



## Melissopalynological analysis of selected samples from Kannur and Wayanad districts of Kerala, India

K. Rimna<sup>1</sup>, K. Sasikala<sup>1\*</sup>, M. Reema Kumari<sup>2</sup>, & Maya C. Nair<sup>3</sup>

<sup>1</sup>Mahatma Gandhi Govt. Arts College, Mahe, U.T. of Puducherry

<sup>2</sup>Maharani Lakshmi Ammanni College for Women, Bengaluru, Karnataka

<sup>3</sup>Govt. Victoria College, Palakkad, Kerala

Manuscript received 2nd June 2017, revised 3rd Aug 2017, accepted 5th Aug 2017

### Abstract

**Background and Aim:** In the present study 5 samples of honey namely sample (S1) - Cheru Then from Wayanad, sample (S2) - Van Then from Kannur, sample (S3) - Rubber honey from Kunduchira, Thalassery, Kannur, sample (S4) - Tea honey from Chirakara, Thalassery, Kannur and sample (S5) - Dabur honey from market were procured from Kannur and Wayanad districts, Kerala for pollen analysis. **Methodology:** The microscopic analyses of the samples were made by following the standard procedures. The microphotographs were also taken. **Results:** A total of 36 pollen types belonging to 21 families were identified and enumerated. Samples S3 and S4 showed the maximum and minimum number of pollen taxa respectively. Unidentified pollen types were found in samples S3 and S4 as minor type and secondary type. The percentage frequency of the sample is greatest in sample S1 and S3 with 58% and 53% respectively and least frequency below 1% is found in S2 and S4. Of the five samples analysed, one sample S2 showed unifloral with *Cocos nucifera* as predominant pollen and the remaining sample S1, S3 and S4 were considered multifloral. The sample S5 showed complete absence of the pollen grains. The frequently occurring pollen types are *Cocos nucifera*, *Syzygium* Type I, *Syzygium cumini*, *Pennisetum polystachyon* and *Urena lobata*. The dominant families include Arecaceae, Asteraceae, Caesalpiniaceae, Elaeocarpaceae, Euphorbiaceae, Loranthaceae, Malvaceae, Mimosaceae, Myrtaceae, Passifloraceae, Poaceae, Sapindaceae and Verbenaceae. Majority of the samples showed dominance of tree species which is mostly preferred by bees for nectar and pollen sources for honey production. **Conclusion:** Pollen analysis in honey samples lead to the identification of vegetation in the area which is important for the survival of bee colonies and in turn helps in conserving the biodiversity of the area.

**Keywords:** Melissopalynology, Pollen analysis, Kannur, Wayanad, Kerala

@2017 BioMedAsia All right reserved

### 1. Introduction

Melittopalynology/ melissopalynology gained considerable importance in India and various parts of the world by the end of 19<sup>th</sup> century. The first scientific information on the geographic origin of honey was reported by Pfister in 1895. The analysis of the pollen grains in the honey is

essential to identify the geographical and botanical origin of honey and pollen preferences by the honey bees for their diet. The vegetation pattern and plant species specificity greatly influence the activities of bee colonies and the quality and quantity of honey production. Across many countries the bee forage plants have been studied through pollen analysis in honey and visual observations.

Abundant research on analysis of pollen from honey samples, pollen loads, bee nutrition and bee keeping were reported in different parts of the country and worldwide. Several studies on bee plant relationship have been carried out by many workers (Deodikar & Thakar, 1955, 1966, Deodikar, 1964, Suryanarayana and Thakar, 1966). Polln analysis in different honey samples in India was carried out by different workers (Sharma & Nair (1965), Suryanarayana *et al.*, (1981), Sivaram *et al.*, 2012, & Ashoke Bhattacharya, 2014).

Pollen analytical studies of Indian honeys are fragmentary, although the pollen and the honey sample have been studied from various parts of India especially north and north eastern states with exception of Tamil Nadu, Kerala, and Karnataka. However, melissopalynological studies from Kannur and Wayanad districts of Kerala, India are scanty. Hence the present study has been under taken with the following objectives.

- To find out the Geographical origin of honey, to

\*Corresponding author

Full Address :

Mahatma Gandhi Govt. Arts College, Mahe, U.T. of Puducherry

Phone no. ?????? ????????

E-mail: drsasikalabot@gmail.com

understand the Botanical source of honey (nectar producing flora), to study the Pollen diversity of honey sample and Pollen percentage frequency.

**2. Methodology**

In the present investigation five samples of honey were procured from Kannur and Wayanad districts of Kerala, for pollen analysis. The microscopic analyses of these samples were made by following the standard procedures (Erdtman, 1960). Details of the samples and methodology adopted are summarized below:

Sample (S1) - Cheru Then from Wayanad; Sample (S2) - Van Then from Kannur; Sample (S3) - Rubber honey from Kunduchira, Thalassery, Kannur; Sample (S4) - Tea honey from Chirakara, Thalassery, Kannur; Sample (S5) - Dabur honey from Market.

Five pollen slides were prepared from each honey sample. Pollen grains were observed under light microscope and were identified with the help of available literature up to the family, genus and species level. Pollen frequency was analyzed for each sample type and categorized into groups based on (Louveaux et al., 1978 & ICBB, 1970). The morphology has been explained based on Punt et al., 2007. The microphotographs of the pollen were made with a Nikon Eclipse 50-i Computer attached anatomical microscope under oil immersion (100x) lens using 10x eye piece. Pollen measurements of polar and equatorial diameters were recorded.

**3. Results**

Melissopalynological studies of five honey samples collected from Kannur and Wayanad districts were recorded. 36 different pollen types were identified belonging to 21 families (Table I). The diversity of pollen types in honey samples is depicted in Table II. 28 pollen types were identified at species level, 4 types at family level, 2 at genus level and 2 were unknown. The pollen characteristics of the species diversity identified from different honey samples are summarized below. The microphotographs of most of the pollen types in honey samples are represented in Plate I.

