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Research Paper / Araştırma Makalesi

Variation in Sugar, Organic Acid and Volatile Flavor Compounds of Watermelon (*Citrullus lanatus*) Grafted on Different Rootstocks at Different Harvest Time

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ABSTRACT

Grafting is used as a viable option for disease control in watermelon production; however, this process can affect quality parameters of the fruits. The aim of this study was to determine sugar, organic acid and volatile flavor compounds of grafted Crisby and Crimson Tide watermelon cultivars at 2 harvest times. In the study, the effect of two watermelon cultivars, three rootstocks and two harvesting time on some of the fruit quality characteristics were determined. Results showed that the quality parameters of samples varied based on the cultivar, rootstock and harvest time. The ranges for glucose, fructose and sucrose contents of fruits were 2.31-2.52%, 3.71-4.01%, 0.70-1.69%, respectively. Organic acids of the samples were composed of citric, acetic, malic, tartaric and oxalic acids and their respective ranges were 135.25-195.13 mg/kg, 97.00-124.13 mg/kg, 67.50-151.50 mg/kg, 61.00-85.38 mg/kg, 22.75-36.25 mg/kg. The main flavor components of samples were *trans*-2-nonenal, *cis*-6-nonen-1-ol, nonanal and 6-methyl-5-hepten-2-one.

Keywords: Watermelon, *Citrullus lanatus*, Grafting, Harvesting time, Quality

Karpuzun *(Citrullus lanatus)* Şeker, Organik Asit ve Uçucu Aroma Bileşimi Üzerine Aşılı Fide Kullanımı ve Hasat Zamanının Etkileri

ÖZ

Aşılı fide kullanımı, karpuz üretiminde hastalık kontrolünde uygulanabilir bir seçenek olarak değerlendirilmektedir. Bununla birlikte, bu işlem meyvenin kalite özelliklerini etkileyebilmektedir. Araştırma, aşılı fide kullanımı ve hasat zamanının Crisby ve Crimson Tide karpuz çeşitlerinin şeker, organik asit ve uçucu aroma bileşimleri üzerine etkisini belirlemek amacıyla yapılmıştır. Çalışmada iki karpuz çeşidi, üç anaç ve iki hasat zamanın etkisi araştırılmıştır. Araştırma bulguları incelenen kalite parametrelerinin kullanılan çeşit, anaç ve hasat zamanına göre değiştiğini göstermiştir. Örneklerin glukoz, fruktoz ve sakkaroz içerikleri sırasıyla %2.31-2.52,%3.71-4.01,%0.70-1.69 aralığında değişim göstermiştir. Örneklerin organik asit kompozisyonu, sitrik, asetik, malik, tartarik ve okzalik asitlerden oluşmakta ve bu bileşenlerin miktarı sırasıyla 135.25-195.13 mg/kg, 97.00-124.13 mg/kg, 67.50-151.50 mg/kg, 61.00-85.38 mg/kg, 22.75-36.25 mg/kg aralığında değişim göstermektedir. Örneklerin ana uçucu aroma bileşenleri, trans-2-nonenal, cis-6-nonen-1-ol, nonanal ve 6-metil-5-hepten-2-one olarak tespit edilmiştir.

Anahtar Kelimeler: Karpuz, *Citrullus lanatus*, Aşılama, Hasat zamanı, Kalite

INTRODUCTION

Watermelon is one of the most cultivated vegetables from the tropics to temperate zones worldwide [1]. Grafting is becoming highly popular in watermelon production and other vegetables [2]. Although watermelon grafting was first adopted to limit effects of *Fusarium* wilt, reasons for grafting have increased over the years. Grafted seedlings have been used to: induce resistance against low and high temperatures; enhance nutrient uptake; improve yield when plants are cultivated in infected soils; increase synthesis of endogenous hormones; improve water use; increase flower and seed production; and enhance tolerance to drought, salinity and flooding [2, 3].

