Optimized Alkaline Pretreatment Technology of Rice Straw for Ethanol Production

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Abstract. As a renewable energy, ethanol from lignocellulosic biomass has developed technologically in many ways. Effective pretreatment can enhance the enzymatic saccharification of cellulose by removing lignin and hemicellulose from raw material. Different alkaline pretreatment technologies were studied to find more effective and economical pretreatment of rice straw for ethanol production. Results showed that heated ammonia pretreatment was more effectively to remove lignin than hemicellulose. Combination of microwaves and heated ammonia enhanced the removal of lignin and enzymatic saccharification. Besides of lignin and hemicellulose, lime pretreatment removed cellulose as well, that decreased the enzymatic saccharification. Take enzymatic saccharification and weight loss into concern, 2% NaOH pretreatment at 100℃ is the most effective alkaline pretreatment technology of rice straw for ethanol production..

Introduction

Ethanol is a renewable, bio-based oxygenated fuel[1]. Developing ethanol as fuel will require developing lignocellulosic biomass as a feedstock because of its abundance and low cost[2,3]. However, the transformation from cellulose of straw to ethanol by microbes is rather complicated[4,5]. Since the protection of hemicellulose and lignin, the rate of enzymatic hydrolysis of cellulose is very low when cellulose is enzymatic hydrolyzed directly [6,7]. Therefore, it is necessary to pretreat the cellulose before hydrolysis by physical, chemical or biological technology[8,9]. The research of effective and low-cost pretreatment technology has been always attractive for years [10]. Acids were adopted in pretreatment popularly [11,12], however, it brought high load to the environment because it is rather difficult to process naturally. Furthermore, acid pretreatment eroded the facilities and brought undesirable by-products[13]. At the same time, alkaline materials attracted more attention recent years, for its lower cost and less environmental disturbances[14,15].

Materials and Methods

Few economical alkaline chemicals, such as ammonia, lime and NaOH, were adopted in varied dosages to pretreat the rice straw in this research. Then the removal rates of lignin and hemicellulose of raw materials, the weight loss of raw materials and the enzymatic saccharification of residue of different pretreatment technologies were analyzed. Finally, the alkaline pretreatment technology of rice straw for ethanol production was optimized.

Materials. Rice straw from suburbs of Chengdu city was smashed and 20 mesh sieved, then was air-dried to constant weight at 60˚C[16].

Buffer solutions included neutral detergent(3% Sodium lauryl sulfate), 2mol/L HCl, 72%H2SO4 and 0.05mol/L Citric acid buffer solutions (pH4.8).

Pretreatment Methods. Heated ammonia pretreatment was operated in following instructions. According to the solid liquid ratio as 1:10, the rice straw powders were mixed with ammonia of concentration of 5%, 10% and 25% in reaction kettles respectively. Mixtures were heated at 100˚C for 2 hours and then leached. The residues were washed to neutral by water, and then dried and weighed. Combined pretreatment of heated ammonia and microwaves was operated in following instructions. According to the solid liquid ratio as 1:10, the rice straw powder was mixed with ammonia of concentration of 10% in reaction kettles. Mixtures was heated at 100˚C for 2 hours in the microwave environment (28 kHz/300W), then leached. The residue was washed to neutral by water, and then dried and weighed. NaOH pretreatment was operated in following instructions. According to the solid liquid ratio as 1:10, the rice straw powders were mixed with ammonia of concentration of 1%, 2% and 5% respectively in reaction kettles. Mixtures are heated at 100˚C for 2 hours and then leached. The residues were washed to neutral by water, and then dried and weighed. Lime pretreatment was operated in following instructions.1g rice straw powders and 10mL water were mixed with 0.05g, 0.1g, 0.2g lime respectively in reaction kettles. Mixtures are heated at 100˚C for 2 hours and then leached. The residues were washed to neutral by water, and then dried and weighed.

Enzyme assays. Carboxymethyl cellulase (CMCase) (20FPU/g) from and cellobiase from Aspergillus niger (10CBU/g) were assayed in reaction mixture (50 ml) with 80g/L substrate concentration and pH 5.0. After 48 h incubation at 50˚C, the reducing sugar liberated in the reaction mixture was measured by the dinitrosalicylic acid (DNS) method [17].

