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Isolation and Characterization of Cellulose Whiskers from Lampung Sugarcane Bagasse, Indonesia

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Abstract: Sugarcane bagasse, the fibrous residue from sugar production, is an abundant agricultural waste in Indonesia, especially in Lampung, one of the country's leading sugarcane-producing provinces. Its high lignocellulosic content makes it a promising alternative source of cellulose. However, effective extraction and conversion into high-value products such as cellulose whiskers require optimized chemical processes. This study aims to isolate cellulose from sugarcane bagasse using alkali and peroxide treatments and convert it into cellulose whiskers through hydrolysis using sulfuric acid (H_2SO_4) at varying concentrations (4–12 M). The objective is to evaluate the optimal acid concentration that produces cellulose whiskers with desirable morphology and crystallinity. Cellulose isolation was achieved through sequential NaOH and H_2O_2 treatments, significantly reducing lignin and hemicellulose content and yielding α-cellulose at 88.37%. Hydrolysis of the purified cellulose was then performed with H_2SO_4. The resulting materials were characterized using FTIR, SEM, XRD, and TGA. FTIR confirmed the removal of non-cellulosic components, while SEM showed that only 10 M H_2SO_4 produced well-defined whiskers with nanoscale dimensions (200–700 nm in length and 10–50 nm in diameter). Lower acid concentrations resulted in incomplete hydrolysis, while excessive degradation occurred at 12 M. XRD analysis revealed an increase in crystallinity to 86.7%, indicating the removal of amorphous regions and successful formation of crystalline whiskers. TGA analysis showed different thermal degradation patterns between isolated cellulose and whiskers, supporting structural transformation. Cellulose whiskers can subsequently be utilized as a reinforcing material in the fabrication of plastic films for water–oil separation and dye adsorption applications in laboratory experiments.

Keywords: Acid Hydrolisis; Cellulose Isolation; Cellulose Whiskers; Sugarcane Bagasse; Sugarcane Waste.

Introduction

In 2020, Sugar factories in Indonesia produced 2190 million tons of sugarcane. Indonesia has five provinces that produce the most sugarcane, namely East Java, Lampung, Central Java, South Sumatra, and South Sulawesi [1]. Lampung is the second-largest producer of sugarcane after East Java, which produced 732.14 thousand tons of sugarcane in 2020 [2]. The increasing production of sugar cane will produce a large amount of sugarcane bagasse. Sugarcane bagasse is a by-product of the extraction process of sugarcane [3]. Sugarcane bagasse content ranges from 25-30% of the total weight of processed sugarcane [4].

In the sugar industry, Sugarcane bagasse is a solid waste that has an impact on environmental pollution; therefore, it is necessary to utilize sugarcane bagasse as an alternative source of cellulose [5]. Fiber from sugarcane bagasse is not soluble in water, and the composition of sugarcane bagasse fiber consists of 45% cellulose, 22% hemicellulose, and 21.7% lignin based on the dry weight [6], [7]. Cellulose can be extracted using alkali treatment to reduce the lignin content, thereby increasing the purity of the cellulose [8]. Cellulose from sugarcane bagasse can be hydrolyzed using acid, which causes the cellulose microfibers to break down in the amorphous part and leave the crystalline part, called cellulose whiskers [9].

Whiskers are a form of single crystals with higher purity that can be formed from nanofibers. The pure crystals of whiskers have high levels of rigidity, surface area, and crystallinity [10]. Due to this characteristic, cellulose whiskers are very well applied in the polymer matrix and as a reinforcement material, such as films for the absorption of methylene blue in wastewater and for the separation of water and oil [11], [12]. The characteristics of cellulose whiskers depend on the cellulose source and conditions of hydrolysis, such as time, temperature, concentration, and purity of the cellulose [13]. The type of acid used during the hydrolysis process plays a crucial role in determining the surface characteristics of cellulose whiskers. Hydrolysis with HCl typically results in whiskers with low colloidal stability, making them more susceptible to particle size variation and aggregation. On the other hand, hydrolysis using H_2SO_4 leads to surface sulfation on certain regions, which introduces negative charges that can enhance dispersion stability in aqueous media [14], [15]. Previous studies have utilized H_2SO_4 for the isolation of cellulose whiskers from sugarcane bagasse originating from Tasikmalaya, West Java. Nevertheless, there has been no investigation into the influence of different H_2SO_4 concentrations or the utilization of sugarcane bagasse from Lampung as a raw material for resulting cellulose whiskers [16].

