

Supplementary Information

Biogenic FeS nanoparticles modulate the extracellular electron transfer and schwertmannite transformation

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Text S1: Preparation of schwertmannite (Sch)

Sch was precipitated from a ferrous sulfate solution using the H₂O₂ method as described previously.¹ Briefly, 5 mL of H₂O₂ (30%) and 10 g of FeSO₄•7H₂O were added to 1 L of deionized water at 25°C and incubated for 24 h. The resultant ochre-colored precipitates were frequently washed with deionized water until the supernatant specific conductance was below 5 µs/cm and then they were freeze-dried.

Text S2: Composition of the modified medium

The modified medium contained (g/L): 0.1 KH₂PO₄, 1.0 NH₄Cl, 0.1 CaCl₂, 0.5 MgCl•6H₂O, 5.6 sodium lactate (60%), 4.76 HEPES (4-(2-hydroxyethyl)-1-piperazine ethyl sulfonic acid) and trace elements (1 mL). Trace elements were prepared as described by Myers and Nealson.² The medium was sterilized for 30 min at 121°C and cooled down before experiment.

Text S3: Sample analyses

Fe²⁺ concentrations were analyzed through the 1,10-phenanthroline method, using an ultraviolet-visible spectrophotometer (UV-160A, Shimadzu, Japan).³ Ion chromatography (DIONEX ICS-1500, 148 Sunnyvale, USA) was applied to determine SO₄²⁻ concentration. Lactate and acetate concentrations were determined using High performance liquid chromatography (HPLC, Agilent LC 1260, USA).⁴

Solid-phase samples were rinsed once with deoxygenated deionized water, dried and preserved under anoxic conditions until further analysis. X-ray diffraction (XRD) with Cu K α radiation (Bruker Advance D8, Germany) was applied to analyze the changes in mineral phases at the end of the incubation. Samples were scanned with a 0.05° 2 θ step-size and 2 s count time from 10° ~ 80° 2 θ . The morphology of secondary minerals was identified by scanning electron microscope (SEM) (ZEISS Merlin3700, Germany). Transmission electron microscopy (TEM; FEI Talos F200X and Hitachi HT7700 Hitachi H-7000FA 100Kv) required with energy dispersive spectroscopy (EDS; Oxford X-Max) was used to observe the morphology and distribution of the solid phase and bacteria.

Text S4: The qPCR reaction system and procedure

The qPCR reaction system and procedure were as described by Ke et al.⁵ The reactions were performed in a total volume of 20 µL with 10 µL 2x SYBR Green

qPCR Master Mix and 0.4 μ l ROX Reference Dye (APExBIO, Houston, USA), 0.5 μ L each primer (10 μ M), and 1 μ L of DNA template. The procedures were 95°C initial denaturation 2 min, 40 cycles of 95°C denaturation 15 s, and 60°C annealing 30 s. The melting curve was obtained by heating the sample from 60°C to 95°C. Every sample was measured in triplicate to guarantee the accuracy.

Table S1 Primers used in this study

| Primer | Sequence (5'-3') | Amplicon Size (bp) | Reference |
|----------------|----------------------------|--------------------|------------|
| <i>recA</i> -F | GTGATGAGGTGGTTGGTAACGAGAC | 117 | 5 |
| <i>recA</i> -R | CGCCAGTACGGTTGATACCTTGAC | | |
| <i>mtrC</i> -F | ATGGTGCACCTATGGTCACTG | 150 | 5 |
| <i>mtrC</i> -R | AGCCTAAGCCTTGCCAGTTAG | | |
| <i>cymA</i> -F | GCGAAATATTCCATCCTAGCGC | 152 | 5 |
| <i>cymA</i> -R | GCCAGCACTTCATTCTTCAAGG | | |
| <i>omcA</i> -F | ATGCCAAGAAAGAACCCG | 115 | 5 |
| <i>omcA</i> -R | TGGTCTACTAAACAAGTGTCGC | | |
| <i>phsA</i> -F | ATGTGGTCAGCGGGTGGAAATTTATC | 103 | This study |
| <i>phsA</i> -R | GCATTGGGTTGGTACGGGACATC | | |

