Pollen and Fungal Aeroallergens Associated with Allergy and Asthma in India

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Abstract: From medical, especially clinical point of view, it is important to know the details about the occurrence of the pollen and spore load in the atmosphere. The flowering time of higher plants are events that come periodically in each season, but the time of blooming may differ from year to year, in different geographic locations. Based on differences recorded in several years of observations in airborne pollen, pollen calendars are drawn as an essential tool to allergy diagnosis and management. Allergic diseases such as bronchial asthma, allergic rhinitis and atopic dermatitis are dramatically increasing all over the world including developing countries. Today, more than 30% of the population is known to suffer from one or other allergic ailment. Major causative agents implicated are pollen grains, fungal spores, dust mites, insect debris, animal epithelia, etc. Several aerobiological studies have been conducted in different parts of the world to ascertain aerial concentration and seasonality of pollen grains and fungi.

In India, first atmospheric survey was initiated in Calcutta in 1873 by Cunningham. Since then, researchers, all over India have conducted exhaustive studies on airborne pollen types and their concentration. An "All India Coordinated Project on Aeroallergens and Human Health" sponsored by the Ministry of Environment and Forests, Govt. of India, was successfully completed by the author and his colleagues to enlist national data base on atmospheric pollen and spores. Important pollen and fungal allergens from 18 different places were identified, quantified and characterized for their allergenic properties. This and work done by other workers provide the most scientific and up-to-date information on aeroallergens of India. The most prevalent pollen allergens are *Amaranthus, Cynodon, Poaceae, Artemisia, Parthenium, Holoptelia, Prosopis, Morus, Eucalyptus, Ricinus, Cannabis* and some others.

Keywords: Pollen, Fungi, Aeroallergens, Allergy, India.

INTRODUCTION

India, with the teaming population of more than one and half billion and with the divergent geographical backdrop ranging from upland plain (Deccan Plateau) in south, flat to rolling plain along the Ganges, deserts in west, Himalayas in north, has a rich aerobiological diversity. This diversity is further enhanced with the climate, which varies from tropical monsoon in south to temperate in north. However, development of civilization often at the expense of the natural environment by pollution or biopollution stimulates the appearance of new health problems, besides increase in allergic diseases. The latter part of the 20th century has seen an increase in the prevalence of allergic diseases, implicating changing environment and lifestyle as significant causes. With the alarming increase in allergic disorders, such as allergic rhinitis, bronchial asthma and atopic dermatitis covering as high as 30% of the population in India, there is an increasing interest in the presence and movement of bioparticulate matter in the earth's atmosphere and their impact on human health. This interdisciplinary approach is known as aerobiology. The bioparticulates implicated to cause allergic symptoms are pollen

grains, fungal spores, insect debris, house dust mites, animal dander, chemicals and foods etc. [1-10]. Among all these agents, pollen grains and fungal spores are the most predominant allergens in the air. However, for the effective diagnosis and therapeutic management of these ailments, a detailed information on the daily, seasonal and annual variations of various pollen and spores in different parts of India is essential [10].

Pollen grains as aeroallergen are well studied from across the world and are important cause of pollinosis. Respiratory system is the direct target organ of airborne pollen taken in by inhalation. This results in immediate hypersensitivity disorders, in genetically predisposed individuals and late hypersensitivity in others causing clinical manifestations of allergic rhinitis, allergic alveolitis, asthma, atopic dermatitis etc. John Bostock [11] was the first to suspect pollen as the cause of hayfever (allergic rhinitis). Later, Blackley [12] established that grasses are important cause of hay fever in U.K. After more than 40 years, Scheppegrell [13] from USA, felt the need for field exploration and aerial surveys to record aeroallergens from the atmosphere. Subsequent studies from all over the world including India established pollen grains as the maior causative agent for respiratory allergic disorderes [8, 10, 14-20].

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Although pollen have been widely studied as aeroallergen through out the world, far less is known about the fungal aerosols, which are present in much higher concentration than the pollen grains in air. The fungi that produce spores and get airborne are called 'aerospores'. These are implicated in the causation of allergic diseases and infections in immuno compromised patients. They are established to cause Type I hypersensitive diseases with IgE mediated response. The first case of fungal sensitivity was reported as early as 1726 [21]. More than a century later, Blackley also suggested the association of species of Chaetomium and Penicillium with attacks of bronchial catarrh [12]. Feinberg reported respiratory allergic reactions to fungi in his patients and attributed outdoor environment as a source of fungi [14]. With the studies establishing the role of fungal spores as a major causative agent for the respiratory allergic disorders [16, 17, 22, 23], the seasonal and annual variations in the bioaerosols have been extensively studied in different parts of the world including India [24-29]. Their knowledge is of paramount importance for diagnosis and therapeutic management of fungal allergy and asthma.

