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Toward the complete utilization of rice straw: Methane fermentation and lignin recovery by a combinational process involving mechanical milling, supporting material and nanofiltration

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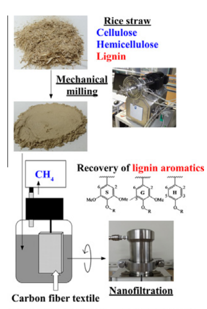
HIGHLIGHTS

- The complete utilization of rice straw was aimed.
- Methane fermentation was performed in a bioreactor containing supporting material.
- Mechanical milling (1.9 MJ/kg-rice straw) increased methane production (3.5 MJ).
- Nanofiltration recovered lignin that was released from the residue.
- 2D NMR showed that lignin aromatic components were recovered.

GRAPHICAL ABSTRACT



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ABSTRACT

Rice straw was mechanically milled using a process consuming 1.9 MJ/kg-biomass, and 10 g/L of unmilled or milled rice straw was used as the carbon source for methane fermentation in a digester containing carbon fiber textile as the supporting material. Milling increased methane production from 226 to 419 mL/L/day at an organic loading rate of 2180 mg-dichromate chemical oxygen demand/L/day, corresponding to 260 mL_{CH4}/g_{VS}. Storage of the fermentation effluent at room temperature decreased the weight of the milled rice straw residue from 3.81 to 1.00 g/L. The supernatant of the effluent was subjected to nanofiltration. The black concentrates deposited on the nanofiltration membranes contained 53.0–57.9% lignin. Solution nuclear magnetic resonance showed that lignin aromatic components such as *p*-hydroxyphenyl (H), guaiacyl (G), and syringyl (S) were retained primarily, and major lignin interunit structures such as the β–O–4-H/G unit were absent. This combinational process will aid the complete utilization of rice straw.

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1. Introduction

The conversion of lignocellulosic biomass to energy sources such as biogas and biofuel is currently receiving attention, both because global energy sources are primarily derived from finite fossil fuels and from the necessity to mitigate greenhouse gas emissions (Lynd et al., 2008; Tuck et al., 2012). Rice straw is an abundant lignocellulosic waste (Binod et al., 2010) and holds promise as a major resource for biogas production. Biogas can be used as a car fuel and for generating heat or electricity (Taherzadeh and Karimi, 2008). Rice straw is mainly composed of cellulose, hemicellulose, and lignin (Binod et al., 2010) and requires pretreatment prior to its conversion into fuel due to its complex structure. Methane production by anaerobic digestion therefore comprises three steps: pretreatment, anaerobic hydrolysis of cellulose and hemicellulose with concomitant methane production, and post-treatment of the liquid effluent fraction (Hendriks and Zeeman, 2009). The transformation of lignin into valuable products is challenging in current biorefineries (Ragauskas et al., 2014), making simultaneous recovery of lignin and biogas desirable.

Lignocellulosic biomass is pretreated to enhance its anaerobic digestion and the efficient conversion of cellulose and hemicellulose into biogas (Hendriks and Zeeman, 2009). Mechanical pretreatment such as milling, grinding, or chipping is environmentally-friendly because chemicals such as acids, bases, or organic solvents are not required (Inoue et al., 2008; Lindner et al., 2015), but these process have the disadvantage of typically being energy intensive (Taherzadeh and Karimi, 2008; Hendriks and Zeeman, 2009; Alvira et al., 2010). Recently, a milling process requiring relatively low energy consumption (0.3–5.4 MJ/kg rice straw) has been exploited (Hideno et al., 2009; Sasaki et al., 2015). In addition, ball milling of a digestate of maize silage/hay/wheat straw provided an increased methane yield (Lindner et al., 2015). These findings suggest that mechanical milling can aid the efficient generation of biogas from the cellulose and hemicellulose in rice straw.

Biogas production has been classified into two processes: wet and dry fermentation. The wet process (with a total solids concentration below 10%) dominates in the agricultural sector (Weiland, 2010). Methane fermentation is typically divided into four phases: hydrolysis, acidogenesis, acetogenesis, and methanogenesis (Ahring, 2003; Weiland, 2010). Different microorganisms are involved in each of these steps due to syntrophic interactions between the microorganisms and their individual environmental requirements (Angelidaki et al., 2003; Stams et al., 2003). The use of carbon fiber textile (CFT) as the supporting material in a continuously stirred tank reactor provided an appropriate environment for microorganisms including methanogenic archaea and consequently enhanced the conversion of rice straw to methane (Sasaki et al., 2010). The current study therefore used CFT in methanogenic bioreactors.

