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ENUMERATING OUTDOOR AEROMYCOTA IN SUBURBAN WEST BENGAL, INDIA, WITH REFERENCE TO RESPIRATORY ALLERGY AND METEOROLOGICAL FACTORS

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Abstract: Aeromycota may act as a reservoir of aeroallergens and upon inhalation may induce IgE-mediated Type I hypersensitivity reaction in pre-sensitized individuals. The total aerospora of an outdoor occupational setting (agricultural farm) in suburban West Bengal was sampled for two years (2002–2004) by a Burkard sampler. Concurrently, the cultivable aeromycota were trapped by an Andersen 2-stage sampler, cultured and tested for allergenic potential by skin prick test. The relationships between various climatic factors (temperature, relative humidity, rainfall and wind speed) and the distribution of aerospora were explored by Spearman correlation test. The antigenic extracts of 15 fungal species belonging to *Alternaria*, Aspergilli/Penicilli, *Cladosporium*, *Curvularia*, *Drechslera*, and *Nigrospora* evoked 10.8–54.8% skin reactivity in subjects with clinical history of respiratory allergy. The aerospora with skin sensitizing potential collectively represented a considerable fraction (52.3–58.4%) of the total aeromycota. The airborne concentration of *Alternaria* spores was higher than its borderline value of 100 spores m^{-3} in May and June, whereas *Cladosporium* spore count exceeded its threshold limit value (3,000 spores m^{-3}) in December, suggesting that this particular time of the year poses allergenic risk for individuals sensitive to these aerospora. Daily minimum temperature and rainfall appeared to be the most important meteorological factors to affect the concentration of aerospora in the study area.

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INTRODUCTION

Fungal spores are universal atmospheric components in both indoor and outdoor environments. Upon inhalation airborne fungal spores or fragments of hypha may incite respiratory allergic symptoms in pre-sensitized individuals [20, 27, 30]. About 20–30% of patients suffering from respiratory allergic disorders are sensitized to various fungal species [30]. Allergic reactions normally occur at the site of allergen deposition. Large spores (>10 μm in size), deposited usually in the nasopharynx, are associated with nasal and ocular symptoms, referred to as allergic rhinitis, [32] whereas smaller spores (<10 μm; specially <5 μm in size) can penetrate the lower airways and trigger bronchial asthma [35]. About 80 fungal genera, mostly belonging to Ascomycetes, Deuteromycetes and then to Basidiomycetes, have been described as the causative agents of airway diseases as yet [8, 27].

The occurrence and dispersion of aerospora in relation to environmental factors have been well documented in

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the mycological literature [12, 23, 25]. However, the concentration of aerospora may also fluctuate for biological reasons such as growth and differentiation of spores or spore-producing parts [22]. Elaborate investigation on the seasonal oscillation of aerospora in relation to the meteorological factors, with the exception of a few studies, is largely lacking in West Bengal [1, 10]. The present study has been conducted in order to specify the time of the year posing allergenic risk for atopic individuals, and to explore the influence of the climatic factors on the occurrence of individual predominant aerospora in the atmosphere of West Bengal.

MATERIALS AND METHODS

The aerobiological sampling was conducted in an agricultural farm located in Madhyamgram (22°42' N and 88°27' E, 6 m above sea level). Madhyamgram is a suburban area situated at about 19 km north of Kolkata. The inhabitants of this locality are in general involved in occupations like agricultural farming and field works. Four distinct seasons namely summer (March to May), monsoon (June to September), post monsoon (October to November), and winter (December to February) have been recognized in West Bengal [5]. The agricultural farm has a variety of fruit tree plantations, namely, sugar cane (*Saccharum officinarum*), mango (*Mangifera indica*), banana (*Musa* sp), coconut (*Cocos nucifera*), betel nut (*Areca catechu*) and papaya (*Carica papaya*). A number of herbs and shrubs such as *Amaranthus viridis*, *Parthenium hysterophorus*, *Cyperus* sp, *Justicia simplex*, *Chenopodium album*, *Catharanthus roseus* also grow abundantly in the farm.

