Carbohydrate Polymers 94 (2013) 38–45

Contents lists available at SciVerse ScienceDirect

Carbohydrate Polymers

journal homepage: www.elsevier.com/locate/carbpol

Wheat straw cellulose dissolution and isolation by *tetra-n*-butylammonium hydroxide

Chao Zhong, Chunming Wang, Fan Huang, Honghua Jia*, Ping Wei

College of Biotechnology and Pharmaceutical Engineering, Nanjing University of Technology, Nanjing 211316, PR China

A R T I C L E I N F O

Article history:
Received 26 August 2012
Received in revised form
12 December 2012
Accepted 17 January 2013
Available online 23 January 2013

Keywords:
Isolation
Wheat straw cellulose
TBAH
XRD
FTIR
Recycling

ABSTRACT

In this article, a novel and high efficient solvent, *tetra-n*-Butylammonium Hydroxide (TBAH), was used for dissolution and isolation of straw cellulose from wheat straw. The composition analysis with gas chromatography (GC) and the spectroscopic characterization analysis conducted by X-Ray diffraction (XRD)/Fourier transform infrared spectroscopy (FTIR) indicated that straw cellulose can be directly dissolved and isolated by TBAH without derivatization. The investigation on the properties of straw cellulose isolation was based on the results of single factor experiments and orthogonal experiments, with optimum conditions for straw cellulose isolation being obtained as follow: reaction temperature 60 °C, reaction time 40 min, concentration of TBAH 50% and ratio of TBAH to straw 10:1 (m/m). In addition, as a solvent for cellulose, TBAH could be recycled several times with high activity being retained.

1. Introduction

Cellulose is one of the most abundant natural polymers (Pinkert, Marsh, Pang, & Staiger, 2009). Cellulose and its derivatives are widely used in the chemical, pharmaceutical, papermaking industry and other areas (Klemm, Heublein, Fink, & Bohn, 2005). Partly because of the increased awareness of environment problems (Mohanty, Misra, & Drzal, 2002), recently there has been an increasing interest in more efficient utilization of agro-industrial residues for the production of various biodegradable and environment friendly cellulose materials (Pandey, Soccol, Nigam, & Soccol, 2000).

Agro-industrial residues primarily consist of cellulose, hemicelluloses and lignin (Zhang, Du, Qian, & Chen, 2010). In order to make full use of the residues, these major components need to be isolated. The cellulose isolated may be used as a raw material for papermaking, fuel alcohol and other industrial applications. With present technologies, efficient methods for isolating cellulose from agro-industrial residues are based on the dissolution of cellulose.

However, it's extremely difficult to dissolve cellulose in most traditional solvents because of the numerous intermolecular and intramolecular hydrogen bonds existing in cellulose (Lin, Zhan, Liu, Fu, & Lucia, 2009). Recently, there have been extensive efforts to find effective solvents for cellulose, with several solvents being reported, such as N-methylmorpholine N-oxide monohydrate (NMMO) (Roder & Morgenstern, 1999), LiCl/N, N-dimethylacetamide (DMAC) (McCormick, Callais, & Hutchinson, 1985), some molten salt hydrates like $LiClO_4{\cdot}3H_2O$ (Fischer, Leipner, Thummler, Brendler, & Peters, 2003) and $LiSCN{\cdot}2H_2O$ (Nehls, Lukanoff, Philipp, & Zschunke, 1983), and some aqueous solution of metal complex like cuprammonium hydroxide (Saalwachter et al., 2000). However, there remain some disadvantages including toxicity, thermal instability, hazardous vapors and high cost (Liu et al., 2007; Zhang, Wu, Zhang, & He, 2005). The other solvents reported recently are neither toxic nor too expensive, among which concentrated phosphoric acid (H_3PO_4) plays the major role (Conte, Maccotta, De Pasquale, Bubici, & Alonzo, 2009; Zhang, Cui, Lynd, & Kuang, 2006). However, cellulose dissolution with phosphoric acid usually results in the degradation of cellulose in lower temperature (Boerstoel, Maatman, Picken, Remmers, & Westerink, 2001; Butera, De Pasquale, Maccotta, Alonzo, & Conte, 2011; Saeman, 1945), which is not appropriate for regenerating cellulose.

Recently, ionic liquids (ILs), such as 1-ethyl-3-methylimidazolium acetate (EMIMAc), 1-butyl-3-methylimidazolium chloride (BMIMCl), 1-allyl-3-methylimidazolium chloride (AMIMCl), have been used as solvents for cellulose (Remsing, Swatloski, Rogers, & Moyna, 2006; Song, Zhang, Niu, & Wang, 2010; Zhu et al., 2006), but there are still some limitations for the application of ionic

---

* Corresponding author. Tel.: +86 25 58139368; fax: +86 25 58139368.
E-mail address: hhjia@njut.edu.cn (H. Jia).

0144-8617/$ – see front matter © 2013 Elsevier Ltd. All rights reserved.
http://dx.doi.org/10.1016/j.carbpol.2013.01.043

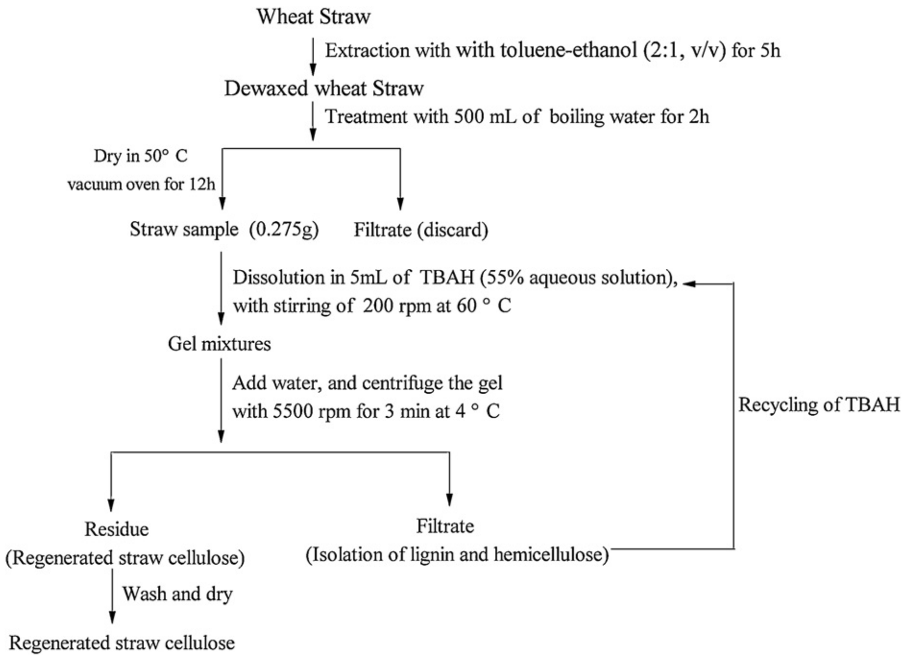

Fig. 1. Scheme for isolation of straw cellulose from wheat straw.

liquids, such as the relatively high viscosity, which slows down the dissolution of cellulose. Besides, the solubility of cellulose may decrease dramatically if a certain amount of water is added (Mazza, Catana, Vaca-Garcia, & Cecutti, 2009). Consequently, it's difficult to use polar ionic liquid as solvent to treat agro-industrial residues without pre-treatment. Furthermore, degradation of cellulose will also occur during the dissolution in ILs (Zhang et al., 2010), which is not appropriate for cellulose regeneration.

