PATHOGENIC MYCOFLORA OF RICE GRAINS AND ITS MANAGEMENT

THESIS SUBMITTED FOR THE DEGREE OF DOCTOR OF PHILOSOPHY IN BOTANY

BY

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DEPARTMENT OF BOTANY

UNIVERSITY OF DHAKA

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DECLARATION

I hereby declare that this dissertation is based on entirely my own work and that, to the best of my knowledge and belief, it holds no material that has been published before or written by another person nor material which to a substantial extent has been accepted for the award of another degree or diploma at any University. From this research work three papers are published in scientific journals.

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LIST OF ABBREVIATIONS

ANOVA	Analysis of Variance
AEZ	Agro Ecological Zone
Append.	Appendix
DAP	Days after planting
DAI	Days after inoculation
°C	Degree Celsius
Fig.	Figure
g	Gram
ha	Hectare
kg	Kilogram
m	Meter
t ha^{-1}	Metric ton per hectare
ml	Milliliter
mM	Millimolar
mm	Millimeter
cm	Centimeter
µm	Milli micron
viz.	Namely
pH	Negative logarithm of hydrogen ion concentration
OMB	Oat Meal Broth Medium
ppm	Parts per million
PDA	Potato dextrose agar
PDB	Potato dextrose broth
PDI	Percent Disease Index
%	Per cent
sp.	Species
et al.	With others
Vol.	Volume

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ABSTRACT

The present investigation is based on diseased rice grains of four commercially cultivated rice varieties namely BRRI 28, 29, Kalijira and Pajam collected from 14 districts of 7 divisions and 40 rice samples *viz*. Hybrid 2, 3, 4, BR 7, 11, 12, 14 ,16, 22, 23, 25, 26 and BRRI 28 to BRRI 55 were collected from Bangladesh Rice Research Institute at Joydebpur. Quality analysis showed that the percentage of pure seed varies from 94-99%. The germination and mortality percentage of rice seeds of different varieties were in the range of 62-100% and 10-30%, respectively. Twenty five species of fungi belonging to 15 genera were associated with these rice varieties. The isolated fungi were *Alternaria alternata, Aspergillus clavatus, A. flavus, A. fumigatus, A. niger, A. ochraceous A. oryzae, A. terreus, Chaetomium globosum, Cladosporium cladosporioides, Colletotrichum gloeosporioides, Curvularia lunata, C. lunata* var. *aeria, Drechslera oryzae, Fusarium moniliforme, F. oxysporum, F. solani, Microdochium oryzae, Nigrospora oryzae, Penicillium* spp, *Pestalotiopsis guepinii, Rhizopus stolonifer, Sarocladium oryzae* and *Trichoderma viride.* Their pathogenic potentiality was tested by seed inoculation technique. Amongst twenty five species nine *viz*., *Alternaria alternata* (Fr.) Keissler, *Aspergillus flavus* Link *Curvularia lunata* (Wakker) Boedijn, *Drechslera oryzae* Breda de Haan (Subramanian and Jain), *Fusarium moniliforme* Sheldon, *F. solani* (Mart.) Sacc., *Microdochium oryzae* (Hashloka and Yokogi) Sam. and Hal., *Pestalotiopsis guepinii* (Desm.) Stay. and *Sarocladium oryzae* (Sawada) W. Gams and D. Hawks were found to be pathogenic. These nine pathogenic fungi were transmitted from seed to seedlings and produced different types of symptoms.

Ten fungicides i.e. Bavistin 50 WP, Capvit 50 WP, Dithane M-45, Greengel, Hayvit 80 WP, Indofil M-45, Ridomil MZ Gold, Salcox 50 WP, MC Sulphur 80 and Tall 25 EC at 100, 200, 300, 400 and 500 ppm were evaluated against the above mentioned nine pathogenic fungi. Tall 25 EC completely inhibited the radial growth of the test pathogens at all the concentrations except *F. moniliforme* and *Microdochium oryzae.*

Antifungal properties of *Allium sativum* L., *Artocarpus heterophyllus* Lamk., *Asparagus racemosus* Willd., *Azadirachta indica* A. Juss., *Citrus medica* L., *Datura metel* L., *Mangifera indica* L., *Nerium indicum* Mill., *Senna alata* (L.) Roxb. and *Tagetes erecta* L. at 5, 10 and 20% concentrations were evaluated against the nine test

pathogens. All the plant extracts completely inhibited the radial growth of the test pathogens at 20% concentration except *Asparagus racemosus* and *Nerium indicum.*

Antagonistic potential of the selected six soil fungi against pathogenic fungi were evaluated. Amongst 48 interactions grade 3 was found in 22 interactions. Volatile substances from soil fungi inhibited radial growth of the test pathogens varied from 8.33-57.36%.The highest inhibition (57.36%) was found due to *T. harzianum* against *P. guepinii*. Non-volatile substances showed inhibition of mycelial growth ranged from 29.05 to 64.5%. The highest inhibition was observed owing to the *T. harzianum* against *C. lunata*. In colony interaction, the highest growth inhibition (88%) was observed by *T. harzianum* against *A. alternata*.

In the field experiment out of 13 treatments, T6 (Bavistin+*Azadirachta indica+Trichoderma harzianum*) and T10 (Bavistin+Tall+*Azadirachta indica+Citrus medica*) treated seeds showed highest per cent of germination and seedling vigor index against all the test pathogens.

CONTENTS

CHAPTER 1

INTRODUCTION

Rice (*Oryza sativa* L.*)* is one of the major cereal crops of the world used as staple food by 60% of the world population. It grows profusely in Bangladesh, India, Ceylon, Burma, Japan, China, Thailand, Malaysia and Philippines. Rice is cultivated in Bangladesh in different seasons namely Aus (March-June), Aman (July-December) and Boro (February-July). About 82% of the cultivated area of this country is used for production of rice (BBS 2009). Bangladesh is the 4th largest rice producer in the world. It covers 93% of the food grains production and 72% of the total crop land (Abedin *et al*. 2012). The average world yield of rice is 3.84 tons/hectre and the average yield of rice in Bangladesh is 2.52 tons/hectre. Approximately 2.5 million tons of rice is lost annually owing to rice diseases.

Most of the major diseases of rice are seed borne, 11 seed transmitted fungal pathogens are responsible for causing diseases in the field (Fakir 1982). These pathogens also play an important role in deteriorating the quality and longivity of seeds (Mia and Mathur 1985, Shahjahan *et al.*1988).

In Bangladesh, 43 diseases are known to occur on the rice crop. Among these 36 are seed borne of which 14 are major importance, 22 are caused by fungi (Fakir 2000). This crop is affected by more than seventy two diseases in all rice growing areas of the world, from which 31 are reported from Bangladesh (Miah *et al.*1985). These diseases are very common in many parts of South East Asia and responsible for considerable losses in rice production. Yield loss due to diseases was reported by Shahjahan (1993) and Chakrabarti *et al.* (1998).

The seeds of rice not only carry pathogens but also abundant microorganisms that act as biological control agents against other rice pathogens. Biological control has now become one of the most exciting and rapidly developing areas in plant pathology because it has great potential to solve many agricultural and environmental problems (Baker and Cook 1983, Hossain *et al* 2005). Antagonistic bacteria isolated from healthy and diseased rice seeds and leaves, was found most effective in controlling rice pathogens (Sharma *et al.* 2004, Akter *et al.* 2003).

Annual loss of crops to world because of diseases has been estimated to be about 25000 million dollars; of this a major part is due to fungal pathogens. Recent studies revealed that more than 50% of the seed saved by farmers in Bangladesh are spotted or discolored which ultimately reduced the market price. Pathogens associated with seed also cause germination failure, post emergence seedling infection and also seedling blight. Aim of my research work is to introduce remedial management of pathogens by recognizing those that would be helpful to solve the issue of crop loss every year. My research work would also cover study of pathogens of aromatic rice to protect their loss and enhance use of those as aromatic rice is closely related to social and cultural heritage of Bangalees and it is consumed during different festivals, wedding and entertaining guests.

A very little information is available on the impact of farmers' seed processing, management practices on the seed associated fungi as well as fungi associated in storage grains and their management. Keeping the above facts under consideration, present work has been undertaken to the following aspects:

- To examine the seed quality of selected rice varieties.
- To detect mycoflora associated with the rice grains collected from different districts of Bangladesh including BRRI.
- Evaluation of pathogenic potentiality of the isolated fungi.
- Determination of seed to seedling transmission of pathogenic fungi.
- Screening of fungicides and plant extracts against the test pathogens.
- *In vitro* screening of antagonists on the radial growth of the test pathogens.
- Integrated approach to control the test pathogens.

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CHAPTER 2

REVIEW OF LITERATURE

2.1. Rice plant

Rice (*Oryza sativa* L.) is the major world's primary food crop mostly grown in tropical and sub tropical climate. It belongs to the family Poaceae. The climate of Bangladesh is most suitable for rice cultivation as well as for the development of diseases but the production of rice in Bangladesh is extremely low as compared to other rice growing countries of the world (Mansur *et al.* 2013).

2.2. Rice seeds

Association of various fungi with rice grains have been reported by various workers. Ganguly (1946) reported *Helminthosporium oryzae, Pyricularia oryzae, Alternaria tenuis, Curvularia lunata, Nigrospora oryzae* and *Epicoccum* sp. as seed borne pathogens. Del Prado and Christensen (1952) isolated several seed borne fungi like *Aspergillus, Penicillium, Fusarium* and *Curvularia* from rice hulls, caryopsis and dehulled seeds.

Fungal invasion can lead to rotting and loss of viability. A group of fungi, especially species of *Aspergillus* and *Penicillium* invade the seed while in the storage under favourable conditions and deterioration of seeds by storage fungi in terms of reducing germination is a common problem (Christensen and Kaufman, Onesirosan 1978).

Noble and Richardson (1968) listed seed borne diseases and observed that largest number of pathogens on seeds are fungi. Severe infection on the boot leaf sheath decreases or completely inhabits the emergence of panicles resulting in grain sterility (Ou 1972, 1985, Amin *et al.* 1974, Hsieh *et al.* 1977, Chien and Huang 1979). Singh (1981) reported the association of *Curvularia lunata, Fusarium semitectum, Helminthosporium oryzae* and *Trichoconis padwickii* on rice seed and concluded that they were responsible for causing serious diseases in the field. Most pathogens causing abnormal seedling of rice are seed borne (Gueretto *et al.* 1972).

In Malaysia, Zainum and Nik (1977) observed that the most common fungal pathogens of rice seeds were *Trichoconis padwickii, Drechslera oryzae, Fusarium moniliforme, Nigrospora oryzae* and *Pyricularia oryzae.* Neergaard (1977) reported that discoloration, necrosis, kernel rot leading to breakage during milling and loss of viability are caused by parasites such as *Fusarium* spp.*, Drechslera oryzae, Sclerospora oryzae* and *Trichoconis padwickii* were pathogenic on rice seeds. Discoloration of filled grain was also observed more by Estrada *et al.* (1979) in inoculated tillers compared to control. He also observed seed borne pathogens affect seed quality.

Abdel-Azim and Khalil (1980) reported that storage moulds *Aspergillus candidus, Aspergillus flavus, Aspergillus fumigatus, Aspergillus niger, Aspergillus ochraceus, Aspergillus wentii* and *Penicillium chrysogenum* are also responsible for the discoloration and deterioration of rice grains. When these fungi invade rice grains, they cause great reduction in germination percentage. Arunyanart *et al.* (1981) found six fungi to cause seed discoloration, *viz. Helminthosporium oryzae, Trichoconis padwickii, Fusarium semitectum, C. oryzae, Sarocladium oryzae* and *Curvularia lunata.* Duraiswamy and Mariappan (1983) isolated *Helminthosporium oryzae, Trichoconis padwickii* and *Curvularia lunata* from diseased grains. Lee *et al.* (1986) demonstrated that *Drechslera oryzae, Curvularia lunata, Trichoconis padwickii, Sarocladium oryzae, Alternaria tenuis,* and *Fusarium solani* isolated from discolored grains were pathogenic to rice in the test. He also found negative correlation between seedling emergence and incidence of *Trichoconis padwickii, Drechslera oryzae, Fusarium moniliforme, Pyricularia oryzae, Cercospora oryzae, Curvularia* spp., *Aspergillus* spp., *Penicillium* spp..

Shamsi *et al.* (1994) reported 11 fungi belonging to 9 genera and 14 fungi belonging to 11 genera were found to be associated with sheath rot symptoms and grains respectively. The fungi isolated from sheaths were *Aspergillus niger, Curvularia lunata, Curvularia lunata* var *aeria, C. pallescence, Drechslera oryzae, Fusarium arenaceum, F. pallidoroseum, Nigrospora oryzae, Penicillium* sp., *Phoma sorghina, Sarocladium oryzae* and *Trichoconis padwickii.* All the above mentioned fungi were also recovered from the sheath rot affected grains. *Alternaria alternata, Nigrospora sphaerica* and *Sclerotium oryzae* were only associated with affected grains.Grain spotting or discoloration is a complex malady in rice; it is an increasing problem in seed production as well as crop production in Bangladesh and elsewhere (Pagmanaghan 1947, Rangonathaiah 1985, Rodriguez *et al.* 1988 and Mia *et al.* 1994). It also causes both quantitative and qualitative losses in rice, resulted lot of breakage during milling and black clean rice, which ultimately reduces the market price (Vidhyasekaran and Ramadoss 1973 and Vidhyasekaran *et al.* 1984). A recent study revealed that more than 50% of the seed saved by farmers in Bangladesh are spotted or discolored (Mia 2004).

Khan *et al.* (2000) reported that flowering to milk stage of the rice plant is the most vulnerable stages for the seed infection with *C. lunata, F. semitectum, Phoma* sp. and *Trichoconis padwickii.* Inoculation of *Alternaria, Curvularia,* and *Penicillium* spp. at the flowering stage reported to produce a large number of discolored grains (Chai *et al.* 1991).

Islam *et al.* (2000) observed that highest lethal seed infection caused by *Fusarium moniliforme*, *Trichoconis padwickii* and *Curvularia* spp.

Haque *et al.* (2002) found that blotter method of seed health testing for three Boro rice varieties, collected from 21 selected farmers after different processing activities *viz.*, harvesting, threshing, cleaning, drying, storing and soaking, revealed the association of 19 fungal species under 15 genera. Storing was found to be most important factor that affected the population of associated fungi significantly. In general, storage fungi were found to increase while the seeds were in the storage and field fungi decreased. Earlier workers reported similar result (Christensen and Kaufman 1965, Miah and Fakir 1989). Cleaning was also found to be effective in reducing *B. Oryzae.*

To detect seed borne pathogens seed health testing is an important step in the management of crop diseases. Ora *et al.* (2011) detected 12 pathogens from 15 varieties of hybrid rice. The pathogens were *Alternaria tenuissima, Aspergillus* spp., *Bipolaris oryzae*, *Chaetomium globosum, Curvularia lunata, Fusarium moniliforme, Nigrospora oryzae, Penicillium* sp., *Phoma* sp., *Tilletia barclyana*, *Rhizopus stolonifer*, *Xanthomonas oryzae.* Of all the pathogens, *Aspergillus* sp., *Bipolaris oryzae, Fusarium moniliforme, Rhizopus stolonifer* were predominant. Sharma *et al.* (1987) and Bhutta and Ahmed (1994) had also reported similar result.

Archana and Prakash (2013) observed 16 genera of fungi comprising 27 species were found to be associated with the 69 rice seed samples were obtained from different states of India and used for testing their health status. Out of 69 samples 57 seed samples carried *Bipolaris oryzae, Curvularia lunata*, *Microdochium oryzae*, *Fusarium moniliforme* and *Rhizoctonia solani* are the causal agents of rice diseases such as black kernel, leaf scaled, bakanae disease and sheath blight respectively. These fungi are known to cause huge economic losses by reducing rice yield by 42%.

Shamsi *et al.* (2010) isolated seed borne fungi, *Aspergillus niger, Aspergillus* sp., *Curvularia* sp., *Cladosporium* sp., *Fusarium* sp., *Pyrenochaeta oryzae* and *Sarocladium oryzae* from three aromatic rice i.e. Kalizira, Kataribhog and Jira dhan (BRRI 34). They also recorded sheath rot on all the three rice varieties. Stem rot on Kalizira and sheath blotch on Kataribhog and the pathogens were *Sarocladium oryzae, Pyrenochaeta oryzae* and *Sclerotium oryzae* respectively.

2.3. Diseases of Rice

The most destructive seed borne fungal diseases of rice are Brown spot (*Bipolaris oryzae*), Blast (*Pyricularia oryzae*), Sheath rot (*Sarocladium oryzae*), Sheath blight (*Rhizoctonia solani*), Leaf scald (Microdochium oryzae), Seed rot and Seedling blight (*Bipolaris oryzae, Sclerotium rolfsi* and *Fusarium* spp.), Grain spot (*Curvularia lunata*, *Nigrospora oryzae, Phoma glumarum* and *Cladosporium* sp.). Most of the diseases of rice are seed borne. In Bangladesh, approximately 2.5 million tons of rice is lost annually due to diseases caused by seed borne pathogens (Fakir *et al.* 2003).

2.4. Chemical control of rice diseases

Hossain and Mia (2001) observed the highest disease incidence and corresponding lowest grain yield were recorded under farmers' practice where close planting spacing (15 cm x 15 cm) with high dose of nitrogen fertilizer was applied. The best treatment in respect of disease reduction and yield increase was found under the treatment when foliar spray with Tilt (0.1%) along with 40kg MP/ha was applied in addition to recommended practices. The positive influence of additional potash in reducing sheath blight disease has also been reported by Nandi and Chakraborty (1977).

Bhalli *et al.* (2001) evaluated 8 fungicides *viz*. Apron, Benlae, Derosal, Copperoxy, Chloride, Ridomil, Score, Topaz and Topsin-M to control mycelial growth of *Fusarium moniliforme.* Derosal was the best to inhibiting the mycelial growth *in vitro.* Farid *et al.* (2002) tested 12 seed samples of rice and all were found infected by *Bipolaris oryzae.* Four fungicides viz. Bavistin, Theophenate methyl, Tilt 250 EC and Dithane M-45 were evaluated against *Bipolaris oryzae.* Dithane-M was best with 100% reduction to the prevalence of the pathogen and inhibited the mycelial growth at 0.3% of the seed weight as seed treatments. All fungicides were effective against *Bipolaris oryzae* at higher concentration and showed highest germination. Mia *et al.* (2002) found that rice seed very often infected by field fungi *Bipolaris oryzae, Trichoconis padwickii, Fusarium moniliforme, F. oxysporum, F. semitectum, Pyricularia grisea, Curvularia lunata.* There is no accurate estimate about yield loss, roughly 10% yield loss of rice due to seed borne diseases.

Farid *et al.* (2002) observed amongst four fungicides *viz.,* bavistin, dithane M-45, hinosan and tilt 250 EC, only dithane M-45 was the best with 100% reduction of the prevalence of the pathogen and inhibited the mycelial growth.

Nghiep and Gaur (2005) found Vitavax, Thiram and Mancozeb were biologically active and have retained fungicidal properties against the test fungus *Bipolaris oryzae* even after 6 months of storage. The result was confirmed by Dharam and Sharma (1986) when they found that fungicides were active even after 20 years of storage.

Nghiep and Gaur (2005) reported that most of the seed borne fungi viz. *Bipolaris oryzae, Alternaria padwickii, Curvularia lunata* were eradicated by vitavax, Thiram and Mancozeb, whereas Bavistin was only effective slightly on *Alternaria padwickii* and *Curvularia lunata* but did not affect *Bipolaris oryzae* and other seed borne fungi like the other chemicals. They also observed that field fungi decreased progressively while storage fungi increased gradually during the increasing period of storage.

Butt *et al.* (2011) isolated four pathogenic fungi *viz., Alternaria alternata, Fusarium moniliforme*, *Curvularia* sp.and *Helminthosporium* sp. from 5 varieties of rice *viz.* KS-282, Basmati-385,370,198 and Kernel. Four chemical fungicides such as antracal, topsin, mancozeb and derosal were used to investigate their effect on seed borne mycoflora. Antracal completely stopped the growth of *Helminthosporium* sp. and *Curvularia* sp. but other 3 fungicides suppressed the growth of these fungi up to 50%.

2.5. Biological control of rice diseases

2.5.1. Plant extracts

.Bashar and Rai (1991) reported antifungal activity of extracts of some plant parts against *Fusarium oxysporum.* Nghiep and Gaur (2005) reported that most of the seed borne pathogens of rice *viz*., *Bipolaris oryzae*, *Alternaria padwickii*, *Curvularia lunata* and other seed borne fungi were controlled by neem oil.

Mohana *et al.* (2011) found that methanol extract of *Acasia nilotica, Caesalpinia coriaria, Emblica officinalis, Mimosops elengi* and *Lawsonia inermis* showed significant antifungal activity at 3500 mg/l concentration on rice pathogens by food poisoned technique.

Yesmin *et al*. (2012) reported that pathogenic fungi of rice were reduced up to 100% over the control, where provax was found best and significantly similar to garlic extract (1:1).

Mansur *et al.* (2013) observed reducing of seed borne infection and increasing germination of rice seed due to the treatment of some plants extracts and also reported that garlic (1:1) extract was most effective.

Shamsi and chowdhury (2016) reported twenty per cent ethanol extract of *Allium sativum, Artocarpus heterophillus, Azadirachta indica, Datura metel, Mangifera indica, Nerium indicum, Senna alata* and *Tagetes ercta* completely inhibited the radial growth of *Sarocladium oryzae.* Ten per cent ethanol extract of *D.metel* and *M. indica* were also completely inhibited the radial growth.

2. 5. 2. Antagonist

Antagonists act as biological control agent against rice pathogens. Biological control has now become one of the most exciting and developing areas in plant pathology because it has great potential to solve many agricultural problems (Baker and Cook 1983, Hossain *et al.* 2005). *Trichoderma harzianum* is known to be capable of producing antibiotics which might have suppressed the growth of the test pathogens (Akter *et al.*2014).

Prince *et al*. (2011) observed interactions at grade 4 between soil fungus *T. harzianum* and sugercane red rot pathogen *Colletotrichum falcatum*. Barakat *et al*. (2013) reported that volatile metabolics produced by an isolate of *T. harzianum* inhibited mycelial growth of *Botrytis fabae* by 39.77% after six days of incubation. The inhbition of the radial growth of the test fungi due to non-volatile metabolites may be attributed to the production of antibiotic substances in the culture filtrates and impoverishment of nutrient (Kexiang *et al*. 2002, Howell 2003 and Wool and Larito 2007).

Bashar and Chakma (2014) reported that volatile substances produced by *T.viride, A. niger*, *A. flavus* and *A. fumigatus* showed 29.75, 20.15, 15.78 and 12.25% inhibition respectively on *F. oxysporum.*Thakur and Harsh (2014) reported that volatile metabolites produced from the culture of *A. niger* showed 42.43% inhibition of mycelial growth of *C. gloeosporioides.* Zaidi *et al.* (2018) reported that duration of environmental stresses has posed a serious threat to global food security. Exploitation of *Trichoderma harzianum* has been hypothesized to play an important role in mitigating stresses and enhancing the yields of stress tolerant rice varieties. IRRI enhanced the efficacy of the *Trichoderma* by improving its BMP (Best Management Practices).

2.5.3. Integrated management

Ashrafuzzaman *et al*. (2011) reported the integratred managemenmt of sheath blight of aman rice. A total of 13 treatment combinations including controls with or without inoculum of the pathogen were tested. Severity increased with the increasing maturity of rice plants under all the treatments. The development was significantly least in plants treated with combined doses. The highest yeild of rice increase was recorded in plants tested with the combined doses.

Waris *et al*. (2016) found complete inhibition of seed borne pathogenic fungi when treated with garlic extracts alone and in combination with datura and neem leaf extracts. Islam and Monjil (2016) observed complete inhibition of sheath blight pathogen of rice when treated with four indigenous medicinal plants extracts *viz.*, tulsi, nishinda, thankuni and biskatali. They also reported that germination failure must have also been caused due to fungal infections. Germination and seedling vigor was increased due to combined effect of plant extracts.

CHAPTER 3

MATERIALS AND METHODS

3.1. Collection of seed samples and their sampling sites

The present study was based on spotted grains of four commercially cultivated rice varieties *viz.* BRRI 28, 29, Pajam and Kalizira collected from Pabna and Sirajgonj (Rajshahi division), Tangail and Gazipur (Dhaka division), Comilla and Laksmipur (Chittagong division), Potuakhali and Barishal (Barisal division), Dinajpur and Gaibandha (Rangpur Division), Satkhira and Chuadanga (Khulna Division), Habiganj and Sunamganj (Sylhet Division) during Boro and aman seasons of 2012 and 2013 and 40 rice samples *viz*. Hybrid 2, 3, 4, BR 7, 11, 12, 14 ,16, 22, 23, 25, 26 and BRRI 28 to BRRI-55 were collected from Bangladesh Rice Research Institute at Joydebpur. Samples were collected after harvesting and placed in clean brown paper bag labeled properly and preserved at 25^0 C ±2 °C) for subsequent use.

3.2. Quality analysis of rice seeds

Quality status of rice seeds were determined by seed quality analysis. Ten gms of seed was weighed by an electrical balance. There are two types of contaminants *viz.* inert matter and weed seeds were present in rice seeds. Four types of abnormal seeds were recorded in the present study. The abnormal seeds were discoloured, wrinkled, spotted and undersized. All of them were weighed separately and listed in Table 1.

3.3. Isolation, purification and identification of fungi associated with diseased rice grains

The fungi were isolated from the samples following the “Tissue Planting method” on PDA medium (CAB 1968) and “Blotter method of ISTA. Two hundred seeds of each sample were placed on three layers of moist blotting paper (Whatman No. 1) in Petri plates. The seeds were washed with sterile water and then surface sterilized by dipping in 10% Chlorox solution for 5 minutes. Seeds were placed in each plate and incubated at 25 ± 2ºC for 5 - 7 days.Fungi grown in the seeds were transferred to separate PDA plates and PDA slants for further studies.

3.3.1. Tissue planting method

Surface sterilized seeds were used to isolate the fungi from the specimen. The inocula were washed in sterile water and then surface sterilized by dipping in 10% Chlorox for 3-5 mins. Then they were transferred into a sterile Petri plate containing sterile blotting paper to remove the surface water.

The inocula were placed in Petri plates containing sterilized potato Dextrose Agar (PDA) medium (Potato 200 g, Dextrose 20 g, Agar 15 g, Distilled water 1000 ml). Each Petri plate contained 15 ml of PDA medium with an additional of 1 drop of lactic acid which was used for checking the bacterial growth. A total number of 30 inocula were transferred in 10 Petri plates. Then the inoculated plates were incubated at room temperature (25 ± 2 °C) for 5-7 days.

3.3.2. Blotter method

In this technique, moist chambers were made by placing two layers of filter paper on the bottom of the Petri plate and covered with upper plates. In each Petri plate surface sterilized (with 10% chlorox for 3-5 minutes) healthy and affected seed samples of rice were placed on the filter paper inside the Petri plates. A total number of 200 inocula were transferred in 10 Petri plates and were moistened with sterilized water (autoclaved at 15 lbs pressure and 120°C temperature) and incubated under room temperature.

The fungi growing out of the inocula were examined and identified whenever possible and transferred to PDA slants. The isolates were purified following dilution plate method (Anon. 1968), maintained on PDA slants and stored at (10 ± 0.5 °C) in an incubator for future studies.

Reading of fungal colonies grown out of the inocula were taken on 5th day of the inoculation and continued for two weeks depending on the medium and the fungal organism associated with the inocula.

Percentage frequency of the occurrence of the fungal isolates was calculated by adopting the following formula (Spurr and Welty 1972):

$$\% \text{ frequency} = \frac{\text{No. of inocula from which fungal isolates were raised}}{\text{No. of inocula cultured}} \times 100$$

3.3.3. Preservation of the fungi

The fungi that were obtained from the inocula were preserved in the PDA slants for the future studies and identification. Those preserved fungi in the slant were stored at 4-10 ºC in refrigerator.

3.3.4. Medium used for isolation of fungi

Preparation of Potato Dextrose Agar (PDA) medium: 1000 ml

Peeled and sliced potatoes	200 g
Dextrose	20 g
Agar (Powder)	15 g
Distilled water	1000 ml
PH	6.0

PDA medium was found suitable for growth of fungi than almost all the media. Most fungi grew best at natural medium at PH 6.0 (Konger, 1971).

3.3.5. Chemicals used in preparation of slides

Preparation of lacto phenol

Composition of Lacto phenol solution used as mounting medium:

Phenol crystals	20 g
Lactic Acid	20 ml
Glycerol	40 ml
Distilled water	20 ml

After weighing, the constituents were taken in a conical flask to which distilled water was added. The flask was shaken well till a homogenous solution was obtained.

3.3.6. Preparation of cotton blue stain

One gram of cotton blue was added to 100 ml of lacto phenol and shaken well till it was dissolved. The solution of Lacto phenol and cotton blue was stored in cool dark place. Generally, it is stored in an amber colored bottle.

3.4. Morphological studies of fungi

Detailed morphological studies of the fungal isolates were made in order to determine their identity. For microscope observations fungal structures like mycelia, spore bearing structures and spores were scrapped off from the surface with a scalpel or blade or picked up with a needle and was mounted in lacto phenol over a clean slide. In case of hyaline structures, a little amount of aniline blue (cotton blue) was added to the mounted fluid. A clean cover was placed over the material; excess fluid was removed by shaking with blotting paper and examined under microscope. The microscope structural view of the fungi was taken by a digital camera. All the specimens, included in the present study were preserved in the Mycology and Plant pathology section, Department of Botany, University of Dhaka, Bangladesh.

The isolated fungi were identified based on morphological characteristics observed under a compound microscope following standard keys (Barnett and Hunter 1972, Booth 1971, Ellis 1971, 1976, Ellis and Ellis 1997, Benoit and Mathur 1970, Booth 1971, Subramanian 1971 and Sutton 1980). Percentage of prevalence of fungi in different specimens was also recorded.

3.5. Pathogenecity test of isolated fungi in test tubes

Pathogenicity of the test fungi was done following seed inoculation technique (Reddy and Subbayya 1989). Four hundred healthy and 400 spotted grains were selected from BRRI 29 and soaked in distilled water in a beaker for three hours then surface sterilized with 10% chlorox for 10 minutes. One hundred milliliter of spore suspension of the test fungi at 10^4 concentrations was prepared in a 250 ml sterilized beaker. Four hundred seeds from each variety were inoculated with spore suspension and then incubated for 30 minutes.

Two hundred of each healthy, spotted and inoculated seeds of BRRI 29 rice varietiy were placed in sterilized 8 inch cotton plugged test tubes containing 5 ml 2% water agar medium. Healthy seeds served as control. Observation was made for 4 weeks at 3 days intervals. Germination percentage of seeds, development of disease symptoms and mortality of seedlings were recorded on healthy, diseased and inoculated seeds of BRRI-29 rice variety. After 10 days of inoculation, pathogenic fungl were re-isolated from diseased and inoculated seeds and the seedlings from those healthy seeds remained fresh.

3.6. Transmission of pathogenic fungi from seed to seedlings in pot experiments

Seeds were artificially inoculated with conidial suspension of the test fungi for 1 hour. Plastic pots were prepared with formalin treated soil. One hundred inoculated seeds for each pathogen were sowing in plastic pots. Observation was made for 4 weeks at 5 days intervals. Percentage of germination of seeds and mortality of seedlings, development of disease symptoms were recorded after 15 to 20 days of germination. Healthy seeds served as control.

Data on different parameters were analysed following computer package MSTAT-C and means were compared using Duncans Multiple Range Test (DMRT). The data were collected and evaluated by analysis of variance (ANOVA) by using STAR statistical program.

3.7. Fungitoxicity of fungicides against the test fungi *in vitro*

3.7.1. Prparation of fungicides used in the experiment

Ten fungicides i.e. Bavistin 50 WP, Capvit 50 WP, Dithane M-45, Greengel, Hayvit 80 WP, Indofil M-45, Ridomil MZ Gold, Salcox 50 WP, MC Sulphur 80 and Tall 25 EC were collected from Krishi Upokoron Biponi Kendro, Khamarbari, Farmgate, Dhaka. For each fungicide, a stock solution having the concentration of 10000 ppm was prepared. Then calculated amount of the stock solution of a fungicide was supplemented with sterilized PDA medium to get the final concentration of 100, 200, 300, 400 and 500 ppm etc. In the control set required amount of sterile water instead of fungicide solution was added to the PDA medium. Five mm mycelial agar disc cut from the margin of actively growing culture of test fungi and then it was inoculated at the centre of the plate. Three replications were maintained in both the cases.

3.8. Effects of plant extracts against the radial growth of test fungi *in vitro*

3.8.1. Preparation of plant extracts used in the experiment

For *in vitro* effect of plant parts extracts on the vegetative growth of test pathogens, ten plants *viz., Allium sativum* L., *Artocarpus heterophyllus* Lamk., *Asparagus racemosus* Willd., *Azadirachta indica* A. Juss., *Citrus medica* L., *Datura metel* L., *Mangifera indica* L., *Nerium indicum* Mill.*, Senna alata* (L.) Roxb. and *Tagetes erecta* L. were selected. The desired parts of each plant were thoroughly washed in tap water, air dried and were prepared by crushing the known weight of fresh materials with ethanol in ratio of (1:1, w/v). The mass of a plant part was squeezed through fine cloth and the extracts were centrifuged at 3000 rpm for 20 min. The supernatants were filtered through Whatman filter paper No.1 and the filtrate was collected in 250 ml Erlenmeyer conical flasks. The requisite amount of the filtrate of each plant extract was mixed with PDA medium in which plant extracts were in 5, 10 and 20% concentrations. In the control set required amount of sterile water instead of plant extract was added to the PDA medium. Five mm mycelial agar disc cut from the margin of actively growing culture of test fungi and then it was inoculated at the centre of the plate. Three replications were maintained in both the cases.