**3.1 Enumeration of Pollen grains**

**Acacia auriculiformis** A. Cunn. ex Benth. (Mimosaceae) (Plate I 1)

Grains in polyads; polyads 16-celled, elliptic; bilateral symmetry, paraisopolar; equatorial outline elliptic-obtuse-plane; polyapantoporate; sculpturing psilate.

**Acanthaceae Type** (Plate I 27)

Grains in monads; 4.55 x 9.47 µm; Tricolporate; sculpturing microreticulate.

**Achyranthes aspera** L. (Amaranthaceae) (Plate I 2)

Grains in monads; 17.68 - 21.72 x 16.45 - 20.96 µm;

**Table I: List of Pollen Types in Honey Samples**

Sl. No.	Pollen Type
1	<i>Acacia auriculiformis</i> A. Cunn. ex Benth
2	Acanthaceae Type
3	<i>Achyranthes aspera</i> L.
4	<i>Adenantha pavonia</i> L.
5	<i>Aporosa lindleyana</i> (Wight) Baill.
6	<i>Areca catechu</i> L.
7	Asteraceae Type I
8	Asteraceae Type II
9	<i>Caesalpinia bonduc</i> (L.) Roxb.
10	<i>Camellia sinensis</i> (L.) O. Ktze.
11	<i>Careya arborea</i> Roxb.
12	<i>Cocos nucifera</i> L.
13	<i>Croton bonplandianum</i> Baill.
14	<i>Dendrophthoe falcata</i> (L.f.) Etting.
15	<i>Dillenia pentagyna</i> Roxb.
16	<i>Elaeocarpus serratus</i> L.
17	<i>Gomphrena celosioides</i> Mart.
18	<i>Helixanthera wallichiana</i> (Schult.) Dans.
19	<i>Hevea braziliensis</i> (Willd. ex A. Juss.) Muell.-Arg.
20	<i>Lagerstroemia speciosa</i> (L.) Pers.
21	Lamiaceae Type
22	<i>Mimosa pudica</i> L.
23	<i>Passiflora foetida</i> L.
24	<i>Peltophorum pterocarpum</i> (DC.) Backer ex Heyne
25	<i>Pennisetum polystachyon</i> (L.) Schult.
26	<i>Schleichera oleosa</i> (Lour.) Oken.
27	<i>Stachytarpheta jamaicensis</i> (L.) Vahl.
28	<i>Syzygium cumini</i> (L.) Skeels
29	<i>Syzygium</i> Type I
30	<i>Syzygium</i> Type II
31	<i>Terminalia paniculata</i> Roth
32	<i>Tridax procumbens</i> L.
33	<i>Urena lobata</i> L.
34	Verbenaceae Type
35	Pollen Type I
36	Pollen Type I

polar and equatorial view spheroidal or circular, radially symmetrical, isopolar, polyapantoporate; pore circular; sculpturing verrucate.

**Adenantha pavonia** L. (Mimosaceae) (Plate I 3)

Grains in polyads; polyads 12-celled, spherical, 41.79 x 39.84 µm; polar outline circular; equatorial outline elliptic obtuse, plane, polyapantoporate; sculpturing psilate.

**Aporosa lindleyana** (Wight) Baill. (Euphorbiaceae)

Grains in monads; 13.46 - 18.26 x 8.77 - 15.10 µm, prolate; polar outline triangular-obtuse-convex; equatorial outline elliptic; trizonocolporate; sculpturing

**Table II: Diversity of Pollen Types in Honey samples**

Sl. No.	S1	S2	S3	S4	S5
1.	+	-	+	-	-
2.	+	-	+	-	-
3.	+	-	+	-	-
4.	+	-	+	-	-
5.	+	-	+	-	-
6.	-	-	+	-	-
7.	+	-	+	-	-
8.	-	-	+	-	-
9.	+	-	+	-	-
10.	-	-	-	+	-
11.	+	+	-	-	-
12.	+	+	+	-	-
13.	+	-	+	-	-
14.	+	-	+	-	-
15.	-	-	+	-	-
16.	+	-	+	-	-
17.	+	-	+	-	-
18.	-	-	+	-	-
19.	+	-	+	-	-
20.	+	+	+	-	-
21.	+	-	+	-	-
22.	+	-	+	-	-
23.	+	-	+	-	-
24.	+	-	+	-	-
25.	+	-	+	+	-
26.	+	-	+	-	-
27.	+	-	+	-	-
28.	+	+	+	-	-
29.	+	+	+	+	-
30.	+	-	+	-	-
31.	+	-	+	-	-
32.	+	-	+	-	-
33.	+	+	+	-	-
34.	+	-	+	-	-
35.	-	-	+	-	-
36.	-	-	+	+	-

S1 = Cheru Then, S2 = Van Then I, S3 = Van Then II, S4 = Rubber Honey, S5 = Tea Honey, S6 = Dabur honey. 1. *Acacia auriculiformis*, 2. Acanthaceae Type I, 3. *Achyranthes aspera*, 4. *Adenantha pavonina*, 5. *Aporosa lindleyana*, 6. *Areca catechu*, 7. Asteraceae Type I, 8. Asteraceae Type II, 9. *Caesalpinia bonduc*, 10. *Camellia sinensis*, 11. *Careya arborea*, 12. *Cocos nucifera*, 13. *Croton bonplandianum*, 14. *Dendrophthoe falcata*, 15. *Dillenia pentagyna*, 16. *Elaeocarpus serratus*, 17. *Gomphrena celosioides*, 18. *Hevea brasiliensis*, 19. *Helixanthera wallichiana*, 20. *Lagerstroemia speciosa*, 21. Lamiaceae Type, 22. *Mimosa pudica*, 23. *Passiflora foetida*, 24. *Peltophorum pterocarpum*, 25. *Pennisetum polystachyon*, 26. *Schleichera oleosa*, 27. *Stachytarpheta jamaicensis*, 28. *Syzygium cumini*, 29. *Syzygium* Type I, 30. *Syzygium* Type II, 31. *Terminalia paniculata*, 32. *Tridax procumbens*, 33. *Urena lobata*, 34. Verbenaceae Type, 35. Pollen Type I, 36. Pollen Type II.

microreticulate.

