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# Evaluation of Pomological Traits and Fatty Acid Composition of Some Persian Walnut (*Juglans regia* L.) Cultivars in Bursa, Turkey

Umran Erturk[*1], Sevil Yucel[2], Dilan Ahı Koşar[1], Mevlüt Batuhan Koşar[1], Hacer Akturk[1]

[1] *Department of Horticulture, Faculty of Agriculture, University of Uludag, Bursa, Turkey*

[2] *Yıldız Technical University, Chemical, and Metallurgical Engineering Faculty, Bioengineering Department, 34210, Davutpaşa/Istanbul, Turkey*

A R T I C L E I N F O

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A B S T R A C T

The pomological properties and nutritional values of walnuts vary according to the cultivars and ecological conditions. This study aims to determine some pomological properties and fatty acid compositions of some domestic ('Bilecik', 'Maraş 12', 'Maraş 18', 'Şebin', 'Şen1', 'Şen 2') and foreign walnut cultivars ('Chandler', 'Fernette', 'Fernor', 'Howard', 'Pedro', 'Serr') grown in Bursa (northwest in Turkey) conditions in 2016-2017. The nut weight of cultivars ranged from 9.19 g (Maraş 12) to 16.20 g (Şen 1). Average nut width varied from 28.84 mm (Maraş 12) to 38.50 mm (Şen 1), nut thickness was between 27.91 mm (Maraş 12) and 37.09 mm (Şen 1). Nut length was between 34.70 mm (Maraş 12) and 41.63 mm (Şen 1). Kernel percent and shell thickness varied from 41.77 (Pedro) to 58.06% (Şebin) and 1.10 mm (Şebin) to 1.92 mm (Fernor), respectively. The total oil content of the samples ranged from 60.95 (Bilecik) to 66.00% (Şebin). The linoleic acid content ranged from 53.76 (Maraş 18) to 65.37% (Bilecik), and the linolenic acid content was between 8.12 (Şebin) and 11.89% (Bilecik). The oleic acid content of the oils ranged from 12.45 (Bilecik) to 23.21% (Şebin) of the total fatty acids. Palmitic (6.71-11.18%) and stearic acid (2.55-3.97%) were a considerable amount of the remaining fatty acids. 'Şebin,' 'Maraş 18', 'Chandler,' 'Fernor,' and 'Howard' were found to be outstanding cultivars for fruit quality traits and fatty acid profiles in Bursa conditions.

## Introduction

Anatolia is the center of origin and one of the oldest cultivation areas of walnut. As the walnut is cultivated from the seed throughout the years, Anatolia has a rich genetic resource of walnuts. The most commonly grown walnut species for fruit production in Turkey is *J. regia* L. (Akca, 2005, 2012; Sen, 2011). Having 111775 hectare fields and 215000 tons production of walnuts, Turkey is among the first four walnut producer countries with a share of 5% in worldwide walnut production (Anonymus, 2018). Walnuts are generally grown as boundary recently been established with the introduced foreign cultivars or promising domestic genotypes selected from different regions of Turkey. Unfortunately, there is a lack of knowledge about the adaptation of those cultivars for different ecological areas of Turkey (Erturk *et al*., 2014). Therefore, the determination of the physical, chemical, and nutritional fruit properties of all the cultivars is extremely important for ongoing walnut breeding programs in the world (Akça et al., 2020; Sadat Hosseini Grouh *et al.*, 2011).

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Walnut is a nutritionally valuable crop that is of high demand around the world (Hassankhah et al., 2017). Fruits of walnut are particularly rich in terms of unsaturated fatty acids and proteins, and with the mineral elements, the content has a considerably important place for human nutrition and health (Jahanbani et al., 2016 and 2018). The walnut kernel (*Juglans regia* L.) generally contains approximately 60% oil (Lavedrine *et al*., 2000; Savage, 2001; Amaral *et al*., 2003). However, it varies from 52 to 70% (Greve *et al.*,1992; Yerlikaya *et al*., 2012). The amount of oil in the walnut kernel depends on the genotype, location, maturation and the irrigation rate (Greve *et al*., 1992; Garcia *et al.*, 1994; Yerlikaya *et al*., 2012). It is important to identify these differences in domestic cultivars and to identify which fatty acids give the best nutritional qualities (Savage *et al*., 1999). The primary fatty acids found in walnut oil are oleic (18:1), linoleic (18:2), and linolenic (18:3) acids. The ratio of fatty acids to each other are important for the economic and nutritional value of the nut. The fatty acid profile of walnut oil varies between cultivars (Greve *et al*., 1992).

