

Characterization of the Rambutan Peel Fiber (*Nephelium lappaceum*) as a Lignocellulosic Material for Technological Applications

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This study evaluated the potential of rambutan peel (*Nephelium lappaceum*), an abundant, renewable and low-cost waste, as a source of lignocellulose for technological applications. FTIR analysis confirmed the achievement of lignin, hemicellulose and cellulose. It has obtained an extract with antioxidant properties by soxhlet extraction with ethanol. The peel has a high lignin content (35.34 ± 2.05 % (w/w)) and a cellulose content of 24.28 ± 2.30 % (w/w). The extraction of lignin and cellulose nanowhiskers by basic pulping and acid hydrolysis, respectively, were accomplished. A stable cellulose nanowhiskers suspension of type I and II was obtained with a 62.2 % crystallinity index. Thermogravimetric analysis showed that the nanoparticles had good thermal stability. TEM micrographs and birefringence of the suspension confirmed the presence of rod-like nanowhiskers with average length 144.93 ± 50.67 nm and average width of 5.59 ± 2.09 nm. The rambutan peel proved to be a good source of lignocellulosic material for applications, for example, in bionanocomposites.

1. Introduction

Development of new technologies to contribute in environmental preservation is a current theme in research field. The use of biomass waste to obtain higher value added products is highlighted in this study because they are abundant, renewable and cheap. In this context, lignocellulosic biomass is a promising source of important products such as cellulose nanowhiskers (Habibi *et al.*, 2010; Portela *et al.*, 2015) which have application as reinforcing agent in polymer matrices, and lignin, which can be used as a plasticizer. In this sense, this study evaluated the rambutan (*Nephelium lappaceum*) peel as a source of those products. It is an exotic tropical fruit from Asia, whose cultivation has occurred in other parts of the world. The fruit is composed of 40 % edible portion and 60 % of peel and seed (Lestari *et al.*, 2014). Its consumption has increased in recent years due to the softness and sweet flavor of its edible portion and their nutritional composition, rich in antioxidant compounds (Lestari *et al.*, 2014), but the few records of technological applications of its peel, mainly as adsorbents (Ahmad *et al.*, 2011), making it an abundant and interesting waste with little use (Figure 1). In this study, the main chemical composition of the peel was determined and the products obtained (cellulose, hemicellulose and lignin) were analyzed by FTIR. The basic pulping and acid hydrolysis produced lignin and cellulose nanowhiskers, respectively, which were characterized by XRD, TGA and TEM.



Figure 1: Rambutan fruit and rambutan peel.

