IJNRD.ORG ISSN : 2456-4184
INTERNATIONAL JOURNAL OF NOVEL RESEARCH AND DEVELOPMENT (IJNRD) | IJNRD.ORG
An International Open Access, Peer-reviewed, Refereed Journal

DETERMINATION OF SEED MYCOFLORA OF FARMERS' STORED SEEDS OF SELECTED *Oryza sativa* VARIETIES

Dangsha Martha Maring[1], Sangbanbi Devi Ningthoujam[2], Bimola Devi Naorem[3], Tahseen Fatimah[4], Raghumani Singh Seram[5]

1. Research Scholar, Arunodaya University, Itanagar, Arunachal Pradesh
2. PGSC, HRDRI, Canchipur, Imphal, Manipur
3. Department of Environmental Science, Kha Manipur College, Kakching, Manipur
4. Department of Botany, Arunodaya University, Itanagar, Arunachal Pradesh, India
5. Department of Botany, Manipur International University, Airport Road, Ghari, Imphal, Manipur, India

E mail ID: marthadangsha8@gmail.com

Abstract - The present study aimed to detect and identify seed-associated mycoflora and assess seed vigor in five rice (*Oryza sativa* L.) varieties—Sona Musori, Thoibi Phou, Chakhao Angangba, Drum Phou, and Chakhao Amuba—collected from the Kakching district, Manipur. Seed-borne fungi were isolated using the Blotter paper and Agar plate methods after ten months of storage. A total of twenty fungal species belonging to eleven genera were recorded, representing both field and storage fungi. The Blotter method detected 624 colonies (51.15%), while the Agar plate method revealed 596 colonies (48.85%), indicating the higher sensitivity of the Blotter technique. Among the identified fungi, *Bipolaris oryzae* (9.06%), *Curvularia lunata* (8.97%), *Magnaporthe oryzae* (8.64%), and *Fusarium moniliforme* (7.69%) were predominant and known pathogens causing brown spot, leaf spot, and blast diseases. Storage fungi such as *Aspergillus* and *Penicillium* spp. were moderately frequent, indicating suboptimal storage conditions. Variety-wise, Chakhao Amuba exhibited the highest fungal load (24.52–25.67%), while Drum Phou showed the least (7.70–8.77%). Seed vigor index (VI) varied significantly among varieties, with Drum Phou exhibiting the highest VI (2832.20), followed by Sona Musori (2745.10), reflecting superior seed quality and germination potential. Lower vigor indices in Chakhao varieties were associated with higher fungal infestation. The study highlights that both field and storage fungi persist in rice seeds during prolonged storage, adversely affecting germination and vigor. Integrated seed health management practices, including fungicidal treatment, controlled storage conditions, and routine seed health testing, are essential for maintaining seed quality and ensuring sustainable rice production.

Keywords: Blotter paper, Agar plate methods, vigor index (VI), pathogens, seed mycoflora

I. INTRODUCTION

Rice (*Oryza sativa* L.) is one of the world's most important cereal crops, serving as the staple food for more than half of the world's population. In India and many other countries, small-holder farmers save seeds from previous harvests for the next planting season, a practice that plays a crucial role in agricultural sustainability, farm economics and seed availability. However, the quality of farmer-stored seed is often compromised by multiple factors, among which seed-borne and storage-associated fungi (collectively referred to as seed mycoflora) play a major and sometimes neglected role.

Seed mycoflora refers to the fungal species that are either internally or externally associated with seeds, including those borne on the seed surface or within the seed tissues; these fungi may be carried over from the field or acquired during harvesting, drying, handling and storage. Such fungi influence seed health by reducing germination, seedling vigour, causing seed discolouration, rot, or becoming a source of inoculum for subsequent field infections. For instance, in rice seeds, a suite of fungi — including *Curvularia lunata*, *Bipolaris oryzae*, *Fusarium sp.*, *Aspergillus flavus* and *Penicillium sp.* have been frequently isolated. For example, in a study of rice seed samples from North East India, eight mycoflora taxa were identified using blotter and agar-plate methods, with *Bipolaris* accounting for 25.76 % and *Fusarium* 20.92 % of incidence under blotter method. However, it was observed that Bipolaris was predominant in blotter method (Umbrey et al., 2021).