The radial growth of the colonies was measured at the 5th day of incubation in both cases. The per cent growth inhibition of each test fungi was calculated by using the following formula described by Bashar and Rai (1991).

$$I = \frac{C - T}{T} \times 100$$

Where, I = per cent growth inhibition

C = growth in control

T = growth in treatment.

Data on different parameters were analysed following computer package MSTAT-C and means were compared using Duncans Multiple Range Test (DMRT).The data were collected and evaluated by analysis of variance (ANOVA) by using STAR statistical program.

3.9. Evaluation of antagonistic potential of some soil fungi against test pathogens

3.9.1 Colony interaction

Six antagonistic soil fungi *viz*. *Aspergillus flavus* Link, *A. fumigatus* Fresen., *A. niger* Tiegh., *Penicillium* sp., *Trichoderma harzianum* Refai *and T. viride* Pers. were isolated from the rhizosphere of the several healthy rice crop fields following serial dilution method and selected to test their antagonistic potential against the pathogenic fungi following dual culture technique described by Bashar and Rai (1994). Five mm blocks of each test pathogen and selected soil fungus were placed 3 cm apart on PDA medium in paired combination.Three replications were maintained in each case.The inoculated plates were incubated at 25+1 temperature for 7 days.The colony growth of the pathogen was measured at both sides, that is towards and opposing each other from their central loci.The radial growth was measured after 3, 5 and 7 days. In dual culture, assessment of colony interactions grading were done based on intermingling and inhibition zone which were determined by the model of Skidmore and Dickinson (1976). The grades and types are follows:

Grade 1: Mutually intermingling growth were both fungi grew into one another without any microscopic sign of interaction.

Grade 2: Intermingling growth where the fungus under observation has ceased the growth and is being overgrown by another colony.

Grade 3: Intermingling growth where the fungus being observed into the opposed fungus either above or below its colony.

Grade 4: Slight inhibition with a narrow demarcation line (1-2 cm).

Grade 5: Mutual inhibition at a distance more than 2 mm.

Per cent inhibition of the growth of the test fungi due to the presence of antagonists were also calculated as follow:

$$I = \frac{r1 - r2}{r1} \times 100$$

Where, I = per cent growth inhibition

r_1 = the radial growth of the test fungus towards the opposite side

r_2 = the radius of the test fungus towards the soil fungus

The experiment was performed twice. The data were collected as inhibition percentage of the radial growth of the pathogen in mm in each replication and evaluated by analysis of variance (ANOVA) by using STAR statistical program.

3.9.2 Effect of volatile substances emanating from the cultures of the soil fungi on the radial growth of the test pathogens

To asses the effects of volatile substances of the soil fungi on the test pathogens ,each selected soil fungus was grown in 9 cm Petri plates on PDA medium. After 7 days, the lid of each Petri plate was replaced by the same size bottom plate, containing 15 ml PDA media, centrally inoculated with a test pathogen and was enclosed by 3 layers of parafilm, to prevent the loss of volatile substances. Control was also preapared in the same way but with the test pathogen at the bottom. After 7 days in all sets colony diameter of the test pathogens were measured.

3.9.3 Effect of non-volatile substances (culture filtrates) of the soil fungi on the radial growth of the test pathogens

To evaluate the effects of non volatile substances present in the culture filtrates of the selected soil fungi on the test pathogens, three equal size blocks (0.5 mm) each of individual soil fungi, cut from the actively growing margins of 5 days old cultures, were inoculated separately into 250 ml conical flasks each containing 100 ml sterilized potato dextrose broth medium. After 10 days of incubation the static cultures were filtered through Whatman filter paper No.44 and then centrifuzed at 3000 rpm for 20 minutes. Ten ml filtrates of each fungus were added in 90 ml sterilized PDA medium separately, the conical flask containing the PDA medium and culture filtrates was moved in different directions gently in the laminar air flow cabinet to get the homogenous distribution. Each Petri plate contained 15 ml of PDA supplemented medium and metabolites with an addition of 1 drop of lactic acid which was used to check the bacterial growth. Each Petri plate was allowed to solidify and inoculated centrally with a 5 mm agar disc, cut from the margin of actively growing culture of a test pathogen. In control set, Petri plate containing PDA medium and requisite amount of sterilized distilled water instead of culture filtrates were inoculated with a test pathogen as described above. Three replications of each treatment were maintained. All the Petr plates were incubated at 25 ± 2ºC and the colonies were measured after 7 days.

Effects of volatile and non- volatile metabolites of the selected soil fungi against the test pathogens were also calculated following the methods described by Dennis and Webster (1971b) and Bashar and Rai (1994). The per cent growth inhibition of radial growth of test pathogen was calculated by the formula given above. The results were evaluated by analysis of varience by using STAR statistical program.

3.10. Integrated management of test pathogens *in vivo*

A pot experiment was conducted in the earthen pot of Botanical garden, University of Dhaka, to assess the single and combined doses of fungicides, plant extracts and antagonists for the management of nine rice pathogens. A total of 13 treatments including controls with or without inocula of the pathogens were tested.The experiment was designed in a randomized block design with 3 replications. Pot soil was prepared by mixing sandy loam soil and decomposed organic fertilizer at the ratio 4:1. The plastic pots (20 cm diameter) were filled with 2 kg soil which was treated with formalin. A high yielding susceptible rice variety BRRI-29 was selected for this study (Anonimous 2004).

The treatments and their combinations were : T1 = Bavistin, T2 = Tall, T3 = *Azadirachta indica*, T4 = *Citrus medica* T5 = *Trichoderma harzianum*, T6 = Bavistin + *Azadirachta indica* + *Trichoderma harzianum* T7 = Bavistin + *Citrus medica* + *Trichoderma harzianum*, T8 = Tall + *Azadirachta indica* + *Trichoderma harzianum*, T9 = Tall + *Citrus medica* + *Trichoderma harzianum*, T10 = Bavistin +Tall + *Azadirachta indica* + *Citrus medica*, T11 = Bavistin +Tall + *Azadirachta indica* + *Citrus medica* + *Trichoderma harzianum* , T12 = Control (with inocula), T13 = Control (without inocula). Seeds were suspended with the inocular suspension of test pathogens for 1 hour and treated with the above mentioned treatments for 2 hours. Then the seeds were sowing in plastic pots. Treatments were applied at 10 days intervals. Datas were recorded after 15, 20, 25 days of germination. Twenty nine days old seedlings of BRRI 29 variety were uprooted carefully to avoid root injury. The shoot and root length were taken separately against all test pathogens. The percentage of germination and mortality was also recorded.

The experiment was performed twice. The data were collected as inhibition percentage of the radial growth of the pathogen in mm in each replication and evaluated by analysis of variance (ANOVA) by using STAR statistical program.

CHAPTER 4

RESULTS AND DISCUSSION

4.1. Seed quality analysis

Quality status of rice seeds were determined by seed quality analysis. Quality analysis showed that the percentage of pure seed varies from 94-99% (Table 1). The germination percentage of rice seeds of different varieties were in the range of 62-100% on the 7th day of incubation and the mortality percentage were in the range of 10-30% on the 20th day of incubation.

4.1.1. Seed contaminants

Seed contaminants and its frequency of occurrence in different varieties are included in Table 1. Two types of contaminants were recorded in the present study. The contaminants were inert matter and weed seeds. The contaminants varied significantly from one another with respect to different varieties. The occurrence of inert matter varied from 0.10-1.80% (Table 1). The highest per cent of the inert matter (1.80%) was found in BRRI 50 and the lowest (0.10%) in BRRI 40. In case of weed seeds, the highest per cent of it (0.90%) was found in BRRI 29 and the lowest (0.10%) in BRRI 38 (Table 1).

4.1.2. Abnormal seeds

Abnormal seeds and its frequency of occurrence in different varieties of rice are shown in Table 1. The highest amount of abnormal seeds (4.20%) was recorded in BRRI-50 whereas the lowest count (0.20%) was recorded in BRRI 54. Four types of abnormal seeds were recorded in the present study. The abnormal seeds were discoloured, wrinkled, spotted and undersized. The highest occurrence of discoloured seed (2.90%) was recorded in BRRI 50 while the lowest (0.2%) in BRRI 16. The highest per cent of wrinkled seed (0.80%) was found in BRRI 50 and the lowest (0.2%) in BRRI 38. The highest per cent of spotted seed (1.20%) was found in BRRI 29 and the lowest (0.1%) in BRRI 42. The highest per cent of undersized seed (0.60%) was found in BRRI 36 and the lowest (0.20%) in BRRI 37 (Table 1).

4.2. Determination of seed germination

The present study revealed that BRRI 29 showed highest seed germination (100%) while Hybrid 3 showed lowest (62%) seed germination (Table 2). Maximum germination of other rice varieties were 98, 97, 96, 95, 94 and 90% which was found in BRRI 28, pajam, BRRI 34, 31 36 and 38. The standard germination percentage was between 92-100% as recommended by Anonymous (1990). Mansur *et al*. (2013) reported that the prevalence of seed-borne infection is also responsible for lower germination (Table 2).

4.3. Determination of seedling mortality

The highest (30%) mortality percentage of rice seedling was found in BRRI 50 and the lowest (10%) value was found in BRRI 28. Maximum mortality of other rice varieties were 28, 27, 25, 24, 22, 21 and 20% etc. which were found in BR 23, 16, 22, 12, 11, 7 and BRRI 35 rice varieties, respectively.These results showed similarity with the findings of Chowdhury *et al.* (2014). He reported that germination percentage of seeds in all impermeable containers was above 80% and seedling mortality was found to be positively correlated with moisture content and storage duration (Table 2).

4.4. Detection of seed borne fungi associated with rice seeds (Table 3)

A total of 96 rice seed samples consisting each of different varieties were obtained from 14 districts under 7 divisions including BRRI of Bangladesh and were used for testing their health status and the results are furnished in table 3. Totally 15 genera and 25 species of fungi were isolated from these samples through Tissue planting and Blotter method .

Table 1. Per cent purity status of different rice varieties

Sl. No.	Name of varieties	Pure seed	Abnormal seeds				Total	Inert matter	Weed seeds
			Discoloured	Wrinkled	Spotted	Undersized			
1	Hybrid 2	97	0.5	0.5	0.8	-	1.8	1.2	-
2	Hybrid 3	98	0.5	0.3	-	0.3	1.1	1.0	
3	Hybrid 4	98	1.0	-	-	-	1.0	1.0	-
4	BR 7	98	0.5	-	0.5	-	1.0	1.0	-
5	BR 11	98	0.8	0.4	-	-	1.2	0.8	-
6	BR 12	99	0.8	-	0.2	-	1.0	-	-
7	BR 14	98	1.0	-	-	-	1.0	1.0	-
8	BR 16	99	**0.2**	0.6	0.2	-	1.0	-	-
9	BR 22	99	1.0	-	-	-	1,0	-	-
10	BR 23	98	1.0	-	0.4	-	1.4	0.6	
11	BR 25	98	1.1	-	-	-	1.1	0.9	-
12	BR 26	97	1.0	-	0.5	-	1.5	1.0	0.5
13	BRRI 28	96	1.0	0.5	0.5	0.5	2.5	1.0	0.5
14	BRRI 29	94	1.0	0.5	**2.9**	0.5	4.9	0.2	**0.9**
15	BRRI 30	97	2.0	-	0.3	-	2.3	0.7	-
16	BRRI 31	95	1.0	0.5	1.0	0.5	3.0	1.3	0.7
17	BRRI 32	97	1.0	0.5	1.0	0.5	3.0	-	-
18	BRRI 33	98	1.4	-	-	-	0.4	0.6	-
19	BRRI 34	99	1.0	-	-	-	1.0	-	-
20	BRRI 35	99	0.5	-	-	-	0.5	0.5	-
21	BRRI 36	98	0.8	0.3	-	0.6	1.7	0.3	-

Table 1 contd.

Sl. No.	Name of varieties	Pure seed	Abnormal seeds				Total	Inert matter	Weed seeds
			Discoloured	Wrinkled	Spotted	Undersized			
22	BRRI 37	98	0.4	-	1.0	**0.2**	1.6	0.4	-
23	BRRI 38	98	0.5	**0.2**	-	0.3	1.0	0.9	**0.1**
24	BRRI 39	97	1.0	-	1.0	-	2.0	1.0	-
25	BRRI 40	99	0.5	-	0.4	-	0.9	**0.1**	-
26	BRRI 41	98	1.2	0.6	-	-	1.8	0.2	-
27	BRRI 42	99	0.6	-	**0.1**	-	0.7	0.3	-
28	BRRI 43	99	1.0	-	-	-	1.0	-	-
29	BRRI 44	97	0.6	0.6	1.2	-	2.4	0.6	-
30	BRRI 45	98	0.8	-	1.0	-	1.8	-	0.2
31	BRRI 46	98	0.8	-	0.2	0.3	1.3	0.4	0.3
32	BRRI 47	97	1.1	-	1.0	-	2.1	0.9	-
33	BRRI 48	97	1.0	-	1.0	-	2.0	0.7	0.3
34	BRRI 49	98	1.0	-	1.0	-	2.0	-	-
35	BRRI 50	94	**2.9**	**0.8**	-	0.5	**4.2**	**1.8**	-
36	BRRI 51	97	1.0	0.5	0.3	0.5	2.3	0.7	-
37	BRRI 52	96	2.0	-	-	-	2.0	1.3	0.7
38	BRRI 53	99	1.0	-	-	-	1.0	-	-
39	BRRI 54	99	0.3	-	-	-	0.3	0.4	0.3
40	BRRI 55	98	1.0	-	-	-	1.0	0.5	0.5
41	Pajam	95	1.0	0.7	1.0	**1.0**	3.7	0.5	0.8
42	Kalijira	95	1.5	0.7	0.7	**1.0**	3.9	0.3	0.8

Table 2. Per cent germination of rice seeds and seedling mortality

Sl. No.	Name of varieties	Per cent Germination		Per cent Mortality
		5th day	7th day	20th day
1	Hybrid 2	60	80	18
2	Hybrid 3	60	**62**	18
3	Hybrid 4	75	80	19
4	BR 7	90	95	21
5	BR 11	85	90	22
6	BR 12	85	90	24
7	BR 14	70	80	25
8	BR 16	65	75	27
9	BR 22	75	80	25
10	BR 23	60	70	28
11	BR 25	91	95	20
12	BR 26	62	70	20
13	BRRI 28	90	99	**10**
14	BRRI 29	92	**100**	12
15	BRRI 30	90	96	12
16	BRRI 31	96	97	12
17	BRRI 32	94	96	14
18	BRRI 33	93	95	17
19	BRRI 34	95	97	15
20	BRRI 35	92	95	20
21	BRRI 36	90	94	15

Table 2 contd.

Sl. No.	Name of varieties	Germination (%)		Mortality (%) at 20 days
		5^{th} day	7^{th} day	
23	BRRI 38	90	95	12
24	BRRI 39	80	95	15
25	BRRI 40	55	70	13
26	BRRI 41	90	95	19
27	BRRI 42	85	97	12
28	BRRI 43	75	94	11
29	BRRI 44	86	92	16
30	BRRI 45	94	96	15
31	BRRI 46	90	95	13
32	BRRI 47	50	70	12
33	BRRI 48	80	93	20
34	BRRI 49	90	95	13
35	BRRI 50	90	95	**30**
36	BRRI 51	95	97	15
37	BRRI 52	90	96	12
38	BRRI 53	90	94	14
39	BRRI 54	82	86	15
40	BRRI 55	94	95	18
41	Pajam	98	99	14
42	Kalijira	84	90	30

Table 3. Fungi associated with diseased rice grains during the tenure of 2012 - 2014

Sl. No.	Name of fungi
1	*Alternaria alternata* (Fr.) Keissler
2	*Aspergillus clavatus* Desm.
3	*A.flavus* Link
4	*A. fumigatus* Fresen.
5	*A. niger* Tiegh.
6	*A. ochraceus* K. Wilh.
7	*A. oryzae* (Ahlb.) Cohn
8	*A. terreus* Thom.
9	*Chaetomium globosum* Kunze ex Fr.
10	*Cladosporium cladosporioides* (Fresen.) G.A. de Vries
11	*Colletotrichum gloeosporioides* (Penz.) Penz. & Sacc.
12	*Curvularia lunata* (Wakker) Boedijn
13	*C. lunata* var *aeria* (BAT., J.A. Lima & C.T. Vasconc.) M.B. Ellis
14	*Drechslera oryzae* Breda de Haan (Subramanian and Jain)
15	*Fusarium moniliforme* Sheldon
16	*F. oxysporum* Schltdl.
17	*F. solani* (Mart.) Sacc.
18	*Microdochium oryzae* (Hashloka and Yokogi) Sam. and Hal.
19	*Nigrospora oryzae* (Berk. & Broome) Petch
20	*Penicillium* sp_1.
21	*Penicillium* sp_2.
22	*Pestalotiopsis guepinii* (Desm.) Stay.
23	*Rhizopus stolonifer* (Ehrenb.) Vuill.
24	*Sarocladium oryzae* (Sawada) W. Gams and D. Hawks
25	*Trichoderma viride* Pers.

4.5. Per cent frequency of association of fungi with diseased seeds of Boro rice varieties collected from BRRI on July 2012

The occurance of different fungi varied among the seasons. During Boro season 23 species of fungi belonging to 13 genera were found to be associated with 11 varieties of Boro rice after collection from BRRI in July 2012 (Table 4).

Highest per cent frequency (25.7) was found in *Drechslera oryzae* and lowest (2.0) in *Pestalotiopsis guepinii.* Both were isolated from BRRI 29 rice variety.Maximum per cent frequency of other fungi, i.e. *Aspergillus flavus* (24.6), *A. fumigatus* (22), *Rhizopus stolonifer* (20.7), *Chaetomium globosum* (20), *Penicillium sp.* (15.7), *Fusarium moniliforme* (14), *Curvularia lunata* (10.7), *F. oxysporum* (9.7), *F. solani* (9.0), *Fusarium sp*. (9.0), *Nigrospora oryzae* (8.7), *Alternaria alternata* (8.6), *A. clavatus* (8.0), *Trichoderma viride* (6.7) and *Sarocladium oryzae* (2.3) were found in different rice varieties. *Pestalotiopsis guepinii* was isolated first time from rice grains in Bangladesh (Table 4).

After 3 months of storage at 25±2°C, frequency of field fungi were decreased while storage fungi increased. Frequency percentage of *Drechslera oryzae* and *Pestalotiopsis guepinii* were 22.7 and 1.2% respectively while frequency of *Aspergillus flavus*, *A. fumigatus, A. niger, Penicillium* sp. and *Rhizopus stolonifer* were increased i.e. 28.6, 24, 23, 18 and 21.7%, respectively.

Frequency (%) of *Alternaria alternata, Curvularia lunata, Fusarium moniliforme, F. oxysporum, F. solani*, *N. oryzae* and *Sarocladium oryzae* were decreased i.e. 7.7, 9.7, 10, 8, 7, 8 and 2% respectively (Table 5). After 6 months of storage at 25±2°C, highest frequency (28%) was observed in *A. niger* in BRRI 29 and lowest (1.0%) frequency in *P. guepinii* in BRRI 29 (Table 6).

Table 4. Per cent frequency of association of fungi with diseased seeds of Boro rice varieties after collection from BRRI (July 2012)

		Boro rice varieties and per cent frequency										
Sl. No.	Name of fungi	Hybrid-2	Hybrid-3	BR-7	BR-14	BRRI-28	BRRI-29	BRRI-35	BRRI-36	BRRI-45	BRRI-47	BRRI-50
1.	*Alternaria alternata*	-	2.7	-	5.3	6.7	5.0	-	8.6	-	-	-
2.	*Aspergillus clavatus*	-	-	-	2.2	4.0	-	8.0	-	-	-	-
3.	*Aspergillus flavus*	8.3	10.3	15.7	10.0	10.0	3.0	24.6	12.0	10.7	11.0	-
4.	*Aspergillus fumigatus*	10.7	5.7	8.3	20.0	10.0	-	12.0	10.7	14.7	22.0	-
5.	*Aspergillus niger*	20.3	10.0	20.7	10.0	20.0	8.0	20.7	18.3	20.3	16.7	15.7
6.	*Aspergillus terreus*	-	-	-	-	8.7	-	-	-	-	-	10.0
7.	*Aspergillus ochraceus*	-	-	-	-	2.3	6.7	-	-	-	-	3.3
8.	*Chaetomium globosum*	-	-	-	-	-	8.0	-	20	-	-	-
9.	*Cladosporium cladosporioides*	-	-	9.7	8.7	-	8.0	-	-	-	-	-
10.	*Curvularia lunata*	-	-	2.6	-	10.7	10.0	-	3.0	-	-	-
11.	*C. lunata* var.*aeria*	-	-	-	3.3	2.3	6.0	-	-	-	-	-
12.	*Drechslera oryzae*	2.0	-	7.7	20.3	-	**27.7**	12.0	14.3	-	-	-
13.	*Fusarium moniliforme*	-	-	-	5.0	14.0	4.0	-	-	-	-	10.00
14.	*F. oxysporum*	-	-	4.0	-	-	9.7	-	-	-	-	-
15.	*F. solani*	-	-	5.0	-	-	9.0	-	-	-	-	-
16.	*Microdochium oryzae*.	-	-	-	5.7		9.0		-	6.7		-
17.	*Nigrospora oryzae*	-	-	-	-	2.3	2.7	8.7	-	-	-	-
18.	*Penicillium* $sp_{1.}$	9.7	15.7	4.7	-	-	-	3.7	-	5.7	3.3	10.7
19.	*Penicillium* $sp_{2.}$	-	-	-	-	-	-	-	-	3.6	-	-
20.	*Pestalotiopsis guepinii*	-	-	-	-	-	**2.0**	-	-	-	5.6	-
21.	*Rhizopus stolonife*r	3.3	-	-	-	-	-	-	-	-	-	20.7
22.	*Sarocladium oryzae*	-	-	-	-	2.3	3.3	-	-	-	-	-
23.	*Trichoderma viride*	-	-	-	-	2.6	-	-	3.0	-	-	6.7

- represents absence of respective fungus

Table 5. Per cent frequency of association of fungi with seeds of Boro rice varieties after 3 months of storage at 25±2 °C

		Boro rice varieties and mean per cent frequency										
Sl. No.	Name of fungi	Hybrid-2	Hybrid-3	BR-7	BR-14	BRRI-28	BRRI-29	BRRI-35	BRRI-36	BRRI-45	BRRI-47	BRRI-50
1	*Alternaria alternata*	-	2	-	4.3	-	4	7.7	-	-	-	-
2	*Aspergillus clavatus*	-	-	-	8.7	4	-	-	-	-	-	-
3	*A. flavus*	12.3	15.3	16.7	16	10	4	25	12	13.7	24.6	10
4	*A. fumigatus*	11.7	8.7	12.3	24	14	-	14	10.7	14.7	22	-
5	*A. niger*	23.3	10	20.7	18	20	10	22.7	18.3	22.3	16.7	18.7
6	*A. terreus*	-	-	-	-	5	-	-	-	-	-	12
7	*A. oryzae*	-	-	4.6								
8	*Chaetomium globosum*	-	-	-	-	12	4	8	-	-	-	-
9	*Cladosporium cladosporioides*	-	-	-	-	8.8	4	-	-	-	-	-
10	*Curvularia lunata*	-	-	2.7	-	9.7	-	-	2.8	-	-	-
11	*C. lunata* var. *aeria*	-	-	-	3	2.2	5	-	-	-	-	-
12	*Drechslera oryzae*	-	-	17	20.3	-	**25.7**	10	14.3	-	-	-
13	*Fusarium moniliforme*	-	-	-	5	8	4	-	-	-	-	10
14	*F. oxysporum*	-	-	3	-	-	8	-	-	-	-	-
15	*F. solani*	-	-	4	-	-	7	-	-	-	-	-
16	*Microdochium oryzae*.	-	-	-	6.7	-	8	-	-	-	-	-
17	*Nigrospora oryzae*	-	-	-	-	2.2	8	-	-	-	-	-
18	*Penicillium* sp$_{1.}$	10	18	6.7	-	-	8	13.7	-	12.7	5.6	14.7
19	*Penicillium* sp$_{2.}$	-	-	-	-	-	-	-	-	9.6	7.8	-
20	*Pestalotiopsis guepinii*	-	-	-	-	-	**1.2**	-	-	-	-	-
21	*Rhizopus stolonifer*	12	-	-	21.7	-	8	-	-	-	-	15.7
22	*Sarocladium oryzae*	-	-	-	-	4	-	-	-	-	-	-
23	*Trichoderma viride*	-	-	-	-	3.6	-	-	-	-	-	-

- represents absence of respective fungus

Table 6. Per cent frequency of association of fungi with seeds of Boro rice varieties after 6 months of storage at 25±2 °C

		Boro rice varieties and mean percent frequency										
Sl. No.	Name of fungi	Hybrid-2	Hybrid-3	BR-7	BR-14	BRRI-28	BRRI-29	BRRI-35	BRRI-36	BRRI-45	BRRI-47	BRRI-50
1	*Alternaria alternata*	-	1.7	-	-	-	-	-	-	-	-	-
2	*Aspergillus clavatus*	-	-	4.7	9	-	5	-	-	-	-	-
3	*Aspergillus flavus*	18.3	18.3	18.7	18	12	14	30	14	16.7	25	20
4	*Aspergillus fumigatus*	14.7	10	18.3	25	15	15	12	12.7	18.7	24	-
5	*Aspergillus niger*	24.3	12	24.7	22	22	**28**	24.7	20.3	24.3	16.7	20
6	*Aspergillus oryzae*	-	-	-	-	2.3	6.7	-	-	-	-	-
7	*Aspergillus terreus*	-	-	-	-	8.7	-	-	-	-	-	3
8	*Chaetomium globosum*	-	-	-	-	5	8	-	-	-	-	-
9	*Cladosporium cladosporioides*	-	-	-	-	-	3	-	-	-	-	-
10	*Curvularia lunata*	-	-	2.6	-	4.7	5	-	3	-	-	-
11	*C. lunata* var. *aeria*	-	-	-	2	2	3	-	-	-	-	-
12	*Drechslera oryzae*	-	-	7.7	10.3	-	4	-	12.3	-	-	-
13	*Fusarium moniliforme*	-	-	-	4	2	4	-	-	-	-	9
14	*F. oxysporum*	-	-	2.3	-	-	3.4	-	-	-	-	-
15	*F. solani*	-	-	-	3	-	6.7	-	-	-	-	-
16	*Microdochium oryzae*.	-	-	-	4	-	5	-	-	-	-	-
17	*Nigrospora oryzae*	-	-	-	-	2.3	6.3	-	-	-	-	-
18	*Penicillium* $sp_{1.}$	-	20	9.7	-	-	8	3.7	-	15	8	15
19	*Penicillium* $sp_{2.}$	4.0	20	8.7	-	4.3	-	-	-	12	10	-
20	*Pestalotiopsis guepinii*	-	-	-	-	-	**1.0**	-	-	-	-	-
21	*Rhizopus stolonifer*	-	-	-	24	-	10.7	-	-	-	-	18
22	*Sarocladium oryzae*	-	-	-	-	2	-	-	-	-	-	-
23	*Trichoderma viride*	-	-	-	-	4.6	-	-	-	-	-	4

- represents absence of respective fungus

4.6. Per cent frequency of association of fungi with diseased grains of aus rice varieties collected from BRRI in July 2012

In the Aus season eleven species of fungi belonging to eight genera were obtained from spotted grains of seven rice varieties. Highest frequency (25.7 %) was observed in case of *Rhizopus stolonifer* isolated from BRRI 42. Lowest frequency (2.7%) was observed in both *Sarocladium oryzae* and *Curvularia lunata* from BR 26 and BRRI 48 respectively. Frequency (%) of other fungi, i.e. *Aspergillus flavus* (24.6), *A. fumigatus* (22), *Rhizopus stolonifer* (20.7), *Chaetomium globosum* (20), *Penicillium sp.* (15.7), *Fusarium moniliforme* (14), *Curvularia lunata* (10.7), *F. oxysporum* (9.7), *F. solani* (9.0), *Fusarium sp.* (9.0), *Nigrospora oryzae* (8.7), *Alternaria alternata* (8.6), *A. clavatus* (8.0), *Trichoderma viride* (6.7), *Sarocladium oryzae* (2.3) were found in different rice varieties.

After 3 months of storage at 25±2°C.Highest frequency (29.7 %) of *Rhizopus stolonifer* was observed in BRRI 42. Lowest frequency (2%) was noticed in *Sarocladium oryzae* from BR 26. After 6 months of storage at 25±2°C. Highest frequency (37.7 %) of *Rhizopus stolonifer* was recorded from BRRI 42. Lowest frequency (1%) was noticed in in *Sarocladium oryzae* from BR 14 (Table 7 - 9).

Table 7. Per cent frequency of association of fungi with diseased grains of aus rice varieties collected from BRRI in July 2012

		Aus rice varieties and per cent frequency						
Sl. No.	Name of fungi	BR 12	BR 14	BR 16	BR 26	BRRI 42	BRRI 43	BRRI 48
1.	*Alternaria alternata*	12.0	6.0	10.6	4.0	4.7	-	-
2.	*Aspergillus flavus*	18.7	10.6	10.0	12.0	-	6.3	10.7
3.	*A. fumigatus*	12.6	10	-	12.0	10.0	-	10.3
4.	*A. niger*	10.7	17.6	20.7	10.7	10.3	24.7	-
5.	*A. terreus*	-	3.3	-	-	-	-	3.7
6.	*Curvularia lunata*	6.0	3.3	7.7	-	6.6	-	**2.7**
7.	*Drechslera oryzae*	16.6	17.6	20.6	10.3	14.6	12	13.6
8.	*Fusarium moniliforme*	-	8.3	10.6	4.7	-	10.3	-
9.	*Penicillium* sp.	10	-	-	9.3	-	5.0	10.7
10.	*Rhizopus stolonifer*	12	14.3	10.7	20.7	**25.7**	-	20.3
11.	*Sarocladium oryzae*	-	7.3	-	**2.7**	-	-	-

- represents absence of respective fungus

Table 8. Per cent frequency of association of fungi with diseased grains of Aus rice varieties after 3 months of storage at 25±2 °C

		Aus rice varieties and mean per cent frequency						
Sl. No.	Name of fungi	BRRI 12	BR 14	BR 16	BR 26	BRRI 42	BRRI 43	BRRI 48
1.	*Alternaria alternata*	-	-	8.6	3.0	2.7	-	-
2.	*Aspergillus flavus*	20.7	15	12.0	22.0	-	16	18.7
3.	*A. fumigatus*	16.6	14	-	14.0	16.0	-	14.3
4.	*A. niger*	22.7	27.6	25.7	14.7	15.3	30.7	-
5.	*A. terreus*	-	12.3	10	-	8.6	-	12.7
6.	*Curvularia lunata*	-	3.0	7.0	-	5.7	-	2.0
7.	*Drechslera oryzae*	6.6	9.6	9.6	8.3	4.6	-	3.6
8.	*Fusarium moniliforme*	-	6.3	8.0	3.7	-	8.3	-
9.	*Penicillium* sp.	14.3	-	-	10.3	-	15.0	20.7
10.	*Rhizopus stolonifer*	20.7	16.3	17	22.7	**29.7**	-	25
11.	*Sarocladium oryzae*	-	2.3	-	**2.0**	-	-	-

- represents absence of respective fungus

Table 9. Per cent frequency of association of fungi with diseased grains of Aus rice varieties after 6 months of storage at 25±2 °C

		Aus rice varieties and mean per cent frequency						
Sl. No.	Name of fungi	BR 12	BR 14	BR 16	BR 26	BRRI 42	BRRI 43	BRRI 48
1.	*Alternaria alternata*	-	-		-	2.0	-	-
2.	*Aspergillus flavus*	21.7	16.6	16.0	24.0	-	16.3	20.7
3.	*A. fumigatus*	19.6	20.0	23	18.0	20.0	10	15.3
4.	*A. niger*	25.7	20.6	30.7	15.7	31.3	33.7	10
5.	*A. terreus*	3.0	12.3	-	-	-	-	2.7
6.	*Curvularia lunata*	-	3.3	3.7	-	2.6	-	3.3
7.	*Drechslera oryzae*	4.6	8.6	9.6	7.3	4.6	-	3.6
8.	*Fusarium moniliforme*	-	6.3	5.6	4.7	-	3.3	-
9.	*Penicillium* sp.	16.0	-	-	10.3	-	5.0	20.7
10.	*Rhizopus stolonifer*	25.7	19	20.7	25.7	**37.7**	-	25
11.	*Sarocladium oryzae*	-	**1.0**	-	4.7	-	-	-

- represents absence of respective fungus

4.7. Per cent frequency of association of fungi with diseased grains of Aman rice varieties collected from BRRI in July 2013

During T. Aman season the numbers of isolated fungi were 15 species belonging to 11 genera from 22 varieties of affected rice grains. Highest frequency (20.7%) was observed in case of *Aspergillus niger* from BRRI 41 and lowest frequency (2.0%) in *Pestalotiopsis guepinii* from BRRI 38. After 3 months of storage at 25±2°C, highest frequency (30.7 %) was in *Aspergillus niger* from BRRI 41 and lowest frequency (1.0%) in *Pestalotiopsis guepinii* from BRRI 38. After 6 months of storage at 25±2°C, highest frequency (40.7 %) was found in *Aspergillus niger* from BRRI 41 and lowest frequency (1.0%) in *Sarocladium oryzae* from BRRI 34 (Table10 - 12).

