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Pollen analysis of summer honeys collected from forest area of Bramhapuri tahsil of Chandrapur district, (Maharashtra state)

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Abstract

The paper incorporates a qualitative and quantitative analysis of pollen contents in two squeezed honey samples of *Apis dorsata* hives collected from forest area of Bramhapuri tahsil of Chandrapur district. *Terminalia sp.* represents the predominant pollen type in one sample (46.58%) is designate as Termanalia honey. The other significant pollen types recorded include *Terminalia sp., Astericantha longifolia, Psidium guajava, Capsicum annumm, Sapindus emarginatus, Mangifera indica, Bombax ceiba, Blumea sp., Alengium salviifolium*. The pollen counts ranged from 410,000/g to 840,000/g. The data reflects the floral situation of the place were particular honey was produced and the identification of geographical origin based on the presence of a combination of pollen types of that particular area.

Keywords: pollen, honey, *apis dorsata*, forest area, bramhapuri tahsil

Introduction

Melittopalynology is an applied branch of palynology dealing with the study of pollen grains in honey samples and its application in Apiculture. Plant produces nectar and pollen both of which are avidly sought after by the bees to provide nutrition to the colony. Melittopalynology is concerned with the identification of pollen in honeys. Evaluation of plants for their utility as sources of bee forage provides the information needed to assess the potential for beekeeping in an area. Melittopalynological studies are thus helpful in bee management and in promoting the beekeeping development. Laboratory studies using Melitto paloynological methods have been made to evaluate sources of pollen and nectar for honey bees in different parts of the country namely Maharashtra (Laxmikant Borkar and Devendra Mate, 2017-18, Bhusari *et al*., 2005; Phadke, 1962; Kumar and Jagtap, 1988), Andra Pradesh (Ramanujam and Khatija, 1991, Kalpana and Ramanujam, 1991, Moses,1987, Karnataka (Yoganarasimhan, 1982; Agashe and Ranjaswami, 1997; Sheshagri, 1985; Bhargava *et al.*, 2009) [12, 20] Lucknow (Suryanarayana, 1976) and Indian honeys (Sen and Banarjee, 1956; Nair, 1964; Seethalakshmi, 1993) [10, 11, 15] Present investigation incorporates a quanlitative and quantitative pollen analysis of five honey sample from forest area of Bramhapuri tahsil of Chandrapur District. In order to identify the chief bee foraging plants recognize the uni and multifloral honeys and identify areas suitable for bee-keeping industry in this area. It is further investigated that a study of this nature would also highlight the geographical source of the honey samples.

