



## Qualitative aeromycological study of the rice agroecosystem

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### Abstract

Rice crop is affected by many foliar diseases by fungal pathogens such as Blast, Brown spot, Narrow brown spot etc. might be polycyclic pathogens which are mainly spread and impacts all-around times of rice cultivation. Considering their impact on significant yield loss every year, a one-year (2020) aerobiological study was carried out in Thiruthani (13°11'N 79°38'E) at Thiruvallur District in two seasons Kharif (April to July 20/2019) and Rabi (August – December 23/2019). The aerobiological study conducted from January 2019 to December 2019 observed a total of  $2414 \times 10^3$  fungal colonies of 20 different species and some unidentified species. Of these,  $812 \times 10^3$  fungal colonies were observed during the Kharif season,  $1283 \times 10^3$  fungal colonies were observed during the Rabi season, and only  $319 \times 10^3$  fungal colonies were observed during barren land conditions (January 2019–March 2019). Most of the airborne pathogens reached 100% percentage frequency excluding *Penicillium* spp. (91.4%), *Rhizopus* spp (83.3%), *Pythium* spp (75%), *Erysiphe* spp (66.6%), *Puccinia* spp (83.3%), *Sclerospora* spp (58.3%), *Ustilagoidea* spp (75%) and *Albugo* spp (75%) were not present all over the months. *Airspora Bipolaris oryzae* percentage contribution was higher than all other organisms (20.7%) followed by *Curvularia* spp. (12.24%), *Alternaria* spp. (11.13%), and *Trichoderma* spp. (10.65%) are considered as Highly Potential microbes (Percent contribution >10%). *Alternaria*, *Bipolaris*, *Curvularia*, *Fusarium*, and *Trichoderma* spp were present all over the year as Potential microbes (Percent contribution 5 – 10 %). *Cylindrocladium*, *Cercospora*, and *Colletotrichum* spp (Percent contribution 2 - 5 %) were present all over the year but as Moderately Potential microbes, while, *Aspergillus* and *Pyricularia* spp (Percent contribution 1-2%) categorized as Less Potential pathogens. *Mucor*, *Rhizopus*, *Pythium*, *Erysiphe*, *Puccinia*, *Sclerospora*, *Ustilagoidea*, *Chaetomium*, *Albugo* and unidentified microbes (Percent contribution <1%) were at Low Potential condition. Among different classes of fungi, Ascomycetes occupied first position with 93.92 airspora percent contribution while other classes contributed below 2%.

**Keywords:** rice agroecosystem, airborne pathogens, percentage frequency, percentage contribution

### Introduction

Rice (*Oryza sativa*) is the third most widely cultivated crop in the world after wheat and maize. Even though many constraints exist in the rice cultivation, diseases are preambing among them, especially by airborne pathogens. Rice crop is vulnerable to various types of airborne diseases, including blast, brown spot, narrow brown spot, false smut, etc., these pathogens could be polycyclic pathogens that are spread widely all through rice crop production. The circadian rhythm followed by these pathogens in the airspora prolongs and sustains their infection nature, making their presence seem ubiquitous. Rice disease has serious negative effects on crop yield (Ruoling Deng *et al.* 2021) [8]. The macro and micro climate present in the Rice agroecosystem should be addressed since, they are the third factor in the disease triangle. For effective research to be conducted, airborne disease spread by the pathogens must also be validated, similar to soil-borne and seed-borne diseases. Using the aerobiological and epidemiological tools together will allow researchers to forecast the airborne diseases in rice agroecosystems and will thereby allow them to create a standardised model. In view of these factors, this research work is having the one-year aerobiological study at Thiruthani, Thiruvallur district, Tamilnadu, India to estimate the percent frequency and percent contribution of the prevailing organisms in the airspora of Rice agroecosystem. Additionally, this study investigated the taxonomical class-

wise and Inoculum potential-based classification of fungi in rice aerosphere.

### Materials and methods

#### Field Layout

A field study was conducted in Thiruthani (Thiruvallur district, 13°11'N 79°38'E) to collect data on airborne microflora. An area of  $20 \times 20$  m<sup>2</sup> was selected for the study. This study area was cultivated with ASD 16 variety of paddy during Kharif season (April 2019 to July 2019) and Co 43 variety of paddy during Rabi season (August 2019 to December 2019), because these varieties are susceptible to most of the airborne disease. Rice was grown on both the north and south sides of the study area, and coconuts, palm trees, and mango trees grew on both the east and west sides. Healthy seeds were carefully chosen to prevent infection through seed spreading. The standard crop management practices are followed without using foliar fungicides or pesticides.