Table 10. Per cent frequency of association of fungi with diseased grains of Aman rice varieties collected from BRRI in July 2013

		Aman rice varieties and per cent frequency										
Sl. No.	Name of fungi	Hybrid 4	BR 11	BR 22	BR 23	BR 25	BRRI 30	BRRI 31	BRRI 32	BRRI 33	BRRI 34	BRRI 37
1	*Alternaria alternata*	-	-	-	-	7.7	-	-	-	-	-	2.2
2	*Aspergillus flavus*	10.3	2.3	10	10	18.3	10.7	15.7	-	16.3	8.7	8.7
3	*A. fumigatus*	-	8.7	-	-	22.7	15.3	2.7	-	20.7	-	-
4	*A. niger*	10.6	10.7	5.6	10	12.3	13	14	9.7	12.7	15.7	-
5	*A. terreus*	-	-	-	-	2.3	-	-	5	-	-	-
6	*Cladosporium cladosporioides*	-	-	2	5.6	7.7	-	-	4.6	-	-	9.3
7	*Colletotrichum gloeosporoides*	-	-	-	-	-	-	-	-	-	4	1.3
8	*Curvularia lunata*	-	-	4.3	3.3	-	-	-	4.0	-	8.7	4.7
9	*Drechslera oryzae*	2.3	12	8.6	-	14	12	3.3	12.3	-	7.6	14.7
10	*Fusarium moniliforme*	-	-	-	-	2.7	-	-	-	4.3	-	5.6
11	*Microdochium oryzae*	-	-	-	-	-	-	-	-	-	4.3	-
12	*Penicillium* sp.	4.7	6.7	6.7	5.7	3.3	-	4.6	4.7	-	-	3
13	*Pestalotiopsis guepinii*	-	-	-	-	2.3		-	-	-	-	-
14	*Rhizopus stolonifer*	8.3	8.3	-	6.3	-	-	-	8.3	12	10	-
15	*Sarocladium oryzae*	-	-	-	-	2.3	-	-	-	-	2	-

- represents absence of respective fungus

Table 10 contd.

Sl. No.	Name of fungi	BRRI 38	BRRI 39	BRRI 40	BRRI 41	BRRI 46	BRRI 49	BRRI 51	BRRI 52	BRRI 53	BRRI 54	BRRI 55
		Aman rice varieties and per cent frequency										
1	*Alternaria alternata*	3.7	2.7	-	-	-	-	2.3	-	-	1.3	-
2	*Aspergillus flavus*	8.7	10	4.6	-	6.7	7.3	-	4.5	8.3	-	-
3	*A. fumigatus*	3.3	7.5	10.3	10.3	-	10.7	10	15	4.7	8.7	16.7
4	*A. niger*	10.3	16	14.7	**20.7**	10.7	-	-	-	-	12.3	8.3
5	*A. terreus*	10.7	-	-	-	-	10.6	5.7	8.7	4.3	8.3	8.3
6	*Cladosporium cladosporioides*	-	3.3	-	-	-	-	4.3	-	-	8.6	6.7
7	*Colletotrichum gloeosporoides*	2	-	-	-	-	-	-	-	-	4	-
8	*Curvularia lunata*	2.3	2.3	-	-	-	-	-	2.3	-	3.7	6.7
9	*Drechslera oryzae*	14.3	12.3	-	-	-	10.7	-	13.7	-	-	13.3
10	*Fusarium moniliforme*	4	2.3	-	-	5.6	-	-	2.7	-	-	-
11	*Microdochium oryzae*	-	-	-	4.7	-	-	5.6	3	-	5.7	-
12	*Penicillium* sp.	-	-	3.3	8.7	10.7	-	5.6	-	-	-	-
13	*Pestalotiopsis guepinii*	**2.0**	-	-	-	-		-	-	-	-	-
14	*Rhizopus stolonifer*	-	-	8.7	10.7	8.7	-	-	-	10.3	8.7	-
15	*Sarocladium oryzae*	-	-	-	-	-	-	2.6	8.3	2.3	-	-

- represents absence of respective fungus

Table 11. Per cent frequency of association of fungi with diseased grains of Aman rice varieties after 3 months of storage at 25±2 °C

Sl. No.	Name of fungi	Aman rice varieties and mean per cent frequency										
		Hybrid 4	BR 11	BR 22	BR 23	BR 25	BRRI 30	BRRI 31	BRRI 32	BRRI 33	BRRI 34	BRRI 37
1	*Alternaria alternata*	-	-	-	-	2	-	-	-	-	-	-
2	*Aspergillus flavus*	12	10	13	14	28	13	15	2 4	18	12	10
3	*A. fumigatus*	10	18.7	11	-	25.7	17.3	12.7	5	24.7	-	-
4	*A. niger*	20.6	16.7	15.6	12	16.3	20	15	11.7	15.7	18.7	-
5	*A. terreus*	-	-	-	1.3	2.3	-	-	-	-	-	-
6	*Cladosporium cladosporioides*	-	-	-	-	-	-	-	-	-	-	4.3
7	*Colletotrichum gloeosporoides*	-	-	-	-	-	-	-	-	-	3.7	-
8	*Curvularia lunata*	-	-	2.3	-	-	-	-	2.6	-	8.7	2.7
9	*Drechslera oryzae*	1.3	10	6.6	-	10.7	2.0	-	12	-	-	12.7
10	*Fusarium moniliforme*	-	-	-	-	2	-	-	-	-	-	2.6
11	*Microdochium oryzae*	-	-	-	-	-	-	-	-	-	2.3	-
12	*Penicillium* sp.	6.7	7.7	11.7	8.7	5.3	-	-	10.7	-	-	10
13	*Pestalotiopsis guepinii*	-	-	-	-	10	-		-	-	-	-
14	*Rhizopus stolonifer*	12.3	13.3	-	16.6	-	-	-	18.3	14	16	-
15	*Sarocladium oryzae*	-	-	-	-	1.3	-	-	-	-	1.5	-

- represents absence of respective fungus

Table 11 contd.

		Aman rice varieties and mean per cent frequency										
Sl. No.	Name of fungi	BRRI 38	BRRI 39	BRRI 40	BRRI 41	BRRI 46	BRRI 49	BRRI 51	BRRI 52	BRRI 53	BRRI 54	BRRI 55
1	*Alternaria alternata*	2	2	-	-	-	-	1.3	-	-	-	-
2	*Aspergillus flavus*	10	12	14.6	16	-	10	11	-	10.3	10	15
3	*A. fumigatus*	6	8	20.3	18.3	10	20.7	14	20	14.7	9.7	20.7
4	*A. niger*	16.3	25	20.7	**30.7**	13.7	10	8	11	-	20.3	10 .3
5	*A. terreus*	18.7	-	-	-	-	12.6	6.7	12.7	5.3	10.3	10.3
6	*Cladosporium cladosporioides*	-	-	-	-	-	-	3.3	-	-	7.6	5.7
7	*Colletotrichum gloeosporoides*	-	-	-	-	-	-	3.7	-	-	3	-
8	*Curvularia lunata*	1.3	2.0	-	-	-	-	-	2.0	-	2.7	4.7
9	*Drechslera oryzae*	12.3	12	-	-	-	8.7	-	10.7	-	-	3
10	*Fusarium moniliforme*	2	2	-	-	4.6	-	-	2.0	-	-	-
11	*Microdochium oryzae*	-	-	-	2.7	-	-	4.6	2	-	4.7	-
12	*Penicillium* sp.	6	8	10	8.7	20.7	-	12.6	4	-	-	-
13	*Pestalotiopsis guepinii*	**1.0**	-	-	-	-		-	-	-	-	-
14	*Rhizopus stolonifer*	10	12	18.7	12.7	15	-	-	8	12.3	18.7	-
15	*Sarocladium oryzae*	-	-	-	-	-	-	1.6	4.3	2	-	-

- represents absence of respective fungus

Table 12. Per cent frequency of association of fungi with diseased grains of Aman rice varieties after 6 months of storage at 25±2 °C

		Aman rice varieties and mean per cent frequency										
Sl. No.	Name of fungi	Hybrid 4	BR 11	BR 22	BR 23	BR 25	BRRI 30	BRRI 31	BRRI 32	BRRI 33	BRRI 34	BRRI 37
1	*Alternaria alternata*	-	-	-		1.6	-	-	-	-	1	-
2	*Aspergillus flavus*	16.3	12.3	16	15	20.3	14.7	18.7	12	18.3	18.7	15
3	*A. fumigatus*	-	8.7	8.6	11.6	22.7	15.3	12.7	-	20.7	12	-
4	*A. niger*	20.6	10.7	15.6	10	20.3	20	13	13.7	12.7	25.7	12
5	*A. terreus*	-		-	-	4.3	-	-	-	-	-	2.0
6	*Cladosporium cladosporioides*	-	-	2	4	-	-	-	-	-	-	6
7	*Colletotrichum gloeosporoides*	-	-	-	-	-	-	-	-	-	2.7	1.3
8	*Curvularia lunata*	-	-	2.3	-	-	-	-	2.0	-	3	2.7
9	*Drechslera oryzae*	4	2	4.6	-	4.7	2.0	-	2.3	-	-	6.7
10	*Fusarium moniliforme*	-	-	-	-	2	-	-	-	-	-	2.6
11	*Microdochium oryzae*	-	-	-	-	-	-	14	-	-	4	-
12	*Penicillium* sp.	18.7	12	14	10	13.3	-	-	14.7	-	-	12
13	*Pestalotiopsis guepinii*	-	-	-	-	1.3	-			-	-	-
14	*Rhizopus stolonifer*	18.3	14.3	-	16.3	-	-	-	18.3	16	16	-
15	*Sarocladium oryzae*	-	-	-	-	-	-	-	-	-	**1.0**	-

- represents absence of respective fungus

Table 12 contd.

		Aman rice varieties and mean per cent frequency										
Sl. No.	Name of fungi	BRRI-38	BRRI 39	BRRI 40	BRRI 41	BRRI 46	BRRI 49	BRRI 51	BRRI 52	BRRI 53	BRRI 54	BRRI 55
1	*Alternaria alternata*	-	2	-	-	-	-	1.3	-	-	-	-
2	*Aspergillus flavus*	13	15	14	12.5	10	7.3	-	11	8.3	-	8.0
3	*A. fumigatus*	-	-	20.3	10.3	-	20.7	10	20	4.7	8.7	20.7
4	*A. niger*	10.3	25	30.7	**40.7**	10.7	-	-	-	-	20.3	8.3
5	*A. terreus*	16.7	-	-	-	-	12.6	9.7	10.7	11.3	8.3	28.3
6	*Cladosporium cladosporioides*	-	-	-	-	-	-	10.3	-	-	9.6	8.7
7	*Colletotrichum gloeosporoides*	-	-	-	-	-	-	1.7	-	-	2	-
8	*Curvularia lunata*	1.3	2.0	-	-	-	-	-	2.3	-	3.7	6.7
9	*Drechslera oryzae*	14.3	14.3	-	-	-	10.7	-	13.7	-	-	3
10	*Fusarium moniliforme*	2	2.3	-	-	5.6	-	-	2.7	-	-	-
11	*Microdochium oryzae*	-	-	-	2	-	-	2.6	2	-	2.7	-
12	*Penicillium* sp.	16	-	13.3	10.7	20.7	-	15.6	-	-	-	-
13	*Pestalotiopsis guepinii*	-	-		-	-		-	-		-	
14	*Rhizopus stolonifer*	15	14	18.7	10.7	18.7	-	13	12	14.3	20.7	-
15	*Sarocladium oryzae*	-	-	-	-	-	-	2	2.3	2	-	-

- represents absence of respective fungus

4.8. Per cent frequency of association of different fungi with Boro, Aus and T. Aman rice varieties at different divisions of Bangladesh

Fifty six (56) rice seed samples representing over four rice varieties collected from 14 different districts of Bangladesh under Barisal, Chittagong, Dhaka, Khulna, Rajshahi, Rangpur and Sylhet divisions.

A total of 23 species of fungi belonging to 15 genera were isolated from these rice samples. The most predominating fungus detected from the grains was *Drechslera oryzae.* Highest frequency (50.3%) of *Drechslera oryzae* was observed in Rajshahi division. Lowest frequency (2.0%) was noticed in *Sarocladium oryzae* from Barisal division (Table 13). After 6 months of storage at 25±2°C, highest frequency (40.0%) was found in case of *Aspergillus niger* from Dhaka division and lowest frequency (1.0%) for *Sarocladium oryzae* from Barisal division. From the results, it appears that predominating fungi associated with grains were *D. oryzae*, *A. niger, A. flavus, Rhizopus stolonifer* and *Penicilium* sp.. Among three seasons frequency of association of fungi was slightly higher in Boro season compared to Aus and T. Aman season. In T. Aman the most frequently occurring fungi were *A. niger, A. flavus, C. lunata, and D. oryzae.*

Table 13. Mean per cent frequency of fungi with Boro, Aus and T. Aman rice varieties at different divisions of Bangladesh

Name of the fungi	Mean per cent frequency of fungi at different divisions						
	Barisal	**Chittagong**	**Dhaka**	**Khulna**	**Rangpur**	**Rajshahi**	**Sylhet**
Alternaria alternata	12.0	6.0	10.7	11.5	10.5	11.0	-
Aspergillus flavus	10.0	10.5	14.0	15.0	10.0	20.3	10.0
A. fumigatus	12.0	16.5	12.0	6.7	8.0	20.6	10.0
A. niger	14.0	20.5	14.0	10.5	10.0	8.0	15.0
A. oryzae	-	-	5.6	-	-	9.0	-
A. terreus	8.0	6.0	8.0	4.0	3.0	10.0	6.6
A. ochraceus	-	-	4.6	4.0	-	5.5	-
Chaetomium globosum	12.4	15.6	20.0	-	8.0	16.6	9.0
Cladosporium cladosporioides	-	6.4	15.0	6.6	4.4	14.3	4.3
Colletotrichum gloeosporioides	-	-	3.0	-	-	2.3	-
Curvularia lunata	-	6.0	9.6	10.3	9.2	11.3	3.0
Drechslera oryzae	30.0	34.0	40.0	35.0	7.0	**50.3**	8.0
Fusarium moniliforme	-	11.0	16.5	12.7	8.5	8.4	6.6
F.oxysporum	-	-	9.6	-	-	10	-
F.solani	4.3	-	7.7	-	2.2	8.7	-
Microdochium oryzae	-	-	13.7	-	7.6	9.7	-
Nigrospora oryzae	2.5	8.7	12.7	4.5	2.6	3.7	3.3
Penicillium sp.1	3.5	10.0	12.6	6.6	6.0	9.6	5.5
Penicillium sp.2	-	-	10.6	-	4.7	8.6	5.6
Pestalotiopsis guepinii	-	-	2.6	-	-	2.5	-
Rhizopus stolonifer	10.5	12.5	20.6	12.3	12.0	15.5	10
Sarocladium oryzae	**2.0**	7.0	4.6	6.6	10.5	18.6	-
Trichoderma viride	-	-	6.3	-	-	8.3	4.4

- represents absence of respective fungus

Table 14. Mean per cent frequency of fungi with Boro, Aus and T. Aman rice varieties at different divisions of Bangladesh after 6 months of storage

Name of the fungi	**Mean per cent frequency of fungi at different divisions**						
	Barisal	**Chittagong**	**Dhaka**	**Khulna**	**Rangpur**	**Rajshahi**	**Sylhet**
Alternaria alternata	2.0	4.3	6.7	1.5	1.5	1.0	-
Aspergillus flavus	15.0	12.5	34.0	10.0	10.0	20.3	18.0
A. fumigatus	2.0	12.5	10.0	8.7	8.0	20.6	16.0
A. niger	12.0	22.5	**40.0**	11.5	10.0	8.0	25.0
A. oryzae	3.3	5.5	10	-	-	12	-
A. terreus	2.0	2.0	8.0	-	-	10.0	-
A. ochraceus	-	-	2.6	-	-	1.5	-
Chaetomium globosum	2.4	5.6	10.0	-	-	16.6	-
Cladosporium cladosporioides	-	2.4	12.0	-	-	14.3	-
Colletotrichum gloeosporioides	-	-	2.0	-	-	1.3	-
Curvularia lunata	-	2.5	5.6	8.3	9.2	11.3	3.0
Drechslera oryzae	30.0	34.0	32.3	25.0	7.0	30.3	6.0
Fusarium moniliforme	-	11.0	12.5	2.7	8.5	8.4	6.6
F. oxysporum	-	-	12	-	-	10	-
F. solani	-	-	9.6	-	-	-	-
Michrodochium oryzae	-	-	13.7	-	7.6	9.7	-
Nigrospora oryzae	1.5	4.7	9.7	2.5	2.6	3.7	-
Penicillium sp.1	8.5	9.5	16.6	16.6	8.0	9.6	16
Penicillium sp.*2*	-	-	7.6	-	4.7	8.6	-
Pestalotiopsis guepinii	-	-	1.6	-	-	10.5	-
Rhizopus stolonifer	20.5	14.5	25.6	16.3	23.0	15.5	20
Sarocladium oryzae	**1.0**	4.0	2.6	6.6	10.5	18.6	-
Trichoderma viride	-	-	10.3	-	-	8.3	-

- represents absence of respective fungus

4.9. Occurrence of seed borne fungi in rice seed samples (96) obtained from different districts of different divisions

A total of ninety six seed samples consisting each of different varieties were obtained from different districts of Bangladesh. Seed samples were used for testing their health status and the results are furnished in Table 15. In total, 15 genera of fungi comprising 25 species were found to be associated with the seed samples. Among them the most predominant was *Drechslera oryzae*, which was associated with 62.5% seed samples followed by *Aspergillus flavus* (45.83%), *A. niger* (39.58%), *A. fumigatus* (34.38%), *Penicillium* sp_1 (25%), *Rhizopus stolonifer* (21.88%), *Alternaria alternata* (18.75%), *Curvularia lunata* (17.71%), *Fusarium moniliforme* (15.63%), *A. terreus* (13.54%), *Cladosporium cladosporoides* (11.46%) *Sarocladium oryzae*, (8.33%), *Microdochium orzae* (5.20%).

The following fungi *viz.*, *Aspergillus clavatus, Colletotrichum gloeosporoides, Penicillium* sp_2 and *T viride* showed a common per cent incidence value of 4.16%. Five of other fungi namely *A. oryzae, Chaetomium globossum, C. lunata* var. *aeria, F. oxysporum, Nigrospora oryzae* showed an incidence of 3.13%. Least incidence (2.08) was observed in both *F. solani and P. guepinii.*

Many workers have reported *Alternaria padwickii, A. longissima, Aspergillus niger, Nigrospora oryzae, Curvularia oryzae, C. lunata, Bipolaris oryzae, Fusarium moniliforme, F. semitectum, F. solani* and species *of Phoma, Cercospora, Chaetomium, Sclerotium, Penicillium, Myrthecium* and *Colletotrichum* from seeds of different varieties of rice (Haque *et al.* 2000, Mew and Gonzales 2002, Ora *et al.* 2011, Shamsi *et al.* 2010).

In the present research, the frequency of *D. oryzae* was 62.5%. Out of 96 seed samples tested, 61 samples carried *D. oryzae*, among them 26 carried 1-10 per cent, 17 carried 11-20 per cent, 7 carried 21-30 per cent, 4 carried 31-40 per cent, 2 carried 51-60 per cent, 1 carried 61-70 per cent, 2 seed samples showed 71-80 per cent and 1 sample showed 91-100 per cent seed infection (Table 15).

Rice seeds were reported to have been associated with 32 genera and 48 species of fungi (Richardson 1972). Twenty different species of fungi were identified on the rice seeds which include 10 genera (Mishra and Dharam 1992). Mew and Gonzales (2002) reported 104 species of fungi which include 39 genera. Shamsi *et al.* (2003) reported association of 15 species of fungi with sheaths and grains of sheath rot affected rice varieties from Bangladesh.

Ora *et al*. (2011) detected and identified 12 seed borne pathogens from nine cultivated hybrid rice varieties in Bangladesh. Habib *et al.* (2012) detected 10 seed borne fungi from 15 varieties of rice (8 coarse and 7 fine) from Rice Research Institute, Kala Shah Kaku, Paksitan. Archana and Prakash (2013) isolated 16 genera of fungi comprising 27 species were found to be associated with 69 rice seed samples obtained from different states of India. Among these fungi the most predominant was *Bipolaris oryzae.* The results of my investigation showed similarity with the findings of above mentioned workers.

Table 15. Occurrence of seed borne fungi in rice seed samples (96) obtained from different districts of different divisions

Sl. No.	Name of fungi	Seed lot infected	Range of infection percentage of seed sample									
		(%)	01-10	11-20	21-30	31-40	41-50	51-60	61-70	71-80	81-90	91-100
1	*Alternaria alternata*	18.75	17	1	-	-	-	-	-	-	-	-
2	*Aspergillus clavatus*	4.16	4	-	-	-	-	-	-	-	-	-
3	*A.flavus*	45.83	19	22	3	-	-	-	-	-	-	-
4	*A.fumigatus*	34.38	12	14	7	-	-	-	-	-	-	-
5	*A.niger*	39.58	5	20	11	1	-	-	-	-	-	1
6	*A. ochraceus*											
7	*A.oryzae*	3.13	3	-	-	-	-	-	-	-	-	-
8	*A.terreus*	13.54	9	4	-	-	-	-	-	-	-	-
9	*Chaetomium globossum*	3.13	2	1	-	-	-	-	-	-	-	-
10	*Cladosporium cladosporoides*	11.46	11	-	-	-	-	-	-	-	-	-
11	*Colletotrichum gloeosporoides*	5.2	5	-	-	-	-	-	-	-	-	-
12	*Curvularia lunata*	17.71	17	-	-	-	-	-	-	-	-	-
13	*C. lunata* var. *aeria*	3.13	3	-	-	-	-	-	-	-	-	-
14	*Drechslera oryzae*	**62.5**	26	17	7	4	-	2	1	2	-	1
15	*Fusarium moniliforme*	15.63	13	2	-	-	-	-	-	-	-	-
16	*F. oxysporum*	3.13	2	1	-	-	-	-	-	-	-	-
17	*F. solani*	**2.08**	2	-	-	-	-	-	-	-	-	-
18	*Microdochium oryzae*	4.16	2	2	-	-	-	-	-	-	-	-
19	*Nigrospora oryzae*	3.13	2	1	-	-	-	-	-	-	-	-
20	*Penicillium* sp.1	25	18	6	-	-	-	-	-	-	-	-
21	*Penicillium* sp.2	4.16	2	2	-	-	-	-	-	-	-	-
22	*Pestalotiopsis guepinii*	**2.08**	2	-	-	-	-	-	-	-	-	-
23	*Rhizopus stolonifer*	21.88	9	8	4	-	-	-	-	-	-	-
24	*Sarocladium oryzae*	8.33	8	-	-	-	-	-	-	-	-	-
25	*Trichoderma viride*	4.16	3	1	-	-	-	-	-	-	-	-

- represents absence of respective fungus

4.10. Pathogenecity test of isolated fungi of different rice varieties

After 10 days of inoculation, among 25 species of isolated fungi, pathogenic fungl were re-isolated from diseased and inoculated seeds and the seedlings from those healthy seeds remained fresh. The isolated pathogenic fungi were *Alternaria alternata* (Fr.) Keissler, *Aspergillus flavus* Link *Curvularia lunata* (Wakker) Boedijn, *Drechslera oryzae* Breda de Haan (Subramanian and Jain), *Fusarium moniliforme* Sheldon, *Fusarium solani* (Mart.) Sacc. *Microdochium oryzae* (Hashloka and Yokogi) Sam. and Hal., *Pestalotiopsis guepinii* (Desm.) Stay. and *Sarocladium oryzae* (Sawada) W. Gams and D. Hawks.

4.11. Effect of Pathogenic fungi on seed germination, seedling mortality and shoot-root length

Different varieties of rice seeds were inoculated with nine pathogenic fungi *viz. Alternaria alternata* (Fr.) Keissler, *Aspergillus flavus* Link *Curvularia lunata* (Wakker) Boedijn, *Drechslera oryzae* Breda de Haan (Subramanian and Jain), *Fusarium moniliforme* Sheldon, *Fusarium solani* (Mart.) Sacc. *Microdochium oryzae* (Hashloka and Yokogi) Sam. and Hal., *Pestalotiopsis guepinii* (Desm.) Stay. and *Sarocladium oryzae* (Sawada) W. Gams and D. Hawks.which showed germination 70, 75, 60, 80, 60, 50, 40, 70 and 60% respectively. But in control set, 100% seeds were germinated. The mortality percentages were 40, 35, 30, 30, 40, 16, 30, 50 and 40% respectively. The highest and the lowest shoot-root length range was 2.0 cm-1.2 cm and 0.3 cm-0.1 cm among all test pathogens. But in control set the highest and lowest shoot-root length was 9.5 cm - 8.5 cm to 8.0 cm - 7.7 cm (Table 16).

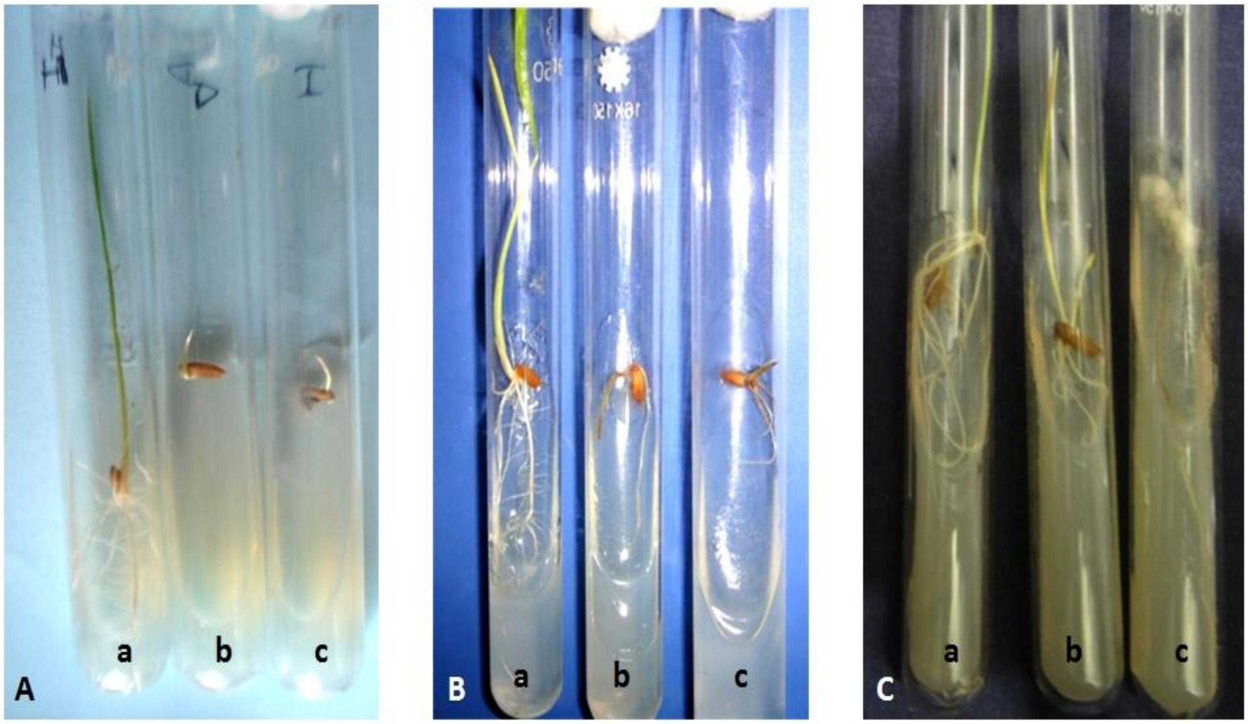

Fig. 1. Pathogenecity test of selected fungi. a, b & c represent controlled, diseased and incoluated

A. Rice variety BRRI 29 inoculated with *Alternaria alternata*

B. Rice variety BRRI 28 inoculated with *Curvularia lunata*

C. Rice variety BRRI 29 inoculated with *Drechslera oryzae*

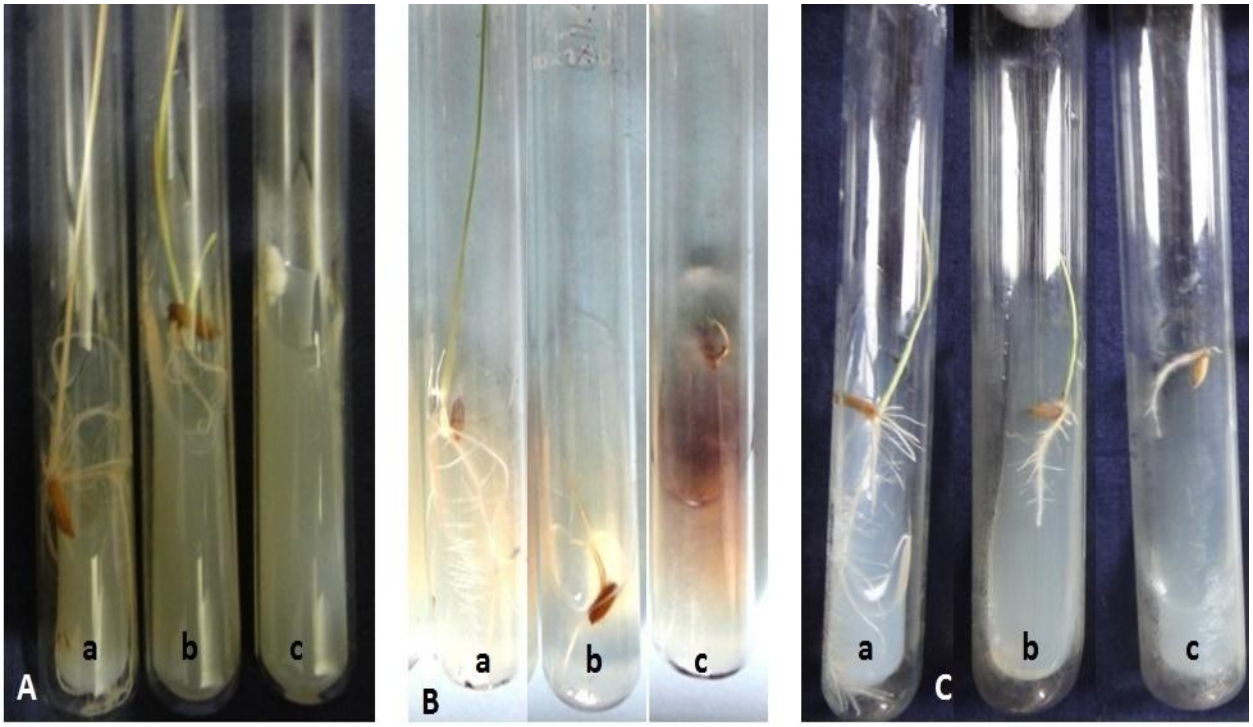

Fig. 2. Pathogenecity test of selected fungi. a, b & c represent controlled, diseased and incoluated

A. Rice variety BRRI 29 inoculated with *Fusarium moniliforme*

B. Rice variety BRRI 28 inoculated with *Fusarium solani*

C. Rice variety BRRI 41 inoculated with *Microdochium oryzae*

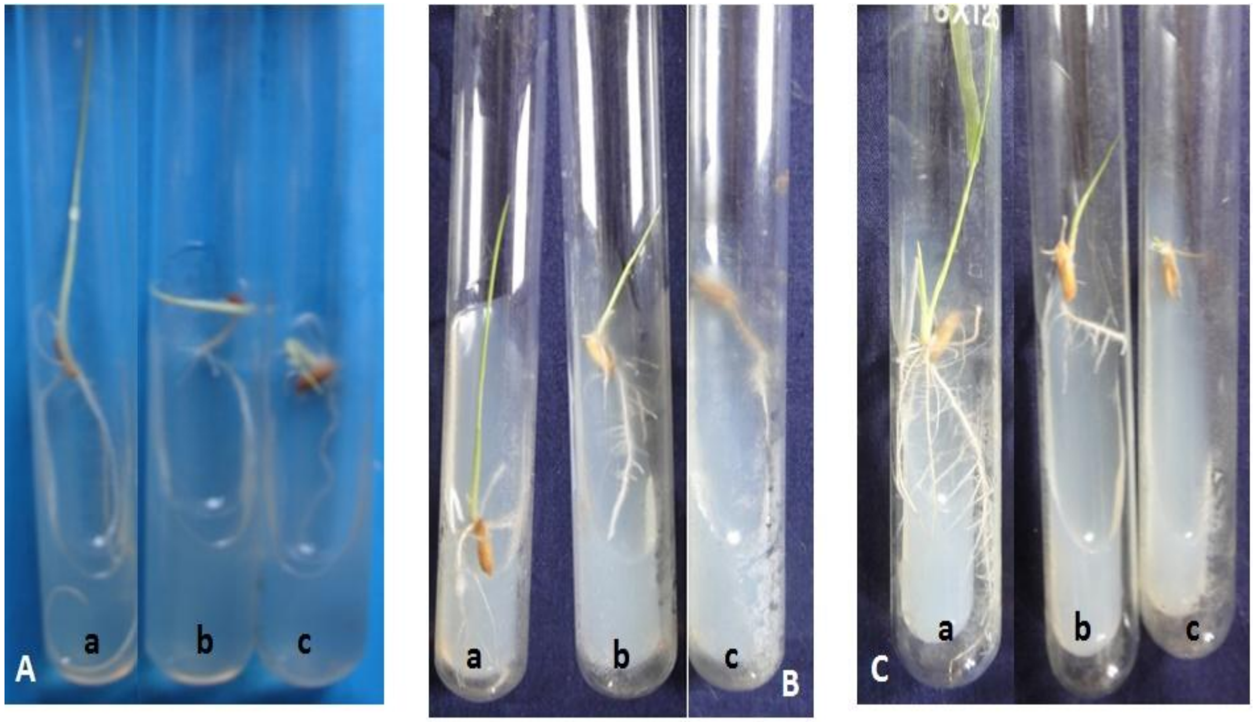

Fig. 3. Pathogenecity test of selected fungi. a, b & c represent controlled, diseased and incoluated

A. Rice variety BRRI 29 inoculated with *Aspergillus flavus*

B. Rice variety BR 25 inoculated with *Pestalotiopsis guepinii*

C. Rice variety BRRI 28 inoculated with *Sarocladium oryzae*

Table 16. Effects of pathogenic fungi on germination, seedling mortality and height of different rice varieties in test tubes

Sl. No.	Name of Fungi	Name of variety	Germination percentage			Mortality percentage			Shoot-Root length (cm)		
			Control	Diseased	Inoculated	Control	Diseased	Inoculated	Control	Diseased	Inoculated
01.	*Alternaria alternata*	BRRI 29	100	80	70	12	40	70	9.0-8.1	2.0-1.4	1.6-1.1
02	*Aspergillus flavus*	BRRI 29	100	80	75	14	35	**72**	8.0-7.5	2.5-1.4	**2.0-1.2**
03.	*Curvularia lunata*	BRRI 28	100	70	60	16	30	50	**8.0-7.7**	4.0-2.5	1-5-2.5
04.	*Drechslera oryzae*	BRRI 29	100	**90**	**80**	14	**50**	63	9.4-8.3	2.3-1.8	1.5-1.0
05.	*Fusarium moniliforme*	BRRI 29	100	70	60	13	40	67	9.1-8.1	2.0-1.6	1.4-1.1
06	*F. solani*	BRRI 29	100	60	50	15	**16**	**20**	9.2-8.1	2.0-1.5	**0.3-0.1**
07.	*Microdochium oryzae*	BRRI 41	100	**50**	**40**	**20**	30	60	9.0-8.0	2.2-1.7	1.3-1.1
08.	*Pestalotiopsis guepinii*	BR 25	100	60	70	**10**	**50**	40	**9.5-8.5**	2.0-1.5	1.5-1.0
09.	*Sarocladium oryzae*	BRRI 28	100	80	60	17	40	67	9.2-8.2	2.1-1.3	1.1-0.9

Table 17. Particulars of fungicides used in the present study

Sl. No.	Trade name	Active ingredient (s)	Recommended dose (ppm)	Ten times less than recommended dose (ppm)	Manufacturer
1	Bavistin 50 WP	50% Carbendazim (methyl Benimidazol-2-ylcarbamate)	1000	100	BASF, Germany
2	Capvit 50 WP	Copper oxychloride	2000	200	Jiangsu HongZe Chemical and Industry Company Limited, China
3	Dithane M-45	80% Mancozeb	2000	200	Dow Agro Science, Brazil
4	Greengel 72 WP	64% Mancozeb + 8% Metalaxyl	2000	200	Green Bangla Agrovet Ltd.
5	Hayvit	Abamactin	2000	200	Synzenta (BD) LTD
6	Indofil	45% Mancozeb	2000	200	Dow Agro Science, Brazil
7	Ridomil MZ Gold	68% Mancozeb + 4% Metalaxyl	2000	200	Synzenta Production, France
8	Salcox 50 WP	Copper oxychloride	2000	200	Chemiski Production
9	MC Sulphur 80 WP	Sulphur 50%	2000	200	Aco B.V., Netherland, France
10	Tall 25 EC	Propiconazole	500	50	Synzenta Crop Production ag, Switzerland

Table 18. Particulars of the plants used in the present study

Sl. No.	**Name of Plants**	**Family**	**Parts Used**
1	*Allium sativum* L.	Amaryllidaceae	Bulb
2	*Artocarpus heterophyllus* Lamk	Moraceae	Leaf
3	*Asparagus racemosus* Willd	Asparagaceae	Leaf
4	*Azadirachta indica* A. Juss.	Meliaceae	Leaf
5	*Citrus medica* L.	Rutaceae	Leaf
6	*Datura metel* L.	Solanaceae	Leaf
7	*Mangifera indica* L.	Anacardiaceae	Leaf
8	*Nerium indicum* Mill.	Apocynaceae	Leaf
9	*Senna alata* (L.) Roxb.	Caesalpinioideae	Leaf
10	*Tagetes erecta* L.	Asteraceae	Leaf

4.12. Transmission of pathogenic fungi from seed to seedlings in pot experiments

Test pathogens exhibited the symptoms were noticed in the form of seed rot after 5 days and seedling blight after 15 days of sowing. More than 20% seedling mortality was observed. Amongst nine pathogenic fungi, highest per cent (70%) of seed germination was observed in seeds inoculated with *A. alternata* and *A. flavus* and the germination was lowest in seeds inoculated with *P. guepinii*. Highest percentage of mortality (18.38%) and seed to seedling transmlssion of pathogen (23.08%) were observed in *D. oryzae.* Symptoms of seedlings were yellowing of leaves and blight. The following percentages of germination were 65, 62, 58, 56, 55 and 52% found in *D. oryzae, F. moniliforme, M. oryzae, F. solani, C. lunata* and *S. oryzae* respectively (Table 19, Plate 1).