To overcome these disadvantages, hydroxide anion is considered as an ideal substitution (Cai, Liu, & Zhang, 2006; Liu & Zhang, 2009; Zhang et al., 2001). As the solvent for cellulose, sodium hydroxides (NaOH) aqueous solution with or without urea/thiourea added have already been reported (Cai & Zhang, 2006; Isogai & Atalla, 1998; Zhang, Ruan, & Gao, 2002). However, the applications of sodium hydroxide are strictly controlled for the conditions required to dissolve cellulose, which prevents it to be an ideal solvent for cellulose (Egal, Budtova, & Navard, 2007). In this article, we chose *tetra-n*-Butylammonium Hydroxide (TBAH) as the solvent for cellulose and isolated cellulose from wheat straw by dissolving it in TBAH. The feasibility of isolating straw cellulose from wheat straw was investigated. In addition, the recyclability of TBAH was also discussed. All the works in this article indicated that TBAH a useful solvent for dissolving and isolating cellulose from agro-industrial residues.

2. Materials and methods

2.1. Materials

Agricultural wheat straw, collected from Siyang, Jiangsu Province, was supplied by the biogas pilot plant of Nanjing University of Technology. It was dried in sunlight and then cut into small pieces with length of 1 cm. *Tetra-n*-Butylammonium Hydroxide (55% aqueous solution), acetic anhydride and potassium bromide were purchased from the Alfa Company. Microcrystalline cellulose, diethyltriamine pentaacetic acid, anthrone, sulfuric acid, hydrogen peroxide (30% aqueous solution) and sodium hydroxide were provided by the Aladdin Reagent Company.

2.2. Dissolution of microcrystalline cellulose in TBAH

The aqueous solution containing 55% TBAH by weight was used as the solvent for microcrystalline cellulose. Measured 5 mL of solvent into triangular flask, and then 0.55 g of microcrystalline cellulose was immediately added and mixed with the solvent. After stirring for 2 min, the microcrystalline cellulose could be completely dissolved in solvent at room temperature, with the resultant solution turning into a transparent gel. After that, 50 mL of water was added into the gel mixtures, with some white substances being precipitated. The white precipitates were obtained by centrifuging the gel system at a speed of 5500 rpm for 3 min at 4 °C, followed by being filtered, washed with 40% ethanol/water, and dried in a vacuum oven for 12 h.

2.3. Isolation of wheat straw cellulose

Based on the results illustrated above, we chose TBAH (55% aqueous solution) as the solvent for isolating cellulose from wheat straw, with a scheme for isolation of cellulose from wheat straw being described below and shown in Fig. 1.

The wheat straw was first dewaxed in Soxhlet apparatus with toluene–ethanol (2:1, v/v) for 5 h (Sun, Lawther, & Banks, 1995), followed by being immersed in 500 mL of boiling water for 2 h, and then dried at 50 °C in vacuum oven for 12 h. The dried wheat straw (0.275 g) was added into triangular flask and mixed with 5 mL of solvent completely. The mixtures were heated at 60 °C and stirred with speed of 200 rpm until the straw samples completely dissolved (actually 15–20 min was enough), and converted into mixture of gel particles. After the gel was cooled to room temperature, 50 mL of water was added with stirring to stop the reaction, with some substances precipitating. The gel mixtures were then centrifuged at a speed of 5500 rpm to obtaining the precipitates (regenerated straw cellulose), which was filtered afterwards, washed with 40% ethanol/water, and dried at 50 °C in a vacuum oven for 12 h.

2.4. Chemical identification of regenerated straw cellulose

Cellulose content of regenerated straw cellulose was determined by using the anthrone method (Frederick & Leslie, 1949) with some modifications. Anthrone, a tricyclic hydrocarbon ($C_{14}H_{10}O$), was generally used for cellulose assay and colorimetric determination of carbohydrates. This method was performed with initial digestion of regenerated straw cellulose (0.1 g) with 60% sulfuric acid (20 mL) in ice bath for 30 min, followed by adding 0.5 mL of anthrone solution (2.0%) to the digestion solution. The mixtures were subsequently heated in boiling water for 10 min. Finally, the absorbance of green color in resultant solution, which was proportional to cellulose content of sample, was measured by a UV–vis spectrophotometer Genesys 10S (Thermo, USA) at 625 nm. The detection was repeated three times. In addition, microcrystalline cellulose was used as a standard for calibration.

The neutral sugar composition of regenerated straw cellulose was detected by gas chromatography (GC) analysis of the corresponding alditol acetates. The regenerated straw cellulose powder (40 mg) was first hydrolysed with 72% sulfuric acid (0.125 mL) for 1 h at 35 °C, followed by a further hydrolysis for 2 h at 100 °C after dilution to 1.475 mL. After hydrolysis completed, the hydrolysate was made neutral by adding ammonia solution. Alditol acetates of neutral sugars in the hydrolysates were prepared according to the methods described by Blakeney, Harris, Henry, and Stone (1983). The alditol acetate derivatives of sugars were quantified by a HP 5890 GC using an Alltech DB-225 capillary column (30 m × 0.25 mm × 0.25 μm film) and the temperature program of initial temperature 190 °C with a hold of 4 min, followed by a temperature rise of 2 °C/min to 230 °C with a final hold of 25 min was designed. Helium was used as the carrier gas, and the sugar derivatives were detected with a flame ionization detector (FID).

2.5. Spectroscopic characterization of cellulose

The crystal structure of cellulose was detected by the X-Ray diffraction (XRD). The regenerated straw cellulose and microcrystalline cellulose were ground into powder for the measurement. The X-ray diffraction pattern of cellulose crystal was recorded on XTRA/3KW X-ray diffractometer (ARL), it was performed with CuKα radiation ($\lambda = 0.1541$ nm) and the diffraction scans were recorded with 2θ ranging from 3° to 50°.

The Crystallographic Index (CI) of cellulose was described by the Segal Equation (Segal, 1959) as Eq. (1):

$$CI = \frac{I_{hkl} - I_{am}}{I_{hkl}} \times 100\% \qquad (1)$$

where I_{hkl} was the diffraction intensity of crystallization area diffraction peak of cellulose; I_{am} was the diffraction intensity of non-crystallization area diffraction peak ($2\theta = 18°$) of cellulose.

