SUMMARY

This study gives an insight into various aspects of aeroallergens especially focused on Parthenium hysterophorus and Holoptelea integrifolia pollens.

Aeroallergens:

Aeroallergens are air-borne particles that induce allergic reactions in sensitized subjects and can cause respiratory, cutaneous, or conjunctival allergy.

They are a subset of various forms of aerosols starting from submicronic particles to relatively larger pollen grains, fungal spores, animal emissions, and biogenic debris. Pollens and fungal spores have been studied most extensively in the outdoor environment because they are relatively easy to sample and identify.

To be clinically significant, aeroallergens must be buoyant, also present in significant concentration, and allergic in nature.

Grass pollens and Alternaria spores are typical examples of frequently encountered aeroallergens.
In general, entomophilous (insect-pollinated) plants produce scanty, heavy, or sticky pollens that do not become air-borne.

Anemophilous (wind-pollinated) plants represent only about one-tenth of the more than 2,50,000 pollen-producing species.

Depending on their season, anemophilous spores can reach concentrations >100 grains/m$^3$, can remain air-borne for days, and can be carried hundreds of miles from their point of origin.

In contrast to entomophilous plants, anemophilous plants possess large stamens borne on long, well-exposed filaments, often organized as catkins. Their flowers usually lack color, scent, and nectar and release large quantities of pollen in warm and dry weather.

Particle size is a critical physical attribute of aeroallergens and an important consideration in the pathogenesis of allergic rhinitis and bronchial asthma.

Protective mechanisms in the nasal mucosa and upper
trachea bronchial passages remove larger particles, so only those of 5 micrometers (µm) or less reach the alveoli of the lungs. Intact pollen grains, range in size from about 15 µm to 75 µm. Thus, the conjunctivae and upper respiratory tract get exposed to the highest dose of aeroallergens. Until recently, pollen-induced asthma remained enigmatic because the available evidence suggested that inhalation of pollen grains encountered naturally could not reach the bronchial tree. But studies done recently, give evidence which supports the importance of pollen allergens associated with submicronic aerosol fractions in asthma.

To detect the aeroallergens we have to understand the environmental conditions that influence aeroallergens and on them depends the sampling equipment. The selection of sampling device for a given study depends on the aeroallergen of interest. Thus knowledge of aerodynamic size, concentration and viability of the aeroallergen is must.
In urban environment samples are taken mostly from unobstructed rooftops of buildings. Airflow patterns between tall buildings may significantly affect pollen concentrations at locations only a few hundred feet apart. In our study the site was a terrace of institute situated on 13m from the ground.

**Pollen Aeroallergens:**

Pollen is a fine to coarse powder containing the microgametophytes of seed plants, which produce the male gametes (sperm cells). Individual pollen grains are small enough to require magnification to see detail.

Most comprehensive keys for pollen identifications rely on size, shape, germinal apertures, sculpture and cell contents. The size of pollen of a particular species is predictable when mounts are prepared fresh with a consistent medium.

It’s observed that pollen grains of small size have wider distribution and are capable of contributing a significant amount of pollen to the atmosphere. Due to small
size the pollen tend to float in the air for a longer duration than the larger sized pollens which tend to settle down on the ground more quickly than the smaller ones.

Trees are more exposed due to their height so chance of pollen loss is greater and this is compensated by the higher production of the pollens.

Pollen is nature’s gift to mankind as it is responsible for pollination, fertilization, seed and fruit setting and multiplication of plants. But some of the pollen after getting into the air stream keeps floating in the air before settling down on the ground. Some of this pollen on coming in contact with human beings punishes them by producing allergic manifestations.

**Pollen Allergy:**

Pollen Allergy is a type of airborne allergies (wind-blown pollen generated by trees, grass, plants and weeds), these are a major cause of illness and disability. For the majority of allergy sufferers in India, inhaling airborne allergens produce a variety of allergic symptoms. Because
pollen grains are small and buoyant, they can remain airborne for hundreds of miles.

Typical allergic symptoms caused by airborne allergens include sneezing, running nose and nasal congestion (allergic rhinitis or hay fever). When an allergic victim's eyes are affected, they may experience itchy, watery and red eyes.

