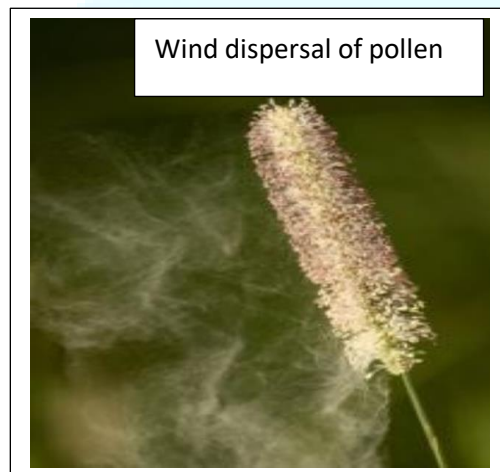


AEROPALYNOLOGY AND POLLEN ALLERGY

- **Erdtman** (1969) defines aeropalynology as the study of palynomorphs found in the atmosphere.
- The term palynomorph encompasses pollen grains, spores and other bioparticles that can not be dissolved in hydrofluoric acid.
- Aeropalynology is a branch of palynology and related to study of pollen and spores that are dispersed in the atmosphere. The study includes their eventual dissemination, deposition and impact on human systems.
- Pollen grains are dispersed more than 400 miles away from the source plants. They can be found more than two miles above the surface.



- The airborne pollen grains originate from anemophilous plants and so they are small, light, smooth walled, colourless, produced in large numbers, dry and lack nectar.
- In other words they are custom made for wind transport.
- The first and foremost requirement in the study of airspora is to identify and detect the airborne palynomorphs and sporomorphs.
- The airborne pollen are trapped for sampling by two major principles:
 - a. Simple gravimetric method:

- where pollen and spores are deposited by normal gravitational force on slides.
- b. Suction method:
- where the atmospheric air is sucked in with the help of instruments -called **samplers**.
-
- There are many types of sampler that can suck certain volume of air according to a known velocity for a definite time.
 - Along with the air pollen and spore are sucked in and they get stuck either on a plastic band or sticky cello tape (fitted according to sampler), which were previously placed there for the purpose of trapping pollen.
 - Mention may be made of **Burkard seven days sampler**, **Burkard personal slide sampler**, **Burkard petri plate sampler**, **Anderson sampler**.
 - The next task is to analyze the trapped grains. Analysis includes identification and quantification of sporomorph and palynomorphs.
 - Microscopic identification is dependable in most cases. The frequency of the identified grains is also calculated.
 - Trapping, identification and quantification are carried out throughout the year and the results are recorded. The data help to prepare the **pollen calendar** of a particular locality.
 - A pollen calendar reveals the name of plant species that release pollen and spore in particular month(s). It also reveals the **approximate amount of grains released in that season**.
 - The term '**pollen count**' is frequently used to represent the concentration of pollen in atmosphere. It is a measure of how much amount of pollen and spore is present in the atmosphere in a particular area at specific time.
 - The count may consist of a particular type of pollen or spore, or all pollen and spore is present in the air. Pollen count is expressed as the number of pollen present in a cubic metre or other standard volume of air over a twenty-four hour period at a particular place. The samplers collect pollen grains.
 - A sampler has a drum or rod coated with silicone grease or sticky cellotape.

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- During trapping the rod/drum is rotated one turn only in an entire 24 hours of a day with the help of a motor fitted with the sampler. The trapped pollen grains are then analyzed for identification and quantification.

Group/Season	Dominant pollen types
Group I (Autumn)	<i>Cassia, Cocos, Eucalyptus, Trema</i> and grasses.
Group II (Winter)	<i>Borassus, Phoenix, Carica, Cassia, Casuarina, Croton, Dillenia, Eucalyptus,</i> Cyperaceae and grasses.
Group III (Spring)	<i>Acacia, Areca, Azadirachta, Bombax, Borassus, Carica, Cassia, Casuarina, Cheno-Amaranths, Cocos, Croton, Madhuca, Mangifera, Phyllanthus, Ricinus</i> and grasses.
Group IV (Summer)	<i>Acacia, Areca, Borassus, Barringtonia, Litchi, Cheno-Amaranthus</i> and grasses.
Group V (Rain)	<i>Cassia, Cheno-Amaranthus,</i> Cyperaceae and grasses.

