

FMR measurements in fire ants: evidence of magnetic material.

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Abstract

Based on the behavioral studies and the localization of iron-containing tissue fire ants were examined by EPR for magnetic material. Results suggest the presence of magnetite particles.

Key words: fire ants, biomineralized magnetic material , EPR

Adaptability of living beings to the environment yields to a complex navigation or orientation systems. This complexity is related to a variety of information sources as the pattern of polarized light, terrestrial landmarks (Wehner et al, 1983), geomagnetic field (Kirschvink, 1985), etc. The geomagnetic field influence has been studied in organisms from bacteria to human beings (Kirschvink, 1985). Results in fire ants, *Solenopsis invicta*, have proposed to add geomagnetic orientation as a mechanism that contributes to the success of this species in foraging (Anderson and Vander Meer, 1993). In addition localisation of subcuticular iron-containing tissue in fire ants was reported (Slowik and Thorvilson, 1996). Although recent attempt to replicate Anderson and Vander Meer's results failed (Klotz et al, 1997), their suggestion of a possible magneto-kinetic effect keeps stimulating further studies on magnetic materials biomineralized by fire ants.

Electron Paramagnetic Resonance (EPR) seems to be a convenient technique to study ferromagnetic materials (Raikher and Shliomis, 1994). As far as we know there is only one paper using this method in ant studies (Krebs and Benson, 1965), where the ant species investigated were divided in two groups: one with a strong, relative narrow EPR signal in the free radical region and the second which gave a strong 6-peak signal attributed to the Mn^{++} ion. There was no attempt to investigate ferromagnetic material.

Fire ants (*Solenopsis sp.*) from Citrolândia, Rio de Janeiro and from Venda Nova, Espírito Santo, both in the Southeast Brazil were used for EPR experiments. Samples of smashed and desiccated ants were transferred to sealed EPR tubes and followed by nitrogen flux. EPR was performed with an X-band spectrometer (Bruker ESP300E) at room temperature. Spectra from literature were digitised on a DaVinci, Digigraf digitiser after amplification. Frequency and field parameters corrections were taking into consideration for spectral comparisons, using the software Winspec.

The complex spectra of fire ants are composed of different lines. The presence and the intensity of these lines depend on the samples: ant origin, colony localisation or preparation. Moreover spectra are dependent on the sample position in the cavity and on the orientation relative to the magnetic field. Experiments were performed on samples of different colonies and repeatedly checked for several batches of the same colony. Fig 1 shows representative spectra of ants from Venda Nova (Fig 1(a)) and from Citrolândia (Fig 1(d)).

A very broad line spread from zero field up is observed in Figs 1(a) and (d). Fig 1(a) is dominated by this line but a resonance in the region of $g=2$, although much less intense, is still observed. In the region of $g=2$ three lines are present. A very narrow at $g=2.02$ is observed in all spectra of ants. It is superimposed on a another one of approximately 350G easily seen in Fig 1(a). A wider one, with g about 2.2 and 1300G linewidth is predominant in spectra of Fig 1(d). Some spectra additionally present a superimposed 6-peak structure with peak-to-peak distances of about 100G as in Fig 1(a).

Most spectra (not shown) contain also a signal in the region of $g=4.3$ which is usually assigned to the presence of not aggregated Fe^{3+} in a site of low simetry. It can be observed in Fig 1(a).

In general, biomineralized magnetic materials in living beings are identified as magnetite particles (Kirschvink, 1985). Natural ferro (ferri)magnetic minerals can be distinguished by their EPR spectra features (Ikeya, 1993). Then the interpretation and

identification of the spectra obtained with fire ants were based on previous EPR studies of mineral and synthesised magnetite. The zero-field component of Fig 1(a) could be related to spectrum of Fig 1(b) of natural mineral magnetite large particles (Ikeya, 1993). Similar signals to the components in the region of $g=2$ were observed for small magnetite particles. A $g=2.013$ line 400G width was observed for samples of magnetite with 3.5nm average diameter (Aharoni and Morton, 1971) (Fig 1(c)) is associated to the component in Fig 1(a). EPR measurements on ultrafine magnetite particles films show that a wide ferromagnetic resonance became to be observed at particle sizes larger than 3.4nm while a narrow one may be related to the precursory state of the latter (Bandow and Kimura, 1991). Fig 1(e) shows the spectra obtained for 3.4nm particles with the magnetic field parallel to the magnetite film ($g\sim 2.1$ and 1000G linewidth) to be compared with the component of Fig 1(d).

The central narrow line in spectra of ants can be due to either a precursory state or/and the presence of free radicals resulting of several biological processes. Finally the 6-peak structure can be associated to a manganese ion (Mn^{++}) as suggested by Krebs and Benson (Krebs and Benson, 1965).

These results strongly suggest the presence of magnetite particles in fire ants. Furthermore it could be inferred coexistence of different particle sizes. Magnetite particles with diameters smaller than 3.5nm are known to present superparamagnetic behaviour (Bandow and Kimura, 1991). Presence of such particles were reported in abdomen of honey bee (*Apis mellifera*) (Schiff, 1991), summing up, the results in fire ants (*Solenopsis invicta*) (3,4) support magnetite to be involved in fire ant geomagnetic sensing, even though their experimental results were not replicated by Klotz et al (Klotz et al, 1997).

