

TITLE: Glutaminase-free L-asparaginase production by *Leucosporidium muscorum* isolated from Antarctic marine-sediments

AUTHORS: FREIRE, RKB¹, FERRARO, RB¹, LOURENÇO, FR², SETTE, LD³, PESSOA-JR, A¹

INSTITUTIONS: ¹Department of Biochemical and Pharmaceutical Technology, School of Pharmaceutical Sciences, University of Sao Paulo, Av. Prof. Lineu Prestes, 580, 05508-900 Sao Paulo, SP, Brazil; ²Department of Pharmacy, School of Pharmaceutical Sciences, University of Sao Paulo, Av. Prof. Lineu Prestes, 580, 05508-900 Sao Paulo, SP, Brazil; ³Department of Biochemistry and Microbiology, Institute of Biosciences, Statal Paulista University “Julio de Mesquita Filho” (UNESP), Av. 24A, 1515, 13506-900 Rio Claro, SP, Brazil.

ABSTRACT:

L-asparaginase (ASNase) is an important therapeutic agent used in the treatment of acute lymphoblastic leukemia (ALL). Bacterial ASNase preparations available on the market have some limitations related to their low stability in serum, which lead to severe immunological reactions in 3-78% of patients. Yeasts are highlighted as important microorganisms for ASNase production since they are able of producing similar enzymes to human congeners and with fewer side effects. In this work, the screening of 145 yeasts isolated from marine-sediments in King George Island, Antarctica, resulted in nine glutaminase-free L-asparaginase producing yeasts. A strain of *Leucosporidium muscorum* was the isolated that yielded the highest ASNase activity (490.41 U.L⁻¹) and volumetric productivity (5.12 U.L⁻¹.h⁻¹). Carbon and nitrogen sources were evaluated by a method of variation in one factor at a time. Sucrose, yeast extract and proline resulted in maximal production of enzyme and were selected for Czapek Dox Medium (CDM) optimization by full factorial design. Optimum media condition for yeast growth and ASNase yield were 20 g.L⁻¹ sucrose, 15 g.L⁻¹ yeast extract and 20 g.L⁻¹ proline and resulted in 4,582.5 U.L⁻¹ of enzyme and 63.64 U.L⁻¹.h⁻¹ of volumetric productivity. This is the first report on production of ASNase by a cold-adapted yeast which may indicate a new potential source of glutaminase-free L-asparaginase for commercial purpose.

Keywords: L-asparaginase . *Leucosporidium muscorum* . Antarctica . psychrophilus . leukemia . factorial design

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