Grafting and rootstock/scion combinations could affect quality parameters of vegetables produced in this manner [4]. Watermelon has been grafted on to pumpkin rootstocks (*Cucurbita* and *Lagenaria*). Use of grafted seedlings on *Lagenaria* rootstock was more successful than *Cucurbita* rootstock. Use of grafted seedlings has positive effects on plant growth and yield but not on water soluble dry matter and fruit weight of watermelon [5]. Alan et al. [6] studied effects of different rootstocks on plant growth, fruit yield, fruit index, peel thickness and water soluble dry matter parameters by comparing grafted with non-grafted plants for 'Crisby'. Water soluble dry matter was ranged from 9.70 to 11.13°brix for non-grafted plants and 9.28 to 10.20°brix for grafted plants. Çandır et al. [7] found that sugar and organic acid composition of 'Crimson Tide' grafted on different rootstocks ranged for: glucose (1.95-3.14%), fructose (2.69-3.79%), sucrose (1.58-4.69%) as sugar and malic (0.327-0.457%), citric (0.042-0.127%) as organic acid components. There have been some studies on flavour compounds of non-grafted watermelons [8-13] that determined aldehyde, alcohol, ketone, furan compounds as main flavour compounds for watermelon. Taste of any fruit is mainly composed by organic acids, sugars and volatile flavour compounds. There is no study on determination flavour compounds for grafted watermelon. Another important parameter for watermelon is harvest time. Growers want to harvest the fruit as possible as early to earn more profit because of the high value of early season product. These fruits could be lacking in taste.

The study was undertaken to determine the influence of cultivars, rootstocks and harvest time on the organic acids, sugar composition, and volatile flavour compounds of watermelon cultivars.

MATERIALS AND METHODS

Materials

This research was carried out on the Food Technology Department of Batı Akdeniz Agricultural Research Institute, Antalya (36º56ˈ north, 30º53ˈ east, altitude: 10 m), Turkey. The cultivars Crisby and Crimson Tide F1, which are widely grown in Turkey, were used. Argentario (*Lageneria*), Maximus and RS841 (*Cucurbita maxima*×*Cucurbita moschata* hybrid) were used as rootstock. Non-grafted Crisby and Crimson Tide were used as controls. Grafting was performed with the side graft technique when seedlings had 4 true leaves. After grafting, these plants were maintained for 20 days in an unheated seedling greenhouse, and then grafted and non-grafted plants were transplanted to the field into loamy soil. Seedlings were established on 5 March 2012 at a 3 m row spacing and 80 cm between plants in the same row. Plants were irrigated with drip irrigation on 3-days intervals. The emitter distance was 50 cm with a mean discharge of 4.0 L/h. Soil infiltration rate for this area was between 8 and 34 mm/h. Average total annual rainfall, daily temperature, total evaporation and relative humidity values were 1132.9 mm, 18.2°C, 1913.5 mm and 63%, respectively. Fruits were harvested on 12 and 22 July 2012 and transferred immediately to the laboratory. The fruits were harvested and pooled from 3 plants. Three fruits were used for each replication and analyses were carried out in parallel.

Methods

To determine effects of grafting; sugar, organic acid contents and volatile flavour compounds were analysed. Analyses were carried out using all edible red parts of the fruit. To extract organic acids, the pulp was mixed with ultra-pure water (1:5 w:v), homogenized with an ultra-turrax (IKA T 25, Staufen, Germany) in an ice bath for 1 min and centrifuged at 3857 *g* (Hettich Universal 32R, Tuttlingen, Germany) for 10 min at 4°C. The supernatant was filtered through a 0.45 µm membrane filter (Macherey-Nagel, Düren, Germany) before injection. Organic acids of this filtrate were analysed by LC 20 AT model HPLC system (Shimadzu, Kyoto, Japan) with a diode array detector (DAD, SPD-M20A) [14]. Separation of organic acids was with an Inertsil C_{18} column (5 µm, 250 × 4.6 mm, GL Science, Tokyo, Japan). The HPLC elution was done at 30°C with isocratic flow of 2% KH_2PO_4 as mobile phase at a 0.9 mL/min flow rate and the chromatogram monitored at 214 nm. Calculation of each organic acid was based on the external standard method from the peak area by analytical interpolation in the calibration curve. Sugar extracts were obtained with the same procedure for organic acids. Sugar contents of these extracts were analysed by HPLC with a refractive index detector (RID 10A). Sugar separation was performed on an Inertsil NH_2 column (5 µm, 250 × 4.6 mm, GL Science). The HPLC elution was with isocratic flow of acetonitrile:water (70:30 v:v) at 25°C [14].

