

Variation in Organic Acid, Sugar and Phenolic Compounds in Fruits of Historical Apple Cultivars

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Abstract

Knowledge about diversity of local apple cultivars is particularly important and helps to ensure that specific cultivars are available when and where farmers need them. The phenolic content, organic acids and sugars were investigated in the four historical apple cultivars ('Aslik', 'Cebegirmez', 'Bey Elmasi' and 'Arapkizi') grown in Van province, located in Eastern Anatolia, Turkey. In the study, ellagic acid (63.034 mg per 100 ml) was found to be higher than other phenolic compounds and followed by vanilic acid, caffeic acid and routine, respectively. When examined the apple cultivars, it has been found that the 'Bey Elmasi' cultivar has the highest content of six phenolic acids (chlorogenic, ferulic, *o*-coumaric, *p*-coumaric, catechin and quercetin). The organic acid contents were as follows: malic>fumaric>succinic>citric>tartaric among cultivars. 'Arapkizi' cultivar possessed the highest value of four organic acids except of tartaric acid. It was determined that fructose was the major sugar for all apple cultivars. There was a positive correlation among gallic, ellagic acid and routine. Similarly, the positive correlation was observed among malic, fumaric and succinic acid. However the negative correlation between citric acid and all sugars was observed. As a result, 'Bey Elmasi' and 'Arapkizi' cultivars with higher phenolic content, 'Arapkizi' cultivar with higher organic acid content and 'Aslik' cultivar with higher sugar content comes to the foreground. Results indicated the importance of local apple germplasm to use of them for future apple breeding activities to increase inner quality.

Keywords: autochthonous Turkish apple cultivars; organic acids; phenolic compounds; sugars

Introduction

Anatolia is accepted the homeland of many wild, wild edible and cultivated fruit species (Kamiloglu *et al.*, 2009; Tosun *et al.*, 2009; Ercisli *et al.*, 2012; Canan *et al.*, 2016; Caliskan *et al.*, 2017). Anatolia has also rich apple genetic resources including mostly historical cultivars selected by humans among wild populations for centuries. There are hundreds historical apple cultivars distributed different agroclimatic conditions in Turkey. This kind of richness gives the choice of different uses of them for the breeders (Kaya and Balta, 2013). In addition Turkey is an important apple producer in the world besides the rich apple genetic resources. World apple production is more than 84 million

tons. Turkey produces around 3.0 million tons and takes fourth place after China (40 million), United States (5 million) and Poland (3.1 million) (FAO, 2017).

In recent years their consumption are increasing as a diet product and for health because advertisement of apple phytochemicals (Boyer and Liu, 2004). They reduce risk of lung cancer (Michels *et al.*, 2006), risk of type II diabet and lost weight (Boyer and Liu, 2004) and prevent risk of cardiovascular disease (Sesso *et al.*, 2003; Boyer and Liu, 2004). The apples are rich in potassium, phosphorus, calcium, magnesium and sodium, respectively (Mordogan and Ergun, 2001). Also, apple fruits have strong antioxidant activity due to contained flavonoid and phenolic acids (Wolfe *et al.*, 2003; Boyer and Liu, 2004). The researchers suggest that the apple fruit is rich in chlorogenic, catechin (Carbone *et al.*, 2011) and phloridzin (Jakobek *et al.*, 2016).

The apple peel is richer in phenolic substance than apple flesh (Wang *et al.*, 2015; Khanizadeh *et al.*, 2008). Apple seeds are also known to be rich in phenolic compounds. It is emphasized by some researchers that the seeds contain the most phloridzin and followed by hyperin and chlorogenic acid (Xu *et al.*, 2016). Studies show that apple growing conditions, storage time and shape (Seçmen *et al.*, 2015) and cultivar affect phenolic compounds (Khanizadeh *et al.*, 2008).

It is important to protect apple genetic resources in order to obtain qualified apple cultivars. The value of the historical cultivars grown in our country must be preserved and the biological characteristics should be determined. Van province is home to many fruit species and the presence of a large number of historical apple cultivars (Özrenk *et al.*, 2011). Although there are some studies on the pomological characteristics of these historical cultivars, no studies on their phytochemical properties have been reported.

Thus, the aim of our study was to reveal the phytochemical properties of 4 well-known historical apple cultivars grown in the Van region. In addition, the correlation between phenolic compounds, organic acids and sugars has been revealed and presented in the statistical distribution between these biochemicals.

