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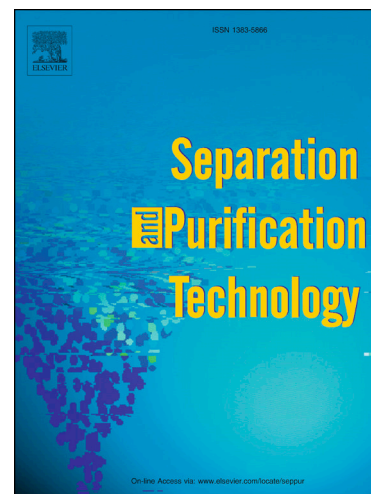
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Solubility and fractionation of Indulin AT kraft lignin in ethanol-water media

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Keywords: lignin, fractionation, solubility, molar mass, phenolic, hydroxyl

Highlights

- **Identified a solubilization maximum at 60 wt% ethanol**
- **The molar mass distribution of the fractions can be adjusted by the solvent's ethanol concentration**
- **The phenolic hydroxyls in lignin can be concentrated by removing the water-soluble fraction**
- **The phenolic hydroxyls in lignin can be concentrated by removing the ethanol-insoluble fraction**

Abstract

Lignin solubility is a varying property, as depending on the type of lignin and its origin, its solubility in different solvents will differ. This is due to the highly heterogeneous nature of lignin. Solubilizing lignin could improve its potential valorization by making it more conducive to chemical reactions. Solvent fractionation could be utilized as the first step in lignin refining to adjust some of its properties before further processing. It is known that kraft lignin is completely soluble in alkaline solvents such as aqueous NaOH. The purpose of this study was to assess the solubility of Indulin AT lignin in aqueous ethanol solvents and to determine the useful properties of the resulting fractions: namely, molar mass distribution and phenolic hydroxyl groups. The highest concentration of lignin was achieved in 60 wt% ethanol solution with 235.89 g/L at a solid-to-liquid ratio of 300 $\text{g}_{\text{Lignin}}/\text{L}_{\text{Solvent}}$. The original lignin had a mass average molar mass of 4.7 kDa. When only water was utilized, the average molar mass of the dissolved fraction was between 1-2 kDa and the molecular size distribution was mostly between 0.1-1 kDa. When using pure ethanol, the molecular size distribution ranged from 0.1 kDa to 10 kDa, with a mass average molar mass between 1-1.3 kDa. With an ethanol content in the solvent higher than 90 wt%, the bigger molar mass molecules (>10kDa) could be separated as the insoluble fraction. Additionally, the polydispersity of both fractions decreased with an ethanol concentration above 80 wt%. The hydroxyl group content of the insoluble fraction was higher when fractionating with less than 20 wt% ethanol, whereas with 20 wt% ethanol and higher, the soluble fraction retained most of the phenolic hydroxyls. Fractionating lignin prior to use or further processing may be potentially beneficial if the lower molar mass molecules can be used as they are, leaving only the higher molar mass molecules to be further processed or burned for energy.

1. Introduction

The solubility of lignin is a varying property. Due to the highly heterogeneous nature of lignin, its solubility in different solvents will differ depending on the lignin type and origin. In simple compounds, solubility can be as straightforward as identifying the polarity or lack thereof of the compounds to assess their solubility (e.g., NaCl in water). In polymers, however, several factors come into play: solubility may be affected by polarity, degree of polymerization, and cross-linking, as well as the pH of the solvent [1]. It is especially challenging to determine solubility in lignin, which is a highly heterogeneous material [2]. It is well known that kraft lignin such as Indulin AT is completely soluble in alkaline solvents, e.g., aqueous NaOH. Indulin AT kraft lignin is also

soluble in DMSO, ethylene glycol, and DMF. It has reduced solubility in acetone, ethanol, and THF, as well as low solubility in water [2-4].

Lignin is composed of polyphenolic molecules with varying degrees of polymerization. These molecules have phenylpropanoid structures with mainly methoxyl and hydroxyl functional groups attached to the aromatic rings, as well as possible primary and secondary alcohols in the propanoid chains. The phenolic hydroxyl (OH_{ph}) can be free (-OH) or etherified (-O-) [5]. The most common etherified bond in lignin is the so-called β -O-4, where β is the second position in the propanoid chain and '4' is the 4th position in the phenyl ring (para) starting from the carbon attached to the propanoid chain [6].

In order to have a more homogeneous material, lignin can be physically fractionated using solvents. By dissolving a portion of lignin and separating it from the bulk, it is likely that more uniform lignin fractions could be obtained. These fractions have the potential of being more homogeneous and having narrower molar mass distributions. Lignin fractionation could be used to adjust some of the properties of lignin, such as molar mass distribution and phenolic hydroxyl content, before further processing. The potential approaches to lignin fractionation for different product streams include solvent extraction, selective precipitation, ultrafiltration, and combinations thereof [7-9].

Chemical reactions benefit from occurring in homogeneous conditions; thus, finding the right area of solubility to work with lignin is essential. Solubility studies have been made on Indulin AT softwood kraft lignin using different solvents. It has been determined by C-NMR that the largest amounts of phenolic hydroxyl groups per unit mass are found in the methanol-soluble fraction and the tetrahydrofuran-insoluble fraction. The lowest average molar masses (M_n and M_w) and polydispersity (PDI) were found in the 2-butanone-soluble fraction [7]. Saito et al. [10] found that the methanol-soluble fraction in softwood kraft lignin had both a lower polydispersity and average molar mass than the insoluble counterpart. However, the polydispersities of both the soluble and insoluble fractions were lower than the PDI of the original lignin.

Organosolv lignin from switchgrass and pine has been sequentially fractionated with aqueous acetone. It was found that the highest solubility was achieved between 60 and 80 % acetone concentration in both pine and switchgrass lignins [11]. Sequential bamboo lignin fractionation was investigated by Li et al. [12] with different organic solvents. They concluded that ethyl acetate extracted a large amount of low molecular weight organosolv lignin, nearly 40 %. A study on the sequential precipitation-extraction of kraft lignin from black liquor found that lower pH values yielded in general lower molar masses [13]. Sequential precipitation-extraction has been carried out by Jääskeläinen et al. [14], starting with an 80% aqueous ethanol concentration and separating the soluble and insoluble lignin fractions by centrifugation, followed by addition of water to the supernatant to obtain a new precipitate. In general, the lowest average molar masses and PDI's were obtained in the last precipitations (when the ethanol concentration was 10%).

