# Biorefinery Process Combining Specel® Process and Selective Lignin Precipitation using Mineral Acids

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Soda black liquors from the Specel® process, which used wheat straw as the raw material, were subjected to an acid precipitation process to recover the lignin. Lignin was isolated by acid precipitation using three different inorganics acids ($H_3PO_4$, $H_2SO_4$, and HCl) at three concentration levels, and at pH values of 2 and 4. Even though the highest lignin yield was achieved using phosphoric acid, the most economical inorganic acid was sulphuric acid. Physico-chemical characterizations of the precipitated lignin samples were performed using differential scanning calorimetry (DSC), thermogravimetric analysis (TGA) (for thermal properties), Fourier transform infrared spectroscopy (FT-IR), and heteronuclear single quantum correlation spectroscopy (HSQC) (for chemical structures). No significant differences were found in the thermal properties and chemical structures of the isolated lignins, except for the lignin obtained with phosphoric acid to lower the black liquor from pH 10.72 to pH 2. Apart from the lignin fraction collected, the soda pulp obtained by the Specel® process using wheat straw could represent a good alternative for packaging industries.

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## INTRODUCTION

Lignocellulosic biomass is one of the largest renewable and economical energy sources in the world. Among other materials, lignocellulosic biomasses are generated by agricultural, forestry, or industrial residues, which are produced in large amounts. These residuals are generally burned in the open, which causes environmental air emission problems.

One of the above-mentioned residues is obtained from the harvesting of cereals grains; these lignocellulosic materials represented 2.542 million tonnes in 2014 according to a FAOSTAT report (www.fao.org 2015). A large quantity of these wastes are generated each year when taking the FAO data into account and assuming that 1 kg of cereal grain can produce the same amount of agricultural residues (0.8 to 1 kg of cereal straw) (Rodríguez *et al.* 2009). Wheat straw accounted for 29% of total cereal straw in the world (FAOSTAT 2015).

To achieve a wide spectrum of bio-based products from the lignocellulosic biomass, an integral exploitation of it is necessary. To best utilize these resources, a

fractionation process is employed. Pulping processes are commonly used to acquire cellulosic fibers. In this regard, delignification mechanisms using alkaline treatments have several advantages, such as being versatile to process diverse raw materials (including agricultural residues), which is a key economic factor for an industrial plant. Besides, alkaline pulping affords a high yield with a short cooking time and a relative low capital investment for plant equipment (Rodriguez *et al.* 2009). In addition, these alkaline pulping processes use sulphur-free delignifying agents, which thereby removes one of the main drawbacks of the traditional pulping process such as the kraft process (González *et al.* 2010). Within these alkaline pulping processes, the Specel® process uses soda as the only chemical reagent. This pulping process has been successfully used by Ecopapel (Écija, Spain) in small and medium enterprises to produce cellulosic pulp from wheat straw (Vargas *et al.* 2012). This cellulose pulp could be used as a raw material for cellulose nanofiber and nanocrystal production (Robles *et al.* 2015) for medical applications (Cherian *et al*. 2011), or could be used for cardboard production (Jeetah *et al*. 2015).

Pulp production generates a large amount of spent cooking liquors, such as black liquor. Part of the lignin and hemicelluloses from the wheat straw are solubilized during soda pulping into the black liquor. A method to recover lignin from the black liquor was performed in order to fully exploit all components fractionated from lignocellulosic biomass in an integrated process. Lignin is one of three main components of lignocellulosic biomass (Toledano *et al.* 2010a), which is one of the most important renewable natural resources. A sustainable process to obtain lignin should have the following characteristics: environmentally compatible, socially acceptable, and economically viable.

There are three methods to separate alkaline lignins from black liquors. One method is acid precipitation, the second one is extraction with selective solvents, and the last method is separation using membrane technology, which is better known as ultrafiltration (Toledano *et al*. 2010a; Hellstén *et al.* 2013; Arkell *et al.* 2014). Selective lignin precipitation is one of the most commonly used processes since the lignin is recovered by the simple addition of an inorganic acid to the black liquor (Toledano *et al.* 2010b). The acidification process consists of decreasing the pH of the black liquor solution until the desired pH value is attained (Mussatto *et al.* 2007).

Many applications could be developed using lignin as the main ingredient and also as a secondary ingredient such as dispersants, binders, emulsifiers, epoxy resins, or even foams (El Mansouri *et al.* 2011; Arkell *et al*. 2014; Tondi *et al.* 2016). In addition, lignin can be used in the synthesis of new, more environmentally compatible polymeric materials, such as hydrogels, which are more suitable for various applications that range from biomedical to automotive (El-Zawawy and Ibrahim 2012; Thakur and Thakur 2015). These applications may be the best option for utilizing recovered lignin in the near future (Tejado *et al.* 2007). Therefore, all the above-mentioned applications could enhance the competiveness of pulp production by recovering lignin as high value-added by-product.

Characterizations of the physico-chemical properties of alkali lignin are very important in valorising this pulping by-product (Ibrahim *et al.* 2004). Characteristics of the different technical lignins depend on the raw lignocellulosic materials used and the extraction process used to obtain them (Tejado *et al.* 2007).

In this work, acid precipitation using three different inorganic acids ($H_3PO_4$, $H_2SO_4$ and HCl) at different levels of concentration was performed to separate dissolved lignin from black liquor by acidification. Lignins obtained from different precipitation processes were characterized by their physico-chemical properties in order to evaluate the impact of the mineral acid used, its concentration, and the final pH of the liquid residues. The black

liquors used in this study where from a soda pulping process, which is called Specel®, using wheat straw as the raw material. The use of cereal straws with this pulping process has afforded a cellulosic pulp with high yield.

## EXPERIMENTAL

### Materials

Wheat straw was provided by Ecopapel S. L. Company (Écija, Spain). Due to its origin as well as the collection method used, a manual screening was performed to separate different components (stones, seeds, dust, *etc.*), from the straw. The cleaned straw had a moisture content of around 10% (at typical room humidity conditions), and it was stored in plastic bags before it was pulped.

