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Improvement of methane production from rice straw with rumen fluid pretreatment: A feasibility study

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ABSTRACT

To overcome the inherent recalcitrance of rice straw during anaerobic digestion process, effective pretreatment is required for promoting methane production. In the present study, a biological pretreatment using rumen fluid was proposed. The rice straw was pretreated with the rumen fluid at 39 °C for 120 h under anaerobic conditions. Various volatile fatty acids, especially acetic acid and propionic acid, were produced by the rumen fluid pretreatment. The methanogenic process was carried out over a 30-day anaerobic digestion. The results indicated that the optimal pretreatment time for anaerobic digestion was 24 h, resulting in a biogas production increase of 66.5%, a methane yield increase of 82.6% and a technical digestion time decrease of 40.0%, compared with the control. At the end of anaerobic digestion, degradation efficiency of total solid and volatile solid was respectively improved by 16.4–33.3% and 14.8–31.7% for rumen fluid pretreatment. The promoted methane production and organic matter degradation could be mainly attributed to the effective hydrolysis of rice straw by the mixed microorganisms in rumen fluid. Methane production could be well explained by modified Gompertz model rather than the first order model, and a higher methane production rate of 29.31 ml/(g_{VS}·d), a rapider hydrolysis rate of 0.09 1/d, and a shorter lag phase of 1.62 d were obtained after 24 h pretreatment. Therefore, the rumen fluid pretreatment is promising for effective production of methane from rice straw and reduction of rumen fluid discharge from slaughterhouse.

1. Introduction

Rice is one of the main esculent cereals in central and southern China, and rice straw is a major by-product of rice production. As one of the biggest agricultural countries, China produces the rice straw of 203 million tons every year (National Bureau of Statistics of China, 2009). However, large quantities of rice straw are often dumped or burned in open environment, which is not a recommended practice in term of environmental and ecological aspects of sustainable development (Chandra et al., 2012a, b). The rice straw is composed mainly of cellulose, hemicellulose and lignin, and can be transformed into renewable energy (biomethane or ethanol) by anaerobic fermentation (Gu et al., 2014), which may alleviate many problems for environment and energy to a certain extent (Song et al., 2013). However, the cellulose, hemicellulose and lignin in lignocellulosic biomass are strongly linked to each other and form complex three-dimensional structures, which resist the accessibility of microorganisms (Malherbe and Cloete, 2002; Monlau et al., 2013). Therefore, suitable pretreatment methods are needed to destroy the structural and compositional barrier of lignocellulosic biomass (Yu et al., 2014). Various pretreatment technologies have been investigated for lignocellulosic biomass to enhance methane yield, such as chemical (Song et al., 2013), mechanical (Chen et al., 2014), thermal (wet oxidation) (Ferreira et al., 2013), biological (Yan et al., 2012) or combinations of them (Bruni et al., 2010). High energy consumption associated with mechanical pretreatment and strong corrosiveness to reactors for chemical pretreatment limit their large-scale application. Compared with these pretreatments, biological pretreatment is environmentally friendly because of its lower energy requirement and milder reaction conditions. Various

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microbial agents, such as white-rot fungi (Zhao et al., 2014), mesophilic lignocellulolytic microbial consortium (BYND-5) (Yan et al., 2012) and thermophilic microbial consortium (MC1) (Yuan et al., 2014) have been applied for the pretreatment of lignocellulosic biomass. These microbial communities can effectively improve the biomass biodegradability and methane yield. Considering the constant supply of these microbial agents and their screening cost, the direct utility of microbial agents might not be economically feasible. Therefore, the microbial agents with high lignocellulose degradation efficiency, low cost and environmentally friendly properties are needed to be explored.

Rumen fluid, including complex microbial population of bacteria, protozoa, fungi and archaea, is formed in the fore-stomach (reticulorumen) of cows and exhibits higher ability and activity to degrade lignocellulosic biomass than other normal anaerobic microorganisms (Yue et al., 2013; Creevey et al., 2014). It has been reported that the cellulose solubilization by rumen microorganisms are significantly faster than that by microbial communities from landfills or anaerobic digesters (O'Sullivan et al., 2006; Song et al., 2005). Hu and Yu (2005) studied anaerobic fermentation of corn stovers with rumen microorganisms as inocula, and found that the volatile solids (VS) was rapidly degraded and a higher production of volatile fatty acids (VFAs) was observed, compared with the conventional acidogenic bacteria derived from sewage (Hu and Yu, 2005). The high lignocellulose degradation rate and effective hydrolytic conversion to VFAs by the rumen fluid make it possible to improve the methane production. Besides, blood and rumen contents are major slaughterhouse wastes, causing high investment and operating costs when they are discharged to sewage treatment plants (Makinde and Sonaiya, 2010; Roy et al., 2013). The use of these natural microbial consortia should be cost-effective, and using the rumen fluid for the lignocellulosic pretreatment is a promising option.

