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Determination of Some Phytochemical Contents of Local and Standard Grape Varieties Grown in Diyarbakır Province

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Abstract

Grape (*Vitis vinifera* L.) has been the subject of many research studies because of its phenolic compounds and antioxidant properties which are known to have positive effects on health. In this research study, some phytochemical contents of local and standard grape varieties widely grown in Diyarbakır province were determined. Amount of total phenolic, total anthocyanin and total flavonoid were examined in the berry seed, berry pulp and berry skin of each variety. The statistical differences among Boğazkere, Öküzgözü and Kızıl Banki were obtained and with the addition of Şire variety all varieties were compared in terms of statistical differences.Content of total phenolic was between 389.15 mg GAE / kg and 4050.17 mg GAE / kg, while content of total anthocyanin was recorded between 25.61 mg / kg and 634.00 mg / kg.Total flavonoid content recorded from 2.34 mg CE /kg to 2402.00 mg CE/kg.With this study, it was determined that some phytochemical contents of different tissues of the grapes vary and this difference also occurs between the grape varieties.

Keywords: Grape, Phytochemical contents, Berry tissues

Introduction

In 2018 grapes are produced on 7.2 million hectares in the world and approximately with 417 thousand hectares area and 6 % ratio Turkey placed 5th row. In the same year, 79 million tons of grape were produced in the world, and with 3.9 million tons and a 5% ratio, Turkey placed 6th row. According to the data of the World Food Organization (FAO), the production amount increased by 8.4% and the area increased by 3.4% in 2018 compared to the previous year. (Tarım ve Orman Bakanlığı Bitkisel Üretim Genel Müdürlüğü, 2020).

The economy of Hittite;Phrygian,Urartian and Lydian which were important cultures of ancient Anatolia in the 2nd and 1st Millennium BC was based on agriculture and livestock which the main agricultural products of the Hittites were wheat and barley as well as peas,beans,onions, flax, figs, olives, grapes, apples and pomegranates(Bülbül, 2017) Viticulture is still one of the most important agricultural branches in Anatolia. (Söylemezoğlu et al., 2018). Viticulture was made in a total area of 175,387 da in Diyarbakır and 103,872 tons of production was realized in 2019. (TÜİK, 2019).

The number of standard and local grape varieties commonly used in grape production in Diyarbakır is 74 and Boğazkere, Kızıl Banki, Öküzgözü and Şire (sin. Mazrumi) take place among the varieties (Karataş et al.,2015). The synonym of the Kızıl Banki variety grown in Diyarbakır is Kızıl Vanki (Gürsöz,1993). Öküzgözü, Boğazkere and Şire grape varieties are widely grown in Diyarbakır, Elazığ and Mardin provinces (Özdemir and Sessiz,2018).

Grape varieties (*Vitis vinifera L.*) are considered to be

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beneficial for the health through their antioxidant effects,fatty acid contents and phenolic contents(Odabaşıoğlu and Gürsöz,2020). Researchers carry out studies in order to search grape positive effects on human health (Özdemir,et al.,2017). Scientists conduct studies to find grapes positive effects on lot of chronic diseases (Özden and Vardin,2009).Grape(*Vitis vinifera L.*)have positive effects on health thanks to their many various bioactive phenolic contents(Lutz,et al.,2011). Grapes are important in terms of sources of nutritional antioxidants and biologically active dietary components at the same time (Eshghi,et al.,2014). Grapes are rich in terms of resveratrol,phenolics and flavonoids that are claimed to be responsible on their benefits for the health(Yang,et al.,2009). This research study is aimed to examine the important phytochemical contents of grapes in terms of human health that are widely grown in Diyarbakır and to make a comparision between varieties and also grape berry organs.

Materials and Methods

In this study, 4 different grape varieties were examined. The varieties examined are Boğazkere, Öküzgözü, Şire and Kızıl Banki grape varieties grown within Diyarbakır province. Grapes were taken from different vineyards (modern and traditional vineyards). Samples of the varieties were taken when each variety reached harvest maturity. The samples for the study were kept at -20 ^{o}C and were taken off deep freezer before being used. The amount of total phenolic, total flavonoid and total anthocyanin were examined as three replications in berry skin, berry pulp and berry seed of each grape variety, and our study was realized in the Central Laboratory of Harran University.

