Barmuda grass

B. Extracts prepared from Barmuda grass

A

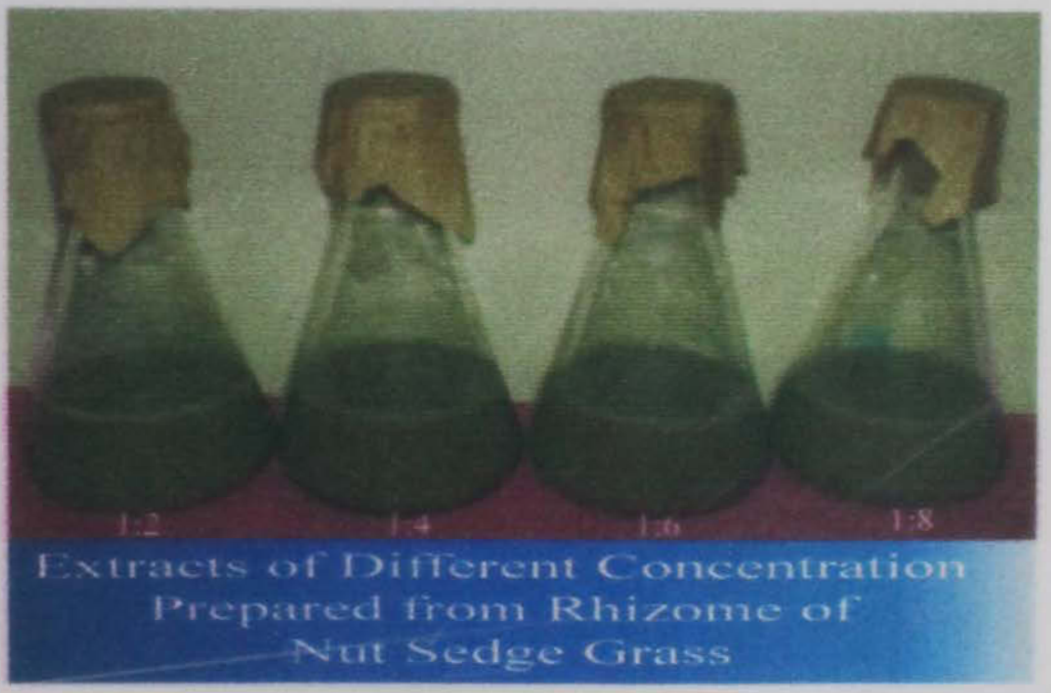
B

Plate 5.

A. Seeds of Black cumin

B. Extracts prepared from Black cumin seeds

A



B

Plate 6.

A. Rhizome of Nut sedge grass
B. Extracts prepared from Nut sedge grass rhizomes

A

B

Plate 7.

A. Neem leaves

B. Extracts prepared from Neem leaves

3.6. Seed health study of treated seed

Seed health study was done following the standard method of ISTA (1999). In this method, three layers of blotting paper (whatman filter paper no.l) soaked in sterilized water and were placed at the base of a 9 cm diameter plastic petridish and there after 25 seeds were placed on filter paper maintaining equal distance and covered with the lid. The petridishes containing seeds were incubated at 25±1 °C for 7 days maintaining 12 /12hr alternative cycle of NUV light and darkness in the Laboratory. Time to time watering was done to keep the filter paper moist.

3.7. Detection of seed-borne *Bipolaris sorokiniana*

After 7days of incubation the incubated seeds were examined under stereo binocular microscope in order to record the incidence of *Bipolaris sorokiniana* (plate-8). Germination of the seeds was also recorded. Identification of *Bipolaris sorokiniana* under the stereoscopic binocular microscope was confirmed by preparing temporary slides and examined under the compound microscope with the help of relevant taxonomic books (Booth, 1971 and Ellis 1971).

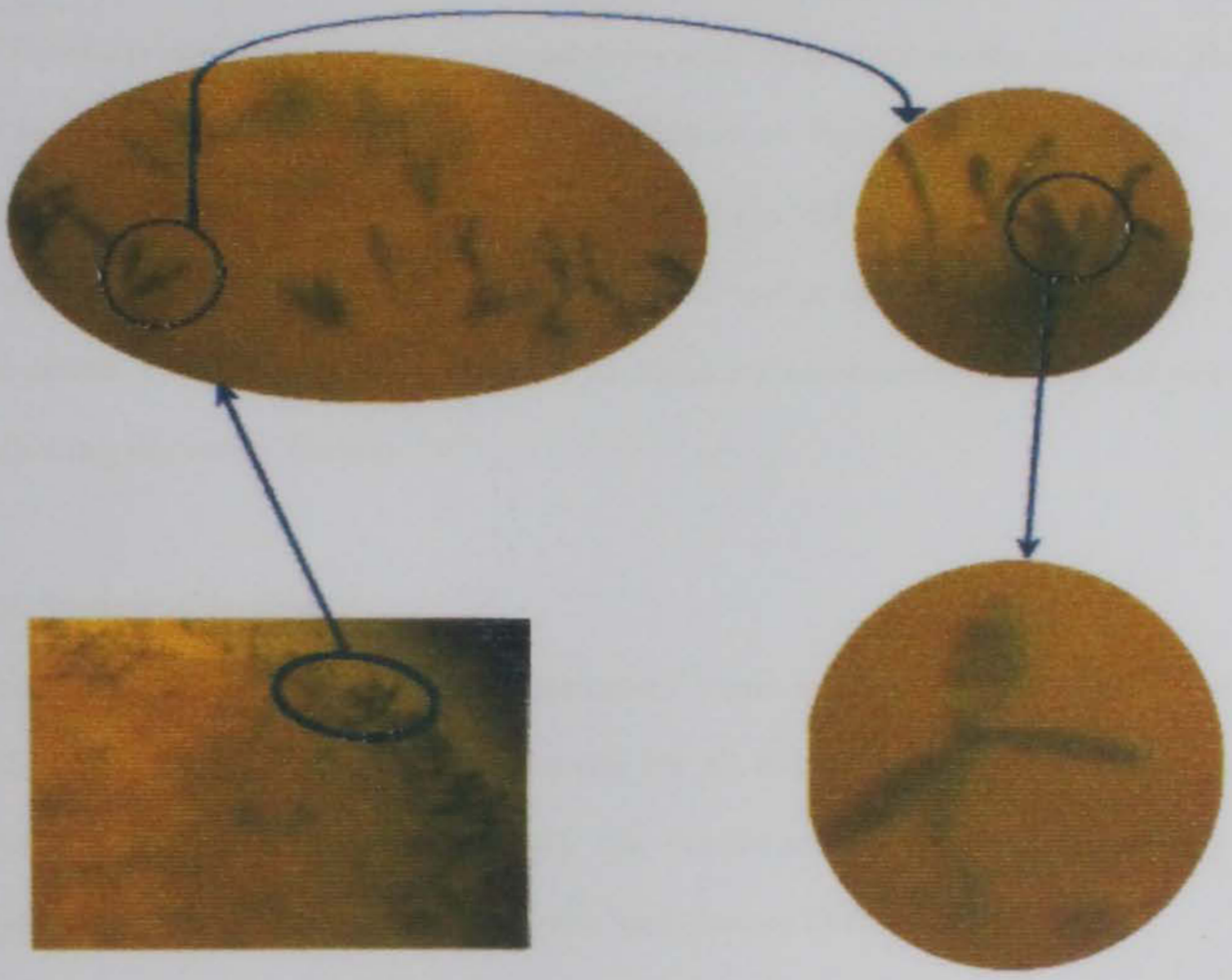


Plate 8.

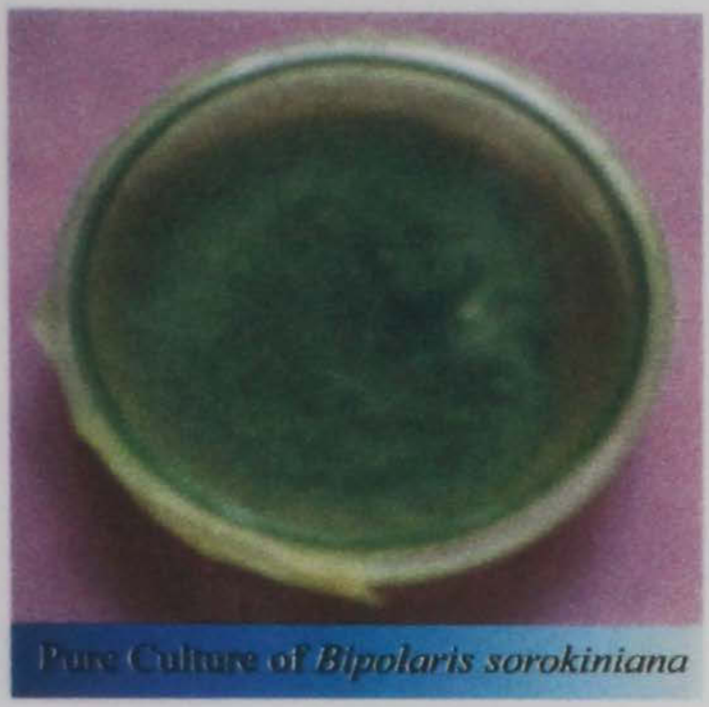
Habit character of *Bipolaris sorokiniana* on incubated wheat under stereomicroscope (45X)

3.8. Isolation and identification of pathogen

The seeds were plated on moistened filter paper and incubated at 25±1^{0}C for 7 days. After incubation seeds were observed under stereo binocular microscope. The conidia of *Bipolaris sorokiniana* were removed from seed surface by needle and were placed on to PDA media in petridish inside the laminar air flow in aseptic condition. The plates were then incubated at 25±1°C for 7days. Later the pathogen was purified using hyphal tip culture method and grown on PDA media at 25+1°C for 2 weeks and identified. The pathogen was identified as *Bipolaris sorokiniana* (plate 8 and plate 9) following the key of Brooth, 1971.

3.9. Statistical analysis:

The collected data on different parameters were analyzed statistically by using MSTAT-C package program. The means for all the treatments were compared by DMRT (Duncan Multiple Range Test). The significance of the difference among the means was calculated by LSD test (Least Significance Difference).



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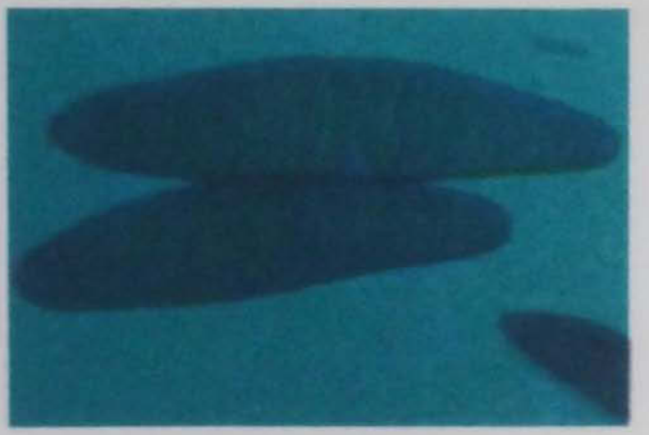
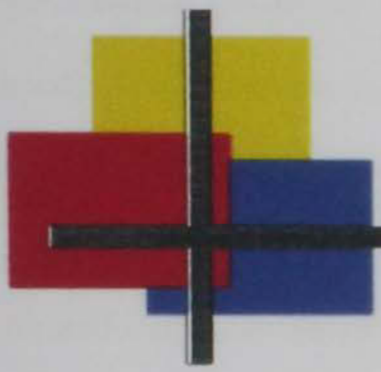


B Conidia of *Bipolaris sorokiniana*

Plate 9.

A. Pure culture of *Bipolaris sorokiniana*
B. Conidia of *Bipolaris sorokiniana* (100X) under compound microscope



Chapter 4
Results

4. RESULT

4.1. EVALUATION OF FUNGICIDE AGAINST *Bipolaris sorokiniana*

4.1.1. Effect of seed treatment with fungicides on germination of farmer's saved seed

Seed treatment with fungicides differed significantly in respect of seed germination of wheat (Table 1, Fig. 1). The highest seed germination (92.33%) was counted in the treatment T_4 (Farmer's saved seed treated with Tilt 250EC @ 0.2%) followed by T_5 (Farmer's saved seed treated with Rovral 50WP @ 0.2%) and the lowest seed germination (74.63%) was counted in the treatment T_1 (Untreated farmer's saved seed). The treatment T_2 (Farmer's saved seed treated with Cupravit 50WP @ 0.7%) and T_6 (Farmer's saved seed treated with Bavistin 50WP @ 0.2%) were statistically similar in respect of seed germination followed by treatment T_3 (Farmer's saved seed treated with Ridomil gold MZ 68 WP @ 0.2%) but they differed significantly from T_1 (Untreated farmer's saved seed).

4.1.2. Effect of seed treatment with fungicides on germination of apparently healthy seed

The treatments differed significantly in terms of percent seed germination of apparently healthy seeds (Table 1, Fig. 1). The highest seed germination (94.33%) was counted in the treatment T_4 and T_5 and the lowest seed germination (80.33%) was counted in the treatment T_1 (Untreated apparently healthy seed). The treatment T_2, T_3

and T_6 were statistically similar in respect of seed germination but they differed significantly from T_1 (Untreated apparently healthy seed).

4.1.3. Effect of seed treatment with fungicides on incidence of *Bipolaris sorokiniana* on farmer's saved seed

Different treatments differed significantly in terms of incidence of *Bipolaris sorokiniana* (Table 1, Fig. 2). All the fungicides reduced the incidence of *Bipolaris sorokiniana* over control. The highest incidence of *Bipolaris sorokiniana* was noted (10.50%) under the treatment T_1 (Untreated farmer's saved seed) followed by T_5. The lowest incidence was noted (3.50%) in the treatment T_3. The effect of treatments T_2 and T_6 were statistically similar.

4.1.4. Effect of seed treatment with fungicides on incidence of *Bipolaris sorokiniana* on apparently healthy seed

The treatments differed significantly in terms of incidence of seed borne *Bipolaris sorokiniana* (Table 1, Fig. 2). All the fungicides reduced the incidence of *Bipolaris sorokiniana* over control. The highest incidence of *Bipolaris sorokiniana* was noted (8.50%) under the treatment T_1 followed by T_2 and T_6 .The lowest incidence was noted (2.33%) under the treatment T_3. The treatment T_4 and T_5 were statistically identical but they differed significantly from T_1 (Untreated apparently healthy seed).

