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# Influence of hydrothermal pretreatment on microalgal biomass anaerobic digestion and bioenergy production

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#### ABSTRACT

Microalgal biomass grown in wastewater treatment raceway ponds may be valorised producing bioenergy through anaerobic digestion. However, pretreatment techniques seem to be necessary for enhancing microalgae methane yield. In this study, hydrothermal pretreatment was studied prior to batch and continuous reactors. The pretreatment increased organic matter solubilisation (8–13%), anaerobic digestion rate (30–90%) and final methane yield (17–39%) in batch tests. The highest increase was attained with the pretreatment at 130 °C for 15 min, which was attested in a laboratory-scale continuous reactor operated at a hydraulic retention time of 20 days with an average organic loading rate of 0.7 g VS/L·day. The methane yield increased from 0.12 to 0.17 L CH<sub>4</sub>/g VS (41%) in the pretreated digester as compared to the control. Microscopic images of microalgal biomass showed that pretreated cells had unstructured organelles and disrupted cell wall external layer, which may enhance the hydrolysis. Indeed, images of the pretreated reactor digestate showed how cells were more degraded than in the control reactor.

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#### 1. Introduction

High rate algal ponds (HRAP) were first developed for wastewater treatment in the 1950's in California (Oswald and Golueke, 1960). This technology consists in shallow ponds with constant mixing provided by a paddle-wheel that enhances phytoplankton photosynthesis, since it allows sunlight to penetrate through the whole system. In these microalgae-based ponds, organic matter and nutrients are removed from the influent wastewater through the symbiotic

\* Corresponding author. Tel.: +34 934016463. E-mail address: ivet.ferrer@upc.edu (I. Ferrer). http://dx.doi.org/10.1016/j.watres.2014.10.015 0043-1354/© 2014 Elsevier Ltd. All rights reserved. relation between heterotrophic bacteria and microalgae. Thus, bacteria degrade organic carbon consuming oxygen, which is synthetized by microalgae photosynthesis. In comparison to conventional activated sludge systems, here no external aeration is needed for bacteria growth. In HRAP treating urban wastewater, biomass is composed by around 90% microalgae and 10% bacteria (García et al., 2000). Harvested microalgal biomass can be treated through anaerobic digestion, a well-known process widely used for sewage sludge treatment in conventional wastewater treatment plants (WWTP). However, microalgal biomass has a low anaerobic biodegradability, mainly due to its complex cell wall structure. Actually, microalgae cell wall varies greatly among species. While some species such as *Dunaliella salina* lack the cell wall, others may differ on the cell wall composition, being a proteinbased cell wall for *Euglena gracilis* and a polysaccharide-based cell wall for *Scenedesmus obliquus*, conferring to the latter a more recalcitrant nature (González-Fernández et al., 2011). Moreover, predominant species in microalgal biomass grown in wastewater generally have a rigid cell wall, due to its adaptability to grow under variable ambient conditions, with predatory organisms and high organic content (Park et al., 2011).

In order to improve microalgae anaerobic digestion, pretreatment methods are currently being studied. So far it has been shown that reactors with a hydraulic retention time (HRT) of at least 20 days, preceded by some pretreatment step are required for reaching a methane yield around 0.30 L CH<sub>4</sub>/g VS (Passos and Ferrer, 2014; González-Fernández et al., 2012). Among the investigated pretreatment techniques, thermal pretreatment has exhibited the most promising results, reaching high methane yields, while attaining positive energy balances (Passos and Ferrer, 2014; Schwede et al., 2013). To date, temperatures from 55 to 170 °C have been applied. When thermal pretreatment is applied at temperatures higher than 100 °C, pressure increases. In this case, thermal pretreatment is so-called hydrothermal pretreatment. Generally, it is applied at temperatures between 100 and 140 °C along with pressures around 1-2 bar. As can be seen in Table 1, the methane yield may increase from 20 to 108% depending on the pretreatment conditions, and most importantly on the microalgae species used in each case.

