Ionic-liquid pretreatment of cassava residues for the cogeneration of fermentative hydrogen and methane

Jun Cheng [a,*], Jiabei Zhang [a], Richen Lin [a], Jianzhong Liu [a], Li Zhang [b], Kefa Cen [a]

[a] *State Key Laboratory of Clean Energy Utilization, Zhejiang University, Hangzhou 310027, China*
[b] *Shandong Botong New Energy Company, Jining 272000, China*

HIGHLIGHTS

- Cassava residues were pretreated by ionic liquid N-methylmorpholine-N-oxide (NMMO).
- NMMO-pretreated cassava residues generated deep grooves (∼4 μm in width) and pores.
- Cellulose I became cellulose II in NMMO-pretreated cassava residues.
- H_2 yield from NMMO-pretreated cassava residues after enzymolysis was enhanced.
- Overall energy conversion increased to 21.4–27.9% in H_2/CH_4 cogeneration.

ARTICLE INFO

ABSTRACT

An ionic liquid of N-methylmorpholine-N-oxide (NMMO) was used to effectively pretreat cassava residues for the efficient enzymatical hydrolysis and cogeneration of fermentative hydrogen and methane. The reducing sugar yield of enzymolysed cassava residues with NMMO pretreatment improved from 36 to 42 g/100 g cassava residues. Scanning electron microscopy images revealed the formation of deep grooves (∼4 μm wide) and numerous pores in the cassava residues pretreated with NMMO. X-ray diffraction patterns showed that the crystallinity coefficient of NMMO-pretreated cassava residues decreased from 40 to 34. Fourier transform infrared spectra indicated that crystal cellulose I was partially transformed to amorphous cellulose II in the NMMO-pretreated cassava residues. This transformation resulted in a reduced crystallinity index from 0.85 to 0.77. Hydrogen yield from the enzymolysed cassava residues pretreated with NMMO increased from 92.3 to 126 mL/g TVS, and the sequential methane yield correspondingly increased from 79.4 to 101.6 mL/g TVS.

© 2017 Elsevier Ltd. All rights reserved.

1. Introduction

Alternative renewable energy has become increasingly important because of the severe depletion of fossil fuels and environmental resources as a result of the growing demand for energy (Navarro-Díaz et al., 2016). Hydrogen is a type of alternative energy carrier that plays an important role in reducing greenhouse gas emissions (Christopher and Dimitrios, 2012). Lignocellulose exhibits good application potential because it can be used as a raw material for bacteria fermentation to produce hydrogen (Gonzales et al., 2016). In southern China, a large number of cassava starch factories and alcohol manufacturing firms operate, and thus, a considerable amount of cassava residues are generated as by-products. According to the data from the Food and Agriculture Organization of the United Nations, China's cassava yield was close to 4.56 million tons in 2012. If not treated appropriately, waste cassava residues would negatively affect the environment (Lin et al., 2016).

Cassava residues are composed of natural lignocellulose fibres, the structure of which is dense and complex. The main components include cellulose, hemi-cellulose and lignin. They are densely cross-linked, they constitute a complex matrix structure, and they demonstrate strong resistance (Kumar et al., 2015). After hydrolysis, only cellulose and hemi-cellulose can be used by bacteria in the fermentation stage; lignin cannot be used (Yi et al., 2014). Therefore, a key step in the microbial fermentation process is the transformation of the cellulose and hemi-cellulose of raw cassava residues into fermentable sugars (Gonzales et al., 2016). As a result of the complexity of their composition, cellulose, lignin and hemi-cellulose are intertwined, and cassava residues cannot quickly hydrolyse (Antonopoulou et al., 2016). Pretreatment methods

should clearly be adopted to accelerate the hydrolysis of cassava residues. The aim of pretreatment is to destroy the complete physical structure of the residues and reduce the crystallinity and degree of polymerisation. The development of efficient and low-cost pretreatment methods has become crucial.

Various pretreatment methods have been explored to improve the digestibility of lignocellulose and further enhance the succeeding biofuel production (Yi et al., 2014). Existing pretreatment methods mainly include physical methods (mechanical polishing, steam explosion, etc.), chemical methods (ozone decomposition, acid decomposition, alkaline decomposition, etc.), synthetic methods (acid catalytic steam blasting, ammonia fibre blasting, carbon dioxide blasting, etc.), biological methods, and so on (Sindhu et al., 2016). In one study, the methane yield of microalgae after beating treatment increased by 37% in three days (Montingelli et al., 2016). When sunflower stalks were pretreated with NaOH (20 g/100 g TS), the hydrogen yield rose to 155.3 L/kg TS (Antonopoulou et al., 2016). After the addition of 0.5% H_2SO_4 in the steam explosion process, the structure of elephant grass was changed, and the sugar yield showed a sevenfold improvement in comparison with untreated biomass (Kataria et al., 2016).

