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Advanced enzyme engineering concepts to create biocatalysis suitable for stereoselective synthesis of chiral intermediates and for carbohydrate conversion

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PURPOSE OF THE ABSTRACT

This lecture will highlight principle strategies and current challenges in enzyme discovery and protein engineering aiming to enhance their usefulness in biocatalytic applications [1] where also the combination of enzymes with chemocatalysts [2] and their incorporation into retrosynthetic concepts [3] play important roles.

For the synthesis of chiral amines, we engineered (S)-selective ATA for the acceptance of bulky ketones in the asymmetric synthesis of chiral amines [4].

Recently, we have shown a distinct metabolic function of P450-monooxygenases in the degradation of agarose or porphyran, where the P450 enzymes (originating from Formosa agariphila or Zobiella galactinovorans) together with appropriate redox partners catalyze the demethylation of 6-O-methyl-D-galactose [5]. Furthermore, we have determined the crystal structure of the P450 enzyme and identified key residues essential for catalysis and substrate recognition [6].

In addition, we could elucidate the entire metabolic pathway involved in the degradation of a major cell wall polysaccharide using a set of enzymes present in a marine bacterium in a distinct and so far unexplored polysaccharide utilization locus (PUL). The pathway consists of carbohydrate-active enzymes, including lyases, sulfatases and glycoside hydrolases that sequentially break down the complex polysaccharide into fermentable monosaccharides. For all previously unknown enzymes we performed a detailed biochemical characterization, determined several crystal structures and could identify the structures of all oligosaccharide intermediates formed during the complex enzymatic degradation by NMR spectroscopy and MS analysis [7].

FIGURES

FIGURE 1

FIGURE 2

KEYWORDS

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