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# **Comparative Studies on Thermochemical Characterization of Corn** Stover Pretreated by White-Rot and Brown-Rot Fungi

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ABSTRACT: The effects of white-rot and brown-rot fungal pretreatment on the chemical composition and thermochemical conversion of corn stover were investigated. Fungus-pretreated corn stover was analyzed by Fourier transform infrared spectroscopy and X-ray diffraction analysis to characterize the changes in chemical composition. Differences in thermochemical conversion of corn stover after fungal pretreatment were investigated using thermogravimetric and pyrolysis analysis. The results indicated that the white-rot fungus Irpex lacteus CD2 has great lignin-degrading ability, whereas the brown-rot fungus Fomitopsis sp. IMER2 preferentially degrades the amorphous regions of the cellulose. The biopretreatment favors thermal decomposition of corn stover. The weight loss of IMER2-treated acid detergent fiber became greater, and the oil yield increased from 32.7 to 50.8%. After CD2 biopretreatment, 58% weight loss of acid detergent lignin was achieved and the oil yield increased from 16.8 to 26.8%.

KEYWORDS: biopretreatment, white-rot fungi, brown-rot fungi, corn stover, FTIR, XRD, thermogravimetric analysis, pyrolysis analysis

## INTRODUCTION

The depletion of fossil fuels and climate changes are regarded as important factors driving researchers to develop sustainable and clean energy sources.<sup>1</sup> Biomass resources including agricultural residues, forestry residues, and energy crops are the most promising energy sources that can be used to produce biofuels and chemicals, which could be logical choices to replace fossil fuels and have great benefit to the whole environment.<sup>2</sup> Pyrolysis is a potential thermochemical conversion route, converting biomass to energy-dense biofuels as well as chemical feedstocks.<sup>3</sup>

Reactor designs and thermal decomposition mechanisms on the pyrolysis of biomass have been extensively studied.<sup>4,5</sup> In contrast, only a limited number of studies have focused on the pretreatment before biomass pyrolysis to promote thermochemical reactions and upgrade oil contents.<sup>3</sup> The influence of different pretreatment processes used for removing the inorganic compounds of sugar cane bagasse on pyrolysis products was reported.<sup>6</sup> Phosphoric acid pretreatment prior to fast pyrolysis had been proved to be a promising method for obtaining 1,6anhydrosaccharides in high yields.<sup>7</sup> More recently, the effect of chemical pretreatments using NaOH,  $H_2O_2$ , and  $Ca(OH)_2$  on empty palm fruit bunches (EPFB) to degrade EPFB lignin before pyrolysis was investigated.<sup>8</sup> It was proposed that pretreatments may modify the chemical structure of biomass or selectively remove lignocellulosic components, thereby improving the thermochemical conversion of biomass during pyrolysis.9,10 However, the usage of acid and alkali in chemical pretreatments might cause environment consequences. Thus, effective, lowcost, and green biopretreatment under mild conditions and low energy consumption has manifest superiority over the aforementioned chemical means.<sup>11,12</sup>

Previous studies showed the potential application of fungi to break down lignocellulosic biomass.<sup>13</sup> Fungal pretreatment has widely been used in the production of bioethanol as well as in the process of wood pulping. Pretreatment with white-rot and

brown-rot fungi has different deconstruction characteristics on biomass chemical composition, which may cause different changes in the production of biofuels. White-rot fungi preferentially decay lignin, thus opening the structure of lignocellulose and increasing access to other biomass components such as cellulose and hemicellulose, which subsequently can be utilized to produce biofuels.<sup>14</sup> Irpex lacteus, as a well-known promising white-rot fungus, has been used in biopretreatment for biomass enzymatic hydrolysis.<sup>11</sup> Brown-rot fungi are often considered as organisms that predominantly decay polysaccharide, whereas lignin decay is limited.<sup>12</sup> The metabolic mechanism of brown-rot fungus *Fomitopsis* sp.<sup>15</sup> and its application for biological treatment of black liquor<sup>16</sup> have been investigated.

