Enhanced Delignification of Oil Palm Empty Fruit Bunch Fibers with *in situ* Fenton-oxidation

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The degradation of lignin in oil palm empty fruit bunch (EFB) fibers by a low concentration of H_2O_2 was observed with the assistance of Fenton oxidation with Fe(III), Fe(0), and Fe_3O_4 as a catalyst. To escalate the oxidation activity toward lignin in the EFB fibers, the uptake of the Fenton reagent on the EFB fibers for *in situ* Fenton oxidation was optimized with fitted Langmuir and Freundlich adsorption models. The efficiency of assisted Fenton reagents was monitored through controlled parameters of H_2O_2 concentration, retention time, and increment of Fenton reagents. The delignification was observed with up to 71.2% of lignin degradation compared to 47.2% without the use of the Fenton reagents. The characteristics of EFB fibers after the oxidation process were changed based on the observation of morphological and chemical properties. The oxidation concurrently dislodged part of the silica bodies and disrupted specific functional groups and the crystallinity of the EFB fibers.

Keywords: EFB; Fe_3O_4; Fenton oxidation; Lignin; Pretreatment; Zero valent iron

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INTRODUCTION

Bioconversion refers to conversion of organic material by biological or chemical processes that reduce the material into fine products. Fine products from bioconversion can be potentially used as biopolymer materials, fine chemicals, or biofuels for energy sources (Jonsson and Martin 2016; Singh *et al.* 2016). The bioconversion process mostly involves underutilized lignocellulosic materials. These materials are attracting interest due to their low cost and renewability. They are conveniently obtainable from nature and as byproducts from the agriculture industry. Malaysia, a major world producer of palm oil, requires 71.3 million tons of fresh fruit bunch every year for extraction processes. The process leads to 19 million tons of residue annually (Mekhilef *et al.* 2011). These residues consist of lignocellulosic material, such as trunks, oil palm frond, and empty fruit bunch (EFB) fibers, which are still underutilized apart from their use for soil enrichment or being burned in incinerators. Therefore, EFB is the greatest contributor of oil palm residue with about 17 million tons annually (Chin *et al.* 2013).

Oil palm EFB fibers are composed of lignin, cellulose, and hemicellulose. These form the major chemical composition in the lignocellulosic fibers (Law *et al.* 2007). The composition of cellulose and hemicellulose can be hydrolyzed into useful simple sugars such as glucose, mannose, galactose, and xylose. These simple sugars are the core substrate to produce a compound such as xylitol, sorbitol, mannitol, *etc*. that is extensively used in pharmaceutical, food, and thin coating applications (Rahman *et al.* 2007; Maki-Arvela *et al.* 2011). In addition to the high conversion of sugar yield, there are several factors that contribute to the hydrolysis process, such as the porosity of the materials, cellulose fiber crystallinity, and lignin and hemicellulose content (Wan and Li 2012). The presence of lignin and silica hinders the penetration of chemicals or biochemical into the hemicellulose matrix and cellulose. Moreover, silica bodies are found in great numbers on EFB fibers; they attach to circular craters and spread uniformly over the strand's surface (Yunus *et al.* 2010). While the focus is to obtain maximum sugar conversion of lignocellulose, the pretreatment process is required to facilitate the hydrolysis of cellulose and hemicellulose.

The pretreatment process of lignocellulose generally refers to the removal of lignin and disruption of the structure of lignocellulose based on several technologies consisting of physical, chemical, and biological methods (Chaturvedi and Verma 2013). Pretreatment consists of rupturing the complex lignin seal, loosening up the crystalline structure of cellulose, and increasing the porosity of the biomass (Tadesse and Luque 2011). An ideal pretreatment process should include producing highly digestible solids that enhance sugar yields during enzyme hydrolysis, avoiding the degradation of sugars, minimizing the formation of inhibitors for subsequent fermentation steps, recovering lignin for conversion into valuable coproducts, and maximizing economic benefits (Sun and Cheng 2002). These changes in lignocellulosic materials make it highly accessible for hydrolysis and result in higher production of fine sugars. This will have a significant impact on the overall yield of the bioconversion process.

In previous research, the usage of a strong oxidizing agent has shown good selectivity towards the lignin structure (Gould 1984; Mishima *et al.* 2006). The oxidative method requires pre-soaking lignocellulosic biomass with an alkaline reagent before proceeding with an oxidizing agent, *i.e.* hydrogen peroxide (H_2O_2). Thus, it has been demonstrated that pretreating the byproduct enhances the sugar yield during the hydrolysis process. However, the limitation of this process is the high usage of chemicals and water due to a high liquid/solid ratio, leading to an increase of pretreatment expenditure process (Cheng *et al.* 2008). Instead, the introduction of a catalyst in the system could improve the reactivity of the degradation process (Canton *et al.* 2003; Kang *et al.* 2009; Zhang *et al.* 2014). For the improvement of reactivity, the catalytic activity of Fenton reagents is attractive for exploitation, depending on whether the catalyst employed is easily accessible and cost effective (Sun *et al.* 2011). Iron-based reagents have importance in the oxidative method because they involve the creation of free radicals. The matter that distinguish different type iron reagents is the reactivity and free radical generation (Cui *et al.* 2009).

