Cuban stingless bee livestock exhibit specialized floral resource use: a palynological study on honey samples from Matanzas and Mayabeque provinces

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Abstract
The knowledge of the different plant species that make up the feeding diet of animals is highly important to develop more efficient strategies. This research aimed to characterize the food potential available for the Cuban stingless bee livestock of the Matanzas and Mayabeque provinces. Palynological analysis was done using 60 g of pollen from sealed pots and 80 mL of honey from the ten randomly selected beehives (five in each province). The results showed that in the honey collected in Matanzas province, the most represented family was Amaranthaceae, followed by Myrtaceae and Fabaceae. Meanwhile, for Mayabeque, the most represented ones were the families Fabaceae and Myrtaceae. Regarding the stingless bee pollen of Matanzas provenance, the family Fabaceae prevailed, followed by Burseraceae and Myrtaceae. The pollen corresponding to Mayabeque coincided in showing Fabaceae as the most representative. In addition, pollen grains of small size (from 10 to 25 µm) were collected, with a marked representation of the pollen type of *Mimosa pudica* in the Mayabeque honey. It was concluded that the Cuban stingless bee livestock of the Matanzas and Mayabeque provinces had a specialist feeding behavior because a low number of plant taxa made up its diet.
With the use of this discipline, the study of the floral sources visited by bees is facilitated to a large extent (Sánchez, 2001).

The antecedents of using trophic resources by bees allow the evaluation of the plant taxa that contribute nectar and/or pollen to the bee diet. And consequently, from these data, the elaboration of floristic lists and phenological calendars can be done, contributing to better management of the beehives by farmers (Flores et al., 2015).

Due to the above-exposed facts, knowing the main plant species that make up the diet of these animals is a priority in developing efficient feeding strategies that guarantee higher production with an optimum health level. This research aimed to characterize the food potential available for the Cuban stingless bee livestock of the Matanzas and Mayabeque provinces and its possible application to other territories with beehives of this species.

Materials and Methods

Study sites

The study was conducted in two Cuban stingless bee apiaries in April 2018. The first one is located in the Matanzas municipality, of Matanzas province, at 23°02′25″ North latitude and 81°30′58″ West longitude, at an altitude of 14 m.a.s.l.; and the second one, in the San Nicolás municipality, belonging to the Mayabeque province, located at 22°47′13″ North latitude and 80°55′05″ West longitude, at an altitude of 34 m.a.s.l.

The characteristic climate of the studied area is classified as tropical, with marked seasonality of the rains in two periods corresponding to the dry season (DS), which extends from November to April, and the rainy season (RS), which lasts from May to October (Academia de Ciencias de Cuba, 1989).

During the sampling period, the Matanzas locality showed a warm climate that caused minimum temperature values and positive anomalies of 0.8°C. The recorded cumulative rainfall was 22.8 mm, lower than the historical mean (52.6 mm). Regarding relative humidity, it was 74%, over the historical values. In the case of Mayabeque, it was also warm, conditioned by the influence of high oceanic pressure that weakly influenced the territory during most of the month. This situation is responsible for the cold air masses not reaching the territory, which provoked temperature values with significant positive anomalies of 1.7 °C compared with the historical average. The minimum temperatures averaged 19.9°C. Regarding rainfall, the cumulative exceeded the historical mean of 171.3 mm, and the mean air relative humidity was 75%, slightly higher than the historical average.

Floristic inventory of each stingless bee apiary

Knowing the foraging area of bees is important to understand the use of floristic resources. Studies on species of the genus Melipona Illiger, 1806, found flight radios close to 2 km, with the studied species having body sizes larger than M. beecheii (Nunes Silva et al., 2020). However, a flight radius of 1 km was estimated for M. beecheii, similar to that of species of the genus Trigona Jurine, 1807 (Araújo et al., 2004), since they share similar biometric parameters (Fonte, 2007).

Considering that both stingless bee apiaries were located in urban areas, where gardens and house backyards prevail, the main existing plant species potentially contributing with pollen and nectar were determined: native, naturalized, and ornamental plants (Pérez-Piñeiro, 2017). In addition, to determine the representativeness of the species in the environment of stingless bee apiaries, four categories (abundant, frequent, little frequent, and rare or scarce) and their corresponding biological form (tree, shrub, and herbaceous) were established, adapting the methodology proposed by García-Abad (2015) to urban environments, which are less diverse from the botanical point of view (Wittig & Becker, 2010).

Construction of the palynotheque

From each inventoried species, floral buds were taken and preserved in a universal fixator preservative composed of a mixture of 95° ethyl alcohol, glacial acetic acid, 37-40% formaldehyde, and distilled water. Afterward, they were dissected with the aid of a surgical blade, extracting the anthers, which were placed in conical test tubes of 10 ml capacity, and the protocol used for the preparation of the samples for the palynological and melissopalynological qualitative analysis was followed.

