

Microbial dissimilatory Fe(III) reduction in subsurface environments

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Dissimilatory Fe(III)-reducing bacteria (IRB), play an important role in the natural cycling of organic matter and minerals in subsurface environments and can be important agents for the bioremediation of both organic and metal contamination.

Two types of IRB, designated strain KNA6-3 and KNA6-5, were isolated from the groundwater of a sedimentary layer (158-160m depth) using Fe(III)-citrate as the sole electron acceptor for anaerobic respiration. The groundwater sample was collected from borehole KNA-6 in Tono Mine operated by Japan Nuclear Cycle Development Institute. The occurrence of the isolated Fe(III)-reducers in the groundwater were detected and quantified by real-time PCR, and it turned out that the isolates account for 0.06% of the in situ microflora.

The 16S rDNA sequences of the isolates showed their close relation to the genus *Pseudomonas* in the gamma-subdivision of Proteobacteria. *Pseudomonas* species are widely distributed in various environments such as marine, terrestrial and subsurface habitats, suggesting that they are highly adapted to various environmental conditions. Versatile microorganisms such as *Pseudomonas* species respond to environmental changes via sensing and transduction of environmental cues (signals). This communication reports that the Tono IRB switch their respiratory modes from aerobic to anaerobic via oxygen is mediated by two distinct regulatory systems composed of the Arc two-component system and anaerobic regulator. The Arc two-component system comprises the ArcB protein as the membrane sensor kinase and the ArcA protein as the response regulator. ArcB is an environmental sensor of the cell growth conditions. ArcB functions to activate ArcA during anaerobic conditions via protein phosphorylation. ArcA, when activated, bind to regulatory DNA sites to mediate both positive and negative control of gene expression. Anr, a transcriptional regulator of each of the respiratory pathway genes, becomes activated when cells are shifted from aerobic to anaerobic growth condition. In order to detect the relevant genes (*arcB*, *arcA* and *anr*), PCR primers were designed. These genes were amplified by PCR and sequenced. Furthermore, expression levels of the relevant genes were compared by real-time RT-PCR of *arcA*. Therefore these results suggest that *arcA* and *anr* can contribute to dissimilatory Fe(III)-reduction.