Inhaling airborne allergens may cause bronchospasm and difficulty in breathing. Contact allergies from touching plants, grass, leaves and even substances such as latex, can trigger hives or a skin rash. Many medical personnel develop a latex allergy from latex protective gloves. The same airborne pollens can also cause contact allergies or skin allergies when pollen comes into contact with the skin.

The major pollen allergens are divided into three subcategories: tree pollen, grass pollen and weed pollen.

Our study was focused on Holoptelea integrifolia and Parthenium hysterophorus.

**Holoptelea integrifolia:**
**Holoptelea integrifolia** belongs to the family Ulmaceae, having 15 genera and about 200 species, distributed over tropical and temperate regions of the Northern Hemisphere including the Indian peninsula to Indo-China and Srilanka.

Indian Elm (**Holoptelea integrifolia**) is a large deciduous tree, growing up to 18 m tall. It has grey, pustular bark covered with blisters that is smooth when young, peeling in corky scales on old trees. Alternately arranged leaves are elliptic-ovate, 8-13 cm long and 3.2-6.3 cm wide, smooth, with entire margins, and a pointed tip (apex acute or acuminate). Leaf base is rounded or heart-shaped, main nerves 5 to 7 pairs; Stipules are lanceolate (lance-shaped). Crushed leaves emit an unpleasant odour. The flowering time of the tree is January to February, whereas, fruiting is seen in April to May.

**Parthenium hysterophorus:**

**Parthenium hysterophorus** is a species of flowering plants in the aster family, Asteraceae. The name is derived from the Greek word *parthenos*, meaning "virgin," or
*parthenion*, an ancient name for a plant. Members of the genus are commonly known as fever few. Notable species include Guayule (*P. argentatum*) and *P. hysterophorus*, a serious invasive species in the Old World. Parthenium weed was first discovered in Queensland Australia. Parthenium is a major weed in Australia.

Parthenium hysterophorus is a deep rooted herb to four feet tall at fully flowered stage. Parthenium is an annual herb. It is also known as carrot grass, congress grass, star weed, and gajar ghass. An erect, much-branched, vigorous growing annual herb. Its stems become woody with age. Its leaves are pale green, deeply lobed and covered with fine soft hairs.

Flowers: tiny, creamy white and in numerous distinctly 5-cornered heads about 4mm across adorn the tips of the many stems in other words the large open branched terminal sprays.

Parthenium bears white flowers which disperse due to wind and water.
Parthenium Weed generally flowers in spring and early summer. It produces flowers and seeds throughout its life and then dies in late autumn. During summer, Parthenium can flower within four weeks and if the conditions are right, it can flower and seed all year long. Seed can be spread by wind, water, contaminated soil and stock feed and harvested material, vehicles, machinery, stock and other animals.

Reference slide preparation:

First of all a general field survey was made within the district. Extensive botanical study of the district was made. For the collection of polliniferous material, an intensive field survey of flora of Jaipur district was conducted, within the range of Jaipur district. Field trips of adjacent places were made frequently for collection purpose. Flowering periods, flower colours, size of flowers, pollination mechanism of collected plants were observed during collection. Periodic field trips to various parts of the city and
the adjoining areas covering various seasons of the year were made.

Plant collections for the present investigation, were made from the adjacent vegetation of the area of Jaipur district. Field trips were made several times in a month to different localities within the area marked for this study. The survey of the vegetation of Jaipur district was conducted.

In the field each polliniferous material was collected in separate polythene bags then brought to the laboratory and anthers or very small flowers were fixed in F.A.A. (Formaline 5%, acetic acid 25% and absolute alcohol 70%) in small vials. A plant specimen for herbarium was also maintained for reference and identification purpose. All the identification has been done in the "Herbarium" Department of Botany, University of Rajasthan, Jaipur. Reference slides were named of the specific species, after identification and confirmation.
Reference slides of acetolysed pollen reference slides were prepared by the method suggested by Erdtman (1952).

Unacetolysed pollen preparations were useful for identification and comparison of the air borne pollen caught on the exposed slides as both had retained the protoplasm, while acetolysed preparations helped in the study of pollen morphology (aperture, ornamentation etc.) in greater detail, because of the dissolution of the pollen protoplasm.

