PREPARATION AND CHARACTERIZATION OF CELLULOSE NANOFIBRES FROM PINEAPPLE LEAF FIBRES AND SUGARCANE BAGASSE

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Abstract

The aim of the study was to extract cellulose nanofibres (CNF) from pineapple leaf fibres (PALF) and sugarcane bagasse (SCB). The CNF was isolated from PALF and SCB by mechanical treatments and chemical treatments using alkaline, acids and inorganic salts. The physicochemical properties of prepared samples such as moisture content, ash content, pH and solubility were determined conventionally. Yield percent of pineapple leaf cellulose nanofibres (PALCNF) was higher than that of sugarcane bagasse cellulose nanofibres (SCBCNF). The raw samples and prepared samples were characterized by XRD, FT IR, EDXRF and SEM analysis to confirm as cellulose nanofibres. Crystallinity index percent of the prepared cellulose nanofibres was investigated by using XRD analysis. According to the XRD investigation, PALCNF and SCBCNF have the sharp diffraction peak is 20 value at 22° and the amorphous diffraction peak is 20 value at 18°. It could be noticed that cellulose was present in the form of cellulose. Based on the FT IR spectrum, there are several peaks in raw samples which are not found in the spectrum of cellulose nanofibres. The EDXRF table shows the elemental constituents of the untreated raw samples and the CNF after chemical treatment. The SEM analysis showed that the PALCNF has fibrous nature and the SCBCNF has irregular aggregated shape fibrils.

Keywords: Cellulose nanofibres, pineapple leaf cellulose nanofibres, sugarcane bagasse cellulose nanofibres, chemical treatment

Introduction

Cellulose is an organic compound with the formula $(C_6H_{10}O_5)_n$, a polysaccharide consisting of a linear chain of several hundred to many thousands of $\beta(1 \rightarrow 4)$ linked D-glucose unit (Lavanya *et al.*, 2011). Cellulose is the most abundant polymer on Earth. It consists of glucose-glucose linkages arranged in linear chains where C-1 of every glucose unit is bonded to C-4 of the next glucose molecule. Cellulose in nanometers or nanocellulose has a size range from 10 nm to 350 nm (Kadla and Gilbert, 2000).

Cellulose, an important structural component of plants, have received much attention because of their low density, nonabrasive, combustible, nontoxic, low cost and biodegradable properties. Cellulose fibres have some disadvantages such as moisture absorption, quality variations, low thermal stability and compatibility with the hydrophobic polymer matrix. However, production of nanoscale cellulose fibers and their application in composite materials have gained increasing attention due to their high strength and stiffness combined with low weight, biodegradability and renewability.

The cellulose fibres can be classified into three groups. They are following.

- (i) Leaf fibre : abaca, cantala, curaua, date palm, henequen, pineapple, sisal, banana;
- (ii) Seed fibre : cotton; bast: flax, hemp, jute, ramie; fruit: coir,kapok, oil palm;
- (iii) Grass fibre : alfa, bagasse, bamboo; stalk: straw (cereal).

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The bast and leaf (the hard fibers) types are the most commonly used in composite applications. Commonly use plant fibres include cotton, jute, hemp, flax, ramie, sisal, coir, henequen and kapok. Cotton contains much of cellulose in it (around 90%), how in wood, plant leaves and stalks, it is found in combination with other materials, such as lignin and hemicelluloses (Chandrahasa *et al.*, 2008).

Pineapple leaf fibres (PALF) are an important natural fiber that exhibit high specific strength and stiffness. The PALF observed to have the high percentage of α -cellulose content (81.27 %) and low percentage of hemicellulose (12.31 %) and lignin (3.46 %) content. The α -cellulose is purified with steam treatment correlated with acid treatment processes (Cherian *et al.*, 2011).

Sugarcane bagasse (SCB) contains cellulose (40-50 %), much of which is in the crystalline structure. Another component in sugarcane bagasse is hemicellulose as much as (25-35 %) which is amorphous polymer and mainly composed of xylose, arabinose, galactose, and mannose. The rest is mostly lignin with about (18-24 %) (Wulandari *et al.*, 2016).