#### ***Areca catechu* L. (Arecaceae) (Plate I 4)**

Grains in monads; 22.61 x 15.59  $\mu\text{m}$ . Pollen monocolpate, elongate; equatorial outline elliptic, bilaterally symmetrical, paraisopolar; sculpturing reticulate.

#### **Asteraceae Type I (Plate I 28)**

Pollen grains in monads; 5.37 - 13.03 x 5.5 - 12.92  $\mu\text{m}$ ; polar and equatorial view circular or spheroidal, radially symmetrical, apolar, trizonocolporate; sculpturing faintly microreticulate.

#### **Asteraceae Type II (Cultivated) (Plate I 29)**

Pollen grains in monads; polar and equatorial view elliptic-circular, hexazonocolporate; sculpturing reticulate-echinate.

#### ***Caesalpinia bonduc* (L.) Roxb. (Caesalpinaceae) (Plate I 5)**

Grains in monads; 34.99 x 34.82  $\mu\text{m}$ , prolate; polar outline elliptic, trizonocolporate, operculate; sculpturing reticulate.

#### ***Camellia sinensis* (L.) O. Ktze. (Theaceae)**

Grains in monads; Tricolporate, triangular; sculpturing faintly reticulate.

#### ***Careya arborea* Roxb. (Lecythidaceae) (Plate I 6)**

Pollen grains in monads; 20.10 x 19.59  $\mu\text{m}$ ; polar outline triangular-obtuse-convex; equatorial outline transversely elliptic; trizonosyncolpate; sculpturing psilate.

#### ***Cocos nucifera* L. (Arecaceae) (Plate I 7)**

Grains in monads; 4.5 - 31 x 29.68  $\mu\text{m}$ ; pollen monocolpate; sculpturing faintly reticulate.

#### ***Croton bonplandianum* Baill. (Euphorbiaceae) (Plate I 8)**

Grains in monads; 38.52 - 52.04 x 37.82 - 50.42  $\mu\text{m}$ , circular, apolar, radially symmetrical, inaperturate; crotonoid pattern.

#### ***Dendrophthoe falcata* (L.) Etting. (Loranthaceae) (Plate I 9)**

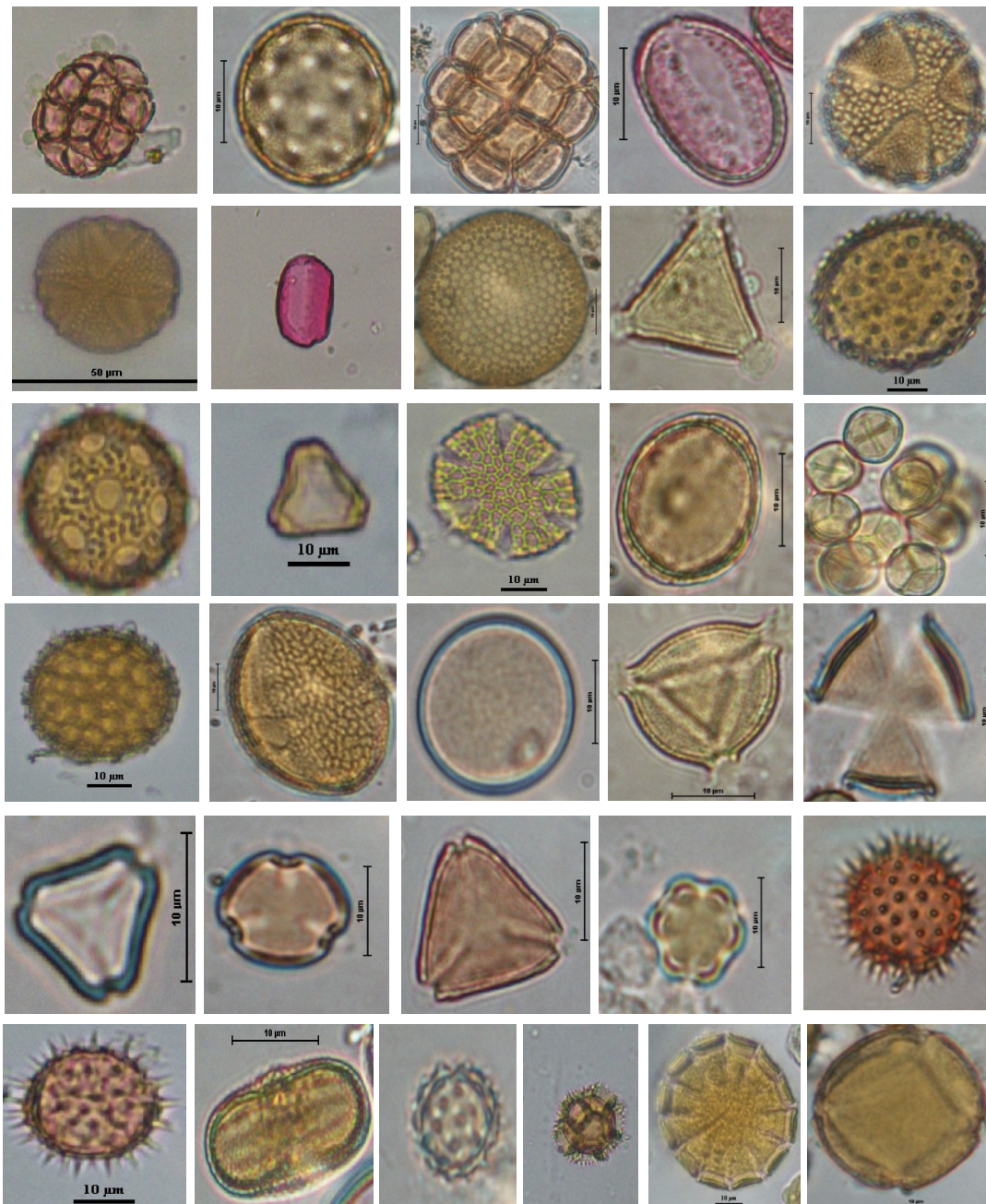
Pollen grains in monads; polar outline triangular-obtuse-concave; equatorial outline elliptic; trizonocolpate; sculpturing psilate.