Dominant pollen

DEFINITION OF ALLERGY

- The term allergy is defined as "an altered or accelerated reaction of a person to a second or subsequent exposures to a substance, usually harmless to the general population, to which he/she has been sensitized during the first exposure" - Shivanna and Agarwal et al.

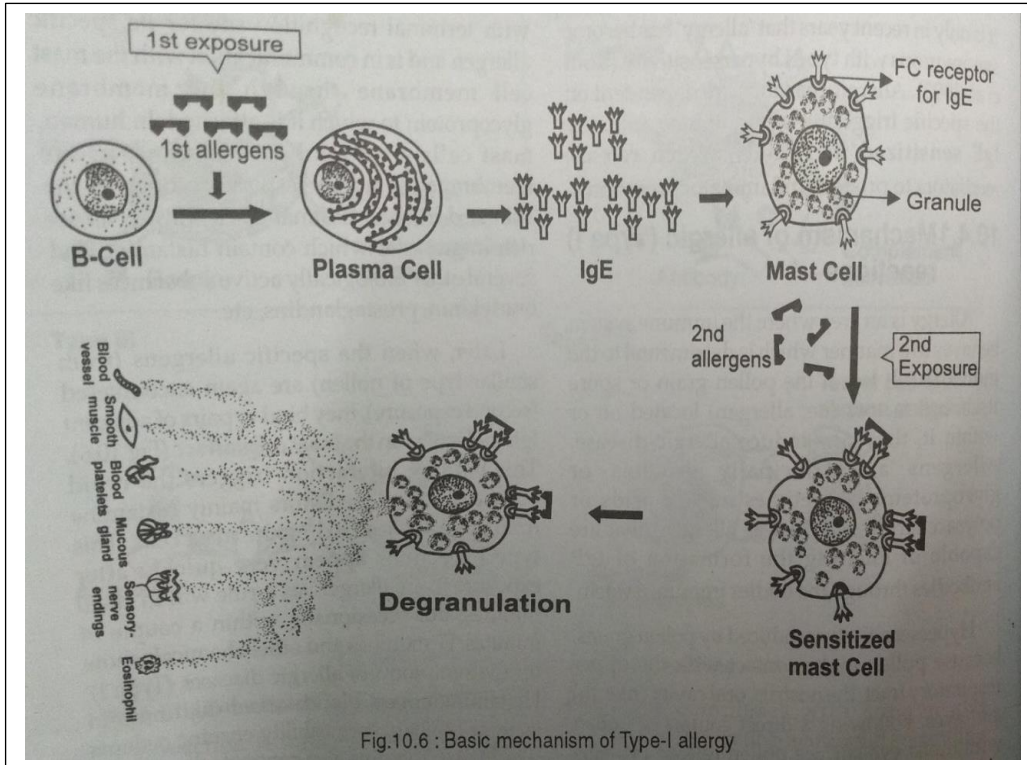


- The sensitizing agents are known as allergens and it is defined as a substance to which a person is allergic (e.g. pollen and spores, food protein, house dust mites, furry animals etc.).
- Sometimes the term allergen is used as a synonym for **antigen or immunogen**. It is protein in nature. A normal adult inhales about 14- 15 cubic meters of air per day, which contains a good number of bioparticles including pollen grains.
- So, the study of airborne allergic pollen is very useful for proper diagnosis and treatment of allergy. From medical, specialty clinical point of view it is important to know the details about the occurrence of the pollen load in the atmosphere.
- The correlation between the onset of different airborne pollen seasons and the occurrence of a patient's symptoms is now well known.
- Pollen grains causing allergy are quite variable in different ecozones and also in a particular place from season to season, year to year depending on changes in ecological and climatic conditions.
- This makes it very important to identify **pollinosis**(The allergic response of body parts to pollen is called "Pollinosis") causing species from every region and to prepare extracts from them for diagnosis and immunotherapy.
- That's why an aerobiological survey is needed to make a pollen calendar of a particular area. **Pollen calendar of an area is essential to test the relevant**

antigens on the patients and to correlate the seasonal occurrence of the pollen types to the patient's allergic symptoms.

