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Pollen spectrum of honey of *Apis mellifera* L. and stingless bees (Hymenoptera: Apidae) from the semi-arid region of Bahia State, Brazil

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Abstract
Pollen in honey reflects its botanical origin and melissopalynology is used to identify origin, type, and quantities of pollen grains of the botanical species visited by bees. This study aimed to identify the pollen spectrum of honeys from *Apis mellifera* and stingless bees produced in the semi-arid region of Bahia, Brazil. We analysed 78 honey samples, which were submitted to the acetolysis process for identification and quantification of pollen types. Fabaceae, Asteraceae and Euphorbiaceae were the most predominant families in pollen types. For Fabaceae, the most representative pollen types were *Chamaecrista* 1, *Mimosa caesalpiniifolia, Mimosa pudica, Mimosa tenuiflora, Prosopis* and *Senna*. The results indicate that the flora explored by the bees to collect nectar is diverse in the semi-arid region of Bahia and the honeys analysed were classified as multifloral.

Keywords: *melissopalynology, beekeeping, melliferous flora, pollen spectrum*

The semi-arid region of Bahia, the Caatinga biome, is known for its characteristic botanical species adapted to dry, hot climatic conditions with rainfall unevenly distributed throughout the year (Velloso et al. 2002; Sousa et al. 2016a). It also contains a diversity of bees that favour honey production.

Drought is one of the main characteristics of the semi-arid region of the Brazilian northeast. This region has an average annual precipitation between 250 and 500 mm, and its vegetation is mainly formed by shrubs (Cirilo 2008). This region covers approximately 1 219 000 km^2 (Cirilo 2008), highlighting the importance and the need to understand the uniqueness of beehive products obtained from this area. Pagano and Araújo (2010) reported on the diversity of the native vegetation of the Brazilian north-eastern semi-arid region. However, the pollen spectrum of beehive products of stingless bees (*Melipona quadrifasciata anthidioides* Lepeletier, 1836, *Melipona scutellaris* Latreille, 1811, *Tetragonisca angustula* Latreille, 1811) and Africanised bees (*Apis mellifera*, Linnaeus, 1758) of this region commonly displays a large number of representatives of Asteraceae, Fabaceae, Rubiaceae and Myrtaceae (Novais et al. 2013, 2015; Costa et al. 2015; Oliveira et al. 2016).

The pollen analyses of beehive products from the semi-arid region of Bahia show a major predominance for pollen stored by the bees (bee pollen) (Novais et al. 2009; Oliveira et al. 2016; Carneiro Neto et al. 2017). However, honey is the best known and most consumed product of bees (Tsutsumi & Oishi 2010). Thus, the honey produced in the semi-arid region may present an attractive sensory profile

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to consumers and these characteristics are directly related to the flora visited by the bee to collect nectariferous resources. Thus, the identification of floral origins explored by bees is relevant to characterise and value beehive products for the market.

When bees visit flowers to collect nectar, they adhere body pollen grains that mix with nectar at regurgitation (Nogueira-Neto 1997; Santana et al. 2011). Pollen in honey reflects its botanical origin as each botanical species has flowers with a specific pollen type, allowing identification of the floral source of honey nectar. The floral source that bees use in producing honey can be identified by melissopalynology, a useful tool to identify the presence, absence, type and amount of pollen of certain botanical species that bees use, indicating their potential for bee rearing (Louveaux et al. 1978; Silva & Santos 2014).

The determination of the botanical origin is used to label honey. The knowledge of the geographic origin are factors that influence considerably the commercial value of the product and can still be used as a screening tool for quality control (Barth et al. 2013; Corvucci et al. 2015). Nevertheless, despite its importance, little information is available on honey from stingless bees of the Bahia semi-arid. Therefore, our study aimed to identify the pollen spectrum of *Apis mellifera* honeys and stingless bees (Meliponinae) produced in the semi-arid region of the state of Bahia, Brazil.

Material and methods

Study site and sampling

Samples were collected in ten municipalities in the semi-arid region of Bahia, Brazil (Figure 1). The semi-arid region covers more than 50% of Bahia State, presenting a highly diversified flora, as recorded by Ramalho et al. (2009). Honeys were harvested from eight bee species, totalling 78 samples, composed of approximately 250 g, in one year of sampling (Table I). Samples were collected in each municipality according to bee species and the availability of honey produced in the period (Table I).

Pollen analysis

The methodology described by Louveaux et al. (1978), following modifications proposed by Jones and Bryant Junior (2004) was used as the basis for preparation of the samples and later submitted to the acetolysis process of Erdtman (1960). The pollen sediment resulting from the acetolysis process was used to prepare slides with glycerine gelatine for identification and counting of pollen grains that comprise the pollen spectrum of the sample.

Figure 1. Geographic location of the study site, where samples of honey samples were collected in the semi-arid region of Bahia, Brazil.

The pollen types were identified using specialised literature such as Barth (1989), Roubik and Moreno (1991), Punt et al. (2007), Lorente et al. (2017), Ybert et al. (2017, 2018) and consulting database and images of the Palinoteca of the Federal University of the Recôncavo of Bahia (Insect Study Group/Insecta). For that purpose, images of each pollen type were captured per sample using an Olympus® optical microscope (CX41) fitted with an Olympus® digital camera (E-330).