Preparation of extract for total phenolic content

With modification, grape berries were selected as 5 g from grape sample and separated as pulp, skin and seed. It was treated with liquid nitrogen and was ground in the mortar. It was extracted by 50 mL of distilled water over 5 g. It was heated in the Soxhlet device until it reached the boiling temperature and 1 hour after the boiling started it was taken. When it came to room temperature they were placed in tubes. Analyzes for skin and seed were made after dilution of 1/10 from the extracted samples(Atak and Uslu,2018).

Preparation of extract for total flavonoid and total anthocyanin content

With modification, , grape berries were selected as 5 g from grape samples and separated as pulp, skin and seed. They were extracted with 50 mL of ethanol over 5 g, heated up to boiling temperature in the Soxhlet device and 1 hour after the boiling started it was taken and kept until it came to room temperature and placed in tubes. Analyzes were made after dilution of 1/10(Atak and Uslu,2018).

Total Phenolic Content Determination Method

Content of total phenolic was determined with some modifications and using gallic acid as standard by Folin-Ciocalteu (Singleton and Rossi, 1965). For the mixture 0.4 mL of diluted extract solution was used to mix with 2 mL of Folin-Ciocalteu reagent (the reagent was pre-diluted by distilled water 10 times) and 1.6 mL of sodium carbonate (7.5% w /v). At room temperature after 60 minutes of incubation, the absorbance was measured at 765 nm against the blank solution prepared using a UV-Visspectrophotometer (Akyurt et al., 2018).Our findings in our study evaluated as mg GAE/kg.

Total Anthocyanin Content Determination Method

Method of pH differential (Giusti and Wrolstad) was used. In the framework of this method,the spectrophotometric absorptions of extracts that incubated at room temperature for 15 minutes in the 0.025 M KCl buffer (pH 1.0) and 0.4 M CH3COONa buffer (pH 4.5) were measured at 520 and 700 nm and the absorbance values found by formula (Özden and Özden,2014).

Total Flavonoid Content Determination Method

Aluminum chloride colorimetric method (Zhishen et.al.,1999) was used. One portion (1 mL) of extract or standard catechin solution (20, 40, 60, 80 and 100 mg /L) was added to a 10 mL volumetric flask containing 4 mL dd H_2O. Also to the flask ,0.3 mL of 5% $NaNO_2$ was added . 5 minutes after, 0.3 mL of 10% $AlCl_3$ was dropped, and at the minute of 6th ,2 mL of 1 M NaOH was added to complete total volume to 10 mL .The solution was mixed good and then the absorbance was measured against the reagent blank which was prepared at 510 nm. The content of total flavonoid was expressed as mg catechin equivalent (CE) / 100 g fresh mass (Srivastava et al.,2013). In our study, the values were converted to mg CE / kg.

Statistical Analysis

Variance Analysis (ANOVA) of the data of the study was done by using the SPSS 20.0 package programme. Then Tukey test was used to determine the level of differences.

Results and Discussion

According to a research carried out with 22 grape varieties that grown in Marmara region of Turkey, total phenolic amount of pulp, seed and skin was respectively found between 9.26- 62.29; 162.29- 326.18 and 96.61- 167.42 mg GAE / 100 g fresh weight (Yılmaz et al., 2015). Furthermore ,some quality and the phytochemical properties of Bertiz Kabarcık grape which is important in Kahramanmaraş viticulture was examined and the total amount of phenolic substance was reported to vary between 44.3 mg GAE 100 g $^{-1}$ and 313.9 mg GAE 100 g $^{-1}$ (Balbaba and Bağcı, 2020). Also, total phenolic content of the seeds of two white (Emir and Gök grape) and one black (Kara Dimrit) grape variety were examined and the content of highest total phenolic was found with the seeds of the Gök grape variety with 87031.32 mg GAE / kg (Akın and Altındişli, 2010). When compared with this study the seed contents difference could be originated due to variety. In our study, the least total phenolic amounts were detected in berry pulp and the highest amounts were found in berry seed of varieties except Boğazkere variety. With this aspect, our findings shows similarity with previous studies .

In another study which anthocyanin, the tannin based phenolic compound profile and other phytochemical properties of Kalecik Karası grape variety which grown in Ankara and Nevşehir conditions were examined, amount of total anthocyanin varied between 323.08 mg kg^{-1}- 202.37 mg kg^{-1} (Toprak ,2011).In a study with sixteen red grape varieties total anthocyanin(TA)content ranged from 40.3 mg/L to 990.8

mg/L fresh weight (Orak,2007). In our study anthocyanin was determined between 25.60-634.00 mg / kg.Our study is in conformity with the studies.