When farmer-stored seeds are contaminated with fungi, several consequences follow. First, the germination percentage may decline, and speeding of germination may be delayed, thus impacting stand establishment in the field. Studies on rice have shown that seedborne fungi can reduce germination by 14-18 % and significantly lower seedling dry weight and vigour indices. The result reveals the diversity and prevalence of fungal pathogens in rice seeds (Haider et al., 2024). Second, discolouration, seed rot, or seedling blight can occur, lowering effective seed quality and affecting certification standards (for example, seeds with more than 50% discolouration often yield germination below the 80% threshold). Discoloured seeds gave rise to seed rot, producing stunted and blighted seedlings (Bala and Pannu, 2017). Third: Seed-borne mycoflora can act as a primary inoculum for field disease outbreaks, thereby reducing yield in the crop season and contributing to the cycle of infection between seasons. The most predominant fungus was *D. oryzae,* which was followed by *A. flavus,* and the least incidence was observed in the case of *F. solani* and *P. guepinni* (Chowdhury et al., 2021). Moreover, some of these fungi may produce mycotoxins or other metabolites that affect stored seed health or consumption value.

Farmer-stored seeds are particularly vulnerable because the storage environment is often sub-optimal: higher ambient temperature, fluctuating humidity, prolonged storage periods, less rigorous drying, and use of traditional packaging (sacks, cloth bags) rather than hermetic or controlled-environment storage. These conditions favour the proliferation of storage fungi (e.g., *Aspergillus*, *Penicillium*, *Rhizopus*), which may succeed field fungi (e.g., *Curvularia*, *Fusarium*) as storage duration increases. Higher incidence of mycoflora and *Aspergillus flavus* were recorded in jute-sack samples throughout the storage period. A study on pigeon pea showed that while field fungi dominated initially, storage fungi increased over time, especially under jute-sack storage. The level of aflatoxins detected in the jute-sack storage system was considerably higher than that occurring in the iron bin system (Bankole et al., 1995).

Given the crucial role of seed quality in crop productivity and the economic risk faced by smallholder farmers, it is imperative to monitor, quantify and characterise the seed mycoflora associated with farmer-stored seeds of rice. Although several studies have been done on seed-borne fungi of rice in various regions (for example, Ghana, Pakistan, India) — e.g., sixteen fungal species from eleven genera were isolated from five rice varieties in Ghana, with *C. lunata* (25.7 %) and *A. flavus* (23.8 %) being most predominant. The most predominant fungus on all five varieties was *C. lunata* (25.7%) and was closely followed in order of predominance by *A. flavus* (23.8%) and *B. oryzae* (21.2%). Seeds need to be treated before planting to reduce field and postharvest contamination of rice seeds. (Aidoo et al., 2015). There remains a need to focus on seeds saved by farmers (farmer-stored seeds) of selected rice varieties in specific agro-ecological zones, to assess the incidence and diversity of mycoflora under local storage conditions, and to relate this to seed health and storage practices.

Therefore, the present investigation aims to determine the seed mycoflora of farmer-stored seeds from selected *Oryza sativa* varieties. The objectives include: (1) to identify and quantify the fungal species associated with these stored seeds; (2) to compare differences among varieties and/or storage practices; and (3) to evaluate the implications of fungal contamination for seed health and seed quality. Such information can help inform seed health management, storage practices, certification processes, and ultimately support farmers in maintaining good seed quality and improving crop performance.

II. MATERIALS AND METHODS

Collection of Seed Samples

In the Kakching region of Manipur, India, five different kinds of rice (*Oryza sativa* L.) were gathered from the fields of farmers. These types are Sona Musori, Thoibi Phou, Chakhao Angangba, Drum Phou, and Chakhao Amuba. Until they were used, the seeds were allowed to air-dry, washed to remove any debris, and then stored in polythene bags that had been sterilized at a temperature of 25 ± 2 degrees Celsius.

Detection and Isolation of Seed Mycoflora

1. Blotter Method

In accordance with the recommendations provided by the International Seed Testing Association (ISTA, 2008), the Blotter method was utilized in order to identify fungi that are transmitted through seeds. Using Petri dishes with a diameter of nine centimeters, three layers of sterile blotter paper that had been dampened were placed within. A total of ten seeds that had been surface-sterilized were distributed evenly throughout each plate. For surface sterilisation, seeds were first submerged in sodium hypochlorite (NaOCl) at a concentration of 1% for a duration of one minute. Subsequently, the seeds were washed three times with sterile distilled water and then dried on sterile filter paper.