OBJECTIVE

In this review, I have made an effort to provide aerobiological perspective of environmental pollen as well as fungi from different parts of India published by different workers. The historical development of aerobiology, prevailing airborne pollen and fungal flora in different ecogeographical regions of India and their clinical significance in the diagnosis and management allergy and asthma has been discussed.

SOURCE OF AEROALLERGENS

Pollen as Aeroallergen

The transport of pollen grains by wind or by the insects, from floral anther to recipient stigma is the critical reproductive event among higher plants. The dispersion of replicate units in massive abundance assures the success of wind pollination as well as its human health effects including asthma, rhinitis, atopic dermatitis etc. Pollen prevalence (grains per cubic meter) at any point reflects (plant) source strength and location as well as the dynamics of the intervening environment conditions such as climatic factors, pollution and degree of exposure. The presence of pollen, profile of species, concentration etc. depends on various climatic factors such as temperature, humidity, wind direction, sunshine, substrate precipitation and other seasonal factors. Because of change in the climatic conditions, the study of variations in the diurnal and seasonal prevalence becomes very important [30].

Fungi as Aeroallergen

Fungi possess highly evolved mechanism of spore liberation due to which the spores remain suspended in the air for a varying duration i.e. few hours to several days. Fungi and fungal particles can clearly induce an allergic response in susceptible individuals. Typical symptoms include wheezing, cough, rhinorrhea, itchy nose, sore throat, sinus congestion etc. [31, 32]. The development of allergies to fungi follows the same biological phenomenon as allergies to other environmental allergens. Dead fungi are able to produce symptoms just as well as live fungi [33]. Hayward and coworkers reported a separation and characterization of antibodies to moulds in human sera and the role of human precipitins to common fungal antigens in allergic reaction, which was later proved by Pepys [34, 35].

Monitoring Airborne Allergens

Sampling Devices

Monitoring of airborne allergens is carried out by various gravimetric, impaction, and filteration sampling devices [36-38]. In addition, new immunochemical techniques are also used for detecting allergenic pollen and measuring of the size of allergen-carrying particles [39, 40].

A. Gravimetric Sampler

This is based on the principle that bioparticulates settle down on a surface due to gravitational force. The Durham gravity-sampling device [36] consists of two horizontal disks with a diameter of 22.1 cm and 8.1 cm. After exposure, the slides are mounted in a drop of molten glycerin jelly, for various allergen types trapped. However, this sampler is no more used in developed world, but developing countries still report the use of gravity settlement method for fungal spores/colonies.

B. Impaction Sampler

Rotorod Sampler

Rotorod sampler developed by Perkins [37] has leucite rods of 1-3 mm coated with adhesive silicon grease are used to collect air borne particles. It is a lightweight portable sampler operated by DC power. The exposure time can be adjusted according to requirement. There are three models available in the Rotorod Aeroallergen Models (40s, 85s and 95s) and the pollen/spore catch obtained by these three models is almost similar. These three samplers are used to study both pollen and fungal spore types on continuous basis.

C. Suction Samplers

The method requires suction of certain volume of air according to a known velocity and for a chosen duration on trapping.

Hirst Trap

The Hirst spore trap was most commonly used in UK. In this trap bioparticulates adhere to slides coated with glycerin jelly and slides are replaced each day with fresh slides and provides quantitative data [38]. The method requires suction of certain volume of air according to a known velocity and for a chosen duration on trapping. It records the atmospheric concentration of pollen grains, fungal spores, and other biological particles as a function of time through morphological identification.

Burkard Seven Day Volumetric Sampler

The hirst trap was later modified to Burkard trap in which slides were replaced with a drum, which can rotate and run continuously for seven days with a definite speed with suction rate of 10 L of air per minute. Adhesive coated tape mounted is used in on a rotating drum Burkard continuous seven days sampler. The drum is connected to a timer and rotated at constant speed. The tape is changed every seven days. Exposed tape is cut in seven strips corresponding to seven days and mounted on a slide. This is one of the most widely used samplers to study diurnal or seasonal trends for pollen grains as well as fungal spores all over the world.