A study carried out with 29 grape varieties in Şanlıurfa province, it was reported that when the tissues of grapes were compared, pulp contained very low flavonoid contents while the values of skin and seed were close to each other. It was stated that the highest amount of flavonoid was in the seed of Kızıl Banki among seed contents (0.371 g. kg^{-1}) (Polat, 2016) . In our study, the highest value was found in the seed of the Kızıl Banki variety and the lowest values were found in the berry pulp. Our study is similar to this study.

Where phenolic contents were examined as well as other parameters in the skin, pulp and seed parts of 15 grape varieties, it was reported that content of the total phenolic was the highest in the seed, then in skin and last in pulp (Harbi et al., 2013). In our study, the highest values were found in the berry seed except Boğazkere and the least values were found in the berry pulp. Our study is supported by the previous study findings.

Phenolic compounds of varieties of 3 table grape (Alphonse Lavallee, Red Globe and Hamburg Misketi) and varieties of 3 wine grape (Boğazkere, Cabernet Sauvignon and Kalecik Karası) were determined in a study, the highest total anthocyanin amount was found with the skin of Boğazkere variety and the total phenolic compound amount was found with Kalecik Karası seed (Tahmaz et.al.,2013). In our study, the highest values of anthocyanin were found in the berry skin of the varieties and are listed as Öküzgözü, Boğazkere and Kızıl Banki, Şire respectively.

A study which the contents of total phenolic and total flavonoid of Öküzgözü and Boğazkere varieties were examined in pulp, seed and skin for 2 years, the flavonoid content was found between 5.08 µg QUE / mg and 111.55 µg QUE / mg.And It has been reported that the contents of total phenolic (µg GAE / mg) and total flavonoid of Öküzgözü and Boğazkere cultivars differ significantly according to skin, seed ,pulp and years (Özdemir et al.,2017).In our study differences were seen as well in total flavonoid values in terms of berry tissues.

However, amount of total phenolic and total flavonoid substance and the antioxidant activities in the skin, seed, pulp and extracts obtained from whole berry were examined with 12 grape genotypes grown in Turkey. When compared the grape samples in terms of substance of total phenolic and total flavonoid, the highest amounts were obtained in grape seeds, then skin and then whole berry and pulp at the last (Bayır Yeğin and Uzun,2018). Our study showed that, the highest content of total phenolic was found with the seed of varieties except Boğazkere. Boğazkere variety has got thick skin ,and we think that made higher skin phenolic content And the lowest values were found with berry pulp. Our findings are generally supported by the findings of this study.

A study with 6 grape varieties obtained from Tunceli province, the highest content of anthocyanin was found with Koşkuran variety (1192.1 mg / kg) the lowest content of anthocyanin was found with Ulaş Siyahı variety (358.5 mg /kg) (Karaca Sanyürek et al.,2018). Also, a study which phytochemical properties and total antioxidant activities of some wine grape varieties that grown in Şanlıurfa conditions were determined it was reported that the total phenolic content, anthocyanin content, antioxidant activities and the phytochemical properties of grape varieties may vary based on the grape variety, growing climate and soil conditions, soil type, levels of ripening, cultural practices and amount of yield. Total anthocyanin contents of the varieties of Merlot, Chardonnay, Cabernet Sauvignon, and Şiraz were respectively 1144.9; 39.48; 723.3, and 1011.6 mg / kg (Özden and Vardin, 2009). Our study shows similarity with the previous findings. The highest anthocyanin value was found in the berry skin of Öküzgözü variety and values vary between 25.60-634.00 mg / kg.

In another study with Shiraz grape variety that grown in the district of Güney of Denizli which carried out in order to research the effects of four different cluster thinning (8- 16- 24 and 32 clusters / grapevine) being applied just after berry retention, on the yield, quality properties and on the biochemical properties of the berry ; highest total phenolic (285.20 mg GAE / 100 g) and total flavonoid (100.68 mg CTE / 100 g) substance amounts were found from 8 cluster / grapevine(Pehlivan and Uzun,2015). Total flavonoid content of Perricone grape variety in the higher peak concentration was 3233.29 ± 347.32 mg/kg in the dates of last harvest , and the total flavonoid content of Nero d'Avola grapes in maximum peak concentration was 2519.22 ± 66.91 mg/kg in the maturation of later stage reported in another study (Gervasi et.al., 2016). In the study carried out with 29 grape varieties in Şanlıurfa province ,total flavonoid contents of berry pulp of white varieties changed between 0.968 mg/kg-10.6 mg/kg and total flavonoid contents of berry pulp of colored varieties changed between 1.87 mg/kg and 20.0 mg/kg in terms of years of average(Polat,2016)In our study, total flavonoid values were between 2.34-2402 mg CE / kg and our findings are in conformity.