#### Meteorological data collection

In order to produce the contextual data on aerobiological study, monthly weather data including temperature, relative humidity, precipitation, and wind speed were collected from the weather database “Weather Atlas” during the study period (Annexure1).

## Estimation of airborne microorganisms contribution in rice fields

### Nutrient Culture Plate Exposure method

To determine contribution of microorganisms to the microflora in our experimental plot, a Nutrient Culture Plate Exposure method (Eeshwary Prasad Chelaky and Sharma, 2012) <sup>[2]</sup> was applied. At 1.0 feet above the crop canopy, three sets of five sterilized Petri plates containing PDA media (mixed with 0.1% streptomycin sulfate) were exposed for two hours. Every time the height of this experiment was changed according to the crop's growth and canopy, to ensure microbes were captured from the aeromicroflora. Petri plates were incubated in a laboratory at 28° c for 24 hours after a predetermined time exposure. Polyvinyl acetate and anelene blue were used as staining media in order to count each fungal colony. Emerged fungal colonies were identified under a microscope based on their color, size, and shape. On the basis of morphological characteristics, visual identification was made by comparison with reference slides (Kadam & Khandia, 2018) <sup>[3]</sup>

### Estimation of aerial fungal flora

To estimate the air-borne fungal flora the percentage frequency and percentage contribution were calculated using the following formula (Eeshwary Prasad Chelak and K. Sharma, 2012) <sup>[2]</sup>.

$$\text{Percentage Frequency} = \frac{\text{Number of observations in which a species is appeared} \times 100}{\text{Total number of observations}}$$

$$\text{Percentage Contribution} = \frac{\text{Total number of colonies of a species in all observations taken together} \times 100}{\text{Total number of colonies}}$$

### Results and Discussion:

During the investigation,  $2414 \times 10^3$  fungal colonies were observed. There were  $812 \times 10^3$  fungal colonies seen in Kharif season (April to July/2019) and  $1283 \times 10^3$  fungal colonies seen in Rabi season (August 10 to December 23/2019), but only  $319 \times 10^3$  fungal colonies were observed on barren land (Table1). It was found that the number of fungal colonies in the aeromicroflora of the Rabi season was significantly greater than that of the Kharif season in this study. A majority of plant pathogens peaked between June and July, and the number drops after December. There is more plant pathogen activity and saprophytes in the Rabi season than in the Kharif season. Because of the weather conditions prevailing from August to December, including the low temperature, high relative humidity, and rain patterns, the RBS pathogen is more likely to establish and infect the rice crop than during the Kharif (April-July) season. In both seasons of the paddy crop, fungal colonies increased in the second month, which often coincided with the time of transplanting. In Kharif (April-July) and Rabi (August-December), microbe populations in the air increased during the maximum tillering stage. According to Michel Almaguer *et al.* (2013) <sup>[4]</sup>, microbe populations were more prevalent during the dry season from November to April. In barren land conditions, the population of fungal colonies was significantly lower than in cultivated seasons. The study concluded that the presence of the host and the canopy of the crop was crucial in attracting the pathogen in the air. A lower number of spores was found on barren land could be the result of collateral hosts such as *Cyanodon dactylon*, *Digitaria*, *Panicum repens*, etc. Sreeramulu and

Ramalingam (1966) <sup>[9]</sup> also observed that, in the absence of crop plants, grass species such as *Cyanodon dactylon*, *Digitaria*, and *Eleusine* serve as collateral hosts.

