Study on structure and morphology (*Boehmeria nivea*) in the irregular and regular parts of the fiber after biodegumming

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Abstract. Ramie is a natural fiber that is very potential to be developed in Indonesia. Decorticated-fiber which has been known as china grass produce different structures irregular part but shows a long straight section in the middle. This study aims to determine differences in chemical components, morphology and microstructure of two different parties after biodegumming process. China grass has been processed to remove gum using pectinolytic fungus. The microstructure of the treated was further tested by Fourier Transform InfraRed (FTIR), X-Ray Diffraction (XRD), and Scanning Electron Microscope (SEM). The FTIR study indicated that during the biodegumming process, chemical bonding of non-cellulose components most removed by the activity of pectinase from the fungus. XRD analysis reflects an increase in the crystallinity of the fiber after biodegumming. Scanning electron microscopy (SEM) was used to confirm a reduction in the size of the fiber after biodegumming either in the irregular and regular part of the fiber after biodegumming.

1. Introduction

Gum removal process (degumming) on natural fiber in plants has been studied by various methods and it is important to increase the content of cellulose if the application will be used in the textile and other industrial. Degummed fiber produced would facilitate for further processing. By knowing the morphology and microstructure of natural fibers of the fiber is expected to obtain the basic knowledge essential for the further development of the natural fiber polymer, considering the utilization can vary widely as well as the need for raw materials with a high content of high purity of cellulose. A novel biodegumming process with pectinases for ramie fiber was developed in combination with oxygen plasma, in order to improve the efficiency of enzymes in impurity degradation with a temperature of 75 ℃, a pH value of 8.0 in subjected solution for 60.0 min [1].

Application of biodegumming had reported by researchers to use *Pectobacterium* sp. [2], a mutant strain T66 from *Bacillus subtilis* [3], with a mixed culture of four bacteria 72 h followed by boiling in 7 g/L NaOH solution for 4 h [4], a large-scale degumming using *Bacillus pumilus* DKS1, a newly isolated strain with high pectate lyase activity, was reported [5]. The galacturonic acid from the ramie previously treated with chemical reagents could be removed by *Grewia optiva* and *Bacillus* sp. [6]. Some enzymes for the degumming of ramie were obtained from Actinomycete [7], Amycolata sp. [8]. A rapid process of ramie biodegumming by *Pectobacterium* sp. CXJZU-120 has been established. Factory practical tests for the rapid process had reported [9].

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Many treatment sets of degumuming and bleaching reduce the extractive content on lignocellulosic fiber to produce microcrystalline, pulp, and cotton. Change in the physical properties of character from the fibers usually investigated by FTIR spectroscopy and X-ray diffraction to estimate the crystallinity and amorphous degree [10] [11]. The crystalline fraction of fiber is given only by cellulose, since the component of hemicellulose and lignin identified as an amorphous. With the purpose of supplying of biomaterial, the factors of physical and chemical properties of the fiber have a significant effect by crystallinity[12].

China grass is coarse fibers produced from the decortication that has different structures with no homogeneous in structure and morphology, especially at the middle and end of the fiber. The structural of fiber with crystallinity and amorphous component could be responsible for making effective a process of degumming. In this study particular objective was to characterize the difference structure and morphology ramie (*Boehmeria nivea*) in the irregular and regular parts of the fiber after biodegumming.

2. Method

2.1. Materials

China grass was supplied from Central of Ramie, Wonosobo, Central Java, Indonesia. The fiber was cut as part regular shape and an irregular shape. Biostarter for bio degumming, a pectinolytic fungus R-apw1 as culture collection from Microbiology Laboratory, Department Biology, University of Padjadjaran, Indonesia.

2.2. Biodegumming

Two part of the china grass as ramie bast fibers was soaked in 3 L medium modified [8] in reactor degumming containing 10% of decorticated dried raw fibers and 10 % of activated biostarter from 3-day-old liquid cultures. The reactors were incubated at room temperature for 42h. Biodegummed fiber was rinsed with water, finally air-dried.