### Methods

*Pulping conditions*

The cellulosic pulp and the black liquor were produced from wheat straw using a 15-L batch reactor that was heated by an external electrical jacket. The reactor was stirred by rotating the reactor *via* a motor; the digester was monitored by a controller unit that maintained pressure and temperature set points. The wheat straw was pulp at the following conditions: 7% NaOH, 100 °C, with a liquor to solid ratio of 10:1 (mL/g) under stirring, and 150 min of cooking. These soda pulping conditions are identical to the conditions employed in the Specel® process used by Ecopapel S. L. Company.

*Selective precipitation of soda lignins*

The soda lignin isolation was performed by precipitating the dissolved lignin from the black liquor by acidifying it to a pH of 2 or 4. Acid precipitation used one of three different inorganic acids (*i.e*., sulphuric, hydrochloric, or phosphoric acid) at three different concentration levels (20%, 60%, and concentrated for $H_2SO_4$ and $H_3PO_4$, and 1, 2, and 3 M for HCl). After lowering the pH of the black liquor to 2 or 4, the solution was allowed to sit for 24 h to allow for the sedimentation of the precipitated lignin. The next step after sedimentation was centrifugation of the samples at 3500 rpm for 12 min; the precipitated solids were washed with water twice to remove possible impurities, such as monosaccharides. Finally, the washed precipitates were dried at 60 °C in an oven for 48 h.

*Characterization of black liquors*

The black liquors obtained from soda pulping of wheat straw were characterized determining their pH, density, total dry solids, and ash content. The pH was analyzed using a Crison GLP 21 pH meter (Crison Instruments, Barcelona, Spain). The measurement of density was determined by dividing its total mass by its total volume. Total dry solids were determined by drying the samples in an oven at 105 °C for 24 h in porcelain crucibles. Afterwards, the samples were heated in a muffle at 575 °C for 3 h to determine the ash content. Additionally, the lignin content was determined by conducting a simple mass balance. The electrical conductivity was analyzed with a Crison GLP 31 conductivity meter (Crison Instruments, Barcelona, Spain). All analytical measurements were performed in triplicate.

The monosaccharides, acetic acid, furfural, and hydroxymethylfurfural (HMF) content in the black liquors were determined by taking an aliquot of the liquor and

degrading it to its monomeric sugars by quantitative hydrolysis with 4% $H_2SO_4$ at 121 °C for 60 min. Afterwards, the resulting supernatant was filtered through a 0.45-µm membrane filter before high performance liquid chromatography (HPLC) analysis. HPLC analyses were performed using an Aminex HPX-87H column (Bio-Rad, California, USA) at 30 °C eluted with 0.01 M $H_2SO_4$ at a flow rate of 0.6 mL·min$^{-1}$ using a refractive index (RI) detector to quantify glucose, xylose, arabinose, acetic acid, HMF, and furfural (García-Domínguez *et al.* 2013).

*Lignin-rich fraction characterization*

Lignin-rich precipitates obtained with the different extraction processes (*i.e.*, acid precipitation) were characterized in order to establish their physico-chemical properties. The total amount of lignin was considered to be the sum of acid-insoluble and acid-soluble lignin. Prior to both determinations, an acid hydrolysis was performed. Firstly, around 300 mg of each sample was subjected to an acid hydrolysis with 72% $H_2SO_4$ (w/w) at 30 °C for 1 h (0.01 mL/mg of sample). After the initial hydrolysis, the mixture was diluted with deionized water to obtain a 4% $H_2SO_4$ solution and heated to 121 °C for 60 min in an autoclave. The resulting suspensions were filtered through a sintered glass crucible (number 3) previously weighed. The crucibles were then washed twice and dried in an oven at 105 °C for 24 h and the acid-insoluble lignin was determined. The hydrolysate solution obtained after the two-step acid hydrolysis was used to determine the amount of acid-soluble lignin. The UV absorbance of the hydrolyzate at 205 nm was measured using a Jenway 7305 spectrophotometer (Bibby Scientific Limited, Sttafordshire, UK). An acid-soluble lignin absorptivity value of 110 g·L$^{-1}$·cm$^{-1}$ was used (Ehrman 1996). In addition to the above characterization, the ash contents of the lignin precipitates were also measured with the following procedure: 800 ºC during 3 h until no black particles were observed (ILI).

Thermogravimetric analysis (TGA) and differential scanning calorimetry (DSC) were conducted with the different samples to study their thermal properties. Thermogravimetric measurements were made using a TGA/DSC 1 thermobalance (Mettler Toledo, Greifensee, Switzerland). Scans were run from 30 to 800 °C at 10 °C·min$^{-1}$ under a nitrogen flow rate of 80 mL·min$^{-1}$. The glass transition temperature ($T_g$) of each sample was measured using DSC 1 differential scanning calorimeter (Mettler Toledo). Scans were run from 30 to 200 °C at 10 °C·min$^{-1}$ under a nitrogen flow rate of 10 mL·min$^{-1}$.

Fourier transform infrared spectroscopy (FTIR) analysis of samples was measured using a Spectrum Two™ instrument by Perkin Elmer (Massachusetts, USA) equipped with an attenuated total reflectance (ATR) module. The spectra of each sample, from 4000 to 450 cm$^{-1}$ wavenumbers, were recorded with a resolution of 4 cm$^{-1}$ and 20 scans. The software used to interpret the results was the Spectrum 10™ (Perkin Elmer, Massachusetts, USA).

The lignin structure was characterized using 2D-NMR spectroscopy. To carry out the analysis, around 40 mg of each sample was dissolved in 750 µL of DMSO-$d_6$. 2D-NMR spectra were performed at 25 °C on a Bruker AVANCE III 500 MHz instrument (Massachusetts, USA) equipped with a cryogenically cooled 5 mm TCI gradient probe with inverse geometry. Spectral widths were 5.8 KHz and 20.8 KHz for $^1$H and $^{13}$C dimensions, respectively; a relaxation delay of 1 s was used.

## RESULTS AND DISCUSSION

### Black Liquor Characterization

The physico-chemical properties of the wheat straw black liquors used in this work are shown in Table 1. The black liquors obtained from wheat straw pulping had a density of 1.0109 g/mL and a pH value of 10.72, which was due to the residual NaOH from soda pulping. Data regarding the total dry solids and ash content are also showed in Table 1. The ash content of this soda black liquor (15.97 g/L) was similar or even lower than the black liquor values obtained by others cooking processes, which used other pulping reagents and different raw materials (Minu *et al.* 2012). The initial amount of dissolved lignin in the black liquors was estimated by a simple mass balance, considering the amount of lignin in the raw wheat straw and in the resulting wheat straw soda pulp. Monosaccharides, acetyl groups, 5-hydroxymethyl furfural (HMF), and furfural that were dissolved in the black liquors after soda pulping were also measured using HPLC analysis. In addition to the organic component analysis, the electrical conductivity of the spent liquor was also measured and showed in Table 1.

