A non-catalytic process to produce levulinic acid from the flesh fruits of *trembesi* (*Samanea saman*) using a subcritical water

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Abstract. Levulinic acid (LA) has been identified as a promising green, biomass-derived platform chemical. A non-catalytic method to produce LA from the flesh fruits of trembesi (*Samanea saman*) using subcritical water has been performed. Effect of reaction time and operating pressure under CO₂ atmosphere on the conversion of glucose and yield of LA has been investigated. The conversion of glucose was found to be affected by the reaction time rather than operating pressure. The longer reaction time from 30 to 480 min. increased the conversion of glucose from 87.72 ± 1.57% to 98.74 ± 1.76% or increased by 12.56% at 200 °C, 4 MPa and ratio of flesh fruit and water of 1/8 (g/ml). Increasing the operating pressure from 4 MPa to 8 MPa, only increased the conversion of glucose by 4.67%. The yield of LA was found to be affected by the reaction time and operating pressure. The yield of LA was increased by 61.88% by increasing reaction time from 30 to 240 min, while extended further reaction time to 480 min. the reduced yield of LA by 24.05%. The highest yield of LA (22.28 ± 0.04%) can be achieved in 240 min at 200 °C and 8 MPa. The result showed that LA could be obtained by a green method.

1. Introduction

Levulinic acid (4-oxopentanoic acid or γ-ketovaleric acid) is an organic compound with short-chain fatty acids containing carbonyl group of ketones and carboxylic acids. Levulinic acid is an important chemical platform for the production of various organic compounds. It can be used for the production of polymers, resins, fuel additives, flavours and others high-added organic substances. This chemical can be produced through several routes [1] and one of the most promising processes is the dehydrative treatment of biomass or carbohydrate with various kinds of acids. Trembesi (*Samanea saman*) has been used as animal feed because the organic compound in the trembesi was high. The carbohydrate in the trembesi composed of cellulose (9.77%), hemicellulose (10.53%) and other glucose (10%), that is indicated to be used as the feedstock to produce levulinic acids [2]. Levuliniv acid (LA) can be used as a precursor, substances or chemical materials as a feedstock of process products in the industry. Several processes have been continuously developed to dissociate cellulose and hemicelluloses from lignocellulosic biomass. The processes commonly used are dilute acid, enzymatic hydrolysis, and ionic liquids [3,4]. Each one of these processes presents some advantages and drawbacks, which influences the feasibility of hydrolysis of lignocellulosic biomass at an industrial scale. Diluted acid
hydrolysis affects the subsequent fermentation due to the high levels of inhibitors produced during the reaction; ionic liquid has a high cost and complex synthesis process, which restrict its use in industrial applications [5], and enzymatic hydrolysis presents high cost and high reaction time [6]. Therefore, special attention has been focused on the development of more sustainable technologies to improve biomass processing [7].

Subcritical water hydrolysis is being used as an efficient alternative for dissociating lignocellulosic biomass. A wide variety of biomass from agro-industrial manufacture is completely processed into fermentable sugars and bioproducts by subcritical water hydrolysis treatment within a few minutes. Therefore, Subcritical water hydrolysis is a promising technology because it has a potential for breaking down biopolymers of hemicelluloses or cellulose into simple sugars like xylose and arabinose and small molecules for downstream fermentation [8]. Depending on the processing conditions and biomass employed, the concentration of compounds (oligosaccharides, monosaccharides, acetic acid, and sugars decomposition products) in the liquid phase may differ [9]. The use of CO₂ as pressurizing gas can be increased the acidity of the water because dissolved CO₂ reacts with water to produce carbonic acid H₂CO₃ [10].

Furthermore, to the best of our knowledge, none study could be found in the main database that reported the integrated evaluation of subcritical water hydrolysis of trembesi (flesh fruit) on yield in the hydrolysed liquid solution. Therefore, the focus of this work was to obtain fermentable sugars (e.g., glucose) and bioproducts (e.g., levulinic acid) from such biomass using subcritical water hydrolysis as a green technology.

2. Methods

2.1 Materials
Trembesi (Samanea saman) was obtained from Surabaya, East Java. CO₂ gas and demineralized water were obtained from a commercial source in Surabaya. 3-5 Dinitrosalicilic acid 98%, glucose 99.5%, and levulinic acid 97% were obtained from Sigma Aldrich. Potassium Sodium Tartrate 99% was obtained from Merck.

2.2 Production of Levulinic Acid (LA) using Subcritical Water
At first, the trembesi pod was sorting to get a good quality of trembesi pod (golden brown). After that, peel the trembesi pod to separate the flesh from others. The flesh of trembesi was kept in the refrigerator to maintain the quality and to avoid their damage. The water content and their sugar composition were analyzed before production of LA from the flesh fruit of trembesi. The water content, glucose and fructose contents in the flesh fruit of trembesi are 21.00%, 12.12%, and 9.90%, respectively. In general, the flesh (5 g) was dissolved in demineralized water (40 mL), the solution was subjected to the subcritical reactor and CO₂ gas is used as a pressurizing gas. The experiment was carried out by the desired variables, the reaction time (t) = 30, 60, 120, 240 and 480 min. at T = 200 °C, P = 40 bar, and the ratio of the flesh fruits of trembesi and water = 1/8 (g/mL). Variable of the operation pressure (P) = 40, 50, 60, 70, and 80 bar at T = 200 °C, t = 240 min. and the ratio of the flesh fruits of trembesi and water = 1/8 (g/mL). After the process was done, turn off the heater and cool down suddenly and let it stand at room temperature. The liquid phase was separated from others by filtration, and it was analyzed by high-performance liquid chromatography (HPLC). The experimental setup for LA production using subcritical water can be seen in Figure 1. The conversion of glucose and yield of LA were calculated according to equations (1) and (2), respectively.

