Characterization of *Meranti* wood Sawdust and Removal of Lignin Content using Pre-treatment Process

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Abstract: *Meranti* wood sawdust (MWS) is consisting of major components such as lignin, hemicellulose, and cellulose. The characterizations of MWS are important to determine the composition in MWS.The objective of this study is to characterize MWS and recover cellulose after pretreatment applied with the removal of lignin content. In order to remove the lignin content from MWS, physical pretreatment, and chemical pretreatment was applied. The physical pretreatment involved process such as drying, grinding, screening a sample for the uniform size of the sample less than 0.5mm and autoclaved samples to make sure that, no microorganisms react with the sample (MWS).Then for chemical pretreatment process, type of chemicals that used to remove lignin content in MWS included sodium hydroxide (NaOH), peracetic acid (CH_3COOH) and acid sulfuric (H_2SO_4).The result shows that, MWS contains - Cellulose: 41.58%, Hemicellulose: 32.81 % Lignin: 33.56 %, Extractives: 3.08 % and Ash: 0.64 %. The result also shows that after using NaOH solution, peracetic acid solution and sulfuric acid solution, the percentage of lignin content in MWS was reduced from 33.56 % to 0.31 % left in MWS. This means that sodium hydroxide (NaOH), peracetic acid (CH3COOH) and acid sulfuric (H2SO4) are categorized as a good agent for treatment. The low lignin content and the high composition of cellulose was important in order to use as energy sources to convert into biochemical products and used as energy sources feedstock for further processing.

Key words: *Meranti* wood sawdust, characterization, pretreatment, lignin

1. INTRODUCTION

Malaysia is a large country that produces many types of wastes. One of it is wood sawdust. These wastes' contribute to a significant environmental problem if not disposed of in a proper manner. Agro-industrial wastes' like sawdust has a great potential as a substrate for sorbitol fermentation and other biochemical products. Besides solving an environmental problem, such utilization of sawdust wastes' would further increase the profitability of the biochemical industries. As a renewable energy source, woody biomass is expected to play an important role in the future metallurgical application too. The wood sawdust is a waste by-product of the timber industry that used as cooking fuel and fuel in manufacturing industries. However, the sawdust can be used as raw material in order to produce a high-value product and low-cost of absorbent for heavy metal due to having lignocellulosic compositions. From the above application of sawdust the environmental problem can be solve and besides that, it is also a good alternative to manage the abundant waste.

Generally, *Meranti* wood sawdust (MWS) is consisting of cellulose, hemicelluloses and lignin content called as lignocellulose materials. Cellulose is a linear homopolymer that composed of D-glucopyranose. The most of the prevalent component in the lignocelluloses is cellulose, where it is predominant component of plant waste and occurs in the plant cell wall as elementary fibrils. The chain of cellulose contains about 10000 D-glucose molecules and made up of the repeating unit of glucose and it is a simple sugar. Hemicellulose is also one type of components in the plant cell wall material where are general interlocks the cellulose microfibrils. It is heterogeneous in nature, short and it has branched polysaccharides which are more complex compared to the cellulose [1]. The chain of hemicelluloses backbone can be a homopolymer that consists of a single sugar repeat unit [2].

Lignin is a very complex molecule compared to cellulose and hemicellulose. It has a molecule that constructed of phenylpropane units where it is networked in a three-dimensional structure where is particularly difficult to biodegrade. Lignin has high contained in the softwoods than hardwoods and most of the agriculture residues [3]. The functions of lignin are to provide rigidity and cohesion to the material cell wall, to form a physic-chemical barrier against microbial attack, resistant to chemical and enzymatic degradation [2].

The purposes of pre-treatment MWS are to alter the lignocellulosic structure in order to removed lignin content and at the same time to increase the rate of enzymatic hydrolysis of primary cellulose. Pre-treatment method of sawdust can be divided into three categories such as physical method, chemical method, and the combination of these two methods [4]. To recover every component in this cellulosic the process pre-treatment should be applied for this studied. The cellulose is an insoluble material, whereas hemicellulose and lignin are soluble material.