There are two basic approaches for creating nanostructurs – bottom-up method and top down method. The bottom up method involves construction on a molecular scale from scratch using atoms, molecules and nanoparticles building blocks. The top down method involves the disintegration of macroscopic material to a nanoscale by the following methods: mechanical (e.g., grinding), chemical (e.g., partial hydrolysis with acids or bases), enzymatic (e.g., treatment with enzymes hydrolyzing cellulose, hemicellulose, pectin and lignin) and physical (e.g., techniques using focused ion beams or high–power lasers). In this paper, the top down method was used for production of cellulose nanofibres.

The top down approach deals mainly with the removal of non-cellulosic compounds to obtain pure cellulose and then mechanically treating it to get cellulose nanofibres. The treatment basically involves alkaline hydrolysis to remove pectin and lignin followed by bleaching to get rid of hemicellulose and lastly acid hydrolysis to remove mineral traces and to hydrolyze amorphous cellulose, providing the required nanofibres (Nohwar *et al.*, 2016).

Materials and Methods

Sample Collection

Pineapple leaf was collected from Shaw Pyar Village, Pathein Township and sugarcane bagasse was collected from Hledan Market. Other requiring chemicals were purchased from chemical store. Distilled water was used as the solvent in all analyses.

Extraction of Pineapple Leaf Fibres

Pineapple leaf fibres from the leaves can be done in two ways, namely the manual and mechanical methods. The most common and effective is the manual method. Firstly, pineapple leaves were washed with water. The manual process used a plate or whittle with no sharp knife to remove the skin leaves still attached to the fibres surface. After that, the fibres were washed with water and dried in sunlight or using the oven. Dry fibres were cut into small pieces (Adam *et al.*, 2016).

Preparation of Cellulose Nanofibres from Pineapple Leaf Fibres

PALF were treated with 2 % (w/v) NaOH (fibres to liquor ratio1:10) in an autoclave and kept under 138 kPa pressure for a further period of 1 hour. Pressure was released immediately. The fibres were removed from the autoclave, and the fibres were washed in distilled water until it was rid of alkali. The steam exploded fibers were bleached using a mixture of NaOH and glacial CH₃COOH (27 g/L and 78.8 g/L, respectively) and a mixture of 1:3 NaClO solutions. The bleaching was repeated six times. After the bleaching the fibres were thoroughly washed in distilled water and dried. The steam exploded bleached fibres were treated with $H_2C_2O_4$ of 11 % (w/v) concentration in an autoclave until it attained a pressure of 138 kPa. The pressure was released immediately. The autoclave was again set to reach 138 kPa and the fibres were kept under that pressure for 15 min. The pressure was released and the process repeated eight times. The fibres were taken out washed until the washings no longer decolorized KMnO₄ solution to make sure that the washings are free from acid. The proceeded nanofibrils were suspended in distilled water and kept stirring with a mechanical stirrer for about 4 h until the fibres are dispersed uniformly (Cherian *et al.*, 2011).

Preparation of Cellulose Nanofibres from Sugarcane Bagasse

Sugarcane bagasse was dried in sunlight and then cut into small pieces. The cut bagasse was milled to become powder. The powder of bagasse was bleached with sodium hypochlorite for 6 h with constant stirring at 45 °C to remove the lignin. The residue was washed with distilled water until a neutral pH. The neutral residue was refluxed with 17.5 % sodium hydroxide for 3 h with constant stirring at 45 °C to remove hemicellulose. The residue of this process was also washed until reach a neutral pH, and it was dried at room temperature for 2-3 days. Isolated cellulose from sugarcane bagasse was hydrolyzed with sulfuric acid with a ratio of cellulose to sulfuric acid 1:25. The hydrolysis of cellulose with 10 % (v/v) H₂SO₄ at 40 °C for 10 min. The hydrolysis process was quenched by adding 10-fold excess distilled water (250 mL) to the reaction mixture. A colloidal suspension which produced was centrifuged at 6500 rpm for 30 minutes. Then, it was dialyzed for 5 days to neutralize and eliminate the sulfate ions. The neutral colloidal suspension was sonicated for 10 minutes to homogenize the generated cellulose nanofibres (Wulandari *et al.*, 2016).

