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Research Article

FUNGAL SPORE EXPOSURE IN THE SCHOOL CLASSROOMS OF SUBURBAN EASTERN INDIA WITH REFERENCE TO RESPIRATORY ALLERGY

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ABSTRACT

A questionnaire survey was conducted among a student population of a suburban school of West Bengal, India, on the basis of frequency and familial history of respiratory allergy, awareness level and absenteeism due to allergic symptoms. In addition, airborne fungal spores were monitored in the classrooms for a year (January-December, 2013) using rotorod sampler (overall spores) and petriplate exposure (viable/culturable fungal spores) with nutrient media.

More than 11.82% of students (n =482, age range 12-14 years) reported respiratory allergic symptoms and 47.36% were aware about their allergic sensitivity before the survey. Due to respiratory allergic symptoms, 78.95% of the allergic group became absent in school. In the air of classrooms, 14 fungal spore types were identified with maximum contribution of *Aspergillus-Penicillium* group (11.7%). Seventeen viable fungal species were recorded by petriplate exposure in nutrient media. Among them, *Aspergillus niger* (12.8%) was recorded as highest contributor, followed by *Aspergillus fumigatus* (9.95%), *Cladosporium cladosporioides* (9.87%), etc. The flare-up of respiratory allergy in school-children may lead to impaired daily function and absence from school. Hence, attention must be given to control the fungal growth and multiplication inside the classrooms to safeguard the health of school students.

INTRODUCTION

In recent times, people spend 80% - 90% of their time in indoors where concentration of many air pollutants are higher than outdoor (Carrer *et al.*1997). Hence, air quality of indoor environment is one of the major factor affecting health, wellbeing and productivity of human being. School students (<18 years of age) constitute a risk group (Daisy *et al.* 2003) and are especially vulnerable to poor air quality in indoors, as they breathe higher volume of air, relative to their body weight and their tissues/organs are in growing phase. Indoor air quality is directly affected by the presence of suspended bio-pollutants like mould spores and bacteria (World Health Organization 2009). Moulds may cause adverse health effects by inducing allergy, infection and toxicity (Cabral 2010). Many respiratory allergic disorders like rhinitis, hypersensitivity pneumonitis and allergic asthma may occur in association with exposure to indoor mould spores (Seltzer and Fedoruk 2007). Thus, concentration and diversity of airborne mould spores, which may trigger allergic reaction, must be taken into account to provide a safe indoor environment, where indoor air of the classrooms of schools is of particular concern (Chakraborty *et al.* 2014).

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Comprehensive information on the composition of the indoor airborne mould spores from suburban schools in state of West Bengal, is rare. In addition, there is also lack of awareness about the extent and management of allergenic mould spore exposure among the students. In the present study, after conducting a questionnaire survey in a student population, the diversity and dynamics of indoor airborne fungal spores in class rooms has been portrayed for a year from a suburban secondary school of West Bengal, which can initiate allergic reaction in susceptible individuals after inhalation.

MATERIALS AND METHODS

Sampling Site

Airborne fungal spores/conidia were monitored inside the classrooms (Classes VI, VII & VIII) of Prafullanagar Vidya Mandir (Boys') school building, of Habra, a suburban town of Nortn 24 Parganas district, West Bengal for the year 2013.

Questionnaire survey

A questionnaire survey was conducted among the students of class VI, VII and VIII (age range 12-14 years, n = 482) to record the number of the students suffering from respiratory allergy, their awareness level and absenteeism due to this particular disorder.

Sampling Procedure

Monitoring of airborne fungal spores was conducted by rotorod sampler (2800 revolutions/minute) for overall fungal spore types for the indoor of the classrooms of the school building. Petri plates containing sterile nutrient media were exposed (5 minutes) to record viable airborne fungal spores inside the classrooms. The sampling frequency was twice per month at an interval of 15 days for a year from January 2013 to December 2013. Exposures were made at approximate human height (1.5 m) at 14.00 pm. The duration and sampling time of the day were determined after standardization trials in pilot studies.

The fungal spores were deposited on the smeared glycerine jelly coated cellotapes stuck over the rotating arms of the rororod sampler. The cellotapes were mounted over glass slide and recorded spores were observed under microscope (Olumpus Magnus). Spore count in each slide was converted into spore count/m^3 air according to standard protocol (Singh and Singh 1994).

