



Article

Cultivar Differences on Nutraceuticals of Grape Juices and Seeds

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Abstract: In this study, nutraceutical properties of fruit juice and seeds, which are important for human health, of green (Kabarcik, Cavus), red (red Globe) and black (Honusu, Yildiz, Yediveren and Helvani) skin colored grape cultivars grown in same ecological conditions were investigated. Harvest period, number of seeds, cluster form, cluster weight, berry weight, berry color and usage area were determined as morphological parameters. The Folin-Ciocalteu method was used for total phenol content analysis. The total antioxidant status of juices and seeds of grape cultivars have been determined by 2,2-diphenyl-1-picryl-hydrazyl-hydrate (DPPH), ferric reducing antioxidant power (FRAP) and trolox equivalent antioxidant capacity (TEAC) assays. Seeds of all grape cultivars exhibited higher nutraceuticals than fruit juices. Total phenolic content of seed samples was found to be quite variable in range of 1.90 mg GAE/g (cv. Yildiz)–3.46 mg GAE/g (cv. Honusu) fresh weight base (FW), indicating 2 folds of differences between green and black grape cultivars. Green and black cultivars also showed the lowest and the highest total phenolic content in juices between 1.69 (cv. Yediveren) and 2.45 (cv. Honusu mg GAE/g FW). Seeds and fruit juices of all cultivars analyzed showed high antioxidant capacity and total phenol content. Of all different colored cultivars, black peel-colored cultivars had the highest values and combined better morphological and nutraceutical traits with an excellent berry qualitative profile for cv. Honusu and Helvani.

Keywords: grape; bioactive content; juice; seed

1. Introduction

Scientific studies on horticultural crops, including fruits, vegetables and grapes, in recent years have revealed that these products have very important functions in terms of human health, and they contain especially high levels of biological active components. Biological activity analyses were previously performed on the whole fruits, berries, etc., but more recent analyses have focused on different parts (seed, juice, flesh, peel) to find better parts for human nutrition and health. The enormous diversity among different cultivars offers great opportunity for researchers to find better ones for both human nutrition and pharmaceutical industry and for future cross-breeding activities to use them as parents [1–5].

Turkey is one of the most important countries in the world in terms of viticulture. Nine agricultural regions in the country have traditional grape cultivars with their unique cultivation styles. The biodiversity on *Vitis* is very high in Turkey, and the other countries in Europe (Italy, France Spain) historically have high biodiversity in *Vitis vinifera*.

It is very difficult to find such a diversity for viticulture in grapevine producing in other countries around the world. On the other hand, the production of fruit juice and molasses are the especially prominent features in terms of viticulture in Turkey. In addition, many traditional products (köfter, sucuk, pestil, etc.) obtained from grape berries make the country quite different in terms of grape growing [6–9]. In Turkey, approximately 4000 thousand tons of grapes are produced from 525 thousand hectares of vineyard area. In Turkey, the produced grapes are equally (30–32%) consumed as fresh, dried or processed into molasses, vinegar, jam, juices or cider, and only 3% of total production is processed into wine. The dried grapes are mostly obtained from seedless cultivars because around 30% production consists of seedless grape cultivars which are more common in the Aegean Region. However, the other regions produce mostly table grapes in particular in the Marmara and Mediterranean regions [7,8].

Grapes are one of the most cultivated fruits in the world with around 80 million annual production, and 85.0% of grapes are used in wine production [10]. This huge amount of berries generating large waste materials includes seeds. More recently, grape seeds have been heavily involved in the markets as dietary supplements. The process to obtain grape seed products requires removing, drying, and pulverizing of seeds. Studies showed that seeds of grape berries are rich in antioxidant substances, including phenolic acids, anthocyanins and flavonoids. Thus, grape seeds are accepted as one of the most important natural antioxidant sources and may have helped to protect humans against oxidative stress, tissue damage, and inflammation [11–13]. Grapes are very popular among consumers due to their unique taste, texture, flavor and aroma. It is morphologically one of the richest groups among horticultural crops and exhibits yellow, green, pink, red, purple and black colored berries. The flesh part of grape berries includes water, carbohydrates and vitamins. In addition, it includes some important biological active components (vitamin C, anthocyanins and phenolics), depending on the cultivar and climatic and cultivation conditions. Due to the functional properties of grape berries and seeds, there is an increased demand for grapes and grape products. The studies revealed that grape berries, including seeds, have numerous beneficial substances that are important for human health in terms of preventing diseases, such as cancer, and other degenerative diseases [14–18].

