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Ascorbic Acid Content and Proteolytic Enzyme Activity of Microwave-Dried Pineapple Stem and Core

Nurul Asyikin Md Zaki^{*,a,b}, Norazah Abd Rahman^{a,c}, Norashikin Ahmad Zamanhuri^{a,c}, Syafiza Abd Hashib^{a,c}

^aFaculty of Chemical Engineering, Universiti Teknologi MARA, 40450 Shah Alam, Selangor, Malaysia.

^bGreen Technology and Sustainable Development, Community of Research (CoRe), Universiti Teknologi MARA, 40450 Shah Alam, Selangor, Malaysia.

^cFrontier Materials and Industrial Application, Community of Research (CoRe), Universiti Teknologi MARA, 40450 Shah Alam, Selangor, Malaysia.

asyikin6760@salam.uitm.edu.my

Solid wastes generated from the industrial processing of pineapple are favourable raw materials for obtaining antioxidants and bioactive compounds because of their low cost and the possibility of reducing environmental problems caused by waste disposal. Hence this study aims to investigate the microwave drying characteristics of pineapple waste and its effect on the ascorbic acid content and proteolytic enzyme activity. Pineapple stem and core were subjected to microwave drying process at three different power intensities (380 W, 530 W and 680 W). Dried samples were then analysed for ascorbic acid concentration and proteolytic enzyme activity. Results show that ascorbic acid concentration in the stem sample dried at 530 W was the highest. The proteolytic enzyme activity was found to be the highest in pineapple core dried at 380 W. As the power level increased, more proteolytic enzyme denaturated and resulted in the reduction of enzyme activity. Microwave drying could remarkably preserve both pineapple stem and core while retaining the ascorbic acid content and proteolytic enzyme activity of dried pineapple wastes.

1. Introduction

Pineapple is mainly exported into varieties of processed products such as canned fruits, fresh juices, concentrates and jams besides freshly consumed. The production of pineapple have increasingly generated substantial wastes consist of pineapple stem, core, leaves, peels, crown and also residual pulp. The generation of great waste by this tropical fruit is mainly due to assortment and removal of parts that are unfit for human consumption. In addition, Upadhyay et al. (2010) found that huge generation of pineapple wastes have incredibly increase the dump site area and cause serious environmental problem as these wastes interfere with BOD and COD of natural systems. Pineapple waste could be exploited more for its natural content such as antioxidants, vitamins, phenolic contains, enzymes and many others nutritional value. Ascorbic acid and proteolytic enzymes are among the important nutrients that can be preserved in pineapple stem and core using drying method.

Drying is a technique employed to improve the stability of the product and decreasing the water activity thus slower the microbiological activity in order to reduce physical and chemical reactions that may occur prior to storage (Layla, 2013). Various drying methods are used in the drying of fruits and vegetables. Microwave drying is one of the methods that have gained interests among researchers as an alternative drying technique for many agricultural products due to increasing concerns about product quality and production costs (Gursoy et al., 2013).

Microwave drying offers several advantages over others conventional drying including increase operation speed, lower energy usage, accurate process control and fast start-up and shut down of the equipment (Datta and Anantheswaran, 2001). Microwave drying may preserve and improve product quality such as aroma, taste, rehydration ability, microbial stability, nutritional value and overall appearance as the detrimental effects

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of high temperature declined during drying period (Maskan, 2001). The drying rate could also be increased substantially as the electromagnetic wave directly infiltrated the material to be dried and the volumetric (from the inside out) heating process occurs throughout the entire material. Quick evaporation of water occurs as the energy absorption by water molecules from electromagnetic wave is rapid (Mullin, 1995). Microwave drying also only requires small floor space as the unique heating process makes it possible to control or compact the design size. This might be a good advantage for large scale production such as in industries as it is cost-effective.

Microwave drying has been proven to be suitable for retention of ascorbic acid and other bioactive compounds in agricultural products such as in banana (Maskan, 2000), potato (Khraisheh et al., 2004) and apricots (Karatas and Kamisli, 2007). Hence this study aims to investigate the microwave drying characteristics of pineapple waste and its effect on the ascorbic acid content and proteolytic enzyme activity.