Therefore, improvising a pretreatment of the oxidative method was conducted with the assistance of Fenton reagents. The predominant process of adsorption and desorption studies was performed in determination of the uptake and recovery of Fenton reagents on EFB fibers. The oxidation process was introduced with a low concentration of Fe(III), zerovalent iron (Fe(0)), and magnetite particles (Fe_3O_4) as the Fenton reagents to enhance the delignification process of EFB fibers. Even though Fe(0) is expensive and unstable, it has been introduced in this study because it can generate hydroxyl radicals without the presence of hydrogen peroxide (Drzewicz *et al.* 2012). The impact of hydrogen peroxide concentration, retention time, and weight ratio of Fenton reagents on delignification was observed to identify the efficiency of the catalyst. Furthermore, the total sugar conversion of EFB fibers was measured through acid hydrolysis in comparison with different Fenton reagents in the delignification process.

EXPERIMENTAL

Materials

Oil Palm EFB fibers were obtained from Szetech Engineering Sdn. Bhd (Selangor, Malaysia). The fibers were milled and sieved to collect fibers with sizes ranging from 106 µm to 500 µm. For the oxidation process H_2O_2 (Merck, Darmstadt, Germany) was used. A stock solution of Fe(III) was prepared from $FeCl_3$ (Merck, Darmstadt, Germany) in deionized (DI) water with aliquots of 1 M HCl (Merck, Darmstadt, Germany) to avoid sedimentation. Zerovalent iron (Fe(0)) in this study was synthesized by an *in situ* reaction of Fe(III) and sodium borohydride, $NaBH_4$ (Merck, Darmstadt, Germany). Magnetite, Fe_3O_4, was synthesized according to a previous study using the chemical co-precipitation method of $FeCl_2$, $FeCl_3$, and NaOH (Chia *et al*. 2006). Briefly, the reaction was achieved with the molar ratio of Fe^{2+}:Fe^{3+}: NaOH = 1:2:8 at 60 °C. After the reaction, the synthesized solution was washed with distilled water to remove excess unreacted chemical.

Optimization and Recovery of Fenton Reagents

Optimization of Fenton reagent uptake on EFB fibers was conducted based on an adsorption isotherm study. Adsorption isotherm was performed in a series of concentrations of Fe(III) ions solutions (5 mg/L to 40 mg/L) at room temperatures. The mixture of 50 mL of Fe(III) ions solution and 1 g of EFB fibers was placed in a water bath shaker at a constant speed of 200 rpm for 4 h. The final concentration of Fe(III) (C_e) at equilibrium was measured using atomic absorption spectroscopy (AAS) to calculate the adsorbed amount of Fe(III) at final equilibrium (q_e) using the following equation,

$$q_e = \frac{(C_0 - C_e)V}{m} \tag{1}$$

where C_0 and C_e are the initial and equilibrium concentrations of the Fe(III) (mmol/L), respectively, V is the volume of the solution (mL), and m is the mass of the adsorbent (g).

The recovery of the Fenton reagent was measured by a desorption study with sufficient amounts of the Fenton reagent (8 mg/L) adsorbed in EFB fibers (1 g). The fibers were washed with deionized water to remove excess amounts of Fe(III) and dried overnight at 105 °C. A desorption experiment was performed using 50 mL of 0.1 M NaOH stirred with 1 g of adsorbed EFB fibers for 4 h. The amount of Fenton reagent desorbed from EFB fibers was calculated using the following equation,

$$\text{Fenton reagent recovery (\%)} = \frac{\text{Concentration desorbed (mg/L)}}{\text{Concentration adsorbed (mg/L)}} \times 100\% \tag{2}$$

and desorbed EFB fibers were washed to remove excess eluent and dried until a constant weight was achieved.

