## Application of the yeast Yarrowia lipolytica for food additives production

Isabel Belo<sup>b,1</sup>, Patrícia Ferreira<sup>1</sup>, Adelaide Braga<sup>1</sup>, Marlene Lopes<sup>1</sup>

<sup>1</sup>CEB - Centre of Biological Engineering, University of Minho, Campus de Gualtar, 4710–057 Braga, Portugal

# <sup>b</sup>:Corresponding author: E-mail:ibelo@deb.uminho.pt, Tel: +351 253 604 413

#### **Introduction:**

Yarrowia lipolytica is a nonconventional, aerobic and dimorphic yeast with many biotechnological applications due to the wide range of substrates that can use as carbon source and the ability to produce a large variety of metabolites with industrial interest. It can usually be found in environments containing hydrophobic substrates, such as alkanes and fats. It can also be isolated from cheeses, yoghurts, kefir, soy sauce, meat and shrimp salads. Y. lipolytica has been proved to be a robust cell for the biotechnological production of compounds that can be used as additives in food industry, such as organic acids (citric acid), enzymes (such as proteases and lipases), biosurfactants, sweeteners (such as erythritol and mannitol), and aroma and fragrances compounds.

#### **Materials and Methods**

Most of the work performed to optimize biotechnological processes with Y. lipolytica was carried with the strain W29 (ATCC 20460). Stirred tank bioreactors were used for batch and fed-batch cultures, as well as airlift type bioreactors. Cells were grown on different culture medium containing low-cost and renewable substrates, such as crude glycerol, waste cooking oils, castor oil, among others. Besides cellular growth and morphology characterization, intracellular compounds accumulation and extracellular metabolites production were monitored. Bioprocesses were optimized to maximize the productivity of the target products.

### **Results and Discussion:**

Lipase production (12000 U·L<sup>-1</sup>) and lipid-rich biomass (48 % of lipids mass per dry cellular mass) enriched in unsaturated fatty acids (oleic and linoleic acids) was obtained with Y. lipolytica W29 from waste frying oils based medium. Biotransformation of castor oil into lactones (fruity aromatic compounds) have been extensivly studied either by comparing the reactors type and operation strategies, either using genetic modified strains derived from W29 strain. Results lead to an improvement in  $\Box$ -decalactone (peach-like aroma) accumulation in the medium (up to 7  $g \cdot L^{-1}$ ) reducing the side products formation.

Using crude glycerol, a by-product of biodiesel industry, citric acid, erytritol and microbial lipids were produced. Besides those substrates, also volatile fatty acids, such as acetic, propionic and butiric acid were sucessfully converted to lipids by Y. lipolytica. Oxygen transfer rate to the aerobic cultures of the yeast has been a major factor of process optimization strategies.

### **Conclusion:**

*Y. lipolytica* W29 has been proven to be a workhorse cell for the development of biobased industry, mainly for the improvement of the sustainability of industrial biotechnology for the production of food additives. Its GRAS status, ability to use wastes and subproducts as feedstocks, and the high titers of interesting compounds obtained by *Y. lipolytica*, turns this yeast into an emerging microrganism for the implementation of circular economy on Food industry.

#### Acknowledgements:

We would like to thank the Portuguese Foundation for Science and Technology (FCT) under the scope of the strategic funding of UID/BIO/04469 unit and through the bilateral cooperation project FCT/NKFIH 2017/2018, COMPETE 2020 (POCI-01-0145-FEDER-006684) and BiotecNorte operation (NORTE-01-0145-FEDER-000004) funded by the European Regional Development Fund under the scope of Norte2020 - Programa Operacional Regional do Norte.