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Digestibility of pollen of grains by worker honeybees, *Apis mellifera* (Hymenoptera, Apidae, Apini) in the microregion of Viçosa, MG, Brazil

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ABSTRACT: The digestibility of pollen grains consumed by *Apis mellifera* workers during one year in the microregion of Viçosa, MG, Brazil was analyzed. In total 80% was digested, 5% was undigested and 15% was broken pollen. Therefore the useful rate by bees was high and there was not significant difference among the three hives sampled suggesting that there is not relationship between the low honey flow period with the use of protein resources at Viçosa microregion. The predominant pollen species during the year were of families Myrtaceae and Poaceae. Sometimes the pollen consumed by worker bees were different from those found in the flowers on a determined year period which is to be expected as young bees feed on pollen grains stored in the hive.

Key words: Pollen grains, digestion, honeybee.

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INTRODUCTION

The nutritional requirements of worker honeybees (*Apis mellifera* L. 1758) vary during their adult lives. Young workers require a rich protein diet, consuming more pollen, essential for the development of their hypopharyngeal glands, while the forager bees consume more honey and nectar, rich in sugars and therefore, supplying energy for the foraging activity (HAYDAK, 1970; SERRÃO & CRUZ-LANDIM 1995). Pollen is the main source of nitrogen for the colony, as it is rich in proteins, and also contains lipids, sugars, mineral salts and vitamins (FREE, 1980). However the pollen grains are covered by a wall of cellulose and esporopollenin, substances for bees difficult to digest, because they do not produce cellulases (GROGAN & HUNT, 1979, SCHUMAKER *et al*., 1993). MAURIZIO (1954) and ROUBIK (1989) suggests that the pollen grains present different nutritional values with the protein content varying of 15 - 65% according to the species.

In the microregion of Viçosa (MG, Brazil), previous studies have identified low honey flow from September to December. Thus, the purpose of this work was to study the digestibility of pollen grains ingested by nurse workers of *A. mellifera* during one year in the Viçosa region to determine whether the bees have different gains of protein resources during this period of low nectar flow.

MATERIALS AND METHODS

Every fortnight (August 1998 to July 1999) five bees were collected from each of three *A. mellifera* hives at the Central Apiary of the Universidade Federal de Viçosa, State of Minas Gerais, Brazil. These bees were collected in the brood rearing area of the hive and are assumed to be nurse workers.

Each of the 390 bees was dissected and the rectum isolated and transferred to 4% paraformaldehyde at phosphate buffer 0.1M, pH 7.2 for 4h. The material was placed on slides, the wall of the rectum was disrupted, its contents stained with 1% methylene blue and mounted in glycerine jelly for observation

under a light microscope. For each rectal content three areas were scanned and an average of 190 pollen grains per bee was classified and quantified according to their digestibility following the classification proposed by CRUZ-LANDIM & SERRÃO (1994)

Fifteen groups of rectum were selected from the samples obtained during the experiment. These were submitted to the acetolysis method for identification of the pollen species. The identification was accomplished by the comparison with photographed pollen grains in the Universidade de Taubaté (UNITAU), Central Apiary (UFV) and PIRANI & CORTOPASSI-LAURINO (1993) which permitted the identification of about 98% of pollen grains found in the samples.

The data were pooled for statistical analysis using Kruskal-Wallis test at a significance of 5%.

RESULTS

The pollen grains observed in the rectum of *A. mellifera* workers were classified as digested (empty or with their contents retracted in the center of the grain), undigested (full grains) or broken (Fig. 1).

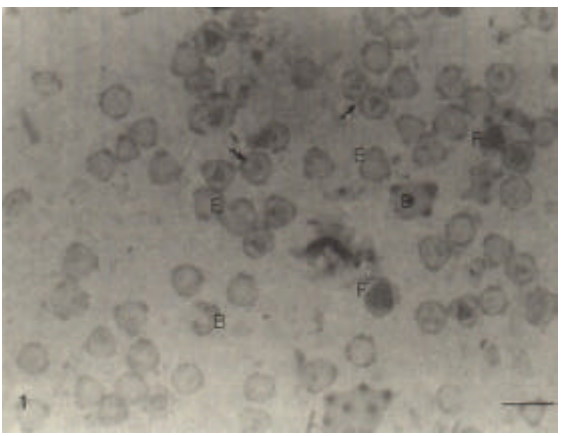

Figure 1. Different aspects of pollen grains found into the rectum of a young worker of *Apis mellifera*.: Broken (B), filled (F), Empty (E) and with content retract to the center of the grain (arrows). Bar = 50 m

Full grains were not considered as useful for bees, because they had not undergone digestion while the grains partially digested and the empty were taken as useful for bees because they had undergone digestion. Broken pollen grains were considered as an indeterminate profit because it was unknown where the release of their content occur, if in the digestive tract of bees or in the storage cells as a consequence of bees manipulation.

