Determination of Bioactive Compounds and Mineral Contents of Seedless Parts and Seeds of Grapes

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In this study, phenolic compounds, minerals, total flavonoids, total phenolic contents and antioxidant activities of the seedless parts (pulp+skin) and seeds of table and wine grapes were determined. Also, the total oil, tocopherol contents and fatty acid composition of seed oils of table and wine grapes were investigated. The highest total phenolic content of the grape pulp was found in Trakya ilkeren (199.063 mg/100 g), while total flavonoid and antioxidant activity of the pulp was determined at a high level in Red Globe (6.810 mg/g, 90.948%). Antioxidant activity, and the total phenolic and flavonoid contents of grape seeds varied between 86.688 and 90.974%, 421.563 and 490.625 mg GAE/100 g, and 90.595 and 145.595 mg/g respectively ($p < 0.05$). Generally, the main phenolic compounds of all grape pulps and seeds were gallic acid, 3,4-dihydroxybenzoic acid, (+)-catechin and 1,2-dihydroxybenzene. In addition, the oil contents of grape seeds ranged from 5.275 (Çavuş) to 13.881% (Çınarlı karası) ($p < 0.05$). The major fatty acids of grape seed oils were linoleic, oleic and palmitic acid. The seed oil of the Trakya ilkeren variety was rich in tocopherols in comparison with the other varieties. The major minerals of both the seedless parts and the seeds were determined as K, Ca, P, S, Mg.

INTRODUCTION

Grapes (*Vitis* spp.) belong to the Vitaceae family and are used mainly for their juice, especially in the wine industry (Lutterodt *et al*., 2011). The bioactive compounds, including simple phenolics, flavonoids, anthocyanins, stilbenes, proanthocyanidins and vitamin E, are important phytochemicals in grapes. Simple phenolic compounds of grapes are hydroxycinnamic acid (p-coumaric, caffeic, sinapic and ferulic acids) and hydroxybenzoic acid (gallic, gentisic, protocatechuic and p-hydroxybenzoic acids) (Georgiev *et al*., 2014; Shi *et al*., 2016). Several phenolic compounds are found in the skins, pulp and seeds of grapes (Nile *et al*., 2013). Most of these compounds have antioxidant properties, which play a significant role in human health because they reduce the oxidation of low-density protein, thereby decreasing the risk of heart disease and preventing cancer (Yılmaz *et al*., 2015; Yalcin *et al*., 2016). The amount of phenolics is affected by the grapevine variety, climatic and geographical factors, cultural practices, and the stage of ripeness (Obreque-Slier *et al*., 2010; Farhadi *et al*., 2016).

A large amount of grape seeds are obtained from the waste of fruit products. The wastes of this industry represent about 50% to 60% of the raw fruit. On the other hand, grape seeds are used as a source of oil, with nutritional and bioactive constituents (Kamel *et al*., 1985; Hassanein & Abedel-Razek, 2009). Grape seed oils obtained from grape by-products can be used for pharmaceutical and nutritional purposes due to their high levels of vitamin E and unsaturated fatty acids (Hassanein & Abedel-Razek, 2009). These by-products contain some valuable substances, such as phenolics, fatty acids, tocopherols and minerals with potential applications in the food industry (Crews *et al*., 2006; Wie *et al*., 2009). Generally, grape seed oil is used in salad dressings, marinades, deep frying, flavoured oils, baking, massage oil and cosmetic products (Akın & Altındişli, 2011).

The aim of this study was to investigate some physicochemical properties (soluble solid content, titratable acidity and maturation index of grapes; total dry matter of grape seeds), antioxidant activities, total phenolic and flavonoid contents, phenolic compounds and mineral contents of grapes (pulp+skin) and seeds, and the oil yields, fatty acid and tocopherol contents of grape seeds due to the importance of grapes and seeds as sources of bioactive compounds, oil, tocopherol and minerals.

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MATERIAL AND METHODS

Grapes (Çavuş, Çınarlı Karası, Kalecik Karası, Red Globe, Trakya Ilkeren, Yalova Incisi and Yapıncak) were obtained from the Viticulture Research Institute of Tekirdağ in Turkey. All varieties belong to *Vitis vinifera*. Their colours and properties are given in Table 1.

Sample extraction

For the phenolic compounds and antioxidants, the seedless parts (pulp+skin) and seeds of the grapes were extracted according to Gomez-Alonso *et al*. (2007) with some modifications. About 2 g of each sample were added to 15 ml of methanol:water:formic acid (5:4.85:1.5, v/v).The mixture was sonicated for 2 min, followed by centrifugation at 4 500 rpm for 15 min, after which the supernatants were collected. Then 10 ml of *n*-hexane were added and mixed using a vortex apparatus. After the extract was concentrated at 50°C in a rotary evaporator, and after the dried extracts had been dissolved in 1.5 ml of methanol, it was injected into HPLC.

Physicochemical analysis

The content of soluble solids, titratable acidity, maturation index and total dry matter were analysed according to Cemeroğlu (1992).

Total phenolic content

The total phenolic content of the grape pulp and seeds was determined by Folin–Ciacueltau (FC) reagent according to Yoo *et al*. (2004). About 10 mL of Na_2CO_3 solution and 1 ml of Folin–Ciacueltau were mixed, and then completed with 25 ml deionised water. After 1 h, the total phenolic content was measured at 750 nm in a spectrophotometer. The results were given as mg gallic acid equivalent/100 g.

Antioxidant activity

The free radical-scavenging activity of the samples was determined using DPPH (1,1-diphenyl-2-picrylhydrazyl) according to Lee *et al*. (1998). The extract was mixed with 2 mL methanolic solution of DPPH. The mixture was shaken vigorously and allowed to stand at room temperature for 30 min. The absorbance was recorded at 517 nm by using a spectrophotometer.