In this study, cellulose was sourced from sugarcane bagasse taken from the waste of the PT. Bungamayang sugar

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factory, Lampung, Indonesia. Cellulose was extracted from sugarcane bagasse using alkali treatment, and cellulose was converted to cellulose whisker form through hydrolysis using sulfuric acid (H_2SO_4) with various concentrations. The results of the cellulose whisker were extensively characterized through Fourier Transform Infrared Spectroscopy (FTIR), scanning electron microscope (SEM), X-ray Diffraction (XRD), and TGA.

Research Methods

Materials

The materials used in this study were sugarcane bagasse from a sugar factory in PT. Bungamayang, Lampung. NaOH, H_2O_2, and H_2SO_4, all sourced from Merck.

Isolation of Cellulose from Sugarcane Bagasse

Sugarcane bagasse was dried and ground to a 100 mesh size. Sugarcane bagasse powder was dissolved in 400 mL of distilled water with sonicated for 30 minutes. The mixture was filtered by washing three times, and the residue was dried at 50°C (A). A total of 5 g of sample was added to 95 mL of 4% NaOH, and it was heated to a temperature of 80 °C for 4 hours. The mixture was filtered under vacuum. The residue was washed with distilled water and dried at 50 °C to constant weight (B). A total of 20 g of sample B was added to 500 mL of 5% H2O2 and heated in a water bath at 70 °C for 3 hours. The mixture was filtered, and then the precipitate was washed with distilled water to neutral pH. The residue was dried in an oven at 60 °C until dry [17]. Isolation of cellulose from Sugarcane bagasse was repeated 3 times. The results of the isolation of cellulose were the determination of hemicellulose, cellulose, α-cellulose, lignin, and the analysis of functional groups by FTIR.

Hydrolysis of Cellulose to Cellulose Whiskers

A total of 5 g cellulose was dissolved in 100 mL of H_2SO_4 by variations in concentration of 4 M, 6 M, 8 M, 10 M, and 12 M at 45 °C, and stirred for 40 minutes. The mixture was added to 500 mL of cold distilled water and centrifuged at 12,000 rpm for 10 minutes. Dialysis used a dialysis membrane to reach pH 6-7. The results suspension was sonicated for 5 minutes [18]. Each treatment variation of H_2SO_4 concentration was repeated 3 times. Thermal analysis was done by TGA, whisker crystallinity was analyzed by XRD, and the morphology of the cellulose whiskers was analyzed by SEM.

Results and Discussion

Isolation of Cellulose from Sugarcane Bagasse

Treatment with alkali can break ester bonds between lignin and cellulose through a hydrolysis reaction. In the early step, isolation of cellulose used NaOH to break bonds between lignin and cellulose or hemicellulose (in Fig. 1) [19]. In these conditions, the presence of hydroxide ions leads to significant swelling of the cell walls of sugarcane bagasse, thereby promoting the removal of hemicellulose [8].

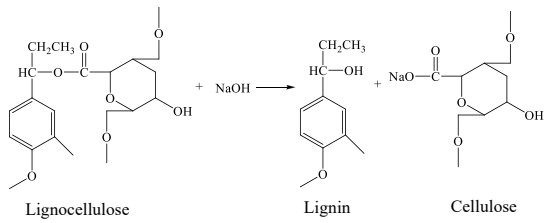

Figure 1. Mechanism of lignocellulose reacting with NaOH [19]

Analysis of the chemical contents of isolated cellulose from sugarcane bagasse was one indicator of the success of the isolation step. The success of isolation was characterized by an increase in the content of α-cellulose and a significant decrease in the content of lignin and hemicellulose, as shown in Table 1. The chemical contents of sugarcane bagasse at the beginning are 46.68% of α-cellulose, 23.72% of hemicellulose, 22.59% of lignin, and 8.05% of water. The results of this research show that the α-cellulose content of Lampung's sugarcane bagasse is higher than that from Gorontalo and East Java [20], [21]. The success of the delignification step in isolating cellulose was evaluated based on the reduction of lignin content in isolated cellulose from 22.59% to 0.19%. This is due to the lignin in the sample having been dissolved in a solution of NaOH; therefore, there is an increased purity in α-cellulose approaching 88.37%. This research achieved a higher α-cellulose content compared to alkali-treated sugarcane bagasse from Gorontalo, which contained 83.67% [21].