The digestibility condition of pollen grains during the low honey flow period from August to December corresponded to 78.1% of digested pollen, 6.7% of undigested pollen and 15.2% of broken pollen, data similar to those obtained during the high honey flow period from January to July (Fig. 2). It is also observed that in the entire period analyzed, undigested pollen corresponded to 14.3%.

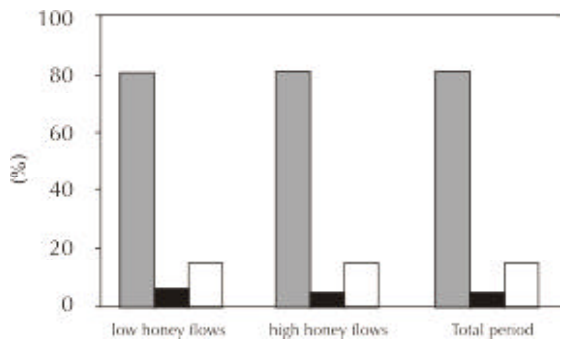

Figure 2. Mean of digestibility condition of pollen grains found into the rectum of young workers of *Apis mellifera* during the periods of low honey flow (August/1998 to Deceber/1999), high honey flow (January/1999 to July/1999) and the total periode (August/1998 to July/1999) at the three nests analyzed. ▒ Digested pollen grains. ■ Undigested pollen grains. □ Broken pollen grains.

Analysis of the digestibility condition of pollen grains per nest was represented by proportions in relation to the totality of the counted grains of each nest and showed that there was not significant difference among the nests, having clearly predominance of pollen grains that undergone digestion at the three nests sampled (Fig. 3).

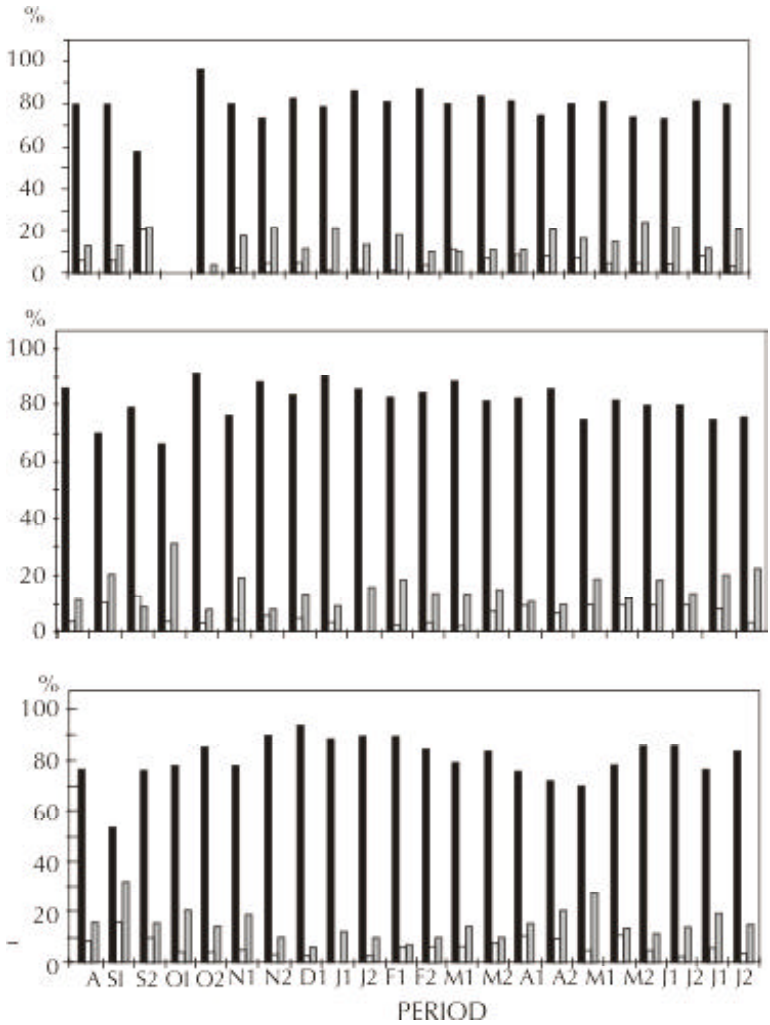

Figure 3. Digestibility condition of pollen grains found into the rectum of young worker of Apis mellifera at the three nests analyzed (A, B, C). ▒ Digested pollen grains. ■ Undigested pollen grains. □ Broken pollen grains. Numbers following the month corresponds to fortnight of collect of bees.

