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PALYNOLOGICAL STUDIES OF POLLEN GRAINS OF SOME BEE FLORA OF UTTARAKHAND, INDIA

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ABSTRACT

Uttarakhand has a great diversity of flowering plants for beekeeping and has a very good potential for commercial beekeeping in the state. The present study was conducted at Govind Ballabh Pant University of Agriculture and Technology, Pantnagar-263145, District Udham Singh Nagar (Uttarakhand) India. Fruit trees provided both pollen and nectar for the honeybees and significant increases in yields are recorded in cross pollinated fruit crops like litchi, apple, peach, pear, plum, etc. due to bee pollination. The honey bees (*Apis* species) have major role and considered widely as pollinating agents. Most of the crops depend on them for pollination services. However, there are many other insects especially Non *Apis* bees also called as native bees or pollen bees, which play an important role in pollination. These efficient pollinators often have a large share in pollinating the crops. Photomicrographs have been taken in different magnification (like X500, X650, X2300, X2000 etc.) on Scanning Electron Microscope (JEOL), films are magnified. Each species is normally illustrated in both polar and equatorial views depicting general and other diagnostic morphological features at different foci.

Key words : Bee-flora, Pollen morphology.

Introduction

The indiscriminate use of pesticides causing the major problems to our environment and also reducing the natural enemies from the ecosystems. Environmental stresses, population explosion and food shortage have caused serious problems to mankind on the globe. Instead of making substantial use of chemical fertilizers, biocides, irrigation, facilitates and heavy machinery for yield enhancement, a shift towards biologically based agriculture becomes necessary to increase food productivity. The apiculture industry plays a big role in generating employment and in increasing family income in the rural areas of the world through different hive products. A beekeeper (or apiarist) keeps bees in order to collect their honey and other hive products (including beeswax, propolis, pollen, royal jelly, bee venom etc.), to pollinate the crops, or to produce bees for sale to other. Beekeeping is a significant sustainable and environmental sound activity involving integration of forestry, social forestry and Agricultural supporting activity since it provides nutritional, economic and ecological balance, while providing employment and income. India has a good potential for beekeeping and to become a major honey exporting nation. Honeybees (Apidae: Apinae) are classified into the genus *Apis* which includes four main species: the common honeybee or Italian honeybee (*Apis mellifera* L.), the giant honeybee (*Apis dorsata* F.), the Asian honeybee (*Apis cerana* F.) and the little honeybee (*Apis florea* F.). There are more than 20,000 species of wild bees. Many species are solitary (*e.g.* mason bees, leafcutter bees (Megachilidae), carpenter bees and other ground-nesting bees). While others rear their young in burrows and small colonies (*e.g.*, bumblebees and stingless bees).

Honeybees pollinate 16% of flowering plant species in the world and nearly 400 species of agricultural plants (Crane and Walker, 1984). Fruits, vegetables or seed production from 87 of the 115 leading global food crops depends upon animal pollination (Klein *et al*., 2007). The value of insect pollination for worldwide agricultural production is estimated to be 153 billion, which represents 9.5% of the value of the world agricultural production used for human food in 2005 (Gallai *et al*., 2009). By investing limited expenses, beekeeping can be practiced to obtain maximum subsidiary income through honey, beeswax and other bee products with agricultural activity. The practice of beekeeping is not only depends on the better strain of honeybees but also on abundance and occurrence of pollen and nectar sources within the surrounding area of an apiary (Free, 1970 and Akrathanakal, 1987).

The pollen grainsare marvelous product evolved by flowering plants to continue their generation. Pollen grains developed and are borne on a flower's stamens, from where they are carried away by external agents, such as wind or an animal (usually an insect or a bird). Honeybees and flowering plants have been considered as an example for co-evolution and mutualism. Bees pollinate flowers, which mean they transfer the pollen from the flower of one plant to the flower of another plant. Bees do not purposely do this. Actually, the bees are trying to collect the pollen to take back to their hives. In the process of going from one flower to another as they collect pollen, some pollen is picked up from one flower and accidentally dropped on another flower. Pollination is the transfer of pollen from male to female reproductive structures of plants. In another words, the process involving transfer of pollen from anthers to the stigma is called as pollination and the agent causing this transfer is called pollinator. Pollen is the fine powder like material consisting of pollen grains that is produced by the anthers of seed plants. When pollen from one flower is carried to the stigma of another, this is called cross-pollination. The sustainable development of agriculture has necessitated the reorientation of the present crop production technologies (Free, 1993; Abrol, 1993; Tikoo and Abrol, 1994). Pollination of flowers requires insect like syrphid flies, honey bees, ants and wasps (Das and Chodhury, 1968).

