TITLE: OPTIMIZATION OF THE β-GALACTOSIDASE PRODUTION BY *TRICHODERMA* sp. UNDER SOLID-STATE FERMENTATION USING CCRD

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ABSTRACT

Lactose is the first source of carbohydrate for mammals. Its absorption depends on the enzyme β-galactosidase (EC 3.2.1.23) that hydrolyzes this disaccharide obtaining glucose and galactose. β-galactosidases have a wide application in different sectors as pharmaceutical and food industries, and in the treatment of effluents. In addition, these enzymes have ability to perform transgalactosylation reactions producing galactooligosaccharides (GOS). Currently, microorganisms are the mains source of β-galactosidases, especially filamentous fungi due to your ability to produce extracellular enzymes facilitating the recovery and purification. In this context, the aim was to optimize the fermentation parameters for the production of extracellular β-galactosidase by *Trichoderma* sp. under Solid-State Fermentation (SSF) using the central composite rotational design (CCRD). Initially, the enzyme production was evaluated using different substrates (sugarcane bagasse, orange peel, rice bran, corn bran, soy bran, wheat bran, oat flour and rye flour) and moistening agent (distilled water, tap water and salt solutions). The SSF was conducted for 120h at 30°C. The highest enzyme activity was obtained using wheat bran as substrate and tap water as moistening. After, the CCRD was applied to analyze the influences of the time of cultivation (120-264h), inoculum concentration (10^4-10^6 spores/mL) and proportion between wheat bran as substrate and tap water as moistening (1:1-1:2, w/v). After fermentation, 50 mL cold distilled water were added into SSF and maintained at 140 rpm for 20 min at 4° C, followed by vacuum filtration. The extracellular cell-free filtrate was used as the enzyme source. The β-galactosidase activity was determined using the glucose oxidase method. The conditions that allowed the best enzyme production (predicted value 2.60 U/g of substrate; experimental value 2.67 U/g of substrate) were inoculum using 10^5 spores/mL, moisture at 1:1.65 (w/v) and cultivation for 184h. The best temperature and pH for the activity of the enzyme obtained under optimized conditions were 55°C and 4.5, respectively. The fungus *Trichoderma* sp. produced β-galactosidase in SSF and after the use of CCRD activity was increased by 8.6 fold. This highlights a great potential of production using FES in optimal conditions, thus enabling future applications of this enzyme.

Keywords: Lactase, galactooligossarídeos (GOS), experimental planning

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