One week was spent incubating the plates at a temperature of 25 ± 2 degrees Celsius, with alternate cycles of 12 hours of light and 12 hours of darkness. The seeds were examined using a stereobinocular microscope after they had been incubated, and the fungi that were growing on the seeds were identified using standard mycological keys based on the colony morphology and microscopic characteristics (Wakil et al., 2014).

2. Agar Plate Method

According to Aneja (2003), the Agar plate method was utilized in order to validate the fungal isolates. Following the preparation of the Potato Dextrose Agar (PDA) medium, it was put into Petri dishes that had been disinfected. After the seeds had been surface-sterilized, they were placed on the medium, with ten seeds per plate, and then they were incubated at a temperature of 25 ± 2 degrees Celsius for a period of seven days. Both Smith, (2012) and Singh and Gumasta (2019) conducted an examination of emerging fungal colonies, purified them by subculturing them on fresh PDA plates, and identified them using microscopy.

Determination of Seed Germination and Seedling Growth

A total of 400 seeds, four duplicates of 100, were planted on damp blotter papers in Petri dishes. These seeds were then placed in an incubator at a temperature of 25 ± 2 degrees Celsius for a period of seven days, with a photoperiod of 12 hours. This was done for germination and vigor analysis.

Calculating the percentage of germination was done as follows:

The percentage of seeds that germinated, also known as germination 100 times the total number of seeds that were tested. The percentage of seeds that germinate is equal to the ratio of the total number of seeds tested to the number of seeds that germinate out of the total number of seeds tested. Total number of seeds that were tested for germination (percentage). The quantity of seeds that have germinated multiplied by 100.

Root length and shoot length (in centimeters) of 10 randomly selected normal seedlings from each replication were measured using a ruler. The seedlings were carefully uprooted, and the measurements were taken.

Determination of Vigor Index (VI)

Seed vigour index was calculated and determined by multiplying germination (%) and seedling dry weight according to Warham, (1990).

Vigour index (VI) = (mean of root length + mean of shoot length)×% of germination……………………………..….(i)

Where, VI = Vigour index

Identification of Fungal Isolates

Both macroscopic (colony color, texture, and development rate) and microscopic (spore and hyphal morphology) traits were utilized in order to identify the various species of fungi. Through the utilization of lactophenol cotton blue, slides were generated and thereafter inspected using a compound microscope with objectives of 40× and 100×. Standard fungal identification manuals were utilized in order to conduct the confirmation of the identification (Barnett and Hunter, 1998).

III. RESULTS AND DISCUSSION

Table 1: Frequency, occurrence of associated fungi with seeds of various rice varieties after 10 months of storage by Blotter paper method.

Fungi	A	B	C	D	E	Total	Frequency (%)
1. *Aspergillus flavus*	6	5	5	4	8	28	4.48
2. *A. niger*	4	6	4	5	6	25	4.06
3. *A. nidulans*	3	4	3	4	5	19	3.04
4. *A. Candidus*	4	7	6	3	6	26	4.17
5. A. Sp.	5	4	5	2	5	21	3.36
6. *Cheatomium globosum*	5	3	7	5	4	24	3.84
7. *Curvularia lunata*	8	9	11	13	15	56	8.97
8. C. Sp.	3	7	5	4	6	25	4.06
9. *Fusarium moniliforme*	9	7	11	9	12	48	7.69
10. *F. oxysporium*	5	4	8	7	6	30	4.80
11. *Alternaria alternata*	6	5	3	2	7	23	3.68
12. *Trichoconis podwickii*	3	5	5	3	6	22	3.52
13. *Magnoprothe oryzae* *	7	9	15	7	16	54	8.64
14. *Bipolaris oryzae***	8	10	17	6	15	56	8.96
15. *Sarocladium oryzae****	5	6	4	7	3	25	4.00
16. *Cladosporium herbarum*	6	7	5	4	6	28	4.48
17. *C. cladosporiodes*	5	6	5	4	7	27	4.32
18. *Penicillium chrysogenum*	5	5	6	4	7	27	4.32
19. *P. glaucus*	6	5	5	7	6	29	4.64
20. P. Sp.	5	6	6	7	7	31	4.96
Total	108	120	136	107	153	624	100
Frequency (%)	17.31	19.23	21.79	17.15	24.52		

A= Sona musori; B= Thoibi phou; C= Chakhao Angangba, D= Drum phou; E= Chakhao amuba; *= Pyricularia grisea; **= Heminthosporium oryzae; ***= Acrocylindrium oryzae

Seed-borne fungi play a crucial role in determining the quality, germination, and vigor of rice (*Oryza sativa* L.) seeds during storage. The Blotter paper method, widely used for seed health testing, revealed a diverse mycoflora associated with seeds of five rice varieties — Sona Musori (A), Thoibi Phou (B), Chakhao Angangba (C), Drum Phou (D), and Chakhao Amuba (E) — after ten months of storage. A total of 20 fungal species belonging to 11 genera were detected, with a total fungal count of 624 colonies, reflecting significant fungal colonization during storage (Table 1).

