DELIGNIFICATION OF UNTREATED ELEPHANT GRASS (EG) USING MICROWAVE ASSISTED BLEACHING PROCESS PRIOR TO NANO-CELLULOSE CRYSTAL ISOLATION: EFFECT OF HYDROGEN PEROXIDE CONCENTRATION ON LIGNIN DEGRADATION

Fathin Nor Faridah Mohamed Nawi, Norazah Abd Rahman, and Syaidatul Akma Mohd Zuki

Faculty of Chemical Engineering, Universiti Teknologi Mara (UiTM) Shah Alam

***Abstract*—NCC isolation involved pretreatment stages which are alkali pretreatment and bleaching. Microwave assisted process for NCC isolation expected to be efficient in the lignin degradation. The objectives are to extract cellulose from untreated EG using microwave assisted bleaching process and determine lignin degradation using different concentration of hydrogen peroxide (H_2O_2) as bleaching agent. Untreated EG will be bleached using different concentration of H_2O_2 in different time. The percentage of total lignin degradation can be determined from the summation of acid soluble lignin (ASL) and acid insoluble lignin (AIL). The result showed that the highest percentage of total lignin degradation is at 120 minutes, 30% concentration of H_2O_2, with 180W. This can be conclude that microwave assisted bleaching process can be an effective pretreatment method for NCC isolation.**

***Keywords*— bleaching, microwave irradiation, elephant grass, lignin, nanocrystalline cellulose.**

I. Introduction

Cellulose, hemicellulose and lignin are the principal components in lignocellulosic biomasses and their auxiliary piece fluctuates with climatic and development conditions [1]. Typically, there are about 40-50% cellulose, 20-30% hemicellulose and 10-25% lignin comprised in agricultural lignocellulosic biomass [2]. From the present perspective, cellulose is the most basic natural polymer which about 1.5 x 10^{12} tons of the yearly biomass creation and is considered an practically unlimited wellspring of crude material for the expanding interest for earth neighborly and biocompatible items [3]. In recent year, attention towards green chemistry principals due to environmental concern have attracted scientists to do research on production of nanocrystalline cellulose (NCC) from lignocellulosic materials due to its miscellaneous application [4] that could replace the non-biodegradable resources [5] and their various peculiar properties make them potentially relevant in a different field of scientific and technological developments [6, 7].

Nanocellulose is a particle with nano-sized dimension which displays structural characteristic of low density, renewability, high crystallinity, high mechanical strength, biocompatible and high surface area connected with modified hydroxyl group [8, 9]. Recently, the nanocrystalline cellulose (NCC) has been recognized for its greatly diverse application as emulsifier, cosmetics, thickener and stabilizer in food and pharmaceuticals applications [10, 11]. Besides, NCC also most being used as filters or reinforcement agents in aerospace, automobile and some biomedical field for manufacturing high performance materials with good thermal stability [12].

Nanocrytalline cellulose (NCC) can be extracted from sources such as animal (tunicates) [13], algae [14], bacteria [15] and lignocellulosic biomasses [16]. There are also a few lignocellulosic biomass that have been done in NCC isolation such as jute fibers [17], sugarcane bagasse [18, 19], olive tree [20], areca palm leave stalk [21], brewers spent grain, hazelnut shell, wheat straw and orange peels [22]. However, this interest also includes the search to find a new biomass sources for the production of nanocrystalline cellulose (NCC) and in this outcome, Elephant Grass found to be possibly engaging profitable source. Elephant grass or its specific name, Pennisetum purpureum is a species of Napier grass which contains 60.20% cellulose, 23.80% hemicellulose and 8.20% lignin. It is a major and one of the highest yielding tropical grass [23]. This natural fiber is derived for use in plastic, automotive, and packaging industries because of their excellent characteristics such as high specific stiffness, low density, eco-friendly, good mechanical properties, good thermal and acoustic insulation, toxicologically harmless and biodegradability [24]. Recently, many researchers had done some research on NCC isolation using a species of Napier grass and the outcomes were very encouraging as the studies showed that Napier grass has a great potential for the industrial productions [5, 24-30].