The structure analysis of cellulose was performed with the Fourier transform infrared spectroscopy (Thermo Nicolet 380) (Kataoka & Kondo, 1998), which was recorded in 4000–400 cm^{-1} range with a resolution of 4 cm^{-1}. A certain amount of regenerated (straw) cellulose and microcrystalline cellulose powder were respectively mixed with dry KBr powder by ratio 1:100 (m/m), with the mixtures being pressed into tablets for infrared spectrometry analysis.

2.6. Design of experiment for wheat straw cellulose isolation

Single factor experiments were designed to investigate the properties of straw cellulose isolation and performed under various conditions. In this article, four predominant factors, including reaction temperature, reaction time, concentration of TBAH and ratio of TBAH to straw (m/m), were investigated respectively. The investigation of each factor's effect on straw cellulose isolation was performed with changing itself, while keeping other variables constant.

The investigation was carried out by measuring 5 mL of TBAH solution with concentrations of 45%, 50%, 55%, 65%, 75%, and 80%, respectively. Wheat straw sample was then added and mixed with the solvent according to mass ratio of TBAH/straw of 6:1, 8:1, 10:1, 12:1, and 15:1. The mixtures were heated at temperatures of 40, 50, 60, 70, and 80 °C with stirring for 20, 30, 40, 50, 60, and 70 min, respectively. After reacting for required times, the resulting solutions were slowly poured into 50 mL of distilled water with stirring to stop the reaction and the mixtures were then centrifuged at a speed of 5500 rpm for obtaining the precipitates (regenerated straw cellulose), which would be filtered afterwards, washed with 40% ethanol/water, and dried at 50 °C in a vacuum oven for 12 h.

In this article, the removal rate of lignin and hemicelluloses, which was calculated as follow: $\alpha = [M - (m_2 - m_1)]/M \times 100\%$, where M was the mass of straw samples, m_1 was the mass of filter paper and m_2 was the mass of filter paper and regenerated straw cellulose, was designed for evaluating the properties of regenerated straw cellulose.

Furthermore, in order to optimize the factors above, experiments based on an orthogonal experimental design (L_9 matrix) with the four variables being investigated were performed (Wu & Leung, 2011). According to the single factor experiments, appropriate levels of the four chosen factors, which were identified to have larger effects on straw cellulose isolation, were determined in the research. An L_9 (3^4) matrix, which was an orthogonal array of four factors and three levels, was employed to assign the considered factors and levels as shown in Table 1. Nine trials were carried out based on the L_9 matrix to complete the optimization process and each test was performed at least two times to guarantee the accuracy of results, with the optimal conditions being obtained by data analysis. Finally, the experiments with optimal conditions were repeated several times for verifying the results.

2.7. Recycling of TBAH

After regeneration of straw cellulose, filtrate was collected and processed with the isolation of lignin and hemicelluloses. The residual TBAH in filtrate was then recovered by simply reducing the pressure and subsequently distilling to remove water under the conditions of vacuum degree 0.1 MPa, temperature 48 °C (Dennewald, Pitner, & Weuster-Botz, 2011; Zhang et al., 2005), with recycled TBAH being applied as the solvent for wheat straw again. Repeated cycles of wheat straw dissolution followed by straw cellulose isolation and TBAH purification were performed to investigate the recyclability of TBAH. The cyclic experiments were carried out under the optimal conditions as explicated above.

2.8. Bleaching of regenerated straw cellulose

In order to make further removal of hemicelluloses, the bleaching treatment of regenerated straw cellulose by alkaline H_2O_2 was introduced in this article (Jiang et al., 2011; Zhang & Huang, 2004) and addition of DTPA (diethyltriamine pentaacetic acid) was designed to improve the stability of H_2O_2 at high temperature. Four factors including bleaching time and temperature, ratio of cellulose to liquid and pH of bleaching solution were researched respectively, with the optimum conditions as follow: bleaching time 1.5 h, temperature 90 °C, ratio 8 g/L and pH 11.5.

According to the conditions illustrated above, 2 mL of H_2O_2 (30% aqueous solution) was initially diluted to 30 mL, followed by being adjusted to pH 11.5 with NaOH, and then 30 μL of DTPA was added afterwards for improving the stability of H_2O_2. The regenerated straw cellulose powder (0.24 g) was measured and mixed with the bleaching solution, which would be heated at 90 °C for 1.5 h. After bleaching completed, the regenerated straw cellulose was filtered and dried in vacuum oven at 50 °C for 12 h.

Table 1
Analysis of L_9 (3^4) orthogonal experiments results.

Run	Factors and levels				Removal rate (%)
	A Temperature (°C)	B Time (min)	C Concentration of TBAH (%)	D Ratio of TBAH to straw (m/m)	
1	1 (50)	1 (20)	1 (50)	1 (8:1)	50.59
2	1	2 (30)	2 (55)	2 (10:1)	46.25
3	1	3 (40)	3 (60)	3 (12:1)	43.92
4	2 (60)	1	2	3	49.95
5	2	2	3	1	51.61
6	2	3	1	2	58.42
7	3 (65)	1	3	2	52.77
8	3	2	1	3	51.99
9	3	3	2	1	51.10
I	140.76	153.31	161.00	153.30	
II	159.98	149.85	147.30	157.44	
III	155.86	153.44	148.30	145.86	
I/3	46.92	51.10	53.67	51.10	
II/3	53.33	49.95	49.10	52.48	
III/3	51.95	51.15	49.43	48.62	
R^a	6.41	1.20	4.57	3.86	

[a] Referred to the results of extreme analysis.

3. Results and discussion

3.1. Characterization of microcrystalline cellulose dissolution in TBAH

Based on the phenomena above, the conclusion that microcrystalline cellulose could rapidly dissolve in TBAH was obtained. It was generally recognized that disrupting numerous intermolecular and intramolecular hydrogen bonds was required for dissolving cellulose. In the case of ionic liquid applied as solvent for cellulose, the relatively high chloride concentration in ionic liquid was thought to play the key role in dissolving cellulose. Similarly, the properties of cations, which would also affect cellulose dissolution, should be considered (Swatloski, Spear, Holbrey, & Rogers, 2002). Consequently, it was suggested that in the solvents for cellulose, which contained strong polarizing cations and large polarizable anions, extensive interactions between ions and cellulose would occur (Mccormick, Dawsey, & Newman, 1990). According to this conclusion, it could be speculated that the free hydroxide anion in TBAH would interact with the cellulose hydroxyl proton and the $[(n\text{-}t\text{-Bu})_4\text{-N}]^+$ cations might attack on the oxygen atom of cellulose hydroxyl during the dissolution process.

Based on the descriptions outlined above, the possible dissolution mechanism of microcrystalline cellulose in TBAH could be speculatively described as follows: at a certain temperature, the ion pairs in TBAH dissociated into individual hydroxide anion and $[(n\text{-}t\text{-Bu})_4\text{-N}]^+$ cation, the free hydroxide anion interacted with the cellulose hydroxyl proton via acid–base reaction and the cellulose hydroxyl oxygen negative charges were neutralized by the free $[(n\text{-}t\text{-Bu})_4\text{-N}]^+$ cations simultaneously, forming the cellulose–$\text{Bu}_4\text{-N}^+$ complex, which disrupted hydrogen bonds in cellulose and led to the dissolution of microcrystalline cellulose.

