innovations from nature

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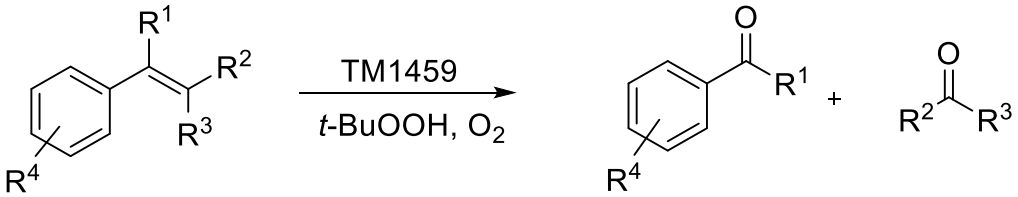
acib Project Proposal

Synthesis of aldehyde and ketones by enzymatic alkene cleavage

SUMMARY
The perfect catalyst for biocatalysis: (i) is easily available in high amounts from expression in *E. coli*, (ii) is solvent and thermostable (iii) reaches high conversion rates (iv) displays high selectivity (v) forms no/few side products and (vi) can be easily optimized by protein engineering for specific target substrates. **Cupin TM1459 combines all these requirements.**

Background

Aldehydes and ketones are accessible by many synthetic routes, one important tool being the **oxidative cleavage of alkenes**. **Chemical approaches** such as ozonolysis or metal-based methods display **several drawbacks**, including **explosive character** of the intermediates generated or **low yield** and poor **chemoselectivity**. Several enzymes are known to catalyze oxidative alkene cleavage reactions to different extent – in many cases as minor (undesired) side reaction. In most cases selectivity is lacking and many other products are observed.



acib-Technology

The **cupin TM1459** from *Thermotoga maritima* is a **small thermo- and solvent stable** β-barrel protein, which **catalyzes the oxidative alkene cleavage reaction in the presence of molecular oxygen and tertiary butyl hydroperoxide at ambient temperature**. We developed and optimized two **high-throughput assays for the detection of product aldehydes or ketones** and applied it during protein engineering of TM1459 (Steiner et al., Front Microbiol, 2016, 7:1511). The currently best variant catalyzes the formation of acetophenone from α-methylstyrene with **97% conversion** and shows a **broad substrate scope**.

acib-Offer

Under protection of a CDA we offer to discuss with you all available options. If a project looks feasible, **we offer evaluation** in a small PoC-study **if an alkene, which will result in the target aldehyde or ketone, is easily available from natural resources or can be easily synthesized.** The currently **available enzyme variants, including but not limited to Cupin TM1459, will be tested for the specific alkene substrate** and a strategy will be developed for the **optimization of the conversion** of the target alkene **including reaction and protein engineering**. Once we have confirmed that the target product can be synthesized, we will offer a **comprehensive project plan** for the reaction and protein engineering strategy.

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