Bioresource Technology 128 (2013) 118–124

Contents lists available at SciVerse ScienceDirect

Bioresource Technology

journal homepage: www.elsevier.com/locate/biortech

Effect of various stress-regulatory factors on biomass and lipid production in microalga *Haematococcus pluvialis*

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HIGHLIGHTS

- Specific growth conditions for biomass yield in *Haematococcus pluvialis.*
- Specific growth conditions for biodiesel feedstock lipids.
- Specific growth conditions for UFA-rich lipids for multiple use.
- Refined biomass proposed for biofuel feedstock.

ARTICLE INFO

Article history:
Received 5 September 2012
Received in revised form 10 October 2012
Accepted 12 October 2012
Available online 23 October 2012

Keywords:
Haematococcus pluvialis
Stress conditions
Biomass
Feedstock lipids
UFA-rich lipids

ABSTRACT

To maximize the biomass and lipid production for applications in food or biofuel feedstock, nine stress conditions were tested considering N and/or P limitations, light intensity & quality, for *Haematococcus pluvialis* SCCAP K-0084 cultivation. Photosynthetically active radiation (PAR), warm white light emitting diode (WWLED), and white light emitting diode (WLED) at illumination of 240 μmol photons m^{-2} sec^{-1} were the best stress-regulatory factors. PAR without P & low N conditions yielded high biomass with 33% lipids containing increased C16:0 and C18:0 saturated fatty acids, and reduced unsaturated fatty acids (UFAs) (oleic, linoleic, and α/γ-linolenic). WWLED and WLED without P conditions also yielded high biomass, but 25% lipids with increased amounts of UFAs. Red light emitting diode (RLED) without P & low N conditions yielded 46% lipids with lowest biomass. PAR and WWLED & WLED illuminated conditions were found suitable respectively for biodiesel feedstock lipids and UFA-rich lipids for multiple applications.

1. Introduction

Haematococcus pluvialis, a freshwater green microalga has been proven to be a significant natural source of astaxanthin, a carotenoid with high antioxidant capacity and has long been used as food coloring agent in aquaculture and poultry (Lorenz and Cysewski, 2000). The microalga grows as motile bi-flagellated green cells under favorable growth conditions, but during extreme environmental conditions, such as nutrient limitation, increased light intensity or high salt concentrations, these cells undergo morphological and biochemical changes as their survival strategies. They form red cysts by losing their motility, increasing cell size, thickening cell walls, increasing lipids biosynthesis and carotenogenesis, particularly accumulation of orange-red pigment astaxanthin esters that may constitute up to 95% of the total carotenoids (Lee and Zhang, 1999). Generally, during most of the stress conditions microalgae accumulate neutral lipids, and particularly in *H. pluvialis*, the accumulation of neutral lipids and astaxanthin esters are simultaneous processes (Boussiba, 2000; Thompson, 1996). The increased neutral lipids under unfavorable conditions were hypothesized to serve as a matrix for solubilizing astaxanthin esters (Boussiba, 2000). This hypothesis was further supported by increased production of oleic acid (C18:1n-9 *cis*) enriched lipids that is essential for astaxanthin accumulation (Zhekisheva et al., 2002). It was reported that the neutral lipids of either nitrogen starved or high light stressed cells possess enhanced palmitic (C16:0) and linoleic (C18:2n-6 *cis*) acids content (Zhekisheva et al., 2002). Therefore,

Abbreviations: BLED, blue LED; DMSO, dimethyl sulfoxide; DW, dry weight; FAs, fatty acids; FFAs, free fatty acids; GLED, green LED; LED, light emitting diode; PAR, photosynthetically active radiation; PUFAs, polyunsaturated fatty acids; RLED, red LED; SFAs, saturated fatty acids; UFAs, unsaturated fatty acids; WLED, White LED; WWLED, Warm white LED.

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http://dx.doi.org/10.1016/j.biortech.2012.10.049