

Commercial Scale Quantification of Lignin

Amudhavalli Victor¹, Indra Neel Pulidindi², Suresh Selvaraj³, Aharon Gedanken^{4,*}

Abstract

Lignin, an aromatic biopolymer, in the terrestrial lignocellulosic biomass constitutes the single largest and sustainable source of biofuels and biochemicals. The recalcitrant nature of lignin is a hindrance to its chemical or biochemical conversion. Moreover, its insolubility in water too is a major challenge in its quantification which is a necessary step prior to and after its conversion. Though there are celebrated quantification methods like the classical Klason lignin method and the acetyl bromide solubilisation (ABSL) method they are not environmentally friendly as harmful chemicals are used in these processes. State of the art analytical techniques based on NMR, HPLC and GC-MS are also being developed for the quantification of lignin owing to the strategic significance of its detection and estimation. Addressing these issues, herein we report a simple and environmentally friendly method for the quantification of lignin (alkali, low sulfonate content) in a two-step process using water as the solvent for the solubilisation of lignin, which is indeed a breakthrough. Clear aqueous solutions of lignin were obtained by high-speed stirring using an ultra-turrax. The aqueous solutions of lignin showed characteristic absorbance at 306 nm. A linear relationship between the amount of lignin and the absorbance at 306 nm is observed that formed the basis of this novel analytical method for the quantification of lignin.

Keywords: Lignin, Ultra-turrax, Sonication, Quantification, UV-Vis spectroscopy, 3-phenyl-2-propen-1-ol

INTRODUCTION

Several patents are being filed related to the processes for the conversion of lignin and also the applications of the derived products. Paper and pulp industries generate huge amount of lignin whose potential remained un-utilized. The challenge is that the decomposition of lignin is very difficult. Huge

amounts of solid residues are often generated during the processes involving the conversion of lignin. Very few studies were devoted to the utilization of the lignin fraction in the biomass owing to the difficulty in the conversion or degradation of lignin. Lignin is an aromatic biopolymer. The chemical structure of lignin indicate that it could be a rich source of several fuels and fine chemicals if it could be depolymerized (broken to small molecules) selectively [1–8].

There is an ancient saying that “we can make anything but not money out of lignin.” It is because of the difficulty or the problem in breaking the rigid aromatic biopolymer, namely, lignin, into low molecular weight compounds of commercial significance. With the increase in knowledge and technological advancement, it is hypothesized that we can make money too from lignin apart from clean fuels and chemicals. Deriving fuels and chemicals

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has double advantage of sustainability and clean environment. Simply stating, success in breaking down lignin selectively to desired target products, leads to making money and alleviates the problem of climate change. As stated above, lignin depolymerization is a challenge that could be turned into an opportunity. Success in this attempt leads to commercially significant products for essential needs (Figure 1) and also result in clean environment by reducing dependence on fossil fuels (Figure 2).



Figure 1. We can make money too from lignin apart from products for essential needs biofuels and biochemical.

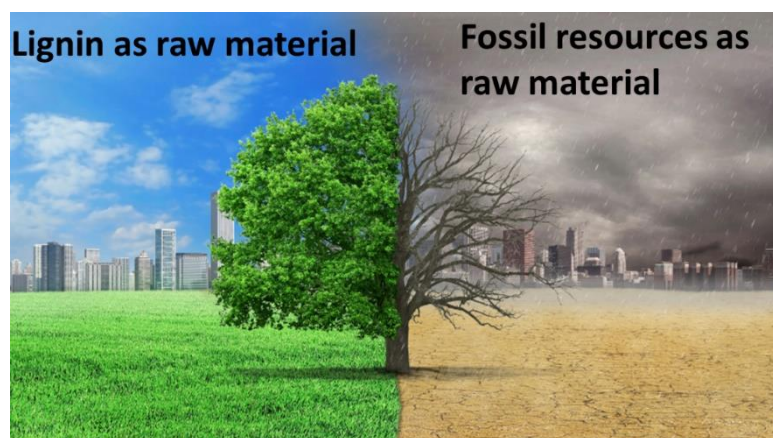


Figure 2. Use of lignin instead of fossil resources for biochemicals and biofuels production leads to clean environment.

