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Comparative analysis of the kinetic and thermodynamic properties of native and recombinant L-asparaginase from *Pseudomonas fluorescens***Hare Ram Singh and Santosh Kr Jha**
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L-Asparaginase (EC 3.5.1.1) is used as an antineoplastic drug in the treatment of Acute Lymphoblastic Leukemia (ALL) and is in high demand all over the world. In the present report, kinetic and thermodynamic properties of the native and recombinant form of L-asparaginase from *Pseudomonas fluorescens* were studied and compared. Both the native and recombinant form of L-asparaginase were produced and purified with quite higher yield. After purification, the specific activity of 1.025×10^4 U/mg and 3.913×10^2 U/mg and fold purification of 9.64 and 2.34 was achieved for recombinant and native L-asparaginase respectively. The kinetic parameters such as K_m , V_{max} , K_{cat} and K_{cat}/K_m for the native L-asparaginase was found to be 4.51 mmol, 28.575 $\mu\text{mol/ml/min}$, 31.74 s^{-1} and 7038.67 mmol^{-1} respectively while 4.481 mmol, 62.5 $\mu\text{mol/ml/min}$, 52.08 s^{-1} and 1296.14 mmol^{-1} respectively were obtained that for recombinant L-asparaginase. This implied that the recombinant L-asparaginase although showed a slightly higher K_m value than that of the native one but have higher V_{max} value as well as the kinetic efficiency. The optimum pH for the native and recombinant enzyme was found to be pH 7 while optimum temperature for the activity was found to be 37°C for both the enzyme. The deactivation energy for the native and recombinant L-asparaginase was found to be 50.54 kJ/mol and 31.27 kJ/mol respectively. The thermodynamic parameters including ΔH , ΔS and ΔG were determined and found to be 47.98 kJ/mol, 0.078 kJ/mol K and 71.22-72.78 kJ/mol respectively for the native enzyme while 28.72 kJ/mol, 0.014 kJ/mol K and -32.89-33.17 kJ/mol respectively for the recombinant form. This indicates that there are no significant processes of aggregation and the enzymatic reaction was exothermic and spontaneous in nature.

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