Toxic methylmercury (MeHg) is produced by certain anaerobic bacteria, and recent study has shown that two genes, i.e., hgcA and hgcB, are required for the methylation process. However, methylation is an intracellular reaction, requiring the uptake and transport of mercury (Hg) across the outer and inner membranes of the bacteria. Cell uptake and transport depend on the Hg speciation, the nature of ligands present in the surrounding media, and the characteristics of proteins on cell surfaces. Studies have shown that the presence of cysteine (Cys) enhances the uptake and methylation of Hg by Geobacter sulfurreducens PCA, but not by Desulfovibrio desulfuricans ND132, and the underlying mechanism and actual role of cysteine on Hg uptake are still not fully understood. Questions remain with regard to whether (i) cysteine is exchanging Hg(II) with the ligands/proteins on cell surface, (ii) Hg-Cys complexes interact with the cell surface through specific binding, or (iii) Hg-Cys complexes directly enter into the cells. Here we systematically investigate the effects of cysteine and selected bis-thiol chelator ligands, such as 2,3-dimercaptopropanol and dipeptides (Arg-Cys), on Hg uptake and methylation. We report that cysteine competes with cells for Hg binding and decreases Hg sorption and methylation initially in a phosphate buffered saline solution but, for unknown mechanisms, over time the G. sulfurreducens PCA cells were able to overcome the initial competition for Hg, leading to uptake and enhanced methylation, even at a high cysteine concentration (1000 µM). The amount of methylated Hg appeared to correspond well with the neutral Hg-complexes (i.e., HgCl₂ and Hg(Cys)₂H₂) in solution, while both MeHg and the neutral complexes were at lowest levels in the presence of ~0.1 µM cysteine. Our results also show that bis-thiol chelators and Arg-Cys peptide strongly inhibit Hg(II) uptake and methylation as compared to control experiments. These results indicate that the effect of cysteine and complexing organic ligands on Hg(II) uptake and methylation is highly complex, and additional research is needed to elucidate the roles of cysteine and natural organic ligands in the Hg methylation process.