

S-Layer – a special way to get in contact with "Heavy Metal"

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The focus of our investigations is the interaction of two mesophilic Acidithiobacilli and two thermophilic strains of the archaeal genus *Sulfolobus* with surfaces of natural pyrite. For a deeper understanding of the leaching process [1-3], we used several scanning and transmission electron microscopy techniques. In order to preserve the ultrastructure of the cells and their interaction site with the mineral in a life-like state, gentle cryo-preparation methods were employed throughout. In particular, this involved freeze-etching, fracture-labeling, and high-pressure freezing, freeze-substitution, embedding in Epon and post-embedding labeling.

Transmission and scanning electron micrographs showed the interaction in all strains to be mediated by S-layers. These are two-dimensionally crystalline surface proteins, containing a considerable carbohydrate moiety (as shown by PAS, periodic acid Schiff staining). Apparently, these macromolecules are equivalent to the EPS – extracellular polymeric substance – observed in other studies [4]. Freeze-etching confirms that the S-layer completely covers the surfaces of the cells. In ultrathin sections, the S-layer is seen to be in direct contact with the mineral surface, when cells are attached to pyrite (Fig. 1). This is directly confirmed via immuno-localization of the surface proteins by post-embedding labeling, confirming the even distribution of the proteins on the cell surface (Fig. 2).

S-layer sheets were isolated by detergent extraction, for both, immunizing rabbits and investigating their fine structure by TEM after negative staining, using correlation averaging [5]. For both strains of Acidithiobacilli, we could identify lattices with rare p2 symmetry, with almost identical lattice dimensions. For both archaeal strains, the proteinaceous surface layer exhibited p3 symmetry, as previously observed for closely related microorganisms of the Sulfolobales [6].

Literature:

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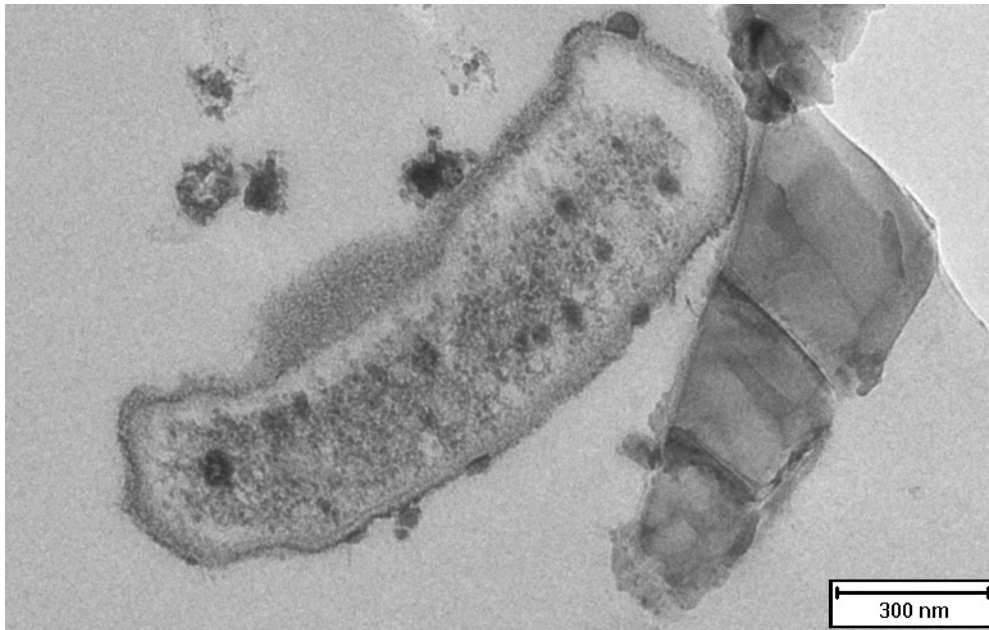


Figure 1. Transmission electron micrographs of an ultrathin section of *Acidithiobacillus ferrooxidans* (left), attached to pyrite (right).