#### ***Dillenia pentagyna* Roxb. (Dilleniaceae) (Plate I 10)**

Pollen grains in monads; polar outline circular, oblate-spheroidal; equatorial outline elliptic; trizonocolpate; sculpturing verrucate.

#### ***Elaeocarpus serratus* L. (Elaeocarpaceae)**

Pollen grains in monads; polar outline circular; equatorial outline elliptic; trizonocolporoidate; sculpturing psilate or faintly reticulate.



**Plate I: Pollen Types.** 1: *Acacia auriculiformis*, 2: *Achyranthes aspera*, 3: *Adenantha pavonina*, 4: *Areca cat-echu*, 5: *Caesalpinia bonduc*, 6: *Careya arborea*, 7: *Cocos nucifera*, 8: *Croton bonplandianum*, 9: *Dendrophthoe falcata*, 10: *Dillenia pentagyna*, 11: *Gomphrena celosioides*, 12: *Helixanthera wallichiana*. 13: *Hyptis suaveolens*, 14: *Lagerstroemia speciosa*, 15: *Mimosa pudica*, 16: *Passiflora foetida*, 17: *Peltophorum pterocar-pum*, 18: *Pennisetum polystachyon*, 19: *Schleichera oleosa*, 20: *Stachytarpheta jamaicensis*, 21: *Syzygium cumini*, 22: *Syzygium* Type I, 23: *Syzygium* Type II, 24: *Terminalia paniculata*, 25: *Tridax procumbens*, 26: *Urena lobata*, 27: Acanthaceae Type, 28: Asteraceae Type I, 29: Asteraceae Type II, 30: Lamiaceae Type, 31: Verbenaceae.

**Gomphrena celosioides** Mart. (Amaranthaceae) (Plate I 11)

Grains in monads; Polar view circular or spheroidal, isopolar, radially symmetrical; sculpturing reticulate.

**Helixanthera wallichiana** (Schult.) Dans. (Loranthaceae) (Plate I 12)

Grains in monads; Polar outline triangular-obtuse-concave; equatorial outline elliptic, trizonocolpate;

sculpturing psilate.

**Hevea braziliensis** (Willd. ex Juss.) Muell.-Arg. (Euphorbiaceae)

Grains in monads. Polar view triangular-acute, straight, trizonocolporate, radially symmetrical; sculpturing psilate.

**Hyptis suaveolens** (L.) Poit (Lamiaceae) (Plate I 13)

Pollen grains in monads; polar outline triangular, obtuse-

convex; equatorial outline elliptic, hexazonocolpate; sculpturing reticulate

**Lagerstroemia speciosa** (L.) Pers. (**Lythraceae**) (**Plate I 14**)

Grains in monads; 19.51 x 18.99  $\mu\text{m}$ , paraoblate; polar outline triangular, obtuse-convex; equatorial view elongated; trizonocolporate; sculpturing microreticulate.

**Lamiaceae Type** (**Plate I 30**)

Pollen grains in monads; 29.19 x 28.97  $\mu\text{m}$ ; polar and equatorial view circular or spheroidal, radially symmetrical, isopolar, 12-zono-colpate; sculpturing reticulate.

**Mimosa pudica** L. (**Mimosaceae**) (**Plate I 15**)

Pollen grains in tetrads; 7.80 - 8.72 x 7.61 - 8.35  $\mu\text{m}$ ; tetrads spherical; polar outline circular; equatorial outline quadrangular, obtuse-plane; tetrapantoporate; sculpturing psilate.

**Passiflora foetida** L. (**Passifloraceae**) (**Plate I 16**)

Grains in monads; 24.80 x 23.23  $\mu\text{m}$ ; polar view oblate-spheroidal; trizonocolporate; sculpturing reticulate.

**Peltophorum pterocarpum** (DC.) Backer. ex Heyne (**Caesalpiniaceae**) (**Plate I 17**)

Grains in monads; polar outline circular, 28.17 - 43.32 x 25.58 - 34.87  $\mu\text{m}$ ; equatorial outline circular to elliptic, 25.58 x 39.49  $\mu\text{m}$ ; trizonocolporate; sculpturing reticulate.

**Pennisetum polystachyon** (L.) Schult. (**Poaceae**) (**Plate I 18**)

Grains in monads; 22.77 - 27.86 x 21.05 - 26.22  $\mu\text{m}$ ; polar and equatorial outline circular; monoporate; sculpturing psilate.

**Schleichera oleosa** (Lour.) Oken. (**Sapindaceae**) (**Plate I 19**)

Grains in monads; 23.42 x 19.15  $\mu\text{m}$ ; polar outline triangular-obtuse, convex to circular; equatorial outline elliptic; trizonoparasyncolporoidate; sculpturing striate-reticulate.

