

## Magnetic Material Arrangement In *Apis Mellifera* Abdomens

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### ABSTRACT

Honeybees are the most studied insects in the magnetic orientation research field. Experiments on the magnetic remanence of honeybees have shown the presence of magnetite nanoparticles, aligned transversely to the body axis on the anterodorsal abdomen horizontal plane. These results support the hypothesis of ferromagnetic sensors for the magnetoreception mechanism. An Electron Paramagnetic Resonance (EPR) study identified isolated magnetite nanoparticles and aggregates of these particles with a low temperature transition (52 K – 91 K). Hysteresis curves of *Apis mellifera* abdomens organized parallel and perpendicular to the applied magnetic field were obtained from 5K to 310K. At low temperatures, the hysteresis curves indicate a preferential orientation of the magnetic easy axis parallel to the body axis. The saturation ( $J_s$ ) and remanent ( $J_r$ ) magnetizations, coercive field ( $H_c$ ) and initial susceptibility ( $\chi$ ) were obtained. Results were interpreted based on the presence of magnetite nanoparticles with 50 K and 120 K mean blocking temperatures.

### INTRODUCTION

The honeybee, *Apis mellifera*, is the most studied insect in the magnetic orientation research field. Different experiments aiming the localisation of sensory magnetic particles in honeybees were reviewed [1]. Studies of the magnetic properties of honeybee have shown the presence of magnetite superparamagnetic or single domain nanoparticles in the anterodorsal abdomen region. Remanence measurements demonstrate that adult honeybees and older pupae possess magnetic material aligned transversely to the body axis on their horizontal plane while dead bees induced remanence tends to be aligned with the magnetic applied field. The transverse horizontal orientation could be the result of crystals that are forced to grow aligned north-south in the geomagnetic field, the most common comb natural orientation. Moreover, each honeybee showed different natural remanence [2-5].

Based on the ferromagnetic hypothesis for magnetoreception, Electron Paramagnetic Resonance (EPR) studies have contributed to the identification of magnetic material [6,7]. The presence of isolated magnetite nanoparticles of about  $3 \times 10^2 \text{ nm}^3$  and  $10^3 \text{ nm}^3$  depending on the hydration degree of the sample, and aggregates of these particles was proposed. A low temperature transition in the 52 K – 91 K region was reported [6].

A lack of hysteresis data yield us to focus on the analysis of the temperature dependence of hysteresis curves of *Apis mellifera* abdomens oriented parallel and perpendicular to the applied magnetic field of the magnetometer.

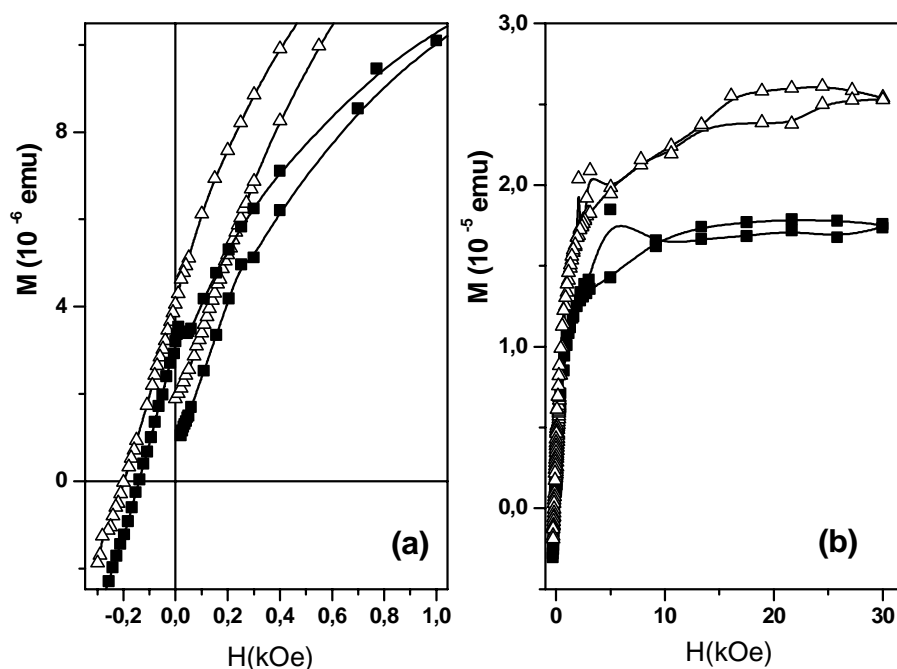
## EXPERIMENTAL DETAILS

Natural dead bees found at the entrance of a hive in Itaborai, Southeast of Brazil were collected and kept in 80%(v/v) ethanol. Bees were extensively washed with this solution. The three first segments of 16 bee abdomens were separated with stainless steel scalpel blades to avoid contamination. Two samples with eight abdomen segments each were oriented one with the axis body parallel and the other perpendicular to the magnetic field.

