Effect of Temperature on Lignin Isolation by Using Organosolv Method from Oil Palm Empty Fruit Bunch

A Pramana¹, Y Zalfiatri¹, E O Sari²

¹Department of Agricultural Industrial Technology, Faculty of Agriculture, Universitas Riau, Indonesia
²Department of Chemical Engineering, Faculty of Engineering, Universitas Riau, Indonesia
Jl. HR. Soebrantas, Km. 12.5, Pekanbaru, 28293, Indonesia
esty@lecturer.unri.ac.id

Abstract. Lignin is the second most abundant biopolymer on the earth as promising raw material for various valuable products. In terms of biorefinery, the organosolv method promotes elevate satisfy isolation due to its ease of purification and environmental friendliness. This study aimed to determine the effect of temperature on lignin isolation from palm oil empty fruit bunch (EFB) obtained through the organosolv method. The isolation was using acetic acid, formic acid, and water (30:60:10( v/v/v)) as the solvent and 0.1% of HCl as a catalyst at various temperatures (60°C, 85°C, 100°C, and 121°C). The heating temperature had significant effect on the yield, purity and pH of lignin. The lignin yield rises when the temperature increase from 60°C to 85°C but starts to decrease at 100°C. The highest yield of 15.87% was obtained at 85°C. However, the purity is inversely proportional to the yield. The higher the yield, the lower the purity correspond to condensation reaction at temperature above 85°C. The highest purity of 94.49% was obtained at the lowest yield at temperature 121°C. The heating temperature also affected pH. The pH is increase with increasing temperature. Functional groups analysis using FT-IR indicated that lignin isolates obtained at a temperature of 60°C, 85°C, 100°C, and 121°C had similar functional groups, consist of ether linkage, aromatic rings, C-H methyl linkage, guaiacyl rings, and syringyl rings indicated that the lignin of oil palm empty fruit bunch classified as SGH-type lignin.

1. Introduction
Palm oil empty fruit bunch (EFB) is the major lignocellulosic by-product of the palm oil industry. The processing of 1 tons of oil palm fresh fruit bunch would give 0.22 tons of EFB, 0.12 tons of fiber, and 0.05 tons of shell, and up to 2.5 – 3.75 tons of palm oil mill effluent (POME) [1,2]. In 2019, Indonesia produced 48.4 billion tons of crude palm oil which is estimated to correspond to 53 billion tons EFB [3]. EFB contains 43-43.47% cellulose, 22.93-23.67% hemicellulose, and 21.28-22.10% lignin which is very potential as a renewable raw material for valuable products [4]. Cellulose has been utilized in many fields such as pulp and paper, nano cellulose, biofuel, and high value add chemicals such as ethanol, acids, saccharides, phenols xylitol, and cellulose acetate [5-7]. Whereas, nowadays there has been a shift from perceiving lignin as a waste to viewing lignin as a promising material for valuable
products. Lignin valorization has received interest in broad applications such as polymers, adhesives, medical and electrochemical energy materials sectors [8,9].

There have been several methods for lignin isolation, namely chemical process, and enzymatic process. The chemical process is the most favorable considering the high cost and long period of enzymatic isolation. Moreover, lignin obtained from the enzymatic process still contains carbohydrates which is couldn’t remove by the common purification method [9]. Kraft pulping is the major chemical process, approximately 85% of the total lignin production in the world (as a by-product that is dilute in black liquor) [10]. However, most Kraft lignin is combusted for heat generation, resulting in low-value utilization [8]. Lignosulfonates, isolated lignins from the sulfate process contain significant amounts of sulfur in the form of sulfonate groups. The sulphur compound still could give a bad impact on the environment. The organosolv method is one of the eco-friendly alternative methods since its process is conducted in the absence of sulfur. The organosolv method is the extraction of biomass components through treatment using an organic solvent, such as ethanol, formic acid, acetone, tetrahydrofuran, ethylene glycol, etc. Furthermore, considering its ease of black liquor recycling and higher purity of lignin this method offers significant opportunities for lignin valorization more than Kraft and sulfite pulping.