**Stachytarpheta jamaicensis** (L.) Vahl. (**Verbenaceae**) (**Plate I 20**)

Grains in monads; 25.47 - 26.40 x 25.85 - 26.13  $\mu\text{m}$ ; polar outline triangular, obtuse-convex; equatorial outline elliptic; trizonocolpate; sculpturing reticulate.

**Syzygium cumini** (L.) Skeels (**Myrtaceae**) (**Plate I 21**)

Grains in monads; Polar outline triangular, obtuse-convex/plane or circular; equatorial outline elliptic-obtuse-acuminate; trizonocolporate; sculpturing psilate.

**Syzygium Type I** (**Myrtaceae**) (**Plate I 22**)

Grains in monads. Polar outline triangular, obtuse-

convex-plane, 10.38 - 12.82 x 10.60 - 12.29  $\mu\text{m}$ ; trizonocolporate; equatorial view elliptic, obtuse, 11.50 x 7.90  $\mu\text{m}$ ; sculpturing psilate.

**Syzygium Type II** (**Myrtaceae**) (**Plate I 23**)

Grains in monads; polar outline triangular acute-straight-plane, radially symmetrical; 16.40 x 15.66  $\mu\text{m}$ , trizonocolporate; sculpturing psilate.

**Terminalia paniculata** Roth (**Combretaceae**) (**Plate I 24**)

Grains in monads; 11.25 - 13.97 x 13.95 - 14.81  $\mu\text{m}$ ; polar outline Triangular, obtuse-convex; isopolar, radially symmetrical; equatorial outline elliptic, trizonocolporate with pseudocolpi inbetween; sculpturing psilate.

**Tridax procumbens** L. (**Asteraceae**) (**Plate I 25**)

Pollen grains in monads; 5.37 - 12.53 x 5.55 - 12.09  $\mu\text{m}$ ; polar and equatorial view circular or oblate, spheroidal, radially symmetrical, isopolar, trizonocolporate; sculpturing echinate.

**Urena lobata** L. (**Malvaceae**) (**Plate I 26**)

Grains in monads; 28.31 x 27.60  $\mu\text{m}$ ; polar outline and equatorial outline circular or oblate-spheroidal, radially symmetrical, isopolar, pantoporate; sculpturing echinate.

**Verbenaceae Type** (**Plate I 31**)

Pollen grains in monads; 28.19 x 28.97  $\mu\text{m}$ ; polar outline rectangular-convex obtuse-plane, radially symmetrical, isopolar, tetrazonocolporate; sculpturing psilate.

**Pollen Type I**

Pollen grains in monads; 17.11 x 15.14  $\mu\text{m}$ ; triangular, Trizonocolporate; sculpturing reticulate.

**Pollen Type II**

Pollen Grains in monads; 19.43 x 15.57  $\mu\text{m}$ ; prolate, elliptic, trizonocolporate; radially symmetrical; sculpturing psilate.

**3.2 Analysis of the samples**

In sample S1, 29 pollen types belonging to 15 families were identified; in sample S2, 6 types belonging to 5 families, in sample S3, 34 pollen types belonging to 18 families and in sample S4, 4 types belonging to 3 families were recorded. In sample S5 no pollen grains were observed. The poor pollen representation or total absence may be due to excessive filtration or falsification (by feeding sugar to bees). Qualitative analysis of the honey samples were carried out by the standard method described (Erdtman, 1960). Pollen grains per sample were identified and placed in one of the frequency classes - predominant pollen type (> 45%), secondary pollen type (16 - 45%), important minor pollen types (3 -

14 %) and minor pollen types (< 3%). Honey sample containing more than 45 % of a single type of pollen were considered as unifloral honey.

Among the five samples one sample S2 collected from Kannur district were recorded as unifloral honey with *Cocos nucifera* belonging to Arecaceae as predominant pollen. The other 3 samples except sample S4 were recorded as multifloral. The sample S5 collected from market (as branded honey) showed the absence of pollen grain. The study revealed secondary pollen types belonging to *Camellia sinensis*, *Careya arborea*, *Mimosa pudica*, *Pennisetum polystachyon*, Pollen Type II, *Stachytarpheta jamaicensis*, *Syzygium* Type I, *Tridax procumbens* and *Urena lobata*. The important minor types include *Achyranthes aspera*, *Adenantha pavonia*, *Aporosa lindleyana*, *Cocos nucifera*, *Lagerstroemia speciosa*, *Stachytarpheta jamaicensis*, *Syzygium cumini*, *Syzygium* Type I & Type II, *Terminalia paniculata* and *Tridax procumbens*. The minor pollen types include *Acacia auriculiformis*, *Achyranthes aspera*, Acanthaceae Type, *Adenantha pavonia*, *Aporosa lindleyana*, *Areca catechu*, *Dillenia pentagyna*, *Hevea brasiliensis*, Asteraceae Type I & Type II, *Caesalpinia bonduc*, *Careya arborea*, *Cocos nucifera*, *Croton bonplandianum*, *Dendrophthoe falcata*, *Elaeocarpus serratus*, *Gomphrena celosioides*, *Helixanthera wallichiana*, *Hevea brasiliensis*, *Hyptis suaveolens*, *Lagerstroemia speciosa*, Lamiaceae Type, *Mimosa pudica*, *Passiflora foetida*, *Peltophorum pterocarpum*, Pollen Type II, *Schleichera oleosa*, *Syzygium cumini*, *Syzygium* Type I & Type II, *Terminalia paniculata*, *Tridax procumbens* and Verbenaceae Type.

