The Isolation of Nanofibre Cellulose from Oil Palm Empty Fruit Bunch Via Steam Explosion and Hydrolysis with HCl 10% 

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Abstract. Cellulose nanofibrils were obtained from oil palm empty fruit bunch using steam explosion and hydrolyzed with 10% solution of HCl. Steam explosion coupled with acid hydrolysis pretreatment on the oil palm empty fruit bunch was very effective in the depolymerization and defibrillation process of the fibre to produce fibers in nanodimension. Structural analysis of steam exploded fibers was determined by Fourier Transform Infrared (FT-IR) spectroscopy. Thermal stability of cellulose measured using image analysis software image J. Characterization of the fibers by TEM and SEM displayed that fiber diameter decreases with mechanical-chemical treatment and final nanofibril size was 20-30 nm. FT-IR and TGA data confirmed the removal of hemicellulose and lignin during the chemical treatment process. 

1. Introduction 
Along with the production of palm oil, OPEFB is produced as a by-product of solids. OPEFB is a fibrous mass left after the separation of fruit due to sterilization (steam treatment at 294 kPa for 1 hour) [1]. It is estimated that for every ton of palm oil produced, yielding 1.1 tons of OPEFB approximately. In year 2011, the world production was 14.5 million tons (dry basis), and half of it was produced by Indonesia [2]. The thing that can be done to overcome the production of OPEFB obtained from palm oil is through its traditional management. OPEFB is commonly used as a source of heat and driving the incinerator palm oil mills. However, this may cause environmental pollution [3]. In addition, there are other things that can be performed to overcome the production of OPEFB, such as; it can be used as materials for the development of science and further utilization that can increase the value of OPEFB. Given the development of nanotechnology, empty palm fruit bunches can be used as an ingredient to isolate the cellulosenanofiber. 

Cellulose is a major component within the cell wall of all plants, and is a linear polymer of a poly-(1→4)-D-glucose unit with a syndiotactic configuration [4]. The cellulose chain is organized into cellulosic microfibers surrounded by a non-cellulose matrix within the cell wall of a plant [5]. Cellulosenanofiber is a very thin fiber; for example, the thickness of the nanofiber contained in the wood cell is 3-4 nm. Their characteristic structures allow them to be used in new applications such as reinforcing nanomaterialson plastics [6] drug delivery systems [7] biosensors, and packaging [8]. Steam explosion is an effective method for separating nanofiber from biomass [9]. High-pressure hot
steam followed by rapid decompression is called a steam explosion. The steam explosive process involves saturation of a dry material with steam at high pressure and temperature, followed by a sudden release of pressure, during which the evaporation of water gives the thermo mechanical force causing the material to break. The steam explosive has been shown to be effective for a variety of ligno-cellulosic biomass, including hardwood, softwood, herbaceous residue and wheat straw [10]. In this case, biomass is heated for a short time with saturated vapor at high pressure, followed by sudden decompression. High-pressure steam provides a means to co-fund biomass, penetrate and modify cell wall structures, ensuring effective hydrolysis especially on hemicelluloses that makes cellulose easier to obtain. Decompression abruptly leads to an adiabatic expansion that makes the material is mechanically disrupted from modified fibers in plant cell walls [11, 12, 13].

The advantages of steam explosion are giving the significantly lower environmental impacts, lower energy consumption, lower capital investment, and less harmful chemical processes[9]. Bibin et al. (2010) have isolated cellulosic nanofibers from pineapple fibers by steam explosion methods[14]. Cherian et al. (2008) explained that the steam explosion method was an effective method for separating the nanofiber from biomass. The method of steam explosion with an alkaline medium resulted the occurrence of hydrolysis of hemicellulose present in the fibers and produces a sugar which can then be washed with water, leaving a residue of cellulose with lignin[9]. Kaushik et al. (2011) reported that the method of steam explosion with alkaline medium and followed by a treatment with hydrochloric acid was an effective method for depolymerization and fiber defibrillation process to obtain the cellulosic nanofiber[15]. Based on research conducted by Bibin et al. (2010), cellulosic nanofibers have a broad uses in biomedical and biotechnology applications, such as tissue engineering, drug delivery systems, wound closure and medical implants [14].

