(GS09) Mellisopalynology

Date: August 28
Place: Room 6326 (poster)

Poster Presentation

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GS09-P01 (271)
Pollen composition of *Apis mellifera* L. honey samples from Bahia, Brazil

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The objective of this study was to determine the pollen composition of *Apis mellifera* L. (Hymenoptera, Apidae) honeys from the municipality of Alagoinhas, Bahia State, Brazil. It is also purposed to contribute to the knowledge of the regional melliferous flora. Honey samples commercialized in 2010 by the Beekeepers' Cooperative and coming from the localities of Sauípe,

Área dos Fornos, Poços, Macaquinho, Boa União, Entrada da Capineira, Lagoa Seca, Ponto do Beiju, Uruba, Matadouro, UNEB and Rio Branco were submitted to standard melissopalynological methods, using acetolysis. Five slides were prepared from each sample. The resulting sediment was analyzed, the pollen types were identified based on reference slide collection of the Laboratory of Palynological Studies at Bahia State University and specialized catalogues. For qualitative and quantitative analyses, a minimum of 1,000 pollen grains were counted per sample. The pollen types were grouped into frequency classes: predominant pollen (PD, > 45%), secondary (AP, 15-45%) and minor (PI, < 15%). The pollen composition of the analyzed honeys showed 51 pollen types distributed among 26 plant families. In this, the families with higher contribution of pollen types in the samples were: Asteraceae, Fabaceae/Mimosoideae (six pollen types each), Myrtaceae (four), Anacardiaceae, Rubiaceae and Lamiaceae (three pollen types each). The pollen types *Eucalyptus* (Myrtaceae) and *Mimosa sensitiva/pudica* (Fabaceae/Mimosoideae) occurred in all analyzed samples. *Eucalyptus* pollen type was predominant pollen in more than 50% of the samples. Some pollen types, as Bignoniaceae, *Citrus*, Cucurbitaceae, *Cuphea tenella*, *Mimosa gemmulata*, *Mimosa misera*, Myrtaceae, Nyctaginaceae, *Salvia*, Simaroubaceae and Verbenaceae were reported to one sample only. The locality with higher diversity of pollen types was Sauípe and the lower diversity was found at Boa União. *Mimosa* was an important genus into the honey samples. It presented five specific pollen types (*M. caesalpiniifolia*, *M. gemmulata*, *M. misera*, *M. quadrivalvis* and *M. sensitiva/pudica*). These most expressive pollen types found in the samples represent different trophic sources visited by *Apis mellifera* in the studied localities.

Keywords: bees, type pollen, melissopalynology, pollen spectrum.

GS09-P02 (378)
First study on pollen concentration in honeys of native bees from Central Amazon, Brazil

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Melissopalynological studies have been carried out in different countries looking for identifying plants species often used as bee pasture. At Brazilian Amazon, Melissopalynology is still incipient. However, that biome presents more than 100 stingless bees' species contributing to pollination services. The purpose of this work is to present the first data on pollen concentration in honeys produced by a native bee from two areas in Central Amazon, Brazil. The samples were harvested in colonies of *Tetragonisca angustula* Latreille (Apidae, Meliponina) sited in the municipalities of Belterra and Santarém, Low Amazon region, Pará State (PA). Acetolysis method was used for all samples. To estimate the pollen concentration, a tablet with spores of an exotic marker, *Lycopodium clavatum* L. (18,583), was added into each sample, before acetolysis. To establish the frequency classes, pollen types found in the processed material were identified and counted (500 per sample, at least). Honey samples from Belterra presented a total number of pollen grains in 10g of honey (TNP-10gr) from 19,388 (minimum) to 149,172 (maximum). Analyzed honeys from this municipality were included among the following Maurizio's Categories: I (< 20,000 pollen grains/10gr), II (20,000 - 100,000) or III (100,000 - 500,000). Honeys from Santarém presented TNP-10gr between 224,507 and 787,072. These samples were included in the Categories III and IV (500,000 - 1,000,000). Results still showed 26 plant families contributing to pollen associations in *T. angustula* honeys from Belterra. Eleven families contributed to honeys' composition in Santarém. Fabaceae was the most representative plant family among the samples, showing 17 different pollen types. *Cecropia* (Urticaceae) and *Spermacoce capitata* (Rubiaceae) pollen types were recognized as

predominant pollen (>50%) in some analyzed samples. *Clathrothropis nitida* (Fabaceae), *Clidemia* (Melastomataceae), *Machaerium* (Fabaceae), *Microtea* (Phytolaccaceae), *Senna* (Fabaceae) and *Tapirira guianensis* (Anacardiaceae) occurred as secondary pollen types (16 - 45%). Most of these plant species can be easily found in the studied areas. Ruderal plants have demonstrated to be useful for bees in different ecosystems around the world. The importance of Fabaceae as a bee plant has been inferred by several authors in palynological studies of honeys and bee pollen. Also native species from Amazon were represented by their pollen types in the analyzed samples. This reflects the contribution of the regional flora with potential to help beekeepers and their apicultural activities. Additional studies are required to increasing the knowledge on pollen content in honeys from Brazilian Amazon stingless bees.