The plant families that were present in majority of the honey samples include Arecaceae, Caesalpiniaceae, Malvaceae and Myrtaceae were found in almost all samples. Acanthaceae, Amaranthaceae, Combretaceae, Lamiaceae and Lythraceae were found in two samples. Dilleniaceae and Theaceae were found only in one

sample (S3). The pollen type *Syzygium* Type I appeared in almost all the samples except S5. *Cocos nucifera*, *Pennisetum polystachyon*, *Syzygium cumini* and *Urena lobata* are present in majority of the samples. The dominance of tree species is followed by herbs, shrubs and climbers in 2 samples S1 and S3. In sample S2 80 % is represented by climbers and 20% by shrubs. In sample S4 77% is represented by trees and 22% by herbs. The percentage frequency of the different vegetation types of identified taxa of bee forage importance is depicted in Figure 1.

In sample S1 (Cheru Then honey) the pollen count is found to be 2074 which contributes 58% of the total pollen grains and is the highest when compared to other samples. No predominant types were observed. The highest contribution of pollen type was *Pennisetum polystachyon* with 33.39% and *Acacia auriculiformis*, Acanthaceae Type, *Adenantha pavonia*, Asteraceae Type I, *Caesalpinia bonduc*, *Croton bonplandianum*, *Passiflora foetida* and Lamiaceae Type are the least types which is represented below 1%. The secondary dominant types include *Pennisetum polystachyon*. The important minor types include *Lagerstroemia speciosa*, *Stachytarpheta jamaicensis*, *Syzygium* Type I and Type II, *Terminalia paniculata* and *Urena lobata*. The minor pollen types include *Acacia auriculiformis*, Acanthaceae Type, *Achyranthes aspera*, *Adenantha pavonia*, *Aporosa lindleyana*, Asteraceae Type I, *Caesalpinia bonduc*, *Careya arborea*, *Cocos nucifera*, *Croton bonplandianum*, *Dendrophthoe falcata*, *Elaeocarpus serratus*, *Gomphrena celosioides*, *Helixanthera wallichiana*, Lamiaceae Type, *Mimosa pudica*, *Peltophorum pterocarpum*, *Schleichera oleosa*, *Syzygium cumini*, *Tridax procumbens* and Verbenaceae Type. The pollen types is represented in 15 families which includes Asteraceae, Arecaceae, Caesalpiniaceae, Combretaceae, Elaeocarpaceae, Euphorbiaceae, Lamiaceae, Loranthaceae, Lythraceae, Malvaceae, Mimosaceae,

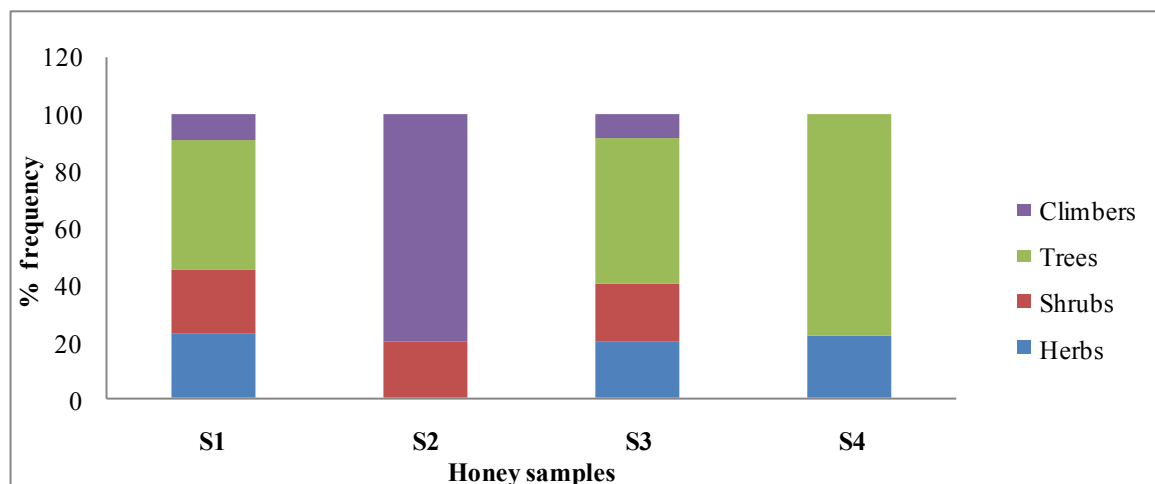


Figure I: % Composition of different vegetation types of identified taxa of bee forage importance

Table III: Comparison of pollen count in different samples

Sl. No.	Samples	Pollen Count
1	Cheru Then (S1)	2074
2	Van Then (S2)	11
3	Rubber Honey (S3)	825
4	Tea Honey (S4)	11
5	Dabur honey (S5)	Nil

Myrtaceae, Passifloraceae and Sapindaceae.

In sample S2 (Van Then), the total pollen count is 11 which are comparatively less contributing below 1% of the total grains. *Cocos nucifera* contribute 54.16% as predominant pollen. The secondary pollen type is represented by *Urena lobata* with 16.6%. The important minor type includes *Careya arborea*, *Lagerstroemia speciosa*, *Syzygium cumini* and *Syzygium* Type I. The pollen types are represented in 5 families which include Arecaceae, Caesalpiniaceae, Lythraceae, Malvaceae and Myrtaceae.

