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Research Article

Isolation and identification of seed borne mycoflora associated with popular rice cultivars in North East India

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Abstract

A total of 32 seed samples collected from 11 locations were tested for associated seed borne mycoflora by using blotter method and Agar plate method. In blotter method, eight mycoflora were identified based on spore morphology namely, *Alternaria alternata, Aspergillus flavus, Aspergillus niger, Curvularia lunata, Fusarium* sp., *Bipolaris oryzae, Penicillium* sp. and *Trichoconis padwickii.* Most frequently occurring fungi genera were *Bipolaris* (25.76%), followed by *Fusarium* (20.92%) and least in case of *Penicillium* (3.33%). While in agar plate (PDA), maximum of 10 seed mycoflora like *A. alternata, A. flavus, A. niger, C. lunata, Fusarium* sp., *Bipolaris* sp., *Nigrospora* sp., *Pencillium* sp., *T. padwickii* and *Trichoderma* sp. were observed. Maximum percent occurrence was observed in genus *Fusarium* (22.67%), *Curvularia* (20.06%) and least in *Nigrospora* (0.15%) and *Trichoconis* (0.15%) respectively. Agar plate method showed maximum number of mycoflora on seeds than blotter method. However, it was observed that *Bipolaris* was predominant in blotter method and *Fusarium* in agar plate method.

Keywords: Agar plate method, blotter method, seed health, seed mycoflora

1. Introduction

Rice (*Oryza sativa* L.) is one of the staple food grain in Asian sub-continent and serves a billion population. In India, West Bengal, Uttar Pradesh, Madhya Pradesh, Punjab, Orissa and Bihar are the major rice producing states. In North east part of India, it is one of the primary diets being consumed daily and is a major source of livelihood for people dependent on rice cultivation. In this zone Rice crop is grown in varied agro-climatic conditions such as, in lower Gangetic plains (parts of West Bengal) and in Eastern Himalayan region (Meghalaya, Tripura and Arunachal Pradesh). But one of the major constrains in rice production is diseases caused by bacteria, fungi, viruses and nematodes; and are responsible for major economic losses in India (Mew and Gonzales, 2003). As such rice, is known to be affected by as many as a total of 153 seed-borne pathogens among which 18% are of quarantine importance, 65% are native pathogen and 17% are storage pathogens (Naveenkumar, 2017) in India. Agrawal (1999) reported more than 50 fungal pathogens to be seed-borne in rice. The mycoflora associated with seeds causes various harmful effects in seeds are; loss of germination capacity, reduced seedling vigor, seed discoloration, decay, increase in fatty acids and utilization of carbohydrates for the synthesis of protein and toxin production etc. (Oh, 2007; Nguefack, 2008; Uma and Wesely, 2013; Mannaa and Kim, 2016). They not only reduce the quality of seed but also transmitted from one season to other and may provoke the introduction of new pathogens in a virgin

area, causing quantitative and qualitative crop losses and permanent contamination of the soil (Ora *et al.*, 2011).

Several fungal pathogens have been isolated from rice grains and have been reported to be responsible for a number of diseases from the nursery to the field (Ibiam *et al.*, 2006). Therefore, using a good quality seed and performing seed health test to detect the presence of seed borne fungi becomes paramount to manage the diseases for healthy crop establishment. Considerable work has been done on seed health and detection of seed borne pathogens in rice seeds from different geographical region of the country (Sharma and Chahal, 1996; Gopalakrishnan and Valluvaparidasan, 2009; Gopalakrishnan *et al.*, 2010; Archana and Prakash, 2013; Sharma and Kapoor, 2016; Singh *et al.*, 2018). But, the information on seed health of rice varieties from different diverse geographical location of this region in particular the north east hilly region is scanty. The extent of discolouration varying with season, locality and variety was reported by Roy (1983), Mian and Fakir (1989) and Sunder *et al.* (1989). This might be due to variation of the weather conditions of agroclimatic zones, especially during harvesting period (Sharma and Kapoor, 2016; Das *et al.*, 2016). Keeping in view the potential threat it may cause to the only staple food grain crop grown in the region, present study was taken up to determine the prevalence and extent of different seed borne mycoflora associated with different popular rice varieties originating from diverse geographical location.

