Renewable Energy 75 (2015) 374–380

Contents lists available at ScienceDirect

Renewable Energy

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

Biogas production from sewage sludge and microalgae co-digestion under mesophilic and thermophilic conditions

M.P. Caporgno [a], R. Trobajo [b], N. Caiola [b], C. Ibáñez [b], A. Fabregat [a], C. Bengoa [a, *]

[a] Departament d'Enginyeria Química, Universitat Rovira i Virgili, Av. Països Catalans 26, 43007, Tarragona, Spain
[b] IRTA Aquatic Ecosystems, Ctra. Poble Nou Km 5.5, 43540, Sant Carles de la Ràpita, Spain

ARTICLE INFO

Article history:
Received 17 January 2014
Accepted 7 October 2014
Available online

Keywords:
Biogas
Biomethane potential (BMP)
Co-digestion
Isochrysis galbana
Selenastrum capricornutum
Sewage sludge

ABSTRACT

Isochrysis galbana and *Selenastrum capricornutum*, marine and freshwater microalgae species respectively, were co-digested with sewage sludge under mesophilic and thermophilic conditions. The substrates and the temperatures significantly influenced biogas production.

Under mesophilic conditions, the sewage sludge digestion produced 451 ± 12 mL_{Biogas}/g_{SV}. Furthermore, all digesters were fed with *I. galbana*, or mixed with sludge, resulting in an average of 440 ± 25 mL_{Biogas}/g_{SV}. On the contrary, *S. capricornutum* produced 271 ± 6 mL_{Biogas}/g_{SV} and in the mixtures containing sludge produced intermediate values between sludge and microalgae production.

Under thermophilic conditions, the sewage sludge digestion achieved yet the highest biogas yield, 566 ± 5 mL_{Biogas}/g_{SV}. During co-digestion, biogas production decreased when the microalgae content increased, and for *I. galbana* and for *S. capricornutum* it reached minimum values, 261 ± 11 and 185 ± 7 mL_{Biogas}/g_{SV}, respectively. However, no evidence of inhibition was found and the low yields were attributed to microalgae species characteristics.

The methane content in biogas showed similar values, independently from the digested substrate, although this increased by approximately 5% under thermophilic condition.

1. Introduction

The industrialisation process and the current population growth have had an immense impact on the environment. The demand on water and petroleum-based fuels are clear evidence of the increase on natural resources.

The wastewater treatment plants (WWTPs) have become an essential component in society to ensure necessary water supplies. A by-product produced in these facilities during the wastewater treatment process is sewage sludge; hence as demand grows so does this by-product. The final disposal of sludge is a problem in WWTPs as this can represent up to 50% of the operating cost [1]. The anaerobic digestion of the sludge is one of the most widespread stabilization processes in WWTPs, which converts sludge into a stable product and simultaneously recovers energy by biogas generation. On the other hand, WWTPs are a potential source of nutrients for microalgae growth: CO_2 generated and released in the atmosphere when biogas is burned; also nitrogen and phosphorus are present in wastewater [2–5].

Microalgae arose as a source of valuable chemical productions, but may be the main feature was as promising feedstocks for renewable biofuels. In 2008, 88% of world energy demand was supplied by fossil fuels, including oil (35%), coal (29%) and natural gas (24%) [4]. Unfortunately, coal supplies depletion is predicted by 2112 and oil and gas reserves depletion by 2042, thus a rapid transition to renewable energy is needed in the near future [6]. Biodiesel production from microalgae appeared as a solution due to the microalgae advantages over the feedstocks currently used in the biodiesel industry; however, the process scale-up is unviable nowadays without a cost-effective dewatering method. Additionally, microalgae cultivation is not simple, although they grow naturally in aquatic environments. The low cell densities required for light penetration to ensure their growth and the small size of the cells are counterproductive to the harvesting step, which can represent between 20 and 30% of the total biomass production costs [4,7].

The anaerobic digestion process creates an alternative for energy recovery from microalgae. The ability to process wet biomass avoids a drying step, thus reducing large amounts of energy input. Besides, all microalgae compounds can be turned into biogas and those species unsuitable for biodiesel production due to their low oil content become potential substrates [8]. During anaerobic

* Corresponding author. Tel.: +34 977 558619; fax: +34 977 559667.
E-mail address: christophe.bengoa@urv.cat (C. Bengoa).

http://dx.doi.org/10.1016/j.renene.2014.10.019
0960-1481/

digestion, the organic nitrogen and phosphorus initially as biomass constituents are converted into ammonium and phosphate, and can be recycled for microalgae cultivation reducing fertilizer needs [7]. Under these circumstances, the possibility of microalgae growth followed by anaerobic digestion in WWTPs is currently being assessed [9].

Since Golueke et al. [10] studied anaerobic digestion of microalgae for the first time, at the end of the 1950s, many microalgae species and process conditions have been evaluated for biogas production. However, not all species have the potential to produce high amounts of methane, and the proper selection of species and operating conditions are the key for biogas production [11]. The cell wall structure and composition, the carbon to nitrogen ratio (C/N), are crucial for microalgae degradability [8]. Cell wall resistance may act as a barrier hampering microorganisms being attacked; on some occasions, pre-treatment methods are required to improve digestibility [12,13]. The C/N balance, low in microalgae derived from their high protein content, increased ammonia and volatile fatty acids concentration and may potentially inhibit anaerobic digestion. Co-digestion with carbonaceous-rich waste is a method to balance this ratio and overcome this disadvantage. Sewage sludge, waste activated sludge, waste paper and corn straw are examples of increasing biogas production from different microalgae species [14–17].