Table 19.Transmission of pathogenic fungi from seed to seedlings in pot experiments

Sl. No.	Test pathogens	Germination of inoculated seeds (%)	Mortality (%)	Seed to seedling transmission of pathogen (%)	Symptoms on seedlings
1	*Alternaria alternata*	**70 b**	14.28 bc	16.66 c	Rots and blight
2	*Aspergillus flavus*	**70 b**	7.14 d	15.38 cd	Rots
3	*Curvularia lunata*	55 f	18.18 a	22.22 ab	Seedling rot
4	*Drechslera oryzae*	65 c	**18.38 b**	**23.07 a**	Yellowing of leaves, blight
5	*Fusarium moniliforme*	62 d	12.9 c	14.80 d	Kernel rot
6	*Fusarium solani*	56 ef	14.28 bc	12.75 e	Stunting, wilting
7	*Microdochium oryzae*	58 e	13.79 bc	**10.00 f**	Seedling blight
8	*Pestalotiopsis guepinii*	**52 g**	9.09 d	20.20 b	Seedling blight
9	*Sarocladium oryzae*	55 cd	18.18 b	22.22 ab	Sheath rot
10	Control	75 a	13.33 bc	-	Healthy
	CV%	2.37	9.21	5.7	

Values within the same column with a common letter (s) do not differ significantly at 5% level by DMRT.

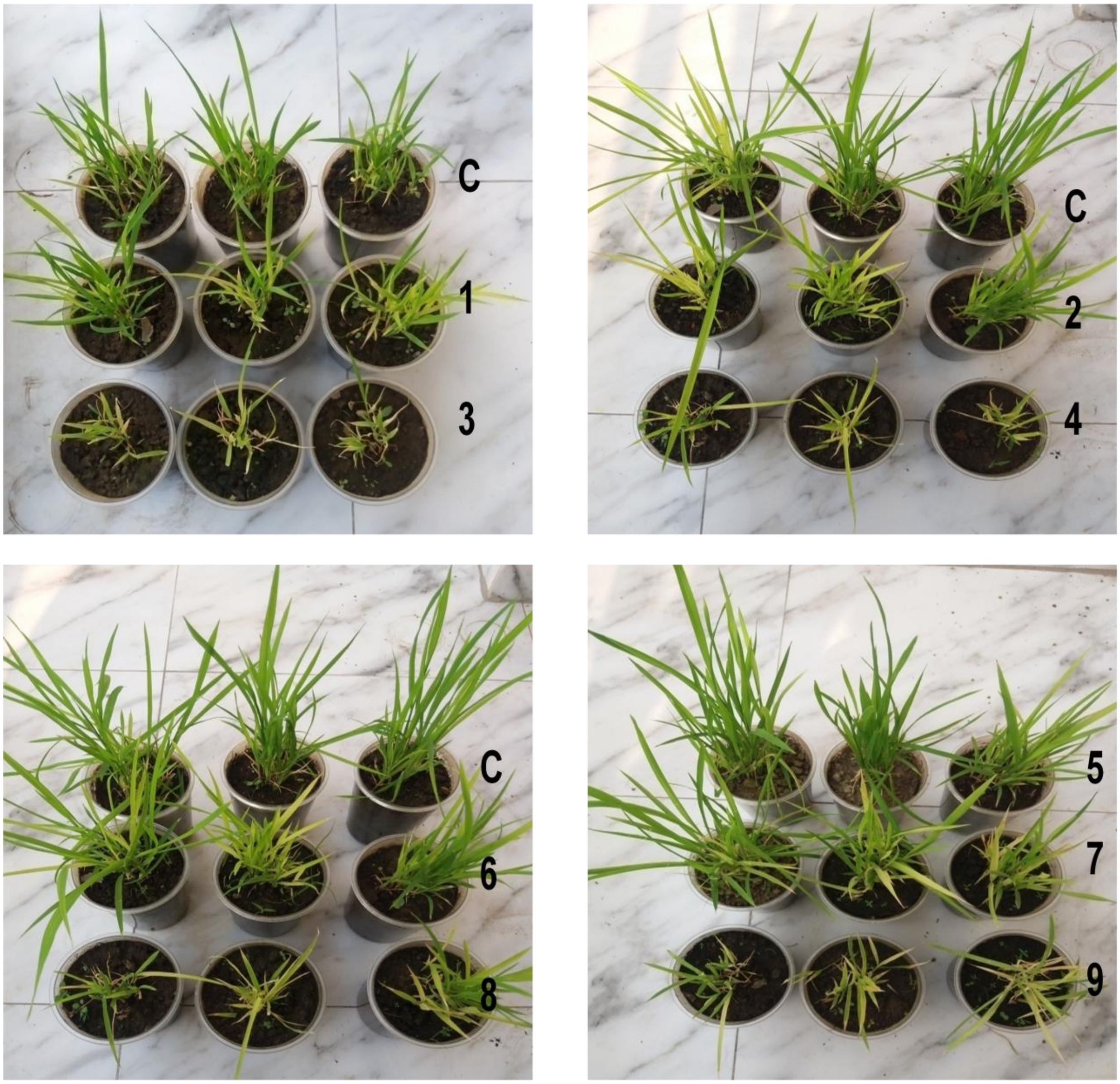

Plate 1. Transmission of pathogenic fungi from seed to seedling 1. *Alternaria alternata,* 2. *Aspergillus fiavus*, 3. *Curvularia lunata*, 4. *Drechslera oryzae,* 5. *Fusarium moniliforme,* 6. *Fusarium solani*, 7. *Microdochium oryzae,* 8. *Pestalotiopsis guepinii,* 9. *Sarocladium oryzae* and C. Control (Healthy seeds without inocula).

In control set percentage of germination and mortality was 75 and 13.33% respectively. No diseased symptoms were found at 21 days old seedlings. Plants remained fresh and healthy.

4.13. Fungitoxicity of fungicides against test pathogens of rice

It was revealed that nine pathogenic fungi were found to be associated with different rice varieties. Isolated fungi were *Alternaria alternata* (Fr.) Keissler, *Aspergillus flavus* Link *Curvularia lunata* (Wakker) Boedijn, *Drechslera oryzae* Breda de Haan (Subramanian and Jain), *Fusarium moniliforme* Sheldon, *Fusarium solani* (Mart.) Sacc. *Microdochium oryzae* (Hashloka and Yokogi) Sam. and Hal., *Pestalotiopsis guepinii* (Desm.) Stay. and *Sarocladium oryzae* (Sawada) W. Gams and D. Hawks.

Amongst the ten fungicides used in the present investigation, Bavistin, Dithane M-45 Greengel, Indofil and Ridomil were systemic while Capvit, Hayvit, Salcox, Sulphur and Tall were protective fungicides. All the fungicides inhibited the radial growth of the pathogens but complete inhibition of the test pathogens were observed with Tall at all the concentrations used except *F. moniliforme* and *M. oryzae* (Plate 3). Bavistin completely inhibited the radial growth of all test pathogens at 500 ppm (Plate 2).

4.13.1. Fungitoxicity of fungicides against *Alternaria alternata* at different concentrations

On the radial growth of *A. alternata*, Bavistin, DithaneM-45, Indofil, Sulphur and Greengel were responsible for complete inhibition at 400 and 500 ppm concentratios. Dithane M-45 also inhibited the radial growth completely at 300 ppm whereas Greengel showed 83.33% and Bavistin showed 71.42% inhibition of growth. Salcox, Sulphur and Capvit showed 66.66% inhibition at 300 ppm, respectively. Bavistin, Capvit, Dithane, Greengel, Indofil and Salcox showed the inhibition of radial growth by 71.42, 63.33, 66.66, 76.66, 64 and 56.66%, respectively, at 200 ppm concentration (Table 20).

Table 20. Fungitoxicity of fungicides against *Alternaria alternata* at different concentrations

Name of fungicides	Per cent inhibition of radial growth at different concentrations (ppm)				
	100	200	300	400	500
Bavistin	65.7 b	71.42 c	71.42 c	**100 a**	**100 a**
Capvit	50 d	63.33 d	66.66 d	70 c	80 b
Dithane	60 c	66.66 d	**100 a**	**100 a**	**100 a**
Greengel	66.66 b	76.66 b	83.33 b	**100 a**	**100 a**
Hayvit	28 h	36 g	52 c	56 e	60 d
Indofil	60 c	64 d	68 cd	**100 a**	**100 a**
Ridomil	41.66 e	50 f	53.33 e	60 d	66.66 c
Salcox	36.66 f	56.66 e	66.66 d	70 c	80 b
Sulphur	33.33 g	50 f	66.66 d	**100 a**	**100 a**
Tall	**100 a**	**100 a**	**100 a**	**100 a**	**100 a**
CV%	2.45	4.23	2.79	2.77	1.43

Table 21. Fungitoxicity of fungicides against *Aspergillus flavus* at different concentrations

Name of fungicides	Per cent inhibition of radial growth at different concentrations (ppm)				
	100	**200**	**300**	**400**	**500**
Bavistin	**100 a**	**100 a**	**100 a**	**100 a**	**100 a**
Capvit	20 f	30 f	50 c	**100 a**	**100 a**
Dithane	50 c	56.66 c	**100 a**	**100 a**	**100 a**
Greengel	56.66 b	66.66 b	**100 a**	**100 a**	**100 a**
Hayvit	5.00 h	10 h	12 f	16 e	20 c
Indofil	5.00 h	25 g	45 d	47.5 c	50 b
Ridomil	28.56 e	35.71 e	38.56 e	42.86 d	50 b
Salcox	14.28 g	28.57 f	42.86 d	**100 a**	**100 a**
Sulphur	33.33 d	50 d	66.66 b	**100 a**	**100 a**
Tall	**100 a**	**100 a**	**100 a**	**100 a**	**100 a**
CV%	3.43	4.13	2.16	2.16	1.34

Values within the same column with a common letter (s) do not differ significantly at 5% level by LSD.

4.13.2. Fungitoxicity of fungicides against *Aspergillus flavus* at different concentrations

The radial growth of *A. flavus* was completely inhibited with Bavistin and Tall at all concentrations used. Capvit, Salcox and Sulphur showed complete inhibition at 200, 300 and 400 ppm. Dithane and Greengel were also responsible for complete inhibition of radial growth at 300 ppm. Capvit Salcox and Sulphur showed 100% inhibition of radial growth at 400 ppm. All fungicides showed complete inhibition at 500 ppm except Hayvit, Indofil and Sulphur (Table 21)**.**

4.13.3. Fungitoxicity of fungicides against *Curvularia lunata* at different concentrations

The complete inhibition of radial growth of *C. lunata* was observed with Dithane and Ridomil at 500 ppm. Bavistin, Greengel, Indofil and Salcox showed 80% inhibition at 500 ppm. Sulphur, Hayvit and Capvit showed 73.33, 82.85 and 73.33% inhibition of growth at 500 ppm, respectively. The growth of *D. oryzae* was completely inhibited with Salcox, Indofil, Ridomil and Sulphur at 400 and 500 ppm. Bavistin, Dithane, Hayvit, Greengel and Capvit were also responsible for complete inhibition of radial growth at 500 ppm. They also showed 56, 80, 80, 84.62 and 66.66% inhibition of growth at 400 ppm, respectively (Table 22).

4.13.4. Fungitoxicity of fungicides against *Drechslera oryzae* at different concentrations

The growth of *D. oryzae* was completely inhibited with Salcox, Indofil, Ridomil and Sulphur at 400 and 500 ppm. Bavistin, Dithane, Hayvit, Greengel and Capvit were also responsible for complete inhibition of radial growth at 500 ppm. They also showed 56, 80, 80, 84.62 and 66.66% inhibition of growth at 400 ppm, respectively (Table 23).

Table 22. Fungitoxicity of fungicides against *Curvularia lunata* at different concentrations

Name of fungicides	Per cent inhibition of radial growth at different concentrations (ppm)				
	100	**200**	**300**	**400**	**500**
Bavistin	33.33 f	50 f	66.66 d	73.33 d	80 b
Capvit	50 d	60 e	63.33 e	66.66 e	73.33 c
Dithane	50 d	66.66 c	73.33 bc	80 c	**100 a**
Greengel	50 d	63.33 d	66.66 d	70 de	80 b
Hayvit	57.14 c	65.11 cd	71.42 c	77.14 c	82.85 b
Indofil	40 e	50 f	56.66 f	60 f	80 b
Ridomil	62.85 b	71.42 b	74.28 b	83.76 b	**100 a**
Salcox	33.33 f	43.33 g	46.66 g	66.66 e	80 b
Sulphur	60 bc	63.33 d	66.66 d	70 de	73.33 c
Tall	**100 a**	**100 a**	**100 a**	**100 a**	**100 a**
CV%	4.71	2.60	2.40	2.64	2.04

Table 23. Fungitoxicity of fungicides against *Drechslera oryzae* at different concentrations

Name of fungicides	Per cent inhibition of radial growth at different concentrations (ppm)				
	100	**200**	**300**	**400**	**500**
Bavistin	22.22 d	40 f	52 f	56 e	**100 a**
Capvit	50 b	60 c	60.33 e	66.66 d	**100 a**
Dithane	50 b	66.66 b	73.33 c	80 c	**100 a**
Greengel	35.38 c	38.46 f	69.23 d	84.62 b	**100 a**
Hayvit	50 b	52 d	70 cd	80 c	**100 a**
Indofil	35 c	47 e	67.65 d	**100 a**	**100 a**
Ridomil	50 b	60 c	66.66 d	**100 a**	**100 a**
Salcox	22.22 d	57.14 c	77.77 b	**100 a**	**100 a**
Sulphur	25 d	40 f	100 a	**100 a**	**100 a**
Tall	**100 a**	**100 a**	**100 a**	**100 a**	**100 a**
CV%	7.31	4.25	2.91	1.63	00

Values within the same column with a common letter (s) do not differ significantly at 5% level by LSD

4.13.5. Fungitoxicity of fungicides against *Fusarium moniliforme* at different concentrations

The complete inhibition of radial growth of *F. moniliforme* was observed with Dithane, Ridomil and Sulphur at 300, 400 and 500 ppm. Bavistin also showed complete inhibition at 400 and 500 ppm. Capvit and Salcox showed complete inhibition at 500 ppm, 71 and 55% at 400 ppm, respectively. Indofil, Hayvit and Greengel recorded 65.5, 46.42 and 60% inhibition of growth at 500 ppm, respectively (Table 24)

4.13.6. Fungitoxicity of fungicides against *Fusarium solani* at different concentrations

The radial growth of *F. solani* was completely inhibited with Bavistin and Tall at all concentrations. All fungicides except Greengel, Hayvit and Indofil showed complete inhibition at 500 ppm. Dithane also showed complete inhibition at 300 and 400 ppm. Ridomil and Sulphur showed complete inhibition at 400 ppm (Table 25).

4.13.7. Fungitoxicity of fungicides against *Microdochium oryzae* at different concentrations

All fungicides except Hayvit were completely inhibited the radial growth of *M. oryzae.* at 500 ppm concentration. Indofil, Ridomil and Sulphur showed complete inhibition at 300 and 400 ppm among all concentrations. Bavistin, Dithane and Greengel also showed 100% inhibition at 400 ppm (Table 26).

4.13.8. Fungitoxicity of fungicides against *Pestalotiopsis guepinii* at different concentrations

Indofil completely inhibited the radial growth of *P. guepinii* at 300, 400 and 500 ppm concentrations. Bavistin and Greengel also showed complete inhibition at 400 and 500 ppm. Ridomil and Sulphur showed 87.5% inhibition, Salcox and Capvit showed 80% inhibition of growth at 500 ppm. Dithane and Hayvit were responsible for 75 and 73.33%

Table 24. Fungitoxicity of fungicides against *Fusarium moniliforme* at different concentrations

Name of fungicides	Per cent inhibition of radial growth at different concentrations (ppm)				
	100	**200**	**300**	**400**	**500**
Bavistin	10.10 h	12 h	60 c	**100 a**	**100 a**
Capvit	14.28 g	18 g	64.28 b	71 b	**100 a**
Dithane	43.33 b	66.66 b	**100 a**	**100 a**	**100 a**
Greengel	26.66 d	33.33 d	36.66 f	40 e	60 c
Hayvit	18 cf	21.41 f	28.6 g	35.7 f	46.42 d
Indofil	34.88 c	41.86 c	53.44 d	58.13 c	65.5 b
Ridomil	20 e	40 c	**100 a**	**100 a**	**100 a**
Salcox	10 h	40 c	45 e	55 d	**100 a**
Sulphur	16.66 fg	40 c	**100 a**	**100 a**	**100 a**
Tall	55.55 d	**80 a**	**100 a**	**100 a**	**100 a**
CV%	5.67	4.34	1.95	2.00	1.09

Table 25. Fungitoxicity of fungicides against *Fusarium solani* at different concentrations

Name of fungicides	Per cent inhibition of radial growth at different concentrations (ppm)				
	100	**200**	**300**	**400**	**500**
Bavistin	**100 a**	**100 a**	**100 a**	**100 a**	**100 a**
Capvit	14.28 g	28 f	44.28 fg	75 b	**100 a**
Dithane	46.33 b	66.66 b	**100 a**	**100 a**	**100 a**
Greengel	20.66 e	35.33 e	46.66 f	50 d	70 b
Hayvit	20 e	21.41 g	28.6 h	35.7 e	46.42 d
Indofil	34.88 c	41.86 cd	53.44 d	58.13 c	65.5 c
Ridomil	30 d	40 de	80 b	**100 a**	**100 a**
Salcox	15 f	40 de	42 g	50 d	**100 a**
Sulphur	10.66 h	40 de	70 c	**100 a**	**100 a**
Tall	**100 a**	**100 a**	**100 a**	**100 a**	**100 a**
CV%	5.38	6.88	2.73	1.54	1.64

Values within the same column with a common letter (s) do not differ significantly at 5% level by LSD.

Table 26. Fungitoxicity of fungicides against *Microdochium oryzae* at different concentrations

Name of fungicides	Per cent inhibition of radial growth at different concentrations (ppm)				
	100	**200**	**300**	**400**	**500**
Bavistin	16.66 f	30 h	60 d	**100 a**	**100 a**
Capvit	20 e	30 h	66.66 c	70 c	**100 a**
Dithane	56.66 c	76.66 e	86.66 b	**100 a**	**100 a**
Greengel	20.66 e	66.66 f	86.66 b	**100 a**	**100 a**
Hayvit	25 d	45.05 g	50 e	55 d	70 b
Indofil	20.66 e	90 b	**100 a**	**100 a**	**100 a**
Ridomil	75 b	82.5 d	**100 a**	**100 a**	**100 a**
Salcox	20 e	50 g	60 d	90 b	**100 a**
Sulphur	75 b	86.66 c	**100 a**	**100 a**	**100 a**
Tall	**80 a**	**100 a**	**100 a**	**100 a**	**100 a**
CV%	3.82	2.75	1.79	1.04	0.65

Table 27. Fungitoxicity of fungicides against *Pestalotiopsis guepinii* at different concentrations

Name of fungicides	Per cent inhibition of radial growth at different concentrations (ppm)				
	100	**200**	**300**	**400**	**500**
Bavistin	37.5 e	52.5 d	75 c	**100 a**	**100 a**
Capvit	33.33 f	40 f	50 e	66.66 b	80 c
Dithane	42.5 c	52.5 d	65 d	70 b	75 d
Greengel	33.33 f	66.66 c	80 b	**100 a**	**100 a**
Hayvit	16.66 h	33.33 g	50 e	66.66 b	73.33 d
Indofil	52.5 b	75 b	**100 a**	**100 a**	**100 a**
Ridomil	12.5 i	25 h	37.5 f	50 d	87.5 b
Salcox	40 d	44 e	50 e	60 c	80 c
Sulphur	25 g	37.5 f	40 e	50 d	87.5 b
Tall	**100 a**	**100 a**	**100 a**	**100 a**	**100 a**
CV%	3.57	2.98	3.63	2.99	1.47

Values within the same column with a common letter (s) do not differ significantly at 5% level by LSD.

4.13.9. Fungitoxicity of fungicides against *Sarocladium oryzae* at different concentrations

The radial growth of *S. oryzae* was completely inhibited with Bavistin, Capvit, Salcox and Sulphur at 300, 400 and 500 ppm concentrations (Table 27). Amongst the ten fungicides, Tall showed best result and Hayvit showed least percentage of inhibition.

The results in this investigation showed similarity with the findings of different workers (Farid *et al.* 2002, Nghiep and Gaur 2005, Butt *et al.* 2011 and Yesmin *et al.* 2012). Farid *et al.* (2002) reported four fungicides *viz.*, Bavistin, Hinosan, Tilt 250 EC and Dithane M-45 against *Bipolaris oryzae* was the best with 100% reduction of the prevalence of the pathogen and inhibited the mycelial growth at 0.3% of the seed weight as seed treatments and 500 ppm as mycelial growth inhibition test followed by Tilt 250 EC, Hinosan and Bavistin. All the test fungicides were effective against *Bipolaris oryzae* at higher concentration (Farid *et al.* 2002).

Nghiep and Gaur (2005) reported that most of the seed borne pathogens of rice *viz. Bipolaris oryzae, Alternaria padwickii, Curvularia lunata* and other seed borne fungi were cotrolled by vitavax, mancozeb and bavistin.

Butt *et al.* (2011) isolated four pathogenic fungi *viz., Alternaria alternata, Fusarium moniliforme, Curvularia* sp.and *Helminthosporium* sp. from 5 varieties of rice *viz.* KS-282, Basmati-385, 370, 198 and Kernel. Four chemical fungicides such as antracal, topsin, mancozeb and derosal were used to investigate their effect on seed borne mycoflora. Antracal completely stopped the growth of *Helminthosporium* sp. and *Curvularia* sp. but other 3 fungicides suppressed the growth of these fungi up to 50% .

Yesmin *et al*. (2012) reported that pathogenic fungi of rice were reduced up to 100% over the control, where provax was found best and significantly similar to garlic extract (1:1).

Table 28. Fungitoxicity of fungicides against *Sarocladium oryzae* at different concentrations

Name of fungicides	Per cent inhibition of radial growth at different concentrations (ppm)				
	100	**200**	**300**	**400**	**500**
Bavistin	30.43 c	52.17 c	**100 a**	**100 a**	**100 a**
Capvit	34.78 b	56.52 b	**100 a**	**100 a**	**100 a**
Dithane	19.04 d	33.33 e	38.09 c	42.85 d	52.38 d
Greengel	14.28 e	28.57 f	40 c	42.85 d	52.38 d
Hayvit	18 d	21.41 g	28.60 d	35.7 e	45.5 e
Indofil	34.88 b	41.86 d	53.44 b	58.13 c	65.5 c
Ridomil	35 b	50 c	55 b	60 b	70 b
Salcox	34.78 b	56.52 b	**100 a**	**100 a**	**100 a**
Sulphur	20 d	50 c	**100 a**	**100 a**	**100 a**
Tall	**100 a**	**100 a**	**100 a**	**100 a**	**100 a**
CV%	3.51	3.98	4.89	1.42	1.43

Values within the same column with a common letter (s) do not differ significantly at 5% level by LSD.

4.14. Per cent inhibition of radial growth of pathogenic fungi owing to plant extracts at different concentrations

Antifungal properties of ethanol extract of *A. heterophyllus*, *T. erecta*, *D. metel*, *S. alata*, *A. indica*, *C. medica*, *M. indica*, *A. racemosus*, *N. indicum*, *A. sativum* at 5, 10 and 20% concentrations were evaluated on 9 pathogenic fungi. All the plant extracts completely inhibited radial growth of the test fungi at 20% concentration except *A. racemosus* and *Nerium indicum*. Ten per cent concentration of *A. indica* and *C. medica* also showed complete inhibition except *F. moniliforme* (Plate 4 & 5).

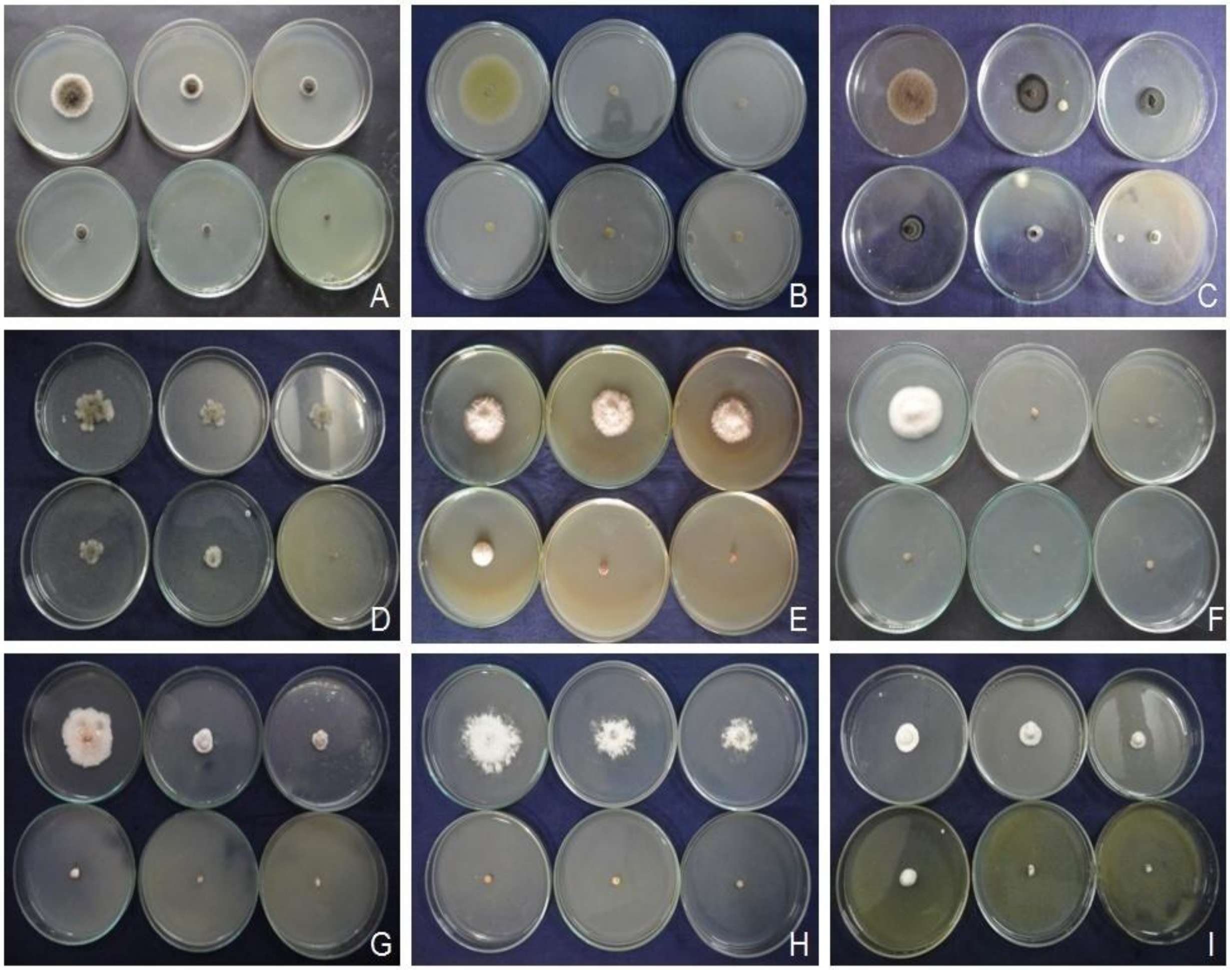

Plate 2. Per cent inhibition of radial growth of A. *Alternaria alternata*, B. *Aspergillus flavus,* C. *Curvularia lunata,* D. *Drechslera oryzae*, E. *Fusarium moniliforme,* F. *Fusarium solani*, G. *Microdochium oryzae,* H. *Pestalotiopsis guepinii* and I. *Sarocladium oryzae* at 100, 200, 300, 400 and 500 ppm concentrations of Bavistin.