To determine the frequency classes of the pollen types in the honey samples, the consecutive counting of up to 1000 pollen grains/sample was performed according to Louveaux et al. (1978), classified as: predominant pollen – PP (> 45% of total grains), secondary pollen – SP (16 to 45%), important minor pollen – IMP (3 to 15%) and minor pollen – MP (< 3%). Additionally, we calculated the frequency of occurrence of each pollen type in the samples using the equation: $FO = (ni/N) \times 100$, where FO is the frequency of occurrence of the pollen type in the sample, ni is the number of pollen grains of the pollen type counted in the sample; N is the total number of pollen grains counted in the sample. According to Feller-Demalsy et al. (1987) we established the frequency classes: > 50% of samples, very frequent (VF); 20–50%, frequent (F); 10–20% infrequent (I); < 10%, rare (R).

Multivariate statistical analysis

The principal component analysis (PCA) and biplot were used to identify the association and prevalence of pollen types and bees species. The cluster analysis

Table I. Sampling of honey from the semi-arid region of Bahia, Brazil.

Species of bee	Number samples	Sampling site (municipality)
Apis mellifera Linnaeus, 1758	(n = 7)	Canarana Itaberaba Rui Barbosa
Melipona asilvai Moure, 1971	(n = 8)	Itaberaba Jeremoabo São Gabriel
Melipona mandacaia Smith, 1863	(n = 24)	Uibaí João Dourado São Gabriel
Melipona quadrifasciata anthidioides Lepeletier, 1836	(n = 12)	São Gabriel Tanque Novo
Melipona quadrifasciata Lepeletier, 1836	(n = 5)	Jeremoabo
Melipona subnitida Ducke, 1910	(n = 12)	Paulo Afonso
Scaptotrigona sp.	(n = 6)	Canarana UibaíRui Barbosa
Tetragonisca angustula Latreille, 1811	(n = 4)	Rui Barbosa Caetité Canarana

was carried out by the unweighted pair group method with arithmetic mean (UPGMA) using Palaeontological Statistics Software (PAST) v.1.34.

The cophenetic correlation coefficient (CCC) was calculated based on the similarity matrix and the dendrogram. To evaluate the adhesion between the dendrogram and the similarity matrix of Jaccard, the cophenetic correlation of Mantel was estimated using the PAST v.1.34 (Hammer et al. 2001).

Results

In the pollen spectrum of the honeys analysed, we identified 64 pollen types distributed in 26 families and two pollen types were not possible to be identified. The indeterminate pollen types occurred as minor pollen (MP < 3%) in the samples (Table II). The richest family in pollen types was Fabaceae with 35.94% of the total identified types, followed by Asteraceae (7.81%) and Euphorbiaceae (6.25%), (Figure 2). For Fabaceae, the most representative (FO > 50% of samples) pollen types in the samples were *Mimosa caesalpiniifolia*, *Mimosa tenuiflora* and *Prosopis* (Table II). The most frequent (FO General > 75%) pollen types in the samples were *Acacia*, *Borreria*, *Mimosa caesalpiniifolia*, *Mimosa tenuiflora, Prosopis*, *Psidium*, *Schinus*, *Solanum* and Rubiaceae type, among which, only *Prosopis* types was identified in the honey pollen spectrum of all bees (Figures 3, 4, Table II).

The pollen type *Mimosa tenuiflora* was found in all samples of *Melipona quadrifasciata anthidioides* and *Melipona quadrifasciata* with frequency also above 70% (very frequent) in the samples of *Melipona asilvai*, *Melipona mandacaia* and *Melipona subnitida*. Additionally, *Mimosa tenuiflora* was observed in the honey of these species of stingless bees as predominant pollen and/or secondary pollen, except for *Melipona quadrifasciata*. In the honey evaluated, there were 11 pollen types as predominant pollen and 22 as secondary pollen (Table III).

The pollen type *Mimosa caesalpiniifolia* was frequent in most honeys of the bee species studied, more frequent in honeys of *Melipona quadrifasciata anthidioides* and *Apis mellifera*. For *Melipona mandacaia*, the *Waltheria* type was present in 100% of the samples of honey (Table II).

The pollen types *Chamaecrista* (1, 2, 3) and *Croton* (1, 2, 3) were separated in three different types because they were identified in samples from different municipalities. Pollen morphology studies show that for *Chamaecrista* Moench. and *Croton* L. pollen size variation is elevated in the same specimen and is not indicative of different species (Carreira et al. 1996; Lima et al. 2007; Nascimento & Del-Claro 2007).

Multivariate statistical analysis

The PCA showed that the first three main components were sufficient to explain 72.8% of the total variation. The first PCA explained 37.3% of this variation, which can be verified in the biplot graph that shows that *Mimosa tenuiflora* was the pollen type that most contributed to the first PCA (Figure 5). We also observed four groups with cluster analysis (Figure 6).

Discussion

In our study, Fabaceae had the highest number of pollen types. Species of this taxonomic group are described as suppliers of trophic resources (nectar and pollen) for bees. In addition, this family is considered the most abundant in the studied region (Alves et al. 2016; Matos & Santos 2016).

The high representation of pollen grains from the Fabaceae in honey samples is because this family is well represented in the Brazilian semi-arid region, with the largest number of species in the Caatinga and Cerrado (Queiroz et al. 2006). Another family also considered important and found in the honeybee pollen spectrum was Asteraceae. Generally reported as potential bee plants species of genera *Baccharis*, *Bidens*, *Centratherum*, *Cosmos*, *Helianthus*,

Table II. Frequency of occurrence (FO%) of pollen types by bee species and frequency of occurrence (general FO%) of pollen types in honey samples of *Apis mellifera* and stingless bees of the semi-arid of Bahia, Brazil.