The photographic shot of each grain from the previously identified botanical species of reference was done through a camera (LEICA DFC290) coupled to the optical microscope (NIKON OPTIPHOT-I) with a magnification of 400. The Leica Application Suite, version 3.3.1 for Windows XP, was used for this purpose.

The obtained photographs and bibliographic references (Aira et al., 2018, Pérez, 2016) served as a basis for determining pollen types found in the honey and pollen samples.

Sample collection

From the ten beehives under study in each province, five were randomly selected in each locality. In Mayabeque, beehives 14, 2, 48, 5, and 6 were chosen, while in Matanzas, beehives 1, 9, 11, 12 and 13 were selected. Sixty grams of stingless bee pollen from sealed pots and 80 mL of honey in each one of them were taken. Sampling was done in April 2018.

Melissopalynological qualitative analysis

The protocol used for sample preparation was that of honey analysis proposed by the Honey Work Group belonging to the Spanish Society of Palynology (APLE, for its initials in Spanish), recently constituted (Sánchez et al., 2018, González-Porto et al., 2018). The protocol is based, with modifications, on García et al. (2001), consisting of the previous treatment
of the samples with 5% acidulated water with their respective washings and centrifugations to obtain sediment that is in turn deposited on the microscope slide and one drop of glycerin jelly with fuchsin is applied before taking it to the microscope for its later analysis.

**Palynological qualitative analysis of stingless bee pollen**

One gram of sample was weighed and was subject to the above-explained protocol, with modifications in the volume of 5% acidulated water that was added. This methodology was used to liken it to the one used in the processing of honey samples, also considering the collection conditions of stingless bee pollen, with different load sizes, variable from small masses of 5-6 mm to dust particles.

The reading of stingless bee honey and pollen samples was done under the optical microscope with a magnification of 400, identifying a minimum number of 300 to 500 pollen grains to guarantee correct botanical and geographical characterization. The samples were done in quadruple.

The observed pollen types were placed in the different class frequencies according to Loveaux et al. (1978), as shown below:

- Dominant pollen (>45%)
- Companion pollen (16-45%)
- Important isolated pollen (4-15%)
- Rare or sporadic pollen (1-3%)
- Present pollen (<1%)

The Shannon-Weaver diversity index was used to determine the browsing strategy deployed by *M. beecheii* from the pollen diversity found in the honey samples (Shannon & Weaver, 1949).

\[
H' = - \sum_{n=1}^{5} p_i \ln p_i
\]

Where:

- **H’** = Shannon-Wiener Index.
- **p<sub>i</sub>** = Proportion of each pollen type found.
- **ln** = Natural logarithm.

Meanwhile, the browsing uniformity of the bees was determined through Pielou’s evenness Index (Pielou, 1984).

\[
J' = \frac{H'}{H'_{\text{max}}}
\]

Where:

- **J’** = Browsing uniformity of the bees.
- **H’** = Shannon-Wiener index.
- **H' <sub>max</sub>** = Napierian logarithm of pollen richness.

The diversity (H’) and evenness (J’) indices were determined through the utilization of the software Diversity Species & Richness 3.02 (Henderson & Seaby, 2002).

**Results**

The results of the floristic inventory in both localities show that in the flight environment of stingless bees, there are 16 plant species in common. We identified 42 plants belonging to 27 families in Matanzas (Table 1). *Alternanthera sessilis* (L.) R. Br. ex DC, *Mangifera indica* L., *Adonidia merrillii* (Becc.) Becc, *Bidens pilosa* L., *Bursera simaruba* (L.) Sarg, *Senna spectabilis* (DC.) H.S. Irwin & Barneby, *Musa x paradisiaca* L., *Psidium guajava* L. and *Coccoloba uvifera* (L.) L. were the most abundant.

