

Article

Identification of Floral Resources Used by the Stingless Bee *Melipona beecheii* for Honey Production in Different Regions of the State of Campeche, Mexico

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Abstract: The stingless bee *Melipona beecheii* is experiencing colony decline due to floral resource scarcity caused by deforestation. A study was conducted to identify the floral resources used by *M. beecheii* using honey samples collected in four regions of the state of Campeche, Mexico. A melissopalynological analysis of sixteen collected honey samples identified 69 plant species from 24 families, and established that Fabaceae was the main plant family visited. Based on botanical origin, seven samples were classified as monofloral and nine as multifloral. The predominant species were *Bursera simaruba*, *Lonchocarpus longistylus*, *Piscidia piscipula*, *Senna pallida* and *Senna racemosa*. Shannon diversity index values (2.06–2.55) indicated moderate diversity in floral resources and Simpson diversity index values (0.82–0.89) indicated a moderate dominance of plant species in the studied regions. The results suggest *M. beecheii* is polylectic with some degree of specialization. The plant species identified as predominant in the studied honey samples are candidates for use in strategies intended to conserve the food resources used by *M. beecheii* on the Yucatan Peninsula.

Keywords: melissopalynology; floral resources; diversity



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1. Introduction

Stingless bees are highly social bees native to tropical and sub-tropical ecosystems [1]. They feed on pollen and nectar from flowers in their habitat [1,2]. Though some are polylectic, they do not collect pollen and nectar from all available flowers. Polyfloral diets are healthier for bees than monofloral diets because they ensure that the bees consume a wide range of nutrients [1,3,4]. The floral diversity available to bees is determined by the environmental and landscape composition of the area surrounding a hive site [3]. In recent years, reductions in native plant diversity, due to deforestation and increasing areas used for intensive agriculture, have been affecting food source quality and availability for bee colonies [2,5]. Stingless bees are strongly affected by deforestation because their small body size reduces their maximum flight distance capacity for obtaining food and nesting resources [6,7]. Stingless bee populations have consequently exhibited substantial declines worldwide [5,8,9].

Deforestation is widespread and ongoing in Mexico. It is particularly notable on the Yucatan Peninsula in the country's southeast. The principal causes in this region are mainly associated with livestock expansion, as well as the expansion of agricultural areas, the latter of which is observed mainly in the state of Campeche [10,11].

The purported human health benefits of honey consumption from the native bee *Melipona beecheii* have driven an increase in demand [12–14]. However, *M. beecheii* is among the stingless bee species experiencing colony decline due to factors such as reduced floral resources and a lack of nesting sites [11]. Stingless bee species tend to exploit the predominant tree species in their environment [15]. Identifying the nectar and pollen source preferences of stingless bees like *M. beecheii* in Campeche can help to promote the use of native plant species that provide floral resources as conservation tools to increase stingless bee populations [16].

Melissopalynological studies are vital to identifying the nectar and pollen sources of bees and supporting improvements in colony and landscape management [17]. Melissopalynological research focused on identifying the nectar and pollen resources used by *M. beecheii* has been limited. One study was conducted in Costa Rica [18] and another in Cuba [19]. Three studies have been conducted in Mexico [20–22], but only one in the state of Campeche [23]; this study focused on a single site, meaning it is unrepresentative of the state as a whole. Much broader melissopalynological analyses are needed to identify *M. beecheii* floral resource preferences and to promote native vegetation reserves for conservation on the Yucatan Peninsula. The present study's objective was to analyze honey samples from four regions in the state of Campeche, Mexico, to identify the floral resources used by *M. beecheii*.

2. Materials and Methods

2.1. Study Area

The state of Campeche is located in the north at 20°50'54", in the south at 17°48'46" latitude north, in the east at 89°07'16" and in the west at 92°28'08" longitude. Campeche borders, to the north, the Gulf of Mexico and Yucatan; Yucatan, Quintana Roo, and Belize to the east; the Republic of Guatemala and Tabasco to the south; and Tabasco and the Gulf of Mexico to the west. The predominant climate is warm–humid in 92.22% of the state, warm sub-wet in 7.73% of the state (eastern part), and semi-dry very warm and warm in 0.05% of the state (north region). The average annual temperature is 26 to 27 °C, and the annual rainfall is 1200 to 2000 mm from the north to the southeast of the state. The main types of vegetation are medium evergreen forest, medium sub evergreen forest, and medium sub deciduous forest, according to the CONABIO geoportal [24].

2.2. Sample Collection Sites

A total of sixteen samples were collected (Figure 1 and Table 1) from March to April 2021, in four regions in Campeche, Mexico: four in the southeast, seven in the north, three in the northeast, and two in the west. The largest number of samples was collected in the north because this is the area in which most of the meliponiculture activity is carried out in the state of Campeche [13]. Honey samples were removed from the honey pots in the hives with sterile Pasteur pipettes and transferred directly into amber-colored jars. Samples were stored at approximately 8 °C until analysis.

