EFFECTS OF DIFFERENT COOKING METHODS AND TIME INTERVALS ON PHYSICO-CHEMICAL AND ANTIOXIDANT PROPERTIES OF GOLDEN PUMPKIN (*Cucurbita maxima*)

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Accepted 4 February 2019, Published online 20 March 2019

ABSTRACT

Effect of different cooking methods with different cooking time on the chemical, physical and antioxidant composition of golden pumpkin (*Cucurbitae maxima*) was evaluated. Pumpkin flesh has been treated with different cooking methods, which were boiling, steaming and stir-frying at different cooking time (5 and 10 minutes). Chemical analysis showed there was no significant difference (p>0.05) detected for all treatments except for the fat content in stir-fried sample. Colour analysis, which was measured using tristimulus colorimeter showed decreasing of lightness (L*) in all treated samples compared to fresh sample. Total phenolic content was determined using Folin-Ciocalteu method and the antioxidant activity was based on the evaluation of free-radical scavenging activity (DPPH assay). Total carotenoids were measured using spectrophotomer by comparing with β -carotene as the standard. Among treated samples, total phenolic content was found to affect the antioxidant activity in all treated samples, as the percentage of DPPH inhibition was slightly lower than fresh sample. Surprisingly, β -carotene was found to be slightly higher in steamed sample compared to other cooking methods and fresh sample. This study showed that different cooking methods gave some affect to the available antioxidant properties in golden pumpkin.

Key words: Pumpkin, cooking method, cooking time, physico-chemical properties, antioxidant, β -carotene

INTRODUCTION

Pumpkin (*Cucurbita maxima*) is defined as a fruit and botanically belongs to the family of Cucurbitaceae (Paris & Brown, 2005). Pumpkin has yellow to orange flesh, naturally contributed by carotenoid compound. It has been reported to be abundant in nutrients especially fiber and carotenoids (Azizah *et al.*, 2009) especially lutein and zeaxanthin (Loy, 2011). As reported by Markovic *et al.* (2002), there are three species of pumpkin; *C. pepo, C. maxima* and *C. moschata* while *C. maxima* was reported with high content of total carotenoid (2120.1g⁻¹)(Carvalho *et al.*, 2014).

Due to its sweet taste, pumpkin has been widely used in Malaysian cuisine. Cooking processess would bring several numbers of changes in physical characteristics and the chemical composition of the vegetable or fruit. Both antioxidant level and activities were normally lower in cooked vegetable compared to fresh (Podsedek, 2007). Most of antioxidants was reported to be reduced with higher temperature (Murador *et al.*, 2014). The degradation of bioactive compounds is due to the absorption of water during cooking process resulting in the dilution of the active compound.

However, both positive and negative effect will occur depending on the differences processing conditions, morphological and the nutritional characteristic of the vegetable species (Nicoli et al., 1999). Interestingly, total carotenoid was reported to be increased in cooked pumpkin (C. moschata) compared to fresh sample (Carvalho et al., 2014). Different temperature and cooking time gave different effects on variety of bioactive compounds (Azizah et al., 2009), the best cooking method to preserve the functional properties of active compound is highly required. Panglossi (2006) reported vitamin C is partially destroyed by cooking but other antioxidant activity increased, which possibly were due to freeing the antioxidant phytochemicals. Turkmen et al. (2005) studied the

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effect of cooking methods on tomato stated that boiling produced small effect on the lycopene, total phenolic, ascorbic acid, and antioxidant activity but frying significantly reduced all of it. Therefore, the aim of this study is to determine the stability of nutrient compounds in pumpkin (*C. maxima*) against different cooking methods and time interval, which may give different results compared to other plants. The output of this study may help in guiding the consumer to obtain sufficient daily intake of nutrients from the selected sample.

MATERIALS AND METHODS

Samples preparation

Pumpkins were collected at similar ripening stage (stage 5) were peeled and the flesh was cut into small pieces (2cm x 2cm). Samples were divided into three different cooking methods with 3 replications; boiling, steaming and stir-frying. In boiling method, 10g of homogenous pieces of pumpkins were immersed in boiling water (100°C) for 5 and 10 minutes, respectively. Steaming was conducted by placing 10g of samples above boiling water (100°C) for 5 and 10 minutes, respectively, in a steamer with lid. For stir-frying method, 2g of cooking oil was pre-heated to 170°C in a pan and 10g samples were cooked for 5 and 10 minutes, respectively. All samples then were drained and cooled at room temperature prior to further analysis.