**Table 1.** Floristic inventory of the Matanzas locality, Cuba, 2018.

| Family            | Scientific name                  | Biological form | Appearance frequency |
|-------------------|----------------------------------|-----------------|----------------------|
| Amaranthaceae     | *Alternanthera sessilis* (L.) R. Br. ex DC. | Herbaceous      | Abundant             |
| Anacardiaceae     | *Mangifera indica* L.            | Tree            | Abundant             |
| Anacardiaceae     | *Anacardium occidentale* L.      | Tree            | Rare or Scarc        |
| Annonaceae        | *Annona muricata* L.            | Tree            | Abundant             |
| Annonaceae        | *Annona squamosa* L.            | Tree            | Abundant             |
| Arecaceae         | *Cocos nucifera* L.             | Tree            | Frequent             |
| Arecaceae         | *Roystonea regia* (Kunth) O.F. Cook | Tree            | Little Frequent      |
| Arecaceae         | *Adonidia merrillii* (Becc.) Becc. | Tree            | Abundant             |
| Asteraceae        | *Bidens pilosa* L.              | Shrub           | Abundant             |
| Asteraceae        | *Viguiera dentata* Spreng.      | Shrub           | Little Frequent      |
| Bignoniaceae      | *Crescentia cujete* L.          | Tree            | Little Frequent      |
| Boraginaceae      | *Cordia collococca* L.          | Tree            | Rare or Scarc        |
| Boraginaceae      | *Bourreria virgata* (Sw.) G. Don | Shrub           | Frequent             |
| Boraginaceae      | *Cordia alliodora* (Ruiz & Pav.) Oken | Tree            | Frequent             |
| Burseraceae       | *Bursera simaruba* (L.) Sarg    | Tree            | Abundant             |
| Cactaceae         | *Nopalea auberi* (Pfeif.) Salm-Dyck | Tree            | Rare or Scarc        |
### Table 1. Floristic inventory of the Matanzas locality, Cuba, 2018. (Continuation)

| Family      | Scientific name                   | Biological form | Appearance frequency |
|-------------|-----------------------------------|-----------------|----------------------|
| Calophyllaceae | *Calophyllum antillanum* Britton | Tree            | Little Frequent      |
| Combretaceae  | *Terminalia catappa* L.           | Tree            | Frequent             |
| Commelinaceae | *Tradescantia pallida* (Rose) D.R. Hunt | Herbaceous      | Frequent             |
| Convolvulaceae | *Ipomoea triloba* L.             | Herbaceous      | Frequent             |
| Convolvulaceae | *Turbina corymbosa* (L.) Raf.   | Herbaceous      | Frequent             |
| Cucurbitaceae | *Cucurbita pepo* L.              | Herbaceous      | Little Frequent      |
| Fabaceae     | *Mimosa pudica* L.               | Herbaceous      | Little Frequent      |
| Fabaceae     | *Vachellia farnesiana* (L.) Wight & Arn. | Tree            | Little Frequent      |
| Fabaceae     | *Senna spectabilis* (DC.) H.S. Irwin & Barneby | Tree            | Abundant             |
| Fabaceae     | *Tamarindus indicia* L.           | Tree            | Rare or Scarce       |
| Fabaceae     | *Trifolium incarnatum* L.         | Herbaceous      | Frequent             |
| Fabaceae     | *Dichrostachys cinerea* (L.) Wight & Arn. | Tree            | Frequent             |
| Lamiaceae    | *Tectona grandis* L. f.          | Tree            | Rare or Scarce       |
| Lauraceae    | *Persea americana* Mill.         | Tree            | Abundant             |
| Malvaceae    | *Hibiscus rosa-sinensis* L.       | Shrub           | Frequent             |
| Malvaceae    | *Hibiscus elatus* Sw.            | Tree            | Rare or Scarce       |
| Meliaceae    | *Swietenia mahagoni* (L.) Jacq.  | Tree            | Rare or Scarce       |
| Moraceae     | *Morus alba* L.                  | Tree            | Rare or Scarce       |
| Moringaceae  | *Moringa oleifera* Lam.          | Tree            | Frequent             |
| Musaceae     | *Musa x paradisiaca* L.          | Herbaceous      | Abundant             |
| Myrtaceae    | *Psidium guajava* L.             | Tree            | Abundant             |
| Passifloraceae | *Passiflora edulis* Sims     | Herbaceous      | Little Frequent      |
| Polygonaceae | *Coccoloba uvifera* (L.) L.      | Tree            | Abundant             |
| Rutaceae     | *Citrus x aurantium* L.          | Tree            | Frequent             |
| Rutaceae     | *Citrus x limon* (L.) Osbeck     | Tree            | Frequent             |
| Sapotaceae   | *Manilkara zapota* (L.) P. Royen | Tree            | Frequent             |

In Mayabeque (Table 2), a lower number of plant species (33) was identified, grouped in 22 families. In the category of abundant are *Mangifera indica* L., *Bursera simaruba* (L.) Sarg, *Ipomoea triloba* L., *Vachellia farnesiana* (L.) Wight & Arn., *Gliricidia sepium* (Jacq.) Kunth, *Persea americana* Mill., *Psidium guajava* L., *Gouania polygama* (Jacq.) Urb. In general, in the two localities, the biological form that prevailed was the tree, followed by shrubs and herbaceous.

