Deep Delignification of Woody Biomass by Repeated Mild Alkaline Treatments with Pressurized O₂

Jing-Xian Wang, Shusaku Asano, Shinji Kudo, and Jun-ichiro Hayashi

ABSTRACT: Delignification is essential in effective utilization of carbohydrates of lignocellulosic biomass. Characteristics of the delignification are important for the yield and property of the resulting carbohydrates. Oxidation with O₂ of biomass in alkaline water can potentially produce high-purity cellulose at high yield. The present authors chose a Japanese cedar and investigated its oxidative delignification at 90 °C. The delignification selectivity was determined mainly by the chemical structures of lignin and cellulose. Treatment conditions, except for temperature, hardly changed the relationship between delignification rate and cellulose retention. During the treatment, dissolved lignin underwent chemical condensation in the aqueous phase. This “unfavorable” condensation consumed O₂-derived active species, slowing down further delignification. Repeated short-time oxidation with renewal of alkaline water suppressed the condensation, enhancing the delignification. Repetition of 2-h treatments four times achieved 96% delignification, which was 8% higher than a single 8-h treatment at 130 °C.

1. INTRODUCTION

Lignocellulosic biomass is a promising carbonaceous resource to produce sustainable fuels, chemicals, and biological products owing to its renewability and availability. Economically feasible chemical production from lignocellulosic biomass has, however, yet to be achieved. Lignin is a critical obstacle to the production of bioethanol1 and carbohydrate-derived chemicals2 as well as materials, while it can be converted into value-added products.3

Alkaline treatment with O₂ has great potential for industrial delignification with economic and environmental feasibilities.4 Dissolving oxidizing agents such as H₂O₂, H₂O, and O₂ into alkaline water leads to formation of active species such as OH− radicals and then decomposition of hemicellulose and lignin by reactions such as electrophilic substitution, side-chain displacement, and oxidative cleavage of aromatic nuclei.5−7 This combination of an alkaline environment and an oxidizing agent is thus effective for the delignification. Due to lower reactivity than H₂O₂, O₂ is normally used with high temperatures over 150 °C to promote the delignification. Klinke et al.8 reported 62% delignification from wheat straw at 195 °C with Na₂CO₃ under 1.2 MPa O₂. Another study by Chang et al. reported 78% delignification from a poplar wood that was treated at 150 °C for 6 h under 1.4 MPa O₂ with Ca(OH)₂.9

Some reports showed that delignification at lower temperatures was effective for reducing the loss of carbohydrates.10,11 For example, Geng et al.11 achieved 50% delignification at 110 °C with a carbohydrate loss of 4.2% from another wheat straw. In those previous studies, the delignification rate was not necessarily maximized because the delignification was investigated as a pretreatment for enzymatic saccharification, which often required a delignification rate as low as 20−65%.13,14 However, deeper removal of lignin is desirable for more production of sugar per consumption of enzyme because the enzyme has a high propensity to be adsorbed to the lignin.15 Minimization of the residual lignin is hence effective for reducing the consumption of enzymes. In addition, rapidly developing technologies of cellulose nanofibers14,16 will demand cellulose with higher purity.

This study aimed to investigate delignification by a low-temperature alkaline treatment with O₂ and its chemical mechanism. A typical Japanese cedar was chosen as feedstock because it had a most refractory type of lignin.17 The hemicellulose of the cedar consists mainly of glucomannan. Removal of glucomannan-linked lignin was more difficult than that of xylan-linked lignin.18 Thus, deep delignification of a Japanese cedar with high carbohydrate retention was challenging. For example, Yoshioka et al. had treated another type of Japanese cedar with an ionic liquid.19 It removed 40% of the original lignin but allowed the loss of hemicellulose (>90%) and that of cellulose (10%). In contrast, Wang et al. removed 98.7% lignin from a red pine with another ionic liquid while preserving 95% of cellulose.20 Such a refractory feature of a Japanese cedar
was appropriate for investigating the possibility of extensive delignification under mild conditions.