Table 1. Findings of Total Phenolic Content(mg GAE / kg)

Berry tissue	Boğazkere		Öküzgözü		Kızıl Banki		Şire	
	Mean	Std. deviation	Mean	Std. deviation	Mean	Std. deviation	Mean	Std. deviation
Berry skin	4004.32^{bc}	373.47	3437.51^{b}	26.89	2718.36^{b}	134.34	3228.35^{b}	108.57
Berry pulp	1200.88^{a}	9.32	457.56 ^{a}	20.93	435.89^{a}	9.07	389.15^{a}	20.13
Berry seed	3616.00^{b}	331.62	3907.86^{c}	243.17	3803.29^{c}	217.74	4050.17^{c}	215.77

The differences between means shown with dissimilar characters in the same column is statistically important (P <0.05). Differences between means shown with similar characters in the same column is not statistically important.

Table 2. Findings of Total Anthocyanin Content(mg/kg)

Berry tissue	Boğazkere		Öküzgözü		Kızıl Banki		Şire	
	Mean	Std. deviation	Mean	Std. deviation	Mean	Std. deviation	Mean	Std. deviation
Berry skin	457.55^{c}	11.69	634.00^{c}	22.24	227.10^{c}	43.80	94.63^{abc}	15.15
Berry pulp	115.78^{b}	2.55	134.15^{b}	25.12	123.57^{b}	24.71	85.16^{a}	7.28
Berry seed	80.15^{a}	6.02	25.60^{a}	8.40	47.87^{a}	5.86	87.95^{ab}	5.37

The differences between means shown with dissimilar characters in the same column is statistically important (P <0.05). Differences between means shown with similar characters in the same column is not statistically important.

Table 3. Findings of Total Flavonoid Content (mg CE/kg)

Berry tissue	Boğazkere		Öküzgözü		Kızıl Banki		Şire	
	Mean	Std. deviation	Mean	Std. deviation	Mean	Std. deviation	Mean	Std. deviation
Berry skin	173.94^{b}	4.87	144.30^{b}	3.57	70.98^{ab}	8.22	14.53^{ab}	3.70
Berry pulp	49.92^{a}	1.35	19.50^{a}	1.35	44.46^{a}	2.34	2.34^{a}	0.05
Berry seed	368.90^{c}	6.21	1457.03^{c}	61.42	2402.00^{c}	22.59	1436.00^{c}	118.05

The differences between means shown with dissimilar characters in the same column is statistically important (P <0.05). Differences between means shown with similar characters in the same column is not statistically important.

Table 4.Total Phenolic Content of Colored Varieties (mg GAE/kg)

Variety	Berry pulp		Berry seed		Berry skin	
	Mean	Std. deviation	Mean	Std. deviation	Mean	Std. deviation
Boğazkere	1200.88 ^{c}	9.32	3616.00 ^{a}	331.62	4004.32 ^{bc}	373.47
Öküzgözü	457.56^{ab}	20.93	3907.86 ^{abc}	243.17	3437.51 ^{b}	26.89
Kızıl Banki	435.89^{a}	9.07	3803.29^{ab}	217.74	2718.36 ^{a}	134.34

The differences between means shown with dissimilar characters in the same column is statistically important (P <0.05). Differences between means shown with similar characters in the same column is not statistically important.

Table 5.Total Anthocyanin Content of Colored Varieties (mg/kg)

Variety	Berry pulp		Berry seed		Berry skin	
	Mean	Std. deviation	Mean	Std. deviation	Mean	Std. deviation
Boğazkere	115.78^{a}	2.55	80.15^{c}	6.02	457.55^{b}	11.69
Öküzgözü	134.15^{abc}	25.12	25.60^{a}	8.40	634.00 ^{c}	22.24
Kızıl Banki	123.57^{ab}	24.71	47.87^{b}	5.86	227.10^{a}	43.80

The differences between means shown with dissimilar characters in the same column is statistically important (P <0.05). Differences between means shown with similar characters in the same column is not statistically important.

