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Determination of fatty acid and tocopherol contents in Chandler × Kaplan-86 F1 walnut population

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Abstarct: Walnut (*Juglans regia* L.) is one of the most common nuts in the world. There are a wide walnut variety of genotypes that differ in forestry, physical and chemical properties. Most of the walnut genotypes, which are natural resources have been evaluated as promising and germplasm sources for breeding. Walnuts are high in many beneficial biochemical compounds, so they have a useful profile for many people in relation to disease risk. Therefore, the demand for production and consumption of walnut is increasing day by day. In the present study, fatty acid and tocopherol profiles of 156 F1 genotypes belonging to Chandler × Kaplan-86 walnut population and their parents were investigated. Fatty acids and tocopherol isomers (α, β, γ) were determined by GC/FID (gas chromatography/flame lonizing detector) and HPLC (high-pressure liquid chromatography) techniques, respectively. The fatty acid content of F1 walnut genotypes and their parents were found to be 0.00%–3.34% for myristic acid, 5.86%–10.05% for palmitic acid, 0.01%–4.46% for stearic acid, 0.00%–0.89% for palmitoleic acid, 10.00%–20.89% for oleic acid, 45.36%–66.20% for linoleic acid and 9.04%–20.55% for α-linolenic acid. The content of α-tocopherol, β-tocopherol and γ-tocopherol from the walnut population were determined to be 1.61–23.65 μg/g, 1.90–12.57 μg/g, 90.22–394.96 μg/g, respectively. The Chandler × Kaplan-86 F1 hybrid walnut population showed a significant variation in terms of the fatty acids and tocopherol contents. This research contains up-to-date scientific data on very wide walnut genetic resources and composition. Our data show that 156 F1 walnut genotypes have a potential source of monounsaturated fatty acids and tocopherols and the results are very important for future breeding studies on walnut.

Key words: *Juglans regia* L., breeding, genetic resources, fatty acids, tocopherol, HPLC

1. Introduction

The nuts are one of the oldest food sources in the world with rich nutritional content. *Juglans* species has an important place among nuts. *Juglans regia* L., known as the English, Persian or Anatolian walnut, is the most known species in the *Juglans* genus, which has more than 20 species due to its superior fruit quality. Walnut is long-lived, deciduous, monoecious, open-pollinated and generally dichogamous characteristics in nature (Şen, 2011; Sütyemez et al., 2019).

Epidemiological studies show that many phytochemical compounds present in nuts (walnut, hazelnut, pistachio and almond) are partly responsible for their beneficial health effects. The phytochemical compounds are also secondary metabolites and having an important physiological and metabolic importance in plants. These compounds play an important role in growth and reproduction, providing protection biotic and abiotic stress, against pathogens, predatorsand specially health (Gundesli et al., 2020; Kafkas et al., 2020). Nuts have high lipid contents but with favorable profiles for promoting cardiovascular health, since they are low in saturated fatty acids and high in mono and polyunsaturated fatty acids. Studies connecting nut consumption and some diseases have focused on fatty acid constituents; however there are other bioactive compounds that may confer additional protective effects. Walnuts with their antioxidant contents and other nutritional values have an important role in human health and nutrition. The walnut kernel represents between 40% and 60% of the total nut weight. The kernels include proteins, lipids, dietary fibers, phytochemicals, microelements and tocopherols (Suburu et al., 2013). Most of the lipid contents of walnut are unsaturated

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essential fatty acids which are beneficial to human health, and the major fatty acids are linoleic, α-linolenic, oleic, palmitic and stearic acid (Maguire et al., 2004; Akca et all., 2005; Kornsteiner et al., 2006; Kafkas et al., 2017). The major vitamin of walnut kernel is γ-tocopherol, which is a vitamin E homologue and used to protect the mucus and skin cell membranes against the harmful effects of free radicals (Iqbal et al., 2004; Şen and Karadeniz, 2015). In addition, there are a significant amount of some other important nutrients like potassium, phosphorus, magnesium, calcium, manganese, iron and selenium in walnut kernels (Lavedrine et al., 2000; Cosmulescu et al., 2010; Özcan and Sütyemez, 2019). In fact, consuming walnut decreases the risk of cardiovascular diseases by increasing high-density lipoprotein (HDL) cholesterol and decreasing low-density lipoprotein (LDL) cholesterol (Spaccarotella et al., 2008; Sánchez-González et al., 2017). It is a traditional remedy commonly used in the treatment of diseases such as cough, stomach ache and cancer (Perry and Merzger, 1980; Duke, 1989; Fukuda et al., 2003). In a study conducted on animals by Orhan et al. (2011), it was stated that walnut consumption reduced the risk of Alzheimer's disease and slowed its progression. Therefore, walnut is going to be consumed as food with rich nutrient content all over the world. In recent years, an increasing number of studies have been conducted on walnut in relation to biochemical, phytochemical and antioxidant characteristics, and their contribution to human nutrition and health. Its consumption trend increases year by year is due to the scientific reports on health benefits (Fukuda et al., 2003; Oliveira et al., 2008; Cosmulescu et al., 2010; Kafkas et al., 2020). The chemical content capacity in walnut is influenced by numerous factors. Especially, the walnut type and cultivar, the growth location, environmental conditions and genotype structure have a great effect on the formation of these substances (Amaral et al., 2003; Tapia et al., 2013; Akbari et al., 2015; Kafkas et al., 2017).

Nowadays, some walnut breeding programs have been carried out to improve these features throughout the world. But, there are a few reports previously published related to fat, fatty acid and tocopherol characteristics of breeding lines. For this reason, it is very important to determine the nutritional contents in addition to the issues such as yield, quality, resistance to diseases in the breeding studies carried out on walnut. As far as in our knowledge, there is no previous report on fatty acid and tocopherol transference from their parents and their distribution in F1 individuals.

The purpose of this study was to identify the profiles of 156 F1 hybrids belonging to Chandler × Kaplan-86 walnut population and their parents based on their fatty acid and tocopherol contents.

2. Materials and methods

2.1. Plant material

The walnut collection and breeding studies were carried out in the Nut Application and Research Center (SEKAMER), Kahramanmaraş Sütçü İmam University, Kahramanmaraş, Turkey. This area is located at 37°35'27"N latitude, 37°03'28"E longitude and 930 m above sea level. Kahramanmaras has a mild climate between the Mediterranean and continental with 727 mm annual precipitation and 16.9 °C average annual temperature. A set of 156 F1 genotypes derived from a cross between Chandler × Kaplan-86 and their parents were used as a material tree. The F1 genotypes were planted in SEKAMER in 2009 with 6 m × 6 m spacing. Chandler is very productive and the most cultivated walnut cultivar in the world. Kaplan-86 nuts are big in size with low lateral fruitfulness and suitable for fresh consumption. Nuts were harvested in the vegetation period of 2018. About 1 kg of walnut fruits were collected from each tree for further experimental use and stored at +4 °C until analysis.

2.2. Oil extraction

Oil was extracted using cold pressing by hand. Oil extraction was performed based on the modified method of Bligh and Dyer (1959). A total of 20 g fruits were extracted for oil within hexane solvent for 1 h using automatic Soxhlet equipment (Gerhardt Soxtherm) in triplicate for each genotype. The extracted oil was used for both fatty acid and tocopherol analysis. The residue was dried until a constant weight. Instead of Boron trifluoride (BF3) potassium hydroxide (KOH) in methanol were used for methylation processprior to the analysis (AOAC, 1990).

2.3. Fatty acids analysis

Fatty acids were analyzed using a Gas Chromatography (Clarus 500) with an autosampler (Perkin Elmer, Shelton, CT, USA) equipped with a flame ionization detector and a fused- silicacapillary SGE column (100 m × 0.32 mm, ID 0.25 µm, BP20 0.25 UM; Perkin Elmer, Austin, TX, USA). The oven temperature was held at 140 °C for 5 min, and then raised to 200 °C at a rate of 4 °C min^{-1} and then to 220 °C at a rate of 1 °C min^{-1}, while the injector and the detector temperatures were set to 220 °C and 280 °C, respectively. The sample volume was kept upto 1 µL, and the carrier gas was controlled at 16 psi. The split ratio was 1:100. The mixture of 37 fatty acid methyl esters (FAMEs) (Supelco, Bellefonte, PA, USA) were used for identification of fatty acids (Kafkas et al., 2017).

2.4. Tocopherols

For each replicate, approximately 1 g of homogenate from each of the genotypes was used. Tocopherols alpha (α), beta (β) and gamma (γ) were analyzed by HPLC technique according to the method developed by Surai et al. (1996) and Surai (2000). In the analysis, 3 µm C18 reversedphase column was used (15 cm × 4.6 mm, Spherisorb ODS2, Phase Separation, Clwyd, UK) and the methanol/water (97:3 v/v; 1.05 ml/min) was used as a mobile phase. Excitation at 325 nm and emission at 490 nm with retinol in the first 5 min followed by excitation at 295 nm and emission at 330 nm were processed with a fluorescence detector for identification of tocopherols (Surai et al., 1996; Surai, 2000).

2.5. Statistical analysis

Excel package program was used to determine the standard deviations and distributions of the data.

3. Results and discussion

It is well known that the chemical composition of fruits varies considerably from genotype to genotype and is affected by the cultivation and storage conditions (Pellegrini et al., 2006). It is important to note that variations in physicochemical parameters composition of genotypes are much wider than those observed for commercial varieties. In the present study, we characterized a total of 156 F1 walnut genotypes (Chandler × Kaplan-86) and their parents in terms of total lipid, saturated and unsaturated fatty acids and tocopherol contents. Stearic acid, myristic acid and palmitic acid which are saturated fatty acids of walnut genotypes were also determined. Due to the increasing trends of healthy nutrition, the determination of nutrient contents in fruits has been one of the most important objective in fruit breeding studies. Therefore, total lipid content and fatty acid profile are some of the most significant parameters for selection of the walnut cultivars. Fatty acid compositions of all extracts, including 156 F1 and their parents, are presented in Tables 1 and 2. The total lipid content of the Chandler was 64.97% and the Kaplan-86 was 44.73% observed. Total lipid contents of F1 walnut genotypes ranged from 27.28% to 84.48%. The distribution of F1 plants in terms of total lipid content is presented in Figure 1. The average total lipid content of this study is 57.45% and it is among the ranges of various studies conducted around the world (Mitrovic et al., 1995; Savage et al., 1999; Kafkas et al., 2017; Gao et al., 2018; Pycia et al., 2019). Our walnut genotypes were found to have a significant variation in terms of the total lipid content. In a study conducted by Beyhan et al. (2017), total lipid contents of 10 different walnut cultivars were evaluated and results revealed that the total lipidcontent of Chandler cultivar was 63% which is similar to our results. In another study, total lipidcontents of thewalnut kernelsranged from 46.4% to 82.1% depending on the genotype and geographic location (Dogan and Akgul, 2005; Li et al., 2007; Pereira et al., 2008; Gharibzahedi et al., 2014; Ünver et al., 2016; Beyhan et al., 2017; Kafkas et al., 2017). In our study, the total lipidcontents of F1 genotypes showed a wide variation compared to other studies. These differences could be resulted from the large population size used in this study.