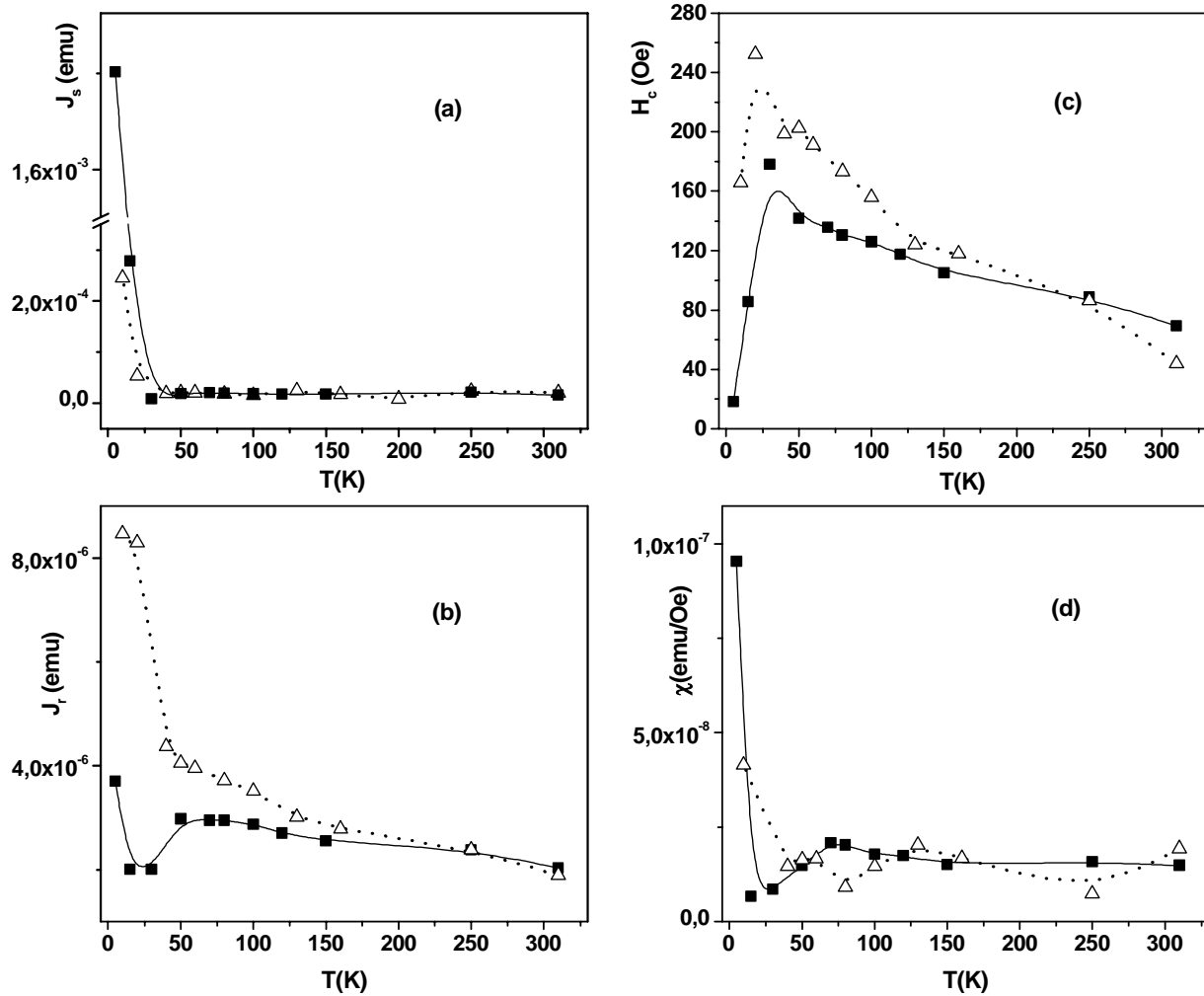
Hysteresis curves were obtained using a SQUID magnetometer (Quantum Design) from 5 K to 310 K for fields up to 30 kOe in the first positive branch, and up to 500 Oe in the negative branch. A straight line dominates the curves with a negative or positive slope depending on the temperature range and orientation of the sample. Linear fits from 10 kOe to 30 kOe were performed using Origin<sup>R</sup> software. The angular coefficient was taken as an effective dia-paramagnetic susceptibility and used to subtract the dia-paramagnetic contribution from the hysteresis curves.