Oxidation Pretreatment Assisted by Fenton Reagents

A preliminary study on EFB delignification by H_2O_2 (200 mmol) had been carried out before oxidation pretreatment assisted by Fenton reagents as a control condition for comparison. The oxidation process assisted by Fenton reagents was performed in an optimum concentration uptake of Fenton reagents, with a mixture ratio of 50 mL prepared Fenton reagent of Fe(III) ion solution and Fe_3O_4 to 1 g of EFB fibers. The sample was placed in a water bath shaker at room temperature with a constant speed of 200 rpm for 4

h. After the adsorption took place, the oxidation process started with the addition of H_2O_2 and was stirred for 24 h. In the case of preparation of Fe(0) as the Fenton reagent in the system, the process was performed under an *in situ* reaction. A similar procedure of the adsorption of the Fe(III) ion on EFB fibers was done for 4 h; an immediate reaction of the reduction agent $NaBH_4$ was carried out, with the molar ratio of Fe(III) to $NaBH_4$ being 1:2. The process was repeated with a controlled parameter of H_2O_2 concentration (50 mmol to 200 mmol), retention time (24 h to 168 h), and concentration of Fenton reagents (2 mg/L to 8 mg/L). All the pretreated fibers were washed and dried overnight at 105 °C, and the effect of pretreatment on the Klason lignin content of EFB fibers was measured using the TAPPI T222 os-06 (2006) standard.

Characterization

Observations of morphological changes after pretreatment were obtained from a field emission scanning electron microscope, FESEM (Merlin Compact, Zeiss Pvt. Ltd., Oberkochen, Germany). The changes in the crystalline region after the pretreatment were verified using an X-ray diffractometer, XRD (Bruker D8 Advance, Bruker, Billerica MA, USA). Characterization for the functional groups of the EFB fibers was performed using attenuated total reflectance Fourier transform infrared spectroscopy, ATR-FTIR (ALPHA FTIR Spectrometer, Bruker, Billerica MA, USA) at a resolution of 1 cm^{-1} in the range of 4000 cm^{-1} to 650 cm^{-1}. The desorption performance for Fenton reagent recovery was measured by atomic absorption spectroscopy, AAS (Perkin Elmer Analyst 800, Perkin Elmer, Shelton, USA) and the Fenton reagent excess on the EFB fibers was observed on energy dispersive X-ray spectrometry, EDS (Hitachi SU8000, Hitachi High-Technologies Co., Tokyo, Japan).

RESULTS AND DISCUSSION

Optimization for Uptake and Recovery of Fenton Reagents

The reactivity of the catalyst reported in a previous study shows the dependence on a number of participating surfaces (Wachs and Goutam 1994). Hence, in the utilization of assisted Fenton oxidation on lignin degradation, the optimum uptake of the Fenton reagent on the surface of EFB fibers was determined through an adsorption study. The maximum uptake of the catalyst and the assumption mechanism of the interaction was determined with correlation adsorption models from Langmuir and Freundlich. In general, the Langmuir model assumes monolayer adsorption between the adsorbate molecules and the adsorbent medium, while the Freundlich model explains the heterogeneous adsorption behavior of the adsorption mechanism. The Langmuir isotherm model can be expressed as (Langmuir 1917),

$$q_\mathrm{e} = \frac{Q_0 b C_\mathrm{e}}{1 + b C_\mathrm{e}} \tag{3}$$

where Q_0 is the maximum adsorption capacity per unit mass of adsorbent (mmol/g) and b is a constant related to the adsorption energy (L/mmol). Whereas, the Freundlich isotherm model is expressed as (Freundlich 1906),

$$q_\mathrm{e} = K_\mathrm{F} C_\mathrm{e}^{1/n_\mathrm{F}} \tag{4}$$

where K_F and $1/n_F$ are the Freundlich constants, with K_F representing the relative adsorption capacity ((mg/g)(L/mg)$^{1/n}$)) of the adsorbent and n_F representing the degree of dependence of adsorption on the equilibrium concentration of Fe(III) ions. The adsorption isotherm models were fitted in non-linearized plots of Langmuir and Freundlich in Fig. 1(a), and the calculated constants are summarized in Table 1. According to the coefficient of determination, R^2 of both models, Langmuir showed highly fitted data (~0.99) that could define the monolayer interaction between the adsorbent to adsorbate in the adsorption mechanism of EFB fibers and Fe(III) ion. In addition, the maximum uptake of Fe(III) on the fibers was 0.352 mg/g, which was lower than the EFB fibers and activated carbon uptake for Fe(III) reported in previous studies, which was 1.33 mg/g and 0.92 mg/g, respectively (Onganer and Temur 1998; Khosravihaftkhany *et al.* 2013). The uptake of Fe(III) on the EFB fibers mostly can be attributed to the electrostatic attraction of positively charges of metal ions toward the negative charges of the hydroxyl group in the lignocellulosic composition (Khosravihaftkhany *et al.* 2013).

