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BIOENERGY AND BIOFUELS

Carotenoid and lipid production by the autotrophic microalga *Chlorella protothecoides* under nutritional, salinity, and luminosity stress conditions

L. Campenni' · B. P. Nobre · C. A. Santos · A. C. Oliveira · M. R. Aires-Barros · A. M. F. Palavra · L. Gouveia

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Abstract Today microalgae represent a viable alternative source for high-value products. The specie *Chlorella protothecoides* (*Cp*), heterotrophically grown, has been widely studied and provides a high amount of lutein and fatty acids (FA) and has a good profile for biodiesel production. This work studies carotenoid and FA production by autotrophic grown *Cp*. *Cp* was grown until the medium's nitrogen was depleted, then diluted in NaCl solution, resulting in nutritional, luminosity, and salinity stresses. Different NaCl concentrations were tested (10, 20, 30 g/L) at two different dilutions. After dilution, a color shifting from green to orange-red was noticed, showing carotenoid production. The best production of both carotenoids and FA was attained with a 20 g/L NaCl solution. The total carotenoid content was 0.8 % *w/w* (canthaxanthin (23.3 %), echinenone (14.7 %), free astaxanthin (7.1 %), and lutein/zeaxanthin (4.1 %)). Furthermore, the total lipid content reached 43.4 % *w/w*, with a FA composition of C18:1 (33.64 %), C16:0 (23.30 %), C18:2 (11.53 %), and less than 12 % of C18:3, which is needed to fulfill the biodiesel quality specifications (EN 14214).

L. Campenni' · B. P. Nobre · C. A. Santos · A. C. Oliveira · L. Gouveia (✉)
LNEG–Laboratório Nacional de Energia e Geologia, Unidade de Bioenergia, Estrada do Paço do Lumiar, 1649-038 Lisbon, Portugal
e-mail: luisa.gouveia@lneg.pt

L. Campenni' · B. P. Nobre · A. M. F. Palavra
IST, Centro de Química Estrutural, DEBQ, Av. Rovisco Pais, 1, 1049-001 Lisbon, Portugal

M. R. Aires-Barros
Department of Bioengineering, Institute for Biotechnology and Bioengineering, Centre for Biological and Chemical Engineering, Instituto Superior Técnico, Av. Rovisco Pais, 1, 1049-001 Lisbon, Portugal

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Introduction

Since the industrial revolution, the infrastructure of our society has relied strongly on petroleum-based products for fuels, materials, and chemicals.

However, the increased consumption of energy leads to a massive influx of carbon into the atmosphere mediated through the burning and consumption of petroleum-based products. Renewable biofuels (e.g., ethanol, butanol, hydrogen, methane, biodiesel) are required to help replace petroleum dependence (Valenzuela et al. 2012).

Microalgae are among the most potentially significant source of sustainable biofuels in the future of renewable energy (Menetrez 2012). A feedstock with unlimited applicability, microalgae can metabolize several waste streams (liquids and gaseous) and produce products with a wide variety of compositions and uses (Menetrez 2012). The high potential of microalgae is due to its high photosynthetic efficiency, high growth rate and consumption of CO_2 with the concomitant production of O_2 and no need of arable land or potable water (Gouveia 2011).

In addition to photoautotrophic growth (i.e., carbon fixation via sunlight), some green algae and diatoms can store carbon and energy in the form of lipids (i.e., triacylglycerides (TAGs)), and this fact has re-invigorated the possibilities of algal oil being used for the production of liquid fuels. Nutrient deficiency or nutrient stress has been well documented to increase TAG accumulation in microalgae (Sheehan et al. 1998) specifically, nitrogen or phosphate limitation (Hu et al. 2008; Rodolfi et al. 2009; Li et al. 2008; Yeesang and Cheirsilp 2011). Lipid productivity is a