2. Materials and Method

2.1 Collection of samples

Thirty-two rice seed samples were collected from 11 different locations *i.e.*, Chakdaha, Jaguli, Gayespur, Kalyani, Hooghly, Purba Medinipur, Rampurhat, Uttar Dinajpur from West Bengal, Umiam (Meghalaya), Sabroom (Tripura) and Anini (Arunachal Pradesh) of North East India (Table 1 and Fig. 1). Seed samples were brought to laboratory in a plastic bag and kept at 4°C until further study. All the seeds samples were subjected to seed health testing following standard blotter method (Don, 2006) and agar plate method.

Table 1. Detailed information of seed samples collected from different locations of North East India

Sl. No.	Varieties	Designation	Location/GPS Coordinate	District/ State	Source
1	IET 4786	CDH 1	CHAKDAHA (CDH) 23.08°N 88.52°E	Nadia, West Bengal	Institutional farm
2	MTU 7029	CDH 2			
3	Shatabdi	JGL 1	JAGULI (JGL) 23.09°N 88.55°E	Nadia, West Bengal	Institutional farm
4	Swarna	JGL 2			
5	Debgiri	GYP 1	GAYESPUR (GYP) 22.98°N 88.48°E	Nadia, West Bengal	KVK farm
6	Maharaj	GYP 2			
7	Shatabdi	GYP 3			
8	Gobindobhog	HGY 1	HOOGLY HGY 22.90°N 88.39°E	Nadia, West Bengal	KVK farm
9	Pratikshya	HGY 2			
10	Swarna Masuri	HGY 3			
11	Swarna Sub	HGY 4			
12	Shatabdi	KYI-AB 1	KALYANI AB KYI-AB 22°58'30"N 88°26'04"E	Nadia, West Bengal	Institutional farm
13	Swarna	KYI-AB 2			
14	Gobindobhog	KYI-C 1	KALYANI- C KYI-C 22°58'30"N 88°26'04"E		
15	Harina Khori	KYI-C 2			
16	Radhuni Pagol	KYI-C 3			
17	Geetanjali	MDN 1	Medinipur MDN 22.424°N 87.319°E	Purba Medinipur, West Bengal	Farmers saved seed
18	Kala Dhan	MDN 2			
19	Paloi Dhan	MDN 3			
20	Sabita	MDN 4			
21	Swarna	RPH 1	Rampurhat (RPH) 24.17°N 87.78°E	Birbhum, West Bengal	Farmers saved seed
22	Hira	UDP 1	Raiganj 25°37'N 88°07'E	Uttar Dinajpur, West Bengal	Farmer's saved seed
23	Swarna	UDP 2			
24	IET 4094	UDP 3			

25	Pusa 1121	MGY 1	Umiam 25.57°N 91.88°E	Meghalaya MGY	CAU, CPGS
26	Sabhagidhan Lr- 23	MGY 2			
27	Hazari	TPR 1	Sabroom 23.00°N 91.73°E	Tripura TPR	Farmer's saved seed and KVK farm
28	Hybrid	TPR 2			
29	Nabin	TPR 3			
30	Puja	TPR 4			
31	Swarna Masuri	TPR 5			
32	Local variety	ARP 1	Anini 27°59'0"N 94°40'0"E	Arunachal Pradesh ARP	Farmers saved seed

Fig. 1. Map indicating the rice seed sample collection locations

2.2 Blotter Method

Two hundred seeds were randomly selected from each sample and placed on three layers of moisten sterilized blotter paper at the rate of 25 seeds per Petri plate (90 mm diameter) using method developed by (Don, 2006). The seeds were surface disinfected with 1% sodium hypochlorite solution for 2 min. followed by rinsing twice with sterilized distilled water and air dried prior to plating onto moistened blotting paper. Seed were placed in 1:8:16 fashion from the center to periphery and incubated at 25±1 °C for 7 days under 12 h alternating cycles of light and darkness. Incubated seeds were examined visually under stereo binocular microscope for the associated mycoflora. Identification was done based on their morphological character and microscopic examination of spores under zoomstar-v stereomicroscope (Barnett and Hunter, 1991; Mathur and Kongsdal, 2003; Mew and Gonzales, 2003). The percent incidence of the seed mycoflora was recorded in each sample.

