

## **Prevalence of fungal spores in rural and urban environments of Pondicherry region during 2013**

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### **ABSTRACT**

*Occurrence of airborne fungal spores in the extramural environments of rural and urban areas of Pondicherry region was carried out by gravitation method from February 2013 to July 2013. It was found that the composition and concentration of fungal spores considerably varied from rural to urban areas as well as from site to site during the study. Rural areas harbored maximum fungal spores (55%) in comparison to urban areas (45%). Occurrence of fungal species was predominated with more number of propagules during March and April in comparison to other months. Qualitatively, *Aspergillus* was found with the highest frequency and had nine members i.e., *A. flavipes*, *A. flavus*, *A. fumigatus*, *A. japonicus*, *A. niger*, *A. sydowii*, *A. sydowii*, *A. terreus* and *A. wentii* and quantitatively *aspergilli* also isolated in highest contribution to total CFUs followed by *Cladosporium*. Out of the 42 isolated fungal taxa, *Aspergillus fumigatus*, *A. niger*, *Cladosporium* spp., *Rhizopus stolonifer* and *Alternaria alternata* were the predominant aeroallergens, which cause different types of respiratory/lung diseases in atopic human beings. *Alternaria alternata*, which is accounted as a human allergen for sporosis inducer and an agent for hay fever and other pathologies, was also intermittently recorded. Few plant pathogenic, saprophytic, field and storage fungi were also recorded during the study period. Similarity coefficient of the fungal diversity was found significant among the localities studied here.*

**Key words:** Rural and urban areas, Pondicherry region, Prevalence of fungal spores, CFUs.

### **INTRODUCTION**

The frequency and incidence of airborne fungi of different localities varies according to the prevailing environmental conditions and local vegetation [1]. Airborne fungi are present in both outdoor and indoor environments, sometimes occupying 70% of total microflora of air [2]. Fungal conidia and spores are liberated in air from plants, animal, soil, decaying organic material and can remain airborne for long time. Airborne fungi and their spores are potential to create health hazards and associated with dermatitis and respiratory diseases along with allergic dysfunctions in human beings [3]. Now a day's allergic respiratory diseases have become so common that it affects over 25% of total population in the world [4]. Many genera of fungi can cause allergic responses, but particularly important are some species of *Cladosporium*, *Alternaria*, *Penicillium* and *Aspergillus*. *Cladosporium* and *Alternaria* are frequent and predominant genera present mainly in the outdoor air [5]. Some moulds also produce mycotoxins and some species become particularly toxicogenic by producing dangerous volatile compounds [6]. Understanding the diversity along with the spatial and temporal distribution of airborne fungal spores is of increasing importance because fungal spores and other propagules can potentially act as sources of plant diseases [3], human allergens [7,8], animal and human infectious agents [7], produce mycotoxins and other deleterious secondary metabolites [2]. The present study was undertaken to carry out a comparative aeromycological study pertaining to the occurrence of airborne fungal spores in rural and urban areas of Pondicherry region during the year 2013.

## MATERIALS AND METHODS

The present study was carried out in two different localities of rural villages; Gopalan Kadai, Porayur and Perambai and urban areas; Lawspet, Rediarpalyam and Muthialpet of Pondicherry region from February up to July, 2013.

### Study sites

Pondicherry is the capital city of Puducherry state situated in the coromandal coast of Bay of Bengal and ruled by the French before 16<sup>th</sup> August 1947, which lies within 11° 46'' and 12° 30'' N latitudes and 79° 36' and 79° 53'' E longitudes. The study sites were selected from rural and urban localities of the Pondicherry region. Among rural village; Gopalan Kadai, Porayur and Perambai were selected, which are situated western parts of Pondicherry city around 10 km from the city area. Among urban localities; Lawspet the education establishment area, Rediarpalyam and Muthialpet within the main city area were selected. Village pattern houses are mostly found in these areas but few recently well constructed houses with good ventilation are also there. Village people are merely dependent on agriculture and few of them are doing service in different ways.

