## **Microorganisms and Biofilms**

## LS.1.P009 3D STXM chemical tomography of cell-mineral aggregates formed by Fe(II)-oxidizing bacteria

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Ferrous iron can be oxidized under anoxic or microoxic conditions at neutral pH by Fe(II)-oxidizing bacteria, forming cell-mineral aggregates. The resulting biogenic cell-mineral aggregates efficiently retain toxic heavy metals as well as other pollutants by sorption processes [1].

To further our understanding of the underlying mechanisms, the mixotrophic, nitrate-reducing Fe(II)oxidizing *Acidovorax sp.* strain BoFeN1 was selected as a model strain to study the Fe(III)-biomineral formation at high spatial resolution and subsequently its influence on the sorption of heavy metals on the submicron scale. BoFeN1 shows initial iron biomineralization in the periplasm followed by cell encrustation in iron minerals that are associated with extracellular polymeric substances (EPS) [2].

Synchrotron-based scanning transmission (soft) X-ray microscopy (STXM) in combination with angle-scan tomography were performed, which enables mapping the distribution of iron and organic macromolecules (e.g. proteins, polysaccharides) in 3D [3]. A substantial advantage of STXM over other tomography approaches such as FIB-SEM or TEM tomography is the combination of microscopy at high spatial resolution (~10-50 nm) with X-ray absorption spectroscopy that provides quantitative chemical speciation information. Strain BoFeN1 was cultured in anoxic mineral medium supplemented with 10 mM Fe(II). Bacterial samples were prepared either by air-drying or in wet cells for STXM tomography of hydrated samples [4]. Organic carbon, protein and iron 3D composition maps were obtained from image sequences acquired across the C1s, O1s and Fe2p X-ray absorption edges. Tomography reconstructions of air-dried BoFeN1 cells revealed 3D biomineralization patterns in form of total encrustation in Fe(III) oxyhydroxides (Figure 1). A BoFeN1 cell in its natural, hydrated state indicated partial iron encrustation with hydrated extracellular "protein" in proximity to the cell (Figure 2).

In contrast to other tomography techniques, STXM in combination with angle-scan tomography allows 3D chemical tomography. Our results also indicated that STXM tomography measurements of pristine bacterial samples in their natural, hydrated state prevent typical preparation artifacts such as dehydration and shrinking [5].

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- 6. We thank the spectromicroscopy beamline team at the Canadian Light Source for help during the STXM measurements. We appreciate also the help of A. Picard, E. Struve, M. Abbas, W. Kuerner and the Geomicrobiology Group of A. Kappler Tuebingen for providing knowledge and samples. This project was supported by the DFG, Emmy-Noether grant (OB 362/1-1) to MO.

<sup>4.</sup> M. Obst et al. in "Canadian Light Source Activity Report 2008", ed. M. Dalzell (Canadian Light Source, Saskatoon) (2009), pp. 142-143







Figure 2. STXM 3D maps of iron and spectral fits for protein and ferrihydrite of a hydrated, partially iron encrusted cell.