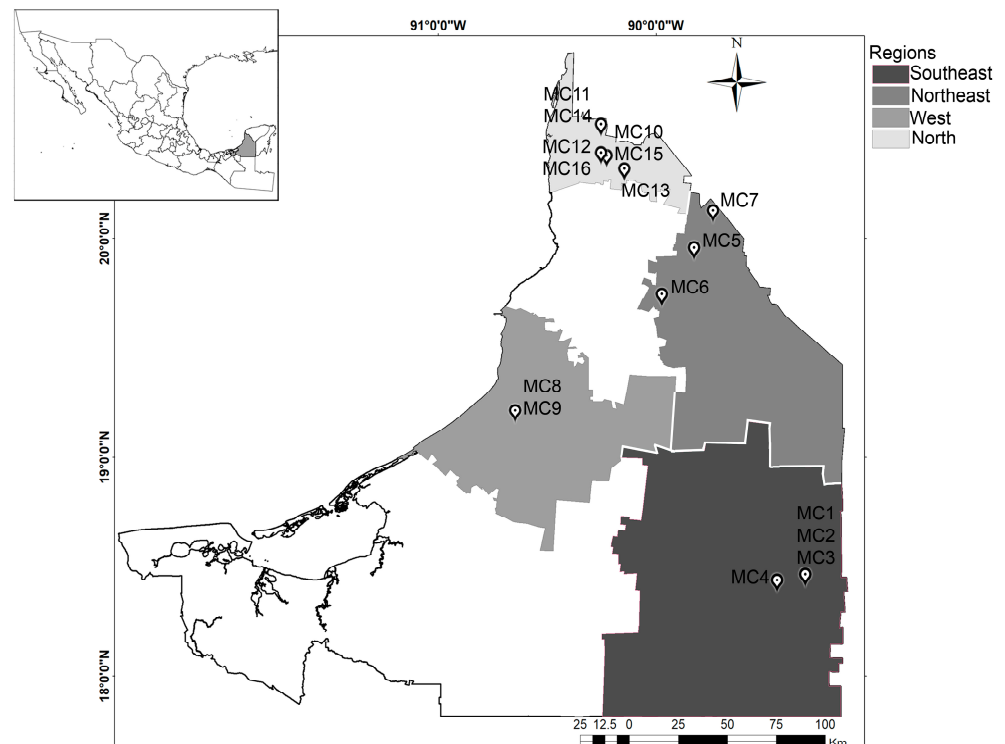


Figure 1. State of Campeche, *Melipona beecheii* honey sample collection sites. MC1–MC3: 20 Noviembre, Calakmul; MC4: La Lucha 1, Calakmul; MC5: Xcalot Akal, Hopelchén; MC6: Ich ek, Hopelchén; MC7: San Antonio, Hopelchén; MC8 and MC9: Sihochac, Champotón; MC10: Pucnachén, Calkiní; MC11: Tankunché, Calkiní; MC12: Santa María, Calkiní; MC13: Sahcabchén, Calkiní; MC14: Tankunché, Calkiní; MC15: Pucnachén, Calkiní; MC16: Santa María, Calkiní.

Table 1. Collection sites of *Melipona beecheii* honey samples and vegetation type.

Regions	Locality	Sample	Vegetation Type
Southeast			
	20 de Noviembre, Calakmul	MC1	Medium sub-evergreen forest
	20 de Noviembre, Calakmul	MC2	Medium sub-evergreen forest
	20 de Noviembre, Calakmul	MC3	Medium sub-evergreen forest
	La lucha I, Calakmul	MC4	Medium sub-evergreen forest
Northeast			
	Xcalot Akal, Hopelchén	MC5	Medium sub-deciduous forest
	Ich ek, Hopelchén	MC6	Medium sub-deciduous forest
	San Antonio, Hopelchén	MC7	Medium sub-deciduous forest
West			
	Sihochac, Champotón	MC8	Medium sub-deciduous forest
	Sihochac, Champotón	MC9	Medium sub-deciduous forest
North			
	Pucnachén, Calkiní	MC10	Medium deciduous forest
	Tankuché, Calkiní	MC11	Medium deciduous forest
	Santa María, Calkiní	MC12	Medium deciduous forest
	Sahcabchén, Calkiní	MC13	Medium deciduous forest
	Tankunché, Calkiní	MC14	Medium deciduous forest
	Pucnachén, Calkiní	MC15	Medium deciduous forest
	Santa María, Calkiní	MC16	Medium deciduous forest

2.3. Palynological Analysis

The samples were processed following an established methodology [25]. Briefly, 10 g of each honey sample was diluted in 10 mL distilled water at near 40 °C. A total of 100 mL 95% EtOH was added and the mixture was centrifuged (5 min at 4000× g rpm). Samples were processed via acetolysis following the Erdtman method [26]. The pollen grains were mounted on glycerin–gelatin and the microscope slides were sealed with paraffin [27]. The slides were viewed using light microscopy with an optical microscope (40×) (Olympus BX41, Olympus Corporation, Tokyo, Japan) fitted with a camera (AmScope MU1803, China, United States of America). Images were processed using the AmScope v3.7 image software (AmScope, Irvine, CA, USA). Pollen grains in the honey samples were identified according to the taxonomic levels of family, genus and/or species. Unidentifiable grains were assigned an “sp.” designation. Pollen grains were identified based on morphological characteristics (polarity, size, shape, surface, and number of apertures) and a comparison with specimens in the Palynological Collection of Mesoamerica Floristic Diversity, Botany Department, Autonomous University of Yucatan (Universidad Autonoma de Yucatán—UADY). Palynological catalogs were also consulted [28–30].

2.4. Honey Botanical Origin

After pollen grain identification, the relative frequencies of the different pollen types or species were quantified by counting out 500 grains from each honey sample. Each sample composition could thus be analyzed in terms of (a) pollen frequency, (b) predominance by plant family and species (based on overall counts among all samples), and (c) species recurrence (number of times a species appears in all samples, divided by the total number of samples) [20]. Pollen frequency was used to classify pollen type dominance into four categories [31]: predominant (>45% total pollen in sample), secondary (15–45%), minor (3–15%), and residual (<3%). The honey samples were classified by botanical origin as either monofloral (dominated by one plant species at ≥45%) or multifloral (three or more secondary percentage pollen types) [31]. The predominance of the different plant families and species identified in the samples was calculated to determine which families were more important for *M. beecheii*, or whether any preference existed between plant families. Family and species recurrence was calculated to identify which species occurred in the largest number of samples, regardless of sampling site. Finally, the identified plant species were classified according to vegetation stratum (arboreal, shrub or herbaceous).

