

EFFECTS OF ROASTING TEMPERATURE AND TIME ON HEALTHY NUTRACEUTICALS OF ANTIOXIDANTS AND TOTAL PHENOLIC CONTENT IN IRANIAN SESAME SEEDS (SESAMUM INDICUM L.)

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ABSTRACT

Sesame seed (*sesamum indicum* L.) is one of the world's most important and oldest oilseed crops with a high level content of antioxidant known to human health. The antioxidant factors responsible for the stability of roasted sesame seeds is highly affected by the conditions of the roasting process. Survey of the roasting temperature and time effects on antioxidants and total phenolic content in Iranian sesame seeds was the aim of this investigation. Spectrophotometer methods based on folin-ciocalteau reagent for determination of total phenolic content (TPC) and Ferric Reducing Antioxidant Power assay (FRAP) technique for total antioxidant activity were used before and after different roasting processes. Some of 8 Iranian sesame seeds cultivares were studied (n = 160), including *Branching Naz*, *Non Branching Naz*, *Dezful*, *Darab*, *Karaj*, *Moghan*, *Varamin* and *Black sesame*. The range of FRAP values was between $0.301 \pm 0.029 \mu\text{M}$ and $1.746 \pm 0.083 \mu\text{M}$ in Moghan and Branching Naz sesame seed cultivares, respectively. The FRAP value increased from $0.974 \pm 0.095 \mu\text{M}$ in unroasted Branching Naz as a control to $1.746 \pm 0.083 \mu\text{M}$ after roasting in 200°C for 20min. Also TPCs increased significantly as the roasting temperature. The amount of TPC varied in different sesame cultivars from $20.109 \pm 3.967 \mu\text{M}$ to $129.300 \pm 3.493 \mu\text{M}$ in Varamin and Branching Naz sesame seed cultivares, respectively; also TPC increased from $70.953 \pm 5.863 \mu\text{M}$ in unroasted Branching Naz sesame seed as a control to $129.300 \pm 3.493 \mu\text{M}$ after roasting in 200°C for 20 min. Branching Naz sesame seed cultivare was at the highest level in total antioxidants and total phenolic contents in comparison to other samples; however Moghan and Varamin cultivares were at the lowest level in total antioxidants and total phenolic contents, respectively. The present study showed that Iranian sesame seed can be considered as a good source of natural antioxidant specially after roasting. The optimum temperature and time roasting to obtain the most antioxidants and total phenolic content was 200°C for 20 min.

Key words: Sesame seed, Phenolic compound, Antioxidant activity, Roasting temperature, Roasting time

INTRODUCTION

The sesame seeds have some potential nutraceutical compounds such as phenolic and tocopherols with antioxidant activity that have significant effect on reducing blood pressure, lipid profile and degeneration of vessels impact reducing chronic diseases. It has been shown that roasting can increase the total phenolic and antioxidants compounds and activity.

The use of natural antioxidants in foods such as flavonoid, tannins, coumarins, phenolics and terpenoids is recently at special attention because of the world wide trend to avoid or minimize the use of synthetic food additives (Mohamed and Awatif, 1998). Some components from fruits, vegetables and seeds extracts have shown strong antioxidant effect in model systems (Salunke and Chavan, 1992; Hochstein and Atallah, 1998; Sadeghi *et al.*, 2009). Phenolic compounds are

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widely distributed in the plant. These compounds are known as important antioxidants because of their ability to donate hydrogen atom or an electron in order to form stable radical intermediate. They prevent the oxidation of various biological molecules. In fact, several oilseeds and their byproducts have been investigated for phenolic compounds for safe sources of natural antioxidant (Namiki, 1995; Jeong *et al.*, 2004).