Petri plates with 2% Potato Dextrose Agar nutrient medium supplemented with streptomycin sulphate (50 µg/ml) were exposed in the classroom. After exposure, Petri plates were incubated at 27 ± 2º C for 3 days for growth and sporulation of fungal colonies. Fungal colonies were identified to the lowest taxonomic rank possible based on their morphological features like colour, size, shape, mycelia type and spores. Different guidelines and published literatures were used for authentic identification (Gilman 1957; Barnett and Hunter 1999; Ellis and Ellis 1988; Pitt 2000). *Aspergillus* and *Fusarium* colonies were further subcultured on Zapek–Dox Agar medium containing streptomycin sulphate (50 µg/ml) to facilitate their identification up to species level (Raper and Fennel 1965; Klitch 2002). The numbers of colonies recorded from both the Petri plates were combined and counted to get the viable/culturable spore records.

RESULTS AND DISCUSSION

Among the study population (n = 482), 11.82% (Fig. 1) reported to have respiratory allergic symptoms like allergic rhinitis (running nose, sneezing, watery eyes, etc) and/or allergic asthma. Among respiratory allergic population (n = 57), 68.42% had familial history of allergy, 26.31% reported to have no such familial history and rest were not sure about their family history of allergic diseases. Regarding awareness level, 47.36% were found to be aware about the causes of allergic sensitivity before conducting the survey. From the attendance record, it was observed that among the allergic students, more than 75% (78.95%) misses their school due to sickness caused by allergic rhinitis/bronchial asthma (Fig. 1). Overall survey in the classroom demonstrated the presence of 14 airborne fungal spore types (Fig. 2a) with maximum concentration of 756 spores/cubic meter air. There were 17 viable/culturable fungal members (Fig. 2b) having total maximum concentration up to 385 colony forming unit. The most important contributor was from *Aspergillus-Penicillium* group (11.7%), followed by *Cladosporium* (10.6%), Ascospores (8.7%), Basidiospores (8.6%), *Alternaria*(7.9%), *Curvularia* (7.5%), *Fusariun* (*6.8%), Ganoderma* (6.3%), *Nigrospora* (5.0%), and *Periconia* (4.7%) and others like *Pithomyces, Periconiella*, *Torula and Tetraploa*, etc. *Aspergillus-Penicillium* group, Basidiospores, *Curvularia* were found round the year. Most of them showed their peak concentration (5.7 to 55 spores/day/cubic meter air) in September-October (*Aspergillus-Penicillium* group, *Cladosporium, Basidiospore, Alternaria, Curvularia, Fusarium, Ganoderma, Nigrospora* and *Periconia* (Fig. 3).

Among the culturable/viable fungal spore types recorded in the air of the classrooms, highest percentage was contributed by *Aspergillus niger* (12.8%), followed by *Aspergillus fumigatus* (9.95%), *Cladosporium cladosporioides* (9.87%) and other fungal members (Fig. 4) like *Rhizopus nigricans, Rhizopus oryzae, Aspergillus flavus, Penicillium*

