ISOLATION AND CHARACTERIZATION OF CELLULOSE AND LIGNIN FROM STEAM-EXPLODED LIGNOCELLULOSIC BIOMASS

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The isolation of cellulose from different lignocellulosic biomass sources such as corn cob, banana plant, cotton stalk, and cotton gin waste, was studied using a steam explosion technology as a pre-treatment process for different times followed by alkaline peroxide bleaching. The agricultural residues were steam-exploded at 220 °C for 1-4 min for the corn cob, 2 and 4 min for the banana plant, 3-5 min for the cotton gin waste, and for 5 min for the cotton stalk. The steamed fibers were water extracted followed by alkali extraction and finally peroxide bleaching to vield cellulose with different degrees of crystallinity. The degree of polymerization of the cellulose fraction ranged from 167.4 to 1615.7. Longer residence time of the steam explosion led to an increase in cellulose crystallinity. The ten isolated cellulose samples were further characterized by SEM, FT-IR, and thermal analysis. Four lignin preparations were also obtained from steam-exploded corn cob, banana plant, cotton stalk, and cotton gin waste after alkali treatment. The SEM micrographs of the lignin showed different morphological structure for the different agricultural residues. The FT-IR and TGA analyses showed that the steam pre-treatment led to an extensive cleavage of ether bonds, condensation reactions, and some demethylation of aromatic methoxyl groups in the lignin structure. The thermal stabilities of the isolated lignins were different for different agricultural residues.

Keywords: Lignocellulosic biomass; Steam explosion: Agricultural residues; Cellulose; Lignin; Crystallinity; Degree of polymerization; Thermal stability

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INTRODUCTION

Lignocellulosic plant biomass is a natural renewable resource, largely unused, and abundantly available source of raw materials that can be converted into useful materials and energy (Klass 1998; Cheng and Zhu 2008). Renewable energy technologies are of interest to the agro-based industries because these industries produce several million tons of waste material, which have to be disposed either through incineration, land applications, or landfilling, However because of the increasing tipping fees and stricter regulations on particulate emissions due to the Clean Air Act, there is renewed interest in producing value-added products from these "apparent waste" materials.

The development of industrial methods to convert biomass into useful materials and products without generating pollutants such as waste gas, wastewater, and solid waste materials will be useful for global environmental protection (Asada et al. 2005).

Lignocellulosic biomass sources range from trees to agricultural residues. Banana plant, cotton stalk, and cotton gin waste are agricultural residues that are not currently used as industrial raw materials. They contain lignin, cellulose, and hemicellulose, which could be used for the production of cellulose, cellulose derivatives, and plastics or could be potentially used as a source of monomeric sugars for fermentation to produce high-valued products. Long ago, these materials were used as firewood, building materials, and animal food. Nowadays, lignocellulosic materials are not just used in their old ways, but also their applications have expanded to include fibers in pulp and paper products.

The treatment of lignocellulosic materials with high pressure steam, for short periods of time, followed by sudden decompression (explosion) represents a simple treatment for biomass that achieves defiberization or "mulching" by a combination of chemical and mechanical action. The parameters controlling the steam explosion technique are reaction temperature (T_r) and retention time (t). The relationship between T_r and t has been defined as severity (log R_o) (Shevchenko et al. 1999; Overend et al. 1987). Steam explosion technology has become a well known method for separating lignocellulosic material into its main components: cellulose, lignin, and hemicellulose (Delong 1981; Nguyen 1989; Josefsson et al. 2002).

Cellulose, the most abundant renewable polymer available, is produced by nature at an annual rate of 10^{11} – 10^{12} tons (Hon 1994). Cellulose is a polymer consisting of unbranched $\beta(1\rightarrow 4)$ D-glucopyranosyl units. The cellulose chains have a strong tendency to aggregate to highly ordered structural entities due to their chemical constitution and spatial conformation. In addition to the crystalline regions (highly ordered), cellulose also contains amorphous regions. More recent models employ architectures of fibrillar units.

Lignin, along with cellulose and hemicellulose, is a major component of the composite-like system comprised entirely of natural polymers responsible for the strength and rigidity of plant stems. As such, lignin is the second most abundant natural polymer on earth (Fox 2006). Complicating the molecular picture of lignin further is the fact that native lignin chemistry can vary depending upon its origin (Fox 2006). Plant species, plant tissue type, and the external environment all have roles in determining the chemistry of a specific lignin sample.

