New types of lignin hybrids from grass- and wood native lignins

Minho Jung and Masamitsu Funaoka
Graduate School of Bioresources, Mie University, Tsu, Japan
Fax: +81-59-231-9521, e-mail: funaoka@bio.mie-u.ac.jp

Through the phase separation treatment with a concentrated acid and a phenol derivative, 80 % of native lignins in rice straw got ether soluble, having high frequency of combined phenols. The successive phase separation treatments of rice straw, followed by western hemlock were carried out. In this process ether soluble fractions from native lignin in rice straw worked as external nucleophiles to native lignin in western hemlock. With increasing the mol ratio of western hemlock- to rice straw lignins, a yield, a weight-average molecular weight (Mn) and a content of methoxyl group of resulting lignin derivatives (acetone soluble, ether insoluble fractions) got higher, having lower and broad absorption at 815 cm⁻¹ in FT-IR spectra, attributed to C-H out of planes skeletal vibration of aromatic ring despite the same contents of combined p-cresol. These results suggested the formation of hybrid types of lignins composed of rice straw- and western hemlock lignins.

Key words: native lignin, phase separation system, lignophenols, lignin hybrid

1. INTRODUCTION

Lignins are the most abundant aromatic polymers in the ecosystem and are important as one of post-petroleum materials. However, it has been very difficult to separate lignin from lignocellulosics without destroying fundamental functions, because of interpenetrating polymer network structures in the cell walls and the sensitivity of lignin for environmental changes. Lignin is biosynthesized from three precursors, coniferyl, sinapyl and p-coumaryl alcohols. The network structure of lignin is constructed through two routes. One is random radical coupling of precursors, giving primary chains. The other one includes the addition of adjacent nucleophiles, H₂O, lignin units and carbohydrates to quinonemethides (C₉ position), giving labile benzyl and 3-dimensional network structures [1-2].

The quick response of lignin for environmental change is due to high reactivity of C₉ positions in the side chain. The phase separation system is a novel multistep functionality control process for native lignin [1-4].

When native lignins (Fig. 1A) contact with acid, benzyl carbonium ions are quickly formed, followed by self-condensation, to form highly condensed lignin with poor reactivity (Fig. 1B). On the other hand, when lignins were solvated with phenol derivatives, followed by acid treatment, benzyl carbonium ions are quickly stabilized by grafting phenol derivatives without self-condensation. This conversion proceeds selectively, and the resulting lignin derivatives (lignophenols) are mainly composed of the primary lignin chains formed by radical coupling during biosynthesis (Fig. 1C).

In our previous works, lignophenols (p-cresol type, ligno-p-cresol) were prepared from western hemlock (Tsuga heterophylla), birch (Betula papyrifera marsh) and rice straw (Oryza sativa) through the phase separation system. The resulting lignophenols reflected the structures of native lignin. 80 % of native lignin in rice straw got ether soluble with high frequency of combined p-cresols. This result indicated that native lignin in rice straw have primary lignin chains of small sizes [6].

This study aims at a design of new type of lignin hybrids from different plant species and a establishment of simple and quick process for the production (Fig. 1D).

2. EXPERIMENTAL

2.1 Conversion of lignin in rice straw

2.1.1 Volume of 72 % sulfuric acid

Rice straw was milled to 80 mesh pass size and was extracted with benzene-ethanol (2/1, v/v) for 48 hrs. Extractives-free rice straw meals were subjected to the phase separation system (2 step process II) as follows : three mol/C₉ (phenyl propane unit of lignin) of p-cresol dissolved in acetone were added to
extractives-free rice straw (1 g). After evaporating acetone, prescribed amount of 72 % H₂SO₄ was added. The mixture was stirred vigorously at 30 °C for 30 min. After dilution to 40 % H₂SO₄, the mixture was added to deionized water. Extractives-free western hemlock (80 mash pass) was added to the mixture. The ether soluble fractions were dispersed into excess amount of deionized water. The ether insoluble and soluble fractions were purified and collected as experimental 2.1.1.

2.2 Preparation of lignin hybrids
Lignin contents of rice straw and western hemlock were 21.1 and 29.4 %, respectively [6]. 1 mol/C₉ of p-cresol dissolved in acetone was added to the extractives-free rice straw (1 g). After evaporating acetone, 3 mL of 72 % H₂SO₄ was added. The mixture was stirred vigorously at 30 °C for 30 min. In order to dilute to 40 % H₂SO₄, the mixture was added to deionized water in ice bath. Extractives-free western hemlock were added to the mixture. The ether soluble fractions (about 80 % of lignin in rice straw) had about 1000 of weight-molecular weight (Mₜ), corresponding to about tetramer [6]. These fractions were assumed as 1 unit of external nucleophile. The mixture was slowly stirred for 1 hour. The mixtures were condensed and then added dropwise to excess -cresol and 10 mL/g of sulfuric acid have been used in standard the phase separation system after the conversion of lignin in rice straw, [2]. However, amount of both p-cresol and sulfuric acid are important for a successive phase separation treatments of rice straw followed by western hemlock. If unreacted p-cresol molecules remain in the reaction system after the conversion of lignin in rice straw, they may affect the properties of the final lignin hybrids.