In Turkey, grape juice (Şıra in Turkish) is very important and has high economic value. It has traditionally been widely used by consumers. Most of the traditional grape products, such as pekmez, pestil and köfter. are obtained by using grape juice in Turkey. There has been an increasing interest in grape seeds recently. Both juices and seeds are accepted as natural antioxidants. However, the issue of dealing with the analysis and comparison of antioxidants in the juices and seeds of *Vitis vinifera* L. cultivars has not been sufficiently studied yet. In the relevant literature, peel, pulp and seed samples of grape cultivars were often compared with each other for total phenolic content and antioxidant activity, and the comparative studies including seeds and juices of different grape cultivars seem to be very rare. Juices and seeds of these seven cultivars have not been compared with each other previously. In this study, three antioxidant determining methods were used to compare seeds and juices. The results in the current study may contribute to determine which grape are is the best cultivar and plant organ to benefit from the maximum pharmacological effects.

2. Materials and Methods

2.1. Plant Material, Sampling and Location

A total of seven grape cultivars with diverse skin color, including green (Kabarcik, Cavus), red (red Globe) and black (Honusu, Yıldiz, Yediveren and Helvani), were harvested from vineyards of Kahramanmaras of Turkey in the 2019 growing season at the commercial

harvest period. The sampling vineyard is 11 years old with a trellis wire training system. Grapevine plants are grown in same vineyards and standard cultural and technical practices (irrigation, fertilization, pruning, pest and disease control) were applied to them. Samples were taken to Laboratories of Cukurova University Agricultural Faculty Department of Horticulture on the same day with frigorific vehicles (+4 °C). The whole berries were used to obtain juice. The removed seeds were grounded by a coffee grinder, whereas whole berry parts were chopped by a blender at room temperature [5].

2.2. Morphological Traits

Morphological traits are important for classification of grape cultivars and done according to OIV requirements. In the full ripening stage of seven cultivars, a representative sample of 4 kg was obtained per cultivar and as morphological parameters, harvest period, number of seed, cluster weight, cluster form, berry weight and berry color were determined. Fifty berries randomly taken from the middle part of the cluster per cultivar were used to determine the berry weight and peel color [5–7,19]. Fruit weight was measured using 0.01 sensitivity electronic balance (Model SPB31, Scaltec Company, Göttingen, Germany).

2.3. Extraction

The harvested berries and seeds belong to seven grape cultivars subjected to ethanolic extraction. For each cultivar, 1 g of sample was used for extraction. To do this, samples were kept for 2 h in 20 mL of 80% ethanol and centrifuged at $4000 \times g$ for 10 min. [5]. Obtained supernatant was used for DPPH (2,2-diphenyl-1-picryl-hydrazyl-hydrate), FRAP (ferric reducing antioxidant power) and Trolox equivalent antioxidant capacity (TEAC) antioxidant determined assays and for total polyphenol content.

All the reagents and chemicals used in the experiments were of analytical grade. The chemicals DPPH, TPTZ (2,4,6-tripyridyl-S-triazine), ABTS, 2-thiobarbituric acid, ascorbic acid and Folin-Ciocalteu reagent were obtained from Sigma Chemical Co. in the U.S. 2,2'-Azobis (2-amidinopropane), dihydrochloride (AAPH), and Trolox (6-hydroxy-2,5,7,8-tetramethyl chromane 2-carboxylic acid) were obtained from Aldrich Chemicals Co. in the U.S. All other chemicals used were obtained from the local suppliers.

2.4. Total Phenol Folin-Ciocalteu Assay

The well-known Folin-Ciocalteu method was used for total phenolic content analysis according to Krawitzky [20] using a UV spectrophotometer (Thermo Fisher Scientific, Waltham, MA, USA). The absorbance was measured at 760 nm against a blank. The results are expressed as mg gallic acid equivalent (GAE)/g fresh weight (FW).