## RESULTS

The hysteresis loop obtained at 50 K for the two sample orientations in the low field region are shown in Figure 1. One branch of each curve including the saturation region is presented in the insert. The saturation ( $J_s$ ) and the remanent ( $J_r$ ) magnetizations, the coercive field ( $H_c$ ) and initial susceptibility ( $\chi$ ) are obtained from these curves.



**Figure 1.** (a) Low Field region of hysteresis curves at 50 K. (b) First branch of hysteresis curves. Abdomen segments of eight bees oriented perpendicular (full square) and parallel (open triangle). Lines are guides to the eyes.



**Figure 2.** Temperature dependence of magnetic parameters. (a) Saturation magnetization,  $J_s$ , (b) Remanent magnetization,  $J_r$ , (c) Coercive field,  $H_c$  and (d) Initial susceptibility,  $\chi$ . Abdomen segments of eight bees oriented perpendicular (full square) and parallel (open triangle). Lines are guides to the eyes.

The temperature dependence of these parameters from 5 K to 310 K is plotted in figure 2 for both orientations. From these data it is evident that the magnetic parameters depend on the orientation of the magnetic field. This suggests that the magnetic particles in bee abdomens are preferentially aligned in specific directions rather than being arranged isotropically. The parallel and perpendicular  $J_s$  values are in the same magnitude range for temperatures above 50 K, but with weak oscillations (not visible in the scale of figure 2a) not observed in the perpendicular direction. Below 50 K there is a splitting of the curves, and at 10 K  $J_s$  in the perpendicular orientation is eight times larger than  $J_s$  in the parallel direction. Similar oscillations are also observed for  $\chi$  (Figure 2d) in the same temperature range with a 2.5 ratio between the two orientations at 10 K (even considering this parameter uncertainty). In contrast,  $H_c$  and  $J_r$  values are anisotropic for a wide range of temperatures (lower than about 200 K) shown in Figures 2b,c). The slope changes (obtained from derivative curves) appear at around 120 K and 50 K for

$H_c$  and  $J_r$ . At 20-30K  $H_c$  curves present a maximum while  $J_r$  curve behaviors are opposite with a minimum for the perpendicular orientation.

## CONCLUSIONS

The almost indistinguishable  $J_s$  values observed above 50 K are consistent with the previous results of dead bee magnetization tracking the applied magnetic field. At lower temperatures, the higher values for  $J_s$  when the magnetic field is applied perpendicular to the body axis suggests that magnetic moments are aligned perpendicular to the body axis as found for live bees [2].

The anomalous increase of  $J_s$  and  $J_r$  could be associated with superparamagnetic particles that stabilize with a low blocking temperature. This is supported by smooth changes on the  $J_r$  and  $H_c$  curves indicating mean blocking temperatures of about 50K and 120 K related to 20 nm and 26 nm particle diameters [8]. Low temperature transitions in smashed bee abdomens were also observed by EPR, 60–91 K for natural samples and 52-72 K for lyophilised ones. At temperatures lower than 25 K the background EPR component changes to an unexpected asymmetric line shape [6]. This result can be related to the peak at around 25 K shown in  $H_c$  curve which is possibly influenced by oxidized surface as observed in nanostructured Fe grains [9]. These magnetic properties presenting several temperature range behaviors could be due to different magnetic nanoparticle system arrangements. This paper presents original results on the magnetic material in bee abdomens it points to the complexity of this system that requires complementary experiments and theoretical developments, especially with respect to 310 K, the bee body temperature.

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