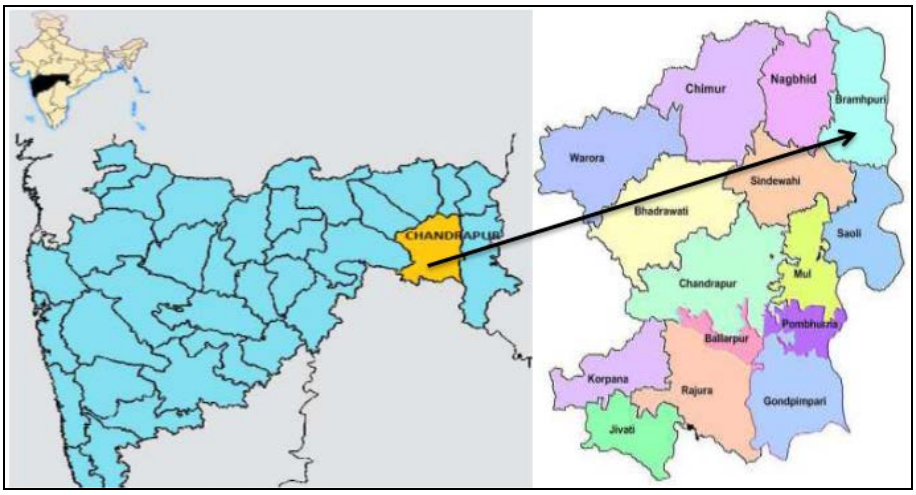

Fig 1: Map of Maharashtra Showing Chandrapur District & Bramhapuri tahsil

Materials and Methods

Two honey samples CHN-BRA-RAM, CHN-BRA-GAN *viz*., were collected during the period 29 April 2012 and 05 May 2013 from Rampuri and Ganeshpur respectively. All the samples represent squeezed honey collected from the natural *Apis dorsata* hives (Map). The squeezing (pressing)

of the honey combs was carried out under personal supervision and only honey bearing portion of the comb was used for this purpose. 1 ml of the honey sample was dissolved in 10 ml of distilled water & centrifuged. The sediment obtained was treated with 5 ml glacial acetic acid. The acetic acid was decanted and the material was subjected to acetolysis (Erdtman, 1960) [17] for analysing the pollen content in honeys qualitatively & quantitatively, three pollen slides were prepared for each sample. The recorded pollen types were identified with the help of reference slides collection & relevant literature for quantification of pollen types recorded, a total of 300 pollen grains were counted at random from the three palynoslides prepared for each samples. Based on their frequencies, the pollen types encountered were placed under the pollen frequency classes recommended by the international commission for bee Botany Louveaux *et al;*(1978) [16] *viz*., predominant pollen type (>45%), secondary pollen type (16-45%), important minor pollen types (3-15%),and minor pollen types (<3%).Non-melliferous (anemophilous) pollen types were excluded while determine the frequencies of melliferous pollen types (International Commission for Bee Botany Louveaux *et al.;* 1978) [16]. The absolute pollen counts of each sample was determined in accordance with the method recommended by Suryanarayana *et al.* (1981). Unacetolysed samples of honey were examined for the study of honeydew elements (fungal spores, hyphal shreads and algal filaments).

Results and Discussion

of the 2 honey samples collected from Bramhapuri tahsil, *Terminalia sp*. ranged from (46.23%) represent the predominant pollen type in one sample (CHN-BRA-RAM***)***. i.e. unifloral. The other significant pollen types recorded includes (secondary and upto minor pollen) *Terminalia sp., Astericantha longifolia, Psidium guajava, Sapindus emarginatus, Capsicum annumm, Melia azadirachta, Blumea sp., Alloum cepa, Alangium salviifolium, Bombax ceiba*, *Pongamia pinnata*.

Table 1: Pollen frequency class & frequencies (%) in *Apis dorsata* summer honey

Sample No.	Date of Collection	Type of Honey	Absolute pollen counts (APC)/g	HDE/P	Pollen Type
CHN-BRA-RAM	29-04-2012	Unifloral	84,000/g	0.01	P – Terminalia sp.(62.58) S - Nil I – Asteracantha longifolia(9.83) Pisidium guajava(9) Sapindus emarginatus(7.25) Capsicum annuum(7.16) Blumea sp.(4.83) Mangifera indica(4.08) Bombax ceiba(4) M – Pon(2.5), Mel(2), Cu(1), Ci(1.66), Ru(1.5), All(0.66), Car(0.5), NMP- Sorghum vulgare(0.86)
CHN-BRA-GAN	05-05-2013	Multifloral	410,000/g	0.01	P –Nil S - Terminalia sp.(40.16) I - Asteracantha longifolia(14.91) Pisidium guajava(9.7) Sapindus emarginatus(8.66) Capsicum annuum(5.5) Melia azadirchta(4) Blumea sp.(3.83) Allium cepa(3.33) Alangium salvifolium(3) M – Cel(2.5), Az(2), Leu(1.66), Br, Ci, Pa(each 1.33), Ju, Por(each 0.66), Ca, Br.(each 0.5) NMP –Nil