Table 6. Total Flavonoid Content of Colored Varieties (mg CE/kg)

Variety	Berry pulp		Berry seed		Berry skin	
	Mean	Std. deviation	Mean	Std. deviation	Mean	Std. deviation
Boğazkere	49.92^{c}	1.35	368.90^{a}	6.21	173.94^{c}	4.87
Öküzgözü	19.50^{a}	1.35	1457.03^{b}	61.42	144.30^{b}	3.57
Kızıl Banki	44.46^{b}	2.34	2402.00^{c}	22.59	70.98^{a}	8.22

The differences between means shown with dissimilar characters in the same column is statistically important (P <0.05). Differences between means shown with similar characters in the same column is not statistically important.

Table 7. Total Phenolic Content of Varieties (mg GAE/kg)

Variety	Berry pulp		Berry seed		Berry skin	
	Mean	Std. deviation	Mean	Std. deviation	Mean	Std. deviation
Boğazkere	1200.88^{d}	9.32	3616.00^{a}	331.62	4004.32^{d}	373.47
Öküzgözü	457.56 ^{bc}	20.93	3907.86 ^{abc}	243.17	3437.51^{bc}	26.89
Kızıl Banki	435.89 ^{b}	9.07	3803.29 ^{ab}	217.74	2718.36 ^{a}	134.34
Şire	389.15 ^{a}	20.13	4050.17^{abcd}	215.77	3228.35 ^{b}	108.57

The differences between means shown with dissimilar characters in the same column is statistically important (P <0.05). Differences between means shown with similar characters in the same column is not statistically important.

Table 8. Total Anthocyanin Content of Varieties (mg/kg)

Variety	Berry pulp		Berry seed		Berry skin	
	Mean	Std. deviation	Mean	Std. deviation	Mean	Std. deviation
Boğazkere	115.78 ^{ab}	2.55	80.15 ^{c}	6.02	457.55^{c}	11.69
Öküzgözü	134.15^{bcd}	25.12	25.60 ^{a}	8.40	634.00^{d}	22.24
Kızıl Banki	123.57 ^{abc}	24.71	47.87 ^{b}	5.86	227.10^{b}	43.80
Şire	85.16 ^{a}	7.28	87.95 ^{d}	5.37	94.63^{a}	15.15

The differences between means shown with dissimilar characters in the same column is statistically important (P <0.05). Differences between means shown with similar characters in the same column is not statistically important.

Table 9. Total Flavonoid Content of Varieties (mg CE/kg)

Variety	Berry pulp		Berry seed		Berry skin	
	Mean	Std. deviation	Mean	Std. deviation	Mean	Std. deviation
Boğazkere	49.92^{d}	1.35	368.90 ^{a}	6.21	173.94^{d}	4.87
Öküzgözü	19.50^{b}	1.35	1457.03^{bc}	61.42	144.30^{c}	3.57
Kızıl Banki	44.46^{c}	2.34	2402.00^{d}	22.59	70.98^{b}	8.22
Şire	2.34^{a}	0.05	1436.00^{b}	118.05	14.53^{a}	3.70

The differences between means shown with dissimilar characters in the same column is statistically important (P <0.05). Differences between means shown with similar characters in the same column is not statistically important.

Conclusion

In our study contents of total phenolic, total anthocyanin and total flavonoid were detected in the berry pulp, berry skin and berry seed of Boğazkere, Öküzgözü, Kızıl Banki and Şire varieties which are commonly grown. The results showed significant differences both between the tissues of the variety and among the varieties. It is thought that the differences are caused by the factors such as cultivation, care conditions, irrigation, variety characteristics, climate, topography. Total phenolic content ranged from 389.15 mg GAE / kg to 4050.17 mg GAE / kg, while total anthocyanin content was recorded between 25.60 mg / kg and 634.00 mg / kg. Total flavonoid content ranged from 2.34 mg CE / kg to 2402.00 mg CE / kg,

Because of contents of these varieties they are considered to be an important resource for the research studies that will be carried out for human health. Phytochemical contents of Boğazkere,Öküzgözü,Kızıl Banki and Şire can be studied again,especially high phenolic content of Boğazkere skin and pulp is important for the upcoming studies. We think the grape varieties examined have an important potential in terms of phenolic compounds.

Compliance with Ethical Standards

Conflict of interest

The author declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

Author contribution

The author read and approved the final manuscript. The author verifies that the Text, Figures, and Tables are original and that they have not been published before.

Ethical approval

Not applicable.

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Data availability

Not applicable.

Consent for publication

Not applicable.

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