



Pollen Analysis of Honeys from Varanasi District, Uttar Pradesh, India

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ABSTRACT: Pollen analysis of six honey samples of *Apis dorsata* collected from Varanasi district has been carried out. Altogether, 37 pollen types were identified. Two honey samples were unifloral and four were multifloral in nature. *Brassica campestris* and *Ageratum conyzoides* were the predominant pollen types in unifloral honeys. In multifloral honey *Brassica campestris*, *Ageratum conyzoides*, *Callistemon citrinus*, *Parthenium hysterophorus*, *Holoptelea integrifolia* and *Lathyrus aphaca* were the secondary pollen types. In total honey samples *Brassica campestris*, *Ageratum conyzoides*, *Callistemon citrinus*, *Coriandrum sativum*, *Eucalyptus globulus*, *Carica papaya*, *Citrus* sp., *Crotalaria juncea* and Poaceae were very frequent in occurrence. Majority of pollen grains recovered from honey samples belonged to entomophilous taxa (54.05%) and amphiphilous taxa (32.48%) while 13.15% pollen were from anemophilous taxa. Present study suggests that *Brassica campestris*, *Ageratum conyzoides*, *Callistemon citrinus* and *Coriandrum sativum* can be regarded as the major bee source plants of the region.

Key words: Pollen analysis, unifloral, multifloral, Varanasi district, Uttar Pradesh

INTRODUCTION

Honey is produced by mutual interaction between bees and nectariferous plants. While foraging honey bees also collect pollen grains along with nectar. Pollen analysis of a honey sample provides information about the plants visited by bees giving relevant information about the nectar and pollen sources of an area and helps to determine geographical and botanical origin of honey (Louveaux *et al.*, 1998; Von Der Ohe *et al.*, 2004; Barth, 2004). Knowledge of botanical sources of honey is prerequisite to beekeepers to stabilize an apiary industry for commercial honey production. In India, investigations on pollen analysis of honey samples are available from various parts of the country viz. Maharashtra (Deodiker & Thaker 1953; Deodiker *et al.*, 1958), Andhra Pradesh (Ramanujam & Kalpana, 1991; Lakshmi & Suryanarayana, 2004; Ramakrisna & Swati, 2013; Devender *et al.*, 2015), Bihar (Suryanarayan *et al.*, 1992), Himanchal Pradesh (Sharma, 1970; Sharma & Raj, 1985), West Bengal (Bhattacharya *et al.*, 1983; Chakraborti and Bhattacharya, 2011; Kamble *et al.*, 2015), Uttarakhand (Garg & Nayer, 1974), Karnataka (Agashe and Rangaswami, 1997; Chauhan & Murthy,

2010; Shubharani *et al.*, 2012; Raghunathan *et al.*, 2013), Orissa (Upadhyay & Bera, 2008, 2012, 2014) and Madhya Pradesh (Chauhan & Quamar, 2010; Sahney & Seth, 2013). However, from Uttar Pradesh melissopalynological reports are available from Lucknow (Sharma & Nair, 1965, Chaturvedi & Sharma, 1973), Shahjahanpur (Chandra and Sharma, 2011) and Allahabad (Sahney & Rahi, 2015) only. Thus, the present investigation is undertaken on the honey samples collected from Varanasi district of Uttar Pradesh to identify source plants of the region for honey production. The paper presents first report on the pollen analysis of honeys from Varanasi.

MATERIALS AND METHODS

A. Study area and collection of materials

Varanasi (latitude-25.3176 & longitude-82.8739) is a religious city situated on the banks of Ganges in Eastern Uttar Pradesh, India. Covering an area 1535 Sq. Km Varanasi is located at an elevation of 80.71 meters. Six squeezed honey samples of *Apis dorsata* were collected during January to March 2014 from six different localities viz. Manduadih (V1), Shivpur (V2),

Chiraiyaon (V3), Lalpur (V4), Danganj (V5) and Goverdhanpur (V6) of Varanasi district (Fig. 1). Samples were filtered with cloth to remove extraneous matter like bee wax pieces, scum and surface dirt and stored in airtight plastic bottles and labeled. Several

field trips were made to survey plants around the honey collection sites to collect the floral material for preparation of reference slides. Reference slides of pollen grains were made following Wodehouse method (1935).

