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Research Article

Carbon concentration and oxygen availability affect lipid and carotenoid production by carob pulp syrup-grown *Rhodospiridium toruloides* NCYC 921

The simultaneous effect of oxygen availability and carbon source concentration on yeast lipid and carotenoid production has never been studied before. In this work, a Doehlert distribution design was used to study the simultaneous effect of carbon concentration and oxygen availability on *Rhodospiridium toruloides* NCYC 921 carotenoid and lipid production. A cheap industrial byproduct was used as carbon source (carob pulp syrup). A total sugar concentration of 106.3 g/L and a medium volume of 0.120 L induced the highest total carotenoid and total fatty acid productivities (4.60 $\mu\text{g/Lh}$ and 0.029 g/Lh, respectively). Flow cytometry was used to assess yeast stress response under different cultivation conditions. The highest proportion of cells with permeabilised membrane (>20%) was induced when the cultivations were carried out at the highest sugar concentration studied (130.0 g/L) or when the culture reached the minimum final medium pH (4.60). The results showed that the total sugar concentration had a positive influence on the yeast biomass and carotenoid content, while the oxygen availability had little influence on the biomass concentration, but had a slight positive influence on the carotenoid content. Regarding the fatty acids, the two factors had a negative impact on the synthesis of these compounds.

Keywords: Carbon concentration / Carotenoids / Lipids / Oxygen availability / Yeast



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Received: January 8, 2015; revised: March 25, 2015; accepted: May 27, 2015

DOI: 10.1002/elsc.201500002

1 Introduction

In the midst of the energy crisis, third generation biofuels (derived from microalgae and other microbes) are considered to be viable fuel alternatives [1, 2]. However, to be economically sustainable, the microbial biofuel production process must valorize the whole biomass fractions to be converted into biofuels and high value added products under the frame of an integrated biorefinery concept.

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Abbreviations: DiOC₆(3), 3,3-dihexylocarbocyanine iodide; MUFAs, monounsaturated fatty acids; PI, propidium iodide; PUFAs, polyunsaturated fatty acids; SATs, saturated fatty acids; TFAs, total fatty acids

The oleaginous yeast *Rhodospiridium toruloides* NCYC 921 (which species is an anamorph of *Rhodotorula glutinis* species) has been widely reported as a potential oil producer yeast [3, 4]. However, the yeast biomass, beyond its high lipid content (that can be converted into biodiesel), is rich in high value added products such carotenoids with commercial interest [5] their commercialization may contribute to reduce the overall process cost of biofuels and carotenoid production [6].

The use of low-cost substrates in media formulations may also reduce the costs of the microbial biofuels production [7]. Carob pulp syrup has been used in media formulations for microbial lipid production [8]. The carob tree (*Ceratonia siliqua* L.) is native to the Mediterranean region, including southern European countries. Carob seeds comprise 10% of the fruit dry weight and are used for gum production for food industry. The remaining 90% of the fruit dry weight (the pulp) contains high content of sugar (sucrose, glucose and fructose). The easily