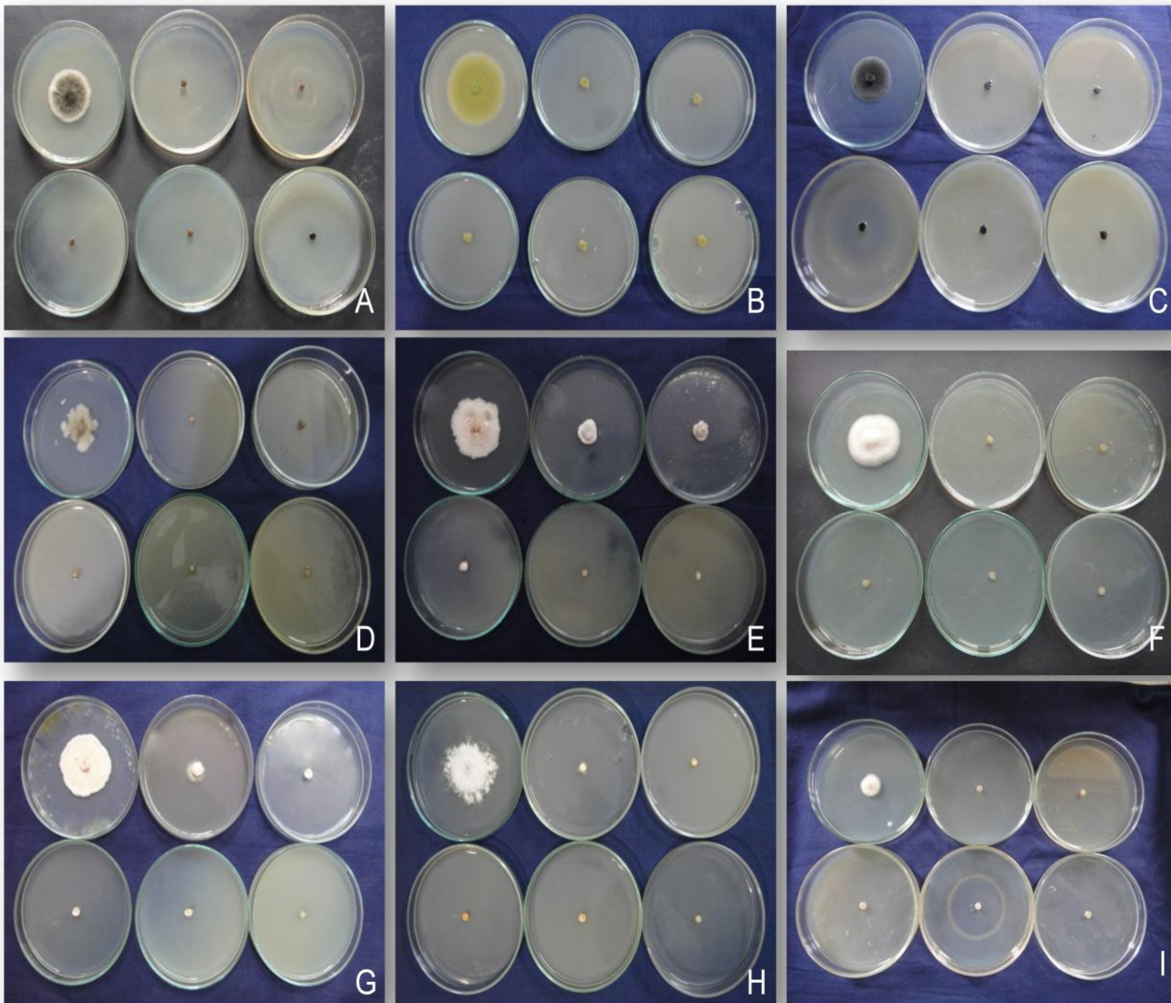

Plate 3. Per cent inhibition of radial growth of A. *Alternaria alternata*, B. *Aspergillus flavus,* C. *Curvularia lunata*, D. *Drechslera oryzae*, E. *Fusarium moniliforme*, F. *Fusarium solani*, G. *Microdochium oryzae*, H. *Pestalotiopsis guepinii* and I. *Sarocladium oryzae* at 100, 200, 300, 400 and 500 ppm concentrations of Tall 25 EC.

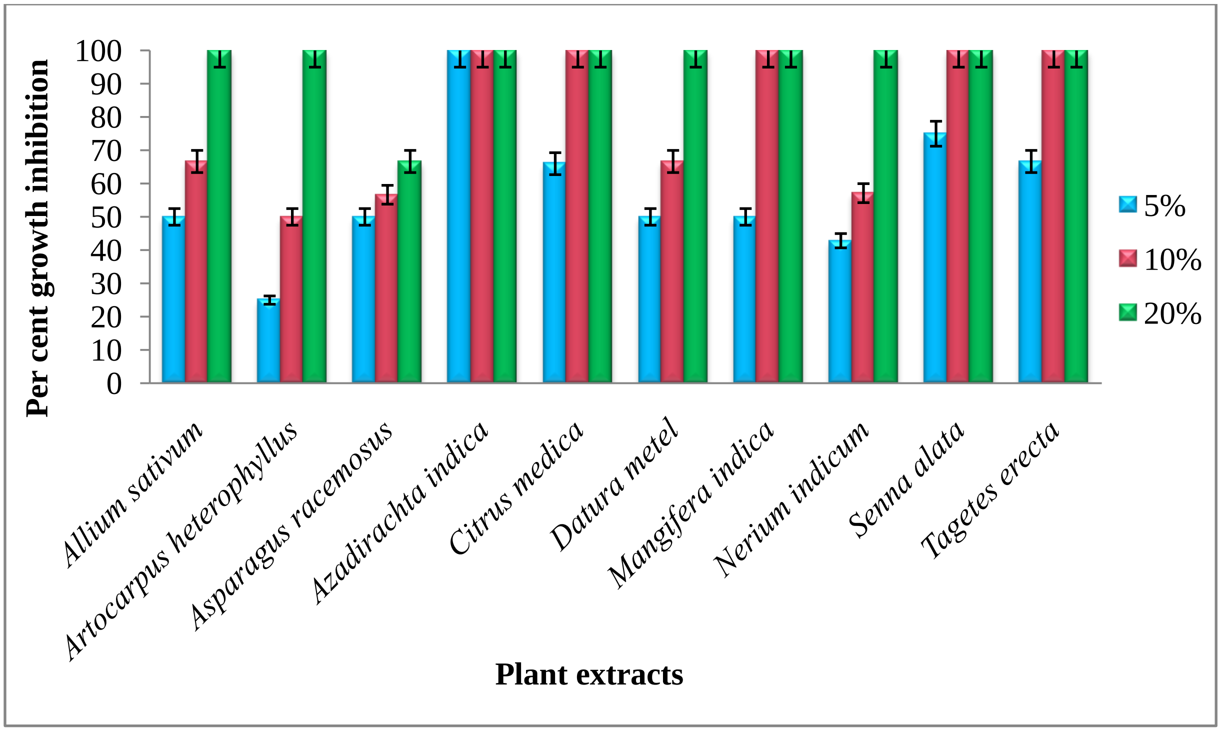

Fig.4. Per cent inhibition of radial growth of *Alternaria alternata* owing to plant extracts at different concentrations

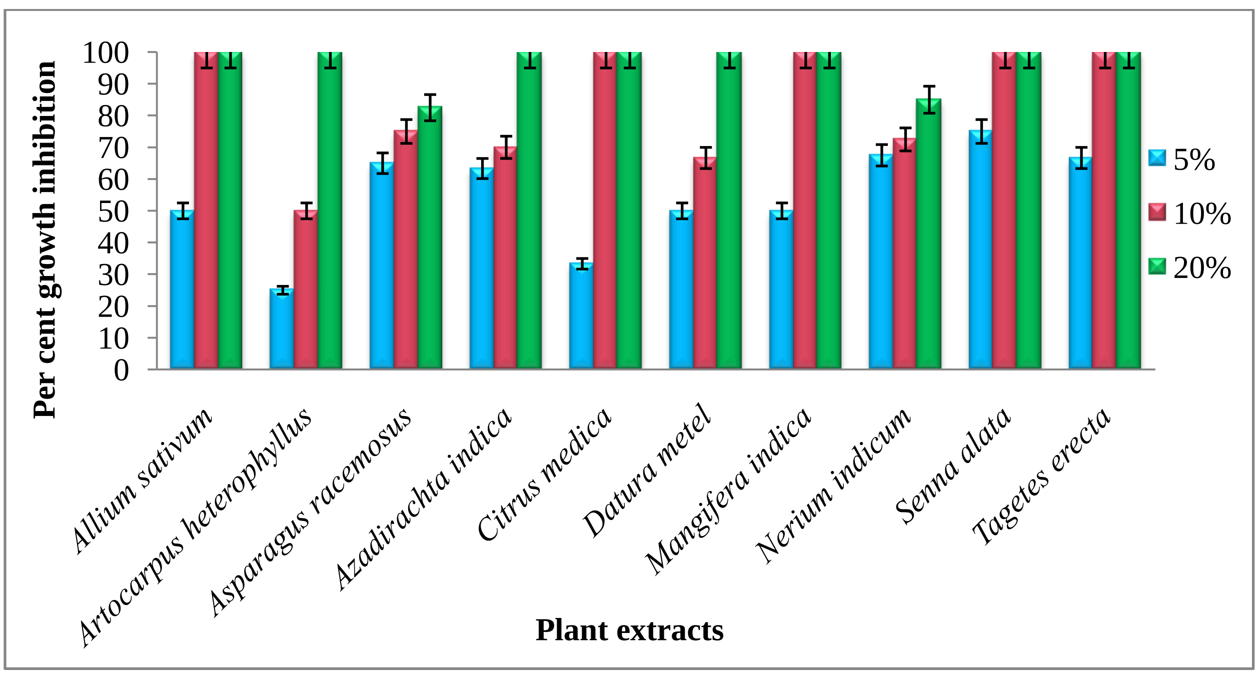

Fig. 5. Per cent inhibition of radial growth of *Aspergillus flavus* owing to plant extracts at different concentrations

4.14.1. Per cent inhibition of radial growth of *Alternaria alternata* owing to plant extracts at different concentrations

Ethanol extract of *A. indica* and *C. medica* at different concentrations also showed complete inhibition of radial growth of all pathogenic fungi. Only 5% concentration of *C. medica* showed 66% inhibition of vegetative growth of *A. alternata*. All the eight plants i.e. *A. sativum, A. heterophyllus, A. racemosus, D. metel, M. indica, N. indicum, S. alata* and *T. erecta* showed 50, 25, 50, 50, 50, 42.85, 75 and 66.66% inhibition of growth of *A. alternata* at 5% concentration, respectively (Fig. 4).

4.14.2. Per cent inhibition of radial growth of *Aspergillus flavus* owing to plant extracts at different concentrations

Twenty per cent leaf extract of all ten plants except *A. racemosus and N. indicum* showed complete inhibition of radial growth of *A. flavus*. Both the plants inhibited only 66.66% of radial growth. At 10% concentration of extract of *A. sativum, C. medica*, *M. indica and T. erecta* were also responsible for complete inhibition of radial growth. Five per cent ethanol extracts of six plants *viz., A. racemosus*, *A. heterphillus, A.indica, D. metel, N. indicum* and *T. erecta* showed 50, 76, 70, 66.66 and 72.5% inhibition, respectively.

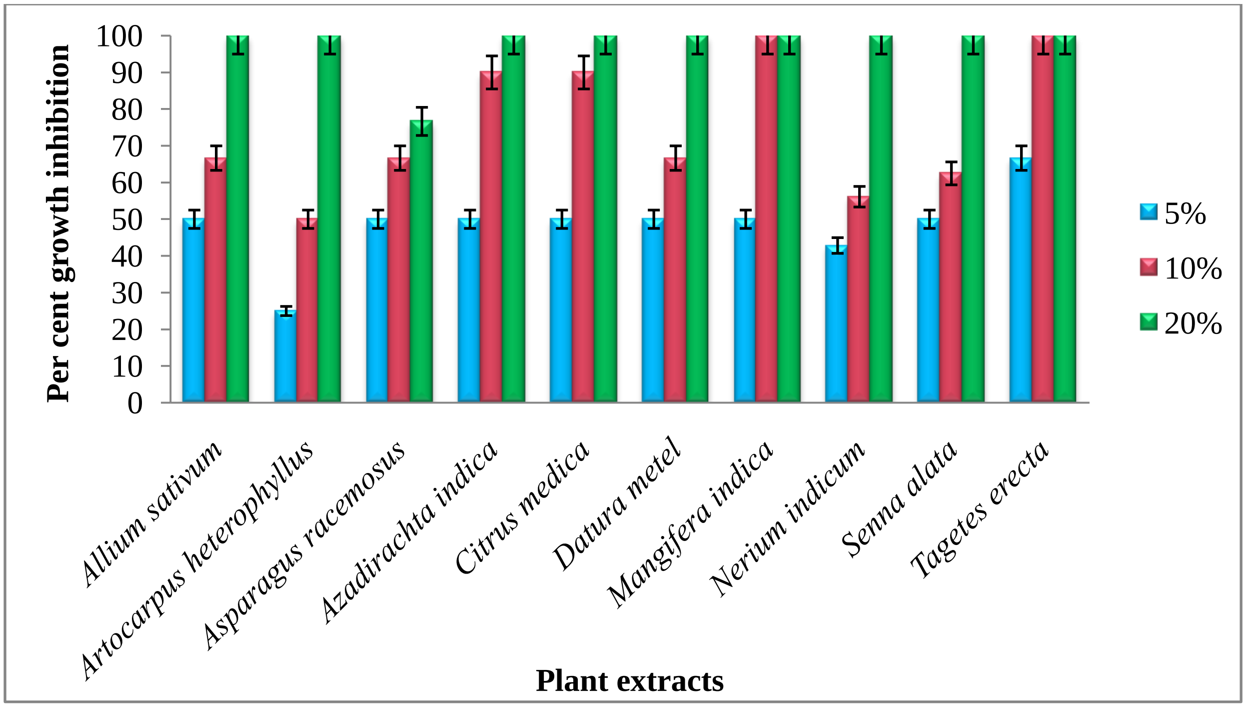

Fig. 6. Per cent inhibition of radial growth of *Curvularia lunata* owing to plant extracts at different concentrations.

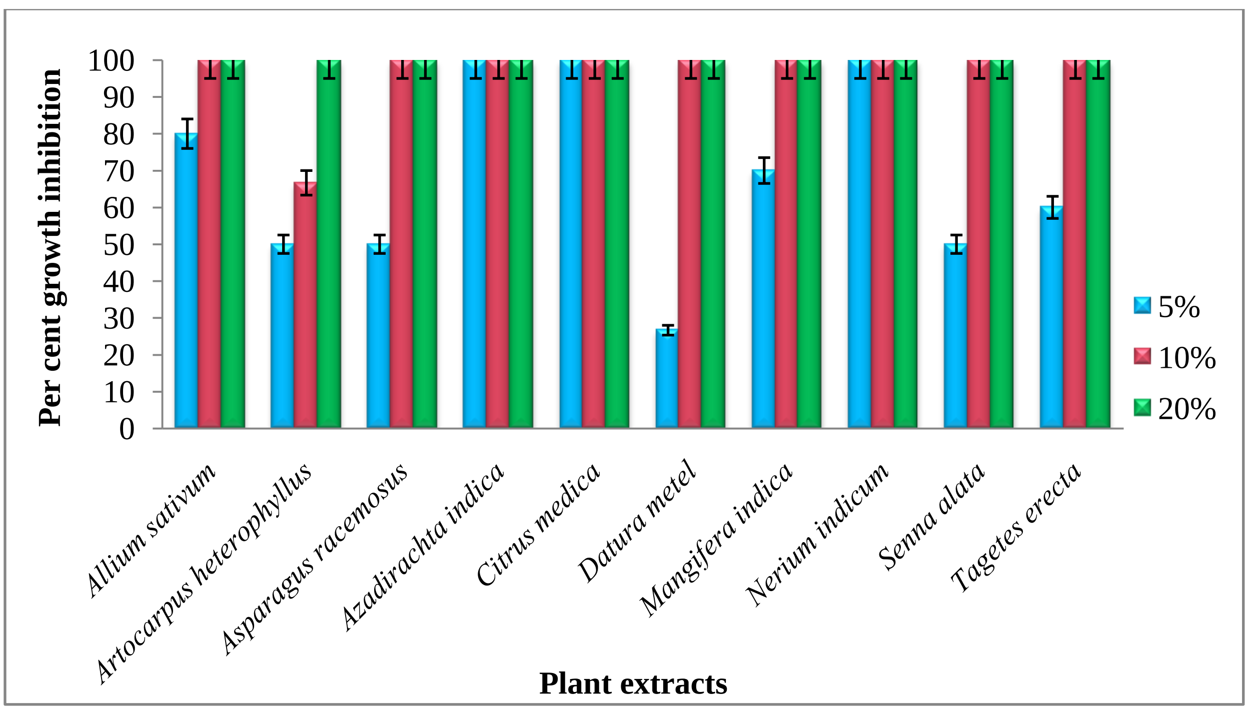

Fig. 7. Per cent inhibition of radial growth of *Drechslera oryzae* owing to plant extracts at different concentrations.

4.14.3. Per cent inhibition of radial growth of *Curvularia lunata* owing to plant extracts at different concentrations

Ten per cent ethanol extract of *T. erecta* and *M. indica* were also responsible for complete inhibition of growth. Ten per cent ethanol extract of *D. metel*, *S. alata*, *A. heterophyllus*, *A. racemosus*, *N. indicum* and *A. sativum* showed 74, 60, 82.9, 50, 50 and 50% inhibition of radial growth respectively. Ethanol extract of 8 plants i.e. *A. sativum, A. heterophyllus, A. racemosus, D. metel, M. indica, N. indicum, S. alata* and *T. erecta* showed 33.33, 34.28, 25, 52, 33.33, 20, 50 and 33.33% inhibition of radial growth of *C. lunata* at 5% concentration, respectively (Fig. 6).

4.14.4. Per cent inhibition of radial growth of *Drechslera oryzae* owing to plant extracts at different concentrations

Ten and 20% ethanol extract of all ten plants showed complete inhibition of radial growth of *D. oryzae* except *A. heterophyllus* which inhibited 66.66% growth at 10% concentration. Ethanol extract of *A. indica, C. medica* and *N. indicum.* were also responsible for complete inhibition of radial growth at different concentrations. Five per cent ethanol extracts of seven plants *A. sativum, A. heterophyllus, D. metel, M. indica, A. racemosus, S. alata* and *T. erecta* were responsible for 80, 50, 26.66, 70, 50, 50 and 60% inhibition of growth, respectively (Fig. 7).

4.14.5. Per cent inhibition of radial growth of *Fusarium moniliforme* owing to plant extracts at different concentrations

Ten and 20% ethanol extracts of all ten plants completely inhibited the radial growth of *F. moniliforme*. *A. indica* and *C. medica* also showed complete inhibition at 5% concentration. *Allium sativum, A. heterophyllus, A. racemosus, D. metel, M. indica, N. indicum, S. alata* and *T. erecta* were also responsible for 48, 40, 50, 33.33, 60, 50 and 50% inhibition of radial growth, respectively at 5% concentration (Fig. 8).

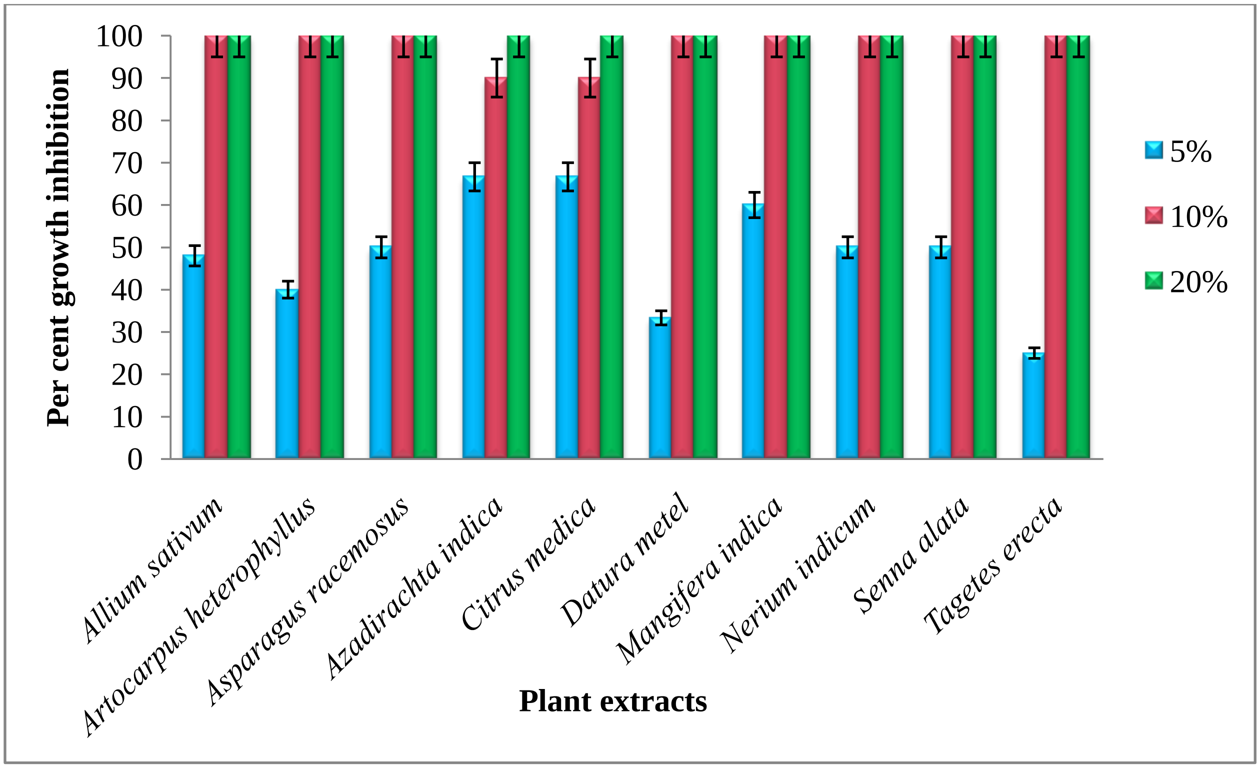

Fig 8: Per cent inhibition of radial growth of *Fusarium moniliforme* owing to plant extracts at different concentrations

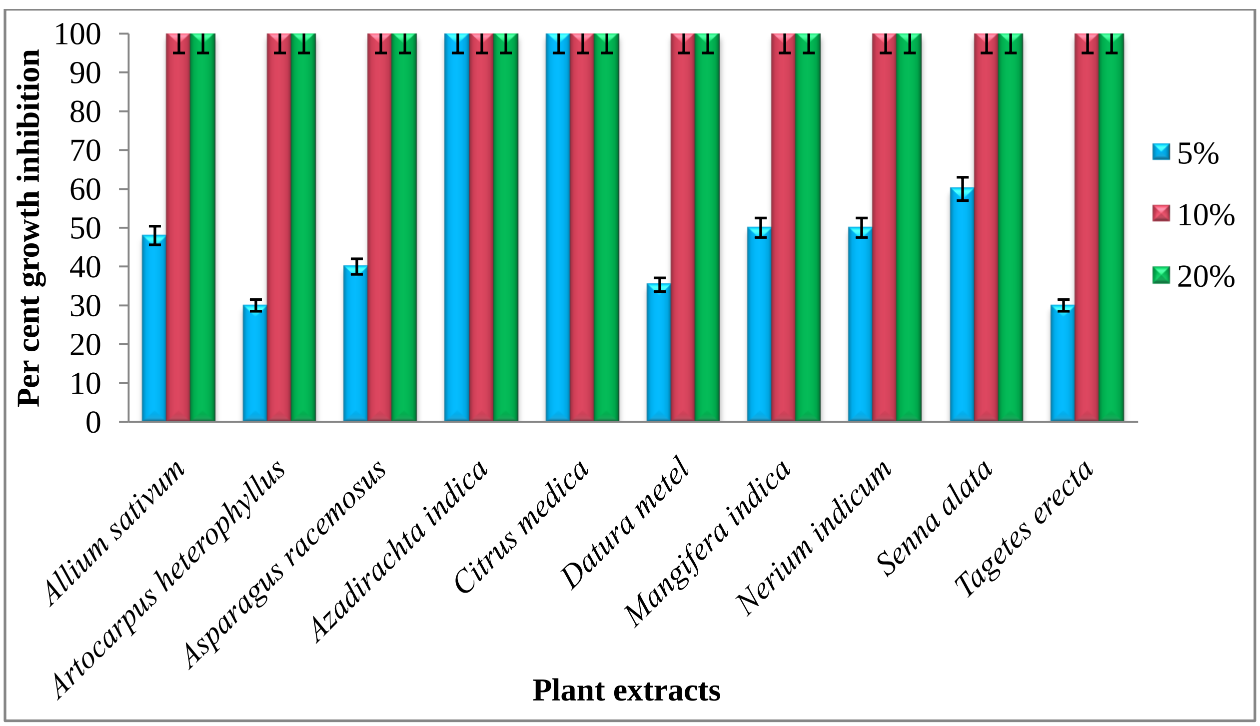

Fig.9. Per cent inhibition of radial growth of *Fusarium solani* owing to plant extracts at different concentrations

4.14.6. Per cent inhibition of radial growth of *Fusarium solani* owing to plant extracts at different concentrations

In *F.solani* 10 and 20% concentration of all plant extracts showed complete inhibition of radial growth. .*A. indica* and C. *medica* also showed 100% inhibition at 5% concentration .*A. sativum, A.heterophillus, A. racemosus, D. metel, M. indica, N. indicum, S. alata* and *T. erecta* were also responsible for 48, 30, 40, 35.32, 50, 50, 60 and 30% inhibition of growth, respectively (Fig. 9)

4.14.7. Per cent inhibition of radial growth of *Microdochium oryzae* owing to plant extracts at different concentrations

Ten and 20% concentrations of all plant extracts showed complete lnhibition of *M. oryzae. A. indica* and *C. medica* were responsible for 100% inhibition at 5% concentration. *A. heterophyllus, A. racemosus, D. metel, M. indica, N. indicum, S. alata* and *T. erecta* were also responsible for 50, 60, 25, 50, 60, 60 and 33.33% inhibition of radial growth at 5% concentration, respectively (Fig. 10)

4.14.8. Per cent inhibition of radial growth of *Pestalotiopsis guepinii* owing to plant extracts at different concentrations

Ethanol extract of all the plants showed complete inhibition of radial growth of *P. guepinii* at 20% concentration. Ten per cent extract of *A. sativum*, *A. indica*, *C. medica*, *M. indica*, *D. metel* and *N. indicum* also showed 100% inhibition. Ten per cent ethanol extracts of *S. alata*, *T. erecta*, *A. heterophyllus and A. racemosus* were responsible for 80, 75, 75 and 80% inhibition of growth, respectively. Five per cent ethanol extract of all the tested plants i.e. *A. sativum, A. heterophyllus, A. racemosus, A. indica, C. medica, D. metel, M. indica, N. indicum, S. alata* and *T. erecta* were also responsible for 60, 47.5, 57.12, 68.42, 83.15, 67.5, 62.5, 71.42, 60 and 62.5% inhibition of growth, respectively (Fig. 11).

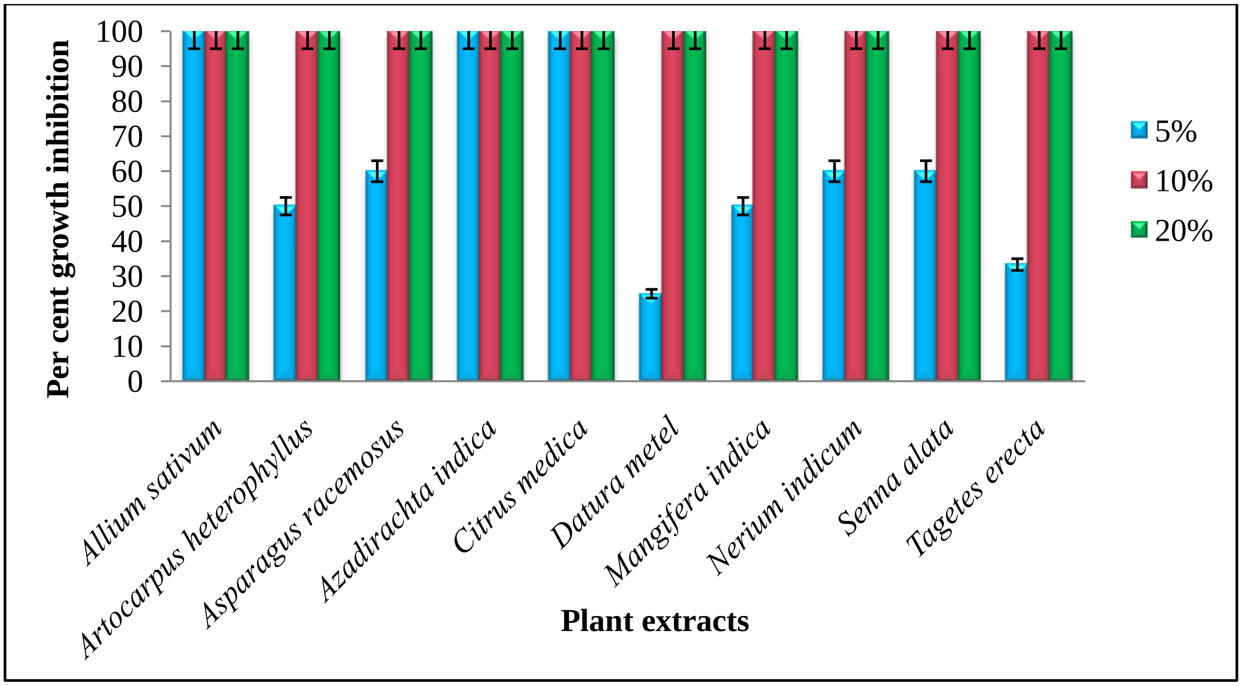

Fig. 10. Per cent inhibition of radial growth of *Microdochium oryzae* owing to plant extracts at different concentrations

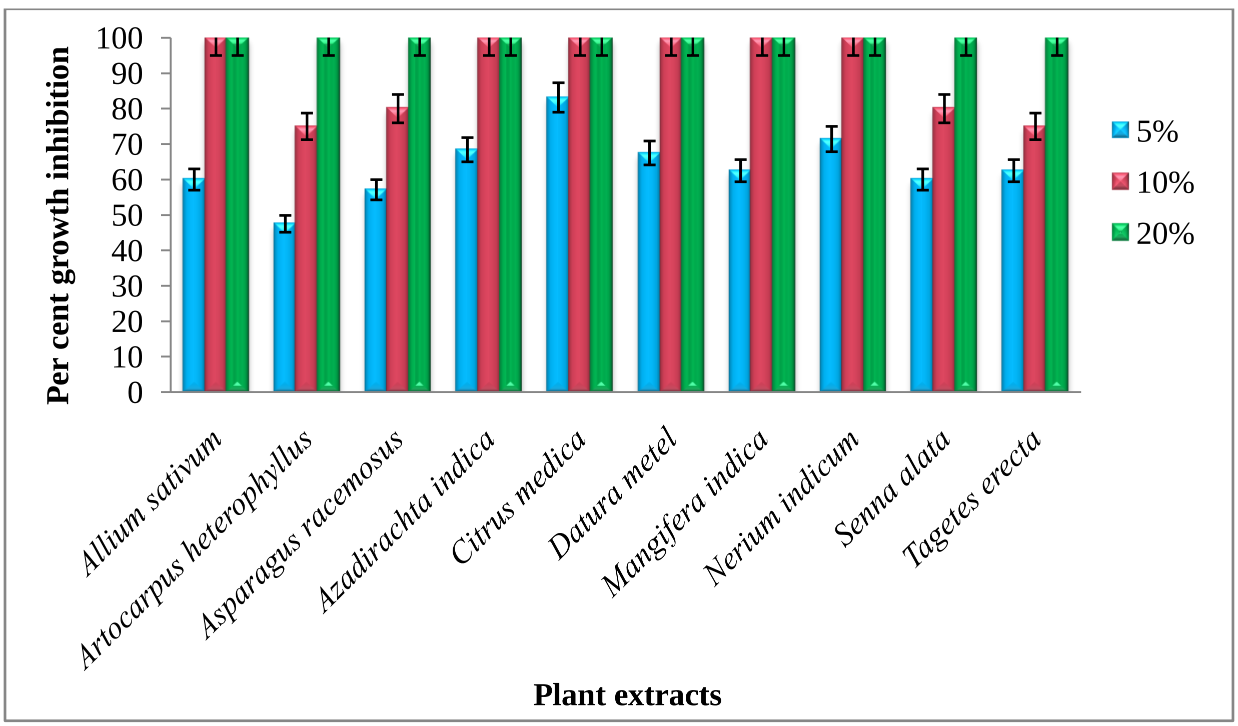

Fig. 11. Per cent inhibition of radial growth of *Pestalotiopsis guepinii* owing to plant extracts at different concentrations

4.14.9. Per cent inhibition of radial growth of *Sarocladium oryzae* owing to plant extracts at different concentrations

Except *A. racemosus*, 20% concentrations of all plant extracts showed complete lnhibition of *S. oryzae*. Only 10% concentrations of *T. erecta* and *M. indica* showed complete inhibition. Rest of all plants *viz., A.sativum, A. heterophyllus, A. indica, C. medica, D. metel, M. indica, N. indicum, S. alata* and *T. erecta* were also responsible for 66.66, 50, 66.66, 90, 90, 66.66, 54.14 and 62.5% inhibition of radial growth at 5% concentration, respectively (Fig. 12). The results in this investigation of management of pathogenic fungi of rice owing to plant extracts at different concentrations showed the similarity with the findings of Mohana *et al.* (2011), Yeasmin *et al.* (2012), Mansur *et al.* (2013), Islam and Monjil (2016).

Mohana *et al.* (2011) from India reported that methanol extract of *Acacia nilotica, Caesalpinia coriaria*, *Decalepis hamiltonii*, *Emblica officinalis, Lawsonia inermis* and *Mimosops elengi* showed significant antifungal activity at 3500 µg/ml concentration on seed pathogens *viz., Alternaria alternata, Aspergillus flavus, Curvularia lunata, Drechslera oryzae*, *D. halodes*, *Fusarium moniliforme, Pyricularia oryzae* and *Trichoconis padwickii* by poisoned food technique.

Yeasmin *et al*. (2012) from India reported that seed borne fungi of rice were *Bipolaris oryzae, Curvularia oryzae, Fusarium oxysporum, F. moniliforme, Nigrospora oryzae, Aspergillus flavus, A. niger* and *Penicillium* sp., where prevalence of *Bipolaris oryzae* (7.5%) and *F. moniliforme* (8.3%) were the maximum. All the treatments significantly reduced the seed borne fungi up to 100% over the control, where Provax was found best and was significantly similar to garlic (1:1) extract against seed borne pathogens of rice.

Mansur *et al.* also (2013) reported that garlic (1:1) extract was most effective in controlling seed borne fungi of rice. Islam and Monjil (2016) observed complete inhibition of sheath blight pathogen when treated with four indigenous medicinal plants extracts *viz.*, tulsi, nishinda, thankuni and biskatali.

