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Analysis of pollen grains in different honey samples from the region of Newasa tehsil in Maharashtra

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Abstract

Pollen analysis is also known as *Melissopalynology* is valuable tool for the identification of the botanical and geographical origin of honey sample. These pollen analytical studies provide information of resources of bee. The present works refers to the pollen analysis of 5 honey samples. The honey sample were collected from different sites of Newasa tehsil, Ahmednagar district. The morphology of pollen contributing as nectar resources were observed using Scanning Electron Microscope (SCM) and the total pollen count were expressed in pollen percentage frequency (PPF) based on the qualitative analysis. The present investigation on the basis of pollen percentage shown multifloral pollen belonging to the family Fabaceae, Asteraceae, Leguminosae, Amranthaceae, Convolvulace, Moringaceae, Cactaceae, Solanaceae.

Keywords: pollen; honey samples; pollen grains; bee; Newasa tehsil

Introduction

Honey is the one of the oldest food of mankind and there have been references to it and the bees that gather it throughout record history. Today honey is regarded as an important food and carbohydrate throughout the world. Honey bee utilizes certain natural raw materials that are identifiable honey. The raw materials are pollen and nectar from flowers of various plants and are the major source of proteins for bees. While nectar is a source of carbohydrates for bees. Identification of pollen found in honey give clear idea honey bees visit to particular plants to the nectar and regarding content into honey. Honey not only provides the nutrient necessary for colony maintenance but also for its development. It is also major source for the colony. 16% of flowers are pollinate by honey bee ^[1]. Pollen is a source of protein and amino acid for the colonies ^[2]. Beekeeping plays a significant role in conserving the natural resources and contributes to the globe through environmental protection like all stocks species, bees require feeds for their production and reproduction. The analysis of honey can help to determine changes in nectar and pollen sources and may help determine the causes of this decline ^[3]. India being a country of varied geography climatic condition at a time with different parts in full of a flora ideal for apiculture. It is essential to study relationship between regional flora and honey bees in order to obtain maximum production of a good quality of honey. However, not much information is available related to pollen analysis in these areas. Thus the present work was carried out to determine the critical analysis of different honey samples and to identify the pollen types in honey samples of Newasa tehsil, District, Ahmednagar, Maharashtra.

Materials and Methods

Study region and period

Pollen analysis of honey included a critical microscopic field study. This involves qualitative and quantitative analysis of the pollen content of the honey and pollen lods from diverse florastic and geographical region. The sample were collected from different villages of Newasa tehsil viz. Bhanashiwara, Handinimgaon, Newasa, Newasa fata, Bhenda etc. Honey sample were collected from period of October 2016 to January 2017.

Sample Preparation

The collected honey sample was stored in plastic bottles, and kept 20-25 °C. The analysis of honey sample was done in accordance with known Louveaux *et al.*, 1987 ^[4] and Suryanarayana *et al.*, 1981 ^[5] work procedure. The qualitative analysis was made to known the specific nature of the pollen grain.

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The percentage representation of different pollen type was determined. The extraction of pollen from honey was achieved by heating honey at 60 °C in water bath, and stirring properly to insure uniform mixing of the pollen. Pollen preparation was made by dissolving 1 gm of honey in 2 ml of water followed by centrifuge as 5000 rpm for 10 min and again washing with distilled water and the centrifuge and decanted for complete removal of sugar [4]. At last sediment was centrifuged in ethyl alcohol to get maximum number of pollen grains from honey [6]. The sediment was divided into two parts, the half part was residue and the supernatant was removed and these sample sediment was dispersed in 0.5 ml ethyl alcohol. The five slides were prepared by simple staining method. The pollen grain sample was dissolved in 1 drop of water and mounted on slide. The prepared slide was used to observe through the light microscope as well as scanning electron microscope (SCM).