However, this biological agent has been seldom tested for lignocellulosic pretreatment to increase methane production. Baba et al. (2013) reported that the waste paper was pretreated by rumen fluid for 6 h and 24 h, and daily methane yield respectively increased by 2.6 and 2.1 times, compared with that of control. The waste paper has been treated both chemically and thermally to remove lignin during paper-making process, and should be easier to be degraded than other lignocellulosic biomass. However, few researches on rice straw pretreatment by rumen fluid for methane production have been reported so far.

The objective of this work is to investigate the feasibility of biological pretreatment of rice straw by rumen fluid to improve the methane production, and determine the optimal pretreatment time. Studying the kinetics of methane production from feedstocks is important when designing and evaluating anaerobic digesters. First-order kinetic (Zhen et al., 2014) and modified Gompertz models (Lu et al., 2014) are most applied to describe the methane production from lignocellulosic materials. The first-order kinetic model is commonly applied to simulate anaerobic digestion process when the hydrolysis is rate-limiting (Gavala et al., 2003). The Gompertz model is commonly used in the simulation of methane and hydrogen production, and is useful to explain lag time and sigmoidal growth curve (Syaichurrozi, 2013). Therefore, the first-order and Gompertz models were used to assist in the interpretation of conclusions.

2. Materials and methods

2.1. Materials

The rice straw was collected from rice fields around Changsha, Hunan. The rice straw was air-dried at room temperature and chopped to 2–3 cm using a paper knife before stored in a refrigerator at 4 °C. After oven-dried at 45 °C for 24 h, the rice straw was ground to a size of 30-mesh by a grinder (HC-700. Huangcheng, China).

The rumen fluid was taken from the fresh stomach of cattles from a local slaughterhouse in Changsha of China, brought to the laboratory in a sealed bottle. The rumen fluid sample was filtered through four layers of gauze with N_2 protection and stored at 39 °C, since this temperature is close to the body temperature of ruminant animals (ranged from 37.8 to 40 °C) (Feng, 2004). The samples were used in experiments within 5 h of being collected from the fresh stomach. The main characteristics of rice straw and rumen fluid are shown in Table 1. With a high C/N ratio of 64.1, the rice straw is not ideal as the sole feedstock for anaerobic digestion. The rumen fluid presented a high concentration of total nitrogen (TN) and NH_4^+-N, thus could serve as nitrogen source during methanogenic process without addition of extra nitrogen.

The seed sludge for methane production was collected from a continuous biogas plant (Changsha, China) with an organic loading rate at about 4.5 kg VS/(m^3·d), a hydraulic retention time of 25 d and a operating temperature of 32 ± 1 °C. The main raw materials of this biogas plant were swine manure and crop straws. After concentrated, the seed sludge was used as the inocula. The characteristics of seed sludge were: 56.2 g/L TS, 34.8 g/L VS. It was cultured in a thermostatic water bath (HH-8. JOYN, China) at 35 °C for a few days until no biogas production, then used as the seed sludge in subsequent anaerobic digestion.

2.2. Rice straw pretreatment with rumen fluid

The rice straw pretreatment was performed in 250 ml conical flasks. Firstly, the rice straw of 3 g, rumen fluid of 60 ml, and deionized water of 60 ml were thoroughly mixed in flasks without adding any nutrient media. The initial pH was maintained at 7.0 by NaOH and HCl. Then the flasks was purged with N_2 for 5 min to remove O_2 and sealed with a rubber stopper. All the flasks were incubated at 39 °C on a incubator shaker (ZHWY-2012C, Shanghai Zanyu Instrument Co. LTD., China) at 120 r/min for 120 h. The constant temperature of 39 °C and initial pH of 7.0 were used, as these conditions are close to the actual rumen environment. Inside of the actual rumen, the temperature maintained between 37.8 and 40 °C and the pH varied between approximately 6.5 and 7.2 (Feng, 2004). The biogas volume generated during the pretreatment was recorded at a certain time interval by water displacement, and the biogas composition was measured by gas chromatography (SP7820,

Table 1
Properties of rice straw and rumen fluid used in experiments.

Samples	Parameter	Value
Rice straw	Total solid (TS) (%)	90.0 ± 0.3
	Volatile solid (VS) (%)	77.8 ± 1.6
	Total organic carbon (TOC) (%TS)	43.6 ± 2.1
	Total kjeldahl nitrogen (TKN) (%TS)	0.7 ± 0.13
	C/N ratio	64.1 ± 3.7
	Cellulose (%TS)	37.2 ± 0.9
	Hemicellulose (%TS)	26.4 ± 1.7
	Lignin (%TS)	6.9 ± 1.0
Rumen fluid	pH	7.1 ± 0.0
	TS (g/L)	10.1 ± 0.8
	VS (g/L)	6.2 ± 1.3
	TN (mg/L)	412.9 ± 22.3
	NH^{4+}—N (mg/L)	288.1 ± 18.9
	Acetic acid (mg/L)	1131.4 ± 37.1
	Propionic acid (mg/L)	414.3 ± 11.3
	Butyric acid (mg/L)	425.6 ± 23.4
	Valeric acid (mg/L)	55.6 ± 13.6