NCC isolation from cellulose materials happens in two stages which are pretreatment of the materials and chemical treatment for extraction process, generally bleaching [31]. Pretreatment process required to amend the shape of cellulosic biomass to make sure cellulose more approachable [27]. Normally, oxidation agent such as hydrogen peroxide (H_2O_2) is used to enhance the effect of bleaching treatment and it is suitable for extraction of cellulose as the oxidative agent is more aggressive on lignin and hemicellulose meanwhile cellulose is hardly decomposed. Some researchers also use chlorine [32], calcium hypochlorite [12], sodium hypochlorite [4], acetate buffer and sodium chlorite [33] as a bleaching agent in order to remove the phenolic chromophores but resulting in a negative environment effects.

Nevertheless, although pretreatment and bleaching are the common and widely used method in delignification process however, this process does not result in significant biomass delignification instead causing repolymerization of some amount of lignin with other degraded products and the requirement of large amount of acid can cause the corrosion problem to the equipment. Besides, the acid or alkali chemical pretreatment methods that used conventional heating method also have disadvantages such as low conversion of biomass components and producing by-products. Hence, the bleaching assisted microwave technique can be used in order to improve the yield of NCC isolation. Lignocellulosic biomass was revealed to the microwave heating as the effect of swelling and fragmentation occurred internally within the biomass materials resulting interference of biomass structures [30].

Microwave technology assisted bleaching process represents as a replacement and genuine option to conventional thermal treatment [34]. They found that these process have become popular due to their low energy consumption, fast reaction time, fully distributed heat convection and effective heat transfer as compared to conventional heating process such as thermo-chemical conversion technologies and biological conversion technologies [35, 36]. Microwave irradiation induces heat at the molecular level by direct conversion of the electromagnetic heat into energy [37]. Several studied have analyzed the use of microwave irradiation preceding to NCC isolation and it has been illustrated that microwave heating is efficient in accelerate the lignin degradation [31, 38].

Therefore, there are two objectives of this research which are to extract cellulose from untreated Elephant Grass (EG) using microwave assisted bleaching process and to determine lignin degradation using different concentration of H_2O_2. In this study, the focus is the microwave-assisted bleaching to pretreat the untreated elephant grass with present of bleaching process. The manipulated parameter is the concentration of H_2O_2 at 20%, 25% and 30%. The bleached elephant grass later will be extracted to find the lignin content.

II. METHODOLOGY

A. *Preparation of raw materials*

The Elephant Grass roughly cut into 2cm and oven dried at 110^{o}C until the moisture content is <10%. The sample was grounded and sieved to obtain 355µm and was sealed and placed in the desiccator to maintain its moisture content before further process.

B. *Bleaching process*

The raw material was bleached using three different concentration of hydrogen peroxide which are 20%, 25% and 30% for seven different time which are 5,10,15, 30, 60, 90 and 120 minutes [39].

C. *Lignin extraction*

0.3g of bleached biomass was added into 3ml of 72% sulphuric acid and was kept for 2 hours in a room temperature. After that, 84ml of distilled water was added into the sample and the sample was being autoclaved at 121^{o}C. The sample was filtered through vacuum using filtering crucible and the residue was dried at 121^{o}C in order to get the percent of acid insoluble lignin (AIL). The acid soluble lignin (ASL) also being measured using UV-VIS [40].

III. RESULTS AND DISCUSSION

A. *The effects of microwave power at 180W in 20, 25 and 30% concentration of hydrogen peroxide on lignin degradation.*

Total of percentage lignin degradation is the summation of acid insoluble lignin (AIL) and acid soluble lignin (ASL) results. Acid insoluble lignin (AIL) is known as an insoluble fraction sample obtained through filtration process using vacuum pump in order to separate the AIL and ASL and it can be calculated manually in order to get the percentage of lignin degrade in the process. Meanwhile, acid soluble lignin (ASL) is known as a hydrolysate from lignin removal process where a mixture of 3 ml 72% H2SO4 with 0.3 g bleached sample was mixed with 84 ml distilled water and has been autoclaved for 1 hour. The graph showed the percentage of AIL, ASL and total lignin degradation from different concentration of H_2O_2 versus time of bleaching process at 20%, 25% and 30% hydrogen peroxide concentration.