2. RESULTS AND DISCUSSION

2.1. Effects of Oxygen on the Delignification and Decomposition of Carbohydrates. Table 1 summarizes the results of alkaline water treatments with or without O₂. The chemical compositions of the resulting solids are listed in Table 2. A positive effect of O₂ on the delignification is evident by comparing run 1 (R1) with O₂ and run 2 (R2) without O₂. R1 and R2 gave delignification rates of 0.536 and 0.084, respectively. The O₂ oxidation of R1 also caused loss of cellulose.

Table 1. Results of Alkaline Treatment

| run ID | conditions              | delignification rate | NPL yield | PRL yield | cellulose retention | xylan | galactan | mannan | pH of suspension after treatment | NaOH consumption, mmol/g-feed |
|--------|-------------------------|----------------------|-----------|-----------|---------------------|-------|----------|--------|----------------------------------|-------------------------------|
| R1     | standard conditions, O₂—8 h | 0.536 | 0.425 | 0.111 | 0.842 | 0.379 | 0.264 | 0.746 | 9.0 | 8.0 |
| R2     | N₂ 2 MPa               | 0.084 | 0.013 | 0.071 | 0.963 | 0.851 | 0.978 | 0.883 | 14.0 | 0 |
| R3     | O₂ 0.5 MPa             | 0.476 | 0.408 | 0.069 | 0.847 | 0.399 | 0.294 | 0.789 | 12.9 | 7.2 |
| R4     | O₂ 3 MPa               | 0.559 | 0.464 | 0.095 | 0.846 | 0.333 | 0.249 | 0.722 | 9.0 | 8.0 |
| R5     | 2 h                    | 0.425 | 0.340 | 0.085 | 0.843 | 0.428 | 0.313 | 0.841 | 13.5 | 4.8 |
| R6     | 4 h                    | 0.486 | 0.387 | 0.099 | 0.847 | 0.402 | 0.295 | 0.809 | 10.1 | 8.0 |
| R7     | 12 h                   | 0.591 | 0.454 | 0.138 | 0.831 | 0.349 | 0.237 | 0.702 | 9.0 | 8.0 |
| R8     | 16 h                   | 0.630 | 0.462 | 0.168 | 0.829 | 0.332 | 0.219 | 0.682 | 8.9 | 8.0 |
| R9     | 20 h                   | 0.666 | 0.481 | 0.185 | 0.833 | 0.317 | 0.217 | 0.65 | 8.7 | 8.0 |
| R10    | 0.5 mol/L              | 0.375 | 0.312 | 0.064 | 0.896 | 0.438 | 0.328 | 0.874 | 7.9 | 5.0 |
| R11    | 1.0 mol/L              | 0.657 | 0.499 | 0.158 | 0.800 | 0.316 | 0.209 | 0.636 | 10.6 | 10.0 |
| R12    | L/S = 5 mL/g           | 0.353 | 0.320 | 0.034 | 0.946 | 0.463 | 0.361 | 0.933 | 8.2 | 4.0 |
| R13    | L/S = 20 mL/g          | 0.666 | 0.476 | 0.181 | 0.757 | 0.323 | 0.21 | 0.601 | 13.8 | 3.4 |
| R14    | 70 °C                  | 0.396 | 0.345 | 0.051 | 0.956 | 0.464 | 0.311 | 0.909 | 13.1 | 6.7 |
| R15    | 130 °C                 | 0.879 | 0.774 | 0.106 | 0.659 | 0.149 | 0.032 | 0.636 | 10.6 | 10.0 |
| R16    | repetitive treatment with solution renewal, 2 × 2-h | 0.701 | 0.472 | 0.229 | 0.736 | 0.152 | 0.105 | 0.364 | 13.9 | 4.9 |