In this work, the effects of biopretreatment using white-rot fungus I. lacteus CD2 and brown-rot fungus Fomitopsis sp. IMER2 on chemical composition and thermochemical conversion of corn stover were investigated. Furthermore, changes in thermogravimetric behavior and pyrolysis product distribution of corn stover varying depending on the different fungal pretreatment were studied for the first time. The overall experimental flowchart is summarized in Figure 1.

## MATERIALS AND METHODS

Fungal Strains and Biopretreatment of Corn Stover. The fungi used for pretreatment of corn stover were isolated from Shennongjia Nature Reserve (Hubei, China). The fungal strains were identified by rDNA internal transcribed spacer (ITS) sequence analysis and were maintained on potato dextrose agar slants at 4 °C. Three to five disks cut from the margin of active fungal cultures on potato dextrose agar plates were inoculated into 250 mL Erlenmeyer flasks with 100 mL

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of potato dextrose broth (PDB) medium (pH 5.5) and incubated at 28 °C and 150 rpm for 5-7 days on a reciprocal shaker. The mycelia in the flasks were gently homogenized, and 10 mL was used to inoculate fresh PDB medium each for 3 days to obtain the secondary seed culture under the same condition.

The corn stover used for the present study was obtained from Henan province in China. The biopretreatment of biomass was carried out in 250 mL flasks containing 10 g (dry mass) of pulverized corn stover (grain diameter = 0.3-0.45 mm) and 15 mL of distilled water. After autoclaving (150 kPa, 45 min), 10 mL of homogenized secondary seed culture was inoculated. Uninoculated control samples were incubated under the identical conditions. After 30 days of incubation at 28 °C and constant humidity, the pretreated corn stover was dried (in an aerated oven at 60 °C), milled, and extracted with a benzene/ethanol mixture (80:20, v/v) in a Soxhlet apparatus.

Isolation and Analysis of Corn Stover Lignocellulose Components. Neutral detergent fiber (NDF), acid detergent fiber (ADF), and acid detergent lignin (ADL) from biopretreated and untreated corn stover were extracted with neutral detergent reagent, acid detergent reagent, and 72%  $H_2SO_4$ .<sup>17–19</sup> Hemicellulose, cellulose, and Klason lignin contents were calculated by the weight differences between ADF and NDF contents, ADF and ADL contents, and ADL and ash contents, respectively.

**Fourier Transform Infrared Spectroscopy.** FTIR spectra were recorded with a NEXUS 670 spectrometer (Thermo Nicolet Corp., Madison, WI). KBr pellets for FTIR spectroscopy were prepared using a Perkin-Elmer pellet die (2 mg of corn stover sample in 40 mg of KBr).<sup>20</sup> Peak height and area were measured using OMNIC software version 1.2a (Nicolet Instrument Corp.).

**X-ray Diffraction Analysis (XRD).** The crystallinity index (CrI) of corn stover was determined by XRD (X'Pert PRO, PANalytical B.V., The Netherlands) by the Analytical and Testing Center, Huazhong University of Science and Technology. The operating conditions of the X-ray diffractometer were 40 kV and 40 mA. Samples were scanned at 1 s/0.02° from  $2\theta = 10^{\circ}$  to  $2\theta = 40^{\circ}$  with a step size of 0.02°. The CrI was defined by eq 1 using the intensities of the crystalline region at  $2\theta = 22.6^{\circ}$  and the amorphous region at  $2\theta = 18.7^{\circ}$ . The crystalline portion of corn stover was determined by the ratio of  $I_{002}$  to the sum of  $I_{002}$  and  $I_{\rm am}^{21}$ 

$$CrI = (I_{002} - I_{am})/I_{002} \times 100$$
(1)

where  $I_{002}$  and  $I_{am}$  are the intensity of diffraction at  $2\theta = 22.6^{\circ}$  and at  $2\theta = 18.7^{\circ}$ , respectively.