2.3 Agar plate method

Surface sterilized seeds were placed at ten seeds per Petri plate containing 20 ml of PDA and incubated for 7 days as described in standard blotter method. The fungi growing out from the seeds were examined based on the colony appearance and identification of spore was done as described under blotter method. The laboratory experiment was conducted following completely Randomized Design (CRD) with four replications each.

3. Results and Discussion

In blotter method, from 32 seed samples collected from different locations of West Bengal, Meghalaya, Tripura and Arunachal Pradesh, eight seed borne mycoflora were observed like *Alternaria alternata* (0-30.25%), *Aspergillus flavus* (0-37.50%), *Aspergillus niger* (0-12.50%), *Curvularia lunata* (6.63-31.25%), *Fusarium* sp. (0-33.41%) *Bipolaris oryzae* (0-37.75%), *Penicillium* sp. (0-37.50%) and *Trichoconis padwickii* (0-25.50%) (Table 2). Also, fungal manifestation over the seed samples was visually examined under stereo binocular microscope and their fungal spore morphology showed different microscopic structure (Fig. 2 and Fig. 3).

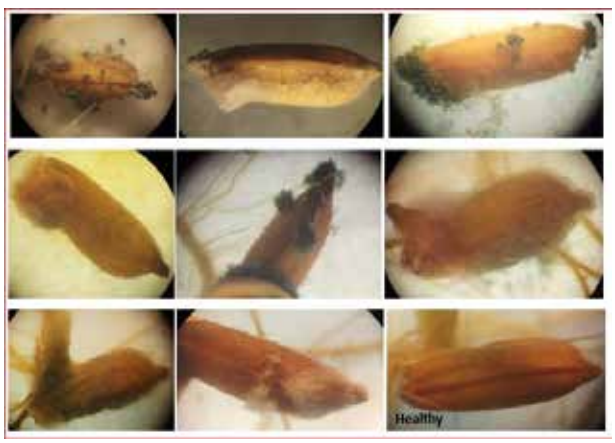

Fig. 2. Visual manifestation of seed associated mycoflora with rice seed along with an apparent healthy seed