2.5. Total Antioxidant Capacity Measurement

2.5.1. DPPH Method

The DPPH activity of samples was done according to Wang et al. [21] spectrophotometrically. In the assay, 0.06 μM of ethanolic DPPH was prepared and 1950 μL of DPPH• was added to 50 μL of sample. Approximately 1 min of shaking was applied on samples and later kept in the dark for 30 min at room temperature. Absorbance was measured against the blank reagent at 515 nm. DPPH activity (radical scavenging activity) was calculated according to the following formula:

$$\% \text{Inhibition} = (1 - ((\text{Abs sample})/(\text{Abs control})) \times 100 \quad (1)$$

2.5.2. FRAP Method

According to [22], the FRAP analysis was conducted spectrophotometrically. Briefly, FRAP reagent at a volume ratio of 10:1:2, sodium acetate-acetic acid buffer (300 mmol/L), TPTZ (10 mmol/L) and ferric chloride solution (20 mmol/L), were prepared. The mixture was left in a water bath at 37 °C before use. Later, 100 microliters of diluted samples were mixed with 3 mL of FRAP reagent. After incubation for 4 min, the absorbance of the

mixture at 593 nm was determined. The results were expressed as $\mu\text{mol Fe (II)}/\text{g FW}$ of grape juices and seeds.

2.5.3. TEAC Method

For TEAC assay, the spectrophotometrical method (ABTS free radical scavenging activity) [23] was used. Firstly, ABTS stock solution was prepared (ABTS (7 mmol/L) plus potassium persulfate (2.45 mmol/L) in a volume ratio of 1:1). After that, stock solution was incubated in dark at room temperature for at least 16 h and stored less than 2 d before use. The solution was diluted with ethanol to an absorbance of 0.710 ± 0.050 at 734 nm. One hundred microliters of diluted sample were mixed with 3.8 mL of ABTS reagent at room temperature, and 6 min. later the absorbance of the mixture was determined at 734 nm. The results were expressed as $\mu\text{mol Trolox equivalent}/\text{g FW}$ of grape juices and seeds.

2.6. Statistical Analysis

Four replications including 10 samples per replicate were used. For statistical analysis, Windows SPSS 20 was used. The ANOVA variance analysis and Duncan multiple comparison test ($p < 0.05$) was used to determine the differences between the means. The Pearson correlation analysis was used between antioxidant activity and total phenolic content.

3. Results and Discussion

3.1. Morphological Traits

Morphological characteristics of seven grape cultivars grown at same location in Kahramanmaraş province in Turkey are given in Table 1. A great variability on most of the morphological traits is evident among seven cultivars.

Table 1. Some morphological traits of used grape cultivars.

Cultivars	Harvest Period	Number of Seed	Cluster Form	Cluster Weight (g)	Berry Weight (g)	Berry Color	Usage
Cavus	Mid-early	1–2	Winged cylindrical	525 ± 17.1	5.86 ± 0.4	Green	White table
Helvani	Mid-late	2–3	Winged cylindrical	1150 ± 43.1	3.82 ± 0.3	Black	Colored Table
Honusu	Late	1–2	Irregular winged conical	587 ± 18.0	6.95 ± 0.7	Black	Colored table
Kabarcik	Mid-season	1–3	Winged conical	433 ± 15.2	3.85 ± 0.3	Green	White wine and Table
Red Globe	Mid-season	3–4	Conical	923 ± 31.1	3.96 ± 0.2	Red	Colored table
Yediveren	Mid-late	1–2	Winged conical	437 ± 14.7	4.42 ± 0.3	Black	Colored table
Yildiz	Mid-season	2–3	Shouldered conical	440 ± 13.9	4.03 ± 0.4	Black	White table

Considering all cultivars used in the experiment, the earlier harvested cultivar was cv. Cavus, followed by Kabarcik, Red Globe and Yildiz. Helvani and Yediveren cultivars were harvested in the mid–late period. Honusu was the latest harvested cultivar (Table 1).