Several factors affected the dissolution of lignin, cellulose, and hemicellulose such as temperature, pressure, and solvent composition. Sun et al. (2005) had been isolated lignin and cellulose from straw using the organosolv method with various composition solvents namely acetic acid-formic acid-water with HCl 0.1 % as a catalyst at 85°C [11]. The highest yield was obtained from solvent composition: formic acid-acetic acid-water (30:60:10 v/v/v). Based on this composition there is still a factor that needs to study to confirm the optimized extraction process. In this paper, the effect of temperature on the yield and composition of organosolv lignin from EFB was determined to explore the optimum extraction temperature.

2. Methodology

The main material was oil palm empty fruit bunch (EFB). The solvent was organic acids of formic acid and acetic acid, distilled water, 0.1%, and HCl as the catalyst. All chemical reagents were pro analyst grade which was purchased from Merck and used with no further purification.

Instruments used in this research were Willey mill, 50 mm sieve, deep fryer, furnace set at 400-600°C, porcelain kars, analytical scale, desiccators, and rotary evaporator. The functional group analysis was conducted by the fourier transform-infrared (FT-IR) (Shimadzu).

2.1 Preparation of Oil Palm Empty Bunch Fiber powder

EFB was torn into fiber and sun-dried for a week. Dry fibers were cut into approximately ± 30 mm length, milled using Willey mill, then sieved using a 50 mesh sieve for uniform powder.

2.2 Lignin Isolation from Oil Palm Empty Bunch Fiber

Isolation was conducted according to Sun et al. (2005). Oil Palm Empty Bunch Fiber was heated in a deep frying instrument to obtain black liquor. Delignification was conducted using 10 gram EFB powder with formic acid-acetic acid-water = 30/60/10 (v/v/v), HCl 0.1% as catalyst with liquor to solid ratio 20:1 (ml/g) for 4 hours heating for each temperature (60, 85, 100, 121°C). After heating, filtrate and residue were separated using a filter cloth. The filtrate was then concentrated and the solvent was removed using a rotary vacuum evaporator to obtain lignin isolate then dried in an oven at 80°C for 6 hours before yield, color, and purity measurement.

2.3 Lignin Analysis

The amount of lignin was conducted by Klasson method, the purity of lignin was obtained from the sum of acid-soluble lignin (ASL) and acid-insoluble lignin (AIL). The yield of lignin was conducted by gravimetric method, the lignin isolate was dried in an oven at 80°C for 6 hours. The yield of lignin
was determined based on a comparison of the total oven-dried solid of black liquor toward an oven-dried mass of EFB.

FT-IR spectra were obtained on fourier transform infra-red spectrophotometer (Shimadzu) using a 0.05 mm thickness of KBr disc containing 1% finely ground isolate of lignin. Then the spectra were measured at a wavenumber of 4000-40 cm⁻¹.

All data obtained then analyzed using analysis of variance (ANOVA) at 95% significance level. Comparison among the means to measure significant difference was analyzed using the Duncan’s multiple range analysis.

### 3. Results and Discussion

In this study, lignin was isolated from black liquor obtained after organosolv treatment at various temperatures. The yield was calculated as the percentage of solid black liquor compare to the oven-dry mass of the sample. The lignin yield and purity at various temperatures are depicted in Table 1 and Figure 1.

#### Table 1. The yield and purity of lignin obtained at various heating temperatures.

| Temperature (°C) | Yield (%) | Purity (%) |
|------------------|-----------|------------|
| 60               | 15        | 93.69      |
| 85               | 15.87     | 90.81      |
| 100              | 15.21     | 91.51      |
| 121              | 14        | 94.49      |

![Figure 1](image-url)  
**Figure 1.** Lignin yield and purity at various organosolv heating temperature.

