Phytochemical Profile and Antioxidant Properties of Vinegars Obtained from Naturally Grown Berry Fruits in the Black Sea Region

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Fruit vinegar samples obtained from six different fruits (hawthorn, strawberry, mulberry, kiwi, grape, and rosehip) were evaluated in terms of total phenolic and flavonoid contents, antioxidant activity (DPPH assay), and selected phenolic and organic acid profiles. The highest total phenolic and flavonoid contents were observed in kiwi vinegar, measuring 676 mg GAE L^{-1} and 694 mg QE L^{-1}, respectively. Regarding DPPH antioxidant activity, the highest values were recorded for kiwi, strawberry, and mulberry fruit vinegars (93%, 92%, and 92%, respectively). Among phenolic acids, mulberry fruit vinegar was rich in protocatechuic acid, rosehip fruit vinegar contained high levels of gentisic acid, and ellagic acid was abundant in both grape and kiwi fruit vinegars. Concerning organic acids, rosehip fruit vinegar exhibited remarkable levels of acetic and tartaric acids. These results demonstrate significant biochemical differences among the studied fruit vinegars and highlight kiwi, strawberry, and rosehip fruit vinegars as particularly valuable sources of phenolic and flavonoid compounds and antioxidant activity.

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Keywords: Wild berries; Biochemical profiling; Phenolic compounds; Flavonoids; Antioxidant activity; Organic acids

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INTRODUCTION

Berry fruits are plant-derived products rich in phytochemicals such as polyphenols, flavonoids, vitamins, and dietary fibers, and they exhibit beneficial effects on human health due to their high antioxidant capacity (Kumar *et al*. 2014; Bouyahya *et al*. 2022; Pepe *et al*. 2023). Owing to these properties, berry fruits hold significant importance in both nutrition and biochemical research. Different agroecological regions of Türkiye provide suitable habitats for both wild and cultivated berry species, including cornelian cherry, strawberry, mulberry, kiwifruit, blackberry, and rosehip (Çolak *et al*. 2022; Ozrenk *et al*. 2020). The phytochemical profiles of samples obtained from these fruits vary considerably depending on species, environmental conditions, genetic background, and processing methods (Skrovankova *et al*. 2015; Alan 2026).

Wild fruit vinegars are particularly important due to their rich bioactive composition, functional food potential, and role as sustainable biological resources. The biochemical compounds in these vinegars, such as phenolic compounds, flavonoids, and organic acids, not only determine antioxidant capacity but also affect the metabolic activities and nutritional quality of the fruits (Granato *et al*. 2017; Rana *et al*. 2022; Çolak *et al*. 2026).

In particular, phenolic compounds are associated with stress responses and defense mechanisms in plants and, in addition to their antioxidant properties, exhibit various biological activities (Zengin 2021; FAO 2025). Similarly, wood is a traditional material frequently used for the maturation of forest-derived food products and contributes to their chemical and sensory characteristics (Cerezo *et al*. 2010). The type and properties of the wood used during maturation can influence phenolic composition, antioxidant activity, and aroma profile (Souza *et al.* 2025). This highlights the importance of forest resources as biological resources both in cultural practices and in the sustainable production of value-added functional foods, establishing a clear link between traditional cultural applications and the findings of this study.

Biochemical variations observed among fruit species and genotypes, in interaction with environmental factors, shape the nutritional value and chemical composition of fruit raw materials and the vinegars produced from them, thereby providing a potential source for functional foods and value-added products (Budak *et al*. 2014; Alan 2026). Furthermore, the Black Sea Region of Türkiye, with its diverse microclimatic conditions and rich wild fruit flora, offers a unique environment for the natural growth and cultivation of these fruits, allowing the production of fruit vinegars with distinct biochemical and organoleptic properties.