Thermogravimetric Analysis. Thermogravimetric experiments were performed by using a thermobalance (PerkinElmer, Diamond, Shanghai, China) at the Analytical and Testing Center, Huazhong University of Science and Technology. The temperature of the furnace and weight were calibrated according to the manufacturer's recommendation. Temperature calibration was performed by measuring curie points of indium, tin, and gold. Prior to thermogravimetric experiments, samples were ground to small chips until the particle size was between 0.15 and 0.2 mm. Initial sample masses of 5 mg were placed in the pan of the thermogravimetric analyzer microbalance. Such sample amount was enough to fill the pan because of the low density of the ground samples. Nitrogen gas was used as carrier gas. Experiments were carried out on a thermobalance at a linear heating rate of 10 °C/min, with the temperature range from 25 to 1000 °C, at a steady nitrogen flow of 100 mL/min. The thermogravimetric experiments were performed three times, and the average results were used for plotting TG-DTG curves.

**Pyrolysis Analysis.** The pyrolysis experiments of corn stover samples were performed by using a fixed bed reactor by Key Laboratory of Green Chemistry and Technology, Sichuan University. Samples were heated from ambient temperature to the final temperature at the heating rate of 10 °C/min and maintained for 1 h. The final temperature was



Figure 1. Flowchart of the overall experiment and analyses.

controlled by Temperature Control Specialists (SKW-1000) at 500  $^\circ$ C. The volatile products were swept out by nitrogen at a flow rate of 66.7 mL/min, and the pyrolysis oil was collected in an ice trap. After pyrolysis, the solid char was removed and weighed. The amount of gas produced was calculated from the material balance.

**Statistical Analysis.** The experimental results were validated by statistical analysis using Microsoft Excel and Origin 8.0 software. Mean values and their standard errors were found. Significant differences between mean values in control and test groups were tested using Student's *t* test (significance levels = 0.05). The corn stover was milled, blended, and replicable tested of randomly drawing sample. The biomass components analysis, XRD analysis, and thermogravimetric experiments were performed three times. Mean values and their standard errors were found. The pyrolysis of biomass was at least duplicated, and the error in the yields of liquid, gas, and char was within 0.5%.

## RESULTS AND DISCUSSION

Components Analysis. The degrees of the changes in chemical components of biopretreated corn stover were found to be different. As shown in Table 1, significant decreases in the hemicellulose content occurred after biopretreatment (p < 0.05). However, the mass reduction of hemicellulose in I. lacteus CD2 pretreated corn stover was almost the same as that of the Fomitopsis sp. IMER2 treated sample (p > 0.05). White-rot fungus I. lacteus CD2 significantly degraded lignin (p < 0.05), whereas the change of lignin content in brown-rot fungus *Fomitopsis* sp. IMER2 treated corn stover was slight (p > 0.05). After 30 days of biopretreatment, the cellulose content in Fomitopsis sp. IMER2 treated corn stover significantly degraded from 43.6 to 37.6%. At the same time, pretreatment with I. lacteus CD2 led to an apparent increase of cellulose percentage in corn stover, which was attributed to the removal of hemicellulose and lignin component during degradation. The results showed that besides the equally significant degradation of hemicellulose, the white-rot fungus I. lacteus CD2 has great lignin-degrading ability, whereas the brown-rot fungus Fomitopsis sp. IMER2 preferentially degrades cellulose. Previous studies of biomass degraded by fungi have shown that significant changes in chemical properties occur before measurable weight loss is detected. This highlights

### Table 1. Component Analysis of Untreated and Biopretreated Corn Stover

		compositional analysis wt % (on dry basis)			
component	corn stover	corn stover pretreated by <i>I. lacteus</i> CD2	corn stover pretreated by <i>Fomitopsis</i> sp. IMER2		
cellulose hemicellulose lignin ash	$\begin{array}{c} 43.60 \pm 0.21 \\ 24.00 \pm 0.56 \\ 11.07 \pm 0.94 \\ 2.00 \pm 0.72 \end{array}$	$\begin{array}{c} 47.40 \pm 0.72 \\ 18.70 \pm 0.71 \\ 8.59 \pm 0.69 \\ 1.40 \pm 0.20 \end{array}$	$37.60 \pm 1.25$ $19.10 \pm 1.11$ $11.60 \pm 0.20$ $2.33 \pm 0.31$		

the necessity of other methods for characterizing the biodegradation patterns of corn stover by different fungi.<sup>22</sup>