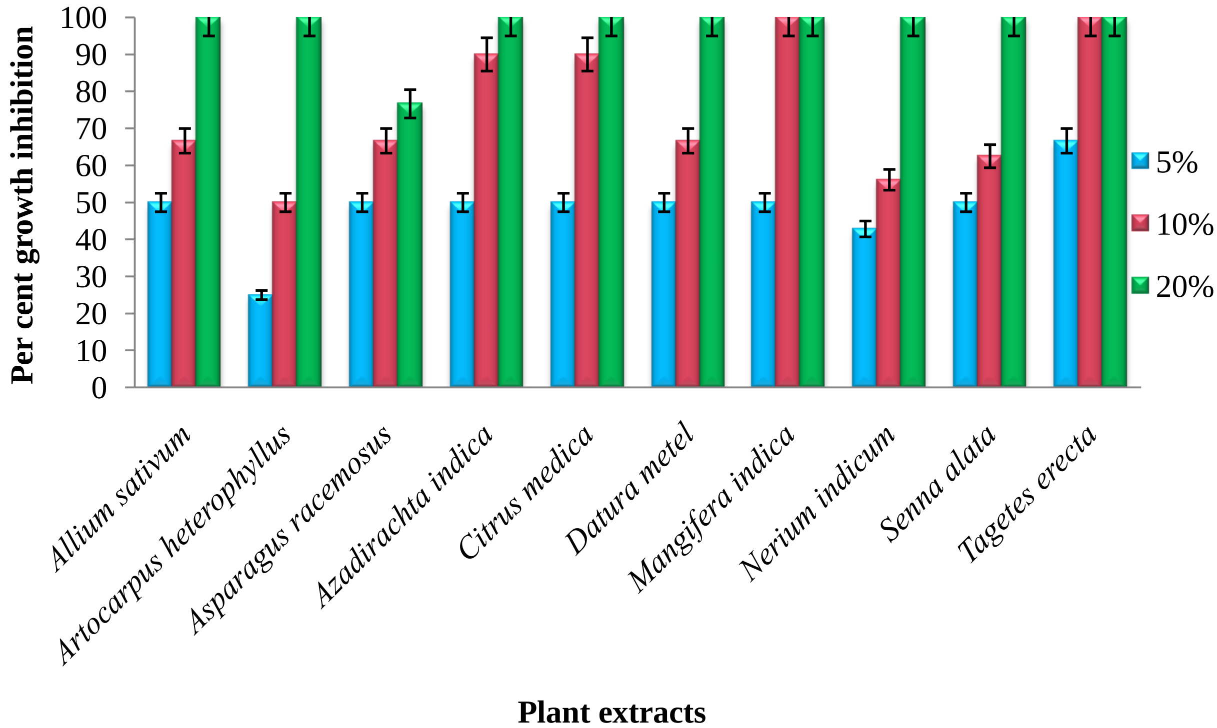

Fig.12. Per cent inhibition of radial growth of *Sarocladium oryzae* owing to plant extracts at different concentrations.

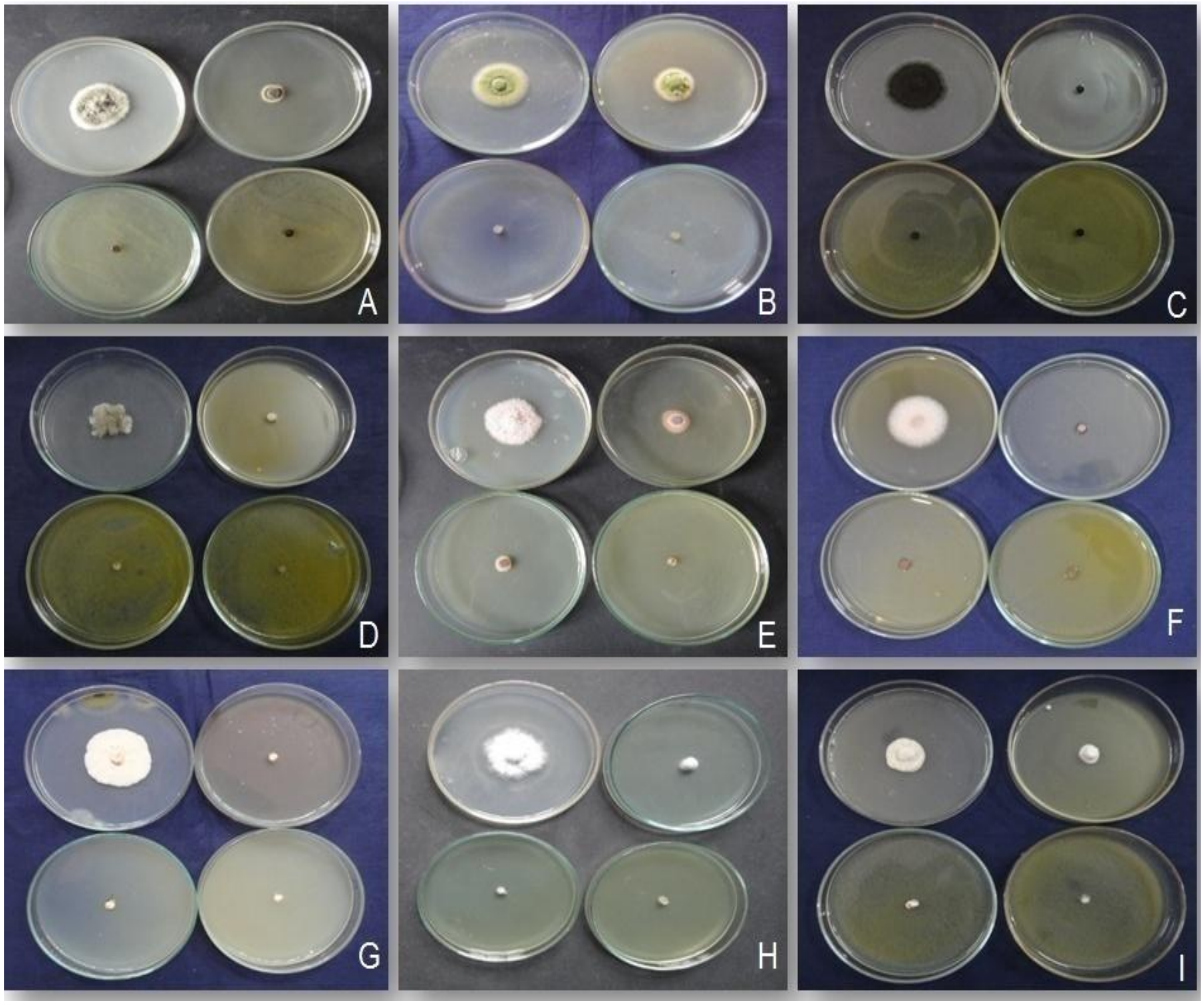

Plate 4. Per cent inhibition of radial growth of A. *Alternaria alternata*, B. *Aspergillus flavus*, C. *Curvularia lunata*, D. *Drechslera oryzae*, E. *Fusarium moniliforme*, F. *Fusarium solani*, G. *Microdochium oryzae*, H. *Pestalotiopsis guepinii* and I. *Sarocladium oryzae* at 5, 10 and 20% concentrations of plant extracts of *Azadirachta indica* .

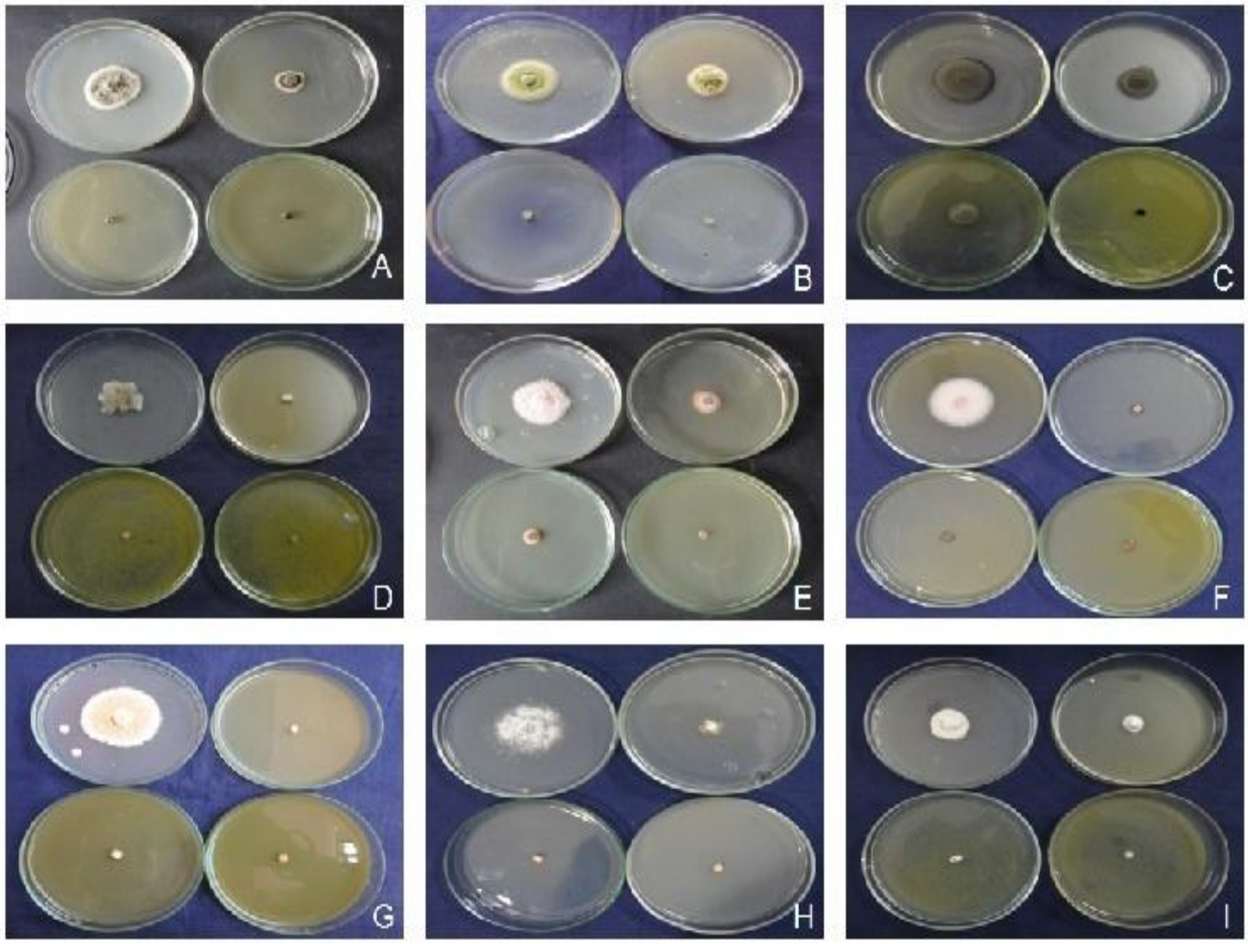

Plate 5. Per cent inhibition of radial growth of A. *Alternaria alternata,* B. *Aspergillus flavus,* C. *Curvularia lunata,* D. *Drechslera oryzae,* E. *Fusarium moniliforme,* F. *Fusarium solani,* G. *Microdochium oryzae,* H. *Pestalotiopsis guepinii* and I. *Sarocladium oryzae at* 5, 10 and 20% concentrations of plant extracts of *Citrus medica.*

The study revealed the presence of nine pathogenic fungi *viz., A. alternata, A. flavus C. lunata, D. oryzae, F. moniliforme F. solani, M. oryzae, P. guepinii* and *S. oryyzae* associated with rice grains were completely controlled *in vitro* at different concentrations of Tall 25 EC. Subsequently antifungal properties of ethanol extracts of all the ten plants completely inhibited the radial growth of all the test fungi at 20% concentration.

4.15. Antagnistic potential of the soil fungi agalnst test rice pathogens

4.15.1. Colony intraction

Antagonistic potential of the selected six soil fungi against the eight test pathogens are presented in Table 29. In this study antagonistic relationship ranged from grade 2 to 4. However, grade 3 was found most commonly encountered type of colony interaction as 20 interactions out of 48 were incorporated in this grade, followed by grade 2 (17 out of 48) and the grade 4 was recorded frequently (11 out of 48). Among 6 soil fungi only *Trichoderma harzianum* showed grade 4 against all the test pathogens except *A. alternata* followed by *Aspergillus niger* (Table 29) which was similar with the observation of Prince *et al.* (2011) and Akter *et al.* (2014). He observed interactions at grade 4 between *T. harzianum* and *Colletotrichum falcatum.*

Results in colony interaction tests showed that the radial growth inhibition of the test pathogens with the soil fungi was in the range of 35.50%- 88.00%. The highest growth inhibition was observed owing to *T. harzianum* against *A. alternata* followed by *F. solani* (86.00%), *Sarocladium oryzae* (82%) and *Drechslera oryzae* (75.25%). The maximum inhibition of *Curvularia lunata* (80%), *Fusarium moniliforme* (76%) and *Pestalotipsis guepinii* (75%) was observed due to *Trichoderma viride* (Table 29, Plate 6 & 7). Prince *et al.* (2011) reported that 53.8% inhibition of *Botrytis fabae* due to *T. harzianum* and Akter *et al.* (2014) reported that 81.48% inhibition was observed due to *T. harzianum* against *F. moniliforme* in colony interaction.

4.15.2. Effect of volatile substances emanating from the cultures of the soil fungi on the radial growth of the test pathogens

Volatile substances from soil fungi inhibited radial growth of the test pathogens varied from 8.33-57.36%. The highest inhibition (57.36%) was found owing to culture filtrate

Table 29. Antagonistic potential of soil fungi against the test pathogens of rice

Name of soil fungi	Test pathogens							
	Aa	Cl	Do	Fm	Fs	Mo	Pg	So
Grades in colony interactions								
Aspergillus flavus	3	2	3	3	3	3	3	2
A. fumigatus	2	2	2	2	3	2	2	2
A. niger	3	4	4	2	2	3	3	2
Penicillium sp.	3	3	3	3	3	2	3	2
Trichoderma harzianum	2	4	4	4	2	4	4	3
T. viride	2	3	4	3	3	3	2	3
Per cent inhibition in colony interaction								
Aspergillus flavus	50.10 d	50.78 d	65.6 c	46.00 d	60.25 b	60.00 b	55.00 b	47.36 cd
A. fumigatus	45.25 e	42.52 f	50.00 d	42.00 e	53.20 c	52.00 c	51.25 c	46.00 d
A. niger	70.66 b	73.87 c	70.25 b	66.66 b	46.66 d	62.00 a	55.15 b	50.00 c
Penicillium sp.	50.25 de	45.53 e	42.15 e	**35.50 f**	48.66 d	54.25 d	45.00 d	50.00 c
Trichoderma harzianum	**88.00 a**	74.55 b	75.25 a	55.25 c	86.00 a	60.25 b	55.25 b	82.00 a
T. viride	60.25 c	80.00 a	75.25 a	76.00 a	55.25 c	50.15 c	75.00 a	56.00 b
CV %	1.98	0.98	1.41	1.91	1.05	1.55	1.02	1.35

Aa = *Alternaria alternata*, Cl = *Curvularia lunata*, Do = *Drechslera oryzae*, Fm = *Fusarium moniliforme*, Fs = *Fusarium solani*, Mo = *Microdochium oryzae*, Pg = *Pestalotiopsis guepinii* and So = *Sarocladium oryzae.*

Grades from 1 to 5 based on Skidmore and Dickinson (1976). Grade 2: Mutual intermingling growth where the fungus is ceased and being overgrowth by the opposed fungus, Grade 3: Intermingling growth where the fungus is growing into the opposed fungus either above or below, Grade 4: Slight inhibition with a narrow demarcation line (1-2 mm).

Values within the same column with a common letter (s) do not differ significantly at 5% level by DMRT.

Name of soil fungi	Test pathogens							
	Aa	Cl	Do	Fm	Fs	Mo	Pg	So
Per cent inhibition owing to volatile substances								
Aspergillus flavus	25.00 d	33.33 c	33.33 ab	22.66 d	8.50 f	11.90 e	20.00 c	21.05 c
A. fumigatus	**8.33 f**	13.33 e	20.00 c	33.33 c	11.90 e	8.50 f	38.46 b	12.00 d
A. niger	40.00 b	37.50 bc	39.85 ab	46.36 a	45.25 a	25.50 d	56.60 a	25.00 b
Penicillium sp.	8.34 f	25.93 d	18.18 c	11.90 e	8.53 f	8.60 f	20.00 c	20.00 c
Trichoderma harzianum	15.00 e	45.00 a	38.85 ab	37.14 b	25.00 cd	36.25 a	**57.36 a**	30.33 a
T. viride	46.00 a	36.66 b	39.00 ab	46.80 a	21.50 b	28.75 b	56.60 a	32.31 a
CV %	5.08	4.73	4.82	2.20	3.10	2.93	3.69	5.46
Per cent inhibition owing to non-volatile metabolites								
Aspergillus flavus	40.32 d	39.00 d	45.20 c	45.75 d	42.00 d	45.00 b	33.45 b	30.00 d
A. fumigatus	35.20 e	30.51 e	40.00 d	39.00 e	38.00 e	38.25 c	32.18 c	**29.05 d**
A. niger	55.55 b	52.50 bc	62.50 a	52.00 b	55.00 a	60.50 a	45.03 b	45.50 c
Penicillium sp.	45.00 d	50.25 c	50.40 e	50.55 c	48.25 c	48.25 d	30.50 d	43.25 c
Trichoderma harzianum	60.50 a	**64.5 a**	62.25 b	53.55 a	52.50 b	61.25 b	45.84 a	50.35 a
T. viride	45.52 c	62.25 b	61.50 b	52.25 a	55.25 a	54.50 c	36.22 b	50.00 b
CV %	1.98	0.99	1.34	1.91	1.05	1.30	1.02	1.35

Aa = *Alternaria alternata*, Cl = *Curvularia lunata*, Do = *Drechslera oryzae*, Fm = *Fusarium moniliforme*, Fs = *Fusarium solani*, Mo = *Microdochium oryzae*, Pg = *Pestalotiopsis guepinii*, So = *Sarocladium oryzae.*

Values within the same column with a common letter (s) do not differ significantly at 5% level by LSD.

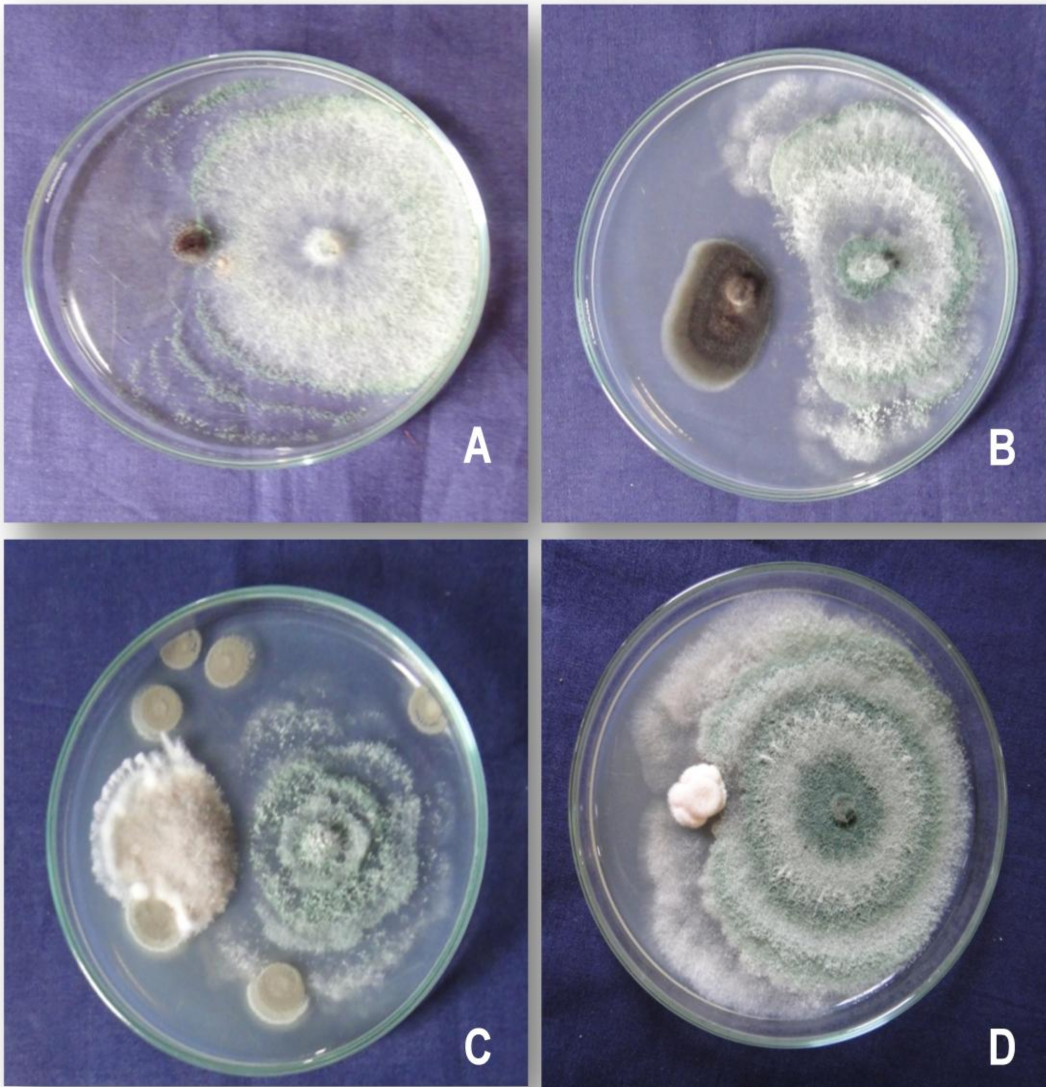

Plate 6. Colony interaction between A. *Alternaria alternata and Trichoderma harzianum*, B. *Curvularia lunata* and *Trichoderma viride*, C. *Drechslera oryzae* and *Trichoderma viride*, D. *Fusarium moniliforme* and *Trichoderma viride.*

of *T. harzianum* against *P. guepinii* followed by *F. moniliforme* (46.80%), *A. alternata* (46%) due to *T. viride.* The 4th highest inhibition (45.25%) was observed by the volatile substances of *A. niger* against *F. solani.* The 5th, 6th and 7th highest inhibition was observed in *C. lunata* (45%), *D. oryzae* (38.85%) and *M. oryzae* (36.25%) due to culture filtrate of *T. harzianum* (Table 29, Plate 8 & 9). Barakat *et al.* (2013) reported that volatile substances of

T. harzianum inhibited 39.77% mycelial growth of *Botrytis fabae* after 6 days of incubation, which showed similarity with my results (Table 29).

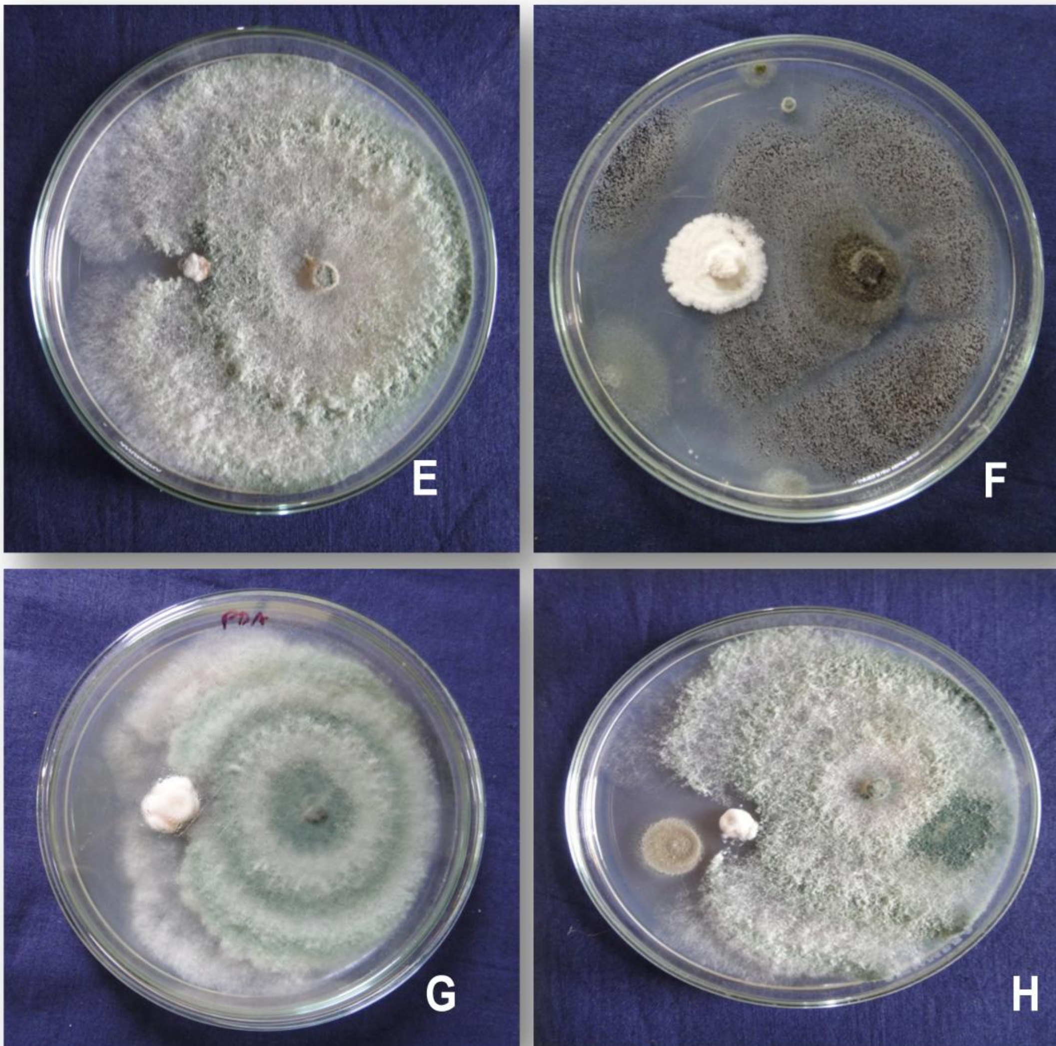

Plate 7. Colony interaction between E. *Fusarium solani and Trichoderma harzianum,* F. *Microdochium oryzae* and *Aspergillus niger,* G. *Pestalotiopsis guepinii* and *Trichoderma viride,* H. *Sarocladium oryzae* and *Trichoderma harzianum.*

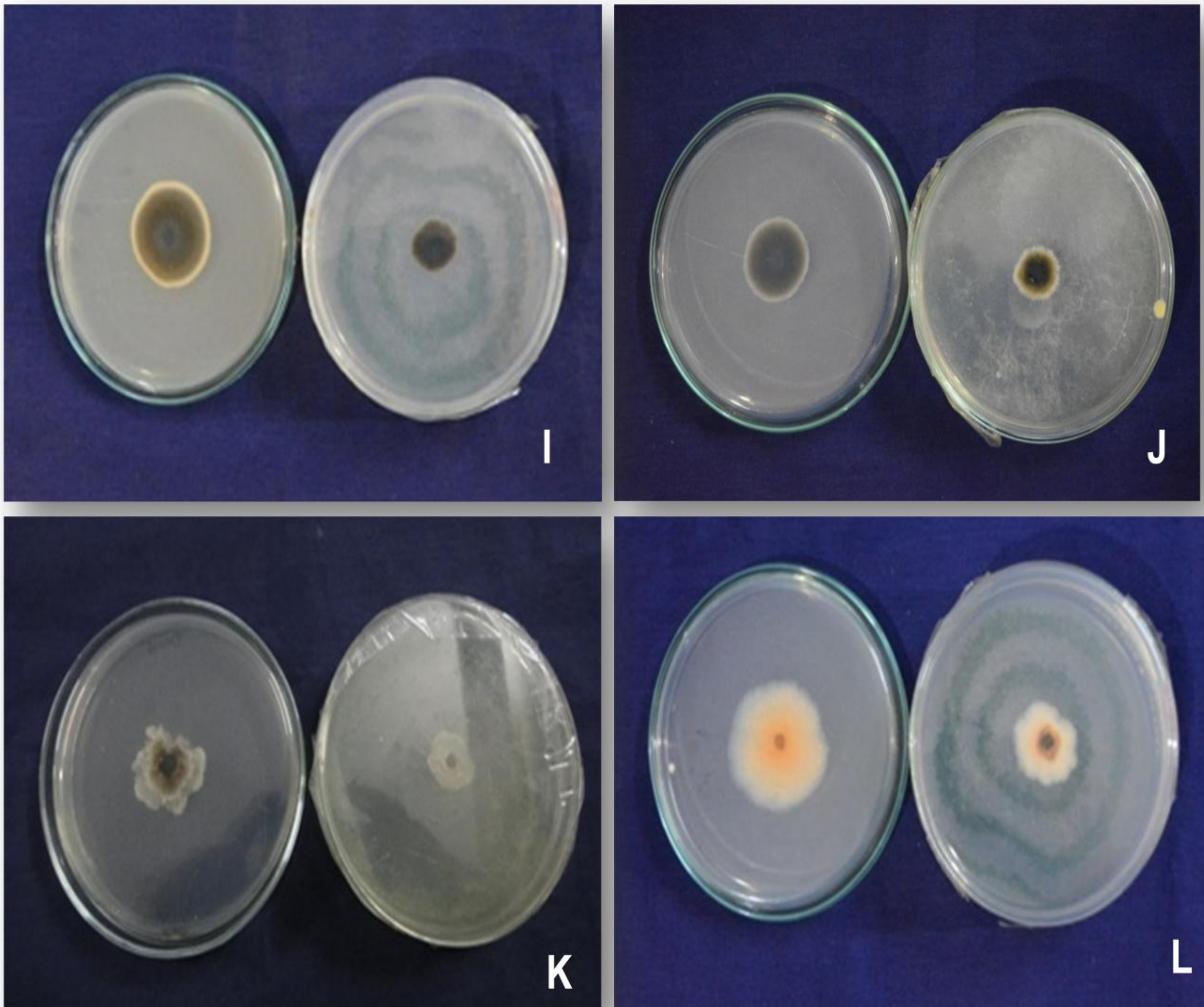

Plate 8. Per cent inhibition owing to volatile substances between I. *Alternaria alternata* and *Trichoderma viride,* J. *Curvularia lunata* and *Trichoderma harzianum, K. Drechslera oryzae* and *Aspergillus niger,* L. *Fusarium moniliforme* and *Trichoderma viride.*

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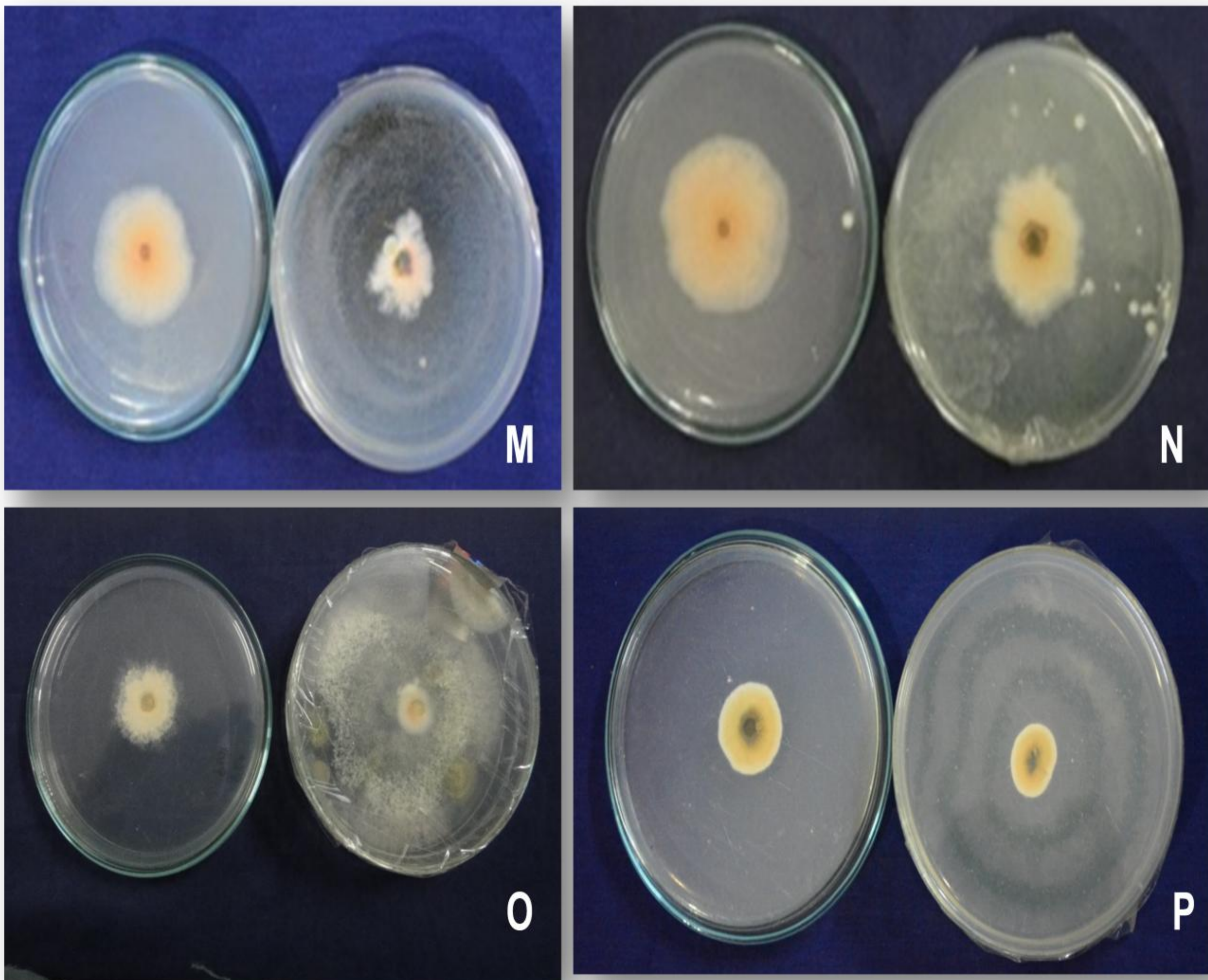