Determination of phenolic compounds

Phenolic compounds were determined by a Shimadzu-HPLC equipped with a PDA detector and an Inertsil ODS-3 (5 µm; 4.6 x 250 mm) column. As mobile phases, 0.05% acetic acid in water (A) and acetonitrile (B) mixture were used. The flow rate of the mobile phase and the injection volume were 1 ml/min at 30°C and 20 µl respectively. The gradient program was as noted: 0 to 0.10 min 8% B; 0.10 to 2 min 10% B; 2 to 27 min 30% B; 27 to 37 min 56% B; 37 to 37.10 min 8% B; and 37.10 to 45 min 8% B. The peak records were carried out at 280 and 330 nm. The total running time for each sample was 60 min. Phenolic compounds were determined according to the retention time and absorption spectra of peaks of standard compounds. The total area under the peak was used to quantify the phenolics.

Total flavonoid content

The total flavonoid content of the samples was determined using the colorimetric method according to Hogan *et al*. (2009). One mL of extract was mixed with 0.3 ml of $NaNO_2$, 0.3 ml of $AlCl_3$ and 2 ml of NaOH respectively. The absorbance of the mixtures was recorded at 510 nm using a spectrophotometer.

Mineral content

After 5 g of grape samples had been dried in a drying cabinet at 70°C, about 0.5 g of the sample was digested in a closed microwave system by using 5 ml of 65% HNO_3 and 2 ml of 35% H_2O_2. After 20 ml ultra-deionised water was added to the digested sample, the sample was analysed by ICP AES (Varian-Vista, Australia) for minerals (Skujins, 1998).

Oil extraction

Oil was extracted from the ground seeds with petroleum ether in a Soxhlet apparatus for 6 h. The solvent (petroleum ether) was then evaporated (AOAC, 1990). The obtained oil was kept in coloured bottles at -18°C until analysis.

Fatty acid composition

The oil sample (50 to 100 mg) was converted to its fatty acid methyl esters (FAME). The fatty acid methyl esters were identified by comparing the retention time of the samples and appropriate standards of fatty acids methyl esters (AOAC,

TABLE 1
Colour of pulp and properties

Grape	Colour of pulp	Properties
Çavuş	Yellow-green	Table grape, light oval, very coarse grained, specific flavour
Çınarlı Karası	Black	Table grape, round-elliptical, coarse-grained, neutral flavour
Kalecik Karası	Blue-black	Wine grape, round, medium grains, specific flavour
Red Globe	Pink-red	Table grape, round-elliptical, very coarse-grained, neutral flavour
Trakya İlkeren	Blue-black	Table grape, round, very coarse-grained, neutral flavour
Yalova İncisi	Green-yellow	Table grape, light-elliptical, very coarse-grained, neutral flavour
Yapıncak	White-yellow	Both table and wine grapes, elliptical, coarse-grained, specific flavour

Ten kg of random grape samples were harvested at the mature stage of the grapes. Grapes were brought to the laboratory in a cooler and cut down the middle with a knife. Pulp+skin (seedless parts) and seeds were separated manually. The seeds of the grapes were cleaned using distilled water and then dried in an oven (Nüve FN055 Ankara, Turkey) at 40°C until a constant weight was obtained. Seeds were stored at +4°C; the pulp and skin parts were frozen at -80°C until analysis.

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1990). The fatty acid methyl esters were injected into a gas chromatograph (Shimadzu GC-2010) with a capillary column, CP-Sil 88 (100 m long, 0.25 mm ID, film thickness 0.2 µm) and flame-ionisation detector (FID).

Tocopherol content

Tocopherol content was measured according to Spika *et al*. (2015). About 0.1 g of oil was dissolved in 10 ml of *n*-hexane and filtered through a 0.45 µm nylon filter. HPLC analyses of tocopherols were done using a Shimadzu-HPLC equipped with a PDA detector and LiChroCART Silica 60 (4.6 x 250 mm, 5µ; Merck, Darmstadt, Germany) column. Tocopherols were separated by isocratic chromatography using a mobile phase of 0.7% propan-2-ol in *n*-hexane. The flow rate of the mobile phase was 0.9 ml/min, and the injection volume was 20 µl. The peaks were recorded at 295 and 330 nm with a PDA detector. The total running time per sample was 30 min. Standard solutions of tocopherols (α-, β-, γ- and δ-tocopherol) were constructed at concentrations of 0 to 100 mg/L. All analyses were done in triplicate.

Statistical analyses

Analysis of variance (ANOVA) using JUMP version 9.0 (SAS Inst. Inc., Cary NC, USA)and the mean ± standard deviation (MSTAT C) of the results were calculated according to Püskülcü and İkiz (1989).

RESULTS AND DISCUSSION

The soluble solid contents (of the pulp), total dry matter (of the seeds), titratable acidity, maturation index, harvest date and some properties of grape cultivars are shown in Table 2. The highest and the lowest soluble solid contents were found in Kalecik karası (22.00°Brix) and Red Globe (15.20°Brix) respectively ($p < 0.05$). The results for titratable acidity ranged from 4.10 g/L to 7.80 g/L. Maximum titratable acidity was determined in Kalecik karası, while the minimum value was found in Yapıncak. The maturation index of the grape varieties at harvesttime varied from 23.40 (Yapıncak) to 47.50 (Çınarlı karası). Water-soluble dry matter, titratable acidity, maturation index and grape colour had an effect on the maturation of the grapes. The grapes can be harvested when the maturation index is a minimum of 20 (OIV, 2008). As can be seen in Table 2, the grapes used in the current experiment were harvested at the maturation stage. According to the total dry matter assay, the highest value was found in Yapıncak seeds (68.27%), while the lowest amount of dry matter was in seeds of Yalova incisi (56.38%). According to Samoticha *et al*. (2017), the soluble solid contents range from 10.6°Brix to 22.3°Brix in white grapes, and from 15.2°Brix to 20.6°Brix for red grapes. The average total acidity of the white and red cultivars was 0.95 and 0.93 g tartaric acid/100 g, respectively.