FTIR Spectroscopy Analysis of Fungal Pretreated Corn Stover. FTIR is a useful technique for describing changes in biomass chemistry following fungal degradation.<sup>23</sup> It has also been used for evaluation of the relative changes in lignin/ carbohydrate composition of wood decayed by fungi.<sup>20</sup> The ratios of peak heights and area values for the lignin-associated band at 1510 cm<sup>-1</sup> to carbohydrate reference peaks at 1373, 1161, and 898  $\text{cm}^{-1}$  were used for providing relative changes in the composition of the structural components.<sup>20</sup> The assignment of bands to structural components according to references is provided as follow: 1373 cm<sup>-1</sup> for C–H deformation in cellulose and hemicellulose, 1161 cm<sup>-1</sup> for C–O–C vibration in cellulose and hemicellulose, and 898 cm<sup>-1</sup> for C–H deformation in cellulose. As shown in Table 2, in corn stover pretreated by I. lacteus CD2 there was a progressive decrease in lignin content relative to carbohydrate, as shown by the decrease of peak heights and areas of lignin-associated bands (at 1510 cm<sup>-1</sup>) relative to corresponding carbohydrate bands (at 1373, 1161, and 898 cm<sup>-1</sup>). It indicated that the fungus *I. lacteus* CD2 has great lignin-degrading ability. In contrast, corn stover pretreated by Fomitopsis sp. IMER2 showed a different degradation pattern with a growth in peak heights associated with lignin relative to carbohydrates, which indicated that the fungus Fomitopsis sp. IMER2 degraded lignin to a limited extent but degraded cellulose and hemicellulose to a significant extent.

Crystallinity Analysis of Fungus-Pretreated Corn Stover. Biomass cellulose is composed of linear chains of  $\beta$ -1,4 linked Danhydroglucopyranose. These chains are arranged into elementary fibrils, consisting of 60-70% crystalline cellulose and 30-40% amorphous cellulose, and surrounded by a hemicellulose and lignin matrix.<sup>22</sup> XRD has been used for decades as a rapid, nondestructive method for observing the crystalline portion of biomass. Crystallinity can be determined by comparing the sharp crystalline diffraction signals to the broad signals created by the amorphous material.<sup>22</sup> As shown in Table 3, both white-rot fungus I. lacteus CD2 and brown-rot fungus Fomitopsis sp. IMER2 have increased overall crystallinity and the crystalline portion of the corn stover. The crystalline degree of corn stover after pretreatment significant increased from 33.22 to 37.20% (*p* < 0.05) for *I. lacteus* CD2- treated sample, and 46.06% (*p* < 0.05) for Fomitopsis sp. IMER2 treated sample. At the same time, the intensity of the crystalline region was greater in the pretreated stover than in the untreated corn stover. The crystalline portion of untreated corn stover was determined to be 59.96%, whereas crystalline portions of the samples pretreated with I. lacteus CD2

Table 2. Ratios of the Intensity of the Lignin-Associated Band
with Carbohydrate Bands for Untreated and Biopretreated
Corn Stover

	relative intensities <sup>a</sup> of aromatic skeletal vibration against typical bands for carbohydrates					
sample	$I_{1510}/I_{1373}$	$I_{1510}/I_{1161}$	$I_{1510}/I_{898}$			
corn stover	0.634 (0.727)	0.329 (0.364)	0.859 (0.696)			
corn stover pretreated by	0.546	0.288	0.791			
I. lacteus CD2	(0.588)	(0.303)	(0.476)			
corn stover pretreated by	1.421	0.840	1.902			
Fomitopsis sp. IMER2	(1.333)	(0.828)	(1.412)			
Relative intensities were c	alculated usi	ng areas (not :	in parentheses)			

and peak heights (in parentheses).

