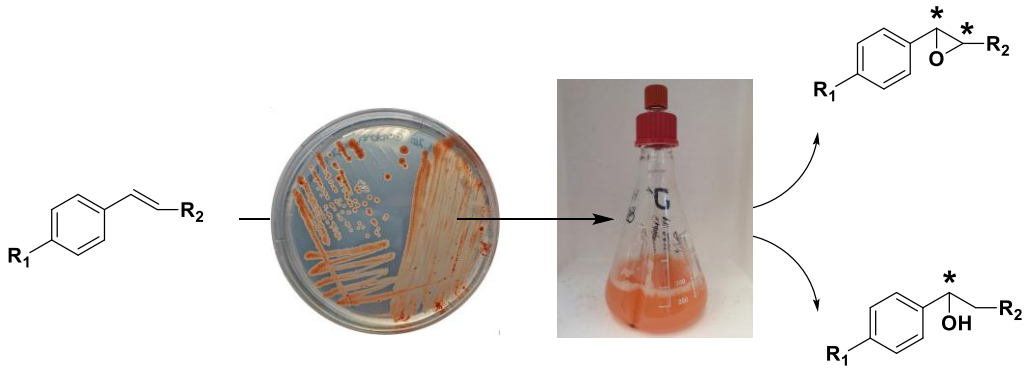
Project Dirk Tischler

Degradative enzymes for the biosynthesis of various aryl-alkyl building blocks

Background and preliminary work: The Tischler group has discovered a new pathway for the degradation of styrene leading to the central intermediate phenylacetic acid. It comprises the activities of already known two-component styrene monooxygenases and phenylacetaldehyde dehydrogenases, but, also of a so far uncharacterized glutathione *S*-transferase. This pathway allows to convert various styrene chemical analogous compounds and to produce respective phenylacetic acid derivatives. However, it is proposed that also here phenylacetaldehydes can be formed and converted to respective alcohols. Enzymes of this pathway can be used in combination with other oxidoreductases to produce related alcohols or epoxides which themselves represent aroma compounds or industrial relevant building blocks as depicted below.



Work planned: As a model organism serves the soil bacterium *Gordonia rubripertincta* CWB2 and its capability to degrade/convert styrene as well as chemical analogous compounds. A detailed biochemical and biocatalytical characterization of the native enzymes involved in this novel styrene degradative pathway as well as adjacent routes will be carried out. A special emphasis will be put on the involved oxygenases and reductases; in order to allow the production of alternative compounds such as selected chiral alcohols or chiral epoxides. Spectrophotometric, GC and HPLC methods need to be established in order to evaluate enzyme performance. Therefore, either the native enzymes of strain CWB2 will be purified and/or the respective gene has to be cloned and recombinantly expressed – in either *E. coli* or *Gordonia*. The produced enzymes will be characterized for general properties, but also with respect to biocatalytic applications. This comprises stability assays, enzyme optimization by mutagenesis as well as enzyme immobilization and bioconversion studies. All these investigations depend on a rapid and online analysis!

Selected references:

D. Eggerichs, C. Mügge, J. Mayweg, U.-P. Apfel, D. Tischler (2020) Enantioselective epoxidation by flavoprotein monooxygenases supported by organic solvents. Catalysts. 10:568.

T. Heine, J. Zimmerling, A. Ballmann, S.B. Kleeberg, C. Rückert, T. Busche, A. Winkler, J. Kalinowski, A. Poetsch, A. Scholtissek, M. Oelschlägel, G. Schmidt, D. Tischler (2018) On the enigma of glutathione-dependent styrene degradation in *Gordonia rubripertincta* CWB2. Appl. Environ. Microbiol. 84:e00154-18.

M. Oelschlägel, S.R. Kaschabek, J. Zimmerling, M. Schlömann, D. Tischler (2015) Co-metabolic formation of substituted phenylacetic acids by styrene-degrading bacteria. Biotechnol. Rep. (Amst.) 6:20-26.