NANOCELLULOSE EXTRACTION FROM SUGARCANE BAGASSE THROUGH ULTRASONICATION-CHEMICAL COMBINATION METHOD

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Abstract
This study focuses on the extraction of nanocellulose from Sugarcane Bagasse by ultrasonication method. Ultrasonic waves can accelerate the dispersion process of nanocellulose particles so that extraction runs faster and environmentally friendly. The baggasse was treated by chemical treatment with ultrasonic waves, and then the nanocellulosa was prepared using acid hydrolysis with ultrasonic waves. The effect of ultrasonication was investigated. X-ray diffraction analysis was used to characterize the crystalline structure of cellulose and nanocellulose. Fourier-transformed infrared spectroscopy analysis was used to characterize the chemical composition of extracted cellulose and nanocellulose that verifying the removal of lignin anda hemicellulose during cellulose extraction process from sugarcane bagasse. Scanning electron microscopy was used to charaterize morphology of bagasse, cellulose, and nanocellulose. Particle size analysis was used to determine particle size of nanocellulose. The result of X-ray diffraction and Fourier-transformed infrared spectroscopy analysis showed that breakages of intramolecular hydrogen bonds and glycosidic bonds occured during the hydrolisis process. Furthermore,. The average size of the nanocellulose praticle was 132.67 nm.

Keywords*: Escerichia coli, Nanocellulose, sugarcane bagasse, synthesis, Ultrasonication*

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INTRODUCTION

Nanocellulose is a new type of cellulose material characterized by an increase in crystallinity, aspect ratio, surface area, and increased dispersion and biodegradability(Abitbol et al., 2016; Mondal, 2017; Oksman et al., 2016). Nanocellulose particles can be used as polymer reinforcing fillers, composites, additives for biodegradable products, membrane reinforcement, thickener for dispersions, and drug carrier media and implants (Ioelovich, 2012). Recently, Nanocellulose has received much attention in its application as a reinforcing filler in antibacterial agents (Szymańska-Chargot et al., 2019). Nanocellulose has a large number of hydroxyl groups so that it has a stable structure in water (Phanthong et al., 2018). Due to wide range application of Nanocellulose, various method of nanocellulose extraction have been developed. Li et al. (2012) succeeded in making nanocellulose with a diameter of 10-20 nm from bagasse through high pressure mechanical methods. Camargo et al. (2016) succeeded in making bagasse nanocellulose using the enzymatic hydrolysis method while Saputri et al. (2018) made nanocellulose from bagasse with a simpler technique, through a blending technique using a household blender. The chemical synthesis method can be done by hydrolysis (Guancha-Chalapud et al., 2020). Several studies have used nanocellulose as the base material for bionanocomposites because nanocellulose can be obtained from renewable and biodegradable natural materials (Effendi et al., 2015). Various methods of nanocellulose extraction have been developed, until now the discovery of an environmentally friendly, energy efficient and cost-effective nanocellulose isolation method remains a challenge. Therefore, it is necessary to conduct research on the method of extraction of nanocellulose-based antibacterial materials that are environmentally friendly, energy efficient, and cost effective.

Nanocellulose can be extracted from agricultural waste, one of which is bagasse (Camargo et al., 2016; Li et al., 2012; Saputri et al., 2018). Nanocellulose extraction methods from bagasse that have been successfully carried out include the high pressure mechanical method (Li et al., 2012), the enzymatic hydrolysis method, the acid hydrolysis method, and the ultrasonication method (Phanthong et al., 2018). Each method has its weaknesses, the high pressure mechanical method requires considerable energy, the enzymatic hydrolysis method requires a lot of cost and the extraction time is longer, the acid hydrolysis method is less environmentally friendly because of the use of high concentrations of acid, while the ultrasonic method is environmentally friendly, however Nanocellulose from acid hydrolysis is better than that of ultrasonication. Based on this, this study used a combination method of acid hydrolysis with ultrasonication.

The resulting nanocellulose then characterized its physical properties using several instruments. X-ray diffraction (XRD) analysis was used to characterize the crystalline structure of cellulose and nanocellulose.

Fourier-transformed infrared spectroscopy (FT-IR) analysis was used to characterize the chemical composition of extracted cellulose and nanocellulose that verifying the removal of lignin anda hemicellulose during cellulose extraction process from sugarcane bagasse. Scanning Electron Microscopy (SEM) was used to charaterize morphology of sugarcane bagasse, cellulose, and nanocellulose. Particle size analysis (PSA) was used to determine particle size of nanocellulose. XRD and FT-IR data analysis using Origin Pro Software.

