



Solid-state anaerobic co-digestion of spent mushroom substrate with yard trimmings and wheat straw for biogas production



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HIGHLIGHTS

- Solid-state anaerobic digestion (SS-AD) of spent mushroom substrate (SMS) was studied.
- SS-AD of only SMS was inhibited due to VFA accumulation and pH dropping.
- Co-digestion of SMS with lignocellulosic biomass feedstock enhanced SS-AD performance.
- Co-digestion of SMS with yard trimmings increased methane yield by 2- to 16-fold.

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ABSTRACT

Spent mushroom substrate (SMS) is a biomass waste generated from mushroom production. About 5 kg of SMS is generated for every kg of mushroom produced. In this study, solid state anaerobic digestion (SS-AD) of SMS, wheat straw, yard trimmings, and their mixtures was investigated at different feedstock to effluent ratios. SMS was found to be highly degradable, which resulted in inhibition of SS-AD due to volatile fatty acid (VFA) accumulation and a decrease in pH. This issue was addressed by co-digestion of SMS with either yard trimmings or wheat straw. SS-AD of SMS/yard trimmings achieved a cumulative methane yield of 194 L/kg VS, which was 16 and 2 times higher than that from SMS and yard trimmings, respectively. SS-AD of SMS/wheat straw obtained a cumulative methane yield of 269 L/kg VS, which was 23 times as high as that from SMS and comparable to that from wheat straw.

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1. Introduction

Spent mushroom substrate (SMS) is the substrate left over after mushroom harvesting. For every kilogram (kg) of mushroom produced, about 5 kg of SMS is generated and, traditionally, has been discarded as waste (Williams et al., 2001). The rapid growth in mushroom production worldwide has resulted in large quantities of SMS (about 13.6 million tons per year) (Uzun, 2004; Williams et al., 2001). These massive amounts of waste can cause environmental problems, which has led to increased research to develop technologies for treating SMS.

Anaerobic digestion (AD) is a well-established and widely applied dual-purpose technology for treating various types of organic wastes and producing biogas as an energy carrier (Chen et al., 2008). AD can be operated at liquid phase with total solids

(TS) content less than 15% or at solid phase with TS content higher than 15%. Compared to liquid AD, solid-state AD (SS-AD), has several advantages such as higher volumetric productivity, fewer moving parts, lower energy cost for heating and mixing, and an end product that is easier to handle (Li et al., 2011). AD of SMS has been studied in liquid AD systems, but there have been no reports on SS-AD of SMS for biogas production (Bisaria et al., 1983; Bisaria et al., 1990; Nguyen and Fricke, 2012; Sharma et al., 1989; Shi et al., 2014).

SMS mainly contains lignocellulosic materials, such as sawdust, straw, and cotton seed hulls, which have been decomposed and permeated by mycelium. According to the literature, in liquid AD of SMS, the degradability of mushroom substrate was significantly increased after long-term mushroom cultivation (4–5 months) compared to untreated mushroom substrate (Bisaria et al., 1983; Bisaria et al., 1990; Sharma et al., 1989). However, this high degradability of SMS may negatively impact the stability of SS-AD. High-degradable feedstocks can be rapidly degraded to volatile fatty acids (VFAs) which may easily accumulate in SS-AD systems due

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to the slow methanogenesis process, which consumes the VFAs. Excessive VFAs can cause a drastic drop in pH and further inhibit the methanogenesis process (Li et al., 2011).

One strategy to address this issue might be co-digestion of SMS and materials with low-degradability to obtain a mixture with balanced overall degradability. Feedstocks with high degradability (such as food wastes) have been co-digested with low degradable materials (such as woody biomass and grass) for enhanced performance of SS-AD (Pagés-Díaz et al., 2014; Xu and Li, 2012). Compared to mono-digestion, co-digestion has balanced nutrients and carbon to nitrogen (C/N) ratios, improved buffer capacity, and diluted inhibitors (Astals et al., 2012; Girault et al., 2012).

Yard trimmings, containing mainly wood chips with small amounts of grass and leaves, are one of the major components of municipal solid wastes. About 33.8 million tons of yard trimmings were generated in the U.S. in 2011 (USEPA, 2011). Wheat straw, the above-ground biomass remaining after wheat harvest, was estimated to be around 70.9 million tons in the U.S (Johnson et al., 2006). Individually, yard trimmings and wheat straw have exhibited low degradability due to their lignocellulosic recalcitrance (Cui et al., 2011; Zhao et al., 2014a; Zhao et al., 2014b). Both of these wastes are abundant and have potential for biogas production through co-digestion with SMS.

In this study, SS-AD of SMS was investigated, and co-digestion of SMS with yard trimmings and with wheat straw was tested for enhanced SS-AD performance. Biogas production from the mixture of SMS/yard trimmings and SMS/wheat straw was evaluated and compared to that from each individual feedstock. Degradation of volatile solids (VS), cellulose, and hemicellulose in the feedstocks was also determined.

