EMPS Annual Report

Project Number: 86687
Project Title : Phytosiderophore Effects on Subsurface Actinide Contaminants: Potential for Phytostabilization and Phytoextraction

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1. Research Objectives

This project seeks to determine the potential of phytosiderophore-producing plants for phytostabilization and phytoextraction of actinides and some metal soil contaminants. Phytosiderophores are secreted by graminaceous plants such as barley and wheat for the solubilization, mobilization and uptake of Fe and other essential nutrients from soils. The ability for these phytosiderophores to chelate and absorb actinides using the same uptake system as for Fe is hereby investigated though characterization of actinide-phytosiderophore complexes (independently of plants), and characterization of plant uptake of such complexes.

2. Research Progress and Implications

This report summarizes work after ~nine months of a three-year project (50% of year-1 budget spent).

Since the beginning of this project in September 2002, we have made significant progress in planning, setting-up and initiating experiments. The first component of our experimental process was to obtain phytosiderophores in order to identify and define their association with plutonium, uranium, and nickel. Mugineic acid, the principal form of phytosiderophore produced by barley plants, can be isolated from barley root exudates, and can also be chemically synthesized. Both methods were undertaken.

Isolating mugineic acid from barley root exudates required germinating and growing barley plants, and establishing an iron-deficient environment to optimize phytosiderophore production. Root washings were then collected every morning for a period of two weeks, and the combined exudates were concentrated down to approximately 500 mL. Chromatography supplies were ordered to carry out the separation.

Mugineic acid synthesis is a twenty-two-step process that has been described in literature. It involves the regioselective reduction of 2,3-epoxycinnamyl alcohol. As the use of sodium azide necessary for this reaction caused safety concerns, a thorough safety analysis had to be done before proceeding. The total synthesis of mugineic acid is now well under way, and we expect to obtain our final product within the next month.

Another component of our project is to characterize plant uptake of actinides and metals. To accomplish this, we needed an experimental set-up where plants could be grown in a controlled environment, in continuously aerated solutions containing plutonium, uranium or nickel. We designed and assembled a system that consists of a 24-port gas manifold connected to the house air line, each port feeding air through a small-diameter plastic tube to bubble solutions that are contained in a 500 mL opaque plastic bottle. Each bottle holds one to four plants, which are placed through a hole in the bottle cap. Plants are germinated on filter paper or fine mesh in water, and then grown in nutrient solutions containing all essential macronutrients and micronutrients, including iron in varying concentrations. Once the plants have attained the desired maturity, uranium, plutonium or nickel are added to the solutions. The extent of actinide/metal uptake is determined by liquid scintillation counting of acid-dissolved plants.

Preliminary nickel uptake experiments were done with barley plants to test the set-up. Currently, further uptake experiments are being performed with uranium and nickel. Results from these experiments are not yet available. We have also started the set up for plutonium plant uptake experiments, which requires a separate system will more safety controls.

3. Planned Activities

Presently, our priority is to obtain phytosiderophores through the two above-mentioned methods, in order to determine the extent and type of uranium, plutonium and nickel chelation. Isolation and synthesis of mugineic acid will be completed within the next month, which will allow us to proceed with actinide/metal complexation many experiments before September 2003. These experiments will include the determination of the rate and amount of actinide solubilization from solid actinide forms (hydroxides, oxides) and from soil-sorbed species, in comparison to the solubilization ability of known common chelators:, such as acetohydroxamic acid, EDTA and citrate. These experiments will be completed in six to nine months.

Hydroponic plant uptake experiments have already begun with uranium and nickel. We have expanded our study to these metals due to the increased interest in remediation of the Savannah River site. Results from this initial set of experiments, expected in August 2003, will give us insight as to the ability of barley plants to adsorb, absorb or mobilize uranium and nickel from solution and if it is correlated to plant-Fe-status and phytosiderophore production. We will also start comparisons to the non-phytosiderophore producing plant Indian Mustard and will perform direct phytosiderophore utilization experiments. These experiments will continue into fiscal year 2004 with different variables and controls, such as comparisons between barley and Indian mustard uptake and solution amendments (additional phytosiderophore, acetohydroxamic acid, citrate, EDTA). We plan to begin the same type of uptake experiments with plutonium before September 2003. The timeline for this set of experiments is six months-one year.

Once results are obtained from hydroponic experiments (~April 2004), other uptake experiments will be performed with synthetic soils that are amended with uranium, nickel or plutonium. These will be performed using the same hydroponic set-up with the soils in nylon or dialysis bags in order to prevent soil particle adherence to the plant roots. This will allow us to determine whether the plants can extract the metals directly from soils (phytoextraction). We will also perform modified column-type experiments to determine if the plants can stabilize the metals in the root zone (phytostabilization). Solution amendments that were found to be effective in previous results will be used here. These experiments will take place between April 2004 and September 2004.

Subsequent experiments planned after July 2004 include tests on contaminated soils obtained from DOE sites, determination of the effect of plant death on metal migration through the soil, and concentration limits in which uptake can be achieved.

4. Information Access:

EMSP-PI meeting presentation:

ARQ (Actinide Research Quarterly)-Actinide Phytoremediation. A review of Phytoremediation of actinides & our research effort. Requested Manuscript in preparation for Sept. publication