

Applications of Self-Immobilizing Decarboxylases in Flow Reactors

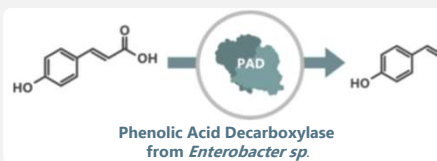
Astrid Winterhalter, Esther Mittmann, Martin Peng, Christof M. Niemeyer and Kersten S. Rabe
Institute for Biological Interfaces (IBG-1), Karlsruhe Institute of Technology

Flow systems are becoming a major focus in organic chemistry, while at the same time the interest in integrating biocatalysis is growing. Novel, flexible reactor concepts based on innovative immobilization techniques and biocatalytic materials are needed.^[1]

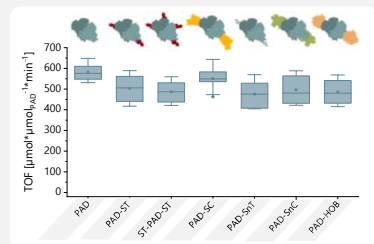
Enzyme Immobilization Techniques



Phenolic acid decarboxylases (PADs) enable the synthesis of industrially relevant styrenes from lignin-derived phenacrylates and thus the valorization of lignin product streams. Therefore, we used a dimeric, highly active and stable PAD from *Enterobacter sp.*



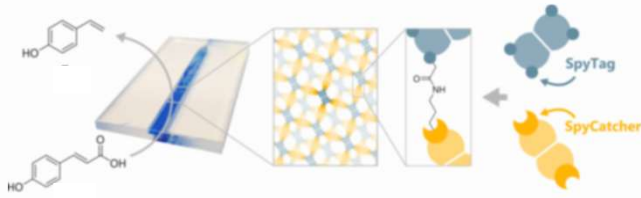
Peptide binding tags can be genetically integrated and allow self-immobilization of PAD without significantly affecting activity.



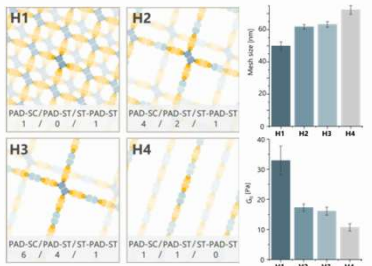
All-Enzyme Hydrogels

All-enzyme hydrogels are covalently crosslinked upon mixing via the genetically encoded SpyCatcher/SpyTag system. Attachment of one or multiple binding sites allow the use of mono- and multimeric enzyme building blocks, while variations in the stoichiometric ratios of multimeric fusion partners allows for the modulation of crosslinking degree and thus rheological properties.^[2]

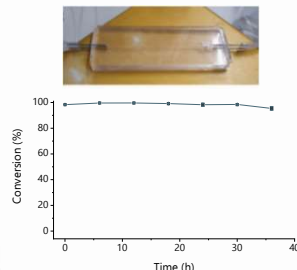
Hydrogel Formation



Variations in crosslinking degree

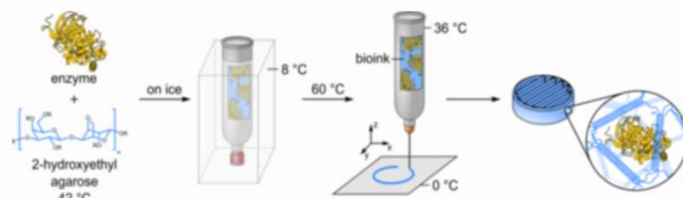


Application of H1 in Flow Reactors



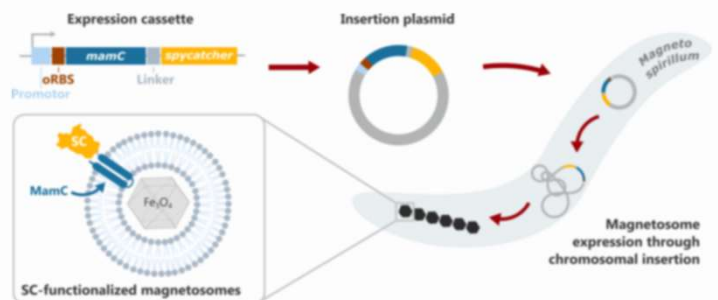
3D-printed Flow Reactor Modules

Unmodified thermostable enzymes are encapsulated in a 3D printed, agarose-based thermoreversible hydrogel and could be applied for continuous production of isopropanol or hydroxystyrenes.^[3]

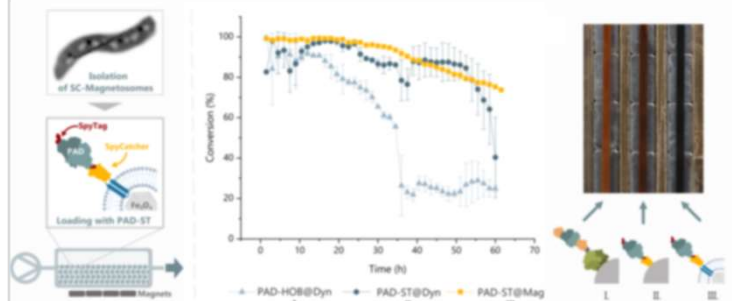


Enzyme Immobilization on Biogenic Magnetic Nanoparticles

The display of SC-connectors on the surface of genetically engineered bacterial magnetosomes generates a multimodal platform for the immobilization of foreign protein cargo.



The immobilized PAD showed superior stability. In flow systems, the PAD-loaded SC-magnetosomes (PAD-ST@Mag) outperformed similarly functionalized commercial particle formulations.^[4]



SC-Magnetosomes were provided by Frank Mikoleit from the lab of Dirk Schüler, University of Bayreuth.

[1] T. Peschke, P. Bitterwolf, S. Hansen, J. Gasmi, K. S. Rabe, C. M. Niemeyer, *Catalysts*, **2019**, 9, 164.

[2] E. Mittmann, S. Gallus, P. Bitterwolf, C. Oelschlaeger, N. Willenbacher, C. M. Niemeyer, K. S. Rabe, *Micromachines*, **2019**, 10, 795.

[3] M. Peng, E. Mittmann, L. Wenger, J. Hubbuch, M. K. M. Engqvist, C. M. Niemeyer, K. S. Rabe, *Chem-Eur. J.* **2019**, 25, 15998.

[4] E. Mittmann, F. Mikoleit, D. S. Maier, M. A. Klein, C. M. Niemeyer, K. S. Rabe, D. Schüler, *ACS Appl. Mater. Interfaces*, **2022**.

Contact us!



Visit our homepage: niemeyer-lab.de Mail to: astrid.winterhalter@kit.edu