

CURRENT RESEARCH AND DEVELOPMENT IN BIOTECHNOLOGY ENGINEERING AT IIUM

VOLUME II

Editors:

Ibrahim Ali Noorbatcha
Hamzah Mohd. Salleh
Mohamed Elwathig Saeed Mirghani
Raha Ahmad Raus



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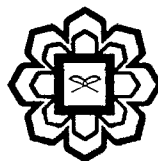
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CHAPTER 17

IMMOBILIZATION OF LIPASE BY CROSS-LINKED ENZYME AGGREGATE (CLEA) TECHNOLOGY

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ABSTRACT

Cross-linked enzyme aggregated (CLEA) is a new method of carrier-free immobilized enzyme for biocatalysis as a replacement for carrier-bound immobilized enzyme. This is to overcome the disadvantages of carrier-bound immobilized enzymes which is associated with large amount of non-catalytic mass and expensive carrier beads. The objective of this project is to optimize the condition parameters for the preparation of CLEA-Lipase for highest lipase activity and comparing its efficiency to the free enzyme system. Experiments are designed according to Response Surface Methodology (RSM) and conducted using central composite design (CCD). Enzyme is precipitated which are then cross-linked. The parameters varied were concentration of precipitant and cross-linker. Two different types of precipitant were used, ammonium sulfate (65-75 % w/v) and acetone (30-60 % v/v). The concentration of cross-linker, glutaraldehyde, was varied from 60 to 80 mM. The results showed that lipase enzyme can be precipitated in both ammonium sulfate and acetone without denaturation. The lipase immobilized in $(\text{NH}_4)_2\text{SO}_4$ showed the highest enzymatic rate of 2.7 $\mu\text{M/s}$ while in acetone the highest rate is 2.15 $\mu\text{M/s}$. The CLEA-Lipase when compared with the free enzyme system showed lower activity. Enzyme reusability tested shows that CLEA residual activity decreased upon usage at the second and third cycle. Results from this study suggested that CLEA-lipase is a promising new low cost enzyme preparation process which can benefit various industrial applications such as food, flavor, pharmaceuticals as well as fine chemicals.

Keywords: immobilization, cross-linked enzyme aggregate (CLEA), lipase, carrier-free, precipitant, cross-linker

INTRODUCTION

Enzymes are the vital component that facilitate or accelerate a chemical and biological reaction without changing their properties at the end of the reaction. An enzyme is a protein in the cell which lowers the activation energy of the reaction to increase the rate of the reaction. For thousands of years, enzymes have been used for domestic and commercial purposes