Pretreatment and Fractionation of Wheat Straw Using Various Ionic Liquids

ARTICLE *in* JOURNAL OF AGRICULTURAL AND FOOD CHEMISTRY · JULY 2013

Impact Factor: 2.91 · DOI: 10.1021/jf401980p · Source: PubMed

5 AUTHORS, INCLUDING:

André M da Costa Lopes
Laboratório Nacional de Energia e Geologia
16 PUBLICATIONS **170** CITATIONS
SEE PROFILE

Karen Andreína João
New University of Lisbon
6 PUBLICATIONS **107** CITATIONS
SEE PROFILE

Ewa Bogel-Łukasik
LAQV, REQUIMTE, New University of Lisbon
64 PUBLICATIONS **1,471** CITATIONS
SEE PROFILE

Luisa Roseiro
Laboratório Nacional de Energia e Geologia
32 PUBLICATIONS **277** CITATIONS
SEE PROFILE

JOURNAL OF AGRICULTURAL AND FOOD CHEMISTRY

Article

pubs.acs.org/JAFC

Pretreatment and Fractionation of Wheat Straw Using Various Ionic Liquids

André M. da Costa Lopes,[†] Karen G. João,[†] Ewa Bogel-Łukasik,[§] Luísa B. Roseiro,[†] and Rafał Bogel-Łukasik*,[†]

[†]Laboratório Nacional de Energia e Geologia, Unidade de Bioenergia, 1649-038 Lisboa, Portugal

[§]Departamento de Química, Faculdade de Ciências e Tecnologia, Universidade Nova de Lisboa, REQUIMTE, 2829-516 Caparica, Portugal

ABSTRACT: Pretreatment of lignocellulosic biomass with ionic liquids (ILs) is a promising and challenging process for an alternative method of biomass processing. The present work emphasizes the examination of wheat straw pretreatment using ILs, namely, 1-butyl-3-methylimidazolium hydrogensulfate ([bmim][HSO_4]), 1-butyl-3-methylimidazolium thiocyanate ([bmim][SCN]), and 1-butyl-3-methylimidazolium dicyanamide ([bmim][$N(CN)_2$]). Only [bmim][HSO_4] was found to achieve a macroscopic complete dissolution of wheat straw during pretreatment. The fractionation process demonstrated to be dependent on the IL used. Using [bmim][SCN], a high-purity lignin-rich material was obtained. In contrast, [bmim][$N(CN)_2$] was a good solvent to produce high-purity carbohydrate-rich fractions. When [bmim][HSO_4] was used, a different behavior was observed, exhibiting similarities to an acid hydrolysis pretreatment, and no hemicellulose-rich material was recovered during fractionation. A capillary electrophoresis (CE) technique allowed for a better understanding of this phenomenon. Hydrolysis of carbohydrates was confirmed, although an extended degradation of monosaccharides to furfural and hydroxymethylfurfural (HMF) was observed.

KEYWORDS: *green solvents, fractionation, cellulose, hemicellulose, lignin, capillary electrophoresis*

INTRODUCTION

The undiscovered potential of lignocellulosic biomass to obtain a variety of value-added products requires broad research to ensure the feasibility of lignocellulosic biorefineries. One of the major limitations of the biorefinery concept is the lack of an efficient biomass processing tool, which could compromise investment in this sector. Therefore, studies on biomass pretreatment and fractionation were developed to efficiently overcome the recalcitrance of lignocellulose and reduce costs of biorefinery processes.[1−4]

The composition of lignocellulose is mainly ascribed to three general components, namely, cellulose, hemicellulose, and lignin. The isolation of each fraction from lignocellulose is required to achieve maximal valorization of a low-cost feedstock by producing valuable commodities, such as hydroxymethylfurfural (HMF) and levulinic acid (cellulose derivatives),[5,6] furfural and xylitol (hemicellulose derivatives),[7,8] and phenolic compounds and styrenes (lignin derivatives).[9] A new and attractive process that allows cellulose, hemicellulose, and lignin as separated fractions is the pretreatment of biomass using ionic liquids.[10,11]

Ionic liquids (ILs) are usually organic salts with melting points below 100 °C. ILs as design solvents demonstrate a great variety of physicochemical properties. The most common properties of ILs are a high polarity,[12] a great thermal stability (even above 300 °C),[13] a high conductivity and a large electrochemical window, a great solvent power,[14,15] a negligible volatility, and nonflammability.[16] The toxicity and biodegradability of ILs is an important issue, and in the past years these topics were studied extensively.[17,18]

The unique properties of ILs allow their use for biomass processing in pretreatment and/or extraction processes.[19−22] ILs have an ability to dissolve biomass by an effective disruption of the complex network of noncovalent interactions between carbohydrates and lignin.[23,24] Generally, a complete biomass dissolution should be attained during the pretreatment to improve the efficiency of the process. After pretreatment with specific conditions (temperature, residence time, and biomass/IL ratio), an antisolvent is added to the solution mixture, promoting carbohydrate precipitation as a recovered material. Lignin and other soluble compounds are partially extracted to the liquid phase.[25] Lignin can be further recovered by acidification of the antisolvent/IL medium.[26−28] From the regenerated material, hemicellulose and cellulose can be obtained as separated fractions using specific solvents to maximize the fractionation process.[26,27,29,30]

1-Ethyl-3-methylimidazolium acetate ([emim][CH_3COO]) is the most often reported IL due to its good solubility properties for biomass. Nevertheless, there is room for additional investigation of new ILs for biomass pretreatment, especially with a specific goal such as integrated biomass selective fractionation and hydrolysis.[31−33]

In a previous work a wheat straw fractionation using [emim][CH_3COO] was successfully carried out.[26] The aim of the present investigation relies on evaluation of the versatility of the previously developed method[26] using the three different ILs

Received: May 6, 2013
Revised: July 20, 2013
Accepted: July 22, 2013
Published: July 22, 2013

1-butyl-3-methylimidazolium hydrogensulfate ([bmim][HSO_4]), 1-butyl-3-methylimidazolium thiocyanate ([bmim][SCN]), and 1-butyl-3-methylimidazolium dicyanamide ([bmim][HSO_4] depicted in Figure 1. The influence of each

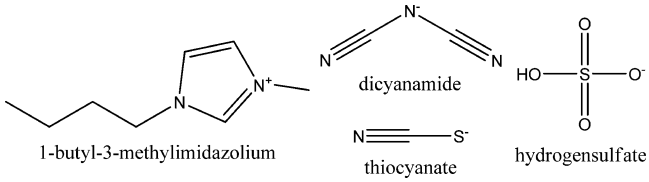

Figure 1. Chemical structures of [bmim] cation and [$N(CN)_2$], [SCN], and [HSO_4] anions used.

IL was verified for the entire process including the pretreatment stage and subsequent fractionation. The effect of ILs on lignin extraction from original biomass and cellulose crystallinity was also studied.

MATERIALS AND METHODS

Materials. ILs selected for this work were [bmim][HSO_4] with 99 mol % purity (5500 ppm of water content), [bmim][SCN] with >98 mol % purity (2100 ppm of water content), and [bmim][$N(CN)_2$] with >98 mol % purity (1500 ppm of water content). The [bmim][SCN] was supplied by Solchemar Lda. (Lisbon, Portugal), and the other ILs were purchased from Iolitec GmbH (Heilbronn, Germany). ILs were dried under vacuum (0.1 Pa) at room temperature for at least 24 h prior to use. The water content of each IL was determined by a volumetric Karl–Fischer titration.

Wheat straw was obtained from Estação Nacional de Melhoramento de Plantas (Elvas, Portugal). The agricultural feedstock was grounded with a knife mill IKA Werke, MF 10 basic (Germany) to particles smaller than 0.5 mm, homogenized in a defined lot, and stored in plastic containers at room temperature. Chemical characterization of wheat straw was previously performed.[34] Moisture content was 8% (w/w).