Burkard Portable (Slide) Sampler

Burkard slide sampler is a compact battery operated sampler. It has a rectangular orifice at the top end and a slit on the slide to insert micro slide. The microslide is coated with glycerin jelly. The sampler sucks in 10 L of air per minute. The particles get impacted on the slide in the form of a streak. The slide is then mounted in glycerin jelly and scanned for pollen grains/fungal spore count under light microscope.

Burkard Portable (Petriplate) Sampler

The Burkard Petriplate Sampler is similar to slide sampler except that it has a stage to hold the Petriplate and a sieve to cover the petriplate and on top a lid to cover the sieve. On the cover is an opening from where the air is sucked in at the rate of 10 liters per minute. These samplers are most convenient for outdoor and spot sampling and places where power connection is not available.

Andersen Sampler

The best device to obtain culturable fungal spore count is the Andersen Volumetric Sampler. It has 2stage to 8 stage sampler. It uses petriplates in which media is kept under different sieve size present in decreasing order of pore size. Each sieve has 400 pores. The sampler sucks in 28.3 L of air per minute. The air passes from the orifice to all the six petriplates before passing out. The particles of similar aerodynamic dimensions are impinged on the same plate. The two stage Andersen Sampler is quiet efficient and less time consuming in number of plates to be examined after exposure.

IMMUNOCHEMICAL ASSAYS

The immunochemical assays for airborne allergens relied on large (e.g. 20X25 cm) fibre glass filters exposed for 24 hr period in high volume sampler. They are rated for continuous operation and adapted to receive filter support.

In a study carried out in Arizona, variability of allergen shedding of airborne cat allergen was carried out by Immunochemical assay. Cats were placed in a lucite chamber with an air sampler attached. Radioallergosorbent (RAST) / Enzyme Linked Immuno Sorbent Assay (ELISA) inhibition type and monoclonal two-site radioimmunosurveys (RIAs) are used to express air concentrations in allergy units (AU) [39,40]. However this is still considered a research tool.

ANALYSIS OF BIOAEROSOL SAMPLES

Regardless of their method of collection, samples of mixed biologic aerosols are analyzed by one of the following techniques

A. Direct Microscopy (Slides)

The microscopic identification of distinctive particles (pollen/fungi) is an approach validated by years of practical applications of both gravimetric and volumetric samplers. The exposed slides are examined directly by microscope because a variety of particles, including pollen grains, certain basidiospores, ascospores and spores of rust, smuts and downy mildew are recognizable but fails to grow on most laboratory media. The particles/ spores are identified based on their characteristics such as shape, size and other morphological features of spores. Therefore it is a most dependable way to identify most of the pollen/fungal spores.

B. Culture Analysis (Petriplates)

This includes the tally of colonies produced in culture or semisolid media. In this the petriplates exposed for sampling are incubated at appropriate temperature (28-30°C) and impacted spores are allowed to grow for a couple of days till colonies start forming. The colonies are identified based on their colony characteristics such as color, shape and other morphological features of the mycelia and spores to the lowest taxonomic rank possible. Each colony represents one spore and considered colony-forming unit (CFU). In addition, different atlases and literature can also be used for authentic identification.

ANALYSIS OF DATA

After suspended particles have been collected on the slide or in a suitable medium, these particles can then be counted and identified to the nearest taxonomic rank. Environmental scientists looking for non- viable particles also used the techniques used to extract viable cells and particles carrying them from the air. The most efficient methods of removing suspended particles from the air, example, filtration through fine pore matrices, might be adequate for resistant forms of microorganisms, such as spores, but can be less damaged environmentally resistant vegetative cells. The total number of cells present can be estimated by microscopic examination, sometimes with the help of stains or fluorescent tags.

The concentration of pollen/fungal propagules are calculated as per the formula given here:

Total number of pollen grain / colonies

----- X 1000

Total volume of air sampled

The counts are expressed as number of pollen grains/m³ or colony-forming units (CFU/m³) as the case may be.