Among the identified fungi, Bipolaris oryzae (syn. *Helminthosporium oryzae*) and Curvularia lunata exhibited the highest frequency of occurrence (8.96% and 8.97%), followed by *Magnaporthe oryzae* (8.64%) and *Fusarium moniliforme* (7.69%). These pathogens are major causal agents of rice seed-borne diseases such as brown spot, leaf spot, blast, and seed discoloration, which reduce seed viability and germination (Ou, 1985; Mew and Gonzales, 2002). The consistent presence of these pathogenic fungi across all varieties highlights their persistence under long-term storage and their capacity for survival even at low moisture conditions (Niaz and Dawar, 2009).

Storage fungi belonging to Aspergillus (*A. flavus, A. niger, A. candidus, A. nidulans*) and Penicillium (*P. chrysogenum, P. glaucus*) were found at moderate frequencies ranging from 3.04% to 4.96%. These fungi are typical indicators of poor storage environments and are known for their ability to thrive under high humidity and temperature, leading to deterioration of seed quality and production of harmful mycotoxins such as aflatoxins and ochratoxins (Christensen and Kaufmann, 1969; Pitt and Hocking, 2009). Their occurrence in all the tested varieties suggests that the storage conditions were conducive to fungal proliferation, particularly in Chakhao Amuba (E), which showed the highest total fungal incidence (24.52%).

Field fungi such as *Alternaria alternata*, *Chaetomium globosum*, *Cladosporium herbarum*, *C. cladosporioides*, and *Trichoconis padwickii* were recorded at moderate frequencies (3.52%–4.48%). These species are typically field contaminants introduced at pre-harvest or post-harvest stages and can persist during storage if proper drying and cleaning practices are not followed (Bhale, 2001; Tandon et al., 2021). The occurrence of *Sarocladium oryzae* (syn. *Acrocylindrium oryzae*), the sheath rot pathogen of rice, also indicates field-to-storage transmission potential (Agarwal et al., 1989).

Variety-wise, Chakhao Amuba (E) exhibited the highest fungal load (24.52%), followed by Chakhao Angangba (C) (21.79%), Thoibi Phou (B) (19.23%), Sona Musori (A) (17.31%), and Drum Phou (D) (17.15%). The aromatic Chakhao rice varieties (C and E) were comparatively more susceptible to fungal invasion, likely due to their pigmented grain surface and higher oil content, which may enhance fungal adhesion and moisture retention. The comparatively lower fungal occurrence in Drum Phou and Sona Musori could be attributed to better husk integrity or lower grain moisture during storage.

The results show that both field and storage fungi can live on stored rice seeds, which is harmful for the seeds' health, germination, and crop establishment. Because pathogenic fungi like *Bipolaris oryzae, Magnaporthe oryzae*, and *Fusarium moniliforme* are present in all varieties, integrated seed health management practices should be used. These include treating seeds with safe fungicides, controlling moisture, and checking on them every so often (Agarwal and Sinclair, 1997). Using the right storage structures and keeping the environment under control may greatly reduce the growth of fungi, which will keep seed lots healthy and make rice production more sustainable.

Table 2: Frequency, occurrence of associated fungi with seeds of various rice varieties after 10 months of storage by Agar plate method.