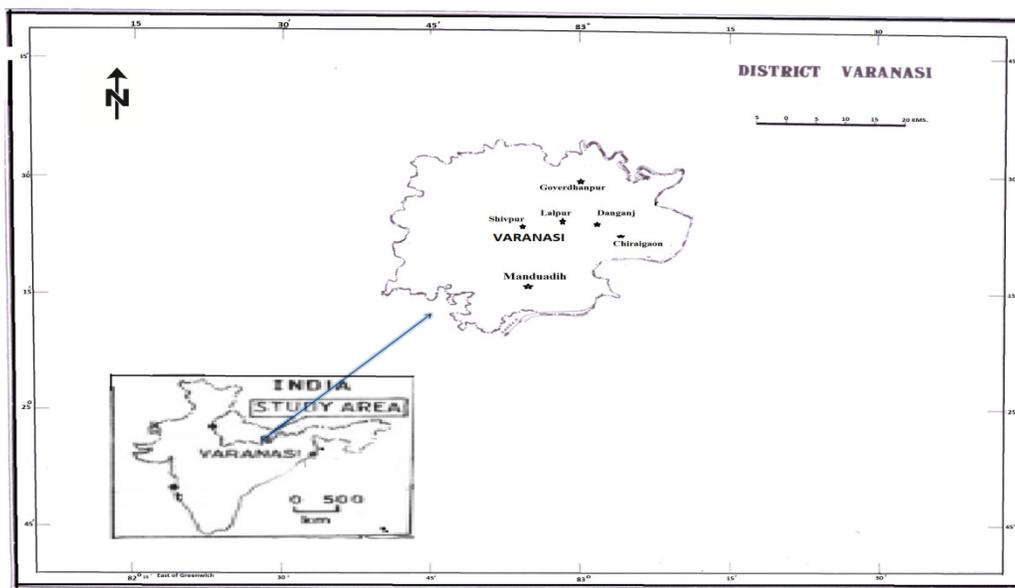


Fig.1. Map showing localities of Varanasi district from where honey samples were collected.

B. Pollen analysis

Methods recommended by International Commission of Bee Botany (Louveaux *et al.*, 1978) were followed to prepare honey slides for the recovery, analysis and quantification of pollen grains. From each honey sample five slides were prepared and scanned thoroughly under the microscope. From each slide 300 pollen grains were counted at random for determining frequency classes *viz.*, Predominant (>45% of the counted pollen grains), Secondary (16-45%), Important minor (3-15%), and Minor (<3%). Identification of pollen grains recovered from honey samples was made with the help of reference slides and relevant literature (Ertzman, 1952; Bhattacharya *et al.*, 2006). Honey sample with single predominant pollen type has been categorized as “Unifloral” and the sample with no predominant pollen type as “multifloral”. Based on frequency of occurrence in total honey samples pollen types have been classified as very frequent (pollen type present in >50% of the samples), frequent (20-50%), infrequent (10-20%) and rare (<10%) (Feller-Demalsy *et al.*, 1987). The absolute pollen count (APC) of honey sample was determined using a haemocytometer as suggested by Suryanarayana *et al.* (1981).

RESULTS AND DISCUSSIONS

Altogether 37 pollen types belonging to 23 families were identified from the microscopic examination of six honey sample collected from six localities of Varanasi district (Plate 1). Pollen types, their frequency classes, absolute pollen count and pollen diversity in the honey sample are presented in Table 1.

Out of 6 honey samples two were found to be unifloral and four multifloral in nature. *Brassica campestris* and *Ageratum conyzoides* were registered as predominant types. Secondary pollen types were represented by *Callistemon citrinus*, *Brassica campestris*, *Coriandrum sativum*, *Parthenium hysterophorus*, *Ageratum conyzoides*, *Holoptelea integrifolia* and *Lathyrus aphaca* while pollen grains of *Benincasa hispida*, *Moringa oleifera*, *Phyllanthus emblica*, *Prosopis juliflora*, *Polygonum aviculare*, *Coriandrum sativum*, *Carica papaya*, *Ageratum conyzoides*, *Eucalyptus globulus*, *Roystonea regia*, *Crotalaria juncea*, *Cajanus cajan* and Poaceae were recorded in important minor frequency class.

Table 1: Pollen types, frequency classes, APC and pollen diversity in honey samples collected from Varanasi district.