2. MATERIALS AND METHODS

A. Chemicals

All chemicals and reagents that used in this study were analytical grades. Chemicals such as sodium hydroxide (NaOH), peracetic acid (CH_3COOH), sulphuric acid (H_2SO4) and others chemical have been purchased from Sigma-Aldrich (M) Sdn Bhd.

B. Cellulosic Materials

In this study, the raw material that used was MWS that was obtained from Kilang Kayu Aman Sdn Bhd, Gambang Kuantan, Pahang. The MWS is of a hardwood type that contains high cellulose content compared to the other types. MWS was taken by bulk from the sawmill plant in order to make sure the quality of the materials is same until the end of the research.

C. Characterization MWS

The MWS that screened in size <0.5mm was used and oven dried in order to characterize and analyzed its prime structural components such as cellulose, hemicellulose, lignin, extractive and ash content. This procedure to analyses compositions of MWS biomass is very important to assess the reactivity of components in biomass.

D. Pre-treatment Process

i) Physical pre-treatments:

The process of physical pre-treatment involves process grinding, screening, autoclaved and drying of MWS

ii) Chemical pre-treatment:

The chemicals pre-treatment which involves the predelignification process using sodium hydroxide (NaOH), first stage pre-treatment process using peracetic acid (CH_3COOH) and second stage pre-treatment using sulfuric acid (H_2SO_4) [5], [6],[7], [8],[9]

E. Determination of lignin content

The concentration of lignin content was determined using Kappa Number. Lignin content was measured for each step in the pre-treatment process in order to know the reduction of lignin contents in MWS. The equation for Kappa Number showed as below:

$$K = \alpha / w \, (Ao - Ae / Ao) \tag{1}$$

α = Volume of Potassium permanganate ($KmnO_4$)
w = Weight of moisture free sample
Ao = Spectra intensities at time zero, before sample added
Ae = Spectra intensities at the end of the reaction

3. RESULTS AND DISCUSSION

A. The Composition of MWS

The composition of MWS in this study was determined by the combination of standard methods [10]. Based on the results, the composition of MWS is shown in Table 1 as below, MWS in this study contains - Cellulose: 41.58%, Hemicellulose: 32.81% Lignin: 33.56%, Extractives: 3.08% and Ash: 0.64%. The previous research on compositions of lignocelluloses by Chinedu has mentioned that lignocelluloses typically contains 35-50% cellulose by weight, 20-35% hemicelluloses by weight and 5-30% lignin by weight [11]. The MWS are hardwood categories' and this hardwood has high cellulose content and hemicellulose content compared to the others like softwood. The high composition of cellulose was important in order to use as a substrate to convert into biochemical products (Krogh, 2008). From the result in Table 1, the composition of cellulose is higher in MWS makes this biomass adequate for further processing of production.

Table 1: The composition of *Meranti* wood sawdust (MWS)

Compositions	**Content (%, w/w)**
Cellulose (alpha)	41.58
Hemicellulose	32.81
Lignin	33.56
Extractives	3.08
Ash	0.64

Besides that, to recover every component in this cellulosic material, the process pre-treatment should be applied. Normally, cellulose is an insoluble material in MWS whereas, hemicellulose and lignin are soluble materials. Based on Table 1, the MWS must be pre-treated first in order to remove lignin, hemicelluloses contents and other compositions in MWS in order to recover the cellulose content.

B. Physical Pre-treatment of MWS

Physical pre-treatment of MWS is quite important because physical properties and cellulose microstructure are among the potential factors influencing the process of direct fermentation of biochemical like sorbitol [12]. For this research, MWS was chopped and milled in order to homogenize the size to less than 0.5 mm. The particle size of MWS must be less than 0.5 mm to make sure the pre-treatment is effective.