In sample S3 (Rubber honey), the total pollen count was 825 which contributes 23 % of the total number of grains. The secondary pollen type includes *Tridax procumbens* with 18.13% and *Pennisetum polystachyon* with 16.69%. The important minor type include *Achyranthes aspera*, *Adenantha pavonia*, *Aporosa lindleyana*, *Cocos nucifera*, Pollen type II, *Stachtarpheta jamaicensis* and *Urena lobata*. The minor types include *Acacia auriculiformis*, Acanthaceae Type, *Areca catechu*, *Caesalpinia bonduc*, *Croton bonplandianum*, *Dendrophthoe falcata*, *Dillenia pentagyna*, *Elaeocarpus serratus*, *Gomphrena celosioides*, *Helixanthera wallichiana*, *Hevea brasiliensis*, Lamiaceae Type, *Mimosa pudica*, *Passiflora foetida*, *Peltophorum pterocarpum*, Pollen Type I, *Schleichera oleosa*, *Syzygium cumini*, *Syzygium* Type I and Type II, *Terminalia paniculata* and Verbenaceae Type.

The pollen type is represented in 18 families which include Acanthaceae, Amaranthaceae, Arecaceae, Asteraceae, Caesalpiniaceae, Dilleniaceae, Elaeocarpaceae, Euphorbiaceae, Lamiaceae, Loranthaceae, Malvaceae, Mimosaceae, Sapindaceae and Verbenaceae and Poaceae.

In sample S4 (Tea honey) the pollen count was 11 which contributes below 1% of the total grains in samples. The pollen types observed were all secondary type with *Camellia sinensis*, *Pennisetum polystachyon*, Pollen Type I, *Syzygium* Type I. *Syzygium* Type I is represented by 33.3 % followed by *Camellia sinensis* (30%). In sample S5 (Dabur honey) the pollen type was totally absent.

#### 4. Discussion

The pollen spectrum of the samples shows a good source of nectar and pollen for bees and also for the development of bee keeping activities. The sample S1 collected from Wayanad showed 29 pollen types. Maximum number of pollen types was observed in S3 and the least number in S4. There is the dominance of tree species with more than 40% in S1, S3 and 77% in S4. The samples studied showed more number of trees and shrubs and these plants ensure sufficient flow of nectar and pollen for colony development.

With respect to total pollen count of all samples it is 2921. Maximum pollen count was found in sample S1 and S3 and the least pollen count was found in sample S2 and S4 and no pollen count was observed in S5. The comparison of pollen count in the analyzed honey samples showed a drastic variation (Table III). The qualitative comparison of honey sample (represented in %) is depicted in Table IV.

The pollen type *Syzygium* Type I is found in all samples except S5. *Cocos nucifera* showed dominance in all 3 samples and is considered as an important pollen and nectar sources to honey bees in the region. Twenty one pollen types were found in 3 samples and can be regarded as the key resources for bee keeping and honey production (Table 2). Six Pollen types were found in 2 samples and 4 pollen types were found in one sample and hence these species are considered as least important for bee forage (Table 2).

The pollen combination in honey samples indicates that there is a mixture of both deciduous and evergreen species. An endemic species of Western Ghats *Helixanthera wallichiana* belonging to Loranthaceae with 1.4% is represented in 2 samples S1, and S3. This shows that honey bees were foraging near the border zones of the forest. In S3 there is a maximum representation of pollen types. The presence of the wild species in the sample is due to the mixing of the sample from forest zone with the apiary sample (Maya C. Nair, 2005). The forest honey sample and market sample can be distinguished on the basis of pollen analysis and differs in having more number of pollen types and count and is found to be a better one for consumption. Bees help in pollination and enhance the crop productivity. Bees are species specific in resource preference and sharing. As per the analysis Bees prefer mostly tree species for nectar and pollen sources for honey production.

On the basis of the pollen types analysed, plants flowering at various seasons of the year can be assessed which indicates the exact season of honey collected (Nov. - March). These species need to be raised for dietary preference of bees for honey production. At times

**Table IV: Qualitative comparison of honey samples (represented in %)**

Sl No	Pollen types	S1	S2	S3	S4
1	<i>Acacia auriculiformis</i>	+		1.07	
2	<i>Acanthaceae type</i>	+		2.21	
3	<i>Achyranthes aspera</i>	1.37		8.84	
4	<i>Adenanthera pavonina</i>	+		4.9	
5	<i>Aporosa lindleyana</i>	1.35		8.56	
6	<i>Areca catechu</i>			+	
7	<i>Asteraceae type 1</i>	+		1.06	
8	<i>Asteraceae type 2</i>			1.01	
9	<i>Caesalpinia bonduc</i>	+		+	
10	<i>Camellia sinensis</i>				30
11	<i>Careya arborea</i>	+	4.16		
12	<i>Cocos nucifera</i>	1.15	54.16	3.15	
13	<i>Croton bonplandianum</i>	+		+	
14	<i>Dendrophthoe falcata</i>	1.23		+	
15	<i>Dillenia pentagyna</i>			1.24	
16	<i>Elaeocarpus serratus</i>	1.36		2.43	
17	<i>Gomphrena celosioides</i>	1		2	
18	<i>Helixanthera wallichiana</i>	1.47		+	
19	<i>Hevea braziliensis</i>			+	
20	<i>Lagerstroemia speciosa</i>	3.83	8.33	+	
21	<i>Lamiaceae type</i>	+		+	
22	<i>Mimosa pudica</i>	1.33		2.73	
23	<i>Passiflora foetida</i>	+		+	
24	<i>Peltoforam pierocarpum</i>	1.21		+	
25	<i>Pennisetum polystachyon</i>	33.3 9		16.69	18.33
26	<i>Schleichera oleosa</i>	2.99		+	
27	<i>Stachytarpheta jamaicensis</i>	11.2 5		4.46	
28	<i>Syzygium cumini</i>	2.17	4.16	1.99	
29	<i>Syzygium type1</i>	9.78	12.5	1.4	33.33
30	<i>Syzygium type 2</i>	3.45		2.76	
31	<i>Terminalia paniculata</i>	5.75		1.61	
32	<i>Tridax procumbens</i>	1.96		18.13	
33	<i>Urena lobata</i>	5.06	16.6	4.17	
34	<i>Verbenaceae type</i>	+		+	
35	<i>Pollen type 1</i>			+	
36	<i>Pollen type 2</i>			6.43	18.33

“+” - Pollen type present below 1%

of scarcity they depend on cultivated taxa like *Areca catechu*, *Cocos nucifera* etc. which flowers throughout the year.

