GOUVEIA e SILVA, C. [2]; BOSCOLO, M. [1]; Da SILVA, R. [1]; LOUREIRO DIAS, M. C. [2]; GOMES, E.[1]; PRISTA, C. [2]; SILVA, R. R. [1]

1. Instituto de Biociências, Letras e Ciências Exatas, IBILCE/UNESP, São José do Rio Preto/SP, Brazil; 2. LEAF, Instituto Superior de Agronomia, Universidade de Lisboa, Tapada da Ajuda 1349-017 Lisboa, Portugal

PROSPECTING PENTOSE SYMPORTERS FROM *Rhodotorula* sp.

Many efforts have been made to hydrolyse the hemicellulosic material and to add value to biorefineries through the production of additional compound *e.g.* organic acids. To date, the fermentative yeast *Saccharomyces cerevisiae*, even modified for the assimilation of pentoses, has not presented satisfactory yield for ethanol production. Consequently, the search for pentose transporters from non-*Saccharomyces* yeasts for heterologous expression in *S. cerevisiae* has been recurrent. Thus, we investigate pentose transporters from a *Rhodotorula* sp. strain isolated from fruit samples. The strain was identified using sequences of the D1/D2 domains of the rDNA, maintained in 15% glycerol and stored at −80 °C. Posteriorly, the *Rhodotorula* sp. **s**train was cultivated in D-xylose (YPX 2% D-xylose) or D-glucose (YPD 2% D-glucose) as sole sugar source, initial pH 4.5 and 6.5. The consumption of D-xylose and D-glucose during fermentation was monitored, and the presence of H^+ symporters for L-arabinose, D-xylose, and D-glucose was evaluated at different time-points during growth: 24 and 48 h. For transport kinetics, the influx of protons/sugar was calculated based on the rate of extracellular alkalinization after the addition of a pulse at different concentrations of sugar to a concentrated and unbuffered suspension of cells. According to the results, kinetic characterization of pentose transport revealed that the highest affinity D-xylose symport was detected in cell cultivated in YPX media, pH 4.5 with K_M = 15.7 mM and V_{Max} = 0.34 mMol.h^{-1}.g^{-1} (24 h), and K_M = 7.7 mM and V_{Max} = 0.22 mMol.h^{-1}.g^{-1} (48 h). As for L-arabinose, the highest affinity for H+/arabinose symporter was observed at 48 h with K_M = 862 mM and V_{Max} = 1.28 mMol.h^{-1}.g^{-1}. These transporters can be potentially used to overcome transport limitations imposed on the yields of metabolic pentose pathways and open an avenue for enabling *S. cerevisiae* and other yeasts for the better use of plant biomass.

Keywords: Fermentation, pentose, symporter, yeast

Development Agency: This study was supported by FCT (Portugal): research unit UID/AGR/04129/2013 (LEAF) and FAPESP: 2017/06399-3 and 2018/09238-3.