Pollen types	Sample code					
	V1	V2	V3	V4	V5	V6
<i>Ageratum conyzoides</i>	M	M	I	P	S	S
<i>Amaranthus caudatus</i>						M
<i>Anthocephalus cadamba</i>						M
<i>Azadirachta indica</i>		M				
<i>Benincasa hispida</i>		I				
<i>Bombax ceiba</i>	M	M				
<i>Brassica campestris</i>	S	M	P	M	I	I
<i>Cajanus cajan</i>					M	I
<i>Callistemon citrinus</i>	S	S			I	I
<i>Carica papaya</i>			I	M	I	I
<i>Cassia nictitans</i>		M				
<i>Cassia sp.</i>	M		M			
<i>Citrus limetta</i>					M	
<i>Citrus sp.</i>			M			M
<i>Coriandrum sativum</i>	S	I	I	M	M	I
<i>Coronopus didymus</i>	M		I			
<i>Crotolaria juncea</i>				M	M	M
<i>Eucalyptus globulus</i>	M	M	I	M		
<i>Holoptelea integrifolia</i>					S	
<i>Ixora pavetta</i>		M				
<i>Justicia procumbens</i>		M				
<i>Lathyrus aphaca</i>		S				
<i>Lathyrus odoratus</i>		S			M	
<i>Madhuca indica</i>				M		
<i>Moringa oleifera</i>				I		
<i>Parthenium hysterophorus</i>	M	S				
<i>Phoenix acaulis</i>	M			M		
<i>Phyllanthus emblica</i>		I		M		
<i>Pisum sativum</i>	M					I
Poaceae	M			M	I	I
<i>Polygonum aviculare</i>		M				
<i>Prosopis juliflora</i>		I				
<i>Roystonea regia</i>			I			
<i>Rumex petientia</i>					M	
<i>Tinospora cordifolia</i>					M	
<i>Wedelia sphagneticola</i>						M
<i>Ziziphus mauritiana</i>					M	
Pollen diversity	12	16	10	11	15	14
APC	Gp-III	Gp-IV	Gp-III	Gp-IV	Gp-II	Gp-IV

26 pollen types registered their presence in minor frequency class viz. *Brassica campestris*, *Eucalyptus globulus*, *Amaranthus caudatus*, *Callistemon citrinus*, *Coronopus didymus*, *Parthenium hysterophorus*, *Cassia* sp., *Phoenix acaulis*, *Azadirachta indica*, *Ixora pavetta*, *Justicia procumbens*, *Phyllanthus emblica*, *Polygonum aviculare*, *Cassia nictitans*, *Carica papaya*, *Lathyrus odoratus*, *Citrus* sp., *Crotalaria juncea*, *Madhuca indica*, *Holoptelea integrifolia*, *Cajanus cajan*, *Citrus limetta*, *Tinospora cordifolia*, *Rumex petientia*, *Ziziphus mauritiana* and *Anthocephalus cadamba*.

With regard to frequency of occurrence of pollen types in total honey samples *Brassica campestris*, *Coriandrum sativum*, *Ageratum conyzoides*, *Callistemon citrinus*, *Eucalyptus globulus*, *Carica papaya*, *Citrus* sp., *Crotalaria juncea* and Poaceae were very frequent in occurrence as they were recovered from more than 50% of the samples. 11 pollen types were recorded as frequent type and rest 16 pollen types were infrequent in occurrence (Fig. 2). With regard to absolute pollen count one sample (V5) belonged to Gp II, two samples (V1 & V3) to Gp III and three samples (V2, V4, V6) to Gp IV.