Table 1. Effect of chemical seed treatment on germination and incidence of *Bipolaris sorokiniana* of wheat

Treatments	% Germination		% Incidence of *Bipolaris sorokiniana*	
	Farmer's saved seed	Apparently healthy seed	Farmer's saved seed	Apparently healthy seed
T_1= Untreated control	74.63e	80.33d	10.50a	8.50a
T_2= Cupravit 50 WP @ 0.7%	87.50c	90.33b	5.50c	3.33b
T_3= Ridomil gold MZ 68 WP @ 0.2%	85.17d	88.17c	3.50e	2.33d
T_4= Tilt 250 EC @ 0.2%	92.33a	94.33a	4.50d	2.73c
T_5= Rovral 50 WP @ 0.2%	89.50b	94.33a	6.50b	2.50c
T_6= Bavistin 50 WP @ 0.2%	87.33c	89.17bc	5.33c	3.167b
LSD (P 0.05)	0.7523	1.194	0.774	0.7022

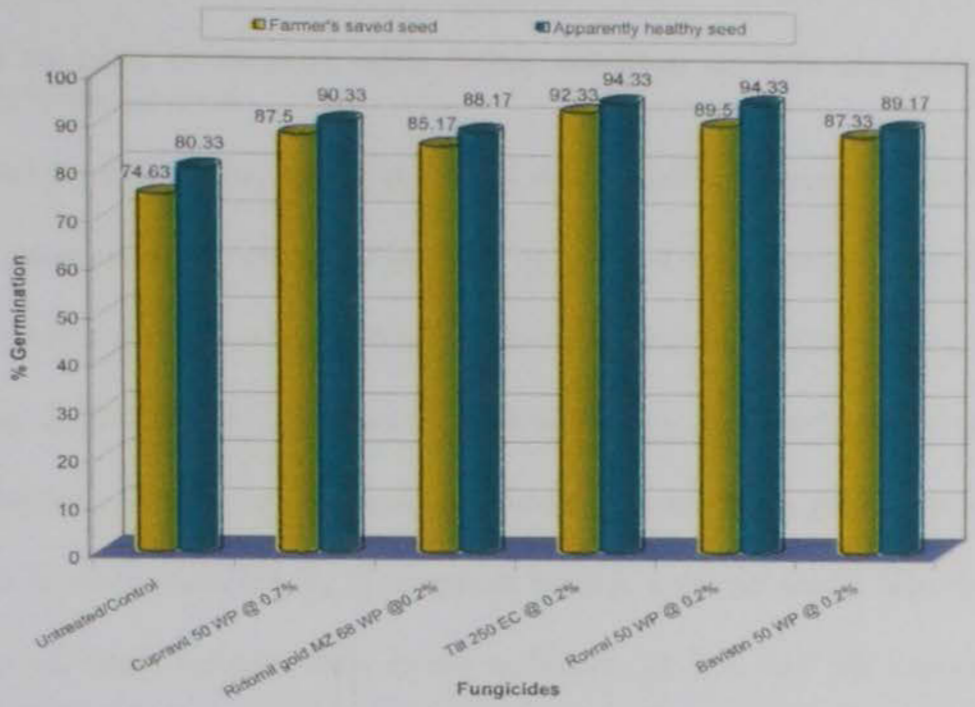


Fig 1. Effect of chemical seed treatments on farmer's saved seeds and apparently healthy seeds

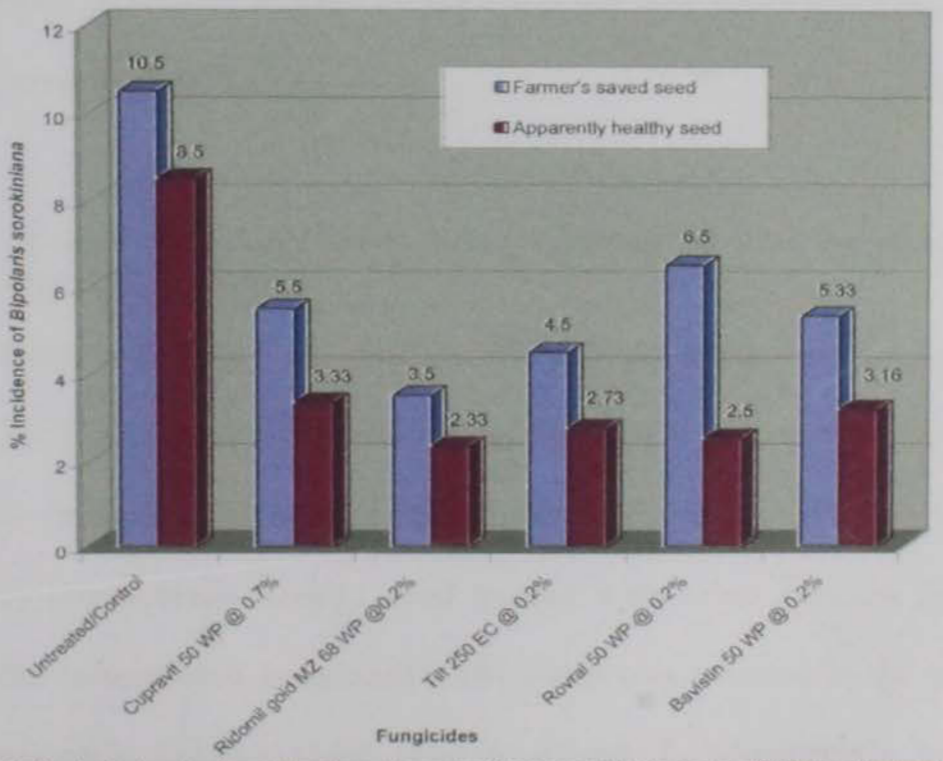


Fig 2. Effect of chemical seed treatments on incidence of *Bipolaris sorokiniana* of farmer's saved seeds and apparently healthy seeds

4.2. EVALUATION OF BRINE SOLUTION AGAINST *Bipolaris sorokiniana*

4.2.1. Effect of seed treatment with brine solution @ 2% on germination of farmer's saved seed

The performance of the brine solution @ 2% was significant over control in terms of percent seed germination (Table 2, Fig. 3). The highest seed germination (91.00%) of farmers saved seed was recorded in the treatment T_3 (Farmer's saved seed treated with brine solution @ 2% for 15 minutes) followed by T_2 (Farmer's saved seed treated with brine solution @ 2% for 5 minutes) and the lowest seed germination (82.00%) was recorded in the treatment T_1 (Untreated farmer's saved seed). The treatment T_4 (Farmer's saved seed treated with brine solution @ 2% for 30 minutes) and T_5 (Farmer's saved seed treated with brine solution @ 2% for 60 minutes) were statistically similar in respect of seed germination but they differed significantly from T_1 (Untreated farmer's saved seed).

4.2.2. Effect of seed treatment with brine solution @ 2% on germination of apparently healthy seed

The effect of brine solution @ 2% was significant over control on seed germination (Table 2, Fig. 3). The highest seed germination (94.17%) was counted in the treatment T_3 (Apparently healthy seed treated with brine solution @ 2% for 15 minutes) followed by T_2 (Apparently healthy seed treated with brine solution @ 2% for 5 minutes) and the lowest seed germination (84.00%) was counted in the treatment T_1 (Untreated apparently healthy seed). The treatment T_4 (Apparently healthy seed treated with brine solution @ 2% for 30 minutes) and T_5 (Apparently healthy seed

treated with brine solution @ 2% for 60 minutes) also differed significantly in respect of seed germination.

4.2.3. Effect of seed treatment with brine solution (2%) on incidence of *Bipolaris sorokiniana* on farmer's saved seed

Different treatments differed significantly in respect to incidence of seed borne *Bipolaris sorokiniana* (Table 2, Fig. 4). Brine solution (2%) reduced the incidence of *Bipolaris sorokiniana* over control. The highest incidence of *Bipolaris sorokiniana* was counted (46.00%) under the treatment T_1 (Untreated farmer's saved seed) followed by T_2. The lowest incidence was counted (19.33%) in the treatment T_5. The treatment T_3 and T_4 also differed significantly over control.

4.2.4. Effect of seed treatment with brine solution (2%) on incidence of *Bipolaris sorokiniana* on apparently healthy seed

Different treatments differed significantly in terms of incidence of seed borne *Bipolaris sorokiniana* (Table 2, Fig. 4). Brine solution reduced the incidence of *Bipolaris sorokiniana* in apparently healthy seeds over control. The highest incidence of *Bipolaris sorokiniana* was counted (39.00%) under the treatment T_1 (Untreated apparently healthy seed) followed by T_2 (30.83%). Significantly lowest incidence was counted and in the treatment T_4 (13.00%), T_3 (14.00%) and T_5 (14.50%) respectively.

Table 2. Effect of seed treatment with brine solution on germination and incidence of *Bipolaris sorokiniana* of wheat

	Treatments		% Germination		% Incidence of *Bipolaris sorokiniana*	
			Farmer's saved seed	Apparently healthy seed	Farmer's saved seed	Apparently healthy seed
T_1=	Untreated control		82.00d	84.00e	46.00a	39.00a
T_2=	Brine solution	5 min	89.50b	92.00b	42.50b	30.83b
T_3=	Brine solution	15 min	91.00a	94.17a	36.50c	14.00c
T_4=	Brine solution	30 min	83.00c	87.00d	28.17d	13.00c
T_5=	Brine solution	60 min	83.50c	89.33c	19.33e	14.50c
	LSD (P 0.05)		1.929	2.231	1.216	4.889

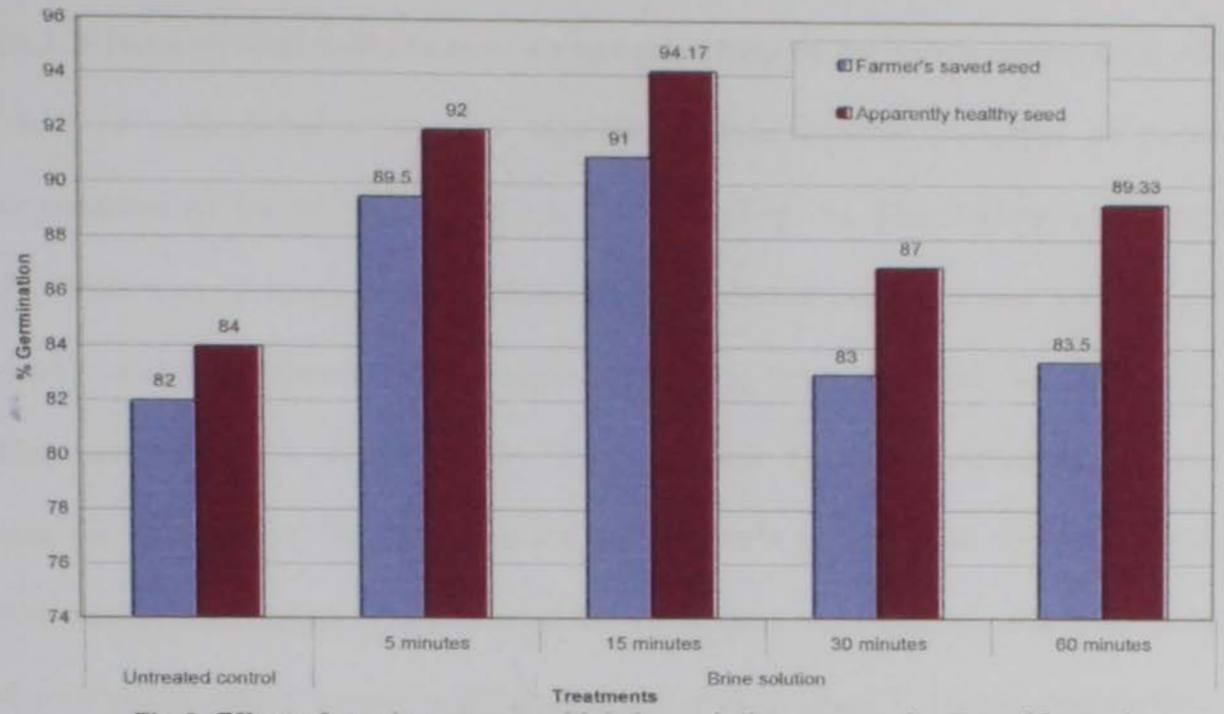


Fig 3. Effect of seed treatment with brine solution on germination of farmer's saved seeds and apparently healthy seeds

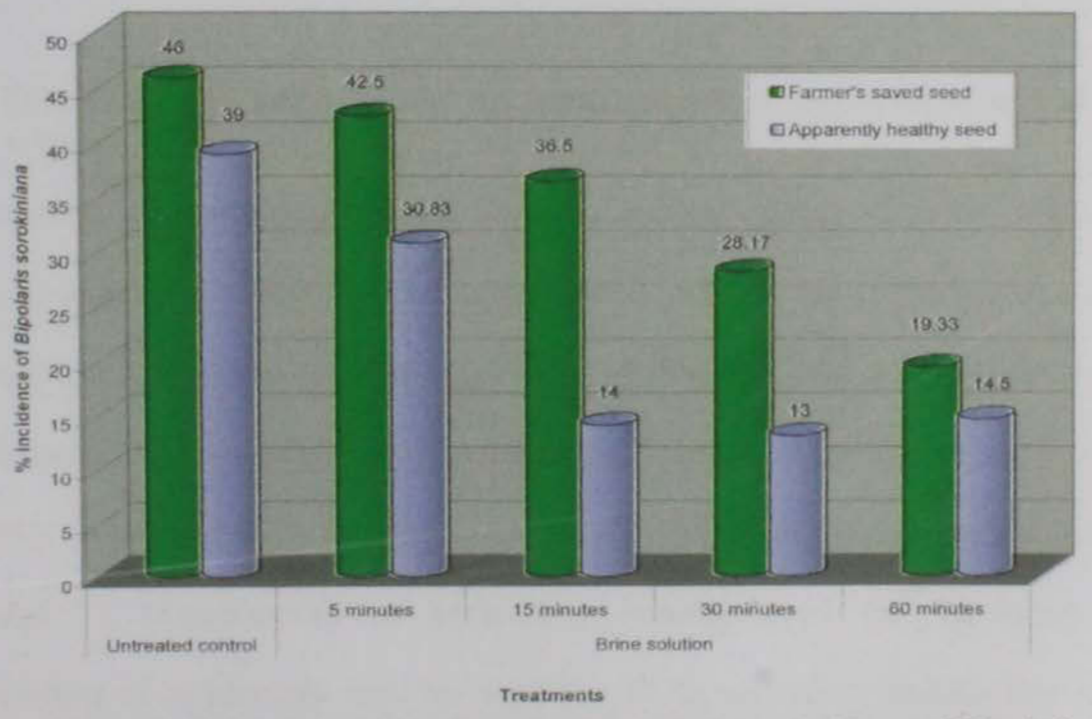


Fig 4. Effect of seed treatment with brine solution on incidence of *Bipolaris sorokiniana* of farmer's saved seeds and apparently healthy seeds

4.3. EVALUATION OF SEED SOLARIZATION AGAINST *Bipolaris sorokiniana*

4.3.1. Effect of seed solarization on germination of farmer's saved seed of wheat

Effect of seed solarization was significant over control in terms of percent seed germination of farmer's saved seeds (Table 3, Fig. 5). The highest seed germination (91.50%) was counted in the treatment T_2 (Solarization of farmer's saved seed for 5 hours) and the lowest germination (81.67%) was counted in the treatment T_1 (Untreated farmer's saved seed). The treatment T_5 (Solarization of farmer's saved seed for 20 hours), T_4 (Solarization of farmer's saved seed for 15 hours) and T_3 (Solarization of farmer's saved seed for 10 hours) were statistically similar in respect of seed germination but they differed significantly from T_1 (Untreated farmer's saved seed).