Sesame seed (*Sesamum indicum* L.) is one of the world's most important and oldest oilseed crops with a high level content of antioxidant known to human health (Abou-Gharbia and Shahidi, 1997). Sesame has long been regarded in the orient as a health food for energy increasing and aging prevention (Hajimahmoodi *et al.*, 2008b). Sesame oil compounds have multiple physiological functions, such as estrogenic activity, providing anti-inflammatory functions, decreasing blood lipids and arachidonic acid levels (Hirata *et al.*, 1996; Kita *et al.*, 1998), and increasing antioxidative ability and γ -tocopherol bioavailability. Sesame seeds are composed of 43% - 50% lipid, 5% - 6% moisture, 10% -15% carbohydrate, 5% - 6% ash, 4% - 5% fiber and 15% - 20% protein.

It is well-known that sesame has many functions for maintaining good health and it has been known for many years that sesame oil is highly resistant to oxidative deterioration (Nagata *et al.*, 1987; Halliwell, 1997). Its remarkable stability is due to the presence of a large quantity of endogenous antioxidants such as sesaminol, sesamol and α -tocopherol (Niwa *et al.*, 1986; Fukuda and Namki, 1988; Abou-Gharbia *et al.*, 2000).

Sesame oil prepared from roasted sesame seeds has a distinctive flavor and longer shelf life and is used as cooking oil and margarines (Yoshida and Takagi, 1997). The roasting process is the key step for making sesame oil, since it affects the color, composition and the quality of sesame oil. The antioxidant factors responsible for the stability of roasted sesame seeds is highly affected by the conditions of the roasting process. Therefore, in order to make good quality, the optimum roasting conditions should be established. The purpose of the present study was to elucidate the relationship between roasting temperature and time on the antioxidant activity and phenolic content of

extract from Iranian sesame seeds when roasted in electric oven.

MATERIALS AND METHODS

Materials

All solvents/chemicals used were of analytical grade and obtained from Merck Co. (Darmstadt, Germany). Double-distilled deionized water was used for the preparation of aqueous solutions.

Sampling of sesame seeds

For 2 different measurements, totally (n=160) samples from eight cultivars of sesame (*Sesamum indicum* L.) including *Branching Naz*(n=10), *Non Branching Naz*(n=10), *Dezful*(n=10), *Darab*(n=10), *Karaj*(n=10),, *Moghan*(n=10),, *Varamin*(n=10), and *Black sesame*(n=10), were donated from Karaj Seed and Plant improvement Institute.

Heat treatment

Whole sesame seeds were placed in Pyrex Petri dish (8.0 cm diameter) and roasted in an oven (Model UNB 100, Schwa Bach, Germany) at 180 °C, 200 °C and 220°C for 10, 15 and 20 minutes.

Sample preparation

200 mg of samples was extracted for 2hr with 2mL of 50% methanol v/v at room temperature on an orbital shaker set at 200 rpm. The mixture was centrifuged (Heraeus Germany) at 1000 g for 15 min and the supernatant was decanted into 4 mL vials. The pellets were combined and used for total phenolic content Measurements and total antioxidant assay.

Total antioxidant activity assay

(Ferric Reducing Antioxidant Power assay) (FRAP) procedure described by Benzie and Strain, was followed (Benzie and Strain, 1996; Hajimahmoodi, Hanifeh *et al.*, 2008a). The principle of this method is based on the reduction of a ferric tripyridyl-triazine (TPTZ) complex to its ferrous colored form in the presence of antioxidant. FRAP reagent contained 5 mL of a (10 mmol/L) TPTZ solution in 40 mmol/L HCL plus 5mL of (20 mmol/L) FeCl₃ and 50 mL of acetate buffer, and at pH=3.6, and was prepared

freshly and warmed at 37°C. 50 mL extract were mixed with 3ml FRAP reagent and the absorbance of reaction mixture at 593 nm was measured by spectrophotometer (Cintra 40 model) after incubation at 37°C for 10 min. For construction of calibration curve, five concentrations of FeSO₄.7H₂O (125, 250, 500, 750 and 1000) were used.