The following reagents were used for pretreatment experiments: 0.1 M NaOH and 3% (w/w) aqueous solutions prepared from NaOH pellets (99% purity) supplied by Eka Chemicals/Akzonobel (Bohus, Sweden) and 1 and 4 M HCl aqueous solutions prepared from HCl fuming 37% (w/w) with a purity grade for analysis bought from Merck (Darmstadt, Germany). Ethanol 96% (v/v) and acetonitrile of HPLC gradient purity were supplied by Carlo Erba Group (Arese, Italy), and acetone (98% purity) was supplied by Valente & Ribeiro, Lda (Belas, Portugal). For NaOH and HCl solutions distilled water (17 MΩ cm^{-1}) and ultrapure water (18.2 MΩ cm^{-1}) were used, both produced by the PURELAB Classic of Elga system. Paper membranes (Whatman GE Healthcare Bio-Sciences Corp., Piscataway, NJ, USA) and nylon filters, 0.45 μm HNPW (Merck Millipore, Billerica, MA, USA), were used.

Acid-hydrolyzed wheat straw (130 °C, 150 min, and 1.5% H_2SO_4), with a known composition (62.6 wt % glucan, 29.9 wt % lignin, 7.5 wt % ash, and others content) was used as a standard for FTIR calibration curves. FTIR samples were prepared with KBr (≥99% trace metal basis) purchased from Sigma-Aldrich Co. (St. Louis, MO, USA).

IL samples for NMR spectroscopy were prepared using chloroform-D (D, 99.8%) + silver foil (Cambridge Isotope Laboratories, Inc., Andover, MA, USA).

An electrolyte solution of 130 mM NaOH and 36 mM $Na_2HPO_4 \cdot 2H_2O$ was prepared for the capillary electrophoresis trials. Monosaccharides (D-glucose, D-xylose, and L-arabinose), furfural, and 5-hydroxymethylfurfural standard solutions with known concentrations were used to perform a qualitative analysis. Furfural and 5-hydroxymethylfurfural solutions were prepared in a 96% (v/v) ethanol.

Biomass Pretreatment and Fractionation. The methodology used for the wheat straw pretreatment with ILs is described as the optimized method published elsewhere.[26] Pretreatments of wheat straw with the aforementioned ILs were performed at 120 °C for 6 h at constant stirring. A 5% (w/w) biomass/IL ratio was chosen. To regenerate carbohydrate-rich material, 40 mL of 0.1 M NaOH was added to the solution mixture. The solid precipitate was recovered by filtration and kept in an oven at 60 °C for later use. The remaining filtrate (filtrate 1) was used to precipitate dissolved hemicellulose (residual) by adding 96% (v/v) ethanol. The resulting solid was filtered, and ethanol of the filtrate (filtrate 2) was evaporated. The pH of the filtrate was adjusted to 2.0 with HCl aqueous solutions (4 and 1 M) and heated at 70 °C for 30 min to precipitate the lignin-rich material. The remaining filtrate (filtrate 3) was later used for IL recovery.

Hemicellulose was extracted from the dried carbohydrate-rich material with a 3% (w/w) NaOH aqueous solution. Filtration was performed to recover the resulting solid as a cellulose-rich material. A 96% (v/v) ethanol was added to the filtrate (filtrate 4) to precipitate the hemicellulose-rich material, and a new filtration was performed. The remaining filtrate (filtrate 5) was acidified with HCl solutions to recover a residual lignin also extracted with a 3% (w/w) NaOH aqueous solution. All recovered solid samples were placed in the oven at 60 °C for 24 h.

For the IL recovery, filtrate 3 was neutralized with NaOH pellets. Then, water was evaporated and a solid containing NaCl and IL was precipitated. Subsequently, 130 mL of acetonitrile was added to dissolve the IL, leaving NaCl as an insoluble residue, which was removed by the filtration. Acetonitrile was removed under reduced pressure, and the recovered IL was dried under vacuum for at least 24 h.

FTIR Spectroscopy Characterization of Fractionated Samples. For the quantitative analysis, 1.0 mg of obtained carbohydrate-rich samples (regenerated material, hemicellulose-rich, and cellulose-rich materials) was added to 50 mg of KBr. The same procedure was performed for the obtained lignin-rich materials: however, only 0.5 mg of sample was used. Milling occurred during 10 min, and samples were pressed with 8.5 tonnes for 5 min. Analogous methodology of the sample preparation was performed for all examined materials to standardize the analysis and minimize potential analysis errors.

All spectra were scanned using an FTIR spectrometer Spectrum BX, Perkin-Elmer, Inc. (San Jose, CA, USA). The instrument was equipped with a DTGS detector and KBr beam splitter. The operating system used was Spectrum software (version 5.3.1, Perkin-Elmer, Inc.). FTIR spectra were acquired in the region of 4000–400 cm^{-1}, with a total of 64 scans and a resolution of 4 cm^{-1} with a strong apodization. These spectra were subtracted against the background of air spectrum and were recorded as absorbance values.

The quantitative FTIR analysis was performed by the preparation of two calibration curves, one for carbohydrates and one for lignin. As a standard with the known composition of carbohydrate and lignin contents, the acid-hydrolyzed pretreated wheat straw was used, changing the amount for each pellet for the curves' construction. To minimize the error, the pellet samples were scanned three times, and an average value was considered for the calibration curve. After spectra acquisition, the applicability of the Beer–Lambert law was verified for the characteristic regions of carbohydrates and lignin. The quantitative analysis of carbohydrates was performed for the band at 898 cm^{-1}, whereas the lignin content was analyzed through the 1503–1537 cm^{-1} band. The validity of calibration curves was checked regularly before each series of analysis. The sample compositions were determined as carbohydrate (cellulose + hemicellulose) and lignin contents.

NMR Analysis of ILs. ^1H NMR and ^{13}C NMR spectra in $CDCl_3$ solution were recorded on a Bruker ARX 400 MHz spectrometer.

FTIR Measurement of Cellulose Crystallinity. The influence of ILs on cellulose structure was evaluated for regenerated materials and cellulose-rich samples. The two infrared ratios were calculated, namely, lateral order index (LOI; $A_{1437cm^{-1}}/A_{898cm^{-1}}$)[35] and total crystallinity index (TCI; $A_{1376cm^{-1}}/A_{2900cm^{-1}}$).[36] Total height was taken for calculations.

Capillary Electrophoresis (CE) Analysis. Capillary electrophoresis was used to evaluate the existence of carbohydrate hydrolysate in filtrate 1 for the process with [bmim][HSO_4]. The

dx.doi.org/10.1021/jf401980p | J. Agric. Food Chem. 2013, 61, 7874–7882

Table 1. Results of Wheat Straw Pretreatment with [bmim][SCN], [bmim][N(CN)$_2$], and [bmim][HSO$_4$]: Regeneration, Liquid Stream Fractionation, and IL Recovery

			SF[a]	LF[b]		
entry	WS,[c] mg	dried WS, mg	RY,[d] % (w/w)	hemicellulose-rich,[e] mg	lignin-rich, mg	IL recovery, % (w/w)
[bmim][SCN]	250.3	230.3	79.5	8.0	13.0	93.5
[bmim][N(CN)$_2$]	250.7	230.6	77.7	15.2	10.1	97.0
[bmim][HSO$_4$]	250.3	230.3	59.6	4.1	7.1	92.1

[a]SF, solid fraction. [b]LF, liquid fraction. [c]WS, wheat straw. [d]RY, regeneration yield. [e]Residual.

employed methodology was adapted from the literature.[37] Separations of standards and samples were performed with an Agilent Technologies CE system (Waldbronn, Germany), equipped with a diode array detector (DAD), ChemStation data software, and a fused-silica uncoated 50 μm i.d. and 56/64.5 cm effective length capillary, also from Agilent. Monitoring was carried out using direct UV detection at wavelengths of 210 and 270 nm with a bandwidth of 10 nm. Samples filtered through a 0.45 μm membrane filter were injected directly under a pressure of 35 mbar for 6 s at the anode (+) of the CE system. The separation voltage was raised linearly within 1 min from 0 to 17 kV. The capillary was preconditioned between runs by flushing with an electrolyte solution for 5 min.

Experimental Error Analysis. Standard deviation errors (u) were calculated for all obtained results. The applied temperature in pretreatment experiments demonstrated a $u(T)$ = 1 °C. All mass determinations were performed using a Mettler Toledo XS205 dual-range scale (Germany) with a given $u(m)$ = 0.1 mg. Pretreatment errors were given as total loss materials for each experiment. An arbitrary error of 5% of the experimental value was considered for the FTIR quantitative analysis.

