Immobilization of selenium by biofilms of *Shewanella putrefaciens*

Y. SUZUKI[1]*, H. SAIKI[1], A. KITAMURA[2], AND H. YOSHIKAWA[2]

[1] Graduate School of Bionics, Tokyo University of Technology, 1404-1 Katakura-cho, Hachioji, Tokyo 192-0982, Japan (*correspondence: yosuzuki@stf.teu.ac.jp)

[2]Geological Isolation Research and Development Directorate, Japan Atomic Energy Agency, 4-33 Muramatsu, Tokai, Naka, Ibaraki 319-1194, Japan

The microbial reduction of the selenite and selenate has been widely studied using planktonic bacteria. Although bacteria are predominantly found within surface-associated cell assemblages, or biofilms in natural settings, there are little information on the interaction between biofilms and selenium. In this study, biofilms of *Shewanella putrefaciens*, which is an iron-reducing bacteria, were formed and their structure was investigated by a confocal laser scanning microscopy (CLMS). Then reduction of selenite by the biofilms was examined.

Biofilms of *S. putrefaciens* were made on circular cover glasses. Formation of biofilms was observed by CLMS. To investigate the reduction of selenite by the biofilms, a solution containing 100 μM sodium selenite as an electron acceptor, 20 mM sodium lactate as an electron donor was added to the biofilms under an anaerobic condition. After 34 h, the selenium concentration in the solution was measured by ICP-AES. Se K-edge XANES spectra of the precipitates appeared on the biofilms were measured at the Beamline 12C, Photon Factory, KEK (Tsukuba, Japan).

The CLMS observation revealed that thickness of the biofilms was about 10-20 μm and the cells were heterogeneously distributed in the biofilms. After 34 h incubation of the biofilms with selenite, the red precipitates were observed at the place where the biofilms were formed. The precipitates were not dissociated from the biofilms by washing with a deionized water indicating that they associated tightly with the biofilms. The selenium concentration in the solution was under detection limit. The Se K-edge XANES spectrum of the red precipitates showed that they were elemental selenium. These results suggest that the biofilms with iron-reducing bacteria in the environment can strongly immobilize the selenium on the biofilms through selenite reduction to elemental selenium.

Prediction of Surface Organic Species at the Mineral-Water Interface vs. Spectroscopy

D.A. SVERJENSKY[1]

[1]Johns Hopkins University, Baltimore, MD 21218, USA (*correspondence: cestrada@jhu.edu, sver@jhu.edu)

Surface complexation models have been widely used to model adsorption data for organic species at the mineral-water interface. However, the goal of developing truly predictive models in which the number of surface species, the nature of the surface species attachments and the variations of the proportions of the surface species as functions of environmental conditions such as pH, ionic strength and surface loading has not been reached. Recent advances in the theory and application of the extended triple-layer model (ETLM), in particular taking into account the electrical work associated with desorption of chemisorbed water molecules released during the formation of inner-sphere attachments [1] have enabled substantial progress.

For example, when the ETLM is applied to batch adsorption data referring to a wide range of environmental conditions, typically only a few reaction stoichiometries are able to fit the data. This is in marked contrast to more traditional surface complexation models that lead to highly ambiguous results. From the reaction stoichiometries, model surface species can be inferred. However, few direct *in situ* tests of such results are available. In the present study, two direct tests are described involving experimental adsorption data for the amino acids glutamate and dihydroxyphenylalanine (DOPA) on titanium dioxide. The predicted glutamate surface species and their behavior established using the ETLM [2] were tested with ATR-FTIR spectroscopy and quantum chemical calculations [3]. For DOPA the ETLM results [4] were tested with surface enhanced Raman spectroscopy (SERS) [5]. Excellent agreement is obtained between the number of surface species, the nature of their attachment to the surface and the dependence of the surface speciation on environmental conditions.

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