Ethanol as a solvent is very practical as it has low toxicity and is relatively low-cost, particularly if it is not needed as absolute-grade ethanol. Ethanol-water mixtures have been utilized in the solubilization of Alcell lignin and it was found that, as the fraction of water or ethanol approached unity, the solubility decreased, with a solubility maximum of about 70 % ethanol [15]. Sameni et al. [2] determined the solubility of Indulin AT in a wide variety of organic solvents (including ethanol) by mixing 100 mg of lignin with 10 mL of solvent. About 6 wt% of the original Indulin AT mass was dissolved in pure ethanol, with a concentration of about 0.6 g/L. However, there is no extensive information on the solubility and fractionation behavior of Indulin AT in aqueous ethanol.

The objective of this work was to assess the solubility of the commercially available, technical lignin Indulin AT in ethanol-water mixtures, as well as determining the useful properties of the resulting lignin fractions, namely average molar mass and phenolic hydroxyl group content.

Materials and methods

2.1. Lignin and solvents

Indulin AT lignin was procured from MeadWestvaco (MWV) with a reported moisture content of 5 wt%, and experimentally determined to contain about 3 wt% of moisture. Absolute ethanol (NORMAPUR) was obtained from VWR Chemicals, and ultrapure water was prepared with a Milli-Q Type 1 purification system.

2.2. Fractionation procedure

Lignin samples were placed in Erlenmeyer flasks with 40 mL of ethanol-water mixtures and magnetic stirrers. The fractionation was carried out overnight at room temperature (about 23 °C). The heterogeneous mixture was then vacuum filtered through 1.2 µm fiberglass filters. The insoluble fractions were dried overnight at 80 °C, cooled at room temperature in desiccators, and subsequently weighed. The soluble fraction was calculated based on the weight of the insoluble fraction.

2.3. Molar mass determination

Just like other polymers, lignin can be represented as a combination of its average molar masses (M_n and M_w) and its polydispersity (PDI), the combination of which can give information about the molar mass distribution of a given lignin sample. The average is used due to the molar mass distribution of a polymer mix, where the molecules can have varying degrees of polymerization. The most common analysis method to determine average molar mass is Gel Permeation Chromatography (GPC), a type of Size Exclusion Chromatography (SEC). However, calibration compounds are normally linear polymers such as polyethylene glycol or polystyrene. These linear compounds vary only in chain length and are best suited for analysis of other linear polymers. In polymer science, the average molar masses are normally expressed in Dalton (Da). However, Da and g/mol units are numerically identical.

To analyze the samples through GPC, the solid samples were dissolved in DMF with 0.05 wt% LiBr to an approximate concentration of 1-2 g/L. The liquid samples were diluted in DMF (0.05 wt% LiBr) as follows. Pure water and 10 wt% ethanol samples were diluted to a 1:1 sample-to-DMF ratio, samples with 20-40 and 80-100 wt% ethanol were diluted to a 1:7 ratio, and samples with 50-70 wt% ethanol were diluted to a 1:15 ratio.

The molar masses were estimated by GPC using an Agilent 1260 series chromatograph equipped with two Phenogel (Phenomenex) 5µm columns (10³ Å and 50 Å) using a UV detector at a wavelength of 280 nm. The mobile phase was composed of DMF with 0.05 wt% LiBr. The flow rate of the mobile phase was 0.5 mL/min and the columns were operated at a temperature of 35 °C. The post-processing of the data was done using polyethylene glycol standards (Agilent EasyVials) of between 10² and 10⁵ Da to estimate molar masses from the retention time data. The polyethylene glycol standards were analyzed using an RID detector, since they did not show up in the UV detector. Additionally, the following formulae were utilized:

$$M_n = \frac{\sum AU_i}{\sum AU_i/M_i} \quad [1]$$

$$M_w = \frac{\sum AU_i M_i}{\sum AU_i} \quad [2]$$

$$PDI = \frac{M_w}{M_n} \quad [3]$$

where M_n is the number average molar mass, M_w is the mass average molar mass, AU_i is the value of absorption units at a corresponding molar mass M_i for a specific data point i , and PDI is the polydispersity of the sample.

2.4. Phenolic hydroxyl group content

The phenolic hydroxyl group content was determined in the soluble and insoluble fractions by a modified method based on the previously reported $\Delta\epsilon$ -IDUS method [16]. In brief, each soluble and insoluble fraction was diluted with the appropriate buffers (pH 6, 12, and 13.3) and immediately analyzed. The difference-spectra were obtained with a Shimadzu double-beam spectrophotometer and the phenolic hydroxyl groups calculated from the difference absorbance values at 300 and 360 nm. The original Indulin AT was also analyzed to serve as a reference. The phenolic hydroxyl group content in the insoluble and soluble fractions was normalized to the measured value of the original Indulin AT (Eq. 4-6):

$$OH_{ph}^{total (calculated)} = x_{soluble} \cdot OH_{ph}^{soluble (measured)} + x_{insoluble} \cdot OH_{ph}^{insoluble (measured)} \quad [4]$$

$$P = \frac{OH_{ph}^{total (measured)}}{OH_{ph}^{total (calculated)}} \quad [5]$$

$$OH_{ph}^{fraction (normalized)} = P \cdot OH_{ph}^{fraction (measured)} \quad [6]$$

where x is the mass fraction of the soluble or insoluble fractions from the original lignin. The mass balance is shown in Eq. 4. The difference between the measured value of total OH_{ph} in Indulin and the calculated value from the fractions can be normalized with a multiplier P (Eq. 5). The multiplier is calculated from the ratio of the measured and calculated OH_{ph} . Thus, the normalized value of OH_{ph} in the soluble or insoluble fraction is adjusted with the multiplier (Eq. 6). The values of P in this study were in average 1.014 ± 0.287 with a C.I. of 95%.

3. Results and discussion

3.1. Lignin solubility in ethanol-water

The solubilization results of lignin in the ethanol-water mixtures are depicted in Fig. 1. The calculation of the soluble fractions was based on the weight of the insoluble fractions. Several solid-to-liquid ratios (S/L) in grams of lignin per liters of solvent ($g_{Lignin}/L_{Solvent}$) were tested. It was found that the maximum solubility was achieved at around 60 wt% of ethanol in the aqueous solvent. This showed the synergistic effect of water and ethanol when dissolving Indulin AT. In other words, at 60 wt% ethanol, the solubility of lignin in the solvent was higher than the solubility in pure water or pure ethanol solvent. The numerical values of the solubility measurements are shown in supplementary material A1.

