Plants Used by Bees as Pollen Sources in the Brazilian "Cerrado"

by

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ABSTRACT

Bee flora is the set of plants that supply food to bees in a given region. The recognition of the plants used by bees for the collection of pollen in order to enable adequate handling by the beekeeper and the improvement of the bee pasture are considered of great importance for a good beekeeping. Some bee pollen samples were obtained monthly during one year from pollen collectors installed in five beehives in two different areas ("cerradão" and "cerrado" *sensu stricto*) in Pirassununga, SP, Brazil. It was noted that species of the Asteraceae family were intensely visited by the *Apis mellifera* bees, indicating the importance of them for an eventual bee flora recomposition. Alternativelly bees visited nearby areas, using mainly *Eucalyptus* sp. and *Citrus* sp. for the collection of pollen.

Key words: *Apis mellifera*, food, protein, bee plant

INTRODUCTION

Bee pollen is the result of the agglutination of flower pollen made by bees by means of the addition of salivary substances and small quantities of nectar or honey (Brasil 2001).

Pollen besides being important for the nutrition of bees due to its physicochemical composition (Funari *et al.* 2003; Barreto *et al.* 2005; Marchini *et al.* 2006). Their quality and composition can change adult bee behavior, their fecundity, life span, brood area size, honey stock, colony population and food larval quality (Toth *et al.* 2005; Human *et al.* 2007, Matttila & Otis 2007) .

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Beekeepers can harvest part of the pollen collected by the worker bees, and for that they use collectors, traps or pollen hunters which withdraw their pollen loads in a pollen basket before the bees penetrate the beehive (Barreto *et al.* 2005).

Bee pollen can be used as food complement by humans and animals. The nutritional importance of the pollen for humans is recognized, as being a protein source of high biological value, also containing carbohydrates, lipids and minerals in its composition. Pollen does not only contain antioxidant vitamins (A, C and E), but also those of the complex B and vitamin D (Oliveira *et al.* 2009, Melo & Almeida-Muradian 2010; Lopes *et al.* 2011).

The botanical origin of the pollen is of extreme importance for the definition of its physicochemical composition, and it is necessary to know the pollen spectrum of the collected product in order to establish the relation between the pollen composition, its physical characteristics and its botanical origin. According to Modro *et al.* (2007) the nutritional components are correlated to the frequency of the specific pollen types and indicate the importance of the collection from several pollen sources for the maintenance of the richest and balanced diets.

The objective of the present study was to identify the pollen sources used by the bees, aiming at a more appropriate handling of the local "cerrado" bee vegetation and to provide to beekeepers alternative botanical species for the recomposition of the bee flora.

MATERIAL AND METHODS

This research was carried out in remaining "cerrado" areas of the Campus of the University of São Paulo, Pirassununga County, State of São Paulo, Brazil, located within the coordinates 21° 57 ' 02 " S, 47° 27 ' 50 " W and 630 m above sea level.

The total area of the Campus contains around 2.300 ha, occupied by different faces of the "cerrado" ecosystem, also known as "savannah", located besides areas of cultivation and pastures. The preserved forests ("cerrado" *sensu stricto*, "cerradão" and riparian forest) correspond to 30 % of the total area (about 705 ha), while the pastures and cropland areas make up approximately 70 % of the total area (1.620 ha).

Two different areas were evaluated: 1- "cerradão" with approximately 56 ha,

being a secondary forest, where the anthropic action has been controlled over more than 20 years, without fire incidence during this period. In this area the soil is classified as a Dark-Red Latosol (Pirassununga-Plio-Pleistocene Formation), textures varying from clayey to sandy-clay, with the canopy of the vegetation reaching heights of 15 to 20 m; 2- "cerrado" *sensu stricto,* characterized by a shrubby flora, with at most 6m height, including lianas and several herbaceous species. This fragment has around 93 ha and the soil is a Red-Yellow Latosol with textures varying from clayey-sandy to sandy.

Fina & Monteiro (2009) observed a quite varied floristic composition in the same "cerradão" area with 730 individuals distributed among 33 families, 60 genera and 80 species. The richest families are Myrtaceae, Lauraceae and Euphorbiaceae; and the most important species are *Copaifera langsdorfi, Anadenanthera falcata* and *Siparuna guianensis* and, according to the literature, several of these plants are sources of nectar and pollen for different bee species.

A total of 5 swarms (in each area) of *Apis mellifera* were collected, transferred to Langstroth beehives, and maintained in the same place of origin. During one year, pollen collections were performed at the entrance of the beehives; collectors of pollen were set, monthly, at the entry of the hives. Pollen samples were homogenized and 1 g of each was taken for preparation of slides.