- The Aerobiological survey of an area involves aeropalynological study, identification of airborne pollen grains, and determination of atmospheric pollen count.
- Although the atmosphere consists of a large number of pollen grains, only a few of them are responsible for allergic manifestations.
- To know the details about the occurrence and concentration of these allergic pollen which can be inferred from the pollen calendar is very essential for the clinicians.
- A pollen calendar of a region is a prerequisite for the immunological treatment of pollen allergies. Pollen grains are generally found during their respective pollination period.
- Wind pollinated plants produce a large number of smooth-walled pollen grain which are often inconspicuous, colourless and lacking nectar.
- Such pollen grains are mostly found in the air and are potentially more allergic than the insect pollination i.e. entomophilous plants.

Mechanism of type I hypersensitivity;



Mechanism of allergic (Type 1) Reactions

- It is not the pollen grain or spore itself, but factors (i.e. **allergen**) located on or within it, that may induce allergic disease.
- Allergens are principally proteins or glycoproteins (sometimes nucleic acids or polysaccharides may act as allergen) that are capable of eliciting the formation of **IgE antibodies** through the body's immune system.
- Sensitization phase-(Hypersensitivity is induced by pollen grains, because pollen make contact with the upper respiratory tract, the nostrils, oral cavity (mouth) and eyes. Following a direct contact of pollen with moist eye surface pollen release proteins to induce **hay fever or rhinitis**.)
- Pollen grains deposited in the uppermost ciliated portion of the respiratory tract, cannot reach to lungs.

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- The nasal cavity filtering them out by inducing a high degree of turbulence in the airflow that are deposited in the trachea and upper bronchi.)For more information and understanding.
- Most pollen grains are swallowed and become accumulated in stomach.
- Pollen discharge their proteins while passing through the stomach and a moderate proportion of proteins are introduced into the bloodstream to start hypersensitivity reaction.
- Antibodies are produced from different antibody forming cells of lymphoid tissues of allergic individuals in response to first exposure to allergens (proteins) obtained from pollen grains.
- These IgE antibodies circulate in the serum in the blood stream and it becomes attached to the surface of mast cells or basophilic granulocytes by its foot piece (by Fc region of IgE).
- About 100,000 IgE molecules may remain bound to the surface of a single mast cell for a period of several weeks.
- Each IgE molecule has two arms with terminal recognition site for its specific allergen and is in communication with the mast cell membrane through the membrane glycoprotein to which it is attached.
- In human, mast cells are found in the lungs, in the membranes of upper respiratory tract, in the skin and in the intestinal tract.
- Mast cells are rich in granules which contain histamine and several other biologically active substances like bradykinin, prostaglandins, etc.

Some important pollen allergies, diseases and their symptoms

- Common pollen/spore allergies Allergic diseases may involve any part of the body, the most frequently involved being the nose, eye and chest with resultant symptoms of hay fever, rhinitis or asthma.
- The skin and eyes also commonly show allergic symptoms.
- Some of the common pollen allergies are described below;

1) Hay fever:

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- This is a seasonal type of allergy.
- The pollen grains of certain grasses, weeds and trees are the main causes of this type of allergy, although mold spores can also cause the symptoms.
- Depending on where the patients live and the pollination period of a particular plant, attacks may occur seasonally either in spring, summer, winter or rainy season.
- The British Scientist, Dr. Blackley in 1873 was the first to prove that pollen grains are the causative agent for hay fever. Later, Wyman (1876), Dunbar (1903) and others directly proved that the pollen grains of ragweed or Ambrosia are responsible to cause the common hay fever of United States. Hay fever or allergic rhinitis also called pollenosis (also spelt as pollinosis) is the best-known allergy of all.
- Various symptoms may occur: The lining of the nose becomes swollen and exudes a runny discharge or clogged nose. Spells of sneezing and itchiness of the throat and palate also occur and the eyes may be similarly affected.