A Burkard 7-day volumetric pollen and spore trap (Burkard Manufacturing Co. Ltd., Hertfordshire, UK) was used for the sampling of aerospora. The spore trap, placed at a height of 1.5 m above ground level amidst the agricultural farm, sampled air (air suction rate = 10 l min^{-1}) uninterruptedly for two consecutive years from November, 2002 to October, 2004. A strip of glycerin jelly coated Melinex tape was exposed to air for trapping the spores and was changed once a week. The exposed tape was cut into 48 mm segments representing 24 h periods. These segments were mounted on microscopic slides using gelvatol as mounting medium. The slides were scanned by a high resolution light microscope (Leitz, Diaplan, Germany) at 400×. The fungal spores were identified following the standard manuals [17, 34] and counted according to the guidelines of the British Aerobiology Federation [7]. The spore counts were then converted (conversion factor = 0.46) into the mean hourly concentration (number of spore per cubic meter of air). The 24-hourly counts were finally summed up to obtain the daily mean concentration.

Concomitantly, an Andersen 2-stage viable sampler (Thermo Andersen, Smyrna, USA) was used for collecting the field samples in order to identify the airborne fungal taxa up to species level, and to culture them for preparing antigenic extract for skin testing. The sampler, usually placed at the same height as that of the Burkard sampler, was operated for 3 minutes at weekly intervals between 12:15–12:45 h round the year. The sampler drew air (air sampling rate = 28.3 l min^{-1}) through the orifice at the top and impinged airborne fungal spores successively onto two Petri plates containing 2% MEA (malt extract agar) medium, supplemented with streptomycin sulphate (concentration = 40 µg ml^{-1}). After incubating the Petri plates at 27 ± 2°C for 4–5 days, the sporulating fungal colonies were identified consulting standard manuals [34, 37].

After culturing the airborne molds (species of *Alternaria*, *Aspergillus*, *Cladosporium*, *Curvularia*, *Drechslera*, *Nigrospora*, *Penicillium*) in 2% malt extract broth with streptomycin sulphate (concentration = 40 µg ml^{-1}) for 4 weeks, the fungal tissue comprising of both mycelia and spores, was harvested and homogenized (1:50 w/v) in phosphate buffered saline (PBS, 0.1 M Na-Phosphate, 150 mM NaCl, pH 7.2) on ice and centrifuged at 12,500 g for 45 minutes at 4°C (Minifuge RF, Heraeus, Germany). The supernatant was collected, dialyzed against PBS for 16 h at 4°C and sterilized by passing through 0.22 µm Millipore filter (Millipore Corp., Bedford, USA). The filtrate was then lyophilized and stored in sterile vials at -20°C for future use. The protein concentration of the crude extract was determined by Lowry assay [31] using bovine serum albumin (BSA) as standard.

About 600 individuals (male – 47.2%, female – 52.8%; mean age 30.2 ± 13.5 years), either employee of the farm concerned or residents of the adjoining area (within 15 km distance from the sampling site), were included in the present study population. The recruitment of study population was conducted as per the criteria followed by Ghosh *et al.* [18]. Among them, the individuals (n = 147/600; 24.5% of the study population) manifesting seasonal/perennial respiratory allergic symptoms (diagnosed by the physicians of the Allergy Section, Institute of Child Health, Kolkata) were selected as subjects for the clinical study (skin prick test). Among the symptomatic subjects, 32.6% were diagnosed as suffering from allergic rhinitis (AR) and 15.2% from bronchial asthma (BA), whereas the remaining 52.2% manifested combined symptoms (AR+BA).

Skin prick test was performed on the volar forearm of the subjects with 20 µl drops of sterile antigenic extracts. Phosphate buffer (0.01 M, pH 7.2) and histamine diphosphate (1 mg ml^{-1}) were taken as the negative and positive controls, respectively. The intensity of reactions was measured after 20 minutes with respect to the wheal and/or erythema responses. The result was considered positive if the wheal diameter was 3 mm or greater according to the International guidelines [14]. The study was approved by the Ethics Committee of the concerned institute and prior informed written consent was obtained from all the subjects.