### Air Samplings

Air samplings were taken for continuous six months from February 2013 to July 2013 at monthly intervals, between 10 to 11 A.M. from outdoors, 20 feet above the ground exposing PDA mediated plates. Three replicated media plates ( $\theta = 9\text{cm}$ ) containing Potato Dextrose Agar (PDA) medium with streptomycin/penicillin ( $50\text{mg}^{-1}$ ) were carried to the study sites with sterilized container and exposed to the air for five minutes to receive the sedimentation of the air borne fungal spores on the media plates. The air sampling was not made in the rainy days. After exposed, each set of plates were brought separately to the Microbiology Laboratory, Department of Botany, KMCPGS (Autonomous), Pondicherry with utmost care and incubated in culture room at  $25\pm 3^{\circ}\text{C}$  upside down for 15 days with constant observation after 3-4 days of incubation. Fungal colonies developed in plates were counted for individual species and to get the total number CFUs. Microscopic slides stained with lacto phenol cotton blue were prepared from each CFU 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/CDA media again and identified later on. The laboratory experience and taxonomic literature were employed to identify the fungal taxa. Annual and monthly percentage occurrence of individual fungus was determined.

### Similarity coefficient analysis

The similarities in fungal community composition between two areas were calculated by following formula.

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

Where, a: Total no. of species isolated from one site, b: Total no. of species isolated from other site, W: Common no. of species.

## RESULTS

During the study period, a total number of 1527 fungal CFUs were isolated from both rural and urban areas of Pondicherry region, of which rural environment contributed (55%) followed by urban environment (45%). Incidence of airborne fungal species, their CFUs contribution and total occurrence recorded in each environment of the region are given on Table 1. Qualitatively, altogether 42 fungal species were isolated comprising of 24 genera from both the areas. Among the total number of isolated fungal species, aspergilli contributed 9 species followed by less numbers of species contributed by penicilli, cladosporia and *Mucor*. Other genera were found one or two in their contributions. *Aspergillus* spp contributed maximum in both rural and urban areas followed by *Penicillium* and *Cladosporium*. Based on the total occurrence, *Aspergillus niger* was found to be the highest (29.27%) in its contribution to the total fungal flora in Pondicherry region followed by *Penicillium citrinum* (13.88%), *Cladosporium herbarum* (11.98%), *Aspergillus flavus* (08.87%) and *Fusarium oxysporum* (03.86%). The month of March contributed the maximum spores followed by April and June. Comparative structure of fungal CFUs recorded in different localities showed that Gopalan kadai was the highest among all and between both the rural and urban sites it was followed by Muthialpet and Porayur and Lawspet (Fig 1). Perumbai from village site and Reddiarpalyam from Urban site contributed the least number of fungal propagules among all. The months starting from February to July didn't show any much difference in the occurrence of fungi in all the areas of rural and urban sites (Fig 1). Matrix showing values of similarity co-efficient and common number of fungal species between one site with other site among the Pondicherry region is given in Table 2, which showed that Muthialpet and Porayur had more similarities and common no. of species but the least similarity was found between Rediarpalyam and

Perumbai. Special group distribution of airborne fungal species isolated from rural & urban environments of Pondicherry showed the Deuteromycetous fungi were more prevalent in both the localities of Pondicherry region