Table 2: Showing pollen morphology of Melliferous taxa

Sr No.	Pollen types	Pollen Size, Shape and Symmetry	Aperture pattern	Pollen wall (Sporoderm) Structure and sculpture
1	Alangium salviifolium			
2	Allium cepa	14-28× 32-48µm, ellipsoidal, Bilaterally symmetrical	Monosulcate, sulcus tenuimarginate	Exine 1.5 µm thick, subtectate, surface faintly reticulate
3	Asteracantha longifolia (Linn.) Nees.	56-59µm, Amb spheroidal or quadrangular; 50-55× 52-59µm, oblate spheroidal; Radially symmetrical	Tetracolporate, colpi long, ends tapering, tips acute, colpi alternating with 4 streak like pseudocolpi, ora more or less circular.	Exine 3.3µm thick, subtectate, surface reticulate, homobrochate, lumina polygonal and psilate.
4	Azadirachta indica	50-54µm, Amb squarish, sides convex; 47-54 38-47µm, subprolate, poles smoothly rounded; Radially symmetrical	Tetracolporate, colpi long, ends tapering, tips acute, ora lalongate	Exine 3 µm thick, tectate, surface psilate to locally granular

5	Blumea sp.	21-24 µm, Amb spheroidal, isopolar, Radially symmetrical	Tricolprate, colpi long	Exine 3 µm thick, surface echinate, spines 5-6 µm long, 4 spines in the inter apertural region interspinal area psilate
6	Bombax ceiba Linn	51 µm (49.5×52.5) µm, peroblate, isopolar, Radially symmetrical	Tricolprate, col. length 12 (10.5-13.5) µm	Exine thick 3 µm, coarsely reticulate, mesh 4.1 µm (3-4.5 µm) in the major part except at the angles showing medium reticulations 1-8 µm (1.5 -3 µm), greater number of baculae are found in the lumen. Muri simplibaculate, faint LO pattern.
7	Brassica sp.(Linn) Koch	30-33 µm, Amb rounded triangular to almost spheroidal; 27-31× 24-27 µm, prolate spheroidal; radially symmetrical	Tricolplate, colpal ends tapering, tips acute	Exine 2.5 µm thick, sub tectate, surface reticulate, heterobrochate, meshes narrow at mesocolpial regions giving a striate look, lumina polygonal.
8	Capparis grandis	10-12 µm, Amb spheroidal; 14-16 ×9-12 µm prolate to subprolate; Radially symmetrical	Tricolporate, colpi linear to narrowly elliptic, ends tapering, tips acute, ora faint lalongate	Exine 1 µm thick, tectate, surface faintly granular to almost psilate
9	Capsicum annuum Linn.	29-34 µm, Amb spheroidal; 29-35× 26-30 µm, subprolate; radially symmetrical	Tricolporate, colpi constricted at oral region, ends tapering, tips acute, ora prominently lalongate	Exine 1.5 µm thick, tectate, surface faintly granular to almost psilate
10	Careya arborea Roxb.	52.1× 40.1 µm (48-54× 37.5 -43.5) µm, subprolate, isopolar, radially symmetrical	Hexacolpate, syncolpate with crassimarginate colpi, col. Length 43.5 (42-46.5) µm	Exine thick, 3 µm, undulating, considerable thick at the poles sexine-nexine not differentiated medium reticulate, more coarse at the poles. Mesh 1.5-3 µm, clear LO pattern
11	Celosia argentea	30-35 µm spheroidal radially symmetrical	Pantoporate, pore No. 15-20, circular. Diam; 4-5 µm, pore membrance flecked with granules, interporal distance 8-11 µm	Exine 2 µm thick, tectate, interporal space coarsely granular