Table 1. The Chemical Content of Sugarcane Bagasse and Isolated Cellulose

Chemical Content	Sugarcane Bagasse (%)	Isolated Cellulose (%)
Water	8.05	4.15
Hemicellulose	23.72	5.38
α-cellulose	46.68	88.37
Lignin	22.59	0.19

FTIR characterization of sugarcane bagasse and isolated cellulose can be seen in Figure 2. The FTIR spectrum of sugarcane bagasse raw material shows absorption at 1508 cm-1, identifying the vibration of lignin carboxyl and lignin rings. Absorption for vibration of acetyl ester groups at wave number 1705 cm-1 and 1508 cm-1 (in Fig. 2.a), which is supported by lignin content [16]. After alkali treatment on sugarcane bagasse (in Fig. 2.b), the typical absorption for lignin compounds decreases and the band at wave numbers at 1103-1033 cm-1 becomes larger, indicating the C-O-C group found in the pyranose ring of cellulose and the appearance of a typical absorption for cellulose at 898 cm-1, which is quite sharp. This absorption indicates the formation of a β-glycoside bond between glucose compounds in cellulose [22]. The presence of absorption at wave numbers 1425-1317 cm-1 indicates the bending vibration of the C-H2 group, and absorption of 2885 cm-1 identifies the stretching vibration of the C-H group [20]. The stretching vibration of the O-H group is at an absorption of 3300 cm-1 and at a wave number of 1201 cm-1, it is predicted that the bending vibration of the O-H group is in the cellulose plane, the absorption of 1161 cm-1 indicates the stretching vibration of the C-O group [23].

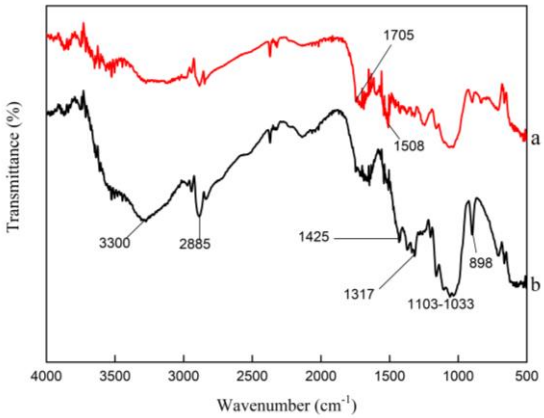

Figure 2. FTIR spectrum of sugarcane bagasse (a) and isolated cellulose (b)

Hydrolysis of Cellulose to Cellulose Whiskers

The results isolating cellulose were hydrolyzed with sulfuric acid concentration variants in Figure 3. Treatment with low acid concentrations did not change the morphology of cellulose fibers, indicating that the cellulose has not been hydrolyzed properly, it can be seen in Figure 3 (a, b, and c). The acid concentration of 10 M could cut the amorphous part, and the morphology of the fiber indicated the size in nanometers; the image is presented in Figure 3d. The H_2SO_4 could break the carbon chain of cellulose [24]; however, a higher acid concentration may excessively degrade the cellulose, resulting in carbon black formation, as shown in Fig. 3e. Morphology measurements of cellulose whiskers were performed using SEM.

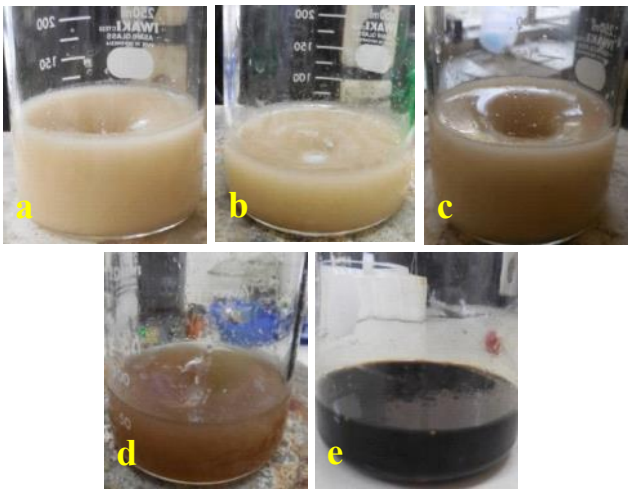

Figure 3. Hydrolysis of cellulose with various H_2SO_4 concentrations (a) 4 M, (b) 6 M, (c) 8 M, (d) 10 M, and (e) 12 M