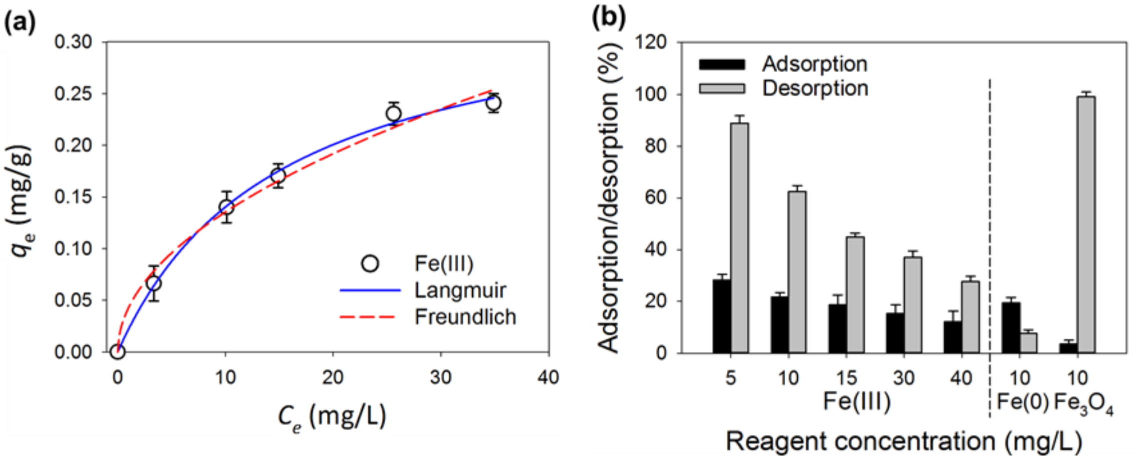

Fig. 1. The optimization of deposited Fenton reagents in (a) the uptake of Fe(III) ion adsorption isotherm of non-linear Langmuir and Freundlich and (b) adsorption/desorption behavior of Fenton reagent on EFB fibers (Fe(III): 5 mg/L to 40 mg/L; EFB fibers: 1 g)

Furthermore, the feasibility to recover Fenton reagents after delignification process was evaluated by a desorption study. This to avoid any excess of Fenton reagents on the EFB fibers for the next stage of bioconversion process. In comparison with the Fe(III) ion recovery from the desorption study recorded in Fig. 1(b), the highest percentage of Fe(III) desorption was 88.9% at 5 mg/L of adsorption uptake. Alkaline eluent can assist the desorption of Fe(III) ions due to ion exchanges of the adsorbate on the EFB fibers. However, a higher concentration of the Fe(III) ion might force more ions to penetrate the pores' site of adsorbent, which has been found to result in a low desorption capability, and showed only 27.6% recovery at 40 mg/L of Fe(III) (Wang and Li 2009). Besides, the desorption performance of 10 mg/L of Fe(0) and Fe_3O_4 showed significant differences in reagent recovery of 7.6% and 99.0%, respectively. Even though the adsorption uptake of Fe(0) with a similar concentration of Fe(III) was comparable, the desorption performance was much less due to the *in situ* formation of Fe(0) particles. The penetration of Fe(III) was on the lumen, hence the introduction of reducing agent of $NaBH_4$ as the formation of an *in situ* Fe(0) particle contributed more to a homogeneous distribution of the particles compared to Fe_3O_4 (Drzewicz *et al.* 2012).

Table 1. Calculated Constants from Fitted Non-linear Adsorption Isotherm Models (Langmuir and Freundlich); (Fe(III): 5 mg/L to 40 mg/L; EFB Fibers: 1 g; NaOH: 0.1 M)

Adsorbate	Langmuir			Freundlich		
	Q_0	b	R^2	K_F	n	R^2
Fe(III)	0.352	0.066	0.997	0.001	1.99	0.979
Note: Q_0 (mg/g) = maximum adsorption capacity; b (L/mg) = constant related to adsorption the adsorption energy; K_F ((mg/g)(L/mg)$^{1/n}$) = relative adsorption capacity; n = degree of dependence of adsorption; and R^2 = coefficient of determination						

Effect of Delignification in Assisted Fenton Reagents

In the authors' preliminary study, the oxidation of EFB fibers with 200 mmol without the assistance of Fenton reagents showed the degradation of lignin from 20.7 wt.% to 10.9 wt.%, which represents a change of 47.2 %. With the assistance of 4 mg/L of Fe(III) and the increasing of concentration of H_2O_2, the delignification of EFB fibers was increased by approximately 9.8% (Fig. 2(a)). Moreover, sufficient amounts of H_2O_2 were predominant in the oxidation activity; the increased H_2O_2 concentration from 100 mmol to 200 mmol led to a noticeable increase of hydroxyl radical ion production (Staehelin and Hoigne 1982). A high oxidation rate can cause reduction in the cellulose crystallinity and disruption between lignin and carbohydrate, hence causing rupture of the lignin seal and degradation of a larger scale compared to EFB that was treated at a lower concentration of H_2O_2 (Jönsson and Martín 2016). The reaction time of the oxidation activity was observed for seven days, and the decrement of lignin content over time is shown in Fig. 2(b). The oxidation reaction seemed to have higher efficiency for the first 24 h, and from 36 h onward the system appeared to be making no further significant change. Although the Fenton reaction still occurred throughout the process, the reduction of lignin content from 24 h to 36 h was approximately 1 wt.%.