Furthermore, various factors affecting the dissolution of microcrystalline cellulose in TBAH were also investigated respectively. In this article, microcrystalline cellulose could be completely dissolved in TBAH (55% aqueous solution) within 2 min at room temperature, with a faster dissolution rate being obtained in higher temperature. In view of the industrial application, the advantage that cellulose samples without any pretreatment or activation could be dissolved in TBAH rapidly was really remarkable. In comparison, in some cases of other solvent systems for cellulose, such as DMAC/LiCl, the pretreatment or activation of cellulose was often needed (McCormick et al., 1985). Besides, higher fraction of TBAH was not always effective for the dissolution of microcrystalline cellulose. A certain amount of hydroxide was needed for cellulose dissolution, but excess hydroxide might decrease the stability of $[(n\text{-}t\text{-Bu})_4\text{-N}]^+$ cation in some extent. Generally, TBAH solution with concentration ranging from 50% to 60% was the best for dissolving microcrystalline cellulose.

After dissolution completed, a transparent gel, which kept its liquid state with enhanced viscosity, was obtained subsequently. The white precipitates (regenerated cellulose) were obtained by adding a certain amount of water into the gel with stirring to stop the reaction. Structure identification of the precipitates was performed with Fourier transformed infrared spectrometry (FTIR), with the spectrum shown in Fig. 2. The absorbance bands at 3385, 2903, 1652, 1432, 1172, 1050, 898 cm^{-1} seen in spectrum were associated with the precipitates (B), which presented the remarkable characteristic peaks of cellulose as follow: A strong band at 3385 cm^{-1} was originated from O—H stretching vibration, and that at 2903 cm^{-1} was attributed to the C—H stretching vibration;

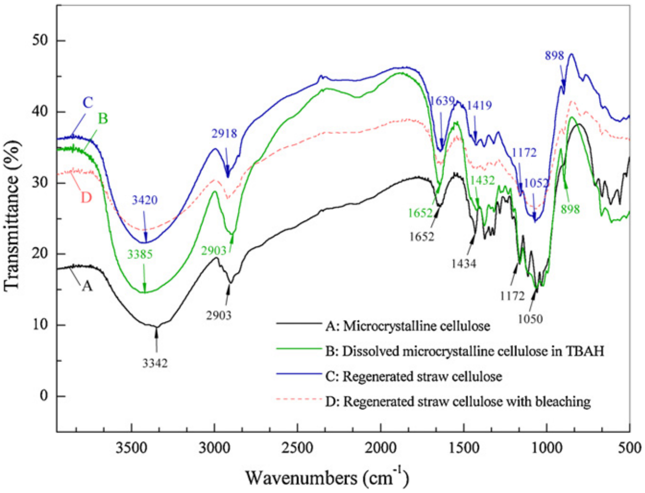

Fig. 2. FTIR spectra of microcrystalline cellulose (A), dissolved microcrystalline cellulose in TBAH (B), regenerated straw cellulose (C) and regenerated straw cellulose with bleaching treatment (D).

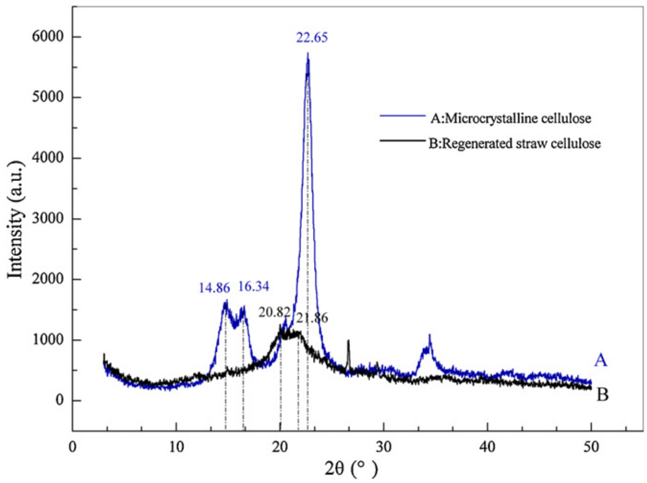

Fig. 3. XRD patterns of microcrystalline cellulose (A) and regenerated straw cellulose (B).

the band at 1652 cm^{-1} corresponded to the bending vibration of absorbed water in cellulose; the absorption band at 1172 cm^{-1} arose from C—O antisymmetric stretching vibration and a strong peak at 1050 cm^{-1} belonged to the C—O—C pyranose ring skeletal vibration; a characteristic peak of cellulose at 898 cm^{-1} was also detected in the spectrum. The descriptions above indicated that no distinct derivatization reaction occurred during the dissolution of microcrystalline cellulose in TBAH. Furthermore, no distinct mass loss of microcrystalline cellulose was observed during the dissolution and regeneration process, based on which the application of TBAH on cellulose dissolution and regeneration would be easier to implement.

3.2. *Chemical properties of regenerated straw cellulose*

In order to identify the chemical properties of regenerated straw cellulose, detections including content of cellulose and sugar composition were performed respectively.

According to the anthrone method, content of cellulose in regenerated straw cellulose was obtained accordingly. In the present study, a standard curve of cellulose with *R*-square of 0.99526 was prepared, based on which the cellulose content in regenerated straw cellulose was obtained according to the absorbance of resultant solution at 625 nm. The results showed that the regenerated straw cellulose, with high cellulose content of about 89.7%, was obtained during the isolation process, indicating that high efficient isolation of straw cellulose from wheat straw could be achieved with TBAH.

Based on gas chromatography (GC) analysis of the corresponding alditol acetate, neutral sugar composition of regenerated straw cellulose was detected, with results showing that glucose was the predominant sugar component, which comprised 91.7% of total sugars, indicating a relatively high content of cellulose in the regenerated straw cellulose, which was consistent with the description of cellulose content analysis above. Furthermore, a small amount of noncellulosic sugars, such as xylose (3.1%), galactose (2.7%), mannose (1.1%), and arabinose (0.9%), were also observed, indicating that minor quantities of residual hemicelluloses still remained in the regenerated straw cellulose, which needed a further reduction in hemicelluloses by bleaching treatment.

3.3. *Structure analysis of regenerated straw cellulose*

XRD spectra of cellulose were illustrated in Fig. 3. As can be seen, the characteristic peaks of microcrystalline cellulose were found with diffractions of 14.86°, 16.34° and 22.65°, which were similar with the characteristic peaks of cellulose I (Zhang & Lin, 2009). The crystallinity of microcrystalline cellulose was 90.3%, which might be attributed to the formation of intermolecular hydrogen bonds in microcrystalline cellulose, forming a stable structure with high crystallinity (Zhbankov, 1992; Zhou, Yuan, Chen, & Bao, 2012). In contrast, the regenerated straw cellulose, with its crystallinity of 48.7%, exhibited the typical diffraction patterns of cellulose II with 2θ of 20.82° and 21.86° (Inagaki, Siesler, Mitsui, & Tsuchikawa, 2010; Nishiyama, Langan, & Chanzy, 2002; Zhao et al., 2007). Compared with microcrystalline cellulose, the crystallinity of regenerated straw cellulose was much lower, indicating that the transformation from cellulose I to cellulose II was performed during the dissolution process (Feng & Chen, 2008).

