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A novel mechanism of electron transfer from iron-reducing microorganisms to solid iron phases

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Dissimilatory iron-reducing microorganisms are facing the problem to deliver electrons to insoluble ferric iron phases during respiration. Several hypothesis on the nature of the electron transfer are currently discussed such as i) a direct contact of cells and a transfer via outer membrane redox proteins, ii) the microbial excretion of quinone electron shuttles and iii) redox active groups in humic acids as natural electron shuttles.

We discovered a novel mechanism of electron shuttling from microorganism to insoluble iron phases via ferric iron nanoparticles. In the first set of experiments, *Geobacter metallireducens* cells were cultivated on ferric citrate, washed carefully to remove all possible electron shuttles and submitted to cell suspension experiments. Iron reduction kinetics was monitored with different electron acceptors such as ferric citrate, ferrihydrite colloids and solid ferrihydrite. The ferrous iron production observed was highest with the soluble electron acceptor but iron colloids were reduced with almost the same kinetics and orders of magnitude faster than the solid phase. The results demonstrate that colloidal iron is probably a very reactive iron phase in nature and much more susceptible to reduction than the solid phases. We furthermore studied if colloids can transfer the electrons to amorphous ferrihydrite and work as a real shuttle rather than a potent electron acceptor only. Therefore, ferrihydrite was added as the main electron acceptor in cell suspension experiments with *Geobacter* and ferric colloids were added in catalytic amounts. Whereas in the first hours a fast production of ferrous iron was only due to the reduction of the colloids itself this changed into a second phase

where the reduction of the ferrihydrite phase was significantly enhanced as compared to controls without colloid amendment. The results show that ferric colloids are not only a very reactive iron phase but can also expose a catalytic function as an electron carrier to less reactive iron phases.

This novel hypothesis for the mechanism of electron shuttling in dissimilatory iron reduction has significant advantages over the above mentioned other hypothesis i) and ii). On the one hand the organism doesn't have to come into near contact with the iron phase where a distance of 12-14 Å would be needed between the electron releasing molecule on the bacterial surface and the iron mineral surface for efficient electron transfer. Furthermore, the microbe doesn't have to invest energy in the production of the electron shuttle, an extremely important factor in oligotrophic environments such as groundwater. However, our shuttling hypothesis doesn't exclude that different organisms developed different strategies to overcome the spatial distance between the mineral surface and the respiratory enzymes.