Bioresource Technology 196 (2015) 606–612

Contents lists available at ScienceDirect

Bioresource Technology

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

Comparing activated carbon of different particle sizes on enhancing methane generation in upflow anaerobic digester

Suyun Xu [a,*], Chuanqiu He [a], Liwen Luo [a], Fan Lü [b], Pinjing He [b], Lifeng Cui [a]

[a] Department of Environment & Low-Carbon Science, School of Environment and Architecture, University of Shanghai for Science and Technology, Shanghai 200093, China
[b] State Key Laboratory of Pollution Control and Resource Reuse, Tongji University, Shanghai 200092, China

HIGHLIGHTS

- Both PAC and GAC could promote the syntrophic metabolism of alcohol and VFAs.
- PAC is superior to GAC on the enhancement of biomethane generation process.
- PAC provides more abundant micropore–mesopore structure for bacteria to colonize.
- Microbial community colonized on PAC and GAC was characterized and compared.

ARTICLE INFO

Article history:
Received 17 June 2015
Received in revised form 3 August 2015
Accepted 8 August 2015
Available online 14 August 2015

Keywords:
Activated carbon
Archaea
Methanogenesis
Syntrophic acetate oxidization
Volatile fatty acid

ABSTRACT

Two sizes of conductive particles, i.e. 10–20 mesh granulated activated carbon (GAC) and 80–100 mesh powdered activated carbon (PAC) were added into lab-scale upflow anaerobic sludge blanket reactors, respectively, to testify their enhancement on the syntrophic metabolism of alcohols and volatile fatty acids (VFAs) in 95 days operation. When OLR increased to more than 5.8 g COD/L/d, the differences between GAC/PAC supplemented reactors and the control reactor became more significant. The introduction of activated carbon could facilitate the enrichment of methanogens and accelerate the startup of methanogenesis, as indicated by enhanced methane yield and substrate degradation. High-throughput pyrosequencing analysis showed that syntrophic bacteria and *Methanosarcina* sp. with versatile metabolic capability increased in the tightly absorbed fraction on the PAC surface, leading to the promoted syntrophic associations. Thus PAC prevails over than GAC for methanogenic reactor with heavy load.

1. Introduction

In China, brewery wastewater is 1.5–2.0% of the total wastewater production in the country (Feng et al., 2008). Due to its high concentration of biodegradable organic matters, biological treatment is a good choice for the treatment of brewery wastewater (Parawira et al., 2005). Generally, aerobic treatment has been successfully applied for the treatment of brewery wastewater and recently anaerobic systems have become an attractive option (Simate et al., 2011). One of the most popular anaerobic processes for wastewater treatment is the Upflow Anaerobic Sludge Blanket (UASB). Simultaneously, methane derived from anaerobic treatment of brewery wastewater in anaerobic digesters is an alternative fuel.

However, as anaerobic bacteria are slow growing microorganisms, the major problem of anaerobic digester is the long start-up periods and difficulty in spontaneous development of granulation. The reduction of start-up time is one of the key parameters to increase the competitiveness of high-rate anaerobic reactors. The presence of porous media such as plastic-, ceramic- or carbon-based material, is known to enhance the performance of anaerobic reactors by offering a rough and fissured surface for microorganisms to settle and colonize easily (Fernández et al., 2007; Kindzierski et al., 1992). Among these materials, bacteria preferably adhered to the solid supports made of carbon material (Kuroda et al., 1988). For instance, anaerobic biofilm reactor packed with Granular Activated Carbon (GAC) has been successfully used to achieve effective decontamination for olive mill wastewater even at high organic loads of 15.7–55.6 g COD/L/d (Bertin et al., 2004). Due to its inherent absorption capacity, AC can help to reduce the organic shock loading impact on the process of biomethane generation (Aktaş and Çeçen, 2007).

* Corresponding author. Tel./fax: +86 21 5527 5979.
E-mail address: xusy@usst.edu.cn (S. Xu).

http://dx.doi.org/10.1016/j.biortech.2015.08.018
0960-8524/

Furthermore, in recent studies, granular activated carbon (GAC) was reported to facilitate direct interspecies electron transfer (DIET) between defined species such as *Geobacter metallireducens* and *Methanosarcina barkeri* (Liu et al., 2012), or mixed culture within methanogenic aggregates (Luo et al., 2015). In fact, electron transfer to methanogens during the syntrophic metabolism can be realized indirectly by taking H_2 or formate as the interspecies electron transfer carries, or directly via biological electrical connections or a combination of biological and abiological electron transfer components (Chen et al., 2014). Related studies suggested that syntrophic metabolism, such as the syntrophic acetate oxidization played a important role in the initiation of methanogenesis, especially in the stressed environmental conditions (Hao et al., 2010). DIET was the primary mechanism of interspecies electron transfer in UASB reactors treating brewery wastewater (Liu et al., 2012; Morita et al., 2011; Shrestha et al., 2014). Thus multiple lines of evidence suggested that the strategy of introducing conductive carbon materials into anaerobic digester may help to strengthen the syntrophic associations between bacteria and methanogens and therefore to enhance the digester's effectiveness (Zhao et al., 2015).