Fig. 1 : Map of study area.

Honeybee species *Apis dorsata* F; *A. mellifera* L; *A. cerana* F. and *A. florea* F. were the most important and efficient pollinators of litchi flowers (*Litchi chinensis* Sonn.). They constituted more than 65% of the total pollinating insects. Between commencement and cessation, the foraging activity of all honeybee species followed the same general pattern as temperature, light intensity, solar radiation, and nectar sugar concentration and inversely with relative humidity. Path analysis revealed that all honeybee species differed in their responses to temperature, light intensity and solar radiation, the three most important factors in foraging behavior (Abrol, 2006). Several horticulture crops cultivated in India derive benefit or are dependent on pollinating insects for effective qualitative and quantitative improvement in crops yield. Many temperate fruit trees have been investigated for their dependence on bees. A number of varieties of apples, pears, plums, peaches and cherries are known to be self–sterile and re-benefited by bee pollination. Orchard growers in Himachal Pradesh initiated the practice of renting bee colonies for keeping in their orchards during the flowering of these crops for enhancing the fruit production (Deodikar and Suryanarayana, 1977).

Study sites

Geographically Pantnagar is located in the sub-tropical zone at 29^0N latitude and 79.3^0E longitude and at an altitude of 243.8 m above the mean sea level in the "tarai" region of Uttarakhand in Northern India. The location has sub-humid tropical climate and is situated in the foot hills of "Shivalik" range of the Himalayas. The meteorological data indicate that the humid climate here is characterized by hot dry summer and cold winter. The temperature rises up to 40^0C in summer, while it falls to 2-10^0C in winter. Approximately, 1400 mm mean rainfall has been recorded and relative humidity fluctuates around 90 ± 5 per cent during rainy season.

Methodology for Palynological study

The study was carried out as per the method developed by Erdtman (1952). At first the pollen masses or anthers separated from the plant material and the fixation of polliniferous material in acetone stored in small vials for at least 12 or more hours before subjecting the further process done. The preserved specimen with acetone were transferred to polythene centrifuge tube and lightly crushed with needle. The dispersed material was removed. The material concentrated through centrifuging and decant off alcohol. The residue was washed with glacial acetic acid and further treated with freshly prepared "acetolysis mixture" (a mixture of 9 parts acetic anhydride and 1 part of concentrated sulphuric acid) in to the tube. Then tube placed with acetolysis mixture in waterbath and boil water for 3-5 minutes, till the time the acetolysis mixture attains a medium brown colour. Thereafter, the mixture decanted by centrifuging leaving a brownish residue presumed to contain the dispersed pollen and other organic debris. The obtained residue was washed with distilled water several times through centrifuging to achieve the clear and neutral preparations. Further pollen grains were picked up with the help of needle and put on small stab. These stabs kept inside the Gold coater for gold coating to the conductivity of pollen grains. Finally, these pollen grains along with stab kept inside the Scanning Electron Microscope for microscopic examination. The measurements of size, exine thickness, sexinous excrescences, such as spines, verrucae etc. have been recorded. Pollen morphological descriptions for the study: Pollen apertural class; shape, size range and both polar (P) and equatorial (E) diameters; aperture (colpus, pore) characters; exine (stratification and ornamentation) characters and any other feature of diagnostic value. Photomicrographs have been taken at different magnifications (like X500, X650, X2300, X2000 etc.) on Scanning Electron Microscope (JEOL), films will be magnified. Each species will be normally illustrated in both polar and equatorial views depicting general and other diagnostic morphological features at different foci.

Results and Discussion

The purpose of this study is to provide the reference information on nectar and pollen source for honeybees and to evaluate the morphological difference in pollen grains of selected bee flora of fruit plants. Chaubal and Kothmire (1980) studied the bee forage plants and presented them in a floral calendar forms based on microscopical and botanical studied in Kolhapur (India). They reported that about 15 important pollen sources sustain at least two hundred colonies during monsoon and about 12 important nectar source plants. Kovacheu (1973) reported that shape of the gladiolus pollens were more or less subprolate with polar to equtorial (P/E) ratio of 1.2739. Rachele (1974) studied the pollen morphology of the papaveraceae in north USA and Canada and found that pollen grains were more or less spherical in shape with spiny exine surface. The results obtained for morphological characteristics of various pollen grains of some important bee flora and other relevant detailed are described below. The present work comprises the detailed pollen morphological study of various bee flora.

Table 1 : Morphological descriptions of various pollen grains.