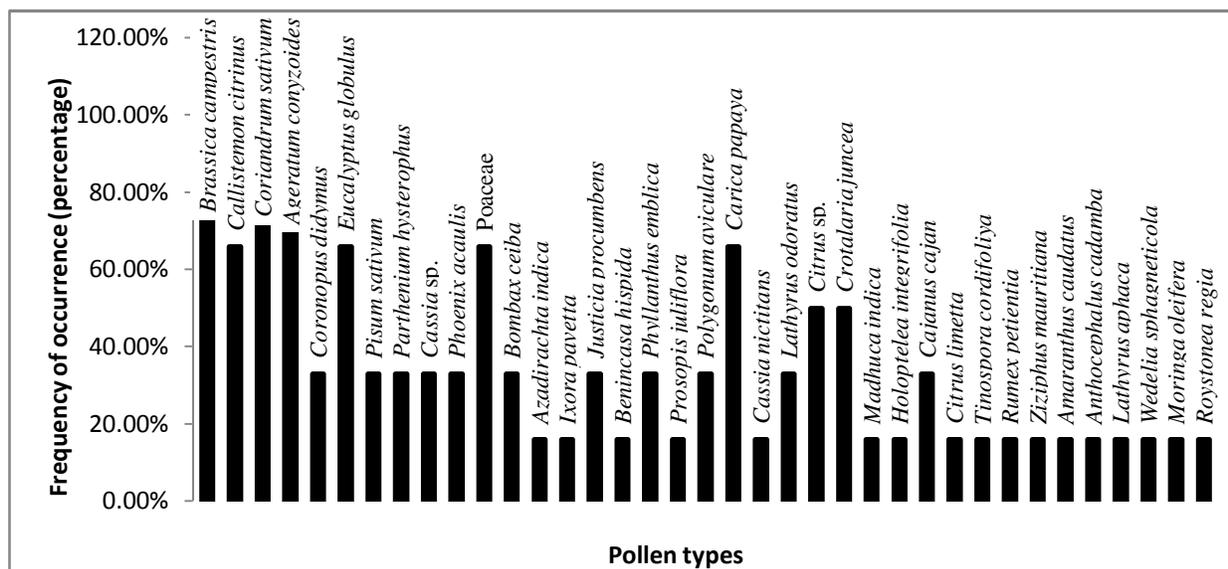


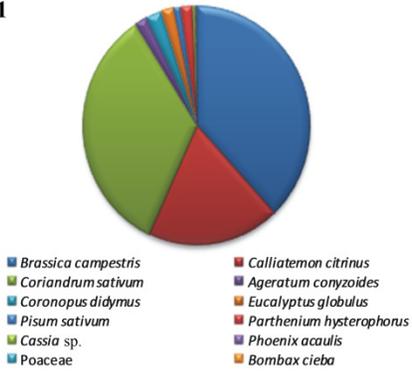
Fig. 2. Frequency of occurrence of pollen types in the honey samples.

Pollen spectra of six honey samples collected from different localities of Varanasi district exhibited diversity in their pollen composition (Fig. 3). Altogether 37 pollen types were recovered from honey samples. Diversity of pollen types ranged from 10-16. Maximum diversity of pollen types was recorded in honey samples V5 (15 pollen types) & V2 (16 pollen types). Both the samples were multifloral in nature. In sample V5 (Danganj) *Ageratum conyzoides* & *Holoptelea integrifolia* were present as secondary pollen types with 34.23% & 35.69% frequency respectively while in sample V2 (Shivpur) *Callistemon citrinus*, *Parthenium hysterophorus* & *Lathyrus aphaca* were present as secondary pollen types with 25%, 21.61% & 23.09% frequency respectively. Both the localities Shivpur (V2) & Danganj (V5) are at the outskirts of Varanasi city.

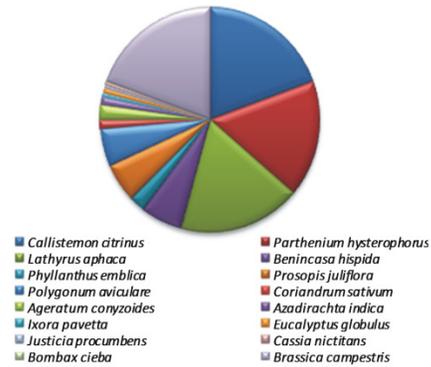
Surveys around the sites of honey collection revealed *Callistemon citrinus*, *Parthenium hysterophorus* and *Lathyrus aphaca* in Shivpur and *Holoptelea integrifolia* & *Ageratum conyzoides* in Danganj (V5) were in bloom.

Minimum diversity of pollen was recorded in samples V3 (10 pollen types) & V4 (11 pollen types). Both the samples were unifloral in nature. In sample no V3 *Brassica campestris* was the predominant pollen type with 50.44% frequency while in Sample V4 *Ageratum conyzoides* was predominant with 80% frequency. Further in both the samples no secondary pollen type was registered. Blooming crops of *Brassica campestris* in Chirraigaon (site of V3 honey sample) and abundance of blooming *Ageratum conyzoides* in Lalpur (site of V4 honey sample) served as good source of pollen /nectar to the bees for honey production.