Lignocellulosic biomass is currently investigated vigorously as a potential feedstock for biofuels, fine and bulk biochemical [1]. Much of the attention is focused on the holocellulose (cellulose + hemicellulose) fraction as this fraction is a rich source of C6 and C5 sugars which could be converted to bioethanol, a potential transportation fuel [2]. The C6 and C5 sugars could also be converted to various chemicals like HMF, levulinic acid, formic acid, furfural, lactic acid and several others [3–9]. Very few studies were devoted to the utilization of the lignin fraction in the biomass owing to the difficulty in the conversion or degradation of lignin [4, 10]. Depending on the type of biomass, the lignin content varies from 10-35 % which is a significant amount [11]. Typical structure of lignocellulosic biomass and the location of lignin is shown in Figure 3 [9].

The structure of lignin, an aromatic biopolymer, comprises of three main building blocks, namely, *p*-coumaryl alcohol, coniferyl alcohol and sinapyl alcohol. Thus, lignin comprises of the major and single largest renewable source of aromatic hydrocarbons on the surface of the earth [5, 6]. The chemical structure of lignin indicate that it could be a sustainable source of several fuels and fine chemicals if it could be depolymerized (broken to small molecules) selectively. Producing biochemicals like phenols, methanol, formic acid, methyl formate, as well as benzene, toluene and xylene (BTX) from lignin is of strategic significance [12–18].

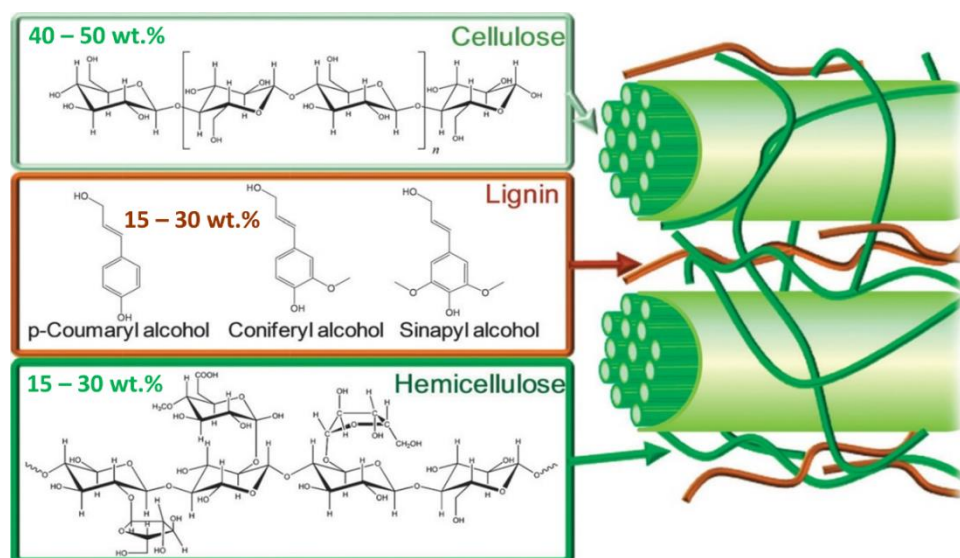


Figure 3. Schematic representation of the distribution of lignin in biomass (cellulose (microfibril), hemicelluloses and lignin) and the location of individual components [Reproduced with permission of the Royal Society of Chemistry from reference 9].