Materials and Methods

Fruits samples

In this study, 'Aslik', 'Arapkizi', 'Bey Elmasi' and 'Cebegirmez' historical apple cultivars were used. These cultivars were grown in the Van region. They were harvested at full maturity stage. The fruit samples picked homogeneously were stored at -80 °C until their laboratory analyses were conducted.

Analysis of phenolic compounds

In the research, the gallic acid, ellagic, catechin, chlorogenic acid, caffeic acid, syringic acid, *p*-coumaric acid, ferulic acid, *o*-coumaric acid, phloridzin, protocatechuic, vanillic acid, rutin and quercetin were determined. In the separation of phenolic acids with HPLC (Agilent 1100 series HPLC G 1322 A, Germany), the method developed by Rodriguez-Delgado *et al.* (2001) was modified and used. The samples collected were distilled with distilled water at the ratio of 1:1 and after they were centrifuged at 15000 rpm for 15 min., the supernatant was filtered with 0.45 µm millipore filters and then injected to HPLC. The chromatographic separation was conducted by using DAD detector (Agilent, USA) and 250*4.6 mm, 4 µm ODS column (HiChrom, USA) in Agilent 1100 (Agilent) HPLC system. Solvent A Methanol-acidic acid-water (10:2:88), Solvent B Methanol-acidic acid-water (90:2:8) were used as the mobile phase. The separation was conducted at 254 and 280 nm and the flow rate was determined as 1 mL/min. and the injection volume was determined as 20 µL.

Analysis of organic acids

In the research, the citric acid, tartaric acid, malic acid, succinic acid, oxalic acid and fumaric acid contents were determined among the organic acids. In the extraction of organic acids, the method developed by Bevilacqua and

Califano (1989) was modified and used. 5 g was taken from the fruit samples obtained and transferred to centrifuge tubes. These samples were homogenized by adding 20 ml 0.009 N H₂SO₄ (Heidolph Silent Crusher M, Germany). Then, it was mixed on the agitator (Heidolph Unimax 1010, Germany) for 1 hour and centrifuged at 15000 rpm for 15 minutes. The aqueous part which was separated at centrifuge was filtered from first coarse filter paper, then 0.45 µm membrane filter (Millipore Millex-HV Hydrophilic PVDF, Millipore, USA) for two times and finally SEP-PAK C18 cartridge. The organic acids were analyzed in HPLC device (Agilent HPLC 1100 series G 1322 A, Germany) by using the method developed by Bevilacqua and Califano (1989). In HPLC system, Aminex HPX - 87 H, 300 mm × 7.8 mm column (Bio-Rad Laboratories, Richmond, CA, USA) was used and the device was controlled with the computers including Agilent package program. DAD detector in the system (Agilent, USA) was set to 214 and 280 nm wavelengths. In the study, 0.009 N H₂SO₄ filtered at 0.45 µm membrane filter was used as mobile phase.

Sugar analysis

The modified method of Melgarejo *et al.* (2000) was used for sugar (fructose, glucose and sucrose) analyses. 5 mL of fruit extracts was centrifuged at 12000 rpm for 2 minutes at temperature of 4 °C. Supernatants were passed by SEP-PAK C18 cartridge. HPLC readings were made with µbondapak-NH₂ column using 85% acetonitrile as liquid phase with refractive index detector (IR). Fructose and glucose standards were used for sugar calculations.

Statistical analysis

R 3.3.4 and SAS/STAT software was utilized for statistical analyzes of the data. Descriptive statistics, normal distribution tests, correlation analysis and one way variance analysis were performed in SAS software. One-way analysis of variance was used to determine the differences between local cultivars averages in terms of phenolic compounds, organic acids and sugars. The Duncan test was used as a multiple comparison test to express the differences between the averages. In R software, Principle component analysis (PCA) was used to for all variables examined using ggplot2 and facto extra packages (Package ggplot2, 2016; Package facto extra, 2017; SAS/STAT, 2012).

Results and Discussion

In this study, it was determined that the main phenolics were ellagic acid, followed by vanillic acid, rutin and caffeic acid in historical apple cultivars (Tables 1, 2, 3, Fig. 1).