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\text{Glucose conversion(%) } = \frac{\text{initial mole of glucose } - \text{final mole of glucose}}{\text{initial mole of glucose}} \times 100\%
\]  

\[
\text{LA Yield(%) } = \frac{\text{mole of LA}}{\text{initial mole of sugar}} \times 100\%
\]
3. Results and Discussions
Non-catalytic biomass decomposition is an environmentally friendly process with high efficiency and it’s suitable for application on an industrial scale. In this work, the production of levulinic acid (LA) was carried out using subcritical water and pressurized with CO$_2$ gas. CO$_2$ gas has another role in this work; it can replace the role of the acid catalyst due to CO$_2$ dissolved in water dissociated into H$^+$ and HCO$_3^-$, and HCO$_3^-$ dissociated further into H$^+$ and CO$_3^{2-}$. Effect of operation pressure on H$_2$O-CO$_2$ composition in the liquid phase and pH are showed in Figure 2 and 3. The atmosphere of acidic conditions can be achieved in the CO$_2$-water system in isothermal by increasing the pressure resulting in a decrease in pH due to mole fraction of CO$_2$ dissolved in water were increases [11].

To predict the solubility of CO$_2$ in the water at 200 °C at the function of pressure (4, 5, 6, 7, and 8 Mpa) by applying the two-liquid non-random thermodynamic model (NTRL) which is the most commonly used to correlated data [12]. The mole fraction of CO$_2$ in the liquid phase was increased with increasing pressure; the mole fraction of H$_2$O in the liquid phase initially decreases rapidly with increasing pressure to the constant value at high pressure [13]. It was caused by the solubility of H$_2$O which is decreases with increasing temperature, but increase sharply with increasing pressure to saturated pressure and there will be little change thereafter [13]. According to The formula above showed the pH at different temperatures and mole fractions of CO$_2$ as the pressure increased, that was indicated the amount of CO$_2$ dissolved in water also increases [14].

The relationship between the mole of the CO$_2$ fraction and the pH of the operating conditions is inversely proportional when the larger mole of CO$_2$ fraction was added, the smaller pH of the operating condition was formed. The pH value in the CO$_2$-water system will decrease with increasing pressure and the pH value will not decrease too much at high temperatures.
In this study, it was expected that glucose reacts in large quantities, which can be associated with the main product (LA). Effects of reaction time and pressure on the conversion of glucose are shown in Figures 4 and 5. The data showed that the highest amount of residual glucose was obtained at the reaction time for 30 min, which was 8.136 g/L and decreased with increasing reaction time. The lowest residual glucose was found at reaction time for 480 min, 0.833 g/L. In general, the amount of glucose remaining decreases with increasing reaction time, this is due to the presence of decomposed or degraded glucose at high temperatures between 180 - 220 °C [15,16,17]. As a consequence, the lowest conversion was achieved at the reaction time of 30 min. and the conversion increases with increasing of reaction time. The highest conversion obtained by 96.25% in the reaction time of 480 min. and this result is following the previous result [18], where the length of reaction time at high temperatures between the range 180-220 °C and the pressure of 10 MPa, the glucose conversion will be higher.

Glucose was easily decomposed into 5-hydroxymethylfurfural (HMF) and LA (Qi et al 2008). Decomposition of glucose has been carried out in 180-200 °C using subcritical water under a pressure of 10 MPa and found that the main products are HMF and LA. In general, reaction time and temperature have an important role in the conversion of glucose into derivative products. Decomposition of glucose at a temperature of 180-200°C and a pressure of 10 MPa results the glucose conversion of 35.2% in 180 min. at 180 °C and the highest conversion of 95.8% was obtained for 90 min. at temperature 220 °C [18].

The influence of operating pressure on the results of glucose conversion can be seen in Figure 5, the conversion was increased in increasing of operating pressure. The highest conversion of glucose was obtained at P= 8 MPa (97.69%).

![Figure 4. Effect of reaction time on glucose conversion. Operation condition: T = 200 °C, P = 40 bar, and ratio of ratio of the flesh fruits of trembesi and water = 1/8 (g/mL)](image)

![Figure 5. Effect of operation pressure on glucose conversion. Operation condition: T = 200 °C, t = 240 min, and ratio of ratio of the flesh fruits of trembesi and water = 1/8 (g/mL)](image)

The correlation of reaction time and pressure on the yield of LA are shown in Figures 6 and 7. The yield of LA increased with increasing reaction time [15]. LA yield of 7.72% was obtained at reaction time for 30 min., and subsequently, the yield of LA increased with increasing reaction time. However, the highest yield of LA was obtained at the reaction time for 240 min. (12.26%). This is because glucose decomposed not only to LA but also to other derivative products such as 5-hydroxymethylfurfural (HMF) and due to the formation of humin which is a side effect of the hydrolysis reaction.

The highest yield of LA was found at a temperature of 200 °C and reaction time for 240 min. at a constant pressure of 4 MPa, therefore to increase the yield of LA carried out by increasing the pressure up to 8 MPa. It was found that the highest yield of LA (22.25%) was obtained at high pressure of 8 MPa.
4. Conclusions
The conversion of glucose increases with increasing reaction time and temperature. In line with this, the yield of LA increases with increasing the operating pressure. The highest yield of LA was obtained at a pressure of 8 MPa which is 22.25% with the conversion of glucose of 97.69%.

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