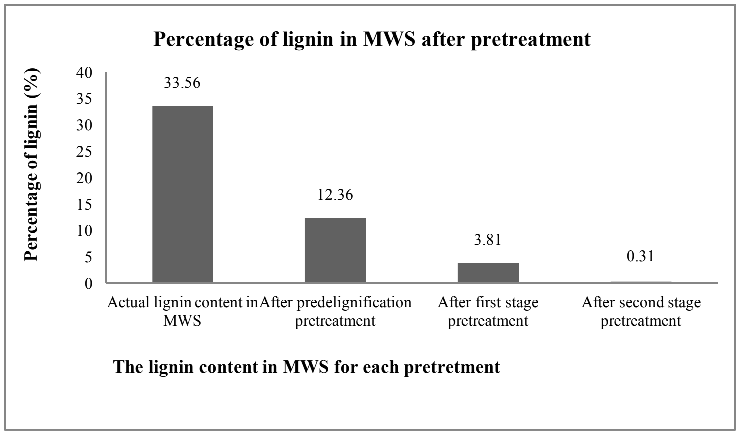

Figure 1: The lignin content in MWS after pre-treatment

The physical pre-treatment method known as milling (cutting the lignocellulosic biomass into smaller pieces) was used in this process. Milling is a mechanical pre-treatment of lignocellulosic biomass. Actually, the purpose of milling in this process is for the reduction of particle size and crystallinity besides causing shearing of the biomass [13]. The reduction in particle size leads to an increase of available specific surface area and reduction of the degree of polymerization (DP). The reduction of DP and shearing, increasing the specific surface area, are all factors that increase the total hydrolysis yield of lignocelluloses and reduces the technical digestion time [3].

C. Chemical Pre-treatment of MWS

Chemical pre-treatment were done to remove hemicelluloses and lignin content in MWS in order to recover cellulose fibers. The hemicellulose and lignin content should be removed in MWS because it can cause inhibition and in the same time, it reduce and decreasing in production for further processing. Basically, chemical pre-treatment is referred to the process of using chemical solutions to modify key chemical components that interfere with biomass cellulose saccharification, mainly hemicelluloses and lignin [14]. The types of chemicals that are used in this pre-treatment are sodium hydroxide (NaOH) for the predeligninfication process, peracetic acid (CH_3COOOH) for the first stage pre-treatment and sulfuric acid (H_2SO_4) for the second stage pre-treatment.

Figure 1 and 2 shows the percentage of lignin content in MWS for each step of the pre-treatment process and removal of lignin content for each step of the pre-the treatment process. The calculation of the percentage lignin content was based on Kappa Number and standard method. The percentage of lignin content in MWS before treatment (untreated) was about 33.56 %. According to the previous research [10], [11] the lignin content in hardwood is 27.8 %. For the untreated MWS, the percentage shows that lignin content is higher than other hardwood types and this means that an effective pre-treatment method is needed to remove lignin content from the MWS. Thus, physical and chemical pre-treatment were the significant factors involved in removing lignin and hemicelluloses contents.