The meteorological data were obtained from the regional office (Dum Dum) of the Indian Meteorological Centre situated 3.5 km from the sampling site. In the present

study, four climatic factors such as temperature (°C), relative humidity (%), rainfall for 24 h (mm) and wind speed (km/h) were taken into consideration. Relative humidity was measured at 08.30 and 17.30 h (local time) and collected data were averaged to obtain a mean value. The relationship between atmospheric fungal spore count and meteorological variables was studied by Spearman's correlation test, with the significance levels of 0.05 and 0.01 (2-tailed) using SPSS statistical software, version 10.0 (SPSS Inc., Chicago IL, 1999).

RESULTS

The total aeromycota concentration in the ambient air of suburban Kolkata attained the main peak in early winter (December) and a subsidiary peak in early monsoon (June) in both sampling years (Fig. 1). The total aerospora count fluctuated between 305.7–3470.1 spores m^{-3} in the first year (2002–2003) and 278.7–3628.1 spores m^{-3} in the second year (2003–2004). Altogether, 37 aerospora were detected in the outdoor agricultural environment of Madhyamgram over 2-year sampling period (Tab. 1).

In the present aerobiological survey 3 categories of aerospora were identified: conidia (*Alternaria*, Aspergilli/Penicilli, *Beltrania*, *Bispora*, *Cercospora*, *Cladosporium*, *Corynespora*, *Curvularia*, *Deightoniella*, *Drechslera*, *Dwayabeeja*, *Epicoccum*, *Lasiodiplodia*, *Memnoniella*, *Nigrospora*, *Periconia*, *Periconiella*, *Pithomyces*, *Spegazzinia*, *Tetraploa*, *Torula*, *Trichoconis*, *Phaeotrichoconis*, and *Pestalotia*); ascospores (*Chaetomium*, *Leptosphaeria*, *Podospora*, *Pleospora*, *Sporidesmium*, *Sporormia*, unidentified ascospores, and xylariaceae ascospores); and basidiospores (*Coprinus*, *Ganoderm*, rust spores, smut spores, and unidentified basidiospores). The atmosphere of Madhyamgram was found to be predominated by conidia (mean annual contribution 63.0%), followed by ascospores (mean annual contribution 23.6%) and basidiospores (mean annual contribution 13.4%) (Fig. 2).

Among the aerospora recorded in Madhyamgram the predominant ones (mean annual contribution >1.0%) were as follows: *Alternaria* spp., Aspergilli/Penicilli group, *Chaetomium* sp., *Cladosporium* sp., *Coprinus* sp., *Curvularia* sp., *Drechslera* sp., *Ganoderma* sp., *Nigrospora* sp., *Periconia* sp., *Pithomyces* sp., unidentified ascospores, and unidentified basidiospores (Tab. 1). The remaining aerospora were detected in low frequencies (<1.0%) in the air of the sampling site. *Cladosporium* species was recorded as the most prevalent aerospora occurring in the ambient air of the study area. A statistically significant difference (t value 2.337, $p<0.05$) was observed between the 2-years' aerobiological data of *Phaeotrichoconis* sp. when the consistency in spore concentration was checked by Student's t test. However, the remaining aerospora, as well as the total aeromycota, exhibited no statistically significant difference in the spore concentrations recorded in the 2 consecutive sampling years (data not shown).

Table 1. Percent contribution of individual fungal spore type to the total aerospora count.