4.3.2. Effect of seed solarization on germination of apparently healthy seed of wheat

The effect of seed solarization was significant over control in terms of percent seed germination (Table 3, Fig. 5). The highest seed germination (99.33%) was counted in the treatment T_2 (Solarization of apparently healthy seed for 5 hours) followed by T_5 (Solarization of apparently healthy seed for 20 hours) and the lowest seed germination (90.00%) was counted in the treatment T_1 (Untreated apparently healthy seed). The treatment T_3 (Solarization of apparently healthy seed for 10 hours) and T_4 (Solarization of apparently healthy seed for 15 hours) were statistically similar but differed significantly over control in terms of seed germination.

4.3.3. Effect of seed solarization on incidence of *Bipolaris sorokiniana* on farmer's saved seeds of wheat

The treatments differed significantly in respect of incidence of seed borne *Bipolaris sorokiniana* (Table 3, Fig. 6). Seed solarization reduced the incidence of *Bipolaris sorokiniana* over control. The highest incidence of *Bipolaris sorokiniana* was counted (47.00%) under the treatment T_1 (Untreated farmer's saved seed). The lowest incidence was noted (7.00%) in the treatment T_4. The treatment T_2, T_3 and T_5 also differed significantly over control.

4.3.4. Effect of seed solarization on incidence of *Bipolaris sorokiniana* on apparently healthy seeds of wheat

Different treatment differed significantly in respect to incidence of seed borne *Bipolaris sorokiniana* (Table 3, Fig. 6). Seed solarization reduced the incidence of *Bipolaris sorokiniana* over control. The highest incidence of *Bipolaris sorokiniana* was recorded (39.00%) under the treatment T_1 (Untreated apparently healthy seed). The lowest incidence was observed (6.00%) in the treatment T_4. The effect of treatments T_2, T_3 and T_5 also differed significantly over control.

Table 3. Effect of seed treatment by sun drying on germination and incidence of *Bipolaris sorokiniana* of wheat

Treatments	% Germination		% Incidence of *Bipolaris sorokiniana*	
	Farmer's saved seed	Apparently healthy seed	Farmer's saved seed	Apparently healthy seed
T_1= Untreated control	81.67b	90.00b	47.00a	39.00a
T_2= Sun drying 5 hours	91.50a	99.33a	13.50b	13.00b
T_3= Sun drying 10 hours	85.33ab	92.50b	12.50b	11.50b
T_4= Sun drying 15 hours	87.00ab	93.00b	7.00d	6.00d
T_5= Sun drying 20 hours	87.17ab	98.00a	10.33c	8.50c
LSD (P 0.05)	7.116	3.455	3.820	3.234

Fig 5. Effect of sundrying on germination of farmer's saved seeds and apparently healthy seeds

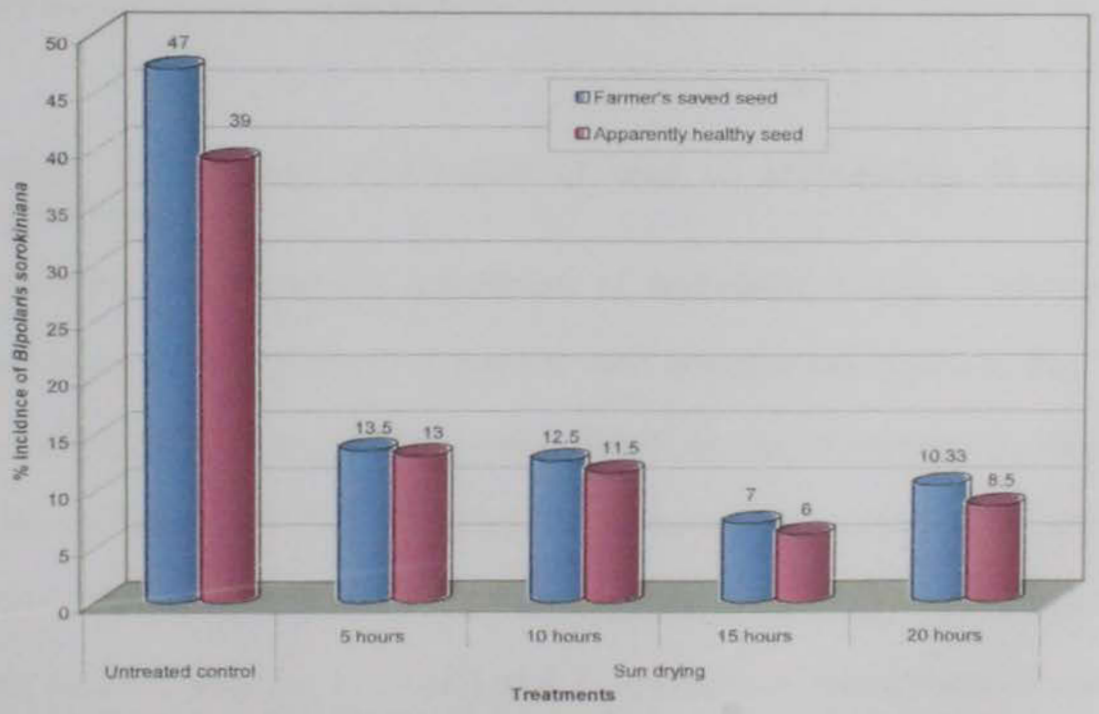


Fig 6. Effect of sundrying on incidence of *Bipolaris sorokiniana* of farmer's saved seeds and apparently healthy seeds

4.4. EVALUATION OF SEED SOLARIZATION COVERED WITH POLYTHENE AGAINST *Bipolaris sorokiniana*

4.4.1. Effect of polythene solarization on germination of farmer's saved seed

Effect of polythene solarization of seed was found significant over control in terms of percent seed germination (Table 4, Fig. 7). The highest seed germination (93.17%) was noted in the treatment T_2 (Polythene solarization of farmer's saved seed for 5 hours) and the lowest seed germination (82.00%) was observed in the treatment T_1 (Untreated farmer's saved seed). The treatment T_5 (Polythene solarization of farmer's saved seed for 20 hours), T_4 (Polythene solarization of farmer's saved seed for 15 hours) and T_3 (Polythene solarization of farmer's saved seed for 10 hours) were statistically indifferent in respect of seed germination but they differed significantly from T_1 (Untreated farmer's saved seed).

4.4.2. Effect of polythene solarization of seed on germination of apparently healthy seed

The performance of polythene solarization of apparently healthy seed was found significant over control in terms of percent seed germination (Table 4, Fig. 7). The highest seed germination (99.00%) was noted in the treatment T_2 (Polythene solarization of apparently healthy seed for 5 hours) followed by T_5 (Polythene solarization of apparently healthy seed for 20 hours), T_4 (Polythene solarization of apparently healthy seed for 15 hours) and T_3 (Polythene solarization of apparently healthy seed for 10 hours). The lowest seed germination (90.00%) was noted in the treatment T_1 (Untreated apparently healthy seed).

4.4.3. Effect of polythene solarization of seed on incidence of *Bipolaris sorokiniana* on farmer's saved seed

The treatments differed significantly in respect to incidence of seed borne *Bipolaris sorokiniana* (Table 4 and Fig. 8). Polythene solarization of seed reduced the incidence of *Bipolaris sorokiniana* over control. The highest incidence of *Bipolaris sorokiniana* was recorded (46.00%) under the treatment T_1 (Untreated farmer's saved seed). The lowest incidence was noted (12.50%) under the treatment T_2. The treatment T_5, T_3 and T_4 are also statistically identical but differed significantly over control.

4.4.4. Effect of polythene solarization of seed on incidence of *Bipolaris sorokiniana* on apparently healthy seed

The treatments differed significantly in terms of incidence of seed borne *Bipolaris sorokiniana* (Table 4, Fig. 8). Polythene solarization of seed resulted lower incidence of *Bipolaris sorokiniana* over control. The highest incidence of *Bipolaris sorokiniana* was recorded (39.00%) under the treatment T_1 (Untreated apparently healthy seed). The lowest incidence was counted (5.00%) under the treatment T_4. The treatment T_2, T_3 are statistically identical and differed significantly over control. The treatment T_5 also differed significantly over control.

Table 4. Effect of seed treatment by polythene solarization on germination and incidence of *Bipolaris sorokiniana* of wheat

Treatments	% Germination		% Incidence of *Bipolaris sorokiniana*	
	Farmer's saved seed	Apparently healthy seed	Farmer's saved seed	Apparently healthy seed
T_1= Untreated control	82.00c	90.00e	46.00a	39.00a
T_2= Polythene solarization 5 hrs	93.17a	99.00a	12.50c	7.00bc
T_3= Polythene solarization 10 hrs	86.50b	92.00d	15.50bc	7.00bc
T_4= Polythene solarization 15 hrs	87.50b	94.00c	13.00bc	5.00c
T_5= Polythene solarization 20 hrs	88.17b	96.00b	16.50b	8.50b
LSD (P 0.05)	4.446	2.793	3.730	2.793

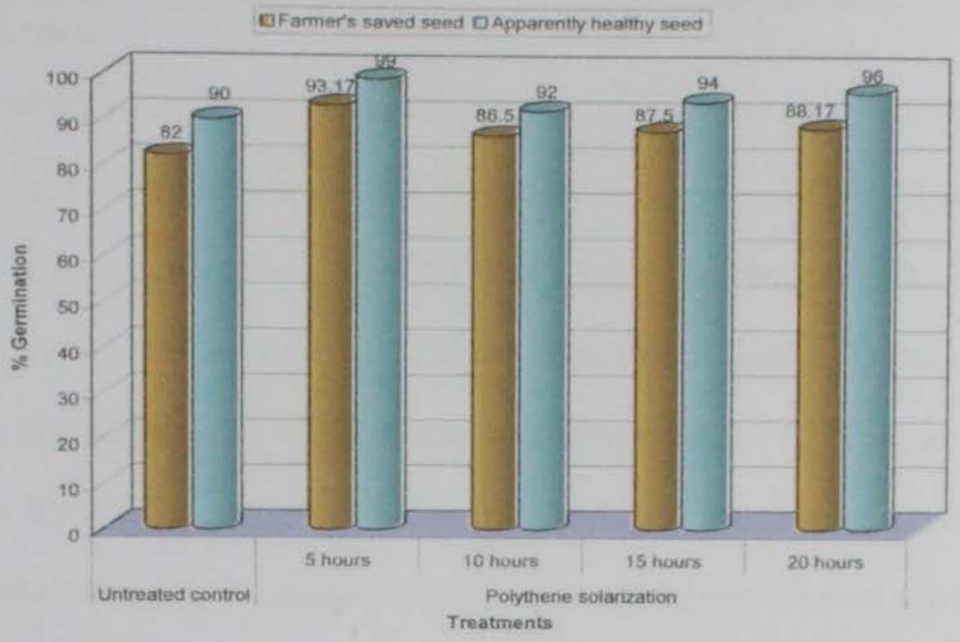


Fig 7. Effect of seed solarization covered with polythene on germination of farmer's saved seeds and apparently healthy seeds

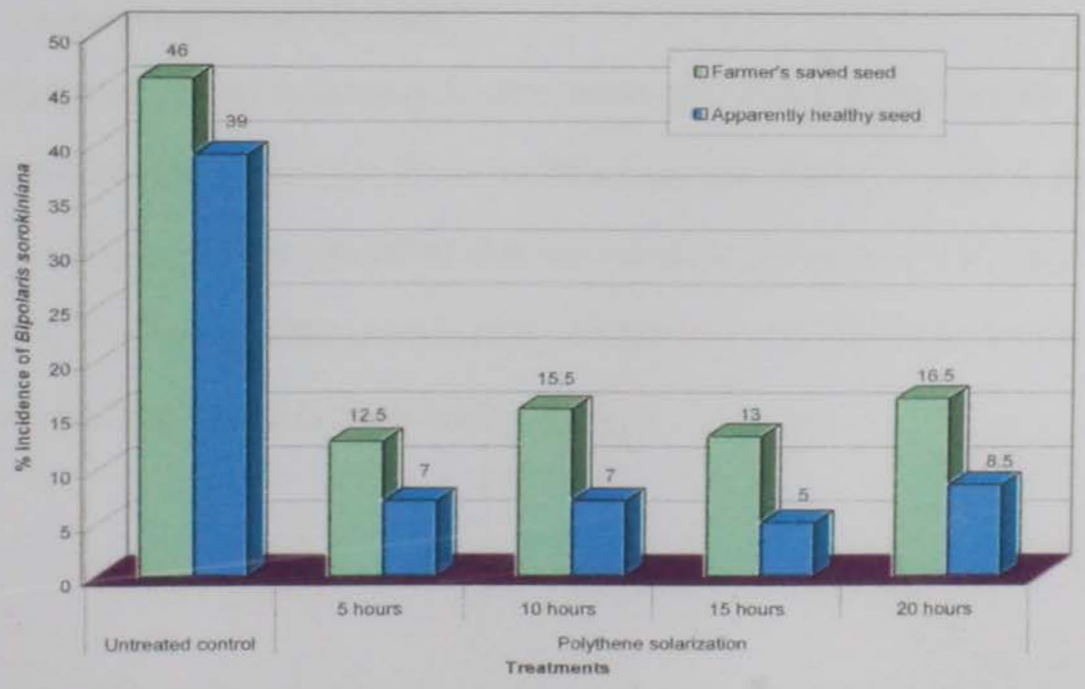


Fig 8. Effect of seed solarization covered with polythene on incidence of *Bipolaris sorokiniana* of farmer's saved seeds and apparently healthy seeds

4.5. EVALUATION OF HOT WATER (52°C) AGAINST *Bipolaris sorokiniana*

4.5.1. Effect of seed treatment with hot water (52°C) on germination of farmer's saved seed

% Seed germination of farmer's saved wheat seed differed significantly in terms of seed treatment with hot water (52°C) for different period of time (Table 5, Fig. 9). The highest seed germination (92.50%) was counted in the treatment T_5 (Farmer's saved seed treated with hot water at 52°C for 20 minutes) and T_2 (Farmer's saved seed treated with hot water at 52°C for 5 minutes) followed by T_3 (Farmer's saved seed treated with hot water at 52°C for 10 minutes) and the lowest seed germination (82.00%) was counted in the treatment T_1 (Untreated farmer's saved seed).