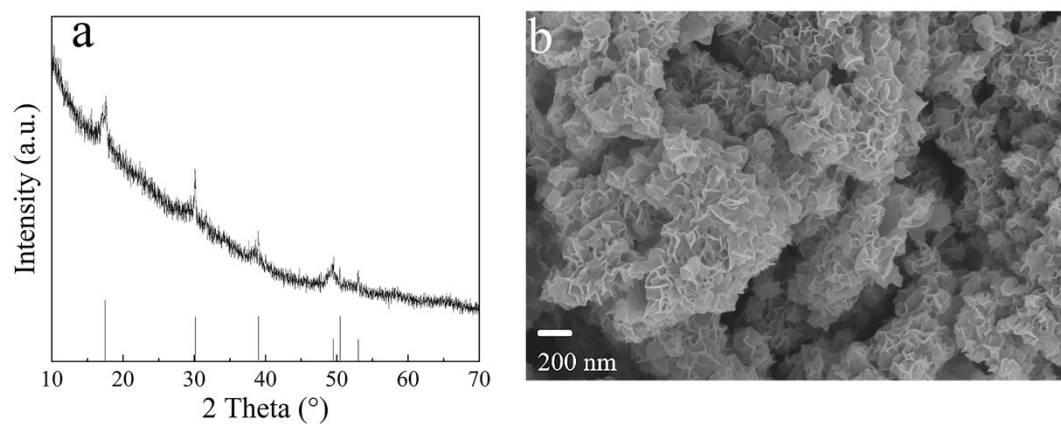


Fig. S1 XRD (a) and SEM images (b) of the FeS NPs.

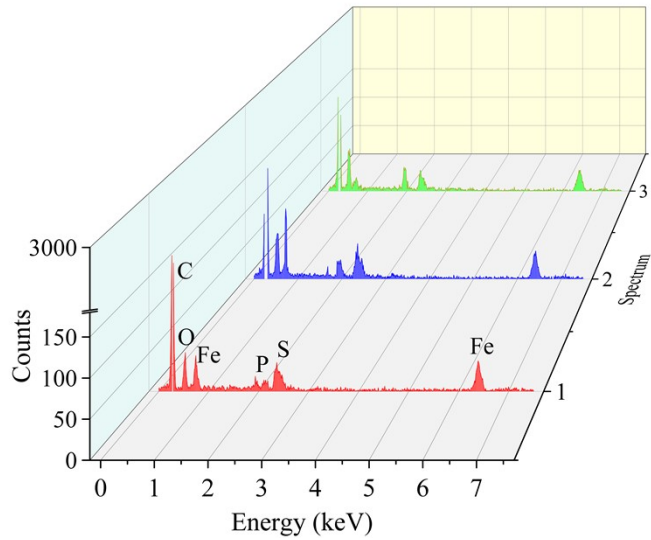


Fig. S2 Determination of element distribution of minerals on cell surface by EDS. The number on the Z-axis corresponds to the number in Fig. 1e.

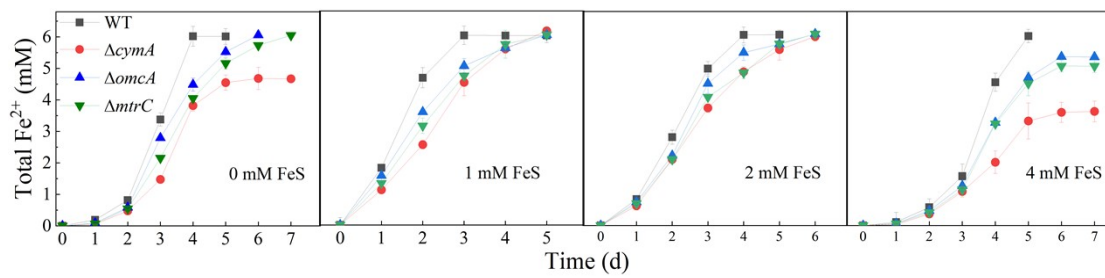


Fig. S3 The total Fe²⁺ concentration during the Sch reduction by *S. oneidensis* MR-1 (WT) and its mutants.