The antioxidant activities, total phenolic and total flavonoid contents of the grape samples are presented in Table 3. Antioxidant activities of the samples varied between 30.298% and 90.948%. The total phenolic contents of the grape samples were found to be between 44.063 and 199.063 mg GAE/100 g. Trakya ilkeren had the highest total phenolic content (199.06 mg GAE/100 g), followed by Red Globe (170.313 mg GAE/100 g) and Çavuş (146.641 mg GAE/100 g). Significant differences were observed between the total phenolic contents of the grape extracts ($p < 0.05$). The total flavonoid contents of the grape varieties ranged from 0.491 to 6.810 mg/g. The total flavonoid content of Red Globe (6.810 mg/g) was found to be close to that of Trakya ilkeren (6.744 mg/g). The minimum flavonoid content was determined in Yalova incisi, with a value of 0.491 mg/g. Hogan *et al*. (2009) observed that the total phenolic and flavonoid contents of wine grapes varied between 0.68 and 1.82 mg GAE/g and 0.48 and 1.19 mg RE/g respectively. In another study, the total phenolic contents of both grape pulp and grape seed varied from 9.26 to 62.29 mg GAE/100 g and from 162.29 to 326.18 mg GAE/100 g respectively (Yılmaz *et al*., 2015). In the experiments reported by Samoticha *et al*. (2017), the total phenolic content of white grapes varied from 1974.9 to 3 884.4 mg/100 g, while it ranged from 1 037.0 to 5 759.1 mg/100 g in red grapes. The degree of maturity and environmental conditions influence the amount of phenolic compounds. Moreover, the fruit parts, such as skin, pulp or seeds, exhibit different polyphenol concentrations. The antioxidant activities of grape varieties range from 93.80% to 95.62% in grape skin, from 88.16% to 94.68% in grape pulp, and from 88.10% to 94.46% in grape seeds (Farhadi *et al*., 2016).

Antioxidant activities, along with the total phenolic and total flavonoid contents of the grape seeds, are shown in Table 3. The antioxidant activities of the seeds varied between 86.688% and 90.974%, and was higher than that found in

TABLE 2
Some Properties of Grape Varieties

Grape varieties	Harvest date	Soluble solid content (°Brix, pulp)	Titratable acidity (g/L)	Maturation index	Total dry matter (%, seed)
Çavuş	2015-09-24	16.90 ± 0.36de*	5.00 ± 0.21b	34.10 ± 0.27c	62.41 ± 1.14d
Çınarlı Karası	2015-09-08	17.20 ± 0.41de**	7.40 ± 0.13a	23.40 ± 0.93g	66.47 ± 0.96c
Kalecik Karası	2015-09-14	22.00 ± 0.67a	7.80 ± 0.93a	28.20 ± 0.48e	66.55 ± 1.09c
Red Globe	2015-09-29	15.20 ± 0.73e	4.60 ± 0.87c	33.20 ± 0.51d	67.03 ± 1.13b
Trakya İlkeren	2015-08-03	19.40 ± 0.27b	7.20 ± 0.69a	26.90 ± 0.38ef	59.29 ± 0.78e
Yalova İncisi	2015-08-14	18.40 ± 0.51d	4.50 ± 0.57c	40.90 ± 0.41b	56.38 ± 0.89f
Yapıncak	2015-10-02	19.60 ± 0.84b	4.10 ± 0.77c	47.50 ± 0.97a	68.27 ± 1.57a

* mean ± standard deviation; ** means in the same raw with the same letters are not significantly different ($p < 0.05$)

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TABLE 3
Antioxidant activity, total phenolic and total flavonoid contents of grape pulp+skin and seeds

Grape pulp+skin	Antioxidant activity (%)	Total phenolic content (mg/100 g)	Total flavonoid content g/g)
Red Globe	90.948 ± 0.001a	170.313 ± 0.014b	6.810 ± 0.005a
Çınarlı karası	82.588 ± 0.007c	127.813 ± 0.017d	3.691 ± 0.003c
Çavuş	88.392 ± 0.005b	146.641 ± 0.023c	4.654 ± 0.017b
Kalecik karası	79.925 ± 0.013e	119.531 ± 0.026ef	3.429 ± 0.001c
Yapıncak	80.085 ± 0.002d	121.016 ± 0.011e	3.848 ± 0.001c
Yalova incisi	30.298 ± 0.001f	44.063 ± 0.003g	0.491 ± 0.000d
Trakya ilkeren	90.735 ± 0.001a	199.063 ± 0.023a	6.744 ± 0.007a

Grape seeds	Antioxidant activity (%)	Total phenolic content (mg/100 g)	Total flavonoid content (mg/g)	Oil content (%)
Red Globe	89.191 ± 0.001*b	473.125 ± 0.020c	130.873 ± 0.005b	9.500 ± 0.300d
Çınarlı karası	86.88 ± 0.002d**	487.813 ± 0.008b	100.039 ± 0.004f	13.881 ± 0.881a
Çavuş	90.096 ± 0.001a	490.625 ± 0.006a	90.595 ± 0.005g	5.275 ± 0.075g
Kalecik karası	90.469 ± 0.000a	421.563 ± 0.032g	145.595 ± 0.005a	8.500 ± 0.300de
Yapıncak	87.966 ± 0.002c	429.688 ± 0.008f	115.595 ± 0.003cd	10.600 ± 0.400c
Yalova incisi	90.841 ± 0.001a	432.813 ± 0.005e	116.289 ± 0.010c	12.369 ± 0.369b
Trakya ilkeren	90.974 ± 0.002a	444.063 ± 0.041d	112.261 ± 0.011e	7.600 ± 0.800f

* mean ± standard deviation; ** means in the same row with the same letters are not significantly different ($p < 0.05$)

the pulp+skin of grape. The total phenolic contents of the grape seeds ranged from 421.563 to 490.625 mg GAE/100 g, and the richest variety in terms of total phenol content was Çavuş (490.625 mg GAE/100 g), followed by Çınarlı karası (487.813 mg GAE/100 g). The total flavonoid content of the grape seeds was considerably higher compared with the rest of the grapes and was found to be between 90.595 and 145.595 mg/g ($p < 0.05$). Concerning the results, the grape seeds contained rich bioactive substances. According to Ky *et al*. (2014), the total phenolic content of grape seed extracts varied from 36.6 to 49.7 mg GAE/g, and the antioxidant activity of the seed extract of grape varieties using DPPH ranged from 117.4 to 536.2 µmol Trolox/g (dry weight). The total phenolic contents of the grape seeds (825.80 to 3 313.5 mg GAE/100 g) were higher than that of the pulp (50.79 to 141.72 mg GAE/100 g) (Anastasiadi *et al*., 2010).

