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Effect of pre-treatment processes to ferulic acid extraction from banana stem waste

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Abstract

The present of ferulic acid (FA) in plants attract the attention of many researchers, especially in food, pharmaceutical and cosmetic industries. Banana stem waste (BSW) was generated from banana plantation. In this study, FA was extracted from BSW by using sugarcane press machine. Effect of two pre-treatment processes to FA extraction were studied namely the ultraviolet (UV) light pre-treatment and heating of BSW before extraction. Response surface method was utilized to design the experiment and to analyze the influences of both pre-treatment processes. The selected ranges for both pre-treatment processes were as follows: UV light (with or without) and heating temperature (25°C-90°C). The FA was analyzed by using high performance liquid chromatography (HPLC) and FA yield ranging from 0.0010mg/g to 0.1204 mg/g. The analysis of the results showed that heating BSW before extraction increased the FA yield however UV light pre-treatment did not give significant effect to FA extraction.

Keywords: mechanical extraction, agricultural waste, ferulic acid, banana stem waste.

INTRODUCTION

Generally, phenolic compounds in plant are bonded to dietary fiber, proteins or two sugars to form the complex structures. The bond needs to be broken in order to release the phenolic compounds during the extraction process. Pre-treatment is often used on plants biomass before the extraction in order to increase the extraction rate. The biomass pre-treatment depended on the combination of several parameters that effected biomass such as temperature, acidity and duration of pre-treatment (Agbor et al., 2011). However, process for the pre-treatment of biomass also one of the challenging process. It is use to maximize the extraction yield while minimizing the cost.

Photochemical pre-treatment is the treatment that initiated by the absorption of energy in the form of light such as ultraviolet (UV) light. UV light is an invisible form of electromagnetic radiation that has a shorter wavelength than the light humans can see. UV light carries more energy than visible light that capable to break the bonds between the atoms and molecules. It also is altering the chemistry of materials when exposed to it. According to Kovacs and Keresztes (2002), the UV photon destroyed the chemical bonds and causing the photochemical reaction. It also damaged the deoxyribonucleic acid (DNA) and physiological processes of the organisms (Hollosy, 2002). UV light stimulated the formation and accumulation of certain phenolic compounds in plants (Treutter, 2010). From studies conducted by Eichholz et al. (2011), the amount of phenolic compounds in blueberries was increased when exposed to UV light. The UV light mediated the stress to increase the phenolpropanoid metabolism that leading to an acceleration of the biosynthesis of phenolic compounds. Interdonato et al. (2011) also found the phenolic compounds in plants increased when exposed to UV light. The phenolic compounds such as FA, *p*-CA and caffeic

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acid in tomatoes showed approximately 20% higher under UV transmission compared when it protected from the UV light (Luthria et al., 2006).

According to Roy et al. (2009), the availability of phenolic compounds was affected with the variation of temperature. The pre-treatment at certain temperature commonly used in the juice manufacturing to obtain higher juice yield and reduced the extraction time (Grimi et al., 2011). The present of heat during pre-treatment can enhanced the recovery of phenolic compounds (Silva et al., 2007). The heat can soften the plant tissues and weaken the phenol-protein and phenol-polysaccharide interactions (Al-Farsi and Lee, 2008). It also can break the polyphenol bounds and cellular constituents of plants cells (Thoo et al., 2010). In the study conducted by Eshtiaghi and Yoswathana (2012), sugarcane was used to study the effect of temperature to sugar extraction. The sugarcane was immersed in water at different temperatures before the extraction process. The extraction of sugar was increased with the increasing temperature. The objective of this research was to study the effect of pre-treatment processes in ferulic acid (FA) extraction from BSW. One factor at time (OFAT) analysis and response surface method (RSM) were used to evaluate the significance of different pre-treatment processes that consisted of ultraviolet (UV) light and temperature.

MATERIALS AND METHODS

Banana stem waste (BSW)

BSW was used as raw material in the extraction of FA. BSW was collected from banana plantation. The BSW sample was obtained from banana plantation in Kuantan, Pahang. The banana trees were cut down during the harvesting process of matured fruits.

