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# Journal of Molecular Catalysis B: Enzymatic

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## Editorial

### Selected papers from the Biotrans 2009 Conference

The 9th International Symposium on Biocatalysis and Biotransformations was attended by over 500 scientists on 5–9 July 2009 in Berne, Switzerland. Recent advances ranging from directed evolution and metabolic engineering to enantioselective synthesis and process engineering were presented. These topics were featured at the conference in the form of 56 oral presentations and more than four hundred posters. Selected authors also accepted our invitation to submit a research paper in view of the present special issue of *The Journal of Molecular Catalysis B: Enzymatic* dedicated to the Biotrans 2009 conference. The manuscripts were subjected to rigorous peer review. A fair number of the submissions could be finally accepted and appear in the following pages.

Structure-based design and mutagenesis studies remains one of the most central themes in biocatalysis research today. Karl Hult and co-workers used a rational design approach to create the mutant *Candida antarctica* lipase B V190A having three times higher catalytic rate constant than the wild type enzyme for the transesterification of methyl methacrylate, and compared its reactivity to chemical acid catalysis and other enzymes showing that the enzyme reactivity is influenced mainly by electronic effects in the substrate. Marianne Graber and co-workers used site directed mutagenesis to redesign the stereospecificity pocket of the same enzyme, and in particular increased the enantioselectivity towards secondary alcohols by a single point isosteric mutations of Ser47 and Thr42. Modification of the hydrophobic tunnel leading to the active site also altered the enantioselectivity compared to the wild type enzyme, showing that accessibility can also influence enantioselectivity. In a more general perspective, Sangeeta Naik and co-workers propose a classification of the substrate selectivity of four groups of lipases which differ by the geometry of their binding site, providing a useful model for selecting enzymes for synthetic reactions. Finally, Bernd Nidetzky and co-workers report kinetic studies and in situ proton NMR analysis of the glucosylation of substituted phenol acceptors by *Leuconostoc mesenteroides* sucrose phosphorylase in the presence of  $\alpha$ -D-glucose 1-phosphate ( $\alpha$ G1P) as glycosyl donor, and the discovery of a mutated phosphorylase with suppressed hydrolase activity and 7-fold higher transfer yield.

The use of enzymes for selective organic synthesis was also prominent at the conference, and is reflected by several contributions in this issue. Czeslaw Wawrzęńczyk and co-workers report an original application of horse liver alcohol dehydrogenase for the regioselective oxidation of alkyl-branched 1,5 and 1,6-diols to form  $\delta$ - and  $\epsilon$ -lactones with 85–99% ee. Vicente Gotor and co-workers report their discovery of a new highly (S)-enantioselective whole cell reduction system for acetophenones in the form of growing cells of the phytopathogen fungus

*Lasiodiplodia theobromae* in potato-dextrose broth. Michael Kotik and co-workers report the identification of two new epoxide hydrolases discovered by screening a metagenomic library from biofilter-derived biomass using PCR with EH-specific degenerate primers followed by genome-walking PCR. Both enzymes show broad substrate specificity and high enantioselectivity, for example in the preparative scale enantioconvergent reaction of racemic cis-1-phenyl-1,2-epoxypropane to the corresponding (1R,2R)-diol with 97% yield and > 98% enantiomeric excess. Raffaele Morrone and co-workers report a new procedure for the highly enantioselective resolution of S-naproxen by esterification of the R-enantiomer with dimethyl carbonate using immobilized CAL-B. Sergio Riva and co-workers report the discovery of biphasic operating conditions for the laccase-catalyzed oxidation of para-alkyl phenols which produces Pummerer's ketones in good yields rather than polymerization products.