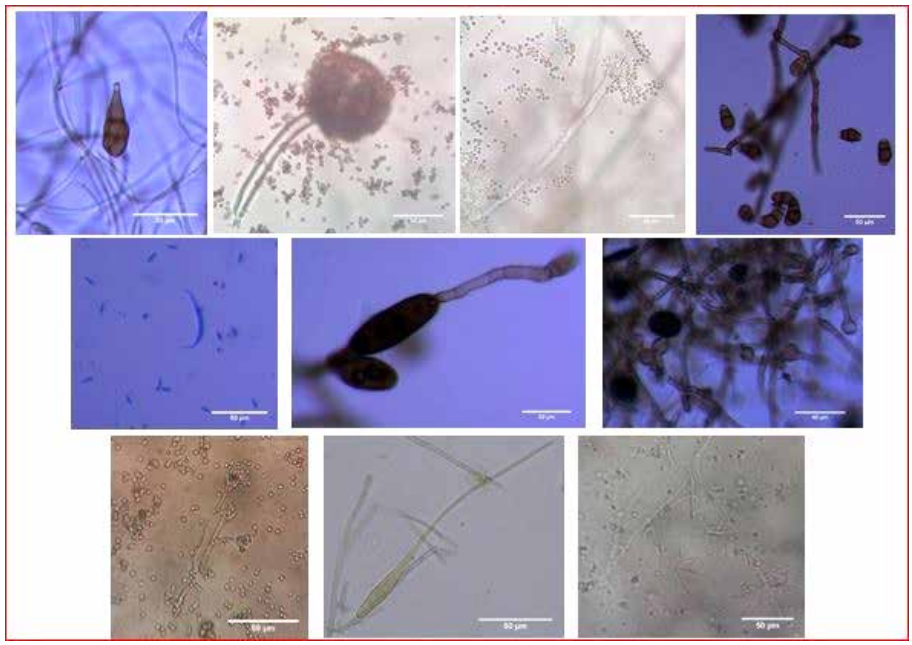

Fig. 3. Microscopic view of Seed borne mycoflora associated with Rice from top left to bottom right namely *Alternaria alternata*, *Aspergillus niger*, *Aspergillus flavus*, *Curvularia lunata*, *Fusarium* sp., *Bipolaris* sp., *Nigrospora* sp., *Penicillium* sp., *Trichoconis padwickii*, and *Trichoderma* sp.

Significant difference was observed with respect to associated seed mycoflora. Maximum per cent infection of *A. alternata* was seen on sample TRP-2 (30.25%) followed by HGY 1 (25.25%) and no infection was observed in samples MGY 1, HGY 3, MDN 2, and MDN 3. Sample MGY 1 (37.50%) had maximum *A. flavus* infection followed by MDN 2 (29.17%) and no infection in case of GYP 1, GYP 2, HGY2, HGY 4 and least 2.78% in JGL 1. None of the samples were free from *Curvularia* infection, sample KYI-C 2 (33.58%) and MGY 1 (6.63%) showed highest and least percent infection, respectively. Fungi *Fusarium* and *Bipolaris* were found to be associated with varying percent in most of the samples tested with an exception for the sample MGY 1. Most of the sample did not show *Penicillium* and *T. padwickii* infection. Overall, irrespective of samples tested *Bipolaris* (25.76%) was found to be the most predominant which is followed by *Fusarium* (20.92%) and *Curvularia* (19.15%). Similar, finding was also reported by many researchers. Mian and Fakir (1989) reported most predominant fungi in order of prevalence to be *Helminthosporium oryzae*, *C. lunata*, *Cladosporium cladosporioides*, *Aspergillus* spp. and *T. padwickii*. Ora *et al.* (2011) reported that among the 12 identified seed borne pathogens, *Xanthomonas* spp., *Rhizopus stolonifer*, *Aspergillus* sp., *B. oryzae* and *F. moniliforme* were pre-dominant on all tested hybrid rice varieties. Habib *et al.* (2012) also reported *B. oryzae* to be dominant fungal pathogen in seeds.

In agar method, ten seed associated mycoflora was observed *A. alternata* (0-9.76%), *A. flavus* (0-20%), *A.niger* (0-26.79%), *C. lunata* (6.67-40.27%), *Fusarium* sp. (0-45%), *Bipolaris* sp. (0-8.47%), *Nigrospora* sp. (0-4.76%), *Penicillium* sp. (0-15.87%), *T. padwickii* (0-4.76%) and *Trichoderma* sp. (0-4.76%). Their fungal spore morphology was examined under zoomstar-v stereomicroscope which showed different structure (Fig. 3). In this test also significant variation with respect to associated seed mycoflora was observed. *C. lunata* infection was observed in all the samples tested with maximum percent recorded in sample MDN 2 (40.27%), followed by KYI-AB1 (34.72%) and least in sample MDN 3 (6.67%). *Fusarium* sp. infection was observed maximum in sample UDP 2 (45%) followed by HGY 2 (40.34%) and KYI-C 2 (39.44%) while no infection was recorded in sample MGY 1 (0%) and least in sample TRP 2 (8.33%). Unlike blotter method *Bipolaris* infection was not predominant in agar method and only few samples were found to be associated with it. Fungi *Nigrospora*, *Trichoconis* and *Trichoderma* sp. were found in few samples of HGY 3 (4.76%), TRP 2 (4.76%) and CDH 1 (4.76%), GYP 1 (3.70%) respectively. Irrespective of all samples tested maximum percent infection was found to be of *Fusarium* sp. (22.67%) followed by *Curvularia* (20.06%)

