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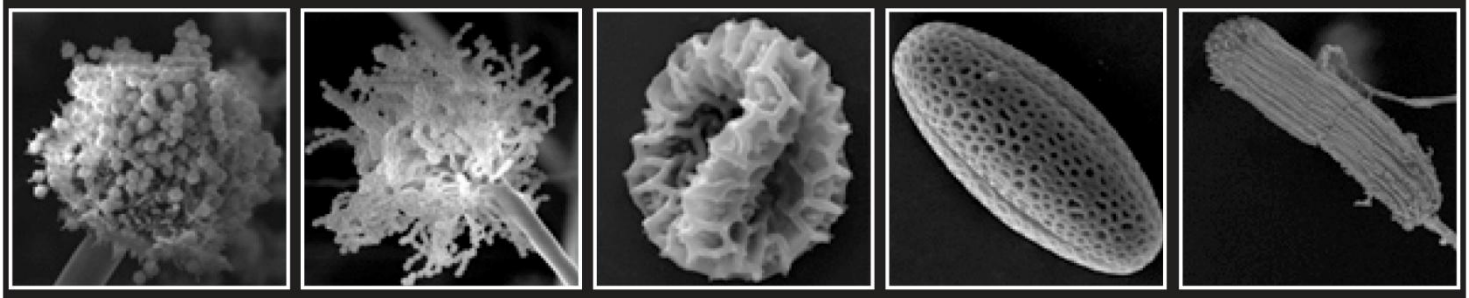

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NEW RECORD OF *IPOMOEA BIFLORA* (L.) PERS. (CONVOLVULACEAE) IN TELENGANA STATE, INDIA

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A new record of *Ipomoea biflora* (L.) Pers., (Convolvulaceae), recently collected from the dry deciduous forest of Ananthagiri Hills, Vikarabad District, Telangana State, is reported here, as a distributional addition for the flora of Telangana State. The detailed taxonomic description and phenology are explicated. The plant is distributed along margins and the paths of tropical dry deciduous forests up to 200-800 m altitude. There are eight plant species: *Tylophora indica, Senna unifloral, Achyranthes aspera, Sida acuta, Ipomoea nil, Apluda mutica, Setaria intermedia* and *Hyptis suaveolens*, which are growing in association with this plant species.

Color photographic plate is furnished to validate the species authenticity. The plant flowers during September-December producing large sized (approximately 66.62 µm diameter) panto-porate and echinate pollen grains.

Keywords: *Ipomoea biflora* (L.)Pers., New record, Ananthagiri hills, Telangana State.

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INTRODUCTION

The Convolvulaceae is known as morning glory family, including of 58 genera and approximately 2,000 species[1]. *Ipomoea* L. is the largest genus of the family Convolvulaceae and represented by 650 species having wide distributional range in the pan-tropical and sub-tropical region[2]. In India, the genus is represented by 63 species[3], with four endemic taxa[4]. In Telangana, there are previous reports that the genus *Ipomoea* is represented by 18 species[5] and 14 species[6] by different authors. During plant collections at the site of Ananthagiri Hills, Vikarabad District, authors found the specimen belonging to the family Convolvulaceae. After critical examination of specimens, inspection of relevant literature[7], it was identified as *Ipomoea biflora* (L.) Pers. This taxon has not been reported from Telangana State and therefore it forms a new distributional record for the state of Telangana, India.

MATERIALS AND METHODS

In November 2020, the authors found few interesting species of *Ipomoea* along margins and the paths of Tropical dry deciduous forest of Ananthagiri Hills, Vikarabad District in flowering and fruiting stage (Plate 1). The plants were collected for preservation in the form of Herbarium according to standard methodology and deposited in Osmania University Herbarium (HY). The complete phenological record of the plant, habitat, associated species, soil type, geographical coordinates, elevation were noted in the field. The photographs of fresh specimens were taken by using Cannon Power Shot SX70 HS and micro-scopic observations were carried out for critical studies by using compound light microscope (for example, to study pollen grains).

RESULTS AND DISCUSSION

Specimen examined: INDIA, Telangana, Vikarabad district, Ananthagiri Hills 16.11.2020, L. Paramesh & A. Vijaya Bhasker Reddy 0538 (HY).

Ipomoea biflora (L.) Pers. Syn. Pl. 1. 183. 1805. *Convolvulus* biflorus L. Sp. Pl. ed. 2. 2. 1668. 1763. *Ipomoea sinensis* (Desr.) Choisy in Mem. Phys. Soc. Geneve 6. 459. 1834

Habit: Annual prostate or twining pilose herbs; stems 2.5 m long.

Leaves: simple, petiole 2-5 cm long, lamina entire, triangular to ovate, 3.5–9.5 cm long, 3.5–6.5 cm wide, sparsely pilose, cordate at base with round basal lobes, acuminate at the apex.

Inflorescence: Cymose, 1-3 flowered

Flowers: small, white, peduncle up to 2.5 cm long, pedicle 1.6 cm long, hairy, bracts 2, small, linear to lanceolate, 5 mm long, hairy.

Sepals: 5 in number, unequal, accrescent, glabrous to pilose, outer 3 with ovate with cordate base, 4.5–7.5 mm long, 2.5– 4.5 mm wide, inner 2 linear to oblong, 5 mm long, 3 mm wide, lanceolate with less ciliate margins.

Corolla: funnel shaped, white or with purple centre, 6-7 mm long, midpetaline areas strigose.

Stamens: 5 in number, 3 mm, anthers ovoid to deltoid with sagittate base,

Pollen grains: Radially symmetrical, approximately 66.62 µm in diameter, Oblate-Suboblate, outline circular, pantoporate, sculpturing echinate with bulbous base, exine ornamentation microreticulate.

Ovary: Conical type, style short, stigma purplish, capsule globose, glabrous, 7-10 mm long, apiculate by the persistent style-base.

Seeds: Sub-trigonous, velvety or grayish pubescent with gray hairs up to 4.5-5 mm long.

Phenology: Plants found to flower in September-December.

Distribution: According to literature, *Ipomoea biflora* is growing in China, India, Indonesia, Japan, Myanmar, Vietnam, Africa, Australia, Thailand, Laos, and Pacific Islands. (Plants of the World Online. Kew Science)[8].

In the present study, the plant is distributed along margins and the paths of Tropical dry deciduous forests up to 200-800 m altitude. The associated plant species are 1. *Tylophora indica* (Burm. f.) Merr., 2. *Senna uniflora* (Mill.) H.S.Irwin & Barne-by, 3. *Achyranthes aspera* L., 4. *Sida acuta* Burm.f., 5. *Ipomoea nil* (L.) Roth, 6. *Apluda mutica* L., 7. *Setaria intermedia* Roem. & Schult., 8. *Hyptis suaveolens* (L.) Poit.

According to the standard literature on pollen size (Walker and Doyle, 1975)[9], pollen grains with diameter <10 µm - minute, 10-24 µm - small, 25-49 µm – medium, 50-99 µm – large, 100-199 µm – very large and more than 200 µm diameter are called gigantic pollen grains. In this respect, pollen of *Ipomoea biflora*, first reported from Telengana state, is large sized. Rajurkar *et al*[10]. reported the pollen of another species of *Ipomoea*, *I. trilobata* with diameter of 68.93-78.9 µm from Maharashtra state of India.

Study of pollen and floral morphology of some *Ipomoea* species in south-west Nigeria reported the pollen of *Ipomoea aquatica* to be largest (60.2-62.5 µm diameter) among species of the genus[11].

As the pollen grains of many species of *Ipomoea* are large sized, these have less chance to be widely airborne. However, in spite of that fact, *Ipomoea fistulosa* pollen grains are already reported to be airborne as well as allergenic from West Bengal, India[12]. In this aspect of human health effect, further studies are necessary to assess the allergenic potential of the pollen grains of this newly found species of *Ipomoea* from Telengana state, India.

ACKNOWLEDGEMENTS

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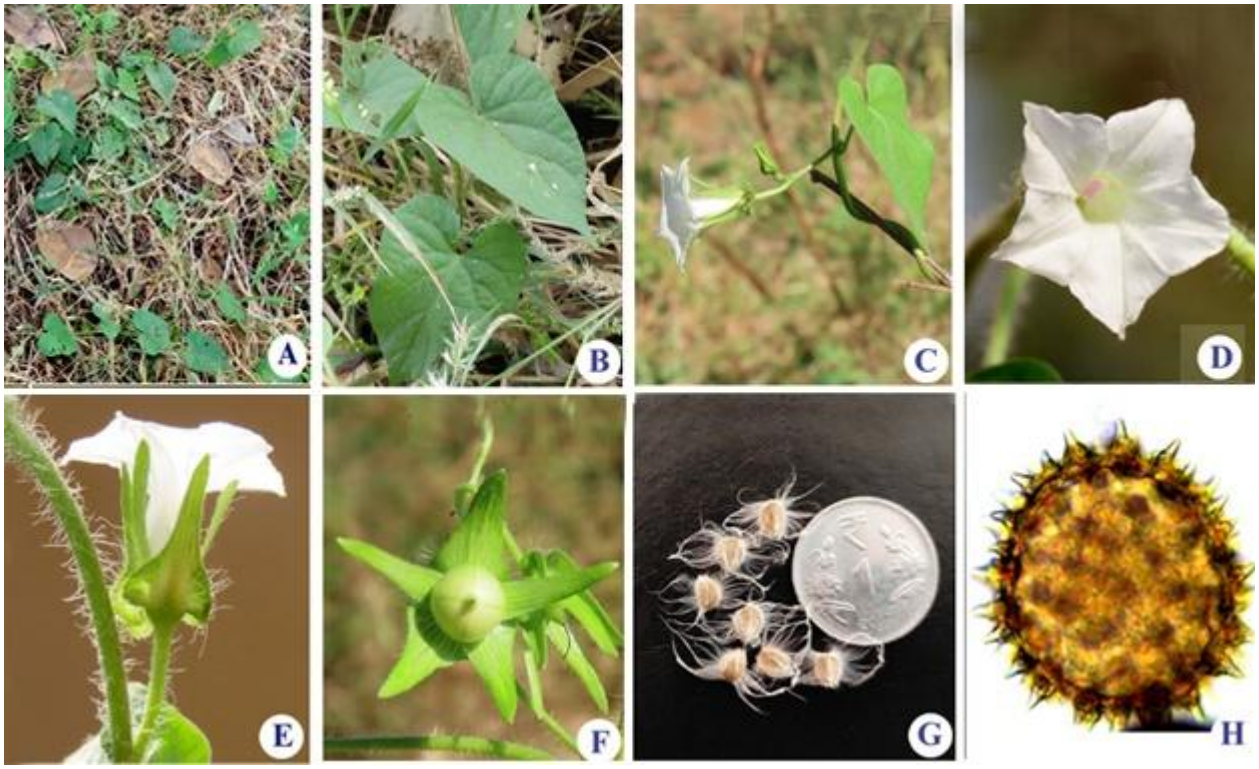

Plate-1: *Ipomoea biflora* (L.) Pers. **A**-Habit; **B**-Leaves; **C**-Inflorescense; **D**, **E**-Front and later views of flower; **F**-Fruit; **G**-Seeds; **H**-Pollen

AEROMYCOFLORA OF PHYTOPATHOGENIC FUNGAL SPORES AT THE PERIPHERY OF RAIPUR CITY

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A Study of Aeromycoflora of phytopathogenic fungal spores at the periphery of Raipur city was conducted for one year during March 2018 to February 2019. The aeromycoflora was isolated by using gravity petriplates method. The atmosphere was rich in propagule of different fungal species. Fungal spores are widely distributed over the area, which constitute the major bioparticle component of the air. The qualitative and quantitative assessment of pathogenic fungal spores would be useful for diseases forecasting system. Survey was carried out in six outdoor sites situated at rural and semi urban areas at the periphery of Raipur city.

A total of 48 species belonging to 20 fungal genera in addition to two sterile types were isolated and identified on the basis of colony morphology, mycelia and spore structure. *Alternaria alternata*, *Aspergillus niger*, *Acremonium strictum*, *Cladosporium cladoporioides*, *Curvularia lunata*, *C. oryzae*, *Fusarium oxysporum* and *Penicillium* species were found in all the sites throughout the year which were known to be the major airborne fungi causing various disease in plant (crop and vegetables) like *Fusarium* wilt, collar rot, *Alternaria* blight, *Colletotrichum* blight, downy mildew, powdery mildew, *Phoma* blight, sheath and collar rot in winter season and blast and brown spot in rainy season. Major fungal diseases harm the crops and vegetables, which also effect the production of crops (5% - 8%) and in vegetable (7% to 10%) in study area.

Keywords: Aeromycoflora, phytopathogenic fungal spores, Crop plant, Raipur city.

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INTRODUCTION

Dispersal in air is one of many mechanisms by which plant pathogens can spread to reach new susceptible plants either within the same field or even in a completely different continent[1-3]. Gregory (1973)[2] predicted that most spores of plant pathogens do not disperse beyond the field in which they were produced. Although this short-distance dispersal can be important for disease epidemics by infection of new plants in the same field or adjacent fields around inoculum sources, longer distance dispersal is also of great significance in allowing spores to reach new locations.

The composition of airborne fungal spores in outdoor environments is influenced by local weather which affects the biology of the fungi and the dispersion of their reproductive propagules. An intensive study and established the relationship between aerobiology and plant diseases and emphasized its role in forecasting the disease[4-5]. It is now the time that, people think and plan to utilize all resources, for aerobiological monitoring prediction and warning into a scheme of optimum value of human and plant population. Studies on fungal components of periphery of Raipur city are useful in understanding plant pathogen and in establishing the forecasting system for diseases control and the survey of aeromycoflora has assumed a great significance. The study of aeromycoflora of a particular region provides the clear view about interaction of fungal spores in the form of disease on plants as well as occurrence of allergy in human being.

