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<AT>Stability of Immobilized Porcine Pancreas Lipase on Mesoporous Chitosan Beads: A Comparative Study

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<PA>*Corresponding author **Highlights**▶.

<ABS-Head><ABS-HEAD>Graphical abstract

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<ABS-P><xps:span class="xps_Image">fx1</xps:span><ABS-HEAD> ▶ Highlights ▶ Use of glutaraldehyde improved chitosan bead porosity by 5 folds. ▶ Enzyme efficiency of combined immobilization was enhanced by 2.6 folds. ▶ Five different techniques of immobilization were thoroughly compared. ▶ Distribution of enzyme in chitosan beads were demonstrated using fluorescence image. ▶ Kinetic parameters for immobilization methods were determined and compared.

<ABS-HEAD>**Abstract**

<ABS-P>Porcine pancreas lipase was immobilized on mesoporous chitosan beads. Glutaraldehyde as coupling agent was used through several immobilization techniques. With the aid of FESEM, BET and BJH analysis, the effect of glutaraldehyde on porosity of chitosan was evaluated. It was observed that the total surface area and pore volume of the carrier were significantly improved by addition of glutaraldehyde as cross-linking agent. The surface area exposure and pore volume were substantially increased (both by 4.4 folds). In addition, distribution of enzyme on the carrier was illustrated by fluorescence image. The characteristics of the immobilized lipases such as immobilization efficiency, enzyme activity, pH stability, thermal stability, reusability, storage stability and enzyme leakage were evaluated. In kinetic studies of enzyme, Michaelis–Menten kinetic coefficients of the hydrolytic activity for the immobilized lipase were defined using Lineweaver–Burk plot. The low value of ionization constant, K_m (~ 0.008 mM) and high value of specific rate, V_{max} (~200 $\mu\text{M}/\text{ml}\cdot\text{min}$) indicate strong affinity and high activity of

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