Table 2. Percent seed-borne mycoflora associated with rice varieties collected from different locations of North Eastern states by blotter method

Variety	*Alternaria alternata*	*Aspergillus flavus*	*Aspergillus niger*	*Curvularia lunata*	*Fusarium* sp.	*Bipolaris oryzae*	*Penicillium* sp.	*Trichoconis padwickii*
CDH 1	5.00	5.00	3.13	13.15	30.50	26.25	0.00	21.25
CDH 2	11.92	9.75	6.90	10.51	14.09	33.14	18.45	0.00
JGL 1	5.55	2.78	2.78	22.36	23.90	25.14	0.00	17.75
JGL 2	18.33	8.83	5.00	15.67	18.57	24.38	0.00	15.25
GYP 1	8.38	0.00	0.00	15.75	27.93	30.00	0.00	0.00
GYP 2	15.56	0.00	0.00	22.71	21.60	29.64	0.00	10.99
GYP 3	6.75	5.75	8.75	19.59	24.58	26.75	0.00	10.00
HGY 1	25.25	6.25	0.00	30.75	22.50	16.25	0.00	0.00
HGY 2	6.73	0.00	0.00	25.50	33.33	27.08	8.81	0.00
HGY 3	0.00	8.58	8.85	16.71	20.84	25.75	8.68	12.50
HGY 4	15.33	0.00	0.00	32.25	22.50	29.17	0.00	0.00
KYI-AB 1	14.66	12.95	0.00	20.96	28.80	16.25	5.50	0.00
KYI-AB 2	3.65	10.92	7.76	14.55	28.58	23.34	12.75	0.00
KYI-C 1	20.83	4.49	4.44	18.59	13.94	32.43	7.62	0.00
KYI-C 2	12.50	0.00	0.00	33.58	16.66	37.75	0.00	0.00
KYI-C 3	20.83	8.46	0.00	14.58	33.41	22.92	0.00	0.00
MDN 1	8.38	0.00	0.00	20.98	20.83	12.25	0.00	25.03
MDN 2	0.00	29.17	0.00	20.83	12.50	37.53	0.00	0.00
MDN 3	0.00	5.63	0.00	23.75	30.83	32.08	0.00	8.33
MDN 4	16.35	13.27	10.00	16.35	13.85	23.27	0.00	19.42
RPH 1	4.17	16.67	12.50	8.41	25.00	20.58	0.00	0.00
UDP 1	12.50	12.50	0.00	27.11	31.25	16.67	0.00	0.00
UDP 2	4.17	26.69	4.17	20.83	21.67	22.75	0.00	0.00
UDP 3	20.83	12.56	8.33	16.67	8.33	33.33	0.00	0.00
MGY 1	0.00	37.50	0.00	6.63	0.00	0.00	37.50	0.00
MGY 2	12.50	12.50	12.50	16.69	20.83	25.00	0.00	0.00
TPR 1	6.53	6.25	9.38	15.63	25.00	37.50	0.00	0.00
TPR 2	30.25	15.00	12.50	9.17	12.50	25.00	0.00	0.00
TPR 3	12.50	12.50	0.00	31.25	12.50	31.25	0.00	0.00
TPR 4	5.10	20.83	8.33	16.25	16.50	18.75	0.00	16.67
TPR 5	12.50	8.33	4.67	19.64	16.32	32.14	7.34	0.00
ARP 1	6.35	3.67	0.00	15.48	19.79	30.06	0.00	0.00
Mean	10.73	9.90	4.06	19.15	20.92	25.76	3.33	4.91

	Variety (V)	Pathogen (P)	VXP
SEm±	0.02	0.01	0.07
CD (5%)	0.07	0.03	0.19

Table 3. Percent seedborne mycoflora associated with rice varieties collected from different locations of North Eastern states by Agar plate method

