# Effect of Roasting and Microwave Heating on the Yield, Quality, Total Phenolics and Antioxidant Capacity of Oil From Red Pumpkin Seed (*Cucurbita pepo* L.) by Mechanical Pressing

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#### Abstract

Pumpkin seeds (*Cucurbita pepo* L.) contain oil, which is considered as a rich source of bioactive compounds beneficial to human health. This work was aimed at evaluation of the effect thermal pre-treatments including microwave heating and roasting on the oil yield and quality properties. The oil was obtained by two mechanical pressing methods. The dried pumpkin seeds were treated under microwave for several minutes or roasted in a coffee roaster for 30 - 60 minutes, at temperature of 90 - 130°C and subjected to hot expelling or cold pressing to get an oil product. The optimal condition for pumpkin seed oil extracted by hot and cold pressing were microwave pre-treatment time in 2 minutes and roasting time in 45 min, respectively. The best oil product by using hot press extraction got a highest oil yield (25.55%) and a good quality with low free-fatty acid (3.17% as oleic acid), peroxide value (9.5 meq  $O^2/kg$ ), high antioxidant capacity (66.89%), total phenolic content (51.4 mg GAE/100g) and total carotenoid content (3361.37 mg  $\beta$ -carotene/kg). Similarly, the best oil product from cold pressing got a quantity at 39.94% with low free-fatty acid value (3.53% as oleic acid), low peroxide value (4.17 meq  $O^2/kg$ ), mild antioxidant capacity (42.15%), total carotenoid content (1671.6 mg $\beta$ -carotene/kg) and high total phenolic content (73.93 mg GAE/100g). The predominant fatty acids found in pumpkin seed oil were oleic, linoleic and palmitic acids.

Keywords: Antioxidant Capacity; Mechanical Pressing; Microwave; Pumpkin Seed Oil; Roasting; Total Phenolic Content

#### Introduction

Pumpkin (*Cucurbita* genus) is a plant belonging to Cucurbitaceae family, consisting of principal species such as *C. maxima, C. pepo, C. moschata, C. ficifolia* and *C. turbaniformis*. Pumpkin fruits are consumed as a food and pumpkin seeds are considered as a rich source of bioactive compounds. The pumpkin seeds are widely utilized as food and folk medicines or for oil production. Pumpkin seed oil is a dichromatic viscous oil, reportedly having strong antioxidant activity, used as a supplement in capsule or liquid form to promote human health [1]. A variety of bioactive components in pumpkin seeds oil has been identified including unsaturated and polyunsaturated fatty acids, sterols, tocopherols and tocotrienols and squalene. The pumpkin seed oil has been reported to have effect in treatment for benign prostatic hyperplasia, which inhibits the growth and size reduction of the prostate [2].

Plant seed oils are obtained by solvent extraction (hexane, methanol, petroleum ether, etc.). This method is inherent with some disadvantages including plant security problems, air pollution with volatile organic compounds, high operation costs and poor quality of products. The environmental regulations, safety and public health concerns made the food industry to find alternatives to organic solvents. Supercritical CO2 extraction (SFE-CO2) is one of the choices [3], but the system is complicated and expensive [4]. Enzyme-assisted

aqueous extraction of oil, considered as environment-friendly and safe, is gaining an attention as an alternative recently. It's limits lie on long processing time, high cost of enzymes and lower oil yield as compared with hexane extraction [5].

The conventional method for oil production is mechanical pressing, of which both cold and hot press process are applied. The screw expelling is a typical hot press process. This method is quite simple, environment-friendly however, it requires much energy and results in low yield and high temperature of oil due to heat of friction [6]. The cold press method is in priority because of little heat generation and process temperature can be kept below 50°C. Even though cold pressing method provides lower oil extraction yield as compared to solvent extraction process [5], but low process temperature allows to preserve bioactive compounds naturally present in oil [7].

Pre-treatments of materials before pressing may help to increase oil extraction yield in mechanical pressing [8]. Traditionally, pumpkin seeds are roasted on a pan. Roasting has a range of effects such as breaking cell membranes to increase oil yield, changing in chemical compositions as well as some bioactive compounds, leading to oxidation stability and development of oil flavor [7]. Pre-treatment may include microwave radiation, which is known for energy and time saving, even distribution of energy throughout the volume of the oil-bearing materials. Microwave can devastate the oil cells' structure in plant tissues. As a result, the cell membranes of oilseeds are ruptured creating a lot of permanent pores, which help the oil moving through the permeable cell walls, so a higher extraction oil yield can be gathered. The effect of microwave treatment could also lead to the change in chemical compositions, foster the oil stability against oxidation by microbial destruction or enzyme inactivation [9].

