

Storage stability of polyphenols and organic acids in cooked and raw grape juice

Pişmiş ve çiğ üzüm suyunda polifenollerin ve organik asitlerin depolama stabilitesi

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ABSTRACT

This study investigated the effects of boiling and short-term refrigerated storage on the physicochemical properties, phenolic compounds, flavonoids, organic acids, and color characteristics of red and white grape juices. Grape juices were prepared from fresh grapes and subjected to boiling treatment, followed by storage at 4 ± 1 °C for 20 days. Samples were analyzed at different storage intervals to determine changes in key quality parameters. Physicochemical analyses, spectrophotometric assays for total phenolics and flavonoids, and HPLC analysis for organic acids were performed. The results showed significant variation in bioactive compounds across grape varieties, thermal processing, and storage duration ($p < 0.001$). Boiling significantly enhanced catechin content, increasing concentrations from 6.81 mg L^{-1} (unboiled) to 10.63 mg L^{-1} (boiled) ($p < 0.001$). Red juices exhibited greater catechin stability compared to white juices throughout the storage period. Quercetin levels increased from 0.83 mg L^{-1} to 1.47 mg L^{-1} following boiling, though a gradual decline was noted over time. Hydroxybenzoic acids and hydroxycinnamic acids were also enhanced under boiling conditions, with notable increases in caffeic acid concentrations ($p < 0.001$). Organic acid contents, including tartaric and citric acids, were significantly impacted by boiling, with tartaric acid rising from 40.17 g L^{-1} to 44.53 g L^{-1} ($p < 0.05$). Storage duration affected total soluble solids (TSS) and lightness, with visual clarity decreasing over time ($p < 0.001$). Fumaric acid remained stable across treatments, while ascorbic acid levels were more variable, demonstrating a rebound at 20 days post-storage. Overall, this study highlights the importance of optimizing thermal processing and storage conditions to preserve the stability of bioactive compounds in grape juice, thereby enhancing its nutritional value, potential health benefits, and sensory quality.

Key Words: Hydroxybenzoic acids; Hydroxycinnamic acids; Juice color stability; Rutin, Quercetin

ÖZ

Bu çalışma, kaynatma ve kısa süreli soğuk depolamanın kırmızı ve beyaz üzüm sularının fizikokimyasal özellikleri, fenolik bileşikler, flavonoidler, organik asitleri ve renk özellikleri üzerindeki etkilerini araştırmıştır. Üzüm suları taze üzümlerden hazırlanmış ve kaynatma işlemine tabi tutulduktan sonra 20 gün boyunca 4 ± 1 °C'de depolanmıştır. Örnekler, temel kalite parametrelerindeki değişiklikleri belirlemek için farklı depolama aralıklarında analiz edilmiştir. Fizikokimyasal analizler, toplam fenolik ve flavonoidler için spektrofotometrik analizler ve organik asitler için HPLC analizi yapılmıştır. Sonuçlar, üzüm çeşitleri, ısı işlem ve depolama süresi boyunca biyoaktif bileşiklerde önemli farklılıklar olduğunu göstermiştir ($p < 0.001$). Kaynatma, kateşin içeriğini önemli ölçüde artırarak konsantrasyonları 6.81 mg L^{-1} 'den (kaynatılmamış) 10.63 mg L^{-1} 'ye (kaynatılmış) yükseltmiştir ($p < 0.001$). Kırmızı üzüm suları, depolama süresi boyunca beyaz üzüm sularına kıyasla daha yüksek kateşin stabilitesi göstermiştir. Kaynatma işleminden sonra kuerşetin seviyeleri 0.83 mg L^{-1} 'den 1.47 mg L^{-1} 'ye

yükseldi, ancak zamanla kademeli bir düşüş gözlemlendi. Hidroksibenzoik asitler ve hidroksisinnamik asitler de kaynatma koşulları altında arttı ve kafeik asit konsantrasyonlarında belirgin artışlar görüldü ($p < 0.001$). Tartarik ve sitrik asitler de dahil olmak üzere organik asit içerikleri kaynatma işleminden önemli ölçüde etkilendi; tartarik asit 40.17 g L^{-1} 'den 44.53 g L^{-1} 'ye yükseldi ($p < 0.05$). Depolama süresi toplam çözünür katı madde (TSS) ve açıklığı etkiledi ($p < 0.001$) ve görsel berraklık zamanla azaldı. Fumaric asit tüm işlemlerde sabit kalırken, askorbik asit seviyeleri daha değişkenlik gösterdi ve depolamadan 20 gün sonra bir toparlanma sergiledi. Genel olarak, bu çalışma, üzüm suyundaki biyoaktif bileşiklerin stabilitesini korumak, böylece besin değerini, potansiyel sağlık yararlarını ve duyu kalitesini artırmak için ısıl işlem ve depolama koşullarının optimize edilmesinin önemini vurgulamaktadır.

Anahtar Kelimeler: Hidroksibenzoik asit; Hidroksisinnamik asit, Meyve suyu renk stabilitesi; Rutin, Kersetin

Introduction

Grape juice, particularly from the red and purple varieties, is well-recognized for its numerous health benefits attributed primarily to its rich content of phenolic compounds (Yadav et al., 2009). These compounds, including flavonoids such as quercetin and catechin, exhibit robust antioxidant properties that contribute significantly to reducing oxidative stress in the body (Dani et al., 2009; Martinez et al., 2021). Grape juice consumption has been associated with improved cardiovascular health, particularly in hypertensive individuals (Karaman et al., 2020; Roura et al., 2016). A controlled trial demonstrated that Concord grape juice supplementation significantly reduced blood pressure in hypertensive men, suggesting its role in cardiovascular health management (Park et al., 2004). Additionally, grape juice appears to positively influence lipid profiles by lowering serum lipid peroxidation markers, which are crucial for cardiovascular risk assessment (Toaldo et al., 2015).

Moreover, grape juice is believed to positively affect gut microbiota due to its polyphenol content, which can enhance the composition of beneficial gut bacteria, thereby promoting digestive health and immune responses (Martinez et al., 2021; dos Santos et al., 2014). Additionally, its protective effects against oxidative stress extend to cognitive health, with research indicating that grape juice can mitigate oxidative damage in brain tissues, offering potential neuroprotective benefits, especially in the context of age-related cognitive decline (Rodrigues et al., 2012; Gabardo et al., 2018). Despite extensive research on the health benefits of grape juice

polyphenols, limited information is available regarding how common household processing methods, such as boiling, combined with refrigerated storage, influence the stability of phenolic compounds and organic acids. In addition to polyphenol content, changes in organic acids, physicochemical properties, and color parameters are also important indicators of grape juice quality during processing and storage. The processing of grape juice significantly affects its health benefits by modulating bioactive compounds and antioxidant activity (Cosme et al., 2018). Techniques such as heating and maceration can increase the concentration of beneficial phenolic compounds, enhancing antioxidant properties. Previous studies have reported inconsistent effects of thermal processing on phenolic stability; while some studies observed degradation of phenolic compounds, others reported increased concentrations due to improved extraction from plant tissues (Silva et al., 2018). While heat may lead to some nutrient degradation, it also facilitates the release of flavonoids and flavonols vital for health benefits (Iguar et al., 2011). Additionally, (Ghafoor et al., 2012) noted that vacuum heating deactivates polyphenoloxidase, preserving nutritional value.

Different grape varieties display distinct phenolic profiles, largely determined by their genomic traits. Studies have shown that the concentration of flavanol compounds, such as catechins, varies significantly among varieties like Zweigelt and Rondo (Stój et al., 2020). The biosynthesis of these phenolic compounds is influenced not only by the grape cultivar but also by climatic conditions during cultivation, which affect the phenolic acid concentration and ultimately the health benefits of the resulting

grape juice (Yilmaz et al., 2024; Aydın et al., 2018). The choice of grape variety and processing methods also influences the juice's phenolic profile, with specific hybrids showing variations in antioxidant capacity (dos Santos et al., 2014; Güler et al., 2023). This variability underscores the importance of understanding the interplay between grape cultivar traits and processing conditions, which can guide the development of juice products with enhanced health benefits (Zubaidi et al., 2023). Red and white grape varieties differ substantially in their phenolic composition due to differences in skin pigmentation and phenolic accumulation, which may lead to different responses to thermal processing and storage conditions (Yang and Xiao, 2013; Derradji-Benmeziane et al., 2014; Krasteva et al., 2023).

Non-thermal extraction methods have become increasingly important in the food industry for enhancing the yield of bioactive compounds while preserving the nutritional and sensory qualities of products like grape juice. Ultrasound-assisted extraction (UAE) utilizes sound waves to create cavitation bubbles that disrupt cellular structures, significantly increasing the extraction of flavonoids and polyphenols while reducing extraction time and preserving quality (Chemat et al., 2017). Pulsed electric field (PEF) extraction applies short electric pulses to induce electroporation, facilitating the release of intracellular components, thereby improving efficiency and selectivity while maintaining the product's integrity (Demir et al., 2023). Similarly, microwave-assisted extraction (MAE) employs microwave radiation to rapidly heat the solvent and food matrix, resulting in higher yields and minimized degradation of heat-sensitive compounds (Nonglait and Jyoti., 2024). Enzymatic extraction leverages specific enzymes to modify cell wall structures, offering targeted extraction of desired bioactive compounds under mild conditions, which is both efficient and environmentally sustainable (Marathe et al., 2019). The supercritical fluid extraction (SFE) employs supercritical fluids, such as carbon

dioxide, to extract a wide range of compounds efficiently and selectively while eliminating the need for organic solvents (Uwineza and Waśkiewicz., 2020). Although these relatively new methods are useful at the industrial level, thermal processing remains the most common method for ensuring microbial stability in fruit juices and is easy to employ at home (Cao et al., 2021).