Throughout the study period, almost 20 fungal colonies as well as some unidentified colonies were found (Table.2). The average percentage contributed by *B.oryzae* (20.79) throughout the year was higher than the average percentage of *Curvularia* spp. (12.24) and *Alternaria* spp. (11.13). *Mucor species* (0.33), *Pythium species* (0.52), *Sclerospora species* (0.45), *Albugo species* (0.8), *Rhizopus species* (0.82), and *Puccinia species* (0.94), were found lesser than 1%. This was well documented by a quantitative study by Anna Maria Picco and Marinella Rodolfi (2002) on fungal air spores in rice fields near Pavia (North Italy) indicated the *Bipolaris oryzae* was detected at the end of June, with a peak in July. Chakraborty *et al.*, 2003 reported that *Bipolaris oryzae/ Drechslera oryzae* frequently contributes to 10.63 -14.58 percent of the total airborne fungal spora. Naim Uddin (2004) conducted an aerobiological survey on aeromycoflora by culture plate exposure technique for dry season rice crop and recorded viable spore counts up to 444 CFU during harvesting. *Pyricularia grisea* was detected in the air for 5 months, since the second week of June until the first week of October, and highest average concentration (25 cfu m<sup>-3</sup>) was observed in August in rice fields of Cuba (Michel Almaguer-Cha´vez *et al.*,2012). Those findings are in strong agreement with the present study's findings.

As far as percentage contributions, *Bipolaris oryzae* represented 20.7%, followed by *Curvularia* species (12.24%), *Alternaria* species (11.13%), and *Trichoderma* species (10.65%), which are all considered Highly Potential microbes(>10%). The presence of *Alternaria*, *Bipolaris*, *Curvularia*, *Fusarium*, and *Trichoderma spp.* was evident all across the year as Potential microbes (>5%). *Cylindrocladium*, *Cercospora*, and *Colletetrichum* species (2-5%) were present all year as Moderately Potential pathogens, while *Aspergillus* and *Pyricularia* species (1-2%) were classified as Less Potential pathogens. *Rhizopus*, *Pythium*, *Erysiphe*, *Sclerospora*, *Ustilaginoidea*, *Chaetomium*, *Albugo*, and unidentified microbes (\*1%) were classified as Low Potential. In terms of taxonomic classification, Ascomycetes occupied first position with 93.92 airspora percent contribution while other classes contributed below 2%. This report shows that most of the airborne pathogens categorised under Ascomycetes group of fungi.

Table.2. shows that Oomycetes are abundant from August to December, but are absent from January to July since the temperature is high (27.7°c on average) and relative humidity is low (60%) in comparison with August to December (26.5°c Temperature, 71.6% Relative humidity). Because of rainfall during August - December, factors such as temperature and relative humidity may vary. While Zygomycetes were absent from August to December, Basidiomycetes were not present in the hot summer months (April - May). Throughout the year, Ascomycetes fungi are ubiquitous, regardless of weather conditions. Most often, their presence is exacerbated during the rainy season in Tamilnadu state due to a combination of low temperatures and high relative humidity during the rainy season, which is from August to December. The study by Eeshwary Prasad Chelak and K. Sharma (2012) <sup>[2]</sup> noted 275 fungal colonies representing 28 species while stating that environmental factors are important to the distribution of fungal spores. A

maximum of 24 fungi were isolated from anamorphic groups, 3 from Ascomycotina, and 1 from Zygomycotina

out of 29. These results are consistent with those we found in our study.

**Table 1:** Estimation of fungal colonies in the rice airspora

Month of Observation (2019)		Total No. of colonies observed (×10 <sup>3</sup> )/ml	
1	Barren Land	January	156 <sup>c</sup>
2		February	085 <sup>c</sup>
3		March	078 <sup>bc</sup>
4	Kharif Season (April5/2019toJuly20/2019)	April	170 <sup>bc</sup>
5		May	185 <sup>b</sup>
6		June	220 <sup>bc</sup>
7		July	237 <sup>c</sup>
8	Rabi Season (August 10 to December 23/2019)	August	218 <sup>bc</sup>
9		September	232 <sup>a</sup>
10		October	269 <sup>a</sup>
11		November	279 <sup>a</sup>
12		December	285 <sup>a</sup>
Total number of colonies		2414	