The morphological characters had been noticed under shape, size aperture, ornamentation, thickness of wall (exine and intine) etc. However, of these, the aperture characteristics are of primary importance.

These reference slides were stored and used for pollen identification.

**Aerobiological sampling:**

Aerobiological sampling was carried out to monitor the qualitative and quantitative prevalence of aeroallergens through Burkard 24 hr. spore trap system, which is a type of

It is a compact unit with built-in pump, designed to sample airborne particles, such as fungus spores and pollens for 24 hrs periods. Air was drawn in at 10 liters/min through a standard orifice size 2 x 14 mm. and airborne particles were deposited on a glass slide coated with adhesive mountant, in our study we used Glycerine jelly as a mountant.

The spore trap system was loaded with a glass slide on which Glycerin jelly was coated. This slide was placed in the sampler and the trapping of the aerobiological material took place from one end to other at a speed of around 2mm/ hr, covering the whole slide in 24hr. The date of slide was noted on the top of the slide and also a small arrow is put on the side of the date, the arrow indicates the direction
in which the slide moves and thus the air gets struck on the glycerine jelly in 24 hr.

The site selected for the entrapment of the air spora was the building of Asthma Bhawan situated at Vidyadhar nagar, Jaipur. Asthma bhawan is on the outskirt of the old city and is on the north western side of it. It is girdled by Aravali ranges, on the north and north eastern side, which have the forest vegetation. North – North West and West side of Asthma bhawan are mostly less populated and have open fields. The areas from West – South and South – East have thickly populated city areas.

The trapping apparatus was installed at a height of 13 meter from ground and 24 hrs of pollen sample was collected from 12:00 noon to 12:00 noon.

Samples were analyzed by direct microscopy. Identification of airborne pollen done by comparing them with the corresponding pollen in the reference slides prepared. Number and distribution of apertures and various patterns of ornamentation of exine to be considered
as the chief characters employed for the identification of atmospheric pollen, as confirmed from standard literature for pollen identification by Erdtman (1952); Faegri and Iverson (1964).

The counting of the pollens has been performed on a daily basis using British Aerobiological Federation manual (Guide to trapping and counting).

Allergic Prevalence:

- Prevalence is a term which means being widespread.
- In science, *prevalence* describes a proportion (typically expressed as a percentage).
- In our study the prevalence (ratio) was = Number of Skin Prick test positive for Holoptelea/Parthenium in a given period (Month)÷ Number of Total Skin Prick test during this period(Month)x100%.

The prevalence of the Holoptelea and Parthenium allergy in Jaipur was also calculated. This was done by collecting the data of the entire Skin prick test done at Asthma Bhawan and out of the total we segregated the data
of Jaipur residing subjects only. Among these data the allergy test positive for Holoptelea/Parthenium in a given period was divided by the total data of Jaipur of the same period to calculate the prevalence of the individual pollens i.e either Holoptelea or Parthenium.

During the study period a total of 36421 pollen grains/m$^3$ were recorded by 29 pollen grain types identified during the study period.

The observations are based on the study of slides exposed every 24 hours.

Among the annual catch the dominance of 8 pollen types (>1% of the total pollen catch) gave 34801 pollen grains/m$^3$ in the 2 years. These were Poaceae > Chenopodium > Amaranthus > Asteraceae > Holoptelea > Brassica > Cassia Spp. > Parthenium hysterophorus > Azadirachta.

Two distinct pollen seasons February to April and August to November can be marked in Jaipur. The second season i.e August to November showed more number of Pollens as compared to the first season i.e February to April.
Highest pollen concentration was recorded in August (9740 pollen/m³air). The peak occurrence of pollen grain during August was due to cheno–amaranthus which was 7544 pollen/m³ of air.

Trees are the chief contributors during February to April, while weeds and grasses are the major pollen producers during August to November.

Pollen of some families like Poaceae, Chenopodiaceae, and Amaranthaceae were represented almost throughout the year.

The grass pollen tops the list of the total Pollen flora.

The city of Jaipur is surrounded by the Araveli ranges, which is richly populated by the trees. Therefore the pollens of trees were trapped in high densities, during their flowering period.