Fungal spore types	Percent contribution (%)	
	2002–2003	2003–2004
Alternaria spp.	2.9	2.5
Aspergilli/Penicilli	5.2	5.5
Beltrania sp.	0.02	0.01
Bispora sp.	0.1	0.2
Cercospora sp.	0.2	0.2
Chaetomium sp.	3.2	3.2
Cladosporium sp.	37	41.5
Coprinus sp.	2.8	1.7
Corynespora sp.	0.2	0.3
Curvularia sp.	0.9	1
Deightoniella sp.	0.04	0.04
Drechslera sp.	2.3	3.2
Dwayabeeja sp.	0.1	0.2
Epicoccum sp.	0.1	0.1
Ganoderma sp.	1.7	1.9
Lasiodiplodia sp.	0.1	0.1
Leptosphaeria sp.	0.04	0.6
Memnoniella sp.	0.1	0.1
Nigrospora sp.	4	4.7
Periconia sp.	4	4
Periconiella sp.	0.5	0.4
Pestalotia sp.	0.1	0.1
Phaeotrichoconis sp.	0.1	0.01
Pithomyces sp.	1.1	1.2
Pleospora sp.	0.1	0.1
Podospora sp.	0.1	0.1
Rust spores	0.9	1.2
Smut spores	0.6	0.7
Spegazzinia sp.	0.1	0.1
Sporidesmium sp.	0.1	0.1
Sporormia sp.	0.1	0.002
Tetraploa sp.	0.2	0.2
Torula sp.	0.5	0.6
Trichoconis sp.	0.2	0.2
Unidentified ascospores	21	18.2
Unidentified basidiospores	9.4	5.9
Xylariaceae ascospores	0.2	0.2

Considering the seasonal oscillation of the predominant aerospora (Fig. 3), airborne *Alternaria*, Aspergilli/Penicilli, *Curvularia*, *Drechslera*, *Nigrospora*, *Periconia*, *Pithomyces*, and unidentified ascospores exhibited perennial occurrence pattern, while the rest showed either seasonal (*Cladosporium*, *Ganoderma*, and unidentified basidiospores) or intermittent (*Chaetomium*, and *Coprinus*) occurrence pattern. *Alternaria* and unidentified basidiospores

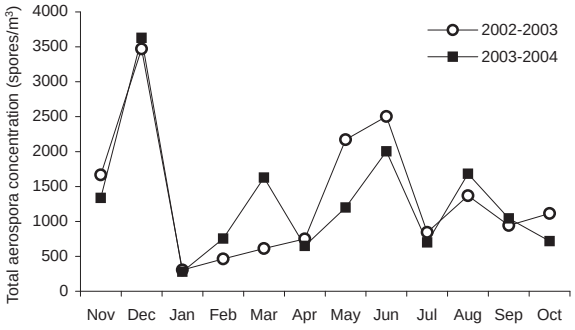

Figure 1. Seasonal variation of the total aerospora in two consecutive sampling years.

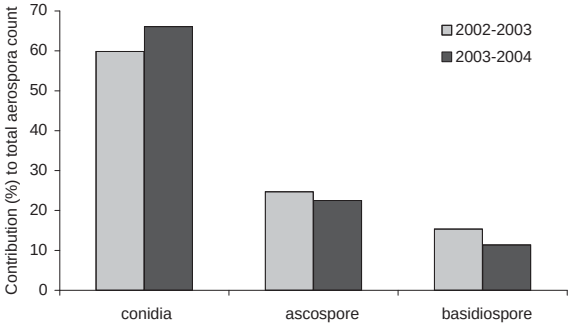

Figure 2. Fractions of conidia, ascospores and basidiospores in the total aerospora count in the study area.

peaked in early monsoon (June); *Curvularia*, *Drechslera*, and unidentified ascospores in mid-monsoon (July–Aug); *Ganoderma* in late monsoon (Sep); and Aspegilli/Penicilli in post-monsoon season (Oct–Nov). Although *Chaetomium*, and *Coprinus* sporadically fluctuated all year round, they attained a somewhat elevated concentration level during monsoon. Seasonally, the spore levels of *Cladosporium*, *Periconia*, and *Pithomyces* peaked towards the early winter season (Dec), whilst *Nigrospora* peaked in late summer.

Table 2. Results of Skin Prick Tests performed with the antigenic extracts of 15 cultivable aeromycota.

