

**Title:** PRODUCTION OF  $\beta$ -GALACTOSIDASE FROM CHEESE WHEY USING *KLUYVEROMYCES MARXIANUS* CBS 6556

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**Abstract:**

The enzyme  $\beta$ -galactosidase produced by microorganisms hydrolyzes sugar present in milk and dairy foods, lactose. The enzymatic hydrolysis of lactose has been an alternative to the food industries, because the result sugars of this process, glucose and galactose, are more soluble and sweet. On the other hand, deficient production of  $\beta$ -galactosidase can cause a syndrome known as lactose intolerance, with symptoms such as bloating, cramping, diarrhea, headaches, among other discomforts. Lactose intolerance is associated with a deficiency on enzyme lactase production, which is an alternative for lactose-intolerant people is ingesting lactase supplements. Due to the increased of the industrial demand of  $\beta$ -galactosidase and most of the  $\beta$ -galactosidase consumed in Brazil is imported, there is a need of to study the agitation and aeration, to obtain a product of high activity. In this context, the aim of this work, was to study the production of  $\beta$ -galactosidase from *Kluyveromyces marxianus* CBS 6556 using alternative sources of carbon and nitrogen aiming in evaluating the effect of concentrations of substrates, temperature, agitation and aeration on enzyme production. Preliminary results from an experimental design (25-1) showed that Prodex-lac® and agitation weren't significant variables. For the purpose of optimizing the enzyme production, a new factorial design was used. The central composite rotational design (CCRD) had cheese whey (100 to 1,000 mL L<sup>-1</sup>) as a carbon source, corn steep liquor (0 to 18 g L<sup>-1</sup>) as an nitrogen source and temperature (25 to 45 °C) as independent variables. The experiments were conducted in Erlenmeyer flasks (1000mL) containing 300 mL of culture medium for 24 h with agitation of 220 min<sup>-1</sup>. Observing the the values of  $\beta$ -galactosidase activity, as well as significant parameters indicated in the treatment of the data, it's conclude that the experiment containing cheese whey (1,000 mL L<sup>-1</sup>) and corn steep liquor (14.36 g L<sup>-1</sup>) carried out at 31°C, was the most suitable for the production of  $\beta$ -galactosidase obtaining 9.8 U mL<sup>-1</sup> for the enzymatic activity. Through response surface methodology, the optimal condition for enzyme production has been established and validated in triplicate under the conditions cheese whey (1000 mL L<sup>-1</sup>) and corn steep liquor (18 g L<sup>-1</sup>) and 31 °C reaching an average volumetric activity enzyme, 9.1% lower than the value predicted by the model generated. In the following tests, under the same condition the influence of the agitation and the aeration was evaluated in the enzyme production by submerged fermentation in Biostat B fermenter of 2 L, the study conditions were: 200 min<sup>-1</sup> together 1.33 vvm (KLa = 19 h<sup>-1</sup>), and 400 min<sup>-1</sup> together 2.67 vvm (KLa = 53 h<sup>-1</sup>). In this study the agitation and aeration, influenced in the  $\beta$ -galactosidase production, and the favorable condition was 400 rpm and 2.67 vvm, respectively, obtaining 6.59 ± 0.20 U mL<sup>-1</sup> for the enzymatic activity.

**Keywords:** *Kluyveromyces marxianus*,  $\beta$ -galactosidase, cheese whey, corn steep liquor, agitation, aeration.