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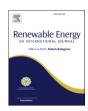
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# Environmental analysis of *Spirulina* cultivation and biogas production using experimental and simulation approach

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#### ABSTRACT

Microalgae is constituted by different compounds, interesting for the production of a wide range of endproducts by using different technologies. Many potential possibilities have been developed under the context of a biorefinery. The aim of this work is to evaluate the environmental performance of biogas production from Spirulina (Arthrospira maxima) through LCA using experimental and simulation results. For this purpose, kinetic models for batch cultivation and anaerobic digestion (AD) were determined from experimental data. Thus, Monod kinetic model and a first order model describe well microalgal biomass growth and AD, respectively. This model was used to simulate growth of Spirulina in a continuous system by using SuperPro Designer 9.5. Calculated results were compared to continuous experimental ones, obtaining good agreement in all cases. On the other hand, the whole process (cultivation, dewatering and AD of Spirulina biomass) was also simulated and the obtained results (material and energy balances) were used to construct LCA inventory data. Thereafter, environmental impacts were quantified through CML-2001 methodology using software Gabi 6.0. LCA results show that abiotic depletion of fossil resources (ADFR) category presents the highest impact, being biomass cultivation the most important contributor (about 56%). This result is directly related to the high energy consumption required for nutrient production, which also leads to increase remarkably the global warming potential (GWP) category. Main conclusion of the work is that the total/partial substitution of mineral fertilizers as nutrient source is the key to improve the environmental performance of the studied process. In this sense, a potential alternative could be the use of nutrients from wastewater or other wastes.

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# 1. Introduction

Biofuels from clean and renewable bioresources have been widely studied as an alternative to fossil fuels in the last decade [1,2]. In this context, microalgae appears as an interesting option because it is an autotroph microorganism that can accumulate lipids, can be grown in non-arable land and their cultivation does not compete with food production [2–4]. Furthermore, this microorganism can fix CO<sub>2</sub> from air by the photosynthetic mechanism similar to the behavior of higher plants, thus reducing carbon dioxide emissions [3,5]. On the other hand, compounds present in

trients, etc, can be also interesting from an industrial point of view [6]. Thus, as recently reported, the use of these compounds for the production of a wide range of end-products (valuable chemicals, human and animal food, etc.) is potentially possible, although a number of drawbacks should be overcome [7,8]. Therefore, regarding to the desired product, different schemes based on biorefinery approach can be used to optimize the use of microalgal sources. These processes consist of a first biomass fractionation step and a further stage in which separated compounds are transformed into a variety of products including biofuels, biomolecules, biomaterials and food [9,10]. Although there are many technologies to convert microalgal biomass, they can be classified in two groups:

thermochemical processes (direct combustion for energy

microalgae composition, such as carbohydrates, protein, vitamins, minerals, carotenoids, long-chain omega-3 fatty acids, phytonu-

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production, gasification for syngas production, liquefaction for biofuel production, pyrolysis for bio-oil production, etc.) and biochemical processes (anaerobic digestion for biogas production, alcoholic fermentation for bioethanol production, transesterification for biodiesel production, etc.) [11].

By taking into account energy related products, biofuels obtained from microalgae has been widely studied [5,12,13]. These works have concluded that current processes must be improved to become competitive because of their large energy requirements. Biogas production, combined biodiesel and biogas production; or extraction of valuable products including biogas production from resulting residues have been reported to improve energy performance of microalgae processing [14–16]. Therefore, biogas production appears as an ideal option to be included in microalgae transformation and use processes.

Biogas production is carried out by anaerobic digestion of algal biomass, which consists of the conversion of organic matter in the presence of methanogenic bacteria, obtaining biogas (mainly composed of methane and carbon dioxide) and a solid waste (digestate). The obtained methane can be used for different purposes, such as electricity production, fuel for internal combustion engines, etc. [11,17,18].

The use of different microalgae for biogas production has been widely reported in the literature. Thus, Jankowska et al., [19] studied different algal biomass (*Scenedesmus*, *Chlorella*, *Nannochloropsis*, *Spirulina*, etc.). Capson-Tojo et al., [20] evaluated *Nannochloropsis gaditana* biomass after the lipid extraction to obtain biogas using a bio-refinery scheme. Ramos—Suárez et al. [21] analyzed *Scenedesmus* extracted residues as substrates for methane production. Microalgae *Spirulina* has been described in literature as an attractive substrate for anaerobic digestion process because of its high growth rate, and fermentability, superior to other microscopic algae [22,23].