The saturated fatty acid composition obtained by Chandler × Kaplan-86 F1 hybrid population and their parents were determined by GC/FID technique, and the results were presented in Table 1. The total saturated fatty acid content of the F1 hybrid population ranged between 7.40% and 17.35%. In a previous study conducted on 70 different walnut genotypes, the highest total saturated fatty acid content among the genotypes was 12.36% (Ghasemi et al., 2010). In the present study, palmitic acid, stearic acid, and myristic acid were determined as saturated fatty acids. Palmitic acid, stearic acid, and myristic acid concentrations of the F1 walnut population were ranged between 5.86% and 10.05%, 0.01% and 4.46%, 0.00% and 3.34%, respectively. The total saturated fatty acid content of Chandler and Kaplan-86 cultivars were 9.44% and 9.98%, respectively. Some of the examined F1 walnut genotypes showed heterosis in terms of these properties (Figure 2). In a previous study carried out on 19 different walnut genotypes, unsaturated fatty acid compositions were examined and its concentrations ranged from 2.57% to 3.37% for stearic acid, 0.00% to 0.05% for myristic acid, 6.42% to 7.92% for palmitic acid and 0.00% to 0.16% for arachidic acid (Beyhan et al., 2017).

Like all nuts, a large proportion of walnuts is composed of lipids. Walnut kernels are rich in unsaturated fatty acids. Unsaturated fatty acids are divided into two groups.Walnut kernels are important nutrients of both polyunsaturated and monounsaturated fatty acids which are beneficial to human health. Unsaturated fatty acids are known to be effective in brain development, reducing risk factors for diabetes type 2 and coronary heart disease (Tapsell, 2009). In our study, we investigated oleic acid and palmitoleic acid as monounsaturated fatty acids, linoleic and α-linolenic acids as polyunsaturated fatty acids (Table 2). The content of monounsaturated fatty acids in F1 walnut genotypes varied between 10.00% and 27.56%. While this monounsaturated fatty acid content in the parents was 16.52% for Chandler and 17.05% for Kaplan-86, respectively. Oleic acid values were found to be the lowest in genotype WCK108 with 10.00% and the highest in genotype WCK56 with 21.78%. Palmitoleic acid values to be ranged from 0.00% to 0.89% in F1 walnut genotypes. In terms of this fatty acid, 133 of the F1 genotypes ranged from 0.00% to 0.14%, 12 genotypes varied from 0.14 to 0.28%, and the rest (11 genotypes) ranged from 0.28% to 0.89% (Figure 3). In Chandler, the oleic acid content was found up to 16.66%, while the palmitoleic acid value was absent, and in the case of Kaplan-86, the oleic acid and palmitoleic acid values were 16.96% and 0.26%, respectively. Similarly, Martínez and Maestri (2008) analyzed the fatty acid composition with eight walnut

Table 1. Total lipid and saturated fatty acids content (%) of F1 walnut genotypes and their parents.

Genotypes	Total lipid (%)	Saturated fatty acids (%)		
		Myristic acid	Palmitic acid	Stearic acid
WCK1	52.94	0.55 ± 0.05	7.45 ± 0.33	0.15 ± 0.03
WCK2	65.16	3.34 ± 0.40	8.88 ± 0,19	3.45 ± 0.53
WCK3	58.04	0.08 ± 0.11	6.45 ± 0.02	1.81 ± 0.01
WCK4	57.09	0.06 ± 0.01	7.16 ± .0.16	2.11 ± 0.06
WCK5	59.30	1.49 ± 0.87	7.72 ± 0.33	3.33 ± 0.93
WCK6	53.15	0.08 ± 0.02	7.22 ± 0.09	2.16 ± 0.04
WCK9	49.38	0.05 ± 0.02	8.03 ± 0.17	1.77 ± 0.04
WCK10	50.46	0.24 ± 0.33	6.26 ± 0.43	2.24 ± 0.27
WCK11	60.51	0.86 ± 0.01	7.79 ± 0.35	2.03 ± 0.24
WCK13	42.23	0.00 ± 0.00	7.26 ± 0.05	2.42 ± 0.06
WCK19	54.44	0.17 ± 0.06	7.25 ± 0.37	2.00 ± 1.38
WCK21	52.57	0.22 ± 0.23	7.57 ± 0.38	1.78 ± 0.01
WCK22	56.32	2.57 ± 0.17	7.21 ± 0.59	2.05 ± 0.58
WCK23	56.20	0.69 ± 0.13	7.52 ± 0.26	2.41 ± 0.53
WCK24	56.25	0.04 ± 0.01	7.00 ± 0.19	2.22 ± 0.01
WCK25	62.32	0.29 ± 0.12	7.24 ± 0.16	2.12 ± 0.25
WCK26	70.64	0.08 ± 0.01	7.27 ± 0.25	2.37 ± 0.08
WCK30	53.00	1.07 ± 0.44	7.43 ± 0.23	2.67 ± 0.64
WCK34	27.28	0.61 ± 0.76	6.63 ± 0.06	2.12 ± 0.39
WCK36	51.00	0.12 ± 0.04	7.70 ± 0.25	1.73 ± 0.02
WCK37	56.67	0.41 ± 0.33	7.37 ± 0.12	3.22 ± 0.66
WCK39	66.31	0.07 ± 0.02	6.87 ± 0.03	2.09 ± 0.08
WCK41	48.15	0.04 ± 0.01	6.29 ± 0.01	2.27 ± 0.11
WCK44	61.56	0.47 ± 0.05	6.62 ± 0.02	2.31 ± 0.01
WCK46	54.57	0.28 ± 0.33	7.42 ± 0.14	2.05 ± 0.74
WCK47	51.37	0.18 ± 0.18	7.61 ± 0.03	2.22 ± 0.25
WCK48	58.04	0.34 ± 0.06	7.38 ± 0.06	2.66 ± 0.01
WCK50	70.44	0.02 ± 0.00	6.66 ± 0.08	1.86 ± 0.01
WCK51	50.07	0.24 ± 0.02	6.49 ± 0.11	2.33 ± 0.03
WCK52	62.59	0.03 ± 0.00	7.28 ± 0.03	2.02 ± 0.08
WCK55	59.23	0.05 ± 0.01	6.73 ± 0.16	2.16 ± 0.07
WCK56	68.58	0.09 ± 0.00	10.05 ± 0.45	3.08 ± 0.32
WCK57	69.87	0.05 ± 0.04	6.08 ± 0.15	2.16 ± 0.12
WCK58	53.49	0.03 ± 0.00	7.12 ± 0.04	2.19 ± 0.03
WCK59	61.55	0.06 ± 0.03	6.53 ± 0.01	2.03 ± 0.11
WCK60	56.97	0.28 ± 0.33	6.09 ± 0.07	2.18 ± 0.21
WCK62	35.27	0.08 ± 0.01	7.57 ± 0.36	1.88 ± 0.11
WCK63	60.17	0.05 ± 0.01	6.09 ± 0.16	1.91 ± 0.03
WCK64	56.54	0.05 ± 0.01	6.09 ± 0.08	1.85 ± 0.11
WCK66	60.84	0.25 ± 0.01	7.02 ± 0.26	2.54 ± 0.21

Table 1. (Continued).

WCK67	65.08	0.11 ± 0.09	7.61 ± 0.02	2.10 ± 0.03
WCK68	70.74	0.07 ± 0.02	7.07 ± 0.22	2.02 ± 0.00
WCK70	60.15	0.23 ± .0.02	6.08 ± 0.19	2.24 ± 0.15
WCK73	50.31	0.03 ± 0.00	7.36 ± 0.07	1.89 ± 0.04
WCK74	61.79	0.03 ± 0.01	7.38 ± 0.17	1.92 ± 0.08
WCK75	70.61	0.08 ± 0.01	7.84 ± 0.16	2.14 ± 0.01
WCK76	54.12	0.01 ± 0.11	6.99 ± 0.04	2.01 ± 0.04
WCK80	69.03	0.08 ± 0.06	6.96 ± 0.21	1.99 ± 0.07
WCK81	50.04	0.97 ± 0.62	6.93 ± 0.15	2.39 ± 0.03
WCK82	45.78	0.07 ± 0.04	7.01 ± 0.48	2.44 ± 0.06
WCK83	65.45	0.05 ± 0.01	7.55 ± 0.25	1.94 ± 0.21
WCK85	63.82	0.08 ± 0.01	7.46 ± 0.08	2.07 ± 0.18
WCK86	47.01	0.05 ± 0.00	7.84 ± 0.04	1.66 ± 0.01
WCK87	68.10	0.04 ± 0.01	6.52 ± 0.11	2.16 ± 0.08
WCK88	68.05	0.02 ± 0.03	7.04 ± 0.02	2.19 ± 0.15
WCK89	58.84	0.06 ± 0.01	7.82 ± 0.04	1.81 ± 0.88
WCK91	64.69	0.05 ± 0.01	6.32 ± 0.08	2.76 ± 0.06
WCK93	63.48	0.05 ± 0.00	7.15 ± 0.23	1.06 ± 0.08
WCK94	51.48	1.04 ± 0.18	7.61 ± 0.57	1.58 ± 0.23
WCK95	70.35	0.18 ± 0.01	7.52 ± 0.79	2.04 ± 0.08
WCK96	46.46	0.16 ± 0.01	7.57 ± 0.07	2.17 ± 0.11
WCK97	45.25	0.48 ± 0.06	7.17 ± 0.01	2.02 ± 0.01
WCK98	56.89	0.39 ± 0.05	7.61 ± 0.16	0.94 ± 0.94
WCK99	54.87	0.28 ± 0.02	7.06 ± 0.02	2.37 ± 0.24
WCK100	50.64	0.04 ± 0.00	7.58 ± 0.08	2.01 ± 0.16
WCK103	65.83	0.00 ± 0.00	7.52 ± 0.42	2.28 ± 0.18
WCK104	46.93	0.05 ± 0.01	6.05 ± 0.35	1.62 ± 0.08
WCK106	60.39	0.05 ± 0.01	7.04 ± 0.04	2.26 ± 0.02
WCK108	69.07	0.03 ± 0.01	7.87 ± 0.01	2.06 ± 0.13
WCK111	69.61	0.03 ± 0.02	6.88 ± 0.02	2.01 ± 0.11
WCK113	51.23	0.59 ± 0.16	7.16 ± 0.33	2.61 ± 0.01
WCK115	61.33	0.02 ± 0.03	6.96 ± 0.23	2.39 ± 0.07
WCK116	65.59	0.05 ± 0.06	7.14 ± 0.01	2.12 ± 0.01
WCK117	60.49	0.03 ± 0.04	7.06 ± 0.12	2.41 ± 0.18
WCK118	71.02	0.05 ± 0.01	6.97 ± 0.05	2.15 ± 0.01
WCK120	63.99	0.03 ± 0.04	7.01 ± 0.13	2.01 ± 0.04
WCK121	57.95	0.45 ± 0.10	8.41 ± 0.23	0.21 ± 0.01
WCK124	42.45	0.00 ± 0.00	7.19 ± 0.15	1.77 ± 0.81
WCK125	59.96	0.03 ± 0.04	7.61 ± 0.93	0.95 ± 0,10
WCK126	54.23	0.10 ± 0.07	6.88 ± 0.25	2.32 ± 0.05
WCK127	38.91	0.03 ± 0.04	5.86 ± 0.19	1.66 ± 0.07
WCK128	64.92	0.00 ± 0.00	7.06 ± 0.01	2.41 ± 0.00
WCK130	52.81	0.01 ± 0.01	7.39 ± 0.19	2.36 ± 0.01