For slide preparation (3 replicates per sample), the Acetolysis method (Erdtman 1952) was adopted, which consists of the chemical treatment of pollen grains, removing their intine, cytoplasm and adhering substances, fossilizing them artificially in order to make the exine most transparent and more appropriate for the study of pollen details. The identification of the pollen types was made based on referential slides made with the pollen of the plants collected in the areas and also in slides already existent in the laboratory of the Apiculture Sector of the ESALQ/USP, Piracicaba, SP, Brazil, in which the work was carried out.

Qualitative analysis – through this analysis the botanical species were determined (or pollen types), considering morphological aspects of the grains comparing them with referential slides.

Quantitative analysis – was performed by means of counting 300 to 500 grains of pollen per sample and grouping them by botanical species and/or pollen types. The pollen types were grouped in four classes of frequency. In

other words, dominant pollen (> 45%) (DP), accessory pollen (15 to 45%) (AP), isolated important pollen (3 to 15%) (IIP) and isolated occasional pollen (< 3%) (IOP) (Louveaux *et al*. 1978).

With the data of pollen types from the "cerradão" and "cerrado" s*ensu stricto)*, the Similarity Index (SI) of Sorensen was calculated according to Silveira Neto *et al.* (1976), by the expression: SI = 2j/(a+b), where:

j= number of collected pollen types common to both: "cerradão" and "cerrado" *sensu stricto.*

a = number of collected pollen types observed in the samples from the "cerradão"

b = number of collected pollen types observed in the samples from the "cerrado" *sensu stricto.*

SI varies from 0 to 1, the closer to 1, more similar are the communities and the closer to 0, less similar they are.

RESULTS AND DISCUSSION

With the quantitative analyses of the samples of the pollen loads it was possible to demonstrate the importance of the plant species in the production of honey, classifying them in the above mentioned frequency classes DP, AP, IIP, and IOP (Tables 1 and 2).

The following types of DP (Figure 1) were observed: *Baccharis* sp. (Asteraceae), *Baccharis dracunculifolia* (Asteraceae), *Brachiaria* sp. (Poaceae) and Asteraceae type only in the "cerradão"; *Alternanthera ficoides* (Amaranthaceae), *Eucalyptus* sp. (Myrtaceae) and *Vernonia polyanthes* (Asteraceae) only in the "cerrado" *sensu stricto, and Eupatorium* sp. (Asteraceae), *Didymopanax vinosum* (Araliaceae), *Mikania* sp. (Asteraceae) and *Citrus* sp. (Rutaceae) in both areas. It was also noted that bees visited nearby areas with *Eucalyptus* sp. and *Citrus* sp. for pollen collection.

The most frequent plant species evaluated through the pollen from the collectors installed in the beehives of the "cerradão" were: *Mikania* sp., which bloomed during four months of the collections; and *Citrus* sp.1 and *Didymopanax vinosum*, for three months. For the "cerrado" *sensu stricto*: *Eupatorium* sp. and *Didymopanax vinosum*, appeared during five months of the collections; *Citrus* sp.1, for four months and *Mikania* sp.*, Alternanthera ficoides* and *Wissadula subpetala*, three months.

Table 1. Pollen spectrum of the pollen samples, originating from the collectors installed from the beehives of the "cerradão" area, Pirassununga, SP, Brazil.