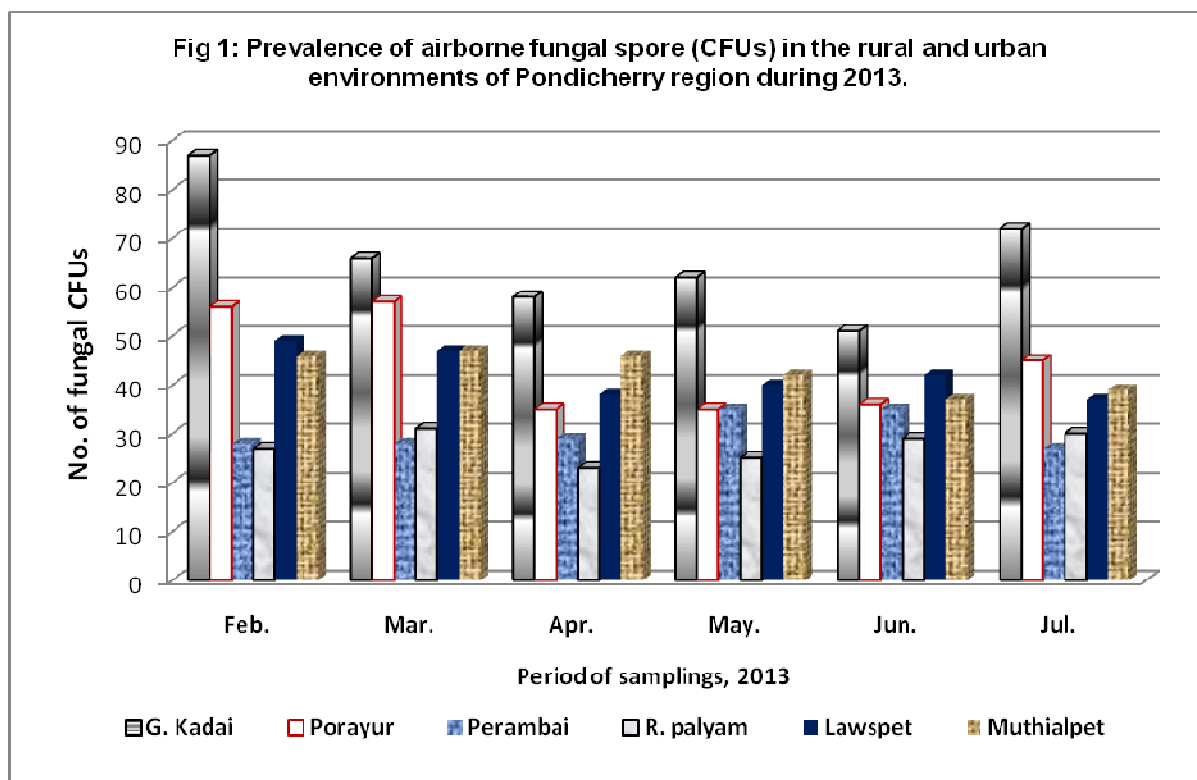
Table 1: Occurrence of airborne microfungi in the rural and urban area of Pondicherry region during 2013

Sl. No.	Name of fungi	Rural area			Urban area		
		G. Kadai	Porayur	Perambai	Lawspet	R. palyam	Muthialpet
1	<i>Absidia corymbifera</i>	-	-	-	6	-	-
2	<i>Absidia spinosa</i>	-	4	-	-	-	4
3	<i>Alternaria alternata</i>	-	-	-	13	12	-
4	<i>Aspergillus flavipes</i>	-	11	3	-	2	-
5	<i>Aspergillus flavus</i>	13	31	33	10	09	36
6	<i>Aspergillus fumigatus</i>	7	3	5	7	5	4
7	<i>Aspergillus japonicus</i>	-	-	-	3	-	-
8	<i>Aspergillus niger</i>	192	62	51	40	31	71
9	<i>Aspergillus sydowii</i>	-	-	-	-	3	-
10	<i>Aspergillus terreus</i>	-	-	5	-	-	-
11	<i>Aspergillus ustus</i>	7	-	-	-	-	2
12	<i>Aspergillus wentii</i>	-	-	-	-	-	7
13	<i>Aureobasidium pullulans</i>	-	-	6	7	-	-
14	<i>Botrytis cinerea</i>	-	14	-	-	-	13
15	<i>Candida</i> sp.	-	7	-	-	-	-
16	<i>Cladosporium cladosporioides</i>	-	-	12	10	-	-
17	<i>Cladosporium herbarum</i>	66	9	12	25	31	40
18	<i>Cladosporium resinae</i>	4	10	-	-	7	-
19	<i>Cladosporium sphaerospermum</i>	-	2	-	4	-	26
20	<i>Curvularia geniculata</i>	-	-	-	-	15	-
21	<i>Curvularia lunata</i>	-	-	8	21	0	-
22	<i>Fusarium moniliforme</i>	-	9	7	-	11	-
23	<i>Fusarium oxysporum</i>	2	8	2	23	8	18
24	<i>Geomyces</i> sp.	-	-	-	-	2	-
25	<i>Geotrichum candidum</i>	6	7	-	-	-	-
26	<i>Gliocladium roseum</i>	5	-	-	4	-	3
27	Grey sterile mycelia	-	-	-	-	-	2
28	<i>Helminthosporium</i> sp.	-	-	-	-	4	2
29	<i>Monilia sitophylla</i>	-	11	11	-	-	-
30	<i>Mucor racemosus</i>	-	7	2	-	4	-
31	<i>Neurospora sitophylla</i>	2	-	-	-	-	2
32	<i>Penicillium chrysogenum</i>	-	-	-	6	-	5
33	<i>Penicillium citrinum</i>	70	55	12	41	12	22
34	<i>Penicillium fellutanum</i>	14	13	3	-	3	-
35	<i>Penicillium frequetans</i>	-	-	-	-	-	4
36	<i>Penicillium oxalicum</i>	-	-	-	2	-	-
37	<i>Rhizopus stolonifer</i>	2	-	3	3	-	3
38	<i>Saccharomyces cerevisiae</i>	2	-	-	-	-	-
39	<i>Syncephalastrum racemosum</i>	-	-	2	-	4	-
40	<i>Trichoderma</i> sp.	1	-	-	-	-	-
41	<i>Wallemia sebi</i>	-	-	-	3	-	3
42	White sterile mycelia	3	4	5	3	2	-