The highest ellagic acid content (63.034 mg/100 ml) was obtained from 'Arapkizi' cultivar and followed by 'Cebegirmez' and 'Aslik' (Table 1). While the highest content of gallic and protocatechuic acid were recorded from 'Arapkizi', the lowest content was determined from 'Bey Elmasi' cultivar. 'Aslik' possessed the highest syringic (2.792 mg/100 ml) and vanilic (31.426 mg/100 ml) acid content (Table 1). Chen *et al.* (2012) determined that the percentage of gallic acid in the whole apple fruit is higher than the peel. He reported gallic acid content of apple

cultivars in followed order: Red Delicious > Royal Gala > Golden Delicious. In a study of red and white fleshed apple cultivars, researchers reported that gallic and syringic content changed among cultivars. Gallic acid was measured as 4.0 mg/kg in red fleshed Hongxun and as 1.5 mg/kg in white flesh Golden Delicious. Also, syringic acid content ranged from 13.8 to 48.1 mg/kg (Wang et al., 2015). Although gallic acid values were comparable with our study, syringic acid values weren't comparable. Karadeniz et al. (2015) reported phenolic compound differences in apple cultivars. The difference with other studies is thought to be due to the cultivars, ecological factors and method differences.

The highest routine content was measured in 'Cebegirmez' as 21.020 mg/100 g, but this cultivar possessed the lowest catechin and phloridzin content (Tables 1, 2, Fig. 1). Catechin content ranged from 4.347-12.619 mg/100 g among apple cultivars (Table 2). It was determined that the highest of phloridzin content was 4.513 mg/100 g as 'Arapkizi' (Table 2, Fig. 1). Jakobek et al. (2016) conducted a study on red and yellow peel colored apple cultivars and

they measured phloridzin as 10.5-30.2 mg/kg in red peeled apple cultivars. Also, researchers found that yellow peeled apple cultivars had 3.0-13.7 mg/kg catechin content. However, contrary our study, some researchers reported that 'Arapkizi' had the lowest phloridzin content as 0.52 mg/kg (Karaman et al., 2010). In another study, some researchers were recorded routine content as 0.37-1.59 mg/g in whole fruit and 2.76-11.4 mg/g in apple peel in different cultivars (Lata et al., 2009). Schieber et al. (2015) conducted a study on phenolic component of apple juice and pomace. They determined that quercetin content measured as 1.2 mg/L in juice and 67 mg/kg in pomace. In another study, researchers reported that the highest catechin content recorded as 27.7 µg/g in SuperMac cultivar (Khanizadeh et al., 2008). It was observed that our values were higher than the results of some researches. It is thought that the difference is caused by ecological factors and cultivars.

The lowest phenolic compound among apple cultivars was measured as *o*-coumaric acid (Table 3, Fig. 1). 'Bey Elmasi' cultivar possessed the highest content of six phenolic

Table 1. Protocatechuic, vanillic, syringic, gallic and ellagic contents of historical apple cultivars (mg/100 ml)

Variable	Historical cultivars	Mean	Low. 95%	Upp. 95%	ANOVA p-value and R-Square
Ellagic	Arapkizi	63.084 ± 2.961 ^A	25.461	100.707	<0.0001 R ² =0.9921
	Aslik	23.859 ± 0.503 ^{BC}	17.474	30.243	
	Bey Elmasi	18.858 ± 0.746 ^C	9.379	28.337	
	Cebegirmez	26.835 ± 0.357 ^B	22.299	31.371	
Gallic	Arapkizi	0.556 ± 0.014 ^A	0.384	0.727	<0.0035 R ² =0.9564
	Aslik	0.327 ± 0.007 ^B	0.238	0.416	
	Bey Elmasi	0.091 ± 0.007 ^C	0.008	0.173	
	Cebegirmez	0.228 ± 0.071 ^{BC}	-0.668	1.123	
Syringic	Arapkizi	0.896 ± 0.007 ^D	0.813	0.978	<0.0001 R ² =0.9964
	Aslik	2.792 ± 0.007 ^A	2.703	2.881	
	Bey Elmasi	2.187 ± 0.079 ^B	1.189	3.184	
	Cebegirmez	1.832 ± 0.023 ^C	1.540	2.124	
Vanillic	Arapkizi	1.110 ± 0.015 ^C	0.919	1.301	<0.0001 R ² =0.9991
	Aslik	31.426 ± 0.608 ^A	23.701	39.151	
	Bey Elmasi	23.751 ± 0.443 ^B	18.128	29.373	
	Cebegirmez	1.953 ± 0.076 ^C	0.993	2.912	
Protocatechuic	Arapkizi	0.298 ± 0.003 ^A	0.260	0.336	<0.001 R ² =0.9951
	Aslik	0.259 ± 0.001 ^B	0.252	0.265	
	Bey Elmasi	0.153 ± 0.006 ^C	0.077	0.229	
	Cebegirmez	0.263 ± 0.004 ^B	0.218	0.307	

Table 2. Catechin, phloridzin, quercetin and routine contents of historical apple cultivars (mg 100/ml)