**Table 1.** Composition and Physical Data for the Untreated Black Liquor

| Black liquor | |
|---|---|
| pH | 10.72 ± 0.16 |
| Density (g/mL) | 1.0109 ± 0.0021 |
| Total Dry Solids (g/L) | 32.55 ± 0.10 |
| Ash (g/L) | 15.97 ± 0.40 |
| Lignin (g/L) | 16.06 |
| Total monosaccharides (g/L) | 3.812 |
| Glucose (g/L) | 0.127 |
| Xylose (g/L) | 1.682 |
| Arabinose (g/L) | 0.706 |
| Acetyl groups (g/L) | 1.297 |
| HMF (g/L) | 0.000 |
| Furfural (g/L) | 0.000 |
| Electrical conductivity (mS/cm) | 14.63 ± 0.01 |

Values given are the average of three measurements and the standard deviation.

### Precipitation Yield and Chemical Composition of Lignin Fractions

Table 2 shows that the yield of the lignin recovered from the black liquor depended upon the acid used for the acid precipitation process. At a given pH precipitation value, the yield of precipitate, as well as lignin, was higher using phosphoric acid, which was followed by sulphuric acid, and then hydrochloric acid with the lowest yield (13.64 to 15.26, 11.96 to 12.95, and 10.49 to 11.87 g/L of precipitate per volume of black liquor, respectively for pH 2; and 14.52 to 16.38, 12.14 to 13.21, and 10.05 to 12.17 g/L of precipitate per volume of black liquor, for pH 4). Similar results were reported by Ibrahim *et al.* (2004), who indicated that the precipitated lignin yield is influenced by the type of acid used, *i.e.,* if it was monoprotic, diprotic, or triprotic.

Table 2 data also showed that the precipitation pH slightly affected the amount of precipitate generated; *i.e.*, the recovered precipitate was higher when the pH of the black liquors was decreased to pH 2. However, the precipitation pH of 2 or 4 had no effect on the amount of lignin that was recovered. The higher precipitate yield at pH 2 was due to the precipitation of non-lignin components, such as low molecular weight carbohydrates

or their degradation products. Based on these results, the majority of the lignin in the soda black liquor was precipitated at pH 4. Finally, the concentration of the acid that was used did not have an effect on the lignin that was recovered.

The chemical composition of the precipitated fractions, shown in Table 2, indicated that the percentage of acid soluble lignin (ASL) was higher when the acidification pH was 2 using sulphuric acid (*i.e.*, 3.32% to 5.10% from 4 to 2 pH) and hydrochloric acid (*i.e.*, 4.65% to 5.49% from 4 to 2 pH). ASL is formed by low molecular weight degradations products and hydrophilic derivatives of lignin (Yasuda *et al.* 2001). Thus, a major proportion of the ASL at pH 2 can be explained by the increase of low molecular weight lignins when the pH value of the black liquor was decreased. Furthermore, a low precipitation pH could also increase the water solubility of black liquor components (Alekhina *et al.* 2015).

Additionally, the proportion of acid-insoluble lignin (*i.e.*, Klason lignin) was different within each acid, being higher when the pH of the black liquors was decreased to 4, excepting in the case of hydrochloric acid. As has been mentioned previously, a lower pH value could increase the amount of other products different from lignin, explaining these results.

Although all the precipitated lignin samples were twice washed thoroughly with deionized water in order to remove possible contaminants, the ash levels in the crude precipitates were higher than was expected (Table 2). However, these results can be explained as being due to the high silica (7.3%) and ash (9.6%) levels contained in the raw wheat straw prior to pulping (Pan and Sano 2005). These high values of ash and silica in the raw material could generate an incomplete incineration during the ash determination caused by fusion of ash and silica (Pan and Sano 2005). Part of the silica in the wheat straw was dissolved into the Specel® pulping liquors, which then precipitates when spent black liquors are acidified; this results in the higher ash levels obtained in the crude precipitates. When phosphoric acid is used for decreasing the pH value to 2, it results in higher ash levels in the crude precipitates, due to lower solubility of the phosphate salts. The amounts of Klason lignin, ASL, and inorganic ash in the black liquor precipitates obtained from using different acids and pH values were slightly different when compared to one another.

## Lignin-Rich Fraction Characterization
*Thermal properties*

Differential scanning calorimetry (DSC) was performed in order to establish the thermal characteristics of the lignin. In order to avoid the influences of trace levels water, all of the samples were dried in an oven for 24 h at 60 °C to remove the moisture.

The glass transition temperatures ($T_g$) of all lignin-rich precipitates were found to be between 119 and 136 °C. These results are similar to observations made with other soda lignins from different raw materials (Tejado *et al.* 2007; Toledano *et al.* 2010a and 2010b). In general, soda lignins show relative low glass transition temperature values; however, as it is observed in our work and in the literature, these values can vary. The glass transition temperature ($T_g$) is different for each polymer. When the polymer is cooled below this temperature, it becomes hard and brittle, just like glass. Some polymers are used above their glass transition temperatures, and some are used below their values. For this reason, this is an important property for polymeric materials, particularly for their applications.