METHOD

Material and Instrumentation

Sugarcane bagasses gathered in Sidoarjo (East java, Indonesia) were used in this study. The chemical reagent use were NaOH (Merck), NaClO (Merck), H_2SO_4 (Merck), Filter paper (Whatmann No.44), demineralized aqua (Bratachem). The equipment uses are glassess equipment, ultrasonicator, Oven (DGG 9053A), anaytical balance (Ohaus px224/E), sentrifuge. Morfology and atomic composition was observed by Scanning Electron Microscopy–Energy Dispersive X-Ray Spectrometry (SEM HITACHI FLEXSEM 1000), functional group of nanocellulose was characterized using Fourier-transformed infrared spectroscopy (FT-IR 8400S/Shimadzu). Cristallinity of nanocellulose was observed using X-Ray Diffraction (XRD Philips Analytical), and nanocellulose's particle size was analyzed using Particle size analyzer (PSA Horiba Scientific SZ-100).

Isolation nanocellulose form Sugarcane Bagasse

The nanocellulose isolation method used was Mandal & Chakrabarty (2011) modified method using an ultrasonicator. Clean and dry sugarcane bagasse (SB) is cut into small pieces and crushed into a finer powder, then oven at 60°C for 16 hours. The dry bagasse is put into the Erlenmeyer flask, then 250 ml of 17.5% w/v NaOH is added. The mixture was ultrasonicated for 2 hours at 70 ° C. Furthermore, the mixture is then filtered. The residue that has been produced is then bleached with a 0.7% v / v NaClO solution, then the mixture is sonicated for 2x1 hours. The residue produced during the bleaching process is washed with distilled water. The residue obtained was then dried in an oven at 60 ° C to obtain cellulose (C-SB). A total of 5 grams of C-SB was reacted with 25 mL of 45% H_2SO_4. Hydrolysis was carried out with the aid of an ultrasonicator at a temperature of 70 ° C for 90 minutes. After that the mixture was neutralized with 28 mL of 0.5 M NaOH and washed with distilled water until the pH was neutral. Then the mixture is centrifuged to obtain nanocellulose (NC-SB). The NC-SB was then dried at 60^{o}C until constant weight (Xu et al., 2019).

Characterization of material

Lignocellulose analysis is used to determine the levels of lignin, cellulose, and hemicellulose in C-SB. This lignocellulose analysis used the Chesson Method (1981). Nanocellulose characterization was carried out to see the composition, morphology, particle size, fuctional group, and the degree of crystallinity. Degree of crystallinity were analyzed from XRD data. The degree of crystallinity is determined through equation (1), the XRD diffraction area is determined using the Origin pro software.

$$\% \text{ Crystallinity} = (\text{Area of crystalline} / \text{Total area}) \times 100\% \quad (1)$$

Particle size was analyzed using PSA, the chemical composition of SB, C-SB,an NC-SB that verifying the removal of lignin and hemicellulose during cellulose extraction process from sugarcane bagasse was characterized using FT-IR. Morfology and atomic composition was characterisized using SEM.

RESULT AND DISCUSSION

Nanocellulose extraction from Sugarcane Bagasse

Extraction of nanocellulose from bagasse consists of 3 stages, alkali treatment, bleaching, and acid hydrolysis. All these steps take place with the help of an ultrasonicator. Starting from raw material (SB), the first stage of extraction was alkali treatment or delignification using NaOH to remove lignin from bagasse. In this process, fiber development occurs so that the hemicellulose, mineral salts, and ash are lost, and produce brownish yellow pulp (Sheltami et al., 2012). The second stage of extraction was a bleaching treatment using 0,7% NaOCl to obtain white cellulose. The third stage of extraction was acid hydrolysis using 45% H_2SO_4. Strong acids can remove the amorphous part of a cellulose chain so that isolation of the crystalline part of

cellulose can be carried out (Effendi et al., 2015). Figure 1 is a picture at different stages of treatment. The color of material changed from brownish yellow (Fig.1a) to light brown (Fig.1b) after alkali treatment and became white after bleaching treatment (Fig.1c). After hydrolysis stage, the colour of material changed form white to brown (Fig.1d). NC-SB produced from 60 grams of bagasse is 6,890 grams, so the yield of nanocellulose isolation from bagasse is 11.483%.

Fig 1. a) Sugarcane bagasse (SB); b) SB after alkali treatment; c) Celullose (C-SB); d) Nanocellulose (NC-SB)

Analysis of Lignoselulose and % moisture content

Analysis of lignocellulose content aims to determine the levels of cellulose, hemicellulose, and lignin in C-SB. The Chesson-data test results showed that the cellulose content in C-SB was 51.27%, the lignin content was 22.33%, the hemicellulose content was 8.49%. The moisture content of C-SB and NC-SB was found to be 1,47% and 3,85%. The moisture content increase slightly after acid hydrolysis treatment. This is due to the existence of three free -OH groups in cellulose that can enchance the rate of moisture absorption (Cherian et al., 2010).