2.5. Ecological Analysis

The palynological data were analyzed using principal component analysis (PCA) and the ecological parameters were calculated. The Shannon–Wiener diversity index (H') was used to determine the diversity of plants that *M. beecheii* visit. In this index, values less than 2 indicate low diversity, those from 2 to 3.5 indicate medium diversity, and those greater than 3.5 indicate high diversity [32]. The Simpson index (D) was used to measure the dominance of the species. Based on a range of 0 to 1, the index measures the probability of two taxa from the same sample, selected at random, belonging to the same species. A value of 0 represents equity (that is, species uniformity) and 1 represents the complete dominance of one taxon [33]. All analyses were run using the PAST program version 3.16 [22,34–36].

3. Results

3.1. Honey Botanical Origin

A total of 69 pollen types belonging to 24 plant families, with 12 indeterminate types, were identified in *Melipona beecheii* honey samples from the state of Campeche (Table 2). The most diverse family was Fabaceae (15 species), followed by Solanaceae (five species), Asteraceae (four species), Myrtaceae (four species), Sapindaceae (four species), and Burseraceae (three species). Pollen grain frequency calculated for the samples as a whole showed Fabaceae (64.4% of samples), Myrtaceae (5.57%), Malvaceae (3.22%), and Asteraceae (3.21%)

to be the most represented. Pollen types lacking botanical affinity occurred in six samples and were grouped into an unidentified class designated “sp.” (Table 2).

Of the sixteen analyzed honey samples, botanical origin was monofloral in seven (MC3, MC4, MC7, MC10, MC11, MC13, and MC14) and multifloral in the remaining nine. The *M. beecheii* that produced the honey samples had visited plants of various genera and with different growth modes or strata; the most visited were arboreal (54.04%) followed by bush (33.87%) and herbaceous (11.33 %) (Table 2).

Quantitative pollen analysis showed that the overall predominant pollen types were *Senna racemosa* (Mill.) H.S. Irwin and Barneby (16.9%), *Piscidia piscipula* (L.) Sarg. (13.9%), *Lonchocarpus longistylus* Pittier, (11.5%), *Bursera simaruba* (L.) Sarg. (9.3%), and *Senna pallida* (Vahl) H.S. Irwin and Barneby (6.9%) (Figure 2). In terms of occurrence in the honey samples, six pollen types were frequent: *L. longistylus* (94% of samples), *S. racemosa* (88%), *P. piscipula* (88%), *Mimosa bahamensis* Benth. (81%), *B. simaruba* (88%), and *Eugenia axillaris* (Sw.) Willd. (75%).