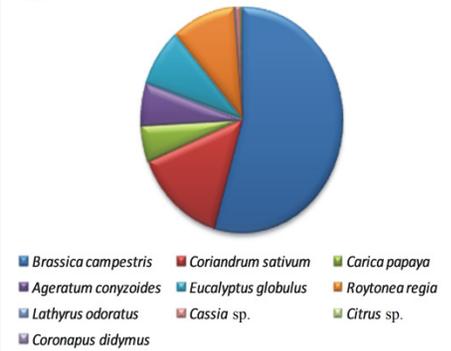
V1



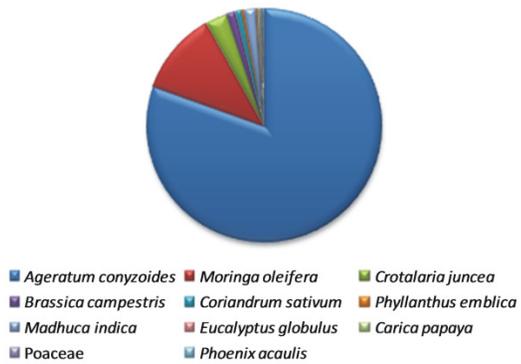
V2



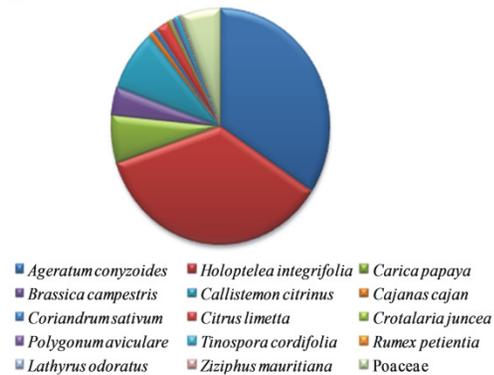
V3



V4



V5



V6

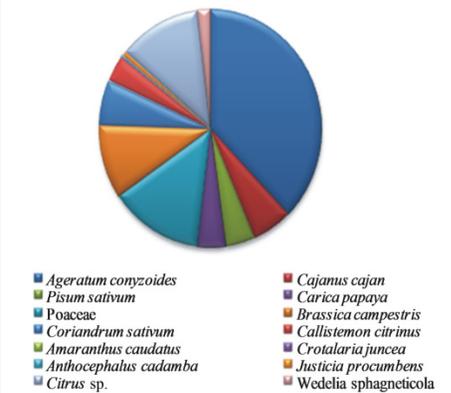


Fig. 3. Pollen spectra of honey samples.