Reagent and Chemical

Chemicals that were used to determine the antioxidant properties in the pumpkin were methanol, hexane, Folin-Ciocalteu, sodium carbonate, gallic acid, diphenyl-1-picrylhydrazyl (DPPH) and β -carotene standard (Sigma Aldrich). The chemicals and reagents that were used in the proximate analysis were hydrochloric acid (HCL), sodium hydroxide (NaOH), acetone, and petroleum ether (R&M Chemicals).

Color analysis

Color of samples was determined by using Conica Minolta chromameter with values of L*, a*, and b*. L* value indicative for brightness, a* value for redness and b* value for yellowness (Caner & Cansiz, 2007).

Chemical analysis

Chemical analysis consisting of moisture content, ash content, crude fiber and crude fat of all samples was done based on the AOAC (2010) method.

Antioxidant analysis

Extraction

A total of 100g of sample was homogenized in 300 mL methanol with three replications. Mixture was left for 2 hours at room temperature. Extract was filtered using muslin cloth and then filter paper (Whattman 1). Extract was evaporated using rotary evaporator at 55°C and kept at 4°C until analysis.

Total phenolic compound analysis

The total phenolic content of the extract was determined by Folin-Ciocalteu method (Choo *et al.*, 2014). 1 mg of crude extract was mixed with 1 mL methanol (75%), 4.5 mL distilled water and 5 mL of Folin–Ciocalteu reagent for 5 min. Then 2 mL of sodium carbonate in distilled water (20%:w/v) was added to the mixture. The mixture was allowed to stand for 90 min in the dark, and absorbance was measured at 750 nm. The total phenolic content was calculated from the calibration curve, and the results were expressed as mg of gallic acid equivalent per g dry weight.

Determination of β -carotene

β-carotene determination was done according to the method by Yuan *et al.* (2008). 1 mL of sample was extracted with 2 mL of ethanol and 3 mL of n-hexane. N-hexane phase was removed after shaking. The extraction was repeated twice and the removed n-hexane phases were pooled. The absorbance of the extract was then measured at 450 nm with UV-vis spectrophotometer (Shimadzu UV-1700 PharmaSpec). The concentration of β-carotene was obtained by referring to a standard curve of β-carotene (10, 20, 30, 40 and 50 µg/mL).

Diphenol-picryllhydroxide (DPPH)

The antioxidant activity of the extract was determined by the 1,1-diphenyl-2-picryl-hydrazyl (DPPH) assay (Shekhar & Anju, 2014). Briefly, 1 mg sample was mixed with 2 mL of 0.1 mM DPPH solution. Mixture was shaken and incubated for 30 min in a dark room. The absorbance of the mixture was then measured at 517 nm. The ability of the sample to scavenge DPPH radical was determined using the formula:

$$\% DPPH = \frac{Abs_{control} - Abs_{sample}}{Abs_{control}} \times 100$$

Statistical Analysis

All the data were analysed using Minitab software. Results were presented in mean \pm standard deviation. Difference between variable was tested for

	Treatment						
Analysis	Boiling		Steaming		Stir frying		Fresh
	5 min	10 min	5 min	10 min	5 min	10 min	
Moisture (%)	91.33±0.06ª	83.10±0.46 ^ª	90.83±0.37ª	82.11±6.31ª	88.51±0.08ª	76.04±1.10 ^ª	90.94±0.47ª
Ash (%)	0.72 ± 0.04^{a}	1.10±0.10 ^a	0.83±0.12 ^ª	0.88 ± 0.06^{a}	1.03±0.05 ^ª	1.13±0.54 ^ª	0.95 ± 0.03^{a}
Fibre (%)	1.13±0.15 ^ª	0.83 ± 0.07^{a}	0.94 ± 0.14^{a}	1.11±0.02 ^a	1.59 ± 0.05^{a}	1.84±0.03 ^a	0.84 ± 0.04^{a}
Fat (%)	0.47±0.25 ^{bc}	0.54±0.18 ^{bc}	0.34±0.13 ^{bc}	0.69±0.70 ^{bc}	1.11±0.07 ^b	3.37±0.67 ^a	0.15±0.06 ^c

Table 1. Effect of different cooking methods and time on chemical composition of golden pumpkin (Cucurbita maxima)

Data are expressed as means ± standard deviations.

a-c Means in the same row with different lowercase letters are significantly different (p<0.05).