This study aimed to evaluate the nut traits and fatty acid content of some domestic and foreign walnut cultivars grown in Bursa, Turkey.

## Materials and Methods

### *Walnut cultivars*

Six domestic ('Bilecik', 'Maraş 12', 'Maraş 18', 'Şebin', 'Şen1', 'Şen 2') and six foreign walnut cultivars (‘Chandler', 'Fernor', 'Fernette', 'Howard', 'Pedro' and 'Serr') which were grafted on *Juglans regia* L. seedlings in the conditions of Bursa, (northwest Turkey) (latitude 40°11' and longitude 29°3') were analyzed in 2016 and 2017. The cultivars were planted in 2008, 7×7 m spacing with four replicates of three trees per replicate.

The area is characterized by a mean maximum temperature of 43.8°C and means minimum -19.2°C and the average annual rainfall is 700 mm. The soil is clay with a content of 38% $CaCO_3$ and a pH of 7.1-7.5. Fertilization has been applied annually using NPK and organic manure.

### *Pomological traits*

After harvest, walnut fruits were dried and stored at 25 °C until the analysis. Fruit traits were analyzed in terms of the weight, length, thickness, and width of nuts, kernel weight, shell thickness, and kernel percentage on 90 nut samples. Also, kernel rottenness and kernel shrinkage (%) were evaluated.

Nut and kernel weight of cultivars were calculated according to the following formula. Kernel Percent (%) = Average kernel weight/ Average nut weight × 100. (TSI, 1991). Kernel percent classification was evaluated according to UPOV (2006); < 40% = Very Low, 40-44% = Low, 45-49% = Medium, 50-55% = High, > 55% = Very High.

### *Determination of total oil and fatty acid composition*

To determine the oil content, the walnuts were ground in a coffee mill for 30 seconds. After grinding, the walnuts were extracted for six h with hexane using a Soxhlet apparatus (Paquat and Houtfenne, 1987).

The fatty acid composition was determined by gas chromatography (GC; VARIAN chromatograph, model 1400; Varian Associates, Walnut Creek, CA), equipped with a flame ionization detector and a 3.0 m × 0.32 cm steel column, packed with LAC-3R-728 (20%; Cambridge Ind. Co., Cambridge, UK) on Chromosorb W/AW(80-100 mesh; Merck, Darmstadt, Germany). Nitrogen was used as a carrier gas (flow rate, 24 mL/min) (ISO 5508:1990, Analysis by gas chromatography of methyl esters of fatty acids) (AOCS, 1993).

### *Statistical analysis*

Analysis of variance (ANOVA) was used for statistical analysis that was performed by Minitab version 16.1. The Tukey test was used to compare means of pomological traits of cultivars. The analysis of pomological traits was performed in triplicate.

while fatty acid composition in duplicate and the data were expressed as averages ± standard errors.

## Results

According analysis of variance, statistically, significant differences were identified between cultivars in terms of nut weight, kernel weight, and kernel percent ($P<0.05$). The nut weight was between 9.19 (Maraş 12) and 16.20 g (Şen 1), kernel weight was between 4.68 g (Pedro) and 8.55 g (Maraş 18), kernel percentage was between 41.77 (Pedro) and 58.06% (Şebin). In terms of kernel percent; Şebin, Serr, Maraş 18, was very high; Maraş 12 was high; Howard, Şen1 and Şen 2 was medium, and the others were low or very low.

Nut width, thickness, and length significantly affected by cultivars. Nut width was between 28.84 mm (Maraş 12) and 38.50 mm (Şen 1), nut thickness ranged from 27.91 mm (Maraş 12) to 37.09 mm (Şen 1), and nut length was between 34.70 mm (Maraş 12) and 41.63 mm (Şen 1) (Table 1).