Figure 1 showed the graph trends of the percentage AIL increased for all the three concentrations. The increment of the percentages can be explained by sufficient hydrolysis of insoluble polysaccharides in the acidic suspension, thus resulting sufficient destruction of the microfibrils structure which later give a higher percentage of degradation [41]. Half of the linkage structures in lignin consists of the ß-O-4 bond and some research has been done stated that depolymerisation of lignin normally targeting the ß-O-4 bond cleavage, thus it will become acid soluble lignin. But, some of them will remain as acid insoluble lignin [42, 43]. The highest percentage was obtained at point of 120 minutes in 30% concentration of H_2O_2 because the reaction of sulphuric acid decreased the lignin content in the biomass and resulting highest percentage of degradation based on the weight of the original biomass [44]. The elimination of lignin groups such as ethylenic and carbonyl groups and solubilisation of hemicellulose are from the reaction of hydroperoxide anions (HOO-) and radical hydroxyl ions (OH-).

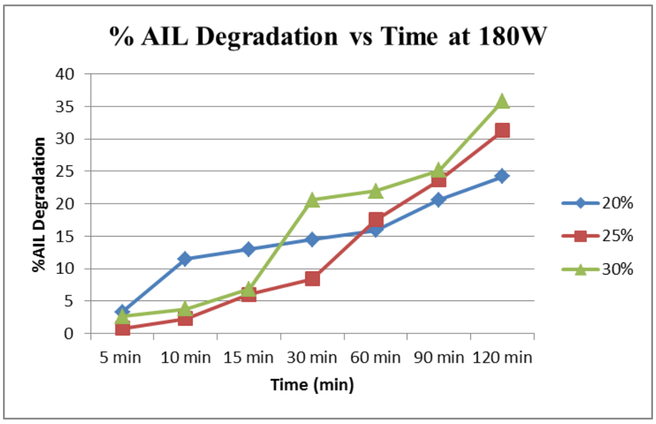

Fig 1: Graph of % Acid Insoluble Lignin (AIL) degradation at 180W

Figure 2 showed the graph trend of percentage ASL degradation for all three concentrations. This indicated that during the bleaching process, the linkage between lignocellulosic materials might be broken evenly with increasing of reaction time hence it will ease the hydrolysis process in the sulphuric acid solution to solubilize the lignin in the liquid fractions and resulting the increasing percentage of the degradation at 30% concentration [45]. Based on the graph, the percentage of the ASL is the highest at 120 minutes in 30% concentration which contributed as a peak towards this process. The utilization of high concentration of sulphuric acid which provide high concentration H_3O^+ ions that in charge of the protonation of ether-bond group in lignin, thus enhance the bonds to eventually hydrolyzed and give the high percentage of the ASL [46]. As is observed in graph of 30% concentration, the power of microwave with increasing time and concentration in bleaching process enhanced the efficiency of lignin degradation due to the disorganized and broken structures of lignocellulosic materials by microwave irradiation [47] that give high surface area for the fast reaction to take place [46].

Meanwhile, the acid soluble lignin showed a significant decreased with respect to reaction time because of the efficient removal of the non-cellulosic components from the sample which resulting in the reduction of percentage ASL in the sample [48]. Besides, the breakage of intermolecular ester linkages between cellulose and lignin during the bleaching treatment does not distributed evenly with existence of heat from microwave irradiation which affected the electromagnetic waves through the sample, thus reducing the percentage of ASL [31]. Furthermore, at concentration 20% and 25%, the percentage ASL reduced can be illustrated by reduction of some activity by the groups contains in the sample that unable to overcome the re-polymerization process and degradation effectively [49] which showed in the figure 2.