RESULTS AND DISCUSSION

Pretreatment of Wheat Straw. The 6 h pretreatment of wheat straw with three 1-butyl-3-methylimidazolium ILs at 120 °C was investigated. The use of [bmim][SCN] and [bmim][N(CN)$_2$] did not allow for a complete dissolution of wheat straw, contrary to [bmim][HSO$_4$], with which a macroscopic complete dissolution was determined. Similar partial biomass dissolution with [bmim][N(CN)$_2$] was already obtained.[38] In the case of [bmim][SCN], its ability to dissolve monosaccharides and sugar alcohols[39] as well as cellulose macromolecule was underscored.[40] The [bmim][SCN] was found to be incapable of dissolving wheat straw completely. This feature could be attributed to the complexity and intricate network of wheat straw material, which [bmim][SCN] is ineffective to disrupt.

Among tested ILs, [bmim][HSO$_4$] was found to be the most efficient in the dissolution process. It is worth mentioning that this IL apart from the others studied is characterized as a Brønsted acidic IL.[41] A strong basic anion of [emim][CH$_3$COO] was already reported as being responsible for a complete dissolution achieved at the same conditions as reported in this work.[26] This means that the dissolution process is certainly performed in a different manner by [emim][CH$_3$COO] and [bmim][HSO$_4$].

In the regeneration process a brown and flocculated material was precipitated after a 0.1 M NaOH addition for experiments with [bmim][SCN] and [bmim][N(CN)$_2$]. These physical and morphological properties are generally noticeable for the regenerated material after the biomass pretreatment using ILs.[20,42] However, in the case of [bmim][HSO$_4$], a black viscous solid was formed. A similar effect was already discovered for the rice straw pretreatment with [emim][HSO$_4$].[20] With the regeneration yields obtained from experiments with [bmim][SCN] and [bmim][N(CN)$_2$] taken into consideration, it can be stated that high regeneration yields (79.5 and 77.7%) were achieved. However, due to an antisolvent used, only the carbohydrate fraction was precipitated (62 wt % carbohydrate content in dried wheat straw). This can be explained by the fact that only partial dissolution in these ILs was noted, where the regenerated material should be mainly composed of insoluble biomass. In the case of [bmim][HSO$_4$], the regeneration yield was determined to be 59.6% as depicted in Table 1. This yield seems to be in good agreement with results found in the literature when a 0.1 M NaOH was used as an antisolvent for sugar cane pretreatment with [emim][Abs] (1-ethyl-3-methylimidazolium alkylbenzenesulfonate), where 46−55% of biomass regeneration was attained.[21] Rice straw pretreatment with cholinium lysinate gave 55.9% of regenerated material.[43] Previously,[26] we sought to obtain from 57.5 to 62.9% (w/w) of regenerated material with [emim][CH$_3$COO] at the same conditions as reported here.

The complete dissolution of wheat straw shows that the most efficient disruption of biomass occurred with [bmim][HSO$_4$]. After the regeneration process, fractionation of the resulting liquid stream and regenerated material was performed. Nevertheless, it should be emphasized that large differences in results were observed for the [bmim][HSO$_4$]-assisted experiment in comparison to other IL-assisted pretreatments.

On the basis of the increasing amount of lignin-rich content in the regenerated material, the ILs can be placed in the following order: [bmim][SCN] > [bmim][N(CN)$_2$] > [bmim][HSO$_4$], demonstrating the best capability of [bmim][SCN] to obtain 13.0 mg of the lignin-rich content material. The highest amount of residual hemicellulose was obtained in the process with dicyanamide IL, which is almost 100% more than in the case of thiocyanate IL and almost 300% more than that of hydrogensulfate IL.

All studied ILs were recovered with a high yield, exceeding significantly 90% of the initial mass used.

Results of the regenerated material fractionation containing cellulose-rich, hemicellulose-rich, and residual lignin-rich materials obtained with the three ILs are presented in Table 2.

Table 2. Results of Wheat Straw Pretreatment with [bmim][SCN], [bmim][N(CN)$_2$], and [bmim][HSO$_4$]: Fractionation of the Regenerated Solid Material

	regenerated material fractionations		
entry	cellulose-rich, % (w/w)	hemicellulose-rich, % (w/w)	lignin-rich,[a] % (w/w)
[bmim][SCN]	83.4	11.7	2.8
[bmim][N(CN)$_2$]	75.8	5.5	7.6
[bmim][HSO$_4$]	58.9	1.2	24.5

[a]Residual.

[bmim][SCN] allowed the highest quantities of carbohydrate fractions (95.1%). The highest recovery of the residual lignin-rich material (24.5%) was verified for the [bmim][HSO_4] process.

Biomass Fractionation from [bmim][SCN] and [bmim][$N(CN)_2$] Processes. The biomass fractionation results demonstrate a higher recovery of dissolved biomass from the liquid stream for [bmim][$N(CN)_2$], where total material of 25.3 mg (lignin-rich plus residual hemicellulose-rich materials) was obtained, when compared to the use of [bmim][SCN]. Therefore, higher biomass conversion and/or degradation can be assumed during the pretreatment with [bmim][SCN], in which highly soluble compounds (low molecular weight) are formed, impairing the recovery.[42,44] Furthermore, the fractionation of regenerated materials allows a higher amount of the cellulose-rich material to be recovered in both [bmim][SCN] (83.4%) and [bmim][$N(CN)_2$] (75.8%) experiments, compared to hemicellulose and lignin fractions. The recovery of hemicellulose-rich fractions was found to be low in both ILs. Such a low value can be a consequence of an incomplete dissolution of biomass in these ILs. In other studies, higher quantities of hemicellulose were obtained due to better fractionation of the carbohydrate-rich fraction.[26,29] Nevertheless, the achieved results indicate that between these two ILs, [bmim][SCN] is a more efficient IL for the biomass fractionation as a larger amount of each fraction (cellulose-, hemicellulose-, and lignin-rich materials) was recovered.

FTIR qualitative analysis of the obtained samples from pretreatments with [bmim][SCN] and [bmim][$N(CN)_2$] demonstrated similarities with the previously presented data for [emim][CH_3COO].[26] Basically, for all of the carbohydrate spectra (regenerated material, cellulose, and hemicellulose spectra) slight differences were observed in the carbohydrate absorption characteristic region in comparison to those presented elsewhere,[26] indicating that hemicellulose and cellulose contents vary in these samples. An interesting fact is that ester bonds between hemicellulose and lignin in the regenerated material from the [bmim][SCN] experiment are still present (presence of a band at 1734 cm^{-1}). Partial biomass dissolution occurring for this IL during pretreatment is responsible for the presence of such bonds. Moreover, [bmim][$N(CN)_2$] seems to contaminate the regenerated material as the presence of the C−N stretching vibration characteristic of $N(CN)_2^-$ is observed at the 2146 cm^{-1} band. Possibly an extended washing step for the regenerated material should be performed in the process with [bmim][$N(CN)_2$]. An analogous problem did not occur in the [SCN] IL, as this band was not detected. With respect to both cellulose rich-materials, bands at 998 and 1235 cm^{-1} were almost undetectable in comparison to the regenerated material spectra. This indicates that celluloses obtained by pretreatments with [bmim][SCN] and [bmim][$N(CN)_2$] are arabinan (998 cm^{-1}) and syringyl lignin type (1235 cm^{-1}) free. The spectra of hemicellulose-rich samples do not exhibit significant differences, contrary to the data presented in the literature.[45,46]

Spectra of lignin samples (including residual lignin) obtained from the [bmim][SCN] experiment show only characteristic bands of lignin materials. However, for the lignin sample (residual lignin) of the [bmim][$N(CN)_2$] assay, a high contamination of carbohydrates, namely, hemicelluloses (897, 1041, and 1079 cm^{-1}), is observed. Furthermore, the presence of a band at 1734 cm^{-1} was observed, indicating the ester linkages between hemicellulose and lignin. Therefore, [bmim][$N(CN)_2$] is demonstrated to be inefficient in separating hemicellulose from lignin during pretreatment.