 
 Table 3. Crystalline Parameters of Untreated and Biopretreated Corn Stover

parameter	corn stover	corn stover pretreated by <i>I. lacteus</i> CD2	corn stover pretreated by Fomitopsis sp. IMER2
crystallinity (%)	$33.22\pm0.05$	$37.20\pm0.38$	$46.06 \pm 1.23$
crystalline portion (%)	$59.96\pm0.02$	$61.42\pm0.14$	$64.96\pm0.52$

and *Fomitopsis* sp. IMER2 were 61.42% (p < 0.05) and about 64.96% (p < 0.05), respectively. Similar results were observed in other biopretreated lignocelluloses.<sup>21,22</sup> This may be due to the fungi, especially brown-rot fungus *Fomitopsis* sp. IMER2, preferentially degrading the amorphous regions of the cellulose microfibrils in biomass during 30 days of pretreatment.<sup>22,24</sup>

Thermogravimetric Analysis of Fungal Pretreated Corn Stover and Lignocellulosic Fractions. Studying the kinetics of biomass pyrolysis is important to a better understanding of the underlying processes and to provide useful information for the design and scaling-up of pyrolysis reactors.<sup>25</sup> The comparison of the thermogravimetric characteristics, both TG (in wt %) and DTG (in  $\sqrt[6]{o}/^{\circ}C$ ) curves, of the untreated corn stover and the biopretreated samples is shown in Figure.2. The main DTG peak is dominated by the decomposition of cellulose, whereas the shoulder at around 275 °C can be attributed mainly to hemicellulose decomposition.<sup>3</sup> The lignin decomposes at a lower rate in a wide temperature range from 200 to 600 °C. The characteristic parameters of the thermogravimetric curves of the corn stover samples are summarized in Table 4 (entries 1-3). It can be observed that the weight loss of biopretreated corn stover samples and the maximum rate of decomposition (DTG<sub>max</sub>) have similar values, and they are all considerably higher than those for the original sample. Results indicated that the biopretreatment favors the thermal decomposition of corn stover. This can be explained by the fact that the component complexity of biomass was simplified and the structural integrity was destroyed by fungi.<sup>26</sup> As mentioned before, the white-rot fungus I. lacteus CD2 significantly degraded lignin, whereas the brown-rot fungus Fomitopsis sp. IMER2 preferentially degraded the amorphous regions of cellulose. In this view, the following study investigated

Weight(%)



600

800

Figure 2. TG and DTG curves of biopretreated corn stover at 10 °C/min in nitrogen atmosphere (*I. lacteus* CD2, corn stover pretreated by white-rot fungus *I. lacteus* CD2 for 30 days, and *Fomitopsis* sp. IMER2, corn stover pretreated by brown-rot fungus *Fomitopsis* sp. IMER2 for 30 days).

Temperature(℃)

400

200

Table 4.	Thermogravimetric Parame	ters" of Untreated	and Biopretreated	Corn Stover,	ADF, and AD	L Samples in N	itrogen
Atmosph	ere, at a Heating Rate of 10	°C/min					

entry	sample	$DTG_{max}$ (%/°C)	$T_{\text{peak}}$ (°C)	char (%)
1	corn stover	0.735	322	33.32
2	corn stover pretreated by I. lacteus CD2	0.996	325	23.41
3	corn stover pretreated by Fomitopsis sp. IMER2	1.121	327	22.38
4	untreated ADF	1.453	337	20.82
5	ADF pretreated by I. lacteus CD2	1.630	335	13.50
6	ADF pretreated by <i>Fomitopsis</i> sp. IMER2	1.857	343	6.81
7	untreated ADL	0.169	326	61.31
8	ADL pretreated by I. lacteus CD2	0.258	316	42.09
9	ADL pretreated by Fomitopsis sp. IMER2	0.202	323	57.35
<sup><i>a</i></sup> The char yi	eld data of the TG curves, the peak height $(DTG_{max})$ , and the	he peak temperature $(T_{peak})$ va	lues of the DTG peaks we	ere calculated.

the dependence of thermal behaviors on different pretreatment fungi via thermogravimetric and pyrolysis analysis of biopretreated biomass fractions including cellulose and lignin components. The methods of sequential detergence used for determining the lignocellulose composition of biomass are based on stepwise extraction of soluble fractions with detergent and 72%  $H_2SO_4$ .<sup>17,18</sup> This allows a convenient means for isolating the lignocellulose fractions with speed, economy, and, most importantly, acceptable purity.<sup>27</sup>

