Separation by Molecular Weight during Purification Process of Lignophenol

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Separations of Hinoki cypress (Chamaecyparis obtusa)-lignophenol (\(p\)-cresol type, HCLC), which are phenolic lignin-based polymers derived directly from lignocellulosics through the phase-separation system, by molecular weight using diethylether (EtOEt), cyclopentylmethylether (CPME), tert-Butylmethylether (TBME) and EtOEt / CPME (1 / 1, v / v) in ordinary purification processes were carried out. After purification, each lignophenol showed different purification yields: HCLC-E, HCLC-C, HCLC-T and HCLC-B showed 63.7 %, 50.3 %, 71.0 % and 62.8 %, respectively. These HCLC samples showed different average molecular weights estimated by GPC. To begin with number average molecular weight (\(M_n\)) were 7 870, 9 960, 11 850 and 8 920, respectively. Next, weight average molecular weight (\(M_w\)) were 17 960, 22 510, 24 770 and 20 370, respectively. These HCLC samples showed almost same chemical properties such as appearance, solubility for solvents. Moreover same structural features were confirmed by FT-IR and \(^1\)H-NMR. Since these HCLC samples also showed almost same thermal properties, there are just a little difference for 5 °C on TMA. In conclusion, Molecular weight separation can contribute to not only new advanced chemical modifications for lignophenols, but to investigate new structural features of both lignophenols and whole native lignins in lignocellulosics.

Key words: lignin, lignophenol, molecular weight, solubility, separation

1. INTRODUCTION

Recently biomass has been expected as a substitution for fossil carbon resources in front of both serious environmental crisis and of apprehension for exhaustion of petroleum. Especially lignocellulosic materials have been tried to be applied for various industrial materials because they are sustainable and common raw materials on the earth. In general, lignocellulosics has been utilized as fuel, woody materials, and pulps as papers or fibers. As lignocellulosics consists of cellulose (50%), hemicellulose (15%) and lignin (30 %), utilization of carbohydrate especially pulp has been often used because of its rich contents. On the conventional separation process with high temperature, high pressure and chemicals, sensitive lignin was destroyed and polymerized at random. Because almost all reactive sites of native lignin were exhausted, the resulting lignins have little chemical reactivity. The low reactivity caused almost all these lignins to be used as thermal sources. But lignins are abundant and important aromatic resources with long circulation loop, it is wrong to be burned out to CO\(_2\). Therefore in order to utilize whole lignocellulosics perfectly, it is important to separate components of lignocellulosics to keep the reactive sites of lignin. The phase-separation system is one of the solutions to utilize whole lignocellulosics [1-2]. On the reaction on the surface between phenols and acids, both separation of components and conversion of lignin to functional materials are realized under 1 atm and room temperature within 60 min (Fig.1). Through this reaction both native lignin and carbohydrates were quantitatively converted into 1,1-bis(aryl)propane-2-O-arylether-3-ol type phenolic lignin based polymers (lignophenols) and hydrolyzed sugars. Lignophenols have appropriate properties for chemical applications such as high solubility for solvents, thermal plasticity, sustainable molecular designs, bio-degradability, bio-compatibility and so on [2]. Based on these appropriate chemical and physical analyses of lignophenols, various applications of lignophenols have been tried and realized [1-3].

In purification processes of lignophenols using ethers shown in bottom of Fig.1, both insoluble and soluble
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moieties were separated. EtOEt-insoluble lignophenols have \( M_r \) around 8 000-20 000. While EtOEt soluble moieties have small \( M_r (1 \text{,} 590, M_r/M_n = 1.81) \) by using nano-TiO\(_2\) collection method. [3]. Interestingly these small molecules showed better performance on several applications [3]. These results implied that it is capable to separate fractions of lignophenols by molecular weight using various solvents. Moreover, it is worthy to separate components by molecular size for various advanced applications. But in general, molecular weight separations are carried out by gel-permeation separation, membrane separation or ultra-filtration membrane mainly in bio-chemical, proteins DNA chemistry, medical [4-5] and waste water treatment [6]. Although these methods can cut-off molecular weights exactly, it is hard to treat large amount of materials.

