PHYSIOLOGY AND INDUSTRIAL POTENTIAL OF NEW SACCHAROMYCES CEREVISIAE STRAINS ISOLATED FROM THE BRAZILIAN BIODIVERSITY

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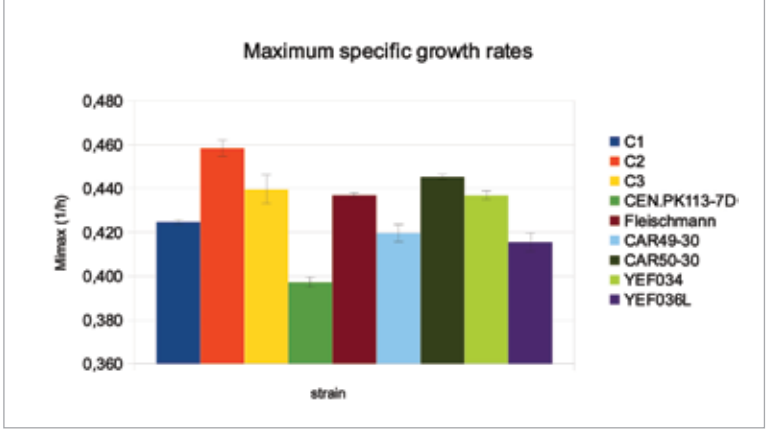

Figure 1. Maximum specific growth rate values of different Saccharomyces cerevisiae *strains, grown on a defined mineral medium (Verduyn et al., 1992), supplemented with vitamins and glucose as the sole carbon source. CEN.PK113-7D is a standard laboratory strain, widely employed by the scientific community for physiological studies (Van Dijken et al., 2000). Fleischmann is a commercial baker's yeast strain. The remaining 7 strains are new isolates, obtained from Atlantic forest environments. The error bars correspond the the deviation of the mean (experiments were performed in duplicate).*

In spite of being the most widely utilized organism in the biotech industry and also the best studied eukaryote, the yeast *Saccharomyces cerevisiae* does not have its natural habitat well characterized. Indigenous strains of this species will be isolated from Atlantic Forest ecosystems by the group of Dr. Carlos Rosa, from the Federal University of Minas Gerais, who will also perform a phylogenetic analysis of these strains. Depending on these results and on some simple fermentation trials, some strains will be chosen to be employed in our project. A detailed and quantitative physiological characterization of these strains will be performed, with the objective of comparing them to well described laboratorial and industrial *S. cerevisiae* strains. For this purpose, the strains will be cultivated both under full aerobiosis and under complete anaerobiosis, during exponential unlimited growth and also during the steady-state of chemostats operated at a 0.1 1/h dilution rate, always using defined media. Finally, the newly isolated strains will have their industrial potential evaluated, which will be carried out using industrial media (sugarcane juice and/or molasses), under conditions that mimic the industrial processes. It is expected that this project will lead to the identification of yeast strains with physiological properties that are different from the strains currently in use and also strains with high industrial potential. The new strains might also be an interesting source for new genes of biotechnological relevance.

SUMMARY OF RESULTS TO DATE AND PERSPECTIVES

The project started recently. For the moment, we have data on the specific growth rates of 7 new *Saccharomyces cerevisiae* isolates, grown on a defined mineral medium, suplemented with vitamins and glucose as the sole carbon source (*Figure 1*).

As may be seen from *Figure 1*, some of the newly isolated strains, e.g. C2, present a higher specific growth rate than not only a laboratorial strain, but also than an industrial strain, when grown on a synthetic medium. This is a quite unexpected result, since C2 went through a small number of duplications in the laboratory and thus has not been adapted or evolved for growth in laboratorial conditions, such as on a synthetic medium. Evolutionary engineering can be further applied to increase the growth rate of this strain, in order to verify to which extent this key physiological parameter can be improved, e.g. by applying the pH-auxostat (Groeneveld et al., 2009).

Another group of 7 isolates will be evaluated, not only on a defined mineral medium, but also in complex media. Furthermore, physiological parameters will be correlated with the isolation niche (e.g. tree species) and phylogenetic data on other known *S. cerevisiae* laboratorial and industrial strains.

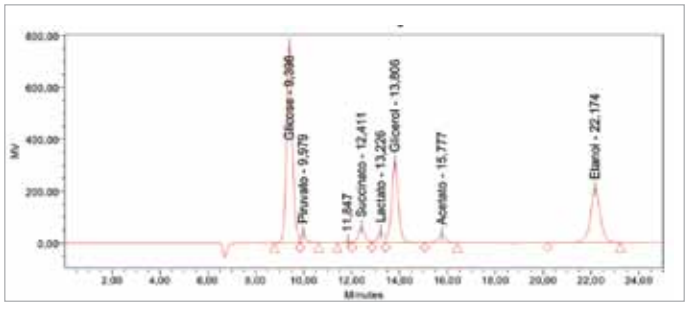

Figure 2. HPLC detection of the main metabolites consumed and produced by the yeast Saccharomyces cerevisiae *during cultivation on a defined mineral medium (Verduyn et al., 1992) supplemented with vitamins and glucose as the sole carbon source*

MAIN PUBLICATIONS

This is the first year of the project and no publication was produced so far. However, a list of recent publications from our group related to sugarcane genomics is shown.

Basso TO, de Kok S, Dario M, do Espirito-Santo JCA, Müller G, Schlölg PS, Silva CP, Tonso A, Daran JM, Gombert AK, van Maris AJA, Pronk JT, Stambuk BU. 2011. Engineering topology and kinetics of sucrose metabolism in *Saccharomyces cerevisiae* for improved ethanol yield. *Metabolic Engineering* (*Print*) **13**: 694-703.

Rocha SN, Abrahão-Neto J, Cerdán ME, Gombert AK, González-Siso MI. 2011. Heterologous expression of a thermophilic esterase in Kluyveromyces yeasts. *Applied Microbiology and Biotechnology*. **89**: 375-385.

Gombert AK, Veiga T, Puig-Martinez M, Lamboo F, Nijland JG, Driessen AJM, Pronk JT, Daran JM. 2011. Functional characterization of the oxaloacetase encoding gene and elimination of oxalate formation in the beta-lactam producer *Penicillium chrysogenum*. *Fungal Genetics and Biology (Print)*. **48**: 831-839.

Rocha SN, Abrahão-Neto J, Gombert AK. 2011. Physiological diversity within the *Kluyveromyces marxianus* species. *Antonie van Leeuwenhoek*. **100**: 619-630.

Basso TO, Dario MG, Tonso A, Stambuk BU, Gombert AK. 2010. Insufficient uracil supply in fully aerobic chemostat cultures of *Saccharomyces cerevisiae* leads to respiro-fermentative metabolism and double nutrient-limitation. *Biotechnology Letters*. **33**: 973-977.

Rocha SN, Abrahão-Neto J, González-Siso MI, Cerdán ME, Gombert AK. 2010. Heterologous expression of glucose oxidase in the yeast *Kluyveromyces marxianus*. *Microbial Cell Factories*. **9**: 4.

Fonseca GG, Heinzle E, Wittmann C, Gombert AK. 2008. The yeast *Kluyveromyces marxianus* and its biotechnological potential. *Applied Microbiology and Biotechnology*. **79**: 339-354

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