Based on the above description, the researcher was interested to study the isolation of cellulose nanofiber by using steam explosion method from empty palm bunches and hydrolyzed with 10% HCl and then characterized by SEM and TEM morphology test, FT-IR functional group analysis and TGA thermal endurance test.

2. Research method

2.1. Equipments
In this study, the equipments used were a set of autoclave tools, a set of glassware, homogenizer, ultrasonic bath, analytical balance, thermometer, hotplate, magnetic stirrer, oven, universal pH, a set of TEM and SEM, a set of FT-IR spectroscopy and TGA machine.

2.2. Materials
The materials used in this study were including, Oil Palm Bunches, NaOH, CH₃COOH(c), NaOCl(c), H₂O₂(c), HCl(c) and distilled water.

2.3. Procedure

2.3.1. Preparation of empty palm bunches. Empty palm bunches were torned apart and dried then chopped into small pieces.

2.3.2. The treatment of OPEFB fiber with steam explosion. OPEFB fibers were immersed with 2% NaOH overnight and then NaOH was disposed after immersion process. Then, it was added with 12% NaOH (with the ratio 1:10) and placed into an autoclave with temperature of 130°C and pressure of 168 kPa. This was performed to reduce the amount of lignin contained in the OPEFB fiber. The fibers were removed from the autoclave and washed with distilled water for several times until they were free from alkaline materials.
2.3.3. **Bleaching process of fiber.** The fibers that have been vaporized and liberated from the alkali were washed with a mixture of 17.5% NaOH solution and 7.4% acetic acid (1: 1 ratio) and with a 1: 3 mixture of NaOCl solution. Bleaching process was performed using H$_2$O$_2$ 8%. Then, the fibers were washed with distilled water and dried in the oven.

2.3.4. **The acid hydrolysis of fiber.** HCl 10% was added into the fiber that has been dried. Then, it was putted in ultrasonicator for 3 hours. After that, it was taken from ultrasonicator and washed until the fiber reached a neutral pH. Then, it was suspended in water and stirred using a homogenizer for 15 minutes until the fiber was agglomerated.

2.3.5. **Morphological analysis.** The morphological characterization of the resultant cellulosic nanofiber was determined using a TEM and SEM analysis equipments. In this case, the TEM equipment used was the JEM-1400 model. Images were taken with the voltage acceleration of 120 kV. Nanofiber cellulose was a dilute liquid and dropped over a carbon coating. As for SEM analysis, the equipment used was ZEIZZ SEM. The analyzed samples were coated with platinum and analyzed using sputtering technique.

2.3.6. **FTIR Spectroscopy analysis.** This analysis was performed to investigate the functional group of fibers before the acid hydrolysis of cellulosic nanofibers. The FTIR spectra were determined using an FTIR ABB 3000 spectrometer and using a triglycerin sulfate detector. Meanwhile, the FTIR spectra obtained were determined in the 4000-450 cm$^{-1}$ absorption area with a resolution of 8 cm$^{-1}$ and using 32 scans for each sample.

2.3.7. **Thermal analysis.** The curve of the thermal analysis for fibers prior to the acid hydrolysis of cellulosic nanofiber was determined using a thermogravimetric analyzer (TGA) type SDT Q600 V20.9 Build 20. Samples were placed on a platinum cup sample. Nitrogen gas was used to the sample and heated at a rate of 10°C/ min from room temperature up to 650°C.

3. **Results and Discussions**

3.1. **The results of cellulose nanofiber from OPEFB**

Empty palm fruit bunches was soaked in clean water, dried and cutted into small pieces have been used as an ingredient in the manufacture of cellulosic nanofibers. The OPEFB fibers used were 50 g and the yield of cellulose nanofiber was 14.54 g. Table 1 shows the data obtained from the results of cellulose nanofiber with a sample of OPEFB. The result of cellulose nanofiber from OPEFB can be seen in figure 1.