 Table 2. Effect of different cooking methods and time on the colour of golden pumpkin (*Cucurbita maxima*)

Treatment	Colour					
Treatment	L*	a*	b*			
Boiling 5 min	49.20±0.26 ^b	11.62±3.76ª	41.66±0.96ª			
Boiling 10 min	47.46±0.31 ^b	9.56 ± 0.53^{a}	38.12±0.52 ^ª			
Steaming 5 min	48.30±0.14 ^b	7.66±0.51 ^a	38.79±1.64 ^ª			
Steaming 10 min	47.64±0.97 ^b	9.50 ± 0.63^{a}	40.37±0.27 ^a			
Stir frying 5 min	50.44±0.39 ^b	14.83±0.26 ^a	42.71±1.52 ^a			
Stir frying 10 min	46.70±1.48 ^b	12.09±0.38 ^a	38.27±2.57 ^a			
Fresh	53.38±1.81ª	15.93±2.19ª	37.30±4.83ª			

Data are expressed as means ± standard deviations.

a-b Means in the same column with different lowercase letters are significantly different (p<0.05).

significance using One-way ANOVA and multiple comparison. Significance difference was defined at p < 0.05.

RESULTS AND DISCUSSION

Effect of different cooking methods on chemical composition of pumpkin

Table 1 shows the chemical composition of moisture, ash, fiber and fat in fresh and cooked pumpkin with different methods and time. It was shown that there was no significant difference (p>0.05) in the moisture, ash and fiber content of all pumpkin before and after cooking. Different cooking methods either with oil or water at different time did not affect the chemical composition except fat content (p<0.05).

Statistical analysis shows that there were significant differences in crude fat analysis and there was an interaction between sample and the cooking method (Table 1). The highest percentage of fat content was in stir-fried pumpkin at 10 minutes of $3.37\pm0.66\%$ followed with stir fried at 5 minutes of $1.10\pm0.07\%$. The lowest fat content in cooked pumpkin was in the boiled and steamed pumpkin, which showed similar content with fresh

sample. The higher fat content is possibly due to the cooking oil used in the method, which increased with increasing time (Ghidurus *et al.*, 2010). The absorption of fat from oil by the sample during cooking contributed to the higher fat in the stir-fried sample. This result is similar to the finding by Gokoglu *et al.*, 2004.

Effect of different cooking methods on colour of pumpkin

The flesh colors of pumpkins and squash generally include a wide range of whites, yellows, and oranges. This color is based on the particular carotenoid types and concentrations that are influenced by genetic and environmental factors (Itle & Kabelka, 2009). Table 2 shows the color value of L* (brightness), a* (redness) and b* (yellowness) of the cooked and fresh pumpkin. There was no difference of a* and b* value for all samples. However, treated samples showed darker (L* value) surface colour than the fresh sample (p<0.05). The darker colour is probably resulting by the browning occurrence due to non-enzymatic reaction of sugar in pumpkin with heat (Adams & Brown, 2007). Moreover, the high sugar in the ripe pumpkin in this study enhances the browning reaction. Besides, the discolouration of yellow

colour in pumpkin also may be resulting by the oxidation and isomerization of β -carotene (Gliemmo *et al.*, 2009) with lipoxygenase. Another study also showed the high degree of browning in yellow vegetable is a result of reaction between dehydro-ascorbic and amino acids in the sample (Adams & Brown, 2007).

Effect of different cooking methods on total phenolic content in pumpkin

The total phenolic content of golden pumpkin is shown in Figure 1. It shows that thermal and time treatment affected the total phenolic compound of the pumpkin. Longer cooking time increased the loss of total phenolic compound (p<0.05). Steamed pumpkin (5 minutes) has the highest total phenolic compound (gallic acid equivalents, g/mg) with mean value 159.46±33.07 g/mg. The lowest phenolic content was in stir-fried pumpkin. The lower total phenolics in stir-fried pumpkin could be due to the breakdown of phenolics or losses (leached out) during cooking as most of the bioactive compounds are relatively unstable when heated and get easily solubilized (Zhang & Hamauzu, 2004).

Effect of different cooking methods on antioxidant activity of pumpkin

Figure 2 shows significant interactions between the cooking methods. It shows that the value of the percentage of DPPH inhibition of all treatment samples was decreased (p<0.05) compared to the fresh pumpkin and synthetic antioxidants (BHT and tocopherol). Stir-fried pumpkin showed the lowest percentage value, with $67.09\pm0.04\%$ of DPPH inhibition. Among all of the cooking methods, steamed pumpkin showed better results. The uniformity of heat circulation of steaming method to the sample may be the reason of this phenomenon. Hence, food cooks faster and fewer nutrients including antioxidants are lost. Heat treatments affect the antioxidant activity of vegetables and in many cases lower antioxidant capacity has been detected in processed samples versus raw vegetables (Chipurura et al., 2010). Cooking factors, including method, temperature and cooking time, strongly affect the antioxidant activity of food (Hwang et al., 2012). Many reports indicated that the thermal treatment affects the antioxidant activity of various food samples (Juaniz et al., 2016; Şengül et al., 2014). Antioxidant capacities were also reported to decrease after thermal treatment of carrots, onions, white cabbage, and other vegetables (Ismail et al., 2004).