2.3 Analyses of lignophenols
2.3.1 Gel permeation chromatography
Average molecular weight was estimated by gel permeation chromatography (GPC) using LC-10 system (Shimadzu Co.) with four columns (KF801, KF802, KF803 and KF804, Shodex Co.). H₃PO₄/ tetrahydrofuran solution (0.015 mol/L) was used as eluent with flow rate of 10 mL/min. Both Mₚ and Mₚ were determined based on standard polystyrenes.

Table 1 Yields of ether insoluble and acetone insoluble fractions

<table>
<thead>
<tr>
<th>Sample</th>
<th>Ratio of western hemlock to rice straw</th>
<th>Sample weight (g)</th>
<th>Rice straw (lignin content)</th>
<th>Western hemlock (lignin content)</th>
<th>Yields of ether insoluble fraction</th>
<th>Yields of acetone insoluble fraction</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>(g)</td>
<td>(wt % of dry sample weight)</td>
<td>(wt % of dry sample weight)</td>
<td></td>
</tr>
<tr>
<td>rice straw only</td>
<td></td>
<td></td>
<td>1.1125 (0.2106)</td>
<td>0 (0)</td>
<td>0.0451</td>
<td>0.1237</td>
</tr>
<tr>
<td>western hemlock/ rice straw (0.3)</td>
<td>0.3</td>
<td>1.1490 (0.2175)</td>
<td>0.0845</td>
<td>21.42</td>
<td>0.0586</td>
<td>0.1999</td>
</tr>
<tr>
<td>western hemlock/ rice straw (0.5)</td>
<td>0.5</td>
<td>1.1752 (0.2253)</td>
<td>0.0453</td>
<td>25.36</td>
<td>0.0584</td>
<td>0.2405</td>
</tr>
<tr>
<td>western hemlock/ rice straw (1)</td>
<td>1</td>
<td>1.1481 (0.2173)</td>
<td>0.0541</td>
<td>20.99</td>
<td>0.0543</td>
<td>0.2405</td>
</tr>
<tr>
<td>western hemlock/ rice straw (5)</td>
<td>5</td>
<td>1.2668 (0.2401)</td>
<td>0.0541</td>
<td>19.66</td>
<td>0.0714</td>
<td>0.3642</td>
</tr>
<tr>
<td>western hemlock/ rice straw (10)</td>
<td>10</td>
<td>1.1983 (0.2286)</td>
<td>0.0564</td>
<td>5.58</td>
<td>0.0764</td>
<td>30.39</td>
</tr>
</tbody>
</table>

2.3.2 FT-IR
FT-IR spectra were obtained using KBr pellet technique by IR Spectrum (Shimadzu Co.). Each spectrum was recorded in the range from 4000 cm⁻¹ to 400 cm⁻¹.

2.3.3 Contents of methoxyl group
Methyl iodide formed by the reaction of methoxyl group with hydroiodic acid was led to the vessel filled with molecular sieve 13X. Contents of methoxyl groups were calculated based on an amount of generated methyl iodide absorbed on molecular sieve 13X.

2.3.4 1H-NMR
1H-NMR spectra were measured by JNM-A500 (JEOL Co.) in 600 μL of CDCl₃/C₆D₆N (3:1, v/v). The amount of grafted p-cresol units was calculated based on the signal intensity of cresolic methyl protons (1.6-2.4 ppm) against aromatic protons (7.8-8.4 ppm) of p-nitrobenzaldehyde (internal standard). The hydroxy group contents were determined from phenolic and aliphatic acetoxyl proton signals (1.6-2.4 ppm) on 1H-NMR spectra of acetylated lignin derivatives.

2.3.5 Thermal analysis
Thermo gravimetric analysis (TGA) was carried out using TG-DTA6200 (SII Inc.) from 50 to 400 °C at a rate of 2 °C/min under N₂ flow. Thermo mechanical analysis (TMA) was carried out using TMA-SS6000 (SII Inc.) from 50 to 280 °C at a rate of 2 °C/min under N₂ flow. A tip of needle was set on the center of an Al plate on the flat surface of a lignin derivative in Al pan.

3. RESULTS AND DISCUSSIONS
3.1 Condition for conversion of rice straw
In order to confirm amount of p-cresol and volume of sulfuric acid for the successive phase separation treatments, conversion experiment of lignin in rice straw rice straw were carried out. In general, 3 mol/C₉ of p-cresol and 10 mL/g of sulfuric acid have been used in standard the phase separation system (2 step process II) [2]. However, amount of both p-cresol and sulfuric acid are important for a successive phase separation treatments of rice straw followed by western hemlock. If unreacted p-cresol molecules remain in the reaction system after the conversion of lignin in rice straw, it may affect the properties of the final lignin hybrids.