Studies of less common oxidases are appearing as new themes for innovation in biocatalysis. Maurice Franssen and co-workers report a new environmentally friendly preparation of 5-aminovaleric acid, a precursor of Nylon 5, from lysine using oxidation by L-lysine  $\alpha$ -oxidase from *Trichoderma viride* immobilized on an epoxy-activated solid support. Florence Husson and co-workers report the cloning and expression of fatty acid hydroperoxide lyase from green bell pepper in the yeast *Yarrowia lipolytica* and its purification by immobilized metal-ion affinity chromatography. The secondary structure content was measured by circular dichroism and differential scanning calorimetry and the results confirmed the theoretical model predicted by the ANTHEPROT software, achieving progress towards the structural understanding of this enzyme.

Biocatalysis under process conditions including ionic liquids and various immobilisation procedures was also intensely discussed at the conference, and is reflected in several contributions in this issue. Anju Chadha and co-workers investigated the performance of *Pseudomonas cepacia* lipase for esterification and transesterification reactions in ionic liquids and observed high yields and operational stability at room temperature for up to 10 months. Bastien Doumeche and co-workers covalently modified the formate dehydrogenase from *Candida boidinii* with hydroxyethyl-methylimidazolium and hydroxypropyl-methylimidazolium cations via carbonyldiimidazole coupling and thereby retained enzyme activity under ionic liquid conditions under which the unmodified enzyme is inactivated. Francis Peter and co-workers studied the reactivity of *Pseudomonas fluorescens* lipase Amano AK immobilized in a sol-gel for enantioselective acylation reactions in ionic liquids and organic solvents,

whereby highest enantioselectivities ( $E > 50$ ) occurred in acetone and tetrahydrofuran. Larissa Freitas and co-workers investigated esterification of glycerol with fatty acids using *Penicillium camembertii* lipase immobilized on epoxy SiO<sub>2</sub>-PVA in solvent free media, and found that the system achieves high specificity for both myristic and palmitic acids and produces a final mixture that fulfills the requirements established by the World Health Organization for food emulsifiers. Maria Ribeiro and co-workers report that the thermal stabilities and activities of the rhamnosidase and glucosidase activities of the naringinase enzyme complex are increased 4-fold respectively 15-fold under high-pressure. In a second paper, the author achieved an even superior stabilisation as well as compatibility with organic solvents by immobilising the enzyme in a tetramethoxysilane + glycerol sol-gel. Percival Zhang and co-workers cloned and expressed in *E. coli* the ORF Cthe0357 from the thermophilic bacterium *Clostridium thermocellum* ATCC 27405 that encodes a putative  $\alpha$ -glucan phosphorylase ( $\alpha$ GP), and discovered conditions for prolonged stability suitable for future process application of this thermophilic enzyme.

Food chemistry applications of enzymes were also a major topic. Ariela Veloso de Paula and co-workers report studies of the enzymatic transesterification of milkfat - soybean oil blends with a sn-1,3 specific lipase from *Rhizopus oryzae* immobilized on polysiloxane-polyvinyl alcohol matrix in a solvent free medium which demonstrate the capacity of system to change the triacylglycerol profile of the blend and obtain cold-spreadable milkfat. Suzana Ferreira-Dias and co-workers report on process stability

of enzymes in the production of human milk fat substitutes by transesterification of tripalmitin with oleic acid or omega-3 polyunsaturated fatty acids in solvent-free media using *Thermomyces lanuginosa* (Lipozyme TL IM), *C. antarctica* (Novozym 435) and *Candida parapsilosis* lipase/acyltransferases immobilized on Accurel MP 1000.

Progress in many other areas including enzyme production and application of enzymes for biofuel production were also discussed. Along these lines, Maristela Peres and co-workers report a study on the extracellular expression of the glycerol 3-phosphate dehydrogenase from *Saccharomyces cerevisiae* in *Pichia pastoris* under the control of the methanol-regulated AOX promoter. Joab Sampaio de Sousa and co-workers characterized a new lipase from germinated physic nut seeds for the production of high quality biodiesel fuel.

I hope that you will enjoy reading these research papers and that the results presented will inspire further contributions to the expanding field of biocatalysis and biotransformations.

Guest Editor, Chairman of Biotrans 2009

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