4.5.2. Effect of seed treatment with hot water (52°C) on germination of apparently healthy seed

% Seed germination of apparently healthy seeds also differed significantly due to seed treatment with hot water (52°C) for different period of time (Table 5, Fig. 9). The highest seed germination (99.00%) was recorded in the treatment T_3 followed by T_2, T_4 and T_5. The lowest seed germination (90.00%) was observed under the treatment T_1 (Untreated apparently healthy seed).

4.5.3. Effect of seed treatment with hot water (52°C) on incidence of *Bipolaris sorokiniana* on farmer's saved seeds of wheat

The incidence of seed borne *Bipolaris sorokiniana* differed significantly in terms of seed treatment with hot water of different period of time (Table 5, Fig. 10). Hot water seed treatment reduced the incidence of *Bipolaris sorokiniana* over control. The highest incidence of *Bipolaris sorokiniana* was noted (46.0%) under the treatment T_1 (Untreated farmer's saved seed) and the lowest incidence was recorded (6.0%) in the treatment T_5 followed by T_4. The treatment T_2 and T_3 also differed significantly over control.

4.5.4. Effect of seed treatment with hot water (52°C) on incidence of *Bipolaris sorokiniana* on apparently healthy seeds of wheat

The treatments differed significantly in terms of incidence of seed borne *Bipolaris sorokiniana* (Table 5, Fig. 10). Hot water seed treatment reduced the incidence of *Bipolaris sorokiniana* over control. The highest incidence of *Bipolaris sorokiniana* was recorded (39.00 %) under the treatment T_1 (Untreated apparently healthy seed). The lowest incidence was noted (1.50%) under the treatment T_5 followed by T_4. The treatment T_3 and T_2 also differed significantly over control.

Table 5. Effect of seed treatment with hot water (52°C) on germination and incidence of *Bipolaris sorokiniana* of wheat

Treatments		% Germination		% Incidence of *Bipolaris sorokiniana*	
		Farmer's saved seed	Apparently healthy seed	Farmer's saved seed	Apparently healthy seed
T_1= Untreated control		82.00c	90.00b	46.00a	39.00a
T_2= Hot water (52°C)	5 min	92.50a	98.83a	15.50b	13.00b
T_3= Hot water (52°C)	10 min	90.17b	99.00a	15.50b	6.00c
T_4= Hot water (52°C)	15 min	83.00c	98.50a	7.00c	3.00d
T_5= Hot water (52°C)	20 min	92.50a	97.67a	6.00c	1.50d
LSD (P 0.05)		1.968	1.945	2.932	2.063

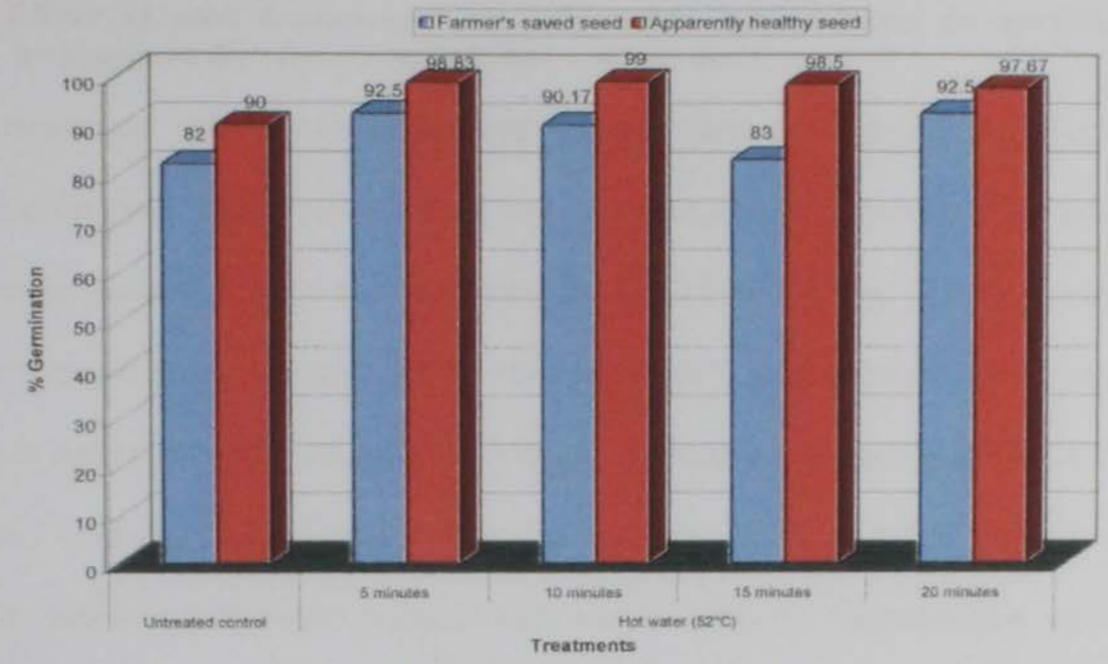


Fig 9. Effect of seed treatment by hot water at 52°C on germination of farmer's saved seeds and apparently healthy seeds

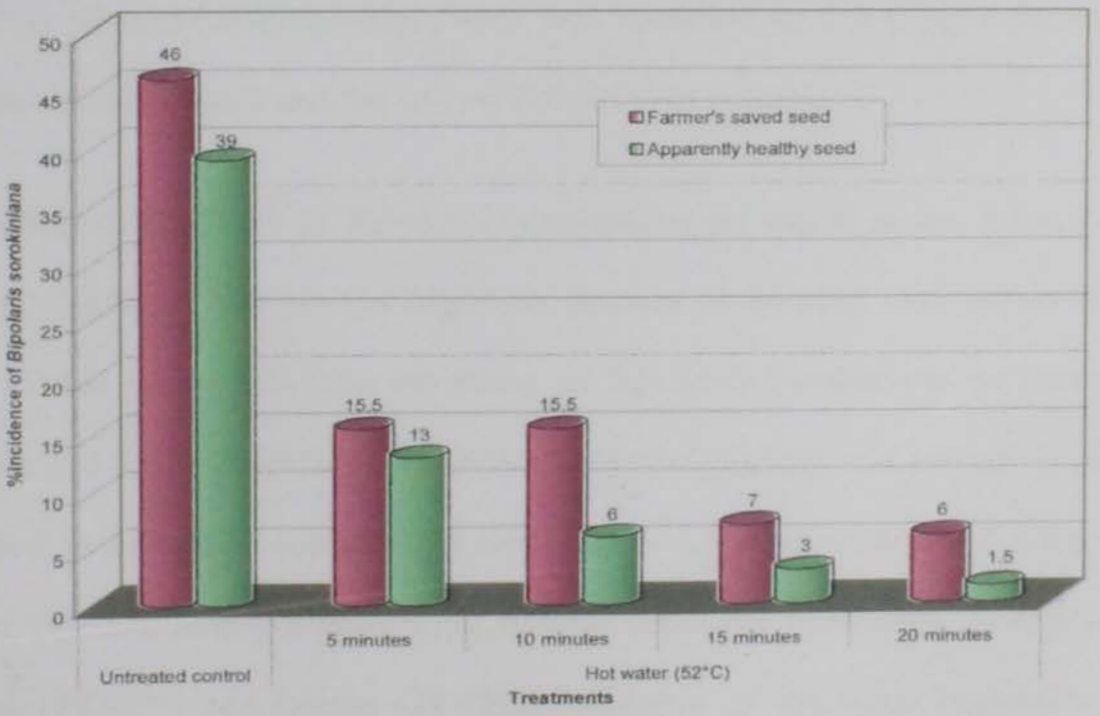


Fig 10. Effect of seed treatment with hot water at 52°C on incidence of *Bipolaris sorokiniana* of farmer's saved seeds and apparently healthy seeds

4.6. EVALUATION OF PLANT EXTRACTS AGAINST *Bipolaris sorokiniana*

4.6.1. Effect of seed treatment with Allamanda leaf extracts on germination and incidence of *Bipolaris sorokiniana* on wheat seed

Seed treatment with Allamanda leaf extract showed significant effect on seed germination and incidence of *Bipolaris sorokiniana* of farmer's saved seed and apparently healthy seeds over untreated control (Table 6, fig 11). % Germination of farmer's saved seed ranged from 76.50% to 82.50% in respect of seed treatment with different doses of Allamanda leaf extract. Highest seed germination (82.50%) was recorded in 1:4 ($^{W}/_{V}$) of Allamanda leaf extract and lowest (76.50%) in untreated control, where seeds were treated with water only. % Germination of apparently healthy seeds was found higher over farmers saved seed. The highest germination (87.67%) of apparently healthy seeds was recorded in 1:4 ($^{W}/_{V}$) concentrations of Allamanda leaf extract and the lowest (78.00%) in control.

Regarding % incidence of *Bipolaris sorokiniana* on wheat seeds, it has been found that the incidence was always higher in farmers saved seed over apparently healthy seeds (Table 6, Fig 12). The incidence of *Bipolaris sorokiniana* on farmer's saved seed ranged from 29.00% to 43.00% where the highest and lowest incidence was recorded in untreated control (T_1) and 1:2 ($^{W}/_{V}$) concentration of Allamanda leaf extract (T_2) similarly for apparently healthy seeds the treatments T_1 and T_2 resulted highest (39.43%) and lowest (24.00%) incidence of the fungi respectively. It was found that apparently healthy seeds had comparatively lower incidence of the fungi (Table 6, Fig 12) over farmer's saved seed.

Table 6. Effect of seed treatment with Allamanda leaf extract on germination and incidence of *Bipolaris sorokiniana* of wheat

Treatments		% Germination		% Incidence of *Bipolaris sorokiniana*	
		Farmer's saved seed	Apparently healthy seed	Farmer's saved seed	Apparently healthy seed
T_1= Untreated control		76.50c	78.00b	43.00a	39.43a
T_2= Allamanda leaf extract	1:2 ($^{W}/_{V}$)	82.07a	87.33a	29.00c	24.00c
T_3= Allamanda leaf extract	1:4 ($^{W}/_{V}$)	82.50a	87.67a	32.00b	27.33b
T_4= Allamanda leaf extract	1:6 ($^{W}/_{V}$)	79.00b	87.17a	32.50b	27.33b
T_5= Allamanda leaf extract	1:8 ($^{W}/_{V}$)	77.00c	86.17a	34.00b	29.00b
LSD (P 0.05)		1.718	4.311	1.975	3.789

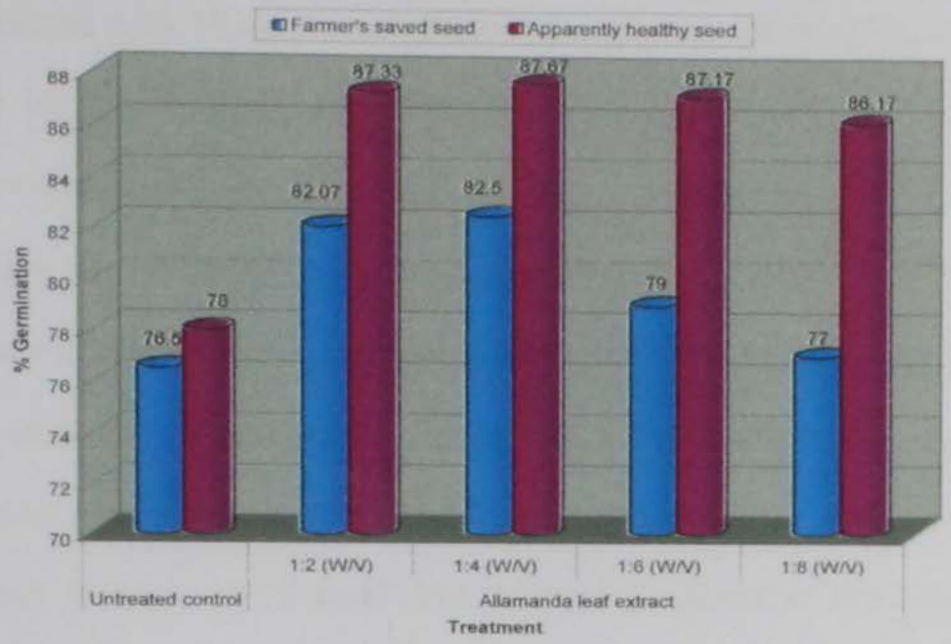


Fig 11. Effect of seed treatment with Allamanda leaf extract on germination of farmer's saved seeds and apparently healthy seeds