2. Experimental Part

2.1. Chemical Composition

The contents of cellulose, hemicellulose and lignin from the rambutan peel were determined by an adaptation of TAPPI methods (TAPPI 2002 and 2009). The TAPPI methodology was chosen because it allows the attainment of the lignocellulosic constituents in sufficient amount to characterize. The rambutan fruit was obtained in South of Bahia (Brazil) local market. At first, the peels were separated from edible portion, manually fragmented and placed to dry in an oven for 48 h at 105 °C. The dry peels were milled in a mill Willye TE-680 supplied by Tecnal. The material was subjected to two successive washings with distilled water for 50 min at 50 °C under stirring, and dried at 105 °C for 48 h producing a Fiber. Then, extractives were removed using Soxhlet (190.0 mL of ethanol 96 °GL for 4 h) producing a 'Fiber and an extract which was used to determine the concentration (% Extractives) gravimetrically. Following TAPPI T-203 cm-99 method (TAPPI, 2009), bleaching was carried out: 3.0 g of 'Fiber were immersed in 120 mL of distilled water, under stirring at 70 °C. 2.5 g of NaClO_{2(s)} and 1.0 mL of glacial acetic acid were added, repeated twice, totaling 7.5 g of NaClO_{2(s)}, 3.0 mL of glacial acetic acid and 3 h of bleaching. This procedure was performed in order to obtain 3.0 g of bleached fiber, known as holocellulose. It was added 1.5 g of holocellulose to a beaker with 75.0 mL of NaOH_(aq) 17.5 % (w/v) for 2 min and slow stirring, and then added another 25.0 mL of NaOH_(aq). The suspension stayed still for 30 min, then 100 mL of distilled water were added and the solution remained for another 30 min. The suspension was filtered under vacuum in order to separate the liquor. The solid material, cellulose, was washed with distilled water and dried in oven at 105 °C for 48 h until it reached constant weight. The hemicellulose dissolved in the liquor was precipitated with ethanol/glacial acetic acid solution 9:1 followed by centrifugation. Hemicellulose was dried in an oven at 50 °C until reach constant weight. To obtain the acid insoluble lignin, following TAPPI T-222 om-02 method (TAPPI, 2002), 1.0 g of the Fiber was macerated with 17.0 mL of H₂SO_{4(aq)} 72 % (v/v) for 15 min, standing still for 24 h at room temperature. Then, the material was diluted with distilled water and it was set in reflux for 4 h, filtered under vacuum, washed with distilled water and dried in the oven at 105 °C until it reached constant weight. The ash content was considered as 4.93 % according to the literature (Ahmad *et al.*, 2011). All the chemical compositions were calculated in duplicate at 95 % of confidence by Eq(1), Eq(2), Eq(3) and Eq(4):

$$\%Holocellulose = \left(\frac{m_{Holocellulose}}{m_{Fiber}} \times 100 \right) \times \frac{100 - \%Extractives}{100} \quad (1)$$

$$\%Cellulose = \frac{m_{Cellulose}}{m_{Holocellulose}} \times \%Holocellulose \quad (2)$$

$$\%Hemicellulose = \%Holocellulose - \%Cellulose \quad (3)$$

$$\%Lignin = \frac{m_{Lignin}}{m_{Fiber}} - \%Ash \quad (4)$$

2.2. Fourier Transform Infrared Spectroscopy (FTIR)

The fractions obtained by the TAPPI method were dried in an oven and placed in the desiccator prior to the analysis. FTIR spectra were obtained in a BRUKER VERTEX 70, using the KBr to form tablets under pressure.

2.3. Acid Hydrolysis

Initially the washed fiber was bleached using NaClO_(aq) 2 % (w/w). The bleached dry fiber was milled in order to increase the contact area during the hydrolysis. Then, 5.0 g of bleached fiber were mixed with 50 mL of H₂SO_{4(aq)} 55 % (v/v). The reaction was maintained for 2 h at 50 °C under vigorous stirring. The reaction stopped by adding cold distilled water at 7 °C. The material was centrifuged to remove acid supernatant, while the solid was washed thoroughly with distilled water under stirring at 4,000 rpm, approximately, for 10 min, when turbidity was observed and the supernatant was collected. The turbid suspension was dialyzed against tap water into a membrane until it reached pH 7 and was vacuum filtered to remove solid waste unhydrolyzed. The concentration of the suspension was made gravimetrically in duplicate, drying an aliquot of 5.00 mL in the oven at 105 °C (Neto *et al.*, 2013) until constant weight. This procedure is schematized in Figure 2.

2.4. Lignin Extraction

Lignin was extracted from the washed fiber using NaOH_(aq) 5 % (w/v) in a proportion of 1.0 g of fiber into 15.0 mL solution. The reaction continued for 2 h in the range of 70 to 80 °C under vigorous stirring. The lignin was precipitated with H₂SO_{4(aq)} 98 % (w/w) at a proportion of 1.3 g of fiber to 1.0 mL of solution. The suspension lied still for 24 h until all the lignin was precipitated, then centrifuged and dried in an oven. This procedure is schematized in Figure 3.