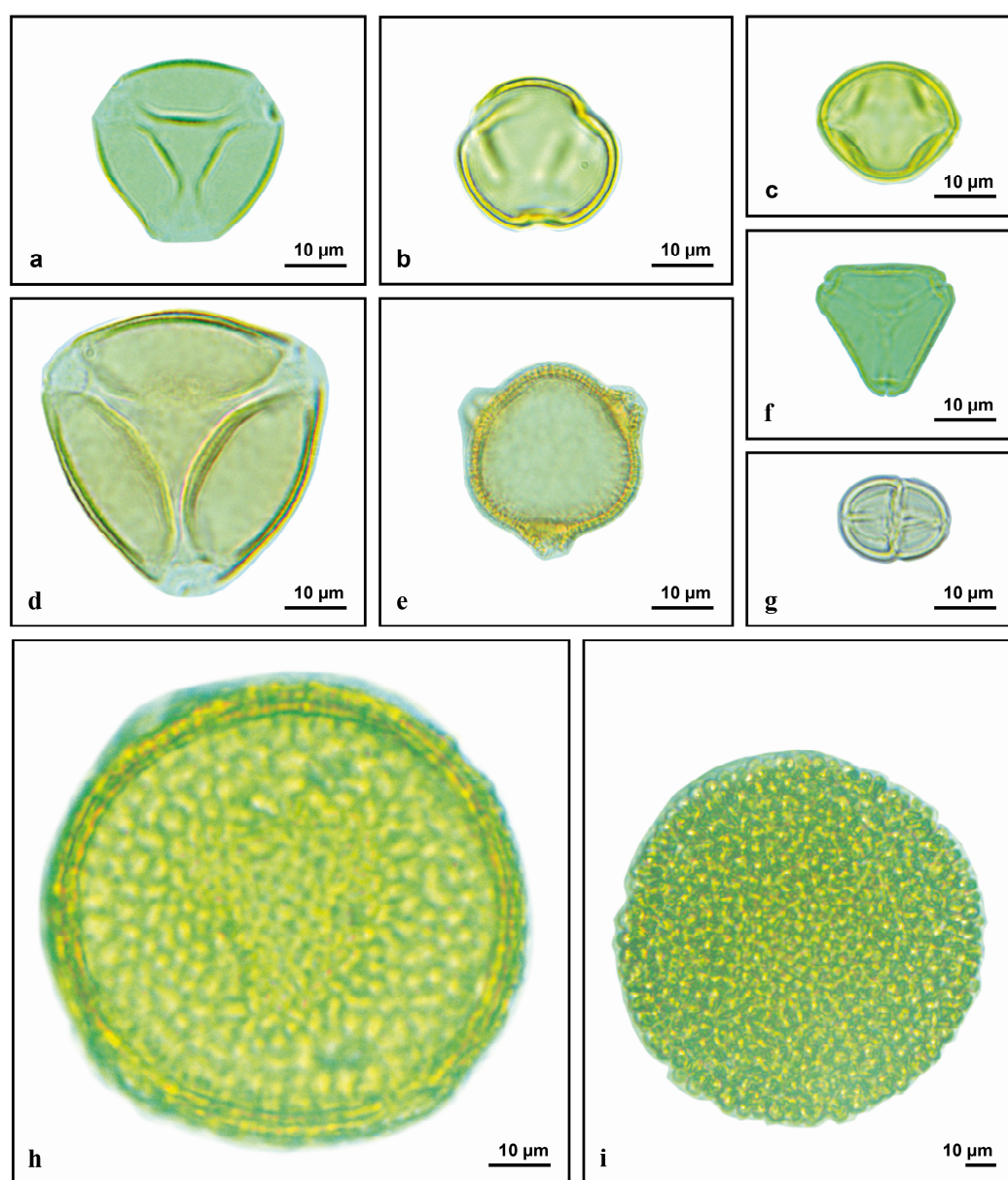


Figure 2. Pollen types identified in *Melipona beecheii* honey samples from Campeche, Mexico. (a) *Senna racemosa*; (b) *Lonchocarpus longistylus*; (c) *Piscidia piscipula*; (d) *Senna villosa*; (e) *Bursera simaruba*; (f) *Pimienta dioica*; (g) *Mimosa bahamensis*; (h) *Waltheria communis*; (i) *Croton* sp. Scale: 10 µm.

Table 2. Pollen types identified in *Melipona beecheii* honey samples from Campeche, Mexico.

Taxa	Mayan Name	Stratum	MC1	MC2	MC3	MC4	MC5	MC6	MC7	MC8	MC9	MC10	MC11	MC12	MC13	MC14	MC15	MC16	PR (%)
Acanthaceae																			
<i>Aoicentzia germinans</i>	Ta'abché	A													1		0.2		13
Antarathaceae																			
<i>Alternanthera ramosissima</i>	Zakmuul	H						11.2										0.8	13
Asteraceae																			
<i>Asteraceae sp.</i>		H				2.3													6
<i>Chaptalia nutans</i>		H											0.6						6
<i>Viguiera dentata</i>	Taj (tajonal)	H	7.2	18.5		0.8	18.7	1.8			0.2								38
Boraginaceae							24												6
Boraginaceae																			6
Brassicaceae																			6
Arabideae sp.		H		4	0.2														13
Burseraceae																			13
<i>Bursera sinarubra</i>	Chakaj	A		3.4	50		0.2	2.4	4.9	5.1	5.5	0.9	35.4	20.9	0.2	18.6	2.6	7.4	88
<i>Protium copal</i>	Sak chakaj	A		9	0.2		2												19
Cactaceae																			6
<i>Cactaceae sp.</i>		H				0.2													6
Combretaceae																			6
<i>Bucida buceras</i>	Pucté	A				0.8													6
Convulvulaceae																			6
<i>Ipomea sp.</i>		H				0.2													6
Cyclanthaceae																			6
<i>Carludovica palmata</i>	Guano	H															0.4		6
Euphorbiaceae																			6
<i>Croton sp.</i>		S		2.1	18.4														13
Euphorbiaceae				1.9															6
Fabaceae																			6
<i>Acacia sp.</i>		S													0.2				6
<i>Acacia collinsii</i>	Subin	S														0.7			6
<i>Caesalpinia gaumeri</i>	Kitim che'	A					2.2	0.6											13
<i>Gliricidia septum</i>		A					-	-						6.6				3.4	13
<i>Leucaena leucocephala</i>	Waaxim	A				3.7	1.5	2.6	2.9	4.2	9						2	-	44
<i>Lonchocarpus longistylus</i>	Baal che'	A	3.6	2.3	3.9	1.2	6.4	22	2.9	-	0.9	19.6	47.2	31.3	1.2	0.2	16.8	24.9	94
<i>Lonchocarpus xukul</i>	K'an xu'ul	A																	13
<i>Mimosa bahamensis</i>	Káatsim blanco	A		1.9	4.5	1.2	1.3	1	6.8	27.5	0.9	0.4		0.9	5.7				81
<i>Mimosa pigra</i>	Je' beech'	S														25.6			6
<i>Mimosa pudica</i>	Múuts'il xiiw	H						0.4		4.2	0.4			0.4	1.4	1.3	0.6	7.8	50
<i>Piscidia piscipula</i>	Ja'abin	A								4.9									6
<i>Senna atomaria</i>		S	20.6			78.1	24.7	26	1	0.2		2.2	0.9	4.2	4.9	47.3	3.2	10.1	81
<i>Senna pallida</i>	Ch'iilib mich	S																	6
