Experimental and bioinformatics study for production of l-asparaginase from Bacillus licheniformis: a promising enzyme for medical application

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Abstract

A Bacillus licheniformis isolate with high L-asparaginase productivity was recovered upon screening two hundred soil samples. This isolate produces the two types of bacterial L-asparaginases, the intracellular type I and the extracellular type II. The catalytic activity of type II enzyme was much higher than that of type I and reached about 5.5 IU/ml/h. Bioinformatics analysis revealed that L-asparaginases of Bacillus licheniformis is clustered with those of Bacillus subtilis, Bacillus haloterans, Bacillus mojavensis and Bacillus tequilensis while it exhibits distant relatedness to L-asparaginases of other Bacillus subtilis species as well as to those of Bacillus amyloliquefaciens and Bacillus velezensis species. Upon comparison of Bacillus licheniformis L-asparaginase to those of the two FDA approved L-asparaginases of E. coli (marketed as Elspar) and Erwinia chrysanthemi (marketed as Erwinaze), it observed in a cluster distinct from- and with validly predicted antigenic regions number comparable to those of the two mentioned reference strains. It exhibited maximum activity at 40 ÁC, pH 8.6, 40 mM asparagine, 10 mM zinc sulphate and could withstand 500 mM NaCl and retain 70% of its activity at 70 ÁC for 30 min exposure time. Isolate enzyme productivity was improved by gamma irradiation and optimized by RSM experimental design (BoxïBehnken central composite design). The optimum conditions for maximum L-asparaginase production by the improved mutant were 39.57 ÁC, 7.39 pH, 20.74 h, 196.40 rpm, 0.5% glucose, 0.1% ammonium chloride, and 10 mM magnesium sulphate. Taken together, Bacillus licheniformis L-asparaginase can be considered as a promising candidate for clinical application as antileukemic agent

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