The antioxidant capacity of fruit vinegars is closely linked not only to their health benefits but also to fruit quality parameters and the ripening process. As a result of genetic and environmental factors, the relationships among fruit size, weight, acidity, and phenolic content exhibit considerable variability (Bekatorou 2019; Cuamatzin-García *et al*. 2022; Alan *et al*. 2025). Therefore, comprehensive analyses are required to understand the variability in the biochemical profiles of fruit vinegars and to optimize their potential. In addition, wood, as a culturally significant material and biological resource, plays an important role in shaping the sensory and biochemical properties of fermented products. The type of wood used during storage or maturation affects organic acid content, phenolic compound profiles, and antioxidant capacity, thereby determining overall fruit vinegar quality. Understanding these properties provides insight into traditional fermentation practices and links regional cultural heritage with the sustainable use of biological resources.

In this context, the present study aimed to comparatively evaluate fruit vinegar samples obtained from hawthorn, strawberry, mulberry, kiwifruit, grape, and rosehip grown naturally in the Black Sea Region of Türkiye in terms of total phenolic content (TPC), total flavonoid content (TFC), antioxidant capacity (DPPH), phenolic compound profiles (HPLC), and organic acid composition (HPLC-UV). By highlighting the biochemical differences among these fruit vinegar samples, the study contributes to understanding their phytochemical potential and provides a scientific basis for the evaluation of agricultural biodiversity. Furthermore, the findings offer valuable insights into the biochemical characteristics of lesser-known or underutilized fruit vinegars, emphasizing the sustainable use of regional biological resources and the development of value-added food products.

EXPERIMENTAL

Materials

Plant material and vinegar production

In this study, six different wild berry fruits (hawthorn, strawberry, mulberry, kiwi, grape, and rosehip) naturally grown in the Black Sea Region of Türkiye were collected from their natural habitats during the ripening period (June to August 2024) and transported to the laboratory in accordance with sustainability principles to avoid harming local biodiversity. Healthy fruits harvested at full ripeness were transported in plastic crates or mesh bags, washed to remove foreign materials, and processed using traditional methods to produce fruit vinegar samples (Akarca *et al*. 2020; Öztürk 2022). The species used and their collection sites were as follows: hawthorn (*Crataegus monogyna* Jacq., OMUB-8379) from the Ondokuz Mayıs and Vezirköprü districts of Samsun province; strawberry (*Fragaria ananassa* Duchesne, OMUB-1818) from the Espiye and Tirebolu districts of Giresun province; mulberry (*Morus alba* L., OMUB-6427) from the Ünye district of Ordu province; kiwi (*Actinidia deliciosa* A. Chev.) from the Sürmene district of Trabzon province; grape (*Vitis vinifera* L., OMUB-8228) from the Çayeli district of Rize province; and rosehip (*Rosa canina* L., OMUB-8273) from the Merzifon district of Amasya province.

Taxonomic identifications were carried out by Dr. Alper Durmaz. Herbarium voucher specimens representing these taxa were deposited in the Herbarium of the Department of Biology, Faculty of Science, Ondokuz Mayıs University (OMUB) under the corresponding accession numbers. Species identifications were performed in accordance with the *Flora of Türkiye*, and current taxonomic information and accepted species names were verified using the Turkish Plants Database, Plants of the World Online (POWO), and relevant recent scientific literature. According to current legal regulations in Türkiye, these fruit species are either cultivated or not under any conservation status; therefore, no official permit is required for collecting them from their natural habitats. Accordingly, no special permit was obtained for the fieldwork. All collection procedures were conducted following sustainability principles and without causing harm to local biodiversity. Vinegar samples from each fruit were prepared using 1 to 2 kg of fresh fruit and stored under cold conditions until further analyses.

For fruit vinegar preparation, fruits were placed in 5 L glass jars, filling approximately one-third of the jar volume, and mashed using a flat wooden stick to extract the juice. To support fermentation, 0.5% honey and molasses were added, and the jars were filled with natural spring water and covered with cheesecloth. Natural fermentation was carried out at room temperature in the dark, utilizing the native fruit microbiota. During fermentation, the fruit vinegar samples were stirred three times daily to ensure process homogeneity and efficiency, and the fermentation process continued for 60 days, resulting in a vinegar mother layer of approximately 0.5 cm on the surface. After fermentation, the fruit vinegars were filtered to remove fruit residues, transferred into new glass jars, tightly sealed, and stored in the dark at 25 °C until chemical and phytochemical analyses were performed.