Keywords: melissopalynology, bee flora, meliponiculture, stingless bee, exotic marker.

GS09-P03 (77)
Pollen diversity in honey samples from Sergipe state, Brazil

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The analysis of the pollen content of bee products has the main objective of identifying their botanical origin. In this study we dealt with the palynological analysis of *Apis mellifera* honey samples from Sergipe state, Brazil. We aimed identifying their botanical origin and findind possible geographical markers. Pollen sediments of 22 honey samples from 22 municipalities were processed using the acetolysis method. To determine the frequency class of the pollen types in each sample, at least, 500 pollen grains were counted and grouped in the following categories: predominant pollen (>45%), secondary pollen (16-45%), important minor pollen (3-15%), minor pollen (1-3%) and trace pollen (<1%). A total of 90 pollen types distributed in 28 plant families were identified. Fabaceae was the family with highest number of pollen types (29 types), followed by Asteraceae (8), Rubiaceae (7), Anacardiaceae and Malvaceae (4 each), and Euphorbiaceae and Myrtaceae (3 each). The presence of predominant pollen was detected in 14 samples: *Mimosa pudica/sensitiva* in five samples, *Alternanthera* in two samples, *Angelonia*, *Cocos nucifera*, *M. arenosa*, *M. tenuiflora*, *Myrcia*, *Prosopis juliflora*, *Protium* and *Schinus* in one sample each. Eleven pollen types were recorded as accessory pollen: *Borreria verticillata*, *M. pudica/sensitiva*, *Tapira* in three samples, *Mitracarpus hirtus* in two samples, *Angelonia*, *Baccharis*, *Cocos nucifera*, Melastomataceae, *Protium*, *Salvia* and *Senna macranthera* in one sample each. Most samples (21) were characterized as heterofloral honey. Pollen types from endemic species to the caatinga vegetation were found in the pollen spectrum from Sergipe state: *Mimosa adenophylla*, *M. ophthalmocentrica*, *M. setuligera* and *M. xiquexiquensis* (Fabaceae), and *Ziziphus joazeiro* (Rhaminaceae). Honey samples analyzed can be labeled as organic honey produced in Northeastern Brazil.

Keywords: melissopalynology, *Apis mellifera,* Brazil, bee products, pollen types.

GS09-P04 (317)
Botanical identification and total phenolic content in propolis from northern coastal region Bahia, Brazil

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This study dealt with propolis samples (eight) from northern coastal region of Bahia, Brazil. Pollen spectrum and total phenolic content in the samples were assessed. Spectrophotometric quantification (740 nm) of total phenol content was obtained by the Folin-Ciocalteau's method. To pollen analyses, propolis was diluted in alcohol, and the organic sediment was acetolysed. Pollen grains were identified by comparison with pollen atlas and pollen slides from the Palynotheca of the Laboratory of Plant Micromorphology at Universidade Estadual de Feira de Santana. Samples from municipality of Inhambupe presented values of total phenolic content from 7.68±2.58 to 36.78±1.52 mg/g EqAG. Samples from municipality of Itapicuru presented values from 9.98±1.34 to 18.43±0.78 mg/g EqAG. Phenolic data are lower than those found in propolis from other Brazilian regions. Samples from Inhambupe presented 25 pollen types (11 plant families) and 34 pollen types (16 families) in those from Itapicuru. The most representative plant family among the samples from both municipalities from northern coastal region of Bahia was Asteraceae with six pollen types. *Eucalyptus* (Myrtaceae) and *Mimosa pudica* (Fabaceae-Mimosoideae) were the most representative pollen types in the samples from Inhambupe and Itapicuru, respectively. The following pollen types were found into all analyzed samples from Inhambupe: *Alternathera ramosissima*, Asteraceae, *Borreria verticillata*, *Eucalyptus*, *Mikania*, *Mimosa pudica* and *Mimosa tenuiflora*. All samples from Itapicuru showed *Mikania*, *Mimosa pudica*, *Mimosa tenuiflora*, *Mitracarpus longicalyx*, *Myrcia* I and *Syagrus* pollen types. Some pollen types were observed in all propolis samples (*Mikania*, *Mimosa pudica* and *Mimosa tenuiflora*). Representatives of resiniferous plant species (Schinus type) is present in 25% of samples.

Keywords: Brazil, pollen, melissopalynology, bee flora.