In brief, H_2O_2 and ferrous ions have the capability to conduct the Fenton reaction in the generation of hydroxyl radicals. The Fenton reaction describes the process of the pathway consisting of the three reagents of oxidant, reducer, and metal ion. These hydroxyl radicals are very reactive and react with hydrogen atoms of lignin (R in Eq. 10 represents the lignin compound), which then reduce the lignin compound (Zhang *et al.* 2014). This process could be shown with the following equations,

$$\mathrm{Fe^{3+} + H_2O_2 \rightarrow Fe^{2+} + H^+ + HO_2^{\bullet}} \tag{5}$$

$$\mathrm{Fe^{2+} + H_2O_2 \rightarrow Fe^{3+} + {}^{\bullet}OH + OH^-} \tag{6}$$

$$\mathrm{{}^{\bullet}OH + Fe^{2+} \rightarrow Fe^{3+} + OH^-} \tag{7}$$

$$\mathrm{HO_2^{\bullet} + Fe^{2+} + H^+ \rightarrow Fe^{3+} + H_2O_2} \tag{8}$$

$$\mathrm{HO_2^{\bullet} + Fe^{3+} \rightarrow Fe^{2+} + H^+ + O_2} \tag{9}$$

$$\mathrm{RH + {}^{\bullet}OH \rightarrow R^{\bullet} + H_2O} \tag{10}$$

where the equation pathways show that the presence of Fe^{3+} in aqueous solution undergoes a redox reaction (Eq. 5), which gives rise to Fe^{2+} and hydroxyl radicals *via* Fenton oxidation (Kang *et al.* 2009). Simultaneously, the re-oxidation of Fe^{2+} and Fe^{3+} by H_2O_2, or hydroxyl radicals (Eqs. 5 through 9) offers an interesting catalytic effect, and a larger amount of hydroxyl radicals can be formed (Canton *et al.* 2003). Hence, the increase of Fe(III) concentration caused a drastic decrement of lignin due to the higher catalytic activity from the generation of free hydroxyl radical ions (Fig. 2(c)). However, throughout the study, it was shown that a higher concentration of Fe(III) may have led the oxidation reaction to reach the saturation limit. This phenomenon may have been due to the re-oxidation reaction reaching equilibrium and limiting the presence of Fe^{2+}, which retards the degradation of lignin in EFB fibers (Suh *et al.* 2003).

The increment of Fe_3O_4 in the system showed an improvement of the oxidation process, which caused the degradation of lignin to be reduced to 5.9 wt.%. This may have been due to the presence of Fe^{2+} (Eq. 6) being adequate for the degradation process, which correlated with the generation of the hydroxyl radical ion. In comparison with Fe(III), Fe_3O_4 has an inverse spinel structure that accommodates both Fe^{2+} and Fe^{3+} and provides effective electron hopping between these sites. This allows the catalytic reaction to occur on the surface site (Suh *et al.* 2003). Because Fe_3O_4 is comprised of Fe^{3+} and Fe^{2+} ions, lignin degrades faster with simultaneous reactions (Eqs. 5 and 6) compared to Fe(III), which is dependent on the oxidation of the Ferric ion, Fe^{3+} (Eq. 6) (Ma *et al.* 2007). Thus, the oxidation reaction was slightly higher in overall catalytic performance of Fe(III) and Fe(0), which was up to 71.2%, 66.7%, and 42.6%, respectively.

In contrast with Fe(III) and Fe_3O_4, Fe(0) is readily oxidized to the ferrous iron, Fe^{2+} by many substances in natural water. This characteristic of Fe(0) leads to dissolution of the solid even in extremely controlled conditions, which is the primary cause of metal corrosion (Agrawal and Tratnyek 1996). This metal corrosion occurs in two different conditions, an aerobic and anaerobic reaction. This reaction was feasible in this experimental process because the process was performed under room temperature and atmospheric pressure. Therefore the aerobic reaction expressed by Eq. 11, is the primary reaction that is possible with aqueous Fe(0) (Wieckowski *et al.* 1983).