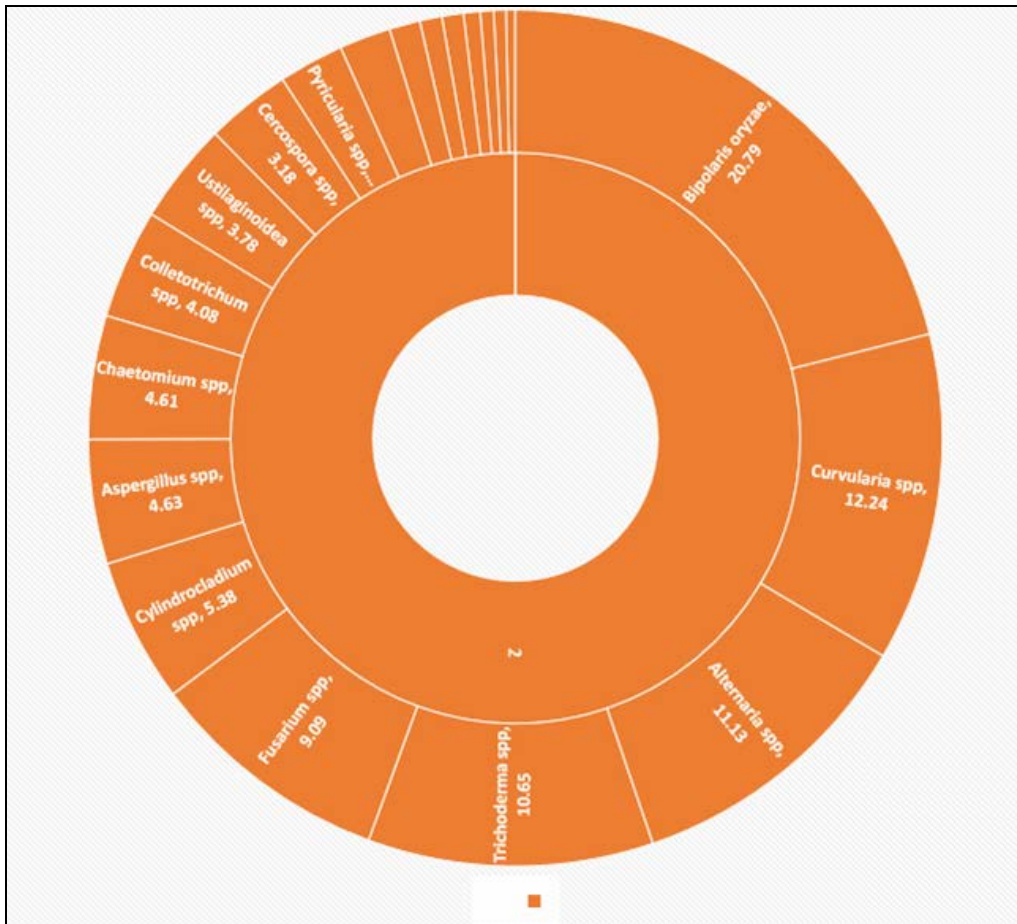
Values in the column followed by same letters not differ significantly by DMRT(p=0.05)