Biomass Fractionation from [bmim][HSO_4] Process. A different behavior was observed for [bmim][HSO_4] compared to the previously discussed ILs. After regeneration of the carbohydrate-rich fraction, only small quantities of lignin-rich and residual hemicellulose-rich materials were recovered from the fractionation of the liquid stream, despite the macroscopic complete dissolution achieved by [bmim][HSO_4]. The pH of the liquid stream containing the IL was determined to be 1.3, even with the addition of a basic 0.1 M NaOH solution that usually should oscillate at a very basic region (pH 12), as determined for other IL solutions. Therefore, this may suggest that due to the acidic property of [HSO_4], acid hydrolysis of lignocellulosic biomass during pretreatment is facilitated.[47] In classical acid hydrolysis, the liquid stream is normally enriched in hydrolyzed hemicelluloses (monosaccharides and oligosaccharides) and the solid material contains mostly cellulose and lignin.[48] As already referred, a small quantity of the lignin-rich fraction was recovered in the liquid stream, extracted with water during the washing step of the regenerated material (neutral pH). A negligible amount of the hemicellulose-rich material was also obtained as a nonhydrolyzed hemicellulose.

Fractionation of the regenerated material into cellulose, hemicellulose, and residual lignin-rich materials in [bmim][HSO_4] gave the results collected in Table 2. The obtained results demonstrate that a regenerated material is essentially fractionated into cellulose- and lignin-rich (24.5%) fractions. Only 1.2% (w/w) of the regenerated material was fractionated into hemicellulose. These results confirm the occurrence of the acid hydrolysis phenomenon, as an insignificant quantity of hemicellulose was extracted from the regenerated material.

In fact, the hydrolytic activity associated with [bmim][HSO_4] can be explained by the high electron attraction of a weak conjugate [HSO_4] base over the [bmim] cation, allowing the hydrolysis of the β-glycosidic bonds. To effectively confirm the existence of the hemicellulose hydrolysis, the FTIR spectrum of the regenerated material was compared to the spectrum of the acid-hydrolyzed wheat straw, as shown in Figure 2. The regenerated material spectrum (red line in Figure 2) confirms the presence of cellulose and lignin in the analyzed samples. In the carbohydrate characteristic region, sharp and characteristic

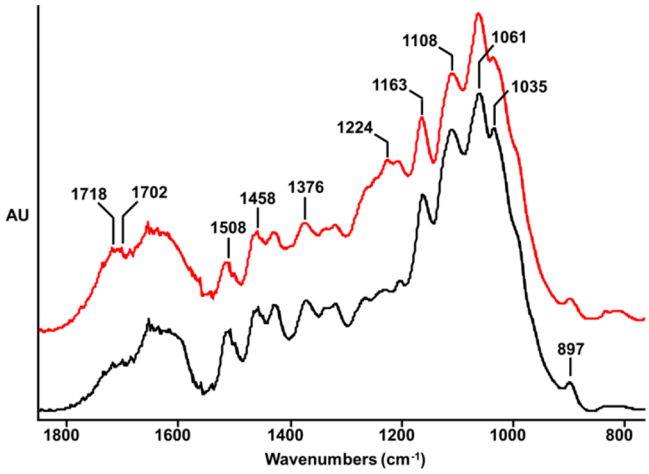

Figure 2. FTIR spectra of the regenerated material obtained in the process with [bmim][HSO_4] (red) and an acid-hydrolyzed wheat straw (black).

dx.doi.org/10.1021/jf401980p | J. Agric. Food Chem. 2013, 61, 7874−7882

absorption vibrations of cellulose were observed (897, 1035, 1061, 1108, and 1163 cm^{-1}). The lignin content is relatively high, because absorption bands at 1458, 1508, 1702, and 1718 cm^{-1} are present with high intensities. Indeed, the spectrum of the regenerated material is practically equivalent to that observed for the acid-hydrolyzed sample (black line in Figure 2), although the band at 1224 cm^{-1} abnormally appears as a strong absorption. It is expected that this strong absorption can be a contribution of the band at 1251 cm^{-1} generally noticeable for hemicellulose (acetyl groups), which emphasizes only a partial hydrolysis of hemicellulose during the pretreatment with [bmim][HSO_4], confirming literature reports.[31−33]

The spectrum of the cellulose-rich fraction obtained in this process demonstrates a high-purity sample. The almost extinguished bands of lignin depict this assumption. Furthermore, the fractionation provides an impressive quantity of the "residual" lignin-rich material from a 24.5% regenerated material. Therefore, it can be stated that cellulose and lignin can easily be separated from the regenerated material. However, a qualitative analysis showed that lignin is a major constituent, despite the presence of contaminants. Nonetheless, the obtained results confirm that vast lignin content in wheat straw was not extracted in the liquid stream after the regeneration process as occurred in pretreatments with other ILs, but it was extracted from the regenerated material, as happened in the acid hydrolysis process.

After the regeneration process, analysis of the liquid stream was performed by the CE technique to identify the compounds resulting from the hemicellulose hydrolysis. The results indicate that only furfural and HMF are present, instead of monosacharides, such as xylose or glucose. The presence of furfural and HMF is due to the degradation process of xylose and glucose, correspondingly. Thus, it is possible to infer that an extended hydrolysis of carbohydrates to degradation products during pretreatment with [bmim][HSO_4] occurred. Therefore, a 6 h pretreatment of wheat straw using [bmim]-[HSO_4] at 120 °C can be considered as harsh conditions because only degradation products were obtained after hydrolysis. In other studies, a 2−5 min pretreatment of corn stalk at 100 °C with [bmim][HSO_4] and (1-(4-sulfobutyl)-3-methylimidazolium hydrogensulfate ([Sbmim][HSO_4]) was applied, achieving 23 and 15% total reducing sugars, respectively.[47] Miscanthus was also pretreated with [bmim]-[HSO_4] mixed with water at 120 °C within 2−22 h. The same trend of the carbohydrate degradation into furfural and HMF was also indicated for long pretreatment times.[33] Herein, the used conditions allowed us to obtain a very extended reaction as no monosaccharides were detected in the CE, which is a highly sensitive technique for the detection of neutral sugars.[37,49] Therefore, mild conditions seem to be more adequate to obtain the hemicellulose hydrolysis to monosaccharides without chemical degradation.

Purity and Composition of Obtained Fractions. The main chemical bond vibrations of lignocellulosic materials are identified in the region of 1800−800 cm^{-1}. Absorption bands at 1161, 1112, and 897 cm^{-1} are attributed to both carbohydrates cellulose and hemicellulose. Cellulose characteristic bands are also assigned to 1376, 1061, and 1035 cm^{-1}. Bands at 1251, 1046, and 996 cm^{-1} can be identified as solely characteristic for hemicellulose.[50−52] Characteristic bands of lignin were observed at 1718, 1702, 1654, 1508, 1458, 1420, 1597, 1261, 1242, 1224, 1127, and 840 cm^{-1}.[53,54] An additional band, such as the vibration band at 1734 cm^{-1}, can be assigned. This band is related to ester-linked groups between hemicellulose and lignin[55] and is generally observed in the spectra of the untreated lignocellulosic biomass. All of the aforementioned bands are characterized in detail elsewhere.[26]

FTIR spectra of the obtained postfractionation samples from the pretreatments with [bmim][SCN], [bmim][$N(CN)_2$] and [bmim][HSO_4] were analyzed. Spectra of the regenerated material obtained from pretreatments with [bmim][SCN] and [bmim][$N(CN)_2$] demonstrated chiefly carbohydrate absorption bands. Slight differences in the region of 1112−896 cm^{-1} observed are dictated by different ratios of the carbohydrate content (cellulose/hemicellulose) in each sample. For the regenerated material from [bmim][SCN] experiment, the band at 1734 cm^{-1} is still observed, although characterized by a lower intensity. Furthermore, an additional band appeared at 2146 cm^{-1} that corresponds to the C−N stretching vibration of [bmim][$N(CN)_2$]. The regenerated material obtained from the pretreatment with [bmim][HSO_4] shows different characteristics. The spectrum demonstrated a well-defined carbohydrate region, showing that cellulose is a major type of carbohydrate present. The presence and the shape of the characteristic bands at 897, 1035, 1061, 1108, 1163, and 1376 cm^{-1} suggest this evidence. Lignin bands are also found (1224, 1458, 1508, 1702, and 1718 cm^{-1}) with a relatively high absorption, which are unexpected for a spectrum of the carbohydrate rich-material.