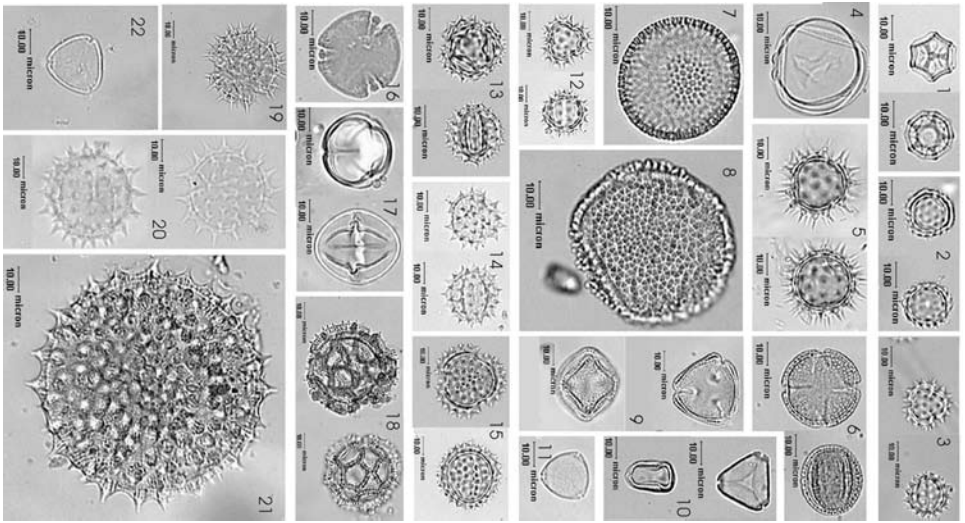
Botanical specie or Pollen type	Pollen spectrum/month											
	Jan.	Feb.	Mar.	Apr.	May	Jun	July	Ago	Sept	Oct	Nov	Dec
Amaranthaceae - *Alternanthera ficoides*	-	-	-	-	AP	-	-	-	-	-	-	-
Araliaceae - *Didymopanax vinosum*	-	-	DP	IOP	IIP	-	-	-	-	-	-	-
Arecaeae - *Astrocaryum aculeatissimum*	-	-	-	-	-	-	-	-	-	-	-	IOP
Asteraceae - *Baccharis dracunculifolia*	-	DP	-	-	-	-	-	-	-	-	-	-
Asteraceae - *Baccharis* sp.	DP	-	-	-	-	-	-	-	-	-	-	-
Asteraceae - *Bidens gardnerii*	-	-	IIP	-	IIP	-	-	-	-	-	-	-
Asteraceae - *Eupatorium* sp.	-	-	-	-	-	DP	-	-	-	-	-	DP
Asteraceae - *Mikania* sp.	-	-	-	AP	-	AP	DP	IOP	-	-	-	-
Asteraceae - *Vernonia cognata*	-	-	-	-	-	-	-	IIP	-	-	-	-
Coniferae - *Pinus* sp.	-	-	-	-	-	-	-	-	-	-	-	AP
Euphorbiaceae- *Alchornea* sp	-	-	-	-	-	-	-	-	-	DP	-	-
Euphorbiaceae - *Croton floribundus*	-	-	-	-	-	-	-	-	-	-	-	AP
Euphorbiaceae - *Croton urucurana*	AP											
Fabaceae-Caesalpinioidea - *Caesalpinia* sp.	-	-	-	-	-	-	IOP	-	-	-	-	-
Fabaceae-Mimosoidea - *Piptadenia moniliformis*	IOP	-	-	-	-	-	-	-	-	-	-	-
Lamiaceae - *Hyptis eriophylla*	-	-	IIP	-	-	-	-	-	-	-	-	-
Malvaceae - *Wissadula subpetala*	-	-	-	AP	-	-	-	-	-	-	-	-
Myrtaceae - *Eucalyptus* sp1.	-	IIP	-	-	-	-	-	-	-	-	-	-
Myrtaceae - *Eugenia aurata*	-	-	-	-	-	IOP	-	-	-	-	-	-
Myrtaceae - *Eugenia bimarginata*	-	-	-	AP	-	-	-	-	-	-	-	-
Piperaceae – *Piper* sp.	-	-	-	-	-	-	-	-	-	AP	DP	-
Poaceae - *Brachiaria* sp.	AP	-	-	-	DP	-	-	-	-	-	-	-
Rutaceae - *Citrus* sp1.	IOP	-	-	IOP	-	-	-	-	DP	-	-	-
Solanaceae - *Solanum* sp.	-	AP	-	-	-	-	-	-	-	-	-	-
Asteraceae type	-	-	-	-	-	-	-	DP	AP	-	-	-
Brassicaceae type	-	-	-	-	-	-	-	IIP	-	-	-	-
Poaceae type	-	-	IOP	-	-	-	-	-	-	-	-	-
Rubiaceae type	-	-	-	IIP	-	-	-	-	-	-	-	-
Rutaceae type	-	-	-	-	-	-	-	-	-	-	IIP	-

DP = dominant pollen (> 45%), AP = accessory pollen (15 to 45%), IIP = isolated important pollen (3 to 15%) and IOP = isolated occasional pollen (< 3%).
-No pollen or pollen type

Table 2 - Pollen spectrum of the pollen samples, originating from the collectors installed from the beehives of the "cerrado" *sensu stricto* area, Pirassununga, SP, Brazil.