$$2Fe^{0} + 4H^{+} + O_2 + 2H_2O \rightarrow 2Fe^{2+} + 2H_2O \tag{11}$$

$$2Fe^{0} + 2H_2O \rightarrow 2Fe^{2+} + H_2 + 2OH^{-} \tag{12}$$

$$Fe^{0} + R + H^{+} \rightarrow Fe^{2+} + RH \tag{13}$$

$$2Fe^{2+} + 4H^{+} + O_2 + 2H_2O \rightarrow 2Fe^{3+} + 2H_2O \tag{14}$$

$$2Fe^{2+} + 2H_2O \rightarrow 2Fe^{3+} + H_2 + 2OH^{-} \tag{15}$$

Interestingly, the reaction of *in situ* Fe(0) with organic compound (R in Eq. 13, represents the lignin compound) in the solution was independent without the existence of H_2O_2. In brief, Fe^{+2} is a primary product from Eqs. 11 and 12, which can undergo further oxidative transformation (Eqs. 14 and 15) (Crane and Scott 2012). However, the reactivity of oxidation is not as efficient as the oxidation process that involves H_2O_2 and assistance from the iron catalyst. However, the lignin in EFB fibers also decreased with increased concentration of Fe(0), as shown in Fig. 2(c). The result showed the lignin content decreased strongly for 2 mg/L of Fe(0) and gradually decreased with further increase in concentration. At 6 mg/L to 8 mg/L Fe(0), the lignin content decreased slowly because the reactivity of the solution reached saturation level.

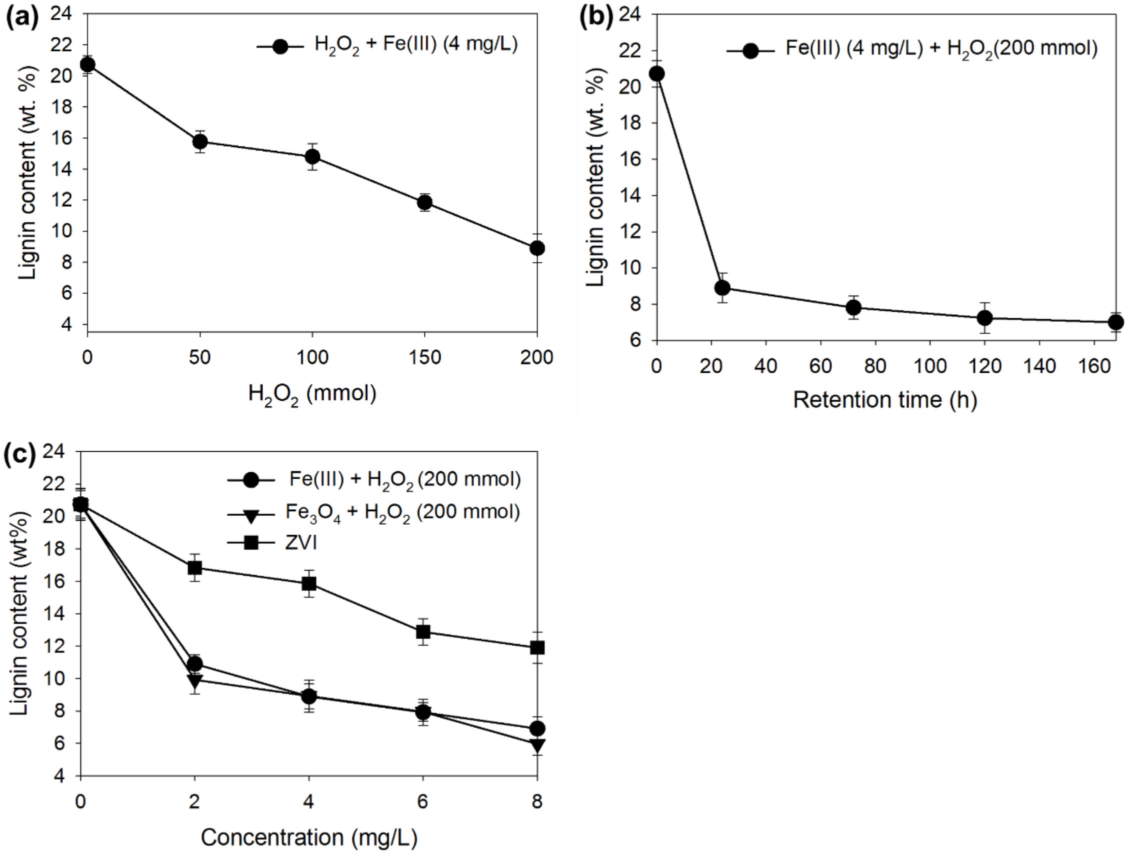

Fig. 2. The delignification of EFB fibers (a) in assisted of Fe(III) with different H_2O_2 concentrations (Fe(III): 4 mg/L; EFB fibers: 1 g; H_2O_2: 50-200 mmol)., (b) oxidation retention time (Fe(III): 4 mg/L; EFB fibers: 1 g; H_2O_2: 200 mmol).and (c) with different Fenton reagents (Fenton reagents: 2-8 mg/L; EFB fibers: 1 g; H_2O_2: 200 mmol)