C. Immunoassay

Immunochemical analysis following descending elution, exposed from filters offers an analytic approach to dust without potential or defined form (e.g. – fungi, pollen, dander, seed pomace, arthropod effluvia etc). One advantage in immunoassay for airborne microorganisms is that the amount of materials needed to measure the concentration of viable air contaminants is much lower than that needed to gravimetrically quantitate nonviable particles where only one or a cluster of cell lands on an appropriate solid nutrient medium, or lawn of host cells, a microscopic fungal or bacterial colony, or viral plaque, will develop. These isolates can then be identified specifically using tests of biochemical and immunological reactions conducted on sub cultures of the original material.

AEROALLERGEN SURVEYS IN INDIA

Pollen

Airborne pollen and their concentration vary in the different seasons depending upon the flowering seasons and climatic factors, which are quite variable not only in different regions of the country but also in different parts of the world.

In India, first atmospheric survey was initiated in Calcutta in 1873 by Cunningham [41]. Since then, researchers, all over India have conducted exhaustive studies on airborne pollen types and their concentration. An "All India Coordinated Project on Aeroallergens and Human Health" sponsored by the Ministry of Environment and Forests, Govt. of India, was successfully completed by Singh and his colleagues [42]. Important pollen and fungal allergens from 18 different places have been identified, quantified and characterized for their allergenic properties. This provides the most scientific and up-to-date information on aeroallergens in India. Recently Singh and Chandni published a review on importance of aerobiology in Allergy and asthma diagnosis and management [10].

From Northern India 43 predominant types of pollen have been recorded. The dominant types are: *Artemesia*, Asteraceae, *Cassia*, *Casuarina*, *Cedrus*, *Eucalyptus*, *Holoptelea*, *Morus*, *Pinus*, Poaceae, *Prosopis Putranjiva*, *Quercus* and *Xanthium* are some important contributors in the air [10,42,43]. From Lucknow important airborne pollen allergens are described recently and *Holoptelia* is one of the dominating pollen in spring [44].



Monthly Pollen Concentration at Delhi (Total Types) for Seven Consecutive Years (1990-97)

Figure 1: Monthly pollen concentration at Delhi.

In an aerobiological survey from Delhi, ninety-four pollen types were recorded and the major contributors included *Morus*, *Cannabis*, Chenopod/Amaranth, *Prosopis*, *Artemisia*, and *Eucalyptus* [45,46]. A significant reduction in pollen concentration was observed in subsequent years (Figure 1). The concentration of *Morus*, *Cannabis*, *Prosopis*, and *Artemisia* pollen decreased considerably. It is suggested that the reduction in pollen numbers from 1990 to 1997 in Delhi could be due to massive clearing of vegetation for developmental activities of the city such as roads, fly overs. Metros and industrial estates.

From Central India, surveys carried out by different workers at Bombay, Gwalior, Nagpur, Bhopal and Kolhapur revealed that the dominant pollen types are from the Poaceae, Asteraceae, Apocynaceae, Rosa, *Ricinus, Ailanthus, Holoptelea, Cyperus, Cicer, Argemone, Cocos nucifera* and *Hibiscus* [10,42,43]. The survey conducted at at Pune [42] revealed *Parthenium* to be the highest contributor to the pollen load with two peak seasons i.e. from September to November and January to April. *Cocos* and *Cassia* were observed throughout the year. *Cocos* pollen were recorded in high concentration in April- May and November-December.

In West Bengal, 59 types pollen were revealed from air - their maximum concentration was recorded in May. Important dominant types are *Areca catechu*, Asteraceae, Chenopodiaceae, *Cocos, Pongamia, Trema orientalis* and *Xanthium*. It is interesting observation that Gupta Bhattacharys and her colleagues have identified pollen of different species of Cassia from the air and assessed their allergenicity [46].

Pollen calendars are very useful for clinicians as well as allergic patients to establish chronological correlation between the concentration of pollen in air allergic and seasonal symptoms. Based on aerobiological data obtained from India, pollen/ flowering calendars have been prepared for Calcutta, Sambalpur, Gulberga, Imphal, Kodai kainal by different workers [46-48]. The Centre for Biochemical Technology (Now CSIR-Institute of Genomics and Integrative Biology) has published a book entitled "Pollen Calendars of 12 different states in India" which provides pollen season for important grasses, weeds and trees prevalent in 12 states of India. The Calendar has proved to be very useful tool for diagnosis and therapeutic of allergic patients by physicians as it contains botanical and vernacular names of the plants in different langueses of the states [49].