The stability of polyphenols and organic acids during processing and storage is critical because these compounds largely determine the antioxidant capacity, nutritional value, and sensory characteristics of grape juice (Cosme et al., 2018). Refrigeration significantly affects the biochemical composition of grape juice, particularly its polyphenolic compounds, antioxidant capacity, and overall quality during storage (Guiné and Barroca., 2014). Beyond phenolic compounds, other quality parameters like color, flavor, and consumer acceptability can vary due to oxidation and non-oxidative reactions (Alaniz et al., 2018). Refrigerated grape juice generally maintains higher antioxidant activity compared to juices stored at ambient temperatures, critical for the health benefits of grape juice consumption, with research showing that refrigerated juices exhibit less degradation over time. Although several studies have examined the effects of storage on fruit juices, information regarding the stability of phenolic compounds and organic acids in grape juice during refrigerated storage remains limited (Abountiolas and do Nunes., 2017). However, while microbial stability is preserved under refrigeration, enzymatic spoilage due to native enzymes remains a concern (Basak et al., 2024). Although previous studies have explored the antioxidant properties and phenolic composition of grape juice, relatively few studies have evaluated how simple thermal processing methods combined with refrigerated storage influence the stability of these compounds in both red and white grape juices. However, the combined effects of thermal processing and refrigerated storage on the stability of polyphenols and organic acids in grape juice

remain unclear. Understanding these changes is important for maintaining the nutritional and functional quality of grape juice during processing and storage.

Considering the importance of processing and storage conditions in determining grape juice quality, this study aimed to evaluate the changes in the quality and biochemical components, particularly polyphenols and organic acids, in white and red grape juices subjected to boiling and different storage durations, in order to better understand their stability and contribute to the optimization of grape juice processing methods for improving nutritional quality and shelf-life in both household and industrial applications.

Materials and Methods

Plant materials

For the red grape juice, the local variety known as Seben Black from Bolu, Türkiye, was used. For the white grape juice, the Etili Beyaz local variety served as the plant material. A total of 15 kg grapes were harvested in late August when they were fully ripe. After harvesting, the grapes were transported directly to the laboratory at Bolu Abant İzzet Baysal University, Horticulture Department, where they were washed with tap water.

Preparation of grape juices

The grapes from both local varieties were separated from their bunches, and their juices were extracted using a juicer (Sinbo SJ-3148, İstanbul, Türkiye). The extracted juice was then filtered through cheesecloth and used as raw grape juice for the study. Additionally, another extraction method involved boiling the grapes. For this process, the grapes were placed in a 10-liter steel pot, crushed, and left to boil. After boiling for 30 minutes, the mixture was filtered with cheesecloth to obtain grape juice. Both juice samples were prepared triplicate, each prepared using 5 kg of grape berries.

Storage conditions

The grape juices collected were placed in 200 ml glass bottles and sealed with cork stoppers. After the initial measurements were recorded, the boiled and unboiled grape juices were stored in a refrigerator at a temperature of 4 ± 1 °C for 20 days. Samples were taken every ten days for analysis.

Physicochemical analyses

Total soluble solids (TSS), pH, and Titratable acidity (TA)

The TSS was measured using a digital refractometer (ATC, 0–32, İstanbul, Türkiye). The pH of the filtrate was determined using a pH meter (Orion Star A211, Thermo Scientific, Waltham, MA, USA). To assess TA, a suitable volume of the filtrate was titrated with 0.1 N NaOH (Ünal et al., 2023).

Color Properties

Numerical color values in the CIE color space, specifically L* (lightness), a* (green-red axis), and b* (blue-yellow axis), along with Chroma and Hue angle values, were obtained using a handheld colorimeter (PCE CSM-4, Southampton, UK). The CIE color space is a standardized color system that quantifies color perception based on three dimensions: The PCE CSM-4 features an LED light source, employs a diffuse/8° measurement geometry, and utilizes the CIE 1964 10° observer type for accurate and consistent color measurement across various applications (Schanda., 2007; Gülüm et al., 2019).

Biochemical Assays

Analysis of polyphenols by HPLC

Phenolic compounds were analyzed using the modified method described in (Kucuker et al., 2025). In summary, a 5 ml grape juice was diluted with an equal volume of distilled water and centrifuged at 15,000 rpm for 15 minutes. The resulting supernatant was filtered through a 0.45 µm Millipore filter before being injected into the HPLC system. Chromatographic separation was conducted on an Agilent 1100 HPLC system

equipped with a diode-array detector (DAD) and a 250 mm × 4.6 mm, 4 µm ODS column. The mobile phase consisted of Solvent A (methanol–acetic acid–water, 10:2:88) and Solvent B (methanol–acetic acid–water, 90:2:8). Separation occurred at wavelengths of 254 and 280 nm, using a flow rate of 1 mL min⁻¹ and an injection volume of 20 µL, and a column temperature of 40 °C. Standard curves were generated using serially diluted HPLC-grade phenolic standards, beginning at 40 ppm (ten times diluted). All chemicals and standards were HPLC-grade and were purchased from Sigma Aldrich (Burlington, Massachusetts, USA).

Analysis of organic acids

The extraction and analysis of organic acids were performed using a modified version of the method reported in (Güler et al., 2025; Bevilacqua and Califano, 1989). Ten milliliters of grape juice were placed in centrifuge tubes and homogenized with 4 mL of 0.009 N H₂SO₄. Following this, the samples were shaken for 1 hour and then centrifuged at 15,000 rpm for 15 minutes. The supernatant was first filtered through coarse filter paper, followed by a double filtration through a 0.45 µm membrane filter and finally through a SEP-PAK C18 cartridge. Organic acids were analyzed using an Agilent HPLC 1100 series G 1322 A equipped with an Aminex HPX-87 H column (300 mm × 7.8 mm). The detection was performed using a DAD set to 214 and 280 nm wavelengths at 25 °C, with the system operated via the Agilent software. HPLC standards were purchased from Sigma Aldrich (Burlington, Massachusetts, USA) in HPLC grade purity and prepared by the serial dilution (ten times diluted) of 100 ppm each.

Analysis of ascorbic acid (Vitamin C)

The content of ascorbic acid was quantified using a modified isocratic HPLC method as outlined in (Cemeroğlu., 2007). Five milliliters of the sample were transferred to a 50 mL volumetric flask and mixed with 10 mL of 6% (w/v) metaphosphoric acid. The mixture was

homogenized at 24,000 rpm for 15 seconds and then centrifuged at 14,000 rpm for 10 minutes at 1°C. Five milliliters of the supernatant were filtered through 0.45 µm PTFE syringe filters and placed in an amber-colored vial. Ascorbic acid levels were determined using a standard external method with an L-ascorbic acid standard purchased from Sigma Aldrich. An Aminex HPX-87 H column (300 mm x 7.8 mm) was utilized with a flow rate set to 1 mL min⁻¹ and an injection volume of 20 µL at 25 °C. The readings were performed at 254 nm.

Statistical analyses

The study was conducted in factorial design with three factors: color, boiling state, and storage time, with three replications for each condition. The collected data were analyzed using a 3-way analysis of variance (3-way ANOVA). To compare the means, Fisher's Least Significant Difference (LSD) test was applied. Results are presented as means ± standard deviations. The analyses and graphics were generated using JMP 16 Pro (SAS, USA).

Results and Discussion

Physicochemicals

Storage duration, boiling, and juice color significantly affected the TSS in grape juice. Boiled samples recorded a mean TSS of 21.98, while unboiled samples had a slightly lower mean of 17.69. The TSS value at day zero was 18.84 and showed no significant changes throughout the storage period, indicating relative stability during storage (Table 1).

Both juice color and boiling significantly influenced the pH of grape juice. Boiled samples exhibited a higher mean pH of 4.16 compared to unboiled samples, which had a mean pH of 3.92. The pH values remained relatively stable throughout the storage durations, with no significant changes observed. The mean pH values were measured at 3.73 at the beginning, 3.85 after 10 days, and 3.83 after 20 days (Table 1).

Storage duration, boiling, and juice color

significantly impacted the TA of grape juice ($p < 0.001$). Both the two-way and three-way interactions were also significant ($p < 0.001$). The mean TA for boiled samples was 2.71, slightly

higher than the mean of 2.57 for unboiled samples. At day zero, the TA measured 2.59, which was comparable to the mean of 2.70 after 10 days (Table 1).

Table 1. Alterations in physicochemical characteristics of red and white grape juices according to the boiling and storage duration (mean \pm std. dev).

Juice Color	Treatment	Storage Time (Day)	TSS	pH	TA
Red	Boiling	Boiled	15.34 \pm 0.28 a	3.92 \pm 0.01 a	3.24 \pm 0.05 a
		Unboiled	15.62 \pm 0.35 a	3.73 \pm 0.01 b	3.02 \pm 0.04 b
	Storage time	0 days	16.35 \pm 0.35 a	3.73 \pm 0.04 b	3.59 \pm 0.31 a
		10 days	16.48 \pm 0.14 a	3.85 \pm 0.02 a	3.05 \pm 0.03 b
		20 days	14.35 \pm 0.39 b	3.82 \pm 0.02 ab	3.14 \pm 0.03 b
	Boiled	0 Days	15.65 \pm 0.23 ab	3.79 \pm 0.06 b	4.29 \pm 0.06 a
		10 Days	15.96 \pm 0.14 a	3.93 \pm 0.02 a	3.23 \pm 0.03 b
		20 Days	14.68 \pm 0.55 b	3.93 \pm 0.02 a	3.10 \pm 0.04 cd
	Unboiled	0 Days	17.04 \pm 0.25 a	3.67 \pm 0.05 b	2.89 \pm 0.04 de
		10 Days	17.01 \pm 0.17 a	3.76 \pm 0.02 b	2.88 \pm 0.03 e
		20 Days	14.02 \pm 0.54 b	3.71 \pm 0.02 b	3.19 \pm 0.06 bc
	White	Boiling	Boiled	21.98 \pm 0.11 a	4.16 \pm 0.02 a
Unboiled			17.69 \pm 0.10 b	3.92 \pm 0.01 b	2.57 \pm 0.02 b
Storage time		0 days	18.84 \pm 0.73 a	3.89 \pm 0.05 b	2.59 \pm 0.02 ab
		10 days	19.92 \pm 0.32 a	4.04 \pm 0.02 a	2.59 \pm 0.02 b
		20 days	19.89 \pm 0.38 a	4.06 \pm 0.02 a	2.70 \pm 0.02 a
Boiled		0 Days	20.43 \pm 0.30 b	3.97 \pm 0.06 b	2.59 \pm 0.04 bc
		10 Days	21.93 \pm 0.13 a	4.16 \pm 0.02 a	2.68 \pm 0.02 b
		20 Days	22.25 \pm 0.14 a	4.18 \pm 0.02 a	2.76 \pm 0.03 a
Unboiled		0 Days	17.24 \pm 0.25 cd	3.82 \pm 0.06 c	2.59 \pm 0.04 bc
		10 Days	17.91 \pm 0.11 c	3.92 \pm 0.02 bc	2.50 \pm 0.02 c
		20 Days	17.53 \pm 0.16 d	3.94 \pm 0.02 b	2.63 \pm 0.03 b
ANOVA					
	Boiling		33.00***	91.88***	101.03***
	Color		169.00***	102.30***	381.02***
	Storage time		13.52***	12.14***	21.40***
	Boiling \times Storage time		5.34**	1.78ns	33.99***
	Color \times Boiling		60.59***	1.05ns	47.88***
	Color \times Storage time		15.87***	1.73ns	18.49***
	Color \times Boiling \times Storage time		0.72ns	0.27ns	40.87***

Different letters in the same column for each factor indicate significant differences at $p \leq 0.05$ according to Fisher's LSD test. ** and *** denotes significance at $p \leq 0.01$ and $p \leq 0.001$, respectively. ns: not significant.