The overall goal of this study is to investigate the effect of thermal pre-treatments including microwave radiation and roasting on oil yield and quality and compare two mechanical methods including cold pressing and hot expelling. Hence, the effective oil extraction process parameters could be established.

#### Materials and Methods Materials

Fresh pumpkin seeds (*C. pepo*) obtained from Okfood Company (Daklak province, Vietnam) were washed, manually cleaned from broken and mold-infected parts and dried under the sun. The seeds were kept in zipper plastic bags and stored in cold, dark, dry environment.

All solvents or chemicals of analytical grade used for the experiment were supplied from local agents of Merck, Sigma or Chinese distributors.

#### Mechanical extraction of oil from pumpkin seeds

Two mechanical extraction techniques were applied for obtaining oil from pumpkin seeds. The first one was mechanical expelling by a screw expeller, which was considered as a 'hot press' method. The second one was by mechanical pressing in a custom-made device under pressure, which was considered as a 'cold press' method.

For the hot press method, a laboratory screw expeller was used (Subor SUB Z608, China), of which the size of the screw was 14.5 cm length and 2.3 cm in diameter, which was operated at a speed of 40 - 60 rpm. In this method, the whole seeds without de-hulling were introduced directly to the expeller without grinding, after being treated under microwave or roasted in a rotating coffee roaster.

For the cold press method, the dried pumpkin seeds were brought to microwave radiation or roasting first, then de-hulled before being milled in a coffee grinder (Akira 5 M520A, Taiwan) and sieved through a sieve of No. 10 mesh ( $\phi$ 2 mm). A custom-made laboratory hydraulic press for cold pressing pumpkin seed was operated at 20.0 MPa in an appropriate duration to obtain the crude oil. The proce-

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dure for cold pressing was introduced elsewhere [10]. For comparison of the yield, the total oil was obtained by Soxhlet extraction using hexane as a solvent for 6h.

Each treatment was done on a triplicate. The crude oil obtained by two methods of pressing were analyzed the quality by determination of physiochemical properties and oil yield.

#### Microwave treatment of the seeds

LG microwave set was applied for pumpkin seed treatment (LG MH 6342D, Korea). The power was kept at 800 W in different time durations (1, 2 and 3 minutes). The sample was then cooled down to ambient temperature before pressing.

#### **Roasting of the seeds**

Roasting seeds was conducted by using a coffee roaster (CBR-101, Korea) instead of pan-roasting where a unique off-axis rotation with a hot air flow ensures quick and even heat transfer. Appropriate conditions were tested as in a modified method from Nederal., *et al* [7]. Specifically, three different roasting durations were tested at a fixed temperature of 110°C (30, 45 and 60 minutes). On the other hand, roasting temperature was tested in three different levels (90, 110 and 130°C) during a fixed duration of 60 minutes.

#### Physiochemical analysis

#### Oil yield

The oil yield (%) was defined as a ratio between the weight of oil obtained and the weight of sample as below:

% Oil yield = 
$$\frac{\text{weight of oil extracted } (g)}{\text{weight of sample } (g)} \times 100\%$$

#### Free fatty acid values

The free fatty acid (FFA) values of the pumpkin seed oil samples were determined by titration method (AOCS Ca5a-40) and expressed as % of oleic acid as follows:

$$Free fatty acid = \frac{Volume of titration (ml)x Normality of KOH x 28.2}{weight of sample (g)}$$

#### The peroxide values

Peroxide values (PVs) were determined by a standard method (AOCS Cd8-53), expressed as mili-equivalent of active oxygen per kilogram of oil (meq  $O_{3}$ /kg), calculated by:

$$Peroxide Value \left(\frac{mEq}{kg}\right) = \frac{V_{Na_2S_2O_3}used \ at \ endpoint \ \times \ Normality \ of \ Na_2S_2O_3 \times 1000}{weight \ of \ oil \ (g)}$$

#### Antioxidant capacity

Antioxidant capacity of the pumpkin seed oil was evaluated as antiradical activity by using the free radical 2,2-diphenyl-1-picrylhydrazyl (DPPH) [11]. The activity of the extract in scavenging DPPH% was calculated as follows:

#### **Total phenolic content**

The total phenolic content (TPC) of oil was determined by using the Folin-Ciocalteau's reagent method [12]. The samples were analyzed at 765 nm wavelength by using spectrophotometer, using Gallic acid as a standard.

