IMMOBILIZATION OF LIPASE PRODUCED BY *RHIZOPUS ARRHIZUS* ON CHITOSAN COATED POLYGALACTURONIC ACID BEADS

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Abstract

Immobilization of enzymes is considered to be a technique with potential to improve protein properties and to broaden enzymes applications. Very often, immobilized enzymes display better thermal stability, and some of their characteristics are changed. Immobilized lipases may find application in catalyzing transesterification reactions in an organic solvent medium. The aim of the present study is to investigate the possibility of immobilization of lipase obtained by solid- state fermentation of *Rhizopus arrhizus* on chitosan coated polygalacturonic acid (PGA) beads and to investigate the biochemical characteristics of the resulting immobilized enzymes.

The effect of time, temperature, pH of the immobilization medium, and enzyme activity on the efficiency of lipase immobilization on chitosan coated PGA beads was investigated. The optimal conditions were determined and the achieved efficiency of immobilization was 23%. The optimum pH for the action of immobilized lipase was pH 9, maintaining about 80% of its activity at pH 10, while the free enzyme was almost completely inactivated at pH 10. The immobilized lipase and the free enzyme had a similar temperature optimum at 40 - 45 ^{0}C, but the immobilized lipase exhibited significant activity of about 70% at 55 ^{0}C while under these conditions the free enzyme was almost inactivated. The thermodynamic parameters of the inactivation of the immobilized lipase were determined, which contributed to its biochemical characterization. The pH and temperature stability at different temperatures and pH were monitored. The immobilized lipase was found to be activated in the presence of some organic solvents such as hexane, Dimethyl sulfoxide (DMSO) and dimethylformamide (DMF), which allowed its use in transesterification reactions. The immobilized enzyme was characterized by good reuse stability. In an aqueous medium, it retained about 50% of its activity after six consecutive cycles of use, and in an organic environment of hexane the immobilized enzyme retained about 80% of its transesterification activity.

Resulting immobilized enzyme differed in its biochemical properties from the free enzyme. The immobilized enzyme exhibited significant activity over a wide pH range of 6 - 10 and a temperature range of 30 - 65 ^{0}C. The immobilized enzyme was significantly more stable than the free enzyme under the tested conditions. As a result of the immobilization, changes in the properties of the enzyme were observed, which broaden the possibility of its application.

Key words: *Lipase, Rhizopus arrhizus, Immobilization, Properties.*