Raipur is the capital of Chhattisgarh state. The major occupation of the rural area of Chhattisgarh is farming, where the major crops are rice and chickpea. The study of aeromycology is essential in plant pathology and in disease forecasting of plant diseases. The bio component occurring in aeromycoflora of rural area adjoining to Raipur needs to be analyzed qualitatively and quantitatively as they are the main cause of diseases in plants. Seasonal variation affects aeromycoflora of the area

Keeping this in view, the present investigation deals with the study of aeromycoflora occurrence at the periphery of Raipur city, its correlation with the diseases in plant for better understanding of the deposition and dissemination patterns of fungal spores.

MATERIALS AND METHODS

Study sites

The study sites of present studies are Chandanidih, Zora, Boriakala, Dhaneli, Baronda and Dumartarai. All the sites are a rural agricultural village, located 10 to 20 km away from periphery of Raipur city. The village occupies 1.0-1.5 km^2 area with a population of 4000-4500 people. The village economy is mostly (75%) dependent on agriculture and cattle farming. Many food crops viz, paddy, wheat, sugarcane, ground nut, maize, along with seasonal crops are cultivated. However, during recent years, the cropland has been decreasing due to urbanization.

Sampling Method

In the present study, six different outdoor sites were selected for sampling Aeromycoflora *i.e.* Chandanidih, Zora, Boriakala and Dhaneli, Baronda and Dumartarai. The study was carried out during March 2018 to February 2019. The culture plate exposure method was adopted for trapping the mycoflora. Potato dextrose agar (PDA) was used as culture medium.

10 ml of sterilized PDA medium was aseptically poured in petriplates and allowed to solidify. Five petriplates containing PDA medium were exposed in the air for 5-10 minutes at 1 meter above the ground level at the sampling sites. The study was conducted in intervals of 15 days in every month. The exposed petridishes were sealed with the help of cello tape and brought to the laboratory and incubated for 3-6 days at 26 ± 1ºC. After incubation fungal colonies were counted, isolated and identified with the help of standard literature[6-7]. For identification at species level, microscopic slides were prepared using glycerin gel as mounting media and lacto phenol and cotton blue stain[8].

A questionnaire survey along with interview was conducted in a population of 300 farmers and 4 agriculture officers to get an overall idea about the fungal diseases on crop vegetation in the study area. Season of crop growing, crop production, crop loss, disease prevalence, precautions taken and treatment process are noted during the survey.

RESULTS AND DISCUSSION

The fungal concentration and its variation in the air of the periphery of Raipur city are represented in Table 1. A total of 48 species belonging to 20 fungal genera in addition to two sterile types were recorded from all six sites. Out of six study sites, highest of 199 fungal species were recorded from Baronda and Dumartarai (118 fungal species) followed by Zora (94 fungal species), Dhaneli (94 fungal species), Chandanidih (87 fungal species) and Broiakala (81 fungal species) during the present study periods (Figure 1). A majority of fungi were members of anamorphic fungi (35 fungal species), while seven fungal species from Ascomycotina, four fungal species from Zygomycotina and two species of mycelia sterilia was recorded in all the site (Figure 2). Seasonal variation of aeromycoflora was also observed. *Aspergillus, Cladosporium, Alternaria, Curvularia, Fusarium, Acremonium and Penicillium* species found dominant throughout the season in every month. *Phoma pomorum* and *Thielavia terricola* was found as least frequent fungi. In summer, 44 fungal species belonging to 17 fungal genera were recorded. In rainy season, 44 fungal species belonging to 17 fungal genera and in winter, 45 fungal species belonging to 18 fungal genera were recorded. The genera recorded mainly comprised with *Cladosporium*, *Aspergillus*, *Curvularia* and *Alternaria* as dominating types in all the sites. In winter, the number of airborne fungal spores increased and climatic condition was favourable to cause disease in some plants. The probable reason for the severity and spread of the diseases was because of continuous rainfall, alternate cloudy weather, humidity and slow wind, soil condition, etc. Such meteorological condi-

tions favour the pathogen attack in plants. The air-borne spore load and infection was reported to have inverse correlation with the mean annual rain-fall[9].

Table 1: Aeromycoflora of phytopathogenic fungi at the periphery of Raipur city

SN.	Name of Aeromycoflora	Name of the study sites																	
		Chandanidih			Zora			Boriakala			Dhaneli			Baronda			Dumartarai		
		Summer	Rainy	Winter	Summer	Rainy	Winter	Summer	Rainy	Winter	Summer	Rainy	Winter	Summer	Rainy	Winter	Summer	Rainy	Winter
	Zygomycotina																		
1.	*Absidia spinosa*	-	+	+	-	-	-	-	+	+	-	-	-	+	-	-	+	+	+
2.	*Mucor cymosus*	-	+	+	-	+	+	+	+	-	+	+	+	+	+	+	+	+	+
3.	*Mucor* sp.	+	+	+	+	+	-	+	+	+	+	+	-	+	+	-	+	+	+
4.	*Gongronella butleri*	-	-	-	-	-	-	-	-	-	-	+	+	-	+	+	+	+	+
	Ascomycotina																		
1.	*Emericella nidulans*	-	-	-	-	+	-	-	+	+	-	+	+	+	+	+	+	+	+
2.	*Khuskia oryzae*	-	-	+	+	+	+	+	+	+	-	+	+	+	+	+	+	+	+
3.	*Phoma destructive*	-	+	+	+	-	-	-	-	-	-	+	+	+	+	+	-	-	+
4.	*Phoma glomerate*	+	-	+	-	-	-	-	+	-	-	-	-	-	+	+	+	+	+
5.	*P. pomorum*	-	-	+	-	+	+	+	-	-	+	+	-	-	+	+	+	-	-
6.	*Phomosis vexans*	-	-	+	-	+	+	-	+	+	-	-	+	+	+	+	-	+	-
7.	*Thielavia terricola*	-	-	-	-	-	+	-	-	-	-	-	-	-	-	+	-	-	-
	Anamorphic fungi																		
1.	*Alternaria alternate*	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
2.	*A. tenuissime*	+	-	-	-	+	+	-	+	-	-	-	+	+	+	+	+	+	-
3.	*A. crassa*	+	+	+	+	-	+	+	-	+	+	+	+	-	+	+	+	+	+
4.	*A. redicina*	-	-	+	-	-	+	+	+	+	-	-	+	+	+	+	+	+	+
5.	*A. citri*	+	+	+	+	+	+	-	-	+	+	+	+	-	+	+	-	+	+
6.	*Aspergillus niger*	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
7.	*A. flavus*	+	+	+	+	+	+	-	+	+	+	+	+	+	+	+	-	+	+
8.	*A. oryzae*	-	-	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
9.	*A. fumigates*	+	+	+	+	+	+	-	+	+	+	+	+	+	+	+	-	+	+
10.	*A. nidulans*	-	-	+	-	+	-	+	+	-	-	+	+	+	-	+	+	+	+

11.	*A. sclerotiorum*	-	-	+	-	+	+	-	+	+	-	-	-	-	+	+	+	+	+
12.	*A. stellatus*	+	-	-	+	-	-	-	-	-	-	-	-	+	-	-	+	-	-
13.	*A. terreus*	-	-	+	-	-	-	-	-	-	-	-	-	+	+	+	+	+	+
14.	*A. versicolor*	-	-	+	+	+	+	-	-	+	+	+	-	+	+	+	+	+	+
15.	*Acremonium strictum*	-	+	+	+	+	+	-	+	+	+	+	+	+	+	+	+	+	+
16.	*A. implicatium*	-	-	+	-	+	-	-	-	-	+	+	-	+	-	+	+	+	+
17.	*A. terricola*	-	-	+	-	-	-	-	-	-	-	+	+	-	+	+	+	+	+
18.	*Cladosporium* sp.	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
19.	*C. cladosporiodes*	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
20.	*C. oxysporum*	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
21.	*Curvularia lunata*	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
22.	*C. clavate*	+	+	+	-	+	+	-	+	+	+	+	+	+	+	+	-	+	+
23.	*C. tritici*	-	-	+	+	+	+	-	+	+	+	+	+	+	+	+	+	+	+
24.	*C. oryzae*	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
25.	*Drechslera tetramera*	-	-	+	-	-	+	-	-	+	-	+	+	+	+	+	-	-	+
26.	*D. australiensis*	-	-	+	-	-	+	-	-	-	-	+	+	-	-	+	-	-	+
27.	*Fusarium* sp.	+	+	+	+	+	+	-	+	+	+	+	+	+	+	+	+	+	+
28.	*F. oxysporum*	-	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
29.	*F. pallidoroseum*	+	+	+	+	+	+	+	-	+	-	-	-	+	+	+	+	+	+
30.	*F. moniliformis*	+	+	+	+	+	+	-	-	-	-	+	-	-	+	+	+	+	+
31.	*Monilia fructigena*	+	-	-	+	-	-	+	-	-	+	+	+	+	+	+	+	+	+
32.	*Penicillium* sp.	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
33.	*P. purpurogenum*	+	+	+	+	+	+	+	+	+	+	+	-	+	-	+	+	+	+
34.	*Nigrospora oryzae*	-	+	+	+	+	+	-	+	+	-	+	+	+	+	+	+	+	+
35.	*Tricoderma* sp.	-	-	+	+	+	+	+	+	-	-	+	+	+	+	+	+	+	+
	Mycelia sterilia																		
1.	*Mycelia sterilia white*	-	-	+	-	-	-	+	-	+	-	-	-	-	-	+	-	-	-
2.	*Mycelia sterilia black*	-	-	-	-	-	-	-	-	-	+	+	+	-	-	-	-	-	+
Total		**22**	**25**	**40**	**28**	**33**	**34**	**21**	**30**	**30**	**25**	**36**	**33**	**36**	**39**	**34**	**37**	**40**	**41**
Grand total		**87**			**95**			**81**			**94**			**199**			**118**		

(+) indicates presence of fungal species, (-) indicates absence of fungal species.

In winter, *Fusarium* wilt, collar rot, downy mildew, powdery mildew, *Alternaria* blight, *Colletotrichum* blight, *Phoma* blight, sheath and collar rot were recorded as plant (crops and vegetables) diseases. However, blast and brown spot diseases were recorded in rainy season and summer in the present survey. The impact of vegetation and crop field on saprophytic microflora in air was well emphasized[10] and application of this knowledge would be one of the best strategies of plant disease control[11].

In this study, *Aspergillus* was observed as most frequent and dominant species. Similar result was

found by earlier researchers in the air at Raipur[12-15] and different places like Aurangabad[16], Kanpur[17] and Nagpur[18]. Similarly, *Cladosporium* were reported as predominant and most frequent fungi at Raipur[19] and different places like Nagpur[18] and Rohtak[20]. Similarly, *Curvularia* was most frequent species observed by earlier workers. Bhatt *et al,* (2009)[21] reported *Curvularia lunata* as dominant causal agent of glume discoloration of rice in Kashmir valley. Sreeramulu and Ramalingam (1966)[22] and Ahire *et.al*. (2007)[23] recorded *Cladosporium* sp as "universal dominant". Khailare and Chitnavis (2002)[24] reported that *Aspergillus niger, Alternaria and Cladosporium* sp. were also the most frequent fungal species of slum and descent area of Kolhapur. Uddin (2004)[25] reported that *Penicillium* and *Aspergillus* were most frequent in airspora, along with *Aspergillus niger, Cladosporium oxysporum* and *Alternaria alternata.* In this way, it is evident that the seasonal variation as well as meteorological conditions affects the fungal flora of the environment both qualitatively and quantitatively. The present study provides the qualitative and quantitative feature on aeromycoflora at the periphery of Raipur city.

Ekhane Fig. 1 & 2

CONCLUSION

Through questionnaire survey along with interview of farmers and field agriculture officers, it was found that fungal spores cause disease in crop plants and vegetables in the study area. *Fusarium* wilt, collar rot, downy mildew, powdery mildew, *Alternaria* blight, *Colletotrichum* blight, *Phoma* blight, sheath and collar rot were the common plant diseases in winter, whereas blast and brown spot disease were common in rainy season and summer.

Pathogenic infection by *Fusarium, Alternaria, Cladosporium, Phoma, Aspergillus* etc. has been recorded in the present study. The spores of anamorphic fungi were the large contributors of the total air borne fungal spores. Thus *Aspergillus, Cladosporium, Alternaria,* and *Curvularia,* which were dominant in all the sites also recorded in higher concentrations the air. Monitoring of air borne fungi can be helpful in prevention of fungal diseases. Experimental survey proves that higher concentration of pathogenic fungal spores in the atmosphere can initiate diseases in plant. The systemic characters will help to develop diagnostic keys supplemented with information on symptoms of diseases, its extent of damage and distribution and management strategies. Present survey showed Carbendazim, Propiconozol and Hexaconozol to be most effective antifungal agents used by farmers in different crop plants. For precaution measure, proper treatment of seed, bund cleaning and use resistant variety, before sowing the seed will be helpful. Moreover, extensive studies should be undertaken to find out the allergenic fungi over such crop field which are implicated in causing different hypersensitive reactions among agriculture workers.

