Journal of Oleo Science

doi : 10.5650/jos.ess19205
J. Oleo Sci. **69**, *(5) 423-428 (2020)*

JOURNAL OF OLEO SCIENCE
Jos

Influence of Roasting on Oil Content, Bioactive Components of Different Walnut Kernel

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Abstract: A study was carried out to evaluate oil contents, fatty acid composition and tocopherol contents of several walnut types in relation to roasting process. The major fatty acid identified was linoleic acid in both roasted and unroasted walnut oils. Linoleic acid contents of unroasted walnut oil varied from 46.44 (Type 9) and 63.59% (Type 7), while the linoleic acid contents of roasted walnut oils at 120℃/h ranged from 55.95% (Type 3) to 64.86% (Type 10). Interestingly, linolenic acid contents of both roasted and unroasted oils changed between 9.43 (Type 10) and 16.29% (Type 8) to 9.64 (Type 10) and 16.58% (Type 8), respectively and were significant ($p < 0.05$) different. γ-tocopherol content of unroasted walnut oils varied between 6.3 (Type 3) and 11.4 mg/100g (Type 1) and γ-tocopherol contents of roasted walnut oils ranged between 28.1 (Type 8) and 38.2 mg/100g (Type 3). The oil could be useful for industrial applications owing to good physicochemical properties. Fatty acid values for oil obtained from roasted walnut were slightly higher than those reported for unroasted walnut oils.

Key words: walnut kernel, roasting, oil, fatty acid, tocopherol, GC, HPLC

1 Introduction

Walnut (*Juglans regia* L.) belongs to Juglandaceae family, and Turkey plays an important role in walnut production in the world[1-4]. Nuts are rich in protein, carbohydrate, unsaturated fatty acid, vitamin and minerals[5, 6]. In several studies, the oil present in walnut kernels varied between 52 and 70%[2, 4, 7, 8]. According to[4, 8, 9] linoleic, linolenic and oleic fatty acids are the prominent fatty acids present in walnut kernel. Roasting is an important operation in nuts processing and majority of physic-chemicals observed in nuts result from roasting operation[10]. Heat treatments including roasting are now commonly used in many food processing operations such as baking, sterilization, drying, cooking, temperingetc[11, 12]. In addition, traditional method of heating often transfer heat slowly from the surface of products to the center and some also have undesirable effects on products quality[13]. Roasting operations have been shown to have desirable effects on the flavour, colour, fatty acid profile and bioactive components of kernel and seeds[14, 15]. The normal temperaure range used for roasting of edible nut ranges from 100-180℃ for 5-60 min and the usual industrial procedure used in roasting in hot air method although different new techniques are also being applied depending on the intended final use[16]. Roasted nuts are being consumed as snacks with or without any additives and spices beside being important industrial raw material for preparing different baked, confectionary and other food products. The roasting process for nuts is indeded for certain desirable changes however, due to high polyunsaturated fatty acids contents, malnut roasting may require mild roasting temperature to avoid oxidantion reaction of fatty acids[17].

The aim of this study was to determine the effects of the roasting process (mild temperature of 120℃ for 30 min) on the oil and tocopherol contents and the fatty acid profile of different types of walnut kernels and their oils. Results were also compared with those of raw walnut kernels.

2 Materials and Methods

2.1 Material

Walnut samples were provided from Ermenek (Karaman) distinct in Turkey. They walnuts were dried in oven at 70℃ and the kernels were then separated from the hulls. The kernels were thereafter cleaned and kept at 4℃ in a poly-

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Accepted February 6, 2020 (received for review August 18, 2019)
Journal of Oleo Science ISSN 1345-8957 print / ISSN 1347-3352 online
http://www.jstage.jst.go.jp/browse/jos/ http://mc.manusriptcentral.com/jjocs

propylene bag for analysis.

2.1.1 Reagents

All reagents used in this present study were of analytical grade. Petroleum ether, HPLC grade tert-butyl methyl ether and heptanes were obtained from Merck, Darmstadt, Germany. Tocopherol and tocotrienol standards used were procured from CalBiochem (Darmstadt, Germany).

2.2 Methods

2.2.1 Roasting

Walnut kernels were roasted at 120℃ for 30 min on an oven. The heating was completed when color of kernels was dark brown, and cooled.