Results and Discussion

Removal rate of main components and weight loss after pretreatment. Effects of different pretreatment technologies on rice straw were evaluated. 2% NaOH changed the surface and inside structure of rice straw in the most remarkable way. The inside structure of straw was swelled fully, and the porosity of surface grew drastically. Thus, the availability of contact of cellulase and cellulose was increased largely (Figure.1 d)[18,19]. The straw pretreated by combined technology of ammonia and microwaves had more porous surface than that pretreated by ammonia only (Figure.1 a ,c). That indicates microwave enhance the effect of pretreatment. Compared with other alkalis, the lime had less noticeable effect on the surface and inside structure of rice straw.

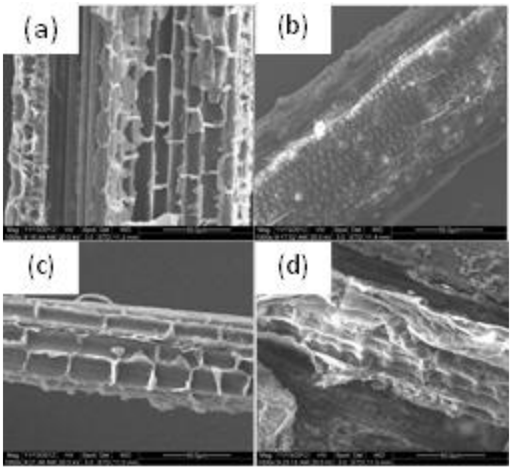

Figure.1 Electron micrographs of residues after different pretreatments (a) Combined pretreatment of heated ammonia and microwaves (100˚C,10%)；(b) Lime pretreatment (100˚C, 0.2g/g)；(c) heated ammonia pretreatmen(100˚C,10%)；(d)NaOH pretreatment(100˚C, 2%)

Heated ammonia pretreatment removed lignin effectively while left most hemicellulose and cellulose. Removal rates of lignin were 62.1% and 44.9%, and removed rates of hemicellulose were 29.9% and 23.3% when straws were pretreated by heated 25% and 10% ammonia respectively. Furthermore, microwaves enhanced the removal of lignin and hemicellulose because that combined pretreatment of heated ammonia and microwaves had more 2.3% lignin removal and 5.5% hemicellulose removal additionally, compared with heated ammonia only. The weight losses were 27.3% and 24.6% when raw materials were pretreated by 25% and 10% heated ammonia respectively, while the combined pretreatment had a slightly higher weight loss as 25.1%.

NaOH pretreatment removed both lignin and hemicellulose powerfully, while the loss of cellulose is rather high. The removal rates of lignin and hemicellulose increased when dose is raised. So did the weight loss of raw materials. 5% NaOH pretreatment removed 91.5% lignin, 74.9% hemicellulose besides of 7.5% cellulose, however, weight loss achieved 49.6%. 2% and 1% NaOH pretreatments had 3.5% and 7.2% less removal rate of lignin than 5% NaOH, 13.3% and 29.3% less removal rate of hemicellulose than 5% NaOH. But the weight losses of 2% and 1% NaOH pretreatments were also less than 5% NaOH as 3.6% and 9.5%.

Lime pretreatment removed more lignin and hemicellulose when dose was increasing. But the loss of cellulose was raised evidently during the process. The removal rate of lignin and hemicellulose were 44.9% and 49.7% of 0.2g/g lime pretreatment, which was the best dosage in this research, while 14.5% cellulose was decomposed into solution and weight loss reached 33.8％.

Analyzing and comparing different pretreatment technologies, it was found that NaOH pretreatment can remove more lignin and hemicellulose and keep more cellulose. Although the weight loss is rather high, it also related with the removal of lignin and hemicellulose closely. Non-specific binding of lignin and cellulase hinders the hydrolysis of cellulose, while hemicellulose decreases the accessibility of cellulase [20]. NaOH pretreatment with appropriate concentration can achieve not only rather ideal removal of lignin and hemicellulose but acceptable weight loss of raw material.

