Palynological Studies of pollen grains of Some Apiculture Importance Bee Flora in Tarai Region of Uttarakhand

Mahendra Singh and Pramod Mall

Abstract
Tarai region of Uttarakhand has a great diversity of flowering plants and has good potential for commercial beekeeping. The area fall under foothills of Himalayas in Uttarakhand state. This region was selected for maximum utilization of flora by beekeepers and recognizes the major pollen and nectar source to honeybees. The present study was conducted at Govind Ballabh Pant University of Agriculture and Technology, Pantnagar-263145, District Udham Singh Nagar (Uttarakhand) India, during the period 2011-2013. 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.

Keywords: Pollen, Bee-flora, Pollen morphology

Introduction
Beekeeping is the maintenance of honey bee colonies, commonly in hives, by humans. The apiculture industry plays an important 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 etc.), to pollinate the crops, or to produce bees for sale to other. 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 E, Walker P. (1984) [4]. Fruits, vegetables or seed production from 87 of the 115 leading global food crops depends upon animal pollination Klein AM, Vaissière BE, Cane JH. (2007) [9]. 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 N, Salles JM, Settele J, Vaissière BE. (2009) [8]. 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 JB. (1970) [6] and Akrathanakal P. (1987) [2].

A pollen grain is a 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).

Study sites
Geographically Pantnagar is located in the subtropical zone at 29°N latitude and 79.3°E 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°C in summer, while it falls to 2-10°C 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 investigations were 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 atleast 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 information about bee forage plants, pollen morphology and types of pollen in tarai region is limited. 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. Chauhal and Kothmire (1980) [3] 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. Waliur-R and Chaudhry (1985) [13] recorded ninety species of forest and fruit trees, ornamentals and agricultural crops providing sources of nectar and pollen for bees at the Pakistan Forest Institute, Peshawar.

Morphological descriptions of various pollen grains

<table>
<thead>
<tr>
<th>Sr. No.</th>
<th>Common name</th>
<th>Scientific name</th>
<th>Polar (P) and Equatorial (E) length</th>
<th>Shape/size of pollen</th>
<th>Colour of pollen</th>
<th>Surface Exine</th>
<th>Flowering Period</th>
<th>Forage Source</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>Coreopsis</td>
<td>Coreopsis tinctoria</td>
<td>P=30.060µm E=27.635µm (P/E=1.087µm) at x2000</td>
<td>white</td>
<td>Spiny</td>
<td>April-June</td>
<td>Herb</td>
<td></td>
</tr>
<tr>
<td>2.</td>
<td>China Aster</td>
<td>Callistephus chinensis</td>
<td>P=27.208µm E=24.167µm (P/E=1.125µm) at X1200</td>
<td>yellowish</td>
<td>Spiny</td>
<td>Feb-April</td>
<td>Herb</td>
<td></td>
</tr>
<tr>
<td>3.</td>
<td>Sunflower</td>
<td>Helianthus annuus</td>
<td>P=32.675µm E=32.326µm (P/E=1.010µm) at X2300</td>
<td>yellow</td>
<td>Spiny</td>
<td>May-June, July-August</td>
<td>Oilseed crop</td>
<td></td>
</tr>
<tr>
<td>5.</td>
<td>Gazania</td>
<td>Gazania rigens</td>
<td>P=37.706 µm E=36.101 µm (P/E=1.044 µm) at X2000</td>
<td>Yellow</td>
<td>Shows different hexagonal structure</td>
<td>over a long period of summer</td>
<td>Herb</td>
<td></td>
</tr>
<tr>
<td>6.</td>
<td>Goldenrod</td>
<td>Solidago nemoralis</td>
<td>P=20.640 µm E=19.959 µm (P/E=1.034 µm) at X4000</td>
<td>yellow</td>
<td>Spiny</td>
<td>October</td>
<td>Shrub</td>
<td></td>
</tr>
<tr>
<td>7.</td>
<td>Californian Poppy</td>
<td>Eschscholzia californica</td>
<td>P=30.399 µm E=27.186 µm (P/E=1.118 µm) at X2500</td>
<td>yellowish</td>
<td>Exine surface with small spines</td>
<td>Feb-April</td>
<td>Herb</td>
<td></td>
</tr>
<tr>
<td>8.</td>
<td>Poppy</td>
<td>Papaver somniferum</td>
<td>P=34.723 µm E=17.443 µm (P/E=1.990 µm) at X1800</td>
<td>dark brown</td>
<td>Exine surface with small spines</td>
<td>Feb-April</td>
<td>Herb</td>
<td></td>
</tr>
</tbody>
</table>

