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ENGINEERING YEAST TOLERANCE TO INHIBITORY LIGNOCELLULOSIC BIOMASS

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In recent years the necessity for biotechnological manufacturing based on lignocellulosic feedstocks has become evident. However, the pre-treatment step in the production of lignocellulosic bioethanol leads to the accumulation of inhibitory byproducts. Robust second generation bioethanol processes require microorganisms able to ferment these inhibitory lignocellulosic hydrolysates. Significant progress has been made in the understanding of the determinants of yeast tolerance to lignocellulose biomass-derived inhibitors, however further knowledge at the genetic level is of essential importance for the improvement of lignocellulose conversion technology. Based on genome-wide results previously obtained [1], two key genes, *PRS3* and *RPB4*, were found to contribute to the maintenance of cell viability in wheat straw hydrolysate and to the maximal fermentation rate of this substrate. Here we describe the outcome, in bioethanol productivity, of fermentations in *Eucalyptus globulus* wood hydrolysate, using recombinant *Saccharomyces cerevisiae* BY4741 overexpressing these genes. Furthermore, we studied their expression in an industrial strain isolated from a Brazilian bioethanol production plant, which was previously demonstrated to have very robust characteristics with outstanding fermentation performances [2]. This expression evaluation was performed during a fermentation mimicking industrial conditions, under the absence and presence of inhibitory compounds, and provides insights into the roles of *PRS3* and *RPB4* in the adaptation to toxic biomass hydrolysates. This study expands our understanding of the underlying molecular mechanisms involved in yeast response to the multiple stresses occurring during lignocellulose fermentations under industrially relevant conditions.

[1] Pereira FB et al. *Identification of candidate genes for yeast engineering to improve bioethanol production in very high gravity and lignocellulosic biomass industrial fermentations.* Biotechnol Biofuels 2011. 4(1):57.
[2] Pereira FB et al. *Robust industrial Saccharomyces cerevisiae strains for very high gravity bio-ethanol fermentations.* J Biosci Bioeng 2011. 112(2):130-6.

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