Table 1. (Continued).

WCK131	69.58	0.02 ± 0.03	6.85 ± 0.25	2.08 ± 0.02
WCK132	44.56	0.14 ± 0.01	7.05 ± 0.83	2.33 ± 0.13
WCK134	43.96	0.02 ± 0.02	7.69 ± 0.07	1.92 ± 0.05
WCK135	43.78	0.18 ± 0.01	6.79 ± 0.18	2.07 ± 0.19
WCK138	71.21	0.00 ± 0.00	7.45 ± 0.04	2.43 ± 0.02
WCK139	43.61	0.00 ± 0.00	7.04 ± 0.31	2.48 ± 0.01
WCK142	53.27	0.07 ± 0.01	8.03 ± 0.02	1.78 ± 0.11
WCK144	51.52	1.00 ± 0.43	7.24 ± 0.18	1.48 ± 0.18
WCK145	59.67	0.04 ± 0.05	7.14 ± 0.21	1.74 ± 0.08
WCK146	50.14	0.03 ± 0.04	6.54 ± 0.18	2.43 ± 0.01
WCK148	84.48	0.03 ± 0.04	7.02 ± 0.01	1.98 ± 0.01
WCK149	70.98	0.25 ± 0.03	7.28 ± 0.07	0.39 ± 0.24
WCK150	67.21	0.67 ± 0.01	7.06 ± 0.05	0.03 ± 0.03
WCK151	69.66	0.00 ± 0.00	7.06 ± 0.16	2.29 ± 0.13
WCK154	45.97	0.03 ± 0.04	7.24 ± 0.02	1.89 ± 0.09
WCK156	54.92	0.02 ± 0.03	7.63 ± 0.47	2.14 ± 0.13
WCK157	53.65	0.00 ± 0.00	7.52 ± 0.39	2.07 ± 0.05
WCK158	39.72	0.02 ± 0.02	7.16 ± 0.11	0.99 ± 0.13
WCK159	65.14	0.03 ± 0.04	7.59 ± 0.01	2.03 ± 0.08
WCK160	70.32	0.54 ± 0.38	6.99 ± 0.17	2.34 ± 0.09
WCK161	49.13	0.08 ± 0.02	6.93 ± 0.14	0.01 ± 0.01
WCK164	54.97	0.05 ± 0.03	7.56 ± 0.06	1.93 ± 0.02
WCK165	66.00	0.18 ± 0.19	7.56 ± 0.26	2.03 ± 0.04
WCK166	56.00	0.02 ± 0.02	7.21 ± 0.06	2.61 ± 0.21
WCK167	62.86	0.02 ± 0.03	7.64 ± 0.86	2.03 ± 0.21
WCK168	72.98	0.03 ± 0.04	7.73 ± 0.16	2.17 ± 0.16
WCK173	37.69	0.03 ± 0.04	8.00 ± 0.01	1.77 ± 0.04
WCK175	67.47	0.03 ± 0.01	7.57 ± 0.07	1.83 ± 0.02
WCK176	53.35	0.66 ± 0.07	9.07 ± 0.45	2.65 ± 0.13
WCK178	44.11	0.06 ± 0.01	7.32 ± 0.14	2.04 ± 0.04
WCK179	66.12	0.08 ± 0.01	8.27 ± 0.09	2.04 ± 0.06
WCK180	62.00	0.07 ± 0.01	6.81 ± 0.23	2.01 ± 0.01
WCK181	38.41	0.05 ± 0.02	6.75 ± 0.11	2.03 ± 0.01
WCK182	44.95	0.39 ± 0.01	7.01 ± 0.18	2.58 ± 0.02
WCK183	58.27	0.33 ± 0.22	7.42 ± 0.02	2.47 ± 0.21
WCK184	79.74	0.05 ± 0.02	6.69 ± 0.06	2.02 ± 0.06
WCK186	46.41	0.07 ± 0.03	7.46 ± 0.03	2.16 ± 0.07
WCK187	57.12	0.02 ± 0.03	7.51 ± 0.05	2.34 ± 0.13
WCK188	58.62	0.04 ± 0.05	7.69 ± 0.33	1.89 ± 0.14
WCK189	61.00	0.04 ± 0.00	7.12 ± 0.08	1.76 ± 0.06
WCK190	51.00	0.26 ± 0.02	7.82 ± 0.27	0.44 ± 0.13
WCK191	65.74	0.00 ± 0.00	7.34 ± 0.02	1.91 ± 0.04
WCK192	63.74	0.41 ± 0.08	7.12 ± 0.14	1.68 ± 0.57
WCK193	84.21	0.05 ± 0.04	7.06 ± 0.25	2.09 ± 0.05

Table 1. (Continued).

WCK195	63.35	0.05 ± 0.01	7.77 ± 0.13	1.92 ± 0.07
WCK196	58.28	0.39 ± 0.33	6.92 ± 0.16	2.03 ± 0.06
WCK197	60.57	0.61 ± 0.13	6.81 ± 0.03	2.31 ± 0.04
WCK198	57.66	0.02 ± 0.00	6.74 ± 0.39	2.02 ± 0.07
WCK199	69.37	0.04 ± 0.00	7.11 ± 0.16	1.94 ± 0.06
WCK201	54.19	0.08 ± 0.06	7.45 ± 0.04	2.16 ± 0.22
WCK202	76.71	0.03 ± 0.01	7.78 ± 0.01	2.09 ± 0.00
WCK203	56.08	0.01 ± 0.01	6.58 ± 0.11	2.06 ± 0.01
WCK204	60.25	0.03 ± 0.04	7.58 ± 0.03	2.07 ± 0.01
WCK205	48.04	0.05 ± 0.00	7.35 ± 0.02	1.62 ± 0.02
WCK206	51.00	0.05 ± 0.00	7.78 ± 0.06	1.97 ± 0.08
WCK207	64.00	0.92 ± 0.22	7.61 ± 0.15	2.14 ± 0.04
WCK211	46.00	0.05 ± 0.04	7.25 ± 0.00	2.11 ± 0.21
WCK212	53.66	0.04 ± 0.01	6.28 ± 0.01	1.89 ± 0.03
WCK213	56.07	0.05 ± 0.00	6.93 ± 0.05	2.09 ± 0.08
WCK214	50.33	0.04 ± 0.01	7.03 ± 0.04	2.03 ± 0.04
WCK217	46.85	0.05 ± 0.00	7.16 ± 0.02	2.26 ± 0.03
WCK218	53.67	0.13 ± 0.00	7.07 ± 0.17	2.01 ± 0.04
WCK219	51.23	0.03 ± 0.33	7.25 ± 0.06	1.98 ± 0.01
WCK220	44.85	0.02 ± 0.00	7.03 ± 0.16	1.74 ± 0.01
WCK221	43.02	0.09 ± 0.00	6.71 ± 0.03	2.43 ± 0.06
WCK222	55.31	0.05 ± 0.00	6.65 ± 0.09	2.24 ± 0.06
WCK227	51.31	0.24 ± 0.22	7.21 ± 0.56	0.75 ± 0.07
WCK229	68.90	0.65 ± 0.52	6.73 ± 0.28	2.01 ± 0.23
WCK230	58.03	0.43 ± 0.46	7.97 ± 0.11	0.41 ± 0.14
WCK231	64.93	0.00 ± 0.00	7.45 ± 0.08	0.92 ± 1.29
WCK232	38.15	2.04 ± 0.00	7.56 ± 0.01	**4.46** ± 0.75
WCK235	52.75	0.02 ± 0.03	7.13 ± 0.12	2.04 ± 0.03
WCK237	76.62	0.04 ± 0.00	6.74 ± 0.07	1.86 ± 0.01
Chandler	64.97	0.04 ± 0.01	7.63 ± 0.28	1.77 ± 0.01
Kaplan-86	44.73	0.62 ± 0.58	7.44 ± 0.11	1.92 ± 0.01
Minumum value	27.28	0.00	5.86	0.01
Maximum value	84.48	3.34	10.05	4.46
Avarage value	57.45	0.22	7.22	2.01

*Each value is expressed as mean ± standard deviation.

cultivars including Chandler and found 16.10% oleic acid in Chandler. We identified palmitic acid as the main saturated fatty acid compound, followed by trace amounts of myristic and arachidic acid in stearic acid and walnut, and this was supported by different studies conducted in previous years (Doğan and Akgül, 2005; Popa et al., 2011; Rabrenovic et al., 2011; Beyazıt and Sumbul, 2012; Beyhan et al., 2017; Kafkas et al., 2017).