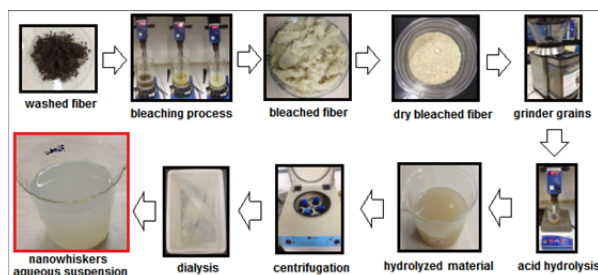


Figure 2: Acid hydrolysis of rambutan peel fiber.

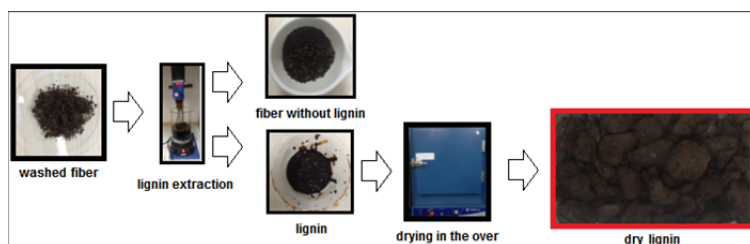


Figure 3: Lignin extraction by basic pulping.

2.5. X-ray Diffractometry (XRD)

The x-ray diffractograms were obtained in a SHIMADZU XRD 6000 at room temperature within a 2θ range from 5 to 80° and a scan rate of 2° min^{-1} . There were used a power of 40.0 kV with a current of 30.0 mA and cooper target ($\text{CuK}\alpha - \lambda = 1.548 \text{ \AA}$). The powdered samples were dried in a oven (4 h and 80° C) in order to remove moisture. The crystallinity index of the material was determined by Segal method (Neto *et al.*, 2013), using peak ($0\ 2\ 0$) and amorphous halo in 18° for calculation.

2.6. Thermogravimetric Analysis (TGA)

The samples were dried in an oven (4 h and 80° C) before the analysis. It was used 6.7 mg of each sample within a $25\text{-}1000^\circ \text{ C}$ range with a heating rate of $10^\circ \text{ C min}^{-1}$. The equipment used was SEIKO EXSTAR6000.

2.7. Transmission Electron Microscopy (TEM)

The samples were analyzed in a THERMO SCIENTIFIC JEOL 1230 microscope. The sample was diluted $1:100$ and deposited in a metallic grid coated with formvar. Uranyl acetate solution 2% (w/w) was used as contrast. The average dimensions were determined from 30 nanowhiskers through software ImageJ.

3. Results and Discussion

3.1. Chemical composition

The main chemical composition of the rambutan peel fiber is shown in Table 1. The visual characteristics of the fractions obtained are in agreement with that reported for cellulose, hemicellulose and lignin. It can be seen that there is a high content of lignin, comparable to other dark fibers such as piassava (Miranda *et al.*, 2015).

Table 1: Main chemical composition of rambutan peel fiber.

Constituent	Content (% w/w)
Cellulose	24.28 ± 2.30
Hemicellulose	11.62 ± 2.31
Lignin	35.34 ± 2.05

An ethanolic extract with intense red color was obtained. According to the literature (Lestari *et al.*, 2014) rambutan peel is rich in phenolic compounds (mainly tannins and flavonoids) with high antioxidant activity, therefore a solution with potential to minimize oxidative degradation in bionanocomposites (Farias *et al.*, 2012). The approximate content of ethanolic extractives (% Extractives) found in rambutan peel was $9.90 \pm 3.36\%$ (w/w).

3.2. Fourier Transform Infrared Spectroscopy (FTIR)

FTIR obtained spectra is shown in Figure 4. All spectra present characteristic bands, confirming the isolation of constituents.