Nevertheless, the colonization of bacteria was affected by the characteristics of the supporting materials. Bacteria preferably adhered to the moderately rough surfaces that have pores measuring a few tenths of a micron in diameter more than the polished surfaces and rough surfaces (Kuroda et al., 1988). It found that the outer pores of AC are inaccessible for the organisms, which can therefore attach only to the external surface and enter only some large macropores and fissures (Kuroda et al., 1988; Voice et al., 1992). Meanwhile, there are intrinsic correlation between microbial community and the conductivity of anaerobic sludge aggregates (Shrestha et al., 2014). Thus it can foresee that the adhesion of microbial communities on AC with different size and macroporous structures might be different, leading to the differentiated efficiencies of DIET and methane conversion. However, to the authors' knowledge, the relevant studies are quite few, especially about the selective colonization of functional microbes on carbon materials with different particular sizes.

Therefore, two kinds of AC with different particle sizes (i.e. 0.84–2.00 mm and 75–177 μm) have been supplemented into UASB reactors and the reactors' performance were compared by studying the stability, substrate bioconversion efficiency and CH_4 productivity under a large range of high organic loading rates. Additionally, microorganism colonized on the surface of AC was characterized by scanning electron microscopy (SEM) and high-throughput 16S rRNA gene pyrosequencing analysis in an attempt to explain the effects of the AC properties on the adhesion and colonization of bacteria.

2. Methods

2.1. Reactor design and operation

Three identical lab-scale upflow anaerobic digesters (internal diameter of 160 mm and height of 360 mm) were used in this study, each of which had a working volume of 5.6 L. The seed sludge was taken from the secondary sedimentation tank of a full-scale wastewater treatment plant (Quyang Sewage Treatment Plant) located in Shanghai, China. After four months acclimation, seed sludge was sieved through 0.5 mm mesh before feeding into UASB reactors. The final concentration of volatile suspended solids (VSS) in reactors was adjusted to 6.0 g/L, the ratio of VSS to total suspended solids (TSS) was 66%.

Two different particle sizes of coal-based AC, i.e. 10–20 mesh (0.84–2.00 mm) of GAC and 80–100 mesh (75–177 μm) of powered activated carbon (PAC) were added into R1 and R2, respectively, each of which received 5 g/L of AC. R0 without AC was operated as a control. Synthetic brewery wastewater was used in this work for feeding the reactors. Ethanol and glucose were used as carbon source, which concentrations were 28.15 g/L and 17.79 g/L, respectively. Additionally, 2.59 g urea, 0.20 g yeast extract, 1.91 g K_2HPO_4, 1.24 g KH_2PO_4 and 2 mL trace element solution were added (per liter) to supply necessary nutrient for microbial growth. The total COD of original solution for synthetic wastewater is 65.3 g/L.

During the experiment, the original solution was diluted with deionized water into different organic loads according to the experiment operation conditions (Table 1). The pH of diluted influent solution was adjusted to 7.2 using Na_2CO_3 and HCl solutions. Reactors were operated in batch mode, and 2 L of diluted synthetic wastewater was fed into each reactor every 48 h. Three reactors were placed in a temperature-control incubator at 35 ± 2 °C without light.

2.2. Analytical methods

2.2.1. Physiochemical analysis

Biogas produced from each reactor was trapped by gas bags, the volume of which was measured with syringe. A cumulative value of biogas generated within two-day batch operation was recorded. Biogas compositions (CH_4, CO_2 and H_2) were determined by a gas chromatograph (GC9890B, Shanghai Linghua Co., China) equipped with a thermal conductivity detector.

Before feeding fresh substrate into reactors every two days, the effluent of each reactor was sampled and analyzed to monitor the variations of pH, total organic carbon (TOC) and volatile fatty acids (VFAs). After filtration (0.45 μm), the concentrations of VFAs were analyzed using high performance liquid chromatography (Waters 2695/2489, USA). TOC was analyzed by using Total Carbon/Total Nitrogen analyzer (Multi N/C 3100, Jena Co., Germany). The microorganism attached on AC was observed by scanning electron microscope (SEM), which detailed method was described in Supporting Information.

2.2.2. Microbiological analyses

2.2.2.1. Spatial fractionation of sludge samples. To study the spatial distribution of bacteria and archaea on AC, the microorganisms in the sludge samples were divided into three parts, i.e. suspended, loosely attached and tightly adsorbed fractions according to the method developed by (Luo et al., 2015). Then the total DNA of three fractions were extracted using the PowerSoil™ DNA isolation kit (Mo-Bio Laboratories Inc., CA), and labeled with "S", "L" and "T" respectively. The concentration of DNA samples were analyzed with UV–Vis spectrophotometer (Nanodrop 2000, Thermo, USA). Each sample has been conducted in triplicate, then the combined DNA solution was stored for the following analysis.

2.2.2.2. High-throughput 16S rRNA gene pyrosequencing. The microbial community of samples collected from suspended sludge and biofilm were analyzed by using high-throughput pyrosequencing

Table 1
Operation conditions for upflow anaerobic digesters.

Stage	Run time (day)	OLR (g COD/L/d)			HRT (d)
		R0	R1	R2	
I	1–10	2.9	2.9	2.9	5.6
II	11–68	5.8	5.8	5.8	5.6
III	69–83	5.8	7.0	7.0	5.6
IV	84–95	5.8	12.0	12.0	5.6