Quantification of pollen

The following term have been used in estimating the pollen grain frequency. Predominant pollen (more than 45%) of pollen counted, secondary pollen (16-45%), important minor pollen (3-15%), minor pollen (less than 3 %) [4]. Pollen percentage for each pollen types was calculated and honey was classified as either unifloral or multifloral.

Observations and Results

In present study, five honey samples were collected from Newasa tehsil, district Ahmednagar. The first honey sample (sample-1) collected from Bhanashiware. In sample first, out of two pollen types the predominant pollen type was *Alternanthera sessilis*, (c.f. Figure 1) and the percentage of these pollen type was 55.14%. The secondary important type observed was *Alternanthera* and *Parthenium*, percentage of these pollen type was 21.49% and 23.36%. The absolute pollen count 107 was observed (Table 1).

Table 1: Analysis details of pollen samples from Newasa Tehsil district Ahmednagar.

S. No.	Sample No.	Absolute pollen count (APC)	Pollen Type	
			Predominant Pollen (> 45%)	Secondary Pollen 16-45%
1.	Bhanashiware	107	<i>Alternanthera sessilis</i> (55.15%)	<i>Alternanthera</i> (21.49%) <i>Parthenium</i> (23.36%)
2.	Handinimgaon	109	---	<i>Cassia bicapsularis</i> (29.35%) <i>Cassia tora</i> (15.59%) <i>Parthenium</i> (30.28%) <i>Pongamia pinnata</i> (24.78%)
3.	Newasa	79	---	<i>Cassia tora</i> (41.77%) <i>Celosia argentea</i> (34.18%) <i>Ipomoea hederifolia</i> (24.05%)
4.	Newasa fata	82	<i>Moringa oleifera</i> (45.13%) <i>Opuntia cochenillifera</i> (54.87%)	---
5.	Ranjangaon	129	---	<i>Pongamia pinnata</i> (25.58%) <i>Parthenium hysterophrous</i> (44.18%) <i>Solanum melongena</i> (30.24%)