		FO (%) Species[a]								
Family	Pollen types	Ame	Mma	Mqa	Mqu	Mas	Msu	Sca	Tan	FO General (%)
Amaranthaceae	*Alternanthera*	14.29	12.50	25.00	—	12.50	—	16.67	25.00	75.00
	Amaranthaceae type	—	4.17	—	—	—	—	—	—	12.50
Anacardiaceae	*Schinus*	14.29	41.67	16.67	—	12.50	33.33	16.67	50.00	87.50
	Spondias	—	4.17	—	—	—	—	—	25.00	25.00
	Anacardiaceae type	—	—	—	—	—	—	—	25.00	12.50
Arecaceae	*Cocos nucifera*	—	4.17	—	—	—	—	—	—	12.50
Asteraceae	*Bidens*	14.29	8.33	—	—	25.00	—	16.67	50.00	62.50
	Centratherum	—	—	—	—	—	16.67	—	—	12.50
	Elephantopus	—	4.17	—	—	—	—	—	—	12.50
	Gochnatia	—	—	—	40.00	—	—	—	—	12.50
	Vernonia	14.29	—	8.33	—	12.50	16.67	—	25.00	62.50
Bignoniaceae	Bignoniaceae type	—	—	—	—	12.50	—	—	—	12.50
Boraginaceae	*Heliotropium*	14.29	4.17	—	—	12.50	8.33	—	—	50.00
Brassicaceae	Brassicaceae type	—	—	8.33	—	—	—	—	—	12.50
Convolvulaceae	*Jacquemontia*	—	—	8.33	—	12.50	—	—	—	25.00
Cucurbitaceae	*Cucurbita* type	14.29	—	—	—	—	—	—	—	12.50
	Cucurbitaceae type	—	20.83	33.33	—	—	50.00	—	—	37.50
Euphorbiaceae	Euphorbiaceae type	—	—	—	20.00	—	—	—	—	12.50
	Croton 1	28.57	—	—	—	12.50	—	—	25.00	37.50
	Croton 2	—	—	—	—	12.50	—	—	—	12.50
	Croton 3	28.57	—	—	—	12.50	—	—	—	25.00
Erythroxylaceae	Erythroxylaceae type	—	4.17	—	—	—	—	—	—	12.50
Fabaceae	*Acacia*	42.86	33.33	8.33	60.00	12.50	91.67	16.67	—	87.50
	Anadenanthera	14.29	—	—	—	25.00	33.33	—	—	37.50
	Caesalpinia	—	—	—	—	—	—	—	25.00	12.50
	Chamaecrista 1	14.29	8.33	—	60.00	—	41.67	16.67	25.00	75.00
	Chamaecrista 2	—	—	25.00	—	12.50	66.67	—	25.00	62.50
	Chamaecrista 3	—	—	8.33	—	12.50	16.67	—	—	37.50
	Delonix	14.29	—	—	—	—	—	—	—	12.50
	Leucaena	14.29	20.83	—	20.00	—	8.33	—	—	50.00
	Leucaena leucocephala	14.29	—	16.67	—	—	41.67	16.67	—	50.00
	Macroptilium	—	—	—	—	—	8.33	33.33	25.00	37.50
	Mimosa	—	—	—	—	12.50	—	—	—	12.50
	Mimosa caesalpiniifolia	42.86	37.50	91.67	60.00	50.00	50.00	16.67	—	87.50
	Mimosa pudica	14.29	—	—	—	50.00	—	—	25.00	37.50
	Mimosa tenuiflora	57.14	91.67	100.00	100.00	75.00	83.33	16.67	—	87.50
	Piptadenia	42.86	4.17	—	—	—	—	—	—	25.00
	Prosopis	57.14	45.83	50.00	60.00	37.50	16.67	66.67	50.00	100.00
	Senna	57.14	29.17	25.00	—	12.50	—	33.33	25.00	75.00
	Fabaceae type 1	—	8.33	—	—	—	—	—	—	12.50
	Fabaceae type 2	—	4.17	—	—	—	—	—	—	12.50

(*Continued*)

Table II. (*Continued*).

Family	Pollen types	FO (%) Species[a]								FO General (%)
		Ame	Mma	Mqa	Mqu	Mas	Msu	Sca	Tan	
	Fabaceae type 3	—	4.17	—	—	—	—	—	—	12.50
Fabaceae	Fabaceae type 4	—	—	—	—	—	8.33	—	—	12.50
	Mimosoideae type	—	—	8.33	—	—	—	—	—	12.50
Lamiaceae	*Hyptis*	—	—	—	—	12.50	—	—	25.00	25.00
Malpighiaceae	Malpighiaceae type	—	—	—	—	—	—	16.67	—	12.50
Malvaceae	*Herissantia*	28.57	4.17	—	—	25.00	16.67	—	—	50.00
	Waltheria	14.29	100.00	58.33	—	37.50	33.33	50.00	—	75.00
Melastomataceae	Melastomataceae type	—	4.17	16.67	—	—	8.33	—	25.00	50.00
	Tibouchina	14.29	—	—	20.00	—	—	—	—	25.00
Moraceae	*Morus*	—	—	8.33	—	—	—	—	—	12.50
Myrtaceae	*Eucalyptus*	28.57	16.67	16.67	—	12.50	—	16.67	—	62.50
	Psidium	57.14	4.17	16.67	100.00	50.00	50.00	16.67	—	87.50
Poaceae	Poaceae type	42.86	4.17	—	—	—	—	—	25.00	37.50
Portulacaceae	*Portulaca*	—	—	—	—	25.00	—	—	—	12.50
Rubiaceae	*Borreria*	28.57	4.17	16.67	—	50.00	8.33	50.00	25.00	87.50
	Rubiaceae type	28.57	54.17	41.67	—	37.50	91.67	33.33	25.00	87.50
Rutaceae	Rutaceae type	—	—	—	—	—	8.33	—	—	12.50
Sapindaceae	*Serjania*	—	4.17	—	—	—	—	16.67	—	25.00
Scrophulariaceae	*Scoparia dulcis*	—	—	8.33	—	12.50	—	—	—	25.00
Solanaceae	*Physalis*	—	—	8.33	—	—	—	—	—	12.50
	Solanum	—	62.50	66.67	80.00	62.50	25.00	33.33	25.00	87.50
	Solanaceae type	—	4.17	—	—	—	—	—	—	12.50
Verbenaceae	*Lantana*	—	—	8.33	—	25.00	—	—	25.00	37.50
Indeterminate	01	14.29	—	—	—	—	—	—	—	12.50
	02	—	—	—	—	—	16.67	—	—	12.50