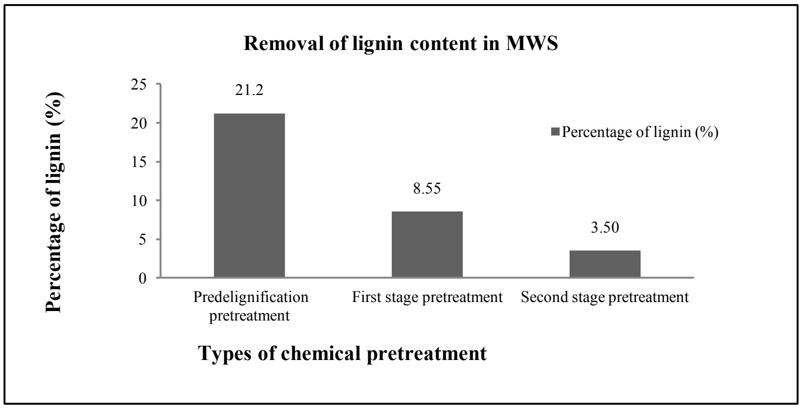

Figure 2: The percentage of lignin content removal in MWS

For the predeligninfication in the pre-treatment process, the 18 % (w/v) sodium hydroxide (NaOH) solution was applied. After using NaOH solution, the removal of lignin content in MWS was higher as shown in Figure 1 where the removal of lignin content was reduced from 33.56 % to 12.36 % respectively. Based on Figure 2, the predeligninfication process during pre-treatment removed about 21.20 % of the lignin content and changed the physical appearance of MWS to a black color. This is because the alkali pre-treatment can cause the solubilization, redistribution, and condensation of lignin, moreover, NaOH acts as a bleaching agent. Furthermore, it can also cause modifications in the crystalline state of the cellulose. From these effects, it can lower and counteract the positive effect of lignin removal and cellulose swelling [12], [13]. During this process, the first reaction taking place is salvation and saponification and it can cause a swollen state of the biomass [10], [19]. Based on Figure 2, the total percentage of lignin content removed from MWS was about 33.56 % leaving some 0.31 % left in MWS.

Generally, the objective of pre-treatment using acid for MWS is to solubilize the hemicelluloses and making cellulose better accessible. The main reaction that occurs during acid pre-treatment is the hydrolysis of hemicelluloses, especially xylan, as glucomannan is relatively acid stable. The solubilized hemicelluloses (oligomers) can be subjected to hydrolytic reaction producing monomer, furfural, and others volatile products in acid environments [20],[21]. In this research, the first stage of pre-treatment using peracetic acid (PAA) was used and conducted under mild conditions (below 90 °C) which mainly caused the removal of hemicelluloses and lignin [10], [18], [22], [23]. For this part, the removal of lignin content in MWS that shown in Figure 1 was reduced from 12.36 % to 3.81 %. This pre-treatment was very helpful in removing lignin and hemicelluloses in order to recover the cellulose content and at the same time increase the enzymatic digestibility [10], [18], [22], [23]. Based on Figure 2, the removal of lignin using peracetic acid for first stage pre-treatment was about 8.55 %. The result indicates that PAA is a good agent for chemical pre-treatment. The physical appearance of MWS also changed from black to yellow. After washing with hot water, the color of MWS will change white, because PAA is also a bleaching agent. Then, the processing proceeds with the second stage of pre-treatment using sulfuric acid (H_2SO_4).

In this study, diluted sulfuric acid was used for the second stage of pre-treatment. The diluted acid can also remove hemicelluloses and lignin contents which increase the accessible surface area of MWS. Based on previous research, the concentration of sulfuric acid was 1.1 % at a temperature of 123 °C and the time of reaction about 118 minutes was applied and it removed most of the lignin content and hemicellulose in MWS. The reduction of lignin content in MWS after the second stage pre-treatment was 3.81 % to 0.31 %. This is because the dilute sulfuric acid is also as an agent to remove lignin. This means that the removal of lignin using sulfuric acid was about 3.50 % respectively. Based on this result, the further process can proceed because of the lower lignin content in MWS.

D. Chemical Structure of MWS

FTIR spectroscopy was used to study the change of chemical structure of MWS and the efficiency of chemical treatment. The spectrum of hardwood showed the same basic structure as all wood samples. The band assignments are listed in Table 2 [24], [25], [26].

Figure 3 below shows the FTIR spectra of MWS before treatment. The major peaks from Figure 3 are considered to be broad bands at 4000cm-1 to 3300cm-1 as attributed to O-H stretching structure, the bands centered between 2800cm-1 to 3000cm-1, predominantly arising from C-H stretching for methyl and methylene group. The region from 1640cm-1 to 1500cm-1 indicates the presence of the C=C alkene and C=C aromatic skeletal vibrations. Then, the band from 1438cm-1 shows the presence of CH2 Cellulose and lignin group. The region from 1346cm-1 to 990cm-1 indicates that the presence of C-H cellulose and hemicelluloses structure, O-H phenolic, O-H alcohol and C=C alkene.

