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Pollens and Allergens - A Threat to Humanity

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ABSTRACT

There is great biodiversity in the air and it varies from place to place. It always contains disease causing agents like Microbes, Allergens, Pollens and some particles. Plants give out millions of pollens through flowers into the ambient air these are causing health hazards in humans, may lead to some chronic respiratory and skin disorders. There is a great need to study Allergens and Pollens dispersed in the air, which are continuously inhaled by human beings in their life. Hence to find them out and have some preventive measures over them is of prime importance Through this study some pathological aspects of these airspora were gathered for further studies, which is of lot importance because as we all know that there is no concrete solution over allergies. Aerobiological survey was carried out over different educational institutes where different types of air borne fungal spores belonging to Zygomycotina, Ascomycotina, Basidiomycotina & Deuteromycotina were recorded. Spore types like pathogenic fungi Alternaria, Cercospora and Curvularia were also recorded.



Keywords: Allergens, Pollens, Airspora, Allergies.

Introduction:

It is well established that clouds of the spores of Fungi, Pollen grains and other organic components consequently cause environmental bio pollution Fienberg (1946), reported the importance of *Alternaria* as a potential allergen everywhere Agarwal Shivpuri (1974) reported some airborne Fungi as allergic to sensitive individuals. Tilak (1988,1998) etc. also recorded serious allergic disorders due to air borne pollen grains.

Due to humid and tropical climatic set up with prolonged monsoon, the diversity of flowering plants in this district is high. Pollen grains, the first cell of gametophyte of angiosperms are disseminated in the atmosphere for the purpose of sexual reproduction. There are many plants in the Hooghly district such as rice, mustard, coconut, grasses, *Alstonia scholaris*, *Parthenium* sp, *Carica papaya*, *Moringa oleifera* etc. the pollen grains of which are responsible for allergy. Due to humid climatic set up of this geographical location, the occurrence of slime moulds in this area is high which may also cause the respiratory troubles. In general, there is no cure for allergies, but certain treatments using antihistamines, decongestants, combination drugs are applied for the same. The medicines as brand name in the market for the treatment of allergy are Allegra, Fexidine, Telfast, Claritin, Tavist etc. Some medicines having chemical formula like Fexofenadine, Levocetirizin, Cetirizin, Salbutamol, Ambroxol etc. are generally used for the treatment of allergic patients.

The prevalence of allergic disease has increased worldwide during the last decades. Pollen allergy is the most typical form of allergic disease. The increase in its frequency during recent years is most evident. Aeroallergens and the environment play an important role in the pathogenesis of respiratory allergies (Ridolo et al., 2007). The environmental factors play an important role in the problem of pollen allergy in large cities. It has been found that the polluted pollen is more effective than non-polluted one, and mature pollen has more allergy potency than immature one. During teaching and research experiences, we have seen that a large number of patients of different parts of Hooghly district, West Bengal, India are suffering from bronchial asthma and other respiratory troubles. There may be several causes such as pollutants, dusts, industrial dusts, fungal spores, pollen



grains etc. for bronchial asthma and related respiratory troubles.

Now a day's study of pollen is an important area of research. Various pollen morphological features such as symmetry, shape, aperture pattern and exine configuration are very conservative features for the taxonomic assessment of the plants (Perveen, 2006; Bera et al., 2007; Keshavarzi et al., 2012). A number of authors Wodehouse (1935), Erdtman (1952), Rowley (1960), Tsukada (1964), Kremp (1965), Faegri and Iversen (1975), Walker and Doyle (1975), Moore and Webb (1978) and other workers studied the pollen morphology of angiospermic plants. Kholer and Lange (1979) distinguished cereal from grass pollen by LM and SEM. Pollen morphology of 49 species of family Gramineae from Venezuelan mountain have examined by Salgado-Labouriau and Rinaldi (1990) and Salgado-Labouriau et al. (1993). Pollen allergies and air borne pollen were monitored at the University of Rome in 1999 in order to determine the concentration and the quality of air borne pollen belonging to allergenic plants by Caiola et al. (2002). Sanchez-Mesa et al. (2005) reported that the occurrence of symptoms in pollen allergy patients in urban areas might be affected by local environmental factors such as sources of pollution, natural and ornamental vegetation etc. Birch (*Betula* sp.) pollen grains are the main cause of seasonal allergies in northern and central Europe.