#### **Total carotenoid content**

The total carotenoid content (TCC) in pumpkin seed oil was determined by using a calibration curve of  $\beta$ -carotene, which concentration in range of 0 - 50 µg/mL in n-hexane. All samples were measured spectrophotometrically at 440 nm wavelength [2].

#### Fatty acid profile

GC-MS is a method used to determine fatty acid profile of pumpkin seed oil. In which, fatty acids are transformed into fatty acid methyl esters (FAMEs) and injected into a Gas chromatography [13]. Specifically, the esterification of fatty acids to fatty acid methyl esters (FAMEs) is performed using an alkylation derivatization reagent. The instrument GC-MS (SCION SQ 456-GC) was used with a Column Rxi-5ms RESTEK (30 m x 0.25 mm (i.d.), 0.25  $\mu$ m df). The column oven temperature was programmed with an initial temperature of 50°C held for 1 min, increased to 150°C in 2.5 minutes, next heated at 10°C/min to 220°C, and at 3°C/min to 230°C, at 25°C/min to 280°C and finally held isothermally for 5 minutes. The carrier gas was Helium, applied at constant flow rate at 1 mL/min. The Injector temperature was kept at 250°C and the split ratio was 30.

#### Statistical analysis

All analyses were done in triplicates. Data are reported as means ± standard deviation obtained and processed with ANOVA at a significance level of p < 0.05 by using SPSS software.

### **Results and Discussions**

#### The oil yields

In the hot expelling, microwave heating in 2 minutes resulted in higher yield than for control sample without treatment. The microwave pre-treatment can devastate the oil cell's structure in plant tissues, especially for oilseeds and promote the oil yield [9]. However, the data showed that shorter time (1 minute) or longer time (3 minutes) resulted in no effect (Table 1). The longer microwave radiation may result in creation of excess of emulsion in a material matrix, preventing oil escape, thus the oil yield decreased.

Treatments		Oil yield (%)		Free fatty acids (% oleic)		Peroxide value (meq 0 <sub>2</sub> /kg)	
		Hot pressing	Cold pressing	Hot pressing	Cold pressing	Hot pressing	Cold pressing
Control	0	$23.50 \pm 0.74^{a}$	$37.52 \pm 2.08^{a}$	$3.53 \pm 0.29^{a}$	$4.36 \pm 0.39^{a}$	$11.17 \pm 1.26^{b}$	$3.17 \pm 0.29^{a}$
Microwave	1 min	$24.76 \pm 0.61^{ab}$	$36.82 \pm 1.43^{a}$	$3.39 \pm 0.38^{a}$	$4.08 \pm 0.09^{a}$	$14.33 \pm 1.15^{bc}$	$5.00 \pm 0.50^{\circ}$
pre-treat-	2 min	$25.55 \pm 0.40^{\rm b}$	$36.89 \pm 0.64^{a}$	$3.17 \pm 0.38^{a}$	$4.89 \pm 0.1^{\mathrm{b}}$	$9.5 \pm 0.87^{a}$	$4.50 \pm 0.50^{\rm bc}$
ment	3 min	$24.88 \pm 0.79^{ab}$	$37.58 \pm 0.98^{a}$	$3.53 \pm 0.19^{a}$	$4.59 \pm 0.23^{ab}$	15.33 ± 1.53°	$3.67 \pm 0.29^{ab}$
Roasting pre- treatment at 110°C	30 min	$25.39 \pm 0.54^{\text{b}}$	$38.38 \pm 0.48^{a}$	$3.79 \pm 0.22^{a}$	$5.04 \pm 0.27^{a}$	$7.33 \pm 0.58^{a}$	$4.50 \pm 0.50^{\rm b}$
	45 min	$24.43 \pm 0.65^{ab}$	$39.74 \pm 0.50^{a}$	$3.55 \pm 0.22^{a}$	$3.53 \pm 0.35^{\text{b}}$	$8.83 \pm 0.76^{ab}$	$4.17 \pm 0.29^{b}$
	60 min	$23.19 \pm 0.98^{a}$	$39.32 \pm 0.52^{a}$	$3.26 \pm 0.38^{a}$	$4.59 \pm 0.10^{a}$	$10.67 \pm 1.15^{b}$	$3.83 \pm 0.29^{ab}$
Roasting pre- treatment in 60 minutes	90°C	$25.34 \pm 0.37^{\text{b}}$	$39.51 \pm 0.61^{a}$	$3.32 \pm 0.39^{a}$	$3.16 \pm 0.34^{b}$	$10.83 \pm 0.76^{\rm b}$	$4.50 \pm 0.50^{\rm bc}$
	110°C	$24.20 \pm 0.66^{ab}$	$39.24 \pm 0.79^{a}$	$3.62 \pm 0.20^{a}$	$3.7 \pm 0.42^{ab}$	$11.17 \pm 1.26^{b}$	$4.17 \pm 0.29^{ab}$
	130°C	$23.91 \pm 0.78^{ab}$	$39.37 \pm 0.24^{a}$	$3.75 \pm 0.18^{a}$	$4.08 \pm 0.46^{ab}$	$16.33 \pm 0.76^{\circ}$	$6.00 \pm 0.50^{\circ}$

