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AOX expression is affected negatively for low concentration of methanol in cultures of yeast *Pichiapastoris*

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In methylotrophic yeasts, which are able to be used as sole carbon source as methanol, the first step is the oxidation of methanol by the alcohol oxidase (AOX1) located in the peroxisome. Thus, the yeast *Pichiapastoris* is an excellent model for the expression of heterologous proteins, which is reflected in the large number of proteins that have been obtained in the biological model, under control of the AOX1 promoter. For this reason, the aim of this study is to determine the most optimal growing condition for *P. pastoris* recombinant allows to obtain high expression of AOX. To conduct the study, we evaluated three critical factors for temperature (14°C, 18°C, 22°C) and methanol concentration (1%, 1.5% and 2%). The quantification of the enzyme AOX was performed by immunofluorescence through flow cytometry. The results show that during 24h of induction all cultures exhibit the same behavior (increased median fluorescence intensity; MIF), subsequent to this period is observed that cultures with high concentrations of methanol exhibit a decrease at 48h and ending at 72h with an increase in MIF; while the culture at 14°C+1%M, MIF shows a peak at 48h and suffers a sharp drop after this; otherwise, the culture to 18°C+1.5%M presents the most stable behavior during the induction. In conclusion, the culture conditions with a concentration of 1% (v/v) of methanol are those with the low concentration of enzyme AOX, which at 72h implies a decrease in the production of heterologous proteins.

Biography

Zepeda A B is Biotechnology Engineer and she is coursing her Sandwich PhD program from the Universidad de La Frontera and the Universidade de São Paulo. She is member of the Sociedad de Farmacología de Chile since 2012. She has worked in the areas of Hypoxia, Cellular and Molecular Biology and Pharmaceutical Biotechnology.

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