One of the key points of these processes is the evaluation of their environmental and energy performance. In this sense, Life Cycle Assessment (LCA) methodology is a suitable tool to quantify positive or negative impacts of processes and products according to different environmental categories. This methodology allows a systematic estimation of the environmental changes related to the examined process, quantification of consumptions and emissions and their effects on human health, eco-systems and resources depletion. There are some LCA studies focused on biogas production from microalgae [17,24]. However, they are mostly based on literature data.

The aim of this work is to evaluate the environmental performance of biogas production from *Arthrospira maxima* through LCA using experimental and simulation results. For that purpose, kinetic models for cultivation and anaerobic digestion of *Arthrospira maxima* were determined from experimental data. Monod kinetic model was found to describe well microalgal biomass growth. On the other hand, experimental data on biogas production were fitted to a first order kinetic model. Thereafter, the whole process, including cultivation, dewatering and anaerobic digestion of *Spirulina* biomass, was simulated by using SuperPro Designer 9.5. This allows the scaling-up of the process. Besides, material and energy results obtained by simulation were used to construct LCA inventory data. Finally, environmental impacts were quantified through CML-2001 methodology using software Gabi 6.0.

#### 2. Methods

#### 2.1. Microalgae cultivation

#### 2.1.1. Batch experiments

Batch cultivation experiments were carried out in 50 ml photo-

bioreactors (schematically represented in Fig. 1(a)). F/2 Guillard medium (provided by Algaenergy) was used to supply the required nutrients.  $CO_2$  was fed into the culture solution at a flow rate of 3.5 l/min. These conditions ensure an excess of  $CO_2$  in the cultivation medium during the whole experiment according to theoretical  $CO_2$  fixation calculated in literature [25–27]. Light requirements were provided by white LED lamps (12 V/24 W) under an irradiance of  $108 \ \mu mol \cdot photons \cdot m^{-2}s^{-1}$ .

Different inoculum:culture medium ratios and light:darkness photoperiods were experimentally tested. In this work, inoculum:culture medium ratio of 5:45 (vol/vol) and light:darkness photoperiod of 12:12 h were selected as the optimal values of both parameters. The distance between the LED light panel and culture medium was 2 cm. These conditions were selected as the optimal ones according to our experimental previous work [28].

*Spirulina* algal cultivation was carried out at 20 °C and pH = 9.4. The biomass growth was monitored by taking culture samples each 24 h. Absorbance of these samples were measured by using a JASCO V-630 spectrophotometer at  $\lambda = 540$  nm. A calibration between absorbance and biomass concentration was used.

#### 2.1.2. Continuous experiments

Continuous culture experiments were performed in a 12 L stirred reactor (Bioflo 110, New Brunswick Scientific Co., Inc.) by using experimental conditions previously optimized in batch experiments (as is schematically represented in Fig. 1b). A reactor volume of 10 L was filled with a solution containing inoculum:culture medium ratio and using the light:darkness photoperiod above mentioned. CO<sub>2</sub> was supplied at flow rate of 3.5 L/min only during light periods. In all the experiments, the system was stirred at 150 rpm.

In continuous experiments, the biomass was firstly grown until finishing the exponential period according to batch results. Thereafter, the continuous experiment was started by feeding the culture medium solution (Guillard's F/2) at different flow rates (0.05, 0.1, and 0.2 L/h). Biomass concentration in the reactor output stream was measured each 24 h until obtaining similar results between consecutive measurements, thus ensuring steady state conditions.

#### 2.1.3. Elementary analysis

Elemental composition of cultivated biomass in batch reactors was determined by using an Elemental analyser (Vario EL III CHNS, Elementar Analysensysteme GmbH, Germany). The method used was that based on the sulphanilic acid standard.

#### 2.1.4. Nitrate content determination

SAN++ Skalar nutrient autoanalyzer was used to determine the nitrate concentration into cultivation medium. Samples were prefiltered, using a 0,45  $\mu$ m nylon filter, before being analyzed by means of a colorimetric method.

#### 2.2. Anaerobic digestion

Data of biogas production used to determine the kinetic model for Spirulina anaerobic digestion (AD) were taken from a previous work [29]. In this work, author used the primary sludge of a municipal wastewater treatment plant as inoculum and the digestions are performed at mesophilic conditions (35  $^{\circ}$ C) in reactors of 1.2 L of capacity containing 0.7 L of liquid phase.

#### 2.3. LCA methodology

# 2.3.1. Goal and scope definition

This study is focused on evaluating the main environmental

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