Table 1: Effect of heat pre-treatments on oil yield, FFA, PV.

\*Values (means  $\pm$  SD) with different index letters are statistically significantly different (p < 0.05) followed by column.

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Similarly, for roasting at 110°C, the roasting duration of 30 minutes was considered as appropriate, resulting in higher oil yield than for the control sample in hot pressing. However, if roasting duration kept for longer (45, 60 minutes), the oil yield was not significantly higher than for the control sample. The longer roasting time may result in creation of excess of emulsion in a material matrix, preventing oil escape. As for the effect of roasting temperature, higher yield was achieved at 90°C pre-treatment in 60 minutes (Table 1). A higher temperature of roasting in the selected duration may result in decline in the yield. This was simply because the higher roasting temperature may create more emulsion so prevent the oil from releasing. Combining the above observations, temperature and roasting duration should be considered in parallel to decide for optimal roasting conditions.

In the cold pressing method, the yield of oil was achieved at 37.5 - 39.7% of the raw material. For comparison in this study, Soxhlet extraction was conducted and the total lipid content collected in pumpkin seeds was about 43.36% (w/w). Thus, the amount of oil (39.74%) that obtained from cold pressing combine with thermal pre-treatment showed that the method could be an effective way to obtain most the whole oil content from pumpkin seeds. This result was consistent with other authors by applied Soxhlet extraction, of which the total lipid content in *C. maxima* seeds was 43.69% [14] or 44.61% [15].

As shown in table 1, the values for oil yield by hot pressing was lower than that by cold pressing, and approximately by a half. It is worthy to note the fact that the hot expelling was done with whole seeds without shelling, whole cold pressing was done with de-hulled seeds.

#### Free fatty acid values

The FFA values of oil were in a range from 3.2-3.8% (Table 1). The effect of three pre-treatment types on free fatty acid in oil by hot expelling were not significant. The vales are quite closed to that reported by from Al-Khalifa [16] for the FFA content in roasted *C. pepo* seed oil at 3.3% as oleic acid. In a report from Jiao., *et al.* [15], the acid value of pumpkin seed oil was 6.97 mg KOH/g oil, equivalent to a value of 3.49% as oleic acid. In general, the trend shows that the FFA values in oil obtained from whole seeds were not sensitive to heat treatments, either microwave heating or conventional roasting in the conditions of this study.

As for cold pressing, the effect of these pre-treatments on FFA was significant. The FFA content in oil may increase by microwave heating durations (Table 1). However, the trend was opposite in oil from seeds treated by conventional roasting at 110°C and 45 minutes or 90°C and 60 minutes: the FFA values decreased. It suggested that 110°C/45 min could be a suitable roasting condition for good oil quality.

The FFA values of the seed oil samples by the two extraction methods in this study was within Codex Alimentarius limitation for crude oils such as virgin palm oil (10.0 mg KOH/g) or coconut oil (4.0 mg KOH/g) [14]. However, the pumpkin seed oil samples in this study seemed to be acidic, since the FFA values were considerably higher than that reported by other authors [14,17].

