SELECTION OF *Saccharomyces cerevisiae* strains TOLERANT TO BAGASSE HYDROLYSATE WITH SUGARCANE MOLASSES, A SUBSTRATE TO SECOND-GENERATION ETHANOL PRODUCTION

Miranda, E.S.[1], Varize, C.S. [1], Christofoleti-Furlan, R.M. [1], Mota, E.C. [1], Silvello, C. [1], Florencio JR, O. [1], Dalia, R.L. [1], Basso, L.C. [1]

[1] Esalq/USP – Escola Superior de Agricultura “Luiz de Queiroz”. Universidade de São Paulo (Av. Pádua Dias, 11 - CEP 13418-900 – Piracicaba – SP)

Abstract:

The search for sustainable solutions, leading to improved energy efficient process, has induced new technologies to the use of cellulolytic biomass as substrate for fermentation. However, the hydrolysis of this material is followed by inhibitors formation, such as acetic acid, furfural and others, of which the first one is known to potentiate the toxic action of ethanol, with deleterious effects on fermentation. The addition of molasses in the bagasse hydrolyzate could allow fermentation with higher alcohol content, contributing to a favorable energy balance of distillation, besides providing mineral and organic nutrients for the yeast. These nutrients could allowa fermentative process with yeast cell recycle, utilizing the structure and knowledge already existing in first generation ethanoldistilleries. The cell recycle enables a rapid fermentation, but imposes repeated stress conditions to yeast cells, which makes difficult to obtain strains with the tolerance profile desired. For this reason, the goal of this project is to select strains with multiple tolerances in relation to the inhibitors present in the hydrolyzate as in molasses. Therefore, 650 strains isolated from distilleries were evaluated for growth (OD 600nm) and maximum specific growth rate (μmax), in 96-well microplates at 30°C for 24 hours, using a selective medium (pH 4.8) consisting of bagasse hydrolyzate and molasses containing 14% TRS (20% TRS derived from hydrolyzate), acetic acid (5 g/L), hydroxymethylfurfural (0.14 g/L), furfural (1.5 g/L) and absolute ethanol (4% v/v). The results showed 45 strains with the highest growth capacity and tolerance to inhibitors as compared to industrial strains of Saccharomyces cerevisiae known for their tolerance to stresses in industrial ethanol production (PE-2 and SA-1).The strains pre-selected will be subjected to fermentation trials with cell recycle, using the same medium, searching for those with ability to withstand the recycle. The best strains will be submitted to genetic modification to obtain lineages able to xylose fermentation.

Keywords: Second-generation ethanol, Selection, *Saccharomyces cerevisiae*, Lignocellulosic biomass

CNPq