Effect of different cooking methods on β carotene content in pumpkin

Result showed the highest β -carotene content in steamed pumpkin (p<0.05) in both 5 and 10 minutes (Figure 3). Surprisingly, the result was higher (p<0.05) compared to the fresh pumpkin. Thermal treatment was found to enhance the availability of the carotenoid (D'Evoli *et al.*, 2013; Hwang *et al.*, 2012). Higher results of lycopene and β -carotene have been reported in cooked carrots and spinach (Rock *et al.*, 1998) compared to the fresh samples. Cooking practices break down the food matrices and loosen the carotene-binding fibers. This may lead to nutrient loss but also may facilitate bioavailability and increase the carotene content (Fernández-García *et al.*, 2012). This study also

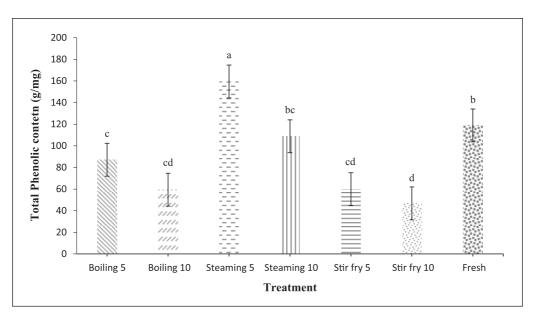


Fig. 1. Effect of different cooking methods and time on total phenolic content (g/mg) of golden pumpkin (*Cucurbita maxima*). ^{a-d}Means with different lowercase letters are significantly different (p<0.05).

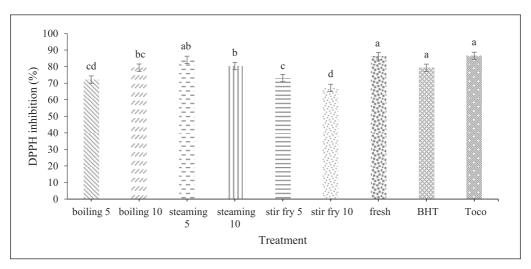


Fig. 2. Effect of different cooking methods and time on antioxidant activity (DPPH %) of golden pumpkin (*Cucurbita maxima*). ^{a-c}Means with different lowercase letters are significantly different (p<0.05).

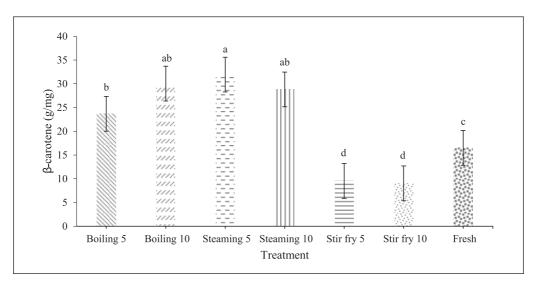


Fig. 3. Effect of different cooking methods and time on \hat{a} -carotene content (g/mg) of golden pumpkin (*Cucurbita maxima*).^{a-d}Means with different lowercase letters are significantly different (p<0.05).

showed that dry cooking method, stir-fring affects the β -carotene, as the value is lower than the fresh one. A study by Azizah *et al.* (2009), reports the reducing value of β -carotene in pumpkin is probably due to the fact that carotenoids are fatsoluble compounds and solubilized readily in oil during stir-frying, and thus resulted in its decrease. In addition, pumpkin was exposed to a much lower temperature during boiling and steaming (100°C) compared to that of stir-frying (180°C).

CONCLUSION

In conclusion, heat treatment with different cooking time influenced the physical and chemical composition of the golden pumpkin. Among all the cooking methods, steaming has the ability to retain nutrients better than other methods as it showed the highest antioxidant properties. In addition, the shorter time of cooking may also be beneficial to retain the maximum nutrients in pumpkin.

ACKNOWLEDGEMENT

The authors would like to thank Universiti Malaysia Terengganu for providing the funds and support towards this research.

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