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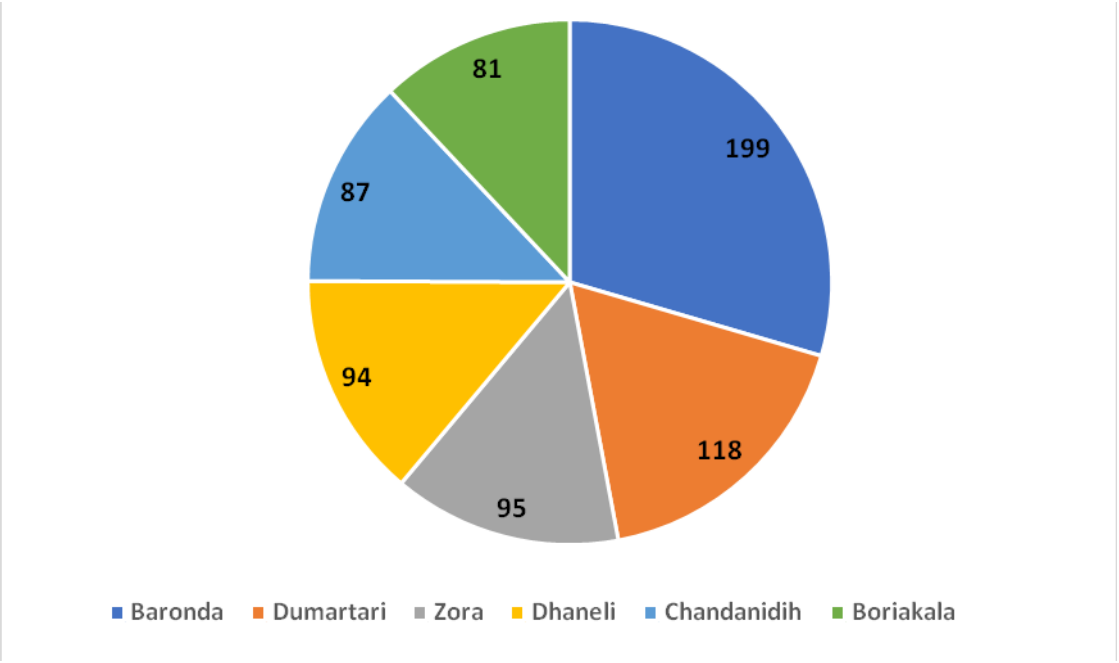

Fig. 1: Occurrence of fungal species in Raipur city

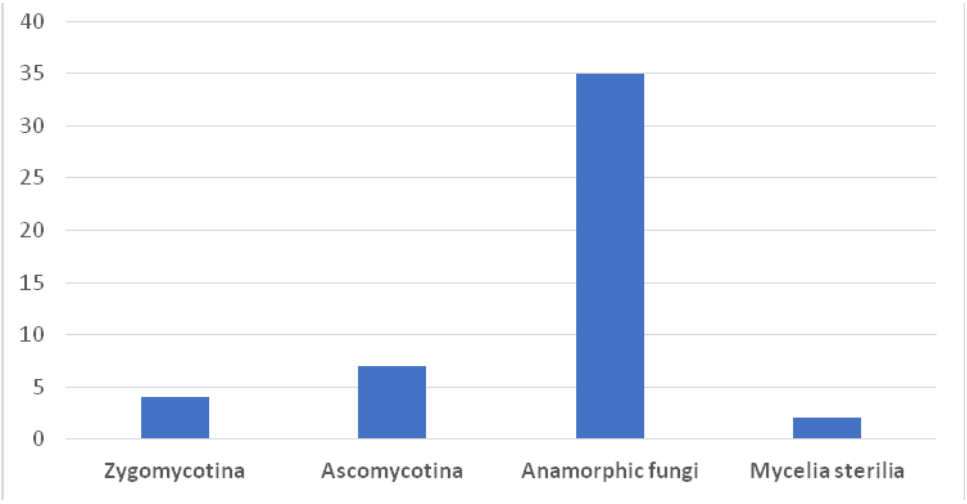

Fig. 2. Classwise distribution of fungal species in Raipur city

QUALITATIVE AND QUANTITATIVE ANALYSIS OF AIRBORNE FUNGAL SPORES AT DIFFERENT HEIGHTS AND GROWTH STAGES OVER A CULTIVATED PADDY FIELD

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Airborne mould fungi are the significant bioaerosols of indoor and outdoor environments at most of the sites. In order to find the solution of the mould risk among atopic human beings and plant disease in the crop fields, we were taken into consideration of this present work to isolate, enumerate and identify the airborne fungal spores by volumetric analysis in the cultivated paddy field at different stages and at ground and 4ft heights by employing Burkard personal sampler on agar plates during 2017-18. During the study period, composition and concentration of fungal species and their distribution considerably varied from heights and stages in the paddy field. In qualitative analysis, altogether 14 fungal species under 6 genera were isolated from the field environments. Quantitative analysis of airborne fungal spores recorded that ground level harboured more spores than 4ft heights. Ground level fungal spores were more (70%) in comparison to 4ft height (30%) in the middle and cutting stages but were found similar in the case of early stages in the paddy field. In composition structure, 4ft height species contribution was found more than ground level in all the stages. Members of Deuteromycotina were mostly prominent in their occurrences in the field environments. It was found that mostly mould spores like aspergilli, penicilli with sterile mycelia, *Fusarium*, *Curvularia* and *Mucor* were recorded from the field. White sterile mycelia and *Aspergillus niger* were found in more numbers. *Penicillium oxalicum* and *Fusarium* sp. were found the dominant fungi in all the stages in the paddy field. Similarity coefficient value showed that the ground level and 4ft height sites of the paddy at early and middle stages were more similar (66.7%) and it was followed by ground level and 4ft height of middle stage of the paddy cultivation (60.0%). The present work would help the plant pathologist and fungal taxonomist those who have interest in analysing the prospect of this work.

Keywords: Airborne fungal spores, cultivated paddy field, Burkard personal sampler

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INTRODUCTION

Majority of the pathogens causing diseases are airborne and make them ubiquitous in their nature. The impact of plant diseases on epidemic scale is enormous and these have far-reaching social implycations[1]. The role of aerobiology in detecting the basic source, release, dispersion and impact of fungal spores on plant surfaces in order to establish the disease on the plants has long been recognized[2]. It is necessary to find out the fungal spore load in the aerial environments of different crops cultivated in India and abroad in order to find the availability of pathogenic fungi in the said environments. So many works have been made worldwide including India to establish the airborne fungi over different crop fields[3-7]. Mehta[3] conducted the experiments with his classical aeroscope and trapped the uredo-spore of three rusts of barley and wheat for detecting the transport of the rust inoculum from hills to plains in India. Wallin and Loonan[4] experimented and found the airborne *Helminthosporium maydis* spores above the corn fields of Amea, Iowa. Couture and Sutton[8]

informed the relation of weather variables and host factors to rate of airborne spores of *Bipolaris sorokiniana* over a barley field in Ontario, Canada. Cammack[5] described differ-rent factors affecting infection gradients of *Puccinia polyspora* while studying the problems relevant to airborne fungal diseases in Africa. In India, Padmanabhan[9] testified that the conidia of *Helminthosporium oryzae* present in the atmosphere acted as primary inoculum to origin the disease over paddy field in Cuttack. Sengupta and Chattopadhyay[10] reported the occurrence of airborne conidia of *Helminthosporium oryzae* over paddy fields near Chinsurah and Uddin[11] ~~(2004)~~ worked on airborne fungal spora in paddy field, West Bengal. Atluri *et al.*[12] studied the circadian periodicity in some airborne fungi over a rice crop in Andhra Pradesh. Singh and Dorycanta[13] studied on Fungal airspora over a maize field in Senapati District of Manipur state. Sreeramulu and Ramalingam[1] conducted a two year survey on the airspora of a paddy field near Visakhapatnam, India. Chakraborty *et al.*[14] reported that the development of brown spot disease is positively correlated with the occurrence of airborne pathogenic conidia of *Bipolaris oryzae* along with the growth stage of paddy for wet season crop in West Bengal.

The present study deals particularly with the environmental biocomponents (mostly airborne fungi) analysis over a cultivated paddy field with employing Burkard volumetric sampler at different heights and stages of the crop. The role of weather in the aerofungal composition and variation would be worked out in detail with reference to each component and comparison with earlier reports. The investigations would further be supplemented in devising useful disease forecasting system for these crops in the region and ultimately in the home states.

MATERIALS AND METHODS

During the present study, quantitative and qualitative analysis of airborne fungal spores was carried out at ground level of the crop and 4ft height (just above the crop) inside the paddy cultivated field of different stages viz., early, middle and cutting times in the premises of Krishi Vigyan Kendra (KVK), Puducherry during 2018-19 by employing Burkard volumetric personal sampler on agar plates.

Sampling of Air

Air samplings were made at 11 AM to 12 noon at ground level and 4 ft above, just above the crop by running the Burkard volumetric personal sampler on agar plates (Plate I) in the cultivated paddy field for five minutes. The media plates (θ = 9cm) con-taining Potato Dextrose Agar (PDA) with strepto-mycin ($50mg^{-1}$) were employed in the Burkard's sampler in the present study to isolate and enumerate the air borne fungal spores. The air samplings were carried out simultaneously in the fields together one after another. After running the sampler for five minutes, the media plates were collected and brought back to the Microbiology laboratory, Department of Botany, Kanchi Mamunivar Govt. Institute for Post-graduate Studies and Research (Autonomous), Puducherry, India, and kept at 25± 3^{o} C upside down for 15 days with constant observation after 3-4 days of incubation in order to visualize the growth of fungal colony forming units (CFUs) and to enumerate them. Growth of fungal colony forming units (CFUs) on the media plates are given in the Plate II. During the sampling time, meteorological parameters like, temperature and relative humidity were recorded together from the indoor environmental sites of the house. The exposure time was standardized to get 50-100 number of fungal colonies/colony forming units (CFUs) per plate. Fungal colonies developed in plates were counted for individual species and to get the total number CFUs.

Identification of fungal spores

Microscopic slides stained with lactophenol cotton blue were prepared from each CFUs and observed microscopically to identify them up to species level. The colony forming units (CFUs) that could not be identified directly from plates were sub cultured in PDA/SDA again and identified later on. The laboratory experience and taxonomic literature were employed to identify the fungal taxa[15-20]. Percentage

Plate I: Air samplings at ground level and 4ft height in the paddy crop field sites.

occurrence of individual fungus was calculated and plotted in the Tables and Figures.

In order to calculate the total number of airborne fungi present in the concerned environments of the study sites, the conversion factor was calculated based on the following principles, since it is mandatory to know the fungal spore load of each environment to assess fungal mass in the said environments. The total number of fungal CFUs counted from the working environment was converted to CFUs m^3 of air.

Observation and calculation of data

Calculation of Percentage contribution of an individual fungus:

$$\% \text{ occurrence of the fungus} = \frac{\text{Total CFUs recorded by the individual fungus}}{\text{Total CFUs recorded by total number of fungi}} \times 100$$

The percentage contribution of spores was found out taking the average of the two readings of each experimental site.

Similarity coefficient analysis

The similarities in fungal composition between two environments were calculated from the recorded data of three sites studied herewith.

By Sorensen's index of similarity or similarity coefficient, the Sorensen's index of similarity was calculated by using the formulae.

Similarity coefficient: $\frac{2W}{a+b} \times 100$

Where, **a:** Total number of species isolated from one site; **b:** Total number of species isolated from other site; **W:** Common number of species.

RESULTS AND DISCUSSION

During the study period, a total number 11920 fungal colony forming units (CFUs) m^{-3} of air were recorded from the paddy cultivated field, of which ground level fungal spores shared the maximum spores in comparison to the aerial (4 ft) height at different stages in the paddy field (Table 1 and Fig. 2). In qualitative analysis, altogether 14 fungal species under 6 genera were isolated from the paddy field environments. Occurrence of airborne fungal spores isolated from two different heights over paddy field is given in Figure 1. Quantitative and qualitative analysis of airborne fungal spores recorded at different heights with their incidence at different stages over the cultivated paddy field is given in Table 1**.**

Ground level fungal spores were more (70%) in comparison to 4 ft height (30%) in paddy field at different stages of the cultivation. Based on the fungal composition, 4 ft height was recorded with more number than the ground level (Fig. 1) but in case of quantitative analysis, ground level harboured a greater number of fungal spores m^{-3} of air (Fig. 2). Among the recorded taxa, members of Deuteromycotina were found as mostly prominent in their occurrences followed by the members of other group in the field environments. It was found that mostly mould spores like Aspergilli and Penicilli, *Curvularia*, *Fusarium* with White sterile mycelia were recorded from the field environments.