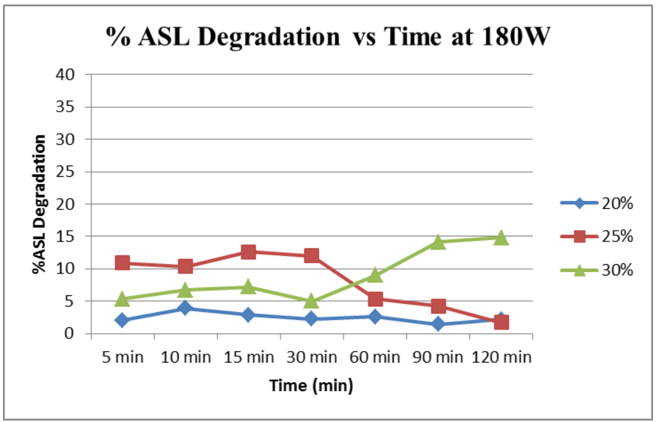

Fig 2: Graph of % Acid Soluble Lignin (ASL) degradation at 180W

Figure 3 showed that the highest percentage of lignin degradation was at 30% hydrogen peroxide concentration for 120 minutes as the retention time gave a significant effect on the degradation process. Furthermore, the enhancement of the cleavage of the chemical bond provides an efficient degradation process. It was said that high retention time presumably assisted more efficient contact time for hydroxide and oxygen radical ions with the lignin content [49]. At microwave power of 180 W, it was found to be the maximum percentage of lignin degradation because of the mechanism of microwave which used electromagnetic waves induction through the sample compared to conventional heating that used only poor conduction process [31]. The amorphous region in the biomass can absorb the chemical solution and become soft and flexible thus resulting the hydrolysis of the amorphous regions into nanocrystalline cellulose.

Besides, microwave heating helps in the dewaxing of the biomass. After the bleaching process is done, the sample will undergo acid hydrolysis process where the cellulosic fiber is exposed to the sulphuric acid. The process introduced to remove the amorphous region and isolation of NCC. As the concentration of bleaching agent is higher, the cellulose amorphous region will going through a hydrolytic cleavage by hydronium ions and released an individual crystallite [50].

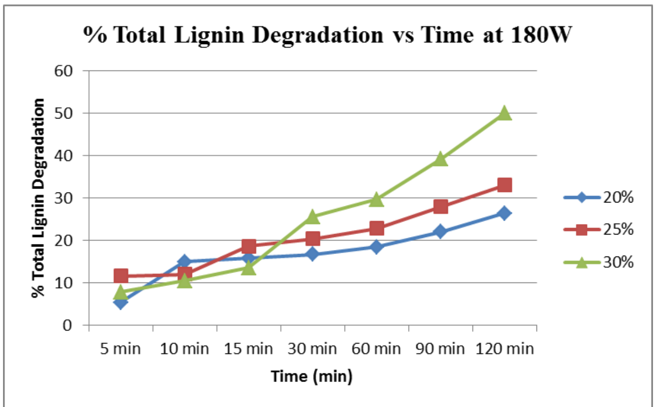

Fig 3: Graph of % total lignin degradation at 180W

IV. Conclusion

In summary, microwave assisted bleaching process could simultaneously degrade the lignin from biomass elephant grass. The bleaching process resulted in 49.93% of total lignin degradation at 180 W of microwave power at 120 minutes. The comparison between different concentration can be seen clearly indicate that concentration hydrogen peroxide for bleaching process at 30% give the best maximum for lignin degradation. Besides, significant lignin degradation could be achieved effectively using microwave irradiation due to their heating mechanism.

Acknowledgment

Foremost, I would like to express my sincere gratitude to my advisors Prof. Dr. Norazah and Cik Syaidatul Akma for the continuous support of my research study, for the patience, motivation, enthusiasm and immense knowledge. Their guidance helped me in all the time of research and writing of this manuscript. I could not have imagined having a better advisor and mentor for my research study.