2.2.2 Oil extraction

Walnut oil was extracted from ground walnut kernel using light petroleum ether in a Soxhlet apparatus for 5 h. Rotary vaccum evaporator was used to recover the remaining oil at 50℃. The walnut oil obtained was kept in colored glass bottles at −18℃ for analysis.

2.2.3 Fatty acid analysis

A gas chromatographic procedure[18] was used to study the fatty acid profile of oil from walnut kernels. Walnut oil drop was added to *n*-heptane (1 mL) in a tube followed by which 50 μg sodium methylate was added. The tube was mixed thoroughly for 1 min at room temperature. Centrifugation 4500 × g for 10 min was carried out after adding 100 μL of water followed by removal of lower aqueous phase. HCL (50 μL) and 1 mol methyl orange was mixed and afterwards the lower aqueous phase was discarded. To the reaction mixture, 20 mg of sodium hydrogen sulphate was added and centrifugation was carried out at 4500 × g for 10 min. The upper phase in *n*-heptane was injected into a gas chromatography (Varian 5890) having a capillary column [CP-Sil 88 (100 m long, 0.25 mm ID, film thickness 0.2 μm)]. The analysis was closely monitored to ensure that injection block and detector temperature was constant at 260℃. Nitrogen served as the mobile phase at a flow rate of 1.51 ml/min, while 80 ml/min was set as total flow rate and 1/40 was split rate. The column temperature was set at 120℃ for 5 minutes and raised to 240℃ at 4℃/min and held at 240℃ for 25 min. The fatty acid present in the walnut kernel oils were determined after comparison of the retention time of sample with those of appropriate fatty acids standards[16].

2.2.4 Determination of tocopherol present in walnut oil

Tocopherol content was determined using HPLC (Merck, Darmstadt, Germany) as described by Balz *et al.*[17]. About 20 μL solution containing 250 mg of walnut oil in 25 mL of *n*-heptane was immediately infused into the Diol phase column (25 cm × 4.6 mmID) of the HPLC at 1.3 mL/min flow rate and tocopherol content was determined accordingly. Wavelength of 295 nm was used for excitation, while 330 nm wavelength was used for the emission. The tocopherols (α, β, γ and δ-tocopherol) were quantified using standard solutions having concentration between 0-100 mg/L[19].

2.3 Statistical analysis

In other to avoid errors due to variation, a completely randomized block design was used and data obtained were analyzed using Analysis of Variance (ANOVA) JMP version 9.0 (SAS Inst. Inc., Cary, N.C.U.S.A). All analyses were done in triplicates and result expressed as mean ± standard deviation[20].

3 Results and Discussion

The effect of roasting on fatty acid composition and oil yield of walnut kernels are shown on Table 1. The result revealed that roasting significantly ($p<0.05$) affected the oil content of walnut kernels. The oil content of the raw (unroasted) and roasted walnut kernels increased from 23.5% (Type 1) and 34.5% (Type 9) to 25.0% (Type 1) and 38.4% (Type 9), respectively. The oil content of roasted walnut kernels increased compared to unroasted walnut kernels. The inccrease in oil content may be attributed to evaporation of water from the walnut during the roasting. Consequently, significant ($p<0.05$) differences was observed between raw and roasted walnut samples ($p<0.05$). With respect to the fatty acids in walnut oil samples, palmitic, oleic and linoleic acids were major fatty acids of walnut oils. Linoleic acid was the key fatty acid for both unroasted and roasted walnut oils. The linoleic acid contents of unroasted walnut oil change between 46.44% (Type 9) and 63.59% (Type 7) and roasting at 120℃/h also changed the linoleic acid contents of the oil from 55.95% (Type 3) to 64.86% (Type 10). Interestingly, the linolenic acid contents of raw and roasted walnut kernel oils changed from 9.43% (Type 10) to 16.29% (Type 8) and 9.64% (Type 10) to 16.58% (Type 8). Additionally, significant ($p<0.05$) statistical difference was observed in the oleic acid present in roasted walnut oil as the values changed from 14.22% (Type 8) to 23.53% (Type 3). However, no significant ($p>0.05$) difference was observed in Type 1, 2, 4 and 6. As shown in Table 1, 80% of fatty acids in walnut samples were polyunsaturated fatty acids. The fatty acids identified in the walnut oil are palmitic (6.88-7.64%), stearic (2.32-3.40%), oleic (14.99%-22.72%), linoleic (57.24%-60.88%) and linolenic acids (5.31-12.13%)[4]. The oleic, linoleic and linolenic acid contents of walnut oil changed between 15.9 and 23.7%, 57.2 and 65.1%, and 9.1-13.6%, respectively[3]. Oil contents of walnut kernels can change between 50% and 70% depending on the agronomic conditions[1, 2, 5, 21, 22]. All these values are in good agreement with previous researches[4, 7]. While our results related to unroasted walnut kernel are found partly similar compared to literature values, the results of roasted walnut fatty acids showed partly differences com-

Table 1 Oil content of fatty acid composition of unroasted (raw) and roasted walnut (Type 1-10) kernels (%).