Variety	*Alternaria alternata*	*Aspergillus flavus*	*Aspergillus niger*	*Curvularia lunata*	*Fusarium* sp.	*Bipolaris oryzae*	*Nigrospora* sp.	*Penicillium* sp.	*Trichoconis padwickii*	*Trichoderma* sp.
CDH 1	3.33	3.33	11.43	25.12	10.00	4.76	0.00	3.33	0.00	4.76
CDH 2	4.17	0.00	13.33	18.77	29.17	3.33	0.00	0.00	0.00	0.00
JGL 1	0.00	4.23	8.36	28.37	31.61	8.47	0.00	0.00	0.00	0.00
JGL 2	0.00	8.33	15.28	24.07	32.87	0.00	0.00	0.00	0.00	0.00
GYP 1	0.00	8.33	11.11	18.98	23.61	7.41	0.00	0.00	0.00	3.70
GYP 2	0.00	7.44	0.00	19.44	22.69	0.00	0.00	7.74	0.00	0.00
GYP 3	3.77	3.80	10.00	17.29	17.41	0.00	0.00	12.86	0.00	0.00
HGY 1	6.67	0.00	4.17	30.28	35.56	0.00	0.00	0.00	0.00	0.00
HGY 2	9.76	0.00	0.00	30.56	40.34	4.17	0.00	4.17	0.00	0.00
HGY 3	3.70	4.17	14.82	18.98	16.20	0.00	0.00	0.00	0.00	0.00
HGY 4	7.45	3.70	7.41	19.58	35.45	0.00	4.76	0.00	0.00	0.00
KYI-AB 1	0.00	7.47	13.10	34.72	17.39	0.00	0.00	0.00	0.00	0.00
KYI-AB 2	4.17	7.50	10.83	19.17	29.17	0.00	0.00	0.00	0.00	0.00
KYI-C 1	0.00	6.73	0.00	32.72	25.56	6.67	0.00	0.00	0.00	0.00
KYI-C 2	5.56	0.00	0.00	31.73	39.44	0.00	0.00	0.00	0.00	0.00
KYI-C 3	0.00	0.00	5.56	13.33	18.89	0.00	0.00	0.00	0.00	0.00
MDN 1	0.00	7.54	18.46	17.86	11.11	0.00	0.00	15.87	0.00	0.00
MDN 2	0.00	6.67	12.22	40.27	25.89	6.67	0.00	12.22	0.00	0.00
MDN 3	0.00	20.00	26.79	6.67	12.50	0.00	0.00	3.33	0.00	0.00
MDN 4	0.00	3.67	6.67	24.29	11.43	8.10	0.00	4.76	0.00	0.00
RPH 1	4.17	4.16	18.45	13.69	31.83	0.00	0.00	0.00	0.00	0.00
UDP 1	0.00	0.00	4.17	13.10	23.21	0.00	0.00	0.00	0.00	0.00
UDP 2	3.33	4.27	7.53	21.78	45.00	3.33	0.00	0.00	0.00	0.00
UDP 3	3.83	4.17	13.23	17.37	24.80	0.00	0.00	0.00	0.00	0.00
MGY 1	0.00	16.67	8.33	8.33	0.00	0.00	0.00	0.00	0.00	0.00
MGY 2	0.00	0.00	5.59	16.33	16.33	0.00	0.00	8.33	0.00	0.00
TPR 1	0.00	9.52	8.67	8.67	12.50	4.76	0.00	4.76	0.00	0.00
TPR 2	0.00	0.00	6.63	8.33	8.33	0.00	0.00	5.56	4.76	0.00
TPR 3	0.00	5.56	9.52	15.87	16.67	4.76	0.00	0.00	0.00	0.00
TPR 4	0.00	4.17	4.17	12.50	14.82	0.00	0.00	0.00	0.00	0.00
TPR 5	0.00	8.33	14.29	12.17	19.33	0.00	0.00	0.00	0.00	0.00
ARP 1	0.00	9.52	13.10	21.45	26.19	4.17	0.00	4.17	0.00	0.00
Mean	1.87	5.29	9.48	20.06	22.67	2.08	0.15	2.72	0.15	0.27