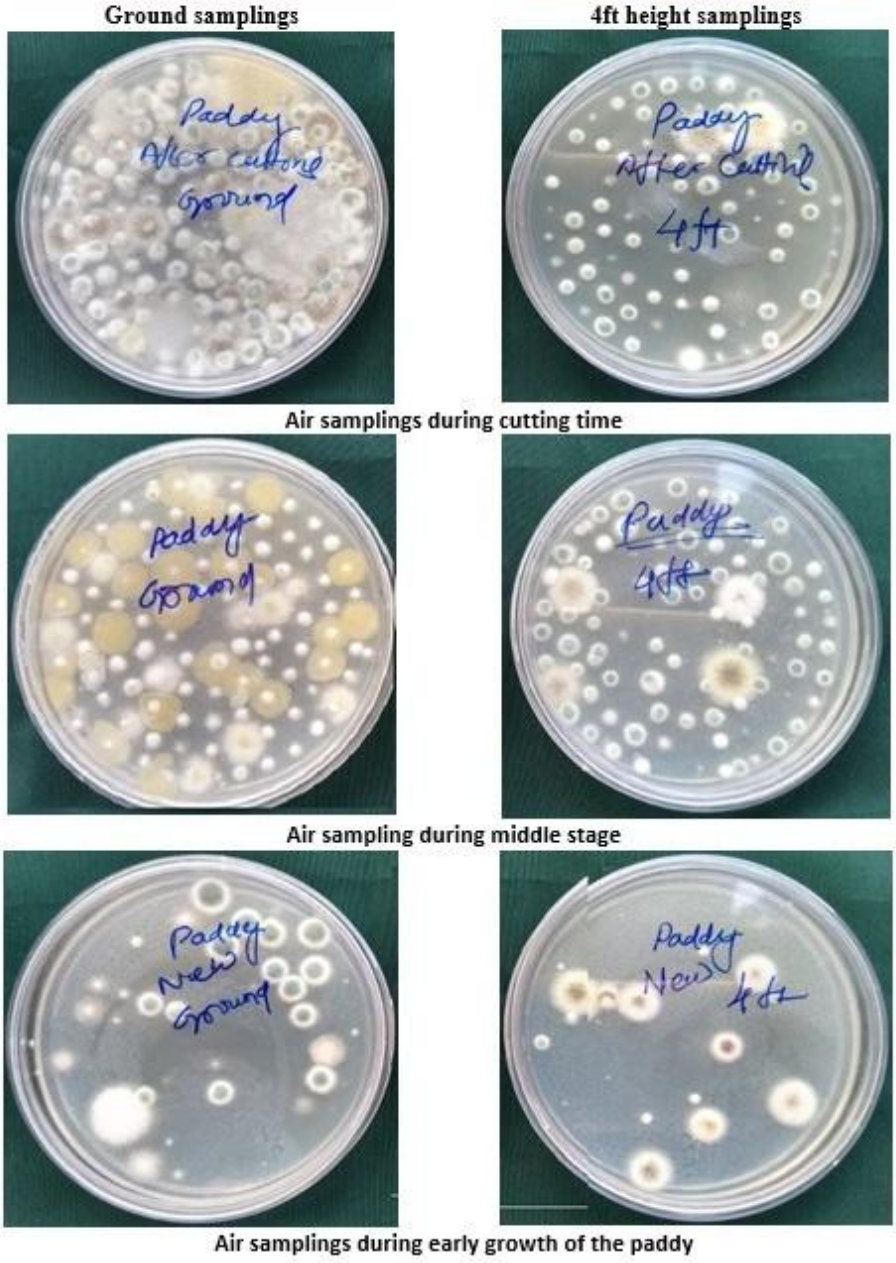

Plate II: Growth of airborne fungal spores (CFUs) on agar plates on the cultivated paddy (*Oryza sativa* L.) field at two heights.

Fusarium sp., *Mucor* and *Penicillium oxalicum* were found as the dominant fungi in paddy field as well as at different stages of the cultivation of paddy (Fig. 3). Percentage of airborne fungal spores at differ-rent heights isolated over paddy field (Table 1) showed different fungi and their occurrence in paddy field at different stages too.

Plant pathogenic fungi viz., *Curvularia* and *Fusarium* were verified but *Helminthosporium* and *Pyricularia* were not recorded in our recent study. It may be attributed that the pathogenic fungi like later ones were not found in the vicinity and heir specific hosts are not available in the environments[4,6,10,11,21].

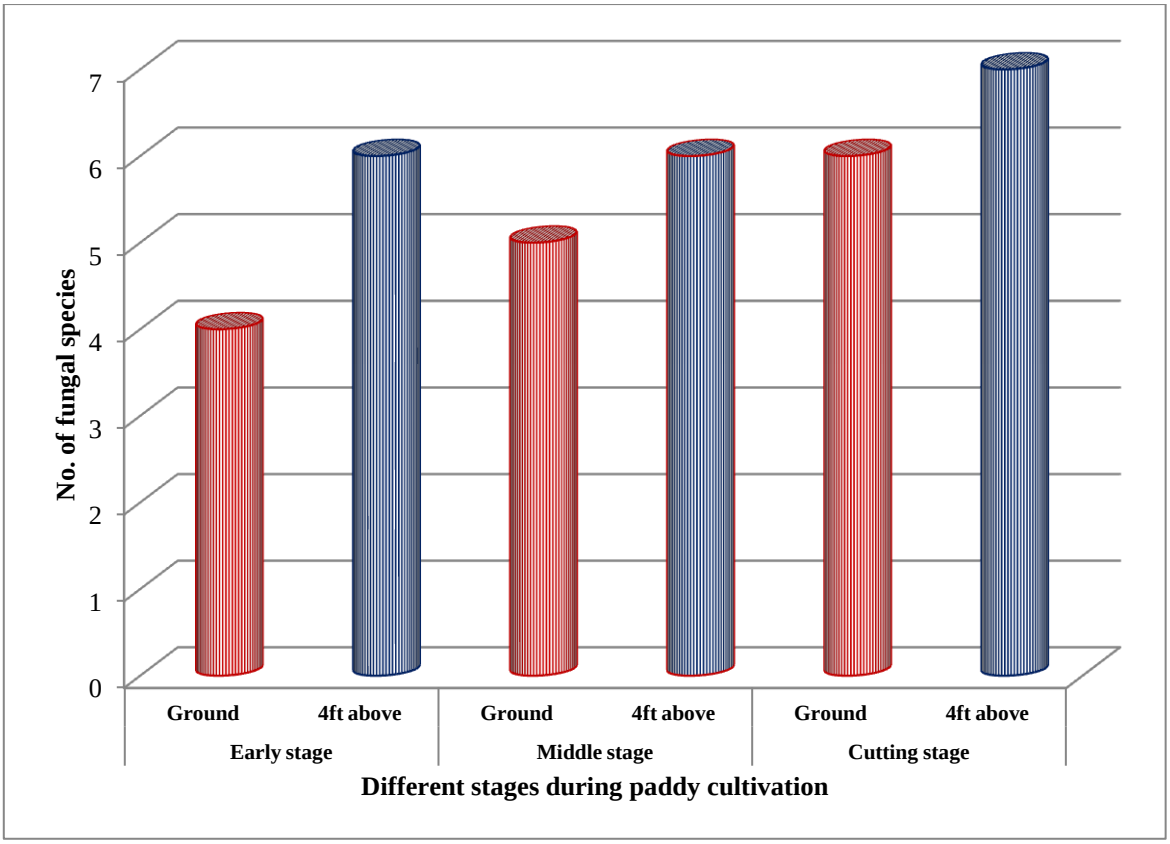

Fig 1: Fungal composition during different stages of paddy cultivation in the field

Uddin[11] (2004) worked on the fungal airspora over paddy field in West Bengal, India. During his work he found *Alternaria, Fusarium, Helminthosporium* and *Nigrospora* as phytopathogenic fungi recorded from the paddy field air, out of which *Alternaria* was recorded as the dominant one. He explained that *Alternaria* was detected as a virulent disease-causing agent in this variety of rice which caused leaf damage to the host plant but in the present study, *Fusarium* was found as one the disease-causing fungus inciting the wilting disease. *Fusarium, Helminthosporium* and *Nigrospora* were found more or less frequently. Fungi those who failed to sporulate were grouped under 'sterile forms' and were found regularly throughout the period of investigation as white sterile mycelia in our work too. Uddin[11] explained that *Cladosporium* was the dominant genus in their work, but in our work, *Penicillium* was the dominant one followed by *Fusarium.*

Similarity coefficient and common airborne fungal species recorded at different heights over the paddy field of various stages is given in Table 2. Similarity coefficient analysis value showed that the ground level and 4ft height sites of the paddy at early and middle stage were more similar (66.7%) and it was followed by ground level and 4ft height of middle stage of the paddy cultivation (60.0%) (Table 2).

Table 1: Percentage occurrence of airborne fungi at different stages and different heights over paddy cultivated field.

Sl. No.	FUNGI	Early stage		Middle stage		Cutting stage	
		Ground	4ft above	Ground	4ft above	Ground	4ft above
1	*Aspergillus awamori*		11.11			15.61	
2	*Aspergillus niger*		27.77		5.95	1.26	1.35
3	*Aspergillus nidulans*		5.55				
4	*Aspergillus oryzae*			7.40		0.84	1.35
5	*Curvularia andropogonis*						4.05
6	*Curvularia* sp.	19.04		1.23	2.38		
7	*Fusarium* sp.	9.52	5.55	32.09	13.79	35.44	21.62
8	*Penicillium oxalicum*	66.67	27.77	43.82	72.61	43.88	66.21
9	*Penicillium fellutanum*	4.76					
10	*Penicillium* sp.		5.55				
11	*Penicillium new* sp.			15.43			1.35
12	*Mucor* sp.					1.26	2.70
13	*Mucor plumbeus*						1.35
14	White sterile mycelia		16.66		4.76	1.68	

In the present study, exposure with Burkard volumetric personal sampler containing agar plates is found quite decent, since it gave us the data of fungal spores per cubic meter per air. On the other hand, the instrument was handy and easy to carry to the places of interest of air samplings. It provided the platform where the settled viable fungal spores germinate and sporulate on the broad-spectrum media and it was easy to enumerate. It facilitated the microscopic study of the colonies and enabled the identification of the species. It was found highly suitable for qualitative study and the result was set forth quantitatively too since it is possible to express volumetrically.

Out of the total isolated fungal species, most of them belong to the members of Deuteromycotina followed by the members of other groups. It was informed by other workers[2,11,22-25] that the airborne fungi mostly dominated by Deuteromycotina or hyphomycetes group of fungi. A number of aspergilli comprises high incidence of *A. awamori*, *A. niger*, *A. nidulans* and *A. oryzae* were reported in the present study similar to findings of many others[7,10,26,27]. Singh[27] worked on aerobiology and diseases of several crops in Manipur, India and their results were found to be similar to the present study.

Table 2: Similarity coefficient index and common number of airborne fungal species isolated from different stages of paddy cultivated field.

		Early stage		***Middle stage***		***Cutting stage***	
		Ground	4ft above	Ground	4ft above	Ground	4ft above
Early stage	Ground	**4***					
	4ft above	1$/20#	**6**				
Middle stage	Ground	3/66.7	1/18.2	**5**			
	4ft above	3/66.7	3/54.6	3/60	**5**		
Cutting stage	Ground	2/40	4/66.7	3/54.5	3/54.5	**6**	
	4ft above	2/36.4	2/30.7	3/50	3/50	4/61.5	**7**

***= Total number of species, #= Similarity coefficient value, $= Common number of species**

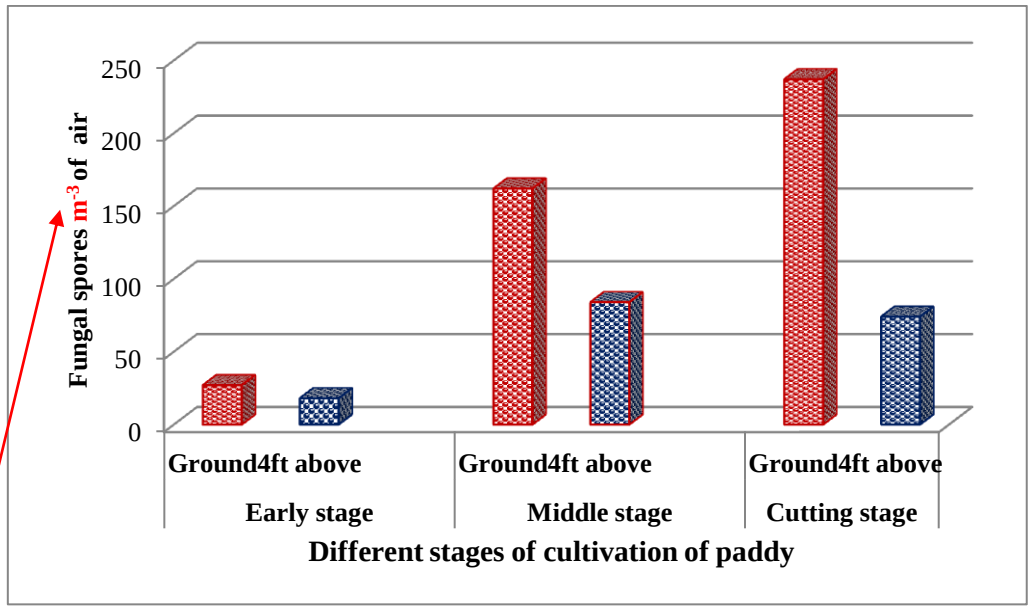

Fig 2: Quantitative structure of fungal spores per m^{-3} of air at different stages of paddy cultivation field

m^{-3} duto eirakom hobe

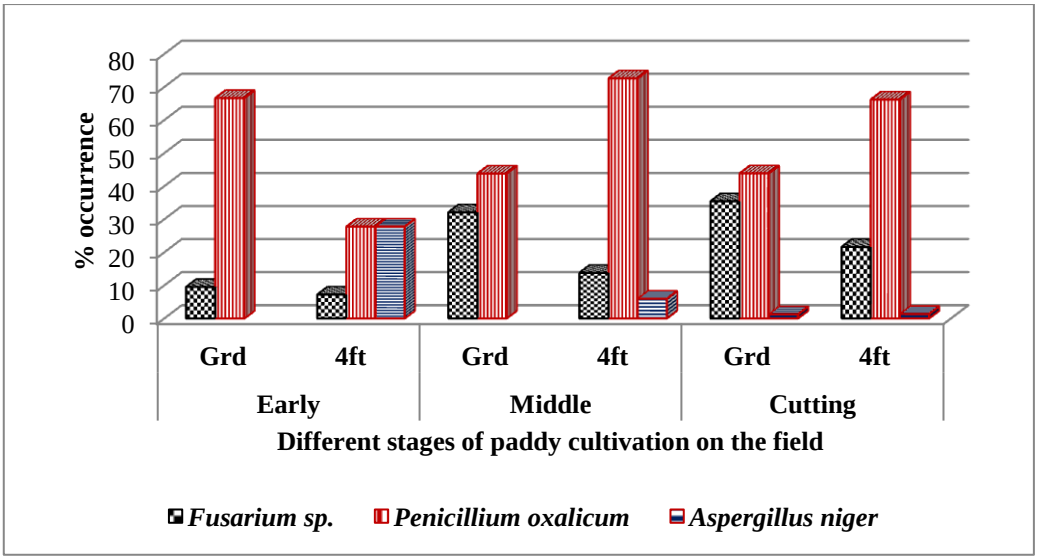

Fig 3: Percentage occurrence of dominant fungi over paddy cultivation field

Recently, attention has been focused on the fungi in relation to plant pathology, human disease and in sick building syndrome[1,11,13,24,28]. However, the presence of fungi in crop and cultivated fields necessarily imply a cause-and-effect relationship with plant diseases[14] and should also alert physicians and healthcare professsionals to do more vigorous environmental testing in order to get the concurrent idea about the airborne fungal plant pathogens and their management.