Plate 9. Per cent inhibition owing to volatile substances between M. *Fusarium solani* and *Aspergillus niger,* N. *Microdochium oryzae* and *Trichoderma harzianum,* O. *Pestalotiopsis guepinii* and *Trichoderma harzianum,* P. *Sarocladium oryzae* and *Trichoderma viride.*

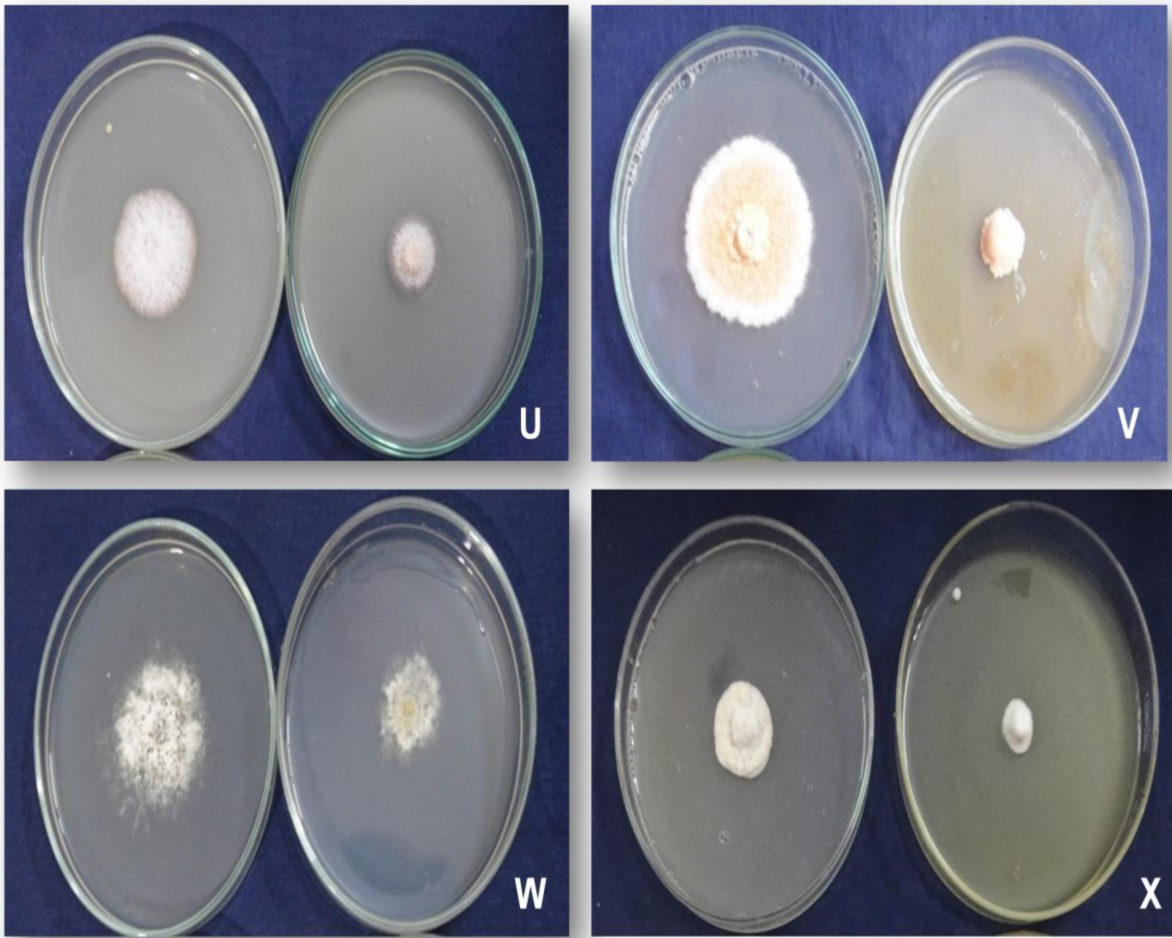

Plate 10. Per cent inhibition owing to non volatile metabolites at 10% cocentrations Q. *Alternaria alternata* and *Trichoderma harzianum,* R. *Curvularia lunata* and *Trichoderma harzianum,* S. *Drechslera oryzae* and *Aspergillus niger,* T. *Fusarium moniliforme* and *Trichoderma harzianum.*

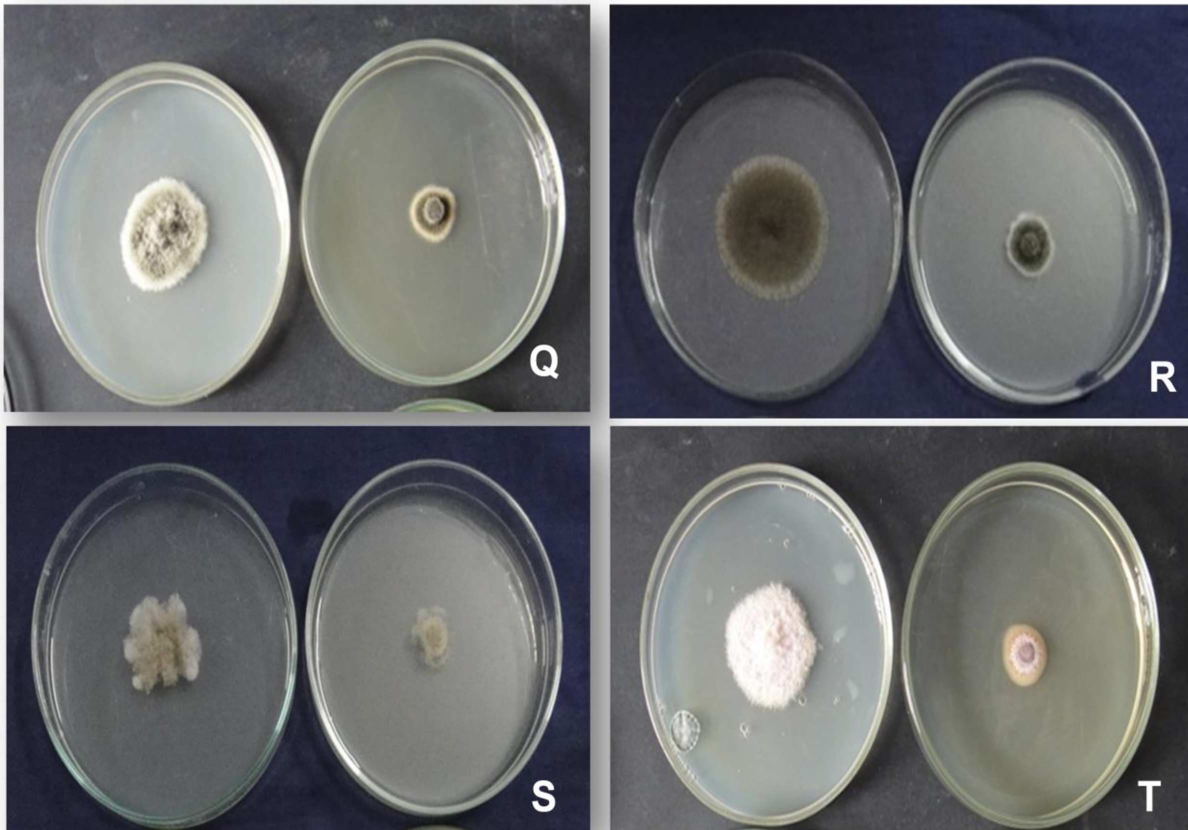

Plate 11. Per cent inhibition owing to non volatile metabolites between U) *Fusarium solani* and *Trichoderma viride,* V. *Microdochium oryzae* and *Trichoderma harzianum,* W. *Pestalotiopsis guepinii and Trichoderma harzianum,* X. *Sarocladium oryzae* and *Trichoderma harzianum* at 10% concentrations.

4.15.3. Effect of non-volatile substances of the soil fungi on the radial growth of the test pathogens

Non volatile substances of the soil fungi showed inhibition of mycelial growth against the test pathogen ranges from 29.05 to 64.5%. The highest inhibition was observed by the culture filtrate of *T. harzianum* against *C. lunata* followed *by M. oryzae* (61.25%), *A. alternata* (60.50%) and *F. moniliforme* (53.55%). The lowest inhibition was observed by the culture filtrate of *A. fumigatus* against *Pestalotipsis guepinii* (28.18%) (Table 29, Plate 10 & 11). These results showed similarity with the findings of Akter *et al.* (2014) and Bashar and Chakma (2014). Akter *et al.* (2014) reported that non-volatile metabolites of *A. flavus, A. fumigatus, A. niger, T. harzianum* and *T. viride* inhibited the maximum radial growth of *Colletotricum* sp. *C. lunata, F. moniliforme, F. semitectum* and *F. oxysporum.* In case of *F. oxysporum*, Bashar and Chakma (2014) also reported 82% inhibition of growth at 20% concentration owing to non-volatile metabolites.

Amongst six soil fungi only *Trichoderma harzianum* exhibited strong antagonistic effect against all the test pathogens. This effect because of its first growing nature, rapid sporulation and toxin producing capacity. It is known to be capable of producing antibiotics which might have suppresed the growth of the test pathogens. These results are in aggrement with the findings of Adriana and Sergio (2001), Krupke *et al.* (2003), Kexiang *et al.* (2002) and Shafiquzzaman *et al.* (2009), Skidmore and Dickinson (1976). This observation suggests that *T. harzianum* may be exploited commercially as a bio-control agent of seed borne pathogens of rice.

4.16. Integrated management of test pathogens *in vivo* (Fig. 13-16)

4.16.1. Combined effect of fungicides, plant extracts and antagonist on seed quality parameters of BRRI 29 rice variety against *Alternaria alternata*, *Aspergillus flavus* and *Curvularia lunata*

Due to combined effect of different treatments with fungicides, leaf extracts and antagonist on seed quality parameters of BRRI 29 rice variety against test pathogens *A. alternata*, *A. flavus and C. lunata* T6 (Bavistin + *Azadirachta indica* + *Trichoderma harzianum*) showed highest per cent of seed grrmination and seedling vigor index amongst 13 treatments. Next to T6, T11 (Bavistin + Tall + *Azadirachta indica* + *Citrus medica* + *Trichoderma harzianum*) and T10 (Bavistin + Tall + *Azadirachta indica* + *Citrus medica*) showed best result repectively (Table 30).

4.16.2. Combined effect of fungicides, plant extracts and antagonist on seed quality parameters of BRRI 29 rice variety against *Drechslera oryzae, Fusarium moniliforme* and *Fusarium solani*

Inoculated seeds with *D. oryzae* and *F. moniliforme* showed highest per cent (74% and75%) of seed germination respectively due to combined effect of T10 (Bavistin+Tall+*Azadirachta indica*+*Citrus medica*). Next to T10, T11 (Bavistin+Tall+ *Azadirachta indica*+*Citrus medica*+*Trichoderma harzianum*) showed best result followed by T6 (Bavistin+*Azadirachta indica*+*Trichoderma harzianum*). Seeds inoculated with *F. solani* showed highest per cent of seed germination due to T3 treatment followed by T8 and T6 treatment (Table 31).

4.16.3. Combined effect of fungicides, plant extracts and antagonist on seed quality parameters of BRRI 29 rice variety against *Microdochium oryzae, Pestalotiopsis guepinii* and *Sarocladium oryzae*

Seeds inoculated with *M. oryzae* and *S. oryzae* showed highest per cent of germination due to combined effect of T10 *viz.* (Bavistin+Tall+*Azadirachta indica+C. medica*) which showed highest per cent (82 and 84%) of seed grrmination and seedling vigor index amongst 13 treatments. Next to T10, T11 (Bavistin+Tall+*Azadirachta indica+Citrus medica+Trichoderma harzianum*) and T6 (Bavistin+*Azadirachta indica+Trichoderma harzianum*) showed best result in both cases (Table 32). Seeds inoculated with *P. guepinii* showed highest per cent germination due to combined effect of T6 treatment followed by T3 and T8 treatments, respectively (Table 32). The results of integrated management of this investigation showed similarity with the findings of Hossain and Mia (2001), Ashrafuzzaman *et al*. (2011) and Islam and Monjil (2016). Hossain and Mia observed two foliar sprays with Aimcozim, Bavistin, Shincar and Tilt at 0.1% and two top dressing of MP with 40kg/ha caused significant reduction of tiller infection of sheath blight disease. They also observed that Tilt was the best fungicide for controlling the disease. Ashrafuzzaman *et al*. (2011) reported the integratred managemenmt of sheath blight of aman rice. A total of 13 treatment combinations including controls with or without inocula of the pathogen were tested. Severity increased with the increasing maturity of rice plants under all the treatments. The development was significantly least in plants treated with combined doses. The highest yield of rice increase was recorded in plants tested with the combined doses. Islam and Monjil (2016) observed complete inhibition of sheath blight pathogen when treated with four indegineous medicinal plant extracts i.e. tulsi, nishinda, thankuni and biskatali. He also reported that germination failure must have also been caused due to fungal infections.

Fig.13. Effects of treatments with fungicides and plant extract.

T1. Bavistin,

T2. Tall and

T3. *Azadirachta indica.*

Fig.14. Combined effects of treatments with fungicides, plant extracts and antagonist.

T4. *Citrus medica*,

T5. *Trichoderma harzianum*,

T6. Bavistin + *Azadirachta indica* + *Trichoderma harzianum*,

T7. Bavistin + *Citrus medica* + *Trichoderma harzianum.*

Fig.15. Combined effects of treatments with fungicides, plant extracts and antagonist,

T8. Tall + *Azadirachta indica + Trichoderma harzianum,*

T9. Tall + *Citrus medica + Trichoderma harzianum*,

T10. Bavistin + Tall + *Azadirachta indica + Citrus medica.*

Fig. 16. Combined effects of treatments with fungicides, plant extracts and antagonist.

T11. Bavistin + Tall + *Azadirachta indica* + *Citrus medica* + *Trichoderma harzianum*,

T12. Control with inocula,

T13. Control without inocula

Table 30. Effect of different treatments with fungicides, leaf extracts and antagonist on seed quality parameters of BRRI 29 rice variety

Treatments	Seed quality parameters against test pathogens														
	Alternaria alternate					*Aspergillus flavus*					*Curvularia lunata*				
	A	B	C	D	E	A	B	C	D	E	A	B	C	D	E
T1	79 c	8.00 dc	2.9 cd	13.0 cd	1256.1 cd	78 cd	10.25 bc	2.90 e	13.0 cd	1240.2 cd	69 def	8.33 d	3.08 de	13.2ab	1123.32 cd
T2	80 c	9.00 c	3.50 a	12.5 de	1280 cd	76 d	8.10 de	3.50 bc	12.5 d	1232 d	65 def	9.09 c	3.2 cde	13.0 cd	1053 bcd
T3	85 b	8.33 d	3.0 bcd	13.0 cd	1360 bc	81 ab	9.87 bcd	3.20 cde	13.0 cd	1312.2 c	72 cde	8.01d	2.50d	14.0 b	1188 bcd
T4	74 e	7.69 ef	3.1 bcd	12.0 e	1117.4 e	72 e	12.5 a	3.00 de	13.5 bcd	1188 d	64 f	9.37 c	2.60c	13.0 cd	998.4 de
T5	72 f	7.84 e	2.90 cd	12.0 e	1072.8 e	62 f	9.67 bcd	3.25 cd	14.0 abc	1069.5 e	62 f	7.69 e	3.25 cd	15.0 ab	1131.5 cd
T6	**87 a**	9.00 c	3.2 abc	14.0 ab	**1496.4 a**	**82 a**	10.97 ab	3.50 bc	15.0 a	**1517a**	**83 a**	9.30 c	3.24 cd	16.0 a	**1596.92 a**
T7	85 b	8.00 de	3.3 ab	13.3 bcd	1411 ab	81 ab	11.11 ab	3.25 cd	14.5 ab	1437.75 ab	72 cde	11.11 a	3.25 cd	14.0 b	1242 bc
T8	77 d	7.40 f	2.80 d	13.8 abc	1278.2 cd	79 bc	10.12 bc	3.80 ab	14.0 abc	1406.2 b	75 bcd	10.66 b	3.01 d	15.0 ab	1350.75 ab
T9	75 e	5.60 g	3.0 bcd	13.9 abc	1267.5cd	77 cd	9.09 cd	3.90 a	14.2 abc	1393.7 b	75 bcd	10.81 b	3.50 bc	14.5 ab	1350 ab
T10	80 c	9.70 b	2.80 d	14.0 ab	1344 bcd	82 a	7.31 e	3.40 c	14.5 ab	1467.8 ab	82 ab	11.63 a	3.80 ab	13.0 cd	1377.6 ab
T11	81 c	8.77 c	3.50 a	14.5 a	1458 a	81 ab	9.87 bcd	3.50 bc	15.0 a	1498.5 a	80 abc	10.01 b	3.90 b	13.5 c	1392 ab
T12	70 g	10.2 a	2.90 cd	13.0 cd	1113 e	70 e	8.97 cde	3.40 c	14.0 abc	1218 d	54 g	9.25 c	3.40 c	12.9 d	880.2 e
T13	74 e	7.69 ef	3.0 bcd	13.8 abc	1243.2 d	72 e	8.75 cde	3.50 bc	13.5 bcd	1224 d	64 f	8.25 d	4.25 a	13.75 c	1152 cde
CV%	1.43	2.58	6.67	3.82	4.36	1.77	9.58	5.37	5.01	3.62	4.69	17.12	13.10	11.07	9.27

T1: Bavistin, T2: Tall, T3: *Azadirachta indica*, T4: *Citrus medica*, T5: *Trichoderma harzianum*, T6: Bavistin + *Azadirachta indica* + *Trichoderma harzianum*, T7: Bavistin + *Citrus medica* + *Trichoderma harzianum*, T8: Tall + *Azadirachta indica* + *Trichoderma harzianum*, T9: Tall + *Citrus medica* + *Trichoderma harzianum*, T10: Bavistin + Tall + *Azadirachta indica* + *Citrus medica*, T11: Bavistin + Tall + *Azadirachta indica* + *Citrus medica* + *Trichoderma harzianum*, T12: Control with inocula and T13: Control without inocula; A: % germination, B: % mortality, C: Root length (cm), D: Shoot length (cm) and E: Seedling vigor index.

Values within the same column with a common letter (s) do not differ significantly at 5% level by DMRT.

Table 31. Effect of different treatments with fungicides, leaf extracts and antagonist on seed quality parameters of BRRI 29 rice variety

Treatments	Seed quality parameters against test pathogens														
	Drechslera oryzae					*Fusarium moniliforme*					*Fusarium solani*				
	A	B	C	D	E	A	B	C	D	E	A	B	C	D	E
T1	72 b	8.33	3.92	11.25	1092.22 de	72 b	9.09	3.1	10.5	979.2 de	80 a	10.01	3.2	13	1296 a
T2	74 a	9.09	3.95	12.5	1217.3 b	76 a	11.76	3.2	9.15	938.6 efg	70 de	11.43	3.6	12.9	1155 b
T3	65 e	8.33	3.64	9.88	878.8 h	70 b	14.29	3.5	10.15	924 ef	**81 a**	9.88	3.5	13.3	**1360.8 a**
T4	60 i	11.25	2.95	11.25	852.0 h	67 c	10.44	2.9	10.25	815.3 gh	75 bc	10.66	2.8	12.2	1125 b
T5	62 h	12.0	3.20	12.0	942.4 g	62 d	12.9	3.01	10.5	837 h	65 f	12.31	2.8	12.5	994.5 cd
T6	70 c	10.5	3.85	12.5	1144.5 c	72 b	11.11	3.25	11.25	1044 bc	78 ab	10.26	3.2	11.75	1166.1 b
T7	64 ef	9.50	3.15	10.25	857.6 h	70 b	11.42	3.5	12	1085 ab	77 ab	10.39	3.01	12.1	1162.7 b
T8	68 d	8.50	3.10	13.25	1111.8 cd	75 a	10.66	3.8	11.25	1128 a	79 ab	10.13	3.3	11.5	1169.2 b
T9	63 fg	12.25	3.30	13.55	1061.55 c	72 b	11.11	3.6	10.5	1015.2 cd	70 de	11.43	3.01	12	1050 c
T10	**74 a**	13.25	3.80	14.0	**1281.6 a**	**75 a**	10.66	3.7	11.5	**1140 a**	66 ef	12.12	2.99	11.5	950.4 d
T11	70 c	14.0	3.90	13.88	1181.6 ab	74 a	10.81	3.6	11.4	1110 a	68 def	11.76	3.2	12.2	1047.2 c
T12	65 e	12.0	4.15	11.25	1001 f	62 d	12.9	3.25	9.3	778.1 I	56 g	14.28	2.9	10.5	750.4 e
T13	70 c	10.5	3.90	11.5	1078 de	68 c	11.42	3.3	10.01	904 fg	72 cd	11.11	3.5	12.2	1130.4 b
CV%	0.006	.0083	.0048	.0072	0.9224	1.72	3.52	.001	.056	0.9828	3.53	16.98	16.53	8.06	6.51

T1: Bavistin, T2: Tall, T3: *Azadirachta indica*, T4: *Citrus medica*, T5: *Trichoderma harzianum*, T6: Bavistin + *Azadirachta indica* + *Trichoderma harzianum*, T7: Bavistin + *Citrus medica* + *Trichoderma harzianum*, T8: Tall + *Azadirachta indica* + *Trichoderma harzianum*, T9: Tall + *Citrus medica* + *Trichoderma harzianum*, T10: Bavistin + Tall + *Azadirachta indica* + *Citrus medica*, T11: Bavistin + Tall + *Azadirachta indica* + *Citrus medica* + *Trichoderma harzianum*, T12: Control with inocula and T13: Control without inocula; A: % germination, B: % mortality, C: Root length (cm), D: Shoot length (cm) and E: Seedling vigor index

Values within the same column with a common letter (s) do not differ significantly at 5% level by DMRT.

Table 32. Effect of different treatments with fungicides, leaf extracts and antagonists on seed quality parameters of BRRI 29 rice variety

Treatments	Seed quality parameters against test pathogens.														
	Microdochium oryzae					*Pestalotiopsis guepinii*					*Sarocladium oryzae*				
	A	B	C	D	E	A	B	C	D	E	A	B	C	D	E
T1	69 e	11.59 c	4.2	14.5	1290.3 d	64 f	12.5 ab	3.2 abc	12.5 bc	1004.8 c	72 de	11.11	3.5	16.5 a	1440 abc
T2	64 f	12.5 b	4.1	15	1222.4 e	68 e	11.76 c	3.5 a	13 abc	1122 d	78abc	10.25	4.01	15.5 ab	1521.78 ab
T3	71 c	11.26 c	4.3	15.5	1405.8 c	85 a	11.62 c	2.99 bcd	14 a	1444.15 ab	81 ab	9.88	4.1	14.6 bc	1514.7 a
T4	62 g	12.90 b	4.01	15.25	1194.12 e	65 f	12.3 b	3.01bcd	12.5 bc	1008.15 c	74 cde	10.81	3.8	14.3 bc	1339.4 abc
T5	61 g	13.11 a	3.9	14.5	1122.4 f	60 g	13.33 a	2.8 d	12.0 c	888 f	78 abcd	10.26	3.9	14.5 bc	1435.2 abc
T6	82 a	9.75 f	4.1	15.2	1582.6 a	**85 a**	11.76 c	3.5 a	14.0 a	**1487.5 a**	82 ab	9.76	4.2	14.6 bc	1541.6 a
T7	81 ab	9.87 f	4.1	14.2	1482.3 b	78 d	10.26 d	3.25 abc	13.5 ab	1306.5 c	73 cde	10.96	3.7	13.9 c	1284.8 cde
T8	78 bc	10.26 d	3.9	14.5	1435.2 bc	80 cd	12.5 ab	3.2 abc	14.0 a	1376 abc	76 bcde	10.53	3.6	13.8 c	1322.4 bcd
T9	75 c	10.66 d	3.8	14.8	1395 c	82 bc	12.2 b	3.3 ab	13 abc	1336.6 bc	79 abc	10.13	3.94	14 c	1417.26 abc
T10	**82 a**	9.88 e	3.9	15.5	**1590.8 a**	85 a	11.76 c	3.3 ab	13.5 ab	1428 ab	**84 a**	9.52	4.01	14.4 bc	**1546.44 a**
T11	80 b	10.00 e	4.01	15.5	1560.8 a	84 ab	11.9 bc	3.2 abc	13.4 ab	1394.4 abc	79 abc	10.13	3.8	14.3 bc	1429.9 abc
T12	58 h	13.79 a	3.2	14.5	1026.6 g	52 h	9.61 e	2.9 cd	12.0 c	774.8 g	64 f	12.5	3.5	14.5 bc	1152 e
T13	60 g	13.33 a	3.8	15	1128 f	60g	11.76 c	3.01 bcd	13.5 ab	990 ef	70 ef	11.43	3.7	13.8 c	1225 de
CV%	1.53	0.895	0.764	1.02	1.38	1.79	9.58	3.89	2.68	2.02	3.53	18.98	16.53	8.06	6.51

T1: Bavistin, T2: Tall, T3: *Azadirachta indica*, T4: *Citrus medica*, T5: *Trichoderma harzianum*, T6: Bavistin + *Azadirachta indica* + *Trichoderma harzianum*, T7: Bavistin + *Citrus medica* + *Trichoderma harzianum*, T8: Tall + *Azadirachta indica* + *Trichoderma harzianum*, T9: Tall + *Citrus medica* + *Trichoderma harzianum*, T10: Bavistin + Tall + *Azadirachta indica* + *Citrus medica*, T11: Bavistin + Tall + *Azadirachta indica* + *Citrus medica* + *Trichoderma harzianum*, T12: Control with inocula and T13: Control without inocula; A: % germination, B: % mortality, C: Root length (cm), D: Shoot length (cm) and E: Seedling vigor index.

Values within the same column with a common letter (s) do not differ significantly at 5% level by DMRT.

Almost all the treatments were significantly increased germination over control plants. The highest germination was recorded in plants grown in pots where the integrated doses of Bavistin , *Azadirachta indica* and *Trichoderma harzianum* were applied followed by plants grown in pots amended with the integrated doses of Bavistin, Tall, *Azadirachta indica* and *Trichoderma harzianum.* Considering the overall performance of the treatments, the integrated use of Bavistin, *Azadirachta indica* and *Trichoderma harzianum* or the integrated use of Bavistin, Tall, *Azadirachta indica* and *Trichoderma harzianum* showed the better performance for reduction of test pathogens and increased germination considerably. These integrated treatments might be useful for the management of rice pathogens for increasing germination.

CHAPTER 5

SUMMARY

Studies on diseased rice grains of four commercially cultivated rice varieties namely BRRI 28, 29, Kalijira and Pajam collected from 14 districts of 7 divisions and 40 rice samples *viz*. Hybrid 2, 3, 4, BR 7, 11, 12, 14 ,16, 22, 23, 25, 26 and BRRI 28 to BRRI 55 were collected from Bangladesh Rice Research Institute at Joydebpur. Quality analysis showed that the percentage of pure seed varies from 94-99%. The germination and mortality of different varieties were in the range of 62-100% and 10-30%, respectively. Twenty five species of fungi belonging to 15 genera were associated with these rice varieties. The isolated fungi were *Alternaria alternata*, *Aspergillus clavatus*, *A. flavus, A. fumigatus, A. niger, A. ochraceous A. oryzae, A. terreus, Chaetomium globosum, Cladosporium cladosporioides, Colletotrichum gloeosporioides, Curvularia lunata*, *C. lunata* var. *aeria*, *Drechslera oryzae*, *Fusarium moniliforme, F. oxysporum, F. solani, Microdochium oryzae, Nigrospora oryzae, Penicillium* spp., *Pestalotiopsis guepinii*, *Rhizopus stolonifer*, *Sarocladium oryzae* and *Trichoderma viride.*

The pathogenicity test proved that nine fungi *viz*., *Alternaria alternata* (Fr.) Keissler, *Aspergillus flavus* Link *Curvularia lunata* (Wakker) Boedijn, *Drechslera oryzae* Breda de Haan (Subramanian and Jain), *Fusarium moniliforme* Sheldon, *F. solani* (Mart.) Sacc. *Microdochium oryzae* (Hashloka and Yokogi) Sam. and Hal., *Pestalotiopsis guepinii* (Desm.) Stay. and *Sarocladium oryzae* (Sawada) W. Gams and D. Hawks were found to be pathogenic. Other fungi were found to be non pathogenic.

The incident of different fungi varied among the seasons. During Boro season 23 species of fungi belonging to 13 genera were found to be associated with 11 varieties of Boro rice. Frequency

(%) was highest in *Drechslera oryzae* and lowest in *Pestalotiopsis guepinii*. Both were isolated from BRRI 29. *Pestalotiopsis guepinii* was isolated first time from rice grain.

In the Aus season 11 species of fungi belonging to 8 genera were obtained from seven rice varieties. Highest frequency was observed in *Rhizopus stolonifer* and lowest in both *Sarocladium oryzae* and *Curvularia lunata*.

During T. Aman season the numbers of isolated fungi were 15 species belonging to 11 genera from 22 varieties of affected rice grains. Highest frequency was observed in *Aspergillus niger* and lowest frequency in *Pestalotiopsis guepinii*. After 6 months of storage at 25±2°C.the pathogenic fungi were decreased but the storage fungi were increased.

Fifty six rice seed samples representing over four rice varieties collected from 14 different districts of Bangladesh under Barisal, Chittagong, Dhaka, Khulna, Rajshahi, Rangpur and Sylhet divisions. A total of 23 species of fungi belonging to 15 genera were isolated from these rice samples. The most predominating fungus detected from the grains was *Drechslera oryzae* .Highest frequency in *D. oryzae* from Rajshahi division and lowest frequency in *Sarocladium oryzae* from Barisal division.

From the results, it appears that predominating fungi associated with grains were *D. oryzae, A. niger, A. flavus, Rhizopus stolonifer* and *Penicilium* sp. Among three seasons frequency of association of fungi was slightly higher in Boro season compared to Aus and T. Aman season. In T. Aman the most frequently occurring fungi were *A. niger, A. flavus, C. lunata,* and *D. oryzae.*

Overall in the present study it was observed that from ninety six seed samples, the most predominant was *Drechslera oryzae*, which was associated with 62.5% seed samples and the least incidence, was found in *P. guepinii* and *S. oryzae* which was associated with 2.08% seed samples.

Among ten fungicides i.e. Bavistin 50 WP, Capvit 50 WP, Dithane M-45, Greengel, Hayvit 80 WP, Indofil M-45, Ridomil MZ Gold, Salcox 50 WP, MC Sulphur 80 and Tall 25 EC, only Tall 25 EC completely inhibited the radial growth of the test fungi at all the tested concentrations except *Fusarium moniliforme and Microdochium oryzae.*

Antifungal properties of all the plants *viz. Allium sativum* L., *Artocarpus heterophyllus* Lamk., *Asparagus racemosus* Willd., *Azadirachta indica* A. Juss., *Citrus medica* L., *Datura metel* L., *Mangifera indica* L., *Nerium indicum* Mill., *Senna alata* (L.) Roxb. and *Tagetes erecta* L completely inhibited the radial growth of the test fungi at 20% concentration except *Asparagus racemosus* and *Nerium indicum.*

Antagonistic potential of selected six soil fungi were evaluated against pathogenic fungi. Amongst 48 interactions, grade 3 was found in 22 interactions. Volatile and non-volatile substances from *T. harzianum* inhibited highest radial growth of the test pathogens. In colony interaction, the highest growth inhibition was also observed due to *T. harzianum* against all the pathogens.

In field experiment out of 13 treatments, T6 (Bavistin +*Azadirachta indica*+*Trichoderma harzianum*) and T10 (Bavistin +Tall +*Azadirachta indica*+*Citrus medica*) treated seeds showed highest per cent of germination and seedling vigor index against all test pathogens.

CHAPTER 6

CONCLUSION AND RECOMMENDATIONS

Based on the findings of the present investigation the following conclusions are drawn:

- Association of 25 species of fungi with 42 rice varieties during 2012 to 2014.
- Three rice varieties i.e. BRRI 28, 29 and Pajam were susceptible.
- Association of *Microdochium oryzae* with rice grains of BRRI 34, 41, 51, 52, 54 in Bangladesh.
- Association of *Pestalotiopsis guepinii* with rice grains is a new record for Bangladesh.
- *Drechslera oryzae* was frequently associated with seeds of different rice varieties in all the seasons.
- After six months of storage prevalence of storage mold i.e., *Aspergillus niger* and *Rhizopus stolonifer* were highest and pathogenic field fungi gradually decreased.
- *Aspergillus niger, Trichoderma harzianum* and *T. viride* showed promising inhibitory effect on the growth of the test pathogens.
- Bavistin 50 WP and Tall 25 EC identified as the best inhibiting chemical fungicides against pathogenic fungi of rice.
- *Azadirachta indica* and *Citrus medica* showed complete inhibition of test pathogens.
- *In vivo* experiment out of 13 treatments, T6 (Bavistin + *Azadirachta indica* + *Trichoderma harzianum*) and T10 (Bavistin + Tall + *Azadirachta indica* + *Citrus medica*) showed highest seed germination, seedling vigor index against *Alternaria alternata, Aspergillus flavus, Curvularia lunata, Pestalotiopsis guepinii* and *Drechslera oryzae, Fusarium moniliforme, Microdochium oryzae, Sarocladium oryzae***,** respectively.