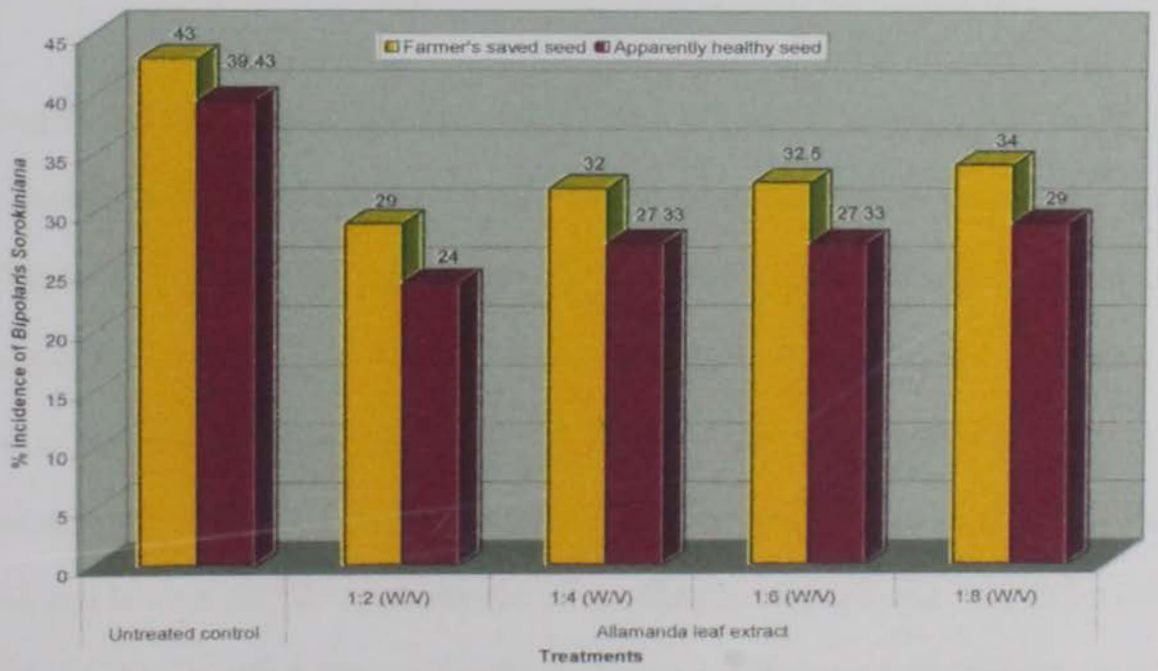


Fig 12. Effect of seed treatment with Allamanda leaf extract on incidence of *Bipolaris sorokiniana* of farmer's saved seeds and apparently healthy seeds

4.6.2. Effect of seed treatment with Neem leaf extract on germination and incidence of *Bipolaris sorokiniana* on wheat seed

Seed treatment with Neem extract showed significant effect on seed germination and incidence of *Bipolaris sorokiniana* of farmer's saved seed and apparently healthy seeds over untreated control (Table 7, fig 13). % Germination of farmer's saved seed ranged from 76.50% to 93.00% in respect of seed treatment with different doses of Neem leaf extract. Highest seed germination (93.00%) was recorded in 1:2 ($^{W}/_{V}$) and 1:4 ($^{W}/_{V}$) of Neem leaf extract and lowest (76.50%) in untreated control, where seeds were treated with water only. % Germination of apparently healthy seeds was found higher over farmers saved seed. The highest germination (99.33%) of apparently healthy seeds was recorded in 1:2 ($^{W}/_{V}$) concentrations of Neem leaf extract and the lowest (78.00%) in control.

Regarding % incidence of *Bipolaris sorokiniana* on wheat seeds, it has been found that the incidence was always higher in farmers saved seed over apparently healthy seeds (Table 7, Fig 14). The incidence of *Bipolaris sorokiniana* on farmer's saved seed ranged from 16.50% to 43.00% where the highest and lowest incidence was recorded in untreated control (T_1) and 1:2 ($^{W}/_{V}$) concentration of Neem leaf extract (T_2) similarly for apparently healthy seeds the treatments T_1 and T_2 resulted highest (39.43%) and lowest (14.00%) incidence of the fungus respectively. It was found that apparently healthy seeds had comparatively lower incidence of the fungus (Table 7, Fig 14) over farmer's saved seed.

Table 7. Effect of seed treatment with Neem leaf extract on germination and incidence of *Bipolaris sorokiniana* of wheat

Treatments		% Germination		% Incidence of *Bipolaris sorokiniana*	
		Farmer's saved seed	Apparently healthy seed	Farmer's saved seed	Apparently healthy seed
T_1= Untreated control		76.50c	78.00d	43.00a	39.43a
T_2= Neem leaf Extract	1:2 ($^{W}/_{V}$)	93.00a	99.33a	16.50c	14.00e
T_3= Neem leaf Extract	1:4 ($^{W}/_{V}$)	93.00a	99.00a	17.50c	15.50d
T_4= Neem leaf Extract	1:6 ($^{W}/_{V}$)	92.00a	97.00b	24.50b	20.33b
T_5= Neem leaf Extract	1:8 ($^{W}/_{V}$)	89.50b	90.00c	26.00b	19.50c
LSD (P 0.05)		2.139	1.566	2.063	0.367

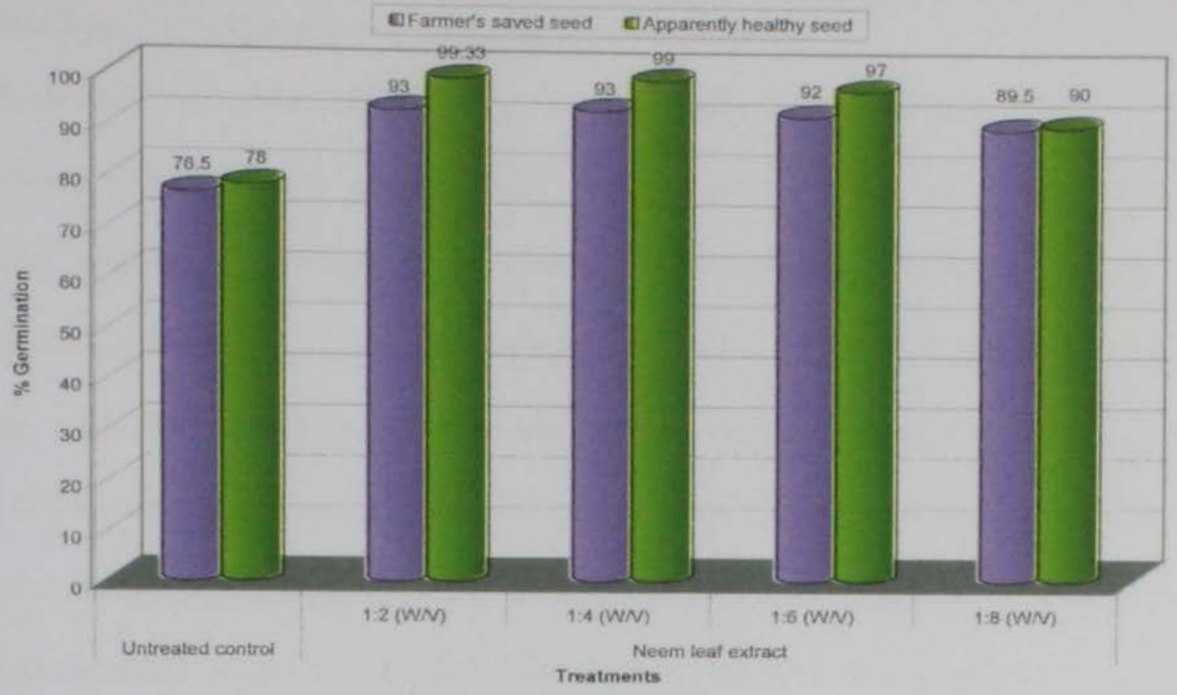


Fig 13. Effect of seed treatment with Neem leaf extract on germination of farmer's saved seeds and apparently healthy seeds

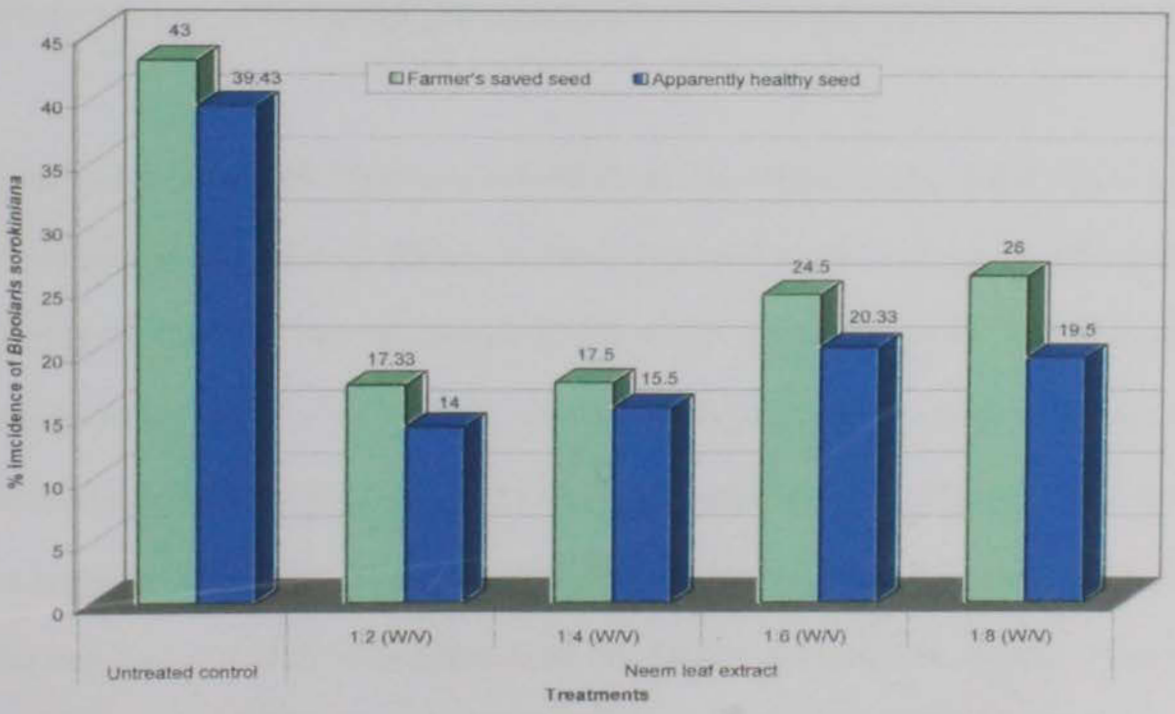


Fig 14. Effect of seed treatment with Neem leaf extract on incidence of *Bipolaris sorokiniana* of farmer's saved seeds and apparently healthy seeds

4.6.3. Effect of seed treatment with Nut sedge grass extract on germination and incidence of *Bipolaris sorokiniana* on wheat seed

Seed treatment with Nut sedge grass extract showed significant effect on seed germination and incidence of *Bipolaris sorokiniana* of farmer's saved seed and apparently healthy seeds of wheat over untreated control (Table 8, fig 15). % Germination of farmer's saved seed ranged from 76.50% to 93.00% in respect of seed treatment with different doses of Nut sedge grass extract. Highest seed germination (93.00%) was noted in 1:2 ($^{W}/_{V}$) of Nut sedge grass extract and lowest (76.50%) in untreated control, where seeds were treated with water only. % Germination of apparently healthy seeds was found higher over farmers saved seed. The highest germination (98.00%) of apparently healthy seeds was observed in 1:2 ($^{W}/_{V}$) and 1:4 ($^{W}/_{V}$) concentrations of Nut sedge grass extract and the lowest (78.00%) in control.

Regarding % incidence of *Bipolaris sorokiniana* on wheat seeds, it has been found that the incidence was always higher in farmers saved seed over apparently healthy seeds (Table 8, fig 16). The incidence of *Bipolaris sorokiniana* on farmer's saved seed ranged from 19.00% to 43.00% where the highest and lowest incidence was noted in untreated control (T_1) and 1:2 ($^{W}/_{V}$) concentration of Nut sedge grass extract (T_2) similarly for apparently healthy seeds the treatments T_1 and T_2 resulted highest (39.43%) and lowest (14.50%) incidence of the fungus respectively. It was found that apparently healthy seeds had comparatively lower incidence of the fungus (Table 8, fig 16) over farmer's saved seed.

Table 8. Effect of seed treatment with Nut sedge grass extract on germination and incidence of *Bipolaris sorokiniana* of wheat

Treatments			% Germination		% Incidence of *Bipolaris sorokiniana*	
			Farmer's saved seed	Apparently healthy seed	Farmer's saved seed	Apparently healthy seed
T_1=	Untreated control		76.50d	78.00d	43.00a	39.43a
T_2=	Nut sedge rhizome extract	1:2 ($^{W}/_{V}$)	93.00a	98.00a	19.00d	14.50e
T_3=	Nut sedge rhizome extract	1:4 ($^{W}/_{V}$)	90.00bc	98.00a	20.33d	15.50d
T_4=	Nut sedge rhizome extract	1:6 ($^{W}/_{V}$)	91.00b	95.00b	23.00c	20.50c
T_5=	Nut sedge rhizome extract	1:8 ($^{W}/_{V}$)	89.17c	91.00c	26.50b	25.00b
LSD (P 0.05)			1.702	0.842	1.702	0.2147

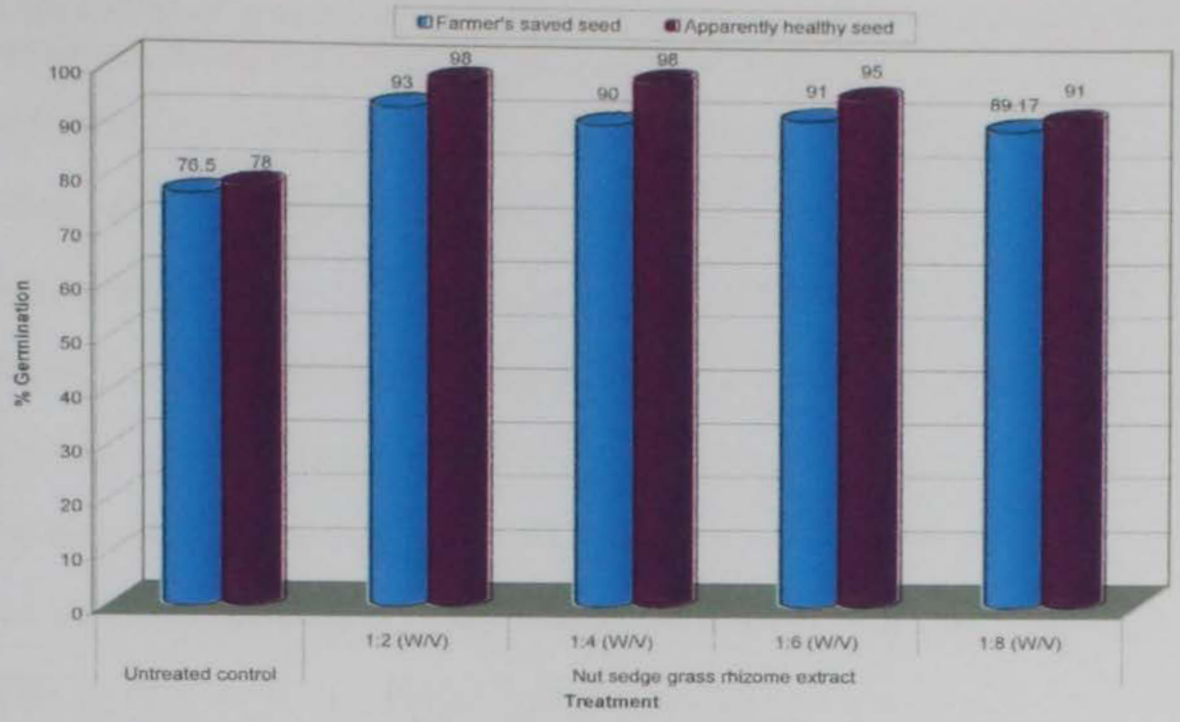


Fig 15. Effect of seed treatment with Nut sedge grass rhizome extract on germination of farmer's saved seeds and apparently healthy seeds