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CONFLICT OF INTEREST

The authors declare no conflict of interest among the authors.

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POLLEN SPECTRUM OF APIARY HONEY FROM AGRO FORESTRY ZONES OF PRAYAGRAJ DISTRICT, UTTAR PRADESH, INDIA

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The present paper deals with the pollen spectrum of honey collected from different agro forestry zones of Prayagraj district during the 2018. A total 52 pollen types were recorded from honey. Among the eight honey samples, three samples were unifloral types and the remaining five were multifloral in nature. *Ageratum conyzoides, Brassica campestris* and *Coriandrum sativum* were the three predominant types in unifloral honeys. Six pollen types were recorded as secondary pollen types include *Ageratum conyzoides, Brassica campestris, Chenopodium* sp., *Coriandrum sativum, Citrus* sp. and *Psidium guajava*. 24 and 41 pollen types were documented as important pollen type and minor pollen type in frequency class. Qualitative and quantitative pollen analyses in the present study demonstrate that agro forestry regions of Prayagraj district are rich in bee flora with a potential of producing adequate honey and thus can be utilized commercially for a moderate to large scale apicultural venture.

Key words: Honey, agro-forestry, Pollen spectrum, Prayagraj district

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INTRODUCTION

The pollen spectrum of honey indicates the plants visited by honeybees. It is examined by quantitative analysis of pollen grains present in honey. Honey-bees are effective and potential pollinators in agri-cultural and natural ecosystem which can enhance the crop productivity improving the socio-economic status of rural communities. Honeybees depends on both nectar and pollen to sustain themselves but pollen is necessary for the raising the brood[1,2]. The absence of pollen grains can affect the strength of the bee colony and honey production as well[3-6]. The knowledge of the pollen flora of an area is a basic tool for the apiculture. India is one of the largest producers and exporters of honey[7,8].

Agroforestry is a dynamic and sustainable land management system of deliberately growing trees along with agricultural crops on the farmlands to secure both tangible and intangible benefits to the farmers. Uttar Pradesh is one of the largest and densely populated states of India in the Indo-Gangetic Plain with large agrarian communities. Uttar Pradesh shows agro forestry practices. Agro-forestry practices vary according to the agro-climatic zones, land capability and socio-economic status of farmers. Generally plants like *Aegle marmelos, Acacia nilotica, Albizia lebbeck, Annona squamosa, Artocarpus heterophyllus, Azadirachta indica, Bambusa balcooa, Bixa orellana, Bombax ceiba, Callistemon* sp., *Carica papaya, Cassia siamea, Citrus* sp., *Dalbergia sissoo, Dendrocalamus strictus, Delonix regia, Emblica officinalis, Eucalyptus* sp., *Populus* sp., *Pongamia pinnata, Jatropha curcus, Mangifera indica, Psidium guajava, Punica granatum, Madhuca latifolia, Moringa oleifera, Syzygium cumini, Tectona grandis, Terminalia arjuna*, and *Zizyphus* sp. are planted for agro-forestry pro-grams. These plants are utilized as pollen and nectar sources by honeybees[9]. Prayagraj districts of Uttar Pradesh shows agro-forestry practices since long time[10, 11].

Pollen analysis of honey samples from Uttar Pradesh has been investigated by several workers from differ-ent regions viz. Lucknow[12-16], Unnao[17], Shahjahan-pur[18] and Varanasi[19]. Prayagraj has predominantly agricultural area. Prayagraj district has a high poten-tial for honey production not yet exploited. Weeds, pastures and exotic trees are used as bee forage in

agricultural areas. Bee forage plants in Prayagraj have been studied previous workers[20,21]. These previous workers worked on natural bee hives honey samples from rural and urban area. However there are no records on pollen analysis of agro-forestry zones of Prayagraj district Uttar Pradesh. Thus, the present work aims to study the pollen spectrum of apiary honey collected from different agro-forestry zones of Prayagraj district of Uttar Pradesh with a focus on the role of agro-forestry plants in good quality honey production, which can improve the economic status of the beekeepers of the study area.

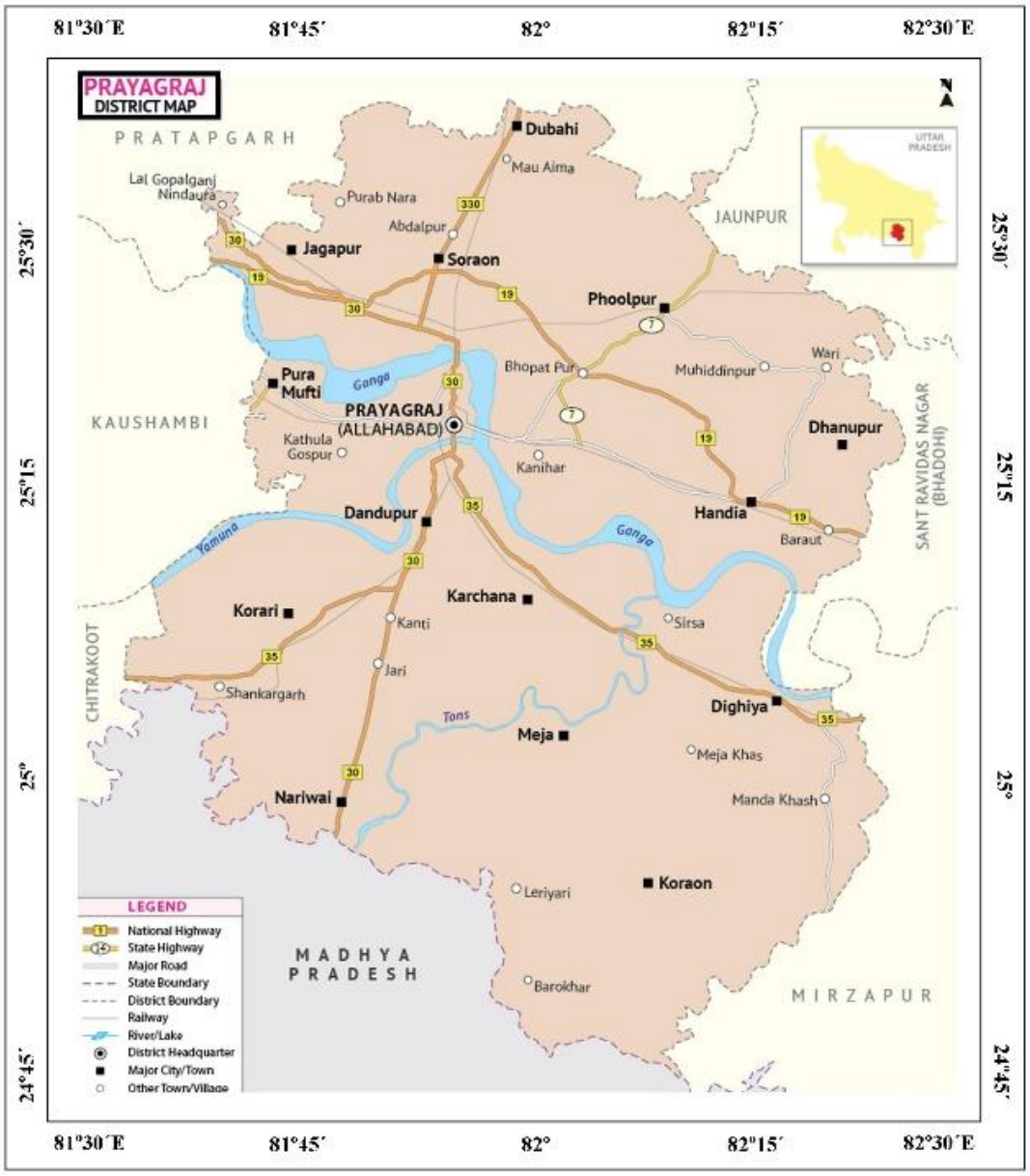

Fig. 1: Map showing the study area in Prayagraj District, Uttar Pradesh.

MATERIALS AND METHODS

Prayagraj is a district of Uttar Pradesh which lies between 24°47' to 25°45' N latitudes and 81°84' to 82°30' E longitudes at a height of about 98 m above sea level (Fig. 1). The agro forestry study sites were selected with the help of the Allahabad Agri-culture Institute and several published literatures[22, 23]. In each agro-forestry zone, one apiary boxes was located with the help of professional beekeepers. Squeezing of the honey comb from apiary boxes was carried out under the personal supervision of the authors.

Eight squeezed honey samples of *Apis cerana indica* were collected from apiary honey boxes collected from agro-forestry zones of Prayagraj district during Janaury to June 2018 (Fig. 2). The honey samples were stored in airtight plastic bottles and labeled. Field surveys were done to survey the plant growing around the sites of honey collection. Flowers were collected during field survey for making reference slides. Reference slides were prepared using Wode-house method[24]. The honey slides were prepared using the Acetolysis method[25]. Qualitative and quan-

titative pollen analysis was based on the methods recommended by International Commission for Bee Botany[26]. Pollen recovered from honey samples were identified by comparing them with the reference pollen slides and published literature[27] [28]. On the basis of quantitative pollen analysis of each honey sample, pollen spectrum was prepared using software from Birbal Sahni Institute of Palaeosciences and identified pollen grains were categorized to various frequency classes[26], such as predominant pollen type (above 45%), secondary pollen type (16-45%), important minor pollen type (3-15%) and minor pollen type (below 3%). On the basis of frequency classes, honey samples with predominant pollen type were categorized as unifloral honey sample. Honey sample with no predominant pollen type categorized as multifloral honey. In total honey samples, frequencies of pollen types were classified as very frequent (>50%), frequent (20–50%), infrequent (10–20%) and rare (< 10%)[29]. Photographs of pollen grains were taken using a Leica DM 2500 microscope with Leica DFC295 camera attachment.

Fig. 2: Apiary boxes located in different agro forestry sites of Prayagraj district, Uttar Pradesh

Table 1. Pollen grains recovered from the honey samples collected from agro forestry site of Prayagraj district Uttar Pradesh.

Locality	Nature of honey	Pollen based on frequency			
		Predominant pollen (> 45%)	**Secondary pollen (16-45%)**	**Important minor pollen (3-15%)**	**Minor pollen (<3%)**
Bara	Multifloral	-	*Brassica campestris*, *Coriandrum sativum*, *Chenopodium* sp.	*Cajanus cajan*, *Holoptelea integrifolia*, *Pisum sativum*, *Murraya koenigii*	*Emblica officinalis*, *Ocimum sativum*, *Madhuca indica*, *Morus alba*, Poaceae, *Prosopis juliflora*, *Ricinus communis*, *Solanum* sp., *Tagetes* sp.
Handia	Unifloral	*Ageratum conyzoides*	-	*Moringa oleifera*, *Brassica campestris*,	*Aegle marmelos*, *Amaranthus* sp., *Feronia limonia* , *Justicia simplex* ,

				Pisum sativum, Cicer arietinum	*Lycopersicon esculantum, Mangifera indica, Prosopis juliflora*
Karchana	Multifloral	-	*Brassica campestris, Coriandrum sativum, Ageratum conyzoides*	*Acacia* sp., *Madhuca indica, Syzygium cumini, Tagetes* sp.	*Azadirachta indica, Cicer arietinum, Datura stramonium, Feronia limonia, Pisum sativum, Ricinus communis, Sonchus arvensis, Tecoma stans, Terminalia* sp., *Ziziphus* sp.
Koraon	Unifloral	*Coriandrum sativum*	-	*Brassica campestris, Datura stramonium, Prosopis juliflora*	*Acalypha indica,Cannabis sativa, Cicer arietinum, Datura stramonium, Feronia limonia, Hygrophila auriculata, Madhuca indica, Ocimum sativum,* Poaceae
Meja	Multifloral	-	*Brassica campestris, Psidium guajava, Coriandrum sativum*	*Amaranthus* sp., *Azadirachta indica, Cajanus cajan*	*Justicia simplex, Delonix regia, Emblica officinalis, Cannabis sativa, Raphanus sativus, Solanum* sp., *Ziziphus* sp.
Phulphur	Multifloral	-	*Ageratum conyzoides, Brassica campestris , Citrus* sp.	*Eucalyptus citriodora, Psidium guajava, Prosopis juliflora*	*Amaranthus* sp., *Combretum indicum, Callistemon* sp., *Ficus* sp., *Chrysanthemum* sp., *Hibiscus rosa-sinensis, Lathyrus sativus, Lycopersicon esculentum, Murraya koenigii,* Poaceae, *Solanum* sp., *Tinospora cordifolia*
Prayagraj	Multifloral	-	*Ageratum conyzoides, Brassica campestris, Citrus* sp., *Psidium*	*Holoptelea integrifolia, Syzygium cumini, Cassia fistula, Bombax*	*Acacia* sp., *Acalypha indica, Aegle marmelos, Callistemon* sp., *Chenopodium* sp., *Eucalyptus*

			guajava	*ceiba*, *Lathyrus sativus*	*citriodora*, *Feronia limonia*, *Hibiscus rosa-sinenesis*, *Parthenium hysterophorus*, Poaceae, *Tecoma stans*
Soraon	Unifloral	*Brassica campestris*	-	*Coriandrum sativum*, *Ficus* sp., *Moringa oleifera*, *Terminalia* sp., *Raphanus sativus*	*Bombax ceiba*, *Delonix regia*, *Emblica offiicinalis*, *Hygrophila auriculata*, *Mangifera indica*, *Parthenium hysterophorus*, *Pisum sativum*