**Table 2:** Percentage Contribution of different fungi to aeromycological observations in rice fields

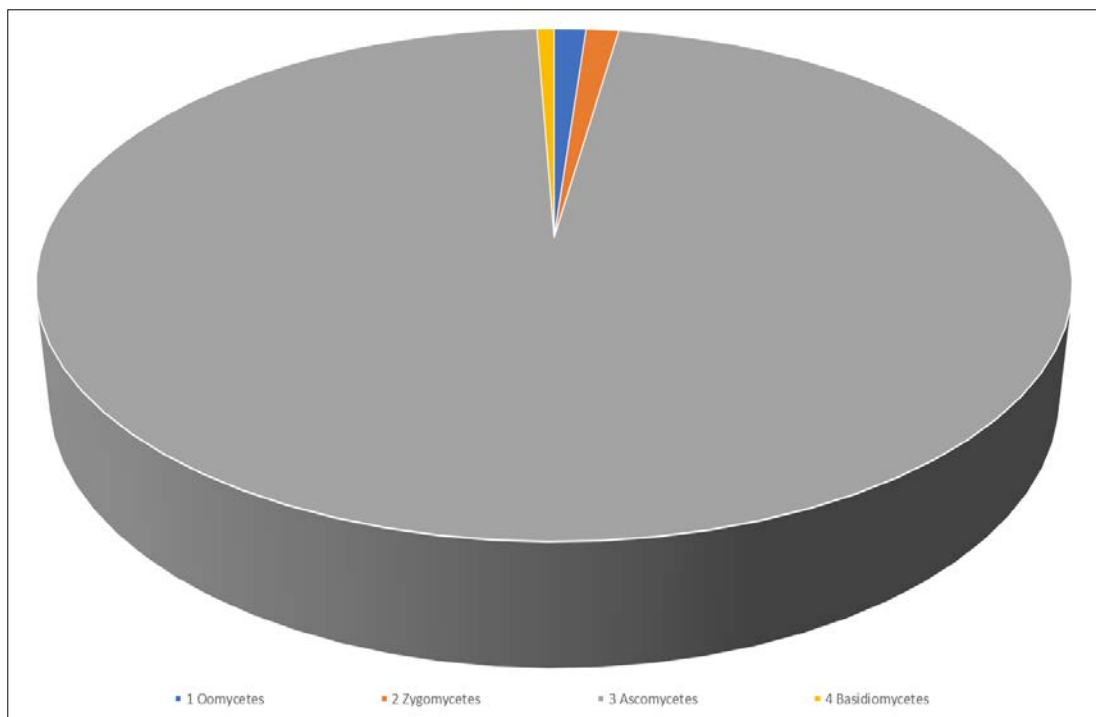
S.No	Microbe	Percentage contribution (Monthwise)												Average Percentage contribution	Percentage frequency
		Barren Land			Kharif season (April/2019 to July/2019)				Rabi Season (August to December /2019)						
		January	February	March	April	May	June	July	August	September	October	November	December		
1	<i>Pythium spp</i>	0.00	0.00	0.00	0.10	0.23	0.45	0.56	0.84	0.96	1.30	1.20	1.00	0.45	75.0
2	<i>Sclerospora spp</i>	1.23	1.35	0.32	0.23	0.00	0.00	0.00	0.00	0.00	0.64	1.32	1.01	0.50	58.3
3	<i>Albugo spp</i>	1.32	1.00	0.00	0.00	0.00	0.05	0.08	1.03	1.02	2.31	1.54	1.35	0.80	75.0
Average Percentage frequency and contribution of OOMYCETES														1.15	69.43
4	<i>Mucor spp</i>	0.56	0.21	1.98	1.08	0.05	0.06	0.05	0.06	0.05	0.04	0.03	0.06	0.35	100
5	<i>Rhizopus spp</i>	2.78	1.98	1.65	1.50	0.08	0.08	0.03	0.80	0.02	0.98	0.00	0.00	0.83	83.3
Average Percentage frequency and contribution of ZYGYMYCETES														1.18	91.65
6	<i>Alternaria spp</i>	5.98	6.83	6.90	6.05	14.34	15.90	16.10	12.88	12.96	10.87	11.88	12.90	11.13	100
7	<i>Aspergillus spp</i>	4.20	6.32	4.86	8.07	9.07	8.65	5.08	2.33	2.87	1.88	1.19	1.09	4.63	100
8	<i>Bipolaris oryzae</i>	10.54	11.84	13.13	14.50	16.07	19.10	22.50	25.50	26.90	28.45	30.87	30.08	20.79	100
9	<i>Curvularia spp</i>	16.32	9.23	9.68	9.86	13.87	14.08	14.40	14.00	13.08	10.75	10.89	10.76	12.24	100
10	<i>Fusarium spp</i>	7.97	11.54	8.54	8.76	7.01	6.98	7.34	7.86	8.80	9.78	11.78	12.80	9.09	100
11	<i>Penicillium spp</i>	4.23	3.60	3.56	2.97	1.65	1.07	1.35	1.87	0.98	0.86	0.90	0.00	1.92	91.6
12	<i>Pyricularia spp</i>	1.02	2.00	1.75	2.05	2.05	2.15	2.98	3.50	3.80	3.05	3.50	1.46	2.44	100
13	<i>Trichoderma spp</i>	10.36	8.05	10.08	10.90	10.20	12.98	10.58	10.00	11.08	10.65	11.00	11.87	10.65	100
14	<i>Cylindrocladium spp</i>	8.10	8.56	8.03	8.30	6.52	5.84	5.02	3.08	4.01	3.05	2.65	1.44	5.38	100
15	<i>Cercospora spp</i>	2.23	3.20	3.10	3.23	2.23	2.94	3.01	4.03	4.54	4.03	3.52	2.10	3.18	100
16	<i>Colletotrichum spp</i>	2.23	3.63	3.45	3.32	3.97	4.87	3.32	4.02	5.68	6.17	4.08	4.23	4.08	100
17	<i>Erysiphe spp</i>	1.63	1.52	1.93	2.61	2.10	1.94	1.48	0.62	0.00	0.00	0.00	0.00	1.15	66.6
18	<i>Ustilaginoidea spp</i>	8.52	7.23	6.35	0.00	0.00	0.00	3.32	4.22	2.18	2.96	4.32	6.23	3.78	75.0
19	<i>Chaetomium spp</i>	10.23	9.46	11.26	13.23	3.25	3.80	1.32	1.25	0.25	0.50	0.54	0.28	4.61	100
Average Percentage frequency and contribution of ASCOMYCETES														93.92	95.22
20	<i>Puccinia spp</i>	0.23	0.56	0.68	0.24	0.00	0.00	0.81	1.53	0.56	0.97	0.87	0.96	0.62	83.3
Average Percentage frequency and contribution of BASIDIOMYCETES														0.62	83.3
21	Unidentified	0.32	1.89	2.75	3.00	1.31	1.06	0.67	0.58	0.26	0.76	0.65	0.33	1.13	100

