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Research Article

Determination of Pomological and Molecular Characteristics of Some Pomegranate (*Punica granatum* L.) Cultivars and Selected Genotypes

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Abstract: In this study, it was aimed to determine the quality, phytochemical contents and molecular characterization of the best thirteen pomegranate genotypes selected as a result of the selection study carried out between 2019-2020 in the İnhisar district of Bilecik province. As a result of the research, the fruit weight of the selected promising genotypes and cultivars was between 208.00 and 601.3 g, fruit width was 74.54-103.47 mm, fruit length was 63.08-92.32 mm, hundred-aril weight was 28.00-66.25 g, aril yield was determined as 35.48-85.00%. The amount of soluble solid was determined between 14.33 and 18.77%, while pH values were between 3.22 and 4.36% and titratable acidity was between 0.23 and 1.72%. The total antioxidant capacity, which was determined with the TEAC method, was 3.28-8.48 µmol TE g^{-1}, while the total amount of phenolic substances was 956.10-2116.10 g GAE kg^{-1}, and the total amount of anthocyanins was 45.50-344 µg Plg-3-glu/g. Seven UBC-ISSR primers were employed to conduct molecular analyses aiming to determine polymorphism levels among the selected thirteen genotypes, along with the comparative Fellahyemez, Katırbaşı, and Hicaznar varieties. The resulting dendrogram is divided into two main clusters at a 25% dissimilarity level, one smaller and the other larger. All local genotypes clustered within the larger group, with Genotype 9 and Genotype 10 exhibiting the closest similarity. When the criteria determined as a result of the study were examined, it was determined that among the selected pomegranate genotypes, there were individuals that could be registered as table and industrial.

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1. Introduction

Pomegranate (*Punica granatum* L.), the most important species of the *Punicaceae* family of the *Myrtales* order, is one of the very old fruit species with subtropical and tropical climate characteristics. It is known that the pomegranate fruit, whose history goes back about seven thousand years, is used by people for food and medicine. In recent years, as a result of the studies carried out in the fields of fruit

growing and breeding techniques, food technology, transportation, and storage, its production and consumption have increased every year (Kahramanoglu, 2019).

The country with the highest pomegranate production worldwide is India and it is followed by Iran, Türkiye, USA, and Iraq. With the establishment of closed pomegranate orchards in Türkiye in the last two decades, production has increased continuously, and in 2022, 681.460 tons of pomegranate have been produced in 58 provinces (TURKSTAT, 2022).

The reason for this increase is that pomegranate is rich in nutritional content, taste, aroma, antioxidants, vitamin C, and phenolic compounds. Studies have shown that pomegranate regulates the digestive system, prevents allergies, cardiovascular, cancer, and diabetes, and reduces blood pressure. It has been reported that it contains a natural source of bioactive components (Teixeira da Silva et al., 2013). It is known that the products with high antioxidant content among the vegetables and fruits that are recommended to be consumed regularly are preferred by consumers, so it has gained importance to determine the phytochemical contents in the selection studies (Montefusco et al., 2021).

It is known that pomegranate cultivars are obtained by selection studies instead of planned breeding studies in countries where there are selection studies for many fruit cultivars cultivated in the world and where pomegranate production is the highest.

Since Türkiye is one of the countries of origin of the pomegranate, there are many cultivars and types of pomegranates. In many regions of Türkiye, pomegranate cultivation is carried out with local cultivars. Various studies have been carried out on some pomegranate cultivars and genotypes in different regions such as the Aegean, Mediterranean region, and Siirt, Hatay, Tokat, Artvin, Hakkari, Bitlis, Şanlıurfa, and Diyarbakır provinces in Türkiye (Caliskan and Bayazit, 2013; Gercekcioglu et al., 2015; İkinci and Kilic, 2016; Akbel, 2017; Öztürk et al., 2019; Kos, 2022).

In recent years, biotechnological applications have also been integrated into breeding studies, known for shortening the breeding duration and enabling the selection of important traits through this method (Simsek and Etik, 2022). Until today, morphological, biochemical, and molecular markers have been extensively employed to define and assess genetic diversity in pomegranates. In many studies, molecular marker techniques such as RAPD, SSR (Orhan et al., 2014; Caliskan et al., 2018), and ISSR have been utilized (Jbir et al., 2014). The ISSR molecular marker technique has been found useful for examining genetic relationships among pomegranate genotypes and varieties, mainly due to its ability to generate a higher number of polymorphic bands (Ismail et al., 2014; Heidari et al., 2016; Almiahy and Jum'a, 2017; Hajiyeva et al., 2018; Al Mousa et al., 2019; Karapetsi et al., 2021).

This study aimed to determine the quality and phytochemical contents, as well as the molecular characterization, of the top 13 genotypes selected using the weighted grading method from among 33 naturally grown pomegranate genotypes in the Inhisar district of Bilecik province, which are well adapted to local climatic conditions.

2. Material and Methods

In 2020, in line with the information received from the Inhisar District Directorate of Agriculture and Forestry in Bilecik, the villages where pomegranate cultivation is carried out were visited and 33 genotypes, which are at the forefront in terms of yield and quality, were determined with the information obtained from the producers. Bilecik Şeyh Edebali University Agricultural Application and Research Center laboratory conducted the determination of pomological and phytochemical characteristics by obtaining three replications from each genotype, with five fruit samples taken in each replication (Akbel, 2017; Cicek et al., 2019; Ozturk et al., 2019).