Characterization of the Prepared Samples

The physicochemical properties (moisture, ash, pH and solubility) of cellulose nanofibres prepared from pineapple leaf fibres and sugarcane bagasse were determined. The crystallinity index was calculated by using XRD analysis. The structural characterization of PALCNF and SCBCNF were characterized using FT IR. The elemental constituents in samples were analyzed by EDXRF analysis. The morphological structure of prepared samples were characterized by SEM.

X-ray diffraction (XRD) analysis was carried out using Rigaku X-ray Diffractometer, RINI 2000/PC software, Cat. No 9240 J 101, Japan. Copper tube with nickel filter was used. The diffraction pattern was recorded in terms of 2 θ in the range of 10-70 °.

FT IR spectrum was recorded in the range of 4000-400 cm⁻¹ by using 8400 SHIMADZU, Japan FT IR spectrophotometer.

Elemental EDXRF analysis on the prepared sample was done by using Shimadzu model EDX-800 EDXRF spectrometer.

The scanning electron microscopy (SEM) images were recorded by using JSM-5610 Model SEM, JEOL-Ltd., Japan.

Results and Discussion

Physicochemical Properties of PALCNF and SCBCNF

Table 1 shows the physicochemical properties (moisture, ash, pH and solubility) of cellulose nanofibres obtained from pineapple leaf fibres and sugarcane bagasse. In this table, PALCNF was a significant higher content of moisture, ash and yield percent than that of SCBCNF. The moisture content of cellulose was within the acceptable range (10 - 13.5 %) for high quality cellulose and standards for food application. The ash content is greater than 0.2 % which is recommended for high quality cellulose. The pH value falls within the acceptable range (6-7) for cellulose used in the pharmaceutical, cosmetics and food industries. Cellulose has strong affinity and hydrogen bonding through the Vander Wal's force itself; hence it is very stable and insoluble in water.

Physicochemical Properties	PALCNF	SCBCNF
moisture (%)	10.11	9.84
ash (%)	0.53	0.01
pН	6.5	6.8
solubility	Insoluble	Insoluble
yield percent (%)	40.26	9.6

Table 1 Physicochemical Properties of PALCNF and SCBCNF

PALCNF = Pineapple leaf cellulose nanofibres

SCBCNF = Sugarcane bagasse cellulose nanofibres

XRD Analysis

XRD analysis was conducted to analyze the crystallinity of raw samples and preparation of samples. The crystallinity index percent (C_I) was calculated using the following equation, by measuring the peak height of the crystalline region (I_{200}) and the amorphous region (I_{am}).

$$C_{I} (\%) = \frac{I_{200} - I_{am}}{I_{200}} \times 100 \%$$

 I_{200} is the maximum intensity of the peak for the crystalline cellulose ($2\theta = 22^{\circ}-24^{\circ}$). I_{am} represent the intensity of diffraction of the noncrystalline material for the amorphous cellulose ($2\theta = 16^{\circ}-18^{\circ}$). Figures 1(a) and (b) show XRD diffractograms of the raw PALF and PALCNF. Figures 2 (a) and (b) show XRD diffractograms of the raw SCB and SCBCNF. From the graph it can be seen that the raw samples are amorphous in nature and after the pretreatment crystallinity in the material is achieved, as also with acid hydrolysis. The amorphous nature of raw samples is due to the presence of large amounts of lignin, hemicellulose and other. After acid hydrolysis, the structural changes in crystallinity.

The crystallinity index percent for all samples was calculated by peak height method and represented as shown in Table 2. From the XRD result, the crystallinity index of the PALF was calculated as 75.12 % and increased in case of PALCNF to 89.05 %. Similarly, the increase the crystallinity index percent from 42.5 % for the SCB to 65.28 % for the SCBCNF. In both samples, it was found that the increase the value of C₁ from raw sample to prepared CNF was undoubtedly attributed to the removal of hemicellulose and lignin in amorphous region and remaining amorphous parts was removed during acid hydrolysis.