Shell thickness, kernel rottenness, and shrinkage were significantly affected by cultivars. The shell thickness of the cultivars varied from 1.10 mm (Şebin) to 1.92 mm (Fernor). Cultivars were also evaluated in terms of kernel rottenness and shrinkage, which are an essential factor affecting internal quality. There was no significant shrinkage in Howard (0.21%), Maraş 18 (1.21%), and Şebin (1.97%), whereas more shrinkage was observed in Pedro (36.79%) and Şen 2 (24.22%). Fernette cultivar did not exhibit any rottenness, whereas the in other cultivars were observed between 1.44% (Pedro) and 29.26% (Şen 1).

### *Total oil and fatty acid composition*

The total oil and fatty acid composition of cultivars were presented in Table 2. The total oil content of cultivars varied between 60.95% (Bilecik) and 66.00% (Şebin). The primary fatty acids in the cultivars were linoleic (C18:2), followed by oleic (C18:1), α-linolenic (C18:3), palmitic (C16:0) and stearic acid (C18:0). Linoleic acid had the most abundant fatty acid in all the analyzed cultivars comprising the values ranging from 53.76 (Maraş 18) to 65.37% (Bilecik).

Oleic acid, the second in order of importance content of the kernels in cultivars ranged from 12.45 (Bilecik) to 23.21% (Şebin) followed by linolenic acid, from 8.12 (Şebin) to 11.89% (Bilecik). Of the remaining fatty acids, only palmitic (6.71-11.18%) and stearic acid (2.55-3.97%) were present in considerable amounts.

The polyunsaturated fatty acid (PUFA) was the leading group of fatty acids in walnut oil extracted from cultivars, with values ranging from 62.33 (Maraş 18) to 77.26% (Şebin). The values for the MUFA group ranged from 12.51 (Bilecik) to 24.01% (Şebin). SFA was a minor constituent, with values ranging from 10.00 (Şen1) to 15.60% (Howard). Şebin had the highest total content, both MUFA, and PUFA. Besides, this cultivar had a high value of oleic acid. In contrast to Sebin, Bilecik had the lowest MUFA and the highest linoleic value (Table 2). The PUFA/MUFA ratio varied from 2.69 (Maraş 18) to 5.30 (Fernette).

## Discussion

The results of nut traits of the walnut cultivars are shown in Table 1. Desirable nut and kernel weight should vary from 12 to 18 g, and 6 to 10 g, respectively, or kernel percent should be at least 50% (McGranahan and Leslie, 1990). Maraş 18 was the cultivar that had these kernel traits. Akca *et al*. (2014) reported that nut weight of the evaluated 13 cultivars varied from 12.79 g (Fernor) to 15.35 g (Midland), kernel weight from 5.80 g (Fernor) to 7.22 g (Fernette) and kernel percentage from 42.80 (Pedro) to 47.33% (Fernette). In respect to kernel percent of Fernette and Pedro cultivars used in both studies, there was a similarity between the results of Akca *et al*. (2014).