The highest oil content was determined in Çınarlı karası (13.881%), while the seeds of Çavuş variety (5.275%) contained the lowest oil content. Sabir *et al*. (2012) reported that the oil content of grape seeds ranged from 7.3% to 22.4%. In addition, the oil content of Red Globe and Kalecik Karası was found to be 142.1 and 123.3 g/kg. Fernandes *et al*. (2013) determined that the highest and lowest oil content of grape seeds was 12.40% and 3.95% respectively. In another study, the oil content in grape seeds of wine varieties averaged at 11.60 g/100 g (Lachman *et al*., 2015).

The phenolic compounds of the grape pulp and seeds are presented in Table 4. Generally, the main phenolic compounds of all varieties were gallic acid, 3,4-dihydroxybenzoic acid, (+)-catechin and 1,2-dihydroxybenzene. The highest gallic acid was determined in Yalova incisi (77.832 mg/100 g), Çınarlı karası (57.325 mg/100 g) and Kalecik karası (49.704 mg/100 g) ($p < 0.05$). The highest content of 3,4-dihydroxybenzoic acid was found in the Çavuş (101.462 mg/100 g) and Yalova incisi (54.093 mg/100 g) varieties. Yalova incisi (53.776 mg/100 g) had also the greatest content of (+)-catechin. The highest amount of 1,2-dihydroxybenzene was also observed in Yalova incisi (95.589 mg/100 g) and Çınarlı karası (56.357 mg/100 g). Significant differences were observed among the gallic acid, caffeic acid and rutin trihidrate contents of grape pulp extracts and between the 3.4-dihydroxybenzoic, resveratrol and isorhamnetin contents of grape seed extracts ($p < 0.05$). Moreover, apigenin 7 glucoside in Çavuş (22.995 mg/100 g), and the caffeic acid and quercetin contents in Yapıncak (20.244 mg/100 g and 22.968 mg/100 g respectively), were found to be higher than in other varieties. The phenolic compounds of grape seeds were determined as significantly high in comparison to in the rest of the grapes. The major phenolic compounds of Red Globe were 1,2-dihydroxybenzene (445.287 mg/100 g), rutin (202.915 mg/100 g), apigenin 7 glucoside (194.638 mg/100 g) and caffeic acid (142.473 mg/100 g). (+)-Catechin (284.424 mg/100 g), gallic acid (221.492 mg/100 g) and quercetin (201.424 mg/100 g) were highest in Çınarlı karası; 1,2-dihydroxybenzene (556.198 mg/100 g and 549.264 mg/100 g respectively) in Çavuş and Kalecik karası; (+)-catechin (480.509 mg/100 g and 309.335 mg/100 g, respectively) in Yapıncak and Yalova incisi; and rutin (231.542 mg/100 g) was found in a considerable amount in Trakya ilkeren varieties. The highest resveratrol content was observed in Çınarlı karası, with a value of 197.508 mg/100 g. Rockenbach *et al*., (2011) studied phenolic compounds of different grape varieties (Cabernet Sauvignon, Merlot, Bordeaux and Isabel), and the rutin, kaempferol, catechin, epicatechin, resveratrol and gallic acid contents ranged from 12.00 to 41.43 mg/100 g, from 0.0 to 15.09 mg/100 g, from 94.28 to 150.16 mg/100 g, from 0.0 to 44.36 mg/100 g, from

TABLE 4
Phenolic compounds of seedless parts and seeds of grapes (mg/100 g)