<i>Senna racemosa</i>	K'an lool	A	10	27.9	6.9		13.6	27.6	66.7	26.8	40	12.1	0.2	1.7	15.8	1.7	19.4		31
<i>Senna villosa</i>	Saal che'	S												5.9	62.1		6.3		88
Gesneriaceae																			19
<i>Achimenes palmata</i>		H															0.2		6
Malvaceae																			6
<i>Luehea speciosa</i>	K'an kaat	A				0.4													6
<i>Waltheria communis</i>		H													5.3	2.2	34.5	7.4	25
<i>Waltheria rotundifolia</i>		H													1.6		1.8	1	19
Meliaceae																			6
<i>Meliaceae sp.</i>								0.8											6
Myrtaceae																			6
<i>Eugenia axillaris</i>		S	6.5	6.6	7.3	6.3	0.9		2.2	0.7	1.7	0.4	-	0.8	0.6				75
<i>Eugenia foetida</i>	Sak loob	A		1.5															6
<i>Pimenta dioica</i>	Boox pool	A	44.7		3.1	0.4													19
<i>Psidium guajava</i>	Pichi	S											0.2						6
Nictaginaceae																			6
<i>Pisonia aculeata</i>	Béeb	H							3.9										6
Onagraceae																			6
<i>Ludwigia octovalvis</i>	Máaskab che'	H	2.2														1.6	2.6	19
Poligonaceae																			6
<i>Coccoloba manzanillensis</i>		A		4.7		0.6													13
<i>Neomillspaughia emarginata</i>	Sak iitsa'	S							0.4		3.7			1.5			1	1.2	31
Poligonaceae sp.		A	0.3							17.6		1.7							19
Primulaceae																			6
<i>Jacquinia aurantiaca</i>		S													0.4				6
Rubiaceae																			6
<i>Psychotria nervosa</i>	K'aanan	S										3.9							6
Sapindaceae																			6
<i>Sapindaceae sp.</i>		H		13.9	1														13
<i>Serjania goniocarpa</i>	Chak sik'ix le'	H		2.4		0.8													13
<i>Serjania lundellii</i>	buy aak'	H				0.8		2.2											13
<i>Thouinia paucidentata</i>	K'an chuunup	A				2.3								4.4					13
Sapotaceae																			6
<i>Sideroxylon foetidissimum</i>	Sibul	A						1.4											6
SOLANACEAE																			6
<i>Cestrum nocturnum</i>	K'an chuunuk	S												4.2					6
<i>Solanaceae sp.</i>		S												12.7					65
<i>Solanum lanceifolium</i>		H												1.1					13
<i>Solanum lanceolatum</i>	Sikil múuch	S															4.6		6
<i>Solanum nudum</i>	Boox kúuts	S						-	3.3		5.5					1.3			9
<i>Solanum tridynamum</i>	Kóon ya'ax iik	H					4.6												6
Unidentified sp.s																			6
sp. 1			1.2																6
sp. 2			0.2																6
sp. 3			0.9																6
sp. 4			2.6																6
sp. 5					3.3														6
sp. 6					1.2														6
sp. 7										0.6									6
sp. 8									6.6										6
sp. 9													0.2						6
sp. 10												3.7							6
sp. 11												0.6							6
sp. 12																1.1			6
Total			100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100