2) Rhinites:

- It is perennial type of allergy.
- The symptoms are similar to hay fever, but appear all the year round.
- The condition is caused by non-seasonal allergens such as pollen grains of grasses and other plants which flower round the year.

- Sometimes house dust components and certain mold spores are also the causative agents for rhinitis.

3) Conjunctivitis :

- The people are more likely to suffer from an allergic condition of the eyes as an adult.
- Allergic conjunctivitis is often associated with allergic rhinitis.
- A general complaint is of itchiness of the eyes which are rubbed frequently.

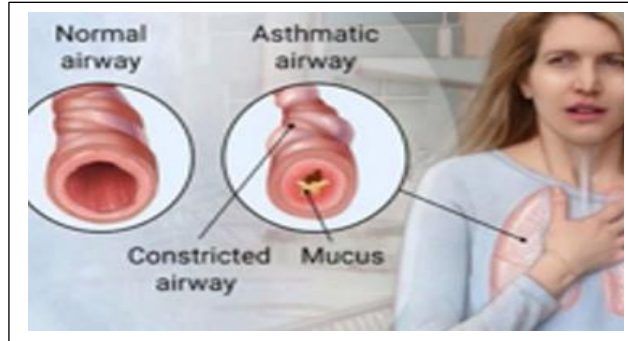


4) Asthma :

- Asthma may be allergic or non allergic in origin.
- In allergic asthma environmental allergens like pollen grains and spores trigger the disease when inhaled.
- The patients may suffer from attacks which obstruct the flow of air to the lungs due to the swollen mucous membrane and formation of mucous in the airway.
- Breathing becomes difficult and forced breathing becomes necessary.
- A wheezing sound appears due to the rush of air through the narrowed airways.

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- At the same time, a troublesome cough can develop. Asthma may begin at any age and trends to become if neglected recur and chronic.



Some important allergic pollen and spores.

Cyanodon dactylon, *Betula verrucosa*, *Cryptomeria japonica*, *Ambrosia artemisifolia*, *Zea mays* etc are some common source of pollen allergens.

Common name	Latin name	Major allergens
1. Birch	<i>Betula verrucosa</i>	Bet v I, II
2. Bermuda grass	<i>Cynodon dactylon</i>	Cyn d I
3. Blue grass	<i>Poa pratensis</i>	Poa p I, V, IX
4. Japanese cedar	<i>Cryptomeria japonica</i>	Cry j I, II
5. Mugwort	<i>Artemisia vulgaris</i>	Art v I – III
6. Orchard grass	<i>Dactylis glomerata</i>	Dac g I, V
7. Ragweed, short	<i>Ambrosia artemisiifolia</i>	Amb a I – VII
8. Ragweed, giant	<i>Ambrosia trifida</i>	Amb t V
9. Western ragweed	<i>Ambrosia psilostachya</i>	Amb p V
10. Rye grass	<i>Lolium perenne</i>	Lol p I – V
11. Timothy grass	<i>Phleum pratense</i>	Phl p I, II, V
12. Maize	<i>Zea mays</i>	Zea m I, Zm PRO 1,2,3
13. Olive tree	<i>Olea europea</i>	Ole e I, Ole e
14. Velvet grass	<i>Holcus latanus</i>	Hol I I, Hol I V
15. A tropical grass	<i>Sorghum halepense</i>	Sor h I
16. Parietaria	<i>Parietaria officinalis</i> <i>Parietaria judaica</i>	Par ol Par j I