Variable	Historical cultivars	Mean	Low. 95%	Upp. 95%	ANOVA p-value and R-Square
Catechin	Arapkizi	5.658 ± 0.229 ^C	2.748	8.568	<0.0001 R ² =0.9971
	Aslik	6.751 ± 0.194 ^B	4.292	9.209	
	Bey Elmasi	12.619 ± 0.138 ^A	10.871	14.366	
	Cebegirmez	4.347 ± 0.070 ^D	3.458	5.236	
Phloridzin	Arapkizi	4.513 ± 0.015 ^A	4.322	4.704	<0.0022 R ² =0.9653
	Aslik	3.332 ± 0.166 ^B	1.229	5.434	
	Bey Elmasi	3.441 ± 0.075 ^B	2.488	4.394	
	Cebegirmez	3.039 ± 0.107 ^B	1.679	4.399	
Quercetin	Arapkizi	0.605 ± 0.025 ^B	0.287	0.923	<0.0211 R ² =0.8918
	Aslik	0.743 ± 0.031 ^A	0.349	1.137	
	Bey Elmasi	0.743 ± 0.011 ^A	0.609	0.876	
	Cebegirmez	0.650 ± 0.007 ^B	0.567	0.732	
Routine	Arapkizi	13.451 ± 1.211 ^B	-1.936	28.838	<0.0006 R ² =0.9816
	Aslik	8.336 ± 0.805 ^C	-1.892	18.564	
	Bey Elmasi	4.150 ± 0.742 ^D	-5.278	13.578	
	Cebegirmez	21.020 ± 0.541 ^B	14.146	27.894	

Table 3. *p*-Coumaric, ferulic, *o*-coumaric, chlorogenic and caffeic contents of apple cultivars (mg 100/ml)

Variable	Historical cultivars	Mean	Low. 95%	Upp. 95%	ANOVA p-value and R-Square
Caffeic	Arapkizi	7.298 ± 0.081 ^B	6.275	8.320	<0.0001 R ² =0.9980
	Aslik	7.740 ± 0.039 ^B	7.244	8.236	
	Bey Elmasi	6.947 ± 0.083 ^B	5.898	7.995	
	Cebegirmez	22.743 ± 0.583 ^A	15.341	30.144	
Chlorogenic	Arapkizi	3.594 ± 0.053 ^C	2.921	4.267	<0.0001 R ² =0.9985
	Aslik	3.919 ± 0.020 ^B	3.665	4.173	
	Bey Elmasi	5.630 ± 0.049 ^A	5.013	6.246	
	Cebegirmez	2.348 ± 0.050 ^D	1.713	2.983	
Ferulic	Arapkizi	0.355 ± 0.008 ^{AB}	0.253	0.457	<0.0253 R ² =0.8814
	Aslik	0.477 ± 0.003 ^A	0.445	0.508	
	Bey Elmasi	0.500 ± 0.035 ^A	0.055	0.945	
	Cebegirmez	0.243 ± 0.067 ^B	-0.602	1.087	
<i>p</i> _coumaric	Arapkizi	1.105 ± 0.085 ^A	0.025	2.185	<0.0001 R ² =0.9958
	Aslik	0.966 ± 0.006 ^A	0.896	1.035	
	Bey Elmasi	1.112 ± 0.003 ^A	1.080	1.143	
	Cebegirmez	0.952 ± 0.005 ^A	0.894	1.009	
<i>o</i> _coumaric	Arapkizi	0.091 ± 0.008 ^C	-0.005	0.186	<0.1022 R ² =0.7558
	Aslik	0.082 ± 0.004 ^C	0.037	0.126	
	Bey Elmasi	0.267 ± 0.005 ^A	0.209	0.324	
	Cebegirmez	0.163 ± 0.002 ^B	0.143	0.182	

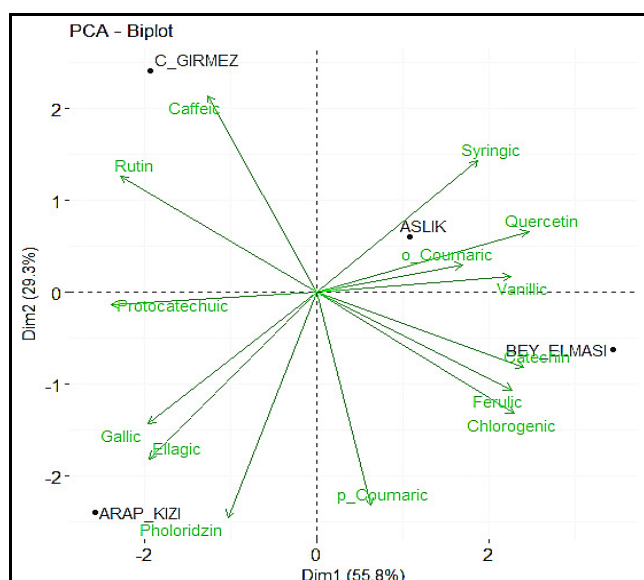


Fig. 1. The distribution of phenolic compounds according to cultivars

acids but this cultivar also had the lowest content of five the other phenolic compounds (Tables 1, 2, 3). In addition ‘Arapkizi’ cultivar had the highest content of five phenolic acids (Tables 1, 2, 3).