The IgE epitopes of Bermuda grass extracts are important local allergen (Potter and Prescott, 2007). D' Amato (2007) of Italy reported that atmospheric pollen grains cause Thunderstorm-asthma. In China the airborne pollen are the major inhalant allergens, and they can elicit type I hypersensitivity such as asthma, rhinitis, and hay fever which result from a serial of complicated immune disorder in atopic individuals who contact with pollen allergens frequently and there are about 10 000 000 patients with pollinosis in China (Zhi-Gang Liu et al., 2010). According to Caimmi et al. (2012) Cupressaceae pollen allergy is a worldwide winter pollinosis. Exposure to cypress pollen has increased enormously during recent decades, and cypress pollen allergy has become a major health problem, especially in Mediterranean countries.

Delhi has published a book on pollen calendars of 12 different states in India (Singh et al., 1992) which is useful for clinicians as well as allergic patients to establish chronological correlation between the concentration of pollen in air and seasonal allergic



symptoms. A pollen calendar is useful for allergy clinics (Tilak, 2012).

Aerobiologists reported that the pollen grains of *Alstonia scholaris*, *Catharanthus roseus*, *Acacia auriculiformis*, *Moringa oleifera*, *Carica papaya*, *Mangifera indica* and *Brassica campestris* are allergenic in nature (Chakroborty et al., 2005; Ghosh et al., 2007; Talukdar et al., 2012). In the agricultural area of Eastern India, *Phoenix sylvestris* Roxb or date sugar palm is grown or cultivated and seasonal allergic rhinitis is common during the pollen season (Chakra borty et al., 2006). The pollen calendar of Agra was recorded with special reference to allergenic significance. Pollen grains of 35 species belonging to 23 angiosperm families have been identified out of a total catch of 24,220/m³ of air annually. High occurrence of pollen grains in air belonged to Asteraceae (5222/m³) and *Parthenium hysterophorus* contributed the maximum (17.91%) of the total airspora. Higher counts of pollen were found in ecozones surrounded by agricultural fields, parks and gardens. Patients of bronchial asthma with rhinitis (62.30%) were maximum followed by bronchial asthma (25.61%) and allergic rhinitis (12.07%). Maximum number of patients had symptoms between the age group of 31-40 years and male were more sensitive than females. Maximum sensitivity was caused by *Amaranthus spinosus*, followed by *P. arthenium*, *P. hysterophorus*, *Chenopodium album*, *Cynodon dactylon* and *Cassia occidentalis* (Chauhan and Goyal, 2006). *Catharanthus roseus* G. Don. (CR) or periwinkle plants are widely grown/cultivated as garden plants in the tropics and subtropics. In spite of its predominantly entomophilous nature, CR pollen had been reported to be airborne and allergenic (Ghosh et al., 2007). The qualitative and quantitative analysis of spore and pollen grains, present in the spider webs was studied in Hyderabad by Reddy et al. (2009). A continuous aeropalynological survey at Central Calcutta for two consecutive years (1985-1987) was done by using a Burkard Seven Day Recording Volumetric Spore Trap. A total of 65 pollen taxa was identified of which pollen of *Trema orientalis* showed a maximum frequency (about 68%) followed by Poaceae and Cyperaceae. A pollen calendar was prepared and seasonal periodicities were recorded. Some entomophilous pollen types e.g. *Delonix regia*, *Bougainvillea spectabilis* were also observed (Barik and Chanda, 2009). A quantitative survey of pollen flora in the atmosphere of Korba-Chhattisgarh, India was done by Shukla and Shukla (2010). Plant



pollen is one of the most common causes of seasonal allergic disease worldwide. Mango flower pollen has allergic effects in animals (Talukdar et al., 2012). The study of atmospheric pollen incidence will be helpful for proper diagnosis and treatment of allergic patients.

Pollen and its allergens

In a dry atmosphere pollen may remain stable for centuries. Anemophilous pollen (in which wind-mediated pollination takes place) has allergenic importance. In general, a pollen grain may be transported for 175 kilometers at a velocity of 10 meters/second and will sediment in still air at an approximate average velocity of 3.1 cm/second.