The relative amount of soluble lignin (percentage of the original lignin) was larger at a lower S/L (Fig. 1.a). At higher S/L values, the relative soluble lignin was lower at almost every ethanol concentration. With water alone, the relative amount of soluble lignin was similar in all cases, as seen from Fig. 2.a. This is probably due to the Indulin AT lignin having a limited water-soluble fraction, which would not easily saturate pure water at the tested S/L values.

The absolute amount of soluble lignin can be seen in Fig. 1.b. The highest concentration of lignin (235.89 g/L) was achieved with S/L 300 $\text{g}_{\text{Lignin}}/\text{L}_{\text{Solvent}}$ and 60 wt% of ethanol. This indicates that the 60 wt% ethanol solvent has a superior capacity to solubilize Indulin AT. However, Fig. 1.a shows that the solvent could be at the point of near-saturation since less of the lignin is being solubilized as the S/L increases. This is exemplified by Fig. 2.b, where the concentration values tend to equilibrium but appear to have not yet reached that point at the tested S/L values.

In pure ethanol, the maximum lignin concentration achieved was 42.0 g/L, and in pure water 26.11 g/L at S/L = 300 $\text{g}_{\text{Lignin}}/\text{L}_{\text{Solvent}}$. Fig. 2.b suggests that the maximum concentration of lignin at 60 wt% ethanol is near-saturation but could still increase with an increased S/L. However, in Fig. 2.a, the relative amount (percentage) of lignin dissolved with 60 wt% ethanol at S/L = 300 $\text{g}_{\text{Lignin}}/\text{L}_{\text{Solvent}}$ has noticeably decreased, which suggests that some of the lignin molecules soluble at this ethanol concentration might have reached saturation, while other different lignin molecules could still be dissolved if the S/L increased. Unlike linear polymers, lignin molecular structure is three-dimensional, highly cross-linked, and heterogeneous. Thus, the physical properties of a group of similar molecules in lignin can make it act as a mixture of materials with similar characteristics rather than a truly homogeneous compound.

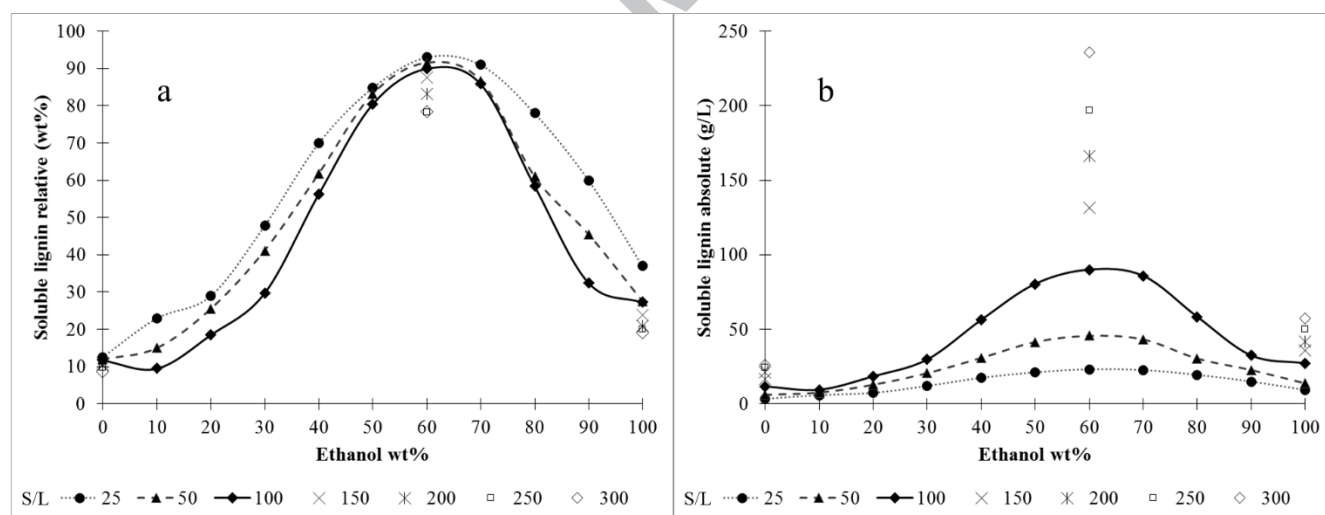


Fig. 1. Soluble lignin at different solid-to-liquid ratios (S/L [=] $\text{g}_{\text{Lignin}}/\text{L}_{\text{solvent}}$): a. percentage of original lignin, b. absolute concentration in solution.

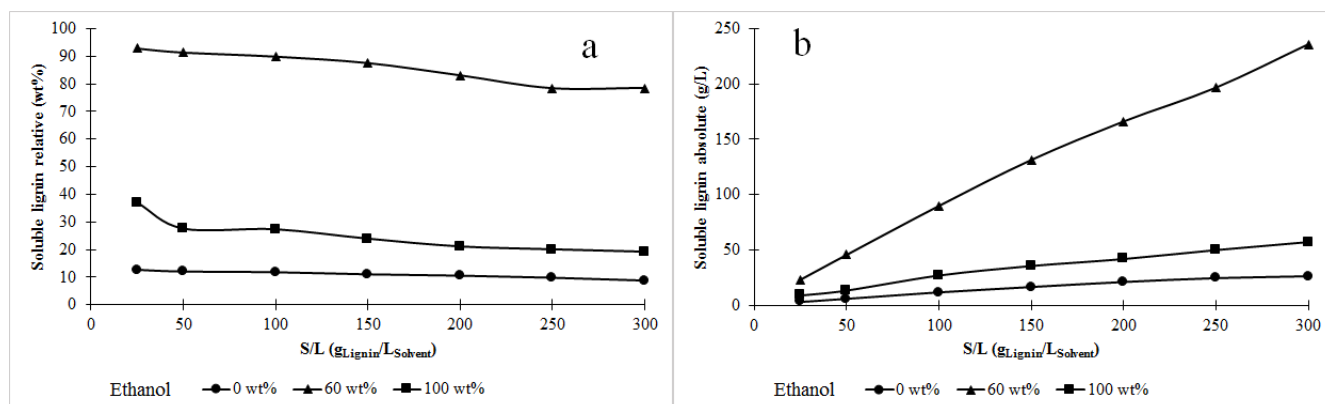


Fig. 2. Soluble lignin as a function of S/L (g_{Lignin}/L_{Solvent}) for three ethanol concentrations a. relative (wt%), b. absolute (g/L)

3.2. Molecular size

The original Indulin AT lignin used in this study had average molar mass values of $M_n = 1336$ and $M_w = 4721$. The reference value of M_w is represented by the dashed lines in Fig. 3.

