“Nutrient limitation alters metabolism, Cr(VI) response, and biofilm matrix composition in *Desulfovibrio vulgaris* Hildenborough”

*PhD Dissertation Defense*

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**ABSTRACT**

Sulfate-reducing bacteria are a diverse group of anaerobic microorganisms that live in anoxic environments and play critical roles in biogeochemical cycling, namely linkages between the carbon and sulfur cycles. *Desulfovibrio vulgaris* Hildenborough (DvH) is a model organism for sulfate-reducing bacteria that has been studied for its ability to reduce toxic heavy metals to insoluble forms and its involvement in microbially induced corrosion in oil pipelines and other industrial settings. The described work investigated how the availability of electron donor carbonsources and electron acceptors affected Cr(VI) reduction, metabolism, and biofilm growth and composition in DvH. DvH was grown planktonically at 20°C and 30°C in batch mode or as a biofilm under continuous flow at 20°C. In the second chapter of this dissertation, it is established that electron acceptor limitation (EAL) predisposes cells to Cr(VI) toxicity compared to a balanced electron donor to electron acceptor (BAL) condition and electron donor-limited (EDL) condition. In chapters 3 and 4, the effect of nutrient limitation on DvH biofilms is investigated, and microscopy revealed unique extracellular membranous structures that have not previously been observed. The extracellular structures were heterogeneously distributed, connected to cells, co-localized with metal precipitates, and were more prevalent under EAL compared to BAL conditions. Differential staining indicated that the structures were composed of lipid, consistent with the observation that these structures are membrane derived. Metabolomic analysis revealed an up-regulation of many fatty acids under the EAL condition, which was confirmed and quantified via GC-MS. Down-regulated metabolites for biofilm grown under the EAL condition included those involved in DNA turnover, N-cycling, and peptidoglycan turnover, indicating that EAL may induce a switch from growth to fatty acid production that may coordinate with alternative electron transfer mechanisms. Outer membrane vesicles (OMVs) were also purified from DvH biofilm grown under EAL condition included those involved in DNA turnover, N-cycling, and peptidoglycan turnover, indicating that EAL may induce a switch from growth to fatty acid production that may coordinate with alternative electron transfer mechanisms. Outer membrane vesicles (OMVs) were also purified from DvH biofilm grown under EAL and BAL conditions. Abundant proteins detected in OMVs included porins, lipoproteins, hydrogenases, and three proteins involved in oxidative stress response. The results presented here show that nutrient limitation and resource ratio affect DvH physiology in both biofilm and planktonic growth modes. The analysis of the DvH biofilm matrix highlights the importance of investigating the extracellular capabilities that are unique to the biofilm growth mode and has implications for activities and physiological states in the environment.