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Effect of heat treatment on the physico-chemical properties of Mengkudu *(Morinda citrifolia)* extract

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Abstract: Extract of mengkudu was heated using a bench-top tube heat exchanger at 30, 50, 70 and 90°C for 0, 5, 10 and 15 min residence time. The treated mengkudu extract was then measured for pH, L^*, a^*, b^* color parameters, clarity, viscosity and total polyphenol content. Results show that heating using temperature of 30 to 90°C for 5 to 15 mins significantly ($p<0.05$) increased the pH compared to unheated samples. However, when compared between the different heating temperature and residence time used, no significant differences were observed. Both temperature and residence time did not significantly affect total soluble solids and color parameters of L^*, a^*, b^*. Using a higher heating temperature resulted in a reduction in clarity and viscosity but no significant effect on total polyphenol content. At higher temperature, increasing the residence time resulted in lower clarity. Residence time did not show a significant effect on total polyphenol content and viscosity.

Keywords: *Morinda citrifolia*, temperature, residence time, flow rate, physico-chemical

Introduction

Mengkudu (*Morinda citrifolia*) is a well known herbaceous plant not only in Malaysia, but also in Polynesia, China, India and Australia. Mengkudu fruit has been used traditionally because it is said to have extraordinary medicinal functions. Mengkudu fruits has been used to treat various types of health conditions, including high blood pressure, menstrual pain, arthritis, cancer, dislocation, depression, and heart disease (Solomon, 2000). Within the last few years, interest in the nutritional and pharmaceutical properties of local herbs, plants and plant materials have increased among the Malaysian consumer. Among the types of products marketed widely are products derived from the mengkudu plant. Currently, there are food companies which produce mengkudu products in the form of juice, powder and capsule.

The production of mengkudu products commonly involves heat processing. Heat processing is an important step in production as it has been reported to reduce the unpleasant smell and taste of the mengkudu juice (Degener, 1973). In addition, heat processing can lower the risk of pathogenic and spoilage microorganisms. Heat processing can also inactivate enzymes (Richardson and Finley, 1985). Hence, heat processing can extend the shelf life of products and lower the risk of food poisoning.

However, heat processing can also affect the sensory properties and nutritional value of food products. During the production of fruit extracts, heat processing can affect pH, colour, viscosity, total acidity, turbidity and vitamin C content. In addition, viscosity has been reported to be highly dependent to the change in heating temperature (Lewis, 1996). The intensity of colour also changes with heating temperature in pea puree (Shin and Bhowmik, 1995). Heat processing also contributes to the turbidity in juice (Daridek *et al*., 1990). Heating at high temperature has been reported to cause flavonoid glycosides to change into other forms subsequently reducing antioxidant activity (Wiel *et al*., 2001).

Although there have been several studies carried out regarding mengkudu extracts such as profiling of volatiles (Farine *et al*., 1996) and optimization of deacidification (Nur Hafiza *et al*., 2010), there are still no scientific studies done on the effect of heat processing on mengkudu extract. Hence, the objective of this study is to determine the effect of heating temperature and time on the physicochemical properties of mengkudu extract.

Materials and Methods

Materials

Mengkudu fruits at a maturity index of 4 which was when the fruits were 80% mature with yellowish and pale color were used to prepare extracts. The fruits were obtained from Kemayan, Pahang. The mengkudu fruits which have been plucked were stored at room temperature for three days before being processed into extract.

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Preparation of samples

Mengkudu fruits were washed using tap water. Subsequently, the fruits were mashed and diluted with water at a ratio of 1:1. The mashed fruit blend was centrifuged at 5000 rpm for 20 minutes to separate its seeds and pulp from the extract. The extract was then subjected to heat treatment at different temperature and flow rate using a fabricated tubular heat exchanger. The tubular heat exchanger consisted of stainless steel tubing with an internal diameter of 0.5 cm and 600 cm in length. The tubing was immersed in a hot water bath heated by an electric heating coil. The required temperature of the water bath was controlled using a digital temperature controller. The processing or residence time of the extract was controlled by controlling its flow rate through the stainless steel tubing. Subsequently, the processed extract were analysed by physical and chemical analysis.

pH

The pH of the mengkudu extracts was measured using a pH meter (WTW pH 422; pH 1-14). pH was measured at room temperature where 30 ml of mengkudu extract was poured into a 50 ml beaker and stirred before the reading was taken. Calibration of the pH meter was carried out using pH 4 and 7 buffers.