#### **Peroxide values**

In hot pressing, under microwave radiation for 2 minutes, the PV was lowest at 9.5 meq  $O_2/kg$ . Microwave radiation seemed to increase the PVs in oil (Table 1). Meanwhile, the PV increased with roasting times for treatments at 110°C (from 7.33 to 10.67 meq  $O_2/kg$  in 60 minutes). Similarly, the PV increased with roasting temperatures for treatments in 60 minutes (from 10.8 to 16.3 meq  $O_2/kg$ ). Hence, roasting at higher temperature or prolong roasting may cause the increase of PVs in oil [7].

In cold pressing with microwave pre-treatment, the PVs decreased with increasing microwave durations (from 5.0 to 3.67 meq 02/kg for a period from 1 to 3 minutes), but still higher than the control sample (3.17 meq  $O_2/kg$ ). Similar trend was observed with roasting, the PVs decreased with roasting durations (4.5 meq  $O_2/kg$  in 30 minutes compared to 3.83 meq  $O_2/kg$  in 60 minutes). Regarding the effect of roasting temperature, the PVs of oil sample increased with temperature, the value was highest (6.0 meq  $O_2/kg$ ) at 130°C. Both roasting and microwave may cause the rise in PVs of oil, as observed in this study or somewhere else [9]. Also, Jiao., *et al.* [15] reported the PVs in oil from microwave-assisted aqueous extraction were lower than that in oil from Soxhlet extraction.

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It was observed that the PVs of pumpkin seed oil by cold pressing was were lower than hot pressing. According to Codex STAN 19-1981, the requirement for peroxide value of cold pressed oil should be less than 10 meq  $O_2/kg$  oil. Thus, the oils having low PVs are tend to more stable and less rancid. Thus, the oils extracted by cold pressing may meet the quality requirements [10].

#### Antioxidant capacity, total phenolic and total carotenoid content

The DPPH scavenging capacity of oil samples from hot pressed oil with microwave treatment decreased with increasing times. Further, the DPPH scavenging capacity of the control sample was lower (60.19%), indicating that microwave heating may cause some change in chemical compositions in oil. On the other hand, the effect of roasting time as well as temperature on antioxidant capacity of oil by hot pressing was not significant. It is un-clear trend in change of the DPPH scavenging capacity with roasting time or temperature (Table 2).

Treatments		Antioxidant capacity (% DPPH scavenging)		Total phenolic content (mg GAE/100g samples)		Total carotenoid content (mg β-carotene/kg)	
		Hot pressing	Cold pressing	Hot pressing	Cold pressing	Hot pressing	Cold pressing
Control	0	$60.19 \pm 5.89^{a}$	$54.57 \pm 2.65^{a}$	$84.00 \pm 2.75^{a}$	$43.37 \pm 3.42^{a}$	3409.91 ± 37.24 <sup>a</sup>	1280.14 ± 99.73 <sup>a</sup>
Micro-	1 min	$69.10 \pm 1.80^{b}$	$50.28 \pm 5.24^{a}$	65.93 ± 7.69 <sup>b</sup>	55.10 ± 6.55°	3273.24 ± 148.94 <sup>a</sup>	1795.07 ± 182.05°
wave pre- treatment	2 min	66.89 ± 2.61 <sup>ab</sup>	$40.55 \pm 1.83^{\text{b}}$	51.40 ± 5.97°	$34.27 \pm 0.87^{ab}$	3361.37 ± 83.98 <sup>a</sup>	1878.68 ± 89.48°
	3 min	62.93 ± 2.97 <sup>ab</sup>	39.73 ± 3.29 <sup>b</sup>	41.30 ± 4.55°	29.17 ± 3.32 <sup>b</sup>	3290.05 ± 107.24 <sup>a</sup>	2360.82 ± 115.69 <sup>d</sup>
Roasting pre-treat- ment at 110°C Roasting pre-treat- ment in 60 minutes	30 min	62.71 ± 3.61 <sup>a</sup>	$35.44 \pm 3.74^{b}$	$28.33 \pm 3.18^{d}$	22.83 ± 0.25 <sup>b</sup>	3385.53 ± 244.08 <sup>a</sup>	2280.46 ± 79.10 <sup>d</sup>
	45 min	$59.03 \pm 5.87^{a}$	$42.15 \pm 1.85^{b}$	$66.53 \pm 7.10^{b}$	73.93 ± 8.08°	3286.89 ± 133.93ª	1671.60 ± 21.06 <sup>b</sup>
	60 min	59.73 ± 2.64 <sup>a</sup>	38.69 ± 2.29 <sup>b</sup>	68.53 ± 7.81 <sup>b</sup>	67.23 ± 3.07°	3308.90 ± 78.01 <sup>a</sup>	1852.83 ± 93.96°
	90°C	64.73 ± 2.82 <sup>a</sup>	$42.05 \pm 3.71^{b}$	$74.63 \pm 8.73^{ab}$	$43.27 \pm 4.25^{a}$	3434.06 ± 126.23 <sup>a</sup>	1540.68 ± 65.90 <sup>b</sup>
	110°C	$63.86 \pm 2.16^{a}$	$43.03 \pm 1.55^{\text{b}}$	$70.53 \pm 7.76^{ab}$	62.07 ± 5.52 <sup>b</sup>	3251.19 ± 213.68ª	1807.21 ± 96.35°
	130°C	$62.02 \pm 2.62^{a}$	28.97 ± 2.86°	61.13 ± 5.22 <sup>b</sup>	$33.77 \pm 2.57^{a}$	3060.27 ± 127.98 <sup>b</sup>	1790.09 ± 47.41°