Botanical specie or Pollen type	Pollen spectrum/month											
	Jan.	Feb.	Mar.	Apr.	May	Jun	July	Ago	Sept	Oct	Nov	Dec
Amaranthaceae - *Alternanthera ficoides*	-	-	-	AP	DP	AP	-	-	-	-	-	-
Araliaceae - *Didymopanax vinosum*	IOP	IIP	-	AP	IOP	DP	-	-	-	-	-	-
Arecaeae - *Astrocaryum aculeatissimum*	-	-	-	-	-	-	-	-	-	IOP	IOP	-
Asteraceae - *Eupatorium* sp.	-	DP	DP	-	-	IOP	-	-	-	-	DP	DP
Asteraceae - *Mikania hirsutissima*	-	-	-		-	-	-	-	-	AP	-	-
Asteraceae - *Mikania* sp.	-	-	-	DP	AP	-	-	IOP	-	-	-	-
Asteraceae - *Montanoa* sp.	-	AP	-	-	-	-	-	-	-	-	-	-
Asteraceae - *Vernonia cognata*	-	-	-	-	-	-	-	-	AP	-	-	-
Asteraceae - *Vernonia polyanthes*	-	-	-	-	-	-	-	DP	-	-	-	-
Asteraceae - *Vigueira* sp.	AP	-	-	-	-	-	-	-	-	-	-	-
Bombacaceae - *Eriotheca gracilips*	-	-	-	-	IOP	-	-	-	-	-	-	-
Convolvulaceae - *Merremia macrocalix*	-	-	-	-	-	-	-	IIP	-	-	-	-
Euphorbiaceae - *Croton floribundus*	-	-	-	-	-	-	-	-	-	IIP	IIP	-
Euphorbiaceae - *Croton urucurana*	AP	-	-	-	-	-	-	-	-	-	-	-
Fabaceae-Mimosoidea - *Piptadenia moniliformis*	-	-	-	-	-	-	-	-	-	IIP	-	-
Malvaceae - *Sida* sp.	-	IIP	-	-	-	-	-	-	-	-	-	-
Malvaceae - *Wissadula subpetala*	-	-	IIP	-	-	-	-	IIP	-	IOP	-	-
Myrtaceae - *Eucalyptus* sp1	-	-	-	-	-	-	DP	-	-	AP	-	-
Poaceae - *Brachiaria* sp.	AP	IIP	-	-	-	-	-	-	-	-	-	-
Poaceae - *Zea mays*	-	-	-	-	-	-	-	-	-	-	IIP	-
Rutaceae - *Citrus* sp1.	IOP	-	-	-	-	-	IIP	-	DP	-	-	IOP
Sapindaceae - *Serjania* sp.	-	-	-	-	-	-		-	-	-	-	-
Solanaceae - *Solanum* sp.	-	-	AP	-	-	-	-	-	-	-	-	-
Brassicaceae type	-	IIP	-	-	-	-	-	-	-	-	-	-
Malvastrum type	-	-	-	-	-	-	-	-	IIP	-	-	-
Myrcia type	-	-	-	-	-	-	-	-	-	-	-	AP

DP = dominant pollen (> 45%), AP = accessory pollen (15 to 45%), IIP = isolated important pollen (3 to 15%) and IOP = isolated occasional pollen (< 3%).
-No pollen or pollen type



1-*Alternathera ficoides* (Amaranthaceae), 2-*Baccharis* sp., 3-*Baccharis dracunculifolia* (Asteraceae), *4-Brachiaria* sp. (Poaceae), 5-*Bidens gardnerii* (Asteraceae), 6-*Citrus* sp1. (Rutaceae), 7-*Croton floribundus*, 8-*Croton urucurana* (Euphorbiaceae), 9-*Didymopanax vinosum* (Araliaceae), *10-Eucalyptus* sp1. (Myrtaceae), 11-*Eugenia bimarginata*(Myrtaceae), *12-Eupatorium* sp (Asteraceae), 13-*Mikania* sp., 14-*Mikania hirsutissima* (Asteraceae), 15-*Montanoa* sp. (Asteraceae), 16- *Alchornea* sp (Euphorbiaceae), *17-Solanum* sp. (Solanaceae), 18-*Vernonia cognata, 19-Vernonia polyanthes* (Asteraceae), 20-*Vigueira* sp. (Asteraceae), 21-*Wissadula subpetala* (Malvaceae) and 22- Myrcia type(Myrtaceae).

Fig. 1. Pollen types which appeared as dominant or accessory pollen in samples of the pollen loads collected in area of "cerradão" and "cerrado" *sensu stricto* of Pirassununga, SP, Brazil.

