

U L₃-Edge XANES Measurements of U(VI) Biologically Reduced by *Shewanella putrefaciens* with Soil Humic Acids

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Introduction

Uranium processing has led to significant soil and groundwater contamination at mining sites and DOE facilities. Biological reduction of soluble U(VI) to an insoluble form of U(IV) [e.g., uraninite or UO₂(s)] has been proposed as a remediation strategy for U subsurface contamination, in which the mobility of U from contaminated areas would be decreased. Various dissimilatory metal-reducing bacteria have been reported to catalyze this reaction under anaerobic conditions. The impact of humic materials on the biological reduction of soluble U(VI) species such as UO₂(CO₃)₂²⁻ is not well understood. The objectives of this study were to examine the effects of humic materials on the biological reduction of U(VI) and to elucidate the mechanism(s) of the humic effects.

Methods and Results

In experiments under nongrowth conditions, biologically reduced materials were produced with uranyl acetate as the electron acceptor and sodium lactate as the electron donor in a solution of PIPES buffer and NaHCO₃ solution with International Humic Substances Society soil humic acid (SHA). The samples were prepared in an electrolyte (BE) of 10 mM Na-PIPES + 28 mM NaHCO₃ equilibrated under an O₂ free atmosphere of N₂:CO₂ (80:20). The samples contained 5 mM sodium lactate as the sole electron donor. The biotic reactors contained *Shewanella putrefaciens* CN32 at 10⁸ cells/mL. Abiotic uninoculated controls were spiked with an aliquot of BE equal in volume to the CN32 spike. The samples were filtered, and XANES measurements were made on the filtrate. Sample UH1 was prepared from a solution containing 100 μM uranyl acetate, 100 mg/L SHA, and CN32, and sample UH5 was prepared from a solution containing 500 μM uranyl acetate, 100 mg/L SHA, and CN32.

Uranium L₃-edge absorption data were collected at the MRCAT beamline [1] with a multielement detector in fluorescence mode. The second harmonic of the insertion device was tapered by 3.5 keV to reduce the variation in the incident x-ray intensity to 20% over the scanned energy range. The incident ion chamber was filled with a mixture of He and N₂ gases (50:50). A Rh mirror with a cutoff energy of 22 keV was used to eliminate higher harmonics. Reference spectra from a uranyl phosphate standard were measured by using scattered x-rays as described elsewhere [2].

The percent U(VI) was determined by comparing the energy value at half edge step in the normalized absorption data with the value for U(IV) and U(VI) standards. The U(IV) and U(VI) standards have energy values (17169.3 eV and 17174.6 eV) that are separated by 5.3 eV. All data sets were aligned accurately in energy by using the derivative of the edge of a simultaneously measured uranyl phosphate standard. The accuracy of this alignment procedure is 0.2 eV or approximately 5%. The XANES spectra indicate that UH1 and UH5 contain approximately 50% U(VI) and 50% U(IV), as shown in Fig. 1.

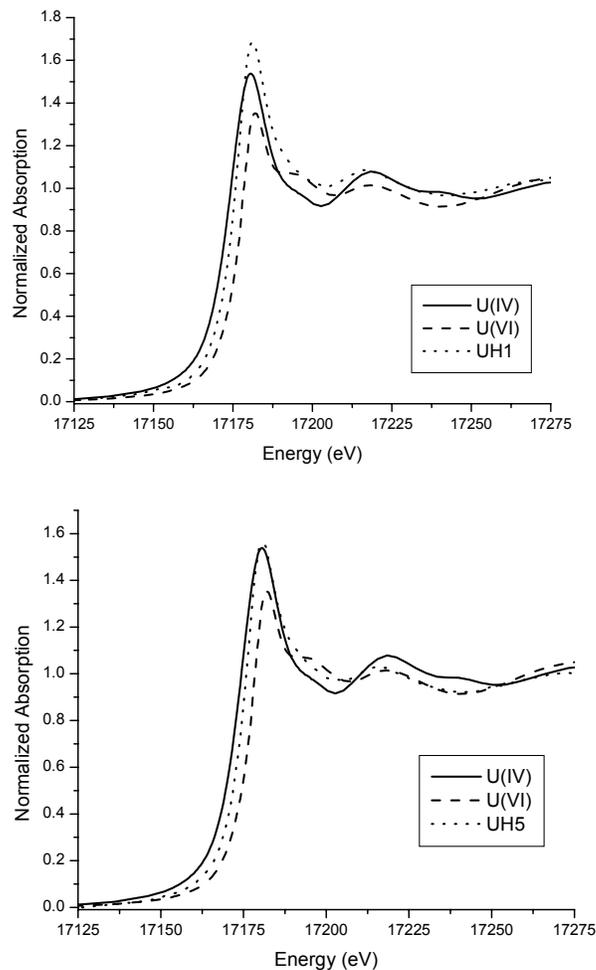


Fig. 1. Comparison of U(IV) and U(VI) standards to the (A) UH1 and (B) UH5 samples.

Discussion

In summary, the percent of U(IV) determined by XAFS in all sample filtrates that contained SHA compared favorably to corresponding kinetic phosphorescence analysis measurements (data not shown). These results suggest that SHA facilitated the transport of U(IV) nanoparticles through the filter or, alternatively, that soluble U(IV)-SHA complexes were formed.

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