S. no.	Common name	Scientific name	Polar (P) and Equatorial (E) length	Shape/size of pollen	Colour of pollen	Surface Exine	Flowering Period	Forage Source
1.	Litchi	*Litchi chinensis*	P=18.911 µm E=17.143 µm	(1.103µm) at X4500	White	Thick	March-April	Tree
2.	Ber	*Ziziphu smauritiana*	P=25.469µm E=17.329 µm	(P/E= 1.469 µm) at X3500	White	Thicker and cellular	September to October	Tree
3.	Pomegranate	*Punica granatum*	P=21.384 µm E=17.300 µm	(P/E=1.236 µm) at X4000	Light yellow	Thick	Feb-April	Tree
4.	Pear	*Pyrus amygdaliformis*	P=39.804µm E=19.936µm	(P/E=1.996 µm) at X2500	Red yellow	Thread like structure	February and March	Tree
5.	Peach	*Prunus persica*	P=43.533µm E=39.881µm	(P/E=1.091 µm) at X1200	Reddish yellow	Thick	February and March	Tree
6.	Apricot	*Prunus armeniaca*	P=59.958µm E=26.667µm	(P/E=2.248 µm) at X1600	Reddish yellow	Smooth	February and March	Tree
7.	Plum	*Prunus cerasifera*	P=41.055µm E=15.945µm	(P/E=2.574 µm) at X2700	light grey	Smooth	February and March	Tree

***Litchi chinensis*, Common name-Litchi, Family-Sapindaceae (Plate 1)**

The blossom period is March-April and a good source of pollen and nectar for honeybees. Pollen grains are white in colour. Pollen3-zonocolporate (synzonocolporate), amb triangular, prolate spheroidal to oblate spheroidal. Pollen grain is more or less spherical in shape. Pollen 3-zonocolporate, oblate spheroidal, 18.911 µm X 17.143 µm (1.103µm) at X4500; ends acute, sexine thicker than nexine.

***Ziziphus mauritiana*, Common name- Ber, Family-Rhamnaceae (Plate 2)**

The flowering season is from September to October. The pollen is white in colour. The exine surface is cellular and thicker. Pollen 3- zonocolporate, amb triangular, oblate spheroidal, 25.469µm X 17.329 µm, (P/E= 1.469 µm) at X3500, colpi long and ora circular.

***Punica granatum*, Common name-Pomegranate, Family-Punicaceae (Plate 3)**

The flowering duration is Feb-April. It is a good source of pollen and minor source of nectar. The pollen grain is light yellow in colour.Pollen3-zonocolporate, subprolate, 21.384 µmX17.300 µm (P/E=1.236) at X4000, ends rounded, sexine thicker than nexine, finely reticulate.

***Pyrus amygdaliformis*, Common name- Pear, Family-Rosaceae (Plate 4)**

The flowering period is Feb-March and it is a good source of pollen and nectar. The pollen is red yellow in colour. Exine surface with long thread like structures. Pollen3-zonocolporate, subprolate, 39.804µm X 19.936 µm (P/E = 1.996 µm) at X2500. Colpi long, almost reaching the poles, tectate, collumella fine, finely reticulate.

***Prunus persica*, Common name-Peach, Family-Rosaceae (Plate 5)**

The flowering period is February and March. It is a major source of pollen. The pollen is reddish yellow in colour. The shape of pollen is triangular,subprolate or ovule. Pollen3-zonocolporate, subprolate to prolate, 43.533µm X 39.881µm (P/E = 1.091 µm) at X1200. Exine surface is thick. Sexine thicker than nexine, angulaperlurate, oblate spheroidal, colpi long, apocolpium, ora circular, granulate.

***Prunus armeniaca*, Common name-Apricot,Family- Rosaceae (Plate 6)**

The flowering period is February and March. It is a major source of pollen and nectar. The pollen is

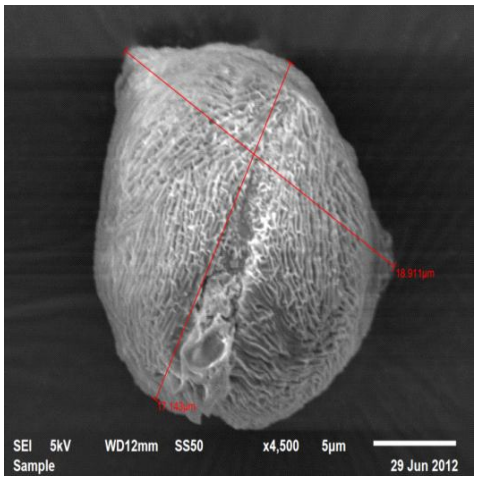

Plate 1 : *Litchi chinensis*.