#### Table 2: Effect of pre-treatments on the antioxidant capacity, TPC and TCC.

#### \*Values (means $\pm$ SD) with different index letters are statistically significantly different (p < 0.05) followed by column.

In cold pressing, the DPPH scavenging capacity decreased with increasing microwave times, and they were lower than that of control sample (54.57%). The value was close to that (52.91%) reported in the literature [18]. Further, the DPPH scavenging capacity the oil samples roasted at 110°C at different durations or roasted in 60 min at different temperatures was moderately lower than the control sample. The sharp drop from 42.05 to 28.97% as temperature increased from 90 to 130°C. Heating would lead to change in some chemical compositions in oil, which play a vital role in antioxidant activities such as tocopherols, tocotrienols, total phenols, etc., leading to decrease in antioxidant capacity. In overall, the DPPH scavenging percentage of cold-pressed pumpkin seed oil (54.57%) was almost taken the highest rate compared to soybean (17.4%), rapeseed (51.2%) except hemp oil (76.2%) [11].

The effect of microwave and roasting pre-treatment on TPC by hot press extraction was significant. The TPC values decreased from 84.00 to 41.30 mg GAE/100g samples with increasing microwave durations from 0 to 3 minutes (Table 2). The heat generated by microwave energy in combination with the heat generated by mechanical shear inside the screw barrel may cause to change chemical compositions of pumpkin seed. The TPC increased with roasting durations but decreased with increasing temperatures (28.33 mg GAE/100g in 30 minutes at 110°C vs. 68.53 mg GAE/100g in 60 minutes). The TPC values were always significantly lower than that in the control sample.

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As for cold pressing, the effect of these pre-treatments on TPC was significant, but a bit in different mode. The TPC increased with increasing microwave durations, however, microwave may help increase the TPC in oil compared to no treatment (55.1 mg GAE/100g at 1 minute compared 43.37 mg GAE/100g, respectively). In roasting, changes in the TPC values with durations or temperature were non-linear. Thus, the effect of roasting pre-treatment was significant, and by choosing suitable conditions of time and temperature, the TPC in oil with roasting could get higher than without it.

In literature, the TPC values have been reported in range of 0.5 - 100 mg GAE/100g (Gohari., *et al.* 2011). In this study, the TPC values in hot expelled oil was from 28.33 to 84.00 mg GAE/100g, while that in cold pressed oil between 22.83 - 73.93 mg GAE/100g, comparable to the results from Parry., *et al.* [12] (98 mg GAE/100g). On the other hand, the TPC values reported by Gohari., *et al.* [19] (6.67 mg GAE/100g), Andjelkovic., *et al.* [20] (2.5 - 5.1 mg GAE/100g from different origins) and Vujasinovic., *et al.* [8] (2 mg GAE/100g in roasted cold pressed pumpkin seed oil), were considerably lower. The difference in TPC values might be explained by some reasons as different extraction solvents used [12], different source of materials, lack of a standardized method [21], extraction systems, conditions of processing and storage [19].

