

# Characterization and Analysis of the Molecular Weight of Lignin for Biorefining Studies

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## ABSTRACT

Cellulose, hemicellulose, and lignin are the three primary constituents of lignocellulosic biomass. While the network polymer has been attributed difficulties in processing cellulose for ethanol, lignin may prove useful in other areas of biorefining. Structural properties of lignin such as its molecular weight holds importance in its future applications in biorefining. Lignin is separated from whole biomass through isolation, pretreatment, or pulping. Following each procedure, changes to the molecular weight of acid soluble or insoluble lignin are often quantified using gel permeation chromatography, or GPC. The data are expressed as number average molecular weight ( $M_n$ ), weight average molecular weight ( $M_w$ ), and polydispersity index (D) and depend on several factors including the severity of processing conditions, the cleavage of intramolecular bonds, genetic modifications, and the presence of recondensation reactions. Depending on its molecular weight properties, lignin can be applied to the production of thermoplastics, carbon fibers, or other materials in biorefining.

## BACKGROUND

Lignin is a biopolymer that is found in plants. It is derived from three monolignol units (p-coumaryl alcohol, coniferyl alcohol, and sinapyl alcohol). The three monolignol units participate in radical polymerization reactions, which are purportedly initiated by an enzyme. The dominant linkage in lignin is the  $\beta$ -O-4 linkage, an ether linkage that is often disrupted during processing as in isolation or dilute acid pretreatment. Lignin typically constitutes between 15 and 40 percent of the woody biomass, but the exact percentage differs by biomass type. The extraction of lignin from biomass is accomplished through a number of routes: isolation, pretreatment, and pulping. The molecular weight (MW) of extracted lignins are commonly analyzed using gel permeation chromatography (GPC), which separates lignins based on size. Isolated lignins such as milled wood lignin are rough representations of the native structure of lignin, even though changes its MW occur. Isolated lignins are commonly used to indicate differences in MW in transgenic plants. Lignins are also recovered from pretreated biomass and pulping. Pretreatments are physical, chemical, or biological means of altering the structure of hemicellulose and/or lignin. Kraft pulping, specifically, removes lignin from wood through the addition of sodium hydroxide and sodium sulfide. Lignins recovered from either pretreatment or kraft effluents are known as technical lignins and can be “upgraded” to more useful applications such as thermoplastics, carbon fibers, and other biomaterials.

## OBJECTIVES

- The purpose of this review is to:
  - Critically evaluate GPC as a method for MW determination
  - Analyze the chemical and structural basis for the modifications to the MW of lignin during isolation, pretreatment, and kraft pulping
  - Identify opportunities for the application of these lignins based on their MWs

## METHODS

The methods listed below describe general procedures for select processing types.

- Isolation method – milled wood lignin (MWL)

Biomass is milled using either a vibratory or rotary ball mill to achieve particle sizes <0.8 mm. Aq. p-dioxane (4% water) is subsequently added to extract the lignin. The dried extracts are dissolved in acetic acid and precipitated with water. The dried precipitate is dissolved in ethylene chloride and ethanol and precipitated in diethyl ether.

- Pretreatment method – ethanol organosolv

Biomass is heated to 180-195°C for 30-90 min in the presence 35-70% ethanol and a mineral acid catalyst. Water is added to the effluent to precipitate the lignin, if desired.