The structural analysis of microcrystalline cellulose and regenerated straw cellulose were performed with Fourier transformed infrared spectrometry (FTIR). As shown in Fig. 2, there was no significant difference between the FTIR spectra of microcrystalline cellulose (A) and regenerated straw cellulose (C).

The strong absorption bands at 2903 cm^{-1} for microcrystalline cellulose and 2918 cm^{-1} for regenerated straw cellulose were assigned to the C—H stretching vibration. The absorption bands at 1639 cm^{-1} in regenerated straw cellulose and 1652 cm^{-1} in microcrystalline cellulose belonged to the bending mode of absorbed water. Besides, the absorption band at 1172 cm^{-1} for both microcrystalline cellulose and regenerated straw cellulose corresponded to C—O antisymmetric stretching vibration (Sun, Xu, Geng, Sun, & Sun, 2005).

For microcrystalline cellulose, the strong absorption band at 1434 cm^{-1} was corresponded with the CH_2 scissoring motion. In comparison, the 1419 cm^{-1} peak of regenerated straw cellulose weakened and shifted to a lower wavenumber, which was attributed to the destruction of intermolecular hydrogen bonds associating with oxygen atom at C_6 (Higgins, Stewart, & Harrington, 1961). A strong peak at 1050 cm^{-1} arose from C—O—C pyranose ring skeletal vibration in microcrystalline cellulose. In contrast, the intensity of the peak at 1051 cm^{-1} for C—O—C stretching in regenerated straw cellulose slightly decreased due to the degradation of cellulose macromolecules during dissolution in TBAH. In addition, an absorbance at 898 cm^{-1}, which belonged to the C—O stretching vibration in the amorphous region, was obviously detected in the regenerated straw cellulose spectrogram. Besides, the broad band at around 3342 cm^{-1} was originated from the O—H vibrations in microcrystalline cellulose. In comparison, the band of O—H vibration in regenerated straw cellulose shifted to a higher frequency (3420 cm^{-1}) with a sharper and narrower profile, which was a sign of breaking down the hydrogen bonds in regenerated straw cellulose (Kataoka & Kondo, 1998; Zhou et al., 2001).

In conclusion, the above results showed that after dissolution of straw samples in TBAH followed by straw cellulose isolation, the isolated substance (regenerated straw cellulose) with a relatively high content of cellulose was obtained, indicating that the dissolution and isolation of straw cellulose from wheat straw could be successfully achieved with TBAH.

In addition, as shown in Fig. 2, the regenerated straw cellulose with bleaching treatment (D), with remarkable characteristic peaks at 3404, 2918, 1646, 1420, 1172, 1050, 899 cm^{-1} respectively, showed a much more similar FTIR spectrogram to native cellulose, indicating that no obvious derivatization occurred during the bleaching treatment. Besides, after the bleaching process, the removal rate of lignin and hemicelluloses increased by about 2–3%, showing that bleaching with alkaline H_2O_2 might be an efficient retreatment for regenerated straw cellulose.

3.4. Factors affecting wheat straw cellulose isolation

Generally, the isolation of straw cellulose from wheat straw was primarily affected by the reaction time and temperature, ratio of TBAH/straw and concentration of TBAH.

In the present study, isolation of straw cellulose at different reaction temperature was investigated. At room temperature, TBAH only swelled wheat straw but could not dissolve it. However, with increasing the temperature, straw samples could be dissolved more rapidly. As shown in Fig. 4(a), at an initial stage of 40–60 °C, the dissolution occurred rapidly with an obvious increase in removal rate of lignin and hemicelluloses, followed by a decline of dissolution rate and slower increase of removal rate at higher temperature. Similarly, the relationship between reaction time and isolation properties was illustrated in Fig. 4(b). The straw samples might be completely dissolved in TBAH with stirring for 20 min at 60 °C. Besides, with the increase of dissolution time, a higher removal rate of lignin and hemicelluloses was achieved accordingly. However, it was characterized by a slight increase of removal rate and decreasing dissolution rate after reacting for 40 min. These phenomena might be explained by the relatively perfect crystalline structure in residual cellulose fibrils and increasing viscosity of resultant solution. With increasing reaction temperature and time, higher concentration of cellulose solution with higher viscosities were prepared in TBAH, weakening the free hydroxide anions' association with hydrogen bonds formed in straw cellulose, which lead to the decrease of dissolution rate.

The changes of removal rate of lignin and hemicelluloses as a function of TBAH concentration was shown in Fig. 4(c). The removal rate sharply increased up to a maximal point of 58.84% at an initial concentration period from 45% to 50%, followed by a decline with increasing the concentration of TBAH. This might be caused by the instability of $[(n\text{-}t\text{-Bu})_4\text{-N}]^+$ cation and the formation of hydrogen bonds between hydroxyl in solution with higher concentration, which resulted in the decrease of free hydroxide anions (Lan, Liu, Yue, Sun, & Kennedy, 2011; Zhen & Zhen, 2012) and weakened hydroxide anions' association with straw cellulose.

The effect of the ratio between TBAH and straw (m/m) on straw cellulose isolation was illustrated in Table 2. As can be seen, the removal rate of lignin and hemicelluloses obviously increased from 30.16% to 71.80% with the ratio of TBAH to straw (m/m) shifting from 6:1 to 15:1, which might be explained by the decline of cellulose concentration with decreasing amount of straw, reducing the viscosity of solution and promoting the TBAH's effects on straw cellulose. However, compared to reaction with ratio 10:1, in reaction with ratio 12:1 and 15:1, the yield of regenerated straw cellulose showed a dramatic decline, which was not appropriate for regeneration of straw cellulose.

The range value in orthogonal experiments reflected the significance of each factor (Liu & Deng, 2009; Wu & Leung, 2011; Yuan, Kong, & Yan, 2006). Based on the comparison of four factors' range values as seen in Fig. 5, the factor influences were listed in a descending order as follows: reaction temperature (6.41) > concentration of TBAH (4.57) > ratio of TBAH to straw (m/m) (3.86) > reaction time (1.20), indicating that the most predominant factor contributing to straw cellulose isolation was reaction temperature (A), followed by concentration of TBAH (C) and ratio of TBAH to straw (D), and lastly reaction time (B).

In addition, as shown in Table 1, based on the data analysis, the optimal operation conditions were obtained as follow: $A_2B_3C_1D_2$ (reaction temperature 60 °C, reaction time 40 min, concentration of TBAH 50% and ratio of TBAH to straw 10:1 (m/m)). Experiments with optimal conditions were repeated several times for confirming the optimized results and efficiencies, with removal rate of lignin and hemicelluloses of around 58% being obtained in a series of confirmatory experiments, which was consistent with the former orthogonal experimental result, indicating that the optimal conditions obtained through orthogonal experiments could in some extent optimize the isolation of straw cellulose.