In the honey samples from Matanzas (Table 3) higher richness of pollen forms is observed, with a mean of 11.2 and a maximum of 12 (beehives 9 and 11). In the analyze honey from this locality, the predominant pollen percentage was that of *Alternanthera* Forrsk., with a mean of 38.26%, with the dominant pollen (>45%) in three of the five samples, followed by *Psidium guajava* L. (17.50%) and the genus *Trifolium* L. (16.07%). The most represented family was Amaranthaceae, followed by Myrtaceae, and finally Fabaceae because only one sample (beehive 9) showed pollen of *Mimosa pudica* L., justifying its inclusion in the companion category, being lower than in the other beehives.

Meanwhile, in the samples of the Mayabeque provenance (Table 4), the quantity of pollen forms or types is considerably lower, with a mean value of 7 and a maximum of 13 for the case of the honey belonging to beehive 6. It should be highlighted that the pollen from *Mimosa pudica* prevailed in all the examined honey samples, with a mean percentage of 74.05%, besides being the dominant pollen (>45%) in the five studied beehives. Fabaceae and Myrtaceae, represented by the species *Psidium guajava*, stand out as companion pollen (16-45%) of the honey obtained from beehives 2 and 48, with a mean of 23.21%.

When analyzing the foraging strategy of *M. beecheii* (Fig 1), we found that in the Matanzas honey, the mean value of the diversity index (H’) was 1.34, with a maximum value in beehive 11 (1.88). On the contrary, Mayabeque showed a lower mean (0.84). These results are in correspondence with the ones exposed in Table 4. Regarding the evenness index (Fig 1), in Matanzas, the mean value was 0.45, and again in beehive 11, the highest value was recorded (0.63). For Mayabeque, this indicator had a mean value of 0.28.
Table 2. Floristic inventory of the Mayabeque locality, Cuba, 2018.

| Family           | Scientific name                        | Biological form | Appearance frequency |
|------------------|----------------------------------------|-----------------|----------------------|
| Anacardiaceae    | Mangifera indica L.                    | Tree            | Abundant             |
| Arecaceae        | Cocos nucifera L.                      | Tree            | Frequent             |
| Arecaceae        | Roystonea regia (Kunth) O. F. Cook     | Tree            | Frequent             |
| Arecaceae        | Adonidia merrillii (Becc.) Becc.       | Tree            | Frequent             |
| Asteraceae       | Bidens pilosa L.                       | Shrub           | Frequent             |
| Boraginaceae     | Cordia gerascanthus L.                 | Tree            | Little Frequent      |
| Burseraceae      | Bursera simaruba (L.) Sarg             | Tree            | Abundant             |
| Cactaceae        | Dendrocerus medius (Engelm. ex Sauvalle) Britton & Rose | Tree | Little Frequent |
| Convolvulaceae   | Ipomoea triloba L.                     | Herbaceous      | Abundant             |
| Convolvulaceae   | Turbina corymbosa (L.) Raf.            | Herbaceous      | Frequent             |
| Ericaceae        | Lyonia myrtilloides Griseb.            | Tree            | Little Frequent      |
| Fabaceae         | Mimosa pudica L.                       | Herbaceous      | Frequent             |
| Fabaceae         | Vachellia farnesiana (L.) Wight & Arn. | Tree            | Abundant             |
| Fabaceae         | Gliricidia sepium (Jacq.) Kunth        | Tree            | Abundant             |
| Lauraceae        | Persea americana Mill.                 | Tree            | Abundant             |
| Malvaceae        | Ceiba pentandra (L.) Gaertn.           | Tree            | Little Frequent      |
| Meliaceae        | Trichilia hirta L.                     | Tree            | Little Frequent      |
| Moringaceae      | Moringa oleifera Lam.                  | Tree            | Little Frequent      |
| Myrtaceae        | Psidium guajava L.                     | Tree            | Abundant             |
| Nyctaginaceae    | Pisonia aculeata L.                    | Shrub           | Frequent             |
| Oxalidaceae      | Averrhoa carambola L.                   | Tree            | Rare or Scarce       |
| Polygonaceae     | Antigonon leptopus Hook. & Arn.        | Herbaceous      | Frequent             |
| Rhamnaceae       | Gouania polygama (Jacq.) Urb.          | Shrub           | Abundant             |
| Rosaceae         | Prunus domestica L.                    | Tree            | Frequent             |
| Rosaceae         | Pyrus communis L.                      | Tree            | Little Frequent      |
| Rubiaceae        | Coffea arabica L.                      | Shrub           | Little Frequent      |
| Rutaceae         | Citrus x aurantium L.                  | Tree            | Frequent             |
| Rutaceae         | Citrus x limon (L.) Osbeck             | Tree            | Frequent             |
| Sapindaceae      | Melicoccus bijugatus Jacq.             | Tree            | Frequent             |
| Sapindaceae      | Nephelium lappaceum L.                 | Tree            | Rare or Scarce       |
| Sapindaceae      | Serjania diversifolia (Jacq.) Radlk.   | Herbaceous      | Little Frequent      |
| Sapotaceae       | Manilkara zapota (L.) P. Royen         | Tree            | Frequent             |
| Sapotaceae       | Pouteria dicryonera (Griseb.) Radlk.   | Tree            | Little Frequent      |

