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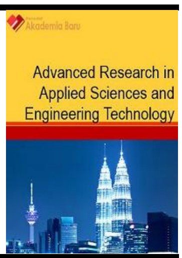

The Comparison of Bioactive Compounds and Antioxidant Activity of Fresh Pineapple and Pineapple Powder

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ABSTRACT

An antioxidant has a possibility to reduce the oxidative damage that caused by free radicals and chelating metals. The objectives of this study were to determine and compare the antioxidant and bioactive compound in fresh pineapple and pineapple powder through spray drying process with 25% maltodextrin addition and inlet temperature 130 °C. In this study, Ananas Comosus.L or pineapple (Morris type) was used and physicochemical analysis like ascorbic acid, flavonoid, phenolic content and 2, 2-diphenyl-1-picrylhydrazil (DPPH Assay) was conducted with methanol concentrations from 20% to100%. The results showed that pineapple powder was found to contain the highest ascorbic acid (32 mg AA/100g fruits) and the highest total flavonoid (79 mg QE/100g fruits). The antioxidant activity in fresh pineapple is higher than pineapple powder based on DPPH analysis at five different methanol concentrations. The highest antioxidant in fresh Morris pineapple fruits and powder was 65% and 53% respectively. In conclusion, the spray-dried pineapple powder could be a good source of natural antioxidants and profoundly increase use of pineapple in value added processing and dietary intake.

Keywords:
Ananas comosus; antioxidant; DPPH; pineapple; spray drying

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1. Introduction

Pineapples were first recorded by Europeans in 1493 on the Caribbean Island and also known botanically as Ananas comosus L. Merr. Pineapples have a wide cylindrical shape, yellow skin and a regal crown of spiny, and green leaves [5]. The fibrous flesh of pineapple is yellow in colour and has a vibrant tropical flavour that balances the tastes of sweet and tart. The fruit has more sugar content and a sweeter taste. However, the fresh fruit is not able to be stored for a long period because it will be bruised and darkened. One of the methods to prevent it from occurring, the spray drying is the best technique to be applied, so that the moisture content can be decreased.

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The most common type of pineapple in Malaysia is Morris that contains high antioxidant. Antioxidant has the possibility to reduce the oxidative damage that caused by free radicals and chelating metals. There are three types of antioxidant that can be found in the fruit that is phytochemicals, vitamins, and enzymes [1]. Mostly, the enzymes come from the protein and minerals of fruit that can be synthesized in the human body, for example iron, copper, selenium, magnesium and zinc [13]. Besides the vitamins includes in the pineapple fruit such as A, C and E, folic acid and beta-carotene that have main functionality which can also maintain immune body system [1].

Hence, this paper aims to evaluate and compare the antioxidant and bioactive compound of fresh pineapple between spray dried pineapple powder and fresh pineapple.

Experimental

Spray drying

The fresh Morris pineapple was bought from local market. Fruits were selected at the maturity stages about 20 to 40% yellowish or 120 days after flower as Figure1.

Fig. 1. Morris Pineapple with 40% Ripeness Stage

The whole pineapple fruits were washed thoroughly and rinsed three times prior, to be cut into small pieces. Then, the pieces of pineapple flesh were blended together to form the slurry using Waring blender. The slurry was filtered to get the pineapple juice. The maltodextrin (MD) was added to the juice with 25% concentration (W/W) to enhance the solid content of the powder during the spray drying process. The pineapple-maltodextrin mixture was stirred to avoid any agglomerates. The experiment was conducted using Basic SD-Plant spray drier at inlet temperature 130°C and speed pump level set at 3 [6]. The powder produced was collected at the dry product collector and weighed using an analytical weight balance.

Sample Extraction

The pineapple was skinned and cut into pieces and washed with distilled water. Then, the pieces of pineapple were blended using a Waring blender to produce pineapple slurry. The slurry was filtered and weighed in a beaker at 25 grams and 20% methanol was added. The mixture was placed in an orbital shaker at a speed 150 rpm at room temperature for 10 minutes. The extract was filtered by using sieve to get solid free extract and centrifuged with a speed 10,000 rpm for 15 minutes. The extracted solution was stored at -20°C for not more than 3 days. After that, the procedure was repeated using 40%, 60%, 80% and 100% methanol concentrations.

