

UNIVERSITY OF FLORIDA
Biocatalytic Lignin Modification for Carbon Sequestration

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Description: After cellulose, lignin is the second most abundant forma of carbon in plants. Lignin's complex structure makes it difficult to use this material in value-added products, and ahte vast majority of lignin is currently burned to provide energy for factory operations. While burning plant derived lignin does not add to global greenhouse gas levels, having options to remove lignin from the global carbon cycle would lead to diminished atmospheric CO₂ levels. This could be accomplished by chemically altering lignin's structure to facilitate long-term terrestrial sequestration or using it in value-added products that would not be discarded immediately. We will use Nature's catalysts (enzymes) to tailor the chemical structure of lignin for both deep-well injection (by using lignin derivatives as drilling "muds") and for materials that can be used in building, packaging, and other manufactured products.)

Budget: \$200,000

Universities: UF

Progress Summary

We have successfully expressed *Pseudomonas putida* toluene dioxygenase in our laboratory and carried out several model studies to establish proof-of-principle. Future studies will utilized either a recombinant strain expressing both toluene dioxygenase and a dehydrogenase (that converts the initial arene *cis*-diols into the corresponding catechols) or blocked *P. putida* mutants that are unable to consume catechols. In addition, once we have obtained access to real lignin samples, we plan to carry out the polymer grafting studies described in our proposal.

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Because laccase enzymes require low molecular weight mediators for catalytic activity, our initial focus has been on preparing these compounds by biocatalytic routes. The goal is to use a dioxygenase (such as toluene dioxygenase) to prepare a mixture of catechols from accessible portions of the lignin, then add laccase to complete the derivatization or degradation. We have successfully expressed *Pseudomonas putida* toluene dioxygenase in our laboratory and carried out several model studies to establish proof-of-principle. Future studies will utilized either a recombinant strain expressing both toluene dioxygenase and a dehydrogenase (that converts the initial arene *cis*-diols into the corresponding catechols) or blocked *P. putida* mutants that are unable to consume catechols.

In addition, once we have obtained access to real lignin samples, we plan to carry out the polymer grafting studies described in our proposal. To save time, this work will utilize exogenous laccase mediators since the major focus is on lignin derivatization and its potential for yielding value-added products from what is now waste material suitable only for burning.