Methods

Chemical characteristics; determination of total polyphenol content

The total phenolic content (TPC) in fruit vinegar samples was determined based on the Folin-Ciocalteu method (Gámez-Meza *et al*. 1999), with some minor modifications during implementation. The fruit vinegar samples were filtered using a 0.45-micron membrane filter before analysis. To construct the calibration curve, gallic acid (Sigma-Aldrich, USA), a known phenolic acid, was used as the standard and methanol-based solutions with varying concentrations (500, 250, 100, 80, 60, 40, 20, 10 ppm) were prepared. Absorbance values at 765 nm were plotted on the Y-axis and the corresponding concentrations on the X-axis to generate the standard curve. The R^2 value of the resulting curve was 0.9997, indicating high sensitivity and precision of the method. Using this curve, the TPC of the fruit vinegar samples was calculated as mg GAE L^{-1} dry matter and plotted. For the analysis, 0.1 mL of the fruit vinegar sample was mixed with 7.9 mL of distilled water, 0.5 mL of Folin-Ciocalteu reagent, and 20% sodium carbonate solution. The mixture was incubated at 40 °C for 30 minutes. Absorbance was then measured at 765 nm using a UV-Vis spectrophotometer. The same procedure was applied to standard gallic acid solutions for calibration. Methanol was used in place of the sample for the blank measurement.

Determination of total flavonoid content

The total flavonoid content (TFC) of fruit vinegar samples was analyzed using the aluminum chloride ($AlCl_3$) colorimetric method developed by Woisky and Salatino (1998). The fruit vinegar solutions were prepared at a concentration of 500 ppm using distilled water. Quercetin (Sigma-Aldrich, USA) (QE) was employed as the standard compound to generate the calibration curve. Standard QE solutions were prepared in methanol at concentrations of 500, 400, 300, 200, 100, 50, and 25 ppm. Absorbance values at 415 nm were plotted on the Y-axis and corresponding concentrations on the X-axis to establish the standard curve. The R^2 value of 0.9996 demonstrated the method’s sensitivity. Based on this curve, the TFC of the fruit vinegar samples was calculated and expressed as mg QE L^{-1} fresh weight (FW). For the assay, 0.5 mL of the 500 mg/L fruit vinegar sample was combined with 1.5 mL of methanol, 0.1 mL of $AlCl_3$ solution, and 0.1 mL of 1M sodium acetate. The mixture was kept at room temperature for 30 min, after which absorbance was measured at 415 nm using a UV-Vis spectrophotometer. The same steps were repeated for the QE standards to construct the calibration curve. Methanol was used as a blank instead of the fruit vinegar sample.

Phenolic compound analysis by HPLC

Phenolic compounds in the fruit vinegar samples were analyzed using an Agilent 1260 HPLC system (CA, USA). Chromatographic separation was performed on an ACE-C18 column (4.6 mm × 150 mm, 5 µm). The mobile phase consisted of water with 0.02% trifluoroacetic acid (TFA) as Phase A and methanol with 0.02% TFA as Phase B. The flow rate was maintained at > 0.5 mL min^{-1}. The gradient program used was as follows: 0–5 min: 25% B; 5–10 min: 25–30% B; 10–16 min: 30–45% B; 16–18 min: 45% B; 18–25 min: 45–80% B; 25–30 min: 80% B; 30–40 min: 80–25% B.