**Table 2.** Yield and Chemical Composition of Lignin Fractions

| Exp. | Acid (conc.) | pH | Acid spent (mL/L BL) | Yield of precipitated fractions | | Chemical composition of precipitated fractions | | |
|------|------|------|------|------|------|------|------|------|
| | | | | (g/L BL) | Yield as pure lignin (g/L BL) | Klason lignin | ASL | Ash |
| | | | | | | [% of o.d. sample] | | |
| 1 | $H_2SO_4$, (20%) | | 55.24 | 17.77 | 11.96 | 62.19 | 5.10 | 28.36 |
| 2 | $H_2SO_4$, (60%) | 2 | 19.05 | 18.01 | 12.95 | 67.99 | 3.90 | 28.65 |
| 3 | $H_2SO_4$, (95%) | | 5.71 | 18.25 | 12.31 | 63.38 | 4.05 | 29.97 |
| 4 | $H_2SO_4$, (20%) | | 28.57 | 16.60 | 12.43 | 71.08 | 3.82 | 28.30 |
| 5 | $H_2SO_4$, (60%) | 4 | 10.00 | 17.04 | 12.14 | 67.95 | 3.32 | 30.30 |
| 6 | $H_2SO_4$, (95%) | | 2.86 | 17.03 | 13.21 | 74.09 | 3.49 | 30.68 |
| 7 | $H_3PO_4$, (20%) | | 185.71 | 22.19 | 13.64 | 58.58 | 2.90 | 46.15 |
| 8 | $H_3PO_4$, (60%) | 2 | 59.05 | 23.99 | 15.26 | 60.56 | 3.07 | 47.15 |
| 9 | $H_3PO_4$, (85%) | | 24.76 | 25.34 | 15.21 | 56.55 | 3.48 | 47.47 |
| 10 | $H_3PO_4$, (20%) | | 62.86 | 21.60 | 16.38 | 72.48 | 3.35 | 29.98 |
| 11 | $H_3PO_4$, (60%) | 4 | 20.00 | 22.46 | 14.88 | 63.10 | 3.13 | 30.11 |
| 12 | $H_3PO_4$, (85%) | | 8.57 | 19.56 | 14.52 | 71.04 | 3.20 | 30.54 |
| 13 | HCl, (1M) | | 180.95 | 15.43 | 11.87 | 71.47 | 5.49 | 25.34 |
| 14 | HCl, (2M) | 2 | 90.48 | 16.15 | 10.49 | 59.65 | 5.33 | 28.23 |
| 15 | HCl, (3M) | | 64.76 | 16.43 | 11.74 | 66.21 | 5.24 | 27.99 |
| 16 | HCl, (1M) | | 112.38 | 13.53 | 10.05 | 69.44 | 4.85 | 27.99 |
| 17 | HCl, (2M) | 4 | 53.33 | 15.63 | 10.17 | 60.17 | 4.91 | 27.16 |
| 18 | HCl, (3M) | | 39.05 | 16.46 | 12.17 | 69.27 | 4.65 | 27.07 |

In addition to the DSC analysis, thermogravimetric analysis (TGA) was also performed on the samples. The results are presented in Fig. 1, which shows the *TG* (weight loss of compounds in relation to the temperature of thermal degradation) and *DTG* (rate of weight loss) curves. These curves yielded different degradation steps. The initial weight loss was due to water evaporation, as well as the gaseous emissions of carbon monoxide, carbon dioxide, and other pyrolysis products. The second weight loss phase is attributed to the lignin, in the temperature interval of 200 to 450 ºC. This extended range is due to the complex structure of the lignin (Domínguez *et al.* 2008). The thermal degradation in this region involved fragmentation of inter-unit linkages between phenolic hydroxyl, carbonyl, and benzylic hydroxyl groups, releasing monomeric phenols (El-Saied and Nada 1993; Domínguez *et al.* 2008; González *et al.* 2009). At temperatures above approximately 450 °C, it was observed that the third and last stage was due to the condensation and/or decomposition reactions of the aromatic rings (Sun *et al.* 2001; Domínguez *et al.* 2008).

The TGA results for each of the samples were virtually identical to one another, which implied that the type of acid used, their concentration, and the final acidification pH did not affect the thermal properties of the recovered lignins. However, the lignin samples precipitated with phosphoric acid at pH 2 exhibited a second stage thermal degradation that began slightly earlier (at 185 °C), which was attributed to the greater quantity of phosphoric acid that was used. This second stage began between 200 and 210 °C for the other samples. Different thermal decomposition stages observed for lignin with TGAs are in accordance with other literature reports (Domínguez *et al.* 2008; González *et al.* 2009; Ibrahim *et al.* 2011; Hussin *et al.* 2014).

The amounts of residues remaining after TGA are also shown in Fig. 1. These results (above 40%) revealed that the obtained soda lignins are stable at high temperature, which is attributed to the formation of highly condensed aromatic structures above 450 °C (Tejado *et al.* 2007; Ibrahim *et al.* 2011).

Additionally, the DTG curves are shown in Fig. 1. The peak of that curve, *DTG*$_{max}$, was similar in all the samples, except for the lignin precipitated at 2 pH using phosphoric acid (85%). The *DTG*$_{max}$ for this simple was 276 °C, while the *DTG*$_{max}$ values for the others samples ranged between 330 and 350 °C. A possible explanation for this observation is the higher levels of monosaccharide degradation products contained in this sample. *DTG*$_{max}$ is expressed as a single temperature value and can be used to characterize the material's thermal stability (Ibrahim *et al.* 2011). The value of this property could be important to determine which applications this lignin can be used, particularly if it is exposed briefly to moderately high temperatures.
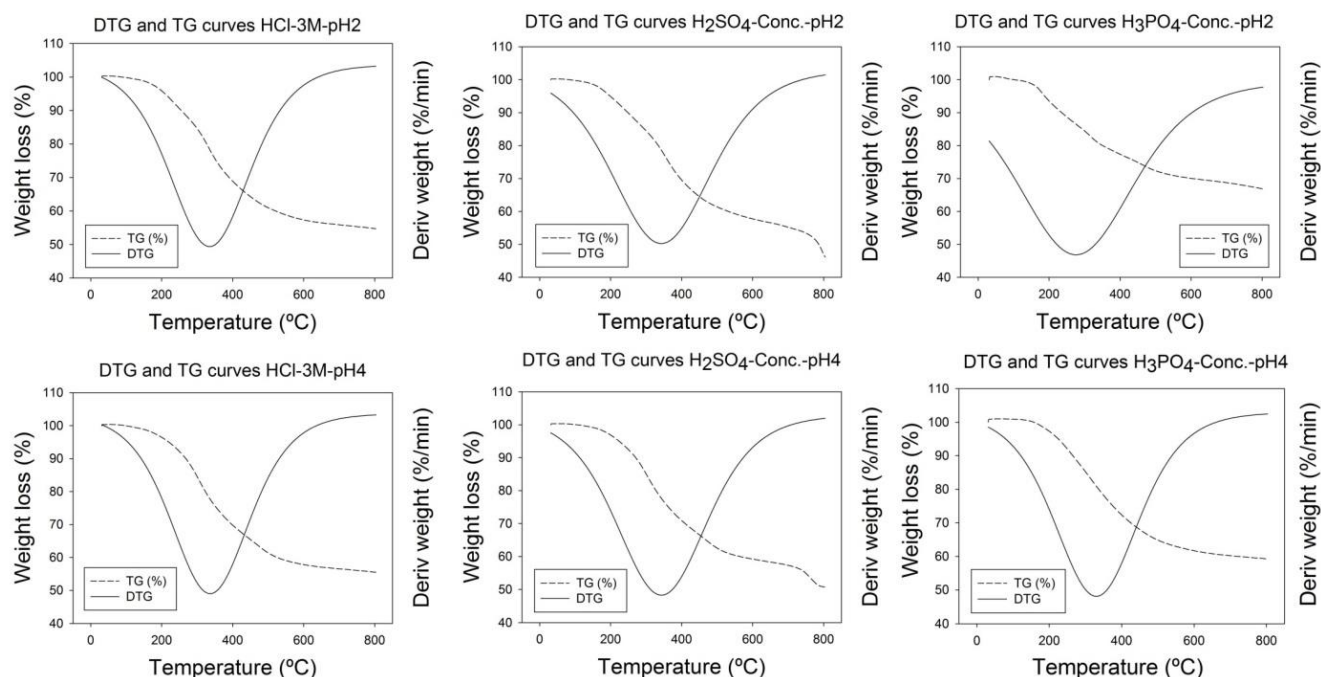