Volatile flavour compounds were determined by SPME (Solid Phase Micro Extraction) headspace methods [15] with a gas chromatograph (Agilent GC 7890A)-mass spectrometry (Agilent MS 5975C, California, USA) instruments using capillary column (HP Innowax Capillary; 60.0 m × 0.25 mm × 0.25 µm). All extractions were carried out at 40°C for 20 min using a 100 µm polydimethylsiloxane (PDMS) SPME filter. Helium (99.9%) was the carrier gas at a constant flow rate of 1 mL/minute. Oven temperature was programmed as follows: 60°C for 10 min, increased at 20°C/min to 250°C, and held at 250°C for 8 min. MS spectra were monitored at 35-450 amu. Percent of components was calculated from GC-FID peak areas, and identified by WILEY NIST, and FLAVOR libraries.

Statistical Analysis

The data were subjected to analysis of variance using SAS (ver. 6.12, SAS, Inc., Cary, NC). The experiment was conducted in a randomized complete block design with three replications. Research findings were evaluated by Duncan's multiple range test. Results were given as mean ± standard error (SE).

RESULTS AND DISCUSSION

Sugar and organic acid contents of vegetables are quality parameters affecting taste for fresh consumption. Their amounts in vegetables are affected by cultivar, harvest time, grafting, ecology, cultural practices and others [16, 17]. The sugars, identified and quantified in all samples, were glucose, fructose and sucrose were identified and quantified in all samples (Table 1).

The major sugar was fructose (Table 1). Yau et al. [18] found that fructose content was higher than glucose and sucrose contents in watermelon. Glucose and sucrose could be dominant sugar components for some other watermelon cultivars [19]. In the present study, differences in fructose content of the samples were significant. Fructose contents of the non-grafted plants were lower than the grafted plants for each rootstock. The highest fructose content was determined for RS 841 followed by Argentario and Maximux rootstocks. The difference was not significant for cultivars. The average ratio of fructose decreased slightly from the first to second harvest, but it was not significant.

Table 1. Changes in the sugar composition (%) of watermelons (mean±SE)*.

Factors	Sample	Glucose	Fructose	Sucrose	Total Sugar
Rootstock	Agentario	2.52^{a}±0.029	3.90^{b}±0.045	1.17^{b}±0.265	7.59^{a}±0.284
	Maximus	2.47^{ab}±0.035	3.82^{c}±0.035	1.12^{b}±0.231	7.40^{b}±0.187
	RS 841	2.47^{ab}±0.031	4.01^{a}±0.068	1.12^{b}±0.194	7.61^{a}±0.140
	Crisby (NG[a])	2.31^{c}±0.049	3.77^{dc}±0.035	1.40^{a}±0.105	7.48^{b}±0.079
	Cr. Tide (NG)	2.43^{b}±0.032	3.71^{d}±0.021	1.34^{a}±0.095	7.48^{b}±0.080
Cultivar	Crisby	2.45^{a}±0.029	3.86^{a}±0.032	1.18^{a}±0.133	7.50^{a}±0.114
	Cr. Tide	2.46^{a}±0.022	3.87^{a}±0.047	1.21^{a}±0.149	7.55^{a}±0.141
Harvest Time	1	2.47^{a}±0.018	3.93^{a}±0.043	0.70^{b}±0.076	7.09^{b}±0.058
	2	2.44^{a}±0.032	3.81^{b}±0.030	1.69^{a}±0.037	7.95^{a}±0.075

[a]NG: Non-grafted, *: Values in a column followed by different superscript letters are significantly (*P <0.05*) different (Duncan's multiple range test)

Another invert sugar was glucose for watermelon samples, and its amount varied with respect to the rootstocks. Glucose contents of the non-grafted Crisby and Crimson Tide were lower than the grafted ones on three different rootstocks. The glucose content differences for cultivars and harvest times were not significant.