Fungi	A	B	C	D	E	Total	Frequency (%)
1. *Aspergillus flavus*	5	5	4	2	8	24	4.03
2. *A. niger*	4	5	3	5	5	22	3.69
3. *A. nidulans*	2	4	3	3	5	17	2.85
4. *A. Candidus*	4	6	5	3	5	23	3.86
5. A. Sp.	4	4	4	2	6	20	3.36
6. *Cheatomium globosum*	5	2	6	4	5	22	3.69
7. *Curvularia lunata*	9	7	11	12	14	53	8.89
8. C. Sp.	3	5	5	3	5	21	3.52
9. *Fusarium moniliforme*	8	10	10	11	13	52	8.72
10. F. *oxysporium*	5	7	9	8	9	38	6.38
11. *Alternaria alternata*	5	3	5	2	6	21	3.52
12. *Trichoconis podwickii*	4	5	6	2	7	24	4.03
13. *Magnoprothe oryzae* *	9	9	13	6	14	51	8.56
14. *Bipolaris oryzae***	7	9	18	5	15	54	9.06
15. *Sarocladium oryzae****	4	4	5	6	3	22	3.69
16. *Cladosporium herbarum*	5	7	3	3	6	24	4.03
17. *C. cladosporiodes*	6	5	6	3	6	26	4.36
18. *Penicillium chrysogenum*	6	5	4	3	7	25	4.19
19. *P. glaucus*	5	4	6	5	7	27	4.53
20. P. sp.	5	5	7	6	7	30	5.03
Total	105	111	133	94	153	596	
Frequency (%)	17.62	18.62	22.32	15.77	25.67		

A= Sona musori; B= Thoibi phou; C= Chakhao Angangba, D= Drum phou; E= Chakhao amuba; *= *Pyricularia grisea*; **= *Heminthosporium oryzae*; ***= *Acrocylindrium oryzae*

Seed-borne fungi are among the major constraints affecting the quality, viability, and germination of rice seeds during storage. The present study revealed the occurrence and frequency of various fungal species associated with seeds of five rice (*Oryza sativa* L.) varieties—Sona Musori (A), Thoibi Phou (B), Chakhao Angangba (C), Drum Phou (D), and Chakhao Amuba (E)—after ten months of storage using the Agar plate method (Table 2). A total of 20 fungal species belonging to 11 genera were recorded, indicating considerable fungal diversity and contamination among the stored rice varieties.

Bipolaris oryzae (also known as *Helminthosporium oryzae*), which causes brown spot of rice, was the most common fungus found (9.06%). *Curvularia lunata* (8.89%), *Fusarium moniliforme* (8.72%), and *Magnaporthe oryzae* (8.56%), which causes rice blast disease, were also common. These harmful fungi are known to cause big losses in the field and in storage, which hurts both the health of the seeds and the output of the crops (Ou, 1985; Mew and Gonzales, 2002). Because these types happen so often, they are likely to get sick and need to be treated properly and stored in clean settings.

It also found a lot of non-pathogenic storage fungi, like *Aspergillus* spp. (*A. flavus, A. niger, A. candidus, A. nidulans*) and *Penicillium* spp. (*P. chrysogenum, P. glaucus*), with individual rates between 2.85% and 5.03%. These fungi are common contaminants of stored seeds when the temperature and humidity are high. They cause the seeds to change color, make mycotoxins, and lose their vigor (Christensen and Kaufmann, 1969; Pitt and Hocking, 2009). The presence of *Aspergillus flavus* and *A. niger* in all cultivars, especially Chakhao Amuba (E) and Thoibi Phou (B), indicates insufficient moisture management during storage.

There were moderate amounts (3.52%–4.36%) of field fungi like *Alternaria alternata, Cladosporium herbarum, Chaetomium globosum,* and *Trichoconis padwickii*. The identification of these illnesses suggests that certain infections may have begun in the field before harvest and continued through storage (Niaz and Dawar, 2009). *Sarocladium oryzae* (also known as *Acrocylindrium oryzae*), which is linked to sheath rot disease in rice, was also identified at a modest rate (3.69%). This shows that seed health monitoring is important for cultivars that are prone to this disease.

In terms of variety, the total fungal frequency was highest in Chakhao Amuba (E) (25.67%), followed by Chakhao Angangba (C) (22.32%), Thoibi Phou (B) (18.62%), Sona Musori (A) (17.62%), and lowest in Drum Phou (D) (15.77%). The greater fungal burden in Chakhao cultivars may be associated with their elongated grain shape, pigmented pericarp, and maybe enhanced surface moisture retention, facilitating fungal colonization (Tandon et al., 2021).

Overall, the data show that both field and storage fungi can live on rice seeds even after ten months of storage, which could cause the seeds to rot and spread disease. The research emphasises the necessity for comprehensive seed health management, incorporating fungicidal seed treatments, regulated humidity and temperature storage, and consistent surveillance of seed-borne fungi to preserve seed quality and guarantee sustainable rice production (Agarwal and Sinclair, 1997; Agarwal et al., 1989).

Table 3: Comparison of seed-associated mycoflora isolated through blotter paper and agar plate methods from paddy seeds.