Unroasted (Raw)	T1	T2	T3	T4	T5	T6	T7	T8	T9	T10
Oil %	23.5 ± 1.13*f	34.1 ± 1.29a	27.9 ± 2.09d	26.5 ± 1.67e	30.0 ± 2.35c	29.3 ± 2.41c	32.5 ± 1.47b	33.4 ± 1.32b	34.5 ± 2.31a	27.2 ± 1.53d
C16:0	7.07 ± 0.17a**	6.27 ± 0.09b	5.65 ± 0.21c	6.45 ± 0.35b	6.49 ± 0.28b	6.26 ± 0.89b	6.31 ± 0.69b	6.01 ± 0.47b	5.10 ± 0.29c	6.74 ± 0.63b
C16:1	0.14 ± 0.03b	− ***	0.01 ± 0.00e	0.01 ± 0.00e	0.15 ± 0.01a	0.02 ± 0.01e	0.13 ± 0.03c	0.04 ± 0.01d	0.06 ± 0.01d	0.02 ± 0.01e
C18:0	2.50 ± 0.13b	2.99 ± 0.11b	2.60 ± 0.09b	2.78 ± 0.17b	2.83 ± 0.19b	2.84 ± 0.21b	2.71 ± 0.09b	3.51 ± 0.16a	2.19 ± 0.13b	2.64 ± 0.17b
C18:1	14.29 ± 1.17f	15.73 ± 0.98e	20.19 ± 0.01b	16.18 ± 1.21d	14.72 ± 1.43f	15.78 ± 1.14e	15.18 ± 0.67e	14.96 ± 0.86f	33.99 ± 1.23a	17.68 ± 1.56c
C18:2	61.40 ± 1.42b	61.49 ± 1.67b	57.62 ± 1.34e	60.63 ± 1.56c	60.69 ± 1.73c	59.93 ± 1.39	63.59 ± 1.68a	59.02 ± 1.91d	46.44 ± 0.68f	63.43 ± 1.27a
C18:3	14.47 ± 0.78c	13.45 ± 0.93d	13.85 ± 0.97d	13.88 ± 0.68d	15.02 ± 1.03b	15.04 ± 1.27b	12.00 ± 0.98e	16.29 ± 1.56a	12.09 ± 0.74f	9.43 ± 0.64g
C20:0	0.07 ± 0.01c	0.07 ± 0.01c	0.07 ± 0.01c	0.07 ± 0.01c	0.07 ± 0.01c	0.13 ± 0.03b	0.06 ± 0.02d	0.16 ± 0.03a	0.13 ± 0.01b	0.06 ± 0.01d
Roasted	**T1**	**T2**	**T3**	**T4**	**T5**	**T6**	**T7**	**T8**	**T9**	**T10**
Oil %	25.0 ± 1.45f	37.1 ± 2.38a	31.3 ± 2.57e	28.8 ± 1.68	34.2 ± 1.77d	34.3 ± 1.59d	35.1 ± 1.61c	36.2 ± 2.34ab	38.4 ± 1.89a	30.6 ± 1.75e
C16:0	7.13 ± 0.56a	6.01 ± 0.93b	5.61 ± 0.86c	6.08 ± 0.89b	6.47 ± 1.03b	6.24 ± 1.13b	6.16 ± 1.21b	5.90 ± 0.94c	5.57 ± 0.83c	6.33 ± 0.79b
C16:1	0.15 ± 0.03a	−	0.14 ± 0.03a	0.13 ± 0.01b	0.02 ± 0.01d	0.07 ± 0.01c	0.01 ± 0.01	0.11 ± 0.03b	0.03 ± 0.01d	0.02 ± 0.01d
C18:0	2.57 ± 0.13b	2.79 ± 0.09b	2.46 ± 0.011b	2.94 ± 0.32b	2.80 ± 0.37b	2.79 ± 0.27b	2.66 ± 0.21b	3.49 ± 0.46a	2.66 ± 0.34b	2.46 ± 0.33b
C18:1	15.11 ± 0.41c	15.68 ± 0.49c	23.53 ± 0.32a	16.82 ± 1.24c	14.46 ± 1.18d	15.22 ± 1.31c	14.90 ± 1.37d	14.22 ± 1.29d	21.62 ± 1.41b	16.60 ± 1.62c
C18:2	61.27 ± 2.17b	61.70 ± 1.89b	55.95 ± 1.56f	60.26 ± 1.49c	60.77 ± 2.40c	60.81 ± 2.69c	64.13 ± 1.58a	59.57 ± 1.45d	58.96 ± 1.72e	64.86 ± 1.49a
C18:3	13.68 ± .98d	13.67 ± 1.03d	12.23 ± 1.16e	13.67 ± 1.21d	15.40 ± 1.53b	14.00 ± 1.47c	12.06 ± 1.32e	16.58 ± 1.30a	11.07 ± 1.13f	9.64 ± 0.87g
C20:0	0.07 ± 0.01c	−	0.06 ± 0.01cd	0.07 ± 0.01c	0.06 ± 0.01cd	0.08 ± 0.01b	0.07 ± 0.01c	0.09 ± 0.03a	0.09 ± 0.02a	0.09 ± 0.01a

