The role of mycorrhizal fungi in the uptake of heavy metals by Salix.

A preproposal for the McIntire-Stennis Research Program

Personnel;

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Research Hypothesis;

Mycorrhizal fungi produce bioactive peptides (e.g. siderophores or phytochelatins) in response to heavy metal stress. The compounds are secreted into the rhizosphere where they serve an important function in the uptake and mobilization of heavy metals in forest soils.

Objectives;

1. Fungi that form mycorrhizal associations with *Salix* species will be grown in artificial culture and tested for their ability to produce siderophores and other metal-binding peptides in response to heavy metal stress.
2. Mycorrhizal fungi that test positive in objective one will be used to inoculated *Salix* plantlets *in vitro.* These plantlets will then be tested for their ability to withstand heavy metal stress.
3. The effect of mycorrhizal fungi, and/or their isolated bioactive peptides, on the uptake of heavy metals will be measured directly using radioisotopes. Microautoradiography will be used to evaluate if peptide formation has a positive or negative effect on the translocation of heavy metals.
4. Preliminary experiments on the formation of mutants, either constitutive or deficient in biopeptide formation will be initiated to confirm if biopeptide formation is a beneficial trait that promotes growth or phytoremediation of metal-contaminated sites by *Salix* sp.

Background and Expected Scientific Benefits;

(1) It is well established that mycorrhizal fungi produce strong metal binding peptides in response to metal limitation. There is also considerable evidence they may produce similar peptides in response to heavy metal stress. The function of these peptides is less clear. They may bind the metal and thus prevent its uptake into the cell. In this scenario, peptide formation would serve a beneficial function and may allow plant growth in metal-contaminated soils that would otherwise be lethal to the plant. However peptide function may also serve an opposite function. If the plant root has the required receptors for the peptide-metal complex, peptide formation would provide a high affinity mechanism for the uptake and hence sequestration of the metal in the plant. In this scenario, peptide formation would be beneficial for phytoremediation of metal-contaminated sites.

(2) The growth of *Salix* for biomass is an area of intense interest at ESF. As part of this project, biosolids are being used as fertilizer. This raises the possibility that heavy metals present in these biosolids may enter the rhizosphere. The use of mycorrhizal biopeptides, either to protect the willow against heavy metal toxicity, or to sequester the heavy metals from the soils so that they do not cause a problem after the land is returned to regular agricultural use, is an area of considerable promise. These studies will also provide valuable information that may be of interest to the current Willow biomass project, and provide the preliminary data necessary for the genetic modification of the Willow-mycorrhizal association to be used in phytoremediation of metal-containing sites.