The polyunsaturated fatty acid content in all F1 genotypes and their parents ranged from 65.91% to 80.88% (Table 2). In many studies conducted on walnut kernel lipid compositions, linoleic acid has been reported as the most abundant unsaturated fatty acid (Bada et al., 2010; Rabrenovic et al., 2011; Beyhan et al., 2017). We also obtained similar results and the linoleic acid content for F1 genotypes ranged from 45.36% to 66.20%. Out of 156

Table 2. Unsaturated fatty acids content (%) of Chandler × Kaplan-86 F1 walnut genotypes and their parents.

Genotypes	Unsaturated fatty acids (%)			
	Polyunsaturated fatty acids		Monounsaturated fatty acids	
	Linoleic acid	α-linolenic acid	Oleic acid	Palmitoleic acid
WCK1	60.39 ± 0.09	13.05 ± 0.35	18.13 ± 0.06	0.00 ± 0.00
WCK2	53.69 ± 1.26	16.61 ± 0.50	13.08 ± 0.47	0.24 ± 0.34
WCK3	65.82 ± 0.03	13.64 ± 0.02	12.23 ± 0.09	0.00 ± 0.00
WCK4	60.53 ± 0.16	14.12 ± 0.26	16.03 ± 0.36	0.02 ± 0.02
WCK5	58.38 ± 0.03	12.39 ± 0.87	16.61 ± 1.17	0.09 ± 0.12
WCK6	59.49 ± 0.83	15.39 ± 0.43	15.69 ± 0.47	0.00 ± 0.00
WCK9	59.76 ± 0.14	12.97 ± 0.14	17.43 ± 0.23	0.00 ± 0.00
WCK10	62.94 ± 0.47	12.78 ± 0.54	15.05 ± 0.11	0.05 ± 0.07
WCK11	59.74 ± 0.46	11.65 ± 0.35	17.37 ± 0.35	0.00 ± 0.00
WCK13	62.26 ± 0.01	12.68 ± 0.05	15.41 ± 0.02	0.00 ± 0.00
WCK19	63.07 ± 2.28	11.53 ± 0.65	16.47 ± 0.71	0.33 ± 0.02
WCK21	62.31 ± 1.87	9.04 ± 0.43	18.97 ± 0.54	0.13 ± 0.18
WCK22	57.38 ± 2.13	12.07 ± 1.23	17.65 ± 0.72	0.00 ± 0.00
WCK23	62.19 ± 1.17	11.68 ± 0.54	14.98 ± 0.42	0.55 ± 0.06
WCK24	61.15 ± 0.74	12.06 ± 0.54	17.53 ± 1.04	0.03 ± 0.04
WCK25	60.95 ± 0.24	13.88 ± 0.06	15.41 ± 0.21	0.13 ± 0.13
WCK26	59.87 ± 1.08	15.04 ± 0.49	15.37 ± 1.89	0.02 ± 0.03
WCK30	61.03 ± 1.52	12.91 ± 0.64	14.56 ± 0.33	0.09 ± 0.03
WCK34	62.26 ± 1.05	14.03 ± 0.45	14.06 ± 0.33	0.05 ± 0.04
WCK36	59.94 ± 0.13	14.24 ± 0.19	16.24 ± 0.18	0.05 ± 0.06
WCK37	58.09 ± 3.74	12.51 ± 0.95	18.04 ± 3.51	0.38 ± 0.33
WCK39	60.37 ± 0.19	14.77 ± 0.69	15.83 ± 0.73	0.02 ± 0.03
WCK41	64.36 ± 0.57	13.07 ± 0.01	13.71 ± 0.25	0.00 ± 0.00
WCK44	62.49 ± 0.56	13.91 ± 0.23	14.16 ± 0.65	0.05 ± 0.01
WCK46	61.43 ± 0.84	11.52 ± 0.17	16.81 ± 0.23	0.06 ± 0.04
WCK47	61.18 ± 0.43	14.21 ± 0.54	14.12 ± 0.03	0.00 ± 0.00
WCK48	63.21 ± 0.03	12.83 ± 0.25	13.04 ± 0.04	0.19 ± 0.04
WCK50	64.52 ± 0.08	11.39 ± 0.37	15.57 ± 0.37	0.00 ± 0.00
WCK51	62.75 ± 1.94	14.55 ± 0.42	13.59 ± 1.58	0.07 ± 0.08
WCK52	63.12 ± 0.64	12.27 ± 0.24	15.06 ± 0.44	0.05 ± 0.06
WCK55	60.44 ± 0.11	15.05 ± 0.44	15.59 ± 0.25	0.00 ± 0.00
WCK56	45.36 ± 2.52	20.55 ± 0.98	20.89 ± 0.94	0.00 ± 0.00
WCK57	63.32 ± 1.21	12.95 ± 0.16	14.67 ± 0.75	0.06 ± 0.01
WCK58	61.59 ± 0.42	13.27 ± 0.19	15.81 ± 0.62	0.00 ± 0.00
WCK59	62.32 ± 0.37	13.97 ± 0.32	14.76 ± 0.48	0.08 ± 0.05
WCK60	63.37 ± 0.66	13.39 ± 0.23	13.82 ± 0.17	0.07 ± 0.02
WCK62	61.62 ± 0.65	12.33 ± 0.02	16.52 ± 0.39	0.02 ± 0.02
WCK63	61.06 ± 0.21	11.07 ± 0.22	17.82 ± 0.28	0.00 ± 0.00
WCK64	60.39 ± 0.64	12.94 ± 0.62	17.89 ± 1.23	0.00 ± 0.00

Table 2. (Continued).

WCK66	61.06 ± 0.06	12.76 ± 0.45	15.63 ± 0.59	0.23 ± 0.04
WCK67	58.91 ± 0.04	15.63 ± 0.35	15.62 ± 0.43	0.03 ± 0.04
WCK68	62.16 ± 0.58	13.04 ± 0.78	15.11 ± 1.11	0.00 ± 0.00
WCK70	60.83 ± 0.04	15.97 ± 0.15	13.82 ± 0.16	0.14 ± 0.04
WCK73	60.56 ± 0.29	12.66 ± 0.95	17.52 ± 1.59	0.00 ± 0.00
WCK74	62.71 ± 0.01	12.98 ± 0.02	14.93 ± 0.03	0.06 ± 0.02
WCK75	60.87 ± 0.57	11.82 ± 0.33	17.26 ± 0.77	0.02 ± 0.02
WCK76	61.28 ± 0.37	14.46 ± 0.01	15.01 ± 0.17	0.07 ± 0.09
WCK80	61.93 ± 0.07	12.75 ± 0.03	16.22 ± 0.23	0.08 ± 0.06
WCK81	60.61 ± 1.29	13.76 ± 0.71	14.47 ± 0.57	**0.89** ± 0.62
WCK82	62.07 ± 0.03	13.32 ± 0.04	15.01 ± 0.68	0.00 ± 0.00
WCK83	60.39 ± 0.93	12.25 ± 1.44	17.56 ± 0.06	0.28 ± 0.39
WCK85	60.63 ± 0.18	12.51 ± 0.37	17.21 ± 0.35	0.05 ± 0.06
WCK86	61.38 ± 0.09	11.68 ± 0.59	17.38 ± 0.22	0.02 ± 0.03
WCK87	63.14 ± 0.02	12.71 ± 0.11	15.43 ± 0.52	0.00 ± 0.00
WCK88	62.03 ± 0.03	13.27 ± 0.72	15.31 ± 0.66	0.15 ± 0.21
WCK89	58.41 ± 1.85	11.53 ± 0.43	20.01 ± 2.72	0.29 ± 0.41
WCK91	62.61 ± 0.57	14.32 ± 0.05	13.81 ± 0.67	0.14 ± 0.12
WCK93	61.67 ± 0.62	15.23 ± 0.26	14.16 ± 0.05	0.17 ± 0.02
WCK94	58.02 ± 2.45	13.44 ± 0.74	17.95 ± 0.31	0.02 ± 0.02
WCK95	59.05 ± 1.07	12.77 ± 0.44	17.85 ± 1.58	0.24 ± 0.08
WCK96	59.86 ± 0.05	12.89 ± 0.67	17.36 ± 0.45	0.00 ± 0.00
WCK97	61.04 ± 0.33	12.45 ± 0.04	18.17 ± 0.76	0.13 ± 0.18
WCK98	58.83 ± 0.04	14.48 ± 0.36	17.66 ± 0.28	0.12 ± 0.16
WCK99	63.09 ± 0.04	12.85 ± 0.28	14.09 ± 0.66	0.26 ± 0.18
WCK100	60.19 ± 1.05	12.97 ± 0.29	17.23 ± 1.41	0.00 ± 0.00
WCK103	59.82 ± 0.48	13.82 ± 0.29	16.57 ± 0.17	0.00 ± 0.00
WCK104	62.07 ± 0.03	13.84 ± 0.43	15.31 ± 0.01	0.00 ± 0.00
WCK106	58.35 ± 0.01	14.34 ± 0.00	17.98 ± 0.10	0.00 ± 0.00
WCK108	60.74 ± 0.61	13.53 ± 0.12	10.00 ± 0.10	0.00 ± 0.00
WCK111	63.83 ± 0.24	13.23 ± 0.16	13.95 ± 0.29	0.00 ± 0.00
WCK113	62.55 ± 0.34	12.48 ± 0.62	14.63 ± 0.23	0.00 ± 0.00
WCK115	61.44 ± 0.22	14.06 ± 0.18	14.58 ± 0.24	0.02 ± 0.03
WCK116	62.65 ± 0.68	14.59 ± 0.08	13.48 ± 0.53	0.00 ± 0.00
WCK117	60.27 ± 0.99	13.86 ± 0.77	16.39 ± 1.08	0.00 ± 0.00
WCK118	61.03 ± 0.21	14.76 ± 0.11	14.08 ± 0.07	0.00 ± 0.00
WCK120	61.62 ± 0.54	12.77 ± 0.05	16.58 ± 0.44	0.00 ± 0.00
WCK121	60.24 ± 0.16	13.25 ± 0.35	17.45 ± 0.04	0.00 ± 0.00
WCK124	60.36 ± 0.32	11.66 ± 0.03	19.03 ± 0.66	0.00 ± 0.00
WCK125	58.02 ± 1.92	13.83 ± 1.41	19.58 ± 3.43	0.00 ± 0.00
WCK126	62.45 ± 0.40	13.87 ± 0.23	14.40 ± 0.04	0.00 ± 0.00
WCK127	**66.20** ± 0.03	14.68 ± 0.11	11.58 ± 0.18	0.00 ± 0.00
WCK128	60.55 ± 0.12	14.66 ± 0.42	14.08 ± 0.55	0.00 ± 0.00

Table 2. (Continued).