The lignin polymer usually undergoes severe chemical modifications during isolation, and these changes restrict many of its potential industrial applications. Therefore, a successful isolation procedure must avoid both fragmentation and condensation reactions. One of the methods commonly used to recover lignin from wood and other lignocellulosic materials is steam explosion followed by mild-alkaline-extraction, which minimizes the condensation of the polymer.

Morphological studies to date have looked at natural and model cellulose. We are interested in understanding how morphology changes after different severities for steam explosion have occurred. In this paper, SEM, TGA, and FT-IR were used to follow the structural changes in cellulose and lignin obtained from corn cob, cotton stalk, banana plant, and cotton gin waste after steam explosion. It was the objective of the present study to investigate the effect of the stem explosion as a pre-treatment process on the fractionations resulted from four biomass sources using a range of practical severity conditions and continued with bleaching method for the resulted fibers.

EXPERIMENTAL

Materials

Cotton stalk (CS) and banana plant (BP) samples were obtained from local Egyptian farmers and dried at room temperature to an equilibrium moisture content of 5 to 6 wt% before the steam explosion experiments. Corn cob (CC) and cotton gin waste (CGW) samples obtained from local Virginia farmers were dried at room temperature to an equilibrium moisture content of 5-6 wt% before being used in the steam explosion experiments.

Steam Explosion

Steam explosion of the CS, BP, CC, and CGW samples was carried out in a 25-L batch reactor located at the Thomas M. Books Forest Products Center, Blacksburg, VA. The procedure reported by Jeoh and Agblevor (2001) was used in these studies. About 1 kg of the sample was weighed and loaded into a 25-L batch steam explosion chamber and the ball valve was closed. Saturated steam was admitted into the chamber, and the biomass temperature was varied from 180 °C to 220 °C in preliminary experiments. The ambient temperature of 220 °C was chosen according to the resultant fibrous mulch. The reactor was allowed to reach target temperature before timing began. Typically, about 20 seconds was required to attain the desired temperature. When the material attained the reaction temperature, i.e. 220 °C, timing of the reaction was started. For these experiments the following retention times were used: 60-240 seconds for CC, 120 and 240 seconds for BP, 180-300 seconds for CGW, and 300 seconds for CS. After the desired time, the ball valve was opened, which caused a rapid explosive decompression and disintegration of the biomass material. The sample was bagged and stored in a cold room until the time of extraction and bleaching.

Water and Alkali Extraction of Steam-Exploded Materials

The steam-exploded fibers (10 g) (oven-dry fiber) were extracted with water at 80 °C for 1 h using a fiber to water ratio of 1:10. The mixture was vacuum filtered in a Buchner funnel and then washed with 500 ml water. The water-extracted fibers were further extracted with 20 wt% sodium hydroxide (NaOH) at 80 °C for 1 h for dissolving the lignin and to obtain a cellulosic fiber using a fiber to liquor ratio of 1:10. The mixture was filtered, the alkali extracted water was collected, and the residue was washed with water until the pH was neutral. The alkali extracted fibers were air-dried under ambient laboratory conditions and stored until the time of bleaching.

Bleaching

The steam-exploded fibers were bleached with a two stage hypochlorite-hydrogen peroxide process for steam-exploded CS, BP, and CGW and one stage hydrogen peroxide for steam-exploded CC. The fibers, CS, BP, and CGW, were first treated with sodium hypochlorite solution equivalent to 60% of the chlorine requirement for 2 h at 40 °C in a temperature-controlled water-bath. The liquor-to-fibers ratio was 20:1, and the pH was maintained at 9 during the hypochlorite process. The fibers were washed with water until neutrality. After the hypochlorite process, the steam-exploded fibers, as well as steam-

exploded CC, were treated with hydrogen peroxide (H_2O_2) . The bleaching liquor consisting of a mixture of H_2O_2 (25wt% on oven-dry fiber (odf)) and NaOH (1.6wt% odf), was added to the fibers in a serum bottle. The serum bottles were closed and placed in a temperature-controlled water-bath at 65 °C for 2 h. The mixture was stirred manually at 10-min intervals to ensure proper liquor-fiber mixing.