Fungal antigenic extracts	Total number of subjects tested	Percentage (%) (+) ve response (Total)	Percentage (%) response (2+/above)
1. *Alternaria* spp.:			
Alternaria alternata	72	22.2	8.3
Alternaria tenuissima	37	10.8	0
2. Aspergilli/Penicilli group:			
Aspergillus flavus	127	29.6	1.3
Aspergillus fumigatus	119	46.3	8.4
Aspergillus japonicus	42	54.8	4.8
Aspergillus nidulans	108	20.3	3.1
Aspergillus niger	45	44.4	2.2
Aspergillus sydowii	96	23.5	0.5
Aspergillus ustus	79	25.1	1.5
Penicillium citrinum	51	37.3	7.8
3. *Cladosporium* sp.			
Cladosporium cladosporioides	97	29.9	2.1
4. *Curvularia* spp.			
Curvularia pallescens	35	17.1	0
Curvularia lunata	85	26.2	4.3
5. *Drechslera hawaiiensis*	69	30.4	5.6
6. *Nigrospora sphaerica*	74	24.1	4.9

Altogether, 15 fungal species belonging to some predominant aerospora, namely, *Alternaria*, Aspergilli/Penicilli group, *Cladosporium*, *Curvularia*, *Drechslera*, *Nigrospora,* were identified from field samples collected by Andersen sampler, and tested for skin sensitizing potential on the subjects with positive clinical history of asthma and/or allergic rhinitis (Tab. 2). The highest skin sensitization was induced by *Aspergillus japonicus* (54.8%). The remainder except for *Alternaria tenuissima* and *Curvularia pallescens,* evoked remarkable skin reactivity (>20.0%) in the symptomatic subjects.

The relationships between the 13 predominant aerospora and climatic factors were explored in the present study. Daily minimum temperature seemed to be the most important factor regulating the seasonal fluctuation of a number of aerospora, namely, *Chaetomium*, *Coprinus*, *Curvularia*, *Ganoderma*, unidentified ascospores, and unidentified basidiospores (Tab. 3). Next to daily minimum temperature, rainfall represented another important weather factor as evidenced by its statistically significant positive association with *Chaetomium*, *Curvularia*, *Ganoderma*, unidentified ascospores, and unidentified basidiospores. Daily maximum temperature (another temperature variable) affected the aerospora concentration to a lesser extent by correlating with only three aerospora, namely, *Alternaria*, *Nigrospora*, and unidentified basidiospores, whereas daily mean temperature seemed to have significant influence on the concentration of unidentified ascospores, and unidentified basidiospores occurring in the air the study area. Increase in daily relative humidity corresponded with increase in the airborne *Ganoderma* spore concentration, but with decrease in the concentration of *Cladosporium* spores in the air. Daily wind velocity had the least regulation on the aerospora concentration as only one aerospora *Alternaria* correlated significantly with it. Aspergilli/Penicilli, *Drechslera*, *Periconia*, and *Pithomyces* did not correlate with any climatic factors.