Sugarcane press machine

Sugarcane press machine is one of the mechanical extractor that commonly used for the extraction of juice (Zahari et al., 2012). In this study, BSW was extracted by using electric sugarcane press machine as showed in Figure 1. The model of this machine was KR3176 with the rotating speed was 80 rpm. This semi-automatic machine consists of three stainless steel roller to extract the juice from BSW. The top or driven roller acted as pinion with the lower rollers. The lower rollers function as feed and discharge roller. The rollers were grooved transversely to their axis with V-shaped grooves to increase the surface area of the rolls during pressing.

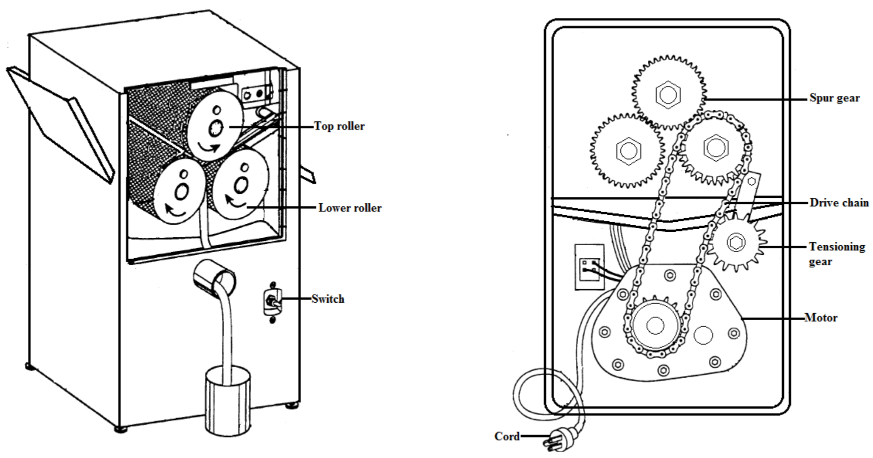

Figure 1. Schematic diagram of sugarcane press machine

BSW extraction method

The extraction of BSW was carried out by using sugarcane press machine. The BSW was extracted after the pre-treatment process. The BSW was pressed pass through the roller pressers and the baggase was thrown away. For the extracted BSW juice, it was collected and centrifuge at 15,000 rpm for 15 min at 4 °C (Thermo Fisher Scientific, NC, USA). After centrifuged, the extracted banana stem juice (EBJ) was filtered using Whatman No. 41 filter paper (20–25 μm) to remove all the remaining debris. Then, the juice was stored at -20 °C before the analysis.

Experimental setup for one factor at time (OFAT) analysis

The UV light pre-treatment of BSW was done to increase the availability of FA during extraction. The amount of phenolic compounds in plants was affected with the exposure of UV light (Treutter, 2010). In this study, BSW were cut into rectangular shaped with the size approximately 2.5±0.5 cm long and 2.5±0.5 cm width. 100 g of BSW was put in the tray and placed in the laminar air flow. The BSW was exposed into UV light for 30 min. After the pre-treatment, the BSW was extracted by using sugarcane press machine.

The pre-treatment temperature of BSW was performed to increase the FA yield during the extraction. It was done by manipulating the temperature during pre-treatment. In this study, water bath was used to control the pre-treatment temperature. 100 g of BSW was weighed and put in the beaker. The beaker was immersed in water bath for 30 min at different temperatures between 20 to 90 °C. After the pre-treatment, the BSW was extracted by using sugarcane press machine.

Experimental setup for response surface method (RSM) analysis

For analysis using RSM, the level of UV light pre-treatment and pre-treatment temperature were (Yes and No) and (25°C, 57.5 °C, 90 °C) respectively. The pre-treatment by using UV light was controlled by exposing the sample in laminar air flow for 30 minutes. The temperature was controlled by using water bath. In this study, BSW was immersed in water bath for 30 minutes. The BSW was extracted by using sugarcane press machine. The EBJ was collected and stored at -20 °C before analyzed.