The soluble fractions start from a low average molar mass at low ethanol concentrations and increase up to a maximum at 60 wt% ethanol, where the values are near-tangential to the values of the original lignin. Considering that most of the lignin is soluble at 60 wt% ethanol, the soluble fraction would likely have similar properties to the original lignin. In the soluble fraction, the pure water solvent retained mostly the lower molar mass molecules (below 1 kDa). At the highest solubility point (60 wt% ethanol), the distribution is practically the same as that of the original Indulin AT lignin (Fig. 4.a). Pure ethanol solvent did not dissolve the higher molar mass molecules (above 10 kDa). Starting at 80 wt% ethanol, the proportion of molecules with molar mass above 10 kDa decreased considerably up to 100 wt% ethanol, where there were practically no molecules above 10 kDa. Numerical values can be found in supplementary material A2; graphical molar mass distributions are depicted in supplementary material A3.

The insoluble fractions exhibited larger average molar masses at all ethanol concentrations and S/L values, compensating for the lower molar masses from the soluble fractions. The average molar masses of the insoluble fraction can be divided into three sections: from 0-50 wt% ethanol, 60-70 wt% ethanol, and 80-100 wt% ethanol. At $S/L = 25$ g_{Lignin}/L_{Solvent}, the proportion of higher molar mass compounds (>10 kDa) in the insoluble fraction increased with increasing ethanol concentration up to 50 wt% ethanol. At 60 wt% ethanol (the maximum solubility point), a large amount of heavy molecules had been dissolved, causing a decrease in the average molar mass in the insoluble fraction. At $S/L = 50$ g_{Lignin}/L_{Solvent}, there was a large increase in the molar mass at 60 wt% ethanol when compared to $S/L = 25$ g_{Lignin}/L_{Solvent}. This is probably due to the limited solubility of the higher molar mass molecules. As the S/L increases, the soluble fraction may become saturated with the larger less-soluble molecules (above 10 kDa), which would cause the average molar mass in the insoluble fraction to increase. In other words, medium and small molecules might still dissolve at higher S/L when using 60 wt% ethanol while larger molecules might not dissolve any further, thus making the average molar mass higher.

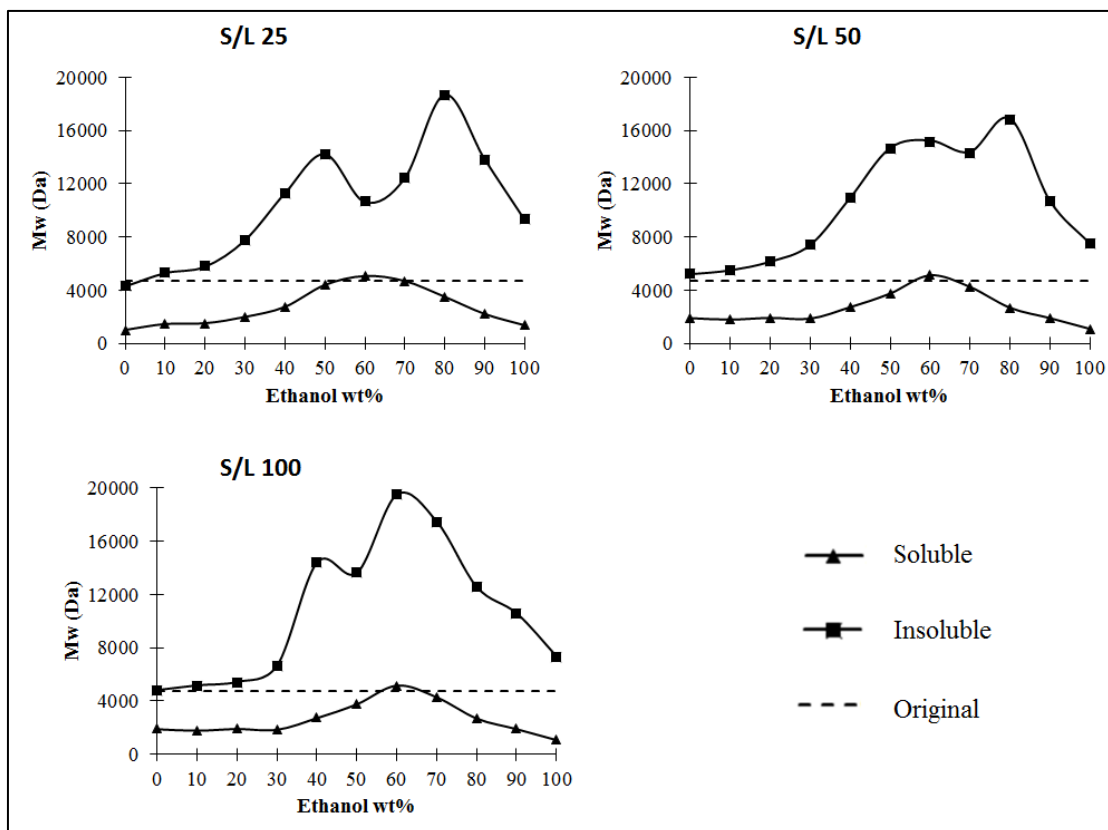


Fig. 3. Mass average molar mass (M_w) of lignin fractions. Dashed line denotes original Indulin AT

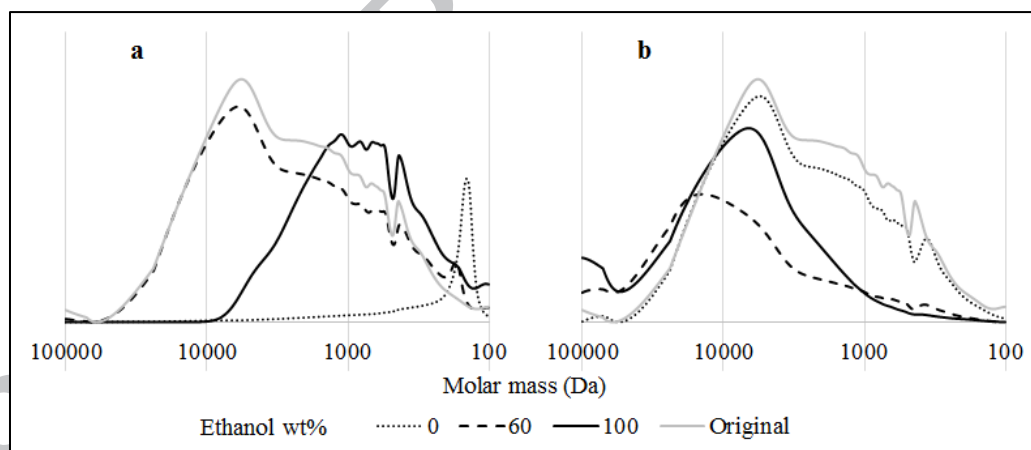


Fig. 4. Molar mass distribution of lignin: a. soluble fraction, b. insoluble fraction. Dashed lines denote the original Indulin AT. Curves not to scale relative to concentration. $S/L = 25 \text{ g}_{\text{Lignin}}/L_{\text{Solvent}}$