Figure 1 XRD diffractograms of

(a) raw pineapple leaf fibres PALF(b) pineapple leaf cellulose nanofibres PALCNF



Figure 2 XRD diffractograms of (a) raw sugarcane bagasse SCB (b) sugarcane bagasse cellulose nanofibres SCBCNF

Table 2 Crystallinity Index Percent of the Raw Samples and Prepared Cellulose Nanofibres

Samples	Crystallinity Index (%)
PALF (raw)	75.12
PALCNF	89.05
SCB (raw)	42.5
SCBCNF	65.28

FT IR Analysis

FT IR spectroscopy was used to show that the lignin and hemicellulose have been removed during the preparation of cellulose fibres through analysis of its functional group. Figures 3 (a) and (b) present the result of the FT IR analysis of raw PALF and chemical treated PALCNF. Figures 4 (a) and (b) also present the FT IR spectra of raw SCB and SCBCNF. Based on the FT IR spectra, there are several peaks in the raw samples which were not found in the spectra of cellulose nanofibres. The peaks are 1244 cm⁻¹, 1248 cm⁻¹, 1514 cm⁻¹, 1587 cm⁻¹, 1729 cm⁻¹ and 1730 cm⁻¹. The absorption peaks of 1244 cm⁻¹ and 1248 cm⁻¹ appeared due to the C-O stretching vibration of aryl group in lignin. The spectrum presented characteristic peaks in the 1514 cm⁻¹ and 1587 cm⁻¹ corresponding to the aromatic skeletal vibration. The C=O stretching vibration of carboxylic groups of hemicellulose and lignin is around 1729 cm⁻¹ and 1730 cm⁻¹. In CNF, the spectral bands of 1428 cm⁻¹, 1426 cm⁻¹ and 897 cm⁻¹ show significantly the presence of cellulose. PALCNF and SCBCNF have the same functional groups. The band assignments of raw samples and chemical treatment of CNF are described in Table 3.



(b) pineapple leaf cellulose nanofibres PALCNF



Figure 4 FT IR spectrum of (a) raw sugarcane bagasse SCB (b) sugarcane bagasse cellulose nanofibres SCBCNF

Observed wavenumber (cm ⁻¹)			cm ⁻¹)	* Literature	Band Assignment
Raw PALF	PALCNF	Raw SCB	SCBCNF	wavenumber (cm ⁻¹)	
3431	3334	3335	3337	3600-3200	O-H stretching
2918	2890	2940	2894	2980-2850	C-H stretching (ketone and carbonyl)
1729	-	1730	-	1765-1715	C=O stretching of ester
1641	1638	-	1635	1665-1620	O-H bending
1514	-	1587	-	1600-1500	C=C stretching (aromatic ring in lignin)
1427	1428	1455	1426	1430-1420	CH ₂ scissoring motion in cellulose
1314	1315	-	1314	1310-1250	C-O-C stretching (antisymmetric)
1244	-	1248	-	1310-1210	C-O stretching (aryl group in lignin)
1158	1160	1148	1158	1160-1000	C-O-C stretching (symmetric)
1031	1054	1033	1027	1050-1000	C-O stretching in cyclic alcohol
897	897	818	897	937-897	β (1-4) glycosidic linkage between the glucose unit in cellulose

 Table 3
 FT IR Band Assignments of the Raw Samples and the Prepared Cellulose Nanofibres

* Silverstein et al., 2003

EDXRF Analysis

Energy dispersive X-ray fluorescence was used for the elemental analysis of raw samples and prepared CNF from PALF and SCB. Figures 5 (a) and (b) show the EDXRF spectra of raw PALF and PALCNF. Figures 6 (a) and (b) also show the raw SCB and SCBCNF. In both raw samples contain many amount of metals. The CNF have the main costituents of (COH) and impurity of sulphur and other the trace metals. Other smaller peaks are Al, Si, K, Ca, Fe, Cu and Zn. They are negligible contents. This elemental impurity is due to the acid (H_2SO_4) hydrolysis of cellulose fibres and remaining after dialysis of CNF having sulphate group to some extent. The elemental constituents in raw samples and prepared CNF are described in Table 4.