Analysis of ferulic acid (FA) by using HPLC

The concentrations of FA in samples were analyzed by using high performance liquid chromatography (HPLC). The analytical HPLC system employed consisted of an Agilent 1100 HPLC equipped with a diode array detector (DAD). The HPLC pumps, column oven, auto sampler, and DAD system were monitored and controlled using the HP Chem Station computer program. The separation and analysis of FA were carried out using a 5 μm Zorbax SB-C18 (250 mm x 4.6 mm, Agilent Technologies, Palo Alto, CA) (Buranov and Mazza, 2009). The temperature of the column was set at 30 °C. The mobile phase consists of water (eluent A) and acetonitrile (eluent B). The gradients of mobile phase were 45 % of water and 55 % of acetonitrile. The mobile phase was pumped with the flow rate at 1.0 ml/min by a quaternary gradient pump (G1311A Quat Pump, Agilent). The injection volume was 10 μl. The FA yields in the samples were identified by comparing the retention times with the FA standards. It was determined under analytical conditions and quantified by the external standard method. (Michaelevski et al., 2010)

RESULTS AND DISCUSSION

One factor at time (OFAT) analysis

BSW was exposed to UV light for 30 min in laminar air flow. Figure 2 shows the FA yield without and with UV light pre-treatment. From the figure, the FA yield was increased when BSW was exposed to UV light. The FA yield when the BSW was exposed into UV light was 0.0149 mg/g while without exposed into UV light was 0.0138 mg/g. It showed the FA yield was only 8.12% higher when the pre-treatment with the UV light was done to the BSW compared to without the pre-treatment. UV light can stimulated the formation and accumulation of certain phenolic compounds in the plants. This was due to the induction of phenylalanine ammonia-lyase (PAL) activity by UV light which mediated the synthesis of the coumaryl CoA (Bakhshi and Arakawa, 2006). Besides that, the photon from UV light also provided enough energy to destroy the chemical bonds in cell walls (Kovacs and Keresztes, 2002).

The pre-treatment temperature of BSW was done to increase the FA yield during the extraction. In this study, the pre-treatment temperature of BSW was done at different temperature between 20 to 90 °C. Figure 4.2 shows the FA yield was increased from 0.0140 to 0.0153 mg/g when the temperature increased from 20 to 30 °C. So, the availability of phenolic compounds increased with the increasing of temperature (Wang et al., 2008). However, the FA yield was decreased when the temperature increased from 30 to 90 °C. The FA yield was decreased from 0.0153 to 0.0072 mg/g. The used of high temperature increased the oxidation of phenolic compounds which decreased the yield during the extraction process (Dai and Mumper, 2010). According to Liyana-Pathirana and Shahidi (2005), the decomposition of phenolic compounds which was already mobilized at low temperature was increased when the temperature increased beyond 25 °C. Besides, it also capable to break down the phenolic compounds that still remained in the plants matrix (Chan et al., 2009).