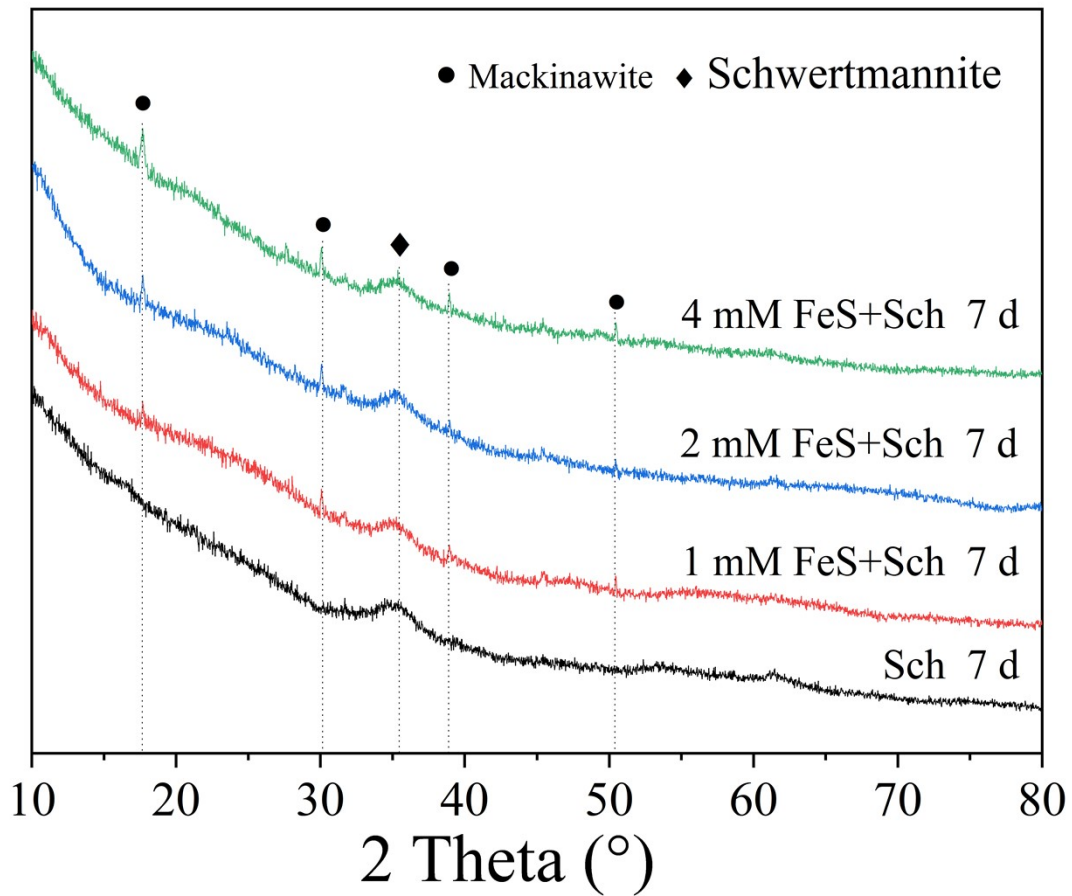


Fig. S4 The interaction of FeS and Sch in the absence of *S. oneidensis* MR-1.

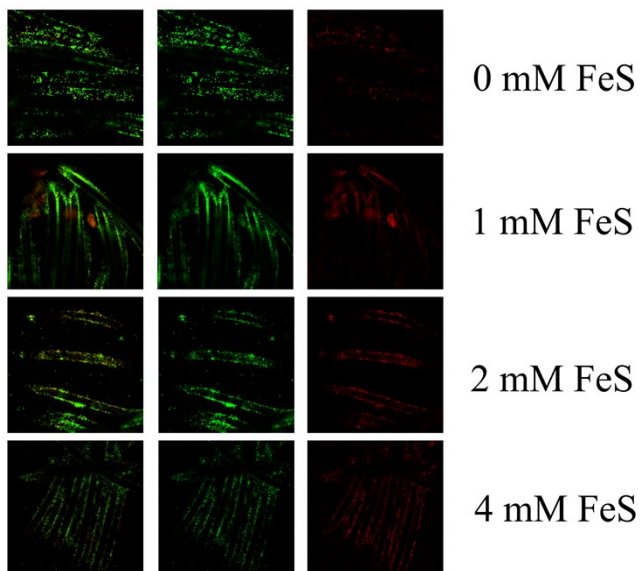


Fig. S5 The cell viability was detected by CLSM.

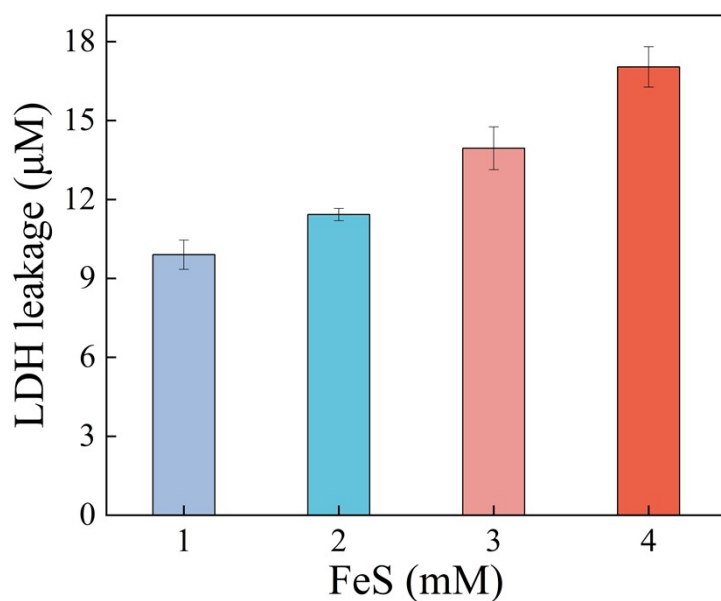


Fig. S6 The cell viability was detected by LDH leakage.

Reference

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