Ascorbic acid (AA) test:

A 5 mL of each extracted pineapple was added with 5 mL 4% metaphosphoric acid, 5 mL of 1M sulphuric acid, and 2 mL of 1% starch solution. The mixture was titrated against 0.001 M iodine solution which was diluted from standardized iodine solution. In this titration, the color of the solution in the conical flask will be changed from blue black to colorless at the endpoint.

Total flavonoid content (TFC) test:

TFC in fruits was determined using spectroscan 2600 UV/Vis spectrophotometer according to the method described by Ali *et al*., [1]. A 0.5 mL of methanol extracts, 1.5 mL of methanol, 0.1 mL of aluminium chloride, 0.1 mL of potassium chloride, 0.1 mL of potassium acetate and 2.0 mL of distilled water was prepared and mixed in an incubation at room temperature for 30 minutes. Next, aluminium chloride was used as a sample blank. The absorbance was measured at 415 nm using HITACHI spectrophotometer. Then, quercetin was used to make the calibration curve and the total estimation was carried out in triplicate and the results was calculated. The results will be expressed in mg QE/100 g fruits.

Total Phenolic Content (TPC) test:

TPC compound was determined by using the Folin-Ciocalteu's reagent method. A 0.3 mL pineapple extract was added into the tubes that contain 1.5 mL Folin Ciocalteu's reagents in water and diluted 10 times. After 10 minutes, 1.2 mL of 7.5% w/v sodium carbonate solution was added to the sample. The mixture was left at the room temperature for 30 minutes before taking an absorbance at 765 nm. The solvent was replaced as a blank sample. The TPC expressed as gallic acid equivalents (GAE) in mg/100 g of fruit juice to the different concentration of polyphenols in ranging from 0.2 to 4 mg/L.

DPPH Free Radical Assay

DPPH was conducted by preparing 10 g of each pineapple extract, then diluted to a total of 2 mL. For each of the sample, 2 mL of 0.15 mM DPPH solution in methanol was added. The mixture was left for 30 minutes before reading the absorbance at 517 nm and methanol was used as the blank sample. Then, 2 mL of solvent was prepared in the extracted sample. The inhibition percentage % of the extract was determined as Eq. (1), where A is absorbance:

$$\%\ inhibition = \left[\frac{\text{Acontrol} - \text{Asample}}{\text{Acontrol}}\right] \text{x } 100\% \quad (1)$$

2. Results and Discussion

2.1 Ascorbic Acid content in the pineapple powder and juice

Ascorbic acid (AA) is one of the most sensitive parts of vitamins in the fruits especially pineapple. That is the reason it is often used to evaluate the influences of food processing on vitamin C contents.

In this study, the spray dried powder was produced using 25% maltodextrin, 130°C inlet temperature and 3 rpm pump speed as performed by Hashib *et al*., [6].

Vitamin C is a bioactive compound found in the pineapple fruit that can be dissolved in water. The highest amount of vitamin C was obtained in orange (10.13± 0.10mg/100ml), next is apple (7.94 ± 0.13mg/100ml) and followed by pineapple (6.40± 0.18mg/100ml) as reported by Nweze *et al*., [13]. The results of AA in this study showed that ascorbic acid content of the spray dried Morris pineapple powder is higher than the fresh type as presented in Table 1.

Table 1
Comparison of ascorbic acid (AA) content

No.	Methanol Concentrations (%)	AA (mg/ 100ml (Powder)	Std Dev	AA mg/ 100ml (Fresh)	Std Dev
1	20	32	1.0	28	1.0
2	40	27.3	1.0	24	0.6
3	60	22	1.0	18	1.0
4	80	17.2	0.5	15.5	0.3
5	100	14.4	1.0	12	1.5

From previous study by Chit *et al*., [2] a higher ascorbic content was observed in Josaphine pineapple extracts compared to other extracts pineapple varieties ie Morris and Sarawak. In another study, which also investigated the ascorbic acid content of Josephine and Sarawak that comparable to that of the 'Nanglae' (6.45mg AA/100g fruit) and 'Phulae' (18.88mg AA/100 g fruit) pineapples as reported by Kongsuwan *et al*., [9].