In spite of the vast untapped potential of lignin as a reserve of biofuels and biochemicals, one of the major bottlenecks is its solubilisation in water as well as its quantification. A web of science search crossing the keywords, namely, lignin and quantification, showed 854 results indicating that the problem of lignin determination is underestimated and not yet fully explored [19–28]. A variety of classical spectroscopic (FT-IR, UV-Vis, and NMR and chromatographic) (GC-FID, GC-MS, HPLC, size exclusion chromatography) techniques as well as advanced versions of the classical techniques [like ^1H - ^{13}C hetero-nuclear single quantum coherence zero (HSQC₀) NMR, direct infusion electron spray ionization tandem mass spectrometry (DI-ESI-MS/MS) were exploited for the quantification of lignin [19–28]. Though TGA was tested, its use for quantification of lignin is not recommended owing to larger discrepancies compared to the classical methods, namely, the Klason lignin method and the acetyl bromide soluble lignin method (ABSL). In the Klason lignin method the residue after carbohydrate solubilization in 72 % H_2SO_4 is estimated and is regarded as a measure of the amount of lignin in a given analyte. Likewise, in the ABSL method the UV absorbance at 280 nm from lignin phenols after complete derivation by solubilisation of acetyl bromide is regarded as a measure of the lignin amount [19].

In the words of Jeremy S Lutherbacher, the precise quantification of the structural features of native lignin is an unmet challenge that is dampening the valorization of lignin [21]. The present research article precisely addresses the challenge raised by Jeremy S Lutherbacher by devising an innovative methodology based on mechanical agitation using ultra-turrax for the solubilisation of lignin in water and subsequently using UV-Vis spectroscopic technique for the quantification of lignin by taking advantage of the unique absorption feature of aq. lignin at 306 nm. Such a report is first of its kind in chemical literature.

EXPERIMENTAL

Lignin (alkali low sulfonate content, CAS 8068-05-1) was procured from Sigma Aldrich Co., (St. Louis, MO, USA) and was used as received. Homogenized aqueous solutions of lignin were prepared using the high-speed stirrer ultra-turrax device (Leroy Somer, Digidrive SK, make ESCO-LABOR). Clear aqueous solutions of lignin were obtained by stirring 1 wt. % lignin suspension in distilled water for 3 h, at a speed of 3000 rpm. UV-Vis spectra of the aqueous solutions of lignin were recorded using a Cary 100 scan Varian UV-Vis spectrophotometer.

RESULTS AND DISCUSSION

High-speed stirring of lignin (1 g) in distilled water (100 mL) using an ultra-turrax resulted in the formation of homogenized and clear aqueous solutions of lignin (1 wt.%). The dissolution of lignin in water was found to be stable for more than three months at room temperature. Thus, a major challenge of solubilizing the recalcitrant lignin is surmounted. In literature, either concentrated acids or organic solvents were used for the solubilization of lignin which are environmentally unfriendly and expensive. UV-Visible spectrophotometry has been identified to be a potential tool for the quantification of lignin in aqueous medium.

Solutions with varying amounts of lignin were obtained by subsequent dilution method, from the afore mentioned stock solution. The concentrations of the solutions were so adjusted that their absorbance values fall in the range of 0-1 (0.13 – 1.21 mg lignin/mL water) in accordance with the Beer-Lambert's law for the quantification of chemical species using UV-Visible spectroscopy (Figure 4). The absorption band observed with a λ_{\max} of 306 nm is characteristic of lignin and its intensity is a measure of the amount of lignin in a given analyte. The calibration plot deduced from the UV-Vis absorption spectra with varying amounts of lignin (0.13 to 1.35 mg/mL) was shown in Figure 5. The limit of detection (LoD) of lignin using the current method is 0.1 ppm.

