



Enhancing the anaerobic digestion of lignocellulose of municipal solid waste using a microbial pretreatment method



Xufeng Yuan¹, Boting Wen¹, Xuguang Ma, Wanbin Zhu, Xiaofen Wang, Shaojiang Chen, Zongjun Cui^{*}

College of Agronomy and Biotechnology, Center of Biomass Engineering, China Agricultural University, Beijing 100193, China

HIGHLIGHTS

- Effect of microbial pretreatment on methane production of LMSW was evaluated.
- Soluble substrates in hydrolysate increased obviously after microbial pretreatment.
- CH₄ production yields and rates significantly increased after microbial pretreatment.

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ABSTRACT

The use of biological pretreatment in anaerobic digestion systems has some potential; however, to date, these methods have not been able to effectively increase methane production of lignocellulose of municipal solid waste (LMSW). In this study a thermophilic microbial consortium (MC1) was used as a pretreatment method in order to enhance biogas and methane production yields. The results indicated that sCOD concentration increased significantly in the early stages of pretreatment. Ethanol, acetic acid, propionic acid, and butyric acid were the predominant volatile organic products in the MC1 hydrolysate. Biogas and methane production yields of LMSW significantly increased following MC1 pretreatment. In addition, the methane production rate of the treated LMSW was greater than that observed from the untreated sample.

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1. Introduction

The quantity of municipal solid waste (MSW) generated in China has increased by 8–10% per year over the past several decades (Shi et al., 2008). For example, in 2007 alone, 150 million tons of MSW were produced in China (Dong et al., 2010). Anaerobic digestion (AD) is often considered one of the more economically, and environmentally sound technologies currently used in the treatment of MSW (Jun et al., 2009). However, use of this technology is not without limitation, especially for MSW. Since approximately 40–50% of landfill space is occupied by paper and cardboard waste (Sufliya et al., 1992), of which lignocellulose of municipal solid waste (LMSW) is a significant component (Béguin and Aubert, 1994). In addition, the solubilisation of cellulose and hemicellulose (both primary components of LMSW) is the rate-limiting step during the anaerobic digestion of lignocellulose of MSW (O'Sullivan and Burrell, 2007). As a result, a number of

studies have examined the use of different pretreatment methods, in an effort to maximize LMSW digestion.

Mechanical pretreatment has been successful in reducing particle size and disrupting the crystalline structure of LMSW (Pommier et al., 2010). Thermal and chemical pretreatments are also effective at enhancing anaerobic digestion of LMSW (Clarkson and Xiao, 2000; Fox and Noike, 2004; Fox et al., 2003; Teghammar et al., 2010; Xiao and Clarkson, 1997). However, these pretreatment methods often require significant energy inputs, and therefore may not be the most economically and environmentally sound technologies (Binod et al., 2010; Sun and Cheng, 2002). To remedy this, the use of biological pretreatment is currently being explored. Biological pretreatment, which is a safe and environmentally-friendly method by using microorganisms, offers some conceptually important advantages such as low chemical and energy use (Binod et al., 2010). However, to date, few biological pretreatment methods have been demonstrated to improve methane production of LMSW. Previous studies have shown that many pure cultures, such as anaerobic bacteria, fungi, and actinomycetes, were effectively able to degrade lignocellulose (Desvaux et al., 2000; Xu and Goodell, 2001). The solubilization of lignocellulose occurs

* Corresponding author. Tel.: +86 10 62733437; fax: +86 10 62731857.

E-mail addresses: acuizj@cau.edu.cn, B08010004@cau.edu.cn (Z. Cui).

¹ These authors contributed equally to this article and are joint first authors.

naturally via the action of multiple microorganisms (Wongwilaiwalin et al., 2010). As such, the use of microbial consortia is often regarded as the most likely successful approach to increasing the methane production rate of LMSW. This is likely a result of the lack of feedback regulation, and metabolite repression that commonly occurs in single strain anaerobic digesters. (Haruta et al., 2002; Soundar and Chandra, 1987). In fact, several studies have directly demonstrated the efficiency of constructed microbial consortia in the hydrolysis of lignocellulose (Guo et al., 2011; Haruta et al., 2002; Wongwilaiwalin et al., 2010; Yang et al., 2011). However, to our knowledge such microbial consortia have not been directly used in the pretreatment of LMSW. Therefore, the objective of this present study was to develop and demonstrate a novel microbial pretreatment method for the effective anaerobic digestion of LMSW. To meet this objective we analyzed the effectiveness of a thermophilic cellulose-degrading consortium (MC1) in enhancing LMSW anaerobic digestion.

2. Methods

2.1. Materials

The lignocellulose from municipal solid waste (LMSW) was obtained by mixing waste office paper, newspaper, and cardboard, all of which were collected from a refuse collection point at the China Agricultural University (Haidian District, Beijing City, China). The mass-mixing ratio of office paper, newspaper, and cardboard was 1:1:1. All paper waste was first cut into 20 × 20 mm squares, and oven dried at 80 °C for 48 h. The lignin, cellulose, and hemicellulose content of this waste were 14.2%, 70.1%, and 12.0%, respectively (Table 1).