At the end of the bleaching period, the fibers were diluted to 1% consistency and then neutralized with sodium metabisulfite to decompose the residual H_2O_2 and to stabilize the brightness. The mixture was vacuum filtered, and the fibers were washed thoroughly with water and air-dried under ambient laboratory conditions.

Isolation of Solubilized Lignin

The black liquor from the alkali extraction of the steam-exploded fibers was in each case acidified to pH 1.5 by a dropwise addition of concentrated sulfuric acid (H₂SO₄). The acidified mixtures were then heated in a water bath at 100 °C for 1 h with continuous stirring. The precipitated lignin fractions were cooled to room temperature, filtered in a Buchner funnel, and then washed with hot water until neutrality. The final products were air-dried under ambient conditions.

The isolated lignins were redissolved in 17.5 (w/w) NaOH, and precipitated with concentrated H_2SO_4 (pH 1.5) using the procedure described earlier. The precipitated lignin was filtered and then washed with hot water until the filtrate was clean and then air-dried under ambient laboratory conditions.

Chemical Characterization of Cellulose Preparations

The chain length of cellulose is generally expressed as the degree of polymerization (*DP*), which is defined as the average number of anhydroglucose units in the chain of the cellulose molecule (Christy and Samfield 1960). There are several methods for determining the *DP* of cellulose, including the intrinsic viscosity method in a suitable solvent (Christy and Samfield 1960). In the present work, the intrinsic viscosities of bleached steam-exploded BP, CS, CC, and CGW were determined in cupriethylenediamine (CED), using a Cannon-Fenske Routine Viscometer. About 3 g of each set of bleached steam-exploded fibers was weighed directly into a 50 ml stoppered bottle, 7.5 mL distilled water was added, and the bottle was shaken for two minutes to "wet" the powder. The bottles were removed from the shaker, and 22.5 mL 1.0M CED solution was added to the samples. The ratio of CED/H₂O was 3/1, and the sample concentration was 0.1 g/ml. The samples were shaken in the CED solution from 10 to 60 minutes until they dissolved.

The intrinsic viscosity was determined using the method of Solomon and Gatesman (Viscotek Application No. 5). Equation 1 was used in calculating the intrinsic viscosity.

$$[\eta] = \frac{\left[2 \left(\eta_{sp} - \ln \eta_r\right)\right]^{\nu_2}}{C}$$
(1)

where, $[\eta]$ is the intrinsic viscosity, η_{sp} is the specific viscosity, η_r is the relative viscosity and *C* is the concentration of the sample. The *DP* was calculated according to the SCAN-CM 15:88 (1988), where:

$$DP^{0.85} = 1.1 \, [\eta] \tag{2}$$

Instrumental Characterization of Cellulose Preparations and Isolated Lignin

The Degree of Crystallinity (*DC*) was measured by using the iodine absorption method (Kortschagin et al. 1991). For this method, the fibers were added to iodine (5 g I_2 , 40 g KI, 50 mL distilled water) and Na₂SO₄ solution and stored and stirred for several hours in darkness. The amounts of Na₂S₂O₃ solution titrated with and without fibers were used to calculate the degree of crystallinity "*DC*" from the following equation:

$$DC = 100 - \frac{37.925}{m} \left[1 - \frac{V_{\text{sample}}}{v_{\text{control}}} \right]$$
(3)

where, m [g] is the mass of the sample, and V_{sample} [mL] and V_{control} [mL] are the volumes Na₂S₂O₃ titrated with and without fibers. Three samples were investigated in each case.

Bleached steam-exploded BP, CS, CC, and CGW samples and lignin fractions were also characterized using techniques described below. Scanning electron microscope (SEM) characterization of the cellulose and lignin preparations was performed using a LEO 1550 electron microprobe analyzer (GEMINI, USA) and a JEOL JXA-840A electron microprobe analyzer (JOEL USA Inc, Peabody, MA). The FT-IR spectra of bleached BP, CS, CC, and CGW samples, as well as lignin fractions were recorded on a Niocolet-670 FT-IR spectrometer, (Thermo Electron, Inc, Madison, WI). Samples (~2 mg) were mixed and thoroughly ground with ~200 mg KBr to reduce particle size and to obtain uniform dispersion of the sample in the disks. All the spectra were recorded in the absorbance mode from 4000 to 400 cm⁻¹ at room temperature. Thermogravimetric analyses (TGA) were performed on the samples using PerkinElmer TGA7 instrument (PerkinElmer Inc, Shelton, CT). The samples were analyzed in a nitrogen atmosphere from 30° to 700°C at a heating rate of 10°C/min and a nitrogen flow rate of 30 ml/min. Data acquisition and analysis was performed using Pyris v 5.0 software.