Thus, homogenization of lignin in water using ultra-turrax and its subsequent quantification using UV-Vis spectrophotometry constitutes a new method for the estimation of lignin. The current methodology is superior to the UV-Vis spectroscopic method of quantification of lignin reported by Jean Michel Lavoie and co-workers in a way that the preparation of aqueous solution of lignin doesn't require the energy intensive single shot steam process that may cause degradation of the biopolymer lignin itself [29]. Molecular level details on the origin of the UV-Vis absorption band at 306 nm are awaited. However, we surmise that the "2-propen-1-ol, 3-phenyl" fragment, common to all the three fundamental building blocks of the lignin structure, namely, *p*-coumaryl alcohol, coniferyl alcohol and sinapyl alcohol, is the origin of the absorption band at 306 nm in the UV-Vis spectrum of the aqueous solutions of lignin.

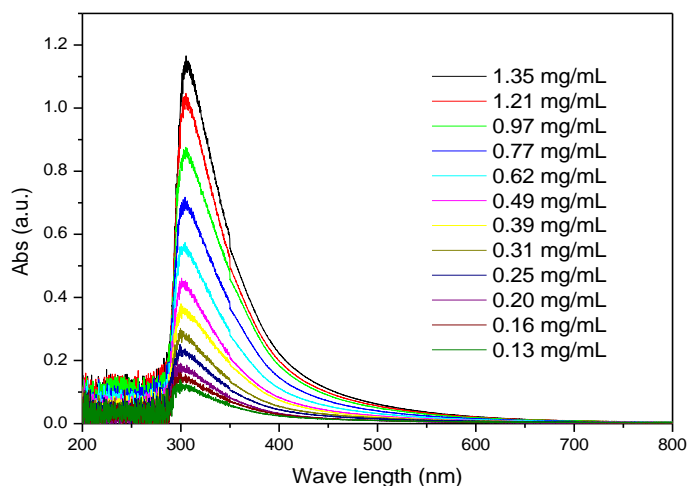


Figure 4. Variation of absorbance ($\lambda_{\max} = 306$ nm) as a function of amount of lignin.

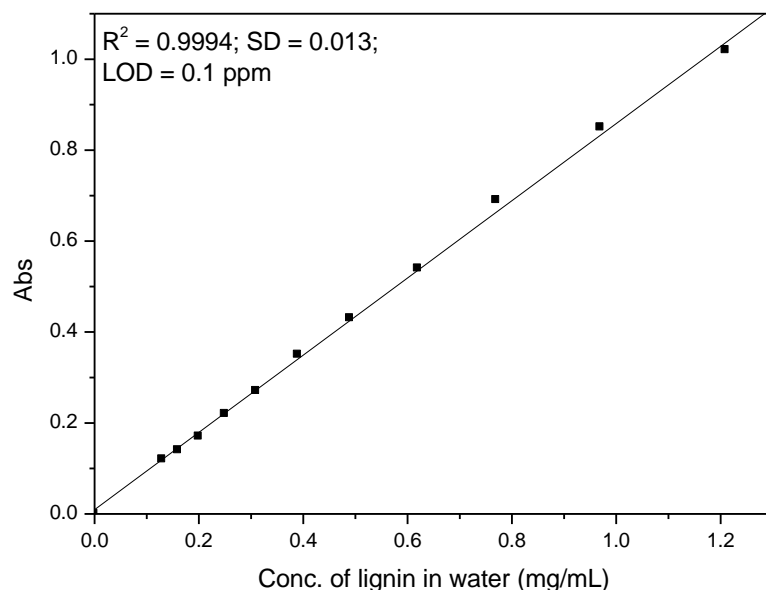


Figure 5. Calibration plot for the estimation of lignin amount in a given analyte.

CONCLUSION

Lignin was solubilized in water using high-speed stirring with ultra-turrax. The aqueous solution of lignin showed unique absorbance peak at 306 nm that showed a linear dependence with the amount of lignin. This formed the basis for the quantification of lignin in the aqueous medium. As the developed methodology uses no sophisticated analytical devices but for a high-speed stirrer and the classical UV-Vis spectrometer, day to day basis in R and D establishments working on the development of processes for the conversion of lignin to biofuels and biochemicals.

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