Cavus, Honusu and Yediveren cultivars had 1–2 seed numbers per berry and Kabarcik had 1–3 seeds per berry, Helvani and Yildiz had 2–3 seeds per berry and Red Globe had 3–4 seeds per berry (Table 1).

Cluster form is also quite variable among cultivars. Cavus and Helvani had winged cylindrical, Kabarcik and Yediveren had winged conical, Honusu had irregular winged conical, Red Globe had conical and Yildiz had shouldered conical cluster form (Table 1). Average cluster weight of cultivars were found between 433 g (cv. Kabarcik) and 1150 g (cv. Helvani) indicating the studied grape cultivars genetically were very diverse from each other (Table 1). The highest berry weight was obtained from Honusu cultivar as 6.95 g while the lowest berry weight was seen in cv. Helvani as 3.82 g (Table 1). Two cultivars

(Cavus and Kabarcik) had green berry color, Red Globe had red berry color and the rest of the cultivars had black berry color.

The low gene flow could be a reason for high dissimilarity between cultivars [24]. Another reason could be natural hybridization and human selection [24]. In the literature, there were several reports indicating a great morphological variability among grape cultivars. For example, Ekhvaia and Akhalkatsi [25], Leao et al. [26], Khadivi-Khub et al. [27] and Abiri et al. [28] reported a high variability in morphological features of grape cultivars including particular cluster weight, berry color and berry weight.

The majority of the cultivars were used for table consuming, and only cv. Kabarcik was processed into white wine in the growing region. All cultivars had cluster weight over 300 g and were classified as big according to [19]. Kok et al. [29] found cluster weight in grape cultivars between 232 g (cv. Balbal)b and 560 g (cv. Antep Karasi). Dilli and Kader [8] reported that the grape cultivars grown in Turkey were harvested in the early, mid and late periods. They also reported that Red Globe (1000 g) and Italia (500–800 g) gave the bigger clusters among searched cultivars.

Leao et al. [26] suggested that cluster and berry size, and berry color could be useful to differentiate cultivars. In grapes, because of sufficient cultivars and easy application, morphological traits might be appropriate for classification and cross-breeding the morphological traits. Grape breeders have been using morphological markers in the cross-breeding programs successfully [30–33]. In fact, humans have successfully used various morphological features to compare cultivars.

In countries such as Turkey, Egypt, Iran, Turkmenistan and Uzbekistan, table grape cultivars have been used as fresh consumption and raisins. In fact, for table grape cultivars, the most desirable traits were thickness of berry peel, lesser number of seeds, bigger cluster and berries. Uniform color formation and suitability to transportation are also desirable traits [8]. The results of the present study are in accordance with studies stated above. Genetic background, environmental conditions and other factors could affect this variation [25–29].

3.2. Total Phenol Content

The total phenol content quantification in both juice and seed of green, red and black grape cultivars is shown in Table 2. Significant differences are presented among cultivars for total phenol content of both juices and seeds ($p < 0.05$) (Table 2).

Table 2. Total phenolic content and antioxidant activity of grape juices and seeds.

Cultivars	Total Phenolic (mg GAE per g fresh weight)		FRAP ($\mu\text{mol Fe (II)/g FW}$)		DPPH (%)		TEAC ($\mu\text{mol Trolox/g FW}$)	
	Juice	Seed	Juice	Seed	Juice	Seed	Juice	Seed
Cavus	1.81 \pm 0.11de	2.13 \pm 0.12e	40.54 \pm 1.1c	860 \pm 17b	73.10 \pm 1.9d	76.22 \pm 1.1c	2.35 \pm 0.19bc	15.23 \pm 0.9bc
Helvani	2.24 \pm 0.13b	3.18 \pm 0.17b	44.24 \pm 1.3b	1002 \pm 13a	80.84 \pm 2.1b	85.44 \pm 1.4a	2.52 \pm 0.11ab	15.99 \pm 0.8ab
Honusu	2.45 \pm 0.13a	3.46 \pm 0.15a	52.15 \pm 1.4a	1013 \pm 18a	82.19 \pm 2.0a	86.77 \pm 1.2a	2.73 \pm 0.15a	18.12 \pm 1.1a
Kabarcik	2.03 \pm 0.10c	3.01 \pm 0.09c	32.02 \pm 0.9d	726 \pm 12d	76.12 \pm 1.6c	79.68 \pm 1.0bc	2.19 \pm 0.14bc	15.01 \pm 0.6bc
Red Globe	1.85 \pm 0.10d	2.45 \pm 0.12d	32.34 \pm 1.0d	795 \pm 20c	74.30 \pm 1.7cd	81.10 \pm 1.3b	2.43 \pm 0.19b	15.56 \pm 0.6b
Yediveren	1.69 \pm 0.09e	1.91 \pm 0.11f	28.66 \pm 0.7e	686 \pm 15e	70.25 \pm 1.5e	79.22 \pm 1.2bc	2.04 \pm 0.11c	12.89 \pm 0.5c
Yildiz	1.76 \pm 0.10de	1.90 \pm 0.08f	28.02 \pm 1.0de	642 \pm 10f	71.95 \pm 1.6de	73.56 \pm 1.4d	2.17 \pm 0.13bc	15.30 \pm 0.7bc