The 33 selected genotypes were evaluated according to the weighted grading method. In the weighted grading method, 20% points were given for fruit weight and fruit taste, 12% for aril yield, ease of graining, and soluble solids, 10% for juice yield, 9% for seed hardness, and 5% for titratable acid. Genotypes were evaluated accordingly, out of a total of 100 points. As a result of this evaluation, 13 genotypes, which were superior and constituted the material of this study, were determined (Table 1). The standard pomegranate cultivars (Hicaznar, Fellahyemez, Katırbaşı) used for comparison were obtained from the Yalova Horticultural Research Institute.

Table 1. The genotypes selected as a result of the weighted grading method and the location where they were selected

	Genotype/Cultivar	Total score	Latitude	Longitude
1	Genotype 3	735	40°03'35.7"N	30°23'18.3"E
2	Genotype 4	795	40°03'36.7"N	30°23'21.7"E
3	Genotype 5	735	40°03'38.2"N	30°23'20.1"E
4	Genotype 8	718	40°02'33.4"N	30°25'38.7"E
5	Genotype 9	724	40°02'34.0"N	30°25'39.4"E
6	Genotype 10	706	40°02'34.6"N	30°25'41.9"E
7	Genotype 14	747	40°02'44.0"N	30°25'14.4"E
8	Genotype 19	720	40°02'45.1"N	30°25'14.1"E
9	Genotype 20	605	40°03'33.5"N	30°22'54.8"E
10	Genotype 22	848	40°03'33.7"N	30°22'54.0"E
11	Genotype 24	813	40°03'29.0"N	30°22'46.7"E
12	Genotype 27	742	40°03'26.4"N	30°22'44.7"E
13	Genotype 33	718	40°03'54.1"N	30°23'02.7"E
14	Hicaznar	Yalova Horticultural Research Institute		
15	Fellahyemez	Yalova Horticultural Research Institute		
16	Katırbaşı	Yalova Horticultural Research Institute		

Fruit width (mm), fruit length (mm), calyx diameter and length (mm), and peel thickness (mm) were measured with a digital caliper (OEM KMP200 Digital caliper) and expressed in millimeters (mm). Fruit weight (g), hundred-aril weight (g), and seed weight (g) were determined by weighing with a precision balance (Kern PNS600) sensitive to 0.01 g. The total soluble solids content of arils was measured using a digital handheld refractometer and expressed as degrees Brix. Titratable acidity was determined by titration of 5 mL of fruit juice with 0.1 N NaOH and expressed as a percentage of citric acid content (Simsek and Etik, 2022). Peel and aril color were determined with the CR 400 Model Minolta Colorimeter (Konica Minolta, CR400) device. pH was measured by a ph-meter (Hanna edge pH parameter).

Total antioxidant capacity was determined according to the TEAC method, which is frequently used for plant materials, according to Özgen et al. (2008). The amount of total phenol was determined according to the method of Singleton and Rossi (1965) using Folin-Ciocalteu's chemical and the results were calculated as µg gallic acid equivalent/g fresh fruit in gallic acid. The total anthocyanin amount was determined according to the pH difference method according to Giusti and Wrolstad (2005), and the values were calculated as µg anthocyanin / g dry matter.

DNA isolation was performed according to the protocol of Doyle and Doyle (1987). DNA samples were quantified using a spectrophotometer, and their concentration was diluted to 10 ng μl^{-1}. In the PCR application, it was initially planned to use 15 primers that had previously successfully amplified in pomegranate. However, due to the inability to achieve amplification with eight of these primers, the study was conducted using seven primers (UBC807; 808; 811; 826; 835; 889; 891) (Jbir et al., 2014; Almiahy and Jum'a, 2017; Amar and El-Zayat, 2017).

The ISSR PCR reaction was performed with a 23 µL reaction mixture of 2 µL DNA (10 ng μl^{-1}) (Kos, 2022). The PCR products were separated for approximately 2 hours at a constant voltage of 100V using 1X TBE buffer and a 2% agarose gel.

The mean minimum and maximum value analyses of the data obtained as a result of the research were made using the SPSS 16.0 package program. Moreover, biplot plots and correlation analysis were performed using the JMP (2020) program. In the molecular section, the data used in the statistical analysis were scored as one (1) in the presence of ISSR bands and zero (0) in their absence. Similarities and differences between genotypes were studied at the molecular level. Principal Coordinates Analysis (PCoA) was performed using similarity coefficients, and analysis was conducted using the Popgene32 version 1.32 (Population Genetic Analysis) and MEGA 5.0 (Molecular Evolutionary Genetic Analysis) software packages. A UPGMA (Unweighted Pair-Group Method with Arithmetic Average) dendrogram was constructed based on the UPGMA method to visualize the relationships among genotypes.

Statistical analysis was performed using the Minitab 19 package program. The data were submitted for variance analysis and the means were tested by the least significant difference ($p<0.05$).

3. Results and Discussions

Fruit weight (g), fruit width (mm), fruit length (mm), hundred-aril weight (g), aril yield (%), juice volume (%), calyx length (mm), calyx diameter (mm), calyx number, peel thickness (mm), flavour and seed hardness in fruits of promising pomegranate genotypes cultivars are given in Table 2. The mean fruit weight of genotypes and cultivars was determined as 332.1 g. While the lowest fruit weight was obtained in Genotype 3 with 208.00 g, the highest fruit weight was determined as 601.25 g in the Fellahyemez cultivar, and the highest fruit weight among genotypes was determined as 399.80 g in Genotype 8 (Table 2). In studies conducted on pomegranate cultivars and genotypes in different regions, fruit weights have been determined as 267.72-650.56 g by Kilic (2014), 251.01-530.25 g by Gundogdu et al. (2015), 267.72-650.56 g by Ikinci and Kilic (2016), 205.44-525.87 by Boguc (2018), 207.30-689.50 g by Ozturk et al. (2019), and 201.55-637.50 g by Dursun (2021).