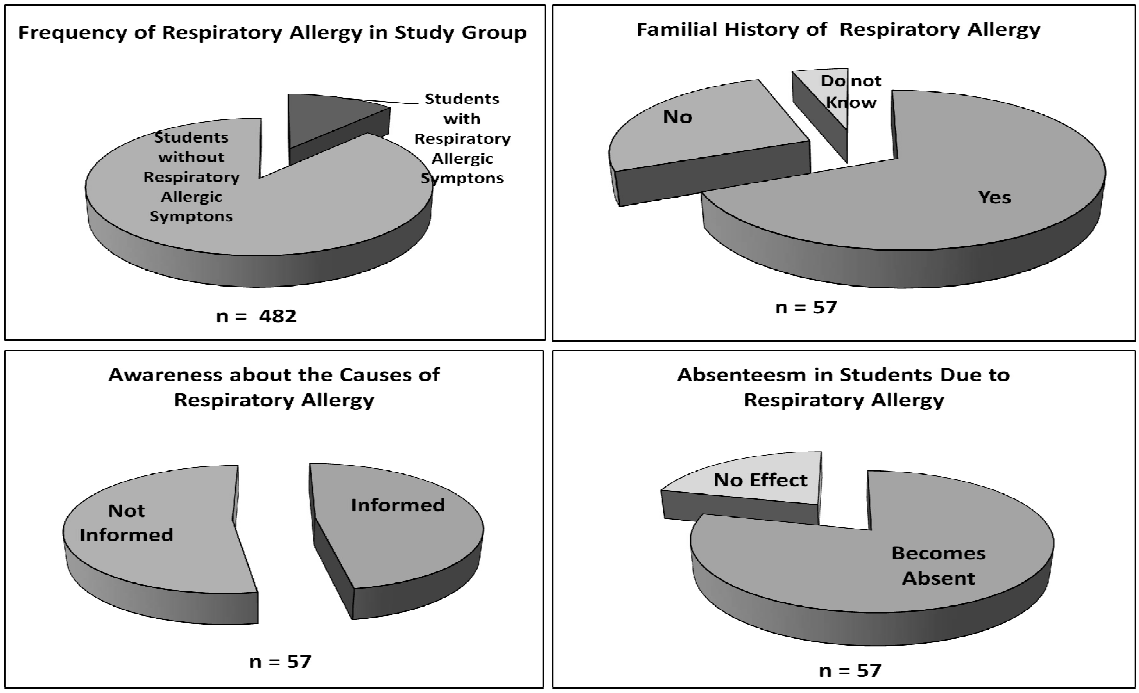

Fig 1 Results of Questionnaire Survey on Respiratory Allergy among the Study Group of school Students.

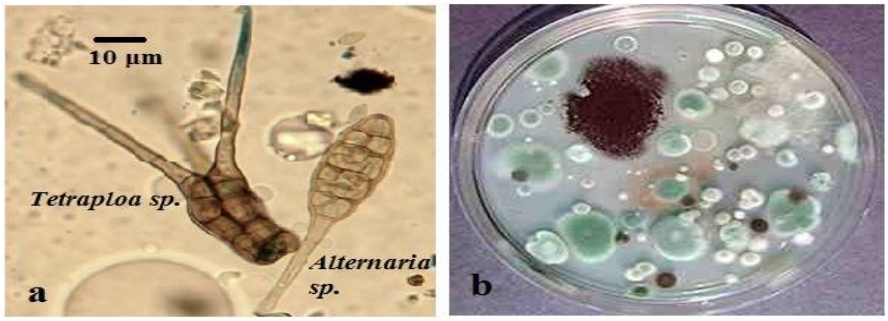

Fig 2 a. Airborne fungal spores trapped by rotorod sampler under microscope [Scale = 10 µm (10^{-2} mm), b. Fungal colonies formed by viable spores after petri plate exposure.