3.3. Polydispersity

Polydispersity (simply the quotient of M_w and M_n) is a measure of how widely distributed the molar mass of a polymer is. A lower PDI means a more homogeneous material, i.e., a narrower molecular size distribution. The original Indulin AT lignin had a PDI of 3.54 (represented by the dashed line in Fig. 5).

At 20 wt% ethanol, the PDIs of both soluble and insoluble fractions were close to the original. The best candidates for low polydispersity in soluble and insoluble fractions simultaneously appeared to be at over 80 wt% ethanol (Fig. 5). Starting at 80 wt% ethanol, the PDIs of both soluble and insoluble fractions decreased and reached a minimum at pure ethanol.

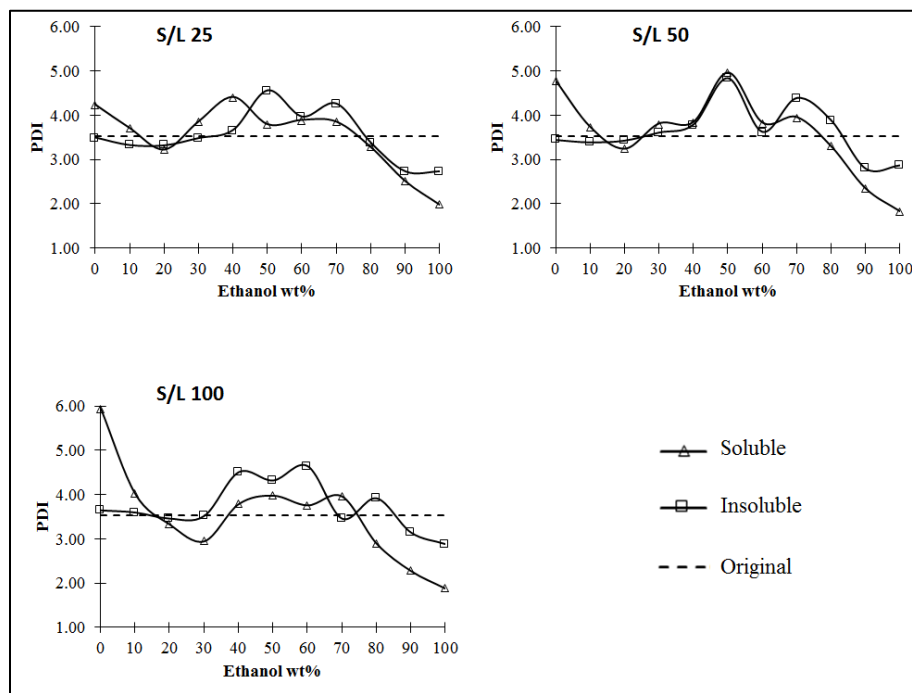


Fig. 5. Polydispersity of lignin fractions. Dashed line denotes the original Indulin AT

3.4. Phenolic hydroxyl group content

The phenolic hydroxyl group content for the original Indulin AT lignin was determined to be 2.07 mmol/g (dashed line in Fig. 6). For $S/L = 25 \text{ g}_{\text{Lignin}}/L_{\text{Solvent}}$, it can be seen that at under 20 wt% ethanol, the soluble fraction retained the molecules with lower OH_{ph} content, even though they are the smaller molecules. However, starting at 20 wt% ethanol, the behavior is reversed, where the majority of the phenolic hydroxyls remain in the soluble fraction. The highest amount for the phenolic hydroxyl group content appears to be in the soluble fraction after 80 wt% ethanol (2.79 mmol/g at 100 wt%). At the highest solubility point (60 wt% ethanol), the soluble fraction had nearly the same amount of OH_{ph} as the original lignin, as the solubility was near complete. Numerical values can be found in supplementary material A4.

The OH_{ph} content in the insoluble fraction was higher than that of the original lignin up to 10 wt%, after which it dropped considerably until reaching a minimum at 60 wt% ethanol. The steep decrease in OH_{ph} content can be explained by large insoluble molecules with a low amount of phenolic hydroxyl groups that are insoluble at 60 wt% ethanol. This behavior mirrors the behavior of the case with pure water, where the soluble fraction retains small soluble molecules with a low amount of OH_{ph} , and the insoluble fraction at that point has similar OH_{ph} content as that of the original Indulin AT.

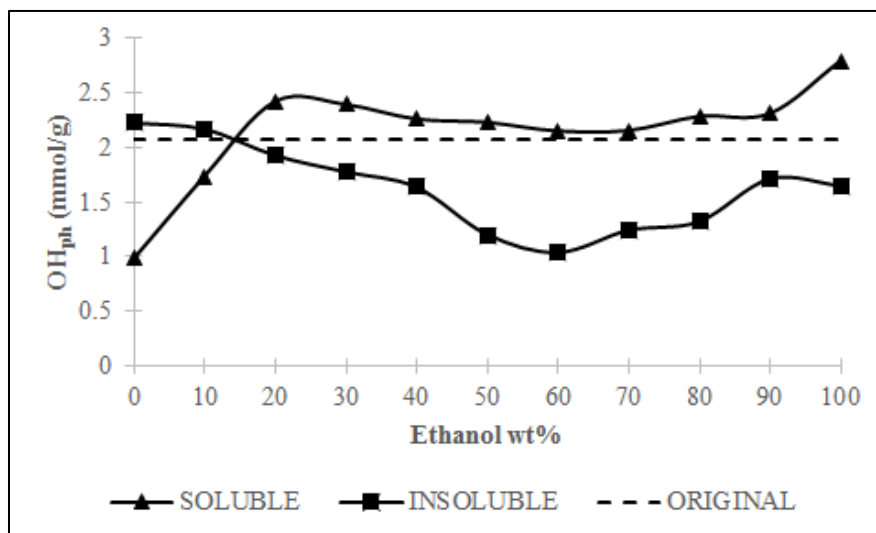


Fig. 6. Phenolic hydroxyl groups at $S/L = 25 \text{ g}_{\text{Lignin}}/L_{\text{Solvent}}$. Dashed line denotes the original Indulin AT