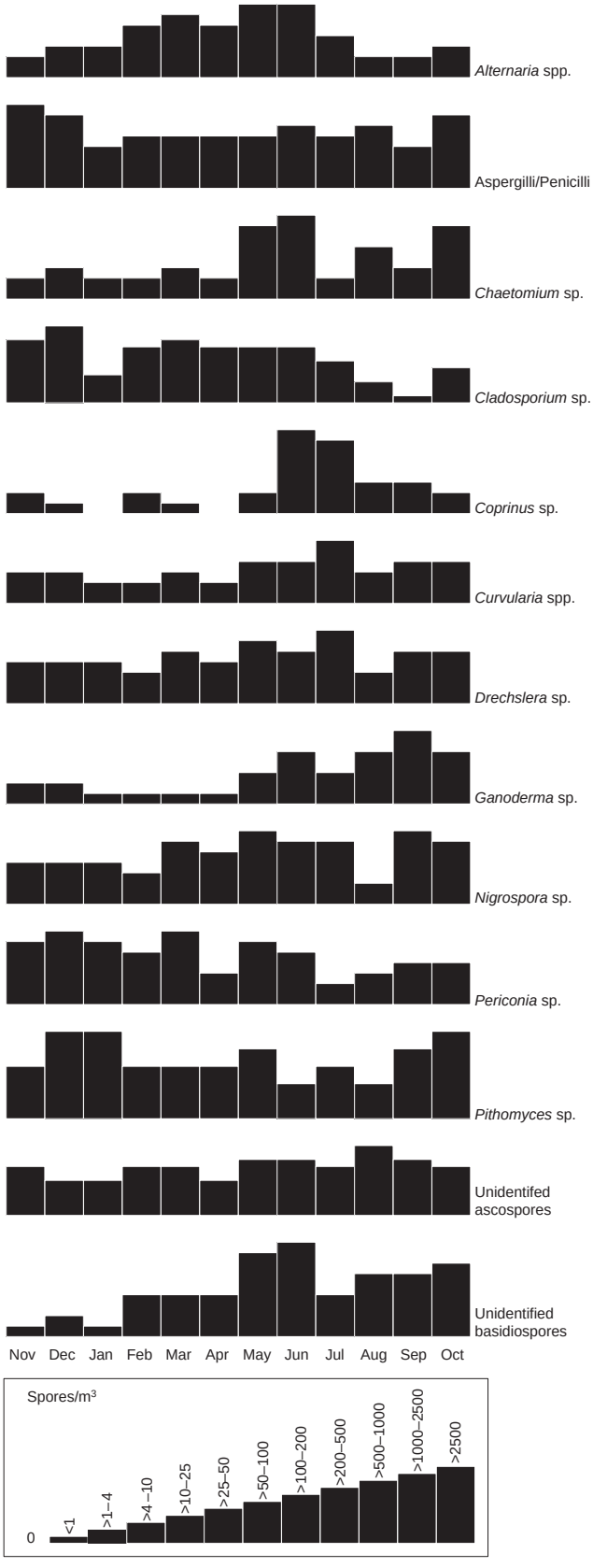

Figure 3. Fungal Spore Calendar representing the seasonal periodicity patterns of 13 predominant aerospora in average values for two-year sampling period (2002–2004) in Madhyamgram region of West Bengal. The scale with the concentration levels of fungal spores is presented.

DISCUSSION

The present aerobiological study was the first of its kind to be performed in West Bengal (India), emphasizing the seasonal fluctuation of some clinically significant aerospora under meteorological influence. The composition of aeromycota (37 aerospora representing 33 genera) recorded in the present study was consistent with the earlier reports in India [1, 38] and other countries [2, 26]. The constituents of aeromycota were more or less homogeneous during two sampling years, except for *Phaeotrichoconis* sp. This observation was almost consistent with the observation of Adhikari *et al.* (2004) where the authors found no statistically significant difference between two years' aerospora count [1]. A slightly elevated (not statistically significant) concentration of aeromycota was recorded in the present study compared to the previous studies conducted in a rural agricultural site [1, 10] and in an urban area [11] of West Bengal. The present survey has recorded the perennial occurrence of the total aerospora with the highest concentration achieved in late post monsoon-early winter (Nov-Dec). The relative humidity and temperature in moderate range, very low wind speed and minimal rainfall during early winter (Dec) facilitated the release and dispersion of dry spore mass; these factors contributed significantly towards achieving the main aeromycota peak during this month. The aerospora count became very low in the late monsoon period as prolonged rain washes the spores from the air. Similar seasonal fluctuation pattern of aeromycota was documented in a rural agricultural area of India [1] and also in an urban area of Athens [4]. The present aerobiological monitoring has recorded *Cladosporium* sp. (a dry aerospora) as the key contributor towards attaining the main peak in December, whereas ascospores and basidiospores (wet or humid aerospora) contributed considerably to the achievement of the subsidiary peak in June.

The predominance of conidia or "dry spores", produced by 24 out of 33 airborne fungal genera, recorded at the present sampling site was consistent with previous reports [1, 9, 36]. *Cladosporium*, the most prevalent aerospora, being a saprophyte thrives on decaying organic materials. In the agricultural fields, an abundance of decomposing leaves and plant debris was noticed, which in turn provided the ideal growth substrata for *Cladosporium*. This phenomenon, perhaps could explain the dominance of *Cladosporium* over other fungal genera found in outdoor environments worldwide [6, 19, 24, 26]. Apart from *Cladosporium*, basidiospores, and ascospores contributed significant proportions to the total aeromycota load, which was consistent with the findings of a previous study conducted in a similar environment [1]. The seasonal variation of various aerospora observed in the present study were in agreement with earlier survey reports from a similar outdoor environment [10, 38], except for *Coprinus*, and *Pithomyces* which exhibited deviation from the earlier reports. This might pertain to the variation in local

Table 3. Spearman's rank coefficients of 13 predominant aerospora and meteorological factors.