The column temperature was kept constant at 25 °C and 10 µL injection volume was used. Detection was performed using a Diode Array Detector (DAD) at four selected wavelengths: 254, 275, 305, and 320 nm. Prior to each run, the system was warmed up and monitored until the baseline stabilized. Detection wavelengths were selected based on the absorption maxima of the targeted phenolic compounds. For example: Syringic acid, protocatechuic acid, and gallic acid (all Sigma-Aldrich, USA) were detected at 280 nm; Vanillic acid at 225 nm; *p*-Coumaric acid (Sigma-Aldrich, USA) at 305 nm; Caffeic acid and chlorogenic acid (Sigma-Aldrich, USA) at 330 nm (Wen *et al*. 2005).

Quantification of organic acids by HPLC-UV

The quantification of organic acids and phenolic compounds in fruit vinegar samples was carried out using an Agilent 1260 HPLC system (Agilent Technologies, CA, USA), equipped with ChemStation software, a quaternary pump, an autosampler and a DAD detector. Organic acids were separated on an ACE-C18 column (4 mm × 150 mm, 5 µm; Hichrom Ltd., Theale, UK) under isocratic elution conditions. The mobile phase consisted of a 10 mM potassium phosphate buffer adjusted to pH 2.2 with ortho-phosphoric acid. The flow rate was kept at > 1.0 mL min^{-1}, and a 20 µL injection volume was used. Detection was performed at 245 nm for ascorbic acid and 210 nm for other organic acids (standards purchased from Sigma-Aldrich, USA) (Fu *et al*. 2015).

Antioxidant activity

Antioxidant activity was determined using the DPPH (2,2-diphenyl-1-picrylhydrazyl) radical scavenging method with minor modifications (Ebrahimzadeh *et al*. 2008). The stock reagent solution (6×10^{-5} M) was prepared by dissolving 0.0024 g of DPPH in 100 mL of methanol. A working DPPH (Sigma-Aldrich, USA) solution (40 mg 100 g^{-1}) was then obtained by diluting the stock solution with methanol. In the assay, 300 µL of the fruit vinegar extract was mixed with 5700 µL of the DPPH working solution in a 10 mL test tube. The reaction mixture was vortexed thoroughly and incubated in the dark at room temperature for 60 min. After incubation, the absorbance of the reaction mixture was measured at 517 nm using a UV-Vis spectrophotometer (Shimadzu UV-1800). A control solution without extract was also prepared and its absorbance was measured at the same wavelength. The antioxidant activity was calculated using the following equation,

$$\text{Antioxidant activity (\%)} = [(AC(O)_{517} - AA(t)_{517}) / AC(O)_{517}] \times 100 \quad (1)$$

where $AC(O)_{517}$ is the absorbance of the control at time $t = 0$ min and $AA(t)_{517}$ is the absorbance of the antioxidant sample after $t = 1$ h.

Statistical Analysis

All statistical analyses were performed using IBM SPSS Statistics version 21.0 (IBM 2013). Descriptive statistics, including mean and standard deviation (SD) were calculated for each experimental group and variable. To assess statistically significant differences between groups, one-way analysis of variance (ANOVA) was performed independently for each measured compound in fruit vinegar samples. Before ANOVA, assumptions of normal distribution and homogeneity of variances were assessed using the Shapiro-Wilk test and Levene's test, respectively. In cases where ANOVA showed significant differences ($P < 0.05$), Tukey's Honestly Significant Difference (HSD) test was used as a post hoc procedure to determine group differences among the vinegar samples. Variables that did not exhibit variance (*i.e.*, constant across all groups) were excluded from inferential statistical tests and reported only in descriptive terms. Results are presented in the format of mean ± standard deviation (Mean ± SD) to ensure clarity in representing both central tendency and variability.

In this study, Principal Component Analysis (PCA) was used as a multivariate statistical technique to reduce data dimensionality and to explain the relationships between variables and fruit vinegar sample groups. Before analysis, the Standard Scaler method standardized the data set to ensure that all variables contributed equally regardless of their original scales. To determine the relative impact of each variable on the principal components, loading scores were calculated and visualized as vectors in the biplot to show the direction and magnitude of each variable's contribution.