Total soluble solids

Total soluble solids were measured using a refractometer (Atago 0 – 32%; Japan). Several drops of the mengkudu extracts were dropped on the surface of the mirror of the refractometer. Total soluble solids were stated in units of ^{o}Brix.

Colour

The colour of mengkudu extracts was measured using a chromameter (Model Minolta CR-300; Japan) which has been calibrated using a standard white plate to determine the value of L^*, a^* and b^*. Mengkudu extracts were half-filled in test tubes during measurement.

Clarity

The clarity of mengkudu extracts was measured according to Floribeth *et al*. (1981). 2 ml of mengkudu extract was poured into a cuvet. Absorbance was measured at 580 nm using UV spectrophotometer (Model UNICAM; England). Distilled water was used as blank. Clarity was measured as O.D.

Viscosity

Brookfield rotational viscometer (Brookfield Digital Viscometer, Model DV-11; USA) was used to measure the viscosity of the mengkudu extracts. Samples were measured using spindle number 3 at 50 rpm. Viscosity was measured in cPs.

Total polyphenol content

Total polyphenol content was determined using the Folin-Ciocalteu reagent (Shahidi and Naczk, 1995), which contains sodium phosphomolibdate and sodium tungstat. The epicatechin stock solution was prepared prior to use.

Statistical analysis

The study was conducted according to a factorial design with 2 factors which consisted of heating temperature and residence time. Three replications were used for all parameters measured. Analysis of the data was carried out using a statistical software (SAS ver. 6.12)(SAS 1998). Statistical tests used were ANOVA and Duncan's Multiple Range test. Level of confidence used was 95%.

Results and Discussion

pH and total soluble solids

pH is an important measurement of acidity. pH affects the flavor or taste of a product and the need for processing. Table 1 shows the pH of mengkudu extracts after heat treatment at different temperature and residence time. The results show that application of heating at 5, 10 and 15 mins significantly ($p<0.05$) increased the pH of mengkudu extracts compared to control. Increase in pH when mengkudu extracts was subjected to heat treatment might be due to the loss of organic acids which caused the decrease in acid content, and hence increased the pH value of the extract (Igual *et al*., 2010). However, no significant difference was observed when heating was prolonged from 5 mins up to 15 mins compared to control. Similarly, the results showed that increasing heating temperature of mengkudu extracts up to 90^{o}C for 5 to 15 mins resulted in a significant ($p<0.05$) increase in pH. Total soluble solids in mengkudu extracts remained the same at 4^{o}Brix even after being subjected to the different heat treatments.

Table 1. pH of mengkudu extract subjected to different heating temperature and residence time

Residence time (min)	Temperature (^{o}C)			
	30	50	70	90
0	3.89^{e}	3.89^{e}	3.89^{e}	3.89^{e}
5	3.97^{d}	4.00^{bcd}	4.02^{abc}	4.06^{a}
10	3.97^{d}	4.00^{bcd}	4.04^{ab}	4.06^{a}
15	3.98^{bcd}	4.02^{abcd}	4.06^{a}	4.06^{a}

$^{a-e}$ Means with different letters showed significant difference ($p<0.05$).