12	Citrus sp.	27-29 µm, Amb squarish, 26-30 ×25-27 µm, prolate spheroidal radially symmetrical	Tetracolporate, colpi linear, tips acute, ora lalongate	Exine 2 µm thick subtectate, surface Reticulate. Heterobrochate, meshes smaller near the apertural regions and larger elsewhere, lumina hexa to pentagonal or irregular, psilate, muri simpli to locally duplibaculate
13	Cucurbitaceae type	50-63 µm, subprolate, isopolar, Radially symmetrical	Tricolporate, col. Length 48.7 (48-49.5) µm	Exine thick 4.5 µm, sexine - nexine not discernible, rather coarsely reticulate, mesh 3.4 (3 – 4.5) µm, baculae distinct, clear LO pattern
14	Justicea procumbence			
15	Leucanaea leucocephala	52-59 µm, Amb spheroidal: 47-49×51-58 µm, sub oblate: Radially symmetical	Tricolporate colpilong, tips acute, ora lalongate	Exine 4 µm thick, subtectate surface microreticulate, homobrochate
16	Mangifera indica Linn.	27-31 µm, Amb subtriangular; 29-32 ×26-28 µm, subprolate; Radially symmetrical	Tricolporate colpi long, tips acute ora prominently lanlongate	Exine 2.5 µm thick, subtectate, surface striatoreticulae, striations more or less parallel in equatorial view, lumen generally elongated in polar direction, murisimplibaculate
17	Melia azadirachta	47.2× 36.7 µm,, 46.5-48× 36.37.5 µm,,subprolate, isopolar, Radially symmetrical	Tetracolporate, col. Length 36 µm, ora 6.5 µm, lalongate, pantoporate, pores, circular 6.9 µm,	Exine thick 3 µm, sexine- nexine not clear, psilate
18	Parthenium hysterophorus	16.6 to 19.8 µm, Amb spheroidal, oblate spheroidal, radially symmetrical	Tricolporate colpi long, ends tapering, tips acute, ora lalongate	Exine 3 µm thick, tectate, surface echinate, spines short 2 µm, to 3 µm,, long 2 µm, in diam at base.
19	Pisidium guajava Linn.	24-25 µm, Amb subtriangular; 13-16× 26-28 µm, oblate; Radially symmetrical	Tricolporate, syncolpate, parasyncolpate, ora lalongate	Exine 1.5 µm thick, tectate surface granular to pailate
20	Pongamia pinnata (Linn) Pierre.	29-31 µm, Amb subtriangular: 27-31× 25-28 µm, subprolate; Radially symmetrical	Tricolporate, colpi linear to narrowly elliptic tips acute, ora lalongate	Exine 1.5 µm thick, subtectate, surface granular to locally faintly microrecticulate
21	Portulaca oleracea	63 µm, spheroidal; radially symmetrical	Pantacolpate, some of the colpi showing a tendency towards pentagonal arrangement around the polar area of each hemisphere. Colpi faint, 15 µm, long with acute tips	Exine 4 µm thick, tectate, surface densely spinulate, spinules 1-2 µm long.
22	Prosopis juliflora	36-39 µm, Amb rounded triangular; 38-42× 30-35 µm, prolate to subprolate; Radially symmetrical	Tricolllporate, occasionally syncolpate, colpi tapering towards poles, tips acute, ora lalongate	Exine 3.2 µm thick, tectate surface faintly reticulate
23	Rungia repens (Linn.) Nees.	40-44 ×25-26 µm, oblong; Bilaterally symmetrical	Diporate, pores circular, 2.5 µm, in diam, margin of the pores densely beset with small processes	Exine 3 µm thick at poles, 4.6 µm at equaltor, subtectate, tectum undulating, distinct rounded to irregular areolae (2-