Total phenolic content (TPC) analysis

Total phenolics were determined colorimetrically using folin-ciocalteu reagent as described by Velioglu *et al.* (Velioglu *et al.*,1998). The extract (100µ) was mixed with 1.5 mL of folin cicalteau reagent and allowed to stand at 22°C for 5 min. 1.5mL sodium bicarbonate solution (60g/L) was added to the mixture. After 90 min at 22°C, absorbance was measured at 725 nm using a UV-visible spectrophotometer.

Statistical analysis

All experiments were carried out in triplicate. Statistical analysis was performed using SPSS software. Data were expressed as Mean±SD or as percentage. Variables were compared by T test, one-way Anova; post hoc Tukey Test and the significance of differences among means were determined at p <0.05.

RESULTS

Precision

The within-run coefficient of variation (CV) was estimated by assay of FRAP and phenol value of standards and the unroasted sesame seed cultivar (sample) three times in the same analytical Run. Between Run was obtained by estimating four different days. The results are shown in Tables 1 and 2.

Effect of seed roasting conditions on the antioxidant activity

The antioxidant activities of sesame seed extracts have been evaluated using FRAP method. The FRAP value of 8 cultivars of Iranian sesame seeds was determined before and after roasting at 180 °C, 200 °C and 220°C for 10, 15, 20 minutes (n = 80). The range of FRAP values was between 0.301±0.029µM and 1.746±0.083µM in *Moghan* and *Branching Naz* cultivars that differ significantly (p<0.001), respectively. The FRAP value increased from 0.974±0.095 µM in unroasted *Branching Naz* as a control to 1.746±0.083µM after roasting in 200°C for 20min that differ significantly (p< 0.001) (Table 3).

Effects of seed roasting conditions on the total phenolic content

Level of total phenolics was determined by the folin-ciocaltea method. The total phenolic compound in methanolic extract of 8 cultivars of Iranian sesame seeds was determined before and after roasting at 180°C, 200°C and 220°C for 10, 15 and 20 minutes (n = 80). TPCs increased significantly with the roasting temperature. The amount of TPC varied in different sesame cultivars from 20.109±3.967µM to 129.300±3.493 in *Varamin* and *Branching Naz* cultivars, respectively; also TPC increased from 70.953±5.863 µM in unroasted *Branching Naz* as a control to 129.300±3.493 µM after roasting in 200°C for 20 min (Table 4).

DISCUSSION

The present study was undertaken to evaluate the effects of seed roasting conditions on the antioxidant activity and the amount of TPC in 8 brands of Iranian sesame seeds (n=160).

Table 1: The precision test for FRAP

Standards/sample (µmol/mL)	FRAP Method	
	Within Run (CV %)	Between Run (CV %)
0.125	0.298	10.600
0.250	1.065	10.521
0.500	2.824	10.644
0.750	3.893	8.382
1.00	3.543	4.231
Branching naz	1.200	8.681

Table 2: The precision test for Phenol Method

Standards/sample (µmol/mL)	Phenol Method	
	Within Run (CV %)	Between Run (CV %)
25	0.321	7.465
50	0.259	8.312
75	0.181	8.579
100	0.389	5.382
125	0.218	8.725
150	0.918	10.132
Branching naz	0.851	7.782

Table 3: The effect of seed roasting conditions on the antioxidant activity of sesame seeds (FRAP value: mean \pm SD, $\mu\text{mol/mL}$)