RESULTS

In the present study 52 pollen types belonging to 23 families were identified from the eight honey samples, collected from different agro-forestry site of Prayagraj district, Uttar Pradesh during six months (January-2018 to June-2018). The pollen analytical data of

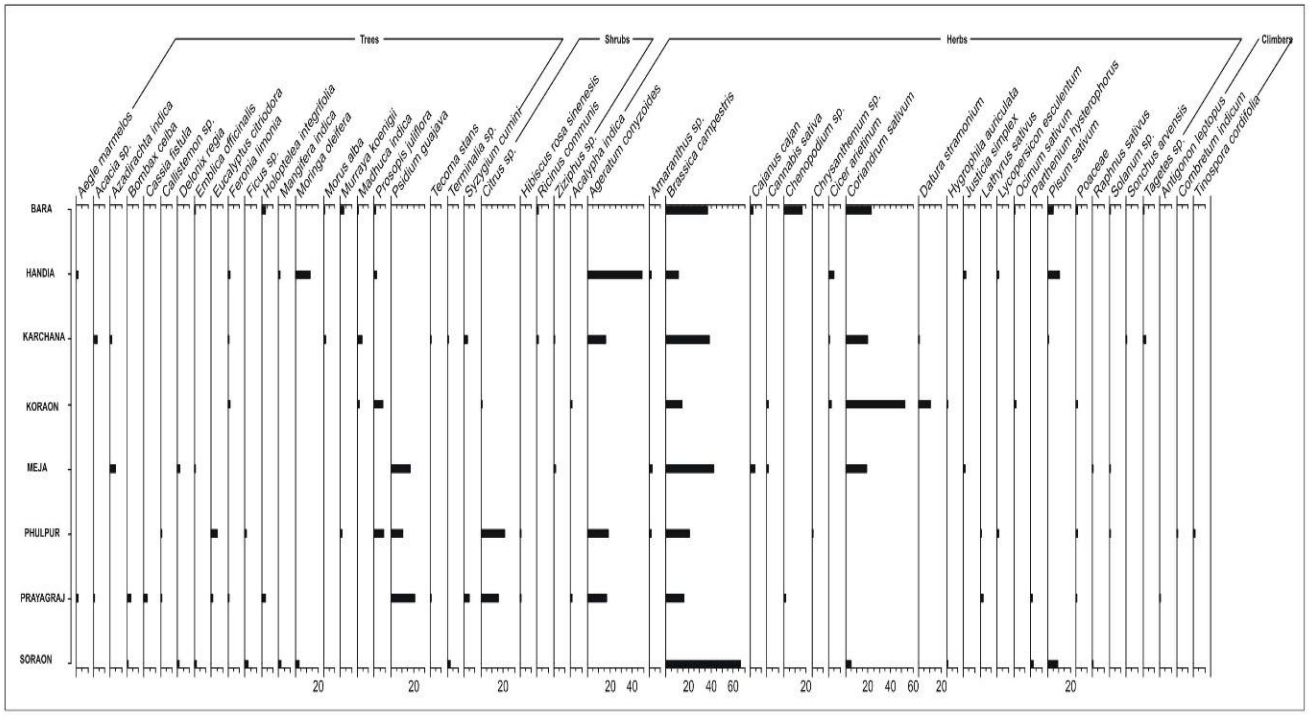

Fig. 3: Pollen spectra of apiary honey collected from agro forestry site of Prayagraj district, Uttar Pradesh.

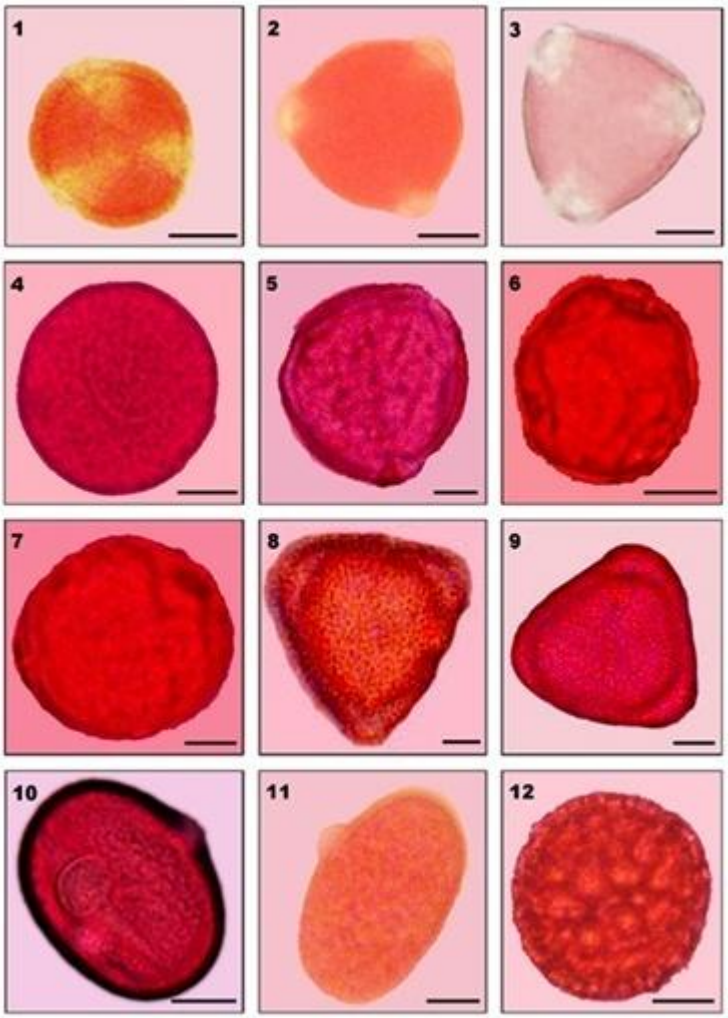

Fig. 4: Photomicrographs (Scale bar = 20µm) of selected pollen grains recovered from the honey of agro forestry site of Prayagraj district, Uttar Pradesh.
(1) Brassica campestris; (2) *Cajanus cajan*; (3) *Cicer arietinum*; (4) *Azadirachta indica*; (5) *Tecoma stans*; (6) *Combretum indica*; (7) *Murraya koenigii*; (8) *Syzygium cumini*; (9) *Callistemon* sp.; (10) *Lathyrus* sp.; (11) *Pisum sativum*; (12) *Delonix regia*.

honey samples are presented in the Table 1. The percentage frequencies presented in pollen spectrum form of the pollen type recovered from the samples are given in Figure 3. Figure 4 depicts the pollen grains recovered from the honey samples from all the studied agro forestry area. Pollen spectra of regional honey samples varied according to the vegetation types utilized by the honey bees with the agro-forestry diverse regions. Three honey samples collected from Handia, Koraon and Soraon were found to be unifloral and remain five honey samples were recorded as multifloral honey. *Ageratum conyzoides, Coriandrum sativum* and *Brassica campestris* are predominant pollen type in unifloral honey samples. In the multifloral honey, six pollen types were constituted the secondary pollen types while 24 and 41 pollen types were recorded as important pollen type and minor pollen type respectively. *Ageratum conyzoides*, *Brassica campestris* and *Coriandrum sativum* were very frequently present in occurrence. Pollen was collected mainly from entomophilous (48 plant taxa) along with the anemophilous (3) and amphiphilous (1) taxa. Anemophilous taxa were *Chenopodium* sp., *Holoptelea integrifolia* and Poaceae while amphiphilous taxa were *Eucalyptus citriodora*. Highest number of pollen types (n = 20) was recorded in the sample Prayagraj and lowest number of pollen type (n = 12) in Handia. The main plant families were documented such as Fabaceae (8 plant taxa), Asteraceae (5), Myrtaceae (4) and Rutaceae (4) (Fig. 5). The main flower color preferred by honeybees for bee forage is white (21 plant taxa) followed by yellow (11 plant taxa) and red (5 plant taxa) (Fig. 6).

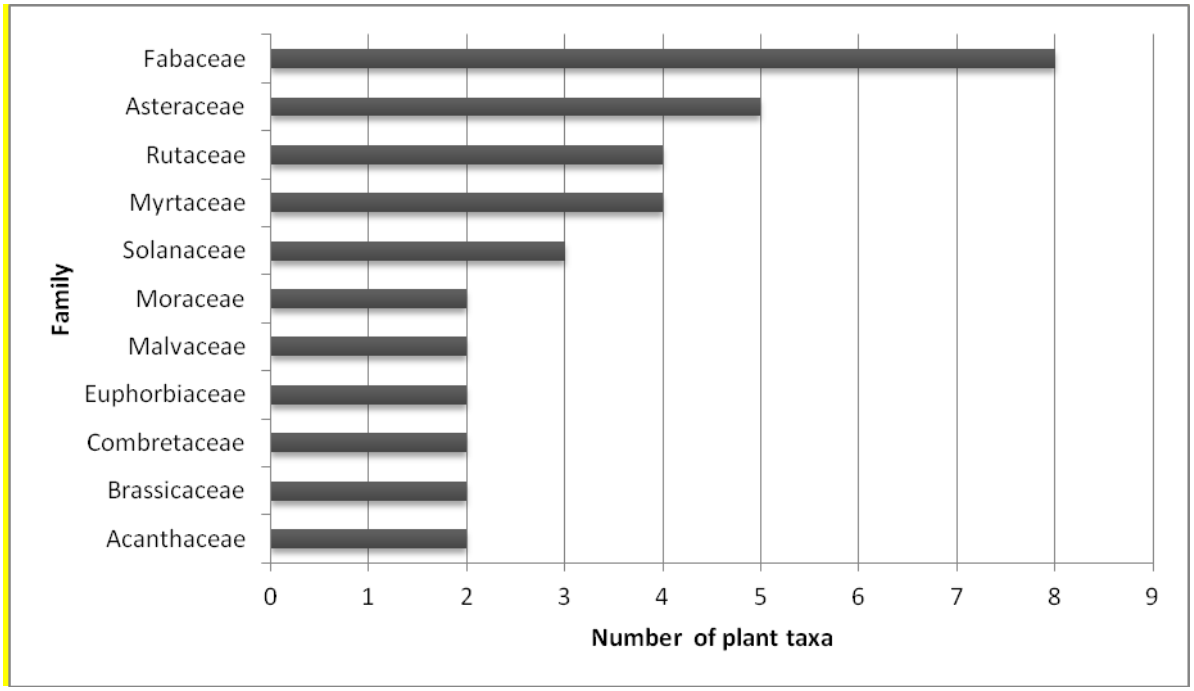

Fig. 5: Main bee forage plant families of Prayagraj district, Uttar Pradesh.

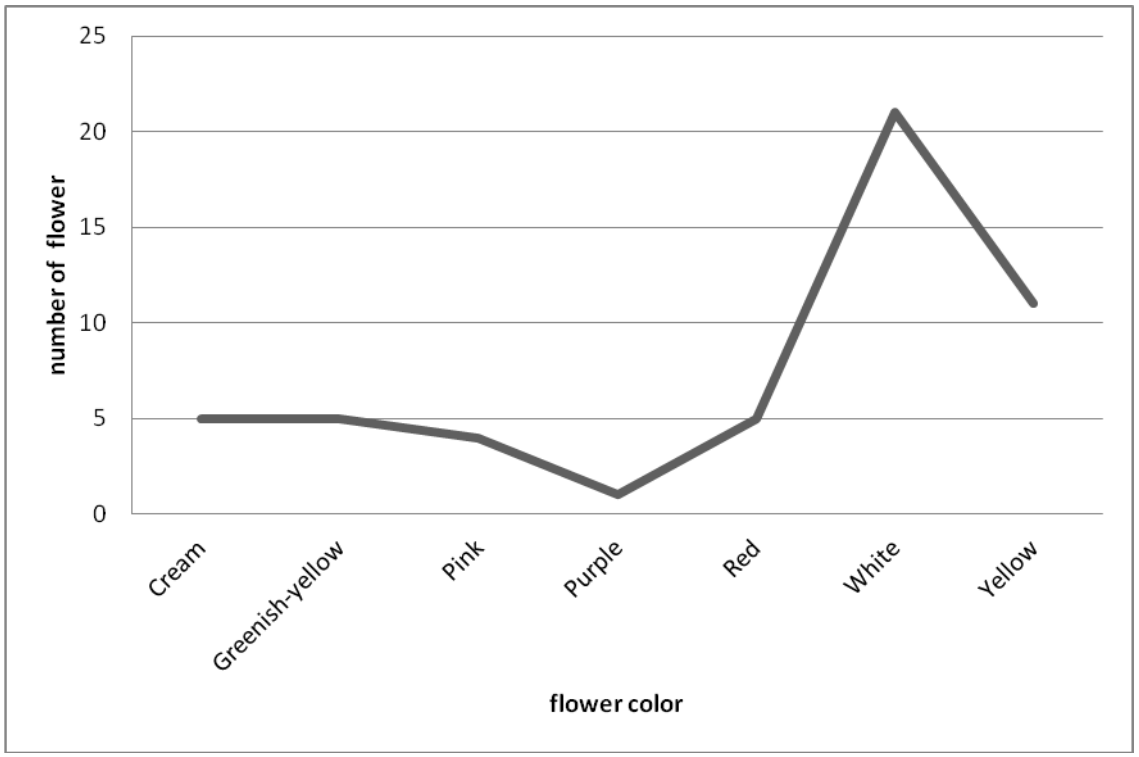

Fig. 6: Line graph showing colour of bee forage plants.