It was observed that the yield of the ether insoluble fraction increased with the increase of p-cresol concentration. The yield of the ether insoluble fraction was 39.4 % at 0.5% p-cresol concentration and 54.2 % at 3% p-cresol concentration. The yield of the ether insoluble fraction also increased with the increase of sulfuric acid concentration. The yield of the ether insoluble fraction was 42.8 % at 0.5 mL/g sulfuric acid and 50.2 % at 3 mL/g sulfuric acid.
In order to confirm formation of hybrid types of lignins, control experiments were carried out. When only rice straw solvated with 1 mol/C₉ of p-cresol and only western hemlock were used as controls in successive phase separation process, the yields of lignin derivatives (acetone soluble, ether insoluble fractions) were 21.4 and 0 %, respectively. No yield with only western hemlock was attributed to self-condensation of lignin in western hemlock. These results indicated that conversion of only lignin in western hemlock without external nucleophiles had generated no lignin derivatives.

With increasing the mol ratio of lignin in western hemlock to lignin in rice straw, the yields of lignin derivatives were increased. These results showed formation of hybrid types of lignins, because the treatment of only lignin in western hemlock did not give lignin derivatives. However, yields of lignin derivatives of western hemlock/rice straw (5) and western hemlock/rice straw (10) were decreased than yields of lignin derivatives of rice straw only (Table 1). It implied that hybrid formation between lignin derivatives of rice straw and lignin from western hemlock or self-condensation of lignin in western hemlock were increased by decrease of low molecular weight fractions as external nucleophiles. These results showed that amount of lignin from western hemlock for the successive phase separation treatments are less than 3 (C₉/unit).

In order to investigate proportion of native lignin, contents of methoxyl group were measured, because amount of methoxyl group are different.
The contents of methoxyl group of rice straw only and Klasson lignin of western hemlock were 10.45 and 15.45 %, respectively. The contents of methoxyl group are increased, with increasing mol ratio of western hemlock lignin to rice straw lignin (Fig. 3), indicating the formation of lignin hybrids between rice straw and western hemlock. In addition, weight-average molecular weights (Mₐ) of lignin derivatives were larger than that of lignin derivatives from only rice straw (Fig. 3).

The contents of combined p-cresol, phenolic OH and aliphatic OH of lignin derivatives from rice straw only, western hemlock/rice straw (0.3) and western hemlock/rice straw (3) are not so different (Table 2). With increasing the mol ratio of western hemlock lignin to rice straw lignin, the yields of lignin derivatives increased. The treatment of only western hemlock did not give lignin derivatives in successive phase separation process. If the increase in the yields of lignin derivatives is due to containing lignin derivatives from only western hemlock without lignin hybrid, the amounts of combined p-cresol of lignin derivatives should be decreased. These results suggested that the hybrid of lignin existed in lignin derivatives.

The FT-IR spectrum of lignophenol (p-cresol type, ligno-p-cresol) from western hemlock had no absorption around 1700 cm⁻¹, assigned to unconjugated C=O. But the FT-IR spectra of both ligno-p-cresol from rice straw and lignin derivatives from rice straw and western hemlock had absorption around 1700 cm⁻¹, assigned to unconjugated C=O. In addition, with increasing the mol ratio of western hemlock lignin to rice straw lignin, this absorption was decreased (Fig. 4). It implied that the ratio of hybrid type lignin derivatives to ether insoluble fraction obtained from only rice straw was increased. The FT-IR spectra of all lignin derivatives had the absorption around 1700 cm⁻¹, assigned to unconjugated C=O. But the FT-IR spectrum of lignophenol (p-cresol type, ligno-p-cresol) from western hemlock had no absorption around 1700 cm⁻¹, assigned to unconjugated C=O. However, that of western hemlock/rice straw (0.3) and western hemlock/rice straw (3) was more decreased and broad than that of rice straw only, despite similar contents of combined p-cresol which was determined by ¹H-NMR (Table 2). These suggested that the hybrid types of lignin were involved in the acetone soluble, ether insoluble fractions prepared from rice straw and western hemlock.

3.3 Thermal characterization of lignin derivatives

TGA and TMA charts of lignin derivatives were shown in Figs. 5 and 6. Five % weight losses of rice straw only, western hemlock/rice straw (0.3), western hemlock/rice straw (3) and ligno-p-cresol from western hemlock were observed at 191.3, 199.7, 188.2 °C and 189.4 °C, respectively. Ten % weight losses of rice straw only, western hemlock/rice straw (0.3), western hemlock/rice straw (3) and ligno-p-cresol from western hemlock were observed at 238.1, 237.1, 232.7 and 255.3 °C, respectively (Fig. 6). With increasing mol ratio of western hemlock- to rice straw lignins, the phase transition temperature [7] of lignin derivatives were getting lower, suggesting the hybridization of western hemlock- and rice straw lignins.

4. CONCLUSION

Hybridization of grass- and wood native lignins was carried out using successive phase separation process, in which low molecular weight fractions from lignin in rice straw works as external nucleophiles to lignin in western hemlock. The characteristics of the resulting lignin derivatives indicated the formation of hybrid types of western hemlock- and rice straw lignins. The successive phase separation process is a simple and rapid tool for preparing hybrid types of lignin polymers from different plant species.

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6. REFERENCES


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