Table 1. The weight of cellulose nanofiber obtained from the isolation of OPEFB.

| Weight of OPEFB (g) | Weight of Cellulose Nanofiber (g) | Yield (%) |
|---------------------|-----------------------------------|-----------|
| 50                  | 14.54                             | 29.08     |

Figure 1. (a) OPEFB fibers (b) cellulosic nanofibers.
3.2. Cellulose nanofiber manufacture
The manufacture of cellulose nanofiber was done by inserting a soaked fiber with 12% NaOH into the autoclave device, then bleached with acetate buffer and NaOCl and using H$_2$O$_2$ as well. This was done to remove the remaining lignin and hemicellulose parts. Then, it was followed by hydrolysis treatment with 10% HCl. It was aimed to increase the degree of crystallinity and change fibers into nanoscale fibers. The acid-treated fibers were then suspended with water and stirred using a homogenizer device. This treatment was aimed to break down the pile (Agglomeration). The resulting cellulose nanofiber was analyzed using TEM (Transmission Electron Microscopy). The TEM result in figure 2 showing that the resulting fiber had a size of 20-30 nm and can be classified as nanofiber because its size was below 100 nm.

3.3. Transmission Electron Microscopy (TEM) analysis
Figure 2 shows the result of TEM analysis of cellulose nanofiber which has treated through chemical and mechanical treatment. Mechanical treatment caused the defibrillation of cellulose nanofiber in the cell wall and the TEM images showed the separation of nanofiber from fiber bundles in micro scale size. In addition, the accumulating fibers may also be observed via Transmission Electron Microscopy (TEM). The average diameter of the nanofiber can be calculated from the electron micrograph by using digital image analysis software. Most of the particles were found in the area (range) diameter from 10-70 nm.

![Figure 2.](image)

Table 2 shows the distribution of diameter dimensions of nanometric fibers after passing the final treatment. A total of 20% fiber had a diameter size of 20-30 nm. While 17.89 and 16.84% were fiber with diameter of 40-50 nm and 10-20 nm respectively. The rest were fibers with diameter size of 30-40 nm, 50-60 nm, and 60-70 nm.

| No | Diameter (nm) | Frequency (percentage) |
|----|---------------|------------------------|
| 1  | 10-20         | 16.84                  |
| 2  | 20-30         | 20                     |
| 3  | 30-40         | 13.68                  |
| 4  | 40-50         | 17.89                  |
| 5  | 50-60         | 11.57                  |
| 6  | 60-70         | 9.48                   |
| 7  | >70           | 10.53                  |
3.4. Scanning Electron Microscopy (SEM)
The steam explosion method with an alkaline medium produced a structural change on the fiber surface. The results on the SEM image of the OPEFB after treated with the steam explosion process can be observed to see the structural changes. SEM micrograph was shown in figure 3.4. In this case it was clear that the fibers became separated individually because of the loss of hemicelluloses, lignin, and pectin after the chemical treatment, and they were adhesives around the bundles of fibers. In addition, the figure explained that the average diameter size of the fibers was about 10-20 μm, which was lower than the average size of the bundles of fibers prior to the chemical treatment. The reduced particle size due to the breakdown of hemicelluloses and lignin are also clearly seen in FT-IR data.

3.5. Fourier Transform Infrared Spectroscopy analysis
The following discussion refers to the results of the study. The fibers, both before hydrolysis and cellulose nanofiber were analyzed with FT-IR to see the changes in the constituents or chemical
constituents of the fiber before and after a chemical treatment (hydrolysis) and mechanics. Figure 3.5 shows the FT-IR spectrum of fibers prior to the treatment of hydrolysis and cellulose nanofibers. The peak in the region of 3348 cm$^{-1}$ corresponds to the O-H stretching that is, due to the vibration of the hydrogen attached to the hydroxyl group. The peak at 2916 cm$^{-1}$ shows vibrational stretching of the lignin polysaccharide (Cellulose and hemicelluloses) in the form of saturated aliphatic C-H. The tendency of hydrophilic fiber before the hydrolysis and cellulose nanofiber is reflected on the occurrence of broad band around the 3700-3100 cm$^{-1}$, which shows the O-H group exists in their main component. The Peak area of 1715 cm$^{-1}$ on fiber before the hydrolysis was caused by the presence of acetyl groups from hemicelluloses and uronat ester linkage or ester of the carboxylic group on ferulic acid and p-coumaric acid from lignin or hemicelluloses.