Color

The colour of tropical fruits and its products is an important quality attribute. Measurement of colour is usually used as a common index for quality identification (Askar and Treptow, 1993). Results from Table 2 indicates that heating temperature up to 90°C and residence time up to 15 mins did not give any significant effect on the L^* value of the mengkudu extracts. The heating treatments used did not show any obvious effects on the a^* (Table 3) and b^* (Table 4) values of the mengkuru extracts. a^* values did not show any significant effects by both heating temperature and residence time on mengkudu extract except for 70°C/5 min, 90°C/5 min and 90°C/10 min (Table 3). Weemaes *et al*. (1999) reported that decomposition of green pigments occurred in broccoli juice when the heating temperature increased to 70 – 80°C. This might be due to chlorophyll in the broccoli changing to pheophytin during heating at high temperature of 70°C. The difference between the results compared to Weemaes *et al*. (1999) may be due to the different sample used. Similar to a^* values, no obvious effects of the heating treatment on b^* values was observed. No significant effect of heating temperature and residence time on b^* values.

Table 2. Degree of lightness (L^*) of mengkudu extract subjected to different heating temperature and residence time

Residence time (min)	Temperature (°C)			
	30	50	70	90
0	33.24[a]	33.24[a]	33.24[a]	33.24[a]
5	33.47[a]	33.48[a]	33.16[a]	33.68[a]
10	32.92[a]	33.73[a]	32.83[a]	33.65[a]
15	33.17[a]	33.57[a]	33.03[a]	33.47[a]

[a] Means with the same letter did not show significant difference.

Table 3. Degree of redness (a^*) of mengkudu extract subjected to different heating temperature and residence time

Residence time (min)	Temperature (°C)			
	30	50	70	90
0	-0.51[de]	-0.51[de]	-0.51[de]	-0.51[de]
5	-0.51[de]	-0.45[bcd]	-0.36[abc]	-0.31[a]
10	-0.51[de]	-0.48[bcde]	-0.47[bcde]	-0.34[ab]
15	-0.51[de]	-0.50[cde]	-0.44[abcd]	-0.37[abcd]

[a-e] Means with different letters showed significant difference ($p<0.05$).

Table 4. Degree of yellowness (b^*) of mengkudu extract subjected to different heating temperature and residence time

Residence time (min)	Temperature (°C)			
	30	50	70	90
0	2.42[a]	2.42[a]	2.42[a]	2.42[a]
5	2.15[abcd]	2.40[ab]	2.0[d]	2.20[abcd]
10	2.33[abc]	2.32[abc]	2.30[abc]	2.11[bcd]
15	2.35[ab]	2.34[ab]	2.05[cd]	2.24[abcd]

[a-e] Means with different letters showed significant difference ($p<0.05$).

Clarity

Table 5 shows the value of absorbance at 580 nm of the heat treated mengkudu extract. A higher absorbance value indicated a lower level of clarity. Based on Table 5, increasing heating temperature from 30 to 90°C resulted in a reduction of clarity when heated for 5, 10 and 15 mins. Reduction in clarity may be due to the formation of haze due to protein-polyphenol complex formation (Siebert, 2006). When heated at 30, 50, 70 and 90°C, extending the residence time from 0 to 15 mins showed a contrasting effect when heated at temperature lower or higher than 70°C. Increasing the residence time from 0 to 15 mins when heated at 70°C did not result in any significant difference. However, when heated at 30 and 50°C, increasing the residence time significantly ($p<0.05$) increased clarity compared to unheated samples. The increased clarity may be due to coagulation of impurities due to heating. Jayabalani *et al*. (2008) also observed no significant effect of heating at up to 68°C in tea drinks. However, heating at 90°C resulted in an opposite effect where increasing the residence time from 0 to 15 mins produced lower clarity which may be due to haze formation as discussed previously.

Table 5. Clarity, based on absorbance at 580 nm, of mengkudu extract subjected to different heating temperature and residence time

Residence time (min)	Temperature (°C)			
	30	50	70	90
0	0.13[bcd]	0.13[bcd]	0.13[bcd]	0.13[bcd]
5	0.12[de]	0.11[ef]	0.13[bcd]	0.135[abc]
10	0.11[ef]	0.11[ef]	0.13[bcd]	0.14[a]
15	0.11[ef]	0.11[ef]	0.13[bcd]	0.14[a]

[a-e] Means with different letters showed significant difference ($p<0.05$).