**Fig. 1.** TG and DTG curves of different lignin samples obtained from thermogravimetric analysis (TGA). Note: Conc. means concentrated values of sulphuric acid (95%) and phosphoric acid (85%).

*Lignin structure*

The obtained lignin samples precipitated from various inorganic acids were analysed by ATR-IR spectroscopy to acquire information about the soda lignin structure. FTIR absorption spectra of the precipitated soda lignin were recorded in the 4000 to 450 cm$^{-1}$ region (Figs. 2, 3, and 4).

All spectra showed a strong wide band between 3500 and 3100 cm$^{-1}$, which was assigned to aromatic and aliphatic OH groups. This band, which was centered at approximately 3290 cm$^{-1}$ in the spectra, is caused by the hydrogen bonding of hydroxyl and phenolic groups (Minu *et al.* 2012).

Absorption bands located at 2920 and 2850 cm$^{-1}$ are related to the symmetrical and asymmetrical C-H stretching of the methyl and methylene groups, respectively (Alekhina *et al.* 2015). The absorption band located around 1460 cm$^{-1}$ is attributed to the asymmetric deformation of C-H vibration. All of these absorption bands were observed in the samples with more or less intensities.

The lignin skeleton was identified in each IR spectra. This skeleton showed bands located at 1595, 1508, and 1422 cm$^{-1}$, which are related to aromatic ring vibrations of the lignin. Figure 3, which illustrates the spectra of the lignin precipitated with phosphoric acid at pH 2, showed a lower intensity of these bands than the spectra of the lignin precipitated with the same acid at pH 4. This was probably related to the existence of high amounts of degradation products and monosaccharides in the sample precipitated at pH 2 with phosphoric acid. Additionally, an absorption signal between 1665 and 1655 cm$^{-1}$ was identified in all the samples; this signal is assigned to stretching vibrations of conjugated carbonyl groups in the lignin structure.

Bands at 1340 to 1330 cm$^{-1}$ are assigned to the vibrations of syringyl rings and stretching vibrations of C-O bonds. Additionally, absorptions bands that are characteristic to the vibrations of guaiacyl rings and stretching vibrations of C-O bonds were located at 1272 to 1220 cm$^{-1}$. In all lignin samples, except for the one obtained with phosphoric acid at pH 2, an absorption band was observed at 1328 cm$^{-1}$, which indicated that the wheat straw soda lignin exhibited S bands. A band at 1100 cm$^{-1}$ was observed in all the lignin samples except for the one obtained with phosphoric acid at pH 2. It was assigned to the deformation vibrations of C-H bonds in the aromatic rings and to the deformations vibrations of C-O bonds in the primary alcohols (Minu *et al.* 2012).

Lignin samples that were precipitated from black liquors with phosphoric acid at any concentration down to pH 2 showed in their spectra different bands, for instance, a signal at 1047 cm$^{-1}$ which is indicative of hemicelluloses. Thus, a lower precipitation pH for the case of phosphoric acid affected the general structure of the black liquor lignin that was precipitated. Additionally, some of the bands related to the deformation vibrations of C-H bonds in the aromatic rings (located at 945, 870 to 850, 817, and 780 cm$^{-1}$) were also identified in each IR spectra of the various lignins.