In this context, this paper attempts to evaluate biogas produced from two microalgae species: the marine species *Isochrysis galbana*, and the freshwater species *Selenastrum capricornutum*, by their co-digestion with sewage sludge. Initially, the influence of the microalgae to sludge ratio and the digestion temperature over the biogas production and its methane content was evaluated.

2. Materials and methods

2.1. Materials

2.1.1. Sewage sludge

The sludge sample consists of a primary and secondary blend, in ratio 65:35 v/v. It was collected from the municipal WWTPs in Reus (Tarragona, Spain) designed to process approximately 25,000 m^3 of wastewater daily. Sludge was received weekly and immediately stored at 4 °C in a fridge prior to use. The experiment involved the use of two different kinds of reactors, semi-continuous reactors for acclimation and batch reactors for co-digestion; the same type of sludge was utilised in both reactors. For the first reactors the maximum storage time was a week; for the second, the maximum storage time was 2 days in order to avoid major changes on its composition or properties.

2.1.2. Microalgae

Two microalgae species were used in the experiments: *Isochrysis galbana*, marine species, and *Selenastrum capricornutum*, freshwater species. Both species were provided by the Institute for Research and Technology in Food and Agriculture IRTA (San Carles de la Ràpita, Spain). For the marine species cultivation, Walne's medium [18] was prepared with filtered (0.45 μm), autoclaved seawater from Alfacs Bay (Ebro Delta, Spain). For the freshwater species cultivation, Woods Hole MBL medium [19] was prepared with deionised water, and autoclaved before inoculation. The microalgae were cultured under batch conditions in 6 L volumetric flasks. The volumetric flasks were kept in an isothermal chamber at 20 ± 3 °C under continuous irradiance of 120–150 μmol photons m^{-2} sec^{-1}, provided by cool-white fluorescent lamps (Philips TLD 58w/865). Mixing was provided by air flow and air was enriched by 0.7% CO_2 addition. The microalgae were grown under these conditions for approximately 2 weeks, until the culture reached a plateau in terms of absorbance (680/800 nm).

2.1.3. Inoculum

From the start, inoculum was provided by the municipal WWTP in Reus (Tarragona, Spain). It consisted of digested sludge from mesophilic and thermophilic anaerobic reactors under continuous operating conditions. Although acclimation to new conditions was not strictly required, an anaerobic semi-continuous plant was set up to adapt inoculum to more stable temperatures, 33 °C and 50 °C. Four reactors (5 L each) were placed in a thermostatic bath for temperature control under magnetic stirring. Biogas production was continuously registered by volumetric gas flow meters. All reactors were daily fed with sludge, following effluent withdrawal.

Prior to the co-digestion experiments, the inoculum had previously been “degassed” by incubation at a constant temperature without feeding, to reduce the residual biodegradable organic material [20]. Observation indicated no significant methane being produced after 5 days incubation.

2.2. Experimental procedure

2.2.1. Microalgae preparation

Once microalgae were received, the biomass was collected by centrifugation using a centrifuge with a capacity of 240 mL sample (Digicen 20, Orto Alresa Centrifuges). Microalgae were recovered after 4 min at 10,304 RCF without temperature control. The supernatant was removed and only the pellet was recovered. Deionised water was added to recover the pellet and the total solid (TS) content of the microalgae suspension was around 10 g/L. The suspension was stored at 4 °C prior to use, and always used before 12 h after microalgae centrifugation finished.

2.2.2. Co-digestion procedure

The batch reactors were set up following the procedure described by Angelidaki et al. [20]. The temperature for the experiments was chosen in accordance to the temperature in the WWTP reactors, 33 °C in the mesophilic range and 50 °C in thermophilic range.

All experiments of co-digestion were conducted in 120 mL serum bottles in triplicate. To carry out the digestion, the microorganisms were provided with 50 mL “degassed” inoculum and optimal environmental conditions were assured by 10 mL anaerobic basic medium addition. The substrates were sewage sludge and microalgae, separately or mixed. Blank assays were prepared without substrate addition, and the biogas production was subtracted from the reactors fed with the substrates. The specific methanogenic activity for inoculum was determined with an initial concentration of 1 g/L acetic acid in the reactor.

Sludge and microalgae were added according to the experimental design. The total substrate amount was decided at 0.12 g volatile solids (VS) equivalent to 2–3 g_{COD}/L, which generates a measurable but not excessive biogas volume. The reactor feed was based on 100% sludge VS for sewage sludge digestion (denoted as Sludge in the figures), and subsequently 25%, 50%, 75% and 100% of the sludge VS were replaced with microalgae VS respectively (denoted as 25%, 50%, 75% and 100% in the figures). Deionised water was added to a final volume of 80 mL and the reactors were closed with a septum and an aluminium crimp. Finally, the reactors were purged with nitrogen to assure anaerobic conditions and placed into an oven.

Biogas production was volumetrically measured by liquid displacement. As a barrier solution, a saline solution consisting of 200 g/L NaCl and 5 g/L citric acid was used. Prior to measurements, the reactors were removed from the oven and left to reach room