Table 2: Matrix showing values of similarity co-efficient and common number of fungal species between one site with other site

	Rural area			Urban area		
	Gopalan Kadai	Porayur	Perambai	Lawspet	Reddiar-palayam	Muthialpet
<b>Gopalan Kadai</b>	(15) #					
<b>Porayur</b>	$\frac{43.24}{8}$ *	(17)				
<b>Perambai</b>	$\frac{45.71}{8}$	$\frac{52.63}{10}$	(18)			
<b>Lawspet</b>	$\frac{52.38}{11}$	$\frac{53.33}{12}$	$\frac{46.51}{10}$	(18)		
<b>Reddiar-palayam</b>	$\frac{32.45}{6}$	$\frac{40.00}{8}$	$\frac{42.10}{8}$	$\frac{45.88}{11}$	(19)	
<b>Muthialpet</b>	$\frac{41.17}{7}$	$\frac{54.05}{10}$	$\frac{62.85}{11}$	$\frac{52.38}{11}$	$\frac{43.24}{8}$	(17)

\* : Similarity coefficient value, \*\* : Common no. of fungal species, # : Total no. of species



## DISCUSSION

Aeromycospora studies employ a number of sampling techniques of which, gravity settling of spores on culture media is one of the widely used technique by different workers [2, 9] both in indoor and outdoor environments but its use in indoors is more appropriate as the sedimentation of spores is less affected by wind turbulence [10]. The present study rightly used this technique expressing the results only qualitatively. Out of the isolated fungal species, most of them belonged to the members of Deuteromycetes followed by members of Zygomycetes, which found by previous authors [11,12]. In the comparative analysis among members of fungal species, *Aspergillus niger* contributed the maximum both in rural and urban areas, but its concentration was more in former than latter, which was unique in its occurrences in Pondicherry environments. Based on the species distribution, *Aspergillus* comprised of nine species, via., *A. flavipes*, *A. flavus*, *A. fumigatus*, *A. japonicas*, *A. niger*, *A. sydowii*, *A. terreus* and *A. wentii* were recorded from Pondicherry environment was in agreement with the findings of many others [2,3,9]. A number of studies on aerospora of indoors and outdoors, particularly of occupational sites are found worldwide [2,9]. Indoor and outdoor mycospora had a greater resemblance with outdoor qualitatively but the concentration greatly depended on atmospheric parameters, both changes with accordance to occurrences, seasonally and diurnally. The present findings found the dense spores in rural areas than urban areas contribute to the prevailing atmospheric conditions and socio economic conditions leading to the degree of cleanness of the house and household activities. Moreover the dumping off cultivated items and paddy straw in the extramural environments of the rural areas made the maximum occurrence of airborne fungi [13]. Lumpkins and his coworkers [13] described the group of Deuteromycotina fungi were reported from both outdoor air and he also reported that there is a common reservoir pool for both indoor and outdoor mycobiota where they represent in the ambient air. *Aspergillus*, *Penicillium* and *Cladosporium* were the most common fungi recorded from both the sites in our study are in agreement with the findings of several other workers [14]. *Aspergillus fumigatus* causes respiratory infections leading to bronchopulmonary aspergillosis [15,16]. Singh and Singh [17] reported *A. fumigatus* as an opportunistic pathogen in immune-suppressed patients. In tropical environments these fungi i.e., aspergilli and penicilli are the dominant ones [18] and are known for allergenicity [19]. *Rhizopus*, *Alternaria* and *Curvularia* are saprophytic fungi and weak pathogen of vegetables and crop plants, were reported in the present study.

## Acknowledgement

The UGC, New Delhi is highly acknowledged for the financial assistance in the form of Major Research Project to the author as Principal Investigator.

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