As seen in Table 2, fruit width and length means were determined as 84.25 and 74.26 mm, respectively. When the genotypes were examined, it was determined that the width and length ratio of Genotype 9 had the highest value. When the previous studies were examined, the fruit width and length values in the present study were found to be similar to other studies (Okatan et al., 2015; Dursun, 2021; Simsek and Etik, 2022).

It was determined that the hundred-aril weight of the genotypes and cultivars was between 27.00-61.00 g, and the aril yield was between 35.48-85.00% (Table 2). In previous studies, while Akbel (2017) weighed hundred-aril with 30 pomegranate genotypes in the Central Sakarya Basin as 17.50-46.60 g, Özden et al. (2017) determined three pomegranate cultivars in Şanlıurfa province as 32.33-61.20 g, Boguc (2018) found as 36.98-61.81 g in their study in Şırnak province, and Simsek and Etik (2022) determined the hundred-aril weight as 19.77-35.07 g in their study in Diyarbakır province. When the hundred-aril weight results obtained in our study were compared with the previous studies, it was determined that they were in a similar value range. While in the study that was carried out on three pomegranate genotypes grown in the Adana region, Gercekcioglu et al. (2015) found the aril yield as 71.33%-81.17%, Ozturk et al. (2019) determined it as 40.50-78.40% in 18 pomegranate genotypes grown in Mardin districts, Dursun (2021) reported that they found the aril yield between 43.55% and 68.98% in the study they conducted with some pomegranate cultivars in Şanlıurfa province. As a result of our study, it was determined that the aril yield value was higher when compared to previous studies.

The mean fruit juice yield of the genotypes and cultivars included in the study was found to be 39.88%. The lowest juice yield was found in Genotype 27 (25.20%) among the genotypes, and in the Fellahyemez cultivar (24.62%) among the cultivars, while the highest juice yield was found in Genotype 3 with 62.12% among the genotypes (Table 2). In the previous studies, Ozturk et al. (2019) reported that the juice yield of local pomegranate genotypes varied between 32-66% in their study in Mardin province. In addition, the fruit juice yield results obtained in our study were found to be similar to previous studies.

The sensory-evaluated fruit flavours were determined as sourish, sweet-sour, and sweet, and the seed hardness was determined as hard, medium-hard, soft, and very soft.

While the lowest calyx length was measured in Genotype 3 with 12.85 mm, the lowest calyx diameter was measured in Genotype 4 with 19.83 mm. Moreover, the highest calyx length and diameter were determined in Genotype 20 with 16.49 mm and 34.78 mm in the genotypes and cultivars in the study, respectively (Table 2). The calyx length and diameter obtained were similar to those of other researchers (Akbel, 2017; Ozturk et al., 2019; Dursun, 2021).

Peel L, a, b, values of genotypes and cultivars are presented in Table 2. The L* value expressing the peel brightness of the fruit was 28.22-56.85, the a value expressing the change of the fruit peel from green to red colour was 6.39-32.51, and the b value expressing the change of the fruit peel from yellow to blue colour was determined between 16.76 and 29.67. Compared to previous studies, L and the values of the peel were similar (Yaman et al., 2015; Akbel, 2017). It is thought that this difference, in which the b value differs from previous studies, is due to the ecological conditions of the region where the fruits are grown (Akbel 2017; Toprak 2019). It was determined that while the L value of aril colour varied between 7.21 and 22.05, the a value was 13.91-24.62 and the b value was between 4.62 and 16.53 (Table 2). The findings obtained in the study were similar to other studies (Akbel 2017; Toprak 2019).

