Algal Research 35 (2018) 76-84

Contents lists available at ScienceDirect

**Algal Research** 



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

# Conversion of biowaste leachate to valuable biomass and lipids in mixed cultures of Euglena gracilis and chlorophytes



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### ARTICLE INFO

Keywords: LC-PUFA EPA DHA Nutrient Bacteria Overvielding

# ABSTRACT

Microalgae are a sustainable alternative for production of valuable omega -3 fatty acids (FAs), but high production costs limit commercialization. Utilization of waste as a nutrient source increases the economics of the cultivation process. Additionally, using mixed algal cultures instead of monocultures makes the cultivation process more flexible and can increase biomass and lipid production. Here, the growth and lipid production of microalgae Euglena gracilis, Selenastrum sp. and, Chlorella sorokiniana were studied in mono- and mixed cultures in small and pilot scale experiments in biowaste leachate. In pilot scale, also nutrient reduction and the number of bacteria were analyzed. Biomass production in the most productive mixed cultures was similar, but not higher than in most productive monocultures. The lipid production was highest in the small-scale monoculture of Selenastrum (10.4% DW) and in the pilot scale culture of Selenastrum with E. gracilis (11.1% DW). The content of alpha-linolenic acid (ALA) increased and eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) remained stable during the cultivation period in all pilot scale cultures. However, increases in biomass and lipid production toward the end of the cultivation resulted in higher EPA and DHA yields in the well growing monoculture of E. gracilis and in the mixed culture of E. gracilis with Selenastrum. Co-cultivation of E. gracilis and Selenastrum also had a positive influence on nutrient uptake and resistance against bacteria. This type of mixed culture may be a good option for commercialization. However, as shown here, minor changes in cultivation conditions can rapidly result in dominance of a subdominant strain, and thus the stability of strain performance and production of desired FAs needs further investigation.

# 1. Introduction

Sustainable production practices and alternatives for fish oils as a source of essential long chain polyunsaturated fatty acids (LC-PUFAs) are needed to satisfy nutritional needs of a growing world population and to protect endangered aquatic ecosystems [1]. Concerns have been raised on pollution effects of growing fish farming and over-exploitation of wild fish populations as fish-feed ingredients [1,2]. Microalgae

produce several health promoting, essential omega -3 FAs such as ALA (18:3n3), EPA (20:5n3) and DHA (22:6n3). The polyunsaturated fatty acid (PUFA) ALA is a common C18 FA (18 carbon atoms in FA chain) in plant oils, while LC-PUFAs EPA and DHA, (C $_{20}$  and C $_{22}$ ) have traditionally been obtained from marine and fresh water food [3,4]. ALA is classified as an essential FA, since it is a precursor of LC-PUFAs in the omega - 3 family [5]. Most animals cannot synthetize precursor FAs de novo, whereas conversion efficiency to LC-PUFAs varies between

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https://doi.org/10.1016/j.algal.2018.08.007

Received 4 October 2017; Received in revised form 24 July 2018; Accepted 6 August 2018 Available online 23 August 2018

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