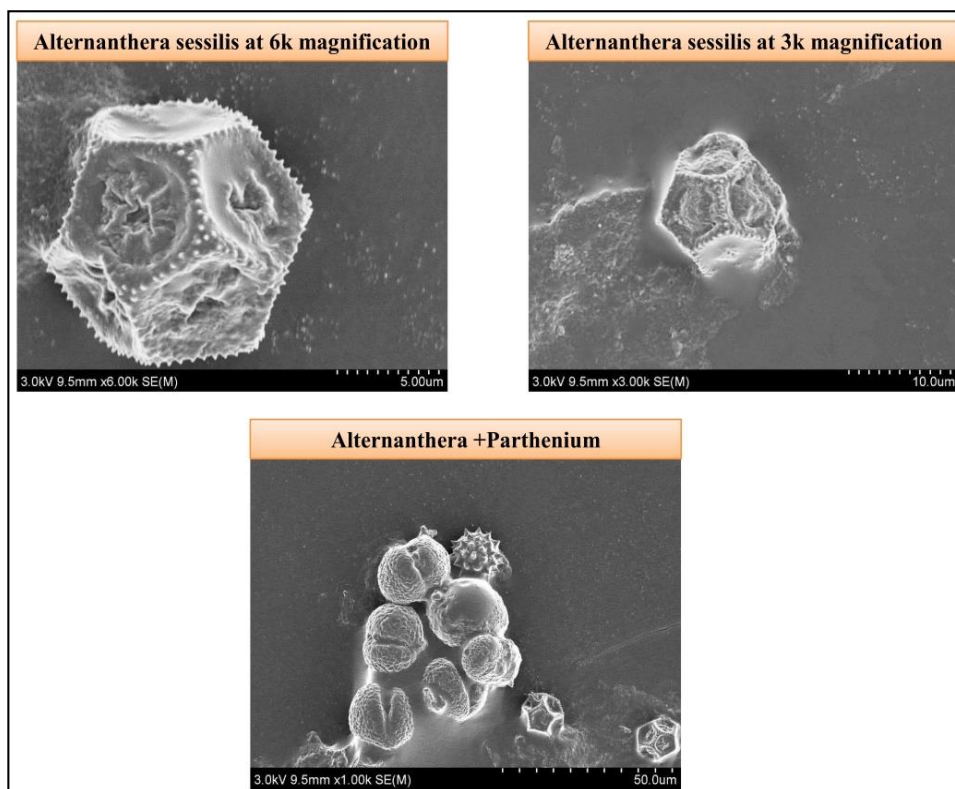


Fig 1: Images of pollen types observed in sample-1 collected from Bhanashiware village

Second honey sample (sample-2) was from Handinimgaon and four plant species were observed in it such as *Cassia bicapsularis*, *Cassia tora*, *Parthenium*, *Pongamia pinnata* (Figure 2) and percentage of these pollen type were

29.35%,15.59% 30.27% and 24.77% respectively. The absolute pollen count 109 was recorded and all the secondary pollen was observed.

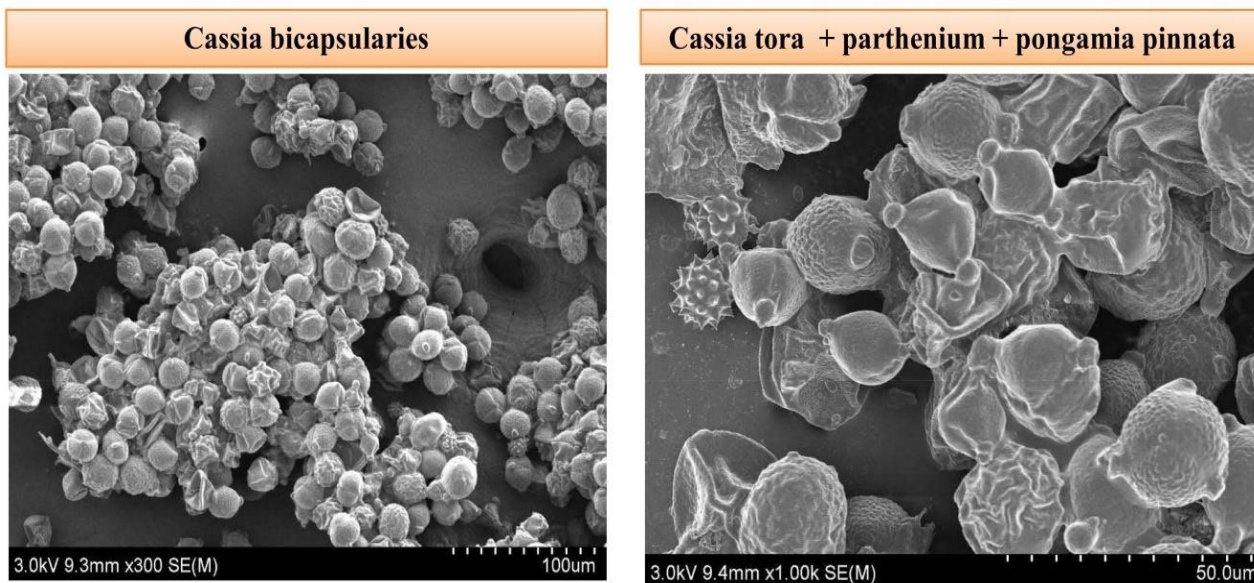


Fig 2: Images of pollen types observed in sample-2 collected from Handinimgaon village

Third sample (sample-3) was collected from Newasa. In this sample single pollen type i.e. secondary pollens were observed such as *cassia tora*, *celosia argentea*, *Ipomea hederifolia* (Figure 3) and percentage of this pollen type was

41.77% 34.17% and 24% respectively. The absolute pollen count 79 was noted. There was no evidence of predominant pollen type.