Spectra of the fractionated cellulose-rich materials from [bmim][SCN] and [bmim][$N(CN)_2$] pretreatments were nearly the same as the characteristic cellulose spectrum obtained in a previous study,[26] with a practical disappearance of the arabinan characteristic band at 998 cm^{-1} and the lignin band at 1235 cm^{-1}. In the [bmim][HSO_4] experiment, the spectrum showed almost complete absence of the lignin component in this sample. The band at 1420 cm^{-1} disappeared, and bands at 1458 and 1508 cm^{-1} are almost extinguished. The lignin absorption bands at 1235 and 1260 cm^{-1} were also very small. The presence of 1718 and 1734 cm^{-1} absorptions indicates the presence of lignin and also hemicellulose as small characteristic bands.

FTIR spectra of hemicellulose-rich materials obtained for [bmim][SCN] and [bmim][$N(CN)_2$] are very similar, with the only difference being the shift of bands from the region of 1112−896 cm^{-1}.

Samples of the residual-lignin material from [bmim][N-$(CN)_2$] and [bmim][HSO_4] experiments demonstrated considerable differences. Lignin absorption bands at 835, 1127, 1031, 1420, 1458, 1654, 1701, and 1718 cm^{-1} were observed for both samples. However, the spectrum obtained for the pretreatment with [bmim][$N(CN)_2$] shows clearly that the residual lignin-rich material was recovered with a high carbohydrate content (carbohydrate bands at 897, 1041, and 1079 cm^{-1}). The absorption band at 1734 cm^{-1} indicates also the presence of hemicellulose that remained linked to lignin.

A quantitative analysis based on FTIR measurements was performed for each recovered solid sample to evaluate the composition and to verify the selectivity of the used ILs, as well as the versatility of the fractionation method. The composition of each phase was determined as carbohydrate (cellulose + hemicellulose) and lignin content, and the percentage of other nonidentified material was calculated by difference. Both cellulose and hemicellulose fractions contain more carbohydrates than raw biomass. Lignin obtained from the liquid fraction exhibits an elevated purity as high as 89 wt %. Results obtained with the use of [bmim][SCN] are shown in Figure 3,

and those for [bmim][$N(CN)_2$] and [bmim][HSO_4] are collected in Tables 3 and 4, respectively.

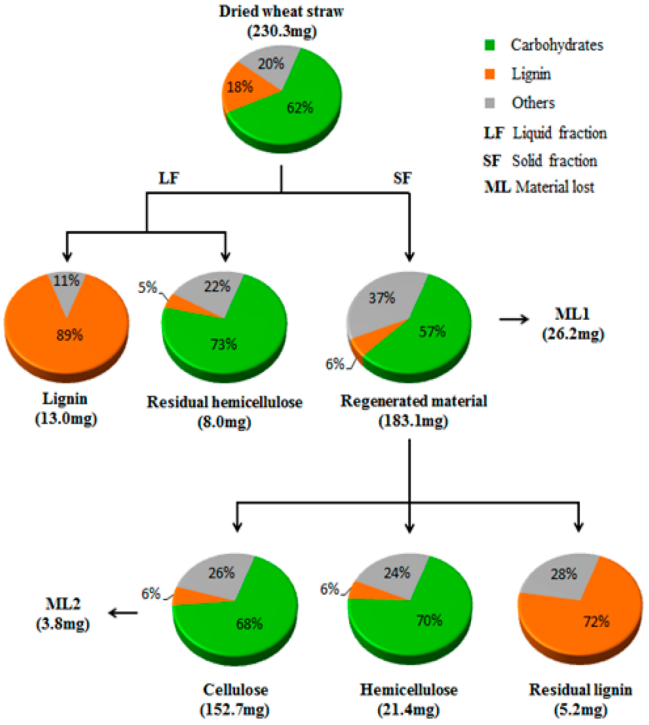

Figure 3. Fractionation of wheat straw using [bmim][SCN].

Pretreatment with [bmim][SCN] allowed us to produce the regenerated material with a carbohydrate content similar to that of the raw material (57 and 62 wt %, respectively) with a 3 times lower concentration of lignin. After fractionation, cellulose and hemicellulose fractions were enriched in the carbohydrate content and reduced in lignin compared to the original biomass or the regenerated solid fraction. The lignin-rich materials obtained did not contain carbohydrates, revealing a lignin content ranging from 72 to 89 wt % depending on the fraction.

Pretreatment with [bmim][$N(CN)_2$] provided a regenerated material with 68 wt % of the carbohydrate content and 11 wt % of the lignin composition. The percentage of other compounds present changed insignificantly when compared to the original dried biomass. The cellulose-rich material was enriched in carbohydrates, achieving 87 wt %. A carbohydrate content of 85 wt % was also quantified for the hemicellulose-rich sample.

In the residual hemicellulose-rich sample, low carbohydrate content was detected due to a large impact of other compounds. Furthermore, as presented in Table 3, the residual lignin is contaminated by hemicellulose and it is constituted by a 1:1 lignin/carbohydrate composition. As aforementioned, these data can be explained by only a partial dissolution of biomass in this IL.

The data depicted in Table 4 obtained from the [bmim][HSO_4] experiment demonstrated a different manner of pretreatment and fractionation of biomass. In the liquid stream small quantities of lignin-rich and residual hemicellulose-rich materials obtained impeded the performance of the FTIR quantitative analysis. The cellulose-rich material shows a 90 wt % carbohydrate content, the most enriched carbohydrate sample achieved in all performed pretreatments in this work. The major lignin fraction is obtained as a "residual" lignin-rich material, once a small quantity was extracted in the liquid phase after the regeneration process. Such an occurrence is related to the similarity to the acid hydrolysis observed for the process with [bmim][HSO_4]. However, only a 62 wt % lignin content was determined for this sample even with no trace of carbohydrates. For the hemicellulose-rich material, an insignificant quantity was recovered; thus, [bmim][HSO_4] catalyzes the hemicellulose hydrolysis and, therefore, a major hemicellulose content was degraded to furfural and dissolved in the liquid stream during the pretreatment process.

Influence of ILs on Pretreatment, Lignin Removal, and Cellulose Crystallinity. Pretreatment of biomass with different ILs shows that each IL can be dedicated to originating different fractions characterized by a high purity. Still, it is important to point out that a major influence of ILs on biomass pretreatment can be associated with the purity/quantity of the regenerated material obtained for each process. Compared to the previously published results,[26] it can be concluded that [emim][CH_3COO] demonstrates the best performance in the regeneration of carbohydrates (81 wt % carbohydrate content) among the ILs tested in this work. However, only 57% (w/w) of the original biomass was recovered, whereas in the case of the other ILs such values varied from 60 to 80% (w/w), although a lower content of carbohydrates is associated. Nevertheless, the results show that each regenerated material had a similar or higher amount of carbohydrates recovered relative to raw wheat straw and is also accompanied by the decreased concentration of lignin.

Herein, a similar effect can be attributed to [emim][CH_3COO], [bmim][SCN], and [bmim][$N(CN)_2$] regarding the extent of lignin removal from the original biomass. The strong basicity of anions allows for the extraction of lignin from

Table 3. FTIR Quantification of Fractionated Samples Using [bmim][$N(CN)_2$]

		composition						
		carbohydrates		lignin		others		
entry		mg	wt %	mg	wt %	mg	wt %	total, mg
dried wheat straw		143.0	62	41.5	18	46.1	20	230.6
RM[a]		121.9	68	19.7	11	37.7	21	179.3
solid fraction	cellulose	102.4	87	9.4	8	5.9	5	117.7
	hemicellulose	7.2	85	0.8	9	0.5	6	8.5
	lignin[b]	4.5	38	4.5	38	2.8	24	11.8
liquid fraction	lignin	NQ[c]	NQ	NQ	NQ	NQ	NQ	10.1
	hemicellulose[b]	8.4	55	0.2	1	6.6	44	15.2

[a]RM, regenerated material. [b]Residual. [c]NQ, not quantified.

dx.doi.org/10.1021/jf401980p | J. Agric. Food Chem. 2013, 61, 7874−7882

Table 4. FTIR Quantification of Fractionated Samples Using [bmim][HSO_4][a]

		composition						
		carbohydrates		lignin		others		
entry		mg	wt %	mg	wt %	mg	wt %	total, mg
dried wheat straw		142.8	62.1	41.5	18.1	46.0	20.0	230.3
RM[b]		87.7	63.9	27.4	20.0	22.0	16.1	137.1
solid fraction	cellulose	66.6	90.0	2.2	3.1	5.2	7.0	74.0
	hemicellulose	NQ[c]	NQ	NQ	NQ	NQ	NQ	1.5
	lignin[d]	0.0	0.0	19.1	62.0	11.7	38.0	30.8

[a]Liquid fraction was not quantified. [b]RM, regenerated material. [c]NQ, not quantified. [d]Residual.

the liquid solution during the pretreatment. In the case of [bmim][HSO_4], an increased lignin content in the regenerated material is observed as a consequence of the acidic property of the IL that not only promotes the hydrolysis phenomenon but also avoids lignin extraction to the liquid stream.

