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Hg accumulation controlled by bacterial cells involved in Hg methylation: Imaging of sulfato-reducing bacteria by nano X-ray fluorescence

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Methylmercury (MeHg) is a potent neurotoxin that bioaccumulates and biomagnifies through aquatic food webs, posing significant risks to ecosystems and human health. Hg methylation by microbes is an intracellular process[1] that saturates with increasing Hg concentrations[2]. We hypothesize that this saturation is the consequence of a limited Hg accumulation in the cells, due to the high toxicity of the metal. In this study, we investigated mercury accumulation in the model *Pseudodesulfovibrio hydrargyri* BerOc1 strain, able to both methylate and demethylate Hg. BerOc1 was incubated with 0.5 µM and 2 µM $HgCl_2$. Mercury localization at the cell level was studied using synchrotron nano X-ray fluorescence with high spatial resolution. For both concentrations, we could evidence Hg into bacterial cytosol, in addition to extracellular Hg-S containing nanoparticles previously observed[2,3]. Interestingly, the intracellular Hg concentration corresponding to the highest cell density was slightly higher in the 2 µM exposure compared to the 0.5 µM exposure (1.61 and 1.40 $pg.mm^{-2}$, respectively), but was not proportional to the spiked concentration, suggesting a regulated uptake and/or accumulation. Among cells, a few Hg-hyperaccumulating cells were observed, and the heterogeneity in Hg concentrations between individual cells was higher at the 2 µM condition. In order to determine the physiological state of these hyperaccumulating cells, we correlated nano X-ray microscopy with optical fluorescence microscopy after Live/Dead staining. Results revealed that Hg-hyperaccumulating cells are dead although not all dead cells exhibited high Hg content. Our results suggest that living cells control Hg uptake or accumulation regardless of the external Hg concentration. These findings provide new insights into the role of intracellular Hg accumulation for Hg methylation.

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