Aerospora	Temperature (°C)			Relative humidity	Rainfall	Wind speed
	Maximum	Minimum	Mean	(%)	(mm)	(km/h)
Alternaria spp.	0.594*	0.196	0.406	-0.573	-0.028	0.650*
Aspergilli/Penicilli	0	0.126	0.056	0.077	0.378	-0.245
Chaetomium sp.	0.483	0.594*	0.559	0.231	0.653*	0.308
Cladosporium sp.	0.112	-0.28	-0.077	-0.643*	0.382	-0.049
Coprinus sp.	0.267	0.611*	0.393	0.498	0.564	0.26
Curvularia spp.	0.375	0.641*	0.466	0.48	0.802**	0.263
Drechslera sp.	0.497	0.434	0.469	0.189	0.55	0.322
Ganoderma sp.	0.175	0.650*	0.42	0.783**	0.743**	0.07
Nigrospora sp.	0.594*	0.448	0.517	0.119	0.487	0.399
Periconia sp.	-0.336	-0.524	-0.566	-0.483	-0.508	-0.503
Pithomyces sp.	-0.413	-0.483	-0.559	-0.189	-0.333	-0.49
Unidentifed ascospores	0.343	0.741**	0.580*	0.413	0.628*	0.343
Unidentified basidiospores	0.601*	0.804**	0.776**	0.399	0.761**	0.545

* Correlation is significant at 0.05 level (2-tailed); ** Correlation is significant at 0.01 level (2-tailed).

environmental variables, edaphic factors, availability of fungal growth substratum, as well as in the vegetations surrounding the trapping site.

Nasal and ocular symptoms have been reported to develop following exposure to 20–500 × 10^3 fungal spores m^{-3}, whereas higher aerospora concentration (>500–17,000 × 10^3 fungal spores m^{-3}) induces development of cough in the sensitized agricultural workers [15]. The total aeromycota concentration in the present study was estimated as 15–16 × 10^3 fungal spores m^{-3}, which was lower than the threshold limit value for clinical significance (as mentioned above), implying that there was no exposure related health risk prevailing in the agricultural farm. But considering the relevant threshold values set for individual aerospora such as *Cladosporium* and *Alternaria* these were 3,000 spores m^{-3} air and 100 spores m^{-3} air, respectively [3]. In the present survey during December of both sampling years *Cladosporium* spores were recorded beyond the threshold limit, which may induce allergic reactions in sensitized individuals. *Alternaria* spores exceeded their borderline value for clinical significance during May 2003 and June of both years (2002–2004), suggesting the risk of allergic sensitization for the individuals sensitive to *Alternaria* spores. Furthermore, the combined concentration of airborne *Cladosporium* and Aspergilli/Penicilli spores (<5 µm in size) was found to be higher than 106 spores m^{-3} throughout the year, except for January, August and September. Exposure to fungal spores (mostly 1–5 µm in size) beyond the level of 106 spores m^{-3} might incite acute symptoms of extrinsic allergic alveolitis (also called Farmer's lung disease) among the agricultural workers, as suggested by Edwards [16], and Lacey & Crook [28].

