	Experiment title: XANES spectroscopy on biologically synthesized magnetite nanoparticles doped with metallic elements (Ni, Zn, Cu)	**Experiment number**: 25-01-1016
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Report:

Magnetosomes are membrane-bound magnetic nanoparticles synthesized by magnetotactic bacteria. In particular, magnetotactic bacteria *Magnetospirillum gryphiswaldense* synthesize 40 nm-sized magnetite (Fe_3O_4) nanoparticles. Aimed at expanding the potential applications of these nanoparticles, bacteria have been cultured in transition metal (M=Ni, Zn, Cu) supplemented media to promote the biosynthesis of metal doped magnetosomes.

In experiment 25-01-1016 we assessed the structure around the metallic elements of the isolated magnetosomes by means of XANES spectroscopy. Due to the low dopant content, measurements on the M K-edge were performed in fluorescence yield mode, but measurements on the Fe K-edge were performed in transmission mode. All measurements were carried out at room temperature.

These results were complemented with other measurements performed in a previous experiment HC-2420 (R. Martín-Rodríguez et al.) on Co and Mn-doped magnetosomes and another experiment in ALBA on the whole magnetotactic bacteria and were recently published in D. Muñoz et al., Sci. Rep. **10** (2020) 11430 [1]. In that work we show that magnetosome-bearing bacteria show an increased tolerance to the presence of metals in the culture medium than bacteria without magnetosomes. XANES spectroscopy has allowed us to assess whether bacteria use magnetosomes as metal storages, or whether they incorporate the excess metal in other cell compartments. Our findings reveal that the tolerance mechanisms are metal-specific: Mn, Zn and Cu are incorporated in both the magnetosomes and other cell compartments; Co is only incorporated in the

magnetosomes, and Ni is incorporated in other cell compartments. In the case of Co, Zn and Mn, the metal is integrated in the magnetosome magnetite mineral core. As an example, Figure 1 shows the Zn K-edge XANES measurements on bacteria cultured in a Zn-supplemented medium and their isolated magnetosomes.

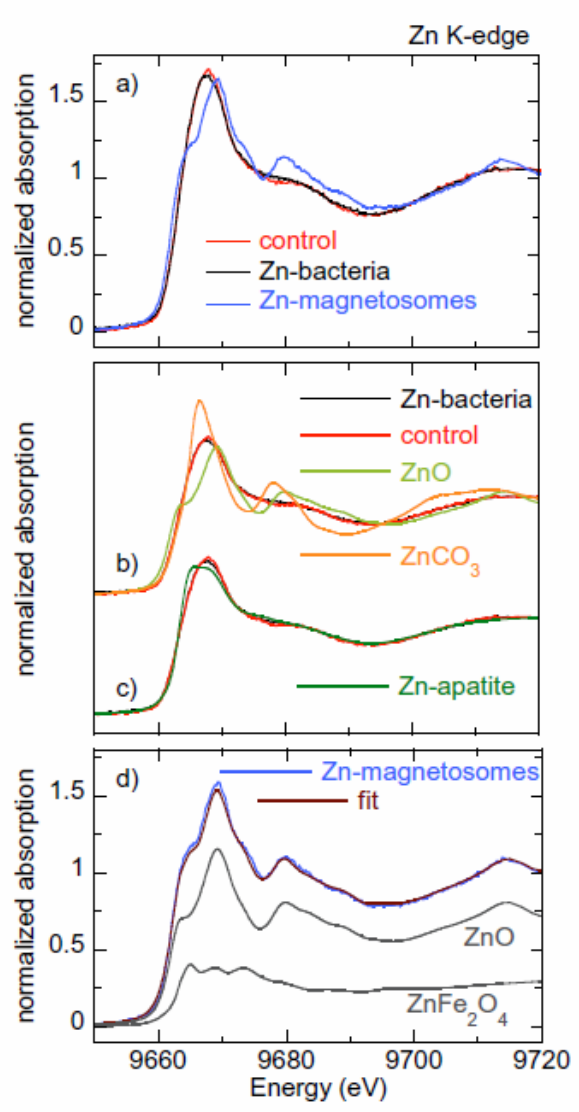

Figure 1: Normalized Zn K-edge XANES spectra of control bacteria, bacteria grown in a Zn supplemented medium (Zn-bacteria) and magnetosomes isolated from Zn-bacteria (Zn-magnetosomes). (**b**) Zn K-edge XANES spectra of Zn-bacteria and control together with two Zn^{2+} standards: ZnO and $ZnCO_3$. (**c**) Comparison of XANES spectra of Zn-bacteria and Zn-control with Zn apatite. (**d**) Linear combination fit of the Zn K-edge XANES spectrum of Zn-magnetosomes with 72% ZnO and 28% Zn ferrite ($ZnFe_2O_4$).

[1] D. Muñoz, L. Marcano, R. MAratín-Rodríguez, L. Simonelli, A. Serrano, A. García-Prieto, M.L. Fdez-Gubieda, A. Muela, *Magnetosomes could be protective shields against metal stress in magnetotactic bacteria*, Sci. Rep. 10 (2020) 11430