All PCA calculations were performed using PAST version 4.03 (Paleontological Statistics Software). The results' graphical representation was prepared using the Python programming language to facilitate clearer interpretation of group clustering and variable importance (Aytar *et al*. 2024).

RESULTS AND DISCUSSION

In this study, the TPC, TFC, antioxidant activity (DPPH assay), and selected phenolic and organic acid profiles of six different fruit vinegars were examined (Table 1). Regarding TPC, the highest value was observed in kiwi vinegar (676 mg GAE L^{-1}), followed by strawberry vinegar (630 mg GAE L^{-1}), rosehip vinegar (503 mg GAE L^{-1}), grape vinegar (503 mg GAE L^{-1}), hawthorn vinegar (459 mg GAE L^{-1}), and mulberry vinegar (404 mg GAE L^{-1}). For TFC, the highest values were detected in kiwi vinegar (694 mg QE L^{-1}) and strawberry vinegar (669 mg QE L^{-1}), while the lowest value was found in rosehip vinegar (350 mg QE L^{-1}). Antioxidant activity, evaluated using DPPH radical scavenging capacity, was highest in kiwi (93%), mulberry (92%), and strawberry vinegars (92%). Regarding phenolic compounds, protocatechuic acid (ProA) content was highest in kiwi vinegar (6.13 mg 100 g^{-1}) and mulberry vinegar (6.10 mg 100 g^{-1}), whereas lower levels were observed in strawberry vinegar (2.84 mg 100 g^{-1}), rosehip vinegar (3.41 mg 100 g^{-1}), and grape vinegar (3.41 mg 100 g^{-1}). This indicates that even vinegars with high TPC do not necessarily exhibit equally high levels of all individual antioxidant compounds. Gentisic acid (GenA) content was highest in mulberry vinegar (52.1 mg 100 g^{-1}) and strawberry vinegar (45.4 mg 100 g^{-1}), while lower levels were recorded in rosehip vinegar (32.3 mg 100 g^{-1}), grape vinegar (32.6 mg 100 g^{-1}), kiwi vinegar (40.4 mg 100 g^{-1}), and hawthorn vinegar (42.4 mg 100 g^{-1}). Ellagic acid (EllA) was most abundant in mulberry vinegar (9.4 mg 100 g^{-1}), with other vinegars exhibiting similar levels ranging from 8.6 to 8.7 mg 100 g^{-1}. In terms of organic acids, acetic acid content was highest in rosehip vinegar (326 mg 100 g^{-1}), followed by kiwi vinegar (308 mg 100 g^{-1}) and grape vinegar (287 mg 100 g^{-1}). Tartaric acid content was also highest in rosehip vinegar (463 mg 100 g^{-1}). Regarding citric acid (CitA), higher values were observed in strawberry vinegar (2.33 mg 100 g^{-1}), mulberry vinegar (2.02 mg 100 g^{-1}), and hawthorn vinegar (2.01 mg 100 g^{-1}), whereas kiwi vinegar (1.05 mg 100 g^{-1}), grape vinegar (1.04 mg 100 g^{-1}), and rosehip vinegar (1.33 mg 100 g^{-1}) exhibited lower levels. These findings demonstrate that each fruit vinegar possesses a distinct biochemical profile, and these differences directly influence the functional properties of the fruit components. Several studies on fruit vinegars have reported similar findings regarding phytochemical composition and antioxidant activity. For instance, Dilimen *et al*. (2021) reported that kiwi vinegars produced from both kiwifruit and kiwi peels exhibited high total phenolic contents, and that during fermentation, pH values decreased while total acidity increased. This indicates the impact of fermentation conditions on vinegar quality. Similarly, Cosmulescu *et al*. (2022) reported that traditional homemade fruit vinegars had a broader range of phenolic compounds and higher bioactive profiles compared to commercial vinegars, including compounds such as gallic acid, catechin hydrate, chlorogenic acid, and syringic acid.

Table 1. Phytochemical Composition and Antioxidant Activities of Six Fruit Vinegars (