2.3. Analysis Sample

China grass and bio degummed fiber were characterized in terms of changes in the functional group's fiber components by using Fourier Transform Infra Red (FTIR). The FTIR spectra of the cellulosic samples were measured on a FTS 2000 spectrometer Series DIGILAB with a frequency range of 4600-400 cm. X-Ray Diffraction (XRD) was used to observe changes in the diffraction pattern of the cellulose /crystallinity. Data diffraction patterns were collected on a RIGAKU RINT 2500 apparatus. To draw main graphics, the instrument was equipped with a transmission type goniometer. Analysis changes in appearance (morphology) of fiber microscopic observation by Scanning Electron Microscope (SEM).

3. Results

3.1. Fibre Morphology

Morphology of ramie fiber can be observed at the macro structure of china grass before and after biodegumming. The microfibrils shape only can be observed by SEM to observe changes in the size and effectiveness of the treatments biodegumming.

3.1.1. Macro fiber

Decorticated fiber (china grass) generally has a length of ± 175 - 215 cm, two distinct parts in the middle with regular long straight and at both ends are tangled because of mistreatment mechanically on the decortication (separation of bark from the trunk. Two type china grass as a regular and irregular part was degummed by pectinase produced by isolate R-apw1 showed differences character microfibril ramie as shown in Figure 1.

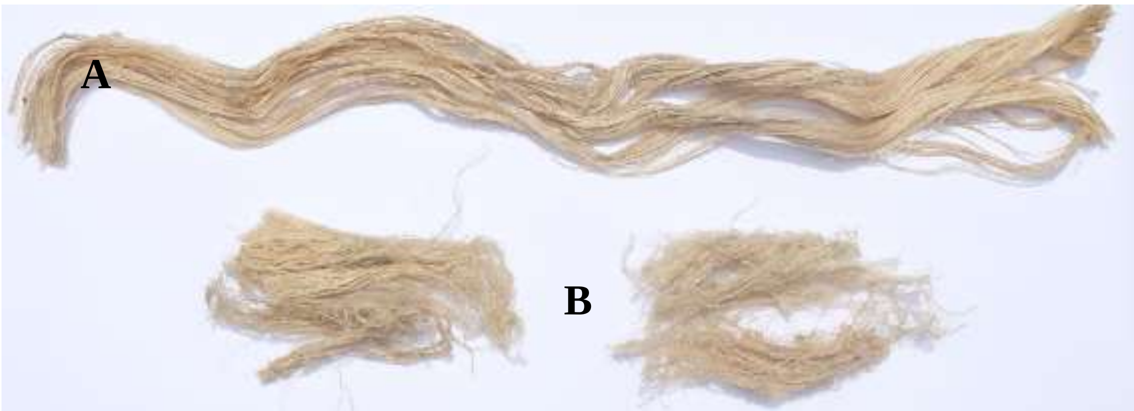


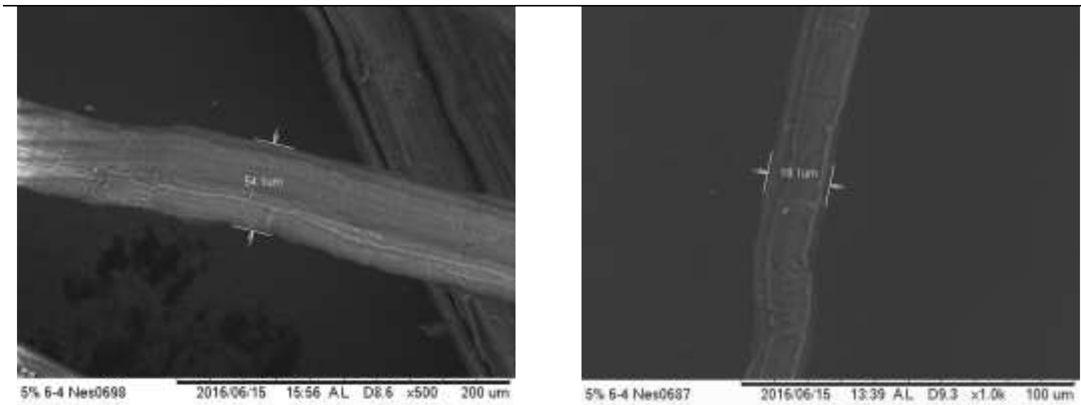
Figure 1. *China grass* (A: middle part- regular and B: the end fiber- irregular)

Gum spreading around the anatomic structure of the bark was composed of pectin as a building block of fiber. Used pectinolytic fungus for biodegumming effective for microfibril degradation cause shrinkage diameter fiber to be microfibrils. As well as physical and visual observations indicate that the results biodegumming fiber become more delicate and brightly colored than before treatment biodegumming. (Figure 2)