Phenolic compounds (pulp+skin)	Red Globe	Çınarlı karası	Çavuş	Kalecik karası	Yapıncak	Yalova incisi	Trakya ilkeren
Gallic acid	29.897 ± 1.768*f	57.325 ± 1.029b	16.740 ± 1.718g	49.704 ± 0.229c	35.674 ± 1.640e	77.832 ± 0.135a	37.976 ± 0.490d
3.4-Dihydroxybenzoic	11.775 ± 0.615e**	10.889 ± 0.615f	101.462 ± 1.223a	49.620 ± 1.839c	49.031 ± 0.791c	54.093 ± 0.317b	19.334 ± 2.236d
(+)-Catechin	10.922 ± 1.348e	15.287 ± 0.975c	8.724 ± 0.386f	14.602 ± 0.533d	20.460 ± 0.194b	53.776 ± 3.836a	15.537 ± 1.645c
1.2-Dihydroxybenzene	8.610 ± 1.033f	56.357 ± 0.898b	39.934 ± 4.238c	8.122 ± 0.520f	18.244 ± 0.752e	95.589 ± 2.318a	32.040 ± 0.649d
Syringic acid	4.478 ± 0.698e	5.738 ± 0.121d	18.655 ± 0.200b	2.867 ± 0.365f	19.169 ± 0.243a	18.799 ± 0.347b	7.997 ± 1.043c
Caffeic acid	2.358 ± 0.219f	6.230 ± 0.411d	20.244 ± 0.283b	0.904 ± 0.086g	24.978 ± 0.282a	13.803 ± 0.158c	4.219 ± 0.394e
Rutin trihidrate	4.346 ± 0.654e	2.389 ± 0.128f	14.479 ± 0.704b	1.039 ± 0.035g	18.514 ± 0.207a	8.928 ± 0.355c	6.857 ± 1.205d
p-Coumaric acid	0.652 ± 0.081e	0.816 ± 0.106d	2.603 ± 0.038a	0.148 ± 0.002f	1.424 ± 0.010b	1.134 ± 0.095b	0.914 ± 0.130c
trans-Ferulic acid	0.368 ± 0.034e	4.851 ± 0.774c	10.305 ± 0.287a	0.477 ± 0.029e	3.498 ± 0.274d	3.111 ± 0.366d	6.187 ± 0.949b
Apigenin 7 glucoside	3.344 ± 0.482e	4.531 ± 0.515d	22.995 ± 0.475a	3.273 ± 0.311e	7.854 ± 0.846c	1.956 ± 0.032f	11.849 ± 0.405b
Resveratrol	0.680 ± 0.010e	1.755 ± 0.197b	1.023 ± 0.002d	0.409 ± 0.029f	1.897 ± 0.153b	1.381 ± 0.134c	2.805 ± 0.398a
Quercetin	5.093 ± 0.173d	4.221 ± 0.266e	22.968 ± 0.050b	4.945 ± 0.023e	23.793 ± 0.105a	3.719 ± 0.378f	11.464 ± 1.426c
trans-Cinnamic acid	0.717 ± 0.020e	1.782 ± 0.101b	2.184 ± 0.064a	1.145 ± 0.059d	1.737 ± 0.109b	0.257 ± 0.012f	1.571 ± 0.121c
Naringenin	2.538 ± 0.145d	3.304 ± 0.314c	5.499 ± 0.436a	1.989 ± 0.107e	5.308 ± 0.507a	2.432 ± 0.314d	4.455 ± 0.523b
Kaempferol	3.264 ± 0.235c	4.455 ± 0.410b	5.137 ± 0.301a	1.620 ± 0.046d	5.680 ± 0.434a	4.244 ± 0.535b	4.221 ± 0.475b
Isorhamnetin	8.371 ± 0.417d	10.818 ± 0.677b	11.976 ± 0.672a	4.425 ± 0.014e	11.883 ± 0.910a	10.241 ± 0.729b	9.031 ± 0.648c
Phenolic compounds (seed)	Red Globe	Çınarlı karası	Çavuş	Kalecik karası	Yapıncak	Yalova incisi	Trakya ilkeren
Gallic acid	44.041 ± 1.624f	221.492 ± 4.628a	81.078 ± 5.590e	44.739 ± 0.241f	106.913 ± 2.127d	128.792 ± 4.475c	150.071 ± 2.850b
3.4-Dihydroxybenzoic	53.865 ± 0.523e	60.476 ± 0.262d	114.838 ± 2.731b	168.733 ± 2.286a	31.471 ± 1.653g	43.772 ± 1.514f	108.277 ± 0.476c
(+)-Catechin	71.689 ± 1.192e	284.424 ± 1.955c	56.423 ± 1.802f	132.597 ± 2.239d	480.509 ± 4.733a	309.335 ± 20.875b	135.255 ± 0.993d
1.2-Dihydroxybenzene	445.287 ± 25.48c	169.022 ± 7.244d	556.198 ± 1.278a	549.264 ± 5.739b	82.626 ± 0.993f	117.822 ± 2.946e	168.389 ± 1.601d
Syringic acid	25.656 ± 2.506g	163.793 ± 0.368c	80.925 ± 0.267d	218.647 ± 0.309a	66.053 ± 2.411e	35.632 ± 2.957f	194.457 ± 0.454b
Caffeic acid	142.473 ± 4.991c	45.422 ± 0.707d	38.817 ± 2.577e	31.152 ± 0.699f	171.839 ± 3.098b	143.010 ± 0.119c	174.288 ± 2.327a
Rutin trihidrate	202.915 ± 1.286b	64.207 ± 0.477f	99.780 ± 2.855e	184.008 ± 1.253c	189.627 ± 0.069c	154.473 ± 1.269d	231.542 ± 6.520a
p-Coumaric acid	9.471 ± 0.225d	22.271 ± 0.402a	10.671 ± 0.151c	9.887 ± 0.321d	10.809 ± 0.338c	8.484 ± 0.103e	11.836 ± 0.701b
trans-Ferulic acid	86.259 ± 2.007c	59.141 ± 0.539f	64.621 ± 2.780e	68.710 ± 1.031d	64.839 ± 0.312e	108.301 ± 1.040b	140.663 ± 1.712a
Apigenin 7 glucoside	194.638 ± 2.078a	85.521 ± 1.979f	87.450 ± 1.526f	45.231 ± 0.790g	90.038 ± 3.123e	112.600 ± 0.103c	137.092 ± 1.165b
Resveratrol	61.388 ± 2.549b	197.508 ± 2.432a	24.998 ± 0.274c	19.086 ± 0.042e	15.732 ± 0.404f	12.072 ± 0.491g	21.206 ± 0.384d
Quercetin	25.728 ± 0.125f	201.424 ± 1.826a	45.458 ± 1.869c	37.748 ± 0.645e	40.332 ± 0.553d	51.706 ± 1.745b	22.604 ± 0.305g
trans-Cinnamic acid	4.617 ± 0.298c	10.914 ± 0.159a	5.268 ± 0.300b	5.081 ± 0.231b	3.404 ± 0.103d	4.393 ± 0.092c	3.920 ± 0.242d
Naringenin	12.529 ± 0.146c	27.087 ± 0.712a	12.212 ± 0.521c	9.988 ± 0.416d	8.880 ± 0.093e	12.780 ± 0.451c	16.679 ± 0.681b
Kaempferol	34.746 ± 0.373b	45.110 ± 0.748a	22.530 ± 0.016c	14.689 ± 0.434f	20.364 ± 0.812d	15.010 ± 0.561e	22.345 ± 0.312c
Isorhamnetin	8.297 ± 0.308g	12.070 ± 0.606e	10.411 ± 0.601f	16.275 ± 0.998b	22.375 ± 0.729a	13.340 ± 0.722d	14.847 ± 0.154c