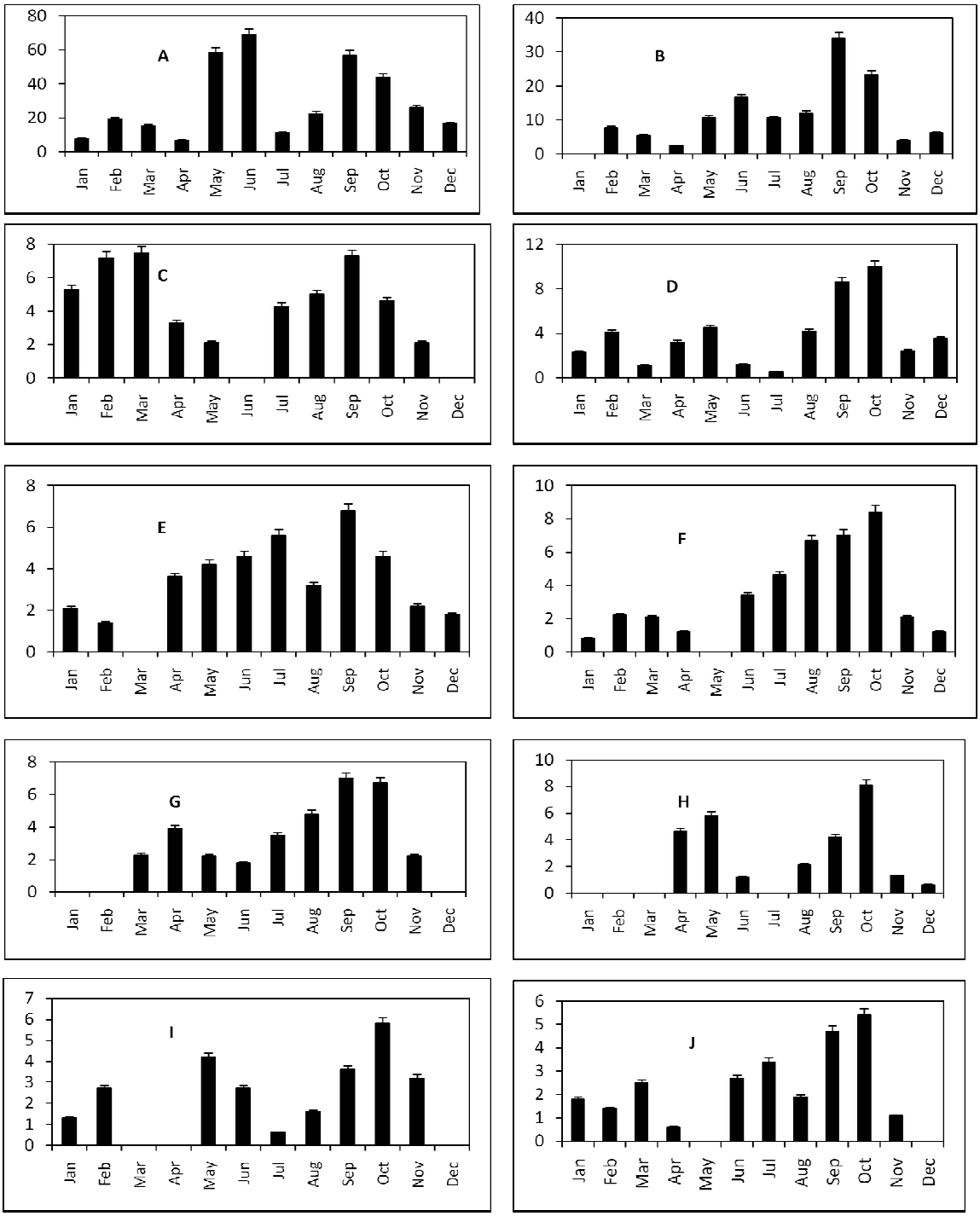

Fig 3 Seasonal periodicity of airborne fungal spores in the class rooms. (a) *Asperigillus-Penicillium* group, (b) *Cladosporium*, (c) Ascospores, (d) Basidiospores, (e) *Alternaris* sp., (f) *Curvularia* sp., (g) *Fusarium*, (h) *Ganoderma*, (i) *Nigrospora*, (j) *Periconia*.(Y-axis denoted spores/day/cubic cm air.; Bars indicate standard deviation.