### *Pomological traits*

**Table 1. Some nut traits of the walnut cultivars (2016-2017)**

| Cultivars | Nut weight (g) | Nut width (mm) | Nut thickness (mm) | Nut length (mm) | Kernel weight (g) | Kernel percent (%) | Shell thickness (mm) | Kernel rottenness (%) | Kernel shrinkage (%) |
|---|---|---|---|---|---|---|---|---|---|
| Bilecik | 11.41±0.21 c* | 33.02 ±0.09 ef | 30.04±0.10 h | 38.40±0.50 c | 4.89± 0.09 fg | 42.80± 1.55 de | 1.43±0.01 ef | 2.19±0.07 fgh | 4.20±0.55 fg |
| Maraş 12 | 9.19±0.19 d | 28.84±0.18 h | 27.91± 0.26 ı | 34.70±0.25 e | 4.81±0.32 fg | 52.34±2.88 abc | 1.18±0.07 gh | 24.02±1.59 b | 3.39±0.16 fg |
| Maraş 18 | 15.46±0.22 a | 33.89± 0.02 cd | 34.11±0.12 b | 40.69±0.05 ab | 8.55±0.28 a | 55.38±1.06 ab | 1.45±0.02def | 6.42±0.81 de | 1.21±0.43 fg |
| Şebin | 9.98±0.22 d | 30.19±0.09 g | 31.80±0.13 ef | 35.30±0.18 e | 5.72±0.32 def | 58.06±2.00 a | 1.10±0.02 h | 1.73±0.15 gh | 1.97±0.31 fg |
| Şen 1 | 16.20±0.28 a | 38.50±0.14 a | 37.09±0.15 a | 41.63±0.19 a | 6.99±0.18 bc | 43.21±1.88de | 1.61±0.03 cde | 29.26±2.11 a | 6.64±0.44 def |
| Şen 2 | 15.11±0.12 a | 33.53 ±0.02 de | 30.98±0.15 g | 41.00±0.13ab | 7.50±0.08 b | 49.66±0.94bcd | 1.62±0.04 cd | 11.33±0.39 c | 24.22±1.88 b |
| Chandler | 11.66±0.11 bc | 33.70 ±0.10 de | 32.87±0.12 cd | 40.01±0.20 b | 5.12±0.06 efg | 44.01± 0.92 de | 1.55±0.02 cde | 5.90±0.40 def | 10.79±0.15 cd |
| Fernor | 11.93±0.12 bc | 32.64±0.20 f | 31.84± 0.18 ef | 38.32±0.14 c | 4.80±0.01 fg | 40.18±0.26 e | 1.92±0.02 a | 2.93±0.23 e-h | 4.87±0.47 efg |
| Fernette | 12.85±0.44 b | 35.60±0.10 b | 33.11±0.12 c | 37.80±0.12 cd | 5.91±0.30 de | 46.10±3.32 cde | 1.73±0.03 bc | 0.00±0.00 h | 10.55±1.47 cde |
| Howard | 12.71±0.27 b | 34.44±0.15 c | 32.24± 0.08 de | 37.08±0.14 d | 5.98±0.25 de | 46.87±1.00 cde | 1.89±0.02 ab | 7.97±0.66 cd | 0.21±0.20 g |
| Pedro | 11.22±0.16 c | 32.77±0.15 f | 31.18±0.07 fg | 38.39±0.19 c | 4.68±0.06 g | 41.77±0.79 e | 1.65±0.04 c | 1.44±0.02 h | 36.79±2.86 a |
| Serr | 11.48±0.18 c | 30.77±0.12 g | 32.24±0.19 de | 37.71±0.16 cd | 6.38±0.21 cd | 55.57±0.96 ab | 1.31±0.02 fg | 5.51±0.21 d-g | 13.13±1.06 c |

*Different letters represent as statistically different groups P (0,05)