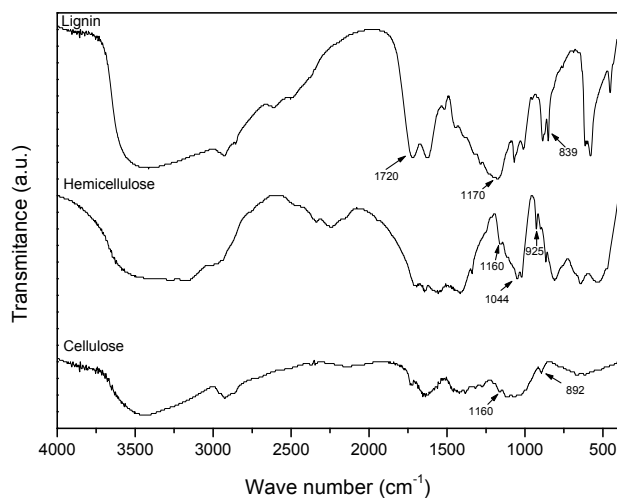


Figure 4: FTIR spectra of lignocellulose constituents.

For the lignin, the band at 839 cm^{-1} corresponding to vibration of syringyl unit; the band at 1170 cm^{-1} corresponding to vibration of the guaiacyl ring; and the band at 1720 cm^{-1} is attributed to C-O vibration of unconjugated ketones, carbonyls and carboxylic acids, which indicates that the lignin has been oxidized during the extraction process. For hemicellulose, the band at 1160 cm^{-1} corresponds the typical C-O-C polysaccharides vibration; the band at 1044 cm^{-1} originates from the C-O bond hemicellulose and the band at 925 cm^{-1} arises from interactions between the sugar units. For cellulose, the same band at 1160 cm^{-1} appears, with another at 1175 cm^{-1} specifically regarding β -glycoside bonds (Sun *et al.*, 2005; Su *et al.*, 2015).

3.3. Acid Hydrolysis

A stable suspension 0.12 % (w/v) was obtained without agglomeration of particles, with turbid and whitish appearance. Figure 5 shows the nanowhiskers evolution over the process of washes and centrifugations until the total dispersion:

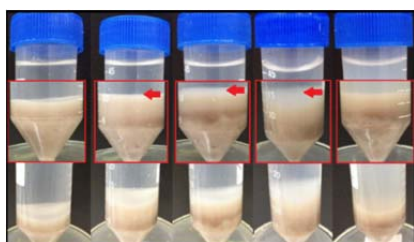


Figure 5: Cellulose nanowhiskers dispersion in distillate water: the red arrows show nanowhiskers.

3.4. Lignin Extraction

With this procedure was recovered about 74 % of the lignin present in the rambutan peel.

3.5. X-ray Diffractometry (XRD)

XRD diffratograms are shown in Figure 6. It appears in both curves typical peaks of cellulose diffraction, although the nanowhiskers peaks have a better resolution. This is consistent because in the fiber there are many amorphous materials that were removed during the bleaching and acid hydrolysis when producing the nanowhiskers. In both profiles it is displayed a mixture of polymorphs of cellulose I and II: $2\theta = 15.08$ e 16.00 (plane 1 0 1 for cellulose I) and $2\theta = 21.74$ and 22.28 (plane 0 0 2 for cellulose II). These results show that the treatment of fiber causes changes in the crystalline structure of cellulose, according to the literature (Neto et

al., 2013). The calculated crystallinity index for nanowhiskers (62.2 %) was higher than the fiber (40.9 %), which is consistent since nanowhiskers are crystalline particles.

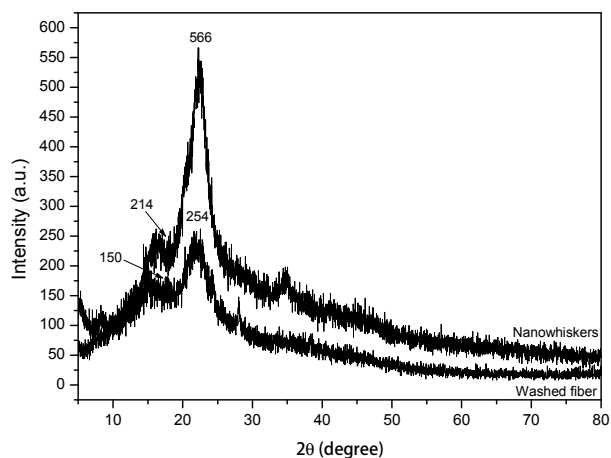


Figure 6: Diffractogram profiles of washed fiber and nanowhiskers.