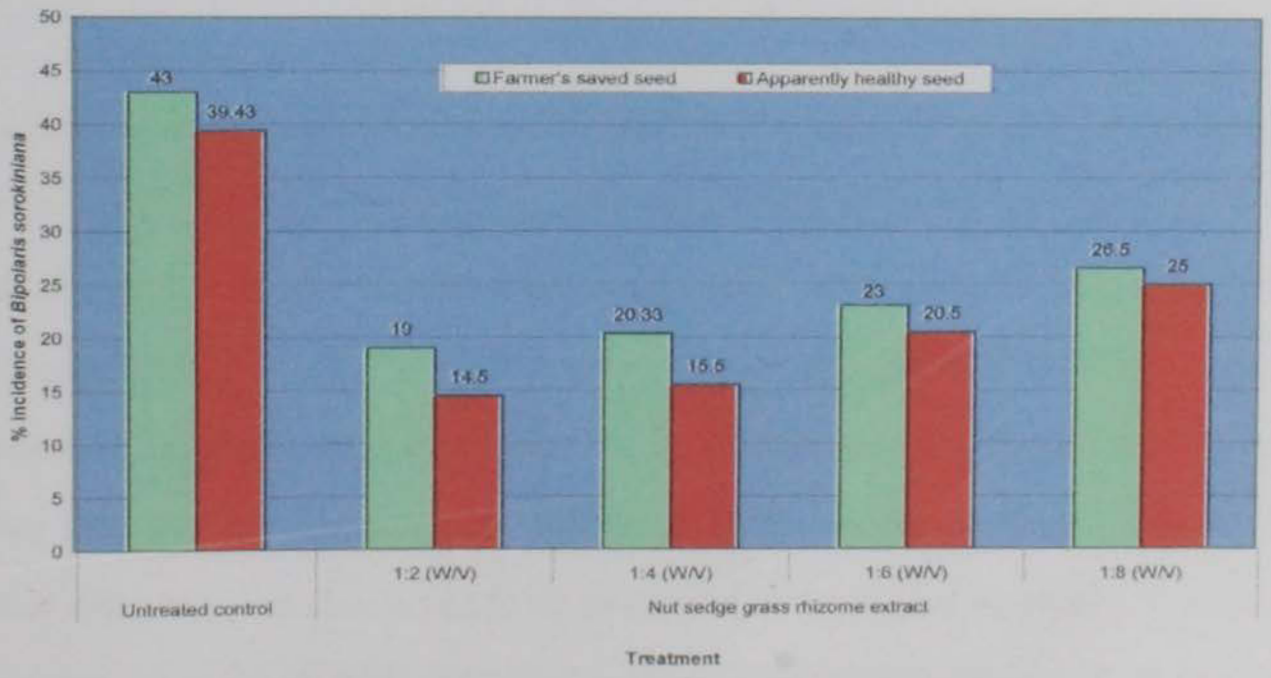


Fig 16. Effect of seed treatment with Nut sedge grass rhizome extract on incidence of *Bipolaris sorokiniana* of farmer's saved seeds and apparently healthy seeds

4.6.4. Effect of seed treatment with Burmuda grass extract on germination and incidence of *Bipolaris sorokiniana* on wheat seed

Seed treatment with Burmuda grass extract showed significant effect on seed germination and incidence of *Bipolaris sorokiniana* of farmer's saved seed and apparently healthy seeds of wheat over untreated control (Table 9, fig 17). % Germination of farmer's saved seed ranged from 76.50% to 95.83% in respect of seed treatment with different doses of Burmuda grass extract. Highest seed germination (95.83%) was observed in 1:2 ($^{W}/_{V}$) of Burmuda grass extract and lowest (76.50%) in untreated control, where seeds were treated with water only. % Germination of apparently healthy seeds was found higher over farmers saved seed. The highest germination (99.60%) of apparently healthy seeds was noted in 1:2 ($^{W}/_{V}$) concentration of Burmuda grass extract and the lowest (78.00%) in control.

Regarding % incidence of *Bipolaris sorokiniana* on wheat seeds, it has been found that the incidence was always higher in farmers saved seed over apparently healthy seeds (Table 9, Fig 18). The incidence of *Bipolaris sorokiniana* on farmer's saved seed ranged from 16.50% to 43.00% where the highest and lowest incidence was recorded in untreated control (T_1) and 1:2 ($^{W}/_{V}$) concentration of Burmuda grass extract (T_2) similarly for apparently healthy seeds the treatments T_1 and T_2 resulted highest (39.43%) and lowest (12.33%) incidence of the fungus respectively. It was found that apparently healthy seeds had comparatively lower incidence of the fungus (Table 9, fig 18) over farmer's saved seed.

Table 9. Effect of seed treatment with Burmuda grass extract on germination and incidence of *Bipolaris sorokiniana* of wheat

Treatments		% Germination		% Incidence of *Bipolaris sorokiniana*	
		Farmer's saved seed	Apparently healthy seed	Farmer's saved seed	Apparently healthy seed
T_1= Untreated control		76.50d	78.00c	43.00a	39.43a
T_2= Burmuda grass extract	1:2 ($^{W}/_{V}$)	95.83a	99.60a	16.50e	12.33e
T_3= Burmuda grass extract	1:4 ($^{W}/_{V}$)	95.50a	99.53a	19.50d	15.57d
T_4= Burmuda grass extract	1:6 ($^{W}/_{V}$)	92.50b	99.40b	23.17c	17.50c
T_5= Burmuda grass extract	1:8 ($^{W}/_{V}$)	90.50c	98.27b	24.40b	18.57b
LSD (P 0.05)		0.4874	1.210	0.4874	0.3622

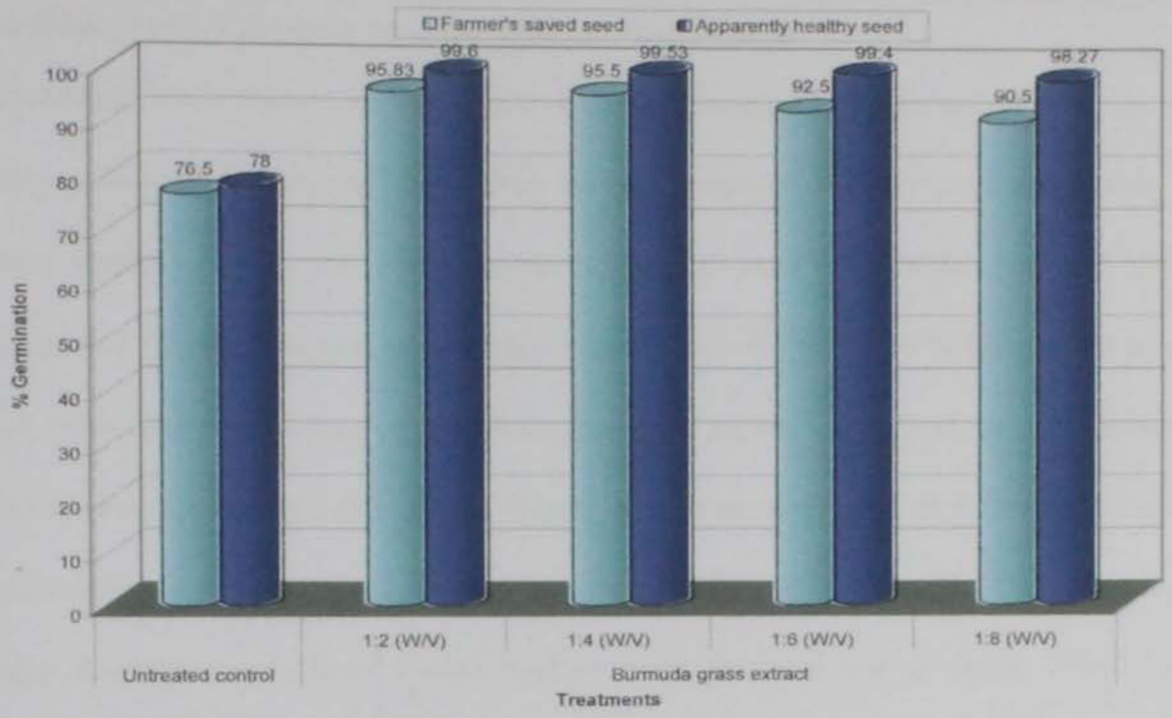


Fig 17. Effect of seed treatment with Burmuda grass extract on germination of farmer's saved seeds and apparently healthy seeds

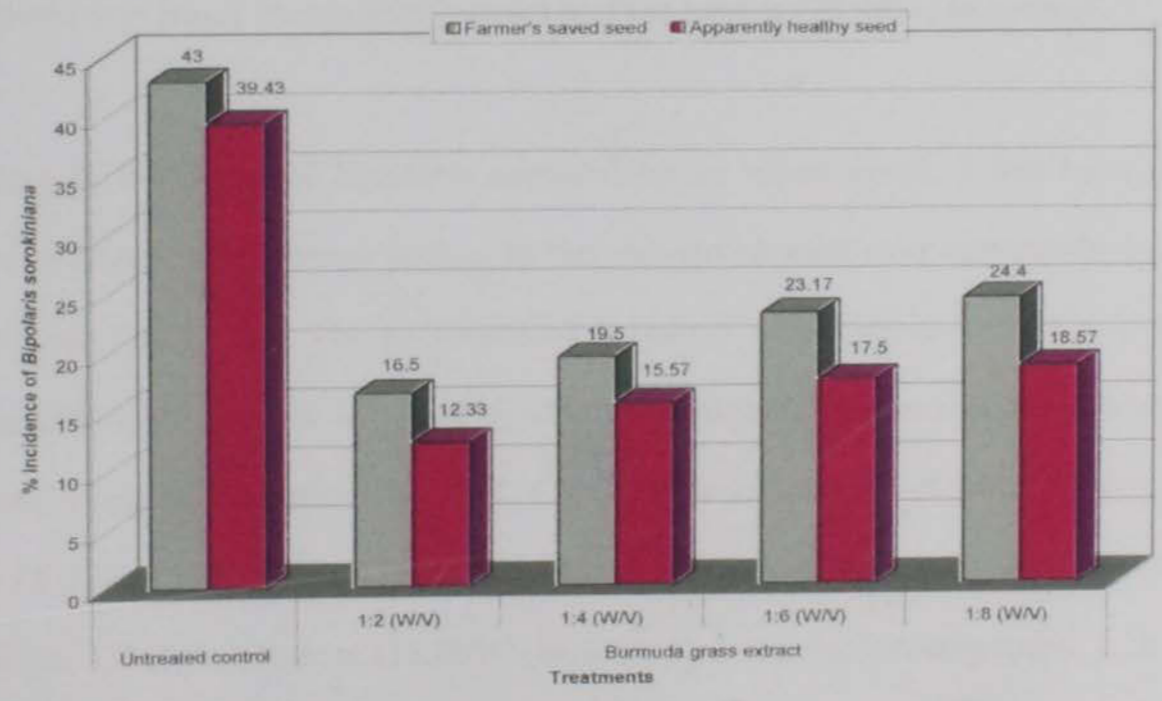


Fig 18. Effect of seed treatment with Burmuda grass extract on incidence of *Bipolaris sorokiniana* of farmer's saved seeds and apparently healthy seeds

4.6.5. Effect of seed treatment with Black cumin seed extract on germination and incidence of *Bipolaris sorokiniana* on wheat seed

Seed treatment with Black cumin seed extract showed significant effect on seed germination and incidence of *Bipolaris sorokiniana* of farmer's saved seeds and apparently healthy seeds of wheat over untreated control (Table 10, fig 19). % Germination of farmer's saved seed ranged from 76.50% to 83.17% in respect of seed treatment with different doses of Black cumin seed extract. Highest seed germination (83.17%) was recorded in 1:2 ($^{W}/_{V}$) of Black cumin seed extract and lowest (76.50%) in untreated control, where seeds were treated with water only. % Germination of apparently healthy seeds was found higher over farmers saved seed. The highest germination (91.00%) of apparently healthy seeds was noted in 1:2 ($^{W}/_{V}$) concentration of Black cumin seed extract and the lowest (78.00%) in control.

Regarding % incidence of *Bipolaris sorokiniana* on wheat seeds, it has been found that the incidence was always higher in farmers saved seed over apparently healthy seeds (Table 10, fig 20). The incidence of *Bipolaris sorokiniana* on farmer's saved seed ranged from 27.17% to 43.00% where the highest and lowest incidence was recorded in untreated control (T_1) and 1:2 ($^{W}/_{V}$) concentration of Black cumin seed extract (T_2) similarly for apparently healthy seeds the treatments T_1 and T_2 resulted highest (39.43%) and lowest (15.20%) incidence of the fungus respectively. It was found that apparently healthy seeds had comparatively lower incidence of the fungus (Table 10, fig 20) over farmer's saved seed.

