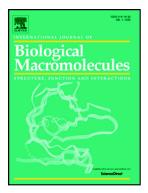
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## **ACCEPTED MANUSCRIPT**

# Catalytic characteristics and application of L-asparaginase immobilized on aluminum oxide pellets

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#### Abstract

L-asparaginase from *Escherichia coli* (L-ASNase) was covalently immobilized on aluminum oxide pellets (AlOPs) using a cross-linking agent, glutaraldehyde. Maximum immobilization yield (85.0%) was obtained after optimizing immobilization parameters using response surface methodology (RSM). Both free and immobilized L-ASNase (AlOP–ASNase) were optimally active at 37°C and pH 7.5. However, the bioconjugate exhibited enhanced activity and stability at different pH and temperatures. It had higher affinity (low  $K_m$ ) and was comparatively more stable in presence of some solvents (ethyl acetate, acetone, acetonitrile), metal ions (Ag<sup>+</sup>, Zn<sup>2+</sup>) and  $\beta$ -mercaptoethanol. AlOP–ASNase was reused in a glass column reactor for L-asparagine hydrolysis upto nine successive cycles without any loss in activity. The AlOP–ASNase was effective in lowering L-asparagine level in blanched potato chips indicating its potential use in mitigating acrylamide formation in starchy foods. This cost-effective enzyme preparation had shelf-life of more than 30 days and can be effectively used in food industry in mitigating acrylamide formation in fried starchy foods.

Keywords: Acrylamide, aluminum oxide, immobilization, L-asparaginase, potato chips, RSM.

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