Cultivar	Unroasted	180°C 10min	180°C 15min	180°C 20min	200°C 10min	200°C 15min	200°C 20min	220°C 10min	220°C 15min	220°C 20min
Darab	0.33 \pm 0.03	0.47 \pm 0.04	0.70 \pm 0.09	0.72 \pm 0.07	0.89 \pm 0.09	1.04 \pm 0.09	1.26 \pm 0.08	1.15 \pm 0.09	1.13 \pm 0.07	1.19 \pm 0.08
Varamin	0.42 \pm 0.02	0.48 \pm 0.05	0.65 \pm 0.02	0.66 \pm 0.08	0.82 \pm 0.09	1.03 \pm 0.09	1.23 \pm 0.12	1.09 \pm 0.09	0.97 \pm 0.08	0.95 \pm 0.04
Non Branching Naz	0.43 \pm 0.04	0.63 \pm 0.08	1.05 \pm 0.09	1.07 \pm 0.09	1.16 \pm 0.05	1.03 \pm 0.07	1.47 \pm 0.09	1.28 \pm 0.08	1.25 \pm 0.09	1.24 \pm 0.09
Branchjng Naz	0.97 \pm 0.09	1.37 \pm 0.06	1.40 \pm 0.05	1.42 \pm 0.08	1.53 \pm 0.06	1.70 \pm 0.05	1.74 \pm 0.08	1.43 \pm 0.07	1.48 \pm 0.07	1.47 \pm 0.07
Karaj	0.31 \pm 0.02	0.50 \pm 0.08	0.57 \pm 0.04	0.58 \pm 0.07	0.81 \pm 0.07	0.92 \pm 0.09	1.08 \pm 0.10	1.02 \pm 0.04	0.94 \pm 0.09	0.92 \pm 0.08
Moghan	0.30 \pm 0.02	0.49 \pm 0.05	0.72 \pm 0.03	0.73 \pm 0.05	0.84 \pm 0.07	1.07 \pm 0.08	1.30 \pm 0.09	1.06 \pm 0.06	0.89 \pm 0.05	0.89 \pm 0.06
Dezful	0.33 \pm 0.03	0.66 \pm 0.06	0.93 \pm 0.03	0.99 \pm 0.05	1.04 \pm 0.05	1.20 \pm 0.07	1.39 \pm 0.08	1.29 \pm 0.08	1.14 \pm 0.06	0.99 \pm 0.08
Black sesame	0.70 \pm 0.01	0.76 \pm 0.03	0.77 \pm 0.04	0.80 \pm 0.04	1.01 \pm 0.09	1.03 \pm 0.08	1.34 \pm 0.08	1.15 \pm 0.09	1.08 \pm 0.09	1.03 \pm 0.07

Table 4: The effect of seed roasting conditions on the phenol content of sesam seeds (mean \pm SD, $\mu\text{mol/mL}$)

Cultivar	Unroasted	180°C 10min	180°C 15min	180°C 20min	200°C 10min	200°C 15min	200°C 20min	220°C 10min	220°C 15min	220°C 20min
Darab	20.31 \pm 2.29	26.40 \pm 2.31	48.84 \pm 1.91	49.71 \pm 1.79	73.70 \pm 2.28	89.04 \pm 1.94	98.41 \pm 2.14	74.30 \pm 1.72	70.41 \pm 2.89	66.53 \pm 2.15
Varamin	20.10 \pm 3.96	34.64 \pm 2.38	53.93 \pm 2.79	56.41 \pm 2.91	76.04 \pm 2.79	87.76 \pm 2.71	102.43 \pm 3.05	88.70 \pm 2.21	78.45 \pm 1.97	71.62 \pm 2.18
Non Branching Naz	22.32 \pm 1.36	44.22 \pm 1.18	60.63 \pm 1.58	63.11 \pm 1.89	96.34 \pm 2.56	105.65 \pm 3.38	106.52 \pm 3.89	104.04 \pm 3.97	90.84 \pm 1.47	86.49 \pm 2.39
Branchj Naz	70.95 \pm 5.89	83.81 \pm 3.79	92.25 \pm 3.41	93.12 \pm 3.71	109.07 \pm 2.27	117.73 \pm 3.47	129.30 \pm 3.49	124.20 \pm 3.84	119.64 \pm 3.71	112.62 \pm 3.56
Karaj	24.53 \pm 3.38	35.91 \pm 2.24	47.57 \pm 2.11	50.38 \pm 2.11	71.95 \pm 2.29	78.92 \pm 2.71	88.97 \pm 2.11	75.77 \pm 2.91	71.28 \pm 2.11	68.60 \pm 2.39
Moghan	25.53 \pm 3.11	37.32 \pm 3.21	52.33 \pm 2.11	56.48 \pm 2.24	76.37 \pm 2.91	89.64 \pm 2.91	93.86 \pm 3.76	83.14 \pm 2.41	74.57 \pm 2.32	69.41 \pm 2.38
Dezful	26.33 \pm 2.51	46.30 \pm 1.71	60.32 \pm 1.42	64.65 \pm 1.41	81.13 \pm 1.79	97.68 \pm 2.11	108.46 \pm 3.11	89.30 \pm 2.78	81.40 \pm 1.90	70.75 \pm 1.84
Black sesame	28.95 \pm 5.48	49.51 \pm 2.47	52.26 \pm 3.24	57.69 \pm 2.35	83.14 \pm 3.71	90.84 \pm 3.28	110.66 \pm 5.41	100.83 \pm 3.21	90.44 \pm 3.21	84.55 \pm 2.99