The numbers correspond to the relative frequency and white space is equal to zero, which is the equivalent of not finding grain. Abbreviations: A, arboreal; S, shrubs; H, herbaceous; PR, percentage of recurrence.

3.1.1. Southeast

In the four samples from the state’s southeast, a total of 32 pollen types were identified from thirteen families. The most frequent species were *E. axillaris*, *L. longistylus*, and *S. racemosa* (100% of samples) (Table 2). Two samples were considered monofloral: MC3 (*B. simaruba*) and MC4 (*P. piscipula*). Two samples were multifloral, one of which (MC1) contained *Pimienta dioica* (L.) Merr. and *P. piscipula* as a secondary pollen, while the other (MC2) contained *S. racemosa* and *Viguiera dentata* (Cavanilles) Spreng as a secondary pollen (Figure 3).

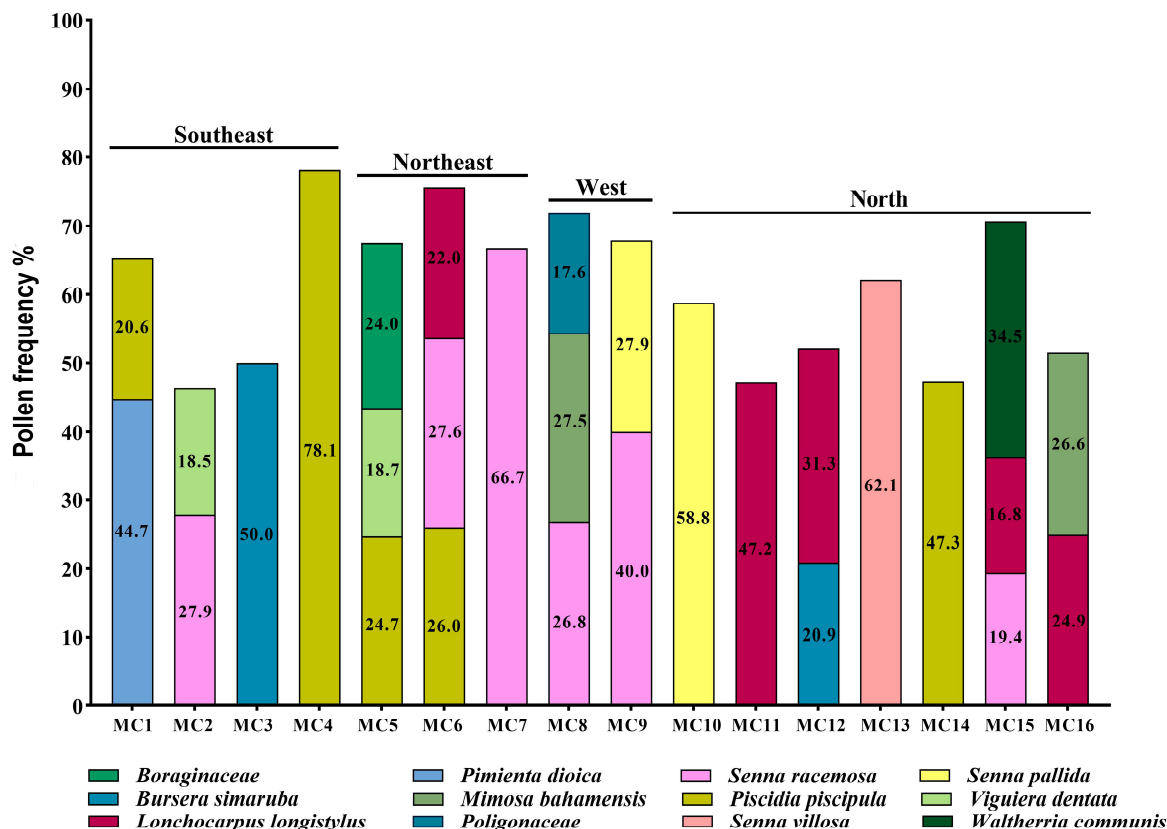


Figure 3. Characterization of the honey samples collected in different regions of Campeche, Mexico, as monofloral or multifloral. Numbers in the bars correspond to the percentages of pollen frequency per sample. Pollen taxa were included only as predominant (>45%) and secondary (15–45%).

3.1.2. Northeast

In the three samples from the state’s northeast, 21 pollen types were identified from 12 families. The most frequent resources were *B. simaruba*, *Leucaena leucocephala* (Lam.) de Wit, *L. longistylus*, *Mimosa bahamensis* Benth., *P. piscipula*, and *S. racemosa* (Table 2). Botanical origin analyses showed one sample (MC7) to be monofloral (*S. racemosa*). Two samples were multifloral: MC5 contained secondary pollen from *Boraginaceae* sp., *P. piscipula*, and *V. dentata*, and MC6 from *L. longistylus*, *P. piscipula*, and *S. racemosa* (Figure 3).

3.1.3. West

In the two samples from the west, 7 families and 16 pollen types were identified. The most frequent resources were *B. simaruba*, *E. axillaris*, *L. leucocephala*, *M. bahamensis*, *S. pallida*, and *S. racemosa* (Table 2). Both samples were multifloral: MC8 also contained *Polygonaceae* sp., *M. bahamensis*, and *S. racemosa*, and MC9 contained *S. pallida* and *S. racemosa* (Figure 3).

3.1.4. North

The seven samples from the north contained pollen from 14 families and from 37 pollen types. The most frequent resources were *B. simaruba*, *L. longistylus*, and *P. piscipula*. Four samples were considered monofloral: MC10 (*S. palida*), MC11 (*L. longistylus*), MC13 (*Senna villosa* (Mill.) H.S. Irwin and Barneby), and MC14 (*P. piscipula*) (Table 2). Three samples were multifloral: MC12 contained secondary pollen from *B. simaruba* and *L. longistylus*, MC15 from *L. longistylus*, *S. racemosa*, and *Waltheria communis* A.St.-Hil., and MC16 from *L. longistylus* and *M. bahamensis* (Figure 3).

3.2. Ecological Parameters

The Shannon diversity index values (H') for *M. beecheii* honey samples collected in the four studied regions exhibited intermediate values (Southeast, 2.51; Northeast, 2.17; West, 2.06; and North, 2.55) (Margalef 1972), indicating that the bees visited several plant species. The Simpson index (D) values showed medium-level dominance in all four regions (Southeast, 0.88; Northeast, 0.82; West, 0.82; and North 0.89).

3.3. Principal Component Analysis (PCA)

The first principal component (PC1) represents 63.79% of the variance and the second (PC2) represents 18.58% (Figure 4); together, they explain 82.36% of the total variance, which is high (eigenvectors, Annex I). PC1 has a greater dimension on the biplot, representing the overall pollen type preference for the geographical region. PC2 is orthogonal to PC1, being the second most preferred. In the biplot, the points represent pollen types and the vectors (green lines) represent the geographical regions. A vector trending in the same direction represents a geographical region with similar pollen type preferences.