It is shown that the highest yield of 15.87% was obtained at 85°C, and over the 85°C the yields were decreased with the increase of temperature to 121°C. But the purity of lignin is inversely proportional to the yield. The higher the yield, the lower the purity. Based on Figure 1, the highest purity of 94.49% was obtained at the lowest yield at temperature 121°C. And vice versa, the lowest purity of 90.81% was obtained at the highest yield at a temperature of 85°C. Meanwhile, the purity of lignin obtained in this experiment was 90.81-94.49%, which has higher purity compared to another method. In the previous study, it has been reported that the organosolv method produces high purity lignin approximately up to 93% [12,13]. Whereas, another method such as alkaline lignins were had higher carbohydrate content (up to 30 wt%) [12].
It is can be notified that heating temperature affects the delignification process. A higher temperature heating process was able to promote lignin dissolution from EFB. However, too high temperature-induced degradation of lignin constituent compound thus reduced isolated yield [14,15]. Lu et al. (2017) reported that the extraction conditions affect the structure of organosolv lignin, that is, severity factor (H-factor) which is the function of time and temperature. The molecular weight of organosolv lignin decreased within the 36-56% range with the increase of the severity. Moreover, the lignin diminishing in higher temperatures indicates the condensation of lignin in the pulp. Condensation during pulping occurs to a greater extent with formic acid than with acetic acid. This happens was due to the use of organic acid higher concentration and tend to encourage the re-lignin polymerization reaction that has been dissolved in the cooking liquid, so that lignin settles on the surface of the pulp then the color becomes darker [12,16]. This is evidenced by the darker pulp color obtained at temperatures higher than 85°C as depicted in Figure 2.

![Figure 2](image)

**Figure 2.** Organosolv pulp obtained at (a) 60°C, (b) 80°C, (c) 100°C and (d) 121°C.

Decreased purity of lignin isolate was presumably caused by non-lignin component degraded and dissolved in black liquor. The low content of lignin indicated that a high amount of non-lignin components were involved. This was caused by the degradation of non-lignin compounds together with polysaccharide separation.

Hydrogen ion concentration plays a very important role in the delignification process. This is because lignin dissolution is expected to be preceded by the acid-catalyzed cleavage of α-aryl and β-aryl ether linkages in the lignin macromolecule, and becomes soluble in the pulping liquor [17]. To break the β-ether bonding it is necessary more aggressive acid conditions, this can explain the reason why when the acid concentration is raised the lignin yield is raised [18-20]. The formic acid and acetic acid can be significate of lignin because it is acidic, so it can degrade lignin well. The amount of Hydrogen ion indicated by pH was affected by temperature. The pH of lignin at the various temperature shown in Tabel 2.
Tabel 2. The pH of lignin obtained at various temperatures.

| Temperature (°C) | pH    |
|-----------------|-------|
| 60              | 6,7   |
| 85              | 8,6   |
| 100             | 10,9  |
| 121             | 11,5  |

Based on Tabel 2, a higher temperature applied to the optimum level increased pH. At a high concentration of hydrogen ion (low pH), more phenolic groups were protonated. This protonation induces decreased electrostatic repulsion among lignin molecules, resulted in less hydrophilic and reduced solubility of lignin molecules [21]. The increase of pH with increasing temperature reveals the occurrence of cleavage of α- and β-aryl bonds at a higher temperature which produce phenolic OH groups [22]. Moreover, higher heating temperature also promotes lignin degradation and condensation which produce hydroxyl ion which makes higher pH.

FT-IR analysis was carried out to identify functional groups contained in lignin isolated from palm kernel shells. Lignin polymer contained several groups generally identified as guaiacyl unit, syringyl, para hydroxy propane, -OH, and several aldehyde groups residues inside chains. Several constituent groups were generally identified in lignin, with absorption at a wavenumber of 3400-345 cm⁻¹ for OH linkage, 2820-2940 cm⁻¹ for C-H methyl linkage, 1600-1515 cm⁻¹ for the aromatic ring, 1460-1470 cm⁻¹ for asymmetrical C-H linkage, 1330-1315 cm⁻¹ for Syringyl ring linkage, 1270-1280 cm⁻¹ for guaiacyl ring, 1030-1085 cm⁻¹ for ether linkage and 870-875 cm⁻¹ for C-H aromatic ring. As seen in Figure 4, FT-IR spectra at wavenumber range of 400-4000 cm⁻¹ indicated that lignin isolated from EFB in this study was among absorption band standard range and relevant with general lignin constituent groups, such as O-H linkage, guaiacyl, and syringyl unit. It was concluded that the isolated compound was lignin.