Color properties

Juice color, boiling, and storage time all significantly influenced the lightness (L) of grape juice, with highly significant interactions observed between color and boiling, color and storage time, boiling and storage time, and a notable three-way interaction. Boiled samples exhibited a mean lightness (L^*) of 24.09, which was significantly lower than the mean of 42.99 for

unboiled samples. Lightness consistently decreased over time, beginning at 45.71 on day zero, declining to 33.63 at 10 days, and further reducing to 31.71 at 20 days, indicating a progressive loss of brightness during storage (Table 2).

The a^* value was significantly affected by juice color and storage time, with significant interactions between color and boiling, color and storage time, boiling and storage time, and a three-way interaction. However, boiling alone did

not exert a significant effect on the a^* value. Unboiled samples exhibited a higher mean a^* value of 15.52 compared to boiled samples, which had a mean of 7.29. Throughout the storage period, the mean a^* value showed minimal and non-significant changes, starting at 11.55 on day zero, slightly increasing to 11.73 at 10 days, and declining to 11.05 at 20 days (Table 2).

Both juice color and boiling significantly influenced the b^* value, with boiling having a highly significant effect. The interaction between color and boiling was also significant, as was the effect of storage time. However, the interactions between color and storage time, boiling and storage time, and the three-way interaction were not significant. Unboiled samples exhibited a higher mean b^* value of 24.65 compared to boiled samples, which had a mean of 14.98. Over the storage duration, the b^* values showed a significant decline, starting at 27.44 on day zero, decreasing to 20.65 at 10 days, and further dropping to 17.90 at 20 days, indicating a progressive loss of yellow intensity (Table 2).

Chroma was significantly influenced by both juice color and boiling, with a significant

interaction between the two factors. Storage time also had a significant effect on chroma; however, interactions involving storage time (color \times storage, boiling \times storage, and the three-way interaction) were not significant. Unboiled samples demonstrated higher chroma (28.59) compared to boiled samples (16.77). Over the storage period, chroma declined significantly, starting at 29.77 on day zero, dropping to 23.18 at 10 days, and further decreasing to 21.17 at 20 days, demonstrating a progressive loss of color intensity (Table 2).

Hue angle was significantly influenced by juice color, while boiling and storage time showed no significant effects. The interaction between color and storage time was marginally significant, but all other interactions (color \times boiling, boiling \times storage time, and the three-way interaction) were not significant. Hue values remained relatively stable, with unboiled samples averaging 57.01 and boiled samples at 60.93, indicating no significant differences. Storage duration also did not significantly impact hue, reflecting consistent color perception throughout the study (Table 2).

Table 2. Changes in color characteristics of grape juices influenced by color, boiling, and storage duration.

Juice Color	Treatment	Storage Time (Day)	L^*	a^*	b^*	Chroma	Hue°
Red	Boiling	Boiled	24.09 \pm 0.43 b	24.63 \pm 0.74 a	3.17 \pm 0.39 b	24.91 \pm 0.79 b	30.45 \pm 13.18 a
		Unboiled	42.99 \pm 0.94 a	17.03 \pm 0.36 b	25.81 \pm 0.72 a	30.92 \pm 0.79 a	56.42 \pm 0.33 a
	Storage time	0 days	45.71 \pm 6.34 a	24.98 \pm 1.93 a	20.32 \pm 5.92 a	34.64 \pm 2.05 a	35.74 \pm 9.92 a
		10 days	33.63 \pm 1.52 b	21.94 \pm 1.00 a	14.93 \pm 1.79 a	29.03 \pm 0.89 b	32.46 \pm 3.79 a
		20 days	31.71 \pm 1.51 b	19.13 \pm 0.56 b	13.21 \pm 1.87 a	25.83 \pm 0.86 c	55.51 \pm 13.64 a
	Boiled	0 Days	31.57 \pm 0.46 c	29.26 \pm 0.43 a	7.09 \pm 0.10 c	30.11 \pm 0.44 bc	13.56 \pm 0.20 ab
		10 Days	24.51 \pm 0.40 d	26.95 \pm 1.15 a	4.24 \pm 0.61 c	27.34 \pm 1.24 c	8.35 \pm 0.72 b
		20 Days	22.60 \pm 0.41 d	21.66 \pm 0.63 b	1.53 \pm 0.24 d	21.74 \pm 0.64 d	54.96 \pm 27.61 a
	Unboiled	0 Days	59.84 \pm 0.87 a	20.70 \pm 0.30 bc	33.55 \pm 0.49 a	39.17 \pm 0.57 a	57.91 \pm 0.84 ab
		10 Days	42.75 \pm 0.97 b	16.93 \pm 0.49 cd	25.61 \pm 1.14 b	30.72 \pm 1.21 b	56.58 \pm 0.56 a
		20 Days	40.82 \pm 0.96 b	16.61 \pm 0.50 d	24.89 \pm 0.84 b	29.93 \pm 0.96 bc	56.05 \pm 0.40 a
	White	Boiling	Boiled	54.50 \pm 1.09 a	7.29 \pm 0.24 b	14.98 \pm 0.47 b	16.77 \pm 0.50 b
Unboiled			43.24 \pm 1.39 b	15.52 \pm 0.21 a	24.65 \pm 0.69 a	28.59 \pm 0.69 a	57.01 \pm 0.87 a
Storage time		0 days	71.31 \pm 2.34 a	11.55 \pm 0.89 a	27.44 \pm 2.15 a	29.77 \pm 2.33 a	66.94 \pm 0.62 a
		10 days	47.05 \pm 1.35 b	11.73 \pm 0.72 a	20.65 \pm 0.87 b	23.18 \pm 1.06 b	61.15 \pm 0.77 a
		20 days	47.48 \pm 1.21 b	11.05 \pm 0.68 a	17.90 \pm 0.94 c	21.17 \pm 1.10 b	55.65 \pm 2.02 b
Boiled		0 Days	76.33 \pm 1.11 a	9.57 \pm 0.14 c	22.67 \pm 0.33 bc	24.61 \pm 0.36 c	66.91 \pm 0.98 ab
		10 Days	54.58 \pm 0.98 c	7.28 \pm 0.36 d	15.59 \pm 0.61 d	17.38 \pm 0.66 d	64.72 \pm 0.99 a
		20 Days	51.30 \pm 0.86 c	6.97 \pm 0.32 d	13.29 \pm 0.30 e	15.04 \pm 0.37 e	56.28 \pm 3.78 bc
Unboiled		0 Days	66.28 \pm 0.97 b	13.53 \pm 0.20 b	32.20 \pm 0.47 a	34.93 \pm 0.51 a	66.98 \pm 0.98 ab
		10 Days	39.52 \pm 0.91 e	16.18 \pm 0.20 a	25.72 \pm 0.39 b	28.97 \pm 0.89 b	57.59 \pm 0.45 bc
		20 Days	43.66 \pm 1.95 d	15.14 \pm 0.32 b	22.51 \pm 1.18 c	27.29 \pm 1.02 bc	55.01 \pm 1.56 c
ANOVA							
Boiling			27.86***	0.61ns	527.05***	103.70***	2.18ns
Color			327.16***	362.80***	64.70***	32.69***	4.31*
Storage time			90.43***	14.42***	35.70***	26.60***	0.83ns
Boiling \times Storage time			5.44**	5.34**	0.63ns	2.38ns	1.25ns
Color \times Boiling			258.84***	179.89***	94.22***	6.32*	3.11ns
Color \times Storage time			9.27***	7.66***	0.97ns	0.45ns	2.39ns
Color \times Boiling \times Storage time			4.66*	7.25***	1.56ns	1.82ns	1.93ns

Different letters in the same column for each factor indicate significant differences at $p \leq 0.05$ according to Fisher's LSD test. *, **, and *** indicate significance at $p \leq 0.05$, $p \leq 0.01$, and $p \leq 0.001$, respectively. ns: not significant.

Polyphenols

Hydroxycinnamic acids

Juice color significantly influenced caffeic acid levels ($p < 0.001$) and boiling also had a substantial impact ($p < 0.001$). Storage duration did not significantly affect caffeic acid levels ($p > 0.05$). The interaction between juice color and boiling was significant ($p < 0.001$), as was the interaction between juice color and storage duration ($p < 0.001$). Conversely, the interaction between boiling and storage duration did not yield significant results ($p > 0.05$), nor did the three-way interaction of juice color, boiling, and storage duration ($p > 0.05$). Analysis of caffeic acid concentrations revealed that white juice contained 1.03 mg L^{-1} , which was 30.4% higher than the 0.79 mg L^{-1} observed in red juice. In comparison, boiled juice exhibited a concentration of 1.11 mg L^{-1} , representing a 56.3% increase over the 0.71 mg L^{-1} found in unboiled juice. The levels of caffeic acid initially peaked at 0.93 mg L^{-1} after 10 days before slightly declining to 0.91 mg L^{-1} at 20 days. Boiled white juice recorded the highest concentration at 1.58 mg L^{-1} on day zero, while unboiled red juice had the lowest concentration at 0.46 mg L^{-1} . Over the storage period, white juice concentrations decreased from 1.58 mg L^{-1} at day zero to 1.43 mg L^{-1} at day 20, while red juice peaked at 0.92 mg L^{-1} after 10 days (Table 3).