*mean ± standard deviation; ** Values within each row followed by different letters are significantly different ($p < 0.05$); ***nonidentified

Table 2 Tocopherol contents of unroasted (raw) and roasted walnut (Type 1-10) kernel oils (mg/100 g).

Unroasted (Raw)	T1	T2	T3	T4	T5	T6	T7	T8	T9	T10
α-tocopherol	5.3 ± 0.46*b	5.8 ± 0.58b	4.9 ± 0.29c	4.7 ± 0.42c	3.9 ± 0.17	5.7 ± 0.61b	6.1 ± 0.27a	4.3 ± 0.13c	4.5 ± 0.24c	4.8 ± 0.45c
β- tocopherol	0.3 ± 0.01d**	0.2 ± 0.01	0.6 ± 0.02a	0.4 ± 0.01c	0.5 ± 0.03b	0.3 ± 0.01d	0.4 ± 0.01	0.2 ± 0.01e	0.5 ± 0.03b	0.3 ± 0.01d
γ- tocopherol	29.8 ± 0.78h	34.5 ± 1.13e	39.6 ± 1.17a	33.4 ± 0.86f	36.1 ± 0.63c	35.8 ± 1.37	32.3 ± 1.29g	28.9 ± 0.56	38.3 ± 0.73b	35.4 ± 0.81d
δ- tocopherol	11.4 ± 0.13a	9.8 ± 0.21c	6.3 ± 0.28f	8.6 ± 0.17d	7.8 ± 0.19e	9.5 ± 0.31c	7.4 ± 0.46e	8.1 ± 0.54d	10.6 ± 0.23b	7.5 ± 0.51e
Roasted	**T1**	**T2**	**T3**	**T4**	**T5**	**T6**	**T7**	**T8**	**T9**	**T10**
α-tocopherol	4.7 ± 0.21b	5.1 ± 0.43a	3.6 ± 0.31c	2.8 ± 0.38d	2.6 ± 0.27d	3.9 ± 0.13c	4.6 ± 0.19b	3.6 ± 0.61c	3.8 ± 0.54c	3.3 ± 0.25c
β- tocopherol	0.1 ± 0.00c	– ***	0.4 ± 0.01a	0.3 ± 0.01b	0.3 ± 0.01b	0.1 ± 0.00c	0.2 ± 0.01c	–	0.3 ± 0.01b	0.1 ± 0.00c
γ- tocopherol	29.1 ± 0.47h	33.4 ± 0.73e	38.2 ± 0.52a	32.7 ± 0.39	35.3 ± 0.41c	32.6 ± 0.36fg	31.8 ± 0.29g	28.1 ± 0.43i	36.4 ± 0.37b	34.8 ± 0.46d
δ- tocopherol	10.7 ± 0.11a	8.6 ± 0.23c	6.1 ± 0.32e	7.3 ± 0.26d	6.5 ± 0.17e	8.3 ± 0.29c	6.8 ± 0.32e	7.5 ± 0.45d	9.3 ± 0.87b	6.6 ± 0.65e

*mean ± standard deviation; ** Values within each row followed by different letters are significantly different ($p < 0.05$); ***nonidentified

pared to literature values. This present study has revealed that lipid profile of walnut kernel oil depends on factors such as walnut kernel type, roasting, locations and growing conditions of the walnut kernels.