WCK130	61.11 ± 0.26	11.91 ± 0.23	17.24 ± 0.03	0.00 ± 0.00
WCK131	61.65 ± 0.04	14.75 ± 0.06	14.67 ± 0.32	0.00 ± 0.00
WCK132	61.01 ± 0.28	14.43 ± 0.83	15.06 ± 0.33	0.00 ± 0.00
WCK134	62.00 ± 0.41	12.52 ± 0.23	15.87 ± 0.18	0.00 ± 0.00
WCK135	60.78 ± 1.01	13.14 ± 0.86	17.05 ± 0.76	0.00 ± 0.00
WCK138	62.31 ± 0.23	12.36 ± 0.15	15.47 ± 0.45	0.00 ± 0.00
WCK139	60.67 ± 0.06	14.18 ± 0.21	15.28 ± 0.04	0.00 ± 0.00
WCK142	59.78 ± 0.54	13.55 ± 0.33	16.08 ± 0.48	0.00 ± 0.00
WCK144	58.55 ± 0.66	12.49 ± 0.27	19.25 ± 0.51	0.00 ± 0.00
WCK145	63.05 ± 0.07	13.68 ± 0.39	13.91 ± 0.13	0.00 ± 0.00
WCK146	60.07 ± 0.11	14.28 ± 0.06	16.04 ± 0.04	0.00 ± 0.00
WCK148	61.48 ± 0.19	14.35 ± 0.00	15.15 ± 0.23	0.00 ± 0.00
WCK149	61.02 ± 0.29	13.32 ± 0.14	17.57 ± 0.23	0.00 ± 0.00
WCK150	60.04 ± 0.13	13.97 ± 0.28	17.08 ± 0.04	0.00 ± 0.00
WCK151	61.16 ± 0.23	13.25 ± 0.68	16.26 ± 0.75	0.00 ± 0.00
WCK154	62.02 ± 0.76	14.05 ± 0.69	14.08 ± 0.32	0.00 ± 0.00
WCK156	59.97 ± 1.02	13.05 ± 0.11	16.75 ± 0.54	0.00 ± 0.00
WCK157	60.61 ± 0.37	14.08 ± 0.72	15.73 ± 0.65	0.00 ± 0.00
WCK158	60.06 ± 0.85	10.53 ± 1.39	20.53 ± 3.53	0.00 ± 0.00
WCK159	60.00 ± 0.36	12.12 ± 0.13	18.26 ± 0.37	0.00 ± 0.00
WCK160	61.65 ± 0.05	14.67 ± 0.15	13.83 ± 0.54	0.00 ± 0.00
WCK161	61.61 ± 0.25	14.01 ± 0.13	17.17 ± 0.18	0.03 ± 0.04
WCK164	61.93 ± 0.16	12.63 ± 0.06	15.89 ± 0.01	0.03 ± 0.04
WCK165	59.72 ± 0.11	12.29 ± 0.37	18.14 ± 0.43	0.01 ± 0.06
WCK166	60.77 ± 0.01	14.36 ± 0.21	14.71 ± 0.63	0.16 ± 0.04
WCK167	62.03 ± 0.37	13.27 ± 0.81	14.76 ± 0.16	0.00 ± 0.00
WCK168	59.30 ± 0.08	15.09 ± 0.22	14.89 ± 0.14	0.00 ± 0.00
WCK173	61.94 ± 0.33	13.02 ± 0.35	15.26 ± 0.11	0.00 ± 0.00
WCK175	64.90 ± 0.01	13.72 ± 0.03	15.36 ± 0.01	0.00 ± 0.00
WCK176	57.77 ± 1.82	13.73 ± 0.08	15.23 ± 0.33	0.28 ± 0.39
WCK178	61.45 ± 0.17	13.99 ± 0.18	15.15 ± 0.09	0.00 ± 0.00
WCK179	61.21 ± 0.23	14.45 ± 0.13	13.97 ± 0.06	0.00 ± 0.00
WCK180	61.58 ± 0.61	14.34 ± 0.57	15.21 ± 0.18	0.00 ± 0.00
WCK181	62.32 ± 1.65	14.49 ± 0.09	14.38 ± 1.73	0.00 ± 0.00
WCK182	60.57 ± 0.07	13.55 ± 0.18	15.19 ± 0.04	0.73 ± 0.02
WCK183	59.01 ± 0.25	13.71 ± 0.04	16.61 ± 0.94	0.38 ± 0.32
WCK184	63.11 ± 0.05	12.00 ± 0.52	16.14 ± 1.16	0.00 ± 0.00
WCK186	61.24 ± 0.62	12.89 ± 0.28	16.02 ± 0.76	0.00 ± 0.00
WCK187	60.12 ± 0.06	15.21 ± 0.57	14.79 ± 0.22	0.04 ± 0.05
WCK188	62.36 ± 0.13	13.76 ± 0.06	14.27 ± 0.49	0.00 ± 0.00
WCK189	61.67 ± 0.78	12.34 ± 0.08	17.08 ± 0.72	0.00 ± 0.00
WCK190	58.26 ± 0.21	13.14 ± 0.03	19.34 ± 0.02	0.77 ± 0.26
WCK191	62.61 ± 0.61	13.99 ± 0.03	14.71 ± 0.16	0.08 ± 0.01

Table 2. (Continued).

WCK192	62.47 ± 1.07	12.26 ± 0.05	15.95 ± 0.55	0.13 ± 0.08
WCK193	60.04 ± 0.45	14.03 ± 1.27	16.02 ± 0.96	0.00 ± 0.00
WCK195	59.78 ± 0.02	14.86 ± 0.07	15.63 ± 0.06	0.00 ± 0.00
WCK196	60.63 ± 0.15	14.97 ± 0.34	14.08 ± 0.04	0.00 ± 0.00
WCK197	59.63 ± 0.27	15.22 ± 0.16	15.13 ± 0.35	0.03 ± 0.01
WCK198	60.32 ± 0.09	15.34 ± 0.15	15.54 ± 0.01	0.00 ± 0.00
WCK199	61.74 ± 0.25	13.87 ± 0.33	15.32 ± 0.01	0.00 ± 0.00
WCK201	62.16 ± 0.45	12.52 ± 0.16	15.06 ± 0.33	0.05 ± 0.04
WCK202	60.71 ± 0.45	13.96 ± 0.13	15.44 ± 0.33	0.00 ± 0.00
WCK203	65.03 ± 0.32	12.49 ± 0.03	13.65 ± 0.16	0.01 ± 0.01
WCK204	61.05 ± 0.18	13.47 ± 0.15	15.82 ± 0.08	0.00 ± 0.00
WCK205	63.53 ± 0.01	12.90 ± 0.00	14.56 ± 0.03	0.00 ± 0.00
WCK206	61.37 ± 0.74	12.65 ± 0.62	16.19 ± 0.01	0.00 ± 0.00
WCK207	57.99 ± 1.25	15.33 ± 0.16	15.94 ± 0.07	0.01 ± 0.00
WCK211	61.15 ± 0.28	13.94 ± 0.36	15.51 ± 0.18	0.00 ± 0.00
WCK212	64.74 ± 0.05	13.36 ± 0.47	13.68 ± 0.00	0.02 ± 0.03
WCK213	61.74 ± 0.01	12.65 ± 0.56	16.51 ± 0.34	0.00 ± 0.00
WCK214	61.17 ± 0.51	11.27 ± 0.18	18.12 ± 0.57	0.08 ± 0.11
WCK217	63.24 ± 0.29	12.69 ± 0.37	14.58 ± 0.02	0.04 ± 0.06
WCK218	60.35 ± 0.12	14.49 ± 0.21	15.87 ± 0.12	0.00 ± 0.00
WCK219	62.35 ± 0.94	13.99 ± 0.64	14.07 ± 1.07	0.08 ± 0.11
WCK220	61.07 ± 0.23	13.39 ± 0.29	16.77 ± 0.01	0.00 ± 0.00
WCK221	60.08 ± 0.34	14.82 ± 0.21	15.66 ± 0.45	0.21 ± 0.03
WCK222	61.82 ± 0.03	15.19 ± 0.62	14.05 ± 0.28	0.02 ± 0.02
WCK227	60.42 ± 3.11	14.63 ± 0.01	16.05 ± 2.64	0.27 ± 0.38
WCK229	62.24 ± 0.38	15.55 ± 1.05	12.28 ± 1.12	0.47 ± 0.03
WCK230	57.51 ± 0.03	16.24 ± 0.11	16.79 ± 0.93	0.67 ± 0.23
WCK231	61.04 ± 0.01	12.71 ± 0.06	17.09 ± 1.32	0.00 ± 0.00
WCK232	55.02 ± 0.04	11.27 ± 0.16	18.91 ± 0.49	0.76 ± 0.29
WCK235	64.13 ± 0.71	12.21 ± 0.49	14.47 ± 0.03	0.03 ± 0.04
WCK237	64.26 ± 0.02	12.47 ± 0.03	14.63 ± 0.04	0.00 ± 0.00
Chandler	62.67 ± 0.02	11.39 ± 0.31	16.52 ± 0.02	0.00 ± 0.00
Kaplan-86	59.51 ± 0.33	13.47 ± 0.14	16.87 ± 0.13	0.18 ± 0.12
Minumum value	45.36	9.04	10.00	0.00
Maximum value	66.20	20.55	20.89	0.89
Avarage value	61.04	13.46	15.8	0.07

*Each value is expressed as mean ± standard deviation.

genotypes, the 115 F1 walnut genotypes were found in the range of 59.24% to 62.71% for this polyunsaturated fatty acid. The content of this monounsaturated fatty acid was found 62.81% and 59.74% for Chandler and Kaplan-86, respectively. In this study, linoleic acid was dominant one of the polyunsaturated fatty acids, followed by linolenic acid. The α-linolenic acid values were found to be the lowest in genotype WCK21 with 9.04% and the highest in genotype WCK56 with 20.55%. However, this α-linolenic acid values for parents and other genotypes were found within this range (Figure 4). In various studies, it was reported that saturated fatty acids were less than from both

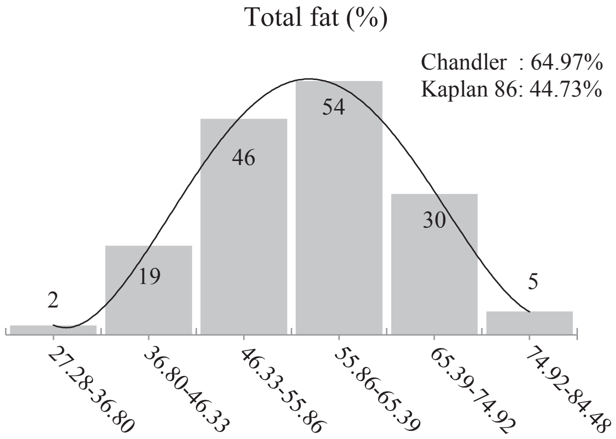

Figure 1. Total fat content distribution of F1 walnut genotypes.

monounsaturated and polyunsaturated fatty acids (Bada et al., 2010; Muradoglu et al., 2010; Ozcan et al., 2020). In a study of Gülsoy et al. (2019) conducted on 20 different walnut genotypes, reported that the major fatty acids were linoleic acid (58.15%–64.07%), oleic acid (12.93%–17.49%), linolenic acid (9.37%–13.61%), palmitic acid (5.60%–8.62%) and stearic acid (4.68%–6.69%). Our results are consistent with previously published the other studies (Dogan and Akgul, 2005; Pereira et al., 2008; Kafkas et al., 2017;). We determined oleic acid was the major fatty acid compound followed by a trace amount of palmitoleic acid, which agrees previous findings from previous studies (Popa et al., 2011; Kafkas et al., 2017).