[a]Ame, *Apis mellifera*; Mas, *Melipona asilvai*; Mma, *Melipona mandacaia*; Mqa, *Melipona quadrifasciata anthidioides*; Mqu, *Melipona quadrifasciata*; Msu, *Melipona subnitida*; Sca, *Scaptotrigona* sp.; Tan, *Tetragonisca angustula*. Frequency of occurrence (FO%) (Feller-Demalsy et al. 1987): > 50% of samples, very frequent; 20–50%, frequent; 10–20% infrequent; < 10%, rare.

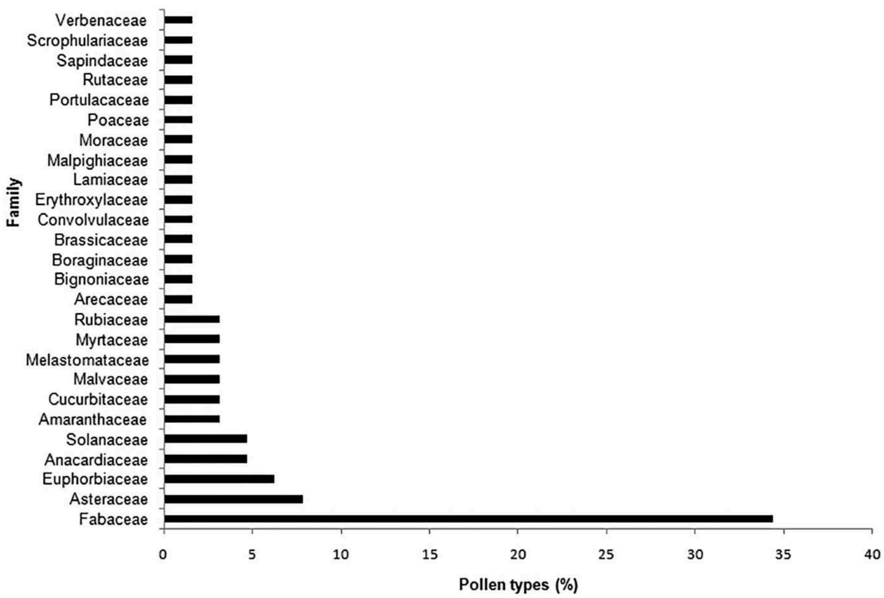

Figure 2. Number of pollen types per family identified in honey samples of *Apis mellifera* and different species of stingless bees from the semi-arid region of Bahia, Brazil.

Mikania and *Vernonia* (Borges et al. 2014; Nascimento et al. 2015; Matos & Santos 2016).

Among the most frequent pollen types in the samples of all bee species, the types *Acacia*, *Prosopis* and *Solanum* are highlighted. *Prosopis*, as well as species of the genus *Acacia* (Fabaceae-Mimosoideae), are very common in the studied region, playing a very important role for bees (Santos & Carneiro 2015). This fact can be verified by the registration of pollen types *Prosopis* and *Acacia* in the samples evaluated (Table II). The species *Prosopis juliflora* (Sw) DC. is very common in the Caatinga and highly used for animal feed (Braga et al. 2009; Oliveira et al. 2010). *Prosopis juliflora* offers both pollen and nectar, becoming attractive to bees (Sousa et al. 2016b).

The pollen type *Mimosa tenuiflora* was present in 100% of honey samples of bee species *Melipona quadrifasciata anthidioides* and *Melipona quadrifasciata*, with frequency above 70% for the other bees of the genus *Melipona* in this study. This result indicates that *Mimosa tenuiflora* is preferred by these bee species. Although the pollen spectrum of the analysed honey is diverse, probably these bees are more attracted by this plant species (Figure 5, Table I). The *Mimosa tenuiflora* (Willd) Poir. species is very attractive to bees, a source of pollen and nectar, being a species characteristic of the Caatinga. Its flowering occurs during a period of little floral abundance, providing great pollen amount to bees. It is also considered a potential species for bee rearing in the semi-arid region of Bahia (Calixto Júnior et al. 2011; Maia-Silva et al. 2012).