![Figure 5. The comparison of FTIR result of OPEFB before and after the hydrolysis process.](image_url)

It can also be seen in figure 5 that this peak is virtually absent in the cellulosenanofiber spectrum, indicating the breaking of the ester bond. At the peak of the 1634 cm$^{-1}$ region there is a possibility of bending due to the absorption of water and the contribution of the carboxylate group [15]. The C=C aromatic stretching of the aromatic ring of lignin gave two peaks at 1542 cm$^{-1}$ and 1428 cm$^{-1}$. It can be observed in the fiber before the hydrolysis treatment [16]. Then, cellulosenanofibers peak of 1542 cm$^{-1}$ has almost disappeared and the intensity at peak of 1428 cm$^{-1}$ was significantly decreased. This corresponded to the loss of the lignin portion of the fiber. The peak area of 1323 cm$^{-1}$ represents an asymmetrical C-H shape change. In the cellulose nanofiber, the peak intensity in the area of 1261 cm$^{-1}$ has diminished in its sharpness, indicating the loss of hemicelluloses. The area between 1103-1026 cm$^{-1}$represents the C-O stretch and band changes in cellulose, lignin and hemicelluloses residues. The band on the 897 cm$^{-1}$ region indicates the type of structure of cellulose (caused by the β-glycoside bond of the glucose ring on cellulose) [15].

3.6. Thermogravimetric Analysis (TGA)

The result of TGA for Oil Palm Empty Fruit Bunch (OPEFB) with different stages is shown in figure 6. This result clearly shows that the thermal stability of OPEFB fiber increases after chemical and mechanical treatment. The degradation temperature in OPEFB fibers increased after chemical and mechanical treatment. This is caused by the amount of non-cellulosic materials that have been lost and the high structural degrees can be well maintained. In this case, it explains the relationship between the structure and thermal degradation in cellulose, where a large crystallinity structure requires a higher degradation temperature.
Figure 6. The TGA curve of: (a) cellulosic nanofiber (b) fiber prior to hydrolysis treatment.

In addition, there is a difference in the amount of residue produced from the residual fibers after heating up to 600°C against the untreated fibers and nanofibers that have undergone the chemical and mechanical treatment. The maximum residue produced on fiber before hydrolysis treatment was 17.41% (1.870 mg). While the cellulose nanofiber produced as much as 12.97% (1.371 mg) residue. This happened due to the high temperatures that cause the occurrence of thermal decomposition and reduced residual mass of the fibers produced after chemical-mechanical treatment. This occurs due to the missing parts of hemicelluloses and lignin in fiber and high degree of crystallinity in cellulose. These results also relate to the results of FT-IR measurements.

4. Conclusion
The process of steam explosion can break up the bundles of fibers, and with the alkaline medium, it will remove some of the lignin in the OPEFB fiber. The hydrolysis treatment using 10% HCl can increase the degree of crystallinity in the fiber and forming a fiber with nanoscale diameter size. The use of autoclave with a temperature of 130°C and pressure of 168.9 kPa was a proper treatment, because high temperature and pressure can affect the fiber of OPEFB, namely the defibrillation of the fiber. SEM analysis results showed the change of structure in fiber. In addition, TEM analysis showed a fiber diameter of 20-30 nm. Moreover, the FT-IR data proved that the release of lignin and hemicelluloses in the fiber and resulting in thermal stability in cellulose nanofiber to increase compared to the untreated fiber.

Acknowledgement
Authors would like to express gratitude to Ministry of Research, Technology, and Higher Education of Indonesia for funding this research via the funding scheme of “Penelitian Unggulan Universitas 2017”.

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