Comparing the two areas, 15 pollen types occurred in both: three belonging to the Asteraceae family, two to the Euphorbiaceae and one of each of the following families, Amaranthaceae, Araliaceae, Arecaceae, Brassicaceae, Fabaceae, Malvaceae, Myrtaceae, Poaceae, Rutaceae and Solanaceae. The other 25 identified pollen types (Tables 1 and 2) were observed either in the "cerrado" *sensu stricto* or in the "cerradão". The Similarity Index of Sorensen for the two areas was the same, IS = 0.545, which indicates that the collection of pollen by bees has a medium similarity between the two forms of vegetation. For other bush areas, located in different Brazilian states, the similarity index was very low for plants visited by solitary bees and stingless bees (Moreti *et al*. 2006).

In the distribution of the pollen types by family (Figure 2), it is noticed that the greatest diversity of species belongs to the Asteraceae family with 12 pollen types, followed by Myrtaceae with four. Costa (2002- unpublished data) investigating pollen sources used by worker bees of *Apis mellifera* in Cruz das Almas, BA, Brazil, also observed the Asteraceae as the family with the greatest number of pollen types and the same was observed by Almeida-Anacleto (2007) in Piracicaba, SP, Brazil. In ten pollen samples of the south region of Brazil, Almeida-Muradian *et al.* (2005) observed that the most frequent families of plants were Arecaceae, Asteraceae and Myrtaceae, with two families of plants coincident with the observed ones in the present study. In "cerrado" areas of Cassilândia, MS, Brazil differently than in the here studied area, the Malpighiaceae and the Fabaceae (Mimosoideae) were the families with the greatest number of species of importance for the bees (Vieira *et al.* 2008).

Through the analyses of the studied pollen samples, a great participation of isolated and occasional pollen types, was found in several samples. Barth (1970) mentions that these species have little importance, when considering the quantity of nectar and pollen supplied by the plant; however, when the interest is the origin and geographical provenance of the samples the presence of these pollen types becomes significant.

CONCLUSION

Species of the Asteraceae family are intensely visited by the *Apis mellifera* bees for pollen collection in the São Paulo state, both in the "cerrado" *sensu*

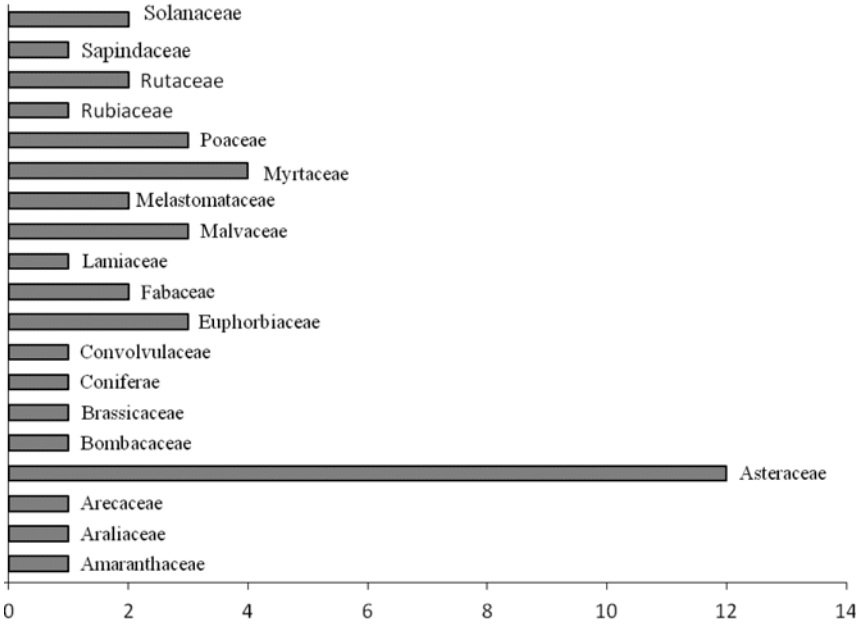


Fig. 2. Number of pollen types by botanical family observed in the samples of pollen loads collected in beehives of *Apis mellifera* in the "cerrado" areas of Pirassununga, SP, Brazil

stricto and in the "cerradão" areas, indicating that this family is very important for a recomposition of the bee flora.

It was noted that besides the existing plants occurring in the "cerrado" s*ensu stricto* and in the "cerradão" areas, the bees visited nearby areas where the beehives were installed, making use mainly of *Eucalyptus sp*. and *Citrus* sp., for pollen collection.

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