Title: STREPTOMYCES GOUGEROTII C1.129 PRODUCER OF L-ASPARAGINASE

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Abstract: L-Asparaginase is an enzyme of great medical importance, due of its chemotherapeutic action by extensive capacity to hydrolyze the amino acid L-asparagine in aspartic acid and ammonia. A wide variety of micro-organisms such as yeast filamentous fungi, bacteria and actinomycetes are capable of producing L-asparaginase. This enzyme has been widely used against several types of cancers, the main choice for the treatment of acute lymphoblastic leukemia. The objective of this study was to evaluate the production of Lasparaginase by Streptomyces gougerotii C1.129 isolated from Caatinga and optimize the conditions of production of this enzyme. Initially a qualitative assay was performed by adding 15 uL of the standardized inoculum of actinobacteria in CZ and M9 media with addition of phenol red as an indicator and L-asparagine as substrates. For the quantitative determination of Lasparaginase, the micro-organism was fermented and some parameters such as carbon sources (M9 media and TGY), fermentation time (24 to 120 hours), pH (5 to 9) and temperature (25 °C to 50 °C) were evaluated for optimization of enzyme production conditions. The quantitative assay parameters were determined by measuring the amount of ammonia formed by nesslerization, where 1 U of L-asparaginase was equal to the amount of enzyme that releases 1 µM of ammonia per minute at 37 °C. The qualitative analysis showed the production of the enzyme through a pink halo around the colony. Through the quantitative assay was determined that the M9 medium was best suited for production of the enzyme, with 101.9 U/ml. a value 3.75 times greater than in the TGY medium. The kinetics showed a peak enzyme production in a 96 hour period with values that reached 189.9 U/ml. The pH of 6 was considered optimal for the production of L-asparaginase as compared to other pH, having an amount of 937.18 U/ml. The 35 °C temperature corresponded to more favorable condition for enzyme production, reaching 273.83 U/ml among the analyzed temperatures. The optimized conditions allow this way, the large-scale production to be successful, with levels of L-asparaginase quite high. The isolated from actinomycetes of Caatinga have shown promise for production of various enzymes including L-asparaginase. The study of bioprospecting of these microorganisms originating from Brazilian biomes, for their great biotechnological potential and possible use of the product by the pharmaceutical industry is essential.

Keywords: Optimization; Asparaginase; Acute Lymphoblastic Leukemia.

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