Rice varieties	Associated fungal colony				Remarks
	Blotter paper method		Agar plate method		
	No.	%	No.	%	
Sona musori	108	8.85	105	8.61	
Thoibi phou	120	9.84	111	9.10	
Chakhao angangba	136	11.15	133	10.90	
Drum phou	107	8.77	94	7.70	
Chakhao amuba	153	12.54	153	12.54	
	624	51.15	596	48.85	1220

The comparative analysis of seed-associated mycoflora isolated through blotter paper and agar plate methods from different rice (*Oryza sativa* L.) varieties revealed notable variation in the total number and percentage of fungal colonies detected (Table 3). A total of 1220 fungal colonies were recorded from both methods, with 624 (51.15%) colonies detected by the blotter paper method and 596 (48.85%) by the agar plate method. Among the tested varieties, Chakhao amuba exhibited the highest fungal association, accounting for 153 colonies (12.54%) in both methods, indicating its high susceptibility to fungal contamination. This was followed by Chakhao angangba (11.15% and 10.90% in blotter paper and agar plate methods, respectively), suggesting that aromatic or pigmented varieties may harbour more diverse mycoflora, possibly due to differences in seed coat pigmentation and biochemical composition influencing fungal colonization (Tandon et al., 2021).

On the other side, Drum phou exhibited the lowest percentage of fungal colonies (8.77% and 7.70% by blotter and agar tests, respectively), which was indicative of a relatively lower fungal load or a higher level of resistance against seed-borne diseases. By using the blotter method, the Thoibi phou and Sona musori cultivars had intermediate levels of fungal incidence. The fungal recovery ranged from 8.85% to 9.84%, and the agar plate method yielded results ranging from 8.61% to 9.10%.

Between the two detection methods, the blotter paper method consistently yielded a higher percentage of fungal colonies compared to the agar plate method. This observation aligns with previous findings by Neergaard (1977) and Mathur and Kongsdal (2003), who reported that the blotter paper technique is more sensitive for detecting a wide range of seed-borne fungi, especially slow-growing or sporulating species. The increased efficiency of the blotter method could be attributed to the minimal nutrient availability and conducive moisture conditions, which promote sporulation and visibility of fungal structures directly on the seed surface. In contrast, the agar plate method, though effective for detecting fast-growing species like *Aspergillus* and *Fusarium*, might suppress the growth of slow-growing or less competitive fungi due to nutrient competition in the medium (George et al., 2019).

The overall similarity in fungal incidence patterns between both methods suggests that they complement each other for comprehensive detection of seed-associated mycoflora. Thus, the combined use of both techniques is recommended for accurate assessment of fungal contamination in paddy seeds to ensure effective seed health management (Haider et al., 2024; Ghosh et al., 2018). These findings highlight the importance of routine seed health testing, as seed-borne fungi not only reduce germination and vigor but also act as primary inoculum sources for field infections, posing significant threats to crop productivity and seed quality.

Table 4: Vigor index of paddy seedlings obtained from samples collected from farmers of different localities.

Paddy varieties	Mean of shoot length	Mean of root length	Germination (%)	Vigor index (VI)
Sona musori	13.80	14.50	97	2745.10
Thoibi phou	12.65	13.97	96	2555.52
Chakhao angangba	12.58	13.85	94	2484.42
Drum phou	14.13	14.77	98	2832.20
Chakhao amuba	12.35	13.83	93	2434.74

The vigor index (VI) of paddy seedlings, calculated based on mean shoot length, root length, and germination percentage, varied significantly among the tested rice varieties (Table 4). The data revealed that Drum phou exhibited the highest vigor index (2832.20), followed by Sona musori (2745.10), indicating superior seed vigor and physiological quality. These varieties also recorded the highest mean shoot lengths (14.13 cm and 13.80 cm, respectively), root lengths (14.77 cm and 14.50 cm, respectively), and germination percentages (98% and 97%, respectively). High seed vigor in these varieties may be attributed to better seed maturity, genetic potential, and lower fungal infestation, which together enhance the metabolic and enzymatic activities during germination (Chowdhury et al., 2014).

In contrast, Chakhao amuba recorded the lowest vigor index (2434.74) with comparatively reduced germination (93%) and shorter shoot and root lengths (12.35 cm and 13.83 cm, respectively). Similarly, Chakhao angangba and Thoibi phou exhibited moderate vigor indices of 2484.42 and 2555.52, respectively. The comparatively lower vigor indices in these aromatic and pigmented rice varieties may be linked to higher levels of seed-associated mycoflora, as observed in previous seed health studies, which can adversely affect seed germination, seedling growth, and overall vigor (Monajjem et al., 2014).