It was identified that there was a positive correlation among gallic, ellagic acid and routine (Table 4, Fig. 2). However there was a negative correlation between gallic and ellagic acid and syringic acid. Therefore, ellagic acid increased as gallic acid increased but syringic acid decreased (Table 4, Fig.2). A similar positive correlation was also identified among ferulic, catechin and chlorogenic acids (Table 4, Fig. 2). But these acids had a negative correlation with routine (Table 4, Fig.2). As shown in Table 1, there

was a statistical difference among historical apple cultivars for phenolic acids.

The highest *o*-coumaric, chlorogenic, ferulic and *p*-coumaric acid content were obtained from ‘Bey Elmasi’ cultivar (Table 3), the lowest content was obtained from ‘Cebegirmez’. Caffeic acid content ranged from 6.947-22.743 mg/100 ml and ‘Cebegirmez’ cultivar possessed this highest content (Table 3). The highest chlorogenic content was measured as 5.630 mg/100 ml (Table 3). Coşkun *et al.* (2016) reported chlorogenic acid ranged between 29.15 mg/kg and 194.45 mg/kg in some local apple cultivars. Also researchers determined the highest caffeic acid was 8.18 mg/kg. In Turkey, chlorogenic and caffeic acid content of ‘Arapkizi’ were measured as 242.3 and 12.14 mg/L, respectively by Karaman *et al.* (2010). In both studies, caffeic acid values are in agreement with our study but chlorogenic acid values are not comparable. In a study by using apple cultivars, the amount of ferulic acid content was found to be higher than *p*-coumaric acid and the ratio of these acids changed for all cultivars (Chen *et al.* 2012). Wang *et al.* (2015) determined that the highest *p*-coumaric, ferulic, caffeic and chlorogenic acid content measured as 0.5, 0.5, 1.3 and 196.0 mg/kg. Khanizadeh *et al.* (2007) measured chlorogenic acid as 183.1 µg/g and phloridzin as 11.8 µg/g in Gala while Tsao *et al.* (2003) measured chlorogenic acid as 153.6 µg/g and phloridzin as 17.96 µg/g in Golden Delicious.

As shown Table 5 and Fig. 3, there was a difference between cultivars in terms of sugar values. It was determined that fructose was the major sugar for all apple cultivars. It was followed by glucose and sucrose. In all sugars, ‘Aslik’ cultivar had the highest sugar content while ‘Arapkizi’ had the lowest sugar content (Table 5, Fig. 3). The content of fructose ranged from 12.315 g/L (‘Arapkizi’) to 16.495 g/L (‘Aslik’) (Table 5, Fig. 3). It was obtained the amount of fructose and glucose was 10 times higher than sucrose