Table 2. Composition of Residues after Alkaline Treatment

| run ID | conditions              | residue composition |
|--------|-------------------------|---------------------|
| R1     | standard conditions, O₂—8 h | lignin | cellulose | hemicellulose | xylan | galactan | mannan | ash |
| R2     | N₂ 2 MPa               | 0.264 | 0.548 | 0.038 | 0.012 | 0.07 | 0.033 |
| R3     | O₂ 0.5 MPa             | 0.393 | 0.46 | 0.063 | 0.022 | 0.062 | 0.016 |
| R4     | O₂ 3 MPa               | 0.29 | 0.538 | 0.039 | 0.013 | 0.072 | 0.033 |
| R5     | 2 h                    | 0.25 | 0.557 | 0.035 | 0.011 | 0.068 | 0.031 |
| R6     | 4 h                    | 0.279 | 0.527 | 0.039 | 0.012 | 0.072 | 0.037 |
| R7     | 12 h                   | 0.243 | 0.566 | 0.037 | 0.011 | 0.069 | 0.027 |
| R8     | 16 h                   | 0.227 | 0.583 | 0.036 | 0.01 | 0.069 | 0.031 |
| R9     | 20 h                   | 0.21 | 0.602 | 0.036 | 0.011 | 0.068 | 0.025 |
| R10    | 0.5 mol/L              | 0.313 | 0.515 | 0.039 | 0.013 | 0.072 | 0.023 |
| R11    | 1.0 mol/L              | 0.221 | 0.59 | 0.036 | 0.01 | 0.068 | 0.029 |
| R12    | L/S = 5 mL/g           | 0.307 | 0.514 | 0.039 | 0.013 | 0.073 | 0.025 |
| R13    | L/S = 20 mL/g          | 0.233 | 0.586 | 0.039 | 0.011 | 0.067 | 0.032 |
| R14    | 70 °C                  | 0.284 | 0.526 | 0.039 | 0.011 | 0.07 | 0.033 |
| R15    | 130 °C                 | 0.12 | 0.746 | 0.026 | 0.002 | 0.051 | 0.006 |
| R16    | repetitive treatment with solution renewal 2-h       | 0.211 | 0.632 | 0.02 | 0.006 | 0.045 | 0.036 |
| R17    | 2 h x 3                | 0.094 | 0.727 | 0.018 | 0.003 | 0.028 | 0.016 |
| R18    | 2 h x 4                | 0.044 | 0.821 | 0.017 | 0.003 | 0.02 | 0.014 |
| R19    | 12 h x 2               | 0.064 | 0.793 | 0.014 | 0.002 | 0.029 | 0.014 |
| R20    | 12 h x 3               | 0.05 | 0.818 | 0.01 | 0.002 | 0.022 | 0.013 |

"Standard conditions in the autoclave were as follows: O₂ 2 MPa, 8 h, NaOH 0.8 mol/L, L/S = 10 mL/g, and 90 °C. Values are based on the steam-treated cedar."
but leaving 84.2% in the solid. The pH after R1, 9.0, was much lower than that after R2, and this was due to the formation of organic acids and CO2. Figure 1 presents a more detailed comparison of the product distribution between R1 and R2. The lignin dissolved into the solution was classified into precipitation recovery lignin (PRL) and nonprecipitated lignin (NPL). As shown in Figure 1a, the introduction of O2 increased the overall rate of dissolution from 0.06 to 0.36 on the cedar carbon basis. The dissolved matter consisted of PRL, organic acids, CO2, methyl ethyl ketone-soluble fraction (MEK-S), and MEK-insoluble material (MEK-IS). The organic acids and CO2 were contributed by not only the oxidative degradation of lignin but also that of carbohydrates. This will be demonstrated later. Some organic acids were also formed in the absence of O2, probably due to reactions preferred in alkaline water such as deacylation and base-catalyzed hydrolysis. The MEK-S was characterized by gas chromatography/mass spectrometry (GC/MS), and the results are shown in Figure S2. The GC/MS detected over 30 types of aliphatic acids, ethers, and esters in MEK-S, but these were not quantified due to very low yields, or the commercial unavailability of the pure standard samples.