- Respiratory allergy is a very common disease among all populations all over the world.
- It is a seasonal and local problem. It is related to the appearance and the concentration of specific airborne allergens in that particular area.
- In India 20 to 30% of the population suffer from allergic rhinitis and 15% suffer from asthma.
- So it is necessary to develop a network of centres in various biozones of India to collect data of the dispersed palynomorphs and sporomorphs. These centres will prepare pollen calendars from the collected data and broadcast the pollen count.
- This will be very useful for clinicians and as well as patients. The centres will also perform the clinical and immunological tests based on local allergens to local patients.

MELISSOPALYNOLOGY / MELLITTOPALYNOLOGY

- Melissopalynology or mellittopalynology is the branch of palynology which deals with the botanical and geographical origin of honey by subjecting honey sediments to microscopic analysis for pollen grains contained therein.
- For over 100 years the literature pertaining to the study of pollen in honey has been termed or spelled in several ways, as mellissopalynology, mellittopalynology.
- The International Commission for Bee Research uses “melissopalynology”, which is therefore the term we adopted.
- While collecting nectar from flowers in the field, pollen grains are occasionally swallowed by bees and carried into the hive. There they are transferred together with the nectar from bee to bee into the storage cells to dry and turn to honey.

HONEY

- Honey has been defined as a splendid sweet substance produced mainly by honey bees from the nectar of flowers.

- It is an easily digestible food stuff containing a range of nutritiously important complementary elements with a high content of a range of saccharides.
- It is water soluble, may granulate between 10° C and 18° C , and is slightly acidic (pH 3.4-6.3). The sugars make honey hygroscopic and viscous.
- Honey is the natural sweet substance produced by the honey bees from the nectar of blossoms or from the secretion of living parts of plants or excretion of plants sucking insects on the parts of plants, which honey bees collect, transform and combine with specific substances of their own, store and leave in the honey comb to ripen and mature.
- Honey bees are social insects, living in colonies of up to about 60,000 individuals. The colony is highly complex and each bee works for the good of the entire hive. The colony centers on its Queen. A fertilized female is capable of laying around a thousand eggs everyday.
- The main source of carbohydrate for bees is honey. Pollen collected from the anthers of flowers provides the essential proteins necessary for the rearing of young bees.
- The chemical composition of the honey is largely influenced by the composition of the pollen. In addition it is also influenced by the weather, soil and other factors. In fact no two honey are identical. Since the composition of pollen changes from species to species variation in absolute amounts of the different compounds can be very high.
- The major components are sugars, protein, amino acids and lipids.
- All amino acids essential to humans (phenylalanine, leucine, valine, isoleucine, arginine, histidine, lysine, metionine, theonine, and tryptophan) can be found in pollen with proline being the most abundant.
- Most of the world production of honey is obtained by the activities of honey bees, viz., *Apis mellifera* (occur mostly in Europe), *Apis cerana* (South East Asia including Indian Sub-continent), *Apis dorsata* and *Apis flora* (Indigenous to South East Asia).
- In India the stingless bees yield meager quantities of honey, but they increase our national wealth by selectively pollinating certain agricultural and forest plants.