A Scanning Electron Microscope (SEM) utilizes a high-energy electron beam to analyze materials with a very high level of precision. The principle of electronic scanning in SEM can provide various information on the nanoscale of a material [25]. The morphological observation of cellulose whiskers is presented in Figure 4. In the treatment with 4M H_2SO_4 concentration, cellulose fibers were not formed as in Figure 4.a. Meanwhile, at H_2SO_4 concentrations of 6 M and 8 M, the cellulose fibers started to form (Fig. 4.b & 4.c). The treatment with 10 M H_2SO_4 resulted in a well-defined and uniform morphology of cellulose fibers or whiskers (Figure 4.d), because the 10M H_2SO_4 was able to cut the amorphous part of the cellulose [26]. Table 2 shows that the cellulose fiber diameter decreases with increasing acid concentration. The results obtained show that hydrolysis using a concentration of 10 M H_2SO_4 can remove the amorphous part of the cellulose fibers; as a result, the form received is worthy of being called whiskers [9]. The whisker size obtained at the 10 M H_2SO_4 concentration indicates the successful hydrolysis of cellulose into cellulose whiskers. Table 2 presents a summary of the dimensions and properties of cellulose whiskers obtained from various sources.

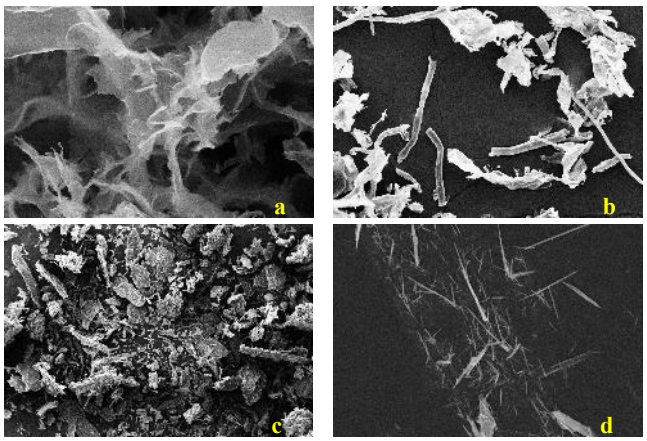

Figure 4. Morphology of cellulose whiskers (a) H_2SO_4 4M, (b) H_2SO_4 6M, (c) H_2SO_4 8M, and (d) H_2SO_4 10M

Table 2. The length and diameter of the fibers of cellulose whiskers

H_2SO_4 Concentration	Length	Diameter
4M	> 100 µm	>20 µm
6M	45–200 µm	5 – 18 µm
8M	30–80 µm	4 – 10 µm
10M	200–700 nm	10 – 50 nm

Cellulose consists of two parts, namely amorphous and crystalline. The amorphous part will be more easily hydrolyzed, leaving the crystalline part. The chemical process begins with the removal of bonds between polysaccharides on the surface of cellulose fibers, followed by the breaking and destruction of the amorphous parts, which subsequently releases the crystalline parts of cellulose [26]. Evaluation of the success of the hydrolysis of cellulose whiskers can be seen from the XRD pattern in Figure 5. The diffractogram of cellulose whiskers shows two main peaks at $2\theta = 12.2°$, d = 7.15 Å, and $2\theta = 22.04°$, d = 4.427, the crystal structure obtained [16]. This is consistent with the study conducted by Wu et al. (2020) using corn stalks, in which the diffractogram of cellulose whiskers exhibited two peaks at $2\theta = 12°$ and $2\theta = 20°$ [27].

The degree of crystallinity can be calculated using the method proposed by Segal *et al.* (1959) [28], which is as follows :

$$\% \text{ Crystallinity} : \frac{I_{Crystalline}}{I_{Crystalline} + I_{amorphous}} \times 100\%$$

The degree of crystallinity of cellulose whiskers obtained in this research was 86.7%, an increase of 15% from the crystallinity of sugarcane bagasse cellulose isolate. As shown in Table 2, the degree of crystallinity of cellulose whiskers obtained from Lampung sugarcane bagasse is comparatively higher than that extracted from cocoa pod husk, Acacia caesia, and date palm fibre. Nevertheless, it remains lower than the crystallinity observed in cellulose whiskers derived from waste jute fiber.