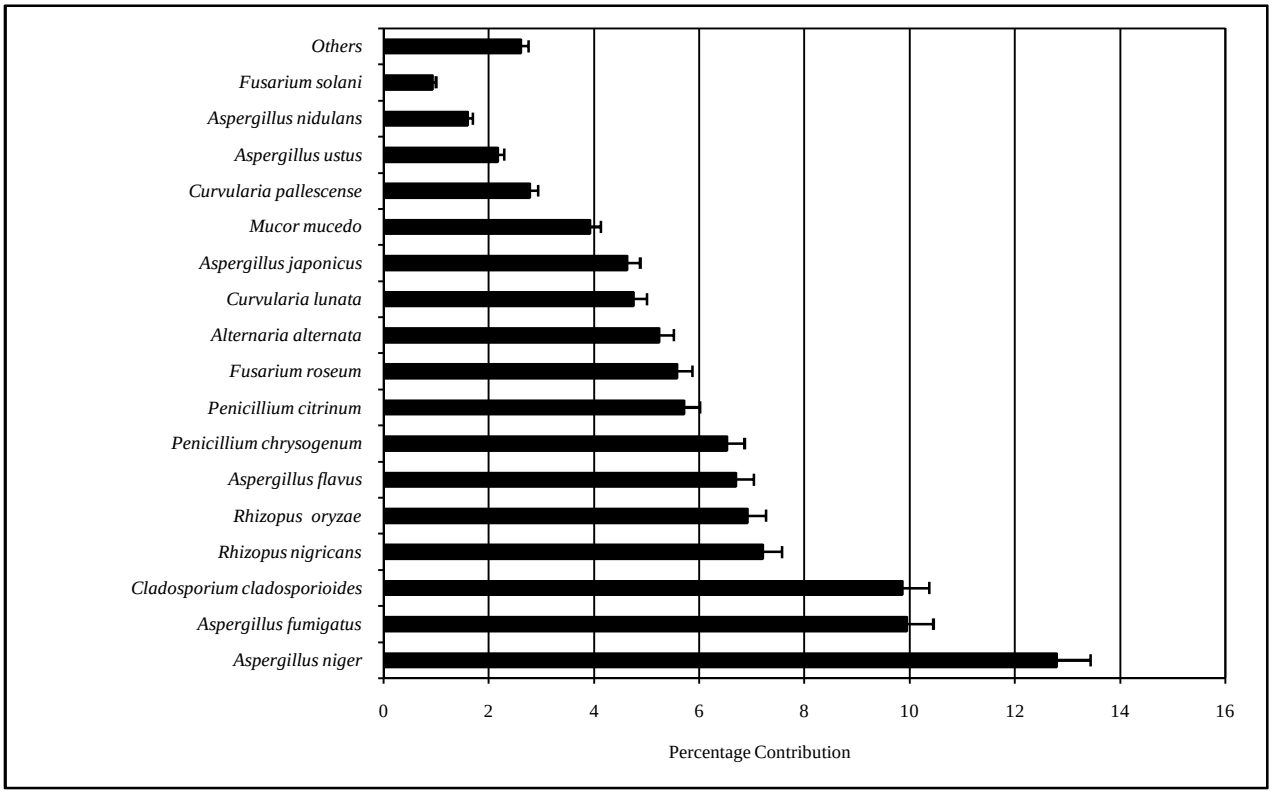

Fig 4 Percentage contribution level of viable/culturable fungal spores inside the classrooms for the year 2013. Bars indicate standard deviation.

chrysogenum, Penicillium citrinum, Fusarium roseum, Alternaria alternata, Curvularia lunata, Aspergillus japonicus, Mucor mucedo, Curvularia pallescens, Aspergillus ustus, Aapergillus nidulans and *Fusarium solani*.

Allergic rhinitis and asthma is a common health problem affecting school students of adolescence. It is a leading cause of school absenteeism all over the world (Moomie *et al.* 2006; Weeke *et al.* 1998). Often this problem has a hereditary link, but exposure to airborne allergens can boost up the hypersensitivity. Complications of respiratory allergy could be influenced by lack of awareness, which is reflected in the questionnaire survey of the present study. Airborne fungal spores are minute particles, which can trigger type 1 allergic reaction in susceptible individuals. These are dominant aeroallergen in the indoors. In the classrooms, students continuously inhale these allergenic fungal spores, which are not good for their health. Other than allergenic reaction, fungal spores can also produce carcinogenic toxins (e.g., aflatoxin from *Aspergillus flavus*), neurotoxins, offensive odours, etc. Sick building syndrome is an example of fungi induced health complications in human being. Excessive school absence disrupts learning and is a strong predictor of premature school drop-out (Baxi *et al.* 2013). The peak of airborne fungal spores are mainly found in September-October, after the end of rainy season, when after luxuriant growth due to high moisture content, these are released – when rain is over. Overall spore count shows the account of all airborne spores, but the members of *Aspergillus* and *Penicillium* genus can be recognized, when we allow the viable spores to grow as colony forming unit in nutrient media. Hence, petri plates were exposed to identify this group and also to recognize the fungal spore types up to species level.

In this way, the aerobiological survey of the class rooms of the school gives us an idea about the fungal spore exposures for the students, about which they are not often aware.

Management of respiratory allergic disorders can be carried out by therapeutic measure. However, avoidance of exposure is the best way to get relief from this health hazard. For this, development of awareness among students is most important (World Health Organization 2009). If awareness is developed, different preventive measures like regular cleaning of the class rooms, improved ventilation, uses of mask during peak period of allergenic fungal spores, etc., can be taken. With these regulations, the air quality inside class rooms can be obviously improved and the achievement level of susceptible students suffering from seasonal/chronic bronchial problem will also be improved.

CONCLUSION

Respiratory problems are important cause of chronic ill-health in school children. The survey results demonstrates that the school students are always exposed to a diverse level of fungal spores in the class room of suburban West Bengal, which are probable predictors of their respiratory health often leading to absenteeism. To overcome this, measures should be taken and awareness development is the most appropriate way in the present situation.

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