TITLE: USE OF GLYCEROL AND SUCROSE AS LOW-COST ALTERNATIVES TO GLUCOSE IN THE PRODUCTION OF PECTINASES BY Aspergillus oryzae IN SUBMERGED PROCESS

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Abstract:

The composition of the cultivation medium is responsible for induction, catabolic repression and inhibition in the production of fungal pectinases. The carbon source is a component that particularly affects both cell growth and enzyme production. Glucose is a most important effector in microbial metabolism and its presence in the cultivation medium could be responsible for the repression of transcription of genes that encode enzymes which are related to the metabolism of other substrates such as pectin. The aim of this work was to assess the influence of glycerol and sucrose, as low-cost alternative carbon sources to glucose, on growth and pectinase formation by Aspergillus oryzae IPT-301 in liquid media. The cultivation media used in the tests had the following compositions (g/L): carbon source, 5.0; wheat bran (aqueous extract), 40; citric pectin (CPKelco), 20; yeast extract, 0.05; (NH₄)₂SO₄, 5.0; MgSO₄, 0.5; KH₂PO₄, 2.5; $FeSO_4 \cdot 7H2O$, 6.3×10^{-4} ; $ZnSO_4$, 6.2×10^{-4} ; $MnSO_4$, 1.0×10^{-5} . The experiments were carried out in duplicate in 500-mL Erlenmeyer flasks containing 100 mL of culture medium, under reciprocal agitation of 300 rpm (Certomat, Sartorious Stedium Biotech), at 28°C and initial pH of 4.0. Initially, 24-hour runs in media without pectin were performed to evaluate the effect of each substrate on the fungal growth. Furthermore, the production of pectinases was assessed in 144-hour cultivations in pectin-containing media. Cell biomass concentration was gravimetrically determined and pectinase activity was estimated by measuring the reduction of viscosity of a standard pectin solution. When glucose was added to the medium, the maximal biomass concentration (3.7 g/L) was lower than those achieved with sucrose (4.1 g/L) and glycerol (4.7 g/L). With the presence of pectin in the medium, cell concentrations close to 9.0 g/L were quantified for any substrate examined. On the other hand, the highest pectinase activities were attained in media containing glycerol and sucrose (11.4 and 11.5 U/mL, respectively), whereas with glucose the activity reached 9.5 U/mL. Although still preliminary, these results indicate that the use of glycerol and sucrose instead of glucose could be advantageous for the production of pectinases by A. oryzae. In the following steps of this research, the process will be evaluated under well-controlled conditions in bench bioreactor in which the achievement of higher enzyme activities is expected.

Keywords: pectinases, *Aspergillus oryzae*, carbon sources.

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