4. Conclusions

The fractionation of lignin using ethanol-water solvents was investigated. It was found that the maximum solubility was achieved at 60 wt% ethanol. The lowest average molar masses were achieved in the soluble fraction when either pure water or absolute ethanol was utilized. An ethanol content above 80 wt% in the aqueous solvent was beneficial with respect to PDI, where the two resulting fractions had a higher homogeneity (lower PDI) than the original Indulin AT. The hydroxyl group content increased in the soluble fraction above 20 wt% ethanol with the highest amounts being reached above 80 wt% ethanol. In the insoluble fraction, there was an increase in the OH_{ph} when using pure water or when using less than 20 wt% ethanol. From a technical and economic point of view, an ethanol concentration that is close to but below the ethanol-water azeotrope could be advantageous for fractionation, as it has been seen that, even at 90% ethanol, not only does a large amount of the lignin dissolve, but also both the soluble and insoluble fractions are considerably more homogeneous. Additionally, at 90 wt% ethanol, the soluble fraction retains most of the lower molar mass molecules and the phenolic hydroxyl groups. Moreover, simple distillation would be sufficient to recover the solvent. Lignin fractionation could be utilized as the first step to optimize lignin properties before further processing and valorization: the lower molar mass fractions could be utilized as they are (e.g., in resoles), while the heaviest fractions could be either depolymerized into smaller molecules or burned for energy. A high solubility (60 wt% ethanol) could be utilized in lignin depolymerization, followed by separation of lower molar mass fractions using a highly selective (90 wt% ethanol) solvent.

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Appendices

A1. Solubility data

Table 1. Soluble fraction data at different S/L

S/L(g _{Lignin} /L _{Solvent})	25		50		100		
	EtOH wt%	wt%	g/L	wt%	g/L	wt%	g/L
0		12.6	3.15	12.0	6.00	11.8	11.75
10		23.0	5.75	15.0	7.50	9.5	9.50
20		29.0	7.25	25.5	12.75	18.5	18.50
30		47.8	11.96	41.2	20.58	29.7	29.75
40		69.9	17.48	61.7	30.87	56.3	56.25
50		84.8	21.20	83.1	41.57	80.4	80.42
60		93.0	23.25	91.5	45.75	90.0	90.00
70		91.0	22.75	86.8	43.38	85.9	85.89
80		78.0	19.50	61.0	30.50	58.5	58.50
90		60.0	15.00	45.5	22.75	32.5	32.50
100		37.0	9.25	27.5	13.75	27.3	27.25

Table 2. Soluble fraction data at different ethanol concentrations

S/L(g _{Lignin} /L _{Solvent})	0 wt%		60 wt%		100 wt%		
	Ethanol	wt%	g/L	wt%	g/L	wt%	g/L
25		12.6	3.15	93.0	23.25	37.0	9.25
50		12.0	6.00	91.5	45.75	27.5	13.75
100		11.8	11.75	90.0	90.00	27.3	27.25
150		11.0	16.50	87.6	131.47	23.8	35.75
200		10.5	21.00	83.1	166.20	21.0	42.00
250		9.8	24.44	78.3	196.77	19.9	50.02
300		8.7	26.11	78.5	235.89	19.1	57.18

A2. Average molar mass data (Indulin AT: $M_n = 1336$, $M_w = 4721$, $PDI = 3.54$)

Table 3. Average molar mass at different S/L. SOLUBLE fraction

S/L ($\frac{g_{Lignin}}{L_{Solvent}}$) EtOH wt%	25			50			100		
	Mn	Mw	PDI	Mn	Mw	PDI	Mn	Mw	PDI
0	236	1002	4.25	398	1909	4.79	394	2340	5.94
10	390	1451	3.72	480	1797	3.74	402	1615	4.02
20	466	1507	3.23	594	1928	3.25	507	1688	3.33
30	512	1984	3.87	490	1872	3.82	756	2230	2.95
40	619	2735	4.42	710	2738	3.86	1263	4787	3.79
50	736	3808	5.17	757	3765	4.97	937	3729	3.98
60	1295	5053	3.90	1339	5143	3.84	1249	4696	3.76
70	1212	4680	3.86	1076	4264	3.96	1055	4179	3.96
80	1065	3497	3.28	817	2696	3.30	978	2837	2.90
90	881	2215	2.52	816	1911	2.34	795	1814	2.28
100	686	1360	1.98	602	1100	1.83	624	1177	1.89

Table 4. Average molar mass at different S/L. INSOLUBLE fraction

S/L ($\frac{g_{Lignin}}{L_{Solvent}}$) EtOH wt%	25			50			100		
	Mn	Mw	PDI	Mn	Mw	PDI	Mn	Mw	PDI
0	1237	4323	3.49	1519	5235	3.45	1319	4796	3.64
10	1602	5325	3.32	1630	5528	3.39	1435	5150	3.59
20	1749	5803	3.32	1801	6178	3.43	1569	5415	3.45
30	2239	7796	3.48	2056	7429	3.61	1897	6661	3.51
40	3087	11321	3.67	2891	10969	3.79	3205	14419	4.50
50	3128	14269	4.56	3030	14685	4.85	3163	13638	4.31
60	2687	10668	3.97	4203	15256	3.63	4214	19531	4.64
70	2918	12435	4.26	3275	14398	4.40	5063	17473	3.45
80	5549	18733	3.38	4345	16897	3.89	3212	12555	3.91
90	5094	13904	2.73	3839	10736	2.80	3367	10567	3.14
100	3429	9358	2.73	2624	7535	2.87	2546	7323	2.88

Table 5. Average molar mass at different ethanol concentrations. SOLUBLE fraction

Ethanol	0 wt%			60 wt%			100 wt%		
S/L (g _{Lignin} /L _{Solvent})	Mn	Mw	PDI	Mn	Mw	PDI	Mn	Mw	PDI
25	236	1002	4.25	1295	5053	3.90	686	1360	1.98
50	398	1909	4.79	1339	5143	3.84	602	1100	1.83
100	394	2340	5.94	1249	4696	3.76	624	1177	1.89
150	381	1717	4.51	930	4257	4.58	583	998	1.71
200	362	1515	4.18	928	3910	4.21	588	1011	1.72
250	285	1194	4.19	1132	4366	3.86	577	1006	1.74
300	285	1163	4.08	1193	4639	3.89	578	1024	1.77

