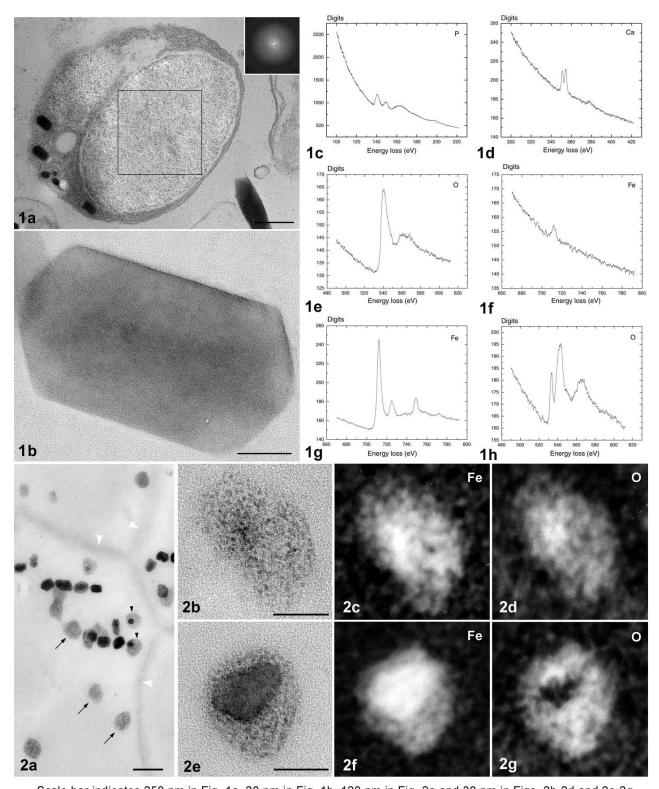
## BIOMINERAL COMPARTMENTALIZATION IN UNCULTURED MAGNETIC BACTERIA

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Magnetic bacteria are a group of motile microorganisms that orient to magnetic field lines because of membrane-bound organelles called magnetosomes. Magnetic bacteria precipitate magnetite (Fe<sub>3</sub>O<sub>4</sub>) or greigite (Fe<sub>3</sub>S<sub>4</sub>) in their magnetosomes. The ability of uncultured magnetic bacteria of accumulating minerals in different cell compartments is still poorly understood. We have been studying natural enrichments of magnetic bacteria from lagoons in Rio de Janeiro, Brazil. Here, we have used energyfiltering transmission electron microscopy to study the element distribution in biocomposites found in magnetic bacteria from both lagoons. In Itaipu lagoon, electron microscopy and phylogenetic position determination by 16S rRNA genes sequencing identified at least four morphotypes of magnetic bacteria[1]. Typical structures found in all gram-negative bacteria, in addition to some internal compartments were observed (Fig. 1a). Amorphous granules (Fig. 1a) and magnetosomes (Fig. 1b) were the two well-defined compartments that accumulated significant amounts of minerals. Granules contained no membrane unit around them and presented no obvious long-range periodic organization as observed by electron diffraction (not shown) and Fourier transform image processing (Fig. 1a, inset is the transform of the boxed region). Electron energy loss spectroscopy indicated that granules contained phosphorus (Fig. 1c), calcium (Fig. 1d) and oxygen (Fig. 1e) as primarily elements and small amounts of iron (Fig. 1f). Additionally, granules contained occasionally variable small amounts of zinc and aluminum. Magnetosomes were of high structural perfection and were composed of magnetite in all samples analyzed. Figure 1b is a high-resolution transmission electron microscopy image of a magnetosome showing lattice fringes corresponding to the [100] direction for magnetite. Electron energy loss spectroscopy of the magnetosome detected iron (Fig. 1g) and oxygen (Fig. 1h) as the only elements in these crystals. Rodrigo de Freitas lagoon contained magnetotactic multicellular aggregates, which consisted of several bacteria connected by intercellular junctions (Fig. 2a white arrowheads). We have observed new amorphous mineral particles (Fig. 2a black arrows) within the cytoplasm of aggregates. Elemental mapping (Fig. 2b-d) and electron energy loss spectroscopy (not shown) detected iron and oxygen in these new particles, which were about the same size of mature iron sulfide magnetosomes (around 50-70 nm). No membranes were observed surrounding the amorphous phases. Magnetosomes of variable sizes also presented amorphous regions around them similar in texture to the amorphous particles (Fig 2a, black arrowheads and 2e). The shape of the regions around magnetosomes followed the shape of the crystalline phase of the magnetosome they enveloped. These regions also contained oxygen and iron (Fig. 2e-2g). The complex process of iron sulfide formation by magnetotactic multicellular aggregates involves several steps. Recent reports suggest that a series of sequential reactions takes place during the formation of iron sulfide magnetosomes [2]. This reaction sequence involves the conversion from mackinawite (FeS) to greigite (Fe<sub>3</sub>S<sub>4</sub>), which requires iron loss. Our data suggest that the amorphous regions and particles observed could be a sink for iron released in cytoplasm during phase transformation in magnetosomes [3].

- [1] S. Spring et al. Arch Microbiol 169 (1998) 136.
- [2] M. Pósfai et al. Science 280 (1998) 880.
- [3] Supported by CAPES-PROCAD, FAPERJ, CNPq (PRONEX), FUJB Brazilian agencies.



Scale bar indicates 250 nm in Fig. 1a, 20 nm in Fig. 1b, 120 nm in Fig. 2a and 30 nm in Figs. 2b-2d and 2e-2g