Figure 2. The Ramie Fiber : (a) middle part (before and after biodegumming) (b) end of the fiber (before and after biodegumming)

3.1.2. *Micro fiber*

Confirmation of loss of pectin and reduced diameter fiber at the regular and irregular fiber, before and after biodegumming shown as results of SEM (Figure 3)



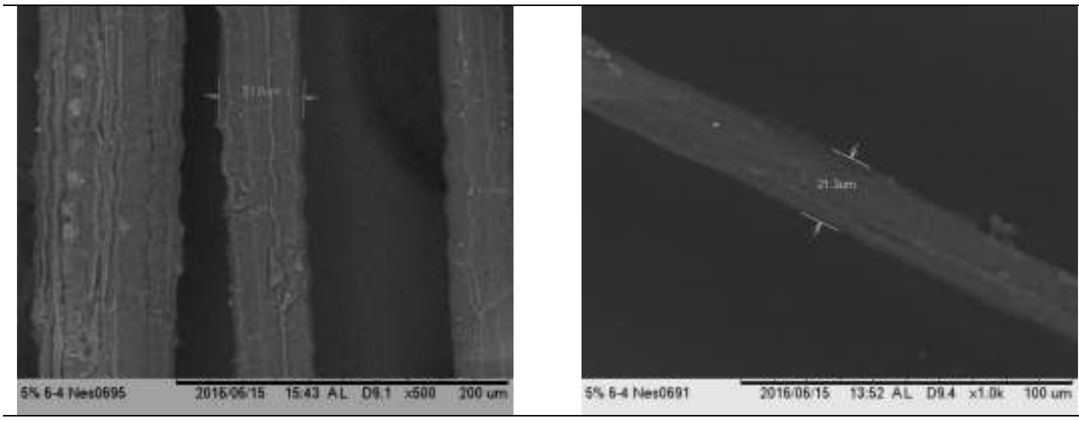


Figure 3. SEM Image Cellulose Microfibrils (A) middle part and (B) the end part (before -(left) and after biodegumming - right))

The size reduction indicated that the fiber fibrillation with a perfect process degradation organic components that change the character of rigid and mutually bonded strongly with each other. the fiber strands become smaller. Activities pectinase produced by pectinolytic fungus capable of hydrolyzing pectin which is a compound of the content of the gum in the fiber.

Changes in the size of the diameter of the microfiber being after biodegumming prived that the process can decipher biodegumming coarse flax fiber decortication result of rigid and mutually bonded strongly with each other, the fiber strands become smaller. It is caused by uses microfungus as biostarter for biodegumming. This fungus was selected with ability pectinolytic to hydrolyze the pectin. Biodegumming of the china grass for both of part of the fiber middle and the ends part leads to remove component non-cellulose and reduced the diameter size china grass before treated by biodegumming had a diameter >50 µM, while after the treatment, the micro fibril of cellulose was average $\underline{+}$ 20 µM.

3.1.3. FTIR Spectra

FTIR is an effectiveness technique for characterizations of surface or interface lignocellulosic fibers. Figure 4 and Figure 5 were shown FTIR spectra of regular and irregular fiber before and after biodegumming.

