Optimization of Fermentation Conditions for Extracellular Production of the Oncolytic Enzyme, L-Asparaginase, by New Subsp. Streptomyces Rochei Subsp. Chromatogenes NEAE-K Using Response Surface Methodology under Solid State Fermentation

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Abstract : L-asparaginase is an important enzyme as therapeutic agents used in combination therapy with other drugs in the treatment of acute lymphoblastic leukemia in children. L-asparaginase producing actinomycete strain, NEAE-K, was isolated from soil sample and identified on the basis of morphological, cultural, physiological and biochemical properties, together with 16S rDNA sequence as new subsp. Streptomyces rochei subsp. chromatogenes NEAE-K and sequencing product (1532 bp) was deposited in the GenBank database under accession number KJ200343. The study was conducted to screen parameters affecting the production of L-asparaginase by Streptomyces rochei subsp. chromatogenes NEAE-K on solid state fermentation using Plackett-Burman experimental design. Sixteen different independent variables including incubation time, moisture content, inoculum size, temperature, pH, soybean meal+ wheat bran, dextrose, fructose, L-asparagine, yeast extract, KNO3, K2HPO4, MgSO4.7H2O, NaCl, FeSO4. 7H2O, CaCl2, and three dummy variables were screened in Plackett-Burman experimental design of 20 trials. The most significant independent variables affecting enzyme production (dextrose, L-asparagine and K2HPO4) were further optimized by the central composite design. As a result, a medium of the following formula is the optimum for producing an extracellular L-asparaginase by Streptomyces rochei subsp. chromatogenes NEAE-K from solid state fermentation: g/L (soybean meal+ wheat bran 15, dextrose 3, fructose 4, L-asparagine 8, yeast extract 2, KNO3 1, K2HPO4 2, MgSO4.7H2O 0.5, NaCl 0.1, FeSO4. 7H2O 0.02, CaCl2 0.01), incubation time 7 days, moisture content 50%, inoculum size 3 mL, temperature 30°C, pH 8.5.

Keywords : streptomyces rochei subsp. chromatogenes neae-k, 16s rrna, identification, solid state fermentation, l-asparaginase production, plackett-burman design, central composite design

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