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 ornamental flowers belonged to family Asteraceae.
1. **Coreopsis tinctoria**, Common name- Coreopsis, Family- Compositae, (Plate-1)
   The blossom period is April- June and it is good source of pollen. Pollen grain is white in colour. The shape of pollen is pantoporate, 30.060µm x 27.635µm (P/E =1.087µm) at x2000, diameter of pollen grain is 15.03µm, spheroidal, without pores, sexine spinate 4.177µm, with broad base and tapering in to acute ends. Mesfin *et al.* (1995) also thoroughly
studied the pollen morphology of North American coreopsis and found that the pollen grains were spherical and suboblate in shape.

2. *Callistephus chinensis*, Common name- China Aster, Family- Compositae, (Plate-2)
The flowering period is Feb-April. It is a minor source of pollen and nectar. The shape of pollen is pantoporate. 27.208µm x 24.167µm (P/E=1.125 µm) at X1200. The pollen is yellow in colour. The diameter is 13.604 µm; exine surface with dense spines, spinate sexine, spine length is 3.064 µm, with broad base and tapering in to acute ends. The distance between acute ends of spine is 6.119 µm.

3. *Helianthus annus*, Common name- Sunflower, Family- Compositae, (Plate-3)
This is an annual crop and duration of flowering is May – August. It is a major source of pollen and nectar. Pollen grain is yellow in colour. The shape of pollen is pantoporate. 32.675µm x 32.326µm (P/E = 1.010µm) at x2300, diameter is 16.163µm; exine surface with dense spines, sexine spinate, spine length is 4.470 µm, with broad base and tapering in to acute ends. The findings are closely similar to Toderich (1992), who studied the pollen morphology of some species of genus Helianthus and reported the shape of pollen is prolate spherical and exine surface with a number of spines.

Blossom period is Feb-March and it is a minor source of pollen for honeybees. The pollen grain is light yellow in colour. The shape of pollen grain is pantoporate. 39.898µm x 39.641 µm (P/E=1.006 µm) at X2000. The pollen is yellow in colour. The shape of pollen is pantoporate, 39.898µm x 39.641 µm (P/E=1.006 µm) at X2000. The diameter is 19.949µm; exine surface with dense spines, spinate sexine, spine length is 5.349 µm, with broad base and tapering in to acute ends. The distance between two broad base of spine and two acute ends is 5.289 and 5.818 µm respectively.

5. *Gazania rigens*, Common name- Gazania, Family- Asteraceae, (Plate-6)
The flowering period is over a long period of flowering. It is a major source of pollen and nectar. The pollen is yellow in colour. Pollen is spherical in shape and shows different hexagonal structure. Pollen 3-zonocolporate, oblate spheroidal to prolate spheroidal, 37.706 µm X 36.101 µm (P/E= 1.044 µm) at X2000.

The flowering period is October and is a good source of pollen and nectar. The colour of pollen grain is yellow. The shape of pollen grain is pantoporate, 20.640 µm x 19.959 µm (P/E=1.034 µm) at X4000. The diameter is 10.32; exine surface with dense spines, spinate sexine, spine length is 2.915µm, with broad base and tapering in to acute ends.

7. *Eschscholzia californica*, Common name- Californian Poppy, Family- Papaveraceae, (Plate-7)
The blossom period is Feb-April. It is a good source of pollen and nectar. The pollen grain is yellowish in colour. Pollen is round in shape. Exine surface with small spines. The size of pollen is 30.399 µm x 27.186 µm (P/E=1.118 µm) at X2500. The diameter is 15.19 µm. These findings are in close agreement with Rachele (1974), who 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.

8. *Papaver somniferum*, Common name- Poppy, Family- Papaveraceae, (Plate-8)
The blossom period is Feb-April and it is a good source of pollen and nectar. Pollen grains are dark brown in colour. Exine surface is with small spines. Pollen is prolate, spherical 34.723 µm X 17.443 µm (P/E=1.990 µm) at X1800. The diameter is 17.3615 µm.

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References