				4 µm) linearly aligned in the vicinity of apertures, rest of the wall microreticulate
24	Sapindus emarginatus vahi.	24-26 µm, Amb triangular, sides straight or even slightly concave; 18-20×26-29 µm, oblate (occasionally suboblate); Radially symmetrical	Tricolporate, colpi narrowly elliptic long, tips acute, ora lOlongate	Exine 2 µm thick on mesocolpia, 1-1.5 µm thick near apertures, surface psilate
25	Terminalia sp.	19-22 µm, Amb spheroidal; 21-24 x20-22 µm, subprolate; Radially symmetrical	Tricolporate, colpi alternating with pseudocolpi colpi linear, tips acute pseudocolpi almost equal the size of colpi, ora more of less circular	Exine 1.5 µm thick, tectae, surface psilate to locally finely granular

Table 3: Showing pollen morphology of Non-melliferous taxa

Sr. No.	Pollen types	Pollen Size, Shape and Symmetry	Aperture pattern	Pollen wall (Sporoderm) Structure and sculpture
01	Sorghum vulagare Pers.	51-55 µm, spheroidal; Radially symmetrical	Monoporate, pore circular provided with annulus, pore diam with annulus 4.1 µm without annulus 3.3 µm	Exine 1 µm thick, tectate, surface faintly granular to almost psilate

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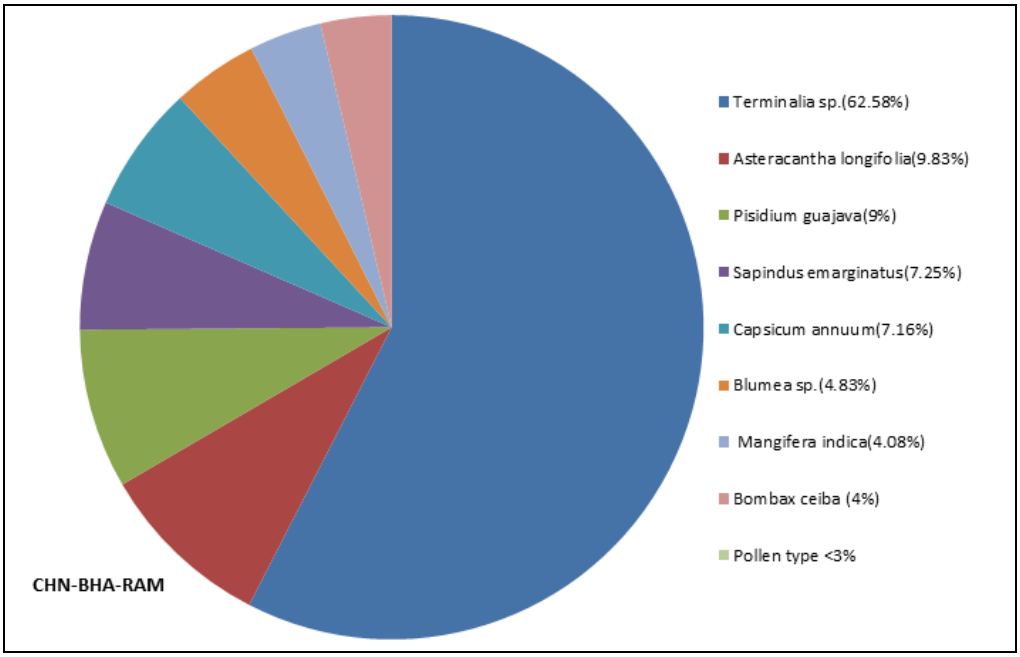