![Graph](image)

**Fig 1.** Values obtained in the diversity (H’) and evenness (J’) indices of the Matanzas (1, 9, 11, 12, 13) and Mayabeque honeys (2, 14, 48, 5, 6).
According to Terrab et al. (2004), pollen grain size is one of the factors influencing higher or lower representation of plant species pollen in honey. Pollen size can be defined by the length of its polar axis and equatorial diameter, and considering the major axis, they are differentiated into several types (Erdtman, 1952). The results indicate that the Cuban

Table 4. Pollen types identified in the honeys from Mayabeque bee hives.

| Beehives | Dominant pollen (>45%) | Companion pollen (16-45%) | Important isolated pollen (4-15%) | Sporadic or rare pollen (1-3%) | Present pollen (<1%) |
|----------|------------------------|---------------------------|-----------------------------------|-------------------------------|---------------------|
| 14       | *Mimosa pudica* (71,29%) | -                         | *Psidium guajava* (11,88%)        | *Acasia farnesiana* (3,96%)   | -                   |
| 2        | *Mimosa pudica* (64,11%) | *Psidium guajava* (22,6%)  | *Moringa oleifera* (5,52%)        | *Acasia farnesiana* (1,54%)   | -                   |
| 48       | *Mimosa pudica* (70,24%) | *Psidium guajava* (23,81%) | *Mimosa pigra* (4,76%)            | *Acasia farnesiana* (1,19%)   | -                   |
| 5        | *Mimosa pudica* (78,03%) | -                         | *Mimosa pigra* (9,85%)            | *Acasia farnesiana* (1,14%)   | -                   |
| 6        | *Mimosa pudica* (86,6%)  | -                         | *Mimosa pigra* (2,9%)             | *Acasia farnesiana* (0,4%)    | Unknown (0,3%)      |

- Acasia farnesiana
- Moringa oleifera
- Psidium guajava
- Coccocuba uvifera
- Arecaceae
- *Adonidia*
- *Nopalia auberi*
- *Primitera coriacea*
- *Mangfiera indica*
- *Myrtaceae*
- *Syzygium*
stingless bee livestock of both provinces has a predilection for collecting pollen grains of small size (varying from 10 to 25 µm) and explain the highest representation of the species *M. pudica* in the Mayabeque honey (it measures 10 µm) (Figure 2).

The main floral resources used by the livestock of Matanzas province (Fig 3) were, first, *M. pudica*, with a mean percentage of 55.24%, dominant in three of the five samples, followed by *B. simaruba* (31.11%) and *P. guajava* (25.50%). Fabaceae was the most represented family in Matanzas and Mayabeque (Fig 4).

![Fig 2. Pollen grains of the prevailing types identified in the stingless bee honey and pollen samples of both provinces seen with a magnification of 400. Genus *Trifolium* (a); Genus *Alternanthera* (b); *Mimosa pudica* (c); *Bursera simaruba* (d) and *Psidium guajava* (e). Scale bar: 20 µm.](image)

![Fig 3. Main floral resources identified in the pollen collected from Matanzas province. Dominant pollen (>45%), companion or secondary pollen (16–45%), isolated pollen (4–15%).](image)
Discussion

If the criterion expressed by Pérez de Zabalza (1989) is followed, the studied honey would be classified as of low palynological richness because they represent less than 20 pollen forms. Nevertheless, this is a parameter established for European bee (Apis mellifera Linnaeus, 1758) honey in a Mediterranean environment which largely differs from the tropics. Studies by Imperatriz-Fonseca et al. (1989) and Ramalho (1990) in stingless bees showed some general foraging patterns for Meliponini, proving that, despite foraging on the wide diversity of available floral resources in the area, they are concentrated only on a few species. This would explain the obtained results.

Ramalho (2004) observed that the bees of the genus Melipona visit flowers of different shapes, sizes, and colors to obtain their food. However, there was a preference for small flowers with radial symmetry and open corollas in which access to pollen and nectar is relatively easy. Studies conducted in 18 colonies of three species of the Meliponini tribe found a prevalence of small pollen grains, suggesting the preference of these bees for small-size flowers with short pistils (Vossler, 2015).

Guimarães et al. (2021) studies confirm the results obtained in the Mayabeque honey regarding the abundance of M. pudica pollen grains (10 µm). These authors detected high representativeness of the Mimosa type in the pollen content of honey samples of Melipona seminigra pernigra Moure and Kerr, 1950, ascribable, according to Ferreira and Absy (2017), to its release in large quantities, their configuration, and the small size of pollen grains (Zappi et al., 2018).