Table 4. Correlations between phenolic compounds

		Pearson Correlation Coefficients, N = 8													
		Protocatechuic	Vanillic	Ellagic	Routine	Quercetin	Gallic	Catechin	Chlorogenic	Caffeic	Syringic	p_Coumaric	Ferulic	o_Coumaric	Phloridzin
Protocatechuic		1.000	-0.532	0.697	0.653	-0.666	0.838*	-0.921*	-0.794*	0.232	-0.451	-0.296	-0.556	-0.902*	0.400
	P-Value		0.175	0.055	0.079	0.071	0.009	0.001	0.019	0.581	0.262	0.477	0.153	0.002	0.326
Vanillic			1.000	-0.653	-0.790*	0.893*	-0.432	0.587	0.639	-0.535	0.875*	-0.078	0.819*	0.141	-0.366
	P-Value			0.079	0.020	0.003	0.285	0.126	0.088	0.172	0.005	0.855	0.013	0.739	0.372
Ellagic				1.000	0.308	-0.781*	0.889*	-0.452	-0.296	-0.205	-0.870*	0.401	-0.320	-0.550	0.896*
	P-Value				0.458	0.022	0.003	0.261	0.476	0.626	0.005	0.325	0.440	0.158	0.003
Routine					1.000	-0.687	0.294	-0.850*	-0.946*	0.853*	-0.437	-0.405	-0.918*	-0.336	-0.095
	P-Value					0.060	0.480	0.008	0.000	0.007	0.279	0.320	0.001	0.416	0.823
Quercetin						1.000	-0.669	0.636	0.600	-0.321	0.871*	-0.017	0.708*	0.392	-0.573
	P-Value						0.070	0.090	0.116	0.438	0.005	0.968	0.049	0.337	0.137
Gallic							1.000	-0.606	-0.397	-0.215	-0.607	0.088	-0.309	-0.823*	0.782*
	P-Value							0.111	0.330	0.610	0.111	0.836	0.457	0.012	0.022
Catechin								1.000	0.962*	-0.570	0.331	0.546	0.735*	0.765*	-0.067
	P-Value								0.000	0.140	0.423	0.161	0.038	0.027	0.874
Chlorogenic									1.000	-0.769*	0.288	0.580	0.828*	0.571	0.120
	P-Value									0.026	0.489	0.132	0.011	0.139	0.776
Caffeic										1.000	-0.064	-0.568	-0.808*	0.060	-0.550
	P-Value										0.881	0.142	0.015	0.887	0.157
Syringic											1.000	-0.443	0.504	0.154	-0.742*
	P-Value											0.271	0.203	0.716	0.035
p_coumaric												1.000	0.336	0.354	0.610
	P-Value												0.416	0.390	0.108
Ferulic													1.000	0.213	0.001
	P-Value													0.613	0.998*
o_coumaric														1.000	-0.364
	P-Value														0.375
Phloridzin															1.000
	P-Value														

*: P≤0.001

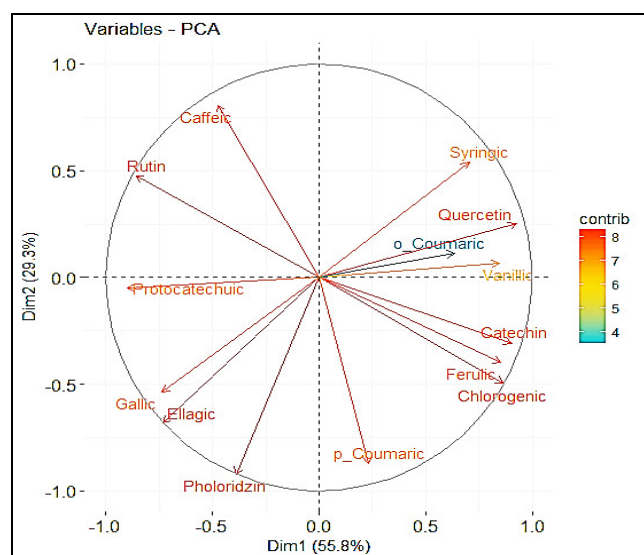


Fig. 2. The correlation of phenolic compounds

content. Referring to Table 7 and Fig. 4, there was a positive correlation among fructose, glucose and sucrose content. So, it was seen that glucose and sucrose content increased as fructose content increased (Table 7, Fig. 4). In Portugal, researchers searched phytochemicals of some apple cultivars. In parallel with our study, it was determined that the major sugar was fructose (4.23-5.62 g/L). Also, researchers measured glucose and sucrose content as 0.73-1.95, 1.17-2.98 g/L respectively (Feliciano *et al.*, 2010). In a different study, sugar concentrations varied depending on the cultivars. Fructose and sucrose concentrations were measured as major sugar. Researchers reported that glucose ranged 5-20 g/kg (Hecke *et al.*, 2006). Our findings are higher compared to Hecke *et al.* (2006) work.