Fig 1: Palynograph of Rampuri

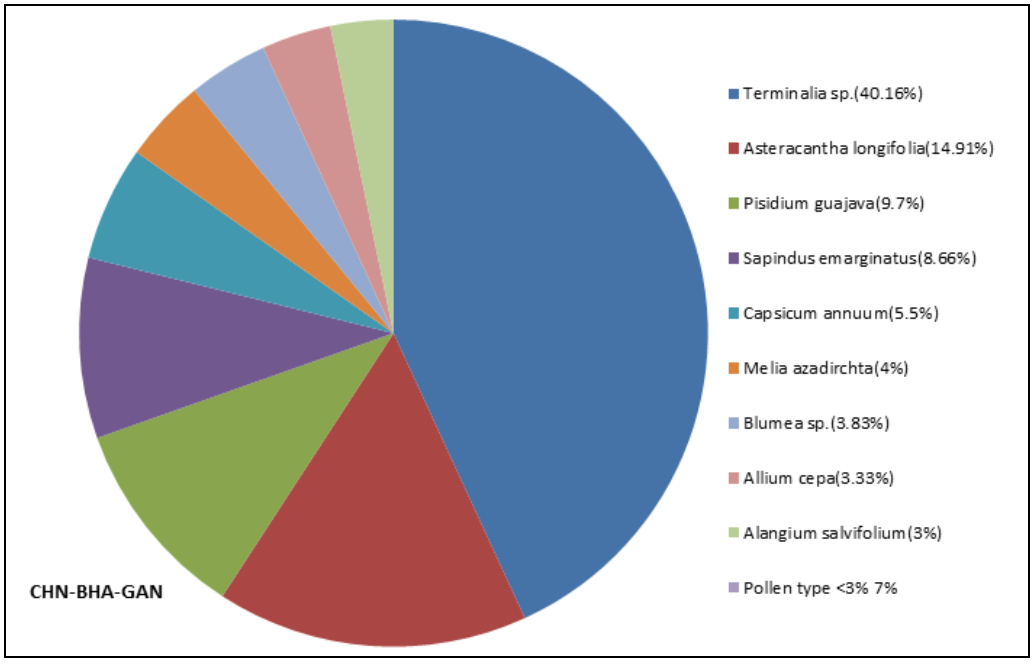

Fig 2: Palynograph of Ganeshpur

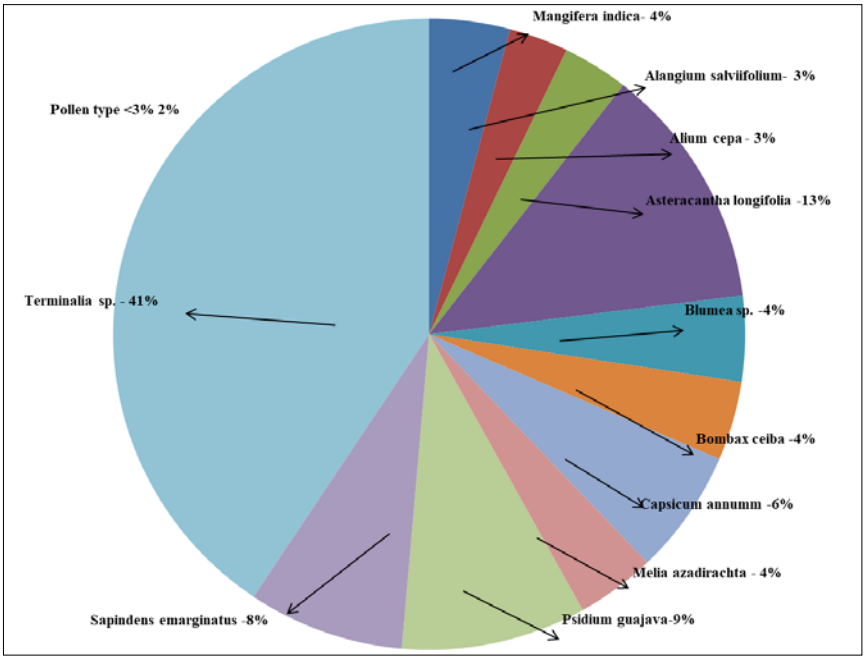

Fig 3: Composite palynograph of summer honeys from Bramhapuri tehsil

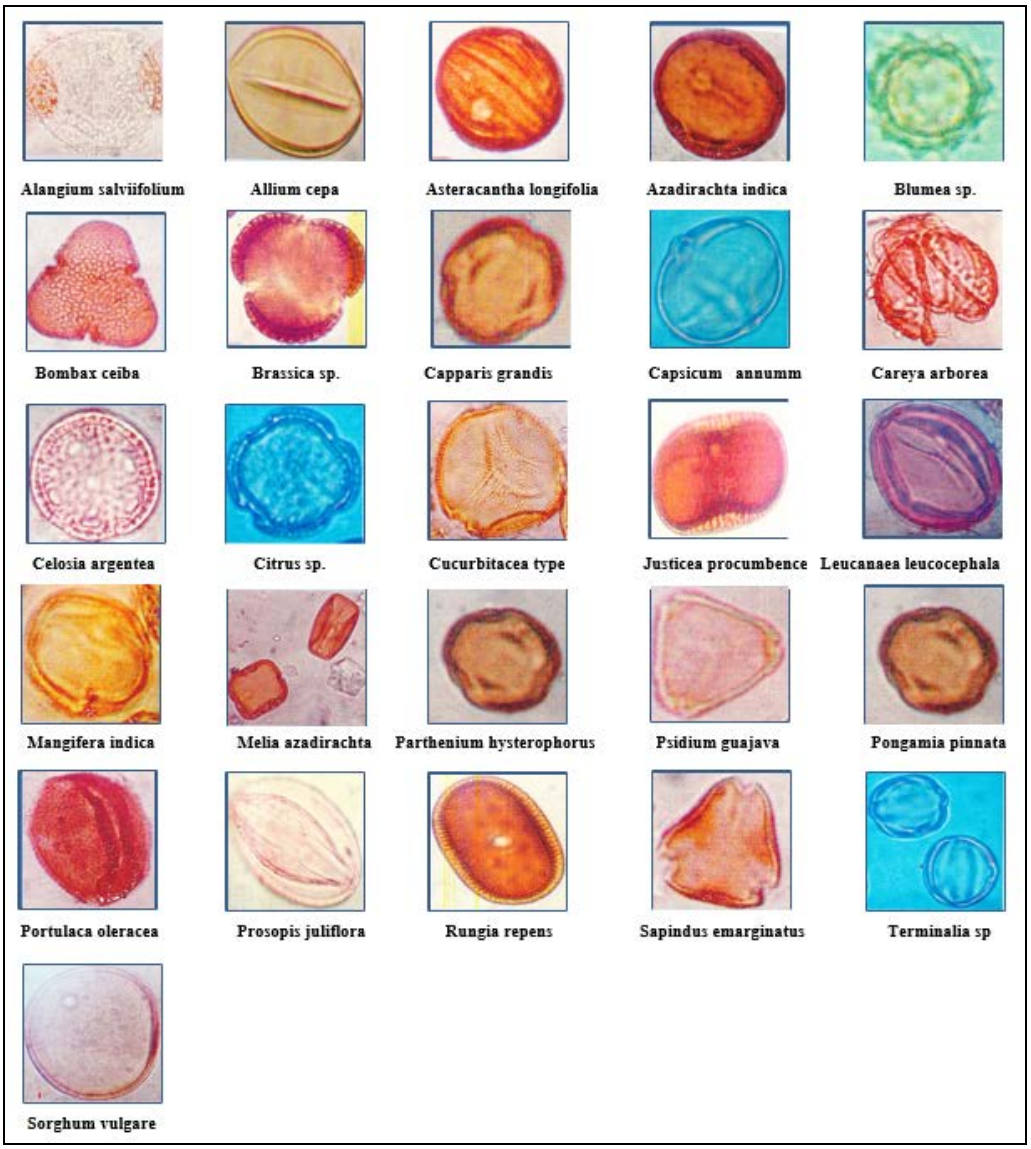

Fig 4: Plate Microscopic photograph of pollen grains found in honey sample

All together 26 pollen types (25 of melliferous and 1 of non-melliferous taxa) referable to 21 families have been recorded from these samples (Photoplate). The sample (CHN-BRA-GAN) showed the maximum number of pollen type (19) and the sample (CHN-BRA-RAM), the minimum number (16). The absolute pollen counts ranged from 410,000/g to 840,000/g and the HDE/P ratio 0.01 and represented by fungal spores (Table 1). The details of the pollen analysis of the 2 honey samples (melliferous/non-melliferous) are represented in table1. Similarly individual palynograph (Pollen spectra) of each honey sample and composite palynograph was also given to show the pollen contents of the samples of Bramhapuri tehsil (Fig. 1.1-1.3). The distinguishing morphological features of the pollen types encountered in the present study are given below. The bee plants of Bramhapuri tahsil are Referable to 3 categories:

1. **Crop plants:** *Allium cepa, Brassica sp., Capsicum annumm and Sorghum vulgare.*
2. **Arborescent taxa/shrub:** *Azadirachta indica, Bombax ceiba, Citrus sp., Leucanaea leucocephala, Mangifera indica, Melica azadirachta, Pongamia pinnata, Psidium guajava, Sapindence emarginatus, Terminalia sp., Capparis grandis, Prosopis juliflora, Alangium salviifolium.*