Table 10. Effect of seed treatment with Black cumin seed extract on germination and incidence of *Bipolaris sorokiniana* of wheat

Treatments		% Germination		% Incidence of *Bipolaris sorokiniana*	
		Farmer's saved seed	Apparently healthy seed	Farmer's saved seed	Apparently healthy seed
T_1= Untreated control		76.50d	78.00d	43.00a	39.43a
T_2= Black cumin seed extract	1:2 ($^{W}/_{V}$)	83.17a	91.00a	27.17d	15.20d
T_3= Black cumin seed extract	1:4 ($^{W}/_{V}$)	82.17b	85.17b	30.17c	23.00e
T_4= Black cumin seed extract	1:6 ($^{W}/_{V}$)	82.50b	84.17c	31.50c	26.50c
T_5= Black cumin seed extract	1:8 ($^{W}/_{V}$)	81.00c	84.50c	34.50b	30.50b
LSD (P 0.05)		0.3622	0.421	1.670	0.2063

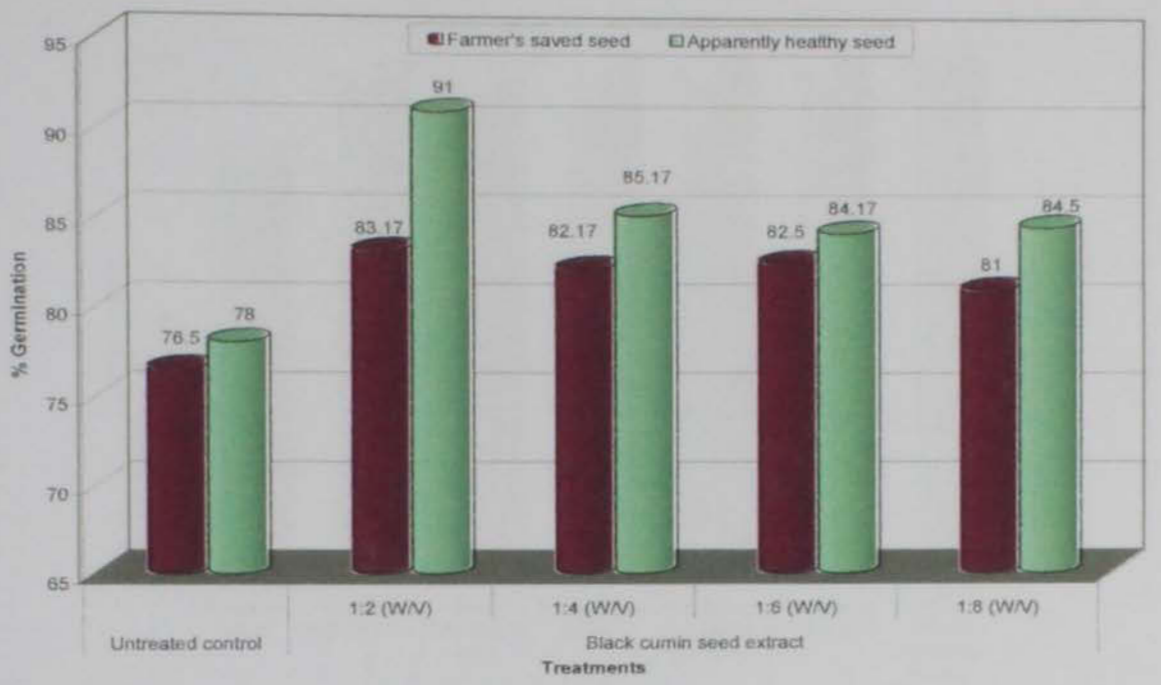


Fig 19. Effect of seed treatment with Black cumin seed extract on germination of farmer's saved seeds and apparently healthy seeds

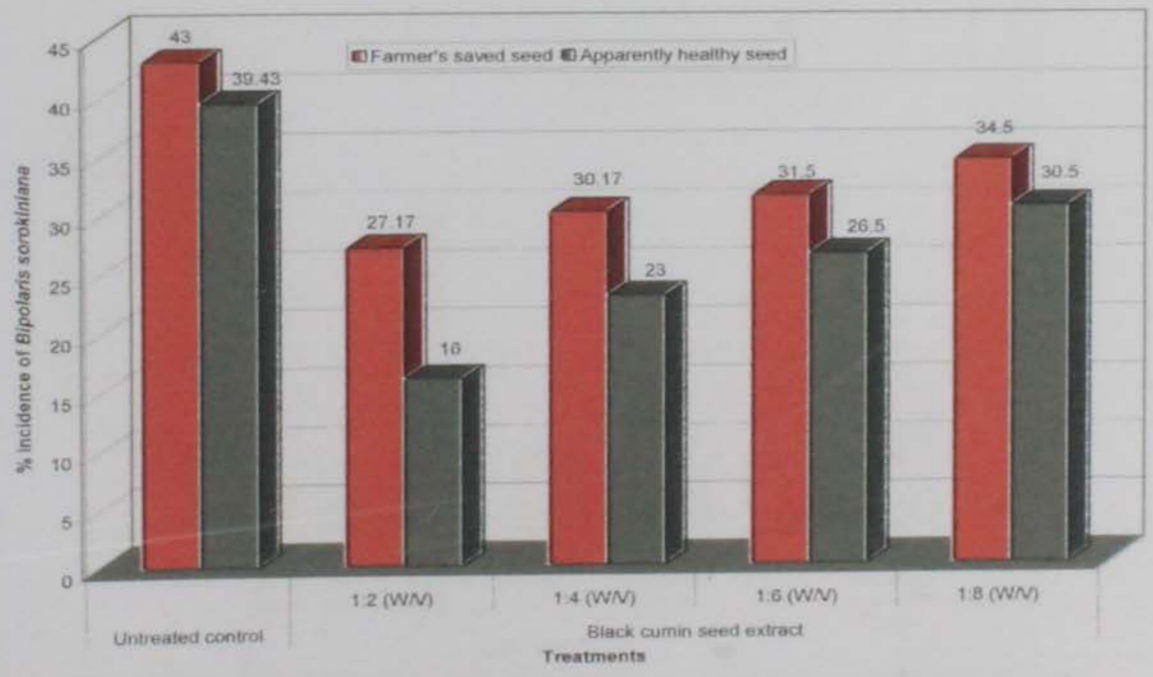


Fig 20. Effect of seed treatment with Black cumin seed extract on incidence of *Bipolaris sorokiniana* of farmer's saved seeds and apparently healthy seeds

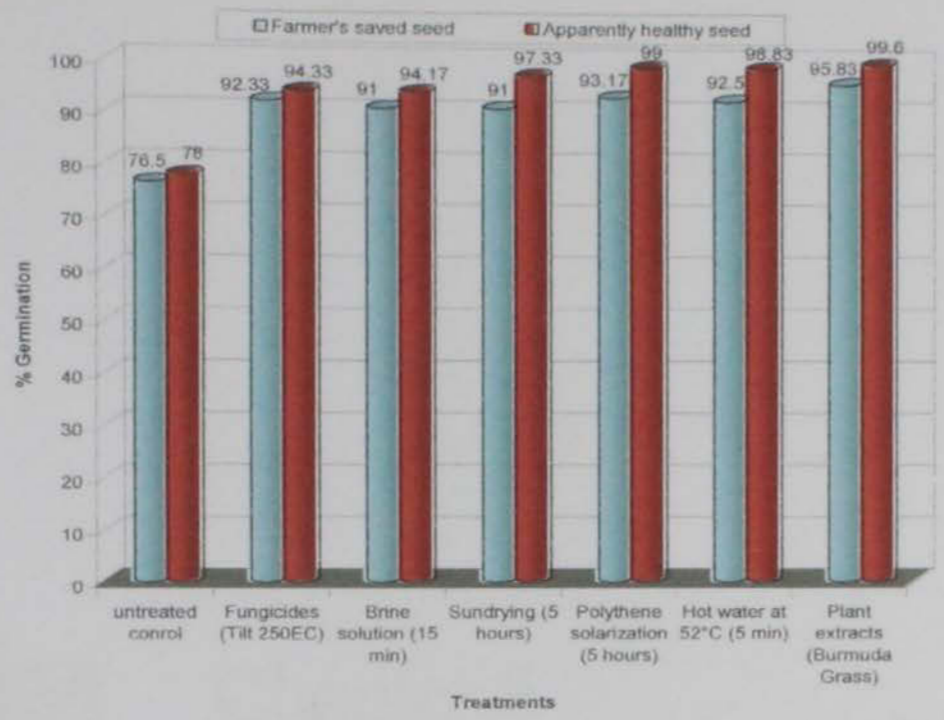


Fig 21. Effect of different seed treatments on germination of farmer's saved seeds and apparently healthy seeds

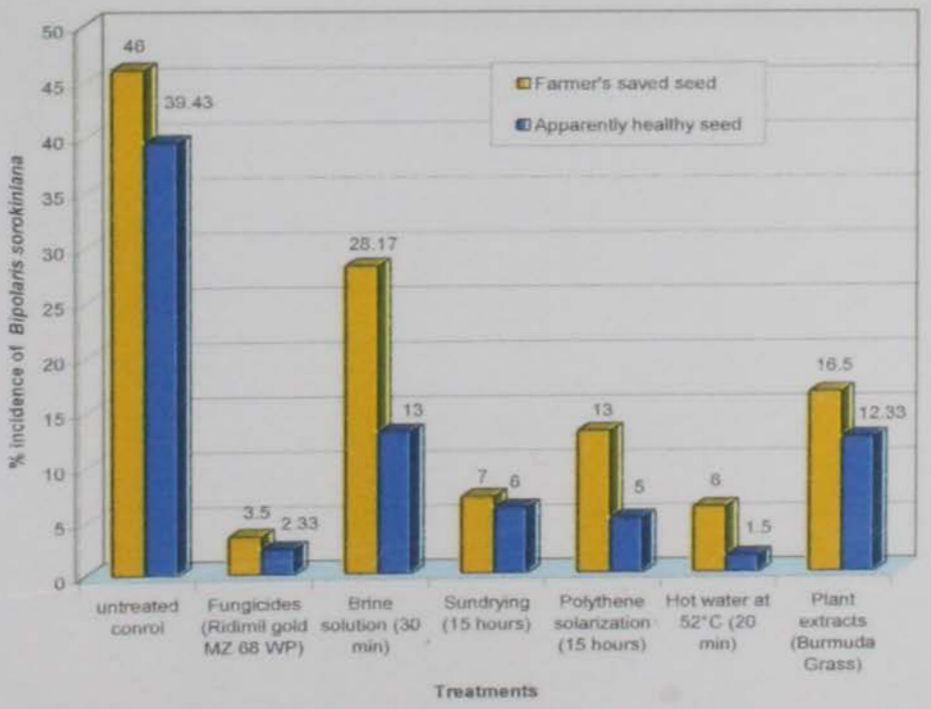
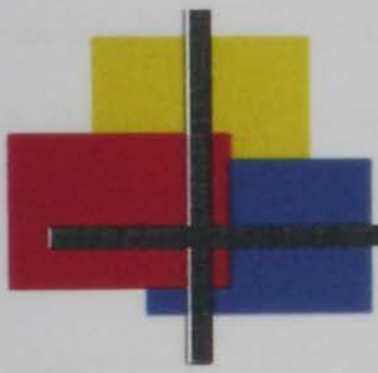


Fig 22. Effect of different seed treatments on incidence of *Bipolaris sorokiniana* of farmer's saved seeds and apparently healthy seeds



Chapter 5
Discussion

5. DISCUSSION

5.1. EVALUATION OF FUNGICIDE AGAINST *Bipolaris sorokiniana*

All the fungicides reduced the incidence of *Bipolaris sorokiniana* over untreated control. In the present study the highest incidence (10.50%) of *Bipolaris sorokiniana* was recorded in farmer's saved seed and the lowest incidence was counted (2.33%) in apparently healthy seed treated with Ridomil gold MZ 68 WP @ 0.2%. The findings of the present study corroborates with the findings of Dey *et al.* (1992). In a similar type of study they reported that seed treatment with Vitavax-200 significantly reduced the incidence of *Bipolaris sorokiniana* and increased seed germination. Pelton and Karjalainen (1992) also found that seed treatment with Tilt 250EC (0.1%) significantly reduced black point severity. In Bangladesh many researchers worked with Tilt 250EC for controlling black point of wheat and found promising result (Anonymous, 1989). In the present study seed treatment with Tilt 250EC significantly reduced the incidence of *Bipolaris sorokiniana* which is also supported by Mondal *et al.* (1994). They found Tilt 250 EC to be the most effective fungicide against *Bipolaris sorokiniana*.

5.2. EVALUATION OF BRINE SOLUTION AGAINST *Bipolaris sorokiniana*

Seed treatment with brine solution significantly increased seed germination and reduced the incidence of *Bipolaris sorokiniana* over control. The highest seed germination (94.17%) was counted in apparently healthy seed treated with brine solution for 15 minutes followed by apparently healthy seed treated with brine

solution for 5 minutes and the lowest seed germination (82.00%) was noted in the untreated farmer's saved seed. In the present study, seed germination increased with increasing the time of seed treatment with brine solution up to 15 minutes but declined gradually at 30 and 60 minutes. On the other hand, brine solution reduced the incidence of *Bipolaris sorokiniana* over control. The highest incidence of *Bipolaris sorokiniana* was counted (46.00%) in untreated farmer's saved seed. The lowest incidence was counted (13.00%) in apparently healthy seed treated with brine solution (2%) for 30 minutes. From the results it has been found that the incidence of *Bipolaris sorokiniana* decreased with increasing the length of seed treatment with brine solution. The findings of the present study corroborates with the findings of Hasan (2000) who reported that seeds become free from *Bipolaris oryzae* when washed with brine solution. The present findings also supported by Kabir (2006). He reported that physically sorted apparently healthy seeds showed higher germination (90.06%) than farmer's saved seed when treated with brine solution (2%). The findings of the present study also supported by the report of Uddin (2005).

5.3. EVALUATION OF SEED SOLARIZATION AGAINST *Bipolaris sorokiniana*

The effect of different levels of seed solarization was significant in increasing seed germination as well as reducing incidence of *Bipolaris sorokiniana.* The highest germination (99.33%) was recorded in solarization of apparently healthy seed for 5 hours and the lowest germination (81.67%) was recorded in untreated farmer's saved seed. Seed solarization significantly reduced the incidence of *Bipolaris sorokiniana*

over control. The highest incidence of *Bipolaris sorokiniana* was counted (47.00%) in untreated farmer's saved seed. The lowest incidence was counted (6.00%) in apparently healthy solarized for 15 hours. The findings of the present study are supported by Zobaer *et al.* (2007). In a similar type of study he found that solar heat treatment reduced 76% incidence of *Bipolaris sorokiniana* over untreated control. The result also closely matched with the report of the Fakir and Jahan (1998) who found that solar heat treatment reduced seed borne infection of *Bipolaris sorokiniana* and increased germination by 9.01%. These findings also corroborated with the findings of Mohindar *et al.* (1994) and Guldhe *et al.* (1985).