Table 3. Dimensions and properties of cellulose whiskers from various sources

Source	Length (nm)	Diameter (nm)	Crystallinity (%)	Maximum degradation Temp. (°C)	References
Lampung's Sugarcane bagasse	200-700	10-50	86.7	324.6	This study
Cocoa pod husk	95 (average)	10–60 (26 average)	67.6	332	[29]
Waste Jute Fiber	800 ± 100 nm	55 ± 10 nm	90.91	260	[30]
Acacia caesia	300	30	79.65	300	[31]
Date palm fibre	146.53	8.94	84.2	322	[32]

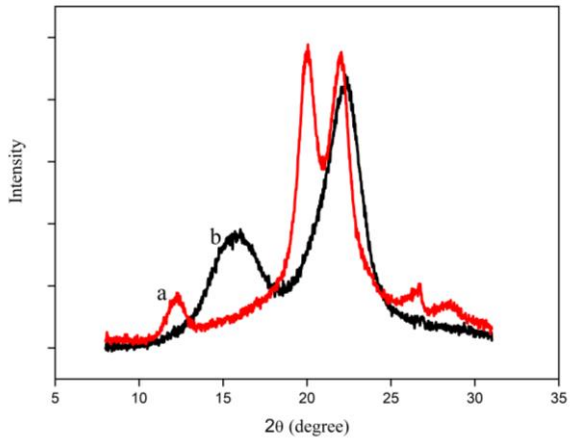

Figure 5. XRD pattern of (a) cellulose whisker and (b) isolated cellulose

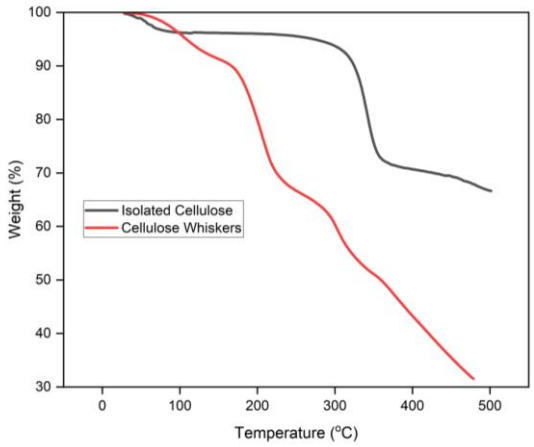

Figure 6. TGA curves of cellulose whisker and isolated cellulose

Thermogravimetric Analysis (TGA) is an analytical technique used to determine the thermal stability of a material and the fraction of volatile components by measuring the weight changes associated with temperature variations [33]. As shown in Figure 6, the TGA profiles of sugarcane bagasse cellulose isolate and cellulose whiskers showed different thermal degradation patterns. The cellulose isolate showed an initial weight loss of 10% between 27 and 305°C, which was due to the decomposition of residual hemicellulose. A sharp weight loss occurred in the second stage up to 374°C, indicating the depolymerization of cellulose, with the overall mass loss reaching about 80%. The final stage of mass reduction by 6% up to a temperature of 500°C is suspected of further damage to cellulose, forming carbon residue. Cellulose isolate degraded at a temperature of 344°C. The degradation temperature of cellulose whiskers starts at 200.4 °C and reaches a maximum of 324.6°C. The degradation temperature in other studies using sugarcane bagasse as a standard shows that nanocellulose begins to degrade at 275°C, indicating that the crystal structure has been damaged [34]. An increased thermal degradation temperature is associated with improved mechanical properties of plastic films. Consequently, cellulose whiskers exhibit significant potential as biocomposite reinforcements in the development of plastic films [35].

Conclusion

In this study, alkali treatment for cellulose isolated from Lampung's sugarcane bagasse successfully obtained a high content of α-cellulose and a low content of lignin, namely 88.37% and 0.19%, respectively. Cellulose was effectively hydrolyzed using different concentrations of H_2SO_4 to produce cellulose whiskers. The best concentration of 10M H_2SO_4 for producing cellulose whiskers with a length of 200 – 700 nm and a diameter of 10 – 50 nm. The results from SEM, XRD, and TGA analyses consistently indicate the successful conversion of cellulose into cellulose whiskers. Further research can utilize cellulose whiskers derived from Lampung's sugarcane bagasse for the development of biodegradable and biocomposite reinforcements of plastic films.

Author's Contribution

Muhammad Ridho Afifi: conceived the study. wrote the manuscript and analyzed data. Zahratul Aini: conceived the study and conducted the experiments. Tun Tedja Irawadi: contribution to this research is as a supervisor, and providing direction. Henny Purwaningsih: analyzed and interpreted the data.

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