Juice color did not exert a significant effect on chlorogenic acid levels ($p > 0.05$); however, boiling significantly influenced chlorogenic acid concentrations ($p < 0.01$). Storage duration also had a significant impact on chlorogenic acid levels ($p < 0.05$). The interaction between juice color and boiling did not significantly alter chlorogenic acid levels ($p > 0.05$). In contrast, the interaction between boiling and storage duration was highly significant ($p < 0.001$). The interaction between juice color and storage duration was significant ($p < 0.05$), whereas the three-way interaction of juice color, boiling, and storage duration did not show significant effects ($p > 0.05$). The concentrations of chlorogenic acid varied among juice types and processing methods. Red juice exhibited a chlorogenic acid concentration of 2.07

mg L^{-1} , compared to 2.26 mg L^{-1} in white juice. Initially, boiled red juice registered at 2.35 mg L^{-1} but decreased to 1.92 mg L^{-1} after 20 days. In contrast, unboiled red juice began at a higher concentration of 4.13 mg L^{-1} on day zero, decreasing to 1.41 mg L^{-1} at 10 days, and subsequently rising slightly to 1.59 mg L^{-1} at 20 days. Boiled white juice recorded a concentration of 2.26 mg L^{-1} at the start, increased to 3.09 mg L^{-1} at 10 days, and then decreased to 2.44 mg L^{-1} by day 20. Unboiled white juice commenced at 2.79 mg L^{-1} , slightly decreased to 2.59 mg L^{-1} at 10 days and dropped to 2.08 mg L^{-1} at 20 days. The average chlorogenic acid concentration for red juice was found to be 1.98 mg L^{-1} , while the average for white juice was 2.53 mg L^{-1} . Additionally, boiled juices had an average concentration of 2.39 mg L^{-1} , in contrast to an average of 2.11 mg L^{-1} for unboiled juices. The levels recorded at the baseline day were 2.81 mg L^{-1} , which changed to 2.33 mg L^{-1} by day 10 and further reduced to 2.01 mg L^{-1} at day 20 (Table 3). Juice color significantly influenced o-coumaric acid levels ($p < 0.05$) and boiling also had a significant effect ($p < 0.01$). Storage duration did not significantly affect o-coumaric acid levels ($p > 0.05$). The interaction between juice color and boiling did not significantly alter o-coumaric acid levels ($p > 0.05$), nor did the interaction between juice color and storage duration ($p > 0.05$). Additionally, the interactions between boiling and storage duration ($p > 0.05$) and the three-way interaction of juice color, boiling, and storage duration were not significant ($p > 0.05$). The concentrations of o-coumaric acid varied between juice types, with red juice containing 1.95 mg L^{-1} , while white juice had a concentration of 1.68 mg L^{-1} . After 10 days, boiled red juice measured 1.25 mg L^{-1} , subsequently increasing to 1.82 mg L^{-1} by day 20. Unboiled red juice displayed a lower initial concentration of 1.35 mg L^{-1} at day zero, with minor increases to 0.31 mg L^{-1} at 10 days and maintaining 1.35 mg L^{-1} at day 20. Boiled white juice began at 1.68 mg L^{-1} , increased to 0.42 mg L^{-1} at 10 days, and then decreased to 1.19 mg L^{-1} at 20 days. In contrast, unboiled white juice

started at 1.10 mg L⁻¹, slightly decreased to 0.38 mg L⁻¹ at 10 days, and rose to 1.01 mg L⁻¹ at 20 days. The average o-coumaric acid levels for red juice were recorded at 1.24 mg L⁻¹, while white juice averaged 0.84 mg L⁻¹. Boiled juices had an average concentration of 1.28 mg L⁻¹, in comparison to an average of 0.82 mg L⁻¹ for unboiled juices. The overall levels were 1.52 mg L⁻¹ at day zero, which dropped to 0.60 mg L⁻¹ at 10 days and subsequently increased to 1.34 mg L⁻¹ at day 20 (Table 3).

Juice color significantly influenced p-coumaric acid levels ($p < 0.05$) and boiling had a significant effect as well ($p < 0.01$). Storage duration did not significantly affect p-coumaric acid levels ($p > 0.05$). The interaction between juice color and boiling showed significant effects ($p < 0.01$), and the interaction between juice color and storage duration was also significant ($p < 0.05$). However, the interaction between boiling and storage duration ($p > 0.05$) and the three-way interaction

of juice color, boiling, and storage duration ($p > 0.05$) were not significant. Red juice contained 0.66 mg L⁻¹, while white juice had 0.39 mg L⁻¹. Boiled red juice showed 0.71 mg L⁻¹ after 10 days and maintained the same level at 20 days, whereas unboiled red juice had lower levels at 0.30 mg L⁻¹ at 0 days, rising slightly to 0.32 mg L⁻¹ at 10 days and increasing to 0.54 mg L⁻¹ at 20 days. Boiled white juice initially measured 0.39 mg L⁻¹, increasing to 0.42 mg L⁻¹ at 10 days and then slightly decreasing to 0.40 mg L⁻¹ at 20 days. Unboiled white juice began at 0.33 mg L⁻¹, increased to 0.60 mg L⁻¹ at 10 days, and then dropped to 0.37 mg L⁻¹ at 20 days. The average levels for red juice were 0.56 mg L⁻¹, while the average for white juice was 0.44 mg L⁻¹. Boiled juices averaged 0.56 mg L⁻¹, while unboiled juices averaged 0.44 mg L⁻¹. At 0 days, levels were at 0.42 mg L⁻¹, dropped to 0.52 mg L⁻¹ at 10 days, and slightly decreased to 0.51 mg L⁻¹ at 20 days (Table 3).

Table 3. Hydroxycinnamic acids according to juice color, boiling status, and storage periods.

Juice Color	Treatment	Storage Time (Day)	Caffeic acid (mg/L)	Chlorogenic acid (mg/L)	o-Coumaric acid (mg/L)	p-Coumaric acid (mg/L)
Red	Boiled	0	0.26 ± 0.01 a	2.07 ± 0.11 b	1.95 ± 0.10 a	0.66 ± 0.03 ab
		10	0.92 ± 0.14 a	2.35 ± 0.18 b	1.25 ± 0.33 ab	0.71 ± 0.02 a
		20	0.88 ± 0.18 a	1.92 ± 0.33 b	1.82 ± 0.18 a	0.71 ± 0.13 a
	Unboiled	0	0.46 ± 0.02 a	4.13 ± 0.22 a	1.35 ± 0.07 ab	0.30 ± 0.02 ab
		10	0.80 ± 0.07 a	1.41 ± 0.14 b	0.31 ± 0.06 b	0.32 ± 0.04 b
		20	0.81 ± 0.05 a	1.59 ± 0.30 b	1.35 ± 0.26 a	0.54 ± 0.05 ab
White	Boiled	0	1.58 ± 0.08 a	2.26 ± 0.12 a	1.68 ± 0.09 a	0.39 ± 0.02 a
		10	1.37 ± 0.09 a	3.09 ± 0.49 a	0.42 ± 0.10 b	0.42 ± 0.04 a
		20	1.43 ± 0.15 a	2.44 ± 0.31 a	1.19 ± 0.19 a	0.40 ± 0.01 a
	Unboiled	0	0.94 ± 0.05 ab	2.79 ± 0.15 a	1.10 ± 0.06 ab	0.33 ± 0.02 a
		10	0.71 ± 0.07 b	2.59 ± 0.29 a	0.38 ± 0.06 b	0.60 ± 0.04 a
		20	0.53 ± 0.12 b	2.08 ± 0.26 a	1.01 ± 0.08 a	0.37 ± 0.11 a
ANOVA						
Color			21.65***	1.82ns	7.39*	5.60*
Boiling			17.55***	0.12ns	11.65**	7.42**
Storage Duration			0.69ns	4.04*	22.71***	0.89ns
Color * Boiling			17.74***	0.72ns	2.16ns	10.93**
Color * Storage Duration			5.67***	3.32*	0.20ns	3.13*
Boiling * Storage Duration			0.67ns	5.66***	0.34ns	0.35ns
Color * Boiling * Storage Duration			0.47ns	1.35ns	1.08ns	2.73*

Different letters in the same column for each factor indicate significant differences at $p \leq 0.05$ according to Fisher's LSD test. *, **, and *** denotes significance at $p \leq 0.05$, $p \leq 0.01$ and $p \leq 0.001$, respectively. ns: not significant.

Hydroxybenzoic acids

Juice color significantly affected vanillic acid levels ($p < 0.001$) and boiling also had a significant impact ($p < 0.001$). However, storage duration did not significantly affect vanillic acid concentrations

($p > 0.05$). The interaction between juice color and boiling was significant ($p < 0.05$). In contrast, the interaction between juice color and storage duration ($p > 0.05$) and the interaction between boiling and storage duration ($p > 0.05$) showed no significant effects. Similarly, the three-way interaction of juice color, boiling, and storage

duration was not significant ($p > 0.05$). Red juice contained 0.80 mg L^{-1} , which was 158.1% more than white juice at 0.31 mg L^{-1} . Boiled juice measured 0.82 mg L^{-1} , representing a 164.5% increase compared to unboiled juice at 0.31 mg L^{-1} . Vanillic acid levels increased from 0.49 mg L^{-1} at 0 days to 0.63 mg L^{-1} at 20 days. Boiled red juice reached 1.42 mg L^{-1} at 20 days, while unboiled white juice measured only 0.14 mg L^{-1} on day zero. Red juice concentrations increased from 0.98 mg L^{-1} to 1.42 mg L^{-1} , whereas white juice fluctuated from 0.53 mg L^{-1} to 0.40 mg L^{-1} (Table 4).

Juice color significantly affected gallic acid levels ($p < 0.001$), and boiling had an even more substantial impact ($p < 0.001$). Additionally, storage duration significantly influenced gallic acid levels ($p < 0.01$). The interaction between juice color and boiling was significant ($p < 0.01$), indicating that the effect of juice color on gallic acid levels depends on the boiling status. However, the interaction between juice color and storage duration ($p > 0.05$) and the interaction between boiling and storage duration ($p > 0.05$) were not significant. The three-way interaction of juice color, boiling, and storage duration was also not significant ($p > 0.05$). White juice contained 4.55 mg L^{-1} , which was 43.5% more than red juice at 3.17 mg L^{-1} . Boiled juice exhibited 6.84 mg L^{-1} , representing a 612.5% increase compared to unboiled juice at 0.96 mg L^{-1} . Gallic acid levels rose from 3.40 mg L^{-1} at 10 days to 4.40 mg L^{-1} at

20 days, with a slight dip to 3.43 mg L^{-1} at 0 days. Boiled white juice reached 8.92 mg L^{-1} at 20 days, while unboiled red juice measured 0.69 mg L^{-1} at day zero. White juice increased from 0.99 mg L^{-1} at 0 days to 8.92 mg L^{-1} at 20 days, whereas red juice increased from 5.21 mg L^{-1} to 6.19 mg L^{-1} (Table 4).