Figure 1b shows the lignin conversions. The presence of O2 substantially increased the lignin conversion from 0.1 to 0.53. The major part of this increase was explained by the formation of NPL. The MW distributions of PRL are compared between R1 and R2 in Figure 1c. The PRL from R1 had a broad MW distribution up to 30 000 Da, unlike that from R2. The following
two hypotheses were developed from the presence of a greater MW component in PRL from R1.

(1) MW of PRL from the oxidative lignin extraction was greater than that from the nonoxidative one.

(2) The oxidative and nonoxidative extractions yielded PRL with the same or very similar MW. However, the PRL from the former underwent particular secondary reactions, i.e., chemical condensation (repolymerization) that caused MW increase.

The above hypotheses will be examined later. Figure 1d shows the yields of major organic acids ranging from formic acid to succinic acid. It seemed that the oxalic acid was formed only by the oxidation. It was difficult to exactly determine the contributions of the carbohydrates and lignin to the individual acids.

Understanding of carbohydrate degradation is important to achieve high cellulose yield along with extensive delignification. The oxidation of reagent cellulose, xylan, and five monosaccharides was therefore investigated. The results of 12-h oxidation are summarized in Figure 2. Both the polysaccharides and monosaccharides were oxidized to organic acids, CO2, MEK-S, and MEK-IS, in apparently similar ways to that of the cedar (see Figure 1). Xylan and all of the monosaccharides were degraded completely to alkaline-water-soluble matter, while the conversion of the cellulose was limited to less than 40%. The major products from the monosaccharides were organic acids. The GC/MS detected and identified up to 20 minor compounds in MEK-S, but these were not quantified due to very low yields, or the commercial unavailability of the pure standard samples. The GC/MS result is shown in Figures S4–S10. The monosaccharides and polysaccharides showed different product distributions, as can be seen in Figure 2b. For every monosaccharide, lactic acid yield accounted for more than half of the total yields of the major organic acids. On the other hand, the degradation of cellulose and xylan resulted in much lower lactic acid yield and selectivity than those of the monosaccharides. This difference strongly suggests that the polysaccharides underwent direct oxidative decomposition of sugar units rather than a sequence of formation and decomposition of monomers. For the polysaccharides, rupturing of C1−C2 (α-scission) and C2−C3 (β-scission), forming C1 and C2 organic acids (formic, acetic, glycolic, and oxalic acid), was likely the main degradation pathway.24 On the other hand, for monosaccharides, the rupture of C3−C4 seemed to be important for generating C3 organic acids such as lactic acid. The organic acids formed in R1 (see Figure 1d) showed a small amount of lactic acid. This result was consistent with the above-mentioned direct oxidation of polysaccharides. It is also noted that the lignin was the other important source of formic, acetic, oxalic, and glycolic acids.25 Morone et al.26 investigated alkaline treatment of rice straw at 161–204 °C with pressurized air and found lower organic acids with substantial yields. They speculated that the polysaccharides were first hydrolyzed into monosaccharides, and then the monosaccharides were further oxidized and degraded into organic acids. However, this was not the case in the present alkaline treatment with O2. In other words, at temperatures as low as 90 °C, the oxidative decomposition of cellulose/hemicellulose was initiated by the direct oxidation of sugar units rather than hydrolytic monomerization or oligomerization.