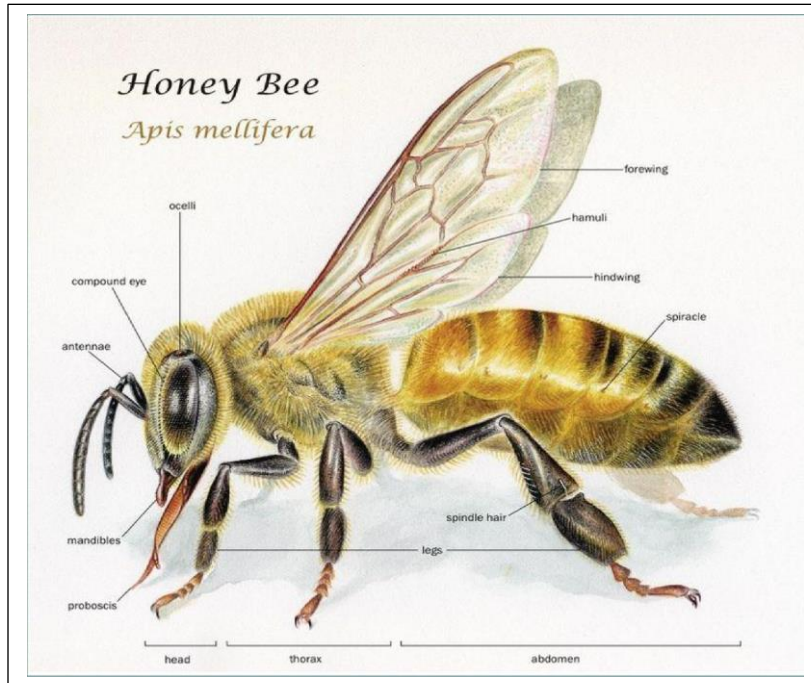
- In India four indigenous bees belong to the highly evolved Apinae, among these *Apis florea* and *Apis dorsata* are primitive members, while *Apis cerana indica* and *A. mellifera* are tetraploid.

HONEY TYPE

- If Honey contains 50% or more pollen of one species, it is called **Uniflorae**.
- If less than 50% then called **Multiflorae**.
- In India Eugenia, Nephalicem, Citrus, Brassica are common nector yielding species, While, Holoptelia, Alnus and Borassus etc are common pollen yielding species.
- The name of honey can be assigned on the basis of pollen Contained in it.

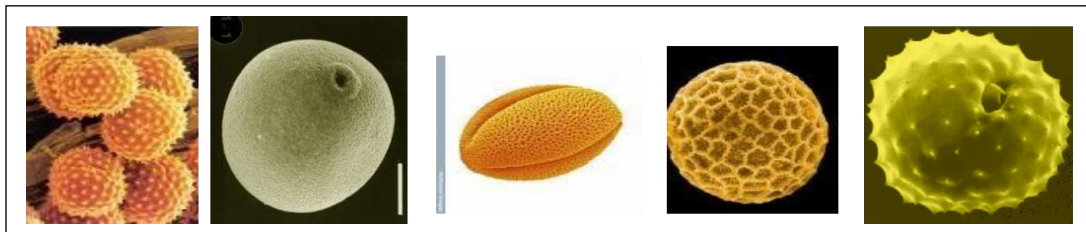
HONEY BEES

- For many insects and specially bees, pollen is the principal source of normal, non-liquid food. Pollen contains excellent nutrients for production of royal jelly, which nourishes the larval queen and young worker larvae.
- The amount of protein & fat in nectar is insignificant.
- Older workers bees use protein directly from pollen, queen images, larval queen and the young larvae of both sexes receive protein
- in the royal jelly produced by nurse bees supplied with pollen. Thus pollen is essential for normal growth and development of individual bees as well as reproduction of colonies.



POLLEN

- It comes in a vast array of shapes and sizes and has complex surface patterns and aperture openings.
- Each plant type produces pollen (or spores) that are quite distinctive from those of other plants.
- Usually, pollen types of species within a single genus look nearly identical.
- Sometimes pollen types of genera within a single plant family will look similar or nearly identical. In some plant families there is a great deal of variation among the pollen genera.



POLLINATION CONSTRAINTS

- Reduction in native pollinator populations due to habitat degradation of pollinators.
- Extensive use of agrochemical like pesticides, insecticides and herbicides with drastic effects on native pollinator population.
- High density of plants in monoculture cropping system limited the availability of native pollinators.
- Crops introduced in regions where natural pollinators are absent.

METHODS OF PACKING AND GATHERING POLLEN:

- Parker (1926) classified them in to following groups:

OPEN FLOWERS:

- The workers bite the anthers with mandibles and pulls them towards its body with its front legs while it runs rapidly over the flowers, all the while packing pollen into the baskets. E.g. *Taraxacum, Rosa, Malus, Ulmus, Acer* etc.