	Variety (V)	Pathogen (P)	VXP
SEm±	0.02	0.01	0.08
CD (5%)	0.07	0.04	0.23

and least of *Nigrospora* sp. (0.15%) and *T. padwaki* (0.15%). Several researchers who have worked on seed heath test have also recorded similar findings. Similarly, Sharma *et al.* (1987) detected 10 fungal species from the rice seeds where they found *F. moniliforme* (*Gibberella fujikuroi*), *C. lunata* (*Cochliobolus lunata*), *A. flavus* to be most common. Ibiam *et al.* (2008) also reported *F. moniliforme* to be the most prevalent among the all the isolated fungi from rice seed in storage and field condition. Butt *et al.* (2011) isolated 12 seed borne pathogens among them *Xanthomonas* spp., *R. stolonifer*, *Aspergillus* sp., *B. oryzae* and *F. moniliforme* was found predominant on all tested hybrid rice varieties. Singh *et al.* (2018) from northeastern state of Mizoram also reported that among 21 fungi isolated from the farmers saved seed *F. moniliforme* was pre dominant in all tested rice samples ranging from 15-35% and 54-82.0% in agar plate and blotter paper method, respectively.

4. Conclusion

In the present study, all the samples subjected to blotter and agar plate method of detection were found to be associated with either one or more fungi and none of the sample was free from the fungal infection. In agar plate method, a greater number of mycoflora was detected than blotter method. Fungi *Bipolaris, Fusarium and Curvularia lunata* was found predominant in blotter method while *Fusarium* sp. and *Curvularia* was dominant in agar method. All the rice seed samples collected from different locations of northeast region showed varying percent of occurrence of seed borne fungi. Since, rice is the staple food crop of this region use of pathogen free seed and seed health test to detect and identify the associated mycoflora becomes vital in order to take up appropriate management strategies for successful rice cultivation.

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Ethical standards

Not applicable with this article.

Conflict of interest

The authors declare that they have no conflict of interest.

Author contributions

SD and SM designed the concept of the article. YU, MD and TD perform the experiments, collect the samples. YU, TD, SM wrote the manuscript, SM communicates to the journal time to time.

5. References

1. Agrawal RL. 1999. Seed Technology. 2nd ed. Oxford and IBH Publishing Co., New Delhi, pp. 87–97.
2. Archana B and HS Prakash. 2013. Survey of seed-borne fungi associated with rice seeds in India. *International Journal of Research in Applied Microbiology* **3**(1): 25-29.
3. Burnett HL and BB Hunter. 1991. Illustrated Genera of Imperfect Fungi, 4th ed., Burgess Publishing Co., Minneapolis, p. 241.
4. Butt AR, SI Yaseen and A Javaid. 2011. Seed borne mycoflora of stored rice grains and its chemical control. *The Journal of Animal & Plant Sciences* **21**(2): 193-196.
5. Das S, A Chakraborty and S Mahapatra. 2016. Effect of fungicidal mixture on severity of brown spot and blast of rice vis -s- vis meteorological factors on diseases progression in field. In: 3rd International Conference on Sustainable Agriculture and Environment (3rd ICSAE) September 26-28, 2016, Warsaw, Poland -475- ISBN: 978-605-9831-95-6.
6. Don, R. 2006. ISTA handbook on seedling evaluation. International Seed Testing Association.
7. Gopalakrishnan C and V Valluvaparidasan. 2009. Seed borne microflora associated with rice seeds in Tamil Nadu, India. *Crop Research (Hisar)* **37**: 207-209.
8. Gopalakrishnan C, A Kamalakannan and V Valluvaparidasan. 2010. Survey of seed-borne fungi associated with rice seeds in tamil nadu, India. *Libyan Agriculture Research Center Journal International*, **1**(5): 307-309.
9. Habib A, N Javed, ST Sahi and M Waheed. 2012. Detection of seed borne mycoflora of different coarse and fine rice varieties and their management through seed treatments, *Pakistan Journal of Phytopathology* **24**: 133-136
10. Ibiam OFA, CI Umemchuruba and AE Arnize. 2006. Seed borne fungi associated with seed of rice (*Oryza*

sativa L.) in storage and from the field in Ohaozara and Onicha local government areas of Ebonyi state. *World Journal of Biotechnology* **7**: 1062-1072.