* mean ± standard devation; ** means in the same row with the same letters are not significantly different ($p < 0.05$)

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0.0 to 6.40 mg/100 g and from 4.59 to 18.68 mg/100 g, respectively. The phenolic acids of wine grapes were found to be gallic acid (16.7 to 72.6 µg/g), chlorogenic acid (2.1 to 3.3 µg/g), vanillic acid (2.2 to 49.4 µg/g), caffeic acid (0.1 to 9.2 µg/g), coumaric acid (2.8 to 4.2 µg/g) and ferulic acid (0.1 to 0.6 µg/g) (Hogan *et al.*, 2009). Anastasiadi *et al*. (2010) reported that the phenolic compounds of grape seeds were gallic acid (7.22 to 161.74 mg/100 g), catechin (143.63 to 1067.0 mg/100 g) and epicatechin (89.21 to 371.65 mg/100 g). The main phenolic compounds in the pulp of grapes were catechin (354 to 514 µg/g), epicatechin (135 to 234 µg/g), gallic acid (87 to 192 µg/g) and quercetin (87 to 198 µg/g). The gallic acid, catechin, epicatechin and quercetin contents of grape seeds were between 67 and 91 µg/g, 122 and 156 µg/g, 103 and 167 µg/g, 25 and 38 µg/g respectively (Farhadi *et al*., 2016).

The fatty acid composition of the grape seed oils is shown in Table 5. Grape seeds are a good source of essential fatty acids, and their linoleic acid contents varied between 66.94% and 77.359%. Significant differences were observed among the linoleic acid contents of grape seed oils ($p < 0.05$). The amount of oleic acids of seed oils ranged from 10.746% to 19.660%. Palmitic and stearic acids varied between 7.176% and 10.358% and 2.983% and 5.435% respectively ($p < 0.05$). The highest linoleic and the lowest oleic acid contents were found in the Çavuş variety. Furthermore, the linolenic acid content of seed oils was below 1%. According to Lachman *et al*. (2015), linoleic acid was the most prominent fatty acid and was observed at between 68.10 and 78.18 g/100 g. In another study, the major fatty acid of seed oils was linoleic acid (53.6% to 69.6%), followed by oleic (16.2% to 31.2%), palmitic (6.9% to 12.9%) and stearic (1.44% to 4.69%) acids. The oils of Red Globe and Kalecik Karası seeds contained high levels of linoleic acid, with values of 68.8% and 63.7% respectively (Sabır *et al*., 2012). The oil contents of the grape seeds ranged from 12.09% (Kalecik karası) to 17.08% (Gamay), and the main fatty acids were linoleic (66.69% to 72.50%), oleic (13.13% to 18.50%) and palmitic acids (6.87% to 9.56%) (Yalcın *et al*., 2016).

The tocopherol contents of seed oils are given in Table 4. The α-tocopherol content of seed oils ranged from 0.137 mg/g (Red Globe) to 0.231 mg/g (Çavuş) ($p < 0.05$). The amounts of β- and γ-tocopherols were 0.00 to 0.220 mg/g and 0.110 mg/g to 0.185 mg/g respectively. Çavuş seed oil had the highest α-tocopherol content, while the highest β- and γ-tocopherol contents were determined in the Trakya ilkeren variety. According to the study of Sabir *et al*. (2012), the α-tocopherol content of seed oils was determined at between 260.5 and 153.1 mg/kg. The α-, β-, γ- and δ-tocopherol contents were 142.2, 1.29, 25.5 and 0.87 mg/kg for Red Globe seed oil, and 172.4, 0.70, 27.5 and 0.63 mg/kg for Kalecik karası respectively. It was reported that grape seed oils are a good source of α-tocopherol, with the value ranging between 85.5 and 244.0 mg/kg (Fernandes *et al*., 2013).

The mineral contents of the seedless parts and seeds of grapes are given in Table 5. The major minerals of the seedless parts of grapes were potassium (K, 1772.615 to 4569.411 mg/kg), calcium (Ca, 187.351 to 498.814 mg/kg), phosphorus (P, 145.038 to 286.042 mg/kg), sulphur (S,

TABLE 5
Fatty acid composition and tocopherol content of grape seed oils

Fatty acids (%)	Red Globe	Çınarlı karası	Çavuş	Kalecik karası	Yapıncak	Yalova incisi	Trakya ilkeren
Palmitic	7.865 ± 0.112*c	8.821 ± 0.030b	7.176 ± 0.093c	10.358 ± 0.163a	7.651 ± 0.144c	7.585 ± 0.188c	8.246 ± 0.404b
Stearic	3.053 ± 0.002b**	3.896 ± 0.005b	2.983 ± 0.020c	3.471 ± 0.017b	3.453 ± 0.033b	3.753 ± 0.053b	5.073 ± 0.234
Oleic	17.402 ± 0.010b	17.698 ± 0.004b	10.746 ± 0.055	17.795 ± 0.056b	19.660 ± 0.153a	13.311 ± 0.137c	13.192 ± 0.602c
Linoleic	70.024 ± 0.012d	68.237 ± 0.038e	77.359 ± 0.007a	66.942 ± 0.153g	67.425 ± 0.047f	74.077 ± 0.011b	72.491 ± 0.062c
Arachidic	0.414 ± 0.284a	0.183 ± 0.001c	0.158 ± 0.004d	0.127 ± 0.003f	0.153 ± 0.008d	0.140 ± 0.006e	0.214 ± 0.001b
Linolenic	0.125 ± 0.020d	0.133 ± 0.001c	0.120 ± 0.012e	0.248 ± 0.121a	0.171 ± 0.013b	0.132 ± 0.015c	0.131 ± 0.005c
Tocopherols (mg/g)	Red Globe	Çınarlı karası	Çavuş	Kalecik karası	Yapıncak	Yalova incisi	Trakya ilkeren
α-Tocopherol	0.137 ± 0.030f	0.142 ± 0.055e	0.231 ± 1.108a	-	0.157 ± 0.013d	0.188 ± 0.019b	0.173 ± 0.033c
β-Tocopherol	0.119 ± 0.006f	0.140 ± 0.143d	-***	0.165 ± 0.110b	0.154 ± 0.030c	0.123 ± 0.058e	0.220 ± 0.242a
γ-Tocopherol	0.112 ± 0.002e	0.122 ± 0.220d	0.122 ± 0.034d	0.142 ± 0.037b	0.139 ± 0.038c	0.110 ± 0.002f	0.185 ± 0.066a