In the case of M. beecheii, it has been reported that the type of opening or dehiscence of the anthers (Vossler, 2015) influences the preference for pollen types among the flora in the foraging area. Such preference favors collecting pollen types from Fabaceae, Melastomataceae, and Myrtaceae, which have poricidal openings (Biesmeijer, 1997). Hence, the higher representation of these families in the evaluated honey of both provenances coincides with the observation made by Novais and Absy (2015) and Costa et al. (2017) in the Meliponini tribe.

The results of the diversity index (H’) for the Matanzas honey (Fig 1) coincide with the value range from 1.23 to 2.00, obtained by Pérez-Sato et al. (2018) in A. mellifera honey produced in the High Plateau of the Puebla State in Mexico. However, they are below the ones reached by Soto (2007) for the stingless bee species Scaptotrigona pectoralis Dalla Torre, 1896, in Guatemala; and the ones reported by Rezende et al. (2020) in three Melipona species in Brazil. For this same index, the Mayabeque provenance was shown in correspondence with those obtained by Novais et al. (2013) for the stingless bee species Tetragonisca angustula Illiger, 1806, in Brazil, with ranges comprised from 0.36 to 2.55.

The diversity index (H’) is a tool that determines the breadth of the trophic niche bees through the number of botanical taxa present in the honey samples. Ferreira and Absy (2017) concluded that the breadth of the niche is determined by the movements of massive foraging of bee species with a higher number of workers, which consequently collect more trophic resources. On the other hand, colonies that have less population tend to compete with lower intensity for trophic resources, as we observed for M. beecheii in Matanzas (Table 2). Yet, both localities show a breadth of trophic niches in correspondence with the results of Hilgert-Moreira et al. (2013) for the genus Melipona Illiger, 1806.

The evenness index (J’) values were low in Matanzas (0.45) and Mayabeque (0.28). According to Espinoza-Toledo
et al. (2018), these results can be explained by the dominance of some species that causes (J’) to show a strong trend toward zero, as observed in the analyzed honey. This fact is markedly proven in the Mayabeque honey (Table 4), where the dominant species in all the samples was *Mimosa pudica*. In addition, the values reached for this index indicate that the bees from both localities showed oligolectic behavior.

According to Biesmeijer (1997), bees classified as oligolectic or specialists behave by visiting and collecting pollen from a very restricted group of plants, which applies to *M. beecheii*. On the other hand, polylectic or generalist bees utilize pollen and nectar from different plant species in their feeding. In turn, Ramalho et al. (1990) and Rezende et al. (2020) ascribe this characteristic to the fact that the bee hives of this stingless bee species are composed of a small number of individuals compared to other species. That is why they focus on few available resources and exploit them intensely.

The most represented family in Matanzas and Mayabeque (Figure 4) was Fabaceae. Similar results were obtained by Ferreira and Absy (2013) and Barquero-Eilizondo et al. (2019) in stingless bee species. They ascribe this fact to the relevance of this family within the ecosystem for its diversity (small-size plants, lianas, shrubs, and trees) and abundant flowerings throughout the year.

Within this family (Fabaceae), the species that stood out was *Mimosa pudica*, reported as a pollen source for bees of the *Melipona* genus in El Salvador (Biesmeijer, 1997), Colombia (Rodriguez, 2005) and other neotropical habitats. In Cuba, in previous studies conducted by Leal-Ramos and Sánchez (2013), they observed a high frequency of the pollen type of *M. pudica* in pollen samples obtained from the species *M. beecheii*.

Conclusions

A specialist feeding habit characterizes the Cuban stingless bee livestock of the Matanzas and Mayabeque provinces. It has a diet based on a few plant species, among which *Bursera simaruba* (L.) Sarg., *Mimosa pudica* L., *Psidium guajava* L., *Trifolium* spp., and *Alternanthera* spp. stand out. In addition, a marked preference for small pollen grains from 10 to 25 µm was observed, especially those of *Mimosa pudica* in the Mayabeque honey.

Acknowledgments

The authors acknowledge the support of the Swiss Cooperation and Development Agency (SDC) for its contribution to funding this research through the project BIOMAS-CUBA, in its stage III.

Likewise, they thank the Pastures and Forages Research Station Indio Hatuey, the University of Matanzas, and the Palynology and Plant Conservation Group (GPCV) of the Instituto Hispano Luso de Investigaciones Agrarias (CIALE) of the University of Salamanca.

Authors’ Contribution

LFC: Conceptualization, methodology, investigation, software, validation, formal analysis, resources, data curation, writing-original draft, writing-review & editing, visualization, supervisión, project administration.