Viscosity

Based on Table 6, it can be observed that there were significant differences ($p<0.05$) in the viscosity of the mengkudu extracts when heated at different temperature and residence time compared to control (without heating), except for 30°C/5 mins and 30°C/10 mins. When temperature was increased from 30 to 90°C, viscosity of the mengkudu extract decreased for each residence time of 5 to 15 mins. According to Lewis (1996), viscosity is very much affected by heat. When heating temperature increases, viscosity of liquid decreases. When the residence time was increased from 5 to 15 mins, no significant effect on viscosity was observed after heating at 50, 70 and 90°C. Hernandez *et al*. (1995) reported that apparent viscosity of orange juice decreased with increasing heating temperature within a Brix range of 44.9 to 66.5°. Giner *et al*. (1996) also observed a reduction in viscosity with increasing heating temperature for cherry juice within a Brix range of 22.0 to 74.0°.

Table 6. Viscosity (cPs) of mengkudu extract subjected to different heating temperature and residence time

Residence time (min)	Temperature (°C)			
	30	50	70	90
0	15.67[a]	15.67[a]	15.67[a]	15.67[a]
5	15.00[a]	13.67[bc]	12.00[d]	10.00[c]
10	15.00[a]	13.33[bc]	12.00[d]	10.00[c]
15	14.00[b]	13.00[c]	11.67[d]	10.00[c]

[a-c] Means with different letters showed significant difference ($p<0.05$).

Total polyphenol content (TPC)

Based on the results as shown in Table 7, it can be observed that increase in residence time from 0 to 15 mins did not result in any significant differences for total polyphenol content when heated at 30 and 50°C. However, when heated at higher temperature of 70 and 90°C, increasing the residence time from 5 to 15 mins showed significantly ($p<0.05$) lower TPC compared to control (0 min). Heating at high temperature can cause the flavonoid glycoside to change into other forms subsequently reducing antioxidant activity (Wiel *et al*., 2001). However, for each residence time, no significant differences were observed between the different heating temperature. Thus, from the results, the period of exposure to heat apparently is more important than the heating temperature.

A few studies has also reported on the reduction of total polyphenols during heat treatment. Crozier *et al*. (1997) reported that quercetin concentration in tomatoes and onions were reduced by 82% and 75%, respectively, after boiling for 15 min. However, the reduction may be due to the leaching of polyphenol compounds into the boiling water (Gil *et al*., 1999). As the mengkudu extract in this study was heated using a tubular heat exchanger, leaching of polyphenols was not possible. The reduction of the total polyphenol observed was thus, probably due to the heating treatment as reported by Faller and Fialho (2009). However, further studies are needed to determine the actual polyphenol compounds affected by the heating.

Table 7. Total polyphenol content of mengkudu extract subjected to different heating temperature and residence time

Residence time (min)	Temperature (°C)			
	30	50	70	90
0	42.26[a]	42.26[a]	42.26[a]	42.26[a]
5	37.29[ab]	36.23[ab]	33.06[ab]	30.01[b]
10	34.47[ab]	35.17[ab]	29.27[b]	28.37[b]
15	33.19[ab]	32.35[ab]	28.49[b]	27.00[b]

[a,b] Means with different letters showed significant difference ($p<0.05$).

Conclusion

Heating using temperature of 30 to 90°C for 5 to 15 mins significantly ($p<0.05$) increased the pH compared to unheated samples. However, when compared between the different heating temperature and residence time used, no significant differences were observed. Both temperature and residence time did not significantly affect total soluble solids and color parameters of L^*, a^*, b^*. Using a higher heating temperature resulted in a reduction in clarity and viscosity but no significant effect on total polyphenol content. At higher temperature, increasing the residence time resulted in lower clarity. Residence time did not show a significant effect on total polyphenol content and viscosity.

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