I thank my fellow labmate: Nur Fatin Syafiqah for the stimulating discussions, for the sleepless nights we were working together before deadlines and for all the fun we have had in completing this research. Also I thank my friends: Azmah Kamaruddin, Afiqah Tasneem and Fatin Najwa for continuously motivate me in finishing this research project.

Last but not the least, I would like to thank my family: my parents for giving extremely supportive motivation for me to be strong in this study and supporting me spiritually throughout my life.

References

[1] D. Ahuja, A. Kaushik, and M. Singh, "Simultaneous extraction of lignin and cellulose nanofibrils from waste jute bags using one pot pre-treatment," *International Journal of Biological Macromolecules,* vol. 107, pp. 1294-1301, 2018/02/01/ 2018.

[2] K. B. Ansari and V. G. Gaikar, "Green hydrotropic extraction technology for delignification of sugarcane bagasse by using alkybenzene sulfonates as hydrotropes," *Chemical Engineering Science,* vol. 115, pp. 157-166, 2014/08/01/ 2014.

[3] D. Klemm, B. Heublein, H.-P. Fink, and A. Bohn, "Cellulose: Fascinating Biopolymer and Sustainable Raw Material," *Angewandte Chemie International Edition,* vol. 44, pp. 3358-3393, 2005.

[4] M. S. Islam, N. Kao, S. N. Bhattacharya, R. Gupta, and H. J. Choi, "Potential aspect of rice husk biomass in Australia for nanocrystalline cellulose production," *Chinese Journal of Chemical Engineering,* vol. 26, pp. 465-476, 2018/03/01/ 2018.

[5] C. O. Aniagor and M. C. Menkiti, "Kinetics and mechanistic description of adsorptive uptake of crystal violet dye by lignified elephant grass complexed isolate," *Journal of Environmental Chemical Engineering,* vol. 6, pp. 2105-2118, 2018/04/01/ 2018.

[6] K. O. Reddy, C. U. Maheswari, M. S. Dhlamini, B. M. Mothudi, V. P. Kommula, J. Zhang, *et al.*, "Extraction and characterization of cellulose single fibers from native african napier grass," *Carbohydrate Polymers,* vol. 188, pp. 85-91, 2018/05/15/ 2018.

[7] F. Beltramino, M. Blanca Roncero, T. Vidal, and C. Valls, "A novel enzymatic approach to nanocrystalline cellulose preparation," *Carbohydrate Polymers,* vol. 189, pp. 39-47, 2018/06/01/ 2018.

[8] L. Brinchi, F. Cotana, E. Fortunati, and J. M. Kenny, "Production of nanocrystalline cellulose from lignocellulosic biomass: Technology and applications," *Carbohydrate Polymers,* vol. 94, pp. 154-169, 2013/04/15/ 2013.

[9] R. A. Ilyas, S. M. Sapuan, R. Ibrahim, H. Abral, M. R. Ishak, E. S. Zainudin, *et al.*, "Sugar palm (Arenga pinnata (Wurmb.) Merr) cellulosic fibre hierarchy: a comprehensive approach from macro to nano scale," *Journal of Materials Research and Technology,* 2019/05/15/ 2019.

[10] L. K. Kian, M. Jawaid, H. Ariffin, and Z. Karim, "Isolation and characterization of nanocrystalline cellulose from roselle-derived microcrystalline cellulose," *International Journal of Biological Macromolecules,* vol. 114, pp. 54-63, 2018/07/15/ 2018.

[11] Z. A. Zianor Azrina, M. D. H. Beg, M. Y. Rosli, R. Ramli, N. Junadi, and A. K. M. M. Alam, "Spherical nanocrystalline cellulose (NCC) from oil palm empty fruit bunch pulp via ultrasound assisted hydrolysis," *Carbohydrate Polymers,* vol. 162, pp. 115-120, 2017/04/15/ 2017.