WLP: Conceptualization, investigation, resources

JDL: Conceptualization, investigation, resources

MDS: Conceptualization, investigation, resources

DMQ: Conceptualization, investigation, software, resources.

DRC: Methodology, investigation, software, validation, formal analysis, resources, writing-original draft, writing-review & editing.

JSS: Methodology, investigation, validation, resources.

ESR: Methodology, investigation, validation, resources.

References

Academia de Ciencias de Cuba. (1989). Nuevo Atlas Nacional de Cuba. Instituto Cubano de Geodesia y Cartografía. La Habana, Cuba. p. 41.

Aira, M.J., Chávez, M.A., Fernández-González, M. & Rodríguez-Rajo, F.J. (2018). Pollen diversity in the atmosphere of Havana, Cuba. Aerobiologia, 34: 389-403.

Araújo, E.D., Costa, M., Chaud-Netto, J. & Fowler, H.G. (2004). Body size and flight distance in stingless bees (Hymenoptera: Meliponini): inference of flight range and possible ecological implications. Brazilian Journal of Biology, 64: 563-568.

Barquero-Eilizondo, A.I., Aguilar-Monge, I., Méndez-Cartín, A.L., Hernández-Sánchez, G., Sánchez-Toruño, H., Montero-Flores, W. & Mesén-Montano, I. (2019). Asociación entre abejas sin aguijón (Apidae, Meliponini) y la flora del bosque seco en la región norte de Guanacaste, Costa Rica. Revista de Ciencias Ambientales, 53: 70-91.

Biesmeijer, J.C. (1997). Abejas sin aguijón. Su biología y organización de la colmena. Elinkwijk BV, Utrecht.

Costa, C.C.A., Silva, C.I., Maia-Silva, C. Limão, A.A.C. & Imperatriz-Fonseca, V.L. (2017). Origem botânica do mel da jandaíra em áreas de caatinga nativa do Rio Grande do Norte. In: V.L. Imperatriz-Fonseca, D. Koedam, M. Hrnčir (Eds.), A abelha jandaíra no passado, no presente e no futuro (pp. 161-166). Mossoró: Ed. Uferas.

Erdtmann, G. (1952). Pollen morphology and plant taxonomy. Angiosperms. Stockholm: Almqvist and Wiksell, 539 p.

Espinoza-Toledo, C., Vázquez-Ovando, A., Torres de los Santos, R., López-García, A., Albores-Flores, V. & Grajales-Conesa J. (2018). Stingless bee honeys from Soconusco, Chiapas: a complementary approach. Revista de Biología Tropical, 66: 1536-1546.

Ferreira, M.G. & Absy, M.L. (2013). Pollen analysis of the post-emergence residue of *Melipona (Melikerria) interrupta*...
La tierra” y sus “tenedores”, las colmenas y la miel que producen, en zonas de las provincias occidentales de Cuba. Degree work. Facultad de Medicina Veterinaria. La Habana: Universidad Agraria de la Habana, 81 p.

García, M., Pérez-Arquillue, C., Juan, T., Juan, M.I. & Herrera, A. (2001). Note. Pollen analysis and antibacterial activity of Spanish honey. Food Science and Technology International, 7: 155-158.

García-Abad, J.J. (2015). Abundancia relativa, frecuencia y riqueza de plantas vasculares a escala local. Metodología de Índices de Ocupación de la Flora (aplicada a La Alcarria Occidental). Estudios Geográficos, 76: 499-530.

González-Porto, A.V., Sánchez Reyes E., De Linares Fernández C., Rodríguez de la Cruz D., Sánchez Sánchez J., Belmonte Guía para autores. AGRO Productividad, 11: 98-104.

Henderson, P.A. & Seaby, R.H. (2002). Species Diversity and Richness 3.02. Lymington, UK: Pisces Conservation Ltd, http://www.pisces-conservation.com. [12/03/18].

Hilgert-Moreira, S.B., Nascher, C.A., Callegari-Jacques, S.M. & Blochtein, B. (2013). Pollen resources and trophic niche breadth of *Apis mellifera* and *Melipona obscurior* (Hymenoptera, Apidae) in a subtropical climate in the Atlantic rain forest of southern Brazil. Apidologie, 45: 129-141.

Imperatriz-Fonseca, V.L., Kleinert-Giovannini, A. & Ramalho, M. (1989). Pollen harvest by eusocial bees in a non-natural community in Brazil. Journal of Tropical Ecology, 5: 239-242.

Louveaux J., Maurizio A. & Vorwhol G. (1978). Methods of Melissopalynology. Bee World, 59: 139-157.

Novais, J.S., Absy, M.L. & dos Santos, F.D.A.R. (2013). Pollen grains in honeys produced by *Tetragonisca angustula* (Latreille, 1811) (Hymenoptera: Apidae) in tropical semi-arid areas of northeastern Brazil. Arthropod-Plant Interactions, 7: 619-632.

