CENTRE OF BIOLOGICAL ENGINEERING
UNIVERSIDADE DO MINHO

INSTITUTE FOR BIOTECHNOLOGY AND BIOENGINEERING

Identification of genes and process conditions required to improve alcoholic fermentation yield under industrially relevant fermentation media

Pedro M.R. Guimarães[1], Francisco B. Pereira[1], Daniel G. Gomes[1], Nuno P. Mira[2], Miguel C. Teixeira[2], Margarida Palma[2], Artur B. Lourenço[2], José A. Teixeira[1], Isabel Sá-Correia[2], Lucília Domingues[1]

[1]*IBB-Institute for Biotechnology and Bioengineering, Centre of Biological Engineering, Universidade do Minho, Campus de Gualtar, 4710-057 Braga, Portugal*
[2]*IBB-Institute for Biotechnology and Bioengineering, Centre for Biological and Chemical Engineering, Instituto Superior Técnico, Av. Rovisco Pais, 1049-001 Lisboa, Portugal*

The successful performance of alcoholic fermentations relies on the ability of *Saccharomyces cerevisiae* strains to cope with multiple stress factors occurring during the fermentation process and on the selection of suitable process engineering strategies [1].

An optimized very high gravity (VHG) glucose medium [2] containing low cost nutrients was developed for stress-tolerant and high fermentation efficiency yeasts selection from “cachaça” and bio-ethanol production plants in Brazil [3]. CA1185 and PE-2 strains, which produce remarkably high ethanol titres (>19%, v/v), are physiologically more prepared to cope with VHG stresses relatively to laboratory strains. Recent results show that this robustness is, most likely, related to specific features of yeast cell wall and plasma membrane. Considering the outstanding properties of PE-2 strain, this yeast strain was genetically modified to introduce flocculation and used in repeated batch fermentations with high cell density resulting in significant improvements on process economics.

A genome-wide screening for determinants of yeast resistance to high sugar [4] and ethanol [5] stresses, relevant in VHG technology, and to inhibitory fermentation compounds from lignocellulosic pre-treatments, including acetic acid [6], was also carried out. Eighteen genes were identified as determinants of yeast resistance to more than three of the above mentioned fermentation relevant stresses. Among these genes, 8 were found to significantly affect ethanol kinetics and/or production in VHG and hemicellulosic liquor fermentations mimicking industrially relevant conditions. Guided by all the gathered information extraordinarily robust strains are being designed to be used in efficient and economic industrial alcoholic fermentation processes.

[1] Mussato et al, *Biotechnol Adv*. (2010) **28**: 817-830; [2] Pereira et al, *Bioresource Technol.* (2010) **101**:7856-7863; [3]Pereira et al., *Biotechnol. Lett.* (2010) DOI 10.1007/s10529-010-0330-9; [4] Teixeira et al, *OMICS* (2010) **14**: 201-210; [5] Teixeira et al, *Appl. Environ. Microbiol.* (2009) **75**:5761-5772; [6] Mira et al, *OMICS* (in press).