Juice color had a significant effect on syringic acid levels ($p < 0.001$) and boiling also had a significant impact ($p < 0.001$). Storage duration significantly influenced syringic acid concentrations ($p < 0.001$). The interaction between juice color and boiling was significant ($p < 0.001$), indicating that juice color affects syringic acid levels in relation to boiling. In contrast, the interactions between juice color and storage duration ($p > 0.05$) and between boiling and storage duration ($p > 0.05$) were not significant. The three-way interaction of juice color, boiling, and storage duration was also not significant ($p > 0.05$). Red juice contained 1.13 mg L^{-1} , which was 109.3% more than white juice at 0.54 mg L^{-1} . Boiled juice had a concentration of 1.12 mg L^{-1} , which was 100.0% higher than unboiled juice at 0.56 mg L^{-1} . Syringic acid levels increased from 0.60 mg L^{-1} at day zero to 0.95 mg L^{-1} at 20 days. Boiled red juice reached 1.77 mg L^{-1} at 20 days, while unboiled white juice measured 0.28 mg L^{-1} at day zero. Red juice rose from 1.40 mg L^{-1} to 1.77 mg L^{-1} , whereas white juice fluctuated from 0.42 mg L^{-1} to 0.67 mg L^{-1} (Table 4).

Table 4. Hydroxybenzoic acids according to juice color, boiling status, and storage periods.

Juice Color	Treatment	Storage Time (Day)	Gallic acid (mg/L)	Syringic acid (mg/L)	Vanillic acid (mg/L)
Red	Boiled	0	5.21 ± 0.27 a	1.40 ± 0.07 ab	0.98 ± 0.05 ab
		10	5.23 ± 0.32 a	1.63 ± 0.09 a	1.02 ± 0.03 ab
		20	6.19 ± 0.43 a	1.77 ± 0.10 a	1.42 ± 0.38 a
	Unboiled	0	0.69 ± 0.04 b	0.31 ± 0.02 c	0.31 ± 0.02 ab
		10	0.60 ± 0.02 b	0.49 ± 0.06 c	0.43 ± 0.05 b
		20	0.80 ± 0.33 b	0.78 ± 0.16 bc	0.40 ± 0.05 b
White	Boiled	0	6.83 ± 0.36 a	0.42 ± 0.02 a	0.53 ± 0.03 a
		10	7.57 ± 0.41 a	0.46 ± 0.09 a	0.43 ± 0.03 a
		20	8.92 ± 0.87 a	0.67 ± 0.07 a	0.40 ± 0.04 a
	Unboiled	0	0.99 ± 0.05 b	0.28 ± 0.01 a	0.14 ± 0.01 bc
		10	0.80 ± 0.02 b	0.54 ± 0.07 a	0.14 ± 0.03 c
		20	1.70 ± 0.38 b	0.59 ± 0.05 a	0.30 ± 0.05 ab
ANOVA					
Color			17.56***	67.84***	16.91***
Boiling			316.61***	64.99***	22.97***
Storage Duration			5.14**	8.33***	0.98ns
Color * Boiling			7.49**	54.79***	5.58*
Color * Storage Duration			0.68ns	0.39ns	0.45ns
Boiling * Storage Duration			1.06ns	0.12ns	0.18ns
Color * Boiling * Storage Duration			0.11ns	0.84ns	1.44ns

Different letters in the same column for each factor indicate significant differences at $p \leq 0.05$ according to Fisher's LSD test. *, **, and *** denotes significance at $p \leq 0.05$, $p \leq 0.01$ and $p \leq 0.001$, respectively. ns: not significant.

Flavonoids

Juice color significantly affected catechin levels ($p < 0.001$) and boiling also had a substantial impact ($p < 0.001$). Additionally, storage duration significantly influenced catechin levels ($p < 0.05$). The interaction between juice color and boiling was highly significant ($p < 0.001$), demonstrating that boiling alters the effect of juice color on catechin levels. The interaction between juice color and storage duration was found to be significant ($p < 0.05$), whereas the interaction between boiling and storage duration showed no significant effects ($p > 0.05$). Notably, the three-way interaction of juice color, boiling, and storage duration was significant ($p < 0.05$). Red juice contained 10.07 mg L^{-1} , representing a 38.5% increase compared to white juice at 7.27 mg L^{-1} . Boiled juice retained a concentration of 10.63 mg L^{-1} , which was 56.1% higher than unboiled juice at 6.81 mg L^{-1} . Over the storage period, catechin levels decreased from 9.97 mg L^{-1} at day zero to 7.96 mg L^{-1} at 10 days, followed by a rebound to 9.03 mg L^{-1} at 20 days. Boiled red juice measured 16.31 mg L^{-1} at day zero, while unboiled white juice reached 5.59 mg L^{-1} at 10 days. Red juice exhibited a trend decreasing from 16.31 mg L^{-1} to 14.02 mg L^{-1} , while white juice fluctuated between 6.56 mg L^{-1} and 7.10 mg L^{-1} (Table 5).

Juice color did not significantly affect quercetin levels ($p > 0.05$), while boiling had a significant impact ($p < 0.001$). Additionally, storage duration did not significantly influence quercetin concentrations ($p > 0.05$). The interaction between juice color and boiling was not significant ($p > 0.05$), nor did the interaction between juice color and storage duration show significant effects ($p > 0.05$). However, the interaction between boiling and storage duration was significant ($p < 0.01$), indicating that the effect of boiling on quercetin levels is influenced by storage duration. The three-way interaction of juice color, boiling, and storage duration was also not significant ($p > 0.05$). In terms of concentration, red juice contained 1.59 mg L^{-1} , whereas white juice had a slightly higher level of 1.81 mg L^{-1} . Boiled red juice measured 1.24 mg L^{-1} at 10 days before dropping to 1.30 mg L^{-1} at 20 days. In contrast, unboiled red juice started at a lower level of 0.21 mg L^{-1} at day zero, increasing to 0.72 mg L^{-1} at 10 days and reaching 1.02 mg L^{-1} at 20 days. Boiled white juice began at 1.81 mg L^{-1} , decreased to 1.59 mg L^{-1} at 10 days, and then rose to 1.66 mg L^{-1} at 20 days. Unboiled white juice measured 0.18 mg L^{-1} at day zero and slightly increased to 0.92 mg L^{-1} at 10 days before reaching 1.01 mg L^{-1} at 20 days. The average quercetin level for red juice was 1.05 mg L^{-1} , compared to 1.25 mg L^{-1} for white juice. Boiled

juices averaged 1.47 mg L⁻¹, while unboiled juices had a lower average of 0.83 mg L⁻¹. Overall, levels were recorded at 0.95 mg L⁻¹ at day zero, increased to 1.10 mg L⁻¹ at 10 days, and remained stable at 1.25 mg L⁻¹ at 20 days (Table 5).

Juice color did not significantly affect rutin levels ($p > 0.05$), although boiling exhibited a significant impact ($p < 0.01$). Storage duration also did not significantly influence rutin concentrations ($p > 0.05$). The interaction between juice color and boiling was not significant ($p > 0.05$), nor did the interaction between juice color and storage duration show any significant effects ($p > 0.05$). However, the interaction between boiling and storage duration was significant ($p < 0.01$), indicating a notable interaction effect on rutin levels. The three-way interaction of juice color, boiling, and storage duration was not significant ($p > 0.05$). In terms of concentration, red juice showed a level of 1.32

mg L⁻¹ compared to 1.47 mg L⁻¹ in white juice. Boiled red juice increased from 1.40 mg L⁻¹ at 10 days to 2.13 mg L⁻¹ at 20 days. Conversely, unboiled red juice measured 0.95 mg L⁻¹ at day zero but decreased to 0.80 mg L⁻¹ at 10 days and then slightly increased to 0.89 mg L⁻¹ by day 20. Boiled white juice began at 1.47 mg L⁻¹, fell to 1.38 mg L⁻¹ at 10 days, and ultimately rose to 2.01 mg L⁻¹ at 20 days. Moreover, unboiled white juice recorded an initial concentration of 0.79 mg L⁻¹, increased to 1.07 mg L⁻¹ at 10 days, and then dropped to 0.98 mg L⁻¹ at 20 days. Average rutin levels were 1.28 mg L⁻¹ for red juice and 1.33 mg L⁻¹ for white juice. Boiled juices averaged 1.70 mg L⁻¹, while unboiled juices averaged 0.93 mg L⁻¹. Overall, rutin levels were recorded at 1.13 mg L⁻¹ at day zero, increased slightly to 1.16 mg L⁻¹ at 10 days, and reached 1.50 mg L⁻¹ at 20 days (Table 5).