RESULTS AND DISCUSSION

Steam explosion was selected as the processing technology to separate the components of the lignocellulosic biomass because it requires little or no chemical input and thus is environmentally benign relative to other technologies, such as acid hydrolysis. To achieve variations of cellulose structures and chemical compositions, different steam explosion experiments, in which the time was varied according to the type of the lignocellulosic biomass, were performed to obtain fibrous mulch. The analysis of the resulting steam-exploded fibers showed a decrease in the holocellulose compared to the untreated fibers, as shown in Tables 1 and 2. This revealed that small amounts of

cellulose were degraded or hydrolysed during the process. On the other hand, the yield of the resulting steam-exploded fibers was decreased with increasing severity parameter (Table 2). This also indicated that the degradation of the fibers depends on the severity.

The lignin contents in both steam-exploded fibers and raw material were measured as Klason lignin, which is the material left after acid hydrolysis of the samples with 72% sulfuric acid, and their results are listed in Tables 1 and 2. As can be seen, the lignin contents in CC1-4 ranged between 17.56% and 25.76% (Table 2), which are higher than the lignin content in the raw material (15.83%, Table 1). A similarly increasing trend of lignin was observed for BP2 and BP4, CGW3-5, and CS, where the lignin for the steam-exploded fibers was higher than the started material (Tables 1 and 2). According to Sun et al. (2005), the higher lignin content is due to the hydrolysis of substantial amount of hemicellulose and a lignin recondensation reaction during the steam explosion process.

In all the experiments, the exploded material was very dark. These color changes during the steam pre-treatment could have resulted from of the degradation of the cell wall components and extractives (Sun et al. 2005). Alternatively, the appearance of brownish products in treated samples could be attributed to sugar degradation products that are produced at higher steam pre-treatment temperatures (Nergro et al. 2003).

		Diolio Bioli	
Raw material	Holocellulose	Lignin	Ash
	(%)	(%)	(%)
CC	85.28	15.83	1.32
BP	57.46	20.31	13.63
CGW	53.89	20.56	12.67
CS	67.80	27.58	1.77

Table 1. Chemical Analysis for Lignocellulosic Biomass (raw material)

Steam-	Yield	Severity	Temperature	Retention	Holo-	Lignin	Ash
exploded	(%)	parameter*	(°C)	Time	cellulose	_	
fibers**					(%)	(%)	(%)
				(min)			
CC1	65.79	3.54	220	1	81.61	17.56	0.36
CC2	44.83	3.85	220	2	61.31	20.00	0.01
CC3	10.76	4.02	220	3	58.53	22.56	0.01
CC4	9.95	4.15	220	4	53.34	25.76	0.01
BP2	38.21	3.85	220	2	53.45	25.58	14.97
BP4	36.76	4.15	220	4	52.76	23.22	19.02
CGW3	78.37	4.02	220	3	51.79	31.99	10.22
CGW4	71.87	4.15	220	4	47.75	35.86	15.23
CGW5	44.23	4.32	220	5	43.87	38.34	17.24
CS	40.81	4.32	220	5	50.14	40.55	4.31

 Table 2. Yield Percent and Chemical Analysis for Steam-Exploded Fibers

* The severity parameter is a relation between the temperature and the retention time.

** CC1, CC2, CC3, and CC4 represent the bleached corn cob cellulose after steam-exploded obtained at 220°C for time from 1 to 4 min, respectively. BP2 and BP4 represent the bleached banana plant cellulose after being steam-exploded, obtained at 220°C for time from 2 and 4 min, respectively. CGW3, CGW4, and CGW5 represent the bleached cotton gin waste cellulose after being steam-exploded, obtained at 220°C for time from 3 to 5 min, respectively. CS represents the bleached cotton stalk cellulose after being steam-exploded, obtained at 220°C for 5 min.