Added this, EtOEt always has high risk of fire or productions of peroxides under aerobic conditions, but EtOEt is the best solvent. In order to overcome these problems, Cyclopentylmethyllether (CPME, Zeon Co.) [7] and other ethers were tried to apply for purification. CPME is known for one of good substitutions for EtOEt or tetrahydrofran (THF) in organic synthesis [8-10]. Properties of CPME were 106 °C of boiling points, 0.809 cP of viscosity, 0.86 gcm\(^{-3}\) density and \( c(25 ^\circ \text{C}) = 8.4 \). As CPME is hard to generate peroxides, it is safe and easy to be recycles and reused, compared to other alkyl ethers [7].

In this study, separations of lignophenol based on solubility through the ordinary purification processes using 4 different ether systems, (A) diethyl ether (EtOEt), (B) cyclopentylmethyllether (CMPE), (C) tert-Buthylmethyllether (TBME) and (D) mixtures of EtOEt and CPME (1 / 1, v / v) in large scales were tried. After purification, comparisons of characteristics of HCLC samples by GPC, \(^1\)H-NMR, FT-IR, TMA and TGA were carried out.

2. EXPERIMENTAL

2.1 The phase-separation system

Synthesis of lignophenol (\( p \)-cresol type) was followed by the phase-separation system (2-step method, process II) [2-3]. Hinoki cypress (\( Chamaecyparis obtusa \)) was used as softwood lignocellulosic material. Milled wood (80 mesh passed, 500 g) was de-fatted by acetone and dried before synthesis. The milled wood was mixed with \( p \)-cresol in acetone. Amount of \( p \)-cresol was 3 mol for \( p \)-cresol type, HCLC) were obtained. Each lignophenol was noted as HCLC-E, HCLC-C, HCLC-T and HCLC-B, respectively.

2.4 Characterization of lignophenols

The structure of HCLC was characterized by Gel Permeation Chromatography (GPC), \(^1\)H-NMR, FT-IR and Thermal Mechanical Analysis (TMA). GPC was carried out by LC-10 system with four columns (KF801, KF802, KF803 and KF804, Shodex Co.). Rectified tetrahydrofran (THF) was used as eluent with 1.0 m\( \text{L}\) min\(^{-1}\) of flow rate at 40 °C. UV-light at 280 nm was used for detection. Both \( M_r \) and \( M_n \) were determined based on standard polystyrene. \(^1\)H-NMR spectrum was also carried out on a FT-IR8400 (Shimazu Co.), using the KBr pellet technique for sample preparation.

3. RESULTS AND DISCUSSION

3.1 The phase-separation system

After the phase-separation system for Hinoki cypress, 97% yield of crude lignophenol based on lignin contents. Using four different solvent systems, purified HCLC showed just a little different characteristics. But almost all the properties were quite same. After purifications, yields of insoluble moieties of HCLC-E, HCLC-C, HCLC-T and HCLC-B were 63.7%, 50.3%, 71.0% and 62.8%, respectively. These all HCLC solids were sufficiently purified by removing un-reacted \( p \)-cresol and small fractions confirmed by GC and TLC. These HCLC solids had light beige appearances and were

\[ M_r = \text{mass of polymer} / \text{mass of monomer} \]

\[ M_n = \text{mass of polymer} / \text{mass of monomer} \]

\[ \frac{M_r}{M_n} \]

\[ \text{Retention Time} / \text{min} \]