**Table 3:** Classification of airborne microbes based on its potential occurrence

S. No.	Percent contribution	Microbes	Category
1	< 1%	<i>Mucor, Rhizopus, Pythium, Erysiphe, Puccinia, Sclerospora, Ustilaginoidea, Chaetomium, Albugo</i> and unidentified microbes	Low Potential
2	1 - 2%	<i>Aspergillus</i> and <i>Pyricularia spp</i>	Less Potential
3	2 -5 %	<i>Cylindrocladium, Cercospora, and Colletotrichum spp</i>	Moderate Potential
4	5-10 %	<i>Fusarium spp</i>	Potential
5	> 10%	<i>Bipolaris oryzae, Curvularia spp, Alternaria spp., Trichoderma spp.</i>	Severe Potential



**Fig 1:** Airspora % contribution of various fungal microbes in Rice Agroecosystem



**Fig 2:** Taxonomic classwise fungi Airspora % contribution of different fungi in Rice Agroecosystem

**Conclusion**

Aerobiology is the science concerned with plant diseases that are affecting crop yields significantly. Combined with epidemiology, aerobiology will provide an excellent analysis of the spread and severity of plant diseases, as many airborne pathogens suddenly lead to epidemic

conditions. With regard to the Rice Agroecosystem as a staple food crop, the research should be designed out well to reach the farmers at their level and to make the profitable agriculture reachable. The present study is an introduction to airborne pathogens that must be provided for each crop against their diseases.

**References**

1. Anna Maria Picco and Marinella Rodolfi. *Pyricularia grisea* and *Bipolaris oryzae*: A preliminary study on the occurrence of airborne spores in a rice field. *Aerobiologia*,2002:18(2):163-167.
2. Eeshwary Prasad Chelak, Sharma K. Aeromycological study of Chandragiri hill top, Chhattisgarh. *International Multidisciplinary Research Journal*,2012:(11):15-16.
3. Kadam S, Rekha Khandia. Studies on composition and components of airspora belonging to phycomycetes over the field of sugarcane. *International Journal of Recent Trends in Science and Technology*, 2018, 130.
4. Michel almaguer, María-jesús aira, Francisco-javier Rodríguez-rajo & Teresa-irene rojas. Study of airborne fungus spores by viable and non-viable methods in Havana, Cuba. *Grana*,2013:52(4):289-298.
5. Naim Uddin. Airspora Studies Over a Rice (High Yielding Variety) Field in Rabi Season in the State of West Bengal, India. *Aerobiologia*,2004:20:127-134.
6. Pampa Chakraborty, Swati Gupta-Bhattacharya & Sunirmal Chanda. Aeromycoflora of an agricultural farm in West Bengal, India: A five-year study (1994–1999). *Grana*,2003:42:248-254.
7. Pampa Chakraborty, Arindom Chakraborty, Swati Gupta Bhattacharya. Dispersal of airborne pathogenic conidia of *Bipolaris oryzae* inciting brown spot disease of paddy in West Bengal, India. *Journal of Tropical Agriculture*,2020:58(2):199-208
8. Ruoling Deng, Ming Tao, Hang Xing, Xiuli Yang, Chuang Liu, Kaifeng Liao and Long Qi. Automatic diagnosis of rice diseases using deep learning. *Frontiers in Plant Science*,2021:12:701038.
9. Sreeramulu T, Ramalingam A. A two year study of the air spora of a paddy field near Visakhapatnam. -*Indian J. Agric. Sci*,1966:36:111-132.