In sum, pretreatment methods mainly focus on alkali, dilute acid and steam blasting methods. Although the above methods have yielded acceptable results, problems such as high cost, low efficiency, and heavy energy consumption remain. The dilute acid method is large in scale, but its reaction temperature is considerably high. Moreover, the separation of the reactor, residual chemical additives and raw materials is difficult. The method is also harmful to the environment because the by-products (furan, furfural, etc.) affect the metabolism of bacteria. Alkaline pretreatment can break down hemicellulose, resulting in the loss of effective components. Moreover, alkali waste liquid is difficult to handle. Steam explosion pretreatment generates a low amount of by-products, but the sugar yield of biomass is low. The method requires high pressure and large-scale application, which is known to be difficult.

Currently, ionic liquids (ILs) are widely used in lignocellulose pretreatment technology (Swatloski et al., 2013). Compared with physical and other chemical pretreatments, IL pretreatment is environmentally friendly. Specifically, ILs have lower boiling points, high thermal stabilities, good recyclability and the potential to improve the thermal properties of lignocellulosic biomass (Ullah et al., 2015). In most previous studies, 1-alkyl-3-methylimidazolium chloride was used to dissolve cellulose in solid form at room temperature. The cellulose showed high viscosity after melting and thus hampered the pretreatment process. Moreover, ILs with chloride ions are corrosive and toxic (Poornejad et al., 2013). In recent years, N-methylmorpholine-N-oxide (NMMO) has emerged as a promising solvent because its use in pretreating biomass results in improved sugar yield and fermentation (Cai et al., 2016). In addition, NMMO is miscible with water, and the functional group N-O possesses strong polarity. It can destroy the lignocellulose hydrogen bonding network and form a new hydrogen bond with fibres, thus reducing crystallinity. The lignocellulosic biomass regenerated from NMMO exhibits a porous texture, an amorphous nature, and a low crystallinity index; these characteristics facilitate the saccharification of biomass (Yi et al., 2014). NMMO is mainly used to produce cellulose membrane and ferment lignocellulose for alcohol production, but its application in fermentative hydrogen production and combined methane production is rare.

In the present study, NMMO was adopted to pretreat cassava residues and thereby promote enzymatical hydrolysis and effectively improve the cogeneration of fermentative hydrogen and methane. Scanning electron microscopy (SEM), X-ray diffraction (XRD), and Fourier transform infrared (FTIR) were used to analyse the surface morphology, crystallinity and functional groups of NMMO-pretreated cassava residues. The hydrogen and methane yields from the enzymolysed cassava residues with NMMO pretreatment increased from 92.3 mL/g TVS to 126 mL/g TVS and from 79.4 mL/g TVS to 101.6 mL/g TVS, respectively.

2. Materials and methods

2.1. Materials

The lignocellulosic substrates used in this work were cassava residues obtained from a cassava processing plant in Guangxi Province, China. Prior to treatment, the cassava residues were oven dried, milled to approximately 0.02 mm particle size, and stored at room temperature before being used in the subsequent experiments.

The microorganisms used were mixed hydrogen-producing bacteria (HPB) and methane-producing bacteria (MPB), which were obtained from a biogas plant in Zhejiang Province, China. The enrichment and isolation of the bacteria were described in previous studies (Xie et al., 2008).

2.2. NMMO pretreatment

An analytically pure NMMO powder purchased from Aladdin was used in the experiments. A solution of 85% (w/w) NMMO was prepared by mixing the powder with deionised water. Then, 5 g cassava residues were soaked in 50 g 85% NMMO solution in round-bottom flasks, which were then placed in an oil bath at 110 °C for 2 h and then stirred continuously throughout the pretreatment. The pretreatment was stopped with the addition of 200 mL boiled deionised water to the suspension, which was stirred for 10 min. The regenerated cassava residues were obtained from the suspension via centrifugation at 6000 r/min for 10 min. Sediments were washed with deionised water for at least three times until a clear filtrate was obtained, and the supernatant liquid was collected to recycle NMMO. The regenerated cassava residues were oven dried until a constant weight was reached. The residues were then stored at room temperature for further experiments.

2.3. Enzymolysis process

Exactly 5 g regenerated cassava residues and raw cassava residues were placed in 250 mL conical flasks, which were added with 100 mL deionised water. The pH of the cassava residue solution was adjusted to 4.5 with NaOH solution. Cellulase was added to the solution at 5 wt% of the cassava residues. After sealing, the conical flasks were placed in an electric-heated thermostatic water bath for 72 h at 45 °C.

2.4. Fermentation methods

2.4.1. Dark hydrogen fermentation

The fermentation assays were carried out in 330 mL glass bottles. Each bottle contained 100 mL enzymolysis liquid that comprised 4 g VS (volatile solids) of untreated or pretreated cassava residues and 0.5 g yeast extract. The initial pH was adjusted to 6.0 ± 0.1 with 6 M HCl and 6 M NaOH. The bottles were then inoculated with 50 mL HPB. The final working volume was adjusted to 250 mL by adding deionised water. The fermentation bottles were sealed with rubber plugs. Nitrogen gas was pumped into the fermentation bottles for 10 min until anaerobic conditions were reached. In the end, the fermentation bottles were placed in an electric-heated thermostatic water bath at 35 °C to produce hydrogen in the following experiments. Gas samples were withdrawn