Different letters in the same columns indicate significant differences ($p < 0.05$).

The highest total phenol content was determined in seeds of Honusu cultivar as 3.46 mg GAE/g FW, and followed by Helvani (3.18 mg GAE/g FW), Kabarcik (3.01 mg GAE/g FW), Red Globe (2.45 mg GAE/g FW), Cavus (2.13 mg GAE/g FW), Yediveren (1.91 mg GAE/g FW) and Yildiz (1.90 mg GAE/g FW), respectively. In terms of juices, the highest total phenol content was observed in cv. Honusu as 2.45 mg GAE/g FW and followed by Helvani (2.24 mg GAE/g FW), Kabarcik (2.03 mg GAE/g FW), Red Globe

(1.85 mg GAE/g FW), Cavus (1.81 mg GAE/g FW), Yildiz (1.76 mg GAE/g FW) and Yediveren 1.69 mg GAE/g FW), respectively (Table 2).

The results confirming that the total phenol content of grape berries vary according to the cultivars [29,34,35], the plant juices and seeds reflect different values and seeds were found to be a richer source of total phenol content than juices of all seven used cultivars (Table 2). In the literature, there are many reports about total phenol content of grape peel and pulp but limited a number of studies have been conducted on seeds and in particular on juices, which makes it possible to notice that the grape cultivars and the plant juice and seeds are factors that exert a great influence on the content of total phenol content. Previous studies conducted on different parts of the world indicated that whole berries of different grape cultivars with diverse colors had total phenol content from 0.29–4.68 mg GAE/g FW [29,34–39]

Shen et al. [40] found that total phenolic content was the highest in seeds, followed by peel, while total phenol content in pulps was less than that of peel and seeds in grapes. Yilmaz et al. [41] observed that the total phenol content of the pulps of grape cultivars was remarkably lower than that of seeds and peel. The results of total phenol content values were consistent with previous studies. Previous studies also indicated that accumulation of the phenolic compounds in grapes vary considerably with respect to the cultivars [42–44]. However, along with genetic background, growing location, climate, soil, temperature, cultural practices, ripening stage, plant tissue and training system could be attributed to the total phenol content in grape berries. Phenolic compounds are known to directly affect the quality characteristics of grape berries, such as color and taste, and significantly contribute to the antioxidant characteristics of grape berries [45].

3.3. Total Antioxidant Capacity

Horticultural crops, such as fruits, grapes and vegetables, include a large number of phytochemicals that contribute to the antioxidant characteristics of these crops. For each laboratory, it is not possible to determine each antioxidant component. Therefore, total antioxidant capacity of horticultural crops and the other plants have been used widely. Among these methods DPPH, FRAP and ABTS (TEAC) have gained more popularity, and are frequently used to determine the total antioxidant capacity of different biological materials [46]. The total antioxidant capacity of juice and seed of seven grape cultivars were determined by DPPH, FRAP and TEAC assays, and the results are given in Table 2.