GS09-P05 (215)
Brazilian Semiarid honeys – melissopalynology study

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The greatest honey producers from Brazil are in semiarid region (Northeast), taking advantage in respect of the others producing regions for its diversity vegetation and presence of native forests, free of pesticides, which enable the production of organic honey with high potential of commercialization. In this context, the pollen analysis research provides data for the identification of botanical and geographical origin of honey, contributing at aggregation of value to the product as well as promoting the improvements of its conditions of production and utilization of resources. This study focused to characterization of the botanical (pollinic) origin of honey samples from the state of Piauí. Honey samples (23) were submitted to dissolution (alcohol) to concentrate pollen sediment, which was acetolysed and analyzed under optical microscope. The countings of pollen grains used the methodology of the exotic marker (*Lycopodium clavatum* L. spores). About the classification according to classes of pollen concentration, 14 samples were classified in category I (<20,000 grains), five in category II (20,000 to 100,000 grains), two in category III (100,000 to 500,000 grains) and two in category V (> 1,000,000 grains), pointing to the low quantity of pollen grains in the semiarid honeys from Piauí. A total of 140 pollen types were identified, belonged to 30 families. Fabaceae was a special family because of its 53 pollen types – 31 of Mimosoideae subfamily. The

pollen types *Borreria verticillata*, *Cecropia*, *Mimosa acutistipula*, *Mimosa arenosa*, *Mitracarpus scabrellus*, *Piptadenia moniliformis* and *Ricinus communis* were classified as predominant pollen in at least one sample; 16 pollen types were pollen accessory by at least one of the samples, including the types *Croton* and *Syagrus* which are characteristic from semiarid vegetation. The most part of pollen types was included at isolated pollen class. The pollen type *Borreria verticillata* was the most frequent, present in 78% of the samples. As an evidence of honeys from Brazilian semiarid region is the presence of two or more of the following types: *Alternanthera ramosissima*, *Borreria verticillata*, *Mimosa pudica*, *Mimosa tenuiflora/verrucosa* and *Piptadenia moniliformis*.

Keywords: Brazil, Piauí, Caatinga, Pollen.

GS09-P06 (422)
Palynological data characterisation of Estonian honey

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There are many reasons why the botanical and geographical origin of a honey may be wanted to be known. For instance, for quality control in marketing and because of legislations that requires correct descriptions on labels. There are around 300 beekeepers in Estonia maintaining about 30000 bee colonies and total honey yield is ~500-600 tons annually. More than 1000 forage plants are known in Estonia. Although the beekeeping tradition has a long history, there has never been a systematic pollen analytical study of Estonian honey. The goal of the present study is to report the results of qualitative pollen analyses from honey samples collected from 2000 to 2011. The main focus is in the floristic spectrum of plants and in the identification of the most important plant sources for honey pollen. Also the changes in the plant compositions and pollen concentrations of honey are considered during the investigation period. Altogether the pollen content of more than 400 honey samples were analysed with an average of 400 pollen grains counted from a sample. Results showed that pollen types *Salix* spp., Rosaceae and Brassicaceae family were the most abundant among the samples. Pollen of *Calluna* type, Apiaceae and Fabaceae, were also present. During the study period, the most marked change observed was that the percentage of the *Calluna* pollen type had decreased, while the proportions of Brassicaceae and *Salix* spp. pollen types showed an increase. The concentration of pollen grains per gram of honey vary a lot from 100 to 100000 (average 6400). Typical Estonian honey is polyfloral, the average number of species is 13 taxa per sample (maximum 28). The current information about Estonian honey may be used to develop analytical standards for pollen content of honey, contributing to quality control of a product offered for export or for the home market. In addition, more research is needed on the Estonian honeys typical for our region in order to investigate their melissopalynological, physico-chemical and organoleptical properties.

GS09-P07 (587)
Combining melissopalynology and flavonoid analysis for the identification of litchi- and longan-honeys in Taiwan

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Litchi (*Litchi chinensis*) and longan (*Euphoria longan*) are two of most important nectariferous plants for monofloral honeys in Taiwan. Owing to high similarity in pollen morphology and overlapping of flowering time in some areas, it is difficult to distinguish either kind of honey. In order to set up a method for identifying them, in this study, we collected 37 litchi honeys, 70 longan honeys, 9 mixtures of litchi and longan honeys, 35 other honeys and 2 foreign longan honeys (from Vietnam and Thailand) from March to May, 2011. Traditional melissopalynological analysis was applied to construct the pollen profiles. In addition, flavonoids were extracted from honey samples by solid-phase extraction, followed by HPLC analysis detected at 280 nm. Data were combined for chemometric analysis to find out the qualitative or semi-quantitative botanical markers. In studied samples, there were 88 pollen types belonging to 43 families. Although it's difficult to distinguish litchi pollen from longan pollen, pollen profiles of litchi/longan honeys were readily differentiated from other honeys. Our results suggest that the threshold for monofloral litchi/longan honeys could be set at 50%. Both litchi- and longan-honeys displayed three characteristic peaks (A, B, D) in HPLC-chromatogram. Semi-quantitative analysis showed that the peak D was lower for Litchi-honeys (0.4%-21.6%, Md = 1.1%), whereas higher for longan- honeys (4.8%-52.0%, Md = 24.9 %). The results of principal component analysis supported that A, B, D peaks could be used as the botanical markers of litchi and longan honeys.

Keywords: melissopalynology, flavonoid, litchi honey, longan honey, botanical marker.