The aim of this study was to evaluate the anaerobic digestion of microalgal biomass grown in wastewater treatment HRAP after hydrothermal pretreatment. To this end, biochemical methane potential (BMP) tests were performed with microalgae pretreated under different temperatures and exposure times. The best pretreatment condition was then studied in continuous reactors. Microscopic images where used to analyse the effect of pretreatment in microalgae cell structure and anaerobic biodegradability. Furthermore, an energy assessment was carried out in order to determine the scalability of this technology.

#### 2. Material and methods

#### 2.1. Microalgal biomass

Microalgal biomass was grown in a pilot HRAP used for secondary treatment of urban wastewater. The experimental setup was located outdoors at the laboratory of the GEMMA research group (Universitat Politècnica de Catalunya) in Barcelona (Spain). The HRAP received the primary effluent from a settling tank which had a useful volume of 7 L and an HRT of 0.9 h. The primary effluent was pumped to the HRAP by means of a peristaltic pump with a flow rate of 60 L/d. The HRAP was built in PVC with a surface area of 1.54 m<sup>2</sup>, a height of 0.3 m, a useful volume of  $0.47 \text{ m}^3$  and a nominal HRT of 8 days. Average surface loading rates were  $\pm 24$  g COD/m<sup>2</sup>day and  $\pm 4$  g NH<sub>4</sub>-N/m<sup>2</sup>day. Microalgae contact with sunlight was enhanced through continuous stirring with a bladed paddlewheel, reaching an approximate mixed liquor flow velocity of 10 cm/s. Further information on the HRAP performance may be found elsewhere (Passos et al., 2013a).

Microalgal biomass was harvested from secondary settlers with a useful volume of 9 L and an HRT of 9 h. Following, biomass was thickened by gravity in laboratory Imhoff cones at 4 °C for 24 h for reaching total solid (TS) concentration of 2.0–2.5 % (w/w). Microalgal biomass macromolecular composition was fairly stable, with 58% ( $\pm$ 2.5) of proteins, 19% ( $\pm$ 1.3) of lipids and 22% ( $\pm$ 2.7) of carbohydrates over a sampling period of four months (Passos et al., 2013a).

#### 2.2. Hydrothermal pretreatment

Hydrothermal pretreatment was carried out in an autoclave (Autester, Selecta, Spain). For the BMP tests, pretreatment conditions were 110 °C (1.2 bar) and 130 °C (1.7 bar) for 15 and 30 min; while for the continuous reactor pretreatment conditions were 130 °C for 15 min, based on previous BMP test results. Relatively low target temperatures were selected not to increase the energy demand for the thermal pretreatment and to avoid Maillard reactions which may lead to the formation of recalcitrant compounds. Exposure times (15 and 30 min) were based on literature results (Table 1). Pretreatment was performed in glass bottles of 250 mL with a useful volume of

Microalgae species	Pretreatment conditions	Reactor	Methane yield (increase)	References
Chlamydomonas sp., Scenedesmus sp. and Nannocloropsis sp.	110 and 140 °C 15 min	BMP	0.32 and 0.36 L CH₄/g VS (19 and 33%)	Alzate et al., 2012
Acutodesmus obliquus and Oocystis sp.	110 and 140 °C 15 min	BMP	0.22 and 0.26 L CH <sub>4</sub> /g VS (11 and 31%)	Alzate et al., 2012
Microspora sp.	110 and 140 °C 15 min	BMP	0.41 and 0.38 L CH <sub>4</sub> /g VS (62 and 50%)	Alzate et al., 2012; Bohutski et al., 2014
Chlorella sp. and Scenedesmus sp.	120 °C 30 min	BMP	0.40 L CH <sub>4</sub> /g VS (20%)	Cho et al., 2013
Nannocloropsis salina	100—120 °C 2 h	CSTR	0.57 L CH <sub>4</sub> /g VS (108%)	Schwede et al., 2013

Note: BMP stands for biochemical methane potential tests and CSTR stands for continuous stirred tank reactors.

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