[12] L. K. Kian, N. Saba, M. Jawaid, and M. T. H. Sultan, "A review on processing techniques of bast fibers nanocellulose and its polylactic acid (PLA) nanocomposites," *International Journal of Biological Macromolecules,* 2018/09/09/ 2018.

[13] M. J. Dunlop, B. Acharya, and R. Bissessur, "Isolation of nanocrystalline cellulose from tunicates," *Journal of Environmental Chemical Engineering,* vol. 6, pp. 4408-4412, 2018/08/01/ 2018.

[14] Y. W. Chen, H. V. Lee, J. C. Juan, and S.-M. Phang, "Production of new cellulose nanomaterial from red algae marine biomass Gelidium elegans," *Carbohydrate Polymers,* vol. 151, pp. 1210-1219, 2016/10/20/ 2016.

[15] H. Ullah, F. Wahid, H. A. Santos, and T. Khan, "Advances in biomedical and pharmaceutical applications of functional bacterial cellulose-based nanocomposites," *Carbohydrate Polymers,* vol. 150, pp. 330-352, 2016/10/05/ 2016.

[16] N. M. Julkapli and S. Bagheri, "Progress on nanocrystalline cellulose biocomposites," *Reactive and Functional Polymers,* vol. 112, pp. 9-21, 2017/03/01/ 2017.

[17] E. Castro-Aguirre, F. Iñiguez-Franco, H. Samsudin, X. Fang, and R. Auras, "Poly(lactic acid)—Mass production, processing, industrial applications, and end of life," *Advanced Drug Delivery Reviews,* vol. 107, pp. 333-366, 2016/12/15/ 2016.

[18] P. L. de Hoyos-Martínez, X. Erdocia, F. Charrier-El Bouhtoury, R. Prado, and J. Labidi, "Multistage treatment of almonds waste biomass: Characterization and assessment of the potential applications of raw material and products," *Waste Management,* vol. 80, pp. 40-50, 2018/10/01/ 2018.

[19] J. Ben-Iwo, V. Manovic, and P. Longhurst, "Biomass resources and biofuels potential for the production of transportation fuels in Nigeria," *Renewable and Sustainable Energy Reviews,* vol. 63, pp. 172-192, 2016/09/01/ 2016.

[20] A. Pérez, M. A. Martín-Lara, A. Gálvez-Pérez, M. Calero, and A. Ronda, "Kinetic analysis of pyrolysis and combustion of the olive tree pruning by chemical fractionation," *Bioresource Technology,* vol. 249, pp. 557-566, 2018/02/01/ 2018.

[21] Z. A. Raza, S. Abid, and I. M. Banat, "Polyhydroxyalkanoates: Characteristics, production, recent developments and applications," *International Biodeterioration & Biodegradation,* vol. 126, pp. 45-56, 2018/01/01/ 2018.

[22] V. Vadivel, A. Moncalvo, R. Dordoni, and G. Spigno, "Effects of an acid/alkaline treatment on the release of antioxidants and cellulose from different agro-food wastes," *Waste Management,* vol. 64, pp. 305-314, 2017/06/01/ 2017.

[23] T. Senthil Muthu Kumar, N. Rajini, K. Obi Reddy, A. Varada Rajulu, S. Siengchin, and N. Ayrilmis, "All-cellulose composite films with cellulose matrix and Napier grass cellulose fibril fillers," *International Journal of Biological Macromolecules,* vol. 112, pp. 1310-1315, 2018/06/01/ 2018.

[24] V. Basso, J. C. Machado, F. J. da Silva Lédo, J. da Costa Carneiro, R. C. Fontana, A. J. P. Dillon, *et al.*, "Different elephant grass (Pennisetum purpureum) accessions as substrates for enzyme production for the hydrolysis of lignocellulosic materials," *Biomass and Bioenergy,* vol. 71, pp. 155-161, 2014/12/01/ 2014.

