Classification of *Saccharomyces cerevisiae* morphology employing image analysis

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Population dynamics of microbial systems can be described by several approaches and in various levels of complexity, each of them arising from specific goals and limitations. From the process-engineering viewpoint there is a need for a comprehensive mathematical model describing population dynamics in terms of measurable entities (microbes) and chemicals involved (limiting substrate, dissolved oxygen, etc.), as well as process configuration (number and type of reactors, interconnections, etc.) and process parameters (inlet flow rate and composition, reactor holdup, and more)^[1].

The description of intricate population dynamics and the inference of cell states lead to complex models with a great number of parameters. Knowledge about whole cell cycle and morphology classification is imperative, since a considerable difference exists between the cell description employed in model formulation and the laboratory reality. As soon as in biological systems exists a relationship between cell morphology and productivity, some authors drive efforts towards the on-line measurement of biomass component to avoid process delays ^{[2],[4]} or to determine cellular characteristics related to its morphology and/or physiology through image processing analysis ^{[5],[6],[7]}.

Saccharomyces cerevisiae size and shape distribution are affected by growth rate, mutation, and environmental conditions (composition, temperature, pressure, presence of oxidant agents, etc.). Although its shape usually assumes an ellipsoid contour it is modified along the cell cycle by bud formation and growing attached to the mother ^[5].

This work deals with *S. cerevisiae* classification based on morphology analysis. Image acquisition was conducted in an optical microscope (x 400 magnification) coupled with a black and white camera and linked to a microcomputer by a frame grabber. Traditional tools generally used for image enhancing were employed. Feature extraction and objects separation were necessary to classify "mothers" and "daughters" and to determine its frequency in the analyzed samples.

Cells were automatically divided in five different classes with respect to bud size compared to the respective mother through image analysis employing Matlab (v.6.1, The Mathworks Inc.). This methodology was validated with distinct samples and employed along *Sacharomyces cerevisiae* growth in different operational conditions. The data herein obtained is being used for morphological structured model formulation.

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[1] Duarte, M.V.E., (2000). M.Sc. Thesis, School of Chemistry, Federal University of Rio de Janeiro, Brazil (in portuguese).

[2] Yamashita, Y. et al. (1993). Journal of Chemical Engineering of Japan, 26(6), 615-619.

[3] Zalewski, K.and Buchholz, R. (1996). Journal of Biotechnolgy, 48, 43-49.

[4] Bittner, C. et al. (1998). Biotechnology and Bioengineering, 60(1), 24-35.

[5] Pons, M.N. et al. (1993). Biotechnology and Bioengineering, 42(11), 1352-1359.

[6] Vicente, A. et al. (1996). Biotechnology and Bioengineering, 51(6), 673-678.

[7] Mas, S. and Ghommidh, C. (2001). Biotechnology and Bioengineering, 76(2), 91-98.

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