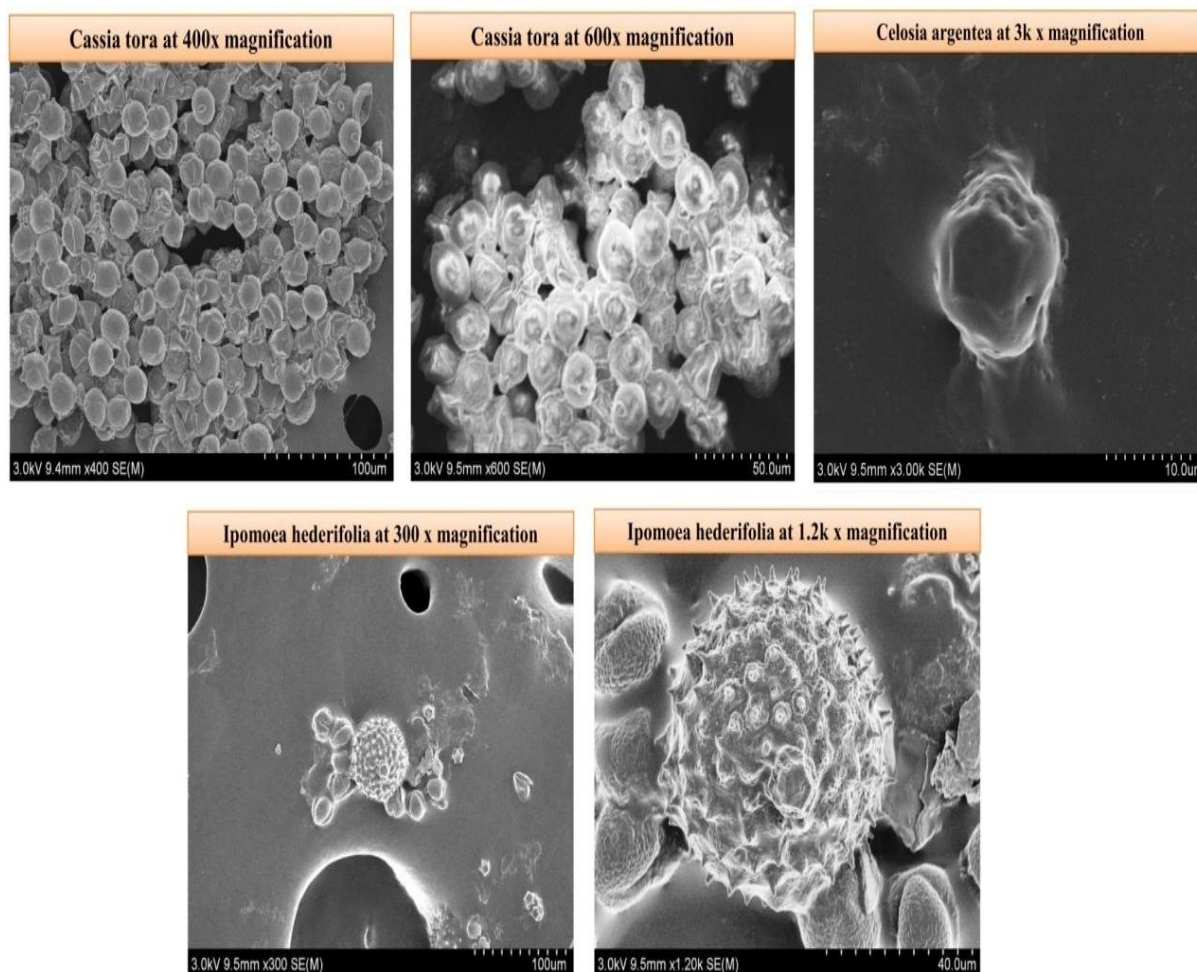


Fig 3: Images of pollen types observed in sample-3 collected from Newasa region

Fourth sample (sample-4) was collected from Newasa fata. In this sample two types of plant species pollen were observed i.e. *Moringa oleifera*, *Opuntia cochenillifera* (Figure 4) and percentage of these pollen type was 45.12%, and 54.47%

respectively. All the predominant pollen was noted and secondary pollen was totally absent. The absolute pollen count 82 was recorded.

