Organic Chemistry: An Indian Journal

Editorial | Vol 15 Iss 6

Multienzymatic Cascade Reactions *via* Immobilization of Enzyme Complexes

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Received: June 03, 2021; **Accepted:** June 14, 2021; **Published:** June 25, 2021

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Multienzymatic cascade reactions are a crucial technology for industrial process development, such as pharmaceutical, cosmetic, and nutritional component synthesis. Various multienzyme structure construction methodologies have been widely described. Fusion proteins, enzyme scaffolds, and immobilisation are three methods for creating enzyme complexes. As a result of substrate channelling, enzyme complexes can boost cascade enzymatic activity. Recent breakthroughs in materials science have led to the creation of a variety of materials that can be used to immobilise enzymes. This review looks at how random coimmobilization, compartmentalization, and positional coimmobilization can be used to assemble multienzyme complexes. Immobilized multienzymes have several advantages, including greater cascade enzymatic activity *via* substrate channelling, improved enzyme stability, and ease of recovery for reuse. We also include the most recent investigations of several model enzyme reactions immobilised on diverse support materials in this review, as multienzyme systems allow for cost-effective product production via bioprocesses. Biocatalysts, which relate to complete cells or enzymes, are natural catalysts. They greatly boost the speeds of chemical processes in living organisms by lowering the activation energy of a reaction. Inside a cell, a variety of chemical processes take occur to enable cellular growth and survival. Enzymes have evolved to catalyse a wide range of chemical processes in cells in order to survive in biological systems' metabolic pathways, resulting in high selectivity and specificity. Many enzyme cascade reactions can be found in a variety of metabolic routes within the cell in nature, and they are used to ensure the integrity of enzyme-catalyzed synthetic pathways that imitate chemical processes. Several enzymes work together to perform multi reaction steps or cascade reactions, resulting in the induction of two or more sequential processes without the need to isolate intermediates. Fermentation with local microbes, such as for beer brewing or vinegar manufacture, is the oldest example of a multi enzymatic reaction. In vitro enzymatic synthesis has distinct advantages over an *in vivo* fermentation technique. Engineering *in vitro* processes has the flexibility to address problems including cell viability, complexity, physiology, and the cell membrane or cell wall. The traditional method for multi enzymatic reactions *in vitro*, on the other hand, is carried out in steps. Low yields, high operating costs, and the employment of numerous chemicals in the separation phases are all downsides of this method. To address these challenges, a new technique is required that can give benefits such as enantioselectivity, stereoselectivity, high yield, low downstream costs, shifting reaction equilibria, and multiple step execution without product recovery. Biotransformations, biosensors, and biomedical engineering are just a few of the scientific and industrial uses of multienzyme reactions that have recently exploded in popularity. According to the Hess group, despite the fact that immobilisation causes multienzymes to be placed in close proximity to one another, activity has been observed to increase in many studies of the Glucose Oxidase (GOD)/Horseradish Peroxidase (HRP) cascade as a result of microenvironment effects on the enzyme and the resulting cascade kinetics. During the coimmobilization process, reduced activity can also be detected, which is attributable to enzyme autolysis and structural alterations. Multienzyme cascade reactions have recently attracted a lot of attention, not only because of their beauty, but also because they allow for the employment of more complicated systems.

Citation: Yuong R. Multienzymatic Cascade Reactions *via* Immobilization of Enzyme Complexes. Org Chem Ind J. 2021;15(6):25