ADF from corn stover was prepared via sequential detergence with neutral detergent reagent and acid detergent reagent, which is mainly composed of cellulose and lignin. Thermal behaviors of ADF samples mainly represent the cellulose thermogravimetric characteristic because the relative content of cellulose in ADF was approximated to 80%. Thermal decomposition of the corn stover ADF samples resembles that of biodegraded and undegraded cellulose as indicated by the features of the TG-DTG curves (Figure 3), as well as by the thermogravimetric parameters (Table 4, entries 4-6). The major thermal decomposition of corn stover ADF samples occurred at temperatures from 300 to 400 °C, and sharp DTG peaks can be observed. When the corn stover samples were pretreated by fungi, the ADF weight loss rates increased and higher weight losses were achieved. This was probably due to the selective degradation of cellulose amorphous regions in corn stover by biopretreatment, leading to the increase of purity and improvement in the structural order.<sup>28</sup> The extents of the increase in weight loss and weight loss rate varied depending upon the different pretreatment fungi. The total weight loss of corn stover ADF pretreated by white-rot fungus I. lacteus CD2 increased from 79.2 to 86.5%, whereas the weight loss of brown-rot fungus Fomitopsis sp. IMER2 treated sample increased dramatically to 93%. Furthermore, it is noteworthy that the weight loss rate of ADF sample pretreated by brown-rot fungus IMER2 was faster than that of the sample pretreated by white-rot fungus CD2, which was implied by the shape of the DTG curves and the calculated values of DTG<sub>max</sub>. The results indicate that by selectively degrading the cellulose amorphous regions, the pretreatment by brown-rot fungus

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Figure 3. TG and DTG curves of biopretreated corn stover ADF at 10 °C/min in nitrogen atmosphere (*I. lacteus* CD2, ADF sample pretreated by white-rot fungus *I. lacteus* CD2 for 30 days; *Fomitopsis* sp. IMER2, ADF sample pretreated by brown-rot fungus *Fomitopsis* sp. IMER2 for 30 days).



Figure 4. TG and DTG curves of biopretreated corn stover ADL at 10 °C/min in nitrogen atmosphere (*I. lacteus* CD2, ADL sample pretreated by white-rot fungus *I. lacteus* CD2 for 30 days; *Fomitopsis* sp. IMER2, ADL sample pretreated by brown-rot fungus *Fomitopsis* sp. IMER2 for 30 days).

*Fomitopsis* sp. IMER2 is more beneficial to the thermal decomposition of cellulose in corn stover.

The lignin is relatively more thermally stable than hemicellulose and cellulose. In comparison to the sharper DTG peaks of corn stover and ADF samples, ADL of corn stover had wide and flat DTG peaks. As shown in Table 4 (entries 7–9) and Figure 4, there were obvious differences in thermal behaviors between untreated and biopretreated ADL samples. From the ambient temperature to 1000 °C, only around 40 wt % of untreated ADL sample was lost at a very slow rate. This might be attributed to the slow carbonization of lignin, and char could be the main product.<sup>29</sup> After biopretreatment, the weight losses of ADL samples became larger and the thermal degradation processes were accelerated. The relationship between the thermogravimetric characteristics of ADL samples and the biodegradation of fungi could be explained by the fact that the deconstruction of lignin and the depolymerization of biomass can make the lignin in corn stover more readily available for thermal degradation.<sup>30</sup> When the corn stover was pretreated by white-rot fungus *I. lacteus* CD2, the ADL weight loss rate increased greatly and a total of 58 wt % maximum weight loss was achieved. The DTG curves of untreated and biopretreated ADL samples presented two main peaks in the 200–350 and 350–500 °C temperature ranges, respectively. The DTG peak heights of biopretreated samples were much higher than those of the control, which may correspond to the low thermal degradation residues compared with the untreated sample. The thermogravimetric analysis results of biomass ADL illustrated that the extents of the increase in weight loss and weight loss rate vary depending upon the different pretreatment

