PRODUCTION, PURIFICATION AND BIOCHEMISTRY CHARACTERIZATION OF A EXTRACELLULAR *KLUYVEROMYCES MARXIANUS* INULINASE

ALVES, M. S¹; REIS, P. A. B²; MARQUES, V. M³; MAGALHÃES, R. D. M.⁴; FARIA, J. A. Q. A.⁵; MAIA, T. A.⁶

Departamento de Bioquímica e Biologia Molecular¹, Centro de Ciências Biológicas e da Saúde¹, Universidade Federal de Viçosa¹.

Inulinase is also known as 2, 1-beta-D-fructanfructanohydrolase EC 3.2.1.7. It is a constitutive enzyme in Kluyveromyces marxianus that catalyzes the hydrolysis of beta-1,2 fructofuranosidic bonds present at sucrose and inulin. Used at sucrose hydrolysis for glucose and fructose syrups, at food and pharmaceutical industries. Here the product obtained has better quality, is more soluble and sweetest than the older one. The goal of this research study is the production, purification and biochemistry characterization of Kluyveromyces marxianus inulinase. The yeast was grown at liquid medium containing sucrose as the only carbon source. The enzymatic activity was carried out utilizing the DNS reactive. Bradford assay was used to measure the protein concentration at the extract. At the purification step, the ionic chromatography (DEAE-Sepharose) was used and polyacrylamide gel electrophoresis was used after. Time, pH, temperature and substrate concentration assays were carried out at enzyme activity. Inulinase was purified 16, 5 times and molecular weight estimated using SDS-PAGE was 107.451 Da. The temperature and the pH assays were 60°C and 4, 41 respectively. Km for sucrose was 10, 27 mM. The project development was consistent with the proposed objectives and the results obtained were coherents with the data at the literature.