# Use of ATR IR Spectroscopy in the Identification and Quantification of Bacterial Surface Functional Groups in Conjunction with Chemical Equilibrium Modelling

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It is well recognised today, that bacteria have high potential for the binding of heavy metal ions due to their great surface area to volume ratio and their overall negative surface charge. To help predict metal distribution in the presence of bacteria in natural systems and to aid the design of bioremediation plants for heavy metal contaminated wastewaters, chemical equilibrium models have been developed. These models are based on acid base titration data and calculate the concentrations and $pK_a$ values of the titratable functional groups which are present on the bacterial surface and likely to be involved in metal binding. However, such models cannot provide direct evidence of the chemical nature of the functional groups. IR spectroscopy is a technique that has been used to identify functional groups in organic molecules and more recently to characterize intact bacteria.

In this study the ionisable surface functional groups of Anoxybacillus flavithermus were investigated. Anoxybacillus flavithermus is a thermophilic bacterium isolated from the effluent of a New Zealand geothermal powerstation. The experimental techniques employed were batch acid base titrations and electrophoretic mobility measurements at different pH values as well as in situ attenuated total reflection infrared spectroscopy (ATR IR) over the pH range from 2-10.

Based on the batch titration and zetapotential data, a chemical equilibrium model was developed in FITMOD, a modified version of FITEQL 2.0. A two site model was able to describe the experimental data suggesting the presence of two types of ionisable functional groups on the bacteria. Additionally, the obtained IR data indicated the presence of carboxyl and phosphodiester moieties and gave direct evidence of carboxyl speciation over the investigated pH range. The results from both of these approaches will be compared and their use in the identification and quantification of bacterial surface functional groups will be discussed.