Characterization Pretreated EFB Fibers

The morphological changes of EFB fibers after the oxidation process assisted with the highest reduction parameters on the Fenton reaction of 8 mg/L of Fe(III), Fe(0), and Fe_3O_4, were observed with FESEM micrograph images, as shown in Fig. 3. Ruptured and rougher surfaces of the EFB fibers were discovered after the oxidation process. In comparison with the raw fibers, the degradation of lignin was observed to cause many cavities and porous structures to form over the strand fibers. Cavities found on the fibers were recognized as being due to dislodgement of the silica bodies, which was consistent with the nature of raw EFB fibers. The reduction of silica on the fibers could be further examined by the EDS spectrum, which identified 10.5 wt.% and 35.2 wt.% of Si and O, respectively. These represent the SiO_2 formation in raw EFB fibers, and subsequently were reduced to 0.4 wt.% to 1.0 wt.% of Si with the assistance of the Fenton reagent (Ghazali *et al.* 2009). Naturally, silica bodies act as an immune defense against the harmful environment of attack by microorganisms. If this defense was capable of acting during the hydrolysis process, it would reduce the fine sugar monomer yields (Law *et al.* 2007). The oxidation process assisted with Fenton reagents showed some improvement of the delignification process along with the removal of silica bodies. Therefore, the removal of silica bodies could expose siliceous pathways and allow more exposure towards an amorphous region, which results in better hydrolysis performance (Omar *et al.* 2014).

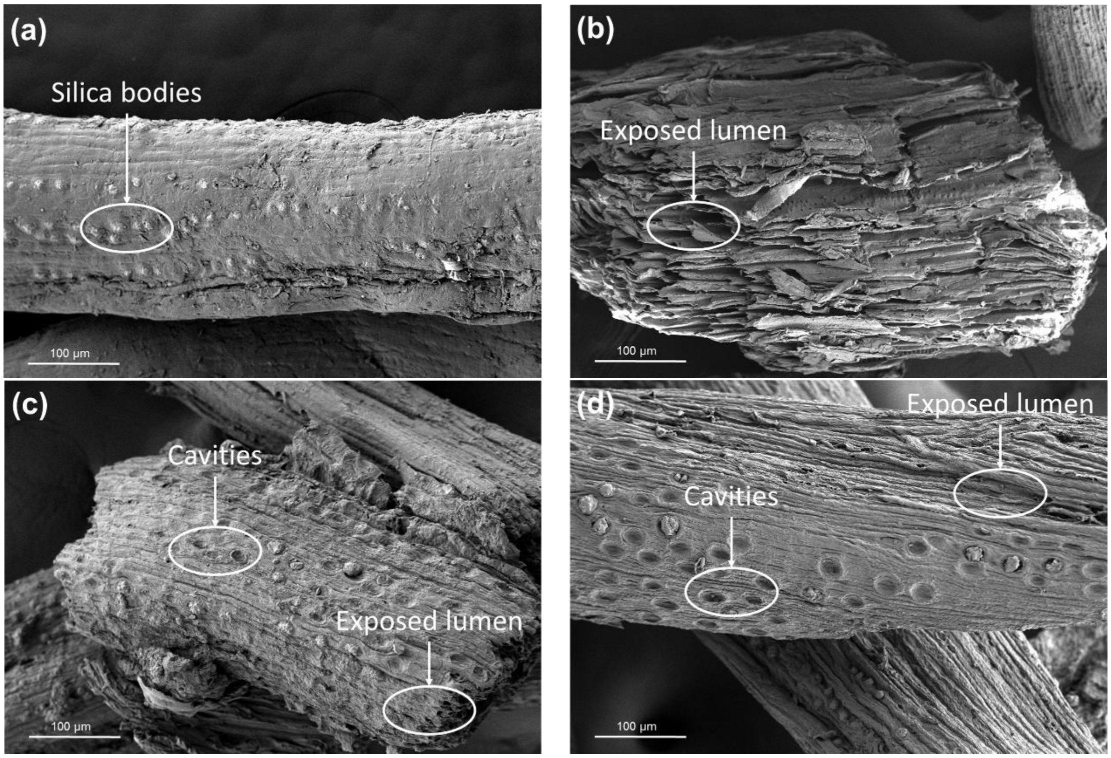

Fig. 3. Surface morphology of (a) raw and delignified EFB by Fenton reagents of (b) Fe(III), (c) Fe_3O_4, and (d) *in situ* reaction of Fe(0) (Fenton reagent: 8 mg/L; EFB fibers: 1 g; H_2O_2: 200 mmol)