**Table 2. Total oil and fatty acids content (mg/g) of walnut cultivars**

| Cultivars | Total Oil | Linoleic Acid (C18:2) | Oleic Acid (C18:1) | Linolenic Acid (C18:3) | Palmitic Acid (C16:0) | Stearic Acid (C18:0) | PUFA | MUFA | SFA | PUFA/ MUFA |
|---|---|---|---|---|---|---|---|---|---|---|
| Bilecik | 60.95±2.34 | 65.37±1.12 | 12.45±0.32 | 11.89±1.18 | 7.36±0,23 | 2.55±0.15 | 63.83±3.04 | 12.51±0.35 | 10.23±0.24 | 5.10±0.12 |
| Maraş 12 | 63.00±6.92 | 60.05±1.98 | 20.94±1.13 | 8.79±0.73 | 6.71±0.25 | 3.51±0.20 | 68.84±1.91 | 20.94±0.72 | 10.22±0.61 | 3.28±0.08 |
| Maraş 18 | 65.24±3.04 | 53.76±1.76 | 23.17±0.97 | 8.57±0.97 | 10.13±0.40 | 3.36±0.13 | 62.33±2.79 | 23.17±0.30 | 14.50±0.74 | 2.69±0.12 |
| Şebin | 66.00±3.46 | 55.71±1.87 | 23.21±0.19 | 8.12±0.64 | 8.09±0.78 | 3.97±0.17 | 77.26±3.21 | 24.01±0.83 | 12.96±1.25 | 3.21±0.11 |
| Şen 1 | 61.74±2.47 | 59.96±1.78 | 21.03±0.65 | 8.95±0.57 | 6.88±0.37 | 3.02±0.32 | 68.91±2.55 | 21.19±0.42 | 10.00±1.18 | 3.25±0.07 |
| Şen 2 | 64.04±0.55 | 60.05±1.98 | 15.00±0.42 | 8.79±0.43 | 7.73±0.49 | 3.53±0.23 | 73.54±2.12 | 15.00±0.34 | 11.46±0.87 | 4.90±0.13 |
| Chandler | 64.21±0.71 | 60.05±1.52 | 17.54±0.20 | 11.33±1.24 | 7.57±0,25 | 3.26±0.08 | 71.38±2.76 | 17.62±0.74 | 11.00±0.22 | 4.05±0.08 |
| Fernor | 65.00±0.57 | 65.12±2.05 | 14.17±0.40 | 9.11±1.05 | 7.91±0,40 | 3.07±0.18 | 74.23±2.51 | 14.32±0.43 | 11.45±0.23 | 5.18±0.10 |
| Fernette | 63.01±2.40 | 61.99±1.85 | 13.45±0.21 | 9.90±0.85 | 10.39±0,34 | 3.50±0.15 | 71.89±2.63 | 13.54±0.33 | 14.57±0.20 | 5.30±0.14 |
| Howard | 64.35±2.10 | 55.16±2.42 | 18.86±1.05 | 10.30±0.97 | 11.18±0.35 | 3.37±0.12 | 65.46±3.01 | 18.94±0.15 | 15.60±0.59 | 3.45±0.06 |
| Pedro | 62.07±0.90 | 60.75±2.31 | 15.01±0.20 | 11.61±1.02 | 8.35±0.34 | 3.36±0.13 | 72.36±3.14 | 15.49±0.39 | 12.49±0.57 | 4.58±0.13 |
| Serr | 61.03±3.04 | 55.13±2.59 | 21.89±0.31 | 8.41±1.25 | 9.85±0.56 | 3.63±0.15 | 63.54±2.07 | 22.06±0.40 | 14.35±0.63 | 2.88±0.06 |

* Results are the mean value ± standard error of two independent experiment

In Australia, nut weights of 'Chandler,' 'Fernette,' and 'Fernor,' were 10.6, 11 and 11.4, respectively (Vanhanen, 2010). In California, USA, 'Chandler,' 'Midland,' and Pedro average kernel weights of 6.4, 6.5 and 7.3 g respectively and kernel ratios of 51, 48, and 51% were reported (Tulecke and McGranahan, 1994). The reason for the differences between Tulecke and McGranahan (1994) and Vanhanen (2010) can be environmental factors.

In the present study, the nut dimension of the cultivars was found to be larger by *Akça et al.* (2014) and *Akça et al.* (2018). Akca *et al.* (2014) in Turkey, evaluated 13 walnut cultivars grafted *J. regia* L. and found nut width about 35.24 mm (Fernor) - 38.47 mm (Fernette); nut length 40.62 (Fernor) - 43.50 mm (Pedro) and nut thickness 33.73 mm (Fernor) - 36.86 (Fernette). Akca *et al*. (2018) reported that nut width of cultivars ranged between 30.55 mm (Maraş12) and 39.97 (Şen1), nut length varied from 32.58 mm (Şen1) to 42.53 mm (Şen 1). The reason for the differences can be the effect of environmental conditions on the cultivar (Vanhanen 2010). In Iran, the average nut widths of 30.5-42.1, nut length of 32.2 - 42.1 mm, and nut thickness of 30.8 - 38.8 mm were reported (Arzani *et al*., 2008). Oguz *et al*. (2008), in Aydın Bozdoğan, nut width of 'Bilecik,' 'Şen 1', and 'Şebin' were found 33.90 mm, 40.79 mm and 31.71 mm respectively. In terms of kernel rottenness and shrinkage, 'Chandler' cultivar gave negative results, and Hendricks *et al*. (1998) also reported that 'Chandler' kernels vary from year to year in the amount of tip shrinkage observed.