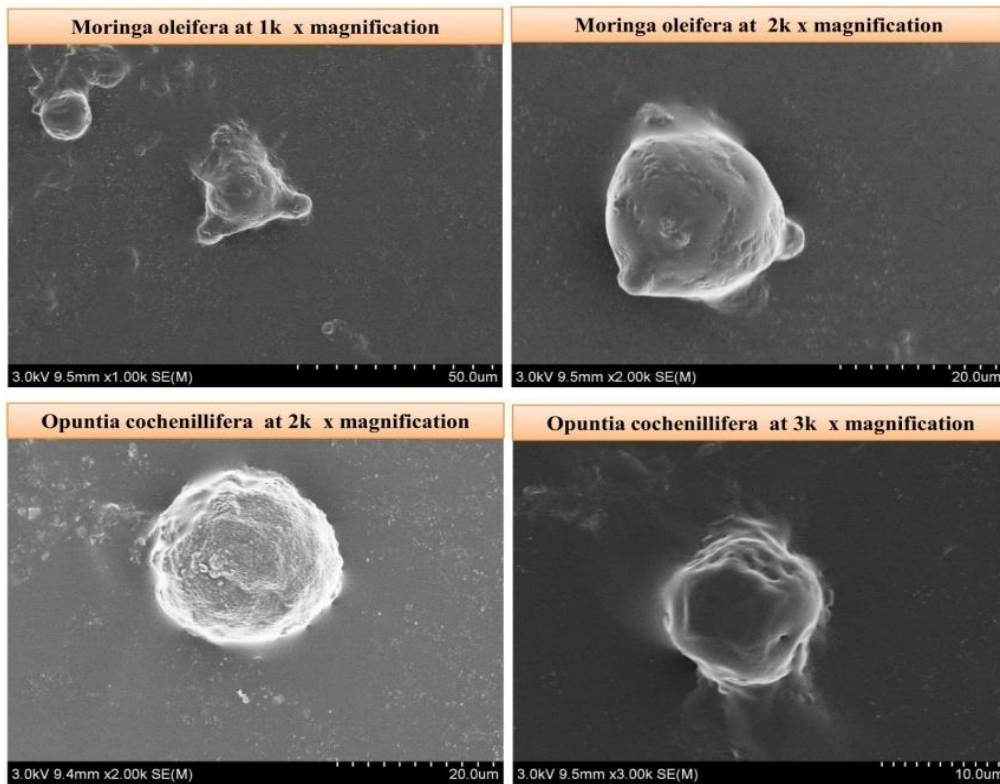


Fig 4: Images of pollen types observed in sample-4collected from Newasa phata region

Fifth honey sample (sample-5) was collected from Ranjangaon village. This sample had three types plant species pollen. These were *pongamiapinnata*, *Parthenium hysterothous*, *Solinium melongena* (Figure 5) and percentage

of these pollen type was 25.58%, 44.18% and 30.23% respectively. The absolute pollen count 129 was recorded. In all five samples multifloral types were recorded.