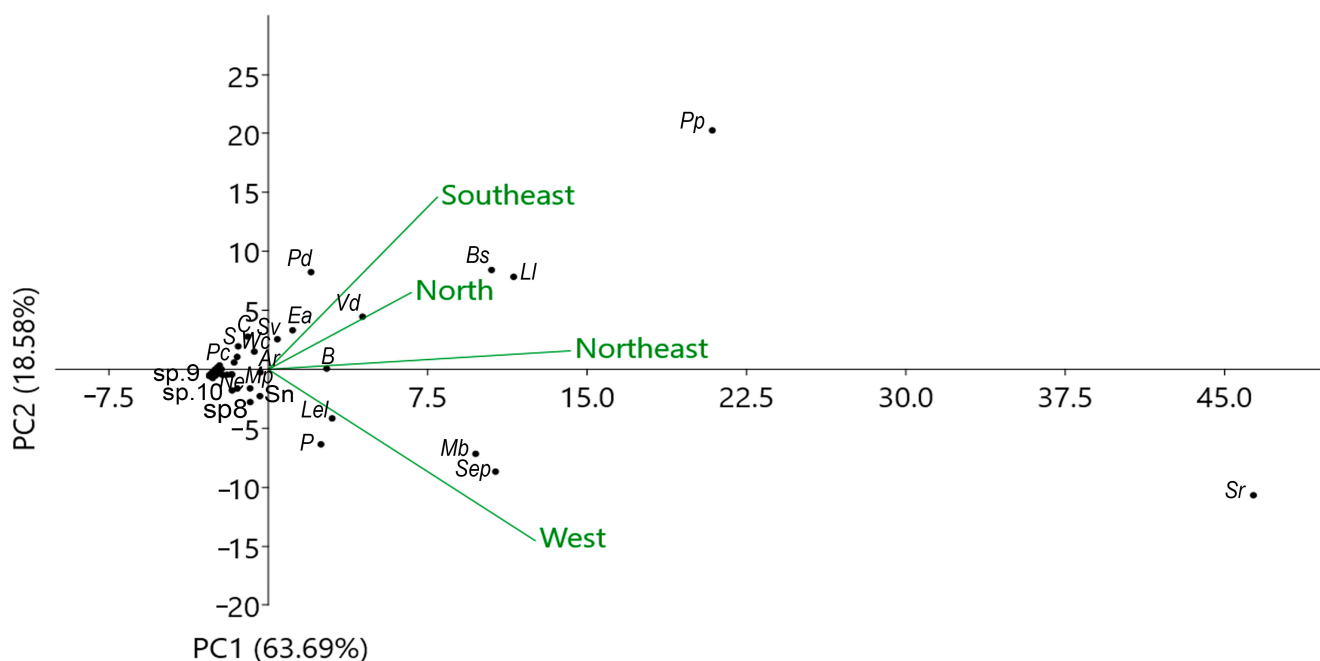


Figure 4. Principal components analysis (PCA) of the floral origin of *Melipona beecheii* honey from the state of Campeche. (Ar) *Alternanthera ramosissima*; (B) *Boraginaceae* sp.; (Bs) *Bursera simaruba*; (C) *Croton* sp.; (Ea) *Eugenia axillaris*; (LeI) *Leucaena leucocephala*; (LI) *Lonchocarpus longistylus*; (Mb) *Mimosa bahamensis*; (Mp) *Mimosa pudica*; (Ne) *Neomillspaughia emarginata*; (P) *Polygonaceae* sp.; (Pc) *Protium copal*; (Pd) *Pimenta dioica*; (Pp) *Piscidia piscipula*; (S) *Sapindaceae* sp.; (Sep) *Senna pallida*; (Sn) *Solanum nudum*; (Sr) *Senna racemosa*; (Sv) *Senna villosa*; (Vd) *Viguiera dentata*; (Wc) *Waltheria communis*.

Based on PC1, it is clear that the samples from the west region are separated on the lower side of PC1, and are characterized by a preference for six pollen types: *S. racemosa*

(most frequent), *Poligonacea* sp., *L. leucocephala*, *M. bahamensis*, *S. pallida*, and *Solanum nudum* Humb. and Bonpl. ex Dunal. The samples from the northeast and north regions are located on the upper side of PC1. The most frequent pollen types in the northeast samples were *B. simaruba*, *L. leucocephala*, *L. longistylus*, *M. bahamensis*, and *P. piscipula*, while in the samples from the north, the most frequent pollen types were *B. simaruba*, *L. longistylus*, and *P. piscipula*. The samples from the southeast are located between PC2 (main load) and PC1, and are characterized by pollen types from *B. simaruba*, *E. axilaris*, *L. longistylus*, *P. dioica*, *P. piscipula*, and *V. dentata*.

The presence of *P. dioica* was characteristic of samples from the southeast, and *S. villosa* was characteristic of those from the north. In contrast, the samples from the northeast were characterized by the presence of multiple pollen types: *B. simaruba*, *M. bahamensis*, *P. piscipula*, *S. racemosa*, and *V. dentata*. The PCA results highlight the preference of *M. beecheii* for certain floral species, and these results coincide with the diversity and floristic composition values produced in the palynological analysis.

The dendrogram (Figure 5), generated based on the most frequent pollen types in each geographical region, classified the four regions into two groups. The first group corresponds to the honey samples from the southeast and north, and the second group includes the samples from the northeast and west. The pollen types among samples in the same group were similar, but differed from those in the other group.

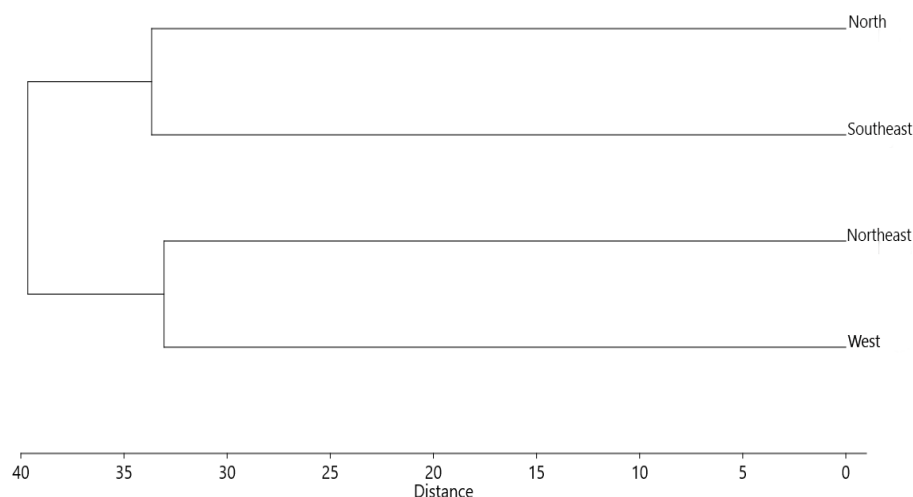


Figure 5. Dendrogram representation of Euclidean distance, based on frequent pollen types in each geographical region.

4. Discussion

The pollen content in *M. beecheii* honey samples from the state of Campeche comprised 69 pollen types belonging to 24 plant families. This total is within the 4 to 71 pollen type range reported in previous melissopalynological analyses of *M. beecheii* honey samples [18–23]. The pollen types identified in the present results are similar to those reported in most of these studies [18–22]. Although a large number of plant species were identified in the present results, of the 69 pollen types, only 14 had relative frequency values greater than 15%, the threshold for deeming a pollen source as being important [31].