3.3.1. DPPH Assay

The obtained results in this study clearly indicated that the grape juice and seed extracts belonging to different cultivars significantly ($p < 0.05$) inhibited DPPH free radicals' generation (Table 2). In DPPH% method, grape juice extracts showed inhibition value from 70.25% (cv. Yediveren) to 82.19% (cv. Honusu). The seed extracts for all cultivars demonstrated higher inhibition values than juices that were in range of 73.56% (cv. Yildiz) to 86.77% (cv. Honusu) (Table 2). Previous studies indicated that in general, grape seed extracts were more effective DPPH radical scavengers than pulp (flesh). Sridhar and Charles [47] used Kyoho grape cultivar (*Vitis labrusca*) and reported that grape peel was the highest DPPH inhibition (95.71%), and followed by seed (92.02%) and flesh (57.78%), respectively. Choi et al. [48] reported higher DPPH radical scavenging activity for Campbell Early grape seed extracts than flesh in Korea. Nile et al. [49] studied a large number of grape cultivars belonging to *Vitis vinifera*, *Vitis labrusca* and *Vitis hybrida* and reported DPPH values from 35.4% (Campbell Early) to 84.5% (Hongiseul) for grape pulps. Fahmi et al. [50] reported that grape cultivar shows DPPH radical scavenging activity between 47 and 60%. Farhadi et al. [51] reported that six grape seed and pulp samples had DPPH inhibition from 88.10–94.46% and 88.16–94.68%, respectively. A similarity between pulp and seed samples was revealed. Mandić et al. [52] found that phenolic compounds and antioxidant capacity of grape berries were strongly affected by species, cultivars, growing condition and ripening time.

3.3.2. FRAP Assay

The results of the FRAP method related to antioxidant capacity of juices and seeds of seven grape cultivars are given in Table 2. The results demonstrated that there was a statistically significant variation ($p < 0.05$) between the antioxidant capacity of juices and seeds depending on the cultivar. Considering the effect of cultivars on total antioxidant capacity values determined by FRAP assay, the juices and seed extracts of Honusu (52.15 $\mu\text{mol Fe (II)}/\text{g FW}$ for seed extract and 1013 $\mu\text{mol Fe (II)}/\text{g FW}$ for juice extract) and Helvani (44.24 $\mu\text{mol Fe (II)}/\text{g FW}$ for juice extract and 1002 $\mu\text{mol Fe (II)}/\text{g FW}$ for seed extract) showed higher FRAP values (Table 2). Seed extracts of all cultivars had higher antioxidant capacity than juice extract. Liu et al. [39] used 30 grape cultivars to determine the antioxidant capacity using FRAP assay and reported that the FRAP values in pulp were in the range of 1.29–11.76 $\mu\text{mol Fe (II)}/\text{g FW}$, respectively. They found that the highest FRAP values were obtained from Pearl Black Grape (11.76 $\mu\text{mol Fe (II)}/\text{g FW}$). The results obtained from this study were in agreement with those of a previous study that used the FRAP analysis of four grape cultivars (1.73 to 10.12 $\mu\text{mol Fe (II)}/\text{g FW}$ in pulp) according to [53]. Fu et al. [54] found that FRAP values of 56 wild edible fruits are variable and varied between 0.67 and 143 $\mu\text{mol Fe (II)}/\text{g FW}$ in pulp. Sochorova et al. [55] reported that grape seeds of different cultivars had high content of antioxidant activity and showed differences in antioxidant activity. Shen et al. [40] reported that the seeds of grape cultivars had higher antioxidant activity than pulp by FRAP assay. Ruiz-Torrallba et al. [35] reported FRAP values were between 14.58 and 15.23 $\mu\text{mol Fe (II)}/\text{g FW}$ in pulp of red and white grape cultivars.