Table 5. Changes in flavonoids according to juice color, boiling status, and storage periods

Juice Color	Treatment	Storage Time (Day)	Catechin (mg/L)	Quercetin (mg/L)	Rutin (mg/L)
Red	Boiled	0	16.31 ± 0.86 a	1.59 ± 0.08 a	1.32 ± 0.07 ab
		10	14.24 ± 0.70 a	1.24 ± 0.08 a	1.40 ± 0.08 ab
		20	14.02 ± 0.77 a	1.30 ± 0.12 a	2.13 ± 0.53 a
	Unboiled	0	4.70 ± 0.25 b	0.21 ± 0.01 b	0.95 ± 0.05 ab
		10	5.70 ± 0.13 b	0.72 ± 0.17 ab	0.80 ± 0.17 b
		20	6.09 ± 1.40 b	1.02 ± 0.21 ab	0.89 ± 0.12 b
White	Boiled	0	6.56 ± 0.35 bc	1.81 ± 0.10 abc	1.47 ± 0.08 a
		10	6.05 ± 0.10 bc	1.59 ± 0.07 ab	1.38 ± 0.04 a
		20	7.10 ± 0.39 bc	1.66 ± 0.13 a	2.01 ± 0.63 a
	Unboiled	0	12.32 ± 0.65 a	0.18 ± 0.01 d	0.79 ± 0.04 a
		10	5.59 ± 0.21 c	0.92 ± 0.18 cd	1.07 ± 0.16 a
		20	8.91 ± 1.29 ab	1.01 ± 0.18 bcd	0.98 ± 0.16 a
ANOVA					
Color			16.23***	2.62ns	0.02ns
Boiling			33.77***	56.41***	8.84**
Storage Duration			4.05*	2.09ns	1.47ns
Color * Boiling			95.25***	1.32ns	0.02ns
Color * Storage Duration			2.72*	0.20ns	0.06ns
Boiling * Storage Duration			1.01ns	5.91**	1.35ns
Color * Boiling * Storage Duration			4.07*	0.14ns	0.11ns

Different letters in the same column for each factor indicate significant differences at $p \leq 0.05$ according to Fisher's LSD test. *, **, and *** denotes significance at $p \leq 0.05$, $p \leq 0.01$ and $p \leq 0.001$, respectively. ns: not significant.

Organic acids

Juice color significantly influenced tartaric acid levels ($p < 0.001$) and boiling also had a significant impact ($p < 0.001$). In contrast, storage duration did not significantly affect tartaric acid concentrations ($p > 0.05$). The interaction between juice color and boiling was significant ($p < 0.001$), indicating that boiling alters the effect of juice color on tartaric acid levels. However, the

interaction between juice color and storage duration ($p > 0.05$) and the interaction between boiling and storage duration ($p > 0.05$) showed no significant effects. The three-way interaction of juice color, boiling, and storage duration was also not significant ($p > 0.05$). Red juice contained 52.72 g L⁻¹, whereas white juice measured 55.92 g L⁻¹. Boiled red juice recorded 45.99 g L⁻¹ at 10 days, increasing to 48.10 g L⁻¹ at 20 days. In comparison, unboiled red juice contained 48.71 g

L⁻¹ at day zero but fell to 37.53 g L⁻¹ at 10 days before rising again to 40.62 g L⁻¹ at 20 days. Boiled white juice started at 55.92 g L⁻¹ but dropped to 41.03 g L⁻¹ at 10 days, further declining to 36.90 g L⁻¹ by day 20. Unboiled white juice initially showed 39.96 g L⁻¹, decreased to 41.07 g L⁻¹ at 10 days and dropped again to 39.06 g L⁻¹ at 20 days. The average levels for red juice were 44.02 g L⁻¹, while white juice averaged 40.55 g L⁻¹. Boiled juices averaged 44.53 g L⁻¹, contrasting with an average of 40.17 g L⁻¹ for unboiled juices. Overall, tartaric acid levels were recorded at 49.33 g L⁻¹ at day zero, slightly reduced to 41.42 g L⁻¹ at 10 days, and measured 41.17 g L⁻¹ at 20 days (Table 6).

Juice color had a significant effect on citric acid levels ($p < 0.001$), and boiling also significantly influenced citric acid concentrations ($p < 0.001$). Conversely, storage duration did not significantly affect citric acid levels ($p > 0.05$). The interaction between juice color and boiling was found to be significant ($p < 0.001$), suggesting that the boiling process alters the effect of juice color on citric acid levels. In contrast, both the interaction between juice color and storage duration ($p > 0.05$) and the interaction between boiling and storage duration ($p > 0.05$) showed no significant effects. Additionally, the three-way interaction of juice color, boiling, and storage duration was not significant ($p > 0.05$). In terms of concentration, red juice contained 35.37 mg L⁻¹, while white juice held a higher concentration of 51.27 mg L⁻¹. Boiled red juice decreased from 32.17 mg L⁻¹ at 10 days to 25.84 mg L⁻¹ at 20 days. Unboiled red juice began at 18.68 mg L⁻¹, increased to 20.35 mg L⁻¹ at 10 days, and then slightly decreased to 19.92 mg L⁻¹ at 20 days. For boiled white juice, the concentration started at 51.27 mg L⁻¹, increased to 64.43 mg L⁻¹ at 10 days, and returned to 51.65 mg L⁻¹ at 20 days. Unboiled white juice measured 20.30 mg L⁻¹ at day zero, declined to 19.29 mg L⁻¹ at 10 days, and fell further to 17.26 mg L⁻¹ at 20 days. The average citric acid level for red juice was 24.88 mg L⁻¹, while white juice averaged 37.00 mg L⁻¹. Boiled juices had an average concentration of 42.82 mg

L⁻¹, in contrast to unboiled juices, which averaged 19.24 mg L⁻¹. Overall, citric acid levels were recorded at 31.40 mg L⁻¹ at day zero, increased to 32.94 mg L⁻¹ at 10 days, and then decreased to 28.66 mg L⁻¹ at 20 days (Table 6).

Juice color significantly influenced malic acid levels ($p < 0.001$) and boiling also had a substantial impact ($p < 0.001$). Additionally, storage duration significantly affected malic acid concentrations ($p < 0.01$). The interaction between juice color and boiling showed significant effects ($p < 0.001$), indicating that boiling modifies the effect of juice color on malic acid levels. The interaction between juice color and storage duration was significant ($p < 0.05$), while the interaction between boiling and storage duration did not exhibit significant effects ($p > 0.05$). Furthermore, the three-way interaction of juice color, boiling, and storage duration was not significant ($p > 0.05$). In terms of concentration, red juice contained 9.18 g L⁻¹, whereas white juice measured 12.09 g L⁻¹. Boiled red juice recorded 7.52 g L⁻¹ at 10 days and increased to 9.67 g L⁻¹ at 20 days. Conversely, unboiled red juice started at 8.38 g L⁻¹ at day zero, decreased to 7.86 g L⁻¹ at 10 days, and then increased to 9.90 g L⁻¹ at 20 days. Boiled white juice was measured at 12.09 g L⁻¹ at day zero, decreased to 12.95 g L⁻¹ at 10 days, and then slightly dipped to 11.97 g L⁻¹ at 20 days. Unboiled white juice began at 8.54 g L⁻¹, dipped to 7.79 g L⁻¹ at 10 days, and rose again to 9.59 g L⁻¹ by day 20. Average malic acid levels were recorded at 8.74 g L⁻¹ for red juice and 10.47 g L⁻¹ for white juice. Boiled juices averaged 10.46 g L⁻¹, while unboiled juices averaged 8.74 g L⁻¹. Overall, levels were 9.55 g L⁻¹ at day zero, decreased to 8.88 g L⁻¹ at 10 days, and increased to 10.28 g L⁻¹ at 20 days (Table 6).

Juice color significantly affected succinic acid levels ($p < 0.001$), and boiling also had a significant impact ($p < 0.001$). In contrast, storage duration did not significantly influence succinic acid concentrations ($p > 0.05$). The interaction between juice color and boiling was significant ($p < 0.001$), indicating that the effect of juice color on succinic acid levels depends on whether the

juice is boiled. Both the interaction between juice color and storage duration ($p > 0.05$) and the interaction between boiling and storage duration ($p > 0.05$) did not show significant effects. The three-way interaction of juice color, boiling, and storage duration was also not significant ($p > 0.05$). In terms of concentration, red juice had 1.36 mg L^{-1} , while white juice recorded a higher level of 3.19 mg L^{-1} . Boiled red juice measured 1.15 mg L^{-1} at 10 days, decreasing to 1.04 mg L^{-1} at 20 days. Conversely, unboiled red juice started at 0.86 mg L^{-1} at day zero, decreased to 0.91 mg L^{-1} at 10 days, and recorded 0.82 mg L^{-1} at 20 days. Boiled white juice began at 3.19 mg L^{-1} , decreased to 3.55 mg L^{-1} at 10 days, and reached 3.51 mg L^{-1} at 20 days. Unboiled white juice measured 1.49 mg L^{-1} at day zero, declined to 1.65 mg L^{-1} at 10 days, and further decreased to 1.11 mg L^{-1} at 20 days. The average levels for red juice were 1.00 mg L^{-1} , while white juice averaged 2.41 mg L^{-1} . Boiled juices had an average concentration of 2.27 mg L^{-1} , whereas unboiled juices averaged 1.13 mg L^{-1} . Overall, the succinic acid levels were recorded at 1.72 mg L^{-1} at day zero, changed to 1.75 mg L^{-1} at 10 days, and registered 1.62 mg L^{-1} at 20 days (Table 6).

Juice color did not significantly affect fumaric acid levels ($p > 0.05$). Similarly, boiling had no significant effect on fumaric acid concentrations ($p > 0.05$). In contrast, storage duration showed a significant impact on fumaric acid levels ($p < 0.05$). The interaction between juice color and boiling was not significant ($p > 0.05$), nor did the interaction between juice color and storage duration show any significant effects ($p > 0.05$). Additionally, both the interaction between boiling and storage duration ($p > 0.05$) and the three-way interaction of juice color, boiling, and storage duration were not significant ($p > 0.05$). In terms of concentration, red juice contained 1.14 mg L^{-1} , while white juice had a higher concentration of 1.73 mg L^{-1} . Boiled red juice registered 1.05 mg L^{-1} at 10 days and increased to 2.78 mg L^{-1} at 20 days. Unboiled red juice exhibited a lower concentration of 0.57 mg L^{-1} at day zero, slightly rose to 0.86 mg L^{-1} at 10 days, and then

decreased to 2.31 mg L^{-1} at 20 days. Boiled white juice measured 1.73 mg L^{-1} at day zero, decreased to 1.91 mg L^{-1} at 10 days, and then rose again to 1.71 mg L^{-1} at 20 days. Unboiled white juice contained 0.57 mg L^{-1} at day zero, decreased to 0.75 mg L^{-1} at 10 days, and increased significantly to 2.46 mg L^{-1} at 20 days. The average fumaric acid level for red juice was 1.64 mg L^{-1} , while white juice averaged 1.63 mg L^{-1} . Boiled juices averaged 1.81 mg L^{-1} , whereas unboiled juices averaged 1.46 mg L^{-1} . Overall, the levels were recorded at 1.00 mg L^{-1} at day zero, increased to 1.11 mg L^{-1} at 10 days, and ultimately registered 2.31 mg L^{-1} at 20 days (Table 6).