Figure 3. FTIR graphic of lignin isolate obtained at various heating temperatures (a) 60°C (b) 85°C (c) 100°C (d) 121°C.
Table 3. FT-IR functional groups identification of lignin isolated from palm kernel shell obtained using various heating temperature.

| Heating temperature (°C) | Wavenumber (cm⁻¹) | Absorption band standard range | Functional groups       |
|--------------------------|-------------------|-------------------------------|-------------------------|
| 60                       | a 2924            | 2820-2950                     | C-H methyl linkage      |
|                          | b 1643            | 1633                          | Aromatic ring           |
|                          | c 1265            | 1278                          | Guaiacyl ring           |
|                          | d 825             | 850-875                       | Ether linkage           |
| 85                       | a 3417            | 3400-3450                     | O-H linkage             |
|                          | b 2924            | 2820-2950                     | C-H methyl linkage      |
|                          | c 1643            | 1600-1610                     | Aromatic ring           |
|                          | d 1368            | 1330-1315                     | Syringyl ring           |
|                          | e 1273            | 1270-1280                     | Guaiacyl ring           |
|                          | f 1018            | 1030-1085                     | Ether linkage           |
| 100                      | a 2947            | 2820-2950                     | C-H methyl linkage      |
|                          | b 1643            | 1633                          | Aromatic ring           |
|                          | c 1273            | 1270-1280                     | Guaiacyl ring           |
|                          | d 1041            | 1030-1085                     | Ether linkage           |
| 121                      | a 2947            | 2820-2950                     | C-H methyl linkage      |
|                          | b 1643            | 1633                          | Aromatic ring           |
|                          | c 1273            | 1270-1280                     | Guaiacyl ring           |
|                          | d 840             | 850-875                       | Ether linkage           |

Based on Figure 3 and Table 3, there was quite a similar functional group profile of lignin isolated at various temperatures of 60, 85, 100, and 121°C, such as ether linkage, aromatic ring, C-H methyl linkage, guaiacyl, and syringyl ring. From the FT-IR spectra, it is also revealed that the lignin type of EFB is SGH-type lignin which is found in almost monocotyledonous [23].

4. Conclusion

Lignin isolate obtained using the organosolv method in this study had characteristics according to lignin standards from previous research. The highest yield of lignin isolate 15.87% was obtained from EFB at a heating temperature of 85°C, while the highest purity of 94.87% was obtained at 121°C. Heating temperature affected pH of lignin isolate, which then affected on isolate amount, yield, and purity of lignin from EFB isolated using organosolv method. The higher temperature of above 85°C promotes lignin condensation. Functional groups analysis of lignin isolate indicated that lignin obtained at 60°C, 85°C, 100°C, and 121°C had similar functional groups, consisted of ether linkage, aromatic ring, C-H methyl linkage, guaiacyl ring, and syringyl ring which indicates that the lignin type is SGH-type lignin.

References

[1] Rupani P F R, Singh P, Ibrahim M H and Esa N 2010 Review of current palm oil mill effluent (pome) treatment methods: vermicomposting as a sustainable practice World Appl. Sci. J. 11(1) 70-81

[2] Visvanathan C, Setiadi T, Herarth G and Shi H 2009 Eco- industrial clusters in urban- rural fringe areas, Asian Institute of Technology, Thailand
[3] Badan Pusat Statistik. Statistic Indonesian 2019 Statistik kelapa sawit Indonesia, Indonesian palm oil statistic 2019 Badan Pusat Statistik Indonesia

[4] Mardawati E, Werner A, Bley T, Kresnowati M and Setiadi T 2014 The enzymatic hydrolysis of oil palm empty fruit bunches to xylose J. Jpn. Inst. Energy 93 973-978. http://dx.doi.org/10.3775/jie.93.973

[5] Julie B, Bikash K N, Ritika S, Sachin K, Ramesh C D, Deben C B and Eeshan K 2018 Recent trends in the pretreatment of lignocellulosic biomass for value-added products Front. Energy Res. 6 https://doi.org/10.3389/fenrg.2018.00141

[6] Lee H V, Hamid S B A, and Zain S K 2014 Conversion of lignocellulosic biomass to nanocellulose: structure and chemical process. Hindawi publishing corporation Sci. World J. 2014 http://dx.doi.org/10.1155/2014/631013

[7] MTAP K, Efri M, and Tjandra S 2015 Production of xylitol from oil palm empty fruits bunch: a case study on biofinery concept Mod. Appl. Sci. 9 (7) 1913-1852

[8] Osbert Y and Kwang H K 2020 Review: Lignin to Materials: A Focused Review on Recent Novel Lignin Applications Appl Sci. 2020 (10) 4626-4635 doi:10.3390/app10134626