There was hardly any period, when atmosphere was free from pollen grains and fungal spores. However, heavy rains reduced the air-borne population. Pollen of Grasses, Amaranthus-Chenopods, occurs in the air almost
throughout the year. The April and May period showed the abundant occurrence of pollen of Azadirachta indica, Cassia spp. and Dalberia sissoo.

In the month of June frequent dust storms caused accumulation of sand on the slides making pollen identification difficult. However, some pollen was identified.

Pollen of Poaceae dominated the study duration. It was observed that several of the pollen grains caught in the air are associated with some characteristic features in shape, size and structure, which are presented to be advantageous for aerial suspension.

Spheroidal shaped pollen grains are the most dominating type. The aperture characters show that 3-zonocolporate pollen is the most dominating type, represented by. Other types Monoporate, Pantoporate, Zonocolpate, Monocolpate. Analysis of the ornamentation types of pollen from air show that psilate is the most
common represented by. The other type of ornamentation is obscure and reticulate.

In the month of March the data shows dominance of pollen grains of *Holoptelea* spp. The average count of the pollens of the study duration in the month of March, was 1233.5 Pollen/m$^3$ of air.

In the month of March of first year of the study period the pollen count for holoptelia was more as compared to the second year and the same was observed for the allergic patient’s number. This shows correlation of the number of pollens in air with the prevalence of allergy in the population.

Analysis of the data showed that during spring of both the years of our study duration, pollen concentration was much higher due to high pollen incidence of a number of tree species eg, *Holoptelea, Morus, Kigelia, Azadiracta* etc.

*Holoptelea* spp. which starts flowering in February has its peak in March. Holoptelea dominates the total pollen count of the March being the highest pollen producer of the
month. Pollen production of certain trees differing from another in a given year may be due to the physiological biorhythm of the particular tree species.

Prevalence of both Holoptelea as well as Parthenium pollen allergy is affected by the presence of the respective pollens in the air.

In our study more males were positive to pollen allergy as compared to females and the 3 peaks of age of positivity on prick test for Holoptelea and Parthenium were (31-35 years, 26-30 years and 11-15 years).

The male number was higher than female this could be probably due to more indoor activities of females as compared to males. As more time is spent indoor by females and this makes them less exposed to the outdoor air and thus also the pollens suspended in the air.

Pollen allergy are quite variable in different ecozones which makes them very important to identify pollinosis causing species from every region and prepare extracts from them for diagnosing and treating the allergy patients.
In our study the following allergens i.e Prosopis juliflora, Ricinus communis, Morus alba, Argemone, Amaranthus, Chenopodium, Holoptelea and Parthenium came out to be as the important allergens from Jaipur.

Pollen calendars are very useful for clinicians as well as allergic patients to establish chronologic correlation between the concentration of pollen in air and seasonal allergic symptoms. Nicely prepared Pollen calendar has to be published that defines out the time when different plant’s pollen peak, causing allergies.

In our study the pollen calendar was compiled for the 18 allergic species present in Jaipur, which has most of the species similar as shown by Centre for Biochemical Technology Institute of Genomics and Integrative Biology (Council for Scientific and Industrial Research) (2012) published book on pollen calendars of 12 different states of India Important pollen season for grasses, weeds and trees prevalent in India are provided in the book.
In conclusion our study aids the pollen allergic cases as well as the clinicians treating such patients to be aware of the season of the specific pollen for which they are allergic. Our study would also allow people planning to visit Jaipur, to plan their visit as per the season of the pollen and avoid the time when the pollen they are allergic to is more in the air.

Even during the course of our study the result came out that the Holoptelea is highly allergic and this would bring up awareness among the jaipurites. People are removing the plant from their areas of vicinity. Even the development authorities of Jaipur came up with a campaign of Holoptelea removal especially from Central park as published in media.

But being botanically important we cannot totally remove this plant so our suggestion to the department of environmental science is that separate areas of Holoptelea plantation should be developed which should be far away from the residential areas.
Regarding Parthenium as it is highly allergic in not only aerobiological manner of Pollens but also by contact of its leaves, stem etc. with skin; as well as it’s of no use at all. Our suggestion to the government authorities is that this weed should be eradicated totally from the environment.