Juice color significantly affected ascorbic acid levels ($p < 0.05$), while boiling did not have a significant impact ($p > 0.05$). Storage duration also significantly influenced ascorbic acid levels ($p < 0.05$). The interaction between juice color and boiling was not significant ($p > 0.05$), and the interaction between juice color and storage duration also showed no significant effects ($p > 0.05$). Furthermore, both the interaction between boiling and storage duration ($p > 0.05$) and the three-way interaction of juice color, boiling, and storage duration were not significant ($p > 0.05$). In terms of concentration, red juice contained 31.51 mg L^{-1} , while white juice had a slightly lower concentration of 29.23 mg L^{-1} . Boiled red juice measured 28.53 mg L^{-1} at 10 days, with a slight decline to 28.89 mg L^{-1} at 20 days. Unboiled red juice started at 32.86 mg L^{-1} at day zero and slightly decreased to 31.77 mg L^{-1} at 10 days, before rising to 32.58 mg L^{-1} at 20 days. Boiled white juice recorded 29.23 mg L^{-1} at day zero, decreased to 27.59 mg L^{-1} at 10 days, and then increased to 31.00 mg L^{-1} at 20 days. In contrast, unboiled white juice had 32.63 mg L^{-1} at day zero, decreased to 21.03 mg L^{-1} at 10 days, and ultimately rose to 29.32 mg L^{-1} at 20 days. The average levels for red juice were 32.85 mg L^{-1} , while white juice averaged 27.70 mg L^{-1} . Boiled juices averaged 31.48 mg L^{-1} , whereas unboiled juices averaged 29.18 mg L^{-1} . Overall, the levels were 31.56 mg L^{-1} at day zero, decreased slightly to 27.22 mg L^{-1} at 10 days, and reached 32.95 mg

L⁻¹ at 20 days (Table 6).

Table 6. Fluctuations in organic acids according to color, storage time and boiling treatment

Juice Color	Treatment	Storage Time (Day)	Ascorbic acid (mg/100g)	Citric Acid (mg/L)	Fumaric acid (mg/L)	Malic Acid (g/L)	Succinic Acid (mg/L)	Tartaric Acid (g/L)
Red	Boiled	0	31.51 ± 1.66 a	35.37 ± 1.86 a	1.14 ± 0.06 a	9.18 ± 0.48 ab	1.36 ± 0.07 a	52.72 ± 2.77 a
		10	28.53 ± 2.41 a	32.17 ± 1.36 a	1.05 ± 0.05 a	7.52 ± 0.24 b	1.15 ± 0.06 a	45.99 ± 2.35 ab
		20	38.89 ± 2.99 a	25.84 ± 3.47 a	2.78 ± 1.29 a	9.67 ± 0.22 a	1.04 ± 0.12 a	48.10 ± 2.63 a
	Unboiled	0	32.86 ± 1.73 a	18.68 ± 0.98 a	0.57 ± 0.03 a	8.38 ± 0.44 ab	0.86 ± 0.05 a	48.71 ± 2.56 ab
		10	31.77 ± 1.70 a	20.35 ± 2.46 a	0.86 ± 0.09 a	7.86 ± 0.36 b	0.91 ± 0.11 a	37.53 ± 2.20 b
		20	32.58 ± 4.38 a	19.92 ± 6.80 a	2.31 ± 0.60 a	9.90 ± 0.44 a	0.82 ± 0.10 a	40.62 ± 2.31 ab
White	Boiled	0	29.23 ± 1.54 ab	51.27 ± 2.70 ab	1.73 ± 0.09 ab	12.09 ± 0.64 ab	3.19 ± 0.17 a	55.92 ± 2.94 a
		10	27.59 ± 2.10 ab	64.43 ± 4.63 a	1.91 ± 0.14 ab	12.95 ± 0.87 a	3.55 ± 0.24 a	41.03 ± 2.80 ab
		20	31.00 ± 1.18 a	51.65 ± 5.75 a	1.71 ± 0.12 ab	11.97 ± 0.98 ab	3.51 ± 0.34 a	36.90 ± 3.38 b
	Unboiled	0	32.63 ± 1.72 ab	20.30 ± 1.07 bc	0.57 ± 0.03 ab	8.54 ± 0.45 bc	1.49 ± 0.08 b	39.96 ± 2.10 ab
		10	21.03 ± 2.67 b	19.29 ± 2.50 c	0.75 ± 0.08 b	7.79 ± 0.25 c	1.65 ± 0.07 b	41.07 ± 1.06 ab
		20	29.32 ± 2.42 ab	17.26 ± 3.08 c	2.46 ± 0.69 a	9.59 ± 0.23 bc	1.11 ± 0.14 b	39.06 ± 1.76 b

ANOVA						
Color		4.49*	15.13***	0.03ns	19.81***	122.08***
Boiling		0.30ns	61.53***	1.19ns	23.41***	84.16***
Storage Duration		5.12*	1.85ns	5.40*	6.25**	1.46ns
Color * Boiling		0.07ns	16.95***	0.02ns	21.51***	44.64***
Color * Storage Duration		0.39ns	0.44ns	0.61ns	2.87*	0.63ns
Boiling * Storage Duration		0.74ns	1.10ns	0.69ns	1.86ns	0.56ns
Color * Boiling * Storage Duration		2.08ns	0.65ns	1.03ns	2.28ns	1.25ns

Different letters in the same column for each factor indicate significant differences at $p \leq 0.05$ according to Fisher's LSD test. *, **, and *** denotes significance at $p \leq 0.05$, $p \leq 0.01$ and $p \leq 0.001$, respectively. ns: not significant.

Pca analysis

The first two principal components (PCs) accounted for 49.61% of the total variation, while the first five represented 74.21%. PC1 explained 26.82% of the total variation, and it was significantly influenced by gallic acid and citric acid, with eigenvectors of 0.41 and 0.41, respectively. On the other hand, PC2 accounted for 22.79% of the variation and was significantly influenced by catechin (0.41) and syringic acid (0.46). The PCA results suggested that boiling and juice color had a more noticeable effect on the

fluctuation of the biochemicals. The boiled juice exhibited a wide range of compounds (green shade), whereas the unboiled samples represented a relatively low variation (purple shade). On the other hand, white grape juice was characterized by relatively pronounced citric, malic, succinic, caffeic, and chlorogenic acids, while the red juice was mainly represented by syringic, vanillic, o-coumaric, p-coumaric acids as well as flavonoids and tartaric and ascorbic acids. Storage durations slightly differed, and 20 days yielded relatively higher contents (Fig. 1).

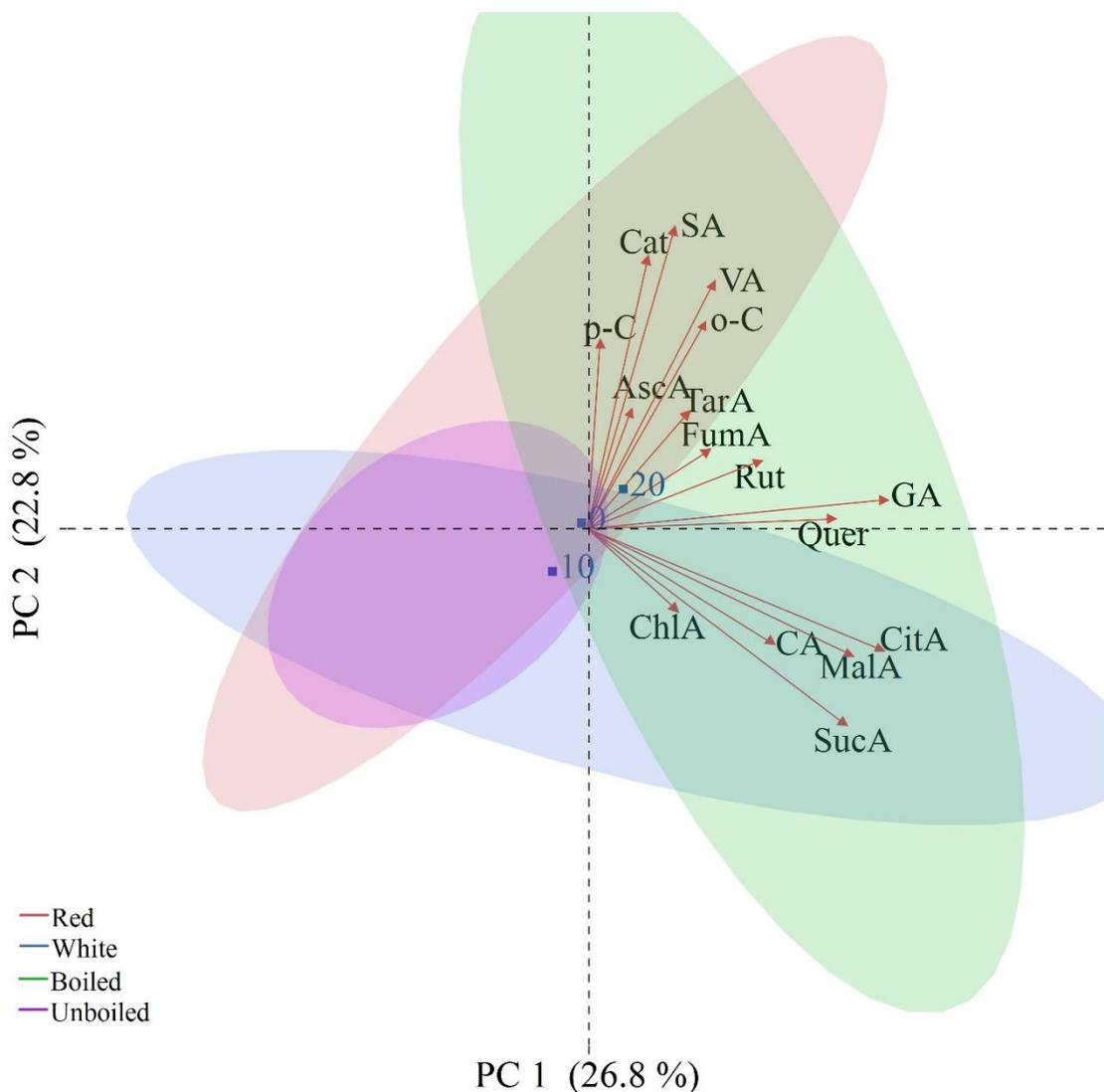


Figure 1. The PCA analysis shows the relationships among factors and biochemical compounds. The density ellipses were created with a 95% confidence level. AscA: Ascorbic acid, CA: Caffeic acid, Cat: Catechin, ChlA: Chlorogenic acid, CitA: Citric acid, GA: Gallic acid, FumA: Fumaric acid, MalA: Malic acid, o-C: o-Coumaric acid, p-C: p-Coumaric acid, Quer: Quercetin, Rut: Rutin, SA: Syringic acid, SucA: Succinic acid, TarA: Tartaric acid, VA: Vanillic acid.