[25] A. de Araújo Morandim-Giannetti, T. S. Albuquerque, R. K. C. de Carvalho, R. M. S. Araújo, and R. Magnabosco, "Study of “napier grass” delignification for production of cellulosic derivatives," *Carbohydrate Polymers,* vol. 92, pp. 849-855, 2013/01/30/ 2013.

[26] A. L. Scholl, D. Menegol, A. P. Pitarelo, R. C. Fontana, A. Z. Filho, L. P. Ramos, *et al.*, "Elephant grass pretreated by steam explosion for inducing secretion of cellulases and xylanases by Penicillium echinulatum S1M29 solid-state cultivation," *Industrial Crops and Products,* vol. 77, pp. 97-107, 2015/12/23/ 2015.

[27] S. A. Nascimento and C. A. Rezende, "Combined approaches to obtain cellulose nanocrystals, nanofibrils and fermentable sugars from elephant grass," *Carbohydrate Polymers,* vol. 180, pp. 38-45, 2018/01/15/ 2018.

[28] S. Montipó, I. Ballesteros, R. C. Fontana, S. Liu, A. F. Martins, M. Ballesteros, *et al.*, "Integrated production of second generation ethanol and lactic acid from steam-exploded elephant grass," *Bioresource Technology,* vol. 249, pp. 1017-1024, 2018/02/01/ 2018.

[29] J. Minmunin, P. Limpitipanich, and A. Promwungkwa, "Delignification of Elephant Grass for Production of Cellulosic Intermediate," *Energy Procedia,* vol. 79, pp. 220-225, 2015/11/01/ 2015.

[30] C. Eliana, R. Jorge, P. Juan, and R. Luis, "Effects of the pretreatment method on enzymatic hydrolysis and ethanol fermentability of the cellulosic fraction from elephant grass," *Fuel,* vol. 118, pp. 41-47, 2014/02/15/ 2014.

[31] R. D. Singh, K. Bhuyan, J. Banerjee, J. Muir, and A. Arora, "Hydrothermal and microwave assisted alkali pretreatment for fractionation of arecanut husk," *Industrial Crops and Products,* vol. 102, pp. 65-74, 2017/08/01/ 2017.

[32] J. Fernández-Rodríguez, O. Gordobil, E. Robles, M. González-Alriols, and J. Labidi, "Lignin valorization from side-streams produced during agricultural waste pulping and total chlorine free bleaching," *Journal of Cleaner Production,* vol. 142, pp. 2609-2617, 2017/01/20/ 2017.

[33] D. M. d. Santos, A. d. L. Bukzem, D. P. R. Ascheri, R. Signini, and G. L. B. d. Aquino, "Microwave-assisted carboxymethylation of cellulose extracted from brewer's spent grain," *Carbohydrate Polymers,* vol. 131, pp. 125-133, 2015/10/20/ 2015.

[34] E. T. Kostas, D. Beneroso, and J. P. Robinson, "The application of microwave heating in bioenergy: A review on the microwave pre-treatment and upgrading technologies for biomass," *Renewable and Sustainable Energy Reviews,* vol. 77, pp. 12-27, 2017/09/01/ 2017.

[35] A. Hivechi and S. H. Bahrami, "A new cellulose purification approach for higher degree of polymerization: Modeling, optimization and characterization," *Carbohydrate Polymers,* vol. 152, pp. 280-286, 2016/11/05/ 2016.

[36] Z. M. A. Bundhoo, "Microwave-assisted conversion of biomass and waste materials to biofuels," *Renewable and Sustainable Energy Reviews,* vol. 82, pp. 1149-1177, 2018/02/01/ 2018.

[37] V. Rigual, T. M. Santos, J. C. Domínguez, M. V. Alonso, M. Oliet, and F. Rodriguez, "Evaluation of hardwood and softwood fractionation using autohydrolysis and ionic liquid microwave pretreatment," *Biomass and Bioenergy,* vol. 117, pp. 190-197, 2018/10/01/ 2018.