Complexity is a characteristic of biological processes that turns magnetic studies in living beings hard to be performed. In this scene these are the first results on magnetic material in fire ants using EPR. This open a wide range of future analysis on properties of biomineralized magnetic material in insects. In particular it can contribute to the understanding of ants behavior which were extensively studied (Hölldobler and Wilson, 1990).

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REFERENCES

- Aharoni S M, Morton H L (1971) Superparamagnetism and Exchange Anisotropy in Microparticles of Magnetite Embedded in an Inert Carbonaceous Matrix. J. Appl. Phys. 42 (1): 352-356.
- Anderson J B, Vander Meer R K (1993) Magnetic Orientation in Fire Ant, *Solenopsis invicta*. Naturwissenschaften 80: 568-570.
- Bandow S, Kimura K (1991) Disappearance of Long-Range Spin-Order in Ultrafine Magnetite Particles. Zeitschrift für Physik D, 19 (1-4): 271-273.
- Hölldobler B, Wilson E O (1990) The Ants. Harvard University Press. Cambridge, Massachusetts.
- Ikeya M (1993) Silica and Silicates: geotherm and Volcanism. In: Zimmerman M R, Whitehead N (eds) New Applications of Electron Spin Resonance Dating, Dosimetry and Microscopy. World Scientific, Singapore New Jersey London Hong Kong

- Kirschvink J L, Jones K S, MacFadden B J (1985) *Magnetite Biomineralization and Magnetoreception in Organism: a New Magnetism*. 1st edn Plenum Press, New York.
- Klotz J H, Lonnie L, Van Zandt L L, Reid B L, Bennett G W (1997) Evidence Lacking for Magnetic Compass Orientation in Fire Ants (Hymenoptera: Formicidae): *J. Kansas Entomol. Soc.* 70 (1), 64 - 65.
- Krebs A T, Benson B W (1965) Electron Spin Resonances in Formicidae: *Nature* 207 (5004), 1412-1413.
- Raikher Y, Shliomis M I (1994) The Effective Field Method in the Orientational Kinetics of Magnetic Fluids in Liquid Crystals. In: William Coffey (eds) *Relaxation Phenomena in Condensed Matter, Advances in Chemical Physics Series*, vol LXXXVII. J. Willey Sons, NY.
- Schiff H (1991) Modulation of Spike Frequencies by Varying the Ambient magnetic Field and Magnetite Candidates in Bees (*Apis mellifera*). *Comp. Biochem. Physiol. [A]* 100 (4), 975-985.
- Slowik T J, Thorvilson HG (1996) Localization of Subcuticular Iron-Containing Tissue in the Red Imported Fire Ants. *Southwestern Entomol.* 21 (3), 247-255.
- Wehner R, Harkness R D, Schmid-Hempel P (1983) In: *Information Processing in Animals*, p.1 (M. Lindauer, ed.). Stuttgart: G. Fischer.

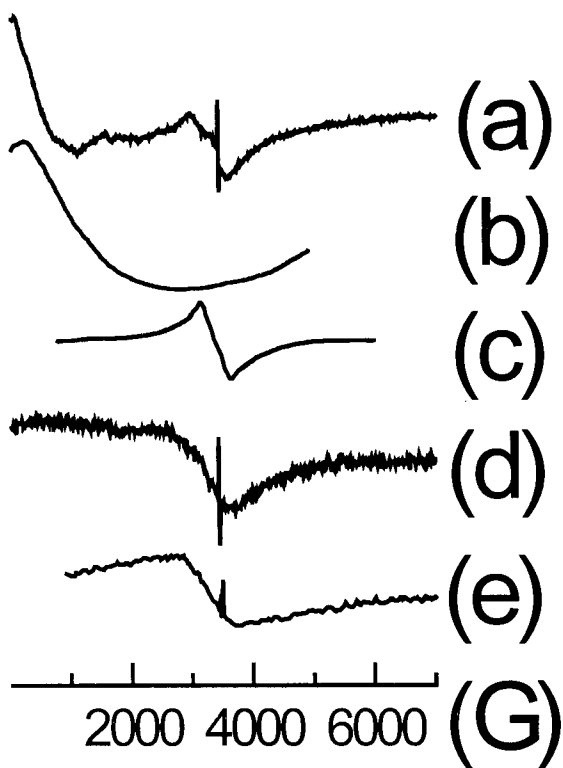


Fig 1. EPR spectra of (a) Venda Nova fire ants, (b) natural mineral magnetite large particles from Ikeya 1993, (c) magnetite particles from Aharoni and Morton 1971 , (d) Citrolândia fire ants and (e) magnetite film from Bandow and Kimura 1991. (a) and (d) spectrometer settings: 20G field modulation, 20 mW microwave power and 9.640 GHz.