From the previous research, the ascorbic acid of Sarawak pineapple decreased from 26.75 to 17.98 mg g-1 recording a loss of 32.79% after spray drying process (George et al., 2009). In another study by Yuris et al., 2014 an ascorbic acid content in the Morris variety (27.8 ± 1.16mg/100ml) is higher than Sarawak (15.0 ± 0.00 mg/100ml) and Josaphine (7.70 ± 0.67mg/100ml).

2.2 Antioxidant activity of pineapple powder and juice

Figure 2 - 4 shows the comparison of total flavanoid content (TFC), total phenolic content (TPC) and DPPH analysis of fresh pineapple and pineapple powder at five methanolic concentrations

In this study, total flavonoid content (TFC) was detected in both Morris fresh pineapple and pineapple powder. Figure 2 shows an analysis of TFC in pineapple powder which resulted a higher reading compared to fresh pineapple juice at five methanol concentrations from 20% - 100%. The results showed an increase in flavonoid content of spray dried pineapple over fresh pineapple extract; it could be due to an increase in the concentration of pineapple at constant temperature and pressure during spray drying [10]. The decreased of TFC in pineapple powder at 80% methanolic compared to 100% methanolic is suspected due to the spray drying process which involved heating of fruits at high temperature and heat sensitivity of the fruit ranged from 20% to 100%.

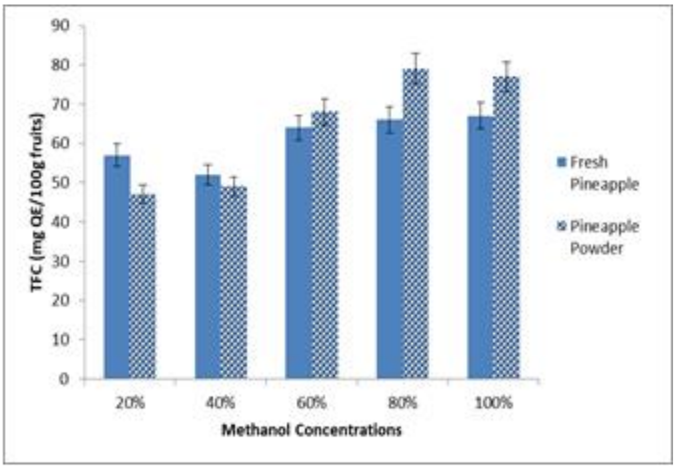

Fig. 2. Total Flavonoid Content (mg QE/100g fruits)

The TFC in fresh pineapple was highest at 100% methanolic extract (67 mg QE/100g fruits) compared to 40% methanolic extract that is 52 mg QE/100g fruit. In the previous study by Silva et al., 2013, the TFC ranged between A. comosus (22.6 ± 0.06 mg QE/100 mL) and the lowest is A. muricata ('guyabano') fruit at 7.06 (± 0.01) mg QE/100 g fruits. However, a previous study by Hashib *et al*., [6], reported that TFC after drying process was ranged between 580.70 (± 20.46) µg of RE100/g sample and this was at its highest level compared to fresh pineapple pulp at 197.10 (± 2.60) µg of RE/100 g sample, dry base and skin is 76.93 (± 11.85) µg RE/100 sample.

The TPC was determined by using the Follin-Ciocalteu's reagent. It has an ability to determine the absorbance of fresh pineapple and pineapple powder in various different concentrations. It also has an ability to reduce by the non-phenolic content in the pineapple fruit such as ascorbic acid, fructose and sucrose. Although this method is acceptable, gallic acid equivalent should be used as a correlation standard. The phenolic content in pineapple fruit was presented in both soluble forms and combined with cell wall complexes.