DISCUSSION

The present study provides new insights into the pollen composition of apiary honey samples collected from agro forestry site of Prayagraj district Uttar Pradesh. The pollen spectrum analysis revealed that *Ageratum conyzoides, Coriandrum sativum* and *Brassica campestris* are predominantly nectar and pollen sources in agro-forestry area of Handia, Koraon and Soraon respectively. These plant taxa are chiefly visited by the honey bees. Similar observations have been made by previous workers[20, 21]. In the multifloral honey, six pollen types were constituted the secondary pollen types. Four species of them are herbs: *Ageratum conyzoides*, *Brassica campestris*, *Chenopodium* sp. and *Coriandrum sativum* while two of them are shrubs and trees: *Citrus* sp. and *Psidium guajava*. 24 and 41 pollen types were recorded as important pollen type and minor pollen type respectively. The important and minor pollen types comprises diverse plant species in which

Acacia sp., *Aegle marmelos, Azadirachta indica, Bombax ceiba, Cassia fistula, Delonix regia, Emblica officinalis, Eucalyptus citriodora, Feronia limonia, Ficus* sp., *Holoptelea integrifolia, Madhuca indica, Mangifera indica, Moringa oleifera, Morus alba, Murraya koenigii, Prosopis juliflora, Psidium guajava, Syzygium cumini, Tecoma stans,* and *Terminalia* sp., are trees and *Acalypha indica, Amaranthus* sp., *Brassica campestris, Cajanus cajan, Callistemon* sp., *Cannabis sativa, Chenopodium* sp., *Chrysanthemum* sp., *Cicer arietinum, Combretum indicum, Coriandrum sativum, Datura stramonium, Hygrophila auriculata, Justicia simplex, Lathyrus sativus, Lycopersicon esculantum, Ocimum sativum, Parthenium hysterophorus, Pisum sativum,* Poaceae, *Raphnus sativus, Solanum* sp., *Sonchus arvensis, Tagetes* sp. and *Tinospora cordifolia* are herbs and *Citrus* sp., *Hibiscus rosa-sinenesis, Ricinus communis* and *Ziziphus* sp. are shrubs.

Brassica campestris was the most present type and best represented in the pollen spectra. *Brassica* sp. has also been reported as very frequent in occurrence by various workers in different region of India[20, 21, 30, 31]. *Ageratum conyzoides* and *Coriandrum sativum* were also registered as very frequent in occurrence in the present study. Similar observations have been documented by other workers[31, 32].

The vegetation cover in this area is thus diverse enough to sustain bee colonies. Local vegetation plays a significant role as to which pollens are collected by bee and it is possible to produce several unifloral honeys in a region with great plant diversity[31, 33]. This indicates that when there is a large diversity of plant resources available honey bees tends to focus their foraging efforts on a few species because the preferred sources are more abundant or because they provide specific nutrients that colonies need at a particular time.

The family Fabaceae consist of 8 species which is highest; followed by Asteraceae 5 species; Rutaceae and Myrtaceae (4 species); Solanaceae consist of 3 species; Acanthaceae, Brassicaceae, Combretaceae, Euphorbiaceae, Malvaceae and Moraceae (2 species each); and the remaining families Amaranthaceae, Anacardiaceae, Apiaceae, Bignoniaceae, Cannabaceae, Chenopodiaceae, Lamiaceae, Meliaceae, Menispermaceae, Moringaceae, Phyllanthaceae, Poaceae, Polygonaceae, Rhamnaceae, Sapotaceae and Ulmaceae were represented by single species each. The main bee forage plants belong to family Fabaceae and Asteraceae in the study area. Similar observations had also been made by previous workers[8, 20, 21].

The flower color preferred by honeybees is played important role in bee foraging for honey productions. In the present study, white color flower was found to be more foraged by honeybees as compared with the other color. The yellow flowers were found to be in the family Asteracese (3 plant species), Fabaceae (3), Brassicaceae (1), Bignoniaceae (1), Combretaceae (1) and Solanaceae (1). The white flowers were seen in Asteraceae, Fabaceae, Moraceae, Myrtaceae, Rutaceae and Solanaceae, which were represented by two species each; while Amaranthaceae, Meliaceae, Chenopodiaceae, Apiaceae, Lamiaceae, Poaceae, Brassicaceae, Euphorbiaceae and Rhamnaceae were represented by single species. The pink color flowers were present in the family Acanthaceae, Combretaceae, Polygonaceae, and Myrtaceae, each representted by single species. Red color flowers were found in Malvaceae with two species and Fabaceae, Myrtaceae and Rutaceae with single species. The families, Anacardiaceae, Cannabaceae, Fabaceae, Moringaceae and Sapotaceae were represented by single species each with cream flower color. Five families Euphorbiaceae, Phyllanthaceae, Menispermaceae, Rutaceae and Ulmaceae were documented with greenish yellow flower. The purple flower was found in Acanthaceae with single species. Colour of flower was found to play an important role during bee foraging by honeybees[34].

Prayagraj district is characterized by having a large variety of plants. Agro forestry cultivated plants like *Aegle marmelos*, *Acacia* sp., *Azadirachta indica*, *Bombax ceiba*, *Callistemon* sp., *Cassia* sp., *Citrus* sp., *Delonix regia*, *Emblica officinalis*, *Eucalyptus* sp., *Mangifera indica*, *Psidium guajava*, *Madhuca indica*, *Moringa oleifera*, *Syzygium cumini*, *Terminalia* sp., and *Zizyphus* sp. are documented as secondary, important and minor pollen types in multifloral honey. Agro forestry cultivated plants play important role as chief bee forage plants in the honey production in the study area. Similar reports have been made by previous workers[9]. They men-

tioned agro forestry cultivated plants are utilized as pollen and nectar sources by honeybees.

CONCLUSION

Qualitative and quantitative pollen analyses in the present study demonstrate that these agro forestry regions of Prayagraj district are rich in bee flora with a potential of producing adequate honey and thus can be utilized commercially for a moderate to large scale apicultural venture. This study will be useful to beekeepers for identifying the pollen and nectar sources to honey bees and is also important for its maximum exploitation.

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INVESTIGATION ON CULTURABLE AEROMYCOFLORA OF UDAIPUR, TRIPURA

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Airborne fungal spores are the causative agent of different diseases triggering respiratory allergy among susceptible individuals. The present study was aimed to identify the airborne fungal spores of different environments of Udaipur of Tripura state. Both viable and non-viable spore counting methods were employed in this study. Altogether 20 different culturable fungal spores were recorded in the air of Udaipur. The results revealed highest number of fungal spores in the library and lowest in the classroom environment. The predominance of *Aspergillus, Curvularia, Rhizopus, Alternaria, Cladosporium* in all the surveyed environments has been attributed to their ability to grow in various substrata. The occurrence of huge number of cellulolytic fungi such as *Aspergillus glaucus, A. niger, A. fumigates, A. ochraceus, Penicillium brefeldianum* and *P.oxalicum* in the library environment ascribed to their ability to decay books, journals and manuscript.

Keywords: Airborne, fungal spores, culturable, Udaipur, Tripura

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INTRODUCTION

Air carries innumerable, invisible biomass or particulate matter of plant origin which remains suspended in the air. These bio-particulate matters include fungal spores, pollen grains, algae, moss and fern spores, etc. Fungal spores constitute a signi-ficant fraction of bioaerosol[1], and their spore size varies from 3 µm to 30 µm in diameter. The amount of bio-particulate matters differs quantitatively and qualitatively at different times of a day depending on weather and seasonal conditions, geographical location and local sources[2]. Some airborne phyto-pathogenic fungi including *Fusarium*, *Alternaria*, *Ustilaginoidea* infect various plants and cause economic loss[3].

Studies carried out in India so far revealed aerospora in outdoor environments. The indoor environment also plays an important role and inhalation of certain airborne spores and other constituents of aerospora within the houses of both well and ill ventilation are now widely accepted as one of the important causes of respiratory disorders[4,.5]. This study will indicate the areas which are drastically affected by fungal spores so that we can take precautionary measures. We spend more time in indoor places than outdoor. So it is very important to know whether College buildings and library, Poultry farms, different small-scale industries, etc are polluted by fungal spores or not. If yes, it is too much necessary to identify the fungal spores and take protective measures. Scientists gave attention to this topic both in developed and developing countries[6-10]. Till now no such research work has been done in Tripura - an important state of North-Eastern part of India. Most of the labours who work both in the outdoor as well as indoor areas of Tripura remain under the B.P.L. (below poverty line) category. They have no such money for their treatment. So, it is very important to know for their health whether the places are affected by fungal spores or not. For young students, this investigation is also very urgent and necessary.

The objective of this study is to monitor the occurrence of culturable airborne fungi in outdoor and indoor occupational environments to explore the species composition of airborne fungal spores in different distinct places at Udaipur, Tripura state, India. A major area of this district falls under rural areas. This will perhaps be the first report from the Tripura state.

MATERIAL AND METHODS

Aerobiological sampling was carried out in the following sites:

[1] An industrial area of Dhajanagar, Udaipur,

[2] A classroom of Netaji Subhas Mahavidyalaya (college),

[3] Library of Netaji Subhas Mahavidyalaya,

[4] A Poultry farm of Udaipur.

In the above-mentioned sites, samples were collected throughout the year, firstly in winter followed by post-winter, summer, monsoon and autumn. For each site, sampling was done using both Burkard Personal sampler and Petriplate method. Burkard sampler was operated for 10 minutes and petriplates were exposed for 5 minutes in a fortnight intervals[11].

Study of aerospora

After air sampling the exposed slides were taken out from Burkard sampler and mounted on 18 sq. mm. coverslip. Then the slides were observed under standard optical microscope using 10x, 40x magnification.

Exposure of agar plates and study of viable fungal spores

Study of viable fungal spores was performed by exposing the agar plates for 5 minutes at different sampling sites[12]. These petriplates were incubated at 27˚C ± 2˚C for three days and then the sporulating fungal colonies were identified following standard manuals[13]. Some colonies were further sub-cultured to facilitate their identification up to species level.

RESULTS AND DISCUSSION

Airborne fungal spores in the indoor and outdoor environments

From indoor and outdoor environments 20 viable fungal species including Mycelia Sterilia were recorded (Table 1). The most dominant colony forming genus was *Aspergillus*. The species without any sporulating stage has been categorised as Mycelia Sterilia. Six species of *Aspergillus* were found to be dominant in all the study sites (Fig.1). Individually, *Aspergillus niger* constituted an average of 13.25%, followed in the degree of preference by Mycelia sterilia (12.5%), *Aspergillus flavus* (11.0%), *Cladosporium cladosporoides* (10.0%), *Aspergillus glaucus* (8.15%), etc. (Table 1) . Some of the fungi were poorly represented such as *Nigrospora*, *Syncephalastrum*, *Trichoconis* and *Candida*.

Among the four study sites, maximum fungal colonies (8560 CFU) were recorded from Library, followed by Poultry (7865 CFU), Industrial area (6846 CFU) and Class room (4628 CFU) (Table 1). The highest concentration of fungi in library is perhaps due to the presence of cellulolytic fungal species namely, *Aspergillus glaucus, Aspergillus niger, Aspergillus fumigates, Aspergillus ochraceus, Penicillium brefeldianum* and *Penicillium oxalicum*,which decay books, journals and manuscripts of the library. The cellulase activity of these fungal species was confirmed earlier by Karak et al[16]. The occurrence of huge cellulolytic fungi in college library was by reason of huge number of old and new books, journals and manuscripts with no regular maintaining system. Poultry farm contributed second highest concentration of airspora (7865 CFU) due to availability of huge amount of poultry feed as well as faecal materials of poultry animals which served as good sources of fungi. In the industrial area, moderate concentration of airborne fungi was recorded (6846). Various substrata and garbages contributed a good source for growth of fungi. Among all the indoor environments, spore concentration is lowest in college classroom because of regular cleaning, proper sanitisation and maintenance of the rooms. Among the poorly represented fungi, *Nigrospora* was absent from both class room and library. No record of *Trichoconis* was observed in class room environment and similarly *Candida* from library (Fig. 1). *Syncephalastrum* was only recorded from industrial area. This study is in accordance with the previous studies of indoor fungal spores by Beggs[14] and Oliver et al.[15] and of outdoor fungal spores by Decco et. al.[6] and Takahasi[7].