2.2. Remarkable Effects of Solution Renewal. The 54% delignification in R1 suggested that the alkaline treatment with O2 worked well even at 90 °C. However, optimization of treatment conditions was required to achieve deeper delignification. The delignification was hence investigated under various treatment conditions with different combinations of O2 pressure, treatment time, alkali concentration, liquid/solid ratio (L/S), temperature, and operation modes (single-batch treatment and repeated one with renewal of alkaline solution). The results are summarized in Tables 1 and 2. Figure 3 plots the cellulose retention against delignification rate for all of the runs. The dotted line illustrates the general trend.
analyzed as shown in Figure 4. It was difficult to correlate the delignification rate to the consumed amount of alkali because 2-h repetitive treatments achieved a high delignification degree with a small alkali consumption of around 5 mmol/g-feed. Thus, neither the final pH nor the alkali amount was a major factor intensified by the solution renewal. It was therefore implied that the dissolved lignin-derived matter, in particular, PRL, hampered the primary reactions between the solid and active oxidative species in the liquid phase by consuming such species, or otherwise, suppressing their formation. Figure 5a displays the apparent MW distributions of the PRLs from runs with single and repeated 2-h treatments or doubled L/S (R5, R16−R18, and R13) on a lignin-weight basis. In contrast to the MW distribution for the 8-h treatment (R1) and the doubled L/S (R13), those for R5 and R16 had no shoulder peaks at the higher MW side. Figure 5b shows the height-standardized chromatograms. Two-hour repetitive treatments (R5, R16−R18) resulted in almost completely identical MW distributions. It was implied that MW distributions of dissolving lignin were constant when undesirable secondary reactions were inhibited. On the other hand, runs with longer treatment times resulted in a larger shoulder peak. This is explained by the progress of chemical condensation of PRL to form that with MW > 2000. Simultaneous decomposition and dissolution of the refractory LCC occur in the early stage. Then, the hemicellulose components free from LCC are decomposed rapidly in the early stage of the delignification. On the other hand, LCC is gradually decomposed and then its fragments are dissolved into the aqueous phase. The higher refractoriness of mannann, which decomposes only about 10% in the early stage, is attributed to its bonding to the lignin.27−29 Glucomannan-linked lignin is more difficult to remove than xylan-linked lignin.19 Thus, less refractory portions of carbohydrates and lignin easily decompose and dissolve in the early stage. Then, the degradation and dissolution of the refractory LCC occur in the mid and late stages. Simultaneous decomposition and dissolution of lignin and hemicellulose were assumed in previous studies.27,29 However, the linear relationships shown in Figure 6 are for the first time demonstrated in the present study.

2.3. Three Stages of Delignification. As illustrated in Figure 3, the delignification process consists of three stages in terms of cellulose retention. In the early stage (delignification rate <0.4), the loss of cellulose is 0.1 or even smaller. This small loss is attributed to that of noncrystalline cellulose.27 In the mid stage (delignification rate 0.4−0.65), the delignification is associated with little loss of cellulose, for which retention is in a narrow range of 0.80−0.85 (entries 1, 3−9). This trend indicates the progress of selective delignification. However, the delignification in the late stage (delignification rate >0.65) is accompanied by significant loss of cellulose. The lignin remaining in the late stage is likely to exist in the deeper regions of the organic matrix of the cedar. Due to poor accessibility of this lignin to oxidizing species, these have to oxidize the cellulose simultaneously with the lignin.