Lignin is a promising source of aromatics and other useful chemicals (Ragauskas et al., 2014) and is composed of several chemically distinct subunits, or monolignols (Weng et al., 2008). Membrane processes, e.g. microfiltration, ultrafiltration and nanofiltration (NF), have been used to recover lignin because of their impressive separation efficiency and low energy consumption (He et al., 2012). For example, a NF membrane process (pore size < 2 nm) (He et al., 2012), was reported to extract lignin from hardwood black liquor (Jönsson et al., 2008). Solution-state two-dimensional (2D) ^{1}H-^{13}C heteronuclear single quantum coherence (HSQC) nuclear magnetic resonance (NMR) spectroscopy has been used to screen the lignin components and polysaccharides in unfractionated plant cell wall materials (Mansfield et al., 2012; Chylla et al., 2013), thus demonstrating the ability of 2D NMR methods to elucidate the lignin components recovered by NF membrane processes.

The aim of this study was to simultaneously produce biogas as methane from rice straw and to recover lignin from the rice straw residues. This was achieved using mechanical milling, CFT as the supporting material, and a membrane process. The rice straw was mechanically milled to increase the organic loading for subsequent methane fermentation. Mechanically-milled rice straw was introduced into a methanogenic bioreactor containing CFT. Following methane fermentation, lignin in the rice straw residue was released into the liquid phase by storing the residue for 30 days; the released lignin was then recovered using an NF membrane process and analyzed by 2D NMR.

2. Materials and methods

2.1. Mechanical milling of rice straw

Rice straw (cv. Nipponbare) was harvested in 2009 in Kansai, Hyogo, Japan. Naturally dried rice straw was shredded into pieces less than 2 mm long using a WB-1 blender (Osaka Chemical Co., Ltd., Osaka, Japan) and used as a control substrate in methane fermentation. The shredded rice straw (150-g aliquots) was mechanically milled in a CMJ01 nano-mech reactor (Techno Eye, Tokyo, Japan) using 1800 g of balls. The samples were pre-milled at 500 rpm for 30 s, then milled for 10 cycles (1 cycle: 1000 rpm for 15 s and 1500 rpm for 45 s) (Sasaki et al., 2015). The energy consumption of the mill was directly measured. The length of milled rice straw was examined by the scanning electron microscopy (JSM-6430F, JEOL Ltd., Tokyo, Japan). The samples were coated with osmium and then analyzed with an accelerating voltage of 5 kV.

2.2. Methanogenic bioreactor with CFT

A synthetic medium using rice straw as the major carbon and energy source (Rs medium) was prepared as follows (g/L): rice straw, 10; KH_2PO_4, 0.1; K_2HPO_4, 0.2; NH_4Cl, 1.0; $NaHCO_3$, 2.0; $MgCl_2 \cdot 6H_2O$, 0.1; $CaCl_2 \cdot 6H_2O$, 0.1; NaCl, 0.6; yeast extract, 1.0. The synthetic medium contained 10 mL of Deutsche Sammlung von Mikroorganismen und Zellkulturen (DSMZ, Braunschweig, Germany) medium 131 trace element solution and 10 mL of DSMZ medium 141 vitamin solution. Two types of rice straw were used: shredded rice straw without milling, and shredded rice straw with 10 cycles of mechanical milling. The dichromate chemical oxygen demand (COD_{Cr}) of the synthetic medium with rice straw was 10,900 mg-COD_{Cr}/L.

Seed sludge was collected from manure compost (black cattle farm, Iwate, Japan). The seed sludge (20% *w*/*v*-water) was inoculated into six glass reactors (250 mL; Duran, Wertheim, Germany), then the sampling port and gas flow line nozzle of the glass reactors were sealed with silicone stoppers (Fig. 1). CFT (75 × 25 × 6 mm) (S-255AH, Osaka Gas Chemicals, Osaka, Japan) was fixed with a steel wire in the reactor. The initial anaerobic conditions were established by replacing the gas phase with nitrogen gas. All bioreactors had a working volume of 250 mL, and microorganisms were cultivated at 55 °C. Bioreactors with the two types of rice straw were operated in duplicate.

To start up the operation, the seed cultures were incubated in the bioreactors at 55 °C to remove any remaining organic materials, then 50 mL of Rs medium was added every 3 days for 12 days by discharging the same volume of seed culture. At the same time, 1.0 N NaOH was added to adjust and maintain the pH above 6.8 throughout the experiment. The bioreactors were operated in semibatch mode as follows: 50 mL of Rs medium was added by discharging the same volume once every 2 days (from day 0 to day 24), then once every day (from day 25 to day 42) (Fig. 2A).