Tocopherols are an important group of natural antioxidant compounds in food preservation technology. They are found in all higher plant which forms as four main types, α-tocopherol, β-tocopherol, γ-tocopherol and δ- tocopherol, differing in their molecular structures and antioxidant activities (Tangolar et al., 2011). Besides these compounds, tocopherols are found in walnut oil and are important in providing some protection against oxidation (Martinez et al., 2010). In the present study, we determined α-tocopherol, β-tocopherol and γ-tocopherol contents of F1 walnut genotypes and their parents by using high-pressure liquid chromatography (HPLC) techniques (Table 3). As seen in Table 3, the α-tocopherol in the analyzed genotypes were ranged from 1.61 µg/g (WCK30)

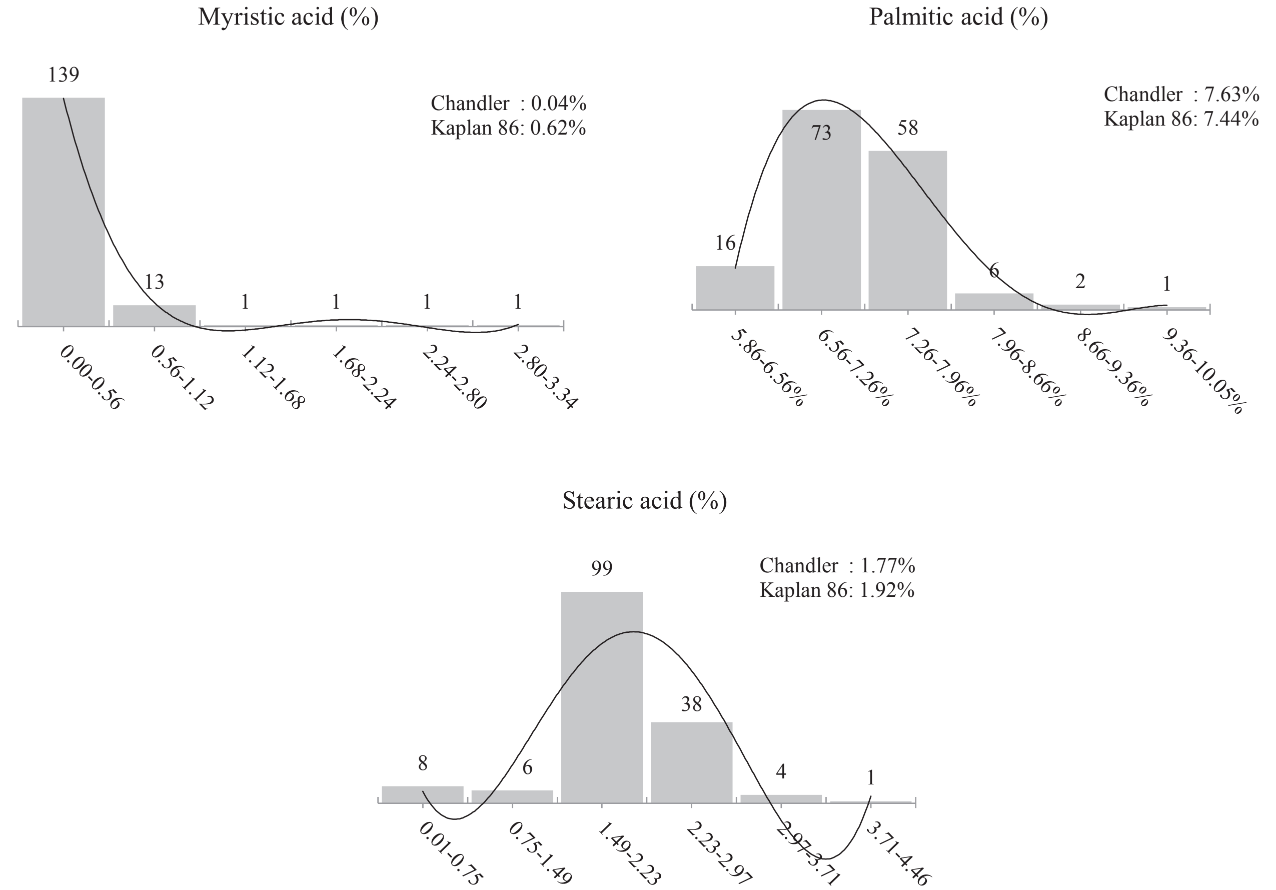

Figure 2. Saturated fatty acid content distribution of F1 walnut genotypes.

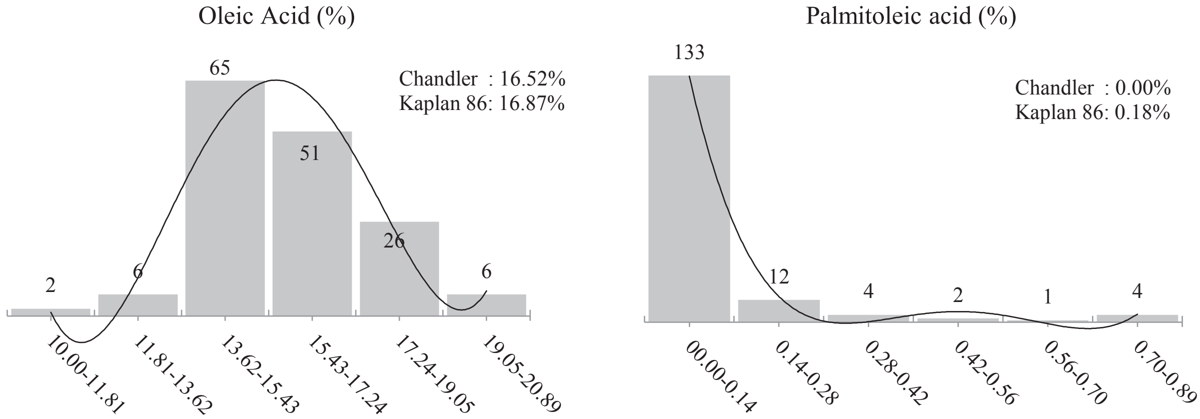

Figure 3. Monounsaturated fatty acid content distribution of F1 walnut genotypes.

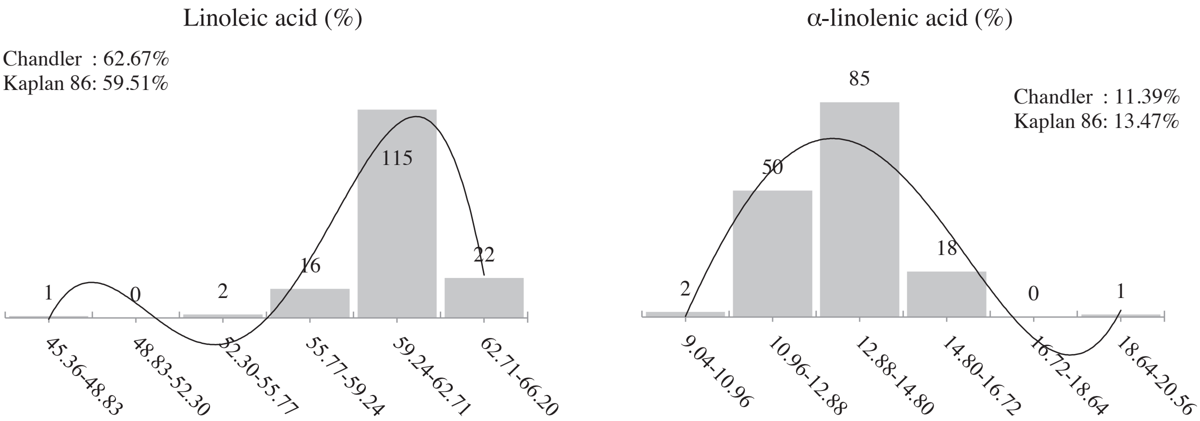

Figure 4. Polyunsaturated fatty acid content distribution of F1 walnut genotypes.

to 23.65 µg/g (WCK138). The content of α-tocopherol was also Chandler 4.44 µg/g and Kaplan-86 4.51 µg/g. It was found that the parents remained below average in terms of α-tocopherol and F1 genotypes showed heterosis. In this research, the lowest value of β-tocopherol content was detected in WCK10 with 1.90µg/g, and the highest value was detected in WCK149 with 12.57 µg/g. All other walnut genotypes for β-tocopherol were included in between these values. The γ-tocopherol contents ranged from 90.22 µg/g to 394.96 µg/g for F1 walnut genotypes, and 298.74 µg/g to 291.62 µg/g for Chandler and Kaplan-86, respectively (Figure 5). Within tocopherols, γ-tocopherol is the main component followed by δ- and α-tocopherol. Amaral et al. (2003) and Kafkas et al. (2017) reported that, the tocopherol content of different walnut (*Juglans regia* L.) varieties and γ-tocopherol was the most abundant tocopherol isomer in all cultivars. Similar to our study and previous reports, Martinez et al. (2010) reported the highest total phenolic values in diffrent walnuts cultivars and ranged from α-tocopherol (8.7 to 40.50 mg 100 kg^{-1}), β-tocopherol (8.20 to 42.30 mg 100 kg^{-1}) and γ-tocopherol (193.70 to 264.63 mg 100 kg^{-1}). In another study, Kafkas et al. (2010) reported that α-tocopherol content of cultivars varied between 28.33 µg/g (Howard) and 38.76 µg/g (Sen), β + γ tocopherol content varied between 161.01 µg/g (Howard) and 312.19 µg/g (Sen) and as for the δ-tocopherol content varied between 17.35 µg/g (Serr) and 40.77 µg/g (Sen). In our study, when tocopherol isomers were compared, it was observed that γ-tocopherol was the highest and similar, while small numerical differences were observed when compared to previous studies (Li et al., 2007; Bada et al., 2010; Beyhan et al., 2017). Significant variation was observed regarding genotype, ecology, cultural practices crop year and geographic origin (Maguire et al., 2004; Li et al., 2007; Martinez et al., 2010). All walnut genotypes showed a significant variation in terms of fatty acid compositions and tocopherol contents. These results provided important information both further breeding studies on these walnut genotypes and in terms of the nutrient content of walnuts.