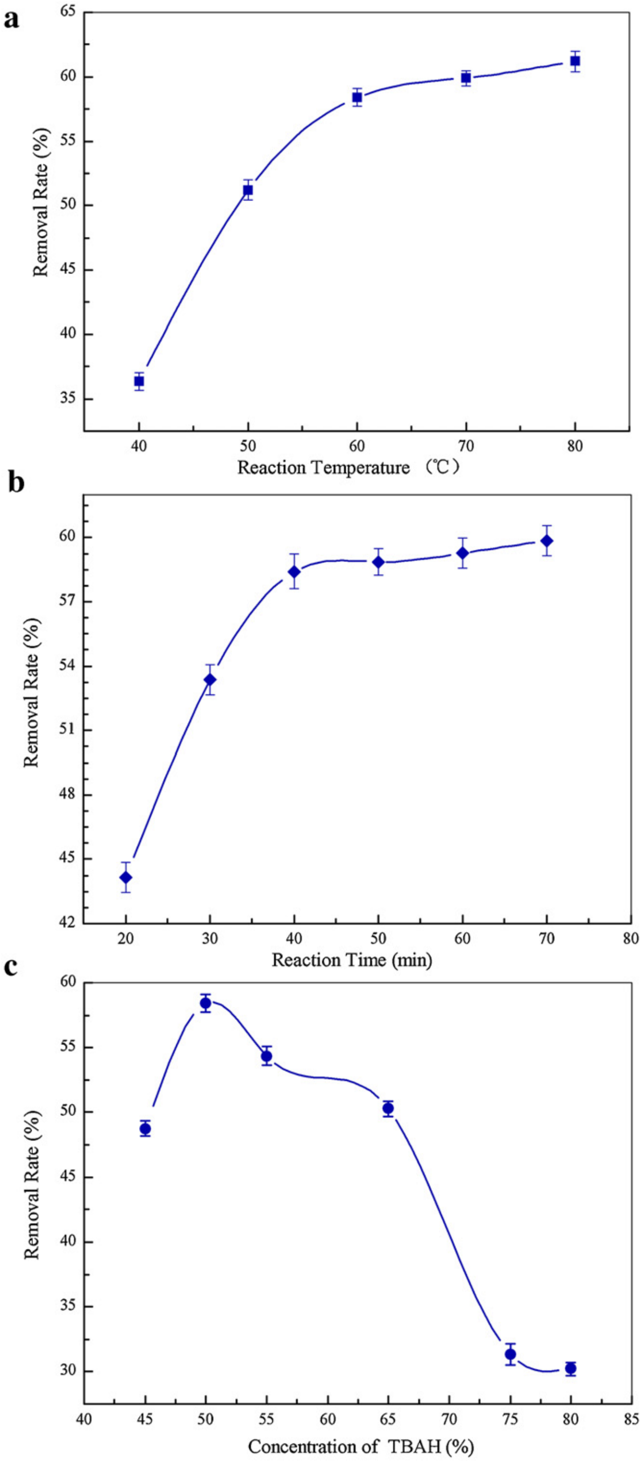

Fig. 4. (a) Effect of reaction temperature on straw cellulose isolation (reaction time 40 min, concentration of TBAH 50%, ratio of TBAH/straw 10:1 (m/m)); (b) effect of reaction time on straw cellulose isolation (reaction temperature 60 °C, concentration of TBAH 50%, ratio of TBAH/straw 10:1 (m/m)); (c) effect of solution concentration on straw cellulose isolation (reaction temperature 60 °C, reaction time 40 min, ratio of TBAH/straw 10:1 (m/m)).

Table 2
Effect of mass ratio of TBAH/wheat straw on straw cellulose isolation (Reaction temperature 60 °C, reaction time 40 min, concentration of TBAH 50%).

Ratio of TBAH to straw (m/m)	6:1	8:1	10:1	12:1	15:1
Removal rate (%)	30.16	30.55	58.42	63.19	71.80
Mass of regenerated straw cellulose (g)	–	–	0.1040	0.0767	0.0470

Table 3
Results of tests with recycling TBAH.

Cycle index of experiments	Volume of TBAH solution (mL)	Loss of TBAH[a] (%)	Mass of wheat straws[b] (g)	Removal rate (%)
1	5	–	0.250	58.42
2	5	2.3	0.244	54.24
3	5	3.4	0.236	51.16
4	5	2.1	0.231	49.18
5	5	3.7	0.222	45.12
6	5	4.5	0.212	41.34

[a] Mass of TBAH would slightly decrease during the reusing experiments due to the operation losses.

[b] Mass of wheat straw was calculated based on ratio of TBAH to straw (m/m) 10:1 with considering of the mass loss of TBAH.

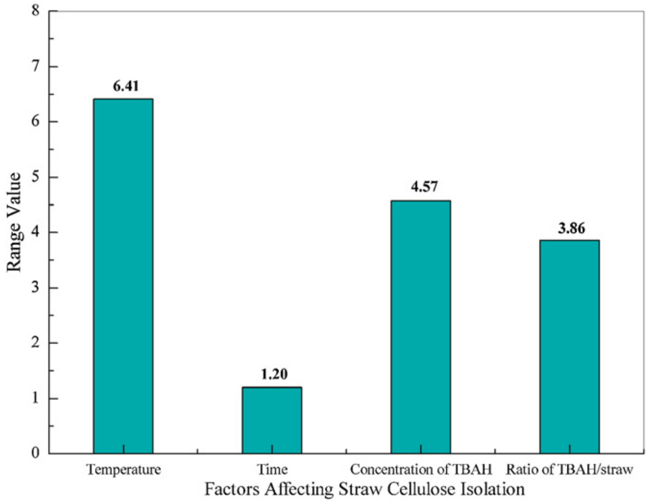

Fig. 5. Range values of factors in orthogonal design experiments.

3.5. The recycling of TBAH

According to the methods above, TBAH might be simply recovered and applied for dissolution and isolation of straw cellulose. The verification tests were designed for checking the recyclability of TBAH, with the results shown in Table 3. In view of operation losses in dissolution and isolation process, the mass loss of TBAH should be considered.

As can be seen, it was noteworthy that the cycles 2–6 employed recycled TBAH as solvent for straw cellulose and exhibited a gradual decrease in removal rate of lignin and hemicelluloses from 54.24% to 41.34%, which met the requirements for isolating straw cellulose from wheat straw, indicating that no significant loss of activity occurred during the recycling of TBAH. It is apparent that this advantage will promote the industrial application of TBAH for cellulose dissolution and isolation in future.