The results show that the antioxidant activity and the amount of TPC increased significantly as the roasting temperature and time until 200°C for 20 min, and then decreased by roasting at 220°C. So, the highest activity and content were achieved by roasting at 200°C for 20 min (Figs 1 and 2) .

Non Branching Naz and *Branching Naz* cultivares had high potential antioxidant activities as the total phenolic content than the others ($p < 0.001$), but other cultivars had no significantly difference with each other.

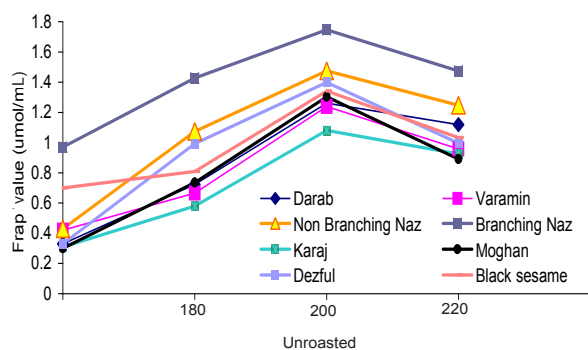


Fig. 1: Antioxidant activity of sesame seed ($\mu\text{mol/mL}$) in 180-220 °C for 20min

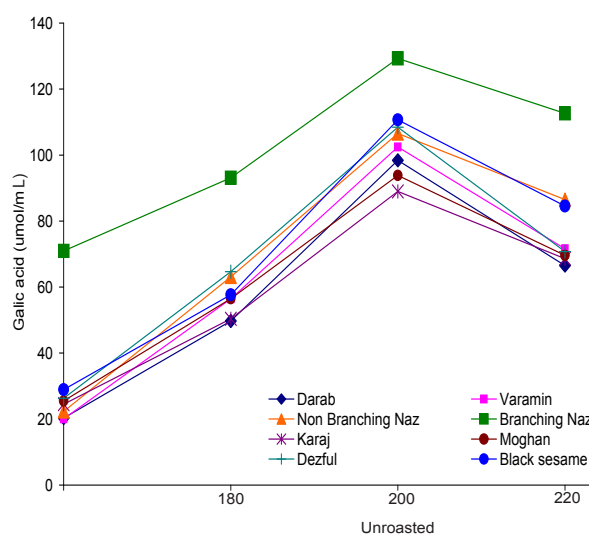


Fig. 2: Total phenolic content of sesame seed ($\mu\text{mol/mL}$) in (180-220) °C for 20min.

The oxidative stability of sesame oil is superior to that of other vegetable oils, even though it contains nearly 85% unsaturated fatty acids. Its remarkable stability is due to the presence of a large quantity of endogenous antioxidants and phenolic compounds, comprised of sesamin, sesamol, and γ -tocopherol (Sadeghi *et al.*, 2009).