Pollen allergens are water-soluble proteins or glycoproteins, which make them readily available biologically, being capable of evoking an IgE antibody-mediated allergic reaction in seconds. Allergenic particles are expelled from the cytoplasm by at least two suggested mechanisms. In the first mechanism, allergens rapidly diffuse when the pollen grain is in direct contact with the mucosa in an isotonic medium, leading to immediate allergic symptoms on the accessible mucosa surfaces such as the conjunctiva and the nose. In the second mechanism a hypotonic medium (such as rain water) allows rapid hydration of the pollen grain which expels allergen-containing inhalable materials that, due to their reduced size, reach lower airways and induce asthma.

There are at least three environmental factors that induce pollen allergen release in the air: a high relative humidity, heavy rain and pollutants. In high air humidity, allergens are released from the pollen grain in a process similar to that which occurs in physiological pollinating conditions. Rarely, such as in thunderstorms, pollen grains may rupture as a result of osmotic shock, releasing allergen-containing particles. This finding, as reported in Australia, may explain the high frequency of asthma crises during heavy rainfall, which has been suggested as a risk factor for asthma crises. Grass pollen grains have diameters between 20 to 55mm, and are unlikely to reach lower airways to cause allergy. Grass pollen allergens have been found in association with smaller particles. These particles are small enough to reach the lower airways and therefore may cause allergic reactions in the distal portions of the lung. Currently, environmental pollutants, especially diesel engine exhaust particles, have been considered as significant pollen allergen releasing factors in the air. These particles contain minerals such as silica, iron, aluminum, magnesium,



manganese, sulphur, and others. According to Knox et al. (1997), pollen allergens associated with carbon particles from diesel engine fumes (DECP) would concentrate many allergic molecules in a single particle, as described with the *L. perenne* Lol p 1 allergen. Pollen grains release allergens in conditions other than high humidity or hydration. Behrendt et al. (2001) showed that pollen grains may secrete significant amounts of eicosanoid-like substances (substances that cross-react with leukotriene B₄ and prostaglandin E₂) depending on the pH, time and temperature. The pollen grain, therefore, could itself activate the airway mucosa epithelium by the secretion of pro-inflammatory mediators.

MATERIALS AND METHODS:

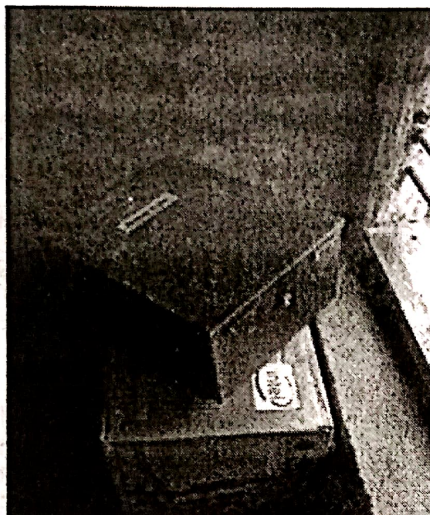
Knowledge of the airspora has depended on developing sound techniques for air sampling. Mathew Luckiest and co-workers (1946) while describing the sampling devices for air borne bacteria have mentioned following four most important characteristics of an ideal air sampler.

- a] High efficiency, collecting a large percentage of all microorganisms entering it, regardless of their origin.
- b] Simplicity of operation, preferably being a complete apparatus requiring no auxiliary equipment's such as vacuum pump, monometer etc.
- c] A constant and known air rate, so that a definite amount of air is sampled in a given time.
- d] A high degree of portability so that samplings can be made conveniently wherever desired.

These four characteristics are also applicable for sampling devices for air borne fungal spores, keeping in view these four factors, Tilak was continuously working to devise air sampler and after several trials for 3-4 years a suitable sampler was devised in 1968. This new (Tilak) Air sampler (Tilak and Kulkarni 1970) in 1971 was awarded government of India prize for import substitution. In the present investigation spore trapping was done by operating continuously this sampler. The technical description and method of working of the sampler is as follows.



Tilak's Air sampler:



The apparatus on electric power supply and helps to obtain a continuous air sampling for 8 days. The instrument has an orifice 0.6 cm in diameter projecting in a copper tube 3.0 cm. in length which is fixed in the side wall of the apparatus. Air is sucked through at a rate of 5 l/min or 0.17 ft³/min, impinging on a transparent cellophane tape 1 cm in breadth and fixed on a circumference of 67.2 cm of the rotating disc. This disc is connected with the clock mechanism arranged inside the instrument. The tape is slightly coated with Vaseline and faces the orifice tube 0.5 cm away from it.