There is a correlation between lignin removal and crystallinity reduction of cellulose.[25] FTIR crystallinity indices LOI and TCI demonstrate a slight decrease of cellulose crystallinity after pretreatment with the tested ILs. LOI is given by a ratio between 1427 cm^{-1} (CH_2 scissoring motion) and 898 cm^{-1} bands. TCI is given by a ratio between 1376 and 2900 cm^{-1} (CH and CH_2 stretching). Pretreatment experiments allowed us to decrease slightly the LOI of cellulose in the fractionated samples (regenerated materials and cellulose-rich fractions) in comparison to the native and acid-hydrolyzed wheat straw and to the standard cellulose. For regenerated materials TCI was observed to have a relevant decrease, whereas for cellulose-rich samples an insignificant change was verified (Table 5).

Table 5. Crystallinity Indices of Regenerated Materials and Cellulose-Rich Fractions Obtained in Pretreatment Experiments as well as Native and Acid-Hydrolyzed Wheat Straws and a Standard Cellulose

		crystallinity index	
entry		LOI[a]	TCI[b]
[bmim][SCN]	RM[c]	1.65	1.03
	cellulose	1.59	1.09
[bmim][$N(CN)_2$]	RM	1.58	1.05
	cellulose	1.54	1.08
[bmim][HSO_4]	RM	1.55	1.01
	cellulose	1.55	1.12
wheat straw	untreated	1.74	1.13
	acid hydrolyzed	1.68	1.07
cellulose	standard	1.69	1.11

[a]LOI, lateral order index. [b]TCI, total crystallinity index. [c]RM, regenerated material.

The highest reduction of cellulose crystallinity could be related to the complete dissolution achieved with [bmim][HSO_4]. For [bmim][SCN] and [bmim][$N(CN)_2$], the ability to reduce the cellulose crystallinity could be compromised by the incomplete biomass dissolution observed for these ILs.

NMR Analysis of ILs. The purity of ILs before and after pretreatments was verified using ^1H and ^{13}C NMR techniques, revealing that pretreatment did not alter the NMR spectra of used ILs. The determined chemical shifts are as follows: [bmim][SCN] ^1H NMR (400 MHz; $CDCl_3$) δ 0.94 (t, 3H, CH_3CH_2); 1.38 (m, 2H, $CH_3CH_2CH_2$); 1.90 (m, 2H, $CH_2CH_2CH_2$); 4.07 (s, 3H, NCH_3); 4.30 (t, 2H, NCH_2); 7.52 (d, 2H, NCHCHN); 9.27 (s, 1H, NCHN). ^{13}C NMR ($CDCl_3$) δ 13.44 (CH_3CH_2); 19.47 ($CH_3CH_2CH_2$); 32.06 ($CH_2CH_2CH_2$); 35.72 (NCH_3); 50.09 (NCH_2); 122.40 (NCHCHN); 123.84 (NCHCHN); 136.56 (NCHN). [bmim][$N(CN)_2$] ^1H NMR (400 MHz; $CDCl_3$) δ 0.95 (t, 3H, CH_3CH_2); 1.36 (m, 2H, $CH_3CH_2CH_2$); 1.88 (m, 2H, $CH_2CH_2CH_2$); 4.00 (s, 3H, NCH_3); 4.22 (t, 2H, NCH_2); 7.41 (d, 2H, NCHCHN); 9.17 (s, 1H, NCHN). ^{13}C NMR ($CDCl_3$) δ 13.28 (CH_3CH_2); 19.35 ($CH_3CH_2CH_2$); 31.85 ($CH_2CH_2CH_2$); 36.47 (NCH_3); 49.94 (NCH_2); 122.28 (NCHCHN, $N(CN)_2$); 123.68 (NCHCHN); 136.33 (NCHN). [bmim][HSO_4] ^1H NMR (400 MHz; D_2O) δ 0.75 (t, 3H, CH_3CH_2); 1.15 (m, 2H, $CH_3CH_2CH_2$); 1.66 (m, 2H, $CH_2CH_2CH_2$); 3.72 (s, 3H, NCH_3); 4.03 (t, 2H, NCH_2); 7.28 (d, 2H, NCHCHN); 8.54 (s, 1H, NCHN). ^{13}C NMR (D_2O) δ 12.55 (CH_3CH_2); 18.67 ($CH_3CH_2CH_2$); 31.19 ($CH_2CH_2CH_2$); 35.54 (NCH_3); 49.20 (NCH_2); 122.13 (NCHCHN); 123.40 (NCHCHN); 135.77 (NCHN).

Capillary Electrophoresis Analysis. As aforementioned almost no hemicellulose was recovered in the process with [bmim][HSO_4]. Therefore, a CE technique was used to verify the existence of hemicellulose hydrolysis mediated by [bmim][HSO_4]. A sample of filtrate 1 from the process with [bmim][HSO_4] was analyzed and compared to pure [bmim][HSO_4] as well as to glucose, xylose, arabinose, furfural, and HMF standards. Electropherograms of all samples were acquired with a UV direct detection at 210 and 270 nm for 30 min. Results demonstrating the examined sample, pure [bmim][HSO_4], furfural, and HMF standards are shown in Figure 4. At 210 nm the signals for pure [bmim][HSO_4] were detected at 5.6 and 10.6 min migration times with regard to $[bmim]^+$ and $[HSO_4]^-$, correspondingly. The sample demonstrates not only the presence of [bmim][HSO_4] detected at 210 nm but also other compounds that are identified at 270 nm. The latter are not related to IL because this is not detected at 270 nm. Furfural (11.0 min) and HMF (12.4 min) were identified as being present in the sample by comparison to the authentic standard solutions at 270 nm. Moreover, co-injection of furfural and HMF standard solutions with the sample was tested, and signals at the same migration times with increased absorption areas were identified. Monosaccharides, such as glucose, xylose, and arabinose, were not detected in the sample.

Despite preliminary reports on biomass dissolution and hydrolysis with [bmim][HSO_4],[33,47] [bmim][HSO_4] was tested in this investigation to evaluate a subsequent fractionation, which could be advantageous due to its inherent acidic properties. Regardless of a relatively low solubility of carbohydrates and wood biomass in [bmim][$N(CN)_2$],[15,38] the effect of this IL on biomass pretreatment was also studied

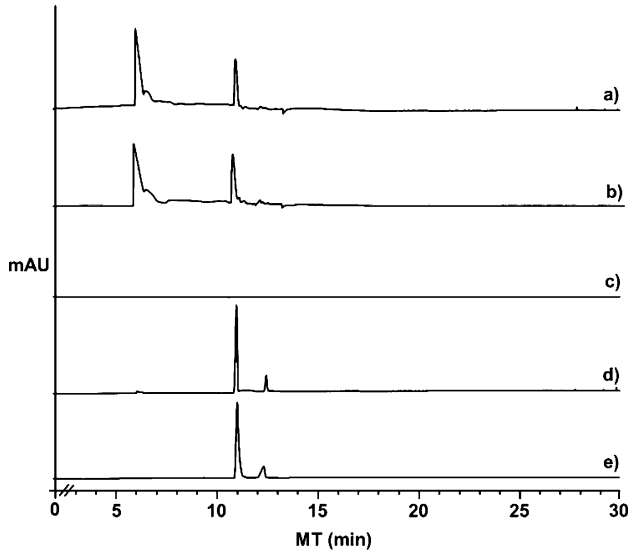

Figure 4. Electropherograms showing the profile for (a) diluted pure [bmim][HSO_4] (1/5) at 210 nm, (b) sample of filtrate 1 from the process with [bmim][HSO_4] at 210 nm, (c) diluted pure [bmim]-[HSO_4] (1/5) at 270 nm, (d) furfural and HMF identified in the sample at 270 nm, and (e) furfural and HMF standard solutions at a wavelength of 270 nm. See text for CE separation conditions.

to verify the synergetic effect of the biomass matrix on the dissolution in this IL. Additionally, [bmim][SCN] was selected to be tested in this method due to promising solubilities of carbohydrates in this IL.[39] To the best of our knowledge, this is the first study of the biomass pretreatment where [SCN]-based IL is applied.