SEM and particle size analysis

Morphological analysis was carried out by SEM and Fig 2 is micrograph of sugarcane bagasse before and after treatment. The surface of SB was smoother than NC-SB. This is due to the existence of the outer non-cellulosic layer on SB, such us pectin, lignin, wax, and hemicellulose, which is acted as cementing material to holds the fibers in bundles (Fig 2a). After ultrasonicating in the presence of H_2SO_4 and drying process, the samples tended to be self assembled into fibrilated fiber (Fig 2b). The dimesion on fiber was decrease because of the removal of the cellulose amorphous region. the NC-SB surface was eroded by acid hydrolysis. Moreover, the erotion of NC-SB may be caused by the emission of heat and exited pecies during ultrasonicating. Ultrasonication in the presence of acid and ionic liquid medium hydrolyzed the amorphous region of cellulose up to certain extent as well as some portion of cellulosic fragment were completely broken to yield soluble oligo- and mono-saccharides (Chowdhury & Hamid, 2016). The average particle size of NC-SB was found to be 132,6 nm. The average length of sample was 205,1 nm while the average width was 42 nm.

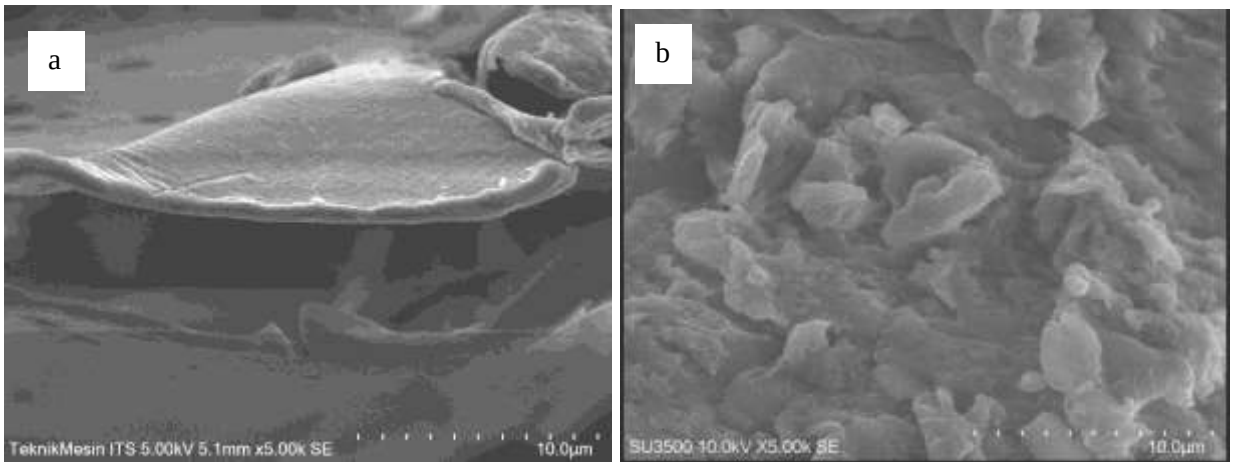

Figure 2. a) Sugarcane Bagasse (SB); b) Nanocellulose (NC-SB)

Analysis FT-IR

FT-IR analysis aims to see the functional groups at each stage of nanocellulose extraction. Changes in infrared absorption at each stage of the extraction treatment indicated a change in the composition of the bagasse cellulose. Figure 3 shows the FTIR spectra obtained for sugarcane bagasse at different stages of treatment. The FTIR band at 1742 cm-1 in SB is the vibration of the carbonyl group (C = O) stretching of the acetyl and uronic acid ester groups, from pectin, hemicellulose, or the ester linkage of carboxylic group of ferulic and p-coumaric acid of lignin or hemicellulose (Alemdar & Sain, 2008; Sun et al., 2005). The FTIR band at 1238 cm^{-1} as present in SB is indicated the C-O out of plane stretching vibration of aryl group in lignin (Le Troedec et al., 2008). The faintly absorption band at 1514 cm^{-1} are associated with aromatic C=C in plane symetrical stretching vibration of aromatic ring present in lignin (Wang et al., 2009). These three band are only found in bagasse before treatment (SB), this indicates that there is a delignification process at the alkaline treatment stage.