In determining functional groups changes, the FTIR spectra of the raw EFB fibers was consistent with the content of lignocellulosic materials, including the presence of carbonyl and carboxyl, with the stretching of 1700 cm^{-1} and hydroxyl groups with a broad band of 3600 cm^{-1} to 3000 cm^{-1} (Fig. 4(a)). The diminishing of bands at 1700 cm^{-1} was clear after the oxidation process due to the loss of C=O through the conjugation of acids, aldehydes, and ketones with C=C bonds (Kent *et al.* 2015). Another distinct disruption of the two main peaks at 1220 cm^{-1} and 1270 cm^{-1} were attributed to C-OH and C-O stretches in $O\text{-}CH_3$ of guaiacyl and syringyl units of lignin (Gnedenkov *et al.* 2015).

Additionally, the crystallinity of the oxidized fibers was monitored through the XRD pattern from Fig. 4(b). The region of the lignocellulosic component for raw EFB fibers shows a peak position of (002) and (040) (Chin *et al.* 2013). Thus, the peak at (040) of oxidized EFB fibers was noticed to be slightly diminished. For further observation, the crystallinity index (CrI) value are calculated using (Park *et al*. 2010):

$$\text{CrI} = \frac{I_{max} - I_{min}}{I_{min}} \times 100\% \tag{16}$$

The crystallinity of oxidized EFB fibers by H_2O_2 showed a reduction from raw EFB fibers by 38.9% to 25.8%. In lignocellulosic material, oxidation reaction will act as amorphous components removal especially from lignin, hemicellulose, and amorphous cellulose, and resulted the structural swelling of EFB fibers (Gharpuray *et al.* 1983).

However, the assistance by Fenton reagents only showed slight increment on crystallinity which rose approximately 1% to 2 % and provided additional ruptured in amorphous region of EFB fibers, which physically can be observe from Fig. 3(b) and 3(c).

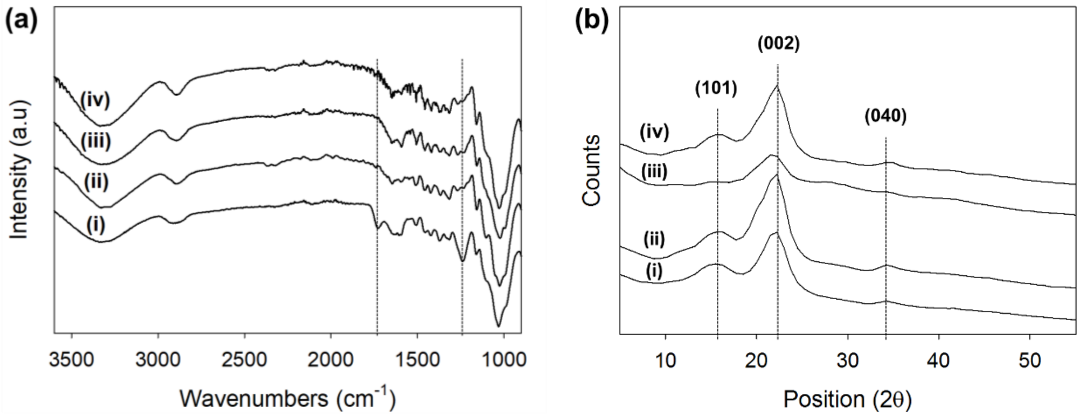

Fig. 4. Characterization of (i) raw and delignified EFB fibers in assisted Fenton reagents [(ii): Fe(III), (iii): Fe(0), (iv): Fe_3O_4] on (a) FTIR spectra and (b) XRD (Fenton reagent: 8 mg/L; EFB fibers: 1 g; H_2O_2: 200 mmol)

CONCLUSIONS

1. In general, *in situ* Fenton oxidation improved the efficiency of the H_2O_2 oxidation. The delignification of EFB fibers increased to 19.4% and 24.0% more with Fe(III) and Fe_3O_4, respectively.
2. Although the *in situ* formation of Fe(0) showed 4.6% less efficiency towards the delignification process, the oxidation performance was comparable even without the existence of H_2O_2 in the system.
3. The delignification process, with the removal of lignin in EFB fiber, was confirmed by FTIR observations. This was supported with the increment of crystallinity for oxidized EFB fibers, which can be attributed to the degradation and removal of amorphous component.
4. In addition to the lignin degradation, the oxidation process concurrently dislodged silica bodies and disrupted the structure of EFB fibers. Together with delignification and simultaneous silica removal, the improvement of an oxidation treatment could be a viable method in promotion of higher yield for the lignocellulosic biorefinery process.
5. Furthermore, the recovery of the Fenton reagents was manageable with possible desorption relative to the uptake of Fenton reagents and feasible for the stage's process, especially for Fe_3O_4, which had magnetic properties.

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