Another pollen type that occurred in most samples studied was *Mimosa caesalpiniifolia*. The species *Mimosa caesalpiniifolia* Benth. presents great potential for afforestation, hedge and wood production (Alves et al. 2002). This species is considered exotic and is used as trophic resources (pollen and nectar) by bees. The use of these plants by bees can be confirmed by the frequency in 75% of the honeys studied. Studies carried out on stingless bee honeys and *Apis mellifera* by Souza et al. (2015) and Nascimento et al. (2015), respectively, identified pollen types of the genus *Mimosa*, for example, *Mimosa caesalpiniifolia*, *Mimosa verrucosa*, *Mimosa arenosa*, *Mimosa quadrivalvis* and *Mimosa pudica*. The diversity of species of the genus *Mimosa* is indicative of the food preference of different species of bees by these plants.

In *Melipona mandacaia* samples, *Waltheria* type had 100% frequency and occurred as predominant pollen (PP) in 52% of the samples (Table II). Although other plant species (pollen types) were found in the honeybee pollen spectrum, which contribute to honey composition, there is possibly a preference for these bees to collect nectar in *Waltheria*. Due to the attractiveness of *Waltheria* spp. (Malvaceae) for bees, beekeepers recognise this plant as excellent to compose the bee flora, considered a nectariferous plant. Silva et al. (2013) and Oliveira and Santos (2014) identified the *Waltheria* pollen type in honeys of stingless bee and *Apis mellifera*.

The abundance of these spontaneous plants (*Waltheria* spp.) can be considered the main attractive factor for bees, since these plants produce small flowers and probably secretion of nectar is low (Kill et al. 2000; Maia-Silva et al. 2012). Castro (1994) defines useful plants for bees with abundant occurrence in the