3. **Herbaceous weeds:** *Asteracantha longifolia, Celosia argentea, Portulaca oleracea.*

   of these three categories the arborersent plants *Terminalia sp.* constitute the chief bee forage plants in this tahsil during summer season. Besides the other arborersent plants *Delonix regia, Pongamia pinnata, Azadirachta indica* represents most preferred nectar sources for the honey bees. Our observation indicate that *Terminalia sp.* represent abundant nectar and pollen sources to *Apis dorsata*. The region selected for the present study has good potential for sustaining bee keeping ventures because of the diversity of nectar and pollen taxa. Since *Terminalia sp.* are member of combretaceae is major sources of forage for honey bees therefore efforts should be made to increase its cultivation. The other plant encountered in these honey samples are the member of families like Acanthaceace, Anacardeaceace, Mimoscae, Caesalpiniaceace, Celastraceace, Myrtaceace, Samydaceae, Menisphermaceace, Liliaceace, Capparidaceae, Amaranthaceae, Cleomaceae, Solanaceae, Papillionaceae and Sapindaceae in this area.

To improve the bee-keeping industry a proper understanding and mutualism between bees and available plant taxa in the region and in a particular season is necessary. The identified taxa were not only the economic crops but also play an important role in the development of bee-keeping in this region. This data reflects the floral situation of the place where particular honey was produced and the identification of geographical origin based on the presence of a combination of pollen types of that particular area.

Conclusion