The results of the total oil and fatty acids content of the cultivars are shown in Table 2. The total oil content of cultivars varied between 60.95% (Bilecik) and 66.00% (Şebin). Korač *et al*. (1988) reported that oil content ranged from 66 to 69% in walnut cultivars. The total oil contents were from 49.30 to 66.10% eighteen cultivars of walnuts by Muradoglu *et al*. (2010). Yarılgac *et al*. (2001) reported that 20 walnut genotypes from Turkey contained 54.89-68.20% oil, Yerlikaya *et al.* (2012) reported that 11 walnut genotypes oil percentages varied from 61.32 to 69.35%, Erturk *et al*. (2014) determined the composition of seven walnut genotypes as follows: total oil 62.17-68.93%.

The walnuts are rich in two unsaturated fatty acids and contain linoleic acid and linolenic acid (Zwarts, 1999). The ratios of fatty acids are important for walnut's economic and nutritional value (Savage *et al*., 1999). The obtained results in this study compared to the previous study results in other geographical origins such as New Zealand (Zwarts *et al*., 1999), Portugal (Amaral *et al*., 2003), Canada (Li *et al*., 2007), Turkey (Yerlikaya *et al*., 2012) and were compatible. Unver *et al*. (2016), identified an oil content of 50.83 and 60.77% for linoleic acid, between 16.14 and 26.44% for oleic acid, between 11.08 and 14.25% for linolenic acid between 5.42 and 7.29% for palmitic acid, and between 1.70 and 2.55% for stearic acid. It has been reported that oleic acid content ranged from 14.3 to 26.1% of the total fatty acids, while the linoleic acid content ranged from 49.3 to 62.3% and the linolenic contents from 8.0 to 13.8% by Zwarts *et al*. (1999). Li *et al*. (2007) reported that the proportion of palmitic, stearic, oleic, and linolenic in walnut was 99.00% of total oil, the proportion of unsaturated fatty acids was 91.00% of oil, the proportion of linoleic and linolenic was 60.00% of oil. Martinez *et al*. (2006) reported that the PUFA/MUFA ratio changed from 2.22 to 4.54 in Franquette, Chandler, and Criolla. In the present study, the highest total content MUFA, PUFA, and oleic acid were obtained from 'Şebin' cultivar. Bayazıt and Sümbül (2012) also stated that, 'Şebin' was very promising for fruit quality traits and fatty acid profiles in the eastern Mediterranean region of Turkey.

In the current study, some walnut cultivars contained higher amounts of linoleic acid, oleic acid, linolenic acid, and palmitic acid than some other walnut cultivars. Walnut oil composition is affected by cultivars, climatic effects, geographical location, and treatment. The form of walnut oil is also affected by the maturity at harvest, its position on the plant, and its handling after harvest (Crews *et al.* 2005).

## Conclusions

This study was carried out in Bursa's ecological conditions to select suitable cultivars in terms of quality, nutritional value, yield increase, and standardized production. Excellent kernel percentage was obtained in some cultivars as the appearance quality criteria. Kernel percentages of 'Şebin,' 'Serr,' and 'Maraş 18' were higher than the other cultivars. Also, a considerable amount of kernel shrinkage and rottenness was not seen on the nuts of Şebin. In point of interior quality, there were differences in the oil content of walnut cultivars. 'Şebin' contained the highest oil (66.0%), whereas 'Bilecik' (60.95%) had the lowest.

In conclusion, Şebin and Maraş 18, in terms of both fruit characteristics and oil content, seem to be superior among the domestic cultivars. Şebin has come into prominence among the domestic and foreign cultivars as it contains more fatty acids that are important for human health. Chandler, Fernor, and Howard are found to be outstanding cultivars, grown in Bursa ecology in terms of fruit properties and oil content, among foreign varieties. The data reported in this paper confirmed that walnuts are one of the richest and the most important nutritional source due to beneficial effects of its oil content for human health.

## Conflict of interests

The authors declare that they do not have any conflict of interest.

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