Sucrose was the lowest sugar component in all samples. Rootstock and harvest time significantly affected sucrose contents of the samples while cultivars had no significant effect (Table 1). Sucrose contents of the non-grafted plants were higher than the grafted plants. The highest value was determined for the non-grafted Crisby, while the lowest value was obtained for Maximus and RS 841 rootstocks. Sucrose content of the samples increased approximately 2-fold from the first to second harvest. For all samples, total sugar contents of the second harvest were higher than for the first harvest, probably due to the ripening stage. Yau et al. [18] also reported sucrose content of watermelon increased throughout the ripening process. Soteriou et al. [20] reported that fructose and glucose content of watermelon decreased by increasing ripening stage. These findings agree with our results. Sucrose content increased with maturity. Çandır et al. [7] found sucrose was plentiful (1.58-4.61%) in watermelon fruit from grafted watermelon. Their reported levels were higher than results here because of the cultivar differences. Yoo et al. [21] found that sucrose, glucose and fructose contents of watermelon genotypes ranged between 0.22-4.15%, 0.86-2.97%, 1.51-5.05%, respectively. Our results were similar with these values.

The acidity in watermelon is generally low but it affects the taste of watermelon. Organic acid composition of cultivars, depends on rootstock cultivars and harvest time (Table 2). Five different organic acids were detected and quantified in all samples. The amounts of citric, acetic and malic acids contents were higher than the amount of tartaric and oxalic acids.

Citric acid was the most common organic acid, and it was higher in the grafted plants than the non-grafted ones. The highest citric acid was determined in Argentario followed by RS 841 and Maximus rootstocks. Citric acid content differences for cultivars were significant. Crimson Tide was higher citric acid than Crisby cultivar. Citric acid contents of the samples slightly decreased from the first harvest to the second harvest time. Similarly, acetic acid contents of the grafted plant's fruits were higher than the non-grafted ones. Crimson Tide had higher acetic acid content than Crisby cultivar. Additionally, acetic acid content of the first harvest samples was higher than the second harvest samples like citric acid. The highest malic acid was determined in the non-grafted Crimson Tide cultivar followed by the grafted on Argentario, Maximus and RS 841 rootstocks. Crimson Tide had higher malic acid content than Crisby like other organic acids. Malic acid amount increased from the first harvest to the second harvest time. Another organic acid, determined in all samples, was tartaric acid and its amount was affected from rootstock, cultivar and harvesting time. The highest tartaric acid was found in Maximus and RS 841 rootstocks. Crimson Tide had higher tartaric acid than Crisby cultivar. There was slight decrease in tartaric acid from the first harvest to the second harvest time. The lowest organic acid was oxalic acid for all samples. The highest amount of oxalic acid was determined in the non-grafted Crimson Tide like malic acid. Differences in oxalic acid contents were not significant for cultivars. Oxalic acid amount increased from the first harvest to the second harvest time. Çandır et al. [7] analysed organic acid composition of Crimson Tide cultivar grown by grafting on different local and commercial rootstocks. They found malic (0.327-0.457%) and citric (0.042-0.127%) acids in those samples and their quantities were affected by rootstock. Liu et al. [19] determined malic (11.68-24.44 mg/g), citric (23.92-64.87 mg/g), tartaric (0.43-1.31 mg/g) and quinic (0.56-3.73 mg/g) acids in watermelon flesh for 12 cultivars (on dry matter base).

Table 2. Changes in organic acid contents (mg/kg) of watermelons (mean±SE)[*].

Factors	Sample	Citric	Acetic	Malic	Tartaric	Oxalic
Rootstock	Agentario	195.13^{a}±12.01	114.00^{a}±17.82	143.88^{b}±3.65	61.13^{c}±3.25	23.63^{b}±2.40
	Maximus	166.63^{c}±13.18	120.88^{a}±12.15	113.75^{c}±14.46	85.38^{a}±3.62	26.00^{b}±3.77
	RS 841	190.75^{b}±12.34	124.13^{a}±6.23	102.13^{d}±6.58	84.25^{a}±3.13	27.13^{b}±3.53
	C (NG)[a]	135.25^{e}±11.98	101.00^{b}±9.33	67.50^{e}±2.72	61.00^{c}±3.24	22.75^{b}±1.11
	CrT (NG)[b]	143.75^{d}±3.61	97.00^{b}±19.93	151.50^{a}±20.28	80.50^{b}±1.19	36.25^{a}±9.70
Cultivar	Crisby	169.21^{b}±8.92	108.88^{b}±7.09	95.94^{b}±6.60	68.63^{b}±3.09	26.19^{a}±2.04
	Cr. Tide	176.69^{a}±10.12	120.13^{a}±9.99	138.69^{a}±8.42	82.13^{a}±3.06	26.94^{a}±3.20
Harvest Time	1	183.88^{a}±7.34	123.69^{a}±9.86	107.69^{b}±6.28	76.75^{a}±3.95	23.00^{b}±2.12
	2	162.13^{b}±10.68	105.31^{b}±6.78	126.94^{a}±11.12	74.00^{b}±3.02	30.13^{a}±2.88