The Present melittopalynological concepts based on 02 honey samples and *Apis dorsata* from seasons clearly brought to light that Bramhapuri tehsil of Chandrapur district with large forest area in which *Terminaliasp*. with extensive plantations of *Asteracantha longifolia*, *Psidium guajava*, *Sapindus emarginatus* and *Capsicum annumm* not only provide the bulk of gaint bee honey but also have the potential for attracting bee keeping industry involving colonies of the Indian Hive Bee, *Apis cerena* on modest commercial scale.

Adequate primary source of nectar and pollen, augmented by secondary or alternate source when require for prolonged periods, availability unpolluted water resources and suitable soil types with enough moisture are prerequisites of paramount importance for successful operation for bee-keeping venture in any area. A consideration of all these parameter clear by indicates that the forest area of Bramhapuri, in particular merit serious attention for establishing bee keeping enterprises for commercial honey production in Chandrapur district. Maintenance of apiaries in the vicinity of agriculture crops, in additional providing enough harvest of honey, also result in enhanced crop yield as aconsequence of higher degree of pollination. Summer represents the honey flow season period with adequate source of bee forage in the forest area with agricultural tracts; successful operation of apicultural activities necessitates of apiary colonies to nearby deciduous forest areas during summer months, for uninterrupted production of commercial honey.

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