

# WESTERN MICHIGAN UNIVERSITY



Department of Chemistry  
College of Arts and Sciences

*The  
Department of Chemistry  
presents*

*Literature Seminar  
by*

**Anna Essenburg**

## **Molecular Basis for a Toluene Monooxygenase to Govern Substrate Selectivity**

P450tol is a Class I P450 monooxygenase that naturally catalyzes the hydroxylation of toluene to benzyl alcohol. This enzyme has a potential use in biosynthesis as benzyl alcohol is an important intermediate in the synthesis of pharmaceuticals and pesticides. The current industrial process is energy-intensive and hazardous, and enzymes can operate under milder conditions in a one-pot-one-step reaction. In this paper, the authors obtained crystal structures of P450tol and its complexes with toluene and benzyl alcohol and determined its catalytic mechanism and key substrate binding residues. They then constructed artificial chimeric enzymes by fusing the heme domain of P450tol with the reductase domain of a self-sufficient P450, RhFRED or CYP116B46, and optimized it by varying the linker region. Extra space in the active site when the substrate toluene was bound suggested that this enzyme would work with halogenated toluenes. The researchers looked at the catalytic activities of their artificial enzymes with halogenated toluenes and found that these could be hydroxylated regioselectively on the methyl group. Finally, P450tol-CYP116B46 was engineered to catalyze the hydroxylation of propylbenzene in a regio- and stereoselective manner, showing its potential for use in pharmaceutical synthesis.

**Monday, April 15th at 4:00 PM  
1260 Chemistry Building**