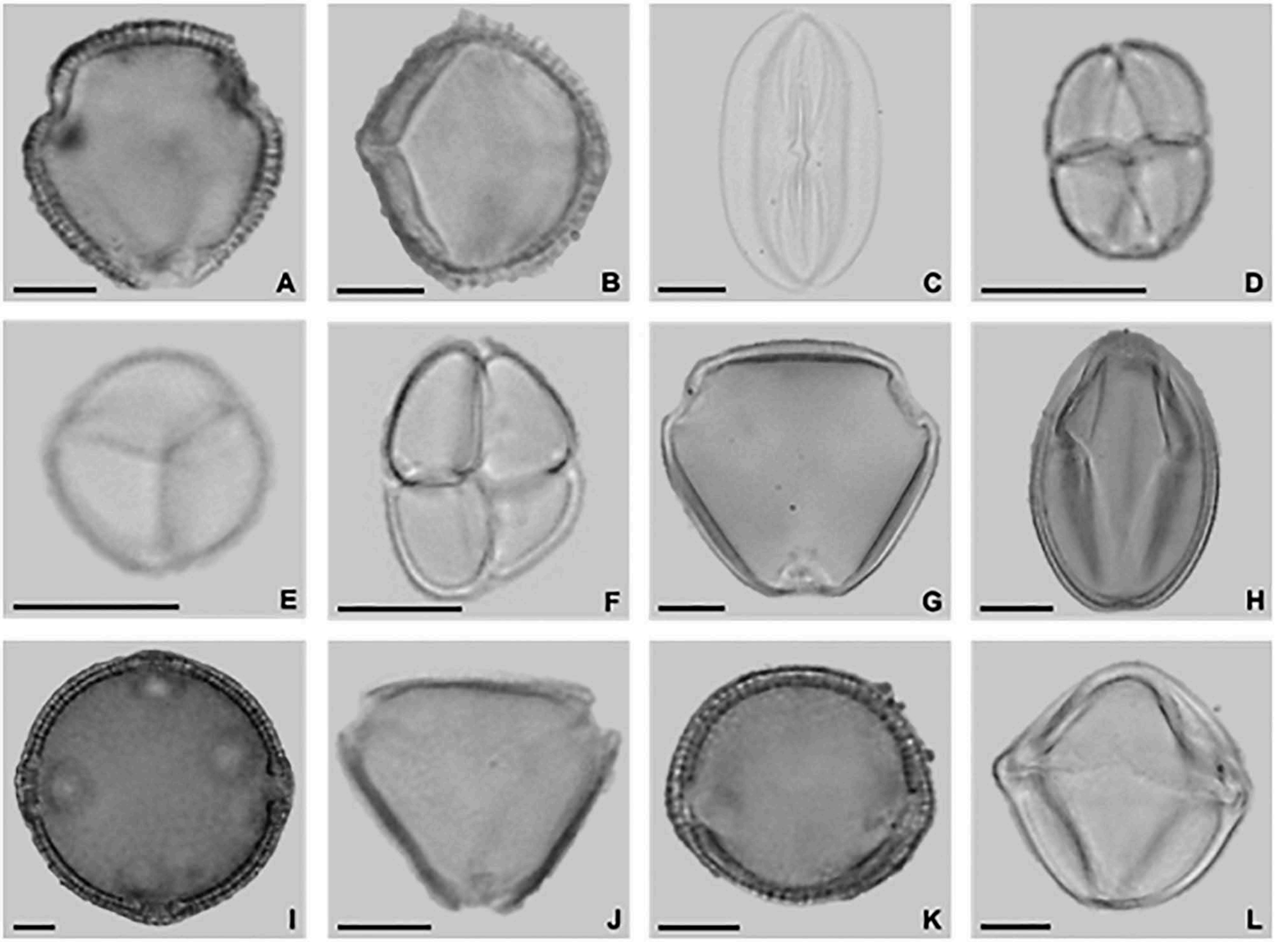

Figure 3. Photomicrography of predominant pollen (PP) types in *Apis mellifera* honey and stingless bees from the semi-arid region of Bahia, being EV (equatorial view) and PV (polar view). **A, B.** Anacardiaceae: *Schinus*, **A.** Polar view. **B.** Equatorial view. **C.** Fabaceae: *Chamaecrista* 1 (EV). **D.** Fabaceae: *Mimosa caesalpiniifolia* (PV). **E.** Fabaceae: *Mimosa pudica* (PV). **F.** Fabaceae: *Mimosa tenuiflora* (PV). **G.** Fabaceae: *Prosopis* (PV). **H.** Fabaceae: *Senna* (EV). **I.** Malvaceae: *Waltheria* (PV). **J.** Myrtaceae: *Psidium* (PV). **K.** Rubiaceae: *Borreria* (PV). **L.** Solanaceae: *Solanum* (EV). Scale bars – 10 µm.

area for breeding these individuals, blooming copiously with nectar and pollen available for bees. Therefore, *Waltheria* can be considered an important plant for beekeeping for the semi-arid region of Bahia.

Frequent presence of *Solanum* type in the honey samples analysed can be a consequence of contamination, as this species of this genus have poricidal anther, through which they release dry pollen, and are pollinated by vibration, besides not secreting nectar (Willmer 2011; Nunes-Silva et al. 2013). Thus, although species of the *Solanum* genus do not contribute to honey composition with nectar volume, their identification in the pollen spectrum of the evaluated honeys helps characterise the flora explored by bees in the sites surrounding the apiaries. The *Solanum* type was also identified in the pollen spectrum of the honeys evaluated by Nascimento et al. (2015), Souza et al. (2015), Falcão et al. (2016) and Matos and Santos (2016).

The honeys studied for *Apis mellifera* or stingless bees are multifloral, except for a sample of *Scaptotrigona* sp. The occurrence of multifloral honeys is due to the great diversity of native flora of the region, which allows bees to explore a range of plants providing trophic resources of the various botanical families. Regardless of the bee species that produce the honey, we observed that they forage several species to form their diet (Table II). This fact can add value to the product, since Brazilian honeys have peculiar sensorial characteristics according to the region of origin (Sousa et al. 2016a).

The search for food by bee *Scaptotrigona* sp. in a single plant species, *Senna* type (Fabaceae), may be related to several factors such as resources available during scarce period of collection, abundance and