Finally, the use of sulphuric acid to precipitate the lignin from soda black liquor showed a characteristic band at 617 cm$^{-1}$, which was attributed to C-S stretching (Ibrahim *et al.* 2004). Except for the precipitated lignin obtained with phosphoric acid at pH 2 (which apparently had a higher level of contamination with hemicelluloses), all the IR spectra were very similar to one another, which suggested that the general compositions of the precipitated soda lignins were not affected by the acids used or their concentration levels. All of these assignments are based on literature reports (Ibrahim *et al.* 2004; Tejado *et al.* 2007; González *et al.* 2009; Minu *et al.* 2012; Hussin *et al.* 2014; Alekhina *et al.* 2015).
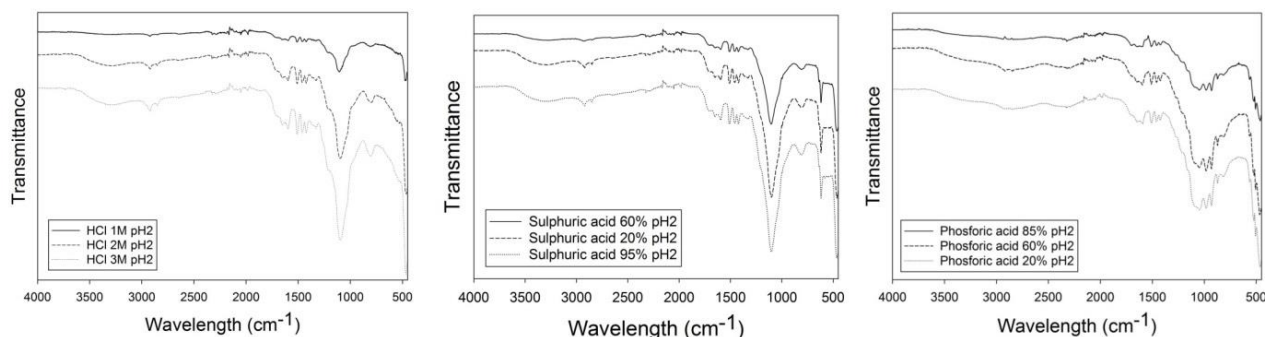
**Fig. 2.** FT-IR spectra of wheat straw lignin precipitated at pH 2 using phosphoric, sulphuric, and hydrochloric acid at the three concentrations levels (wavenumbers of 450 to 4000 cm⁻¹)
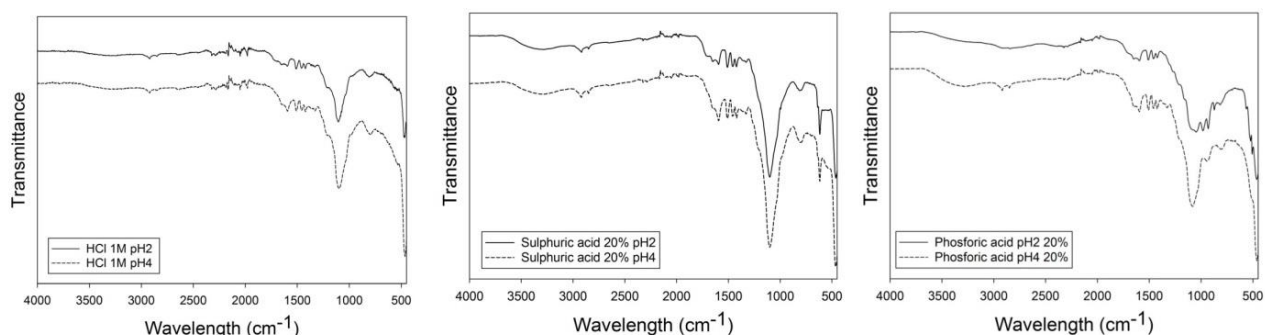


**Fig. 3.** FT-IR spectra of wheat straw lignin precipitated at pH 2 and 4 using phosphoric, sulphuric, and hydrochloric acid at the lowest concentration level (wavenumbers of 450 to 4000 cm⁻¹)
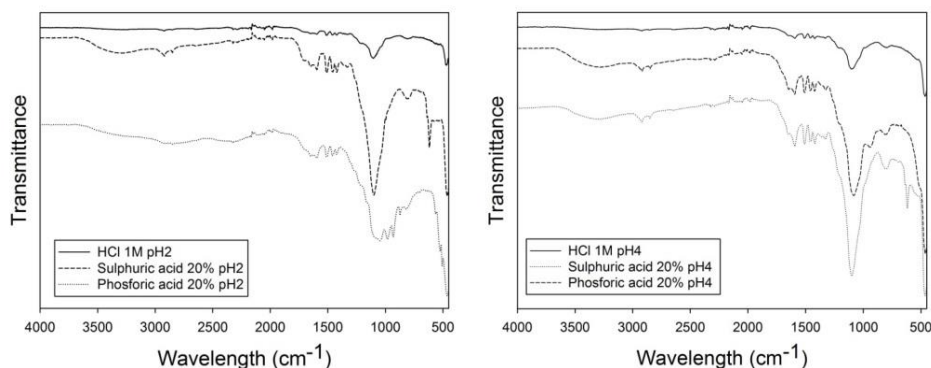


**Fig. 4.** FT-IR spectra of wheat straw lignin precipitated at pH 2 (left) and 4 (right) using phosphoric, sulphuric, and hydrochloric acid at the lowest concentration level (wavenumbers of 450 to 4000 cm⁻¹)

Two-dimensional HSQC-NMR spectroscopy was conducted to further characterize the structure of the various precipitated lignin samples obtained from the soda black liquor. The 2D HSQC spectra of the lignin samples were characterized by three clearly differentiated regions: an aliphatic region (non-oxygenated), an oxygenated aliphatic region, and an aromatic region. The non-oxygenated aliphatic region represents lignin degradation products, as well as other unidentified compounds; thus, this region is generally the least interesting portion of the spectra regarding the precipitated lignin structure.

The type of acid used, as well as the acidic precipitation pH value used, did not have any influence on the major structural features of the precipitated lignin from the soda black liquor. Figure 5 illustrates the HSQC spectra recorded for lignin precipitated using $H_2SO_4$ (95%) at a pH value of 2. Figure 5 is representative of the spectra recorded for the others samples analyzed. All lignin samples showed similar spectra, with no appreciable differences among them.

The two-dimensional HSQC-NMR analysis more deeply focused on the oxygenated aliphatic and the aromatic regions of the spectra (Fig. 5). The assignments of the main cross-peaks are based on the lignin studies reported in the literature (del Río *et al.* 2011, 2012; Rencoret *et al.* 2011; You *et al.* 2013; Savy *et al.* 2015), and are presented in Table 3.

The oxygenated aliphatic region of the HSQC spectra contained the signals corresponding to $\beta$-$O$-4' ether units (Substructure A). The $C_\gamma$-$H_\gamma$ correlations in $\beta$-$O$-4' substructures were observed at $\delta_C/\delta_H$ 59.8/3.2 to 3.6 ppm. Furthermore, the $C_\alpha$-$H_\alpha$ correlations in $\beta$-$O$-4' substructures in syringyl (S) units were observed at $\delta_C/\delta_H$ 71.6/4.8 ppm. In addition, $C_\beta$-$H_\beta$ correlations in $\beta$-$O$-4' substructures were observed at $\delta_C/\delta_H$ 85.7/4.1 ppm, and approximately at $\delta_C/\delta_H$ 82.3/4.3 ppm for both $\beta$-$O$-4' substructures linked to guaiacyl (G) units and to $\gamma$-acetylated $\beta$-$O$-4' substructures linked to S units. Signals corresponding to C-H in the methoxyl groups were observed at $\delta_C/\delta_H$ 55.5/3.7 ppm. The correlations approximately at $\delta_C/\delta_H$ 86.7/5.5 ppm and 79.1/5.6 ppm were assigned to $C_\alpha$-$H_\alpha$ in phenylcoumaran ($\beta$-5') substructures (C) and to $C_\alpha$-$H_\alpha$ in $\alpha,\beta$-diaryl substructures (E), respectively.