Table 2. Pomological characteristics of pomegranate genotypes and cultivars

Genotype	Fruit						Calyx		Peel							Aril	
	Weight (g)	Width (mm)	Length (mm)	Hundred-aril weight (g)	Aril yield (%)	Juice volume (%)	Diameter (mm)	Length (mm)	Thickness (mm)	L	a	b	L	a	b	Flavor	Seed hardness
	**	**	**	**	**	**	**	*	**	**	**	**	**	**	**		
Genotype 3	208.0^{i}	79.4^{efg}	71.5^{def}	33.0^{fg}	85.0^{a}	62.1^{f}	20.1^{g}	12.9^{c}	6.4^{bc}	53.5^{ab}	6.4^{g}	29.7^{a}	21.2^{a}	21.4^{cd}	10.7^{c}	Sweet-sour	Medium-hard
Genotype 4	255.0^{h}	77.0^{fg}	63.9^{g}	60.0^{ab}	78.0^{b}	58.4^{d}	19.8^{g}	13.1^{bc}	4.6^{cde}	43.6^{cd}	19.4^{c}	24.9^{bcd}	16.0^{d}	18.3^{ef}	11.4^{bc}	Sweet	Medium-hard
Genotype 5	209.0^{i}	75.3^{fg}	63.1^{g}	28.0^{g}	82.5^{a}	56.8^{g}	20.9^{fg}	12.9^{c}	4.4^{cde}	44.7^{cd}	32.5^{a}	24.4^{cd}	19.9^{ab}	24.0^{ab}	13.8^{ab}	Sweet-sour	Medium-hard
Genotype 8	399.8^{c}	79.6^{efg}	71.8^{def}	57.0^{abc}	43.3^{l}	32.3^{g}	23.4^{def}	15.4^{abc}	4.2^{de}	52.5^{b}	10.9^{f}	27.7^{ab}	22.1^{a}	14.9^{gh}	14.0^{ab}	Sweet	Soft
Genotype 9	338.0^{e}	88.0^{bc}	77.2^{c}	41.0^{ef}	51.1^{fgh}	33.3^{i}	33.5^{a}	14.5^{abc}	4.4^{cde}	43.4^{cd}	17.2^{c}	23.6^{d}	17.5^{bcd}	19.1^{def}	10.6^{c}	Sweet-sour	Medium-hard
Genotype 10	323.4^{f}	84.0^{b-e}	76.7^{c}	45.0^{de}	52.8^{f}	31.3^{h}	28.6^{b}	14.3^{abc}	5.1^{cde}	52.2^{b}	19.2^{c}	26.7^{abc}	16.0^{d}	20.1^{cde}	11.5^{bc}	Sweet	Hard
Genotype 14	275.6^{g}	81.7^{c-f}	73.1^{d}	48.0^{cde}	62.6^{e}	41.1^{g}	28.2^{b}	15.6^{ab}	5.1^{cde}	41.7^{d}	17.1^{cd}	23.0^{d}	13.1^{e}	21.0^{cd}	10.7^{c}	Sweet	Medium-hard
Genotype 19	214.8^{i}	74.5^{g}	68.9^{f}	43.0^{def}	48.6^{hij}	28.6^{l}	26.1^{bcd}	15.4^{abc}	4.6^{cde}	45.5^{c}	13.2^{ef}	24.3^{cd}	11.5^{e}	22.1^{abc}	10.4^{cd}	Sweet	Soft
Genotype 20	313.2^{f}	86.8^{bcd}	76.9^{c}	45.0^{de}	46.4^{jk}	36.8^{j}	34.8^{a}	16.5^{a}	5.9^{bcd}	56.9^{a}	24.1^{b}	28.3^{a}	17.3^{bcd}	21.2^{cd}	11.3^{bc}	Sweet-sour	Medium-hard
Genotype 22	246.6^{h}	79.3^{efg}	69.4^{ef}	42.0^{def}	74.8^{c}	53.0^{e}	26.5^{bc}	14.0^{abc}	4.8^{cde}	53.2^{b}	23.1^{b}	28.3^{a}	19.4^{abc}	15.5^{gh}	9.6^{cd}	Sweet	Soft
Genotype 24	269.0^{g}	79.8^{d-g}	72.4^{de}	44.0^{def}	66.0^{d}	59.0^{c}	22.4^{efg}	15.3^{abc}	4.4^{cde}	53.7^{ab}	7.5^{g}	27.5^{ab}	20.4^{a}	24.6^{a}	16.5^{a}	Sweet	Soft
Genotype 27	345.2^{de}	88.0^{bc}	74.8^{cd}	40.0^{ef}	51.4^{fg}	25.2^{g}	22.7^{efg}	13.5^{bc}	3.5^{e}	46.5^{c}	14.3^{de}	27.7^{ab}	17.2^{cd}	21.8^{bc}	10.7^{c}	Sweet	Soft
Genotype 33	354.0^{d}	79.7^{efg}	69.6^{ef}	53.0^{bcd}	35.5^{m}	27.4^{k}	25.0^{cde}	13.5^{bc}	4.4^{cde}	54.9^{e}	17.8^{c}	29.9^{d}	16.9^{bcd}	16.0^{h}	8.6^{d}	Sweet	Soft
Hicaz Nar	568.2^{b}	101.4^{a}	92.3^{a}	38.0^{efg}	49.4^{ghi}	36.5^{a}	24.9^{cde}	14.3^{abc}	7.8^{ab}	28.2^{f}	30.5^{a}	16.8^{e}	7.2^{f}	14.1^{h}	4.6^{e}	Sweet-sour	Medium-hard
Fellahyemez	601.3^{a}	103.5^{a}	91.6^{a}	66.0^{a}	46.9^{ij}	24.6^{b}	24.0^{cde}	13.1^{bc}	8.7^{a}	35.4^{e}	17.8^{c}	22.8^{d}	17.3^{bcd}	13.9^{h}	7.7^{d}	Sweet	Medium-hard
Katırbaşı	393.0^{c}	90.1^{b}	83.3^{b}	60.0^{ab}	44.1^{kl}	31.6^{hi}	28.1^{b}	16.3^{a}	9.8^{a}	46.0^{c}	10.8^{f}	29.1^{a}	21.3^{a}	16.7^{fg}	9.5^{cd}	Sweet	Soft
Mean	332.1	84.3	74.8	45.8	57.4	39.9	25.6	14.4	5.5	45.8	17.6	25.5	17.2	18.9	10.7		
Min.	208.0	74.5	63.1	27.0	35.5	23.6	19.8	12.9	3.5	28.2	6.4	16.8	7.2	13.9	4.6		
Max.	601.3	103.5	92.3	61.0	85.0	63.2	34.8	16.5	9.8	56.9	32.5	29.7	22.1	24.6	16.5		

*: Significant at the p<0.05 probability level, **: Significant at the p<0.01 probability level.

It is known that the amount of soluble solid is important in terms of quality criteria since it determines the amount of sugar in the fruit content and fruits with high sugar content are demanded by consumers. In the study, the amount of soluble solids varied between 14.33-18.77%, and while the lowest amount of soluble solids was determined in Genotype 33 (14.67%), the highest was observed in Genotype 22 (18.77%). Among the cultivars, the lowest amount of soluble solid was found in the Fellahyemez cultivar with 14.33%, and the highest amount of soluble solid was found in the Hicaznar cultivar with 15.7% (Table 3).