3.3.3. TEAC Assay

The TEAC values in seven grape cultivars' juice and seed samples are presented in Table 2. Statistically significant differences among cultivars for both juice and seed extracts are evident ($p < 0.05$). The TEAC values of juice and seed samples varied from 2.04 (Yediveren) to 2.73 $\mu\text{mol Trolox}/\text{g FW}$ (Honusu) and 12.89 (Yediveren) to 18.12 $\mu\text{mol Trolox}/\text{g FW}$ (Honusu). High TEAC values were also found in juice and seed extracts from Helvani (2.52 $\mu\text{mol Trolox}/\text{g FW}$ for juices and 15.99 $\mu\text{mol Trolox}/\text{g FW}$ for seed extract) (Table 2). The cv. Yediveren showed the lowest TEAC value both in juice and seed extract (2.04 $\mu\text{mol Trolox}/\text{g FW}$ for juices and 12.89 $\mu\text{mol Trolox}/\text{g FW}$ for seed extract) among cultivars. Results indicated that of all horticultural crops, grape juice and seeds are the best sources of antioxidants. The TEAC results showed that grape seed extract had higher antioxidant capacity than juice extracts. Costa et al. [56] found high antioxidant activity on grape pulp using the TEAC method. Liu et al. [39] used the TEAC assay on pulp of 30 grape cultivars in China and showed that TEAC values were between 0.339 and 4.839 $\mu\text{mol TE}/\text{g FW}$ indicating lower values than the current study. Sochorova et al. [57] found that the grape seed extract was a rich antioxidant source due to high oligomeric and polymeric flavanols. Weidner et al. [58] used seeds of European and Japanese grapevine species (*Vitis vinifera* and *Vitis coignetiae*) and reported that seeds contained great amounts of tannins and detectable levels of catechins and *p*-coumaric, ferulic and caffeic acids. They also reported that total phenolic substances were higher than in Japanese ones. Previous studies indicated a great variability in antioxidant activity among grape berries, seeds and peels [59–64] using DPPH, FRAP and TEAC assays. Spigno et al. [62] indicated that this variation could be the result of genetic background, extraction method and antioxidant activity assay used.

The correlation between the total phenol content (TPC) with antioxidant activity (FRAP, DPPH and TEAC) of grape juices and seed extracts is shown in Table 3. According to Table 3, TPC of grape juices showed the highest correlation coefficient ($0.8 < r < 1$) with DPPH (0.89), followed by TPC and FRAP with moderate positive correlation ($0.5 < r < 0.8$) (0.72) and TPC and TEAC (0.66). For seed extracts, TPC and DPPH interaction showed the highest correlation (0.80), followed by TPC and FRAP (0.76) and TPC and TEAC (0.62). On the other hand, interactions between DPPH and FRAP were strong (0.83 in juice) and

moderate (0.78 in seed extracts). TEAC and FRAP showed moderate positive correlation (0.70 in juices and 0.52 in seeds). The interaction between TEAC and DPPH presented a moderate positive correlation as well ($0.5 < r < 0.8$) (0.74 in juice and 0.64 in seed extracts). When this correlation is considered, it can be stated that total phenol may have contributed much to the antioxidant capacity of grape juice and seeds. In fact, previous studies also indicated this fact [65–67]. In addition, Clarke et al. [68] reported high correlation of DPPH, FRAP, TEAC and TPC.

Table 3. Correlation between the TPC and antioxidant capacities by FRAP, DPPH and TEAC.

	TPC		FRAP		DPPH		TEAC	
	Juice	Seed	Juice	Seed	Juice	Seed	Juice	Seed
TPC	1.0	1.0						
FRAP	0.72 *	0.76 *	1.0	1.0				
DPPH	0.89 **	0.80 **	0.83 **	0.78 *	1.0	1.0		
TEAC	0.66 *	0.62 *	0.70 *	0.52 *	0.74 *	0.64 *	1.0	1.0

TPC: total phenol content; *: ($p < 0.05$); **: ($p < 0.01$).

4. Conclusions

In conclusion, the multi cultivar experiment presented a great diversity in terms of nutraceuticals in both seeds and juices of grape samples. In view of the total phenol content and the antioxidant properties identified in the juices and seeds, this study contributes to a better understanding of the influence of different cultivars and plant organs on nutraceuticals. The results demonstrated that the Honusu and Helvani cultivars are the most promising cultivars in terms of nutraceuticals. The current study indicated that the results of the DPPH, FRAP, TEAC and TPC assays provide essentially identical information in regard to the antioxidant capability of juice and seed extracts of grape cultivars. The results also showed how skin color affects nutraceuticals of grape berries and reveals importance of dark berry skin-colored cultivars for human health-promoting substances.

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