Analysis of enzymatic saccharification of different residues. 48-hours enzymatic saccharification of residues from different pretreatments technologies indicated that different alkalis and their doses impact on the enzymatic saccharification rate greatly. Residues from NaOH pretreatment contributed to much better saccharification effect (Figure.2) than those from ammonia and lime. 1%, 2% and 5% NaOH pretreatment yielded 88.6%, 95.3％and 96.6％saccharification rates at last. Heated ammonia pretreatment did not work as well as NaOH. 5%, 10% and 25% heated ammonia pretreatment yielded 48.7%, 70.1％and 72.5％ saccharification rates eventually. The combination of microwaves and heated ammonia increased additional 1.8% saccharification rate compared with 10% heated ammonia pretreatment. Residues from lime pretreatment had their saccharification rates varied in a broad range, from 39.3% to 73.8%. Although the largest dose of lime seemed promoting the saccharification of residue , it also took higher environmental risk[11].

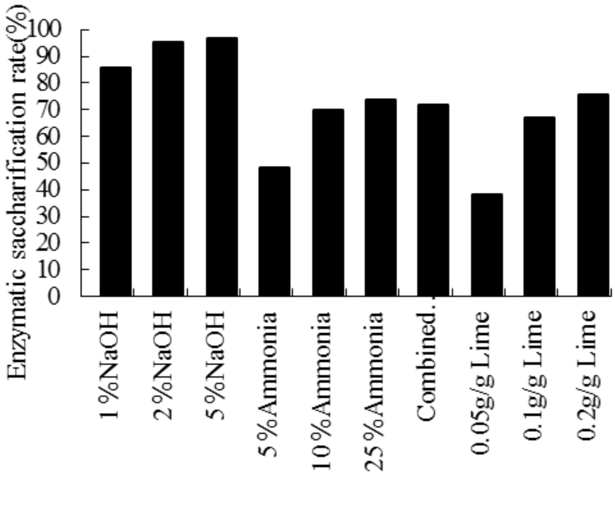

Fig.2 Enzymatic saccharification rates of residues from different pretreatment

There was a positive relationship between enzymatic saccharification rate and removal rate of lignin (Table 1and Figure 2). That probably results from that lignin blocks the sufficient contact of cellulase with cellulose substrate and then decreases the enzymatic effect [21,22]. Moreover, the efficient pretreatment technologies increased the surface porosity and water binding capacity of the raw materials by swelling that raised the efficiency of enzymatic saccharification too[23,24]

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Table 1 Removal rates and weight loss of different pretreatment technologies

Pretreatment technology	Removal rate(%)			Weight loss (%)
	Cellulose	Hemicellulose	Lignin	
25% ammonia	4.0	29.9	62.1	27.3
10% ammonia	2.1	23.3	44.9	24.6
10% ammonia +Microwaves	1.8	28.8	47.2	25.1
5% NaOH	7.5	74.9	91.5	49.6
2% NaOH	5.8	61.6	88.0	46.0
1% NaOH	3.5	45.6	84.3	40.1
0.2g/g lime	14.5	49.7	44.9	33.8

Conclusion

Heated ammonia pretreatment was more effectively to remove lignin than hemicellulose. Because of remaining of hemicellulose, heated ammonia lost less weight of raw materials. Combination of microwaves and heated ammonia enhanced the removal of lignin and also raised enzymatic saccharification rate.

Besides of lignin and hemicellulose, lime pretreatment removed cellulose as well, that decreased the enzymatic saccharification finally.

NaOH removed lignin and hemicellulose in the most effective way. Furthermore, because of the loss of most lignin and hemicellulose, the enzymatic saccharification was enhanced as a result.. However, higher dose of NaOH brought a rather high weight loss of raw material. It is necessary to balance the removal rate and weight loss in this pretreatment.

Take enzymatic saccharification and weight loss into concern, 2% NaOH pretreatment at 100˚C is the most effective alkaline pretreatment technology of rice straw for ethanol production.

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