In the previous studies on the soluble solid value of pomegranate, while Akbel (2017) found the soluble solid value between 15.60-24.00% in the study conducted in the Central Sakarya Basin, Boguc (2018) determined the soluble solid value between 15.90-18.20% in the study conducted with pomegranate cultivars and genotypes in Sırnak province. Furthermore, while Dursun (2021) found the soluble solid value between 14.60-16.60% in a study conducted with different pomegranate cultivars in Sanlıurfa province, Cicek et al. (2019) found the soluble solid value of ten pomegranate genotypes between 15.00-21.00% in their study in the districts of Diyarbakır province, and Öztürk et al. (2019) found that the soluble solid values of 18 pomegranate genotypes between 15.00-18.00% in their study in the districts of Mardin province. The values we found are similar to previous studies.

Table 3. Amount of soluble solid (%) of selected promising genotypes and cultivars

Genotype	TSS	pH	TA	TAA	TMA	TPA
	**	ns	**	**	**	**
Genotype 3	15.9^{b-e}	3.48	0.47^{cde}	5.22^{c-f}	104.7^{f}	1917.8^{cd}
Genotype 4	16.4^{a-e}	3.37	0.50^{bcd}	5.60^{cd}	72.4^{g}	1927.8^{c}
Genotype 5	17.8^{abc}	3.38	0.47^{cde}	6.80^{b}	116.6^{f}	2116.1^{a}
Genotype 8	16.8^{a-e}	3.33	0.58^{bc}	4.36^{efg}	53.6^{h}	1663.6^{fg}
Genotype 9	17.3^{abc}	3.39	0.59^{bc}	5.38^{cde}	142.2^{de}	2014.5^{abc}
Genotype 10	16.7^{a-e}	3.27	0.61^{b}	5.20^{c-f}	160.7^{bc}	1779.5^{ef}
Genotype 14	18.4^{ab}	3.41	0.54^{bc}	8.48^{a}	108.7^{f}	2104.5^{a}
Genotype 19	16.9^{a-e}	3.35	0.58^{bc}	6.16^{bc}	131.0^{e}	1973.6^{bc}
Genotype 20	15.8^{b-e}	3.43	0.53^{bcd}	5.56^{cd}	152.5^{cd}	1802.0^{de}
Genotype 22	16.7^{a-e}	3.37	0.46^{cde}	5.19^{c-f}	172.7^{b}	1567.0^{g}
Genotype 24	17.0^{a-d}	3.37	0.54^{bc}	6.65^{b}	83.8^{g}	2079.5^{ab}
Genotype 27	18.8^{a}	3.98	0.40^{de}	4.27^{fgh}	75.5^{g}	1430.3^{h}
Genotype 33	14.7^{de}	3.44	0.52^{bcd}	4.63^{d-g}	81.7^{g}	1390.3^{h}
Katırbaşı	14.5^{de}	3.24	0.36^{ef}	3.28^{gh}	149.8^{a}	1119.5^{ef}
Fellahyemez	14.3^{e}	4.36	0.23^{f}	6.15^{bc}	47.5^{h}	956.1^{j}
Hicaznar	15.7^{cde}	3.22	1.72^{a}	3.61^{h}	344.6^{cd}	1729.5^{i}
Mean	16.5	3.46	0.57	5.41	124.9	1723.2
Min.	14.3	3.22	0.23	3.28	47.5	956.1
Max.	18.8	4.36	1.72	8.48	344.6	2116.1

*: Significant at the p<0.05 probability level, **: Significant at the p<0.01 probability level, ns: non-significant, TSS: Total soluble solid (%); pH; TA: Titratable acidity (%); TAA: Total antioxidant amount (µmol TE g^{-1}); TMA: Total anthocyanin content (µg Plg-3-glu g^{-1}); TPA: Total phenol amount (g GAE kg^{-1}).

In the study, pH values were found to be between 3.22 and 4.36. The lowest pH value was found in the Katırbaşı cultivar as 3.22, while the highest pH value was found in the Fellahyemez cultivar as 4.36 (Table 3). In the previous studies conducted on the pH value of pomegranates are examined, while Gündogdu et al. (2015) observed as 3.45-4.71 in the study they conducted with Silifke aşı and Hicaznar cultivar, and Boguc (2018) stated that the pH value of pomegranates cultivars between 3.57-3.96 in the study conducted with Hicaznar and four local cultivars.

Titratable acidity (TA) values of genotypes and cultivars were found to be between 0.23-1.72%. The lowest TA value was found in the Fellahyemez cultivar with 0.23%, while the highest value was found in the Katırbaşı cultivar with 1.72% (Table 3). In the previous studies on the titratable acidity value of pomegranate, Dursun (2021) observed between 0.67 and 2.74 in a study conducted with Hicaznar, Katırbaşı, Devedişi, Suruc, and Suruc Karası cultivars in Şanlıurfa province. The taste of

pomegranate juices is associated with titratable acidity values. The titratable acidity of pomegranate juice is known as Sweet Pomegranate with less than 1%, Sourish Pomegranate with 1-2%, and Sour Pomegranate with more than 2%. Accordingly, in the results we found, the taste of pomegranate genotype and cultivars can be evaluated as sweet.