Fabaceae was clearly the main plant family visited by *M. beecheii* in the four studied regions. This is to be expected, since *Melipona* sp. bees generally exhibit, across different biomes, an affinity for plants within the Fabaceae, Melastomataceae, Myrtaceae, and Solanaceae families [37–40]. Fabaceae family plants have also occurred at high levels in *M. beecheii* honey in other studies [18–22,41]. In addition, this family has been reported in honey from the stingless bee *Tetragonisca angustula* [42]. Fabaceae is the third most diverse angiosperm family in the world with over 19,400 species and 730 genera [43]. It is one of the most abundant families in Mexico, with 93 genera and 1274 species; the Yucatan

Peninsula alone is home to 78 genera and 228 species [44]. In the present results, the Fabaceae genera *Lonchocarpus*, *Mimosa*, *Piscidia*, and *Senna* were the most frequent pollen species in the honey samples. The *Mimosa* genus has been reported as a predominant pollen in *M. beecheii* honey [18–22] and is cited as an important pollen resource on the Yucatan Peninsula [45]. The *Lonchocarpus* genus has also been reported in *M. beecheii* honey and is considered a nectar source [20,21]. The genera *Piscidia* and *Senna* have been reported in *M. beecheii* honey [20] and are nectar-pollen sources on the Yucatan Peninsula [45]. It would be safe to infer that the Fabaceae family is a vital food source for *M. beecheii* in Campeche.

Another predominant pollen type in *M. beecheii* honey is *Bursera simaruba*. It has been identified frequently in *M. beecheii* honey in the state of Quintana Roo, Mexico [20]. It grows in a variety of habitats and is cited as a nectar-pollen resource on the Yucatan Peninsula [45]. The genera *Bursera*, *Lonchocarpus*, and *Piscidia* are considered abundant in the state of Campeche [46,47]. They belong to the arboreal stratum, considered typical of semi-evergreen tropical forests [48], which was the predominant strata identified in the pollen types in the *M. beecheii* honey samples (Table 2).

The floral resource analysis by geographic region indicates medium floral diversity, as corroborated by the medium-level Shannon–Wiener diversity index values (2.06–2.55) [32]. These values are similar to those reported elsewhere [18–22]. The Simpson index values (0.82–0.89) showed moderate plant species dominance in the studied regions, a result corroborated by the variable botanical origins in the four regions based on established criteria [31]; this can be seen in the fact that nine of the honey samples were multifloral and seven monofloral. Two of the monofloral samples were from the southeast region, one from the northeast, and four from the north. In previous studies, monofloral *M. beecheii* honeys were based on *Eugenia* sp., *Andira inermis* (W. Wright) DC, *Mimosa pudica* L., and *Tabebuia ochracea* A.H. Gentry [21,22,49]. In the present study, the monofloral samples were based on *B. simaruba* (southeast), *L. longistylus* (north), *S. racemosa* (northeast), *S. palida* (north), *S. villosa* (north), and *P. piscipula* (southeast and north). These results indicate regional similarities in vegetation. The southeast and north regions shared *P. piscipula*, suggesting they have similar plant community profiles. The PCA corroborated this, in that both regions were found on the positive axis and shared species such as *B. simaruba*, *L. longistylus*, and *P. piscipula*. Both also have a high degree of vegetation conservation; the samples in the southeast were collected from an area adjacent to the Calakmul Biosphere Reserve [50], and those in the north were collected near the Los Petenes Biosphere Reserve [13]. In contrast, the samples from the northeast and west regions were collected near agricultural areas [51]. Of particular note is that floral resources in the samples from the southeast and north regions, in which anthropogenic activity is minimal, exhibited greater diversity, as shown in their relatively higher Shannon–Wiener values (2.51 and 2.55, respectively) compared to the those from the northeast and west (2.17 and 2.06, respectively). These results support the conclusion that *M. beecheii* is a polylectic or generalist species, as previously reported [22]. However, some floral resources are fairly constant, though observed in different proportions in the samples; for example, *B. simaruba*, *L. longistylus*, *P. piscipula*, *Mimosa*, and *Senna*. Initially, it may seem contradictory that a generalist like *M. beecheii* would specialize, but some generalists do just that [52]. Stingless bees can be generalists in their search for resources, but they tend to consistently visit the same flower species, particularly those with dense flowers that offer high rewards [53]. The present results support this in that the different regions had medium-level Simpson index values, indicating a moderate plant species dominance. A diversity of native plant species dominated the resources identified in the *M. beecheii* honey analyzed here, but no cultivated forage plants were found, in contrast to previous reports [19,22]. The present study is the first to address the feeding habits of *M. beecheii* in different regions of the State of Campeche, Mexico, and the results strongly suggest a close relationship between *M. beecheii* and the native flora.

5. Conclusions

Melipona beecheii visited several floral resources. Overall, the predominant pollen types in the honey samples were *B. simaruba*, *L. longistylus*, *P. piscipula*, *S. pallida*, and *S. racemosa*. However, *M. beecheii* can apparently expand or alter its feeding niches in response to resource availability, since both the Shannon–Wiener diversity and Simpson indices indicated that floral resource variability was moderate in the studied geographic regions. The results confirm *M. beecheii* as being polylectic, with a preference for vegetation of the tree stratum. Especially important to note is that the *M. beecheii* honey samples indicated that this bee collects from a diversity of native plants, suggesting it has a close relationship with the native flora. The plant species identified as predominant in this study could be promising candidates for use in promoting vegetation that supports *M. beecheii* feeding strategies, and thus its conservation on the Yucatan Peninsula, specifically in the state of Campeche.

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