The Fourier Transform Infrared (FTIR) Spectroscopy was used in this study in order to check the functional group at every step of the pre-treatment process. Figure 3 and Figure 4, shows the FTIR spectra of MWS before pretreatment and FTIR spectra of MWS after using chemical pre-treatment of sulfuric acid, H_2SO_4 for the second stage of pre-treatment/ last stage of pre-treatment to recover cellulose. Figure 4 shows that the band intensities for lignin peaks of all samples that were pre-treated are lower than untreated MWS and lignin content is also low after the second stage of pre-treatment. This means that the cellulose that was produced after the second stage pre-treatment/ last stage of pre-treatment was pure because it has the same intensity and peak as the standard cellulose. From the pretreatment process, it shows that the cellulose produced/recover after pretreatment process is higher and pure, whereby the intensity and peak in FTIR spectra that showed in Figure 4 are pure cellulose when compared with cellulose standard.

Table 2: Assignments of IR Bands of Ash-Tree Wood

Band Position (Cm^{-1})	Functional Group
3300-4000	O-H Stretching
2800-3000	C-H Stretching in methyl and methylene groups
1750-1730	C=O Stretching in carbonyl
1640-1618	C=C alkene
1510-1504	C=C aromatic 603keletal vibrations
1462-1425	CH_2 cellulose, lignin
1384-1346	C-H cellulose, hemicelluloses
1260-1234	O-H phenolic
1170-1153	O-H alcohols (primary and secondary) and aliphatic ethers
910	C=C alkane