The relationship analysis performed is given in Table 4. According to the analysis, there was a significant and positive relationship between weight and width (r=0.911**), length (r=0.885**), juice yield (r=0.648**), and peel thickness (r=0.599*). Besides, there was a negative and significant relationship between weight and aril yield (r=-0.606*), peel colour L value (r=-0.615*), peel colour b value (r=-0.512*), aril colour a value (r=-0.701**), aril colour b value (r=-0.593*), and total soluble solid (r=-0.605*) (Table 4). Furthermore, a significant and positive relationship was determined between peel thickness (PT) and weight (r=0.599*), width (r=0.692**), and length (r=0.760**). The priority feature of pomegranate in both table consumption and processing in the food industry is the juice yield (Gündogdu et al. 2015). A significant and positive relationship was found between juice yield, which was obtained by dividing juice weight by fruit weight, weight (r=0.648**), width (r=0.685**), and length (r=0.611*).

It is known that TSS, pH, and titratable acidity ratios can be caused by characteristics of the variety as well as being affected by climate, soil, and cultural practices. It was expected that there were differences between the cultivars and genotypes examined in the study. There was a significant and positive relationship between TSS and aril colour a value (r=0.679**) and aril colour b value (r=0.606*) while there was a significant and negative relationship between TSS and weight (r=-0.605**), width (r=-0.511**), length (r=-0.549*), and peel thickness (r=-0.658**).

When Table 4 is examined, there was significant and positive relationship between the peel colour L value (PCL) and the peel colour b value (r= 0.895**), the aril colour L value (r=0.623*), the aril colour a value (r= 0.511*) and aril colour b value (r= 0.721**) (Table 4).

When the total phenolic substance content of the genotypes and cultivars was examined, it was determined that it ranged from a minimum of 940.3 g GAE kg^{-1} to a maximum of 2205.30 g GAE kg^{-1}. While the lowest amount of phenolic substance was observed in Genotype 33 (1390 g GAE kg^{-1}) among the genotypes, it was observed in the Fellahyemez cultivar (956.11 g GAE kg^{-1}) among the cultivars. Moreover, the highest value was determined in Genotype 5 (2116.11 g GAE kg^{-1}). Ozgen et al. (2008) reported that the total phenolic content of six pomegranate cultivars grown in Türkiye was between 1245-2076 g GAE kg^{-1}, Akhavan et a. (2015) found between 943-2931 g GAE kg^{-1}, Okumus (2016) determined that the total phenolic substance content of Wonderful and Hicaznar cultivars was 1156.67-1428.1 g GAE kg^{-1}, and Akbel (2017) reported that the total phenolic content of pomegranate genotypes in the Central Sakarya Basin ranged from 551 to 3282 g GAE kg^{-1}.

Like other fruits, the physical and chemical properties of pomegranate, its phenolic content, and therefore its antioxidant activity can vary according to many factors such as variety, maturity, climate, growing region, and cultural practices. For these reasons, it is thought that the results obtained from the genotypes and cultivars in the study are partially similar as they are in the range of values in the literature.

In the study, the TEAC method was used to determine the antioxidant capacity of pomegranates. The total antioxidant capacity of genotypes and cultivars was determined at a mean of 5.40 µmol TE g^{-1}. The lowest antioxidant capacity was found in Katırbaşı (3.28 µmol TE g^{-1}) cultivar among the cultivars and in Genotype 27 (4.27 µmol TE g^{-1}) among the genotypes. Furthermore, the highest antioxidant capacity was determined in Genotype 14 (8.48 µmol TE g^{-1}).

The total anthocyanin content of the genotypes was determined as 124.88 µg Plg-3-glu g^{-1} ta between genotypes and cultivars. The lowest amount of anthocyanin among genotypes was observed in Genotype 8 (53.6 µg Plg-3-glu g^{-1} ta), and the highest amount of anthocyanin was in Genotype 22 (172.67 µg Plg-3-glu g^{-1} ta). Moreover, among cultivars, the Hicaznar cultivar (344.55 µg Plg-3-glu g^{-1} ta) was observed with the highest amount of anthocyanin (Table 3).

The classification of the traits examined in the study according to genotypes and the change of genotypes according to the traits are given in Figure 1. In the biplot graph, if the angle between the vectors is less than 90, it shows that the performance of that genotype is better than the mean, if the angle between the vectors is greater than 90, the performance of the genotype is lower than the mean, and if the angle is equal to 90, it is close to the mean (Yan and Tinker, 2006).

Table 4. Correlation coefficients and significance levels of the analyzes

	Wg	Wd	L	HAW	AY	JY	CD	CL	PT	PCL	PCA	PCB	ACL	ACA	ACB	TSS	pH
Wd	0.911**																
L	0.885**	0.963**															
HAW	0.480	0.290	0.287														
AY	-0.606*	-0.454	-0.519*	-0.472*													
JY	0.648**	0.685**	0.611*	0.192	0.168												
CD	0.110	0.246	0.308	0.041	-0.503*	-0.306											
CL	0.001	0.010	0.179	0.192	-0.467	-0.312	0.645**										
PT	0.599*	0.692**	0.760**	0.370	-0.214	0.468	0.128	0.165									
PCL	-0.615*	-0.531*	-0.457	-0.194	0.304	-0.372	0.131	0.317	-0.326								
PCA	0.168	0.198	0.076	-0.296	0.098	0.182	0.154	-0.212	0.008	-0.387							
PCB	-0.512*	-0.434	-0.384	0.007	0.236	-0.35	-0.007	0.202	-0.133	0.895**	-0.550						
ACL	-0.290	-0.325	-0.338	0.111	0.227	-0.122	-0.171	-0.049	-0.081	0.623*	-0.440	0.789**					
ACA	-0.701**	-0.498	-0.48	-0.579*	0.465	-0.322	-0.050	0.108	-0.425	0.511*	-0.146	0.356	0.119				
ACB	-0.593*	-0.629**	-0.604*	-0.153	0.362	-0.233	-0.168	0.190	-0.533*	0.721**	-0.346	0.585*	0.588*	0.677**			
TSS	-0.605*	-0.511*	-0.549*	-0.401	0.342	-0.333	-0.012	0.091	-0.658**	0.419	-0.111	0.324	0.147	0.679**	0.606*		
pH	0.427	0.477	0.336	0.288	-0.127	0.419	-0.191	-0.426	0.139	-0.228	-0.092	-0.043	0.069	-0.127	-0.194	-0.09	
TA	-0.056	-0.034	0.089	0.281	-0.230	-0.295	0.252	0.541*	0.449	0.164	-0.314	0.374	0.310	-0.042	0.055	-0.024	-0.428

*p<0.05 and **p<0.01 are significant.