[a]C (NG): Crisby (Non-grafted), [b]CrT (NG): Crimson Tide (Non-grafted), [*]: Values in a column followed by different superscript letters are significantly (*P <0.05*) different (Duncan' s multiple range test)

Volatile flavor components affect watermelon aroma [13]. Volatile flavor compounds in fresh vegetables can be formed during maturation and postharvest processing [22, 23]. Volatile flavor compounds of watermelon fruit varied for Crisby (Table 3) and Crimson Tide combinations (Table 4). Rootstock, harvest time, and cultivar affected volatile flavour compounds.

Table 3. Volatile flavour compounds of Crisby fruits with respect to rootstock and harvest time (%).

Compound	Argentario		Maximus		RS841		Crisby (NG[a])	
	1st HT[b]	2nd HT	1st HT	2nd HT	1st HT	2nd HT	1st HT	2nd HT
6-methyl-5-hepten-2-one	16.01	1.89	8.01	7.24	11.86	8.01	7.43	4.51
Nonanal	-	9.91	9.75	10.75	8.93	8.52	7.85	9.31
6-methyl-5-hepten-2-ol	6.19	-	-	4.17	-	-	8.92	7.40
trans-2-nonenal	38.08	23.80	34.63	38.02	45.50	37.53	53.20	38.44
trans,trans-2,6-Nonadienal	12.97	7.95	-	-	-	-	-	-
trans,trans-3,6-Nonadienal	-	-	1.55	1.74	1.58	1.44	2.02	1.20
cis-6-nonen-1-ol	13.18	49.51	37.04	29.23	25.11	38.08	10.25	33.44
Ethylbenzaldehyde	-	-	1.30	-	1.92	1.22	2.26	1.18
trans,trans-2,4-nonadienal	3.51	1.28	2.65	3.72	3.19	3.03	4.03	2.35
Geranyl acetone	2.50	0.45	3.54	1.63	1.92	2.17	2.29	0.99

[a]NG: Non-grafted, [b]HT: Harvest time

Table 4. Volatile flavour compounds content of Crimson Tide fruits with respect to rootstock and harvest time (%).

Compounds	Argentario		Maximus		RS841		Cr. Tide (NG[a])	
	1st HT[b]	2nd HT	1st HT	2nd HT	1st HT	2nd HT	1st HT	2nd HT
6-methyl-5-hepten-2-one	6.24	2.58	3.99	3.54	9.95	9.03	8.18	6.06
Nonanal	4.97	8.45	-	7.32	-	-	5.70	12.65
6-methyl-5-hepten-2-ol	-	-	4.42	7.20	4.47	4.32	-	7.66
trans-2-nonenal	46.74	38.05	41.31	31.89	35.09	46.16	38.92	25.84
trans,cis-2,6-Nonadienal	18.87	16.16	29.62	23.05	10.16	10.89	25.13	9.40
trans,trans-3,6-Nonadienal	1.29	1.17	1.47	1.31	1.68	2.12	1.49	1.08
cis-6-nonen-1-ol	11.70	26.83	6.03	16.96	30.59	14.52	12.06	28.14
Ethylbenzaldehyde	1.26	1.23	1.78	1.10	-	-	1.28	-
trans,trans-2,4-nonadienal	3.22	2.46	2.92	2.06	2.64	3.91	2.57	2.06
Geranyl acetone	1.31	1.15	1.04	1.32	1.61	3.39	0.76	2.18