In the present study, *Alternaria* (2 species), Aspergilli/Penicilli (8 species), *Cladosporium* (1 species), *Curvularia* (2 species), *Drechslera* (1 species), and *Nigrospora* (1 species) were found to evoke remarkable skin reaction on the subjects tested. The percentage of skin reaction induced by the fungal antigenic extracts in the present survey was comparable to the results obtained in previous studies conducted in West Bengal [1, 9]. The climate of West Bengal, a tropical humid region, might be congenial for profuse fungal growth and spore production, thereby resulting in an increase in the aerospora burden in the ambient air. Thus, exposure to such an elevated concentration level of aerospora might be the reason behind the occurrence of higher fungal sensitization in people residing in West Bengal. The fungal species causing positive response in the present *in vivo* test (i.e. skin prick test) have been previously described as important inhalant allergens [8, 27, 30]. Moreover, Green *et al.* (2005) has demonstrated allergic serum-IgE binding to the immobilized expressed antigens of a number of aerospora including *Alternaria*, Aspergilli/Penicilli, *Cladosporium*, and *Curvularia* [20]. The 6 aerospora, tested for allergenic potential in the present study, accounted for a significant fraction (52.3–58.4%) of the total aeromycota count, indicating the presence of potential biohazards in the agricultural work place in India as far as the health of the workers is concerned.

An increase in the concentration of airborne *Chaetomium*, and unidentified ascospores (also called "wet" or

"damp" spores) was recorded in response to the increasing rainfall. This finding of the present survey was in agreement with observation made by Davies [13]. *Ganoderma*, and unidentified basidiospores, the other wet spores recorded in the present monitoring, correlated positively with rainfall. These phenomena can be well explained by the importance of water in fungal spore discharge and dispersion. In addition to rainfall, the airborne concentration of *Ganoderma* was also significantly associated with the moisture content of the ambient air, i.e. relative humidity. This observation was consistent with previous reports [4, 23]. On the contrary, the distribution of airborne *Cladosporium* spore, being a dry aerospora, was negatively associated with rainfall. Daily temperature also represents another important weather factor. Mean daily minimum temperature in tropical countries like India are in the range of optimum growth temperature for fungi, and perhaps this physiological phenomenon causes an increase in aerospora (*Chaetomium*, *Coprinus*, *Curvularia*, *Ganoderma*, unidentified ascospores and unidentified basidiospores) concentration in connection with the rising daily minimum temperature, as documented in the present study. Wind is an important factor for the dispersion of aerospora. High wind speed might facilitate the release and/or dispersal of multicellular large-sized spores such as *Alternaria* [29], thus resulting in a positive association between wind speed and *Alternaria* spores observed in the present study. This observation was consistent with the findings of earlier workers [29, 33]. Thus, *Alternaria* and *Cladosporium*, two aerospora with distinct allergic significance [8, 27, 30], were found to be correlated with several climatic factors, thereby implying that a slight change in the meteorological conditions in this region might have a health hazardous effect on sensitized individuals. This view has been supported by a previous study [21] where a mere change in ambient air temperature was found to provoke asthma symptoms in some patients.

A fungal spore calendar worked out on the basis of seasonal periodicity patterns of aeromycota can provide information about the concentration of aerospora in an individual region, as well as aiming at elaborating the forecasts for the occurrence of airborne fungal taxa in different geographic conditions. Furthermore, this calendar can be helpful in the clinical prevention of seasonal allergic diseases and plant diseases induced by fungal pathogens.

CONCLUSIONS

A rich and diverse aeromycota was recorded in an agricultural farm located in suburban Kolkata. The total aerospora comprising of conidia (63%), ascospores (23.6%), and basidiospores (13.4%) varied seasonally with the peak achieved in early winter (December). *Cladosporium*, ascospores (unidentified), basidiospores (unidentified), Aspergilli/Penicilli, *Nigrospora*, and *Periconia* were the major constituents of the total aerospora. Among them, *Cladosporium* was the most abundant. The cumulative concentration of 6 predominant aerospora having considerable skin sensitizing potential accounted for >50% of the total aeromycota count. Daily minimum temperature and rainfall were found to be the most important climatic factors to regulate the seasonal distribution of aerospora in the study area.

The airborne concentration of *Alternaria* and *Cladosporium* spores exceeded their respective threshold limit values (potential to trigger allergic reactions) during late summer-early monsoon (May–June), and early winter (December), respectively. Accordingly, risk of allergic sensitization prevailed in that time of year for individuals allergic to these two spore types. The fungal spore calendar constructed in the present study would be useful in designing preventive measures against exposure-related respiratory allergic disorders. It is recommended that any allergen forecast, prevention and/or avoidance guidelines(s) for a region should also take meteorological information into consideration.

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