Table 6. Table 6. Average molar mass at different ethanol concentrations. INSOLUBLE fraction

Ethanol	0 wt%			60 wt%			100 wt%		
S/L (g _{Lignin} /L _{Solvent})	Mn	Mw	PDI	Mn	Mw	PDI	Mn	Mw	PDI
25	1237	4323	3.49	2687	10668	3.97	3429	9358	2.73
50	1519	5235	3.45	4203	15256	3.63	2624	7535	2.87
100	1319	4796	3.64	4214	19531	4.64	2546	7323	2.88
150	1535	5309	3.46	2009	11287	5.62	2294	6947	3.03
200	1548	5345	3.45	1616	9935	6.15	2268	6993	3.08
250	1433	4945	3.45	2182	10220	4.68	2202	8429	3.83
300	1382	4922	3.56	1780	7883	4.43	2216	8824	3.98

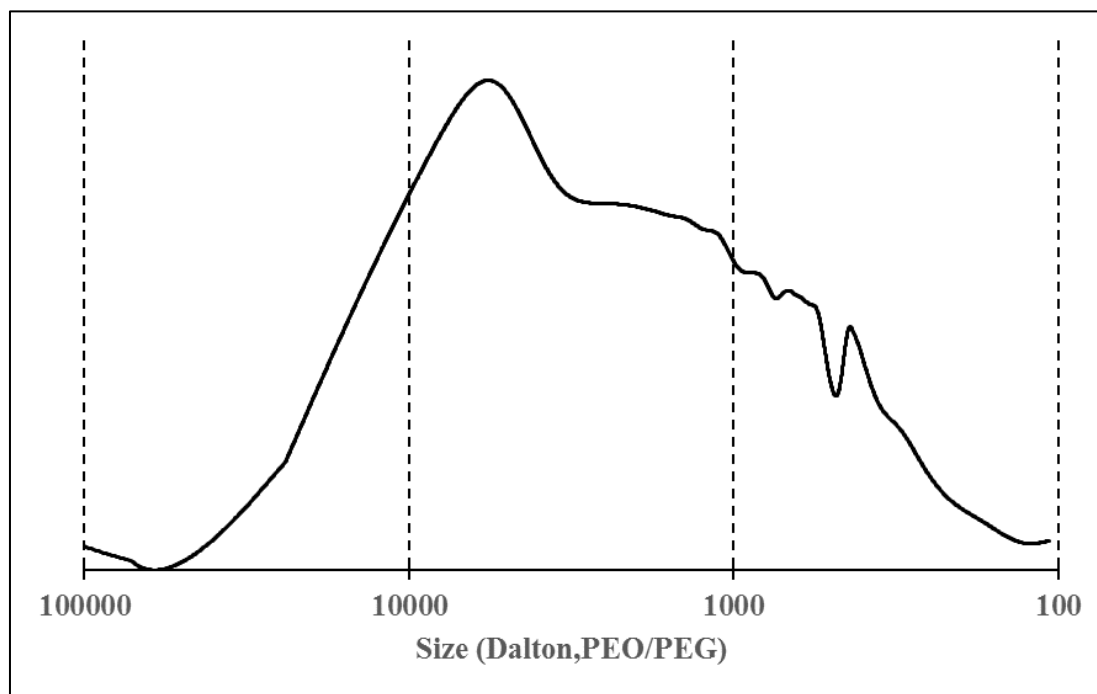
A3. Molecular size distribution

Fig. 7. Molecular size distribution for Indulin AT lignin

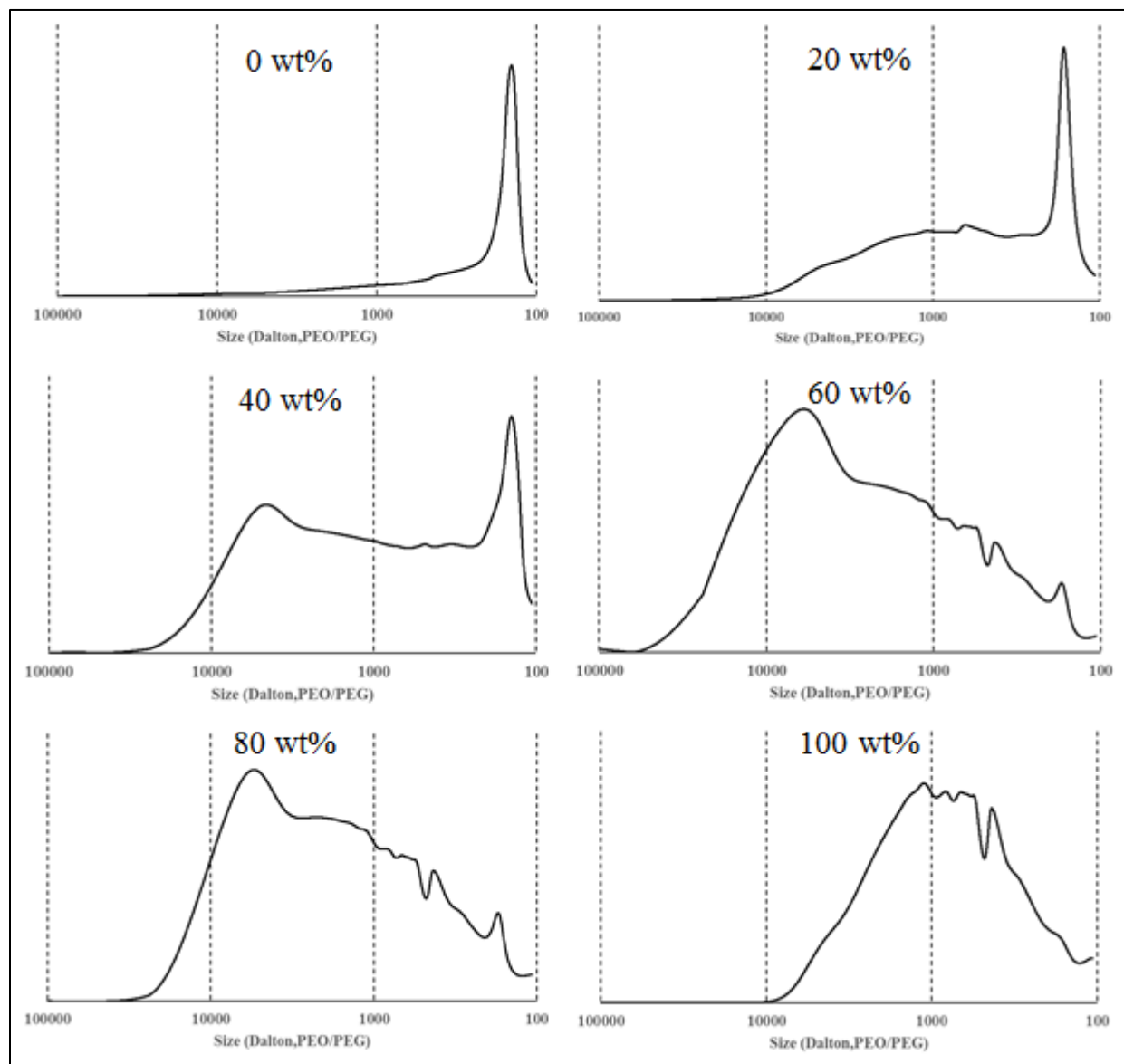


Fig. 8. Molecular size distribution for SOLUBLE fractions at different ethanol concentrations ($S/L = 25$ $\text{g}_{\text{Lignin}}/\text{L}_{\text{Solvent}}$)