Fungi

Fungi are ubiquitous in nature and are reported to be prevalent from different parts of India, both in indoor and outdoor environments. Airborne surveys of fungal allergens have been reported from different parts of the world including India.

OUTDOOR AEROBIOLOGY

In India, many reports provide information of prevalence of fungi in ambient air [16, 29, 50, 51] *Alternaria* is reported as the dominant fungal type from Delhi [29, 52]. A survey conducted for culturable and non-culturable fungi reported 98 fungal forms with *Cladosporium* contributing 25-40% of total airborne fungi followed by *Ustilago* (smuts) (24%) *Aspergillus flavus* (10-13%), *Alternaria* (11%) and *A. niger* (8%). Basidiomycetes contributed 7-13% at different sites [23, 53].

Studies carried out in Gaya, Gauhati and Kolkata revealed that *Cladosporium*, *Alternaria*, *Aspergillus*, *Penicillium*, *Curvularia*, *Helminthosporium*, *Aureobasidium*, *Neurospora*, *Mucor* and *Nigrospora* are the major types recorded from Eastern India. From Pune and Kolhapur, the dominant fungal forms isolated are *Cladosporium*, *Alternaria*, *Curvularia*, *Nigrospora*, *Periconia*, *Helminthosporium*, smuts, rust, *Aspergillus* and *Penicillium* [42].

A volumetric paired assessments of airborne viable and non-viable fungi in five outdoor sampling stations [42] in a rural agricultural area of India concluded that: (i) a rich fungal airspora existed in the rural study area, (ii) to achieve representative information on the total airborne fungal spores of an area, the monitoring in multiple sampling stations is preferable over a single sampling station; for viable fungi, however, one station can be considered, (iii) the percentage of airborne fungal viability is higher in rural agricultural areas.

INDOOR AEROBIOLOGY

The spectrum of indoor airborne mold spores, such as in homes, offices, and other workplaces, differ from place to place due to the influx of spores from outdoor air through ventilation and air exchanges.

In India, indoor fungal survey has been reported from several cities across different geographical region. At Vishakhapatnam, a total of 8909 and 9327 CFU/m3 were recorded from inside and outside house, respectively with 47 types identified. The dominant fungal types were *Cladosporium*, *Penicillium nigricans*, *Aspergillus versicolor*, and *Aspergillus oryzae*. While in Solan and in Shimla sampling conducted in a wet house revealed *Penicillium* as the most dominant types contributing 30.7% followed by *Aspergillus* sp. (15.4 %), *Alternaria* sp (10.5%). In the same place sampling conducted in a mud house revealed *Aspergillus* sp. at 35.1% as the most dominant contributor followed by *Penicillium* (26.9%). The other dominant types were *Alternaria*, *Cladosporium*, *Curvularia* and *Fusarium* [42].

In West Bengal (Kolkata), the volumetric assessment of airborne culturable and nonculturable fungal spores showed higher frequencies of *Aspergilli* /*Penicilli, Cladosporium, Alternaria*, and smut spores by Burkard Sampler whereas Andersen Sampler showed the prevalence of *Aspergillus niger, Aspergillus flavus and Cladosporium cladosporioides* in large rural indoor cattle shed [39, 54].

In Delhi, an indoor survey of fungi in the homes of asthmatic / allergic children [29, 55] revealed highest fungal load in the month of January while the lowest in June in indoors A high viable mold concentration was observed in the homes of asthmatic children in Delhi. The predominant fungal types observed were *Aspergillus niger, A. flavus, A. fumigatus, A. nidulans, Alternaria* spp, *Cladosporium* spp, *Penicillium* spp, *Rhizopus* spp, and *Curvularia* spp etc. The houses in Delhi contain rich and varied concentration of fungi, almost parallel to what is encountered just outside the air.

The indoor work environments are greatly influenced by fungi especially occupational sites employing organic raw materials e.g. granary, poultry, flour mills, bakery, sugar factory etc. Survey conducted at working environments by Singh and his students in bakery, poultry, sugar factory and libraries in Delhi revealed, *Aspergilli-Penicilli* and smut spores as significant contributors in indoor air [52-55].

CLINICALLY IMPORTANT ALLERGENS

Pollen as Allergens

Pollen causing allergy are quite variable in different ecozones which makes it very important to identify pollinosis causing species from every region, and prepare extracts from them for diagnosis and immunotherapy for the benefit of allergy sufferers.