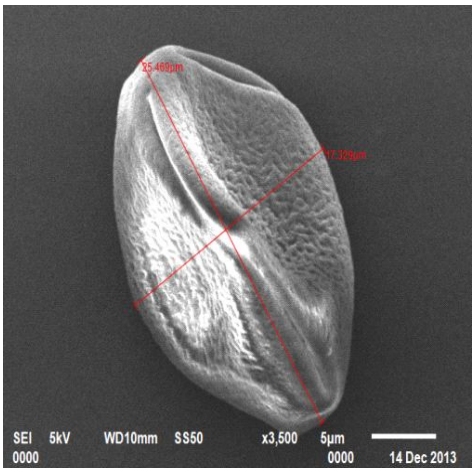

Plate 2 : *Ziziphus mauritiana.*

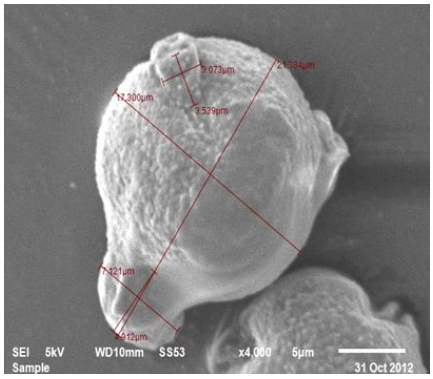

Plate 3 : *Punica granatum.*

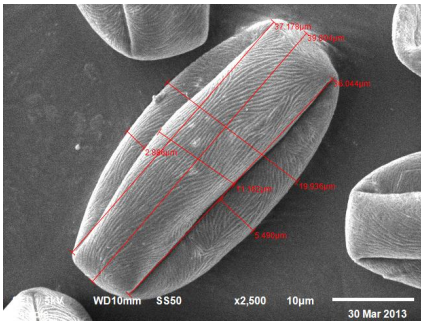

Plate 4 : *Pyrus amygdaliformis*.

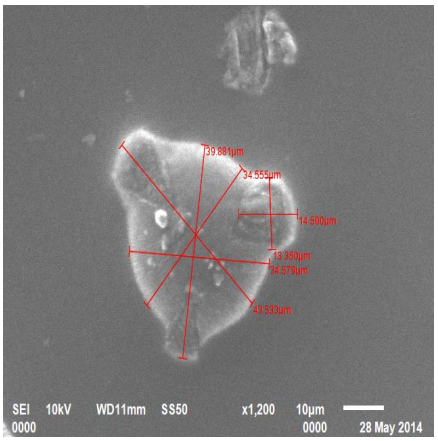

Plate 5 : *Prunus persica*.

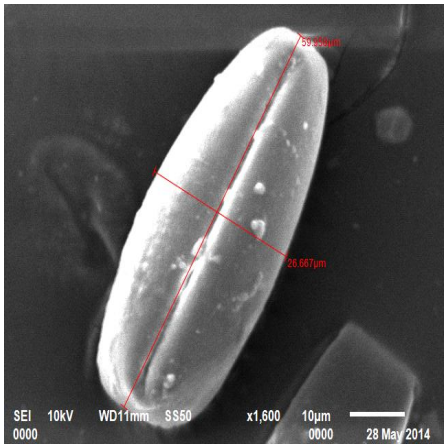

Plate 6 : *Prunus armeniaca.*

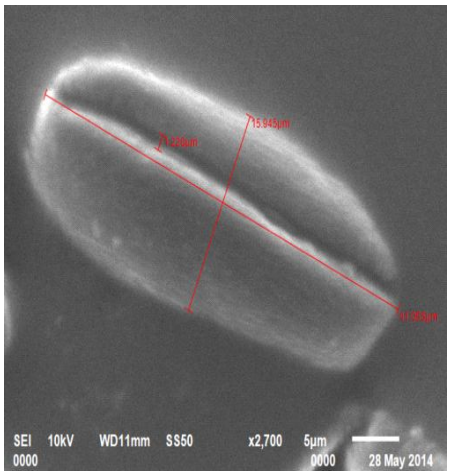

Plate 7 : *Prunus cerasifera*.

reddish yellow in colour and cylindrical in shape. Pollen3-zonocolporate, subprolate, 59.958µm X 26.667µm (P/E = 2.248 µm) at X1600. Colpi long, almost reaching the poles, tectate, collumella fine, finely reticulate.

***Prunus cerasifera*, Common name-Plum, Family-Rosaceae (Plate 7)**

The flowering period is February and March. It is a major source of pollen. The pollen is light grey in colour. Pollen 3-zonocolporate, subprolate, 41.055µm X 15.945µm

(P/E = 2.574 µm) at X2700.Colpi long, almost reaching the poles, tectate, collumella fine, finely reticulate.

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