[a]NG: Non-grafted, [b]HT: Harvest time

The main volatile flavour components were *trans*-2-nonenal, *cis*-6-nonen-1-ol, 6-methyl-5-hepten-2-one, nonanal, *trans,trans*-2,4-nonadienal and geranyl acetone for Crisby combinations. The highest *trans*-2-nonenal was determined in the first harvest of non-grafted Crisby; the lowest value was in the Argentario × Crisby combination at the second harvest. This compound is an aliphatic aldehyde classified as fatty, and it is a characteristic compound for watermelon [13]. Its content was decreased from the first to the second harvest for Crisby combinations except for the Maximus × Crisby combination. The second highest volatile flavour compound was *cis*-6-nonen-1-ol in 'Crisby'. This compound has fruity, sweet and with a fresh aroma [12]. Contrary to *trans*-2-nonenal, *cis*-6-nonen-1-ol increased from the first to the second harvest for all samples except the Maximus × Crisby combination. Differences in volatile flavour compounds between harvests were different for each rootstock/scion combination. One other predominant compound was 6-methyl-5-hepten-2-one (oily green, fruity, herbaceous odor). This compound is a ketone type component. Lowest and highest values of this compound were determined in Crisby × Argentario combinations. The variation in this compound for other Crisby samples was lower than in the Argentario × Crisby combination between harvests. *Trans,trans*-2,4-nonadienal and geranyl acetone were also determined in all samples at lower concentrations. The amount of *trans*-2-nonenal and *trans,trans*-2,4-nonedienal content decreased from the first to the second harvest for Crisby combinations except for Maximus × Crisby combination. Rootstock and harvest affected Geranyl acetone content. Other flavour compounds such as nonanal, 6-methyl-5-hepten-2-ol, *trans,cis*-2,6-nonadienal, *trans,trans*-3,6-nonadienal and ethylbenzaldehyde were identified and quantified in some samples. Rootstock/scion combination and harvest affected the amount of these compounds.

The same volatile flavour compounds were present in Crimson Tide. The predominant component was *trans*-2-nonenal in Crimson Tide combinations and Crisby. The concentration of this decreased from the first to the second harvest except for the Argentario × Crimson Tide combination. Other dominant flavour components were *cis*-6-nonen-1-ol, *trans,cis*-2,6-nonadienal and 6-methyl-5-hepten-2-one for Crimson Tide combinations. Yajima et al. [8] identified 52 flavour compounds in watermelon juice distillate. Additionally, *cis*-3-nonen-1-ol and *cis,cis*-3,6-nonadien-1-ol was the predominant component in their samples. Pino et al. [9] determined volatile components of watermelon by simultaneous steam distillation/solvent extraction (diethyl ether) method. They found ethyl acetate (23.0%), acetaldehyde (18.0%), methyl acetate (11.4%) and ethanol (8.2%) as major volatile components in watermelon, and detected 2-nonenal and 2,6-nonedienal. Beaulieu and Lea [10] determined some aldehydes, ketones, alcohols and furans in their samples. The compounds of 3-nonen-1-ol/*trans,cis*-2,6-nonadienal (16.5-28.2%), *trans*-2-nonenal (10.6-22.5%), *cis*-6-nonenal (2.0-11.3%), hexanal (37.7%) and 6-methyl-5-hepten-2-one (2.7-7.7%) were dominant in RML8129, RML8154, Palomar, Tri-X-313, Pure Heart cultivars. Saftner et al. [11] investigated flavour of fresh-cut watermelon treated with 1-MCP and ethylene. Sixteen volatiles were listed in their study but those were not quantified. Most were aldehydes and ketones. Genthner [12] used static the headspace method and found that *cis*-3-hexenal, *cis,cis*-3,6-nonadienal, *cis*-3-nonenal, *cis*-6-nonenal, *trans*-2-nonenal, *cis*-2-nonenal, *trans,cis*-2,6-nonadienal, *trans, trans*-2,4-nonadienal and *trans,trans,cis*-2,4,6-nonatrienal were important key odorants for fresh-cut watermelon. There were 68 volatile flavour compounds in mini-watermelon [13] with *cis*-3-nonen-1-ol, *trans*-2-nonenal and *cis,cis*-3,6-nonadien-1-ol having the highest values. Some volatile flavour compounds in the previous studies were also determined in the present study, but some differences were observed in their quantities probably due to differences in cultivar, cultural practices, climate and method of analysis.

CONCLUSIONS

Although environment and cultural practices influence fruit quality attributes, rootstock selection for any cultivar, and ripening stage appear to be important to achieve desired physical, chemical and flavour characteristics. Volatile flavour compounds differed for cultivars. Watermelon quality parameters were affected by rootstock/scion combination and harvest time. Appropriate rootstock selection for each cultivar and harvest time for each hybrid combination needs to be determined to meet quality parameters in watermelon.

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