2. Methods

2.1. Feedstock and inoculum

SMS was provided by a mushroom grower located in Burbank, OH, USA. The original substrate used for mushroom production contained sawdust with a small amount of barley grain. Yard trimmings, which contained tree branches and leaves, were obtained from the Ohio Agricultural Research and Development Center (OARDC) in Wooster, OH, USA. Wheat straw was obtained from a local farm near Wooster, OH, USA. After drying at 40 °C to a moisture content of less than 10%, SMS was processed with a hand blender (Black & Decker, New Britain, CT, USA), while yard trimmings and wheat straw were ground using a hammer mill (Mackisik, Parker Ford, PA, USA) with a 10 mm screen. The processed materials were stored in air tight containers at room temperature until use. Their mixtures, SMS/yard trimmings (1:1 based on VS) and SMS/wheat straw (1:1 based on VS), were also prepared as feedstocks for co-digestion.

Effluent from a mesophilic liquid AD system (Montpelier, OH, USA) fed with dairy manure and food wastes was collected and filtered through a 20-mesh screen. The solid residue of the effluent, which had a TS content of $8.52 \pm 0.06\%$, was kept in air-tight buckets at 4 °C, and acclimated at 37 °C for 5 days before use as inoculum.

2.2. Solid-state anaerobic digestion

SS-AD reactors were set up by mixing feedstock with the inoculum (the solid residue of effluent after filtration) to obtain feedstock to effluent (F/E) ratios of 2, 3, and 4 (based on VS), respectively. Tap water was added, if necessary, to obtain a TS content of 20%. SS-AD trials were carried out in 2-L glass reactors that contained 800 g of mixed feedstock, inoculum, and water. Each reactor was sealed with a rubber stopper and connected to a 5-L

gas bag (CEL Scientific Tedlar gas bag, Santa Fe Springs, CA, USA) via a gas outlet on the stopper. All the reactors were incubated in a walk-in thermostat chamber at 37 °C for 62 days. Duplicate reactors were run at each condition. After AD, the weight of digestate in each reactor was determined, and digestate samples were taken for composition analysis.

2.3. Analytical methods

TS and VS were measured using gravimetric analysis (APHA, 2005). Total carbon (TC) and total nitrogen (TN) were determined using an elemental analyzer (Vario Max CNS, Elementar Americas, Mt. Laurel, NJ, USA) to calculate the C/N ratio. Extractives were measured using the method reported by Sluiter et al. (2005). Cellulose, hemicellulose, and lignin contents in the feedstock and digestate after AD were determined using the NREL method (Sluiter et al., 2008). Briefly, extractive free biomass was hydrolyzed into monomers by a two-step acid hydrolysis, and the concentrations of these monomers were measured using high performance liquid chromatography (HPLC) (Shimadzu LC-20AB, Columbia, MD, USA). Acid-soluble lignin was measured by UV-Vis spectroscopy, and acid-insoluble lignin was determined by gravimetric analysis. The degradation of VS, cellulose, and hemicellulose during SS-AD was defined as the percentage decrease of each component with respect to its initial amount in the reactor, and calculated as:

$$\text{Degradation (\%)} = \frac{W_{\text{final}} \times C_{\text{final}} - W_{\text{initial}} \times C_{\text{initial}}}{W_{\text{initial}} \times C_{\text{initial}}} \times 100 \quad (1)$$

where W_{initial} and W_{final} are initial and final dry weights of the mixture in the reactor before and after SS-AD, respectively. C_{initial} and C_{final} are initial and final contents of a component in the mixture before and after SS-AD, respectively.

Degradability of feedstocks was evaluated using an enzymatic hydrolysis method described by Selig et al. (2008) with slight modifications. Cellulase (Cellic Ctec2, Novozymes, Denmark) and xylase (Cellic Htec2, Novozymes, Denmark) were loaded into a sodium citrate buffer (0.1 M, pH 5.3) at loading ratios of 10 FPU/g DW (dry weight) and 60 IU/g DW, respectively. The suspension was supplemented with 1% (v/v) of sodium azide (20 g/L), and incubated at 50 °C with shaking at 130 rpm for 72 h. The mixture was then filtered with a 0.2 μm nylon membrane filter, and the filtrate was analyzed for glucose and xylose contents using HPLC. The glucose and xylose yields were calculated using the following equations:

$$\text{Glucose yeild (\%)} = \frac{\text{glucose released} \times 0.9}{\text{glucan in the feedstock}} \times 100 \quad (2)$$

$$\text{Xylose yeild (\%)} = \frac{\text{xylose released} \times 0.88}{\text{xylan in the feedstock}} \times 100 \quad (3)$$

The volume of biogas collected in the Tedlar bags was measured using a drum-type gas meter (Ritter, TG 5, Bochum, Germany) and the composition of biogas (CO₂, CH₄, N₂, and O₂) was analyzed by a gas chromatograph (GC) (HP 6890, Agilent Technologies, Wilmington, DE, USA) equipped with an alumina/KCl deactivation column (30 m × 0.53 mm × 10 mm) and a Thermal Conductivity Detector (TCD). Helium was used as a carrier gas at a flow rate of 5.2 mL/min. Temperatures of the injector, column, and detector were set at 150, 40, and 200 °C, respectively.

Samples for pH and alkalinity measurements were prepared by suspending 5 g of sample in 50 ml of deionized (DI) water. The suspensions were analyzed with an auto-titrator (Mettler Toledo, DL22 Food & Beverage Analyzer, Columbus, OH, USA). To measure the VFAs, the digestate was first diluted with DI water (1:1, w/w). Then, the suspension was centrifuged at 8000 rpm for 5 min, and the supernatant was acidified to pH ~3 with 2 N hydrochloric acid.

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