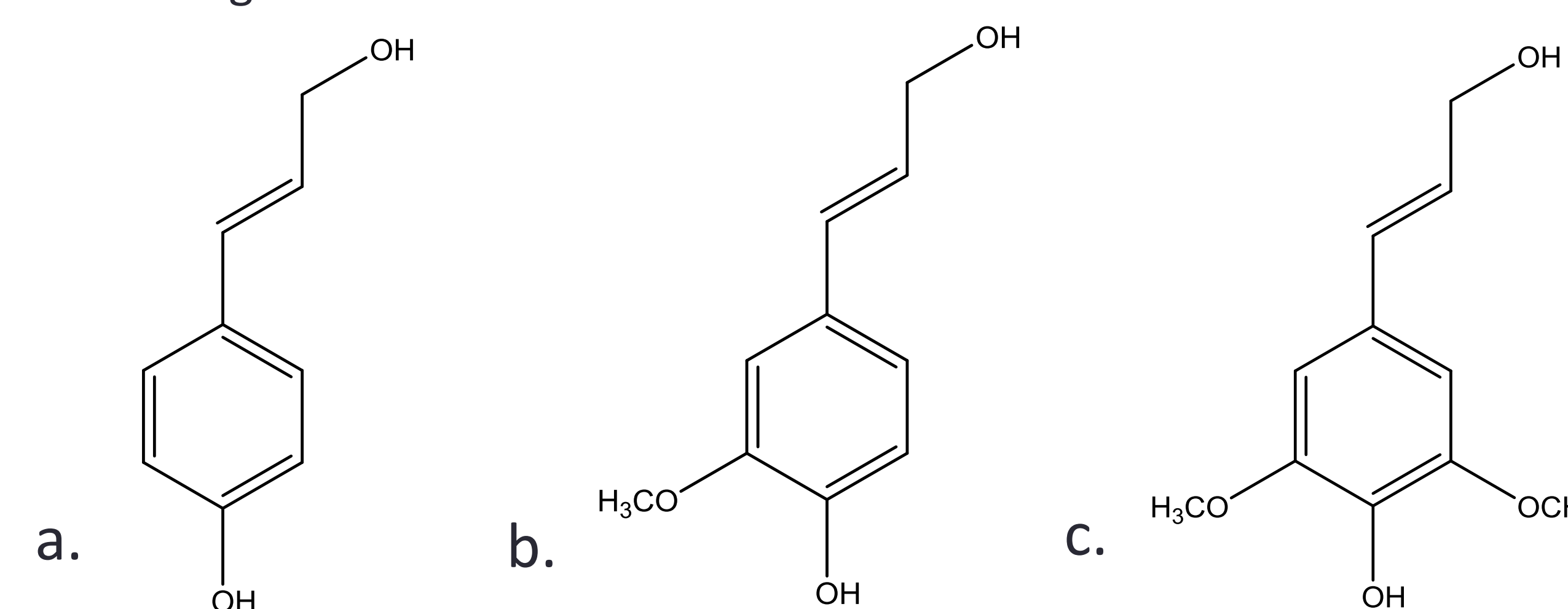
- Pulping method – Kraft pulping

Wood chips are submerged in an aq. solution of sodium hydroxide and sodium sulfide for delignification to take place between 160 and 180°C.

Table 1. Composition of Cellulose, Hemicellulose and Lignin in Various Biomasses<sup>17, 18</sup>

Biomass	Component (% dry weight)		
	Cellulose	Hemicellulose	Lignin
Switchgrass	34	27	17
Monterey pine	42	20	26
<i>Eucalyptus saligna</i>	48	13	27
Corn stover	37	24	18
<i>Miscanthus</i>	37	36	25

<sup>a</sup>Klason lignin



- The images reflect the three monolignol units that undergo radical polymerization to produce lignin (a) para-coumaryl alcohol, or the H monomer in lignin (b) coniferyl alcohol, or the G monomer in lignin and (c) sinapyl alcohol, or the S monomer in lignin.

## RESULTS

- The MW of ball-milled lignins depends on the biomass. Hence, the biomass type may be an important factor in future applications of lignins.
- The MW of ethanol organosolv lignins demonstrate a minimal relationship with the amount of ethanol used. Therefore in future applications of organosolv lignins, ethanol concentration is not a major contributor to changes in MW.
- The MW of hardwood kraft lignins and softwood kraft lignins appear to influence the final  $M_w$ . The molecular weights influenced the solubility of the lignins in methanol.

## RESULTS

Table 2. The Average Molecular Weights and Polydispersity Indices of Milled Wood Lignin from Various Biomasses.<sup>18, 27, 32, 45</sup>

Biomass	$M_w$ (g mol <sup>-1</sup> )	$M_n$ (g mol <sup>-1</sup> )	D
Norway Spruce <sup>a</sup>	23,500	6400	3.7
Douglas Fir <sup>b</sup>	7400	2500	3.0
Redwood <sup>b</sup>	5900	2400	2.5
White Fir <sup>b</sup>	8300	2800	3.0
<i>E. globulus</i> <sup>b</sup>	6700	2600	2.6
Southern Pine <sup>b</sup>	14,900	4700	3.2
Bamboo	12,090	5410	2.23
<i>Miscanthus</i>	8300	13,700	1.65

<sup>a</sup>Vibratory-milled; <sup>b</sup>Ball mill for 28 days

Table 2. The  $M_n$ ,  $M_w$  and D of Ethanol Organosolv Lignins Treated at Variable Ethanol Concentrations

Biomass	Pretreatment	Conditions	$M_n$ (g mol <sup>-1</sup> )	$M_w$ (g mol <sup>-1</sup> )	D
<i>Miscanthus giganteus</i> <sup>122</sup>	Ethanol organosolv	65% ethanol/5% 0.2 M HCl, 4 h, reflux	1520	2720	1.79
		75 % ethanol/5% 0.2 M HCl, 4 h, reflux	1450	2540	1.75
		85% ethanol/5% 0.2 M HCl, 4 h, reflux	1400	2460	1.76
		95% ethanol/5% 0.2 M HCl, 2 h, reflux	1350	2210	1.64
		95% ethanol/5% 0.2 M HCl, 4 h, reflux	1240	2250	1.81
		95% ethanol/5% 0.2 M HCl, 8 h, reflux	1170	2380	2.03

EtOH: ethanol; H<sub>2</sub>SO<sub>4</sub>: sulfuric acid; HCl: hydrochloric acid; NaOH: sodium hydroxide

Table 3. The  $M_n$ ,  $M_w$  and D of Softwood and Hardwood Kraft Lignin

Sample/Isolation	$M_n$ (g mol <sup>-1</sup> )	$M_w$ (g mol <sup>-1</sup> )	D
Softwood kraft lignin Indulin AT <sup>23</sup> /Björkman process	1600	6500	4.1
Softwood kraft lignin Indulin AT <sup>23</sup> /Björkman process	1700	8000	4.7
Hardwood kraft lignin PC-1369 <sup>58</sup> /Björkman process	1000	3300	–
Hardwood kraft lignin PC-1369 <sup>58</sup> /Björkman process	1000	3900	–

## CONCLUSIONS

- The MWs of lignin vary with respect to extraction conditions and biomass types used.
- These observations hold importance when designing the biomaterials whose performance is based upon the molecular weight.