To investigate the characteristics of the three stages from other viewpoints, the relationship between lignin and hemicellulose retentions was analyzed. Figure 6a−c illustrates the relationships of the delignification rate with xylan, galactan, and mannan retentions, respectively. By comparing the xylan retentions in Figure 6a and the product yields from the treatment of the isolated xylan shown in Figure 2, it is clear that xylan units in the cedar were much more refractory than the isolated xylan. For example, R7 (12-h oxidation) gave xylan retention as high as 0.35, while the isolated xylan was completely decomposed. It is also noted that the retentions of xylan as well as the other hemicellulose components are linearly related to the lignin retention in the mid and late stages. Such refractoriness and the linear relationship are explained reasonably by the presence of so-called lignin−carbohydrate complexes (LCC). Hemicellulose components free from LCC are decomposed rapidly in the early stage of the delignification. On the other hand, LCC is gradually decomposed and then its fragments are dissolved into the aqueous phase. The higher refractoriness of mannan, which decomposes only about 10% in the early stage, is attributed to its bonding to the lignin.19,28 Glucomannan-linked lignin is more difficult to remove than xylan-linked lignin.19 Thus, less refractory portions of carbohydrates and lignin easily decompose and dissolve in the early stage. Then, the degradation and dissolution of the refractory LCC occur in the mid and late stages. Simultaneous decomposition and dissolution of lignin and hemicellulose were assumed in previous studies.27,29 However, the linear relationships shown in Figure 6 are for the first time demonstrated in the present study.
As shown in Figure 1, the presence of O₂ greatly increases the NPL yield. Figure 6d presents the relationship between the NPL yield and the delignification rate. Two master curves fitted for the early stage and the other stages are drawn by a dashed line and a dotted line, respectively. The slope for the early stage is clearly larger than that for the mid and late stages, indicating that the selectivity to NPL is higher at the early stage. In the early-stage delignification, the lignin is likely to be decomposed into small molecules (categorized into NPL for convenience) and thereby dissolved into the alkaline solution. Such rapid lignin removal in the early stage was reported by Shi et al. However, no discussion was found in the literature on the clear difference in the product distribution between the early stage and mid/late ones.

Figure 7 proposes the reaction scheme of the delignification and decomposition of coexisting carbohydrates in alkaline treatment with O₂. The delignification continues in the mid stage but more slowly than that in the early stage, while the lignin is still accessible to oxidizing species. The dissolution of lignin accompanies the dissolution of hemicellulose components as they are involved in LCC. The LCC fragments undergo secondary reactions in the aqueous phase, forming heavy condensed products. The late-stage delignification requires decomposition of at least a portion of cellulose, which makes the remaining lignin accessible to oxidizing species. Lower operating temperatures can reduce the loss of cellulose, for which decomposition has a higher activation energy than delignification.

3. CONCLUSIONS

This study investigated extensive delignification at low temperatures and its chemical mechanism. Even at 90 °C, O₂ greatly promotes the lignin dissolution while decomposing carbohydrates. Polysaccharides are decomposed into small molecules without forming monosaccharides. The delignification process, in terms of the delignification rate against cellulose degradation, is mostly dominated by the chemical structure of the cedar. However, the delignification rate is greatly influenced by the operating conditions. The chemical condensation of dissolved
lignin fragments, PRL, greatly impedes the delignification. With the renewal of the aqueous phase every 2 h, the undesirable chemical condensation was successfully avoided. The 4 × 2-h repetitive treatments achieved a delignification rate as high as 96%, which was clearly higher than that achieved by a single treatment at 130 °C. Use of a percolator, to which a solution is continuously fed, would be a promising approach for further research and development of low-temperature delignification.

Lignification consists of three stages (early, mid, and late stages). The late-stage delignification (rate >0.65) requires cellulose decomposition for making the lignin accessible to oxidizing agents. The target purity of the product cellulose should be determined in consideration of the unavoidable trade-off relationship between lignin and cellulose retentions.

4. EXPERIMENTAL SECTION

4.1. Materials. Cellulose, monosaccharides (glucose, xylose, arabinose, galactose, mannose), lower organic acids, NaOH, and Na2CO3 were purchased from Wako Pure Chemical Industries. A type of xylan, derived from beech wood, was purchased from SERVA Electrophoresis. Ultrapure water-soluble material. It was subjected to a steam pretreatment at 220 °C under atmospheric pressure. This pretreatment was performed to cause structural relaxation of hemicellulose and thereby promote the delignification. The procedure of the pretreatment is described in detail in the Supporting Information. The compositions of the original and pretreated cedars are listed in Table 3.