CLOSED FLOWERS :

- The bee alights on the wing of the flower and separates the petals by forcing its fore-legs between them on either sides. The pollen is gathered on the mouth parts and fore legs and packed in the usual way. Eg. *Acacia, Robinia, Trifolium* etc.



TUBULAR FLOWERS :

- The bee alights on the corolla & inserts its proboscis into the tube in search of nectar. Collecting pollen is incidental to nectar gathering. The quantity of pollen obtained is small, it adheres to the mouth parts as forelegs.



SPIKE OR CATKIN FLOWERS:

- The bee may alight at the base of the lower part of the staminate flower, run up the catkin a short distance, then fly away to pack the gathered pollen and return to gather more. It may repeat the process several times.
- In many cases the bee does not actually touch the catkin, but depends itself of Lasting towards the catkin and away from it.



PRESENTATION FLOWERS :

- Flowers of this type present free pollen to the visiting insects. *Apis bomes* & Many solitary bee species press the abdomen against the inflorescence, causing a pollen mass to be pushed out of the disc flowers. Tubular flowers retract activity so that pollen is exposed. The collecting insect clamps to the corolla and the receptacle scale & in this way presses its body into corolla lead. Eg. *Echinops* sp.



TIME OF POLLEN COLLECTION :

- It varies with different species and time of opening of flowers.
- Three categories of opening: 1. Morning 2. Afternoon and 3. Evening.
- Pollen collection may be different in different localities in same species. This is due to weather and climatic conditions.
- The daily rhythm of pollen collection by bees is reasonably well Related to pollen availability of individual species.

COLOR OF POLLEN LOAD

Uniform :

- It is of same color when pollen belong to same species.

Mixed :

- may be mixed in case of different species.

Segregated :

- Pollen is packed in different colors resulting in striped loads. The plant source can often be identified from the color of pollen loads.

STORAGE OF POLLEN IN HIVE

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- Adult bees store pollen primarily to consume in their own diets and to feed the larvae.
- On returning to the hive the worker bee seeks out an empty or partially filled cell.
- The hind legs are dangled into the selected cell and loads are stripped off by the middle legs.
- The pollen gathering workers have now finished its duty. Another bee generally a young one comes to break the load and stamps them down firmly into the bottom of the cell.
- A small amount of honey is deposited on the pollen to prevent spoiling. This store of pollen is called “Bee Bread” by the bee keepers.
- It requires about 18 loads to fill one cell. Because different pollen loads are mixed in storing the pollen, the filling of the cells is stratified.

POLLEN PELLETS

- The hundreds or sometimes millions of pollen grains collected by honey bees is usually mixed with nectar or regurgitated honey and are packed with the help of special combs and hairs into structures called pollen pellets which stick on to their hind legs.



- Size & weight of pollen basket loads collected by bees varies widely. The average weight of a load is about 7.5 mg which may be maximum upto 15 mg.
- A very large amount of pollen can be collected by a colony in one day. In July about 250 gm of pollen requiring some 17000 flights were collected between

8 and 10 a.m. The total amount of pollen collected by one hive in a season varies between 15 and 40 kg.

- Eckert (1942) calculated the pollen demand of one colony to be about 50 kg a year.

POLLEN SPECTRUM

- The pollen analysis of honey provides qualitative information about the forage plants at the collecting sites and depends on the soil type and seasonal flowering pattern. Adjacent colonies in a bee hive do not always produce honey with an identical spectrum.
 - One of the goals of melissopalynology is to determine the floral sources and the geographical origin utilized by honey bees in the production of honey.
 - Honey and its related products are rich in protein, in which these become partial source of vitamins, minerals and amino acids.
 - These will aid in the construction of a pollen spectrum of a region, thus enabling to understand the source and establishing the authenticity of honey.