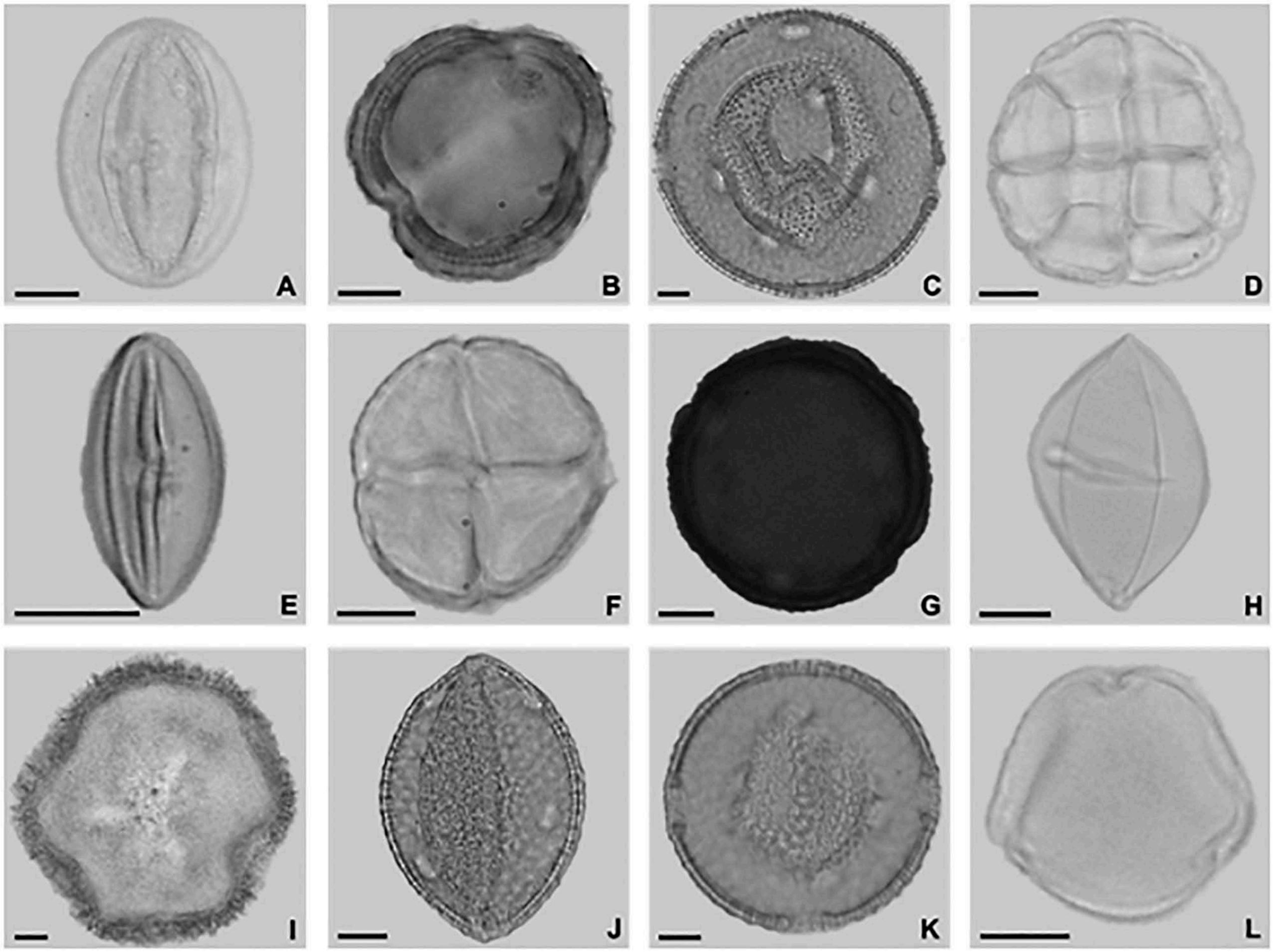

Figure 4. Photomicrography of secondary pollen (SP) types in *Apis mellifera* honey and stingless bees from the semi-arid region of Bahia, being EV (equatorial view) and PV (polar view). **A.** Anacardiaceae: *Spondias* (EV). **B.** Asteraceae: *Gochnatia* (PV). **C.** Cucurbitaceae: Cucurbitaceae type (PV). **D.** Fabaceae: *Acacia* (PV). **E.** Fabaceae: *Chamaecrista* 2 (EV). **F.** Fabaceae: Mimosoideae type (PV). **G.** Malpighiaceae: Malpighiaceae type (PV). **H.** Poaceae: Poaceae type (EV). **I.** Portulacaceae: *Portulaca* (PV). **J.** Rubiaceae: Rubiaceae type (EV). **K.** Rubiaceae type (PV). **L.** Verbenaceae: *Lantana* (PV). Scale bars – 10 μm.

proximity of plant species of the genus *Senna* within the radius of action of this bee. Osterkamp and Jasper (2013) report that the large number of pollen types identified in honeys hinders determination of the exact species that contributed to honey composition. This was also observed in our study.

The multivariate statistical analysis revealed that the pollen type *Mimosa tenuiflora* was collected mainly by bees *Melipona quadrifasciata* and *Melipona mandacaia*, and this pollen type contributed more to the first PCA. This component is also influenced by the *Acacia* type, mainly collected by *Melipona subnitida*. The second PCA explains 20.9% of the variation and is influenced by pollen type *Chamaecrista* 2, which was also collected by *Melipona subnitida*. The first and second PCA are presented in Figure 5.

The third component explains about 14.6% of the total variation and was influenced by *Chamaecrista* 1 and by the collection in plant species represented by the pollen type *Leucaena leucocephala* for *Scaptotrigona* sp., *Prosopis* for *Apis mellifera* and *Mimosa pudica* for *Melipona asilvai*. In addition, *Scaptotrigona* sp. and *A. mellifera* show preference for *Piptadenia* type. The cluster analysis, using the UPGMA method, corroborates the results obtained in the PCA (Figure 6).

Additionally, the cluster analysis showed a cophenetic correlation of 86% and formation of four groups, two groups formed by only one bee species, *Scaptotrigona* sp. and *Melipona subnitida*. The separation of these species of bees occurred due to food preference for an unusual floral resource by the other bee species.

Table III. Predominant pollen (PP) and secondary pollen (SP) types identified in *Apis mellifera* honeys from stingless bees, from the semi-arid region of Bahia.

	Pollen types	
Bee species	PP	SP
Apis mellifera	*Mimosa caesalpiniifolia*; *Mimosa tenuiflora*	*Acacia*; *Borreria; Mimosa caesalpiniifolia; Mimosa tenuiflora*; *Prosopis*; Poaceae type; *Waltheria*
Melipona asilvai	*Mimosa caesalpiniifolia*; *Mimosa pudica*; *Solanum; Waltheria*	*Borreria*; *Chamaecrista* 2; *Lantana*; *Mimosa tenuiflora*; *Portulaca; Prosopis*; *Psidium; Solanum*
M. mandacaia	*Mimosa caesalpiniifolia*; *Mimosa tenuiflora*; *Prosopis; Waltheria*; *Solanum*	*Chamaecrista* 1; *Mimosa tenuiflora; Prosopis; Senna*; *Solanum; Schinus*; Cucurbitaceae type; Rubiaceae type; *Waltheria*
M. quadrifasciata anthidioides	*Mimosa caesalpiniifolia*; *Mimosa tenuiflora*; *Waltheria*	*Lantana*; *Mimosa caesalpiniifolia; Mimosa tenuiflora*; *Senna*; Mimosoideae type; *Solanum; Waltheria*
M. quadrifasciata	*Mimosa caesalpiniifolia*; *Chamaecrista* 1; *Solanum*; *Psidium*	*Gochnatia*; *Mimosa tenuiflora*
M. subnitida	*Mimosa caesalpiniifolia*; *Mimosa tenuiflora*	*Acacia*; *Chamaecrista* 1; *Chamaecrista* 2; *Macroptilium*; *Mimosa tenuiflora*; Rubiaceae type
Scaptotrigona sp.	*Mimosa caesalpiniifolia*; *Prosopis; Senna*	*Mimosa tenuiflora*; *Prosopis; Solanum*; *Schinus*; Malpighiaceae type; *Waltheria*
Tetragonisca angustula	*Borreria*; *Solanum; Schinus*	*Lantana*; *Prosopis; Spondias*; Rubiaceae type; *Waltheria*

Note: PP (more than 45% of the pollen grains counted) and SP (16–45%) (Louveaux et al. 1978).