Scanning electron micrographs (SEM) obtained at high magnifications showed that the bleached steam-exploded CC particle morphology varied with different severities (Fig. 1 a-d). The bleached steam-exploded CC sample prepared at severity parameter 3.54 consisted of lamellar-shaped particles (Fig. 1a). In contrast, the bleached steam-exploded CC prepared at severity parameter 4.15 (Fig. 1d) had round shaped particles and appeared to have a micro-recticulated structure. This clearly showed that the shape and size of the steam-exploded particles can be influenced by the retention time of the steam explosion. The SEM images (Fig. 1a and b) of one individual macrofibril show that the surface of bleached steam-exploded CC at severities of 3.54 and 3.85 were almost free of trenches. After increasing the retention time of explosion, the CC particles showed significant changes on the surface. A closer look at the macrofibril surface (Fig. 1 e-h) at larger magnification shows that many terraces, steps, and kinks had formed after increasing the time of explosion, i.e. at severities 4.02 and 4.15. We propose that these changes resulted from the removal of the amorphous cellulose from the surface.

Figures 2a–d are SEM images of bleached steam-exploded banana plant material under various magnifications. Figure 2a clearly shows the shape and size distribution of the macrofibrils in bleached steam-exploded banana at severity 3.85. The SEM image (Fig. 2a) of one individual macrofibril at larger magnification shows that the surface of bleached steam-exploded banana at severity 3.85 was almost free of trenches, but there were obvious boundary edges in different regions. Increasing the time of explosion, Fig. 2b, shows that some macrofibrils remain separated, but other macrofibrils agglomerate. The higher magnification shows again the presence of trenches, terraces, steps, and kinks.

Figures 3a–f show the SEM images of bleached steam-exploded CGW at different severities under various magnifications. Fig. 3a clearly shows the shape and size distribution of the macrofibrils in bleached steam-exploded CGW at severity of 3.85. These well-separated macrofibrils had lengths of 48.84–57.05 μ m and diameters of 8.25–10.59 μ m. The SEM image (Fig. 3b) of one individual macrofibril at larger magnification shows that the surface was almost free of trenches. After increasing the time of explosion to 3 minutes, the SEM image (Fig. 3c), shows that the macrofibrils were still well separated and their diameters decreased to 5.66-7.97 μ m. Figure 3d displays one individual macrofibril that shows significant changes on the surface, probably due to the removal of the some fibrous layers.

Figure 3e-f shows the SEM images for the bleached steam-exploded CGW after increasing the time of explosion to 4 min. The diameters of the macrofibrils (Fig. 3e) were similar to that exploded for 3 min, but the length decreased considerably. A closer look at the macrofibril surface (Fig. 3f) at larger magnification shows many terraces, steps, and kinks form after increasing the time of explosion. Again, we propose that these changes resulted from the removal of the amorphous cellulose from the surface.



Fig. 1. SEM images for bleached steam-exploded CC under different magnifications for (a) CC1, (b) CC2, (c, e & g) CC3 and (d, f & h) CC4

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Fig. 2. SEM images for bleached steam-exploded BP under different magnifications for (a & c) BP2 and (b & d) BP4

Figures 4a–b show the SEM images of bleached steam-exploded CS at severity of 4.32 under various magnifications. Fig. 4a clearly shows the shape and size distribution of the macrofibrils in bleached steam-exploded CS, which had lengths of 58.82-68.82 μ m and a diameter of 8.71 μ m. The SEM image (Fig. 4b) of one individual macrofibril at larger magnification shows that the surface had many terraces, steps, and kinks that had formed during the process.

The changes in cellulose structure at different severities of treatment were also corroborated by other physico-chemical properties, such as the degree of polymerization and degree of crystallinity (DC). It is known that amorphous cellulose, as one part of natural fibers, forms hydrogen bonds at 60°C and recrystallizes at 150°C (Yano et al. 1976). The degree of crystallinity for the bleached steam-exploded CC, CGW, and BP increased with increasing the time of explosion (Table 3). This result is in agreement with that of the SEM, where increasing the time of steam explosion resulted in the removal of the amorphous cellulose and hence increased the degree of crystallinity. Glasser and Wright (1998) reported that steam explosion raises the crystallinity of the cellulose component. This suggested that the main reactions that occurred during the process were the cleavage of the amorphous part of the cellulose, while the crystalline fraction was preserved. The intrinsic viscosity (η) and average degree of polymerization (DP) for the bleached steam-exploded CC, BP, CGW, and CS are shown in Table 3. As can be seen, the DP decreased with increasing steam explosion severity parameter. Josefeeon et al. (2002) reported that during steam explosion at 180° and 190°C, the molecular weight of cellulose was not greatly affected by the reaction time. At higher temperatures (200220°C), a severe decrease in molecular weight of cellulose with increasing reaction time was observed for aspen wood. The trend in our DP data is in agreement with the cited work.



