A genetic system for Geobacter metallireducens: role of the flagellin and pilin in the reduction of Fe(III) oxide

Geobacter metallireducens is an important model organism for many novel aspects of extracellular electron exchange and the anaerobic degradation of aromatic compounds, but studies of its physiology have been limited by a lack of techniques for gene deletion and replacement. Therefore, a genetic system was developed for G. metallireducens by making a number of modifications in the previously described approach for homologous recombination in Geobacter sulfurreducens. Critical modifications included, among others, a 3.5-fold increase in the quantity of electrotransformed linear DNA and the harvesting of cells at early-log. The Cre-lox recombination system was used to remove an antibiotic resistance cassette from the G. metallireducens chromosome permitting the generation of multiple mutations in the same strain. Deletion of the gene fliC, which encodes the flagellin protein, resulted in a strain that did not produce flagella, was non-motile, and was defective for the reduction of insoluble Fe(III). Deletion of pilA, which encodes the structural protein of the type IV pili, inhibited the production of lateral pili as well as Fe(III) oxide reduction and electron transfer to an electrode. These results demonstrate the importance of flagella and pili in the reduction of insoluble Fe(III) by G. metallireducens and provide methods for additional genetic-based approaches for the study of G. metallireducens.