11. Ibiam OFA, CI Umemchuruba and AE Arnize. 2008. Seed borne fungi associated with seed of rice (*Oryza sativa* L. Faro 12, 15, and 29) in storage and the field in Afikpo North local government area of Ebonyi state. *Scientia Africana*, **7**: 1-4.
12. Mannaa M and KD Kim. 2016. Microbe-mediated control of mycotoxigenic grain fungi in stored rice with focus on aflatoxin biodegradation and biosynthesis inhibition. *Mycobiology*, **44**(2): 67-78.
13. Mathur SB and O Konsdal. 2003. Common laboratory seed health testing methods for detecting fungi. Bassersdorf, Switzerland, ISTA, 425pp.
14. Mew TW and P Gonzales. 2003. A Handbook of Rice Seedborne Fungi. Laguna, Philippines/Enfield, NH, USA: International Rice Research Institute Science Publishers, Inc. 83pp.
15. Mian IH and GA Fakir. 1989. Fungi, Moisture Content and Germinability of Rough Rice Grains during Storage. *Seed Research* **17**: 169-173.
16. Naveenkumar R, A Muthukumar, G Sangeetha and R Mohanapriya. 2017. Developing eco-friendly biofungicide for the management of major seed borne diseases of rice and assessing their physical stability and storage life. *Comptes Rendus Biologies*, **340**(4): 214-225.
17. Nguefack J, V Leth, JL Dongmo, J Torp, PA Zollo, and S Nyasse. 2008. Use of three essential oils as seed treatments against seed-borne fungi of rice (*Oryza sativa* L.). *American-Eurasian. Journal of Agricultural and Environmental Science,* **4**(5): 554-560.
18. Oh JY, SN Jee, Y Nam, H Lee, MI Ryoo and KD Kim. 2007. Populations of fungi and bacteria associated with samples of stored rice in Korea. *Mycobiology*, **35**(1): 36-38.
19. Ora N, NAN Faruq, MT Islam, N Akhtar and MM Rahman. 2011. Detection and identification of seed borne pathogens from some cultivated hybrid rice varieties in Bangladesh. *Middle-East Journal of Scientific Research* **10**: 482–488.
20. Roy AK. 1983. Rice grain discoloration in Assam, India. *International Rice Research News* **9**: 10.
21. Sharma A and AS Kapoor. 2016. Detection of seed borne mycoflora associated with some rice varieties grown in Himachal Pradesh. *The Bioscan*, **11**(4): 2733-2739.
22. Sharma HL, HS Randhawa, A Kapur and S Singh. 1987. Seed Discoloration in Rice. *Seed Research and Production Unit* **24**(1): 37-41.
23. Sharma RC and HS Chahal. 1996. Multilocational evaluation of seed health of parental lines of hybrid rice. *Plant Disease Research* **11**(2): 169- 172.
24. Singh AR, T Boopathi, SB Singh, SK Dutta, LS Singh, SL Saha and NH Singh. 2018. Seed Borne Mycoflora of Tribal farmers' Saved Hill Rice, *Oryza sativa* in Northeast of India. *Indian Journal of Hill Farming*, Special issue: 58-65.
25. Sunder S, DS Dodan and SC Ahuja. 1989. Chemical control of glume discolouration of rice. *Indian Journal of Plant Pathology* **7**(1): 69-71.
26. Uma V and EG Wesely. 2013. Seed borne fungi of rice from South Tamil Nadu. *Journal of Academia and Industrial Research* **1**: 612-614.