3.6. Thermogravimetric Analysis (TGA)

Thermogravimetric analysis are shown in Figure 7. DTG profile of the washed fiber has three events: The first event below 100 °C refers to the removal of water or volatile compounds; the second event at 242 °C refers to the decomposition of hemicellulose and the third event at 324 °C relates to the decomposition of the cellulose. It was not possible to observe the lignin related event since it decomposes over a wide temperature range of 80 to 900 °C. For nanowhiskers the DTG profile exhibited only water loss event and decomposition of cellulose, which shows that other non-cellulosic components have been effectively removed by extraction. The thermal resistance of the nanowhiskers was slightly lower than the fiber, 311 °C, which was expected due to the sulfates groups that are introduced in cellulose during hydrolysis. Nevertheless the decomposition temperature is consistent with the temperatures related to nanowhiskers obtained from other sources (Neto et al., 2013). Furthermore, the observed decomposition temperature is higher than the average temperature processing polymer matrices, about 200 °C, which enables its application as a reinforcing agent without thermodegradation.

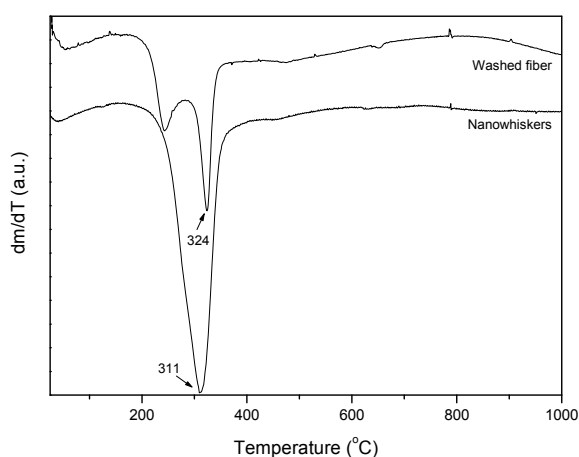


Figure 7: DTG profiles of washed fiber and nanowhiskers.

3.7. Transmission Electron Microscopy (TEM)

Visualization by TEM (shown in Figure 8) confirmed the obtaining of rod-like cellulose nanowhiskers, which is according with a birefringence of suspension when observed between two polarized light sources .The

particles had an average length of 144.93 ± 50.67 nm and average width of 5.59 ± 2.09 nm. It was found only one paper showing the attainment of nanofibers from rambutan, but by the visualization through atomic force microscopy (Niimura *et al.*, 2010). These dimensions are next to dimensions of commercial NCC™ by Cellulforce.

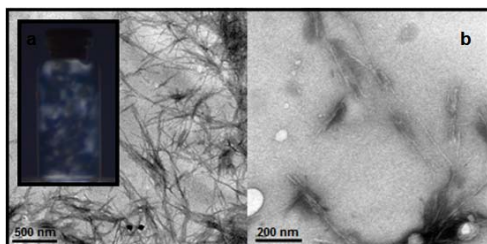


Figure 8: a) Birefringence of rod-like cellulose nanowhiskers; b) TEM micrographs of cellulose nanowhiskers.

4. Conclusions

The present work shows that lignin and rod-like cellulose nanowhiskers can be obtained from rambutan peel. It was possible to recover lignin in significant amount and to prepare a stable aqueous suspension, with nanoparticles having good crystallinity and thermal stability. Because these materials have been widely studied for use as additives in bionanocomposites and rambutan peel is an abundant, low-cost and renewable biomass waste, is proposed use it as a source of lignocellulosic material.

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