IL Recovery. From an evaluation of the IL recovery of tested ILs, it can be concluded that the established recovery process was sufficient for the examined ILs. The obtained IL recovery yields were high, exceeding 90% (w/w) of the initial IL mass used. Special attention should be drawn to the [bmim][$N(CN)_2$] experiment that allowed a 97% (w/w) IL recovery to be achieved.

In conclusion, [bmim][SCN] and [bmim][$N(CN)_2$] demonstrate only a partial dissolution of biomass, whereas complete dissolution was attained for [bmim][HSO_4]. By using [bmim]-[SCN] and [bmim][$N(CN)_2$] in wheat straw pretreatment, the biomass recovery and the purity of each fractionated sample can be tuned, demonstrating a great flexibility of the developed process. Pretreatment with [bmim][SCN] can be used to produce a high-purity lignin, whereas [bmim][$N(CN)_2$] is more adequate to perform a more selective recovery of carbohydrate materials such as cellulose and hemicellulose fractions.

A different behavior was observed for [bmim][HSO_4], which shows a great capability to hydrolyze hemicellulose; thus, an extended degradation was detected for this process. It can be assumed that optimized conditions allow hemicellulose hydrolysis without degradation to furfural and HMF to be performed.

After pretreatment, ILs were recovered up to 97% (w/w) of the initial mass. Therefore, the developed process is advantageous as a high IL recovery yield is obtained for all tested ILs.

AUTHOR INFORMATION

Corresponding Author

*(R.B.-Ł.) Phone: + 351 210924600, ext. 4224. Fax: + 351 217163636. E-mail: rafal.lukasik@lneg.pt.

Funding

This work was supported by the Fundação para a Ciência e a Tecnologia (FCT, Portugal) through Grants SFRH/BPD/26356/2006, PEst-C/EQB/LA0006/2011, and Programme Ciência 2008 and by LNEG through financing of the ERA-IB project (Products from Lignocelluloses-EIB.10.013).

Notes

The authors declare no competing financial interest.

REFERENCES

(1) Dale, B. E. Process for the treatment of lignocellulosic biomass. U.S. Patent 20,120,325,202, 2012.

(2) Holtzapple, M. T.; Davison, R. R.; Lowery, L. L., Jr.; Granda, C. B. Methods and systems for pretreatment and processing of biomass. U.S. Patent 20,120,258,509, 2012.

(3) Galbe, M.; Zacchi, G. Pretreatment: the key to efficient utilization of lignocellulosic materials. *Biomass. Bioenergy* **2012**, *46*, 70−78.

(4) Mtui, G. Y. S. Recent advances in pretreatment of lignocellulosic wastes and production of value added products. *Afr. J. Biotechnol.* **2009**, *8*, 1398−1415.

(5) Rinaldi, R.; Schüth, F. Acid hydrolysis of cellulose as the entry point into biorefinery schemes. *ChemSusChem* **2009**, *2*, 1096−1107.

(6) Zakrzewska, M. E.; Bogel-Lukasik, E.; Bogel-Lukasik, R. Ionic liquid-mediated formation of 5-hydroxymethylfurfural − a promising biomass-derived building block. *Chem. Rev.* **2011**, *111*, 397−417.

(7) Binder, J. B.; Blank, J. J.; Cefali, A. V.; Raines, R. T. Synthesis of furfural from xylose and xylan. *ChemSusChem* **2010**, *3*, 1268−1272.

(8) da Silva, S. S.; Chandel, A. K. *D-Xylitol: Fermentative Production, Application and Commercialization*; Springer: Berlin, Germany, 2012.

(9) Holladay, J. E.; Bozell, J. J.; White, J. F.; Johnson, D. Top value-added chemicals from biomass. Volume II − Results of Screening for Potential Candidates from Biorefinery Lignin. DOE Report PNNL-16983, 2007; http://chembioprocess.pnl.gov/staff/staff_info.asp.

(10) Brandt, A.; Gräsvik, J.; Hallett, J. P.; Welton, T. Deconstruction of lignocellulosic biomass with ionic liquids. *Green Chem.* **2013**, *15*, 550−583.

(11) da Costa Lopes, A. M.; João, K.; Morais, A. R. C.; Bogel-Lukasik, E.; Bogel-Lukasik, R. Ionic liquids as a tool for lignocellulosic biomass fractionation. *Sustain. Chem. Process.* **2013**, *1*, 3.

(12) Reichardt, C. Polarity of ionic liquids determined empirically by means of solvatochromic pyridinium N-phenolate betaine dyes. *Green Chem.* **2005**, *7*, 339−351.

(13) Domanska, U.; Bogel-Lukasik, R. Physicochemical properties and solubility of alkyl-(2-hydroxyethyl)-dimethylammonium bromide. *J. Phys. Chem. B* **2005**, *109*, 12124−12132.

(14) Bogel-Lukasik, R.; Goncalves, L. M. N.; Bogel-Lukasik, E. Phase equilibrium phenomena in solutions involving tannins, flavonoids and ionic liquids. *Green Chem.* **2010**, *12*, 1947−1953.

(15) Zakrzewska, M. E.; Bogel-Lukasik, E.; Bogel-Lukasik, R. Solubility of carbohydrates in ionic liquids. *Energy Fuels* **2010**, *24*, 737−745.

(16) Yoon, H.; Lane, G. H.; Shekibi, Y.; Howlett, P. C.; Forsyth, M.; Best, A. S.; MacFarlane, D. R. Lithium electrochemistry and cycling behaviour of ionic liquids using cyano based anions. *Energy Environ. Sci* **2013**, *6*, 979−986.

(17) Coleman, D.; Gathergood, N. Biodegradation studies of ionic liquids. *Chem. Soc. Rev.* **2010**, *39*, 600−637.

(18) Pham, T. P.; Cho, C.-W.; Yun, Y.-S. Environmental fate and toxicity of ionic liquids: a review. *Water Res.* **2010**, *44*, 352.

(19) Fu, D.; Mazza, G.; Tamaki, Y. Lignin extraction from straw by ionic liquids and enzymatic hydrolysis of the cellulosic residues. *J. Agric. Food Chem.* **2010**, *58*, 2915−2922.

