Mn K-edge and Pb L_3- edge XAFS studies of *Leptothrix discophora*.

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Introduction

The importance of the geochemical matrix of surficial sediments in mediating the availability of metals to sediment ingesting organisms is now a well-documented phenomenon [1]. For example, sediments high in iron oxides have been shown to reduce metal uptake by these organisms. In contrast, trace metals associated with sediments that are high in oxides of manganese that are also concurrently low in oxides of iron have been shown to be highly bioavailable.

Two bivalve species, *Macoma balthica* and *Protothacia staminea* will be fed samples of sediment with differing trace metal compositions and then analyzed for trace metal bioaccumulation. In addition, the bivalve species *Mytilus edulis* will be fed a manganese-oxidizing bacterium, *Leptothrix discophora* that is also carrying differing trace metal compositions. The bioavailability of these metals as determined from tissue residues will be related to the trace metal speciation within the bacterium and the sediments as determined by XAFS. This will allow identification of the role of sediment geochemistry in influencing metal bioavailability to sediment ingesting organisms. Importantly, the XAFS characterization will allow us to examine the association of the trace metal impurities within naturally occurring biological systems and trophic levels.

This report describes our first preliminary results of the XAFS study on the bacteria *Leptothrix discophora*. Four different growth conditions were investigated. All four samples were exposed to aqueous Mn^{2+} at the natural environmental level (1mM $MnSO_4$). Sample #1 was the control; it was exposed to Mn^{2+} only. The other three samples were exposed to varying concentrations of Pb^{2+} (lead acetate $Pb(OOCCH_3)_2 \times 3H_2O$). Sample #2 was exposed to 10 ppb (the natural environmental level), sample #3 to 10^3 ppb (the upper end of naturally occurring Pb concentrations) and sample #4 to 10^4 ppb (significantly above naturally occurring Pb concentrations). It should be noted that the highest Pb concentration did not inhibit the biological process significantly.

Methods and Materials

The bacteria cultures were grown according to methods described elsewhere [2,3]. They were then freeze dried and stored at room temperature.

Fluorescence XAFS data were collected at the undulator beamline, ID-20, PNC-CAT. The samples were mounted on kapton tape for exposure to the x-ray beam. The Mn K edge was measured using an ion chamber containing argon. Because of the lower Pb concentration, the Pb L_3 fluorescence signal was measured with the 13-element solid state detector of the PNC-CAT.

Results and Discussion

The x-ray fluorescence spectrum of sample #2 is shown in Figure 1. In addition to the prominent Mn K_α and K_β and the weak Pb L_α lines there are features caused by other metals in the sample, notably Fe, Ni and Zn. The ratio in the peak area between the Mn K_α and Pb L_α lines is 32, which corresponds to an approximate relative concentration of Mn to Pb of 278:1. This low Pb concentration of sample #2 made it difficult to obtain good Pb L_3 XAFS spectra. In consequence, larger aqueous Pb concentrations were used for the preparation of samples #3 and #4 in an effort to increase the Pb concentration of the bacteria sample, and thus facilitate the Pb L_3 XAFS analysis.

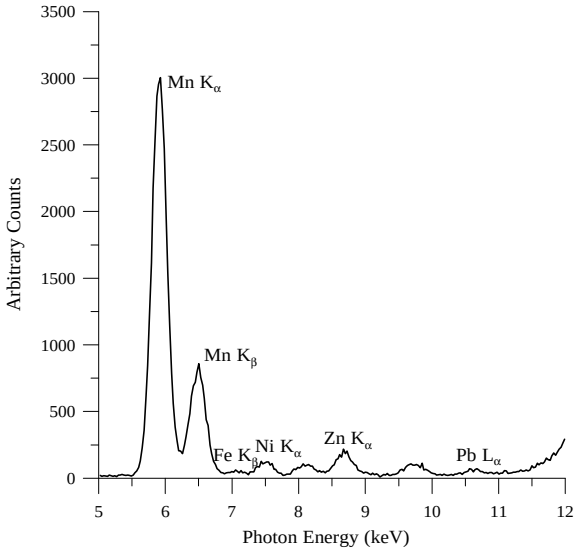

Figure 1: X-ray fluorescence lines of sample #2. Incident photon energy = 13.165 keV, 130 eV above Pb L_3 edge.

The Mn concentration, in contrast, is sufficiently high to permit analysis of the XAFS spectra. The Mn K-edge XAFS interference function $\chi(k)$, weighted by k, is shown in Figure 2 for samples #1 and #2.

Oscillations extend out to k = 12 $Å^{-1}$, at which point the Fe K-edge interferes with the Mn XAFS. The range of the Mn XAFS spectrum can be extended beyond the Fe K-edge through the use of the 13-element detector. The *kc(k)* and Fourier transforms of samples #1 and #2 are fairly similar.

The magnitude of the Fourier Transform (FT) of *kc(k)* is shown for both samples in the inset of Figure 2. The transforms indicate that the adsorbed Pb in sample #2 causes little change in the first and second neighbours of a central Mn atom, but it significantly affects the extended structure from the third neighbours onward. The data will be analyzed to determine the identity, number and bond lengths of the atoms coordinating to the Mn.