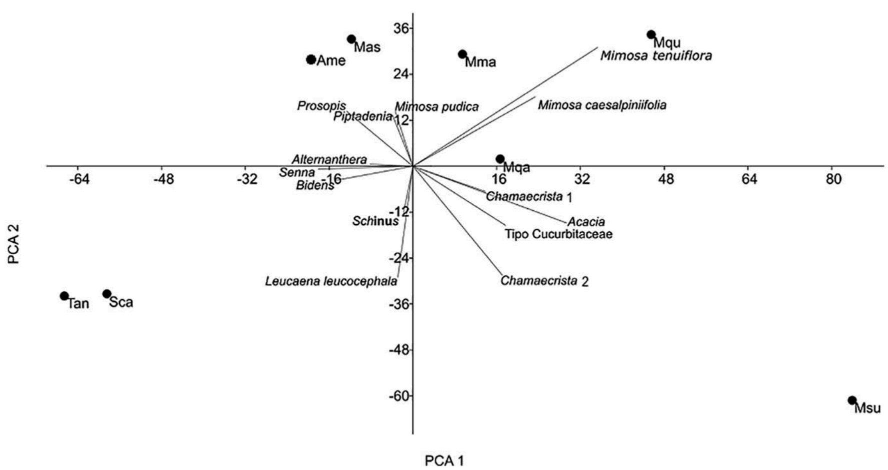

Figure 5. Scatter plot from the principal component analysis (PCA) and biplot, which relates the pollen types of food preference of the bee species under study. Abbreviations: Ame, *Apis mellifera*; Mas, *Melipona asilvai*; Mma, *Melipona mandacaia*; Mqa, *Melipona quadrifasciata anthidioides*; Mqu, *Melipona quadrifasciata*; Msu, *Melipona subnitida*; Tan, *Tetragonisca angustula*; Sc, *Scaptotrigona* sp.

Scaptotrigona sp. as food resource species was represented by the pollen types *Physalis* and *Jacquemontia* and *Melipona subnitida* by *Macroptilium* type.

The cluster analysis revealed similarity in the nectariferous resources visited by *Apis mellifera*, *Melipona quadrifasciata anthidioides* and *Melipona quadrifasciata* in the semi-arid region of Bahia (Fig-Figure 6). These social bees share pollen types of Fabaceae as the most frequent plants for honey composition (Figure 5 and Tables II and III). Another group is formed by *Melipona asilvai* and *Melipona mandacaia* showing greater similarity in the composition of its pollen spectrum. This indicates that if these bee species are managed in the same environment, there may occur trophic niche overlap. Gostinski et al. (2018) conducted a study in the Amazon region in Brazil and observed high niche overlap for stingless bees.

Melipona subnitida and *Scaptotrigona* sp. showed no evidence of group formation with the other species of stingless bees, possibly due to the volume contribution of nectar of each plant visited by the bees. The separation of these species of bees occurred due to food preference for an unusual floral resource by the other bee species. *Scaptotrigona* sp. as food resource species was represented by the pollen types *Physalis* and *Jacquemontia* and *Melipona subnitida* by *Macroptilium* type.

Conclusions

The identification of pollen types in honeys from the semi-arid region of Bahia allowed their classification as

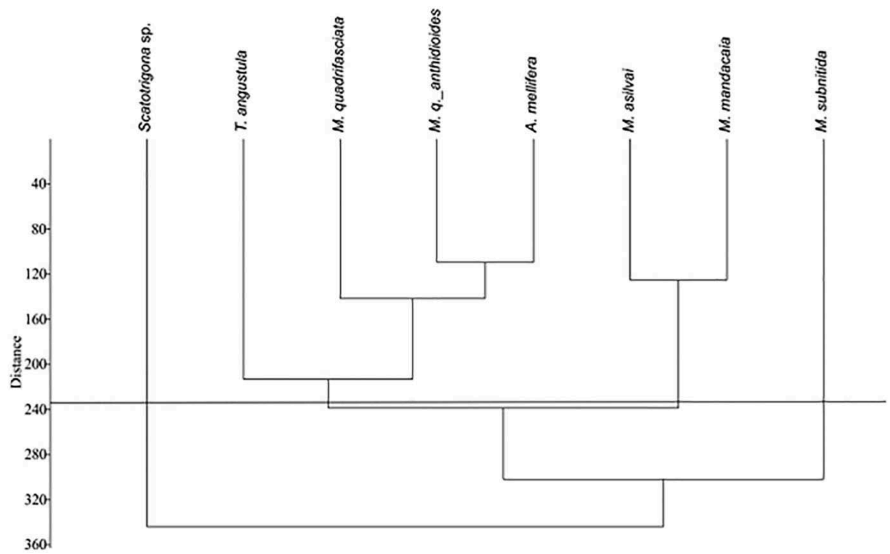

Figure 6. Grouping analysis by the UPGMA method showing preference of different bee species to the flora.

multifloral honeys, with an expressive contribution of pollen types of the Fabaceae family, predominantly, *Chamaecrista*, *Mimosa tenuiflora*, *Mimosa caesalpiniifolia*, *Prosopis* and *Senna*. The large number of pollen types identified in honey samples demonstrates the great vegetation diversity in the semi-arid region of Bahia, Brazil.

Disclosure Statement

No potential conflict of interest was reported by the authors.

Supplementary material

Supplemental data for this article can be accessed here.

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