2. Materials and methods

2.1 Materials and sample preparation

Fresh pineapples were obtained from nearest market around Shah Alam, Malaysia. Fresh pineapples were washed, peeled and cut into sample size of 2 cm length and 1 cm thick. The stem and core of the pineapple were used as samples. All fresh samples were stored at low temperature (4 ± 0.5 °C) in refrigerator until the drying process. On the other hand, the potassium phosphate, acetonitrile and the L-ascorbic acid were used in the analysis to determine the concentration of ascorbic acid in dried pineapple samples. Furthermore, casein, L-tyrosine, trichloroacetic acid, Folin - Ciocalteu's phenol reagent, sodium carbonate, sodium acetate, calcium acetate and phosphate were used in the analysis of proteolytic activity.

2.2 Microwave drying process

Drying process was performed in microwave oven (Samsung GE71M, Korea) with the following specifications. It has a rated power output of 680 W (50 Hz and 240 V). The microwave oven has the capability of operating at three microwave stages which are low power, medium power and high power ranges from 380 W to 680 W. It consists of rotating glass plate with diameter of 254 mm and also equipped with digital clock for time adjustments.

Drying process was carried out at three different microwave powers being 380 W, 530 W and 680 W. The samples to be dried were 50 ± 0.10 g each from the core and stem of pineapples. The moisture content was determined by weighing the samples at interval time using analytical balance (Mettler Toledo) with 0.01 g precision. All drying and weighing process were continued until the moisture content of the samples decreased to 0.1 ± 0.01 g on dry basis.

The experimental moisture content data was determined using Eq(1) where Eq(1) can be further expanded to Eq(2) (Bi et al., 2015) in which MC (X_t) is the moisture content at time t; W is weight of wet solid in total water plus dry solid (kg water) and W_s is the weight of dry solid (kg dry solid).

$$MC(X_t) = W/W_s \tag{1}$$

 $MC(X_t) = (W - W_s)/W_s$ (2)

Drying rate as in Eq(3) (Bi et al., 2015) was determined using the moisture content data from Eq(2).

Rate of drying = $dX/dt = (X_{t+dt} - X_t)/dt$

where X_{t+dt} is the moisture content at time t+dt and X_t is the moisture content at time t, which is the drying time.

(3)

2.3 Analysis on ascorbic acid content

The ascorbic acid content in the sample was determined using HPLC (Perkin Elmer) based on method by Ramallo and Mascheroni (2012) with C18 150 × 4.6 mm column. Analytes were test in a single run, using isocratic elution with a mobile phase consisting of acetonitrile (Solvent A) and buffer (potassium diphosphate 0.02 M, pH 2.5 by phosphoric acid) (solvent B). The gradient profile (A:B) used was 10 % : 90 % at a flow rate of 1.0 mL/min. Detection of L-ascorbic acid was carried out at wavelength 254 nm. Identification and quantification were carried out by comparison of the retention time (min) and the peak area of the sample with those of a standard reference. Direct injection of pure L-ascorbic acid standards was done by dissolving L-ascorbic acid (A-0278, Sigma) in the prepared buffer solution. Different concentration (5 ppm, 10 ppm, 15 ppm and 20 ppm) of L-ascorbic acid standards were prepared and the reference peak area was obtained by injecting 0.4 μ L of each standard solution.

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2.4 Analysis on proteolytic enzyme activity

Dried pineapple stem and core samples were analysed for their proteolytic enzyme activity based on method described by Ketnawa et al. (2012). The samples were mixed with the solution of casein together with the trichloroacetic acid and later on undergo incubation for 30 min. After the incubation, the samples were filtered to new vials using 0.45 µm polyethersulfone syringe filter. Next, Folin reagent and sodium carbonate were added to the vials with samples. After mixing, the samples undergo another 30 min of incubation and the optical density of the samples were measured using the spectrophotometer at wavelength of 660 nm.

3. Results and discussion

3.1 Drying characteristics

Drying curves for stem and core were generated on dry basis as shown in Figure 1 and Figure 2.



Figure 1: Drying curve of pineapple stem at various microwave power



Figure 2: Drying curve of pineapple core at various microwave power

Obviously, drying time reduced with the increasing microwave power levels from 380 W to 530 W and lastly to 680 W. Based on Figure 1, the time required to reduce the moisture content of the pineapple stem from 6.0 kg H_2O/kg dry solid to 0.1 kg H_2O/kg dry solid varied between 8 min to 18 min subjected to the microwave power level. The shortest drying time to reduce the moisture content of pineapple stem occurred at 680 W with 56 % and 33 % reduction compared to 380 W and 530 W. Meanwhile, the time required to achieve desired moisture content for pineapple core was between 6 min to 22 min, as shown in Figure 2. Additionally, microwave drying of pineapple stem and core generally shows steeper drying curve for the first 5 min at all power levels. The

observed drastic or sudden drying curve at the initial stages of microwave drying may be triggered by opening of the sample's structure physically which allowing rapid vaporisation and passage of water molecules (Kostaropoulos and Saravacos, 1995).