4. Conclusion

The results presented in our a significant variation was observed in F1 walnut genotypes in terms of fatty acid and

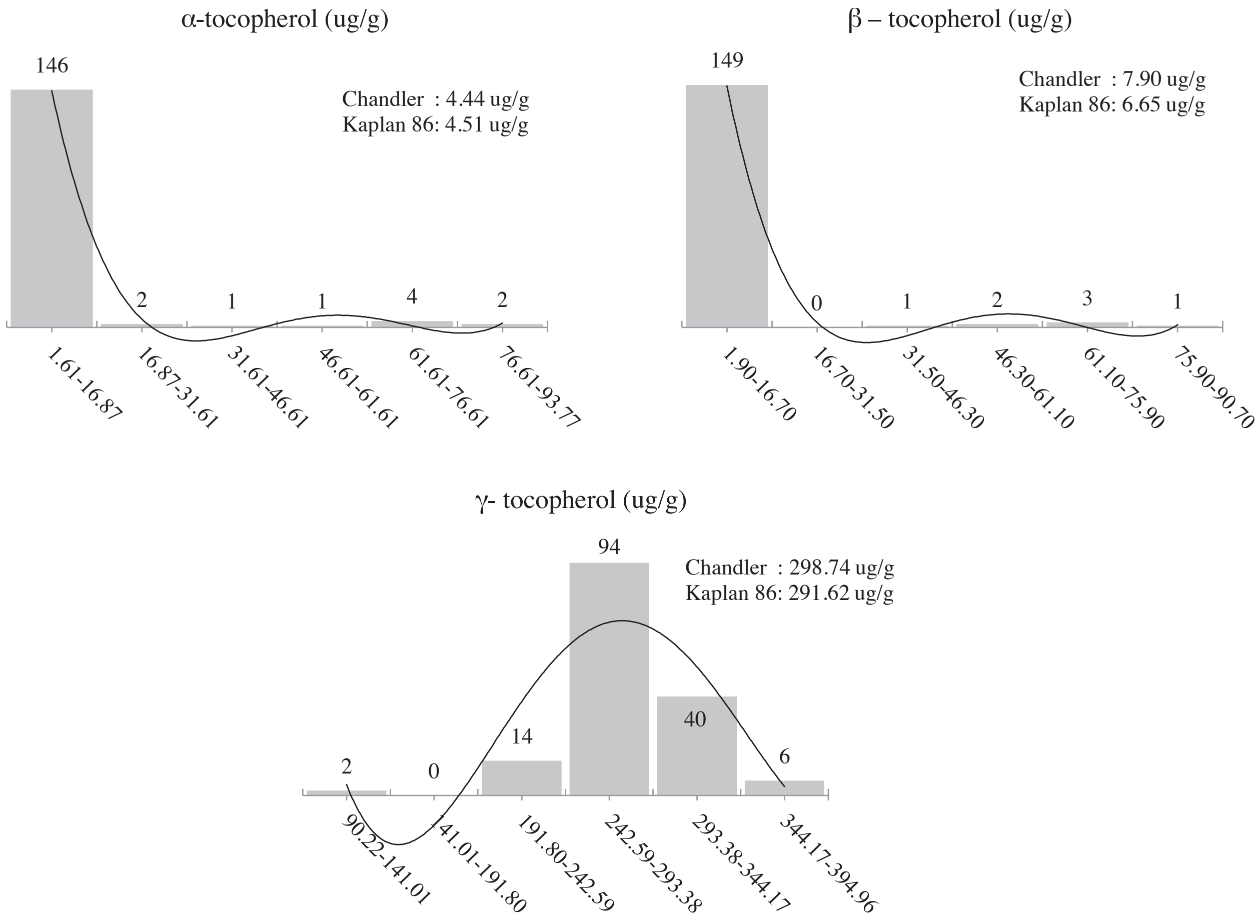

Figure 5. Tocopherol contents distribution of F1 walnut genotypes.

Table 3. Tocopherol contents of Chandler × Kaplan-86 F1 walnut genotypes and their parents

Genotypes	α-tocopherol (ug/g)	Β-tocopherol (ug/g)	γ-tocopherol (ug/g)
WCK1	2.75 ± 1.10	3.02 ± 0.65	304.12 ± 24.12
WCK2	9.65 ± 2.98	8.46 ± 0.66	318.32 ± 26.02
WCK3	3.21 ± 0.12	6.03 ± 0.08	263.00 ± 32.26
WCK4	10.06 ± 1.31	9.09 ± 1.19	308.82 ± 39.64
WCK5	4.32 ± 0.70	8.80 ± 0.91	292.07 ± 6.54
WCK6	7.48 ± 4.10	5.31 ± 0.54	283.88 ± 24.06
WCK9	3.76 ± 0.53	5.43 ± 0.29	348.06 ± 24.06
WCK10	11.16 ± 1.69	1.90 ± 2.24	233.08 ± 5.28
WCK11	3.54 ± 0.70	3.50 ± 0.61	265.22 ± 8.78
WCK13	4.34 ± 0.16	6.63 ± 1.14	230.32 ± 20.02
WCK19	11.60 ± 1.56	5.44 ± 0.85	324.68 ± 18.92
WCK21	11.19 ± 0.85	2.20 ± 0.20	335.07 ± 21.32
WCK22	8.02 ± 1.02	4.03 ± 1.04	296.32 ± 28.26
WCK23	5.04 ± 2.94	5.01 ± 0.50	292.56 ± 1.84
WCK24	4.92 ± 2.68	2.97 ± 1.06	267.74 ± 20.56

Table 3. (Continued).

WCK25	10.27 ± 0.24	3.87 ± 1.18	281.84 ± 16.88
WCK26	9.38 ± 0.24	6.23 ± 0.06	287.54 ± 5.00
WCK30	**1.61** ± 1.12	5.64 ± 0.69	277.02 ± 88.54
WCK34	14.68 ± 1.31	5.70 ± 0.33	315.34 ± 6.42
WCK36	6.62 ± 3.73	6.79 ± 0.26	242.24 ± 2.06
WCK37	3.35 ± 0.69	9.00 ± 0.77	276.36 ± 4.12
WCK39	9.24 ± 0.98	10.37 ± 1.91	296.12 ± 19.86
WCK41	2.38 ± 0.17	4.02 ± 0.20	246.08 ± 1.78
WCK44	5.85 ± 0.32	4.34 ± 0.11	266.60 ± 1.32
WCK46	3.91 ± 0.23	6.37 ± 0.03	342.06 ± 0.88
WCK47	9.50 ± 0.43	7.90 ± 0.46	297.36 ± 12.01
WCK48	8.33 ± 0.40	7.40 ± 0.31	251.96 ± 4.84
WCK50	5.02 ± 2.88	6.23 ± 0.44	328.54 ± 22.09
WCK51	14.53 ± 1.83	6.36 ± 0.43	311.64 ± 38.56
WCK52	13.99 ± 2.10	5.54 ± 0.51	329.92 ± 33.06
WCK55	11.89 ± 0.07	9.45 ± 0.08	237.08 ± 1.07
WCK56	14.56 ± 0.09	7.65 ± 0.09	251.04 ± 0.72
WCK57	11.25 ± 1.94	5.30 ± 0.38	208.64 ± 15.18
WCK58	3.45 ± 0.16	9.23 ± 0.01	364.02 ± 1.68
WCK59	3.64 ± 0.83	9.34 ± 1.08	386.07 ± 36.42
WCK60	8.48 ± 0.03	9.66 ± 0.17	291.02 ± 0.48
WCK62	8.26 ± 0.52	5.99 ± 1.06	394.96 ± 33.84
WCK63	13.37 ± 0.02	9.22 ± 0.24	251.04 ± 2.26
WCK64	11.16 ± 2.81	8.00 ± 0.63	273.04 ± 27.58
WCK66	4.77 ± 0.71	3.21 ± 2.01	277.09 ± 4.98
WCK67	6.83 ± 3.85	8.04 ± 0.55	269.78 ± 3.60
WCK68	4.53 ± 0.78	5.81 ± 0.98	297.98 ± 12.58
WCK70	6.44 ± 0.66	6.54 ± 0.48	273.08 ± 14.70
WCK73	2.77 ± 0.12	7.46 ± 0.08	288.08 ± 0.62
WCK74	6.02 ± 3.35	10.84 ± 0.53	317.16 ± 13.58
WCK75	6.95 ± 0.50	7.98 ± 0.61	299.96 ± 21.00
WCK76	8.61 ± 1.85	6.45 ± 0.43	279.92 ± 1.22
WCK80	8.92 ± 3.09	5.45 ± 0.65	311.01 ± 29.02
WCK81	3.86 ± 0.20	4.63 ± 0.28	297.66 ± 61.16
WCK82	12.16 ± 1.14	6.53 ± 0.41	284.32 ± 10.06
WCK83	5.20 ± 0.41	2.15 ± 0.95	135.74 ± 6.52
WCK85	9.40 ± 0.69	4.87 ± 3.25	244.84 ± 22.08
WCK86	6.16 ± 1.79	5.80 ± 0.77	282.32 ± 13.18
WCK87	8.68 ± 0.52	7.46 ± 0.54	280.42 ± 16.84
WCK88	11.52 ± 0.04	3.22 ± 0.09	259.08 ± 0.66
WCK89	5.20 ± 0.41	2.15 ± 0.95	275.32 ± 1.54
WCK91	5.88 ± 0.62	8.72 ± 0.70	337.36 ± 37.16
WCK93	4.45 ± 0.03	7.98 ± 0.06	308.34 ± 2.26

Table 3. (Continued).