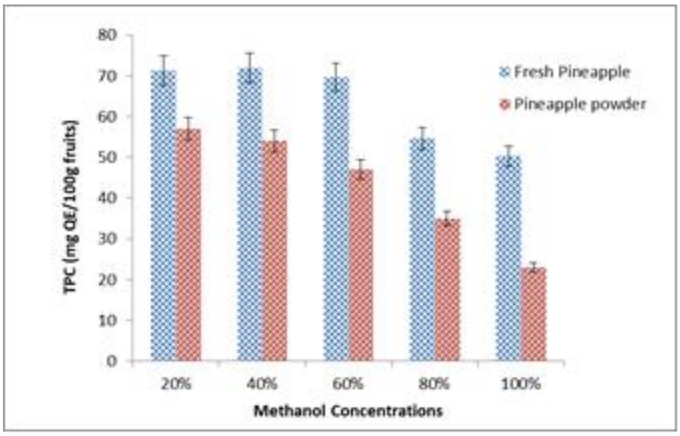

Fig. 3. Total Phenolics Content (TPC) (mg GAE/100g fruits)

Fig. 3 shows the TPC (mg GAE/100 g Fruit) values in this experiment which within range between 23 mg – 71 mg GAE/100g fruit compared with 306.45 ± 14.41 µg GAE/g fruit, which was reported by [5] and 26.20 ± 0.49 mg GAE/100 g fruit that was reported by Hashib *et al*., [6]. These show that the experiments were fairly conducted. Besides, the TPC decreased due to spray drying in pineapple powder. The spray dried pineapple showed the highest TPC, 58 mg GAE/100g fruits compared to with fresh pineapple. In previous studies on comparative analysis of antioxidant analysis of pineapple, the results also reported the range of 53.8 mg GAE/100g fruits for phenolic content in Morris pineapple [8].

DPPH free radical scavenging assay values of fresh pineapple were ranged from 48-65% and pineapple powder between 16-53% in five different methanolic concentrations as Figure 4.

Comparison between fresh and pineapple powder for DPPH assay analysis had shown significant difference between 20% and 100% methanol extracts. The amount of antioxidants in the highest amount of methanol concentrations showed 53%, almost three times multiplier than antioxidants in 20% methanol concentrations. The spray dried pineapple powder showed good retention of antioxidant compared with fresh pineapple extract through DPPH activity. Other studies on fruits like blueberry, acai and peanut had also shown the comparison between processed and soluble extract of fruits characterized by higher concentration effect due to the removal of other insoluble factors [17,12].

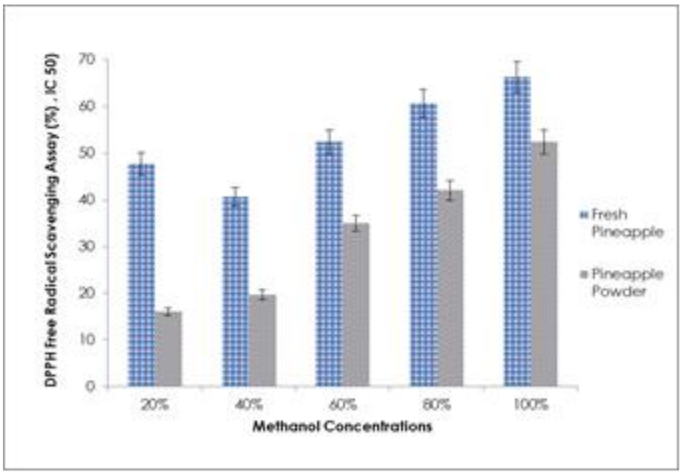

Fig. 4. DPPH Free Radical Scavenging Assay (%)

3. Conclusion

In conclusion, spray drying is potentially processed for a large-scale production of pineapple powders in order to preserve its antioxidant content. The spray dried pineapple powder indicates good antioxidant activity. Furthermore, the numerous applications of pineapple powder are not limited to, commercial products likes jam and jellies but can be used as health supplements too. It is also possible that the resulting powder could be used in a wide range of functional food applications, delivering antimicrobial properties in those foods.

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