[9] Qianqian T, Yong Q, Dongjie Y, Xueqing Q, Yanlin Q and Mingsong Z 2020 Lignin-Based Nanoparticles: A Review on Their Preparations and Applications Polymers 2020 (12) 2471

[10] Chen H 2015 Lignocellulose Biorefinery Engineering: Principles and Applications Woodhead Publishing: Cambridge, UK

[11] Sun J X, Xu F, Sun X F, Xiao B and Sun R C 2005 Physico-chemical and thermal characterization of cellulose from barley straw Polym Degrad Stab 88 521–531 doi: 10.1016/j.polymdegradstab.2004.12.013

[12] Nitsos C, Stoklosa R, Kamaori A, Voros D, Lange H, Hodge D, Crestini C, Rova U, and Christakopoulos P 2016 Isolation and characterization of organosolv and alkaline lignins from hardwood and softwood biomass ACS Sustain. Chem. Eng. 4(10) 5181–5193

[13] Kinanthi M, Jorge A F, Ria M, Wiratni B, Claes N and Mohammad J T 2020 Recovery of high purity lignin and digestible cellulose from oil palm empty fruit bunch using low acid-catalysed organosolv pretreatment Agronomy 2020(10) 674

[14] Mihai B and Cornelia V 2010 Thermal degradation of lignin – A Review Cellul. Chem. Technol. 44(9) 353-363

[15] Yao L, Yong C L, Hong Q H, Feng J X, Xian Y W and Xing F 2017 Structural characterization of lignin and its degradation products with spectroscopic methods J Spectrosc (Hindawi) 2017 1-15 https://doi.org/10.1155/2017/8951658

[16] Berlin A and Balakshin M 2014 Industrial lignins: analysis, properties, and applications,” in Bioenergy Research: Advances and Applications, V K Gupta, M G T P Kubicek and J S Xu, Eds, 315–336, Elsevier, Amsterdam

[17] Sri H, Ahmad S Z and Wisnu S 2017 Effect of acetic acid: formic acid ratio on characteristics of pulp from oil palm empty fruit bunches (opefb) ARPN J. Eng. Appl. Sci. 12(12)

[18] Marcelo P D R, Paulo H B, Daniele G M, Juliana B M, Jessica S D S and Ana M M D 2017 Extraction of organosolv lignin from rice husk under reflux condition J. biol. chem.res. 2017 87-98

[19] Edita J, Matej H, Miha G B L 2020 Acid-catalysed α-O-4 aryl-ether bond cleavage in methanol/(aqueous) ethanol: understanding depolymerisation of a lignin model compound during organosolv pretreatment Sci. Rep. 10(1) 11037 DOI: 10.1038/s41598-020-67787-9

[20] Feng H, Yanlei W, Yu X, Xin T, Suojiang Z and Hongyan H 2019 Theoretical Elucidation of β-O-4 Bond Cleavage of Lignin Model Compound Promoted by Sulfonylic Acid-Functionalized Ionic Liquid Front. Chem. 2019 https://doi.org/10.3389/fchem.2019.00078

[21] Zhu W 2013 Equilibrium of Lignin Precipitation Effects of pH, temperature, Ion Strength and Wood Origins Thesis Department of Chemical and Biological Engineering, Chalmers University of Technology Gothenburg, Sweden
[22] Hochegger M, Boitte B C, Cezard L, Schober S and Mittelbach M 2019 Influence of ethanol organosolv pulping conditions on physicochemical lignin properties of European larch Int. J. Chem. Eng. 2019 https://doi.org/10.1155/2019/1734507

[23] Poovaiah C R, Nageswara R M, Soneji J R, Baxter H L and Stewart T R 2014 Altered lignin biosynthesis using biotechnology to improve lignocellulosic biofuel feedstocks Plant Biotechnol. J. 12 1163–1173

[24] Yáñez-S M, Matsuhiro B, Nuñez C, Pan S, Hubbel C A, Sannigrahi P and Ragauskas A J 2014 Physicochemical characterization of ethanol organosolv lignin (EOL) from Eucalyptus globulus: effect of extraction conditions on the molecular structure Polym. Degrad. Stab. 110 184–194

[25] Lourenco A and Pereira H 2018 Chapter 3: Compositional variability of lignin in biomass Intech open http://dx.doi.org/10.5772/intechopen.71208