Regarding the effect on total carotenoid content (TCC) in pumpkin oils from hot expelling method, both microwave heating and roasting durations have no significant effect. However, roasting the pumpkin seeds at high temperature about 130°C may result in negative effect on TCC (Table 2).

As for cold pressing, microwave pre-treatment yielded significantly higher TCC as compared to no treatment (2360.82 vs. 1280.14 mg/ kg, respectively). Also, microwave duration had significant effect on the TCC increase. Roasting at 110°C resulted in higher TCC than without roasting. However, a minimum with roasting duration suggested that the roasting time should be within 30 minutes at 110°C. On the other hand, a maximum in TTC with roasting temperature suggested that temperature should be kept within 110°C, not to excess 130°C.

Pumpkin seeds are considered as a good source of carotenoids, which are present in all seed oils. The contents of tocopherols and carotenoids were found abundant in pumpkin seed oil [2]. The TCC in pumpkin seed oil were found in differentiation range away from other researches. For instance, the total carotenoids in pumpkin seed (*C. pepo*) oil was about 55.9 mg/kg [8] measured by using UV-spectrophotometer at 455 nm; 27.48 mg/kg measured at 670 nm for *C. maxima* seed oil [4] and 6.84 mg/kg [18], which were much lower than the values in this study. On the other hand, Durante., *et al.* [3] found the TCC in pumpkin oil (*C. moschata*) were much higher than the values in this study (23920 and 11710 mg/kg of oil extracted by Soxhlet and supercritical CO2 method, respectively). The different results might be as a result of several reasons such as raw materials, pressing methods, wavelength variation, etc.

## Fatty acid composition

The fatty acids profiles of pumpkin seed oils obtained by two methods were quite similar. In the cold-pressed oil, the largest proportion is oleic acid (35.48%), followed by linoleic (31.63) and palmitic acid (19.94%).

Therefore, it could be seen that the pumpkin (*C. pepo*) seed oil was rich in unsaturated and short chain fatty acids. This result was similar to other studies such as Rabrenović., *et al.* [23] and Jafari., *et al.* [22] (Table 3). However, the amount of linoleic acid in this study was lower than oleic acid, in contrast to observation from other authors [2,17]. The high content of total USFA in pumpkin seed oil (~70%) makes it desirable for health benefits.

	Oil yield (%, w/w)							
Name of fatty acids	This study		Vujasinovic., <i>et al</i> . [17]	Siano., <i>et al</i> . [2]	Jafari., <i>et al</i> . [22]	Rabrenović., <i>et al.</i> [23]		
Name of facty actus	Hot pressing	Cold pressing	Screw pressing	Soxhlet extraction	Soxhlet extraction	Cold pressing		
Myristic acid (C14:0)	0.13	0.13	-	-	-	0.2		
Palmitic acid (C16:0)	19.68	19.94	11.88	17.58	10.56	11.8		
Linoleic acid (C18:2)	31.17	31.63	52.15	47.45	28.06	40.8		
Oleic acid (C18:1)	36.24	35.48	30.35	25.54	52.65	40.7		
Stearic acid (C18:0)	8.27	9.424	6.93	7.62	8.54	6.2		

Table 3: Effect of extraction methods on the fatty acid composition

#### Conclusions

In this study, both thermal pre-treatment and mechanical pressing method caused the change in quantity and quality of pumpkin seed oil.

Both microwave radiation and roasting at certain degree increased the yield of oil, provided the appropriate treatment conditions are selected (duration or duration and temperature in combination).

Also, both microwave radiation and roasting may cause the increase in PV of the cold-pressed oils, however, their effect on FFA was not noticeable in hot-pressed oils and in the cold-pressed oils, while roasting may lead to decrease in FFA in hot-pressed oils.

Microwave radiation caused the increase in antioxidant capacity but decrease in both TPC and TCC in hot-pressed oil. Nevertheless, microwave treatment lead to decrease in antioxidant, TPC but increase in TCC of the cold-pressed oils.

Similar trend of the effect of roasting on TPC, TCC was observed for oils obtained from the two pressing methods.

Both microwave radiation and roasting showed no effect on the fatty acid profile of the oils obtained from the two pressing methods.

In general, the oils from cold pressing were of better quality than that from hot pressing. Also, the oil yield from cold pressing was higher than that from hot screw expelling.

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