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Efficient use of bicarbonate for mass production and carbon isotopic labelling of the green alga *Ulva ohnoi* under natural conditions

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Abstract

Mass aquaculture production of the green seaweed *Ulva* is advancing under the stimulus of many interesting applications. Increasing the efficiency of carbon use in *Ulva* mass cultures is important to improve its production and diversify biomass applications. Given the high capacity of *Ulva* species to use bicarbonate as a C source, mass cultures of *Ulva ohnoi* carried out under natural conditions and controlled pH were stoichiometrically supplied with sodium bicarbonate on demand. The specific growth rate (average 0.108 ± 0.008 day^{-1}) and the biochemical composition of *U. ohnoi* remained stable over three consecutive production cycles, in which C conversion from added bicarbonate into net produced biomass ranged between 88.5 ± 4.2 and $95.9 \pm 0.9\%$. C conversion was significantly higher ($p < 0.05$) than conversion of nitrogen (73.4 to 87.4%) and phosphorus (68.7 to 80.2%). By enriching the total bicarbonate at 4% with $H^{13}CO_3^-$, the algal isotopic signature $\delta^{13}C$ increased from -7.5 ± 0.38 to 1505 ± 56‰ after 8 days of growth. Uptake of ^{13}C was steady throughout the culturing period and resulted in biomass production estimates very similar to those calculated from the growth rate. The use of bicarbonate in *Ulva* cultures represents an important improvement in C conversion efficiency. It is also an easy alternative to produce biomass sufficiently enriched in ^{13}C, which is demanded in studies on the nutritional value of algae. Reliable production estimates in mass cultures can be expected from the ^{13}C uptake rate in algae with high bicarbonate utilization capacity.

Keywords Bicarbonate · Carbon stable isotope · Carbon use efficiency · Chlorophyceae · Mass culture · *Ulva*

Introduction

Seaweeds represent over one half of total marine and coastal aquaculture production with eight red and brown species cultured in Asia accounting for 96.5% of world seaweed production (Chopin and Tacon 2021). Although abundant in nature and frequently involved in coastal eutrophication issues (Smetacek and Zingone 2013), green macroalgae have contributed very little to global seaweed aquaculture. This situation occurs even despite the high growth capacity and resistance to external conditions of green algae (Taylor et al. 2001). *Ulva* is the most studied green macroalga, but knowledge acquired over decades about *Ulva* ecophysiology, reproduction and culture management has not yet been reflected in the development of the range of applications that have been claimed for this macroalga (Dominguez and Loret 2019). The natural tendency of *Ulva* to bloom in nutrient-enriched coastal waters and generate nuisance green tides could be exploited as a new resource (Smetacek and Zingone 2013; Dominguez and Loret 2019). However, more stable biomass production is required for many commercial applications and fluctuations due to the environment have thus to be minimized. This is of particular importance in nutritional applications and justifies why tank land cultivation of *Ulva* has been extensively studied (Tsubaki et al. 2017; Revilla-Lovano et al. 2021). Land-based seaweed production allows precise culture control, depends less on regulatory issues than extractive processes and competition with other near-shore uses is diminished. Initial land-based seaweed aquaculture was later diversified with the use of macroalgae production for nutrient recycling in integrated multi-trophic aquaculture (IMTA) systems (Neori 2008; de Paula Silva et al. 2008). *Ulva* biomass from different aquaculture systems has been successfully tested as a complementary component in aquaculture feeds (Shpigel et al. 2017; Moutinho et al. 2018) and economic issues are key in deciding whether a widespread nutritional application is developed.

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