- T3 treatment (Bavistin + Tall) showed highest seedling vigor index against *Fusarium solani.*
- Amongst all treatments T6 and T10 showed significant result over control.

Recommendations

- Since rice a staple food, better seed health management is a prerequisite for successful rice production because pathogenic fungi are known to cause huge economic losses by reducing rice yield.
- Application of Bavistin 50 WP and Tall 25 EC at 300 and 100 ppm concentrations respectively may be commercially used for managing pathogens of rice seeds.
- For more confirmation the above mentioned fungicides also need to 2-3 years trial in nursery bed and in field condition.
- In small scale, *Azadirachta indica* and *Citrus medica* at 10% concentration can be used for controlling diseases and production of healthy seeds.
- *Trichoderma harzianum* may be exploited commercially as a bio-control agent against pathogens of rice.
- Combined application of Bavistin, Tall, *Azadirachta indica, Citrus medica* and *Trichoderma harzianum* in seed treatments may be used commercially to control rice pathogens. For more confirmation the above mentioned treatments also need to test 2-3 years in field condition.
- Findings of this research work will be helpful for designing a proper management of pathogenic fungi of rice.

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APPENDICES

Appendix 1. ANOVA of transmission of pathogenic fungi from seed to seedlings in pot experiments

Table 1A. Germination percentage of inoculated seeds

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Test pathogens	9	1518.3000	168.7000	76.68	0.0000
Error	20	44.0000	2.2000		
Total	29	1562.3000			

CV(%) 2.37

Table 1B.Mortality percentage of inoculated seeds

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Test pathogens	9	271.2074	30.1342	19.87	0.0000
Error	20	30.3376	1.5169		
Total	29	301.5450			

CV(%) 9.21

Table 1C.Percentage of seed to seedling transmission of pathogen

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Test pathogens	9	1389.8856	154.4317	188.59	0.0000
Error	20	16.3775	0.8189		
Total	29	1406.2631			

CV(%) 5.70

Appendix 2. ANOVA for fungitoxicity of fungicides against *Alternaria alternata*

Per cent inhibition of radial growth at 100 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Name	9	12170.8851	1352.3206	764.87	0.0000
Error	20	35.3608	1.7680		
Total	29	12206.2459			

CV(%) 2.45

Per cent inhibition of radial growth at 200 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Name	9	8237.8644	915.3183	127.05	0.0000
Error	20	144.0836	7.2042		
Total	29	8381.9480			

CV(%) 4.23

Per cent inhibition of radial growth at 300 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Name	9	7620.9739	846.7749	205.86	0.0000
Error	20	82.2652	4.1133		
Total	29	7703.2391			

CV(%) 2.79

Per cent inhibition of radial growth at 400 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Name	9	9351.0917	1039.0102	196.04	0.0000
Error	20	106.0000	5.3000		
Total	29	9457.0917			

CV(%) 2.77

Per cent inhibition of radial growth at 500 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Name	9	6680.8801	742.3200	463.95	0.0000
Error	20	32.0000	1.6000		
Total	29	6712.8801			

CV(%) 1.43

Appendix 3. ANOVA for fungitoxicity of fungicides against *Aspergillus flavus*

Per cent inhibition of radial growth at 100 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	33743.9268	3749.3252	1874.66	0.0000
Error	20	40.0000	2.0000		
Total	29	33783.9268			

CV(%) 3.43

Per cent inhibition of radial growth at 200 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	25829.0526	2869.8947	667.42	0.0000
Error	20	86.0000	4.3000		
Total	29	25915.0526			

CV(%) 4.13

Per cent inhibition of radial growth at 300 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	28570.2845	3174.4761	1587.24	0.0000
Error	20	40.0000	2.0000		
Total	29	28610.2845			

CV(%) 2.16

Per cent inhibition of radial growth at 400 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	26927.2339	2991.9149	1068.54	0.0000
Error	20	56.0000	2.8000		
Total	29	26983.2339			

CV(%) 2.16

Per cent inhibition of radial growth at 500 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	24480.0000	2720.0000	2266.67	0.0000
Error	20	24.0000	1.2000		
Total	29	24504.0000			

CV(%) 1.34

Appendix 4. ANOVA for fungitoxicity of fungicides against *Curvularia lunata*

Per cent inhibition of radial growth at 100 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	10012.6730	1112.5192	173.83	0.0000
Error	20	128.0000	6.4000		
Total	29	10140.6730			

CV(%) 4.71

Per cent inhibition of radial growth at 200 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	6572.5787	730.2865	270.48	0.0000
Error	20	54.0000	2.7000		
Total	29	6626.5787			

CV(%) 2.60

Per cent inhibition of radial growth at 300 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	5139.2003	571.0223	211.39	0.0000
Error	20	54.0267	2.7013		
Total	29	5193.2270			

CV(%) 2.40

Per cent inhibition of radial growth at 400 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	3442.8511	382.5390	98.09	0.0000
Error	20	78.0000	3.9000		
Total	29	3520.8511			

CV(%) 2.64

Per cent inhibition of radial growth at 500 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	3155.9289	350.6588	116.89	0.0000
Error	20	60.0000	3.0000		
Total	29	3215.9289			

CV(%) 2.04

Appendix 5. ANOVA for fungitoxicity of fungicides against *Drechslera oryzae*

Per cent inhibition of radial growth at 100 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	15808.7904	1756.5323	177.43	0.0000
Error	20	198.0000	9.9000		
Total	29	16006.7904			

CV(%) 7.31

Per cent inhibition of radial growth at 200 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	8998.2841	999.8093	175.41	0.0000
Error	20	114.0000	5.7000		
Total	29	9112.2841			

CV(%) 4.25

Per cent inhibition of radial growth at 300 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	6508.6908	723.1879	157.21	0.0000
Error	20	92.0000	4.6000		
Total	29	6600.6908			

CV(%) 2.91

Per cent inhibition of radial growth at 400 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	6967.9205	774.2134	387.11	0.0000
Error	20	40.0000	2.0000		
Total	29	7007.9205			

CV(%) 1.63

Per cent inhibition of radial growth at 500 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	6967.9205	774.2134	387.11	0.0000
Error	20	40.0000	2.0000		
Total	29	7007.9205			

CV(%) 1.63

Appendix 6. ANOVA for fungitoxicity of fungicides against *Fusarium moniliforme*

Per cent inhibition of radial growth at 100 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	19078.9992	2119.8888	731.00	0.0000
Error	20	58.0000	2.9000		
Total	29	19136.9992			

CV(%) 5.67

Per cent inhibition of radial growth at 200 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	15571.7203	1730.1911	494.34	0.0000
Error	20	70.0000	3.5000		
Total	29	15641.7203			

CV(%) 4.34

Per cent inhibition of radial growth at 300 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	22329.0587	2481.0065	1378.34	0.0000
Error	20	36.0000	1.8000		
Total	29	22365.0587			

CV(%) 1.95

Per cent inhibition of radial growth at 400 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	19756.2720	2195.1413	954.41	0.0000
Error	20	46.0000	2.3000		
Total	29	19802.2720			

CV(%) 2.00

Per cent inhibition of radial growth at 500 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	12061.8533	1340.2059	1489.12	0.0000
Error	20	18.0000	0.9000		
Total	29	12079.8533			

CV(%) 1.09

Appendix 7. ANOVA for fungitoxicity of fungicides against *Fusarium solani*

Per cent inhibition of radial growth at 100 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	18834.1152	2092.6795	740.70	0.0000
Error	20	56.5054	2.8253		
Total	29	18890.6206			

CV(%) 5.38

Per cent inhibition of radial growth at 200 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	14880.3454	1653.3717	177.54	0.0000
Error	20	186.2543	9.3127		
Total	29	15066.5997			

CV(%) 6.88

Per cent inhibition of radial growth at 300 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	16655.0491	1850.5610	656.68	0.0000
Error	20	56.3608	2.8180		
Total	29	16711.4099			

CV(%) 2.73

Per cent inhibition of radial growth at 400 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	18505.8900	2056.2100	1468.72	0.0000
Error	20	28.0000	1.4000		
Total	29	18533.8900			

CV(%) 1.54

Per cent inhibition of radial growth at 500 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	10700.3333	1188.9259	566.16	0.0000
Error	20	42.0000	2.1000		
Total	29	10742.3333			

CV(%) 1.64

Appendix 8. ANOVA for fungitoxicity of fungicides against *Microdochium oryzae*

Per cent inhibition of radial growth at 100 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	25672.5883	2852.5098	1057.99	0.0000
Error	20	53.9232	2.6962		
Total	29	25726.5115			

CV(%) 3.82

Per cent inhibition of radial growth at 200 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	22759.8876	2528.8764	847.96	0.0000
Error	20	59.6458	2.9823		
Total	29	22819.5334			

CV(%) 2.75

Per cent inhibition of radial growth at 300 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	10670.1203	1185.5689	564.56	0.0000
Error	20	42.0000	2.1000		
Total	29	10712.1203			

CV(%) 1.79

Per cent inhibition of radial growth at 400 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	6907.5000	767.5000	852.78	0.0000
Error	20	18.0000	0.9000		
Total	29	6925.5000			

CV(%) 1.04

Per cent inhibition of radial growth at 500 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	2430.0000	270.0000	675.00	0.0000
Error	20	8.0000	0.4000		
Total	29	2438.0000			

CV(%) 0.652

Appendix 9. ANOVA for fungitoxicity of fungicides against *Pestalotiopsis guepinii*

Per cent inhibition of radial growth at 100 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	16137.8135	1793.0904	911.24	0.0000
Error	20	39.3550	1.9678		
Total	29	16177.1685			

CV(%) 3.57

Per cent inhibition of radial growth at 200 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	13620.0675	1513.3408	615.98	0.0000
Error	20	49.1362	2.4568		
Total	29	13669.2037			

CV(%) 2.98

Per cent inhibition of radial growth at 300 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	13266.7147	1474.0794	258.32	0.0000
Error	20	114.1272	5.7064		
Total	29	13380.8419			

CV(%) 3.63

Per cent inhibition of radial growth at 400 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	12364.1069	1373.7897	264.19	0.0000
Error	20	104.0000	5.2000		
Total	29	12468.1069			

CV(%) 2.99

Per cent inhibition of radial growth at 500 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	3266.1043	362.9005	214.81	0.0000
Error	20	33.7881	1.6894		
Total	29	3299.8924			

CV(%) 1.47

Appendix 10. ANOVA for fungitoxicity of fungicides against *Sarocladium oryzae*

Per cent inhibition of radial growth at 100 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	16307.5575	1811.9508	1271.32	0.0000
Error	20	28.5050	1.4253		
Total	29	16336.0625			

CV(%) 3.51

Per cent inhibition of radial growth at 200 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	12603.7703	1400.4189	367.54	0.0000
Error	20	76.2048	3.8102		
Total	29	12679.9751			

CV(%) 3.98

Per cent inhibition of radial growth at 300 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	25825.6500	2869.5167	1572.34	0.0000
Error	20	36.5000	1.8250		
Total	29	25862.1500			

CV(%) 1.89

Per cent inhibition of radial growth at 400 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	21706.1094	2411.7899	2198.21	0.0000
Error	20	21.9432	1.0972		
Total	29	21728.0526			

CV(%) 1.42

Per cent inhibition of radial growth at 500 ppm

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	15017.8531	1668.6503	1329.86	0.0000
Error	20	25.0952	1.2548		
Total	29	15042.9483			

CV(%) 1.43

Appendix 11. ANOVA for antifungal activity of plant extracts against *Alternaria alternata*

Radial growth inhibition at 5 per cent

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	11293.5063	1254.8340	623.81	0.0000
Error	20	40.2312	2.0116		
Total	29	11333.7375			

CV(%) 2.46

Radial growth inhibition at 10 per cent

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	12967.2509	1440.8057	900.50	0.0000
Error	20	32.0000	1.6000		
Total	29	12999.2509			

CV(%) 1.59

Radial growth inhibition at 20 per cent

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	1519.1260	168.7918	2.27	0.0604
Error	20	1484.0741	74.2037		
Total	29	3003.2001			

CV(%) 8.91

Appendix 12. ANOVA for antifungal activity of plant extracts against *Aspergillus flavus*

Radial growth inhibition at 5 per cent

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	6913.3085	768.1454	253.93	0.0000
Error	20	60.5000	3.0250		
Total	29	6973.8085			

CV(%) 3.19

Radial growth inhibition at 10 per cent

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	9379.7491	1042.1943	613.06	0.0000
Error	20	34.0000	1.7000		
Total	29	9413.7491			

CV(%) 1.56

Radial growth inhibition at 20 per cent

The data for the response variable '20 per cent' is constant. So there was no significant difference among treatments.

Appendix 13. ANOVA for antifungal activity of plant extracts against *Curvularia lunata*

Radial growth inhibition at 5 per cent

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	22106.2914	2456.2546	1444.86	0.0000
Error	20	34.0000	1.7000		
Total	29	22140.2914			

CV(%) 2.56

Radial growth inhibition at 10 per cent

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	13904.5470	1544.9497	671.72	0.0000
Error	20	46.0000	2.3000		
Total	29	13950.5470			

CV(%) 1.98

Radial growth inhibition at 20 per cent

The data for the response variable '20 per cent' is constant. So there was no significant difference among treatments.

Appendix 14. ANOVA for antifungal activity of plant extracts against *Drechslera oryzae*

Radial growth inhibition at 5 per cent

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	17881.6801	1986.8533	794.74	0.0000
Error	20	50.0000	2.5000		
Total	29	17931.6801			

CV(%) 2.30

Radial growth inhibition at 10 per cent

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	3001.2001	333.4667	833.67	0.0000
Error	20	8.0000	0.4000		
Total	29	3009.2001			

CV(%) 0.6543

Radial growth inhibition at 20 per cent

The data for the response variable '20 per cent' is constant. So there was no significant difference among treatments.

Appendix 15. ANOVA for antifungal activity of plant extracts against *Fusarium moniliforme*

Radial growth inhibition at 5 per cent

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	4953.8636	550.4293	107.40	0.0000
Error	20	102.5000	5.1250		
Total	29	5056.3636			

CV(%) 4.62

Radial growth inhibition at 10 per cent

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	480.0000	53.3333		
Error	20	0.0000	0.0000		
Total	29	480.0000			

CV(%) 0.09

Radial growth inhibition at 20 per cent

The data for the response variable '20 per cent' is constant. So there was no significant difference among treatments.

Appendix 16. ANOVA for antifungal activity of plant extracts against *Fusarium solani*

Radial growth inhibition at 5 per cent

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	18095.5205	2010.6134	487.89	0.0000
Error	20	82.4200	4.1210		
Total	29	18177.9405			

CV(%) 3.74

Radial growth inhibition at 10 per cent

The data for the response variable '10 per cent' is constant. So there was no significant difference among treatments.

Radial growth inhibition at 20 per cent

The data for the response variable '20 per cent' is constant. So there was no significant difference among treatments.

Appendix 17. ANOVA for antifungal activity of plant extracts against *Microdochium oryzae*

Radial growth inhibition at 5 per cent

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	20368.1100	2263.1233	419.10	0.0000
Error	20	108.0000	5.4000		
Total	29	20476.1100			

CV(%) 3.64

Radial growth inhibition at 10 per cent

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)

name	9	480.0000	53.3333
Error	20	0.0000	0.0000
Total	29	480.0000	

CV(%) 0.00

Radial growth inhibition at 20 per cent

The data for the response variable '20 per cent' is constant. So there was no significant difference among treatments.

Appendix 18. ANOVA for antifungal activity of plant extracts against *Pestalotiopsis guepinii*

Radial growth inhibition at 5 per cent

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	2428.9455	269.8828	149.58	0.0000
Error	20	36.0856	1.8043		
Total	29	2465.0311			

CV(%) 2.10

Radial growth inhibition at 10 per cent

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	3720.0000	413.3333	187.88	0.0000
Error	20	44.0000	2.2000		
Total	29	3764.0000			

CV(%) 1.63

Radial growth inhibition at 20 per cent

The data for the response variable '20 per cent' is constant. So there was no significant difference among treatments.

Appendix 19. ANOVA for antifungal activity of plant extracts against *Sarocladium oryzae*

Radial growth inhibition at 5 per cent

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	2372.3041	263.5893	49.12	0.0000
Error	19	101.9619	5.3664		
Total	28	2474.2660			

CV(%) 4.85

Radial growth inhibition at 10 per cent

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	8482.5685	942.5076	559.61	0.0000
Error	19	32.0000	1.6842		
Total	28	8514.5685			

CV(%) 1.75

Radial growth inhibition at 20 per cent

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
name	9	1465.2047	162.8005	1546.60	0.0000
Error	19	2.0000	0.1053		
Total	28	1467.2047			

CV(%) 0.3325

Appendix 20. ANOVA for antagonistic potential of soil fungi against the pathogens of rice

Table 20A.Per cent inhibition in colony interaction of *Alternaria alternata*

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Name.of.fungi	8	3917.3697	489.6712	339.74	0.0000
Error	9	12.9718	1.4413		
Total	17	3930.3415			

CV(%) 1.98

Table 20B. Per cent inhibition in colony interaction of *Curvularia lunata*

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Name.of.fungi	8	1520.7100	190.0888	574.82	0.0000
Error	9	2.9762	0.3307		
Total	17	1523.6863			

CV(%) 0.98

Table 20C. Per cent inhibition in colony interaction *Drechslera oryzae*

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Name.of.fungi	8	2766.3128	345.7891	621.87	0.0000
Error	9	5.0045	0.5561		
Total	17	2771.3173			

CV(%) 1.41

Table 20D.Per cent inhibition in colony interaction of *Fusarium moniliforme*

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Name.of.fungi	8	3651.8226	456.4778	439.96	0.0000
Error	9	9.3378	1.0375		
Total	17	3661.1604			

CV(%) 1.91

Table 1E.Per cent inhibition in colony interaction of *Fusarium solani*

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Name.of.fungi	8	3167.3638	395.9205	1057.27	0.0000
Error	9	3.3703	0.3745		
Total	17	3170.7340			

CV(%) 1.05

Table 20F. Per cent inhibition in colony interaction of *Microdochium oryzae*

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Name.of.fungi	8	2766.3128	345.7891	621.87	0.0000
Error	9	5.0045	0.5561		
Total	17	2771.3173			

CV(%) 1.55

Table 20G. Per cent inhibition in colony interaction of *Pestalotiopsis guepinii*

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Name.of.fungi	8	1520.7100	190.0888	574.82	0.0000
Error	9	2.9762	0.3307		
Total	17	1523.6863			

CV(%) 1.02

Table 20H. Per cent inhibition in colony interaction of *Sarocladium oryzae*

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Name.of.fungi	8	2766.3128	345.7891	621.87	0.0000
Error	9	5.0045	0.5561		
Total	17	2771.3173			

CV(%) 1.35

Table 20I. Per cent inhibition owing to volatile substances of *Alternaria alternata*

Source	DF	Sum of Square	Mean Square	F Value	Pr (> F)
Name	9	3954.9617	439.4402	300.91	0.0000
Error	8	11.6829	1.4604		
Total	17	3966.6446			

CV(%) 5.08

Table 20J. Per cent inhibition owing to volatile substances of *Curvularia lunata*

Source	DF	Sum of Square	Mean Square	F Value	Pr (> F)
Name	7	1834.6888	262.0984	114.60	0.0000
Error	10	22.8708	2.2871		
Total	17	1857.5597			

CV(%) 4.73

Table 20K. Per cent inhibition owing to volatile substances of *Drechslera oryzae*

Source	DF	Sum of Square	Mean Square	F Value	Pr (> F)
Name	8	1439.7974	179.9747	78.83	0.0000
Error	9	20.5474	2.2830		
Total	17	1460.3448			

CV(%) 4.82

Table 20L. Per cent inhibition owing to volatile substances of *Fusarium moniliforme*

Source	DF	Sum of Square	Mean Square	F Value	Pr (> F)
Name	10	2819.8132	281.9813	533.44	0.0000
Error	7	3.7003	0.5286		
Total	17	2823.5134			

CV(%) 2.20

Table 20M. Per cent inhibition owing to volatile substances of *Fusarium solani*

Source	DF	Sum of Square	Mean Square	F Value	Pr (> F)
Name	10	2979.3486	297.9349	763.81	0.0000
Error	7	2.7305	0.3901		
Total	17	2982.0790			

CV(%) 3.10

Table 20N. Per cent inhibition owing to volatile substances of *Microdochium oryzae*

Source	DF	Sum of Square	Mean Square	F Value	Pr (> F)
Name	8	2100.9850	262.6231	769.90	0.0000
Error	9	3.0700	0.3411		
Total	17	2104.0550			

CV(%) 2.93

Table 20 O. Per cent inhibition owing to volatile substances of *Pestalotiopsis guepinii*

Source	DF	Sum of Square	Mean Square	F Value	Pr (> F)
Name	11	4939.3435	449.0312	191.08	0.0000
Error	6	14.1001	2.3500		
Total	17	4953.4436			

CV(%) 3.69

Table 20 P. Per cent inhibition owing to volatile substances of *Sarocladium oryzae*

Source	DF	Sum of Square	Mean Square	F Value	Pr (> F)
Name	7	832.4746	118.9249	72.47	0.0000
Error	10	16.4108	1.6411		
Total	17	848.8854			

CV(%) 5.46

Per cent inhibition owing to non volatile substances of *Alternaria alternata*

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Name.of.fungi	8	3917.3697	489.6712	339.74	0.0000
Error	9	12.9718	1.4413		
Total	17	3930.3415			

CV(%) 1.98

Table 20 Q. Per cent inhibition owing to non volatile substances of *Curvularia lunata*

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Name.of.fungi	8	1520.7100	190.0888	574.82	0.0000
Error	9	2.9762	0.3307		

Total	17	1523.6863

CV(%) 0.99

Table 20 R. Per cent inhibition owing to non volatile substances of *Drechslera oryzae*

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Name.of.fungi	8	2766.3128	345.7891	621.87	0.0000
Error	9	5.0045	0.5561		
Total	17	2771.3173			

CV(%) 1.34

Table 20 S. Per cent inhibition owing to non volatile substances of *Fusarium moniliforme*

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Name.of.fungi	8	3651.8226	456.4778	439.96	0.0000
Error	9	9.3378	1.0375		
Total	17	3661.1604			

CV(%) 1.91

Table 20 T. Per cent inhibition owing to non volatile substances of *Fusarium solani*

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Name.of.fungi	8	3167.3638	395.9205	1057.27	0.0000
Error	9	3.3703	0.3745		
Total	17	3170.7340			

CV(%) 1.05

Table 20 U. Per cent inhibition owing to non volatile substances of *Microdochium oryzae*

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Name.of.fungi	8	2766.3128	345.7891	621.87	0.0000
Error	9	5.0045	0.5561		
Total	17	2771.3173			

CV(%) 1.30

Table 20 V. Per cent inhibition owing to non volatile substances of *Pestalotiopsis guepinii*

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Name.of.fungi	8	1520.7100	190.0888	574.82	0.0000
Error	9	2.9762	0.3307		
Total	17	1523.6863			

CV(%) 1.02

Table 20 W. Per cent inhibition owing to non volatile substances of *Sarocladium oryzae*

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Name.of.fungi	8	2766.3128	345.7891	621.87	0.0000
Error	9	5.0045	0.5561		
Total	17	2771.3173			

CV(%) 1.35

Appendix 21. ANOVA for integrated management of *Alternaria alternata*

Per cent germination

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	2	49.8462	24.9231	19.84	0.0000
Treatment	12	1011.2308	84.2692	67.07	0.0000
Error	24	30.1538	1.2564		
Total	38	1091.2308			

CV% 1.43

Per cent mortality

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	2	0.0001	0.0001	0.00	0.9985
Treatment	12	47.7103	3.9759	88.12	0.0000
Error	24	1.0829	0.0451		
Total	38	48.7933			

CV% 2.58

Root length

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	2	0.1538	0.0769	1.83	0.1813
Treatment	12	2.0631	0.1719	4.10	0.0016
Error	24	1.0062	0.0419		
Total	38	3.2231			

CV% 6.67

Shoot length

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	2	0.5554	0.2777	1.08	0.3563
Treatment	12	22.7077	1.8923	7.34	0.0000
Error	24	6.1846	0.2577		
Total	38	29.4477			

CV% 3.82

Vigor index

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	2	43427.8108	21713.9054	6.93	0.0042
Treatment	12	615245.0026	51270.4169	16.35	0.0000
Error	24	75242.9559	3135.1232		
Total	38	733915.7692			

CV% 4.36

Appendix 22. ANOVA for integrated management of *Aspergillus flavus*

Per cent germination

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	2	0.0000	0.0000	0.00	1.0000
Treatment	12	1269.2308	105.7692	57.69	0.0000
Error	24	44.0000	1.8333		
Total	38	1313.2308			

CV% 1.77

Per cent mortality

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	2	1.2496	0.6248	0.72	0.4981
Treatment	12	66.1214	5.5101	6.33	0.0001
Error	24	20.8954	0.8706		
Total	38	88.2664			

CV% 9.58

Root length

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	2	0.1538	0.0769	2.32	0.1200
Treatment	12	2.8327	0.2361	7.12	0.0000
Error	24	0.7962	0.0332		
Total	38	3.7827			

CV% 5.37

Shoot length

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	2	1.3846	0.6923	1.43	0.2600
Treatment	12	21.4800	1.7900	3.69	0.0032
Error	24	11.6554	0.4856		
Total	38	34.5200			

CV% 5.01

Vigor index

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	2	3824.6117	1912.3058	0.84	0.4453
Treatment	12	707970.5326	58997.5444	25.82	0.0000
Error	24	54838.6367	2284.9432		
Total	38	766633.7809			

CV% 3.62

Appendix 23. ANOVA for integrated management of *Curvularia lunata*

Per cent germination

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	4	183.7538	45.9385	4.16	0.0056
Treatment	12	41608615	206.3077	31.44	0.0000
Error	48	529.4462	11.0301		
Total	64	4874.0615			

CV% 4.69

Per cent mortality

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	4	76.5189	19.1297	7.36	0.0001
Treatment	12	77.8493	6.4874	2.49	0.0125
Error	48	124.8253	2.6005		
Total	64	279.1935			

CV% 17.12

Root length

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	4	38.4524	9.6131	42.68	0.0000
Treatment	12	13.6937	1.1411	5.07	0.0000
Error	48	10.8117	0.2252		
Total	64	62.9578			

CV% 13.10

Shoot length

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	4	19.0205	4.7551	2.04	0.1042
Treatment	12	58.0374	4.8364	2.07	0.0376
Error	48	112.08650000	2.3351		
Total	64	189.1444			

CV% 11.07

Vigor index

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	4	650654.3177	325327.1588	16.55	0.0000
Treatment	12	1182606.3623	98550.5302	15.04	0.0000
Error	48	6902.3542	287.5981		
Total	64	1840163.0342			

CV% 9.27

Appendix 24. ANOVA for integrated management of *Drechslera oryzae*

Per cent germination

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	2	26.0491	13.0246	807800.78	0.0000
Treatment	12	698.1757	58.1813	3608482.18	0.0000
Error	23	0.0004	0.0000		
Total	37	724.2252			

CV% 0.006

Per cent mortality

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	2	25.9891	12.9945	16227385.17	0.0000
Treatment	12	130.9508	10.9126	13627454.76	0.0000
Error	23	0.0000	0.0000		
Total	37	156.9398			

CV% 0.0083

Root length

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	2	26.0021	13.0010	441268650.80	0.0000
Treatment	12	5.8088	0.4841	16429847.28	0.0000
Error	23	0.0000	0.0000		
Total	37	31.8109			

CV% 0.0048

Shoot length

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	2	25.9894	12.9947	17274273.17	0.0000
Treatment	12	64.2159	5.3513	7113695.03	0.0000
Error	23	0.0000	0.0000		
Total	37	90.2054			

CV% 0.0072

Vigor index

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	2	585035.6776	292517.8388	3058.26	0.0000
Treatment	12	760725.4876	63393.7906	662.78	0.0000
Error	23	2199.9115	95.6483		
Total	37	1347961.0767			

CV% 0.9224

Appendix 24. ANOVA for integrated management of *Fusarium moniliforme*

Per cent germination

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	2	0.0000	0.0000	0.00	1.0000
Treatment	12	1269.2308	105.7692	57.69	0.0000
Error	24	44.0000	1.8333		
Total	38	1313.2308			

CV% 1.72

Per cent mortality

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	2	1.2496	0.6248	0.72	0.4981
Treatment	12	66.1214	5.5101	6.33	0.0001
Error	24	20.8954	0.8706		
Total	38	88.2664			

CV% 3.52

Root length

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	2	26.0000	13.0000	NaN	NaN
Treatment	12	2.7559	0.2297	NaN	NaN
Error	24	0.0000	0.0000		
Total	38	28.7559			

There was no significant difference among roots length

Shoot length

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	2	26.0000	13.0000		
Treatment	12	26.2514	2.1876		
Error	24	0.0000	0.0000		
Total	38	52.2514			

CV% 0.056

Vigor index

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	2	622497.9837	311248.9919	3315.57	0.0000
Treatment	12	478444.7205	39870.3934	424.72	0.0000
Error	24	2253.0003	93.8750		
Total	38	1103195.7045			

CV% 0.9828

Appendix 25. ANOVA for integrated management of *Fusarium solani*

Per cent germination

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	4	122.0923	30.5231	4.15	
Treatment	12	1311.3538	109.2795	14.85	
Error	48	353.1077	7.3564		
Total	64	1912.7692			

CV% 3.53

Per cent mortality

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	4	11.0603	2.7651	0.81	0.5257
Treatment	12	37.6221	3.1352	0.92	0.5375
Error	48	164.0829	3.4184		
Total	64	212.7654			

CV% 16.98

Root length

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	4	25.7385	6.4346	13.33	0.0000
Treatment	12	8.9104	0.7425	1.54	0.1436
Error	48	23.1768	0.4828		
Total	64	57.8257			

CV% 16.53

Shoot length

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	4	99.4745	24.8686	19.74	0.0000
Treatment	12	21.9514	1.8293	1.45	0.1761
Error	48	60.4655	1.2597		
Total	64	181.8914			

CV% 8.06

Vigor index

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	4	1061318.8474	217929.0621	16.55	0.0000
Treatment	12	946600.7718	78883.3976	9.56	0.0000
Error	48	395883.2663	8247.5680		
Total	64	2403802.8855			

CV% 6.51

Appendix 26. ANOVA for integrated management of *Microdochium oryzae* Per cent germination

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	2	49.8462	24.9231	19.84	0.0000
Treatment	12	1011.2308	84.2692	67.07	0.0000
Error	24	30.1538	1.2564		
Total	38	1091.2308			

CV% 1.53

Per cent mortality

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	2	0.0001	0.0001	0.00	0.9985
Treatment	12	47.7103	3.9759	88.12	0.0000
Error	24	1.0829	0.0451		
Total	38	48.7933			

CV% 2.58

Root length

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	2	0.1538	0.0769	1.83	0.1813
Treatment	12	2.0631	0.1719	4.10	0.0016
Error	24	1.0062	0.0419		
Total	38	3.2231			

CV% 6.67

Shoot length

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	2	0.5554	0.2777	1.08	0.3563
Treatment	12	22.7077	1.8923	7.34	0.0000
Error	24	6.1846	0.2577		
Total	38	29.4477			

CV% 3.82

Vigor index

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	2	43427.8108	21713.9054	6.93	0.0042
Treatment	12	615245.0026	51270.4169	16.35	0.0000
Error	24	75242.9559	3135.1232		
Total	38	733915.7692			

CV% 9.38

Appendix 27. ANOVA for integrated management of *Pestalotiopsis guepinii*

Per cent germination

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	2	0.0000	0.0000	0.00	1.0000
Treatment	12	1269.2308	105.7692	57.69	0.0000
Error	24	44.0000	1.8333		
Total	38	1313.2308			

CV% 1.77

Per cent mortality

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	2	1.2496	0.6248	0.72	0.4981
Treatment	12	66.1214	5.5101	6.33	0.0001
Error	24	20.8954	0.8706		
Total	38	88.2664			

CV% 9.58

Root length

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	2	26.0000	13.0000		
Treatment	12	1.6617	0.1385		
Error	24	0.0653	0.00212		
Total	38	27.6617			

CV% 3.89

Shoot length

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	2	26.0000	13.0000		
Treatment	12	18.3969	1.5331		
Error	24	0.0043	0.0023		
Total	38	44.3969			

CV% 2.68

Vigor index

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	2	683714.1995	341857.0998	583.87	0.0000
Treatment	12	2087656.4573	173971.3714	297.13	0.0000
Error	24	14051.9327	585.4972		
Total	38	2785422.5896			

CV% 2.02

Appendix 28. ANOVA for integrated management of *Sarocladium oryzae* Per cent germination

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	4	122.0923	30.5231	4.15	
Treatment	12	1311.3538	109.2795	14.85	
Error	48	353.1077	7.3564		
Total	64	1912.7692			

CV% 3.53

Per cent mortality

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	4	11.0603	2.7651	0.81	0.5257
Treatment	12	37.6221	3.1352	0.92	0.5375
Error	48	164.0829	3.4184		
Total	64	212.7654			

CV% 16.98

Root length

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	4	25.7385	6.4346	13.33	0.0000
Treatment	12	8.9104	0.7425	1.54	0.1436
Error	48	23.1768	0.4828		
Total	64	57.8257			

CV% 16.53

Shoot length

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	4	99.4745	24.8686	19.74	0.0000
Treatment	12	21.9514	1.8293	1.45	0.1761
Error	48	60.4655	1.2597		
Total	64	181.8914			

CV% 8.06

Vigor index

Source	DF	Sum of Square	Mean Square	F Value	Pr(> F)
Replication	4	1061318.8474	217929.0621	16.55	0.0000
Treatment	12	946600.7718	78883.3976	9.56	0.0000
Error	48	395883.2663	8247.5680		
Total	64	2403802.8855			

CV% 6.51

Published paper from this research work

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