Fig. 3. SEM images for bleached steam-exploded CGW under different magnifications for (a & b) CGW3, (c & d) CGW4 and (e & f) CGW5

Table 3. Intrins	ic Viscosity (η), Deg	ree of Polymerization	ו (<i>DP</i>), and Degree of
Crystallinity (D	C), of the Produced	Bleached Steam-Ex	ploded Cellulose Fibers

Sample no.	Severity parameter	η	DP	DC
CC1	3.54	484.9	1615.7	36.8
CC2	3.85	462.4	1527.6	40.0
CC3	4.02	385.8	1234.6	50.0
CC4	4.15	304.6	934.8	60.2
BP2	3.85	138.6	370.2	69.3
BP4	4.15	98.6	247.9	80.0
CGW3	4.02	253.4	752.7	62.4
CGW4	4.15	208.0	596.8	70.1
CGW5	4.32	173.9	483.4	80.1
CS	4.32	70.6	167.4	70.1



Fig. 4. SEM images under different magnifications for bleached steam-exploded cotton stalk

Although dramatic morphology changes on cellulose macrofibrils were observed with increased time of steam explosion, no change in the X-ray patterns and FT-IR data were detected, as shown in Figs. 5 and 6. The X-ray patterns for bleached steam-exploded CCs, Fig. 5, showed that all samples exhibited similar crystalline patterns. The widths at half height for the peaks at $2\theta = 17^{\circ}$ and 26° were similar for all samples, which suggested that their crystallite sizes were similar (Garnier et al. 1990). The same observation was noticed for bleached steam-exploded BP and CGW (data are not presented).



Fig. 5. X-ray diffractogram for beached steam-exploded (a) CC1, (b) CC2, (c) CC3, and (d) CC4 (The arrows indicate the conditions of $2\theta = 17^{\circ}$ and 26°)

The FT-IR spectra of bleached steam-exploded corn cob residue CC1 (spectrum a), CC2 (spectrum b), CC3 (spectrum c), and CC4 (spectrum d) (Fig. 6) were similar, indicating similar structure of the residues. They were characteristic of cellulose peaks and showed the absence of characteristic lignin peaks at 1726 and 1513 cm⁻¹.



Fig. 6. FT-IR for bleached steam-exploded (a) CC1, (b) CC2, (c) CC3, and (d) CC4

The absorption band at 3345 cm⁻¹ was attributed to stretching vibrations of hydroxyl (OH) groups. The OH groups may include absorbed water, aliphatic primary and secondary alcohols found in cellulose, hemicellulose, and carboxylic acids in extractives (Khan et al. 1993; Kolboe and Ellefsen 1962). The shoulder near the OH stretching vibrations, 2901 cm⁻¹, was attributed to CH stretching vibrations and corresponded to the aliphatic moieties in polysaccharides (cellulose and survived hemicelluloses). The absence of absorption bands in the 1750-1700 cm⁻¹ region and at

1513 cm⁻¹ in the spectra of the steam-exploded CCs may be explained by the fact that the compounds rich in carbonyl (C=O), lignin, hemicelluloses, and extractives were removed during the alkaline peroxide bleaching process. The bands in the 1430-1318 cm⁻¹ region, for steam-exploded CCs were assigned to CH in-plane deformation of CH₂ groups, while the peaks at 1165-1059 cm⁻¹ were assigned to the linkage present in the cellulose (Ibrahim 2002). The absorbances at 1431, 1372, 1319, 1165, 1059, and 896 cm⁻¹, which are typical of pure cellulose, can be seen in the steam-exploded CC1, CC2, CC3, and CC4 spectra.