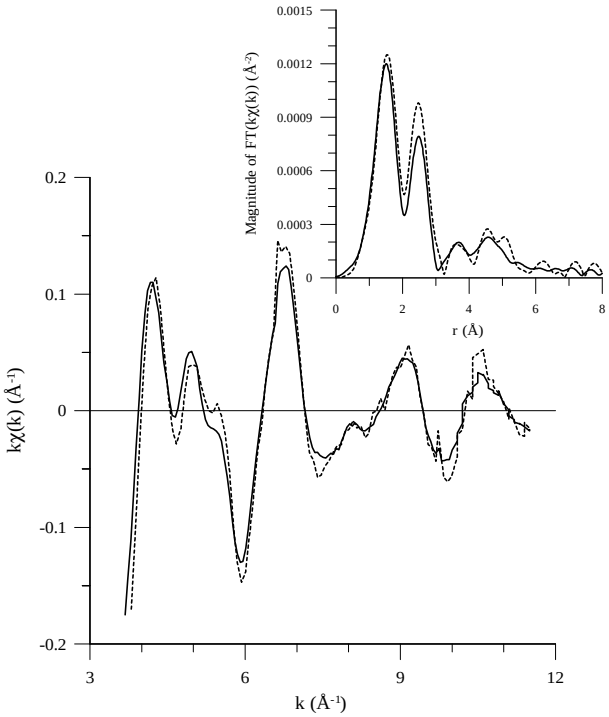

Figure 2: The Mn K-edge XAFS of samples #1 (solid) and #2 (dashed). The inset shows the magnitude of the Fourier transform of these Mn K-shell XAFS spectra, using a 30% Gaussian window over the range 3.74 to 11.51 $Å^{-1}$.

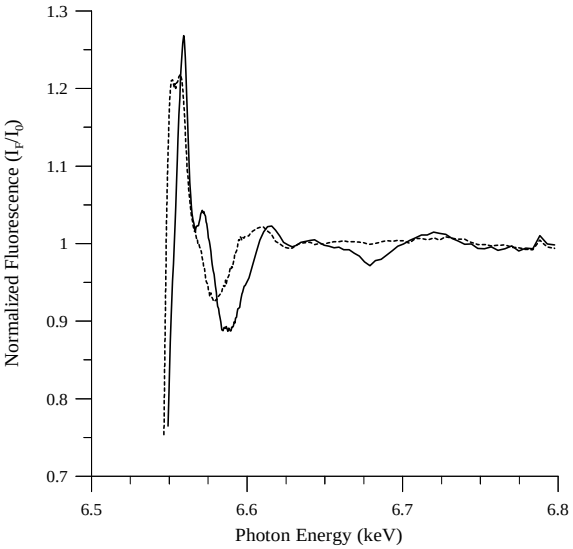

Figure 3: The effect of radiation damage on the Mn K edge spectrum. The 1st (solid) and the 7th (dashed) spectrum of sample #4 taken at the same beam position.

The bacteria samples containing Mn and/or Pb experience radiation damage that is noticeable at room temperature in successive XANES spectra collected at the same position of the sample. This is illustrated in Figure 3 above.

To reduce the radiation damage, the sample was distributed along the circumference of a disk of diameter 4" and rotated at 1200 rpm. After 10 hours of exposure there was no evidence of radiation damage in the Mn K-edge XAFS spectra. Using this approach, along with the increased Pb concentration of samples #3 and #4, it was then possible to obtain absorption spectra of Pb, which is present in the samples at lower concentrations than Mn. Figure 4 shows the Pb L_3 XANES of a single scan of sample #4, however multiple scans must be averaged to obtain high quality low signal/noise XAFS spectra. We will then analyze the local structure about the Pb to attempt to determine how it is coordinated to the Mn.

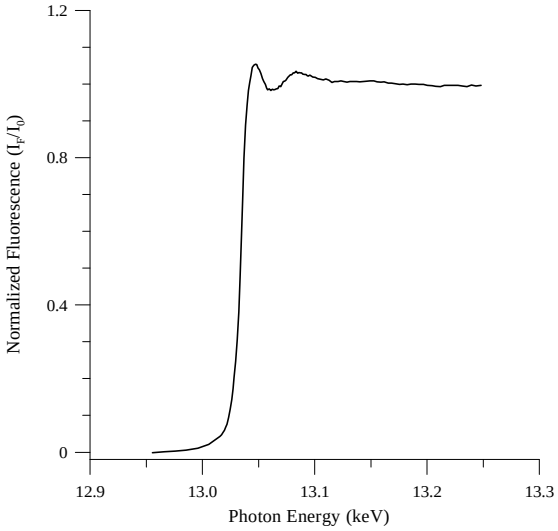

Figure 4: The Pb L_3 edge XANES of sample #4.

The samples are inherently heterogeneous with possible spatial distribution of Mn in different oxidation states. A microprobe study is dictated. But in rotating the sample, the XAFS spectra average over all sites. In subsequent experiments we will determine if cooling a stationary sample to liquid nitrogen temperature will reduce the rate of radiation damage sufficiently to permit micro-XAFS analysis

Acknowledgments

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