Figure 5 EDXRF spectra of (a) raw pineapple leaf fibres PALF (b) pineapple leaf cellulose nanofibres PALCNF



Figure 6 EDXRF spectra of(a) raw sugarcane bagasse SCB

(b) sugarcane bagasse cellulose nanofibers SCBCNF

Table	4	Relative	Abundance	of	Elements	in	Raw	Samples	and	Prepared	Cellulose
Nanofibres from Pineapple Leaf Fibres and Sugarcane Bagasse											

	Relative Abundance (%) of Elements in Raw Samples and Cellulose Nanofibres								
Elements	Raw PALF	PALCNF	Raw SCB	SCBCNF					
Al	1.190	-	1.239	1.215					
Si	-	-	0.528	0.526					
Κ	0.059	-	0.103	0.018					
S	0.066	0.68	0.080	0.064					
Ca	0.135	0.322	0.044	-					
Fe	0.004	0.002	0.005	-					
Mn	0.003	-	0.002	-					
Cr	-	-	0.001	-					
Cu	0.001	0.001	0.001	-					
Zn	0.001	0.001	0.001	-					
НО	0.002	-	-	-					
Ti	0.002	-	-	-					
СОН	98.537	99.606	97.996	98.057					

SEM Analysis

In order to further investigate the structural changes in the fibres, SEM micrographs of the PALF and PALCNF are shown in Figures 7 (a) and (b). According to the SEM image, it is clear that the average diameter of PALCNF is lower than that of PALF. Because PALCNF is the removal of hemicellulose, lignin and pectin after chemical treatment and then to convert nanocellulose after acid hydrolysis. They have fibrous nature.

Figures 8 (a) and (b) also show SEM micrographs of the SCB and SCBCNF. The diameter of the original sugarcane bagasse fibre was much bigger than that of the chemical treatment sample. On treating with the bleaching agent the lignin is removed through complex formation. On subsequent treatment with alkali the hemicellulose is hydrolyzed and becomes water soluble. Finally, the sulphuric acid hydrolysis usually could cleavage the remaining amorphous regions of the cellulose.





(b)





(0)

Figure 8 SEM micrographs of (a) raw sugarcane bagasse SCB (b) sugarcane bagasse cellulose nanofibres SCBCNF

Conclusion

In this study, cellulose nanofibres can be obtained from prepared cellulose from pineapple leaf fibres and sugarcane bagasse followed by acid hydrolysis. The extracted of CNF and raw samples are investigated to determine the physicochemical properties (moisture, ash, pH and solubility) and the spectrophotometric properties (XRD, FT IR, EDXRF and SEM). The pH values of PALF and SCBCNF to be 6.5 and 6.6 respectively, were found. The pH value falls within the acceptable range (6–7) for cellulose used in the pharmaceutical, cosmetics and food industries. From XRD analysis, the crystallinity index of raw samples and CNFs was calculated. It was found that the increase of crystallinity index of the prepared CNF was due to the partially removal of hemicellulose and lignin. FT IR spectroscopy helps to identify the changes in chemical compound of natural fibres before and after the chemical treatments. Analyzing the

spectra, it is evident that there was no remaining lignin in the obtaining cellulose nanofibers. This arises from the absence of the absorption bands related to the aromatic ring vibrations (1600-1500 cm⁻¹). The spectral bands observed at 1428 cm⁻¹, 1426 cm⁻¹, 897 cm⁻¹ represent significant cellulose. Moreover, the bands at 1160 cm⁻¹, 1158 cm⁻¹, 1054 cm⁻¹ and 1027 cm⁻¹ are observed in pure cellulose. According to the SEM image, the surface morphology of PALF and PALCNF composed of several microfibrils. Elemental analysis (EDXRF) showed 0.068 % and 0.064% sulfur impurity in CNFs along other main components in PALCNF and SCBCNF. PALF has the thread-like structure. PALCNF exhibits rod-like structure. SCB is also exhibits plate like lamella structure but SCBCNF has irregular aggregated shape fibrils. From these observation, the yield percent of cellulose nanofibres obtained from pineapple leaf fibres (40.26 %) is greater than that of sugarcane bagasse (9.6 %). Moreover, the cellulose nanofibres process for the sugarcane bagasse (65.28 %). This investigation shows that this chemical process was more efficient and effective process for the preparation of cellulose nanofibres from PALF and SCB.

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