4. Conclusion

In this article, the solubility of cellulose in *tetra*-*n*-Butylammonium Hydroxide (TBAH) and an efficient approach for isolating straw cellulose from wheat straw were investigated. Composition and structure analysis of regenerated straw cellulose indicated that the dissolution and isolation of straw cellulose might be successfully achieved with TBAH. In the investigations of straw cellulose isolation, four factors, including reaction temperature and time, ratio of TBAH/straw and concentration of TBAH, were researched respectively, with results indicating that reaction temperature had greater influence on straw cellulose isolation, followed by concentration of TBAH, ratio of TBAH/straw and reaction time. Optimal conditions for straw cellulose isolation were obtained through orthogonal experiments and listed as follows: reaction temperature 60 °C; reaction time 40 min; concentration of TBAH 50%; ratio of TBAH/straw 10:1. Furthermore, as a solvent of cellulose, TBAH could be recycled several times with high activity retained. In addition, bleaching with alkaline H_2O_2 might be an efficient retreatment for regenerated straw cellulose.

In conclusion, the above results indicated that TBAH might be a potential green solvent for dissolution and isolation of wheat straw cellulose.

Acknowledgment

The research was financially supported by the State Key Basic Research and Development Plan of China (2009CB724700), National Key Technology R&D Program (2011BAD15B02), National Hi-Tech R&D Program (2012AA021405), Jiangsu Key Technology R&D Program (BE2010359), PAPD, Jiangsu Province Natural Science Foundation Innovation Project (SBK200910195), and PCSIRT (IRT1066).

References

Blakeney, A. B., Harris, P. J., Henry, R. J., & Stone, B. A. (1983). A simple and rapid preparation of alditol acetates for monosaccharide analysis. *Carbohydrate Research*, *113*(2), 291–299.

Boerstoel, H., Maatman, H., Picken, S. J., Remmers, R., & Westerink, J. B. (2001). Liquid crystalline solutions of cellulose acetate in phosphoric acid. *Polymer*, *42*(17), 7363–7369.

Butera, G., De Pasquale, C., Maccotta, A., Alonzo, G., & Conte, P. (2011). Thermal transformation of micro-crystalline cellulose in phosphoric acid. *Cellulose*, *18*(6), 1499–1507.

Cai, J., Liu, Y., & Zhang, L. (2006). Dilute solution properties of cellulose in LiOH/urea aqueous system. *Journal of Polymer Science Part B-Polymer Physics*, *44*(21), 3093–3101.

Cai, J., & Zhang, L. (2006). Unique gelation behavior of cellulose in NaOH/Urea aqueous solution. *Biomacromolecules*, *7*(1), 183–189.

Conte, P., Maccotta, A., De Pasquale, C., Bubici, S., & Alonzo, G. (2009). Dissolution mechanism of crystalline cellulose in H_3PO_4 as assessed by high-field NMR spectroscopy and fast field cycling NMR relaxometry. *Journal of Agricultural and Food Chemistry*, *57*(19), 8748–8752.

Dennewald, D., Pitner, W., & Weuster-Botz, D. (2011). Recycling of the ionic liquid phase in process integrated biphasic whole-cell biocatalysis. *Process Biochemistry*, *46*(5), 1132–1137.

Egal, M., Budtova, T., & Navard, P. (2007). Structure of aqueous solutions of microcrystalline cellulose/sodium hydroxide below 0 °C and the limit of cellulose dissolution. *Biomacromolecules*, *8*(7), 2282–2287.

Feng, L., & Chen, Z. (2008). Research progress on dissolution and functional modification of cellulose in ionic liquids. *Journal of Molecular Liquids*, *142*(1–3), 1–5.

Fischer, S., Leipner, H., Thummler, K., Brendler, E., & Peters, J. (2003). Inorganic molten salts as solvents for cellulose. *Cellulose*, *10*(3), 227–236.

Frederick, J. V., & Leslie, S. (1949). Determination of starch and cellulose with anthrone. *Analytical Chemistry*, *21*(8), 950–953.

Higgins, H. G., Stewart, C. M., & Harrington, K. J. (1961). Infrared spectra of cellulose and related polysaccharides. *Journal of Polymer Science*, *51*(155), 59–84.

Inagaki, T., Siesler, H. W., Mitsui, K., & Tsuchikawa, S. (2010). Difference of the crystal structure of cellulose in wood after hydrothermal and aging degradation: A NIR spectroscopy and XRD study. *Biomacromolecules*, *11*(9), 2300–2305.

Isogai, A., & Atalla, R. H. (1998). Dissolution of cellulose in aqueous NaOH solutions. *Cellulose*, *5*(4), 309–319.

Jiang, M., Zhao, M., Zhou, Z., Huang, T., Chen, X., & Wang, Y. (2011). Isolation of cellulose with ionic liquid from steam exploded rice straw. *Industrial Crops and Products*, *33*(3), 734–738.

Kataoka, Y., & Kondo, T. (1998). FT-IR microscopic analysis of changing cellulose crystalline structure during wood cell wall formation. *Macromolecules*, *31*(3), 760–764.

Klemm, D., Heublein, B., Fink, H. P., & Bohn, A. (2005). Cellulose: Fascinating biopolymer and sustainable raw material. *Angewandte Chemie-International Edition*, *44*(22), 3358–3393.

Lan, W., Liu, C., Yue, F., Sun, R., & Kennedy, J. F. (2011). Ultrasound-assisted dissolution of cellulose in ionic liquid. *Carbohydrate Polymers*, *86*(2), 672–677.

Lin, C., Zhan, H., Liu, M., Fu, S., & Lucia, L. A. (2009). Novel preparation and characterization of cellulose microparticles functionalized in ionic liquids. *Langmuir*, *25*(17), 10116–10120.

Liu, C., Sun, R., Zhang, A., Qin, M., Ren, J., & Wang, X. (2007). Preparation and characterization of phthalated cellulose derivatives in room-temperature ionic liquid without catalysts. *Journal of Agricultural and Food Chemistry*, *55*(6), 2399–2406.

Liu, S., & Zhang, L. (2009). Effects of polymer concentration and coagulation temperature on the properties of regenerated cellulose films prepared from LiOH/urea solution. *Cellulose*, *16*(2), 189–198.

Liu, Y. H., & Deng, Y. (2009). Technical parameters of dissolution and separation of cotton stalk cellulose in ionic liquid. *Transactions of the Chinese Society of Agricultural Engineering*, *25*(9), 259–263.

Mazza, M., Catana, D., Vaca-Garcia, C., & Cecutti, C. (2009). Influence of water on the dissolution of cellulose in selected ionic liquids. *Cellulose*, *16*(2), 207–215.

McCormick, C. L., Callais, P. A., & Hutchinson, B. H. (1985). Solution studies of cellulose in lithium-chloride and *N*,*N*-dimethylacetamide. *Macromolecules*, *18*(12), 2394–2401.

Mccormick, C. L., Dawsey, T. R., & Newman, J. K. (1990). Competitive formation of cellulose para-toluenesulfonate and chlorodeoxycellulose during homogeneous reaction of para-toluenesulfonyl chloride with cellulose in *N*,*N*-dimethylacetamide lithium-chloride. *Carbohydrate Research*, *208*, 183–191.