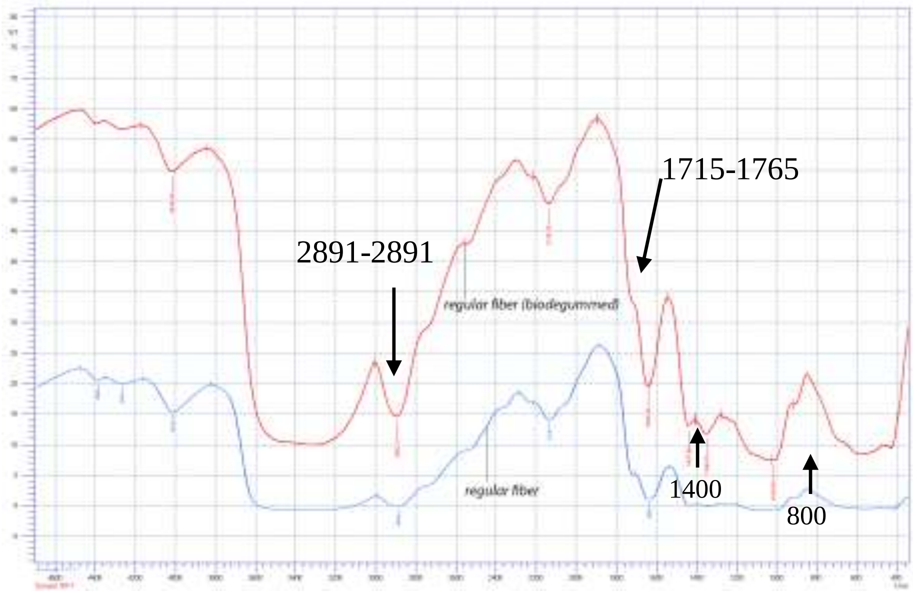


Figure 4. FTIR Spectra of regular fiber before and after biodegumming

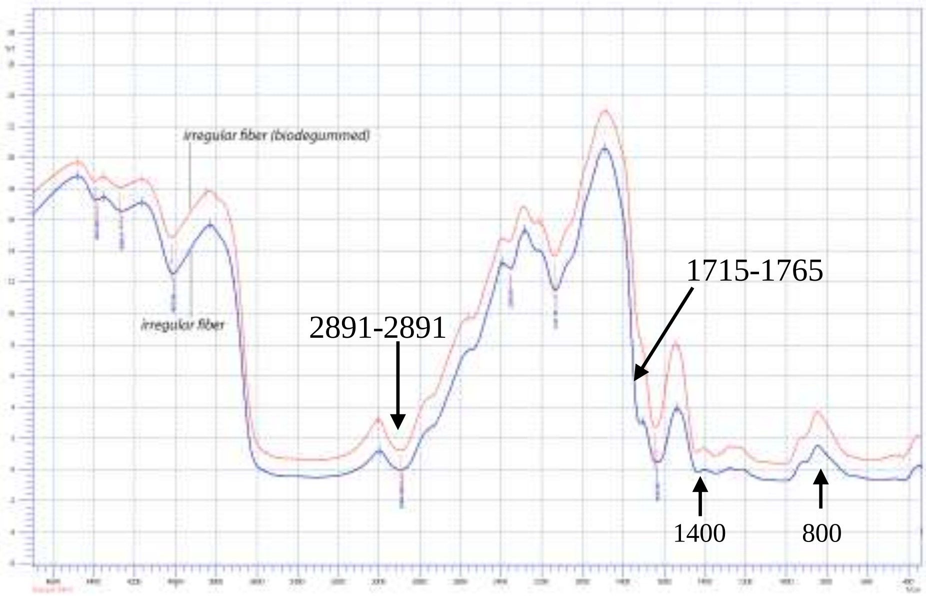


Figure 5. FTIR Spectra of irregular fiber before and after biodegumming

The FTIR was conducted to determine the presence of a functional group of cellulose and no cellulosic, also its changes after biodegumming. Cellulose molecule consists of the elements C, H, and O which has the chemical formula $(C_6H_{10}O_5)n$ hydrogen bonding. Based on data from FT-IR spectra of the fiber (middle and end part) either on the fiber before and after in-biodegumming (Figure 4 and 5), there is the C-H bond in the area of wave numbers between 2891 to 2894 cm-1 were identified as a constituent cellulose. FTIR spectra show that the biodegumming did not damage the chemical bonds biodegumming constituent of cellulose.

C = O at 1765-1715 cm-1 known as chemical bonding of non-cellulose components. FT-IR spectrum the fibers either in the middle or end one showed on the infrared absorption wave number region between 1765-1715 cm-1 in the FT-IR spectrum is an area of vibrational strain C = O of the acetyl group and the ester group of compounds pectin, uronat , hemicellulose or carboxylate ester bond ferulic acid (ferulic) and p-coumaric of lignin or hemicellulose. Disorder structure was examined by the IR absorption after biodegumming this region showed increasing in the intensity. It was found that the absorption spectra both in fiber regular and irregular part of the fiber had changed with the loss of pectin components.

Biodegumming treatment produced some disposition absorption bands in the 1400-800/cm-1 region, especially in the 1000-1200 cm-1 region was found to be very sensitive to change amount of ratio crystalline and amorphous structure of cellulose.

