

Interactions of *Pseudomonas fluorescens* with Uranyl (UO_2^{2+}): Implications for Mobility of Uranyl in the Subsurface

Rizlan Bencheikh-Latmani (latmani@stanford.edu), Jim Leckie (leckie@ce.stanford.edu) &
Alfred Spormann (spormann@ce.stanford.edu)

Department of Civil and Environmental Engineering, Stanford University, Terman Engineering Building,
Stanford, CA 94305, USA

Uranium is a contaminant of concern in the subsurface of the majority of former nuclear fuel and weapons manufacturing facilities. Uranyl is often found in sites co-contaminated with chelators. Some chelators (notably citric acid) are known to significantly alter U sorption to mineral surfaces (Redden et al., 1998). In addition, organic chelators are also subject to biodegradation by indigenous micro-organisms (Joshi-Tope et al., 1995; Bolton et al., 1996; Witschel et al., 1999). Therefore, to understand the sorption behavior of uranyl in the subsurface, it is necessary to consider the potential effect of chelators as well as their susceptibility to biodegradation in uranyl-contaminated environments. There is little systematic knowledge about the effect of complexation with metals on biodegradation of organic substrates, about the interactions of uranyl with bacterial cells, or about the relative affinity of uranyl to cells and mineral surfaces. This study addresses those questions for a well characterized gram negative bacterium (*Pseudomonas fluorescens*) in a simple chemical system of known speciation. We investigated the toxicity of uranyl to *P. fluorescens*, determined the fate of uranyl upon citrate degradation and evaluated the effect of citrate biodegradation on the citrate-enhanced uranyl sorption onto goethite.

P. fluorescens degrades citrate ($1 \mu\text{M}$) rapidly ($> 90\%$ in 5 hours) under non-growth conditions. In the presence of equimolar uranyl, the rate and extent of degradation are slightly lower but 87% degradation is achieved in 10 hours (Figure 1). This result clearly shows that citrate complexed to uranyl is biodegradable under these conditions. At increasingly higher uranyl concentrations, citrate degradation is slower and reaches an asymptotic behavior at lower values of citrate degraded. In the presence of $3 \mu\text{M}$ uranyl, only 45% degradation of citrate is achieved after 25 hours whereas 18% is reached in the presence of $5 \mu\text{M}$ uranyl (Figure 1). The explanation for the decrease in activity is that uranyl is toxic to *P. fluorescens* and that increasing uranyl concentrations shut down citrate metabolism with increasing efficiency. The fate of uranyl in the presence of cells was evaluated. In less than one hour, 90% of uranyl disappears from solution in the presence of cells. In the absence of cells, uranyl remains in solution over the time frame of the experiment. The association of uranyl with cells is not an active process. Heat-killed cells scavenge uranyl from solution to the same extent as live cells. Thin-section TEM (transmission electron microscopy) further confirms the localization of uranyl. Micrographs clearly show that uranyl is clustered on the

membrane(s). In the presence of goethite and under the conditions of the study, 90% of citrate and 100% of uranyl are initially sorbed respectively. We find that *P. fluorescens* cells are able to degrade citrate initially sorbed in the presence and in the absence of uranyl. Moreover, after citrate degradation, uranyl remains sorbed to the goethite-cell mixture. This result indicates that cells are acting as a sorbent for uranyl and are scavenging the uranyl not associated with goethite.

The implication for subsurface environments contaminated with uranyl and citrate, is that the biodegradation of citrate may not affect uranyl mobility significantly. However, the occasional sloughing of uranyl-covered cells may produce discrete spikes in uranyl solution concentrations.

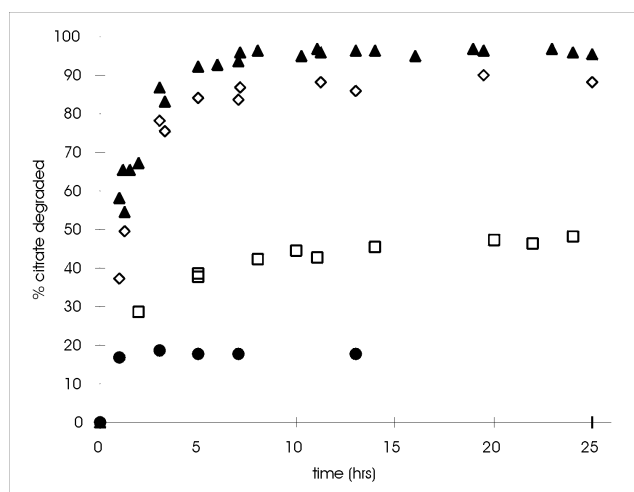


Figure 1: Effect of uranyl on the degradation of citrate ($1 \mu\text{M}$) by *P. fluorescens*.

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