[38] H. Wang, M. L. Maxim, G. Gurau, and R. D. Rogers, "Microwave-assisted dissolution and delignification of wood in 1-ethyl-3-methylimidazolium acetate," *Bioresource Technology,* vol. 136, pp. 739-742, 2013/05/01/ 2013.

[39] M. Rajinipriya, M. Nagalakshmaiah, M. Robert, and S. Elkoun, "Homogenous and transparent nanocellulosic films from carrot," *Industrial Crops and Products,* vol. 118, pp. 53-64, 2018/08/01/ 2018.

[40] A. O. Ayeni, O. Adeeyo, O. Oresegun, and T. Oladimeji, *Compositional analysis of lignocellulosic materials: Evaluation of an economically viable method suitable for woody and non-woody biomass* vol. 4, 2015.

[41] S. Yasuda, K. Fukushima, and A. Kakehi, "Formation and chemical structures of acid-soluble lignin I: sulfuric acid treatment time and acid-soluble lignin content of hardwood," *Journal of Wood Science,* vol. 47, pp. 69-72, 2001/02/01 2001.

[42] J. Dai, A. F. Patti, and K. Saito, "Recent developments in chemical degradation of lignin: catalytic oxidation and ionic liquids," *Tetrahedron Letters,* vol. 57, pp. 4945-4951, 2016/11/09/ 2016.

[43] G.-Q. Chen and X.-R. Jiang, "Engineering microorganisms for improving polyhydroxyalkanoate biosynthesis," *Current Opinion in Biotechnology,* vol. 53, pp. 20-25, 2018/10/01/ 2018.

[44] P. Verma, T. Watanabe, Y. Honda, and T. Watanabe, "Microwave-assisted pretreatment of woody biomass with ammonium molybdate activated by H2O2," *Bioresource Technology,* vol. 102, pp. 3941-3945, 2011/02/01/ 2011.

[45] X. Meng, F. Liu, Y. Xiao, J. Cao, M. Wang, and X. Duan, "Alterations in physicochemical and functional properties of buckwheat straw insoluble dietary fiber by alkaline hydrogen peroxide treatment," *Food Chemistry: X,* p. 100029, 2019/05/14/ 2019.

[46] F. Avelino, K. T. d. Silva, M. d. S. M. de Souza Filho, S. E. Mazzetto, and D. Lomonaco, "Microwave-assisted organosolv extraction of coconut shell lignin by Brønsted and Lewis acids

catalysts," *Journal of Cleaner Production,* vol. 189, pp. 785-796, 2018/07/10/ 2018.

[47] H. Bakhshabadi, H. Mirzaei, A. Ghodsvali, S. M. Jafari, A. M. Ziaiifar, and V. Farzaneh, "The effect of microwave pretreatment on some physico-chemical properties and bioactivity of Black cumin seeds' oil," *Industrial Crops and Products,* vol. 97, pp. 1-9, 2017/03/01/ 2017.

[48] J. Xie, C.-Y. Hse, C. F. De Hoop, T. Hu, J. Qi, and T. F. Shupe, "Isolation and characterization of cellulose nanofibers from bamboo using microwave liquefaction combined with chemical treatment and ultrasonication," *Carbohydrate Polymers,* vol. 151, pp. 725-734, 2016/10/20/ 2016.

[49] Y. Cheng, P.-X. Zhao, M. H. Alma, D.-F. Sun, R. Li, and J.-X. Jiang, "Improvement of direct liquefaction of technical alkaline lignin pretreated by alkaline hydrogen peroxide," *Journal of Analytical and Applied Pyrolysis,* vol. 122, pp. 277-281, 2016/11/01/ 2016.

[50] F. I. Ditzel, E. Prestes, B. M. Carvalho, I. M. Demiate, and L. A. Pinheiro, "Nanocrystalline cellulose extracted from pine wood and corncob," *Carbohydrate Polymers,* vol. 157, pp. 1577-1585, 2017/02/10/ 2017.