When the organic acid values were examined, statistically differences between the cultivars were significant ($p < 0.05$) (Table 6). Malic acid was measured as the major organic acid while tartaric acid was measured as the lowest (Table 6, Fig. 3). The organic acid contents were as follows: malic>fumaric>succinic>citric>tartaric (Table 6, Fig. 3). ‘Arapkizi’ cultivar possessed the highest value of four organic acids except of tartaric acid (Table 6). On the contrary, ‘Bey Elmasi’ cultivar had the lowest content of organic acids (Table 6). The highest malic acid and fumaric acid content was recorded as 4.613 mg/100 ml and 1.252 mg/100 ml (‘Arapkizi’) (Table 6), respectively. It was determined that ‘Cebegirmez’ cultivar had the lowest succinic (0.144 mg/100 ml) and malic acid content but the highest tartaric acid as 0.214 mg/100 ml (Table 6). As shown Table 7 and Fig. 4, the positive correlation was observed among malic, fumaric and succinic acid. No relationship was found between these acids and tartaric and malic acid. However the opposite correlation between citric acid and all sugars was obtained. That is, fructose, glucose and sucrose decreased as citric acid increased (Table 7, Fig. 4). According to researchers, the highest organic acid content was measured as 7106.05 mg/kg (as malic acid) of some local apple cultivars. They determined that tartaric acid ranged between 84.00-382.55 mg/kg (Coşkun *et al.*, 2016). Feliciano *et al.* (2010) reported that citric acid content ranged from 2.8-8.7 mg/100 g and the major organic acid was malic acid in apple. In a study conducted in Austria, it was determined that malic acid was a major acid and ranged from 5.64-17.8 g/kg. Also, fumaric acid was identified at the lowest organic acid (Hecke *et al.*, 2006). Malic acid was found major acid in all studies, in parallel with our research. However there are numerical differences with other studies. This difference is thought to have originated from the cultivars and ecology.

Table 5. Fructose, glucose and sucrose content of apple cultivars (g l⁻¹)

Variable	Historical cultivars	Mean	Low. 95%	Upp. 95%	ANOVA p-value and R-Square
Fructose	Arapkizi	12.315 ± 0.019 ^D	12.074	12.556	<0.0001 R ² =0.9957
	Aslik	16.495 ± 0.156 ^A	14.513	18.477	
	Bey Elmasi	15.497 ± 0.125 ^B	13.915	17.078	
	Cebegirmez	13.818 ± 0.054 ^C	13.132	14.504	
Glucose	Arapkizi	10.461 ± 0.067 ^D	9.616	11.305	<0.0002R ² =0.9908
	Aslik	12.739 ± 0.045 ^A	12.173	13.304	
	Bey Elmasi	12.073 ± 0.142 ^B	10.275	13.870	
	Cebegirmez	11.255 ± 0.028 ^C	10.899	11.611	
Sucrose	Arapkizi	0.179 ± 0.005 ^D	0.115	0.243	<0.0001 R ² =0.9918
	Aslik	0.423 ± 0.012 ^A	0.271	0.575	
	Bey Elmasi	0.337 ± 0.010 ^B	0.210	0.464	
	Cebegirmez	0.243 ± 0.004 ^C	0.198	0.287	

Table 6. Organic acid content of apple cultivars (mg/100 ml)

Variable	Local cultivars	Mean	Low. 95%	Upp. 95%	ANOVA p-value and R-Square
Citric	Arapkizi	0.447 ± 0.002 ^A	0.422	0.472	<0.0001 R ² =0.9996
	Aslik	0.109 ± 0.002 ^D	0.089	0.128	
	Bey Elmasi	0.376 ± 0.005 ^B	0.331	0.420	
	Cebegirmez	0.275 ± 0.002 ^C	0.255	0.294	
Fumaric	Arapkizi	1.252 ± 0.128 ^A	-0.374	2.878	<0.0081 R ² =0.9337
	Aslik	0.972 ± 0.004 ^B	0.921	1.023	
	Bey Elmasi	0.634 ± 0.009 ^C	0.520	0.748	
	Cebegirmez	0.718 ± 0.007 ^C	0.629	0.807	
Malic	Arapkizi	4.613 ± 0.008 ^A	4.511	4.715	<0.0001 R ² =0.9935
	Aslik	3.915 ± 0.045 ^B	3.343	4.487	
	Bey Elmasi	3.195 ± 0.045 ^C	2.623	3.767	
	Cebegirmez	3.095 ± 0.075 ^C	2.142	4.048	
Succinic	Arapkizi	0.511 ± 0.004 ^A	0.466	0.555	<0.0001 R ² =0.9990
	Aslik	0.419 ± 0.002 ^B	0.394	0.444	
	Bey Elmasi	0.361 ± 0.007 ^C	0.278	0.443	
	Cebegirmez	0.144 ± 0.003 ^D	0.106	0.182	
Tartaric	Arapkizi	0.130 ± 0.007 ^B	0.047	0.212	<0.0002 R ² =0.9889
	Aslik	0.140 ± 0.003 ^B	0.102	0.178	
	Bey Elmasi	0.075 ± 0.003 ^C	0.037	0.113	
	Cebegirmez	0.214 ± 0.007 ^A	0.125	0.303	