Seed vigor is a key indicator of the potential field performance of crop plants, reflecting both germination efficiency and early seedling growth capacity (ISTA, 2014). The strong correlation between vigor index and germination percentage in the present study indicates that both morphological and physiological seed quality parameters are influenced by the inherent varietal characteristics and seed health status (Powell 2024; Yari et al., 2012). The higher vigor indices recorded in Drum phou and Sona musori suggest these varieties possess better seedling establishment potential and stress tolerance under field conditions, consistent with the findings of earlier works on rice seed quality evaluation (Islam, 2021).

Overall, the results highlight the importance of assessing seed vigor alongside germination percentage to accurately determine seed quality. Variations among varieties may also be influenced by genetic factors, environmental conditions during seed development, and post-harvest handling practices. Therefore, maintaining seed health through proper storage and disease management is crucial for sustaining high vigor and productivity in paddy cultivation (Haider et al., 2024; Kuistary, 2014).

IV. CONCLUSION

The present study concludes that farmer-stored seeds of selected *Oryza sativa* varieties harbor a diverse range of seed-borne mycoflora, comprising both field and storage fungi, which significantly influence seed health, germination, and vigor. Among the twenty fungal species identified, *Bipolaris oryzae*, *Curvularia lunata*, *Magnaporthe oryzae*, and *Fusarium moniliforme* were predominant and pathogenic, while *Aspergillus* and *Penicillium* species indicated poor storage conditions. The blotter paper method proved slightly more sensitive than the agar plate method in detecting total fungal incidence. Variety-wise, Chakhao Amuba and Chakhao Angangba exhibited higher fungal loads and lower vigor indices, whereas Drum Phou and Sona Musori maintained superior seed vigor and germination, reflecting better physiological quality and lower fungal contamination. These findings emphasize the persistence of seed-borne fungi during prolonged storage and the necessity for integrated seed health management practices—including seed treatment, moisture control, improved storage systems, and routine seed health testing—to safeguard seed quality, ensure better crop establishment, and promote sustainable rice production.

REFERENCES

Abdul-Baki, A. A. and J. D. Anderson. 1973. Physiological and biochemical deterioration of seed. In: Kozlowski, T. T., (2nd Ed.) Seed Biology. Academic Press, New York, London. Pp. 283-315.

Agarwal, P. C., Mortensen, C. N., & Mathur, S. B. 1989. Seed-borne diseases and seed health testing of rice.

Agarwal, V. K., & Sinclair, J. B. 1996. *Principles of seed pathology*. CRC Press.

Aidoo, A. K., Appiah-Kubi, Z., & Bam, R. K. 2015. Mycoflora associated with seeds of five rice (Oryza sativa) varieties in Ghana.

BALA, A., & Pannu, P. P. S. 2017. Status of seed discoloration of rice, mycoflora associated and its impact on seed health and quality of farmer saved seeds. *Seed Research*, *45*(2), 131-135.

Bankole, S. A., Eseigbe, D. A., & Enikuomehin, O. A. 1995. Mycoflora and aflatoxin production in pigeon pea stored in jute sacks and iron bins. *Mycopathologia*, *132*(3), 155-160.

Barnett, H. L., & Hunter, B. B. 1998. Illustrated genera of imperfect fungi.

Chowdhury, P., Shamsi, S., Begum, H. H., & Bashar, M. A. 2021. Mycoflora associated with diseased rice grains in Bangladesh and their pathogenic potentiality. *Bioresearch Communications-(BRC)*, *7*(1), 932-940.

Chowdhury, S., Chowdhury, M. A., Bhattacherjee, S., & Ghosh, K. 2014. Quality assessment of rice seed using different storage techniques. *Journal of the Bangladesh Agricultural University*, *12*(2), 297-305.

Christensen, C. M., & Kaufmann, H. H. 1969. *Grain storage: The role of fungi in quality loss*. U of Minnesota Press.

Christensen, C. M., & Kaufmann, H. H. 1969. *Grain storage: The role of fungi in quality loss*. U of Minnesota Press.

George, T., Dania, V. O., & Ikotun, T. 2019. Evaluation of seed-borne fungi associated with paddy rice (Oryza sativa L.) varieties using different isolation methods. *Nigerian Journal of Mycology*, *11*, 46-58.