The cross-peaks in the aromatic region of the HSQC spectra correspond to the aromatic structures in the lignin. The main signals corresponding to $C_{2/6}$-$H_{2/6}$ correlations for S units and for $C_\alpha$-oxidized S units (S') were identified at approximately $\delta_C/\delta_H$ 103.6/3.7 ppm and 106.1/7.2 ppm, respectively. Furthermore, G units had different correlations corresponding to $C_2$-$H_2$ ($\delta_C/\delta_H$ 110.7/7.0 ppm), $C_5$-$H_5$ ($\delta_C/\delta_H$ 116.3/6.5 and 6.9 ppm) and $C_6$-$H_6$ ($\delta_C/\delta_H$ 118.4/6.8 ppm). Additionally, signals corresponding to $C_{3/5}$-$H_{3/5}$ and $C_{2/6}$-$H_{2/6}$ correlations for *p*-hydroxyphenyl (H) units were also identified at around at $\delta_C/\delta_H$ 114.9/6.7 and 128.7/7.0 ppm, respectively.

Additional cross-signals in this aromatic region suggested the existence of esterified aromatic lignin structures, such as *p*-coumarate (PCA) and ferulate (FA) (Savy *et al.* 2015). Signal corresponding to $C_2$-$H_2$ correlation in ferulate (FA) was identified in the approximately at $\delta_C/\delta_H$ 111.3/7.4 ppm, while signals at 116.5/6.4 ppm and 129.5/7.5 ppm were attributed to $C_{3/5}$-$H_{3/5}$ and $C_{2/6}$-$H_{/6}$ correlations for *p*-coumarate (PCA), respectively. Moreover, the cross-signal at $\delta_C/\delta_H$ 143.7/7.5 ppm was assigned to the $C_\alpha$-$H_\alpha$ correlation for PCA and FA substructures. Additionally, other signals observed in the aromatic spectra correspond to $C_2$-$H_2$ correlation (at $\delta_C/\delta_H$ 113.2/7.4 ppm) and $C_6$-$H_6$ correlation (at $\delta_C/\delta_H$ 121.8/7.1 ppm) for cinnamyl aldehyde end-groups (J). Cross-signals for flavone tricin were also found in the HSQC spectra. These tricin signals correspond to $C_6$-$H_6$ (at $\delta_C/\delta_H$ 98.6/6.3 ppm), $C_8$-$H_8$ (at $\delta_C/\delta_H$ 93.7/6.4 ppm), and $C_{2/6}$-$H_{2/6}$ (at $\delta_C/\delta_H$ 103.8/7.3 ppm). This also confirmed that the flavone substructure is linked to the soda lignin from wheat straw (del Río *et al.* 2012). This observation supports the assertion that the linkages between the flavone tricin substructure and lignin are alkaline-stable, as has been reported in the literature (You *et al.* 2013; Savy *et al.* 2015).

**Table 3.** Assignments of the Lignin $^1$H -$^{13}$C Correlation Signals in the HSQC Spectra of Lignin-Rich Precipitate Isolated from Wheat Straw Black Liquor