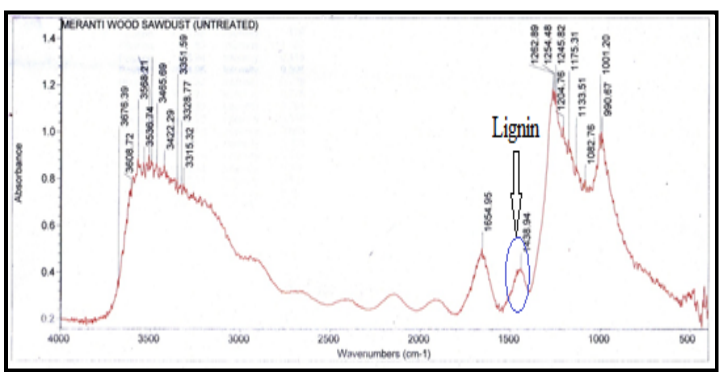

Figure 3: FTIR spectra of MWS before treatment

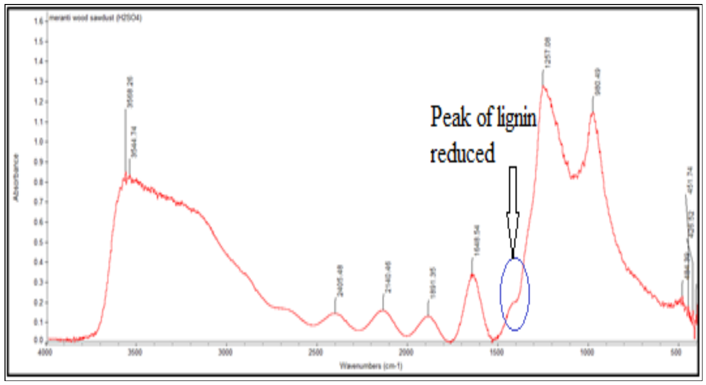

Figure 4: FTIR spectra of MWS after second stage pretreatment using H_2SO_4

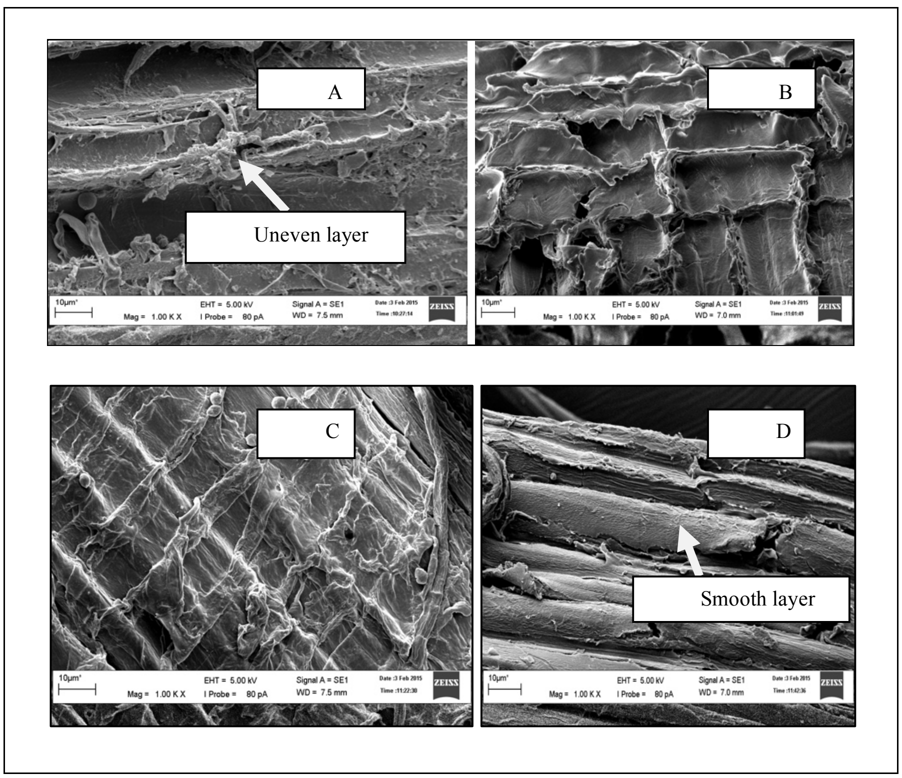

Figure 5: SEM image of MWS: A-untreated; B-after pretreatment using NaOH; C- after pretreatment using PAA; D- after pretreatment using H2SO4

E. Scanning Electron Microscope (SEM) of MWS

Scanning Electron Microscope (SEM) was used to observe the miscibility or otherwise between matrix and fiber at the fractured surface. Figure 5 shows the SEM image for untreated MWS and SEM image of MWS that were treated using solutions sodium hydroxide (NaOH), peracetic acid (PAA) and sulfuric acid (H_2SO_4). From the images of B, C, and D, the surface structure of treated MWS was smoother than that of untreated, A. This is because of others composition like lignin and hemicellulose was removed during pretreatment process. Besides that, the other reason also because of the monomer mixture was penetrated into the cell wall of MWS, that why its look like smooth structure compared untreated. In addition, the uneven layer of lignocellulosic material from untreated A was reduced after using chemical treatment and it also will lead to better mechanical properties.

The scanning electron microscope (SEM) is considered as an important tool in order to observe the morphological characteristic of degraded of MWS at the level of the cell wall, evaluating their damage and identifying the causal agent of decay patterns [27].

4. CONCLUSIONS

The analysis/characterization of cellulosic material like MWS showed that lignin content was higher around 33.56 %. MWS was known to contain a high level of lignin .In order to reduce lignin content in MWS, treatment must be done. The chemical pretreatment such as pre-deligninfication using sodium hydroxide, first stage using peracetic acid and second stage pretreatment using sulfuric acid will be reduced lignin content in MWS in order to improve for further analysis as energy sources.

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