* mean ± standard devation; ** means in the same row with the same letters are not significantly different ($p < 0.05$); *** not detected

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TABLE 6
Mineral contents of seedless parts and seeds of grapes (mg/kg)

Grape pulp+skin	Al	Mo	Ca	B	Cd	Cu	Fe	K
Red Globe	1.048 ± 0.037*d	-***	241.653 ± 3.51c	4.357 ± 0.028a	0.052 ± 0.017c	0.822 ± 0.065b	8.825 ± 0.058e	2090.084 ± 11.00e
Çınarlı karası	1.141 ± 0.021d**	0.133 ± 0.020c	498.814 ± 10.52a	4.841 ± 0.056a	0.031 ± 0.008e	0.88 ± 0.070b	6.164 ± 4.584f	2038.985 ± 6.00e
Çavuş	0.861 ± 0.055e	0.016 ± 0.006	188.596 ± 3.08e	1.854 ± 0.061	0.04 ± 0.010d	0.567 ± 0.060d	5.832 ± 0.040g	1772.615 ± 8.03f
Kalecik karası	7.14 ± 0.020b	0.232 ± 0.023b	187.351 ± 7.03e	3.064 ± 0.061bc	0.062 ± 0.026b	0.388 ± 0.040e	11.152 ± 0.056c	2944.953 ± 6.00b
Yapıncak	2.748 ± 0.181c	-	188.555 ± 1.26e	3.243 ± 0.042b	0.043 ± 0.007d	0.732 ± 0.055c	10.273 ± 0.086d	2135.029 ± 6.00d
Yalova incisi	2.861 ± 0.183c	0.349 ± 0.028a	303.016 ± 6.24b	4.381 ± 0.040a	0.069 ± 0.012a	1.27 ± 0.046a	12.576 ± 0.061b	2476.507 ± 8.67c
Trakya ilkeren	13.55 ± 0.061a	0.022 ± 0.012d	193.147 ± 6.32d	4.266 ± 0.025a	0.033 ± 0.007e	1.238 ± 0.045a	24.205 ± 0.041a	4569.411 ± 8.50a

	Mg	Mn	Na	P	Pb	S	Zn
Red Globe	84.105 ± 6.197d	0.153 ± 0.026d	112.359 ± 4.507b	167.461 ± 4.501e	-	155.057 ± 6.001f	2.771 ± 0.080b
Çınarlı karası	98.291 ± 5.512c	0.014 ± 0.005f	77.207 ± 4.529e	145.038 ± 6.00g	0.277 ± 0.04	154.673 ± 3.513f	7.165 ± 0.090a
Çavuş	82.317 ± 4.511e	0.218 ± 0.040b	103.195 ± 5.525c	183.158 ± 6.006c	0.275 ± 0.045	173.462 ± 5.500d	1.244 ± 0.065e
Kalecik karası	84.222 ± 6.518d	0.174 ± 0.055e	111.666 ± 4.726b	170.72 ± 6.02d	-	177.2 ± 4.530c	1.827 ± 0.060c
Yapıncak	77.638 ± 4.049f	0.014 ± 0.005f	79.507 ± 4.090d	151.499 ± 5.50f	-	161.658 ± 6.029e	1.524 ± 0.075d
Yalova incisi	133.09 ± 6.002a	0.249 ± 0.030b	62.485 ± 4.50f	222.365 ± 4.506b	-	211.982 ± 3.000a	1.634 ± 0.045c
Trakya ilkeren	106.43 ± 2.900b	0.337 ± 0.031a	133.401 ± 4.06a	286.042 ± 5.557a	-	192.423 ± 4.502b	1.768 ± 0.060c

Grape seeds	Al	Mo	Ca	B	Cd	Cu	Fe	K
Red Globe	63.99 ± 1.942a	0.148 ± 0.001d	6500.015 ± 134.413a	9.077 ± 0.318b	0.052 ± 0.001b	7.131 ± 0.145b	27.463 ± 0.833c	3537.591 ± 35.733f
Çınarlı karası	0.346 ± 0.015e	0.209 ± 0.008c	4435.351 ± 25.142d	8.321 ± 0.184c	0.027 ± 0.006d	6.546 ± 0.029c	21.531 ± 0.126e	4857.359 ± 7.087d
Çavuş	1.446 ± 0.029d	3.148 ± 0.027a	6189.102 ± 6.010b	8.497 ± 0.016c	0.06 ± 0.027a	7.357 ± 0.045b	26.482 ± 0.065d	4936.271 ± 14.505c
Kalecik karası	1.348 ± 0.024d	0.439 ± 0.038b	3567.776 ± 26.430e	8.203 ± 0.045c	0.017 ± 0.011f	6.726 ± 0.045c	30.887 ± 0.100b	5555.265 ± 4.026b
Yapıncak	1.832 ± 0.016c	0.208 ± 0.007c	5320.186 ± 49.863c	5.585 ± 0.035e	0.035 ± 0.013c	6.764 ± 0.055c	21.043 ± 0.133e	4194.617 ± 8.071e
Yalova incisi	0.241 ± 0.028e	0.167 ± 0.016d	5993.295 ± 5.670c	14.925 ± 0.055a	0.036 ± 0.012c	9.061 ± 0.095a	27.077 ± 0.115c	8042.746 ± 7.384a
Trakya ilkeren	2.198 ± 0.016b	0.174 ± 0.022d	5329.002 ± 18.734c	7.547 ± 0.050d	0.023 ± 0.006e	7.574 ± 0.080b	31.167 ± 0.112a	5092.413 ± 6.502c