Wg: weight (g); Wd: width (mm); L: length (mm); HAW: Hundred-aril weight (gr); AY: Aril yield (%); JY: Juice yield (%); CD: Calyx diameter (mm); CL: Calyx length (mm); PT: Peel thickness (mm); PCL: Peel color L; PCA: Peel color a; PCB: Peel color b; ACL: Aril color L; ACA: Aril color a; ACB: Aril color b; TSS: Total soluble solid (%);Ph (%); TA: Titratable acidity.

When Figure 1 is examined, it can be seen which genotypes have higher values in terms of the characteristics discussed, and whether these characteristics are positively or negatively related to each other. In Biplot analysis, Major component 1 was 37.2% and Major component 2 was 18.2%, constituting 55.4% of the variation in total (Figure 1).

Hicaznar and Fellahyemez cultivars, which are high in weight, came to the fore in terms of width, length, juice yield, pH, peel thickness, and hundred-aril weight. As seen in Figure 1, in the correlation analysis performed between the features in the same group, it was determined that the relationship between these features was significant and positive at the 1% and 5% levels (Table 4). A strong positive correlation was found between titratable acid amounts, total phenol amount, calyx diameter, and calyx length for the Katırbaşı cultivar included in the study. Besides, it was determined that there was a very strong positive correlation (<90) between total antioxidant amount and aril yield for Genotype 3, Genotype 4, and Genotype 5.

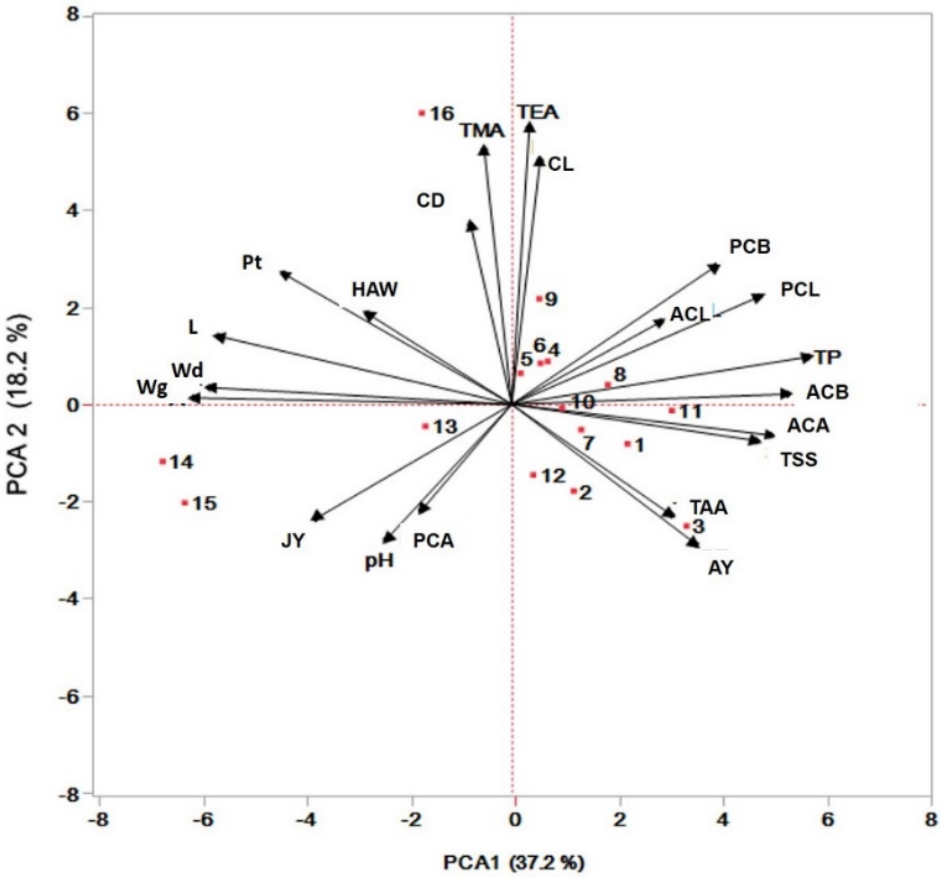

Figure 1. Grouping the examined traits with the biplot analysis method and the relationship of genotypes with the examined traits.

(Wg: weight (g); Wd: width (mm); L: length (mm); HAW: Hundred-aril weight (gr); AY: Aril yield (%); JY: Juice yield (%); CD: Calyx diameter (mm); CL: Calyx length (mm); PT: Peel thickness (mm); PCL: Peel color L; PCA: Peel color a; PCB: Peel color b; ACL: Aril color L; ACA: Aril color a; ACB: Aril color b; TSS: Total soluble solid (%); pH; TA: Titratable acidity (%); TAA: Total antioxidant amount; TMA: Total anthocyanin content; TPA: Total phenol amount).

