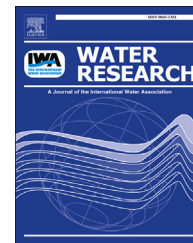




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Influence of temperature and pretreatments on the anaerobic digestion of wastewater grown microalgae in a laboratory-scale accumulating-volume reactor

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ABSTRACT

This laboratory-scale study investigated the performance of a low-cost anaerobic digester for microalgae. Low (~2%) solids content wastewater-grown microalgal biomass (MB) was digested in an unmixed, accumulating-volume reactor (AVR) with solid and liquid separation that enabled a long solids retention time. AVRs (2 or 20 L) were operated at 20 °C, 37 °C or ambient temperature (8–21 °C), and the influence of two pretreatments – low-temperature thermal (50–57 °C) and freeze-thaw – on algal digestion were studied. The highest methane yield from untreated MB was in the 37 °C AVR with 225 L CH₄ kg volatile solids (VS)⁻¹, compared with 180 L CH₄ kg VS⁻¹_{added} in a conventional, 37 °C completely stirred tank reactor (CSTR), and 101 L CH₄ kg VS⁻¹_{added} in the 20 °C AVR. Freeze-thaw and low-temperature thermal pretreatments promoted protein hydrolysis and increased methane yields by 32–50% at 20 °C, compared with untreated MB. Pretreatments also increased the mineralisation of nitrogen (41–57%) and phosphorus (76–84%) during digestion. MB digestion at ambient temperature was comparable with digestion at 20 °C, until temperature dropped below 16 °C.

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Abbreviations: AD, anaerobic digestion; AVR, accumulating-volume reactor; COD, chemical oxygen demand; CSTR, completely stirred tank reactor; d, days; h, hours; HRAP, high rate algae pond; HRT, hydraulic retention time; MB, microalgae biomass; P, phosphorus; OLR, organic loading rate; SCOD, soluble chemical oxygen demand; S_D, solubilisation degree; SRT, solid retention time; VFA, volatile fatty acids; VS, volatile solids; TKN, total kjeldahl nitrogen; TS, total solids; TVFA, Total volatile fatty acids.

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1. Introduction

The sustainability of microalgal biofuels has been questioned if chemical fertilizers are needed for cultivation (Lam and Lee, 2012). However, microalgae have been used to assimilate nutrients from wastewaters in high rate algal ponds (HRAP), e.g., in the USA and New Zealand (Craggs et al., 2012). The microalgae could also use CO₂ from the burning of fuels for their growth. Despite recent research, microalgal biofuels remain unviable because of low energy conversion efficiencies. Thus, simplified algal biofuel processes without extensive energy inputs are needed (Lam and Lee, 2012).

Methane production via anaerobic digestion (AD) is a simple way to utilise microalgal biomass (MB) for energy production. However, there are several challenges for the AD of microalgae, including: (1) low solid content of algal biomass, causing unwanted short hydraulic retention time (HRT) in completely stirred tank reactor (CSTR); (2) difficult and slow degradability of algae, leading to low methane yields, compared to theoretical values (Alzate et al., 2012); and (3) high nitrogen concentration of algae, potentially inhibiting the AD process (Sialve et al., 2009).

For CSTR digesters, the low solid concentration of harvested algal biomass, leads to short HRTs (<15 d) and/or low organic loading rates (OLR) (<1.0 g VS L⁻¹ d⁻¹). Laboratory AD of microalgae using CSTR, have typically had a HRT of 15–30 days (d) (Ras et al., 2011; González-Fernández et al., 2012a). However, algae have been found to be slowly degradable, and Ras et al. (2011) reported 63% higher methane yield from *Chlorella* at a HRT of 28 d compared with 16 d. Effluent is continuously removed from CSTR digesters, meaning that this digestate contains a portion of recently added and undigested material. Therefore, at HRTs <30 d, CSTR digestate may have significant residual methane potential, which has been found to be mainly (e.g., 88–93%) due to the solid fraction (Angelidaki et al., 2006). Achieving longer solid retention times (SRTs) (>30 d) by e.g., using an unmixed, accumulating-volume reactor (AVR) presented in this study, could offer more complete degradation and higher methane yields for microalgae.