(20) Nguyen, T. A. D.; Kim, K. R.; Han, S. J.; Cho, H. Y.; Kim, J. W.; Park, S. M.; Park, J. C.; Sim, S. J. Pretreatment of rice straw with ammonia and ionic liquid for lignocellulose conversion to fermentable sugars. *Bioresour. Technol.* **2010**, *101*, 7432−7438.
(21) Tan, S. S. Y.; MacFarlane, D. R.; Upfal, J.; Edye, L. A.; Doherty, W. O. S.; Patti, A. F.; Pringle, J. M.; Scott, J. L. Extraction of lignin from lignocellulose at atmospheric pressure using alkylbenzenesulfonate ionic liquid. *Green Chem.* **2009**, *11*, 339−345.
(22) Wu, H.; Mora-Pale, M.; Miao, J.; Doherty, T. V.; Linhardt, R. J.; Dordick, J. S. Facile pretreatment of lignocellulosic biomass at high loadings in room temperature ionic liquids. *Biotechnol. Bioeng.* **2011**, *108*, 2865−2875.
(23) Kilpeläinen, I.; Xie, H.; King, A.; Granstrom, M.; Heikkinen, S.; Argyropoulos, D. S. Dissolution of wood in ionic liquids. *J. Agric. Food Chem.* **2007**, *55*, 9142−9148.
(24) Remsing, R. C.; Swatloski, R. P.; Rogers, R. D.; Moyna, G. Mechanism of cellulose dissolution in the ionic liquid 1-*n*-butyl-3-methylimidazolium chloride: a ^{13}C and $^{35/37}Cl$ NMR relaxation study on model systems. *Chem. Commun.* **2006**, 1271−1273.
(25) Lee, S. H.; Doherty, T. V.; Linhardt, R. J.; Dordick, J. S. Ionic liquid-mediated selective extraction of lignin from wood leading to enhanced enzymatic cellulose hydrolysis. *Biotechnol. Bioeng.* **2009**, *102*, 1368−1376.
(26) da Costa Lopes, A. M.; João, K.; Rubik, D.; Bogel-Lukasik, E.; Duarte, L. C.; Andreaus, J.; Bogel-Lukasik, R. Pre-treatment of lignocellulosic biomass using ionic liquids: wheat straw fractionation. *Bioresour. Technol.* **2013**, *142*, 198−208.
(27) Magalhães da Silva, S. P.; da Costa Lopes, A. M.; Roseiro, L. B.; Bogel-Lukasik, R. Novel pre-treatment and fractionation method for lignocellulosic biomass using ionic liquids. *RSC Adv.* **2013**, DOI: 10.1039/C3RA43091J.
(28) Sun, N.; Rahman, M.; Qin, Y.; Maxim, M. L.; Rodríguez, H.; Rogers, R. D. Complete dissolution and partial delignification of wood in the ionic liquid 1-ethyl-3-methylimidazolium acetate. *Green Chem.* **2009**, *11*, 646−655.
(29) Lan, W.; Liu, C. F.; Sun, R. C. Fractionation of bagasse into cellulose, hemicelluloses and lignin with ionic liquid treatment followed by alkaline extraction. *J. Agric. Food Chem.* **2011**, *59*, 8691−8701.
(30) Yang, Q.; Shi, J.; Lin, L.; Zhuang, J.; Pang, C.; Xie, T.; Liu, Y. Structural characterization of lignin in the process of cooking of cornstalk with solid alkali and active oxygen. *J. Agric. Food Chem.* **2012**, *60*, 4656−4661.
(31) Long, J.; Guo, B.; Teng, J.; Yu, Y.; Wang, L.; Li, X. SO_3H-functionalized ionic liquid: efficient catalyst for bagasse liquefaction. *Bioresour. Technol.* **2011**, *102*, 10114−10123.
(32) Long, J.; Li, X.; Guo, B.; Wang, F.; Yu, Y.; Wang, L. Simultaneous delignification and selective catalytic transformation of agricultural lignocellulose in cooperative ionic liquid pairs. *Green Chem.* **2012**, *14*, 1935−1941.
(33) Brandt, A.; Ray, M. J.; To, T. Q.; Leak, D. J.; Murphy, R. J.; Welton, T. Ionic liquid pretreatment of lignocellulosic biomass with ionic liquid−water mixtures. *Green Chem.* **2011**, *13*, 2489−2499.
(34) Carvalheiro, F.; Silva-Fernandes, T.; Duarte, L. C.; Gírio, F. M. Wheat straw autohydrolysis: process optimization and products characterization. *Appl. Biochem. Biotechnol.* **2009**, *153*, 84−93.
(35) Hurtubise, F. G.; Krässig, H. Classification of fine structural characteristics in cellulose by infared spectroscopy. Use of potassium bromide pellet technique. *Anal. Chem.* **1960**, *32*, 177−181.
(36) Nelson, M. L.; O'Connor, R. T. Relation of certain infrared bands to cellulose crystallinity and crystal lattice type. Part II. A new infrared ratio for estimation of crystallinity in celluloses I and II. *J. Appl. Polym. Sci.* **2003**, *8*, 1325−1341.
(37) Rovio, S.; Simolin, H.; Koljonen, K.; Sirén, H. Determination of monosaccharide composition in plant fiber materials by capillary zone electrophoresis. *J. Chromatogr., A* **2008**, *1185*, 139−144.
(38) Brandt, A.; Hallett, J. P.; Leak, D. J.; Murphy, R. J.; Welton, T. The effect of the ionic liquid anion in the pretreatment of pine wood chips. *Green Chem.* **2010**, *12*, 672−679.
(39) Conceiçao, L. J. A.; Bogel-Łukasik, E.; Bogel-Łukasik, R. A new outlook on solubility of carbohydrates and sugar alcohols in ionic liquids. *RSC Adv.* **2012**, *2*, 1846−1855.
(40) Swatloski, R. P.; Spear, S. K.; Holbrey, J. D.; Rogers, R. D. Dissolution of cellose with ionic liquids. *J. Am. Chem. Soc.* **2002**, *124*, 4974−4975.
(41) Gupta, N.; Kad, G. L.; Singh, J. Acidic ionic liquid [bmim] HSO_4: an efficient catalyst for acetalization and thioacetalization of carbonyl compounds and their subsequent deprotection. *Catal. Comm.* **2007**, *8*, 1323−1328.
(42) Wang, X.; Li, H.; Cao, Y.; Tang, Q. Cellulose extraction from wood chip in an ionic liquid 1-allyl-3-methylimidazolium chloride (AmimCl). *Bioresour. Technol.* **2011**, *102*, 7959−7965.
(43) Hou, X. D.; Smith, T. J.; Li, N.; Zong, M. H. Novel renewable ionic liquids as highly effective solvents for pretreatment of rice straw biomass by selective removal of lignin. *Biotechnol. Bioeng.* **2012**, *109*, 2484−2493.
(44) Yoon, L. W.; Ngoh, G. C.; Chua, M.; Seak, A.; Hashim, M. Comparison of ionic liquid, acid and alkali pretreatments for sugarcane bagasse enzymatic saccharification. *J. Chem. Technol. Biotechnol.* **2011**, *86*, 1342−1348.
(45) Kacurakova, M.; Capek, P.; Sasinkova, V.; Wellner, N.; Ebringerova, A. FT-IR study of plant cell wall model compounds: pectic polysaccharides and hemicelluloses. *Carbohydr. Polym.* **2000**, *43*, 195−203.
(46) Peng, F.; Ren, J. L.; Xu, F.; Bian, J.; Peng, P.; Sun, R. C. Comparative study of hemicelluloses obtained by graded ethanol precipitation from sugarcane bagasse. *J. Agric. Food Chem.* **2009**, *57*, 6305−6317.
(47) Li, C.; Wang, Q.; Zhao, Z. K. Acid in ionic liquid: an efficient system for hydrolysis of lignocellulose. *Green Chem.* **2007**, *10*, 177−182.
(48) González, G.; López-Santín, J.; Caminal, G.; Sola, C. Dilute acid hydrolysis of wheat straw hemicellulose at moderate temperature: a simplified kinetic model. *Biotechnol. Bioeng.* **1986**, *28*, 288−293.
(49) Vaher, M.; Koel, M.; Kazarjan, J.; Kaljurand, M. Capillary electrophoretic analysis of neutral carbohydrates using ionic liquids as background electrolytes. *Electrophoresis* **2011**, *32*, 1068−1073.
(50) Sun, R. C.; Tomkinson, J. Characterization of hemicelluloses obtained by classical and ultrasonically assisted extractions from wheat straw. *Carbohydr. Polym.* **2002**, *50*, 263−271.
(51) Sain, M.; Panthapulakkal, S. Bioprocess preparation of wheat straw fibers and their characterization. *Ind. Crop. Prod.* **2006**, *23*, 1−8.
(52) Liu, R.; Yu, H.; Huang, Y. Structure and morphology of cellulose in wheat straw. *Cellulose* **2005**, *12*, 25−34.
(53) Yang, Q.; Wu, S.; Lou, R.; LV, G. Structural characterization of lignin from wheat straw. *Wood Sci. Technol.* **2011**, *45*, 419−431.
(54) Kubo, S.; Kadla, J. F. Hydrogen bonding in lignin: a Fourier transform infrared model compound study. *Biomacromolecules* **2005**, *6*, 2815−2821.
(55) Wang, B.; Feng, H.; Wang, X.; Zhou, B.; de Frias, J. A. Deconstructing lignocellulosic biomass with a two-step method. U.S. Patent Appl. 12/642601, 2012.