Fig.2 Normalized GPC profiles for (A) HCLC-E, (B) HCLC-C, (C) HCLC-T and (D) HCLC-E/C. Dotted lines were fixed on \( t = 30 \) min.
structures such as C5-C5 biphenyl type (10-11%) in native lignins have rich conjugated network type phenol grafting reactions [3]. In general, softwood (benzyl) aryl ether structures by both acid treatments and α major linkages of lignins, after cleavages from Cβ and CβC cell. through endwise polymerization on inner wall in a plant linear type lignin synthesized on the final lignifying step characterized by second wall lignin, which contains rich network type lignin generated on the first growing step in bio-synthesis through bulk polymerization in rich native lignin in intercellular layer, which contains low molecular weight moiety in native lignin. The higher molecular weight was mainly due to native lignin in intercellular layer, which contains rich network type lignin generated on the final lignifying step in bio-synthesis through bulk polymerization in plant cell. On the other hand, the lower one was characterized by second wall lignin, which contains rich linear type lignin synthesized on the final lignifying step through endwise polymerization on inner wall in a plant cell.

HCLC has rich Cβ-O-C4 structures, which are most major linkages of lignins, after cleavages from C6 (benzyl) aryl ether structures by both acid treatments and phenol grafting reactions [3]. In general, softwood native lignins have rich conjugated network type structures such as C5-C5 biphenyl type (10-11%), Cβ-O-C3 phenylcoumaran type (9-12%) and C4-O-C3 diarylether type structures (4%) [11]. These structures remained through the phase-separation system. Therefore due to these two different type molecular structures, GPC patterns were divided.

As compared Fig.2 (A) and (B), HCLC-E included perfectly dissolved in acetone, ethanol, THF, pyridine and 1.0 M NaOH. Moreover the yield of HCLC-B was similar to HCLC-E. This result implied that properties of the solution were much dominated by EtOEt.

3.2 Average Molecular Weight

As shown in Fig.2, molecular weight distributions were different by using different ethers. All HCLC samples showed bimodal peaks with valley around t = 30 min. These bimodal peaks were due to main structures of native lignin. The higher molecular weight was mainly due to native lignin in intercellular layer, which contains rich network type lignin generated on the first growing step in bio-synthesis through bulk polymerization in plant cell. On the other hand, the lower one was characterized by second wall lignin, which contains rich linear type lignin synthesized on the final lignifying step through endwise polymerization on inner wall in a plant cell.

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As compared Fig.2 (A) and (B), HCLC-E included lower molecular weight moieties than HCLC-C. In fact, average molecular weight (Table 1) Mw (Mn) of both HCLC-E and HCLC-C were 17 960 (7 870) and 22 510 (9 960), respectively. This result showed solubility of HCLC for CPME was larger than EtOEt. Moreover HCLC-T showed larger both Mn and Mw (Fig.2, Table 1) with high yield. This result implied that TBME dissolved more linear type molecules selectively.

3.3 Chemical features

As shown in Fig.3, spectra of FT-IR were quite same. This result clearly demonstrated that all HCLC samples had same functional groups such as grafted p-cresol (815 cm-1), ether linkages of lignin units (1 300 - 1 000 cm-1), aromatic skeltones (1 650 - 1 450 cm-1), no carbonyls ( 1 700 cm-1) no aldehydes (2 000 cm-1), rich methylene linkages (2 900 cm-1) and rich hydroxyl groups (3 400 cm-1). The same chemical features can be contributed to various advanced modifications.

As 1H-NMR spectra showed also same chemical features such as methylene groups of grafted p-cresol (2.5-2.0 ppm), methoxyl groups (4-3 ppm), aromatic rings (8-6 ppm) and no carbonyl and carboxyl groups (> 9 ppm) were obtained (Fig.4). For example, HCLC-E and HCLC-C had 1.59 mol/C9 and 1.69 mol/ C9 of p-cresol, respectively. This comparison indicated that low molecular fractions included rich aliphatic-OH groups. Although there were same amounts of grafted p-cresol, total phenolic groups were decreased in HCLC-C. This showed natural phenolic moieties existed in lignophenols tends to be dissolved into CPME. But there were few differences on other parameters between HCLC-E and HCLC-C.