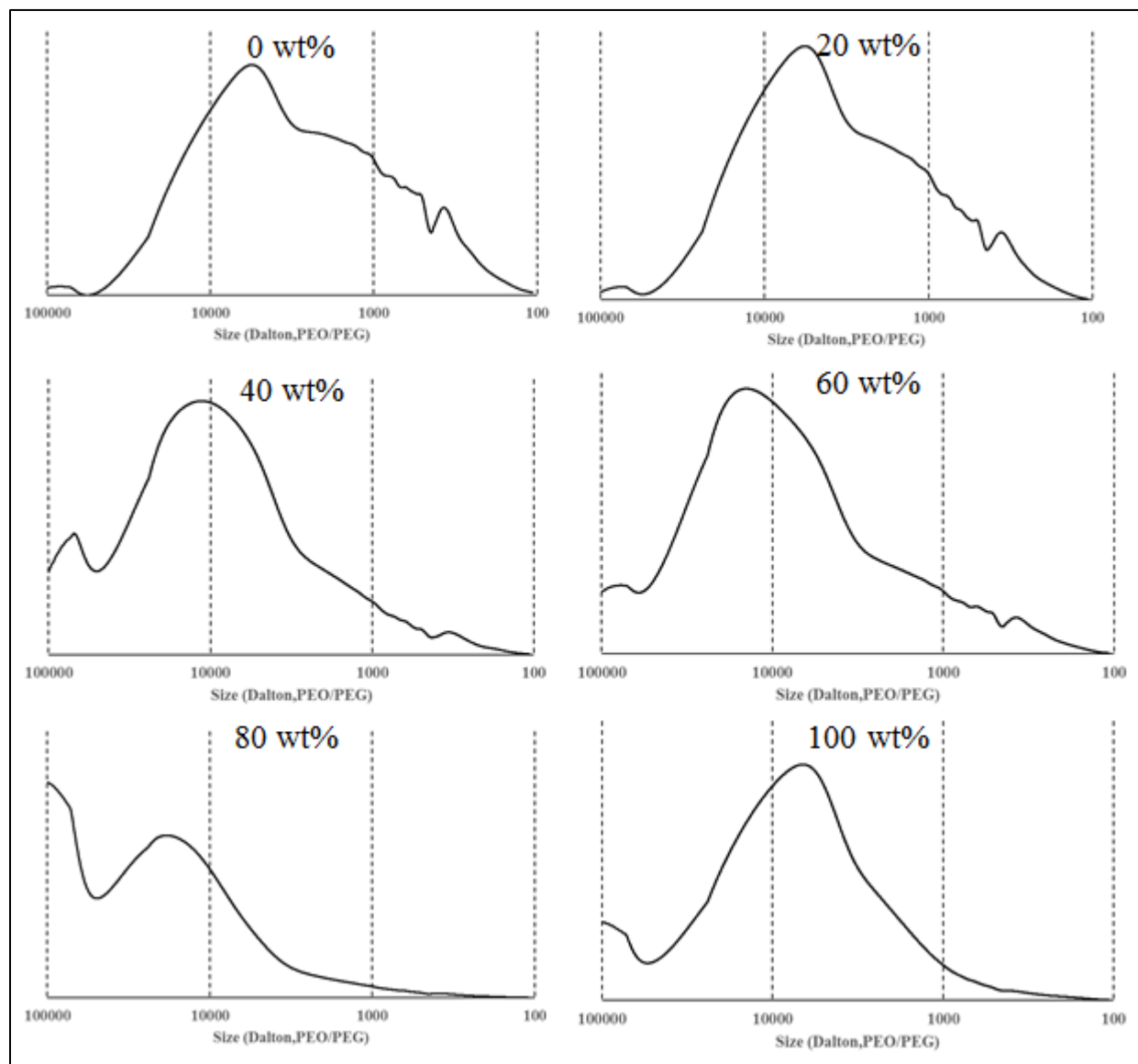


Fig. 9. Molecular size distribution for INSOLUBLE fractions at different ethanol concentrations ($S/L = 25$ gLignin/LSolvent)

A4. Phenolic hydroxyl group data

The original Indulin AT lignin had a total amount of $\text{OH}_{\text{ph}} = 2.07$ mmol/g, of which 1.64 mmol/g were uncondensed and 0.43 mmol/g were C5-substituted.

Table 7. Phenolic hydroxyl group content for S/L = 25 (mmol/g)

EtOH%	SOLUBLE			INSOLUBLE		
	TotOH	C5-substituted	Uncondensed	TotOH	C5-substituted	Uncondensed
0	0.98	0.02	0.99	2.23	0.50	1.73
10	1.74	0.15	1.60	2.17	0.52	1.65
20	2.42	0.10	2.24	1.93	0.57	1.39
30	2.39	0.18	2.10	1.78	0.67	1.21
40	2.26	0.27	1.86	1.64	0.82	1.12
50	2.23	0.46	1.77	1.20	0.33	0.88
60	2.15	0.43	1.71	1.03	0.49	0.67
70	2.16	0.38	1.72	1.24	1.00	0.74
80	2.29	0.43	1.83	1.32	0.45	0.94
90	2.31	0.41	1.89	1.71	0.47	1.26
100	2.79	0.45	2.30	1.65	0.43	1.24