

# **The role of cell surface sulfhydryl and amine binding sites in the reduction of Cr(VI) from solution by *Bacillus subtilis* bacterial cells**

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A range of bacterial species can reduce soluble Cr(VI) to Cr(III), causing the precipitation of Cr(III) phases from solution. However, the mechanism responsible for the initial binding of anionic aqueous Cr(VI) onto the negatively charged cell surface has not been identified. We measured the kinetics of Cr(VI) removal in suspensions containing the Gram-positive soil bacterial species *Bacillus subtilis* as a function of pH and in systems with different Cr(VI):cell ratios, and we measured the effect of pH on the extent of Cr(VI) removal at a fixed time in experiments with *B. subtilis* biomass. The roles of sulfhydryl and amine binding sites in Cr(VI) removal were constrained using site-specific blocking molecules, and the impact of the site blocking on Cr(VI) removal was measured as a function of pH.

Our results indicate that Cr(VI) removal by *B. subtilis* is at least partially non-reversible and is dependent on binding site concentration, pH, and metal loading. Our results are consistent with a two step removal process: Cr(VI) first adsorbs reversibly onto cell wall binding sites, followed by Cr(VI) reduction to Cr(III) likely via electron transfer from cell wall electron transport chain molecules. The presence of either sulfhydryl-specific blocking molecules or amine-specific blocking molecules or both in the experimental systems dramatically reduces the extent of Cr(VI) reduction, especially under circumneutral pH conditions, strongly suggesting that both sulfhydryl and amine binding site types participate in the initial attachment of Cr(VI) onto the cell surface. The experiments with either sulfhydryl or amine sites blocked both exhibited a similar reduction in Cr(VI) removal to the experiments with both types of sites blocked, suggesting that both types of sites are involved simultaneously in binding Cr(VI) species to the cell surface either by forming a bidentate bond with a Cr(VI) species, or by neighboring sulfhydryl and amine binding sites forming a more favorable binding environment. The results of our work are the first to propose a viable mechanism that can explain the binding of anionic Cr(VI) onto an overall negatively charged cell surface as a first step in the reduction to Cr(III) and subsequent removal of Cr from solution.