Table 7. Correlations between organic acids and sugars

		Pearson Correlation Coefficients, N = 8							
		Citric	Tartaric	Malic	Succinic	Fumaric	Fructose	Glucose	Sucrose
Citric		1.000	-0.328	0.224	0.239	0.179	-0.749*	-0.781*	-0.783*
	P-value		0.428	0.593	0.568	0.671	0.032	0.022	0.022
Tartaric			1.000	-0.182	-0.652	0.018	-0.289	-0.263	-0.291
	P-value			0.666	0.080	0.967	0.488	0.530	0.485
Malic				1.000	0.844*	0.950*	-0.422	-0.388	-0.300
	P-value				0.008	0.000	0.298	0.342	0.470
Succinic					1.000	0.718*	-0.072	-0.066	0.030
	P-value					0.045	0.865	0.877	0.943
Fumaric						1.000	-0.482	-0.453	-0.365
	P-value						0.227	0.260	0.375
Fructose							1.000	0.990*	0.983*
	P-value							<.0001	<.0001
Glucose								1.000	0.981*
	P-value								<.0001
Sucrose									1.000

*: P ≤ 0.001

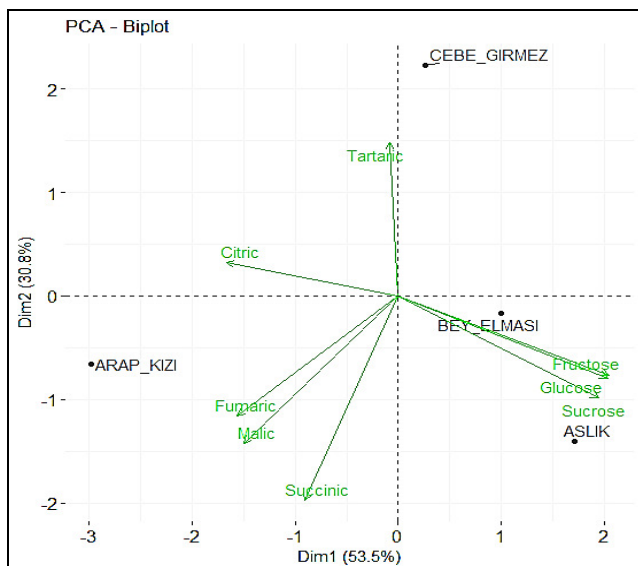


Fig. 3 The distribution of sugar and organic acid according to cultivars

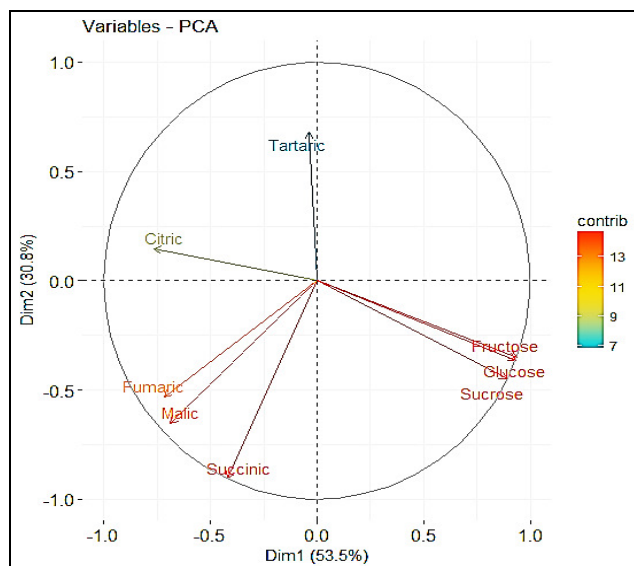


Fig. 4. The correlation of sugar and organic acids

Conclusions

As a result of the study, it was determined that ‘Arapkizi’ cultivar came into prominence the organic acid and phenolic contents. In the study, it was found that phenolic compounds explain 85.1 % of the total variation with two basic components. In terms of the cultivars, phloridzin, ellagic and chlorogenic acids were found to be the most potent compounds while vanillic and *o*-coumaric acid were found to be the lowest potent compounds. Catechin, ferulic and chlorogenic acids have been found to be highly correlated compounds among themselves, and negative correlation with caffeic and routine. In this study, 83.3% of the variation in organic acid and sugar content of the cultivars was explained by two main components. Positive correlations exist between organic acids and the highest positive correlations were observed between succinic, malic and fumaric acids. ‘Aslik’, ‘Bey Elmasi’ and ‘Cebegirmez’ local cultivars have the highest content of sugar while the ‘Arapkizi’ cultivar has been identified rich in organic acids. It was concluded that these two cultivars (‘Aslik’ and ‘Arapkizi’) would be useful for human health. As a result, the production of these cultivars should be increased.

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