3.4 Thermal Properties

There are some relationships between thermal properties and molecular weights. Thermal behaviors of lignin materials are often measured by TMA using penetration method [12]. All HCLC samples were perfectly plasticized around 170 °C (Fig.5). The Solid-Liquid phase transition temperatures of HCLC-E, HCLC-C, HCLC-T and HCLC-B were 174.2 °C, 178.2 °C, 179.2 °C and 177.0 °C, respectively. The TMA patterns showed that HCLC molecules had almost same thermal properties because all HCLC samples plasticized smoothly within 30 °C with low viscosities.

Although there are large differences of both Mn for 5 000 and Mw for 6 000 among HCLC samples, TMA temperatures showed slight dependences on the molecular weights. This result implied there were strong

<table>
<thead>
<tr>
<th>Solvents</th>
<th>Mn</th>
<th>Mw</th>
<th>Mw/Mn</th>
</tr>
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<tbody>
<tr>
<td>EtOEt</td>
<td>7870</td>
<td>17 960</td>
<td>2.28</td>
</tr>
<tr>
<td>CPME</td>
<td>9860</td>
<td>22 510</td>
<td>2.26</td>
</tr>
<tr>
<td>MTBE</td>
<td>11 850</td>
<td>24 770</td>
<td>2.09</td>
</tr>
<tr>
<td>Blend 1)</td>
<td>9 820</td>
<td>20 370</td>
<td>2.28</td>
</tr>
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1) EtOEt / CPME = 1 / 1 (v. / v.)

THF, pyridine

Table 1 Average molecular weights of HCLC.

![Fig.3 FT-IR spectra of (A) HCLC-E, (B) HCLC-C, (C) HCLC-T and (D) HCLC-E/C.](image_url)

![Fig.4 1H-NMR spectra of (A) HCLC-E and (B) HCLC-C.](image_url)
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and dominant both inter- and intra-molecular interactions such as hydrogen bonds due to both rich phenolic and aliphatic -OH groups.

As shown in Fig.5, all HCLC samples showed good plasticity, but there were little decompositions in HCLC molecules. As Fig.6 demonstrated, HCLC-E, HCLC-C and HCLC-T had only little weight losses under 200 °C. There was also little difference among these three HCLC samples. As described in our previous report [13], weight loss observed under 250 °C was mainly due to release of small fractions such as HCHO or H2O detected using TG-GC/MS. Therefore 5 % weight loss under 250 °C depended on releasing these small fractions or solvents. Over 250 °C same TG curves were obtained to 500 °C for all HCLC samples. Since amounts of remained moieties which were carbon rich materials obtained around 500 °C, all HCLC samples had same amounts (65%). This result indicated that there were same carbon skeletons in polymeric structures after removed small fractions by solvents.

4. PERSPECTIVE FOR APPLICATIONS

Base on this separations, two important perspectives on applications of lignophenol were obtained. First, these separations of contents in these bimodal peaks will contribute to discuss structures of native lignin, because analyses of whole lignin without heat or pressure have never been investigated. Although milled wood lignin is regarded as better sample of native lignin, yield of this lignin is very low. On the other hand, lignophenol is derived directly from native lignin under mild physical condition almost quantitatively. Therefore lignophenol can be applied for the first lignin derivatives to analyze whole lignin in lignocellulosics. Moreover by separation of contents using these solvents, new information about two different types lignin such as lignophenol of bulk polymerization and endwise polymerization can also contribute to investigate the structures of native lignin in cell walls.

Secondly, using EtOEt, CPME and TBME, average molecular weight of HCLC was devied by 2 000 for both Mn and Mw. These exact division can be contributed to various chemical applications.

5. CONCLUSION

Using EtOEt, CPME, TBME and EtOEt / CPME solution for purification of HCLC, components were separated by molecular weight in the ordinary purification process of lignophenols. Not only these ethers, but changing structures of ethers characteristics of lignophenols can be controlled. Though there were few differences in chemical properties, a little difference of thermal properties was observed. As each component of the bimodal peaks in the GPC profiles was separated, new advanced investigates on both lignophenols and native lignin are expected.

6. REFERENCES


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