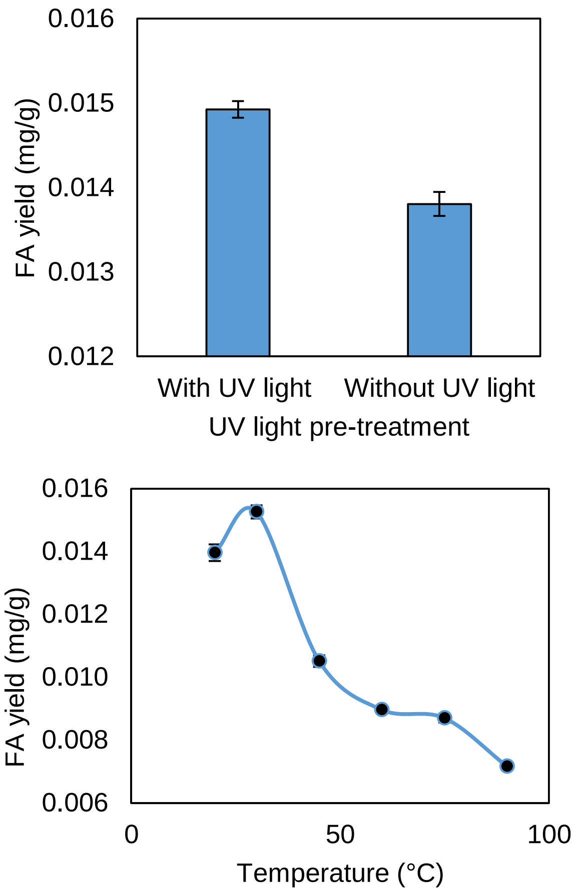

Figure 2. Results from OFAT analysis of two different pre-treatment processes

Response surface method (RSM) analysis

One of the aspects that were studied in the RSM analysis was the main effect. This analysis was studied in order to determine the factors that most contributed to the mechanical extraction of FA from BSW. FA yield from this analysis was between 0.0010 to 0.1204 mg/g Table 1 shows the contribution of each main factor to FA extraction. From Table 1, pre-treatment temperature) proved to be most contributing factor with 23.59%. This seems legit as temperature play important part for the pre-treatment of BSW. In BSW, FA was attached to the lignin and hemicellulose via ester and ether bonds as bridges and formed LCC structure (Sun et al., 2002). The ether bonds between lignin and hydroxycinnamic acid were broken by using steam, hot water and dilute acid (Buranov and Mazza, 2008). So, heat was used in this study to increase the FA yield by breaking the ether and ester bonds. From the study conducted by Vanbeneden et al. (2008), the released of FA yield increased when the pre-treatment temperature increased due to the additional of chemical hydrolysis by enzyme activity. But, when the temperature increased beyond certain values, it may promote the decomposition of phenolic acids. It was supported by Mcmurrough et al. (1996) that stated when the temperature was increased above 65 °C, the FA yield decreased due to the enzyme that released FA from the bonds almost completely denatured. The pre-treatment at different temperature influenced the released of phenolic acids from the cell (Xu et al., 2005). This indicated that this factor was important to determine the phenolic acids. The UV light pre-treatment showed low contribution on FA extraction with only 0.22% contribution. The results from RSM analysis showed similarity to OFAT analysis where increased temperature will decreased the FA yield (Fig. 2 and Fig. 3) and UV light pre-treatment was not a significant factor in BSW extraction process.

Table 1. The contribution of main factors to FA extraction

Factor	Contribution (%)
UV light pre-treatment	0.22
Pre-treatment temperature	23.59

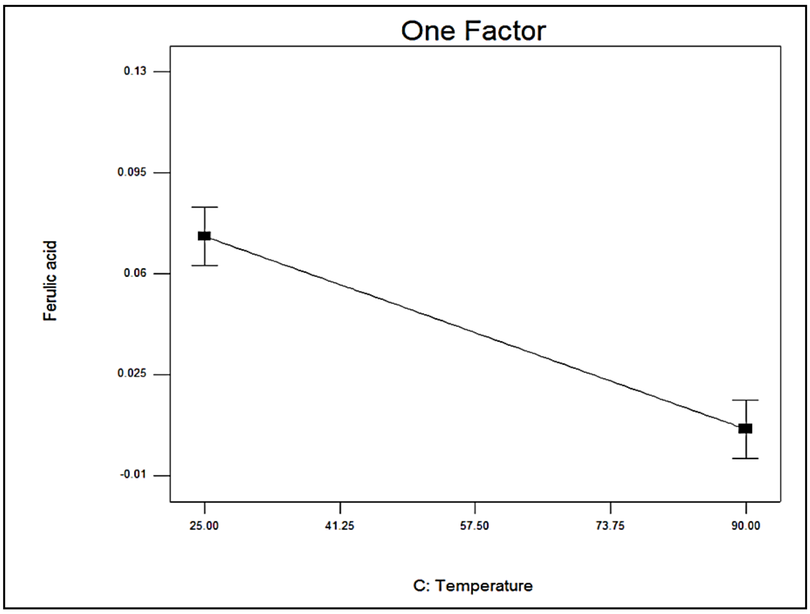

Figure 3. Results from RSM analysis on effect of temperature to BSW extraction

CONCLUSIONS

The following conclusions can be drawn from the study:

- Two types of data analysis has been utilized to study the effect of pre-treatment processes to FA extraction from BSW namely one factor at time (OFAT) and response surface method.
- The analysis was involving two pre-treatment processes which were UV light and temperature.
- The FA was analyzed by using HPLC and FA yield ranging from 0.0010 mg/g to 0.1204 mg/g.
- The analysis of the results showed that heating BSW before extraction increased the FA yield however UV light pre-treatment did not give significant effect to FA extraction.

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