Role of Microbial Growth on Hg(0) Uptake and Production of Methylmercury

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Introduction: Anaerobic bacteria play a central role in the mercury (Hg) biogeochemical cycle through their catalysis of Hg methylation. Several species of anaerobic delta-proteobacteria have now been shown to produce MeHg when provided with dissolved Hg(0) as their sole Hg source. In this study, we examined the role of microbial growth on the uptake of Hg(0) and production of methylmercury. Using growing, resting, and heat-inactivated cells of *Desulfovibrio desulfuricans* ND132, we conducted experiments to investigate the physiological factors that affect the conversion of dissolved gaseous Hg(0) to Hg(II) and MeHg.

Materials and Methods: *D. desulfuricans* ND132 was grown under strict anaerobic conditions in defined media. Experiments with resting cells were conducted by suspending washed cells in deoxygenated phosphate buffer. Heat-killed cells were inactivated at 80°C for 30 min. Hg(0) oxidation experiments were carried out by reacting growing, resting, and heat-inactivated cells of *D. desulfuricans* ND132 with a continuous source of Hg(0) gas. Samples were periodically collected for non-purgeable Hg and MeHg analysis. Optical density ($O.D._{600}$) of the culture was measured at the beginning and end of of the incubation. Epifluorescence microscopy was performed using the LIVE/DEAD *Bac*Light Bacterial Viability Kit and a Zeiss Axioskop 20 microscope.

Results and Discussion: Experiments with metabolically active, resting and heat-inactivated cells indicated that both live and dead cells oxidize Hg(0) to Hg(II). In the live cells experiments, high MeHg concentrations were observed in most replicate experiments, but 3 out of 10 independent replicate samples did not show production of MeHg. Cell density measurements and epifluorescence microscopy revealed that MeHg production was strongly correlated to microbial growth, and was also related to the aggregation state of the cells. In samples where MeHg production was low, the cells were not growing and predominately subsisted in a planktonic state. Conversely, in samples where MeHg production was high, significant growth was observed and cells were tightly aggregated in biofilm-like communities. These results suggest that cellular oxidation of Hg(0) to Hg(II) can have toxicity effects on anaerobic methylating bacteria.