

Towards Optimization of Squalene formation by *Saccharomyces cerevisiae*: effect of Inoculum Size and Fermentation Time

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Current interest in potential chemopreventive activity of squalene (C₃₀H₅₀), though non conclusive, dressed this unsaturated hydrocarbon with the veil of a functional compound and attracted the interest in new methods of its production (1). Taking into account food safety requirements for products of biotechnology, our interest was focused on *Saccharomyces cerevisiae*. Optimizing the industrial performance of *S. cerevisiae* in terms of squalene production aims at establishing conditions that favor both active growth and a relatively high squalene content in yeast. Semianaerobic conditions were found to be a suitable aeration strategy for squalene synthesis at the expense of ergosterol and related intermediates (2).

The present work aimed at directing, under the above aeration conditions, the inoculum size and fermentation time to achieve high squalene yields. A central composite statistical design was used to select the experimental conditions under which the effect of inoculum size (0.75-9.24%, v/v) and fermentation time (5-52h) on squalene yield (µg/L of culture medium), productivity (µg/Lh), and biomass (g/L of culture medium) were investigated for two commonly used *S. cerevisiae* laboratory wild type strains (BY4741 and EGY48). RP-HPLC was employed to monitor changes in the squalene yield and intermediates formation. Results allowed also assessment of factor interactions. Optimization of the bioprocess indicated that maximum squalene yield and productivity were, respectively, 2967.6±118.7 µg/L of culture medium and 104±4.2 µg/Ld for BY4741 (5%, v/v) and 3129±109.5 µg/L of culture medium and 155.9±5.5 µg/Ld for EGY48 (8.1%, v/v). Experimental verification showed that the optimized bioprocess is competitive with regard to those reported in the literature.

Reference: 1) Bhattacharjee, P., Shukla, V. B., Singhal, R. S., and Kulkarni, P. R. *World Journal of Microbiology* **2001**, 17, 811-816, 2) Mantzouridou, F.; Naziri, E.; Tsimidou, M., *J. Agric. Food Chem.* **2009**, DOI:10.1021/jf900673n