Mohanty, A. K., Misra, M., & Drzal, L. T. (2002). Sustainable bio-composites from renewable resources: Opportunities and challenges in the green materials world. *Journal of Polymers and the Environment*, *10*(1–2), 19–26.

Nehls, I., Lukanoff, B., Philipp, B., & Zschunke, A. (1983). C-13-NMR investigation of cellulose in various solvents. *Acta Polymerica*, *34*(2), 105–108.

Nishiyama, Y., Langan, P., & Chanzy, H. (2002). Crystal structure and hydrogen-bonding system in cellulose I beta from synchrotron X-ray and neutron fiber diffraction. *Journal of the American Chemical Society(UNSP JA025731931)*, *124*, 9074–9082.

Pandey, A., Soccol, C. R., Nigam, P., & Soccol, V. T. (2000). Biotechnological potential of agro-industrial residues. I: Sugarcane bagasse. *Bioresource Technology*, *74*(1), 69–80.

Pinkert, A., Marsh, K. N., Pang, S., & Staiger, M. P. (2009). Ionic liquids and their interaction with cellulose. *Chemical Reviews*, *109*(12), 6712–6728.

Remsing, R. C., Swatloski, R. P., Rogers, R. D., & Moyna, G. (2006). Mechanism of cellulose dissolution in the ionic liquid 1-N-butyl-3-methylimidazolium chloride: A C-13 and Cl-35/37 NMR relaxation study on model systems. *Chemical Communications*, *12*, 1271–1273.

Roder, T., & Morgenstern, B. (1999). The influence of activation on the solution state of cellulose dissolved in N-methylmorpholine-N-oxide-monohydrate. *Polymer*, *40*(14), 4143–4147.

Saalwachter, K., Burchard, W., Klufers, P., Kettenbach, G., Mayer, P., Klemm, D., et al. (2000). Cellulose solutions in water containing metal complexes. *Macromolecules*, *33*(11), 4094–4107.

Saeman, J. (1945). Kinetics of wood saccharification – Hydrolysis of cellulose and decomposition of sugars in dilute acid at high temperature. *Industrial & Engineering Chemistry*, *1*(37), 43–52.

Segal, L. G. J. M. A. (1959). An empirical method for estimating the degree of crystallinity of native cellulose using the X-ray diffractometer. *Textile Research Journal*, *29*, 768–788.

Song, H., Zhang, J., Niu, Y., & Wang, Z. (2010). Phase transition and rheological behaviors of concentrated cellulose/ionic liquid solutions. *Journal of Physical Chemistry B*, *114*(18), 6006–6013.

Sun, J. X., Xu, F., Geng, Z. C., Sun, X. F., & Sun, R. C. (2005). Comparative study of cellulose isolated by totally chlorine-free method from wood and cereal straw. *Journal of Applied Polymer Science*, *97*(1), 322–335.

Sun, R. C., Lawther, J. M., & Banks, W. B. (1995). Influence of alkaline pretreatments on the cell-wall components of wheat-straw. *Industrial Crops and Products*, *4*(2), 127–145.

Swatloski, R. P., Spear, S. K., Holbrey, J. D., & Rogers, R. D. (2002). Dissolution of cellose with ionic liquids. *Journal of the American Chemical Society*, *124*(18), 4974–4975.

Wu, X., & Leung, D. Y. C. (2011). Optimization of biodiesel production from camelina oil using orthogonal experiment. *Applied Energy*, *88*(11), 3615–3624.

Yuan, Z. H., Kong, X. Y., & Yan, Y. J. (2006). An orthogonal experiment on echnical needs for Chinese silvergrass hydrolysis process with dilute sulphuric acid. *Acta Energiae Solaris Sinica*, *27*(6).

Zhang, H., Wu, J., Zhang, J., & He, J. S. (2005). 1-Allyl-3-methylimidazolium chloride room temperature ionic liquid: A new and powerful nonderivatizing solvent for cellulose. *Macromolecules*, *38*(20), 8272–8277.

Zhang, J. Q., & Lin, L. (2009). XPS analysis of cellulose treated with two ionic liquids. *Journal of South China University of Technology*, *37*(6), 17–21.

Zhang, L. N., Ruan, D., & Gao, S. J. (2002). Dissolution and regeneration of cellulose in NaOH/thiourea aqueous solution. *Journal of Polymer Science Part B-Polymer Physics*, *40*(14), 1521–1529.

Zhang, L. N., Ruan, D., & Zhou, J. P. (2001). Structure and properties of regenerated cellulose films prepared from cotton linters in NaOH/Urea aqueous solution. *Industrial & Engineering Chemistry Research*, *40*(25), 5923–5928.

Zhang, Y., Cui, J. B., Lynd, L. R., & Kuang, L. R. (2006). A transition from cellulose swelling to cellulose dissolution by o-phosphoric acid: Evidence from enzymatic hydrolysis and supramolecular structure. *Biomacromolecules*, *7*(2), 644–648.

Zhang, Y., Du, H., Qian, X., & Chen, E. Y. X. (2010). Ionic liquid–water mixtures: Enhanced K-w for efficient cellulosic biomass conversion. *Energy & Fuels*, *24*, 2410–2417.

Zhang, Z., & Huang, G. Q. (2004). Oxygen-containing radicals and hydrogen peroxide delignification. *Transactions of China Pulp and Paper*, *19*(1), 186–188.

Zhao, H., Kwak, J. H., Zhang, Z. C., Brown, H. M., Arey, B. W., & Holladay, J. E. (2007). Studying cellulose fiber structure by SEM, XRD, NMR and acid hydrolysis. *Carbohydrate Polymers*, *68*(2), 235–241.

Zhbankov, R. G. (1992). Hydrogen-bonds and structure of carbohydrates. *Journal of Molecular Structure*, *270*, 523–539.

Zhen, D. D., & Zhen, C. (2012). Theoretical and experimental investigation on dissolution and regeneration of cellulose in ionic liquid. *Carbohydrate Polymers*, *89*(1), 7–16.

Zhou, S., Tashiro, K., Hongo, T., Shirataki, H., Yamane, C., & Ii, T. (2001). Influence of water on structure and. mechanical properties of regenerated cellulose studied by an organized combination of infrared spectra, X-ray diffraction, and dynamic viscoelastic data measured as functions of temperature and humidity. *Macromolecules*, *34*(5), 1274–1280.

Zhou, W. H., Yuan, S. S., Chen, Y. W., & Bao, L. (2012). Morphology and hydrogen-bond restricted crystallization of poly(butylene succinate)/cellulose diacetate blends. *Journal of Applied Polymer Science*, *124*(4), 3124–3131.

Zhu, S. D., Wu, Y. X., Chen, Q. M., Yu, Z. N., Wang, C. W., Jin, S. W., et al. (2006). Dissolution of cellulose with ionic liquids and its application: A mini-review. *Green Chemistry*, *8*(4), 325–327.