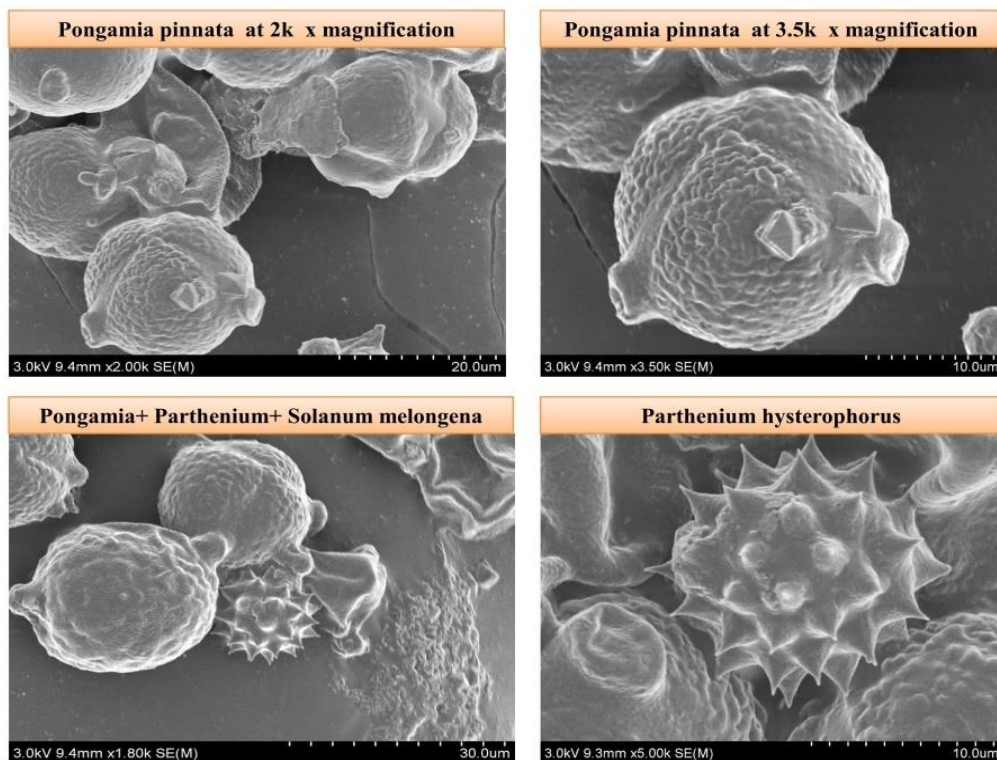


Fig 5: Images of pollen types observed in sample-5 collected from Ranjangaon

Discussion

Present Investigation revealed the presence of good potentials of pollen from honey samples. It indicate that bee used pollen grains from flowers for a growth of colony nectar source in honey would help beekeepers maintaining their colonies [7]. The result were found in each specimen of *Euphorbia coeruleascens*, *E.ledienii* and *E.terrains*. There is no microscopic characters that distinguish pollen grains of three species of Euphorbia. The analysis of exine structural pattern find specific identifying marks in different general Euphorbiaceae via SEM [8]. The pollen analysis carried out during year 2016-2017 by qualitative analysis of different honey samples. It is observed that the regions have wide diversity of nectar producing plant contaminations, tainted honey and mixtures. In present investigation 5 different samples investigated have 12 different pollen types belonging to 8 different families. Four types belonging to Fabaceae (leguminosae). Each single type belonging to family Asteraceae, Amranthaceae, Convolvulaceae, Moringaceae, Cactaceae, Solanaceae. In present study the pollen type such as *Opuntea cochenillifera*, *Mornings olifera*, *Parthenium hysterophrous*, and *Cassia tora* found to be higher amount of protein content.

Conclusion

During the present investigation analysis of honey and honey pollen were carried out. The honey samples were collected from different sites of Newasa tehsil, Ahmednagar district. This work indicate that the pollen analysis of honey would be provides information about quality of pollen texa present in study region. These pollen analytical study provides information of bee resources. The pollen grain from honey can be used for the taxonomic identification of honey yielding plants. The pollen analysis of different honey samples collected shown that the study region have wide diversity in bee flora. From the qualitative analysis of the honey, it is concluded that majority of pollen lodes is found to be family of Fabaceae, Asteraceae, Leguminosae, Amaranthaceae, Convolvulaceae, Moringaceae, Cactaeae, Solanaeae.

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