Novais, J.S. & Absy, M.L. (2015). Melissopalynological records of honeys from *Tetragonisca angustula* (Latreille, 1811) in the Lower Amazon, Brazil: Pollen spectra and concentration. Journal of Apicultural Research, 54: 11-29.

Pérez, K. (2016). Determinación del origen botánico de mieles de *Melipona beecheii* Bennett (Apidae, Meliponini) en meliponarios de la provincia de Mayabeque. Degree work. La Habana: Facultad de Agronomía. Universidad Agraria de La Habana, 51 p.

Pérez-Piñeiro, A. (2017). La apicultura en cuba y su situación actual. Agroecología, 12: 67-73.

Pérez-Sato, M., Flores-Garrido, A.F., Castro-González, N.P., Escobar-Hernández, R., Soni-Guillermo, E. & Pérez-Hernández, H. (2018). Análisis palinológico de la miel de *Apis mellifera* L., producida en el Altiplano del estado de Puebla, México. Guía para autores. AGRO Productividad, 11: 98-104.

Pérez de Zabalza, A. (1989). Estudio palinológico de las mieles de Navarra. Doctoral Thesis. Navarra: Universidad de Navarra. 268 p.

RAE. (2017). Ganado. Madrid: Real Academia Española. http://dle.rae.es/?id=IpmVKGf.

Ramalho, M. (1990). Foraging by stingless bees of the genus *Scaptotrigona* (Apidae, Meliponinae). Journal of Apicultural Research, 29: 61-67.

Ramalho, M., Kleinert-Giovannini, A. & Imperatriz-Fonseca, V. L. (1990). Important bee plants for stingless bees (*Melipona*...
and *Trigonini* and Africanized honeybees (*Apis mellifera*) in neotropical habitats: a review. Apidologie, 21: 469-488.

Ramalho, M. (2004). Stingless bees and mass flowering trees in the canopy of Atlantic Forest: a tight relationship. Acta Botanica Brasiliaca, 18: 37-47.

Rezende, A.C.C., Absy, M.L., Ferreira, M.G. & Marinho, H.A. (2020). Honey botanical origin of stingless bees (Apiidae Meliponini) in the Nova América community of the Sateré Mawé indigenous tribe, Amazon, Brazil. Grana, 59: 304-318.

Rodríguez, A. (2005). Estudio del forrajeo de polen por obreras de *Melipona fasciata* (Hymenoptera, Apidae, Meliponini) en el piedemonte llanero colombiano (Acacias-Meta-Colombia). IV Seminario Mesoamericano sobre abejas sin aguijón. Chalatenango, El Salvador.

Sáenz, C. & Gómez, C. (2000). Mieles españolas. Características e identificación mediante el análisis de polen. Madrid: Ed. Mundi prensa. 129 p.

Sánchez, L. (2001). Métodos palinológicos. Curso de capacitación. PROMABOS. San Salvador, El Salvador.

Sánchez, E., Rodríguez, D., Sánchez, S., Vega, E. & Sánchez, J. (2018). Sobre la metodología en los estudios melisopalinológicos. Coimbra: V Congreso Ibérico de Apicultura.

Shannon C.E. & Weaver W. (1949). The mathematical Theory of Communication. University of Illinois Press, 117 p.

Soto, M.A.V. (2007). Recursos polínicos utilizados por la abeja nativa Shuruya (*Scaptotrigona pectoralis*) (Apiidae: Meliponini) en un meliponario de la parte baja de los cipresales en Pachalum, Quiché, durante la época seca y lluviosa. Doctoral Thesis. Facultad de Ciencias Químicas y Farmacia, Universidad de San Carlos de Guatemala. 73 p.

Terrab, A., Pontes, A., Heredia FJ. & Diez, M.J. (2004). Palynological and geographical characterization of avocado honeys in Spain. Grana, 43: 116-121.

Vossler, F.G. (2015). Small pollen grain volumes and sizes dominate the diet composition of three South American subtropical stingless bees. Grana, 54: 68-81.

Wittig, R. & Becker, U. (2010). The spontaneous flora around street trees in cities – A striking example for the worldwide homogenization of the flora of urban habitats. Flora-Morphology, Distribution. Functional Ecology of Plants, 205: 704-709.

Zappi, D.C., Gastauer, M., Ramos, S., Nunes, S., Caldeira, C. F., Souza-Filho, P.W., Guimarães, J.T.F., Giannini, T.C., Viana, V.P.L., Lovo, J., Mota, N.F.O. & Siqueira, J.O. (2018). Plantas nativas para recuperação de áreas de mineração em Carajás. Belém, PA: Instituto Tecnológico Vale (ITV). 282 p.