Based on clinico-immunological evaluation of pollen antigens, important allergenic plants of India have been identified (Table 1). The work on pollen allergy was initiated in the 1950's by Shivpuri in Delhi. Subsequently, Kasliwal and his colleagues reported important pollen allergens of Jaipur [56]. Shivpuri and

Table 1: Common Allergenic Plants of Different Seasons in India

Spring Feb-April	Autumn SeptOct	Winter NovJan.
GRASSES Cynodon dactylon Dicanthium annulatum Imperata cylindrica	Bothriochloa pertusa Cenchrus ciliaris Hetropogon contortus	Cynodon dactylon Eragrostis tenella Phalaris minor
Paspalum distichum Poa annua Polypogon monspeliensis	Pennisetum typhoides Sorhgum vulgare	Poa annua
WEEDS		
Cannabis sativa	Amaranthus spinosus	Ageratum conyzoides
Chenopodium murale	Artemisia scoparia	Argemone mexicana
Parthenium hysterophorous	Cassia occidentalis	Chenopodium album
Suaeda fruiticosa Plantago major	Ricinus communis Xanthium strumarium	Asphodelous tenuitolius Ricinus communis
TREES		
Ailanthus excelsa	Anogeissus pendula	Cassia siamea
Holoptelea integrifolia	Eucalyptus sp.	Salvadora persica
Prosopis juliflora Putranjiva roxburghii	Prosopis juliflora Cedrus deodara	Mallotus phillipensis Cedrus deodara

Parkash [57] observed *Prosopis juliflora* as a major cause of pollinosis with 12% patients showing a positive skin reaction. Later, important pollen allergens were identified for Delhi by Shivpuri and his colleagues. They were: *Ageratum*, *Ailanthus*, *Amaranthus*, *Anogeissus pendula*, *Artemisia*, *Cassia siamea*, *Cenchrus*, *Chenopodium*, *Cynodon*, *Ipomoea fistulosa*, *Paspalum distichum* and *Poa annua* [58]. We recorded positive skin reactions in 16.9% patients to *Pinus roxburghii* from the foothills of Himalayas [59].

From Northern India, important allergens identified are: Prosopis juliflora, Ricinus communis, Morus, Mallotus. Alnus, Quercus. Cedrus. Argemone, Amaranthus, Chenopodium, Holoptelea, and grasses. From Central India the important pollen allergens are: Argemone, Brassica. Cannabis, Asphoedelus, Parthenium, Cassia, Azadirachta, grasses, Alnus, Betula, Malotus, Trewia nudiflora. From Eastern India, allergenically significant pollen types were found as: Lantana, Cucurbita maxima, Cassia fistula, Cocos nucifera and Calophyllum inophyllum. Recent studies based on clinical and immunologic parameters reported Phoenix, Ricinus communis and Aegle marmelos as causative agents of allergy in this region [42].

From South India *Cassia*, *Ageratum*, *Salvadora*, *Ricinus*, *Albizia lebbeck* and *Artemisia scoparia* have been reported as important aeroallergens [60, 61]. Subbarao *et al.* [62] recorded allergenicity to *Parthenium hysterophorus* pollen extracts in 34% of allergic rhinitis and 12% bronchial asthma patients from

Bangalore. Agashe and Soucenadin [63] recorded high skin reactivity to *Casuarina equisetifolia* in patients from Bangalore.

Clinical studies undertaken by author and his colleagues at various medical centres under the All India Coordinated Project (AICP) on Aeroallergens and Human Health [42] sponsored by the Ministry of Environment and Forest, revealed important allergenic pollen for various regions in India. At Chandigarh, skin sensitivity was highest against Rumex acetosa and Ailanthus excelsa (17.6%), followed by Trewia nudiflora (9.7%), Argemone mexicana (9.5 %), and Cedrus deodara (9.3%). In Delhi, 12.6% of the atopic population was positive to Amaranthus spinosus, 8.5% to Populus deltoides and 7.5% to Dodonea viscosa, Bauhinia vareigata. In Calcutta, 28.8% of the patients were sensitive against Solanum sysimbrifolium, 21.1% to Crotalaria juncea and 18.2% each to Ricinus communis and Ipomea fistulosa. In Trivandrum. maximum skin reactivity was recorded to Mallotus phillipensis (12.1%), followed by Prosopis juliflora (6.3%) [19]. For the first time, Cedrus deodara (Pinaceae) pollen has been recognized as a new allergen from India in the patients from the Himalayan region, where Cedrus deodara occurs naturally [64].