The physicochemical characteristics such as pH, TSS, and turbidity reveal notable variations between processed and nonprocessed juices. Studies have shown that thermally processed juices typically maintain a more favorable TSS to TA ratio, which is critical for juice sensory quality and microbiological stability (Wojdyło et al., 2014; Nguyen et al., 2024). This emphasizes the role of processing in extending shelf life and preserving desirable sensory attributes.

The interaction between boiling, juice color, and storage significantly impacted the visual properties of grape juice. The lightness value consistently declined over the storage period, indicating a progressive loss of visual clarity. This loss corresponds with findings from (Basak et al., 2024), where color parameters are affected by

storage temperature, emphasizing the need for optimal packaging and storage conditions to maintain sensory quality. While boiling did not significantly affect the a^* value, it substantially altered the b^* value, reducing yellow intensity. The decline in chroma and vibrancy observed in our study is consistent with findings reported by (Mendes et al., 2016), where prolonged storage was linked to color loss due to degradation of anthocyanins and other pigments. The slight impact on the hue angle further suggests that while color stability remains an issue, boiling provides a temporary improvement in clarity and visual appeal.

Thermally processed juices have been reported to exhibit significant changes in flavonoid profiles due to pasteurization, which often leads to a

reduction in flavonoid content, particularly in heat-sensitive flavonoids. Saikia et al., (2016) demonstrated that conventional pasteurization results in greater degradation of phenolic compounds, notably flavonols, compared to milder heat treatments like pulsed electric field assisted thermosonication. High temperatures induce thermal degradation, negatively impacting TPC, which correlates with flavonoid levels (Ramírez-Moreno et al., 2017). Nonprocessed juices, which remain unaltered by thermal treatment, typically maintain their flavonoid levels during initial storage periods. However, they are more susceptible to environmental factors, such as light exposure, oxygen interaction, and temperature fluctuations, leading to rapid oxidative degradation of bioactive compounds (Topuz et al., 2014; Nayak et al., 2018). Nayak et al (2018) also documented that fresh star fruit juice maintained its flavonoid content initially, but substantial losses occurred during extended storage at ambient temperatures. This susceptibility is attributed to oxidative mechanisms that are accelerated in the absence of processing. Thermal processing causes the inactivation of antioxidant enzymes like polyphenol oxidase and may extend the stability of grape juices (Umair et al., 2022). A study conducted by Silva et al (2019) reported that grape juices obtained from hot pressing and hot breaking contained more than twice the catechin content compared to those made through cold pressing. Additionally, the researchers found significantly higher levels of total flavanols in the heat-treated juices. In contrast, Mrmošanin et al (2014) observed a decrease in catechin content, which fell from 0.132 mg g^{-1} to 0.113 mg g^{-1} between the 7th and 30th days in cocoa. They also noted a decline in total flavonoid content, which decreased from 14.2 mg g^{-1} to 12.3 mg g^{-1} after heating at 95°C for 25 minutes and then storing the juice at 4°C for 7 days. In our study, we also observed a decline in both red and white grape juices; however, the decrease in white juice was significant, while the reduction in red juice was minimal. This difference may be attributed to the

varying thermal stability of the flavonoid classes present in red and white grape juices.

Over the storage period, we observed an initial decline in catechin levels, followed by partial recovery, indicating susceptibility to degradation followed by possible stabilization. Similar patterns of flavonoid degradation and recovery were observed in other studies, including (Klimczak et al., 2007), which documented fluctuations in polyphenol concentrations during storage. The initial degradation can be attributed to oxidation and interaction with oxygen present in the juice, as supported by the mechanisms outlined in Abountiolas and do Nunes., (2017), where oxidative reactions negatively impacted the stability of anthocyanins and other phenolics. Quercetin levels were significantly affected by boiling, showing a notable increase, although a gradual decline was observed over time, suggesting limited storage stability. This aligns with the findings of (Saikia et al., 2016), which indicated that thermal processing enhances the extractability of antioxidants, although prolonged storage can lead to their degradation. Furthermore, Igual et al (2011) noted improved storage stability of flavonoids following thermal treatment in grapefruit juice, which supports our findings. However, the changing trend, particularly for quercetin, differed significantly between red and white grape juices, likely due to variations in the kinetics of degradation and polymerization of polyphenols in the juices, the phenomenon reported by Danisman et al (2015), and Oancea (2021).

Rutin levels increased significantly with boiling; however, it remained stable across both grape juices and storage durations. Both red and white samples demonstrated comparable extraction efficiency, indicating that rutin's stability is less affected by processing conditions, a finding supported by (Hafizov and Hafizov., 2020). The mechanisms by which rutin is stabilized can include its formation of complexes with other compounds in the juice, which may protect it from degradation.

Our findings regarding organic acids

demonstrated notable impacts of boiling and storage time on tartaric, citric, and malic acids. Tartaric acid concentrations increased from 40.17 g L⁻¹ unboiled to 44.53 g L⁻¹ boiled, aligning with previous findings from (Puzovic et al., 2024), indicating that boiling enhances the extraction of organic acids through evaporation of water, concentrating flavor and acidity. This increase can also lead to a lower pH, promoting anthocyanin stability, as high acidity is known to protect these pigments from degradation (Coelho et al., 2018). Conversely, citric acid demonstrated higher sensitivity to boiling and color conditions, peaking in boiled samples. The significant increase in citric acid reflects the findings of (dos Santos., 2014), which noted that heating not only promotes extraction but also can alter the composition of the juice matrix, affecting overall flavor and stability. On the other hand, even though the preservation of tartaric acid is vital, the notable increase in citric acid by boiling can alter the taste of grape juice, resulting in undesirable taste (Mato et al., 2005).

Fumaric acid exhibited notable thermal and compositional stability across treatments, with concentrations remaining consistent between boiled and unboiled samples, indicating resistance to thermal degradation. This observation parallels results from (Klimczak et al., 2007), where stability was noted in certain organic acids across varying processing conditions. The lack of significant differences in fumaric acid concentrations between different color juices suggests that its stability may derive from its inherent chemical properties, which offer resilience to thermal degradation. In contrast, ascorbic acid levels were more variable and sensitive to experimental conditions. Our results showed that boiling slightly increased ascorbic acid concentrations, but storage influenced its levels significantly, with an initial decline followed by a rebound at 20 days, particularly in red samples. The higher retention of ascorbic acid in red juice corroborates findings by Silva et al., (2018), suggesting that red pigments may help preserve or enhance ascorbic acid stability during

storage. This rebound in ascorbic acid, as noted in our study, might be attributed to the breakdown of other compounds that release bound ascorbic acid during storage.

The biochemical changes in grape juice are complex and influenced by numerous factors. Boiling is known to deactivate enzymes that would otherwise degrade anthocyanins, temporarily enhancing color intensity but potentially leading to pigment degradation through oxidative reactions if exposed to prolonged heat (Mokhtari et al., 2018). High temperatures can generate undesirable compounds that affect both taste and color, as noted by Düsman et al., (2014). Therefore, finding a balance in boiling time and temperature is crucial to maximize the extraction of beneficial compounds while minimizing the adverse effects on color and flavor. This study's findings on the integrated effects of boiling, color treatment, and storage duration contribute to understanding juice processing and the mechanisms behind the stability of nutrients and sensory attributes. Optimizing conditions for boiling and storage can enhance both the nutritional profile and sensory qualities of grape juice, ensuring a product that meets consumer demands for health and quality.

Conclusions

This study demonstrated that both boiling treatment and storage duration significantly influence the biochemical composition of grape juice. The results revealed that thermal processing plays an important role in modifying the levels of several phenolic compounds and organic acids. Boiling generally increased the concentrations of certain bioactive compounds such as catechin, gallic acid, and several hydroxybenzoic acids, suggesting that heat treatment may enhance the extraction or release of these compounds from grape tissues. The findings also showed that grape juice color was an important factor affecting phytochemical composition. Red grape juices generally exhibited higher levels of several phenolic compounds, including catechin and syringic acid, while white

grape juices were characterized by higher concentrations of certain organic acids such as citric, malic, and succinic acids. These differences reflect the natural variation in phenolic and organic acid profiles between grape varieties. Storage duration had varying effects on the measured compounds. While some phenolic compounds showed gradual fluctuations during storage, certain organic acids remained relatively stable. Fumaric acid showed limited sensitivity to processing conditions, whereas ascorbic acid and several other organic acids exhibited changes during storage. PCA analysis further confirmed that boiling and grape color were the primary factors influencing biochemical variation in grape juices. Overall, the results highlight the complex interactions between thermal processing, grape variety, and storage conditions in determining the stability of phenolic compounds and organic acids in grape juice. These findings provide valuable insights for both household juice preparation and industrial processing, suggesting that controlled thermal treatment and proper storage conditions can help maintain the nutritional and functional quality of grape juice.

From a practical perspective, the results suggest that optimizing thermal processing and storage conditions may represent an effective strategy for preserving phenolic compounds and organic acids in grape juice, thereby improving product quality, nutritional value, and shelf-life in both domestic and industrial applications.

Declarations

Ethical Approval Certificate

Ethics committee approval is not required for this study.

Author Contributions

Conceptualization, E.G., L.G.; methodology, E.G., L.G.; software, E.G. L.G.; validation, E.G., L.G.; and Y.T.; formal analysis, E.G., F.M.; investigation, E.G., L.G., and F.M.; resources, E.G.; data curation, E.G., L.G., and F.M.; writing—original draft preparation, E.G., L.G., and F.M.; writing—review

and editing, E.G., L.G., and Y.T.; visualization, E.G., L.G.; funding acquisition, E.G., Y.T. All authors have read and agreed to the published version of the manuscript.

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