The disc rotates continuously with the clock mechanism giving a continuous traces for 8 days. Before the tape is mounted on the glass slides at the end of 8 days, it is divided into 8 equal parts measuring 8.4 cm in length which are again subdivided into two parts measuring 4.2 cm in length and cut each piece of the tape now obtained represents the 12 hours sampling area for a day or night accordingly. The tape for 12 hours is mounted on a slide in glycerin jelly. Scanning is done by dividing this tape into 12 equal parts, each part representing one hours trace area.

The air is sucked through the tube. With the help of a small fan having three prongs and fixed in the circular opening in the cover of the sampler, so as to force air out of the collection chamber causing a negative pressure. The fan is run on 1.5 -6V D.C. motor. An exhaust hole measuring 6 X 2.7 cm is kept in a lid of the apparatus.



The instrument devised in a modified form of spore clock model and Panzer's 24 hours slide spore collector when it was compared with other spore traps, it was found that the rotor sampler (Perkins 1957) is useful only for spot sampling, although its collection efficiency is 85 %. The Hirst trap (Hirst 1952) with minimum 45 % collection efficiency has disadvantages of capital cost, power requirement and unsuitably both for identification in culture and for trapping splash, dispersed spores. The Panzer's slide spore collector (Panzer 1957) with 70 % collection efficiency has less retention capacity and requires attention after every 24 hrs; whereas the present sampler has 75 % collection efficiency greater retention capacity and is also economical.

Sampling site:

Air sampling was carried out by installing "Tilak Air Sampler" in the outskirts at various directions of Jalna, during different seasons. Jalna City is situated in the centre of the map of Maharashtra. Jalna district is located at 19.83°N 75.88°E. It has an average elevation of 489 meters (1604 feet), it is situated on the banks of the Kundalika River. Jalna district covers an area of 7,612 square kilometers (2,939 sq mi), which is 2.47% of the total area of Maharashtra state. Jalna is situated approximately at the center of the state, in the northern part of Marathwada region. Total area of Jalna district is 7718 Sq.km. population is 16,12,357 and density of population is 209.59 kms. Its MSL is 498.60 mts. It is located on the North latitude between 19° 17', 30" and 20° 40' and last longitude between 75° 40' and 77° 40'. River Godavari flows on the Southern boundary of the district, from West to East. It is called 'Dakshin Ganga', River Purna flows to the Northern part of the district, from West to East and Dudhna flows through central part of the district from West to East. Rivers Kundalika and Kalyan are its tributaries.

The climate of Jalna district is generally hot and dry. The annual average rain fall is moderate i.e. 70 cm. The Western part of the district receives low rain fall. It gradually increases towards the east. The climate is cool during the rainy season. The summer is hot. There are few forests in scattered form in the Bhokardan and Jafrabad talukas, which are not dense.

The district has a sub-Tropical climate, in which the bulk of rainfall is received from the southwest monsoon, between June to September. The average annual rainfall of



the district ranges between 650 to 750 mm. The district often experiences drought with rainfall recording as low as 400 to 450 mm. The rainy season is followed by winter, which last up to February, during which the minimum temperature ranges between 9 to 10 Celsius and maximum temperature ranges between 30 & 31 Celsius. The winter is followed by hot summer, which continues up to June. The maximum day temperature ranges between 42 & 43 Celsius during summer.

According to the Proposed Census-2011, Jalna district have 16,12,357 as a total population & density of population is 209 persons / Sq km while in 1991 census it was 177 persons / Sq km.

POPULATION			AGE GROUP 0-6			LITERATE			
RURAL	6,66,694	6,38,147	13,04,841	1,06,785	97,542	2,04,327	4,36,478	2,44,195	6,80,673
URBAN	1,59,283	1,48,233	3,07,516	24,585	22,547	47,132	1,13,475	83,946	1,97,421
TOTAL	8,25,977	7,86,380	16,12,357	1,31,370	1,20,089	2,51,459	5,49,953	3,28,141	8,78,094

Sampling methods:

Sampling was carried out by operating continuously above described air sampler, with its orifice kept at a constant height of 3 feet above the ground level with orifice towards west. The apparatus was protected from rain by a glass cover, which did not impair the sampling efficiency. Air was sampled at the rate of 5 liters a minute and the transparent cellophane tape coated with adhesive glycerol mixed with Vaseline was changed every 8 days at about 8 am the exposed tape was cut into 6 equal parts each part representing 24 hrs trace area. These 6 parts of tape were again cut into two, each representing 12 hrs trace area of day and night accordingly. The tape pieces were mounted on slides, using glycerin jelly as mount ant. Glycerin jelly has the best optical properties for visual examination.