Fungal Allergens

The prevalence of respiratory allergy to fungi is estimated at 20 to 30% among atopic individuals and

upto 6% in the general population [2, 51, 65]. The major allergic manifestations induced by fungi are asthma, rhinitis, allergic bronchopulmonary mycoses, and hypersensitivity pneumonitis [3, 27].

Acharya [60] investigated on 300 nasobronchial allergy patients in Andhra Pradesh, important fungi wereAspergillus flavus, Helminthosporium, Neurospora, Candida, Cladosporium etc. From Bangalore, Mucor mucedo, Fusarium solani, Curvularia, Nigrospora etc are observed to be important allergens [61].

The atmospheric concentration of *Fomes* was recorded by Gupta *et al.* [52] the maximum counts (67 spores/m3) were observed from the North Delhi site in the month of July 1989, compared with 550 spores/m3 in the South Delhi site. Marked skin positivity (2+ and above) varied from 9.8% to spore to 22% to whole body of *Fomes pectinatis* [66] *Ganoderma lucidum*, has been reported as an important allergen from Basidiomycetes in Delhi [65].

MOLECULAR CHARACTERIZATION OF ALLERGENS

The need for standardization of allergenic extracts has been recognized since the advent of immunotherapy [66,67]. The early attempts towards standardization were based on consistency in crude w/v ratio or estimation of PNU content of the extracts. But it was found to be a poor indicator of allergen content.

The standardization of allergenic extracts can be done by biological standardization. The concept of biologic equivalence test was established by Northern Society of Allergology and adopted by European investigators [68]. One HEP unit was defined as a positive skin prick test which gave a wheal diameter equivalent to that given by a prick with 1 mg/ml concentration of histamine hydrochloride. Later on Dirksen *et al.* [69] designated 1 HEP as concentration of extract that gives a wheal diameter equivalent to 1 mg/ml of histamine hydrochloride.

ENVIRONMENTAL MANAGEMENT OF POLLEN ALLERGENS

- 1. All the allergenically significant trees need to be deleted from the list of Recommended tree plantation in gazette of India.
- 2. Ornamentals, insect/bird pollinated and medicinally important trees or others which can

help in controlling pollution, should be encouraged in various tree plantation and afforestation programs.

- 3. The existing allergenically significant trees need to be replaced with non-allergenic trees in a phased manner.
- 4. On medical ground, citizens should have the right to cut or demand removal of allergy causing trees in close vicinity. However, these should be replaced with some non-allergenic trees.
- 5. A genuine beginning needs to be made by sensitizing tree lovers/ horticulturist/ foresters/ botanists and other associated with tree plantation so that the share of allergenically significant plants could be minimized in the near future. The earlier it is done the better.

PREVENTION OF FUNGAL ALLERGENS

- 1. It is important to isolate and identify important airborne fungi that cause allergic disorders.
- 2. Knowledge of life history of these fungi is essential in identifying their source and environmental conditions that help in getting airborne in significant concentration.
- 3. Prevention is better than cure, therefore it is suggested that substrate on which microorganism build up their numbers should be removed from the work place.
- 4. Reduction of moisture inside the work place.
- 5. Air quality can be enhanced by periodic maintenance of air treatment plant, fumigation, application of antifungal agents' etc.
- 6. Personal filter masks of the pore size sufficient for entry of respirable size particles may be beneficial at the work place.
- 7. Environmental factors like air pollution and smoking also can aggravate the existing symptoms. These should be controlled.

CONCLUSIONS

India is a tropical country and its fauna and flora is very variable from western world. Important airborne pollen and fungal allergens prevailing in different geographic regions of India are enumerated in the current review. Based on seasonality and concentration of atpospheric pollen a Calendar from different states was prepared as diagnostic and therapeutic aid to the allergy patients. This is of direct significance to allergy practitioners. The detailed information on indigenous pollen and fungal allergens is of paramount importance and very useful in diagnosis and management of allergic patients in the country.

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Received on 20-06-2014

Accepted on 16-07-2014

Published on 31-08-2014

DOI: http://dx.doi.org/10.14205/2310-6980.2014.02.01.4

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