Ghosh, T. A. N. M. A. Y., Biswas, M. K., Guin, C. H. I. R. A. N. J. I. B., Roy, P. R. A. D. I. P. T. A., & Aikat, K. A. U. S. T. A. V. 2018. A review on seed-borne mycoflora associated with different cereal crop seeds and their management. *Plant Cell Biotechnology and Molecular Biology*, *19*(3), 107-117.

Haider, M. W., Mishra, M. K., Fayaz, F., Ahmad, D., & Mehmood, A. 2024. Evaluating the Impact of Seed-Borne Mycoflora on Seed Quality and Health of Various Oryza sativa Varieties. *International Journal of Microbiology*, *2024*(1), 1263308.

ISLAM, M. S. 2021. *Evaluation of growth and yield of four modern rice varieties as influenced by seedling age* (Doctoral dissertation, DEPARTMENT OF AGRICULTURAL BOTANY, SHER-E-BANGLA AGRICULTURAL UNIVERSITY, DHAKA, BANGLADESH).

ISTA. 1966."International Rules for Seed Testing", Seed Science & Technology 4 : 3-49.

ISTA. 1976. "International Rules for Seed Testing", Seed Science & Technology 4 : 3-49.

KUSTARY, J. 2014. Seed borne fungi associated with four selected inbreed rice varieties of Bangladesh and their influence on seed health and seedling vigor (Doctoral dissertation, Dept. of Plant Pathology).

Mathur, S. B., & Kongsdal, O. 2003. Common laboratory seed health testing methods for detecting fungi.

Mew, T. W., & Gonzales, P. 2002. *A handbook of rice seedborne fungi*. Int. Rice Res. Inst.

Monajjem, S., Zainali, E., Ghaderi-Far, F., Soltani, E., Chaleshtari, M. H., & Khoshkdaman, M. 2014. Evaluation seed-borne fungi of rice [Oryza sativa L.] and that effect on seed quality. *Journal of Plant Pathology & Microbiology*, *5*(4), 1-7.

Neergaard, P. 1977. McMillan Press, London, UK. *Seed pathology*, 1-2.

Niaz, I., & Dawar, S. 2009. Detection of seed-borne mycoflora in maize (Zea mays L.). *Pak. J. Bot*, *41*(1), 443-451.

Ou, S. H. 1985. *Rice diseases*. IRRI.

Pitt, J. I., & Hocking, A. D. 2009. *Fungi and food spoilage* (Vol. 519, p. 388). New York: Springer.

Powell, A. A. 2024. Seed vigor and its assessment. In *Handbook of seed science and technology* (pp. 603-648). CRC Press.

Reddy, Y. T. N., & Khan, M. M. 2001. Effect of osmopriming on germination, seedling growth and vigour of khirni (Mimusops hexandra) seeds.

Singh, S. M., & Gumasta, R. 2019. Some clinically significant genera of dematiaceous Hyphomycetes: an update. In *Recent Trends in Human and Animal Mycology* (pp. 99-142). Springer, Singapore.

Smith, J. E. (Ed.). 2012. *Aspergillus* (Vol. 7). Springer Science & Business Media.

Tandon, A., Anand, V., Kaur, J., Srivastava, S., Katiyar, R., Singh, P. C., & Srivastava, S. 2021. Microbiome Role in Control of Sustenance of Rice Health and Production. *Microbial Biotechnology in Crop Protection*, 335.

Umbrey, Y., Divya, M., Das, T., Das, S., & Mahapatra, S. 2021. Isolation and identification of seed borne mycoflora associated with popular rice cultivars in North East India. *Journal of Cereal Research*, *13*.

Wakil, W., Usman Ghazanfar, M., & Yasin, M. 2014. Naturally occurring entomopathogenic fungi infecting stored grain insect species in Punjab, Pakistan. *Journal of Insect Science*, *14*(1), 182.

Warham E.I. 1990. "Effect of Tilletia Indica Infection on Viability, Germination, and Vigor of Wheat Seed", Planr Disease 74, no. 2 : 130-135, https:// doi.org/10.1094/pd-74-0130.

Yari, L., Zareyan, A., Sheidaie, S., & Khazaei, F. 2012. Influence of high and low temperature treatments on seed germination and seedling vigor of rice (Oryza sativa L.). *World Appl. Sci. J*, *16*(7), 1015-8.