The pollen Type I and Pollen Type II were unknown. The pollen Type I is found in Sample S4 as minor type with 1.06 % frequency. The pollen type II is found in S3

as minor type with 6.4% and in S4 as Secondary type with 18.3%. This can be identified with the help of reference slides prepared from the region. The geographical and botanical origin of the honey samples can be determined by melissopalynological analysis. But there are some limitations for the identification of botanical source of honey. If the honey is subjected to excessive filtration through diatomaceous earth or other similar process the botanical aspects cannot be determined.

Adulteration may be due to contamination of wind pollinated pollen, mixing of forest honey and apiarist honey. When during famine the bees depend on ripe fruits for forage, High fraction Corn Syrup is commonly used as an adulterant (Kashinath Bhattacharya, 2006). The extraction of the bark of *Pterocarpum marsupium* is added to honey to enhance the color.

### 5. Conclusion

Melissopalynological analysis helps to determine the botanical characteristics of honey, geographical origin, pollen diversity and frequency, pollen preference of bee forage and adulteration if any. It also provides a useful guide to bee keeping of the region. Pollen analysis of honey samples indicated the presence of 36 different plant species, of which *Syzygium* Type I were present in samples S1, S2, S3, & S4. 21 pollen types were frequent in 2 samples (S1, and S3), 6 types were of least occurrence in 2 samples and 4 types were found only in one sample. Of the five samples analysed, one sample S2 was recorded as unifloral with *Cocos nucifera* as predominant pollen having 54.16% and the samples S1, S3 and S4 were recorded as multifloral. There was a total absence of pollen in S5. *Cocos nucifera*, *Syzygium cumini*, *Syzygium* Type I, and *Urena lobata* was represented in almost all the samples studied and is apparent that these species are important bee plant species for pollen and nectar sources. The predominant and secondary dominant type represented offer better resources for honey production. Based on the frequency of pollen grains, the degree of purity can be assessed. The quality and quantity of honey are related to plant species specificity and the vegetation pattern around the bee colonies. Honey collected from monoculture plantation or from the apiarist showed that there is a mixture of native, wild and cultivated taxa. Even though the Western Ghats region is with more representation of flowering plants, bees are species specific and hence the apiarist should go for cultivation of those species preferred by bees which is the best performer in relation to pollination service and thus helps in conservation of biodiversity. Pollen analysis in honey led to the identification of vegetation in the area which is important for the survival of bee colonies.



### Acknowledgement

The authors are thankful to the Dr. V. Jayachandran, former Principal, MGGA College, Mahe, for encouragement; Prof. C.P. Ravindran, Head, P.G. Dept. of Plant Science for facilities; Dr. G.V.S. Murthy, the Addl. Director, Botanical Survey of India, Southern Regional Centre, Coimbatore, for providing facilities for the microphotography of the pollen grains and valuable suggestions; the timely help rendered by the colleagues are also gratefully acknowledged.

### Conflict of interest

The author's declares none.

### References

- Ashoke Bhattacharya. (2014). Melissopalynological study of Noida district, West Bengal, India *International J. of current Research* 6(95): 6521 - 6526.
- International Commission for Bee Botany (ICBB). (1970). Methods of Melissopalynology, *Bee World* 51: 125 - 138.
- Deodikar, G.B and Thakar, C. V. (1955). A pollen study of major honey yielding plants of Mahabaleshwar hills. *Apicultural Laboratory, Mahabaleshwar, Bull.* 1: 1 - 6.
- Deodikar, G.B and Thakar, C. V. (1966). Utilization and improvement of local floras as bee pasturage. *Proc. Autumn School Bot. Mahabaleshwar.* Dept. Bot., Poona Univ. 122 - 128.
- Deodikar, G.B and Suryanarayana, M. C. (1973). Bees enhance crop yields. *Lnd. farming.* 23: 31 - 36.
- Erdtman, G. (1960). The Acetolysis method - A revised description. *Sven. Bot. Tidskr.* 54: 561 - 564.
- Kashinath Bhattacharya, Manas Ranjan Majumdar and Swati Gupta Bhattacharya. (2006). *A Text Book of Palynology.* New Central Book Agency, Calcutta.
- Louveaux, J., Maurizio, A. and Vorwohl, G. (1978). Methods of Melissopalynology. *Bee World* 59 (4): 139 - 157.
- Maya, C. Nair. (2005). Palynological identification of resources for development of Apiculture in Kerala - A case study *J. Palynology* 41: 115 - 138.
- Punt, W., Hoen, P. P., Blackmore, S., Nilsson, S and Thomas, A. (2007). Glossary of Pollen & Spore Terminology. *Review of Paleobotany and Palynology* 143(1-2): 1 - 81.
- Sharma, M. and Nair, P K.K. (1965). Pollen analysis of some honeys from Uttar Pradesh. *Ind. J. Hor.* 22(1): 46 - 51.
- Sivaram, V., Roopa, P. Shubharani, R. Guntimasuwannapong. (2012). Pollen analysis in honeys collected from Karnataka region of Nilgiri Biosphere, South India. *J. of Apiculture.* 27(3): 223 -231.
- Suryanarayana, M. C., Seethalakshmi, T. S. and Phadke, R. P. (1981). Pollen analysis of Indian honeys- 1. Honeys from Litchi (*Nephelium litchi*) and Jamun (*Syzygium cumini*), *IV Palynol. Conf., Lucknow:* 491 - 498.