Some studies were undertaken to evaluate the effects of seed roasting conditions on the antioxidant activity. Jeong study agrees with our results, in which the total phenolic content, radical scavenging activity, reducing powers, and antioxidant activity of sesame meal extract increased; and several low-molecular weight phenolic compounds such as 2-methoxyphenol, 4-methoxy—3-methylthio-phenol, 5-amino-3-oxo-4-hexenoic acid, 3,4-methylenedioxyphenol (sesamol), 3-hydroxy benzoic acid, 4-hydroxy benzoic acid, vanillic acid, filicinic acid, and 3,4-dimethoxy phenol were newly formed in the sesame meal after roasting sesame seeds at 200°C for 60 min (Jeong *et al.*, 2004).

The effects of sesame seed pretreatment, including roasting (R), roasting plus steaming (RS) and microwaving (M) on crude oil quality from intact Egyptian and Sudanese seeds were investigated by Abou-Gharbia.

Oxidative stability of sesame seeds was determined by monitoring changes in peroxide value and para-anisidine value. The oils from raw seeds and RS seeds showed higher oxidative stability than other processed oils. The influence on all components after RS was more pronounced than M treatment (Abou-Gharbia *et al.*, 2000). The relatively greater oxidative stability of oils from RS treatment which was observed in seeds may be resulting from the formation of some antioxidants (sesamol) from the degradation of other native compounds; sesaminol (Yen, 1990). The amount of chlorophyll and sesamol decreased with increasing roasting temperature. However, the highest level of sesamol and γ -tocopherol was found in oils prepared with a 200-220° roasting temperature. The sesame oil prepared at a 200°C roasting temperature had the best flavour score when compared with the others. Yoshida and Takagi (Yoshida and Takagi 1997) reported that sesamol a potent phenolic

antioxidant increased as the roasting temperature of sesame seed increased to 180°C or higher, but sesamol was almost depleted after 25 min of roasting. Burning and bitter tastes were found in the oils prepared at roasting temperatures over 220°C. The results suggested that a high-quality product would be obtained by roasting for 25 min at 160 or 180°C, 15 min at 200°C and 5 min at 220°C. Sesame oil prepared from roasted sesame seeds has a distinctive flavor and longer shelf-life. Kim believes that the storage stability of unroasted sesame oil is low, but roasting of sesame seed at 170°C or higher significantly increased stability of sesame oil. The highest stability was achieved by roasting at 200°C (Kim, 2000).

Shahidi *et al.*, (1997) have shown that the effect of processing (R, S, and RS) on changes in the sesamin content in oils from coated seeds was low and generally did not exceed 20% of the original values. On the other hand, oils from hulled seeds underwent a more pronounced decrease in their sesamin content than the oil from coated seeds (Shahidi *et al.*, 1997)

Duh has reported that reducing properties of antioxidants are generally associated with the presence of reductions (Duh, 1998). The reducing power of defatted sesame meal extracts increased with heat treatment at 150°C for 40 min. Also the induction time of lard increased from 0.74 h to 0.86 h when roasted at 100° C for 60 min and 0.74h to 1.04 h when 150°C 40 min was used (Chen and Ho, 1997).

In conclusion, the results indicate that antioxidant activity of sesame extracts was significantly affected by roasting temperature and time. To obtain the highest antioxidant activity and total phenolic content from sesame extracts, sesame seeds should be roasted at 200°C for 20 min. From ancient times to today, sesame has been considered to be a valuable oil-seed, not only because of its high oil content, but also because of its medical effects. Some valuable components in sesame contribute to a nutritional and functional food for humans. Our results indicated that Iranian sesame seeds possess antioxidant, so the seeds can be easily incorporated into a normal diet at a level that might benefit health as a natural antioxidant.

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