WCK94	3.56 ± 0.02	6.56 ± 0.08	288.09 ± 1.36
WCK95	4.12 ± 2.30	9.25 ± 1.59	329.34 ± 59.46
WCK96	2.81 ± 0.03	7.54 ± 0.12	245.12 ± 1.00
WCK97	7.46 ± 0.43	7.22 ± 0.55	278.54 ± 11.96
WCK98	7.59 ± 0.88	12.57 ± 1.45	336.38 ± 36.86
WCK99	5.77 ± 3.17	7.13 ± 0.53	301.38 ± 32.32
WCK100	11.54 ± 0.91	8.91 ± 0.57	280.36 ± 3.07
WCK103	7.05 ± 1.80	7.29 ± 0.89	253.04 ± 13.56
WCK104	8.84 ± 0.75	4.20 ± 0.84	240.68 ± 4.26
WCK106	7.67 ± 1.09	6.45 ± 0.60	271.06 ± 19.58
WCK108	3.45 ± 0.11	4.56 ± .0.04	255.12 ± 1.48
WCK111	4.77 ± 0.17	7.44 ± 0.06	263.12 ± 0.42
WCK113	8.35 ± 0.62	6.57 ± 2.48	282.42 ± 7.34
WCK115	8.48 ± 0.94	7.52 ± 0.94	248.08 ± 27.06
WCK116	4.69 ± 0.47	7.26 ± 0.36	297.42 ± 14.32
WCK117	8.15 ± 3.12	7.84 ± 1.52	264.48 ± 20.42
WCK118	3.55 ± 0.11	5.75 ± 0.11	223.96 ± 0.52
WCK120	6.80 ± 3.27	4.93 ± 0.43	288.78 ± 10.92
WCK121	7.24 ± 0.54	7.83 ± 0.23	322.06 ± 9.08
WCK124	3.84 ± 0.16	5.46 ± 0.15	256.09 ± 1.32
WCK125	13.03 ± 2.14	7.88 ± 1.11	253.84 ± 30.30
WCK126	8.95 ± 0.07	4.52 ± 0.44	242.06 ± 7.22
WCK127	12.74 ± 0.53	6.09 ± 0.35	348.62 ± 16.26
WCK128	9.28 ± 1.52	7.98 ± 0.79	299.94 ± 32.52
WCK130	5.44 ± 0.53	7.25 ± 0.36	275.00 ± 15.08
WCK131	3.55 ± 0.11	6.66 ± 0.02	254.09 ± 0.88
WCK132	6.73 ± 4.79	7.20 ± 0.41	278.21 ± 7.34
WCK134	3.80 ± 0.22	4.67 ± 0.09	288.42 ± 0.36
WCK135	4.81 ± 0.07	6.69 ± 0.07	274.88 ± 0.64
WCK138	23.55 ± 4.76	5.58 ± 1.24	224.62 ± 15.05
WCK139	2.44 ± 0.16	5.81 ± 0.33	294.94 ± 1.26
WCK142	6.27 ± 0.11	5.77 ± 0.48	233.32 ± 0.84
WCK144	7.54 ± 0.12	7.78 ± 0.22	277.12 ± 0.28
WCK145	10.33 ± 1.08	6.46 ± 5.12	265.32 ± 10.22
WCK146	7.51 ± 2.31	6.66 ± 8.46	256.78 ± 3.90
WCK148	8.45 ± 0.18	9.65 ± 0.07	290.72 ± 2.24
WCK149	7.06 ± 1.47	9.07 ± 0.81	275.64 ± 4.76
WCK150	6.68 ± 0.16	8.77 ± 0.06	227.72 ± 0.22
WCK151	6.00 ± 0.14	6.66 ± 0.89	298.01 ± 25.16
WCK154	6.69 ± 0.69	5.49 ± 0.84	341.38 ± 16.56
WCK156	8.96 ± 0.44	7.70 ± 0.52	289.44 ± 0.52
WCK157	8.22 ± 0.11	8.15 ± 1.30	250.88 ± 12.01
WCK158	6.91 ± 0.45	7.67 ± 0.09	267.12 ± 2.22

Table 3. (Continued).

WCK159	8.56 ± 0.56	7.40 ± 0.34	310.04 ± 10.48
WCK160	8.74 ± 0.32	4.68 ± 0.07	294.84 ± 1.14
WCK161	7.55 ± 0.33	3.85 ± 0.23	277.08 ± 2.36
WCK164	5.15 ± 3.87	5.35 ± 0.20	248.07 ± 4.74
WCK165	7.86 ± 0.45	5.74 ± 0.05	278.09 ± 6.65
WCK166	13.04 ± 0.88	6.07 ± 0.83	324.78 ± 14.38
WCK167	7.12 ± 0.93	7.07 ± 0.40	90.22 ± 6.17
WCK168	7.68 ± 0.08	6.36 ± 0.08	283.22 ± 4.55
WCK173	7.67 ± 3.38	5.62 ± 4.10	300.68 ± 7.64
WCK175	4.20 ± 2.24	5.29 ± 0.48	292.86 ± 5.62
WCK176	7.67 ± 0.11	4.65 ± 0.81	269.612 ± 6.50
WCK178	11.89 ± 0.33	6.87 ± 0.22	277.09 ± 6.04
WCK179	8.97 ± 0.44	4.71 ± 0.18	254.09 ± 13.63
WCK180	7.98 ± 1.56	4.82 ± 1.27	258.06 ± 18.34
WCK181	9.68 ± 0.13	6.93 ± 1.06	285.94 ± 12.97
WCK182	5.88 ± 0.56	6.53 ± 0.15	287.03 ± 14.86
WCK183	7.77 ± 0.16	5.45 ± 0.07	255.05 ± 11.37
WCK184	6.43 ± 0.27	4.19 ± 0.50	298.24 ± 10.44
WCK186	17.40 ± 2.25	5.84 ± 0.95	291.18 ± 21.58
WCK187	8.48 ± 0.17	6.64 ± 0.23	259.32 ± 12.86
WCK188	10.50 ± 1.94	10.06 ± 1.17	293.04 ± 25.88
WCK189	6.56 ± 0.35	3.21 ± 2.67	218.84 ± 12.40
WCK190	6.62 ± 0.12	5.70 ± 0.18	234.09 ± 21.06
WCK191	8.69 ± 0.17	5.47 ± 0.19	275.74 ± 14.45
WCK192	7.71 ± 0.15	4.51 ± 0.16	257.07 ± 14.63
WCK193	9.37 ± 25.76	4.46 ± 0.56	245.08 ± 18.59
WCK195	10.46 ± 1.05	5.19 ± 0.71	346.26 ± 28.21
WCK196	8.49 ± 0.01	5.57 ± 0.63	268.72 ± 15.71
WCK197	7.86 ± 0.20	8.72 ± 1.30	298.03 ± 8.21
WCK198	7.86 ± 0.06	5.55 ± 0.18	275.03 ± 12.63
WCK199	7.04 ± 1.23	4.56 ± 0.94	244.32 ± 13.67
WCK201	7.8 ± 1.15	5.72 ± 1.86	280.52 ± 10.73
WCK202	7.10 ± 0.83	6.53 ± 0.59	311.66 ± 5.30
WCK203	3.94 ± 0.62	3.60 ± 1.29	287.02 ± 14.42
WCK204	4.77 ± 1.48	7.50 ± 1.02	289.42 ± 15.14
WCK205	3.3 ± 0.49	5.04 ± 1.32	309.14 ± 15.07
WCK206	5.85 ± 0.63	7.23 ± 0.49	261.86 ± 6.58
WCK207	6.29 ± 0.95	3.58 ± 0.08	264.18 ± 15.42
WCK211	5.31 ± 0.25	5.48 ± 0.33	277.84 ± 16.41
WCK212	10.69 ± 1.16	6.73 ± 0.43	300.46 ± 13.73
WCK213	10.45 ± 0.31	4.68 ± 0.20	257.05 ± 8.87
WCK214	6.43 ± 0.06	4.56 ± 0.08	257.72 ± 15.05
WCK217	5.95 ± 3.69	5.05 ± 0.36	285.48 ± 8.04
WCK218	5.41 ± 0.21	6.51 ± 0.06	243.12 ± 16.44

Table 3. (Continued).

WCK219	13.78 ± 0.99	7.00 ± 0.36	285.03 ± 10.93
WCK220	7.57 ± 1.82	5.22 ± 0.17	248.26 ± 7.51
WCK221	8.49 ± 0.07	8.48 ± 0.06	275.26 ± 13.47
WCK222	5.52 ± 1.39	3.27 ± 0.20	314.56 ± 9.25
WCK227	13.29 ± 1.37	9.51 ± 0.33	265.08 ± 14.78
WCK229	7.45 ± 0.11	5.71 ± 0.04	253.26 ± 12.56
WCK230	7.56 ± 0.03	4.62 ± 0.11	258.48 ± 12.62
WCK231	8.53 ± 0.21	5.54 ± 0.09	270.02 ± 13.65
WCK232	7.45 ± 0.06	7.58 ± 0.13	237.05 ± 11.11
WCK235	6.48 ± 0.01	8.59 ± 0.12	251.78 ± 13.23
WCK237	9.09 ± 0.29	3.83 ± 0.11	298.09 ± 15.67
Chandler	4.44 ± 0.31	7.90 ± 0.45	298.74 ± 12.56
Kaplan-86	4.51 ± 0.27	6.65 ± 0.07	291.62 ± 11.83
Minumum value	1.61	1.90	90.22
Maximum value	23.65	12.37	394.96
Avarage value	7.55	6.32	279.04

*Each value is expressed as mean ± standard deviation.

tocopherol content and variation distubuiton also differed based on the components. It is clear that this study showed that F1 genotypes showed heterosis based on the tocopherol contents and the fatty acid levels. According to the results of many studies, it is thought that walnut fruits, which are an important source of tocopherol and fatty acid, have a positive effect on human health in increasing their consumption in terms of nutrition and health. This will not only benefit the producer and the consumer, but also contribute to the development of improvement studies and sectors related to these fruit species. The results obtained in this study are thought to be important in terms of being a source for further studies and revealing the nutritional values of world walnut gene pools. In conclusion, in this study, some F1 walnut genotypes were evaluated in terms of some biochemical features, and those with superior characteristics among these genotypes will have the chance to be a cultivar for walnut cultivation.

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Conflict of interest

The authors confirm that they have no conflicts of interest with respect to the work described in this manuscript.

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