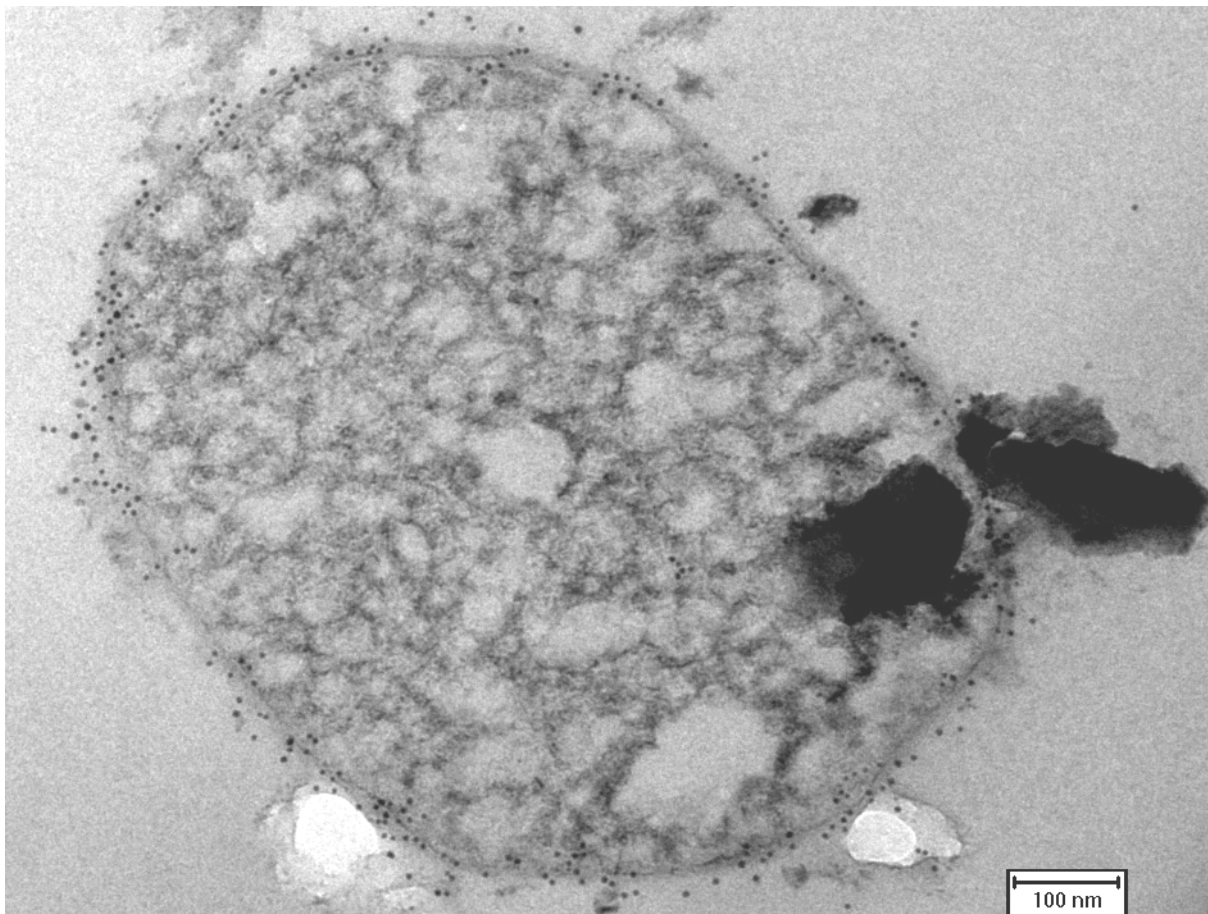


Figure 2. Ultrathin section of the archaeal pyrite leaching strain *Metallosphaera sedula*. Post-embedding labeling with an antiserum raised against the S-layer protein. Secondary antibody: goat anti-rabbit globulin, 6 nm Gold.