2.2. Microbial consortium and culture medium

The microbial consortium (MC1) capable of effectively degrading various cellulosic materials (e.g. filter paper, cotton and rice straw) under aerobic static conditions was constructed via a succession of enrichment cultures as in Haruta et al. (2002). The high stability of the consortium's degradation ability was demonstrated by its ability to tolerate several rounds of subculture in medium with/without cellulosic material, and being heated to 95 °C or frozen at –80 °C (Haruta et al., 2002). MC1 was cultured in a peptone cellulose solution (PCS) containing 1% (w/v) filter paper for three days at 50 °C, and stored at –20 °C in 20% glycerol. Although MC1 has not been fully characterized, it is known to contain *Clostridium straminisolvans* CSK1, *Clostridium* sp. FG4b, *Pseudoxanthomonas* sp. train M1-3, *Brevibacillus* sp. M1-5, and *Bordetella* sp. M1-6 (Kato et al., 2005).

Culture medium: The peptone cellulose solution (PCS) was composed of 2 g peptone, 1 g yeast extract, 2 g CaCO₃, 5 g NaCl, and 1 L H₂O (pH 8.0). All medium was autoclaved at 121 °C for 20 min and cooled prior to inoculation.

2.3. Pretreatment with the microbial consortium MC1

The primary purpose of pretreatment with MC1 was to increase cellulose and hemicellulose availability, and thus digestibility. Previously prepared and frozen MC1 was inoculated into 125 ml sterile peptone cellulose solution (PCS) with a 1% (w/v) carbon source (filter paper), and allowed cultured at 50 °C for 3 days. Following this 3 days culture, 2, 4, 10 and 20 g of LMSW were mixed with 400 ml PCS medium (final LMSW concentrations = 0.5%, 1.0%, 2.5% and 5.0%, respectively) and each inoculated with 20 ml of this 3-day-old MC1 culture. The ratio of inoculum to PCS culture medium was 1:20 (Table 2). All mixtures were subsequently incubated at 50 °C for 14 days.

Samples were obtained at: 0 (immediately after inoculation), 1, 2, 4, 6, 8, 10, and 14 days (Table 2). The pretreatment experiment consisted of 62 digesters. Experimental digesters (32) were sampled at the eight pretreatment post-inoculation times for the four substrate concentrations of 0.5%, 1.0%, 2.5% and 5.0%. Samples were analyzed for soluble chemical oxygen demand (sCOD), pH, volatile organic products (VOPs), and substrate final weight (each measurement was repeated three times). The remaining 30 digesters were used for subsequent anaerobic digestion for only the 2.5% and 5.0% substrate concentrations.

2.4. Anaerobic digestion

The residual LMSWs with 400 ml hydrolysates pretreated by MC1 were respectively digested in batch anaerobic digesters at the pretreatment times of 2, 4, 6, 8, and 10 days for the 2.5% and 5.0% substrate concentrations (Table 2). Untreated LMSWs with 400 ml PCS medium were used as the control. The volume of each anaerobic digester was 1 L, with a working volume of 750 ml. Each digester was seeded with the anaerobic sludge taken from a mesophilic anaerobic digester from the Deqinyuan Biogas Plant (Beijing, China). The sludge contained 57.2 g/l total solids (TS), 31.5 g/l volatile solids (VS), and 39.6 g/l mixed liquor suspended solids (MLSS). The ratio of substrate to inoculum (anaerobic sludge) was 1:1 in each anaerobic digester. All anaerobic digesters were purged with N₂ for 5 min to remove O₂, and then sealed with a rubber stopper. Each digestion was repeated three times at mesophilic temperature (35 °C) Average values were used in the blank test (CK) in which biogas production only resulted from the 400 ml PCS medium, and the seeded anaerobic sludge. The purpose of the blank test (CK) was to obtain the biogas and methane yield of the 400 ml PCS medium and anaerobic sludge alone. The biogas and methane yields of LMSW were calculated as follows:

$$\text{Biogas yield, (ml/g VS)} = \frac{(\text{Biogas volume})_{\text{total}} - (\text{Biogas volume})_{\text{CK}}}{\text{VS of substrates added}}$$

$$\text{Methane yield, (ml/g VS)} = \frac{(\text{Methane volume})_{\text{total}} - (\text{Methane volume})_{\text{CK}}}{\text{VS of substrates added}}$$

Table 1
Characteristics of the substrates used in the experiments.

Parameter	Office paper	Newspaper	Cardboard	Mixture (LMSW)
TS (%)	95.3 ± 0.2	93.2 ± 0.4	95.4 ± 0.3	94.6 ± 0.3
VS (%TS)	98.6 ± 0.2	96.1 ± 0.3	87.2 ± 0.2	94.0 ± 0.2
Ash (%TS)	1.4 ± 0.0	3.9 ± 0.1	12.8 ± 0.2	6.0 ± 0.1
Lignin (%TS)	1.4 ± 0.5	23.4 ± 0.5	17.8 ± 0.5	14.2 ± 0.5
Cellulose (%TS)	84.9 ± 1.3	68.5 ± 1.1	56.9 ± 0.8	70.1 ± 1.1
Hemicellulose (%TS)	12.3 ± 0.6	13.1 ± 0.3	10.7 ± 0.3	12.0 ± 0.4
COD _{substrate} (g O ₂ /g TS)	1.07 ± 0.02	1.21 ± 0.03	1.10 ± 0.03	1.13 ± 0.03

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