Evaluation of the simultaneous production of lutein and lipids using a vertical alveolar panel bioreactor for three *Chlorella* species

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**A B S T R A C T**

The concept of a biorefinery improves the economic efficiency of a biofuel production process from microalgae by recovering high value added compounds. Lutein is a carotenoid currently extracted from petals of *Tagetes erecta* with an established market in poultry and in human nutritional supplements. For the very first time, an extended study on the lipid and lutein production over three *Chlorella* species as well as cell disruption methods was performed. *Chlorella vulgaris*, *Chlorella zofingiensis* and *Chlorella protothecoides* were grown in an indoor vertical alveolar panel photobioreactor with continuous illumination, and two cell disruption methods were assessed at a laboratory scale: glass bead vortexing and ball mill grinding. For *C. vulgaris*, *C. zofingiensis* and *C. protothecoides* the intracellular lutein content was measured as: 3.86, 4.38 and 3.59 mg L⁻¹ respectively. Lipid contents vary slightly among microalgae with a value close to 9% w/w. Biomass and lutein productivities were found to be higher for *C. vulgaris* (0.131 g L⁻¹ d⁻¹, 0.51 mg L⁻¹ d⁻¹) and for *C. zofingiensis* (0.122 g L⁻¹ d⁻¹, 0.53 mg L⁻¹ d⁻¹) compared to *C. protothecoides* (0.103 g L⁻¹ d⁻¹, 0.37 mg L⁻¹ d⁻¹). *C. vulgaris* 1803 and *C. zofingiensis* B 32 were found to be promising organisms for simultaneous production of lutein and lipids. Although all the microalgae under study belong to the same genus, a species-specific response was observed for each of the cell grinding methods tested.

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1. Introduction

In the last decade, biofuel production from microalgae has been intensively investigated. This is evident by the rapidly increasing number of research articles published and patents issued [1]. A number of advantages of microalgae over conventional oilseed crops explain this research effort: fresh water consumption can be reduced using wastewater or seawater microalgae; its production does not require agricultural land and a high biomass and oil productivity per acre can be attained [2,3]. A theoretical ceiling of 94 to 155 m³ oil ha⁻¹ yr⁻¹ was calculated for microalgae assuming oil contents of 25% or 50% respectively [4] and a 10% photosynthetic efficiency [5]. Although these figures represent a theoretical value, when a realistic lipid productivity is considered (cf. supplementary material in ref. [4]), oil productivity of microalgae culture is still 4.65 and 23.4 times higher when compared to palm or sunflower oil productivities [3]. However, using realistic productivities, biofuel production can only be economically feasible when high value products are concomitantly produced [4]. A biorefinery approach in which both fuels and multiple value-added compounds are produced, or even microalgae production is synergistically coupled with carbon sequestration and wastewater treatment [6], might support the development of the microalgae energy industry. The potential for valuable co-products in algae processing has been cited as one of the key reasons for exploring this source of biofuels [7]. Besides polar and non-polar lipids, microalgae produce pigments and sterols with established market values [1,8]. Among the colored compounds, carotenoids are lipid-soluble molecules that play essential roles in photosynthesis. They contribute to light harvesting, scavenge reactive oxygen species and dissipate excess energy [9]. Lutein, a carotenoid found in flowers, food and human serum [10] has also been reported to be available in microalgae [11], has an established market in poultry and human nutritional supplements with a world market valued at 233 million USD in 2010 [12]. Along with zeaxanthin, lutein is present in the macula lutea, a small area in the retina [13] and in the crystalline lens [14]. Oral intake of lutein has been linked to a reduced risk of diseases such as age related macular degeneration [13,15], cataracts [16–18] and retinal degeneration [19].

Currently, the lutein production process involves the extraction of this compound from dried *Tagetes* sp. petals with organic solvents, saponification of the extract to remove waxes and fatty acids, and finally crystallization [20]. The product contains lutein as the major component and a smaller proportion of zeaxanthin. Lutein production from petals is

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