| | | $^1$H-$^{13}$C HSQC Spectrum |
|---|---|---|
| **Label** | **δ$_C$/δ$_H$ (ppm)** | **Attribution** |
| OMe | 55.5/3.7 | C-H in methoxy groups |
| A$_\square$ | 59.8/3.2 and 3.6 | C$_\gamma$-H$_\gamma$ in β-O-4' substructures (A) |
| X$_5$ | 63.1/3.40-3.72 | C$_5$-H$_5$ in β-D-xylopyranoside (X) |
| Aα$_{(S)}$ | 71.6/4.8 | C$_\alpha$-H$_\alpha$ in β-O-4' in S units erythro (A) |
| X$_2$ | 72.5 | C$_2$-H$_2$ in β-D-xylopyranoside (X) |
| X$_3$ | 73.7 | C$_3$-H$_3$ in β-D-xylopyranoside (X) |
| X$_4$ | 75.4 | C$_4$-H$_4$ in β-D-xylopyranoside (X) |
| Eα | 79.1/5.6 | C$_\alpha$-H$_\alpha$ in α,β-diaryl substructures (E) |
| Aβ$_{(G)}$ and A'β$_{(S)}$ | 82.3/4.3 | C$_\beta$-H$_\beta$ in β-O-4' structures linked to G units and in (A) $\square$-acetylated β-O-4' units linked to a S unit (A') |
| Bα | 84.9/4.6 | C$_\alpha$-H$_\alpha$ in resinol (*b-b*) substructures (B) |
| Aβ | 85.7/4.1 | C$_\beta$-H$_\beta$ in β-O-4' substructures (A) |
| Cα | 86.7/5.5 | C$_\alpha$-H$_\alpha$ in β-5' (phenylcoumaran) substructures (C) |
| T$_8$ | 93.7/6.4 | C$_8$-H$_8$ in tricin (T) |
| T$_6$ | 98.6/6.3 | C$_6$-H$_6$ in tricin (T) |
| S$_{2/6}$ | 103.6/6.7 | C$_{2/6}$-H$_{2/6}$ in etherified syringyl units (S) |
| T$_{2'/6'}$ | 103.8/7.3 | C$_{2'/6'}$-H$_{2'/6'}$ in tricin |
| S'$_{2/6}$ | 106.1/7.2 | C$_{2/6}$-H$_{2/6}$ in oxidized (C$_\square$=O) phenolic syringyl units (S') |
| G$_2$ | 110.7/7.0 | C$_2$-H$_2$ in guaiacyl units (G) |
| FA$_2$ | 111.3/7.4 | C$_2$-H$_2$ in ferulate (FA) |
| J$_{2(G)}$ | 113.2/7.4 | C$_2$-H$_2$ in cinnamyl aldehyde end-groups (J) |
| H$_{3/5}$ | 114.9/6.7 | C$_{3/5}$-H$_{3/5}$ in *p*-hydroxyphenyl units (H) |
| G$_5$ | 116.3/6.5 & 6.9 | C$_5$-H$_5$ in guaiacyl units (G) |
| PCA$_{3/5}$ | 116.5/6.4 | C$_{3/5}$-H$_{3/5}$ in *p*-coumaric units (PCA) |
| G$_6$ | 118.4/6.8 | C$_6$-H$_6$ in G units |
| J$_{6(G)}$ | 121.8/7.1 | C$_6$-H$_6$ in cinnamyl aldehyde end-groups (J) |
| H$_{2/6}$ | 127.8/7.0 | C$_{2/6}$-H$_{2/6}$ in *p*-hydroxyphenyl units (H) |
| PCA$_{2/6}$ | 129.5/7.5 | C$_{2/6}$-H$_{2/6}$ in *p*-coumarate (PCA) |
| PCAα and FAα | 143.7/7.5 | C$_\alpha$-H$_\alpha$ in PCA and FA |
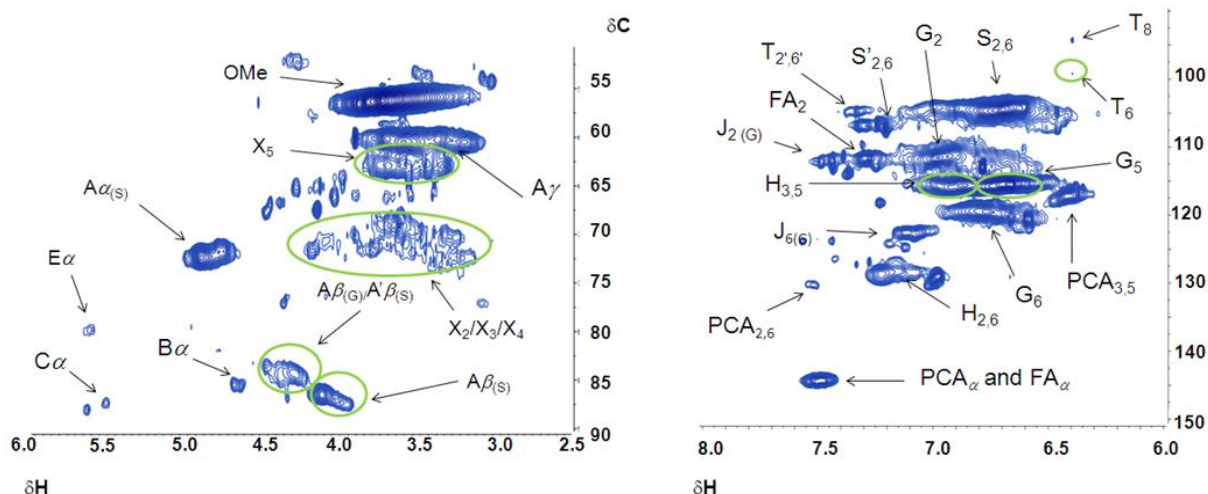
**Fig. 5.** Two-dimensional HSQC-NMR spectra of oxygenated aliphatic ($\delta_C/\delta_H$ 50–90/2.5–6.0) and aromatic ($\delta_C/\delta_H$ 90–106/6.0–8.0) regions for lignin isolated from soda black liquors (Specel® process) using wheat straw (sample precipitated with $H_2SO_4$ (95%) to pH 2). Labels refer to identified lignin sub-units, whose signal assignment is reported in Table 3.

Finally, some carbohydrate-related cross-peaks in the spectra have been attributed to $\beta$-D-xylopyranoside substructures (Fig. 5). The resonance of the $C_2$-$H_2$ may be found at 72.5/3.1 ppm, while that of $C_3$-$H_3$ is located at 73.7/.2 ppm. Cross-signals at 75.4/3.6 ppm and 63.1/3.40-.72 ppm are assigned to the $C_4$-$H_4$ and $C_5$-$H_5$ correlation, respectively. Such carbohydrates substructures could be derived from the carbohydrates present in the soda black liquors (see Table 1), that have been left in the lignin-rich precipitate the acidic precipitation.

## Economic Evaluation

The economics of the acid precipitation process has been evaluated with regards to the costs of the inorganic acids employed (Data were obtained from Brenntag company.). Acid cost data were calculated based on the highest concentration for each acid. The total amounts of lignin, which is part of the lignin-rich precipitates, as well as the final pH values, were taken into account in the economic evaluation.

The most cost-effective option for acidic precipitation of lignin was using sulphuric acid. In this case, the cost to obtain 1 kg of lignin ranged between 0.30 and 0.64 € for pH 4 and pH 2, respectively. Acid precipitation using phosphoric acid was the most expensive option; the amount of this acid required to obtain 1 kg of lignin had a cost that ranged between 1.44 and 3.97 € (for pH 4 and pH 2, respectively). Hydrochloric acid was an intermediate cost option. The cost to obtain 1 kg of lignin ranged between 0.90 and 1.53 €. Economic data for the inorganic acid used (HCl, $H_2SO_4$, and $H_3PO_4$) were 1.11, 1.37, and 2.43 €/L obtained from our laboratory provider.

Even though the highest yield was achieved with $H_3PO_4$, the most economical acid to use was $H_2SO_4$. This is largely due to the high amounts of $H_3PO_4$ needed to neutralize the alkali in the black liquor and lower the pH value from pH 10.72 to pH 4 or 2 (Table 2). As has been mentioned previously, the cost of $H_3PO_4$ is almost twice the cost of $H_2SO_4$. Based on all these factors, $H_2SO_4$ is the most suitable acidifying agent for the precipitation of lignin from Specel® soda black liquor; there were no appreciable differences observed in the chemical structures of the isolated lignins obtained using various inorganic acids. The only exception to this observation was the case of $H_3PO_4$ at pH 2.

## CONCLUSIONS

1. The highest yield of lignin was achieved using phosphoric acid as the precipitation agent; however, it was the most expensive option. Thus, the best economical option is to use the sulphuric acid. The chemical structures of the precipitated lignin with sulphuric acid observed in this study were very similar to those reported in the literature for wheat straw.

2. The optimum precipitation pH value was 4 for sulphuric, phosphoric, and hydrochloric acids in terms of getting the maximum lignin yield.

3. The lignin obtained by lowering the pH value of the black liquor from pH 10.72 to pH 2 using phosphoric acid had different thermal properties than the other precipitated lignins examined in this study; this was attributed to the high amounts of phosphoric acid used in the precipitation process to pH 2.

## ACKNOWLEDGMENTS

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