Anaerobic digestion is most commonly conducted at a mesophilic temperature range of 30–40 °C, which requires heating of the reactors in most climate zones. However, with low total solids (TS) substrates such as microalgae, what is actually being heated is mostly water. Digestion in low temperatures (<20 °C), conducted by psychrophilic or acclimatised mesophilic microorganisms, could be a feasible option to decrease energy input (Kashyap et al., 2003). In low temperatures, however, metabolism of microorganisms is reduced; specifically, the hydrolysis step is considered rate limiting (Halalshah et al., 2011).

To improve algae degradability and thus methane yield without lowering the overall energy balance, there is a need for low-cost pretreatment techniques. Among the pretreatments, low-temperature thermal treatment (<100 °C) could be a sustainable option, since excess heat (50–70 °C) is commonly available, e.g., from combined heat and power units or industrial processes. Another potential low-cost pretreatment could be freeze-thaw of algae biomass, which to our

knowledge, has not previously been reported for microalgae, but has been researched for wastewater sludge conditioning (Gao, 2011).

The results of previous studies on low-temperature thermal pretreatment of microalgae have been contradictory. Passos et al. (2013) showed that low-temperature pretreatment for <10 h increased microalgae methane production. Conversely, Alzate et al. (2012) reported no effect, or even decreased methane production after >12 h pretreatment at 55–60 °C. Indeed, a short treatment time may be favourable for microalgae, for example 4-h at 55–80 °C was found to achieve >80% volatile solids (VS) and chemical oxygen demand (COD) solubilisation (González-Fernández et al., 2012a; Passos et al., 2013). Solubilisation is an important factor for methane production, since hydrolysis is often the limiting step of the AD process.

Freeze-thaw improves wastewater sludge dewaterability and settleability (Örmeci and Vesilind, 2001; Hu et al., 2010; Gao, 2011), and increases COD solubilisation (Gao, 2011) and methane yield (Montusiewicz et al., 2010). As wastewater sludge and microalgal-bacterial biomass share the same unicellular physical properties, freezing could also be considered as a pretreatment method for MB. However, freeze pretreatment may be costly in terms of energy use. In countries with cold winter seasons, nature could provide this energy at a low cost, if suitable freezing infrastructure could be developed.

The objective of this study was to investigate a low-cost AD process for wastewater-grown microalgae. For this purpose, a novel AVR was used and operated at low temperatures. Low-cost pretreatments (low-temperature thermal and freeze-thaw) were also studied to improve algal digestion.

2. Materials and methods

The experimental set up is shown in Fig. 1. During the nine-month experimental period (June 2011–March 2012), two experiments were conducted. In experiment 1 (five months), MB was digested in AVRs at 20 °C and 37 °C, and as a reference, in a CSTR at 37 °C. In experiment 2 (four months), pretreated (low-temperature thermal and freeze-thaw) MB was digested in AVRs at 20 °C, with untreated MB as the control. During experiment 2, untreated MB was also digested under ambient temperature conditions. The experiments were conducted in duplicate or triplicate.

2.1. Microalgal biomass and inocula

The HRAP that produced the microalgae used in this experiment were fed with primary settled sewage and were operated with a HRT of 4–8 days, water depth of 0.3 m and horizontal water velocity of 0.15 m s⁻¹. Further details of the HRAP operating conditions are described in Park and Craggs 2010. For reactor feeds, HRAPs were harvested 1–5 times per week over the experimental period, using gravity-settling cones. The TS, VS and pH of each harvest were measured. The dominant species during the nine-month period, as analysed every third week with a microscope, was found to vary over time, consisting of *Pediastrum* sp., *Micractinium* sp. and

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