3.1.4. X-ray Diffraction Analysis

Figure 6 showed the spectra-X-ray Diffraction (XRD) showed a pattern of the character of cellulosic fiber as a natural fiber. Both spectra of fiber regular and irregular part before and after bio degumming exhibited a primary peak at the diffraction angle 2θ = 23.12° and two peaks at 2θ = 15.70° and 16.68°, however, those small peaks tended to merge.

XRD overview of fiber central part of the regular and irregular in the end showed the same diffraction pattern, however, high intensity are different, with the presence of amorphous components in part irregular. These figures also featuring high-peak difference crystallinity between china grass and biodegummed fiber. Figure 6 (A) in the regular shape in the middle position of the fiber, the maximum relative intensity of the crystallinity of the fiber at position 2θ - 23° was 11,142.68 cuts, while after biodegumming, increased only 12,106.66 cts. The middle part of the fiber is basically a homogeny polymer cellulose compared to the end of the fiber.

In the irregular, increased fiber crystallinity at the position of 2θ -22.68 ° height peak at 4480.80 cts, while after biodegumming was observed changed position spectra be come 2θ-23.76 cause increasing crystallinity with height peak 6543.73 cts. The results of the analysis of X-ray diffraction pattern showed an increasing crystallinity on the fiber after biodegumming, due to the loss of the gum which is one component of the non-natural cellulose fibers, which incidentally is the cause of formation properties of amorphous (non-crystalline) on the fiber. Part amorphous more easily hydrolyzed than the crystalline portion, so that the hydrolysis treatment causing the fibers to become more crystalline. Therefore it is necessary to do the next stage of fiber processing to completely eliminate non-cellulose components in order to produce pure natural fibers with high level of isolated cellulose.

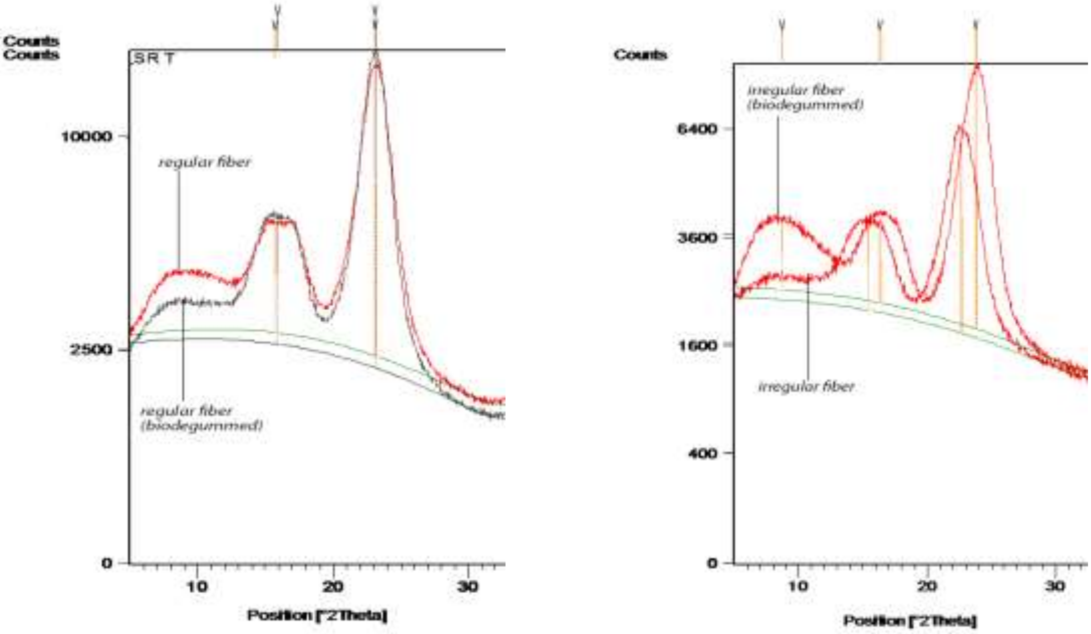


Figure 6. XRD Spectra: (A) Regular part and (B) Irregular part of the fiber before and after biodegumming

Biodegumming of the china grass for both of part of the fiber middle and the ends part leads to remove component non-cellulose proved by characteristic morphology and absorption spectra by FTIR dan XRD.

Acknowledgement
The present work was supported by a Grant from the Ministry of Research Technology and Higher Education, with skim funding Penelitian Unggulan Nasional (PUSNAS), 2016.

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