The FT-IR spectra of cellulose preparations obtained by alkaline peroxide posttreatment of the corresponding steam-exploded residues of CGW, BP, and CS, were similar to those for CCs. The absence of the band at 1726 cm⁻¹ and the band relating to lignin aromatic ring vibrations at 1513 cm⁻¹ revealed that lignin was removed during the peroxide bleaching step. Also, the absorbances at 1431, 1372, 1318, 1281, 1164, 1059 and 897 cm⁻¹, which are typical of pure cellulose, can be seen in the steam-exploded BP, CGW, and CS spectra (Fig. 7).



Fig. 7. FT-IR for bleached steam-exploded (a) BP, (b) CS, and (c) CGW

Thermal Stability

Thermal analysis is a convenient and useful method for characterizing heterogeneous organic materials. In particular, it is a valuable analytical method to investigate the physico-chemical properties of macromolecules such as cellulose. Figure 8 shows typical TGA curves of bleached steam-exploded CCs at severity parameters of 3.54 to 4.15 (Figs. 8 a-d). The onset of weight loss (T_s) for the four bleached steam-exploded CCs samples occurred at about 225°C. The maximum decomposition temperature (T_{max}) occurred at 352, 352, 355, and 359°C, respectively, for CC1, CC2, CC3, and CC4. These T_{max} values indicated that the four bleached steam-exploded CCs samples had similar thermal stability.



Fig. 8. TGA for bleached steam-exploded (a) CC1, (b) CC2, (c) CC3, and (d) CC4

Figure 9 shows typical TGA curves of bleached steam-exploded BP, CS, and CGW at severity parameters of 4.15, 4.32, and 4.15, respectively (Figs. 9 a-c). The onset of weight loss (T_s) for the three bleached steam-exploded BS, CS, and CGW samples occurred at about 210, 231, and 238°C, respectively. The maximum decomposition temperature (T_{max}) occurred at 365, 368, and 389°C, respectively, for BP4, CS, and CGW4. These T_{max} values indicated that the bleached steam-exploded CGW sample had higher thermal stability. Moreover, the thermograms for the bleached steam-exploded BP, CS, and CGW showed that there was another peak for derivatives (Fig. 9). This other peak may due to the presence of alkali metal impurities after alkali peroxide bleaching. However, the samples were neither dialyzed nor analyzed for alkali metal content.

A linear correlation was observed between the DP and T_{max} (Fig. 10). This trend could be attributed to the decrease in the amorphous fraction of the cellulose with decrease in DP. The more crystalline fraction of the cellulose appeared to have a lower DP (Table 3). This variation in T_{max} with DP is in agreement with Kim et al. (2005) and Chatterjee and Schwenker (1972), who reported an increase in T_{max} with increasing crystallinity.



Fig. 9. TGA for bleached steam-exploded (a) BP4, (b) CS, and (c) CGW4



Fig. 10. Relation between the maximum decomposition temperature (T_{max}) and degree of polymerization (*DP*) for the bleached steam-exploded fibers

SEM of Isolated Lignin

The morphologies of the lignin particles from the alkali extracted steam-exploded CC, BP, CS, and CGW are shown in Fig. 11. The representative images show different morphological structure for these samples. The lignin from CC showed a sponge-like structure (Fig. 11 a & c), whereas a rock-like structure was observed for the steam-exploded BP lignin (Fig. 11 b). A closer look at the surface of the BP lignin (Fig. 11d) at larger magnification shows that many steps were formed on the surface of the lignin.

The morphology of the lignin particles from steam-exploded CS is shown in Fig. 11 e & g. The SEM micrograph at lower magnification showed a fluff of lignin particles, whereas at higher magnification the surface appeared as lines with dots (Fig. 11 g). The SEM micrograph of CGW lignin showed a morphology that appeared as agglomerated particles (Fig. 11 f), while at higher magnification (Fig. 11 h) it revealed a honeycomb structure. This structure was significantly different from the structure of the lignins isolated from the other samples. This is in agreement with Fox (2006) that the lignin differs according to the plant species and plant tissue type facing the external environment, which make sense for the different morphological structure for the different agricultural biomasses.

