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Subcellular localization, speciation and imaging of mercury during bacterial methylation process

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Microbial Hg methylation is a key process in the Hg cycle occurring in soils, sediments and anoxic waters and controlling the neurotoxic methylmercury (CH_3Hg) concentrations in aquatic environments [1]. Despite our improved understanding of the microbiological mediated Hg methylation in the environment and the identification of the involved bacteria (e.g. sulfate-reducing bacteria (SRB)) [2], the cellular mechanisms leading to CH_3Hg remain not well understood. Because methylation extent seems to be controlled by cellular uptake [3], the assessment of the localization of mercury at the subcellular level is critical to provide mechanistic insights of such processes.

In this work, a kinetic study of Hg methylation was conducted with a model SRB strain, *Pseudodesulfovibrio hydrargyri* BerOc1, during 24 hours and for different mercury concentrations (0.05 µM to 5 µM). The quantification of CH_3Hg and inorganic Hg in different subcellular fractions at different kinetic times was performed by GC-ICP-MS. We used Hg L3-edge High energy Resolution X-ray Absorption Near Edge Structure (HR-XANES) spectroscopy to determine Hg ligands and coordination. Finally, cells were imaged by High resolution Scanning Transmission Electron Microscopy (HR-STEM) and Synchrotron Radiation based nanoscopic X-Ray Fluorescence (SR nano-XRF) during the Hg methylation to locate Hg and other metals during Hg methylation.

The results showed that Hg methylation reaction rate depended on Hg concentration, and that a saturation of the methylation process occurred. At the Hg concentrations tested, a maximal threshold value of intracellular CH_3Hg was reached, explaining why CH_3Hg is mainly found in the extracellular medium. On the other hand, intracellular inorganic Hg content increased during the incubation whatever the added Hg concentration. Tetra-coordinated Hg-S species were identified as the dominant Hg species in the bacteria. Chemical imaging validated that Hg is both extracellular and intracellular. Interestingly, we observed a high heterogeneity between bacterial cells with Hg 'hyperaccumulating' cells and low accumulating cells. These outcomes open new perspectives for studying Hg methylation mechanisms at the cell level.

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[3] Hsu-Kim H.et al. (2013), Environ. Sci. Technol. 47(6), 2441-2456