Table 1 summarize the pollen species found in the rectum of bees during the period studied. At the low nectar flow period, in August and September was found greater amount of pollen grains of Myrtaceae, Euphorbiaceae (*Mabea fistulifera*) and

Asteraceae (*Vernonia* sp). In October and November the pollen grains eat by bees were represented by Leguminoseae, Myrtaceae and Asteraceae, being the later found only in November. In December was observed higher amount of pollen grains of Asteraceae, Leguminoseae and Poaceae with a decrease of pollen grains of Myrtaceae. On the other hand, analysis of the entire period studied showed that Myrtaceae and Poaceae were the plant families with higher amount of pollen grains eat by bees.

Table 1. Pollen species found into the rectum of young workers of *Apis mellifera* during one year (August/1998 to July/1999).

Plant	Aug/ 98	Sep/ 98	Oct/ 98	Nov/ 98	Dec/ 98	Jan/ 99	Feb/ 99	Mar/ 99	Apr/ 99	May/ 99	Jun/ 99	Jul/ 99
Asteraceae		++		++	++	+			+	+	+	++
Convolvulaceae				+		+	+	+	+	+	+	+
Convolvulaceae - *Ipomea* sp.	+	+			+			+				
Euphorbiaceae - *Croton* sp.				+	+		+		+			
Euphorbiaceae - *Mabea fistulifera*		++	+						++	++	+	
Labiateae - *Hyptis* sp.									+		+	
Leguminoseae	+	+			++	+			+			
Leguminoseae - *Crotalaria* sp.			++	++								
Leguminoseae - *Desmodium* sp.												+
Leguminoseae - *Mimosa velosiana*						++			+		+	
Leguminoseae (Mimosoideae)			++	++							+	
Malvaceae											+	++
Moraceae							++	+				
Moraceae - *Cecropia* sp.							++	+	+		+	
Myrtaceae	++	++	++	++	+	+	+	++	++	++	++	++
Poaceae			+		++	++	++	++	++		++	+
Poaceae- *Panicum* sp.	+	+								+		
Rubiaceae	+	+							+			
Sapindaceae		+										+
Solanaceae				+							+	
Solanaceae - *Solanum* sp.					+		++		+	+		
Tiliaceae - *Triunpheta* sp.							+	+	+			
Verbenaceae- *Lantana* sp.	+											

+ few pollen grains. ++ many pollen grains.

DISCUSSION

Our results showed that digestibility rate of pollen grains during one year was high, contrasting with data obtained by CRUZ-LANDIM (1985) who observed that 50% of the pollen grains ingested by *A. mellifera* pass the digestive tract intact, which may be due to the species of pollen grains consumed, because we have observed that some types of pollen grains were not digested by the bees. On the other hand, FERNANDES-DA-SILVA & SERRÃO (2000) and ZERBO et al. (2001) found that in the stingless bee *Scaptotrigona postica*, digestibility rate of pollen grains was about 90%, a similar rate found by us.

Pollen grains can be found empty in the field, yet in the flowers and ones could hypothesizes that the high rate of digested pollen in the bee gut would be a misinterpretation. In this sense, in *A. mellifera* stored pollen undergoes a process of lactic acid fermentation (CHEVTCHENIK, 1950) and seems to have an associated microflora, suggesting that microorganisms are involved in this process (HEBERT & SHIMANUKI, 1978; GILLIAM, 1979; SOMMEIJER & BRUIJIN, 1994) which helps in enhancing the digestibility of pollen through production of some enzymes by the microorganisms (MACHADO, 1971; GILLIAM et al., 1990). However, in preliminary studies we observed that pollen grains stored in the hive were almost all filled (about 90%) without digestion signs, as observed in *S. postica* by FERNANDES-DA-SILVA & SERRÃO (2000) who have pointed out that pollen fermentation acts to the pollen's acquisition of certain organoleptic properties and/or serve to enhance preservation of stores in tropical environments. This hypothesis is corroborated by results herein presented, because digestibility rate was constant along of the year.

It is interesting to notice that not always the pollen consumed by the bee coincides with those found in the flowers in a period, e. g., the pollen of *Mabea fistulifera* was present into the bee rectum on September, while the time of blooming of this plant occurs between March and June. This results was already expected because the young bees do not realize external flights, feeding with those pollen grains stored in the cells of the beehive.

Analysis of pollen species consumed by bees showed that Myrtaceae and Poaceae were the main plant source visited by *A. mellifera*. This may be due to the localization of the three hives in the apiary, because they were placed near to a fragment of secondary rain forest Mata Atlântica, where many species of these families are present.

Variation of bloom and nectar flows period in the Viçosa region has few relation with the uses of the protein resource, because the different types of pollen grains consumed presented a digestibility rate always high (about 80%) without significant difference between pollen grains consumed during the both low and high honey flow periods.

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