In the PCR analysis, seven ISSR primers were utilized, resulting in the formation of 51 bands, of which 41 were found to be polymorphic. The number of bands obtained from the ISSR primers ranged from five to twelve, with an average of 7.29 bands and an average of 5.86 polymorphic bands. Among the used primers, the highest band count was obtained from primer 808 (12 bands), while the lowest band count was from primers 811 and 891 (5 bands each). The lowest polymorphism rate was determined to be 60.00% in primer 811. The average polymorphic band ratio among the seven primers was found to be 80.39%.

As seen in Figure 2, the dendrogram depicts two main clusters at a similarity level of 25%, one larger and the other smaller. The smaller cluster includes the varieties Hicaznar and Fellahyemez. The

larger cluster further divides into two subgroups at a 19% level of dissimilarity. Within these subgroups, the Katırbaşı variety forms one branch, while the other branch comprises various genotypes.

In the study, the closest similarity (2%) among the used genotypes was determined between Genotype 9 and Genotype 10. It was observed that these genotypes were selected from neighboring locations.

During the survey conducted in the İnhisar district, it was established that the grown pomegranates differed from one another. Pomegranates referred to by local farmers as Devedişi were designated as Genotypes 14, 19, and 20, and it was noted that these genotypes exhibited similarity in the dendrogram.

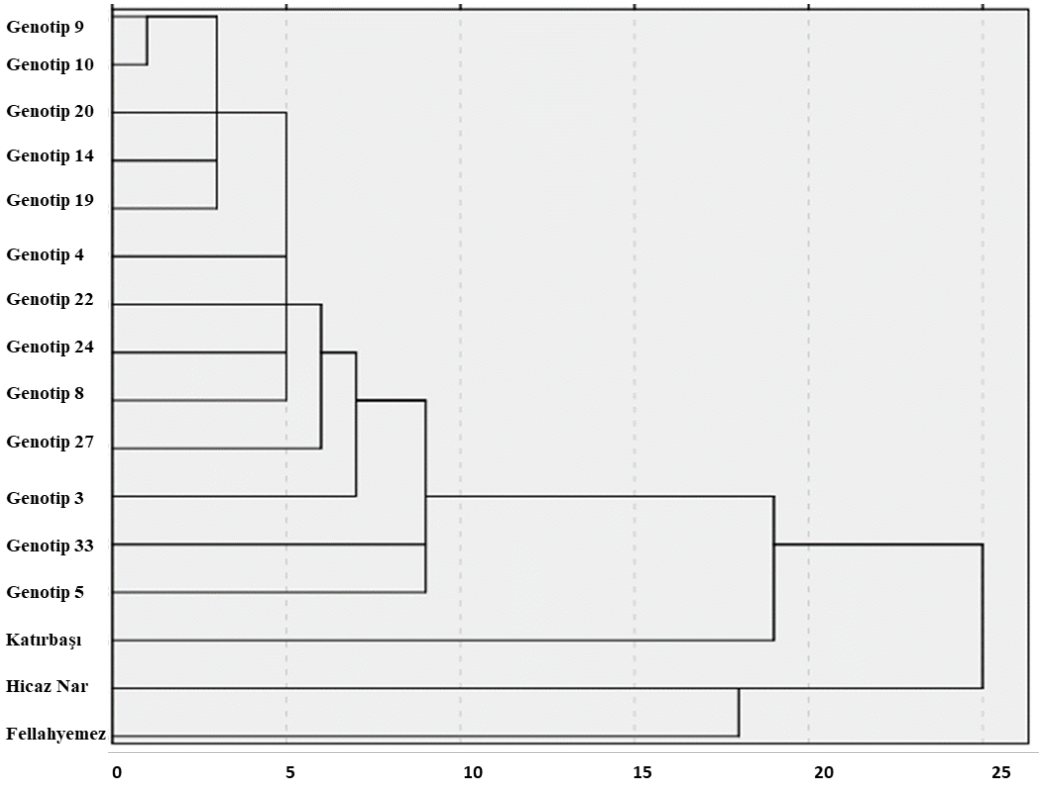

Figure 2. Dendogram obtained by cluster analysis.

Conclusion

It is known that many fruit cultivars cultivated in the world are found by selection studies and in countries where pomegranate production is most common, pomegranate cultivars are obtained by selection studies instead of planned breeding studies. In this study, it was aimed to determine the quality and phytochemical analyzes of selected pomegranate genotypes.

It is known that weight, taste, and seed hardness are important criteria in genotypes selected as promising in previous studies. When the fruit weights and sizes were examined in the study, it was determined that the standard pomegranate cultivars were heavier and more voluminous than the genotypes on the mean. Moreover, fruit weights and sizes are affected by environmental factors and cultural practices as well as depending on the cultivar.

Besides, Genotype 8, Genotype 33, and Genotype 27 can be considered promising table genotypes since they have higher fruit weights compared to other genotypes, as well as being sweet and having soft seed hardness.

Although the fruit sizes of the genotypes are generally smaller than the standard cultivars, the high fruit juice yields of the genotypes allow these types to be used effectively in the fruit juice processing industry. The priority feature of pomegranate in its industrial use is its fruit juice efficiency. As a result of the study, the aril and juice yields of Genotype 3, Genotype 4, and Genotype 5 were found

to be higher than the other genotypes. According to these results, the use of genotypes in the juice processing industry can be evaluated.

When the fruit taste and seed hardness of the genotypes and cultivars in the study are compared with the studies in the literature, it is thought that the fruit taste is between sweet and sweet-sour, the seed hardness is medium and soft, and it is suitable for both table consumption and fruit juice production.

The study is thought to be a guide for researchers and producers in the process of standardizing promising genotypes and expanding their commercial production.

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