Table 1: Aeromycology of culturable fungi in Udaipur, Tripura

Sl. No.	Major fungal species	Incidence of fungi (%) in Study sites				
		Industrial area	Class room	Library	Poultry	Average % (species wise)
01.	*Aspergillus flavus*	7.50	9.50	12.00	15.00	**11.0**
02.	*A. fumigates*	8.50	7.50	9.50	5.00	**7.5**
03	*A. glaucus*	7.00	6.50	11.00	8.00	**8.15**
04.	*A. japonicas*	1.00	1.50	5.00	2.00	**2.4**
05.	*A.oryzae*	1.00	2.50	3.50	2.00	**2.25**
06.	*A.niger*	9.50	10.50	15.00	18.00	**13.25**
07.	*Aspergillus* sp.	7.00	3.50	3.00	2.00	**3.95**
08.	*Candida* sp.	1.00	1.50	—	2.00	**1.1**
09.	*Cladosporium* sp.	2.00	3.50	2.00	0.50	**2.0**
10.	*C. cladosporoides*	11.50	8.50	12.00	8.00	**10.0**
11.	*Curvularia lunata*	7.00	1.50	0.50	3.50	**3.1**
12.	*Curvularia pallescens*	2.50	3.50	1.00	3.00	**2.5**
13.	*Fusarium* sp.	6.50	6.00	0.50	3.00	**4.0**
14.	*Nigrospora* sp.	1.00	—	—	0.50	**0.4**
15.	*Penicillum oxalicum*	2.50	4.50	3.00	2.00	**3.0**
16.	*P. brefeldinum*	5.00	6.50	8.00	7.00	**6.6**
17.	*Rhizopus stolonifer*	8.00	6.50	0.50	6.00	**5.25**
18.	*Syncephalastrum* sp.	0.50	—	—	—	**0.4**
19.	*Trichoconis sp.*	0.50	—	0.50	1.50	**0.65**
20.	Mycelia sterilia	10.50	16.50	13.00	10.00	**12.5**
Total No. of spores (CFU)		**6846**	**4628**	**8560**	**7865**	**27,899**

Seasonal Variations

As per earlier records temperature and humidity are the two important factors for fungal growth[2,5]. In the present investigation during monsoon period the growth of *Aspergillus* was highest. But in winter season the rate of colony growth was delayed than other seasons (data not provided). The genus *Rhizopus stolonifer* was found especially in the monsoon period. Spores were usually in least number in dry season. The genus *Cladosporium* was more or less equal in number in all season. In this investigation, *Fusarium*, *Curvularia*, *Penicillium*, *Rhizopus* and *Cladosporium* were recorded and according to Shivpuri[17], the above mentioned fungi were allergenic. These allergenic fungal spores are regularly and continuously inhaled by human being and these spores are important causative agent of respiratory allergy[1].

CONCLUSION

One year monitoring of air showed that a total of 20 different types of culturable fungal spores were recorded for the first time from Tripura state. Among them *Aspergillus* spp. and *Cladosporium* spp. were most common and mostly recorded in maximum concentration from college library. These fungi undergo cellulolytic activity and ultimately result in the decay of old and new books, journals and manuscripts.

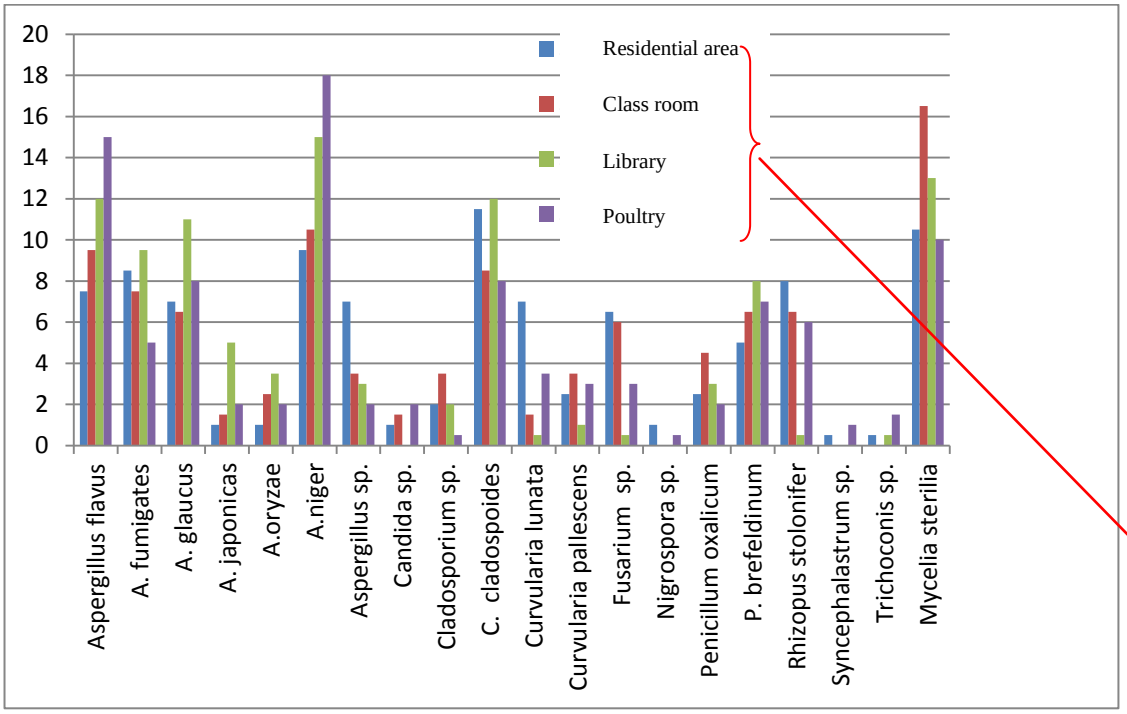

Fig.1: Airborne culturable fungi of four selected sites of Udaipur, Tripura

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FLOWERING PHENOLOGY, POLLEN MORPHOLOGY AND NECTAR-SECRETION DYNAMICS OF THREE SELECTED MANGROVE PLANTS OF SUNDARBAN BIOSPHERE RESERVE, WEST BENGAL, INDIA

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The Sundarban Biosphere Reserve — one of the largest mangrove forests of the world and the second largest Biosphere Reserve (SBR) in India – is located on the southern perimeter of West Bengal. The delta of Sundarban holds most widespread halophytic mangrove forest with rich reservoir of flora and fauna because of its extremely dynamic physicochemical factors. The present study was conducted in Lothian Wildlife Sanctuary, a model tiger free Island of SBR covering an area of 36 sq.km. Most of the true mangroves and mangrove associates are present in this Island. So, it provides a valuable opportunity to understand the plant diversity related study. The present study was aimed to know the flowering phenology, pollen morphology and nectar-secretion dynamics of three mangrove plants viz, *Avicennia officinalis*, *Bruguiera gymnorrhiza* and *Sonneratia apetala*. This study help to understand the importance of these three mangrove plants as honey yielding plants.

Keywords: Mangroves plants, Sundarban, flowering phenology, pollen, nectar secretion dynamics.

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INTRODUCTION

Indian Sundarban is situated within the longitude 88°37' E -89°09' E and 21 °31' N-21°53' N, under the jurisdiction of both North and South 24 Parganas districts of West Bengal. Total length of the coast line in Indian Sundarbans is about 120 km. and it is spreading over 50 km to 70 km inside the main land. At present Indian Sundarban is a Biosphere Reserve comprising three wildlife sanctuaries (Sajnekhali wildlife sanctuary, Lothian wildlife sanctuary and Halliday island wildlife sanctuary).

The Sundarban Biosphere Reserve - rich in mangrove vegetation - is highly productive and provides a safe habitat for bees. Hence the associations between bees and mangroves have been exploited by humans for thousands of years. Lothian Wildlife Sanctuary was chosen as our experimental station, because the Royal Bengal tigers are not present in this island, thus it represents itself as a model Island for flora and fauna. Most of the true mangroves and mangrove associates are present in this Island. So, it provides a valuable opportunity to understand the plant diversity of mangroves.

The present project aims to study the nectar secretion dynamics and the honey production potential of three selected mangrove plants namely, *Avicennia officinalis* L., *Bruguiera gymnorhiza* (L.) Lam. and *Sonneratia apetala* Buch-Ham., which are found to be important for both in pollination perspective and honey production potential.

MATERIAL AND METHODS

(a) **Study site:** The study was conducted in the Lothian Wildlife Sanctuary of Sundarban which is situated at 21°34′45″ N and 88°16′06″ E, covers an area of 38 sq.km and is surrounded by the river Saptamukhi and the Bay of Bengal.

(b) **Flowering period distribution:** For each species, the peak flowering time was taken into account when more than 50% of flower buds were in blooming stage[1].

(c) **Flowering Phenology:** Phenology is the timing of biological events such as growth and repro-

duction. Genes and their interaction with the environment determine the timing and duration of these events. Environmental influences on the timing of growth and reproduction events allow organism's flexibility to adjust to spatially and temporally variable environments. For the study of flower phenology of three plant species, each with ten flower head buds per plant were studied which was monitored every two hours from 04.00-18.00hr. The time of opening of flowers and maximum pollen release were observed and continued until the flowers are wilted[2].

(d) **Pollen morphology:** Reference pollen slides were prepared by acetolysis technique[3]. The identification of pollen grains was done in consultation with the standard literatures such as Erdtman[4] and Bhattacharya et al[5].

(e) **Determination of nectar-secretion dynamics:** Nectar secretion and its dynamics were determined for each plant species following Farkas and Orosk-Kovack[6]. Five mature flower buds were randomly selected and bagged one day before their flower opened. Then the nectar was collected by using micropipette, and the total mean volume was calculated, and nectar sugar concentration was measured by Refractometer.

RESULTS

(a) **Flowering period distribution:** The flowering periods of three plant species are varied. All the plant species (Plate 1) showed a common blooming period from March to July. However, *Bruguiera gymnorhiza* showed its long flowering period during February to September and *Avicennia officinalis* bloom from April to July (Table 1).

Table 1. Flowering period of three selected mangrove plants

Mangrove plants	Local name	Family	Habit	Flowering period
Avicennia officinalis	Jatbaen	Acanthaceae	Tree	April – July
Bruguiera gymnorhiza	Kankra	Rhizophoraceae	Tree	February – September
Sonneratia apetala	Keora	Sonneratiaceae	Tree	March – July

Bruguiera gymnorhiza (x ¼) ***Sonneratia apetala*** (x ½) ***Avicennia officinalis*** (x ¼)

Plate 1 : Flowering twig of the investigated mangrove plants

(b) **Flowering phenology:** In the present study, it was observed that *Avicennia officinalis*, *Bruguiera gymnorrhiza* and *Sonneratia apetala* open their flowers between 05:00 – 18:30hr, 07:00 – 10:00hr and 15:00 – 18:00hr respectively and remain open for 2-5 days (Table 2). *A. officinalis* produce maximum number of flower per tree (14040) with 12 ±2 flower per inflorescence where an average of 1170 inflorescences are formed in each plant. The minimum number of flower output (30) was observed in *B. gymnorhiza* (Table 2). In *S. apetala*, the floral output per plant was moderate (1430)

(c) **Pollen morphology:** Pollen morphology of three plants described following the terminology of Erdtman[4].

Avicennia officinalis: Pollen grains 3-colporate, prolate-spheroidal, PA/ED 35.5/32.0µm, Colpi 30.0 µm in length, slightly curved, ora large, lalongate, exine 2.5 µm thick, sexine and nexine demarcated, surface reticulate (Plate 2).

Bruguiera gymnorhiza: Pollen grains 3-colporate, oblate-spheroidal, PA/ED 35.0/38.0µm, colpi 28.0 µm in length, curved, ora small, lalongate, exine 3.0 µm thick, sexine and nexine demarcated, surface reticulate (Plate 2).

Sonneratia apetala: Pollen grains 3-porate, prolate-spheroidal, PA/ED 34.5/32.5µm, ora circular, large with aspidote, exine 2.5 µm thick, sexine and nexine demarcated, surface reticulate (Plate 2).

Bruguiera gymnorhiza ***Sonneratia apetala*** ***Avicennia officinalis***

Plate 2: Pollen morphology of *Bruguiera gymnorhiza, Sonneratia apetala* and *Avicennia officinalis* (x 750)

(a) **Nectar-secretion dynamics:** The results of nectar secretion dynamics are given in Table 2. Among the three studied mangrove plants highest average nectar was recorded in *Avicennia officinalis* (9126 µl/plant) followed by *Sonneratia apetala* (7178.6µl/plant) and lowest average nectar was recorded in and lowest average nectar was recorded in *Bruguiera gymnorrhiza* (60.3 µl/plant). Nectar sugar concen-tration was also highest in *Avicennia officinalis* (39%) followed by *Sonneratia apetala* (30%) and *Bruguiera gymnorrhiza* (21%).

Table 2: Nectar-secretion dynamics of three mangrove plants

Name of plants	Amount of nectar(µl) per flower	Average amount of nectar (µl) produced per plant	Nectar sugar concentration (%)
Avicennia officinalis	0.65	9126	39
Bruguiera gymnorrhiza	2.01	60.3	21
Sonneratia apetala	5.02	7178.6	30

DISCUSSION

In the presence study, we observed that among the three selected species, *Avicennia officinalis* with panicle inflorescence contain maximum number of flower per plant (approx. 14040/plant). There was a peak flowering period between March to July. Palynologically, *Bruguiera gymnorhiza* and *Sonneratia apetala* showed similar type of pollen aperture (3-colporate), however they differed in the shape and size of pollen as well as size of pore (ora). *Avicennia officinalis* pollen showed completely different aperture (3-porate) with larger pore size. Thus pollen is found to be the important character by which all the three species could be identified.

Flowers of *Avicennia officinalis* opened early in the morning at 05:00 hour and stayed for three days, but flowers of *Sonneratia apetala* opened in the afternoon at 15:00 hour and stayed for 2 - 3 days. *Avicennia officinalis* showed highest average nectar (9126 µl) followed by *Sonneratia apetala* (7178.6 µl), but *Bruguiera gymnorrhiza* showed lowest average nectar (60.3 µl). Maximum amount of nectar sugar concentration was recorded in *Avicennia officinalis* (39%) followed by *Sonneratia apetala* (30%).

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