Table 3. Compositions of Cedar before and after Pretreatment (Unit; wt %, Dry)

| sample             | mass loss from raw cedar | lignin | cellulose | xylan | galactan | mannan | ash |
|--------------------|--------------------------|--------|-----------|-------|----------|--------|-----|
| original cedar     | 36.6                     | 43.5   | 6.11      | 4.13  | 7.49     | 0.38   |
| steam-treated cedar| 3.29                     | 38.6   | 44.3      | 6.90  | 2.97     | 6.36   | 0.34 |

4.2. Alkaline Treatment with O2. The steam-treated cedar was subjected to oxidation in an autoclave (Taiatsu Techno, TVS-N2, internal volume; 120 mL). In a typical run, 2 ± 0.1 g of the dry steam-treated cedar and 20 ± 1 mL of a 0.8 mol/L NaOH aqueous solution were charged in the autoclave. The air was subjected to oxidation in an autoclave (Taiatsu Techno, TVS-N2, internal volume; 120 mL). In a typical run, 2 mL of a 0.8 mol/L fresh NaOH aqueous solution at 60 °C under vacuum. Then, 2 ± 0.1 g of residual solid collected from several 2-h treatments was subjected to the next 2-h treatment with 20 ± 1 mL of a 0.8 mol/L fresh NaOH aqueous solution.

4.3. Characterization of Products. The workflow of the analyses of gas, liquid, and solid products is shown in Figure 8.

Figure 8. Workflow of product separation, collection, and analysis.

The gaseous product was collected in a gasbag, and analyzed by gas chromatography with an Agilent micro GC 490. The solid and liquid of the slurry were separated by vacuum filtration. The solid was washed with water at 80 °C to remove the residual water-soluble material completely, dried at 60 °C under vacuum, and then subjected to analyses to quantify glucan, xylan, arabinan, galactan, mannan, and lignin, according to a report by the National Renewable Energy Laboratory (NREL).

The content of cellulose was represented by the amount of glucan, assuming that most of the glucan units belonged to the cellulose.

The pH of the liquid (filtrate-1) was measured with a pH meter (D-71; Horiba Ltd., Japan). Consumption of alkali was calculated from the pH value. It was assumed that the remaining NaOH concentration was the same as log10(PH-14) mol/L. Inorganic and organic carbons in filtrate-1 were quantified using a total organic carbon (TOC) analyzer (Shimadzu, TOC-5000A). A portion of filtrate-1 was acidified to pH <2 to precipitate light brown-colored lignin (PRL). PRL was isolated by centrifugation, filtration, washing with water, and vacuum drying at 60 °C. SEC was applied for measurement of the apparent molecular weight (MW) distribution of PRL. The analytical procedure is described in detail in the Supporting Information. Formic, acetic, oxalic, glycolic, lactic, malonic, succinic, and 3-hydroxypropionic acids dissolved in aqueous solution.
glycolic acid, malonic acid, succinic, and 3-hydroxypropionic acid, which had already been determined by the previous HPLC analysis, were subtracted from the carbon-based yield of MEK-S. It was assumed that lactic acid, acetic acid, and formic acid had been evaporated together with MEK. No further analysis was performed for the MEK-IS left in the aqueous phase. It was speculated that the MEK-IS consisted mainly of derivatives of hemicellulose and cellulose.\(^\text{23}\)

The lignin after the treatment was classified into unconverted lignin in the residue (RL), PRL, and NPL in solution. Their yields, on the basis of the mass of lignin in the original cedar, were calculated using the following equations

\[
\begin{align*}
RL &= \frac{\text{mass of lignin in residue}}{\text{mass of lignin in initial feed}}, \text{ PRL} \\
NPL &= \frac{\text{mass of precipitated lignin}}{\text{mass of lignin in initial feed}}, \text{ NPL} = 1 - RL \\
&= PRL, \text{ delignification rate} = 1 - RL
\end{align*}
\]

NPL was believed to consist of low-molecular-mass aromatic compounds; however, yields of lower organic acids and CO\(_2\) originating from lignin were included in that of NPL inevitably.

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