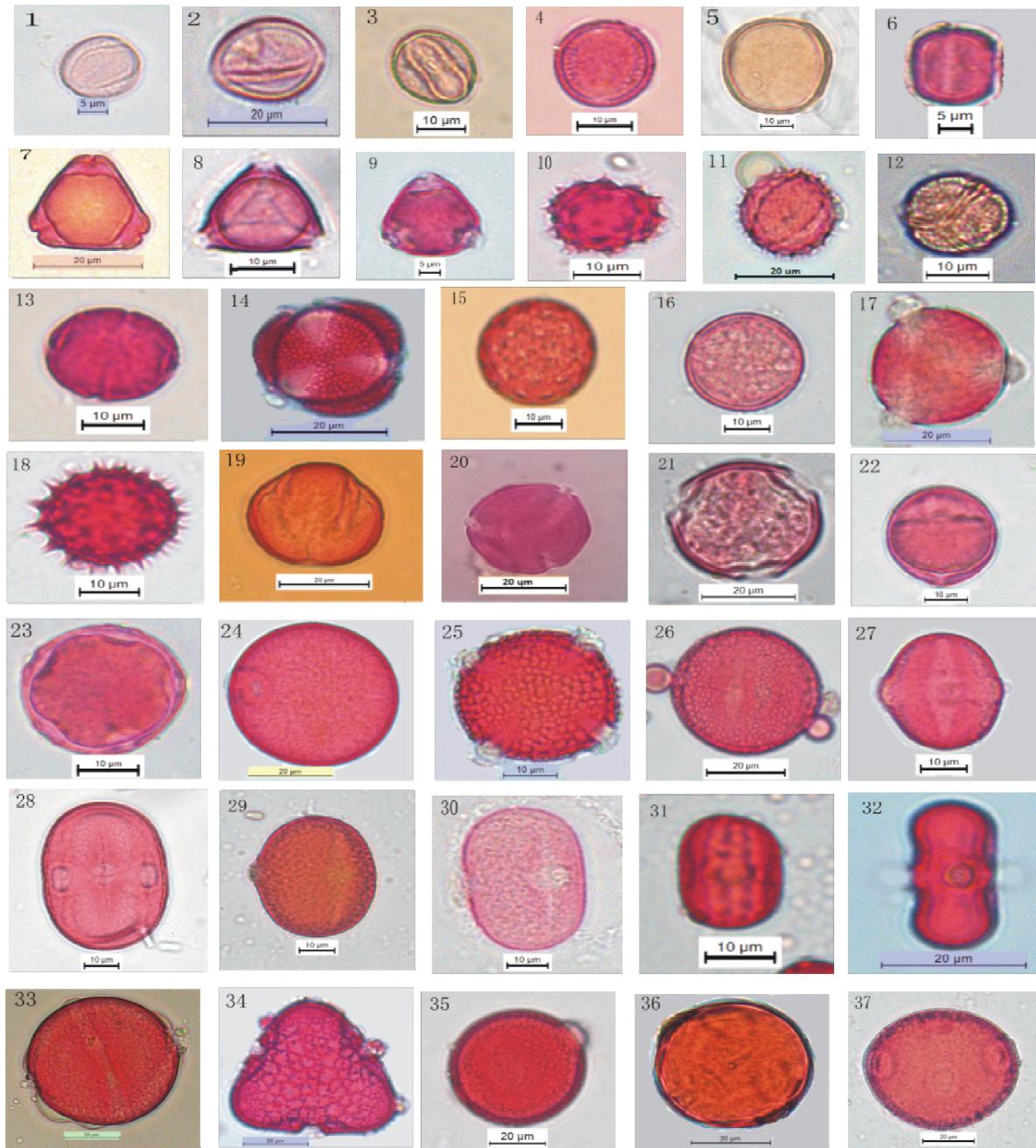


Plate I. Pollen types recovered from the honey samples: 1. *Anthocephalus cadamba* 2. *Phoenix acaulis* 3. *Ixora pavetta* 4. *Coronopus didymus* 5. *Roystonea regia* 6. *Polygonum aviculare* 7. *Eucalyptus globulus* 8. *Callistemon citrinus* 9. *Ziziphus mauritiana* 10. *Parthenium hysterophorus* 11. *Ageratum conyzoides* 12. *Tinospora cordifolia* 13. *Phyllanthus emblica* 14. *Brassica campestris* 15. *Amaranthus caudatus* 16. *Rumex petientia* 17. *Prosopis juliflora* 18. *Wedelia sphagneticola* 19. *Cassia* sp. 20. *Cassia nictitans* 21. *Moringa oleifera* 22. *Carica papaya* 23. *Holoptelea integrifolia* 24. Poaceae 25. *Citrus limetta* 26. *Citrus* sp. 27. *Crotalaria juncea* 28. *Lathyrus odoratus* 29. *Lathyrus aphaca* 30. *Pisum sativum* 31. *Justicia procumbens* 32. *Coriandrum sativum* 33. *Azadirachta indica* 34. *Bombax ceiba* 35. *Cajanus cajan* 36. *Madhuca indica* 37. *Benincasa hispida*.

Majority of pollen grains recovered from honey samples belonged to entomophilous taxa (54.05%) and amphiphilous taxa (32.48%) while 13.15% pollen were from anemophilous taxa. Among entomophilous taxa *Callistemon citrinus*, *Ageratum conyzoides*, *Parthenium hysterophorus* & *Lathyrus aphaca* and among amphiphilous, *Coriandrum sativum* & *Brassica campestris* were represented in predominant/secondary frequency class in the honey samples. While out of five anemophilous taxa viz. *Holoptelea integrifolia*, Poaceae, *Polygonum aviculare*, *Rumex petientia* and *Amaranthus caudatus* only *Holoptelea integrifolia* was recorded in secondary frequency class. High representation of pollen grains of *Holoptelea integrifolia* in the honey sample -V5 which was collected during March may be correlated with its flowering period (February-March) and its high pollen production capacity (Sahney and Chaurasia, 2008). Thus it is very possible that the abundant pollen grains present in the atmosphere may get into honey comb through wind current as stated by Sahney & Rahi (2015).

Based on findings of present melissopalynological investigation *Brassica campestris*, *Coriandrum sativum*, *Ageratum conyzoides* and *Callistemon citrinus* can be regarded as major bee source plants in the investigated localities of the Varanasi region as their pollen have been recorded in predominant/secondary frequency classes and are also very frequent in occurrence in the honey samples.

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