Figure 3 and Figure 4 show the effect of drying conditions on the drying rate of pineapple stem and core. All drying rate (kg H_2O /kg dry solid min) was inconsistent which consists of a period of constant drying but mostly the entire drying process occurred in the range of falling rate. The high moisture foods can be expected to have a period of constant rate drying (Maskan, 2000). However microwave drying occurred mostly at the falling rate period (Gowen et al., 2008). The drying rates observed were higher at power 680 W for each sample as expected because the drying rate was exponentially dependent on microwave power levels. In the entire samples, the higher drying rates were observed at the initial phase of drying, as at this point the samples contained very high moisture which resulted in higher absorption of heat. As the drying period progressed, the loss of moisture from the sample decreased and resulted in the reduction of drying rate.



Figure 3: Drying rates of pineapple stem at various microwave power



Figure 4: Drying rates of pineapple core at various microwave power

3.2 Ascorbic acid content

The ascorbic acid content of pineapple stem and core dried at different microwave power levels were presented in Figure 5. Significant values of ascorbic acid were detected in the stem. Increase in power level from 380 W to 530 W resulted in higher retention of ascorbic acid content. This might be due to longer drying period for microwave drying at 380 W. Many studies reported significant reduction of vitamin C with the extending drying period, such as during microwave drying of spinach (Ozkan et al., 2007), broccoli (Zhang and Hamauzu, 2004), and asparagus (Nindo et al., 2003). However, the lowest ascorbic acid content was found in

pineapple stem dried at the highest power level 680 W which is 0.69 mg ascorbic acid per 100 mL. This might be due to drastic changes in power level which can be related to high temperature that adversely affect the degradation of ascorbic acid. The highest amount was observed for pineapple core dried at microwave power level 530 W which is 0.84 mg ascorbic acid per 100 mL.



Figure 5: Ascorbic acid content of microwave-dried pineapple stem and core at various power

Ascorbic acid was only detected in pineapple core samples dried at power level 380 W. No ascorbic acid was found in core samples dried at 530 W and 680 W. Extreme moisture removal at higher power levels could mean higher microwave absorption and this might cause detrimental effect towards ascorbic acid content in the core. Haruenkit (2003) also reported on unsuccessful quantitative determination of ascorbic acid using HPLC method. Core samples contained higher amount of moisture content compared to pineapple stem. This could relate to water activity (a_w) which is one of the important parameters that affect the degradation of ascorbic acid. The relationship between moisture content and water activity is very complex and sometimes higher water activity enhances the distribution of the ascorbic acid throughout the core samples, hence other factors such as temperature, pH, light, oxygen, enzymes and metallic catalysers contributed to the degradation of the ascorbic acid (Santos and Silva, 2008).

3.3 Proteolytic enzyme activity

The proteolytic enzyme activity of pineapple stem and core dried at different microwave power levels was shown in Figure 6. The highest proteolytic enzyme activity was found in pineapple core dried at 380 W. As the microwave power increased, the proteolytic enzyme activity constantly decreased for both stem and core samples. Ketnawa et al. (2012) analysed the proteolytic enzyme activity of pineapple and reported the reduction of proteolytic enzyme activity at the highest drying temperature. The enzyme molecules were having enough kinetic energy to undergo reaction with water molecules. Thus, the structure of the enzyme started to decompose (Switzer and Garrity, 1999). For this study, the higher the microwave power, the higher chances of enzyme molecules disrupted.



Figure 6: Proteolytic enzyme activity of microwave-dried pineapple stem and core at various power

4. Conclusions

From the present work, it was concluded that microwave power levels influenced the drying characteristics of pineapple stem and core. Drying conditions such as power level and processing time influenced the ascorbic acid content and proteolytic enzyme activity of the dried samples. Increase in power level resulted in shorter

drying time. Even though higher microwave power contributed to faster drying but it also resulted in loss of ascorbic acid content and proteolytic enzyme activity from the samples. Effects of other parameters such as pH, light and oxygen should be studied to further optimize the microwave drying process of pineapple stem and core.

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