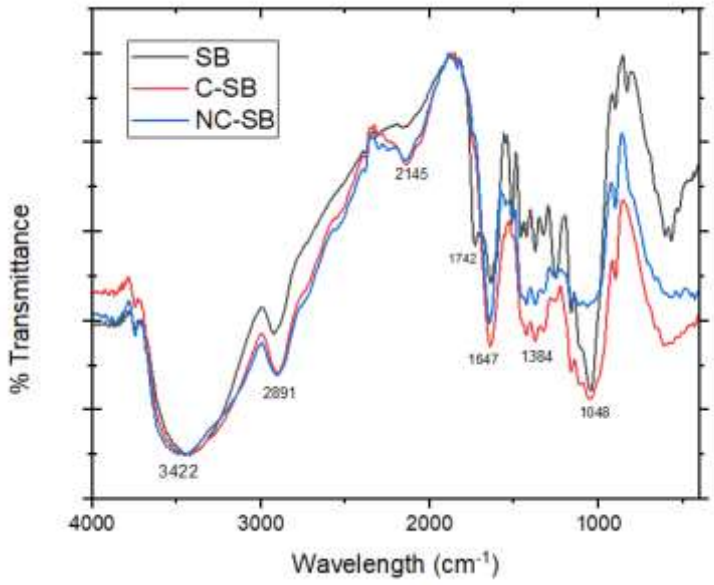

Figure 3. FT-IR Spectra of sugarcane bagasse (SB), Cellulose (C-SB), Nanocellulose (NC-SB)

Overall, The FTIR spectrum of C-SB dan NC-SB have similar peak in all wave number. The differences between both of FTIR spectrum was slightly intensity changes in the peaks. All of the spectra have the broad band in the region of 3300 cm^{-1} to 3400 cm^{-1} that indicates the O-H stretching vibration of OH group in cellulose. Moreover, the absorbance band around 2900 cm-1 indicated C-H stretching and the absorbance peak in region between 1630 cm^{-1} to 1650 cm^{-1} reflect the O-H bending of absorbed water (Alemdar & Sain, 2008; Le Troedec et al., 2008). The band around 1057 cm^{-1} reflect the C-O-C pyranose ring skeletal vibration and the increase in intensity of this peak showed an increase in crystallinity of the samples (Elanthikkal et al., 2010). The band around 895 cm^{-1} reflect the β-glycosidic linkage between the anhydroglucose units in cellulose (Alemdar & Sain, 2008).

XRD Analysis

Figure 4 shows the XRD patterns for sugarcane bagasse at different stages of treatment. All of XRD patterns showed peaks around 2θ of 22° indicating the typical cellulose I structure. The only difference is slight intensity change in the peaks that indicating some change in the crystallinity index of samples (Subramanian et al., 2005). The most defined peak at 22° is NC-SB peak, while C-SB peak at 22° more defined than SB peak at It can be seen in NC-SB peak at 22° were more defined than SB peak. This is indicated that acid hydrolisis and ultrasonis wave can increase cristanillity index of cellulose. During the acid hydrolisis, hemicelluloses and lignin were dissolved and the remaining pure of crystallin were isolated. The particle can increases peak intensity and give narrower crystalline peaks (Rosli et al., 2013). Ultrasonication in presence of ionic liquid and acid hydrolizing medium effectively dissolved lignin and hemicellulose from SB (Chowdhury & Hamid, 2016). % Cristallinity for SB, C-SB, and NC-SB was found to be 68%; 77,2%; 92,62%.

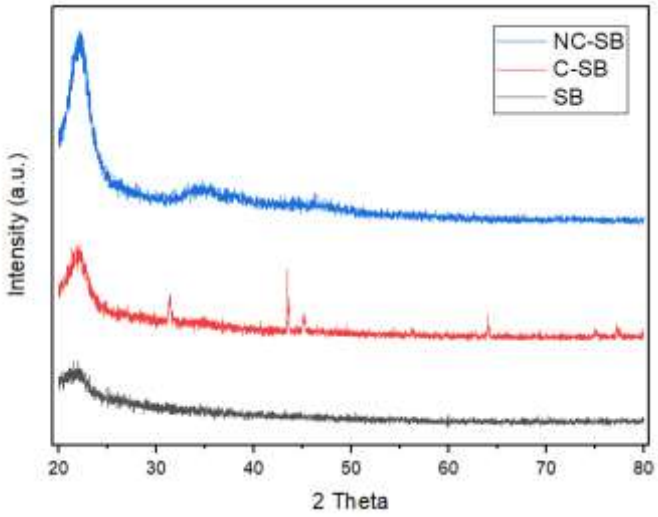

Figure 4. Diffractogram SB, C-SB, and NC-SB

CONCLUSION

Based on the result of this research, it can be concluded that the nanocellulose with high % crystallinity was succesfully extracted from sugarcane bagasse using a ultrasonication-chemical combination method. The FTIR spectrum showed the broad band at 3300-3500 cm^{-1} that foud to be the vibration of cellulose OH group. The band around 895 cm^{-1} reflect the β-glycosidic linkage between the anhydroglucose units in cellulose. % crystallinity of nanocellulose reached up to 92,62%. The average particle size of nanocellulose was found to be 132,6 nm that consisting of the average length of sample was 205,1 nm while the average width was 42 nm.

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