	Mg	Mn	Na	P	Pb	S	Zn
Red Globe	1403.564 ± 35.440b	15.678 ± 0.236a	69.172 ± 0.284e	2395.649 ± 25.93a	0.07 ± 0.010c	1039.52 ± 4.901a	12.66 ± 0.251b
Çınarlı karası	1157.78 ± 11.099d	10.604 ± 0.388d	247.756 ± 11.081a	1921.268 ± 10.51d	-	1007.501 ± 5.68b	8.856 ± 0.090f
Çavuş	1233.787 ± 6.153c	10.731 ± 0.092d	56.268 ± 1.615f	1907.32 ± 7.507d	-	937.535 ± 6.711c	7.319 ± 0.061g
Kalecik karası	1121.404 ± 11.501d	10.364 ± 0.251d	234.563 ± 6.048b	2307.98 ± 9.00a	0.369 ± 0.055b	1024.356 ± 7.088a	14.294 ± 0.085a
Yapıncak	1194.747 ± 7.805d	11.471 ± 0.105c	178.089 ± 7.002c	2205.186 ± 8.799b	-	1007.281 ± 5.664b	9.142 ± 0.063e
Yalova incisi	1516.708 ± 8.117a	10.642 ± 0.105d	109.482 ± 5.293d	2109.14 ± 8.004c	-	1025.548 ± 13.60a	11.108 ± 0.081c
Trakya ilkeren	1435.561 ± 12.731b	12.272 ± 0.071b	179.791 ± 4.664	1953.38 ± 10.28d	0.718 ± 0.056a	818.12 ± 4.548d	10.405 ± 0.162d

* mean ± standard deviation; ** means in the same row with the same letters are not significantly different ($p < 0.05$); *** not detected

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154.673 to 211.982 mg/kg), magnesium (Mg, 77.638 to 133.086 mg/kg) and sodium (Na, 62.485 to 133.401 mg/kg) ($p < 0.05$). Other minerals, e.g. Fe, Al, B, Cu, Cd, Mo, Zn, Mn and Pb were found at lower levels. The highest potassium content was observed in the Trakya ilkeren variety (4 569.411 mg/kg), followed by Kalecik karası (2 944.953 mg/kg). Çınarlı karası had the highest Ca content (498.814 mg/kg), while Yalova incisi had the maximum Mg content (133.086 mg/kg). The greatest amounts of P, S and Na were determined in Trakya ilkeren (286.042 mg/kg) and Yalova incisi (211.982 mg/kg). Moreover, Trakya ilkeren was the variety with the highest Fe (24.205 mg/kg) and Al (13.550 mg/kg) contents. Cd shows a toxic effect and was found below 1%, while Ni and Cr were not detected in any of the varieties.

Macro-elements such as K, Ca, P, Mg, S and Na in the grape seeds varied between 3 537.591 and 8 042.746 mg/kg, 3 567.776 and 6 500.015 mg/kg, 1 907.320 and 2 395.649 mg/kg, 1 121.404 and 1 516.708 mg/kg, 818.120 and 1 039.520 mg/kg, and 56.268 and 247.756 mg/kg respectively ($p < 0.05$). The mineral contents of the grape seeds were higher than in the rest of the grape. The highest Ca, P and S were observed in Red Globe seeds, with values of 6 500.015 mg/kg, 2 395.649 mg/kg and 1 039.520 mg/kg respectively ($p < 0.05$). The seeds of Yalova incisi had the highest K (8 042.746 mg/kg) and Mg (1 516.708 mg/kg) contents. In addition, grape seeds did not contain Ni and Cr, although Cd was found at levels below 1%, similar to in the grape pulp+skin. In the experiments of Aykut (2002), the mineral composition of the different grape varieties (Sultani, Hamburg Misketi and Alicante Bouschet) was as follows: K (1 255 to 1 750 mg/kg), Ca (24 to 34 mg/kg), Mg (53.75 to 102.5 mg/kg), Na (31.32 to 53.30 mg/kg), P (97.20 to 168.2 mg/kg), Fe (0.71 to 1.88 mg/kg), Cu (0.8 to 2.5 mg/kg), Zn (0.35 to 0.79 mg/kg) and Mn (0.49 to 1.45 mg/kg).

Some differences were observed in the results when compared with the literature. These differences could probably be due to variety, cultural factors and analytical conditions. Grape pulp and seeds are very important for human health because of phenolics, fatty acids, tocopherols, minerals and bioactive properties.

CONCLUSIONS

Grape and grape products are important foods due to their richness in bioactive components. Many studies emphasise the importance of grapes and their products, which are sources of natural antioxidants, for health. Therefore, it is necessary to investigate the physicochemical properties and bioactive components of local varieties as well as international varieties. For this purpose, this study was performed to determine the bioactive compounds of both the seeds and the seedless parts of different grape varieties. In addition, the oil content, fatty acid composition and tocopherol content of seeds were investigated. Of all the grape varieties, Red Globe and Trakya Ilkeren have the highest antioxidant activities (90.948%, 90.735%), and total phenolic (170.313 mg/100 g, 199.063 mg/100 g) and total flavonoid (6.810 mg/g, 6.744 mg/g) contents. The seedless parts of the grapes (pulp+skin) exhibited lower phenolic, flavonoid and mineral contents than the seeds of the grapes. Furthermore, the main fatty acid was linoleic acid (66.942 to 77.359%), and α-tocopherol was the most abundant isomer (0.137 to 0.231 mg/g). The mineral contents of the grape varieties also were compared. Grapes are a significant source of K, Ca and P.

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