FT-IR Spectra

FT-IR spectra of lignin from CC, BP, CS, and CGW, are shown in Fig. 12 a, b, c, and d. Clearly, the spectra varied only with the type of the agricultural residue. Depending on the agricultural residue, the spectra showed a difference in the 1708-1654 cm⁻¹ region. In the case of spectrum d, for CGW, an intensive band at 1710 cm⁻¹ was observed, while it was a weak peak at 1700 cm⁻¹ for spectrum a, for CC The peak at

1700-1710 cm⁻¹ was absent in spectra b and c, for BP and CS respectively. This band was assigned to unconjugated carbonyl ketone and/or aldehyde groups in the lignin fractions.



Fig. 11. SEM images under different magnification of lignins from (a & c) CC, (b & d) BP, (e &g) CS and (f & h) CGW

The absence of the band at 1745 cm⁻¹ suggested that the labile ester bonds were completely cleaved during the steam explosion. It should be noted that in spectra b and c

the bands due to conjugated carbonyl groups (α -carbonyl groups) were observed at 1620 and 1601 cm⁻¹ respectively, whereas this absorption was detected at 1653 cm⁻¹ for spectrum d. These results suggested that the lignin fractions from different agricultural residues contained different amounts of unconjugated carbonyl groups depending on the type of the agricultural residue. In addition, the effect of the steam explosion can be seen if the absorption bands in each spectrum are normalized to the band at 1518 cm⁻¹, which is attributed to aromatic skeleton vibrations. Although the intensity of this band varied minimally between lignin fractions CC (spectrum a) and CS (spectrum c), it decreased in BP lignin (spectrum b) and CGW lignin (spectrum d).



There was also a difference in the peak at 1459 cm⁻¹ due to methoxyl groups for spectra a and c, as wells as for spectra b and d. A decrease in intensity of this band in

spectra b and d (BP and CGW lignins) indicated a decrease in methoxyl groups. Furthermore, there was also a decrease in the relative intensity at 1032 cm⁻¹, due to C-H deformation of the guaiacyl units (Thring et al. 1990), in the spectra of b and d. This indicated that either the cleavage of ether linkage and demethoxylation of lignin during the steam explosion were greater for BP and CGW or that the proportions of the methoxyl groups in the two lignins were different.

Thermal Analysis for the Isolated Lignin

The thermal degradation pattern of lignin preparations CC, BP, CS, and CGW gave additional evidence of the thermal stability of the samples. Pyrolytic degradation of lignin involved fragmentation of inter-unit linkages, releasing phenolic compounds into the vapour phase. The evolution of methanol from the lignin is due to the cleavage of methyl-aryl ether bonds near 400 °C (Sun and Tomkinson 2001); and the decomposition or condensation of aromatic ring occurs at 400–600 °C (Yoshida et al. 1987). The TGA of lignin fractions, Fig. 13 a-d, indicated that the starting decomposition temperature for the lignin prepared from CC, BP, CS, and CGW occurred at 105, 142, 186, and 135°C, respectively. Beyond these temperatures, the degradation of the lignin from CGW was faster than the others. The maximum thermal degradation rate of CC, BP, CS, and CGW lignins occurred at 352, 392, 315, and 282 °C, respectively. As the isolated lignins from BP required higher temperature to reach complete degradation, (541 °C), it seems that the higher thermo-stability of this lignin was due to the occurrence of more condensed polymeric structures in its composition.



CONCLUSIONS

The results obtained in this study suggested that the steam explosion pretreatment of CC, BP, CS, and CGW followed by alkaline post-treatment is a promising alternative separation process that allows the fractionation of lignocellulosic materials into polymeric fractions with reasonable purity. It was found that the steam explosion degraded substantial amounts of hemicelluloses, while post-treatment with alkali under the conditions used removed all the lignin. In addition, the cellulose obtained at longer steam explosion residence times and sequential alkaline peroxide bleaching had a higher degree of crystallinity than those obtained after a short time of steam explosion. Furthermore, steam explosion produced cellulose products with different crystallinity, depending on treatment conditions. This leads to the possibility of different chemical applications, for example, dissolving pulp, microcrystalline cellulose, or powdered cellulose.

The results also showed that the lignin fractions obtained from the steamexploded materials had different morphological structure and thermal stability, depending on the type of the agricultural residues used. This is important for those who plan biomass strategies, especially in terms of chemical uses of lignin.

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