3.0 SIGNIFICANCE OF WORK

The work described within examines metal speciation and the interactions between metals and microbiology in both laboratory settings and the natural environment. The approach is unique for its examination of metals using advanced microscopic and spectroscopic tools. This combination of methods, along with comparisons to traditional analytical techniques, provides information that would be missed when examining these phenomena at the bulk scale. In particular, analytical electron microscopy (AEM) allows the visualization of particle morphologies and the elemental associations therein. This allows characterization of different colloid classes and shows on which classes the metals are concentrated. Since this is an individual particle analysis, one can extract specific information that will be missed when examining the bulk system. X-ray absorption spectroscopy (XAS) provides a complimentary picture to the AEM results. This method uses X-rays to examine the coordination environment of the element of interest. Although extensively used in material science, it potential is just now beginning to be realized for application to natural systems. An important feature of XAS is that is can probe elements that are traditionally referred to as spectroscopically silent, i.e., metals which have a full valence shell, such as zinc and cadmium. These methods also provide a degree of *in situ* measurement. While it is currently impossible to take the instruments required to make these measurements directly into the field, care can be taken to preserve the samples so no chemical change occurs before analysis. The methods themselves then examine the sample with minimal change or alteration of the sample.

Another significant portion of this work is the way in which the XAS data are gathered, processed, and analyzed. XAS spectra were all measured using the continuous scan (CS-XAS) mode. Described in detail later, this technique involves collecting the spectra continuously as the X-ray energy is smoothly varied between the starting and ending energies. This approach has many advantages. First, since time to collect a spectrum is small, *e.g.* average time around 120 seconds, the sample is given minimal exposure to the X-rays. Traditional step-by-step spectra employ scan lengths of typically thirty minutes. This minimizes damage caused by heating or photoreduction from the beam. In addition, the sample throughput is greatly increased. When examining sediment cores, one desires to collect spectra at many depths within an individual sample. Since collection times are small, many more samples can be processed. This is especially important, as beamtime allotments at synchrotron sources are often sparse. Second, since the scan time is small, many replicate spectra can be collected in a relatively short amount of time. This improves the statistics of the overall, averaged spectrum. Additionally, the first scan and last scan can be compared to ensure that no alteration of the sample has occurred. Finally, the collection of CS-XAS data allows the measurement of the experimental error. The error that is calculated is the error from the actual experimental setup, not only the error of taking replicate measurements. This is accomplished by the oversampling of the data from each data point. These errors can then be carried through each step of the data processing. This involves data normalization, background subtraction, and spectral deconvolution. Each step involves a complicated process that is often non-linear. Thus, a Monte-Carlo approach has been taken to propagate the errors through each step in the data reduction and analysis.

The approach of the CS-XAS analysis is also novel. Traditionally, XAS experiments have been performed on compounds consisting of single crystals or relatively pure alloys. The analysis is done by performing *ab initio* calculations and extracting parameters such as the bond length and number of atoms in a given coordination shell. This technique is not well suited to samples originating from the natural environment, as there are likely to be numerous phases in which a metal may exist. For this analysis, spectral deconvolution using quadratic linear programming was performed to resolve the composition of the metal in the sediment mixture. Again, the errors calculated from the data processing are carried through in this step as well, so that the error of the final composition of the sample is also determined. The deconvolution was based upon a set of geochemically relevant standards that may be expected in sedimentary environments. Components such as carbonates and sulfides are obvious choices, due to the production of CO_2 and H_2S from bacterial metabolism. Additionally, adsorption to mineral substrates, such as clay particles and iron oxides, must be considered. Further additions to the basis set were chosen using the insights of results from AEM. Data from AEM give elemental associations on individual particles that can be used to infer their composition. From this information, the appropriate standard compounds can be chosen for XAS to verify the observations from AEM. The completeness of this basis set is critical in order to achieve and accurate result.

As the collection, processing, and analysis of the data involves new approaches, a software package was written in order to facilitate these calculations. SAMXAS runs on a Unix-based platform and uses Scilab as its primary computing environment. A graphical user interface was also written using Tk/Tcl widgets for ease of use. The details of the program and its use are given in Appendix B.

By making these observations on an *in situ* microscopic and macroscopic basis, the factors that affect metal speciation can be input into geochemical models. These types of models can subsequently be used to determine the effectiveness of remediation strategies, such as natural attenuation. The success of these strategies is strongly dependent on the quality and completeness of data and observations collected from the field. Thus, the approaches used herein to determine metal speciation can be used to better the understanding of metal fate and transport in the natural environment.