5.4. EVALUATION OF POLYTHENE SOLARIZATION OF SEED AGAINST *Bipolaris sorokiniana*

Polythene solarization of seed is a new idea for reducing the incidence of seed borne *Bipolaris sorokiniana* increasing percent seed germination. In the present study the highest germination (99.00%) was counted in polythene solarization of apparently healthy seed for 5 hours and the lowest germination (82.00%) was counted in untreated farmer's saved seed. Polythene solarization of seed reduced the incidence of *Bipolaris sorokiniana* over control. The highest incidence of *Bipolaris sorokiniana* was counted (46.00%) under the treatment untreated farmer's saved seed. The lowest incidence was counted (5.00%) in polythene solarization of apparently healthy seed for 15 hours. The present findings corroborates with the findings of Zobaer *et al.* (2007). He found that polythene solarization of seed reduced 62.13% incidence of

Bipolaris sorokiniana over untreated control. The findings also corroborated with Mohindar *et al.* (1994) who found that loose smut of wheat can be completely controlled by solar heat treatment.

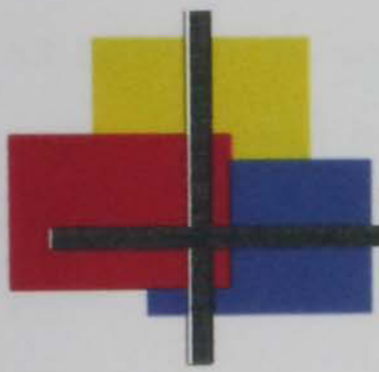
5.5. EVALUATION OF HOT WATER (52°C) AGAINST *Bipolaris sorokiniana*

The highest seed germination (99.00%) was counted in apparently healthy seed treated with hot water at 52°C for 10 minutes and the lowest seed germination (82.00%) was counted in untreated farmer's saved seed. Hot water seed treatment effectively reduced the incidence of *Bipolaris sorokiniana* over control. The highest incidence of *Bipolaris sorokiniana* was counted (46.00%) in untreated farmer's saved seed and the lowest incidence was counted (1.50%) in apparently healthy seed treated with hot water at 52°C for 20 minutes. The present findings corroborated with the findings of Zobaer *et al.* (2007) who reported that hot water treatment of wheat seeds at 52°C for 5 minutes increased seed germination up to 74.90%. The findings also supported by Muniz (2001) who found that hot water treatment at 50°C for 30 minutes eradicated fungi associated with tomato seed. Khaleduzzaman (1996) found that wheat seed treated at 49°C, 52°C, 55°C, for 5-10 minutes gave highest control of *Bipolaris sorokiniana* and increased seed germination. Prabhu and Prasada (1970) also found that leaf blight of wheat can be controlled by soaking seed in water at 52°C-54°C for 10 minutes.

5.6. EVALUATION OF PLANT EXTRACTS AGAINST *Bipolaris sorokiniana*

Seed treatment with plant extracts had profound effect on seed germination and incidence of *Bipolaris sorokiniana*. The highest seed germination (99.60%) was counted in Burmuda grass extracts (1:2 $^{W}/_{V}$). The lowest germination (76.40%) was counted in untreated farmer's saved seed. All the plant extracts reduced the incidence of *Bipolaris sorokiniana* over control. The highest incidence (43.00%) of *Bipolaris sorokiniana* was counted in untreated farmer's saved seed and the lowest incidence was counted (12.33%) in Burmuda grass extract (1:2 $^{W}/_{V}$). Neem leaf extracts (1:2 $^{W}/_{V}$) and Nut sedge grass extracts (1:2 $^{W}/_{V}$) resulted 14.00% and 14.50% incidence of *Bipolaris sorokiniana* respectively which were significantly lower over control. The present findings keep in with the findings of Rahman (2006) who found that botanicals have strong effect against *Bipolaris sorokiniana*. Islam (2006) also reported that kalizira, Neem, Onion and Zinger extracts significantly reduced the incidence of *Bipolaris sorokiniana* on wheat seeds over control. The findings was also supported by Alice and Rao (1987), Fakir and Khan (1992), Khan and Kumar (1992), Hossain and Schlosser (1993), Khan and Fakir (1995) and Hossain *et al.* (1997). Ashrafuzzaman and Hossain (1992) found Neem extract effective against *Bipolaris sorokiniana* and inhibited the growth of the fungus also reduced its pathogenecity on wheat leaves. Hossain and Schlosser (1993) reported that germination rate of wheat seeds increased while treated with extract of Neem seed and Neem oil cake. Hossain *et al.* (2005) reported that Neem extract reduced the incidence of *Bipolaris sorokiniana* significantly and increased seed germination. They also found that out of

six plant extracts, Neem extract was proved superior followed by garlic, Bishkatali and Vatpata. Alice and Rao (1987) evaluated plant extracts against seed borne infection of fungi and found increased germinability of the treated seeds. Khan and Kumar (1992) also observed the antifungal activity of leaf extracts of Neem against seed mycoflora of wheat. They found remarkable reduction of seed mycoflora and increased seed germination of treated seeds.



Chapter 6
Summary and Conclusion

6. SUMMARY AND CONCLUSION

Wheat suffers from as many as 26 seed borne pathogens causing 14 seed borne diseases. Among them leaf blight and black point caused by *Bipolaris sorokiniana* has become a serious concern. The fungus reduced the yield and seed quality. The present study was undertaken to evaluate some selected fungicides and eco-friendly treatments against *Bipolaris sorokiniana* of wheat.

All the fungicides reduced the incidence of *Bipolaris sorokiniana* over untreated control. The lowest incidence of *Bipolaris sorokiniana* (2.33%) was observed in apparently healthy seed treated with Ridomil gold MZ 68 WP @ 0.2%, over untreated control.

The highest seed germination (94.17%) was recorded in the treatment apparently healthy seed treated with brine solution (2%) for 15 minutes followed by apparently healthy seed treated with brine solution for 5 minutes and the lowest seed germination (82.00%) was recorded in the treatment of untreated farmer's saved seed. On the other hand brine solution reduced the incidence of *Bipolaris sorokiniana* over control. The lowest incidence of *Bipolaris sorokiniana* (13.00%) was observed in the treatment apparently healthy seed treated with brine solution for 30 minutes.

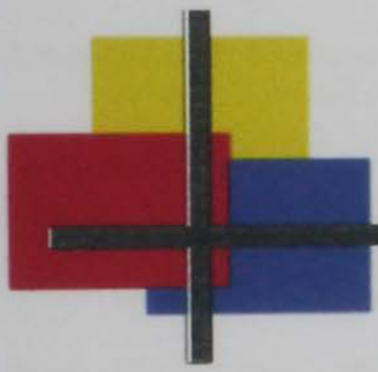
The effect of different levels of seed solarization was found promising in increasing seed germination as well as reducing incidence of *Bipolaris sorokiniana.* The highest seed germination (99.33%) was noted in solarized apparently healthy seed for 5 hours and the lowest seed germination (81.67%) was counted in untreated farmer's saved seed. Seed solarization significantly reduced the incidence of *Bipolaris sorokiniana* over control. The highest incidence of *Bipolaris sorokiniana* (47.00%) was observed under the treatment of untreated farmer's saved seed. The lowest incidence (6.00%) was counted in solarized apparently healthy seed for 15 hours.

The highest seed germination (99.00%) was recorded in polythene solarization of apparently healthy seed for 5 hours. Polythene solarized seed reduced the incidence of *Bipolaris sorokiniana* over control. The lowest incidence of *Bipolaris sorokiniana* (5.00%) was counted in the treatment polythene solarized apparently healthy seed for 15 hours.

The highest seed germination (99.00%) was noted in apparently healthy seed treated with hot water at 52°C for 10 minutes and the lowest seed germination (82.00%) was noted in untreated farmer's saved seed. Hot water at 52°C effectively reduced the incidence of *Bipolaris sorokiniana* over control. The lowest incidence was observed (1.50%) in apparently healthy seed treated with hot water at 52°C for 20 minutes.

The highest seed germination (99.60%) was counted in case of Burmuda grass extract (1:2 $^{W}/_{V}$) followed by 99.53% (1:4 $^{W}/_{V}$) and 99.40% (1:6 $^{W}/_{V}$) of Burmuda grass extract. The lowest seed germination (76.40%) was counted in untreated farmer's saved seed. All the plant extracts reduced the incidence of *Bipolaris sorokiniana* over control. The lowest incidence of *Bipolaris sorokiniana* (12.33%) was counted in case of Burmuda grass extract (1:2 $^{W}/_{V}$), followed by 14.00% and 14.50% in Neem leaf extract (1:2 $^{W}/_{V}$) and Nut sedge grass rhizome extract (1:2 $^{W}/_{V}$) respectively.

From the above findings of study it is well exposed that Ridomil gold MZ 68 WP is very effective in controlling *Bipolaris sorokiniana*. Tilt 250 EC is effective in increasing seed germination. Among the physical seed treatments brine solution 2% for 15 minutes, Sun drying (solarization) and Polythene solarization of seeds for 5 hours and hot water treatment for 10 minutes had excellent performance in controlling seed borne *Bipolaris sorokiniana* and increasing seed germination. Among the plant extracts Burmuda or Durba grass (1:2 $^{W}/_{V}$) proved promising against *Bipolaris sorokiniana* with increasing seed germination of wheat.



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APPENDICES

Appendix 1. Analysis of variance of the data on chemical seed treatment on germination and incidence of *Bipolaris sorokiniana* of wheat

Source	Degrees of Freedom	Mean Sum of Square			
		% Germination		% Incidence of *Bipolaris sorokiniana*	
		Farmers saved seed	Apparently healthy seed	Farmers saved seed	Apparently healthy seed
Replication	2	5.551	3.097	5.681	3.749
Treatment	5	111.749**	79.989**	17.814**	16.607**
Error	10	0.171	0.431	0.181	0.149

** Significant at 1% level of probability

Appendix 2. Analysis of variance of the data on treatment with brine solution on germination and incidence of *Bipolaris sorokiniana* of wheat

Source	Degrees of Freedom	Mean Sum of Square			
		% Germination		% Incidence of *Bipolaris sorokiniana*	
		Farmers saved seed	Apparently healthy seed	Farmers saved seed	Apparently healthy seed
Replication	2	33.800	10.550	20.000	4.467
Treatment	4	81.375**	20.067**	352.792**	618.142**
Error	8	0.425	1.467	0.417	6.342

** Significant at 1% level of probability

Appendix 3. Analysis of variance of the data on treatment by sun drying on germination and incidence of *Bipolaris sorokiniana* of wheat

Source	Degrees of Freedom	Mean Sum of Square			
		% Germination		% Incidence of *Bipolaris sorokiniana*	
		Farmers saved seed	Apparently healthy seed	Farmers saved seed	Apparently healthy seed
Replication	2	12.117	10.617	1.950	3.617
Treatment	4	37.808**	46.567**	826.725**	517.767**
Error	8	4.283	3.367	3.450	2.679

** Significant at 1% level of probability

Appendix 4. Analysis of variance of the data on treatment by polythene solarization on germination and incidence of *Bipolaris sorokiniana* of wheat

Source	Degrees of Freedom	Mean Sum of Square			
		% Germination		% Incidence of *Bipolaris sorokiniana*	
		Farmers saved seed	Apparently healthy seed	Farmers saved seed	Apparently healthy seed
Replication	2	0.617	3.950	1.800	0.450
Treatment	4	47.850**	401.100**	608.475**	623.850**
Error	8	5.575	2.200	3.925	2.200

** Significant at 1% level of probability

Appendix 5. Analysis of variance of the data on treatment with hot water (52^0C) on germination and incidence of *Bipolaris sorokiniana* of wheat

Source	Degrees of Freedom	Mean Sum of Square			
		% Germination		% Incidence of *Bipolaris sorokiniana*	
		Farmers saved seed	Apparently healthy seed	Farmers saved seed	Apparently healthy seed
Replication	2	7.217	0.150	0.800	2.450
Treatment	4	79.642**	44.142**	796.125**	717.000**
Error	8	1.092	1.067	2.425	1.200

** Significant at 1% level of probability

Appendix 6. Analysis of variance of the data on treatment with Allamanda leaf extract on germination and incidence of *Bipolaris sorokiniana* of wheat

Source	Degrees of Freedom	Mean Sum of Square			
		% Germination		% Incidence of *Bipolaris sorokiniana*	
		Farmers saved seed	Apparently healthy seed	Farmers saved seed	Apparently healthy seed
Replication	2	1.763	6.117	0.600	22.217
Treatment	4	23.731**	50.442**	84.150**	97.520**
Error	8	0.833	5.242	1.100	4.050

** Significant at 1% level of probability

Appendix 7. Analysis of variance of the data on treatment with Neem leaf extract on germination and incidence of *Bipolaris sorokiniana* of wheat

Source	Degrees of Freedom	Mean Sum of Square			
		% Germination		% Incidence of *Bipolaris sorokiniana*	
		Farmers saved seed	Apparently healthy seed	Farmers saved seed	Apparently healthy seed
Replication	2	6.067	0.774	5.313	6.104
Treatment	4	189.667**	270.551**	320.425**	346.559**
Error	8	0.067	0.413	0.067	0.037

** Significant at 1% level of probability

Appendix 8. Analysis of variance of the data on treatment with Nut sedge grass extract on germination and incidence of *Bipolaris sorokiniana* of wheat

Source	Degrees of Freedom	Mean Sum of Square			
		% Germination		% Incidence of *Bipolaris sorokiniana*	
		Farmers saved seed	Apparently healthy seed	Farmers saved seed	Apparently healthy seed
Replication	2	1.817	1.817	7.200	4.513
Treatment	4	128.692**	284.079**	118.500**	295.508**
Error	8	0.817	0.817	0.200	0.013

** Significant at 1% level of probability

Appendix 9. Analysis of variance of the data on treatment with Burmuda grass extract on germination and incidence of *Bipolaris sorokiniana* of wheat

Source	Degrees of Freedom	Mean Sum of Square			
		% Germination		% Incidence of *Bipolaris sorokiniana*	
		Farmers saved seed	Apparently healthy seed	Farmers saved seed	Apparently healthy seed
Replication	2	1.865	0.317	0.200	5.017
Treatment	4	151.067**	244.167**	339.375**	311.525**
Error	8	1.290	0.692	1.200	0.038

** Significant at 1% level of probability

Appendix 10. Analysis of variance of the data on treatment with Black cumin seed extract on germination and incidence of *Bipolaris sorokiniana* of wheat

Source	Degrees of Freedom	Mean Sum of Square			
		% Germination		% Incidence of *Bipolaris sorokiniana*	
		Farmers saved seed	Apparently healthy seed	Farmers saved seed	Apparently healthy seed
Replication	2	4.517	4.550	1.517	5.425
Treatment	4	21.400**	62.150**	109.650**	115.586**
Error	8	0.037	0.050	0.787	0.012

** Significant at 1% level of probability