It was prepared by mixture.

Gelatin - 1 gm

Glycerol - 7 gm

Water - 6 ml

Phenol - 1 gm

Calculation to obtain conversion factor:



1. Sampled Area
8.4 cm x 1 cm
8.4cm²
84000000mm²
2. Scanned Area
20x20x24 = 9600mm
3. Volume of air sampled per minute = 5 liters
4. Volume of air sampled in 24 hours
5x60x24 = 7200 liters
5. To convert one liter of air into 1 cubic meter by 0.001000028
6. Volume of air sampled in 24 hours in terms of cubic meters
= 7200x0.001000028
7. Volume of air sampled in the Scanned area in 24 hours
$$\frac{9600 \times 7200}{1000000}$$
$$= 69.12 \text{ Liters}$$
$$\frac{1000}{69.12}$$
$$= 14.20/ \text{m}$$

Scanning :

Scanning was done regularly. Area 9600 sq. microns of the total area of the trace obtained in a day was scanned under 10x and 45x eye piece objective combination of the microscope on 24 transverses corresponding to hourly intervals on each slide. Assuming the trapping efficiency to be 75 % the counts were converted into number per m³ of air. All times are given in Indian standard time (I.S.T.).

The identification of the spores caught was based on 1] Microscopical characters 2] Comparison with the parasitic and saprophytic fungal material collected in and around the field and studied microscopically and 3] Comparison with the cultural characters. In all possible cases specific and generic counts were made which are based on the color shape and other diagnostic features of the spore.



Conclusions:

Pollen grains contributed (2.20%, 2.34% and 2.31 %) to the total airspora during three different seasons of years 1999-2000 and even after a one and half decade there is no significant reduction in their no. and the no. of patients. Their incidence in the air was quite high not only due to the flowering but also due to short duration seasonal including grasses in the form of weeds. In general release of pollen grains from flowering takes place during forenoon and afternoon period. Nevertheless the exact correlation of pollen incidence & weather conditions has still remained unsolved problem. It may be due to just the weed Flora growing nearby premises at Jalna.

In the present investigation pollen grains of grasses, solanum, acacia, Parthenium, Mellia, Cassia, Eucalyptus, Lantana, Argemone, Brassica, Cannabis, Asphodelus, Parthenium, Azadirachta, Cocos-nucifera, Albizia lebbek and Hibiscus etc. were trapped which are of allergenic importance, has been recorded.

Grass allergy is generally related to hay fever because symptoms and causes are somewhat similar, it includes rhinitis which causes sneezing and runny nose, allergic conjunctivitis (watering and itchy eyes). Depending on the season severity of symptoms vary it includes coughing, wheezing, irritability, depression, losing appetite, sleeping problems and headaches. In the random survey it has been found that if both parents are suffering from allergies there is 66% chances for the individual to suffer from seasonal allergies and it is 60% if just one parent had suffered from allergies.

Allergy immunotherapy (AIT) involves increase of long term tolerance of the body against respiratory allergies, discovered by Leonard Noon and John Freeman in 1911. One of the most popular is the European herb butterbur (*Petasites hybridus*). A group of British researchers gave approval to butterbur's effectiveness in battling symptoms of grass allergy. Possible Control and Preventive measures for Pollen Allergens.

- 1) Allergenically significant trees should not be recommended.
- 2) Growing non allergenic trees to replace allergenic once.
- 3) People should cut or remove the allergy causing trees which are in close vicinity.
- 4) Botanists should think of reducing allergenic plants in near future.



The diversity of angiospermic plants in the district Jalna Maharashtra, India is high. The pollen calendar can be utilized for enhancement of crop yield. Pollen morphological data will be helpful for the taxonomic assessment of the plants. Monitoring of atmospheric pollen incidence will be helpful for diagnosis and management of allergic patients in this region. There is ample scope for investigation of allergic pollen grains in this region. The exact reason for allergic diseases in human body is yet to be explored.

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