 Table 5. Pyrolysis Product Distribution of Untreated and

 Biopretreated Corn Stover, ADF, and ADL Samples

		product distribution						n		
	C	corn stover			ADF			ADL		
sample	oil	gas	char	oil	gas	char	oil	gas	char	
untreated	40.2	28.0	31.8	32.7	55.3	12.0	16.8	23.5	59.7	
I. lacteus CD2 treated	33.2	39.2	27.6	39.2	37.1	23.7	26.8	32.6	40.6	
Fomitopsis sp. IMER2	44.6	27.8	27.6	50.8	27.5	21.7	20.6	35.7	43.7	
treated										

fungi. White-rot fungus *I. lacteus* CD2 is more beneficial to the thermal decomposition of lignin in corn stover compared to the brown-rot fungus *Fomitopsis* sp. IMER2. The great lignin-degrading ability and structure alteration of *I. lacteus* CD2 might contribute to the promotion of thermal characteristics.

Pyrolysis Products Distribution of Fungal Pretreated Corn Stover and Lignocellulosic Fractions. The effects of biopretreatment on pyrolysis product distribution of corn stover and the ADF and ADL samples are shown in Table 5. Comparisons were made using Student's t test. The difference between the variables was taken to be significant if the p value was <0.05. Because of the error in the yields of liquid, gas, and residue was within 0.5%, the differences in the pyrolysis product distribution between untreated and biopretreated sample were statistically significant. In the case of corn stover, maximum liquid product (bio-oil) was achieved as 44.6% for Fomitopsis sp. IMER2 treated sample, slightly higher than that from untreated corn stover (40.2%). Although the oil yield of I. lacteus CD2 treated corn stover was the lowest (33.2%), the conversion rate of this sample was found to be as high as that of IMER2 sample (72.4%). This was because the gas product increased dramatically from 28.0 to 39.2% despite the lower oil yield from I. lacteus CD2 treated corn stover. The results of corn stover ADF sample pyrolysis analysis showed that the yield of both the oil and the solid product (char) were increased, whereas the yields of the gas product were decreased after biopretreatment. The oil yield obtained in this study for the untreated ADF of corn stover was 32.7%, whereas the I. lacteus CD2 treated ADF sample increased to 39.2% and the Fomitopsis sp. IMER2 treated sample increased dramatically to 50.8%. The char yield of untreated corn stover ADF sample was 12.0%, whereas the char yields of the sample pretreated with I. lacteus CD2 and Fomitopsis sp. IMER2 were 23.7% and about 21.7%, respectively. The yields of gas product of ADF sample after pretreated significant decreased from 55.3 to 37.1% for I. lacteus CD2 treated sample and to 27.5% for Fomitopsis sp. IMER2 treated sample. Compared to corn stover and ADF samples, ADL of corn stover gave the lowest liquid yield and the highest char yield at the same time. After biopretreatment, the yield of the oil and that of the gas product were increased, whereas the yield of the char was decreased. The increase in the oil yield of I. lacteus CD2 treated ADL sample was by about 159% (based on original untreated ADL sample), whereas in the case of Fomitopsis sp. IMER2 treated ADL sample the increase was by about 123% (based on original untreated ADL sample). The yields of gas product of ADL samples after biopretreatment significant increased from 23.5 to 32.6% for I. lacteus CD2 treated sample and to 35.7% for Fomitopsis sp. IMER2 treated sample. The char product from untreated ADL sample was 59.7%,

whereas the yields were decreased to 40.6 and 43.7% for *I. lacteus* CD2 and *Fomitopsis* sp. IMER2 treated samples, respectively. Briefly speaking, the trends of liquid product (bio-oil) yields in pyrolysis analysis were coincident with the changes in thermo-gravimetric characteristics, indicating that the brown-rot fungus *Fomitopsis* sp. IMER2 is favorable to the thermochemical conversion of cellulose, whereas the white-rot fungus *I. lacteus* CD2 is more beneficial to the thermochemical conversion of lignin in corn stover. This work may demonstrate the potential of fungal pretreatment in thermochemical conversion of corn stover to obtain biofuels.

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### ABBREVIATIONS USED

ADF, acid detergent fiber; ADL, acid detergent lignin; TG, thermogravimetric analysis; DTG, derivative thermogravimetry analysis; FTIR, Fourier transform infrared spectroscopy; XRD, X-ray diffraction analysis.

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