Tocopherol contents of unroasted (raw) and roasted walnut (Type 1-10) kernel oils are shown in Table 2. While α-tocopherol contents of unroasted walnut oils changed from 3.9 mg/100g (Type 5) to 6.1 mg/100g (Type 7), α-tocopherol contents of roasted walnut oils ranged from 2.8 mg/100g (Type 4) to 4.7 mg/100g (Type 1). In addition, the highest β-tocopherol was found in unroasted walnut Type 3 oil (0.6 mg/100g). Also, γ-tocopherol content of unroasted walnut oils varies between 6.3 mg/100g (Type 3) and 11.4 mg/100g (Type 1). Additionally, roasting significantly changed γ-tocopherol present in walnut oil from 28.1 mg/100g (Type 8) and 38.2 mg/100g (Type 3), while δ-tocopherol of oil prepared from raw walnut kernel changed from 6.3 mg/100g (Type 3) to 11.4 mg/100g (Type 1). The highest δ-tocopherol was found in roasted walnut Type 1 (10.7 mg/100g). γ-Tocopherol dominates in tocopherol fraction for all tested oils followed by δ-tocopherol and α-tocopherol. The highest γ-tocopherol in raw and roasted walnut oils were found in oils extracted from Type 9 and Type 3, respectively. This has an impact on the increase in oxidative stability of the oils extracted from sample. β-Tocopherol was not found in the tested Type 2 and Type 8 walnut oils. Uzunova *et al.*[23] reported that walnut oils growing at the different years contained 4.4-5.7% α-tocopherol, 0.0-0.2% β-tocopherol, 85.1-88.2% γ-tocopherol and 6.1-9.7% δ-tocopherol. β-Tocopherol was not found in the tested walnut oils, which is confirmed by other authors for different varieties of nuts. However, lower levels of β-tocopherol were found in walnut kernel used in this present study. Generally, tocopherol contents of roasted walnut oils partly decreased compared to tocopherol results of unroasted walnut oils. This decrease may be due to effect of heat applied during processing. The content of (γ + β)-tocopherol contents of oils extracted by the soxhlet method changed from 29.6 mg/100 g (Jupiter) to 38.4 mg/100 g (G-139)[3]. Konsteiner *et al.*[24] reported that cashew, hazelnut, peanut, pecan, pistachio and walnut kernel oils contained 0.3, 0.1, 1.8, 0.2, 0.5, and 3.8 mg/100 g δ-tocopherol, respectively. However, α-tocopherol present in raw walnut oil changed from 28.9 to 38.3 mg/100 g and this is comparable with values reported for walnut oil[3, 24, 25, 26]. The results obtained are in partly agreement with the previously published data although some differences. Tocopherol contents of unroasted and roasted walnut oils varied depending on walnut types. These changes may have been influenced by some climatic factors, walnut types, harvest time and analytical conditions. Hence it can be inferred that roasting treatments may affect certain physicochemical properties of walnut kernels and their oils while compared with the unroasted

ones. These properties were however, also noted to vary with the type of walnut cultivars. Hence, more studies may be carried out to monitor the effects of different roasting temperatures and times and hence optimum roasting conditions may be established for the walnut kernels.

4 Conclusion

All the kernel analysed exhibited differences in their oil content and fatty acid compositions and tocopherol contents depending on walnut types and roasting. The results of the experiment presented here show that walnut kernel oil have a distinctive fatty acid and tocopherol profiles. Especially, linoleic acid had dominant fatty acid for both processing. Generally, fatty acid values of roasted walnut oils were found partly high compared with unroasted results. The reason for this, fatty acids of walnut oils probably increased due to removing of the relative humidity during roasting of walnuts. Generally, tocopherol contents of roasted walnut oils partly decreased compared to tocopherol results of unroasted walnut oils. This decreasing can be probably due to heating process. The oil could be useful for industrial applications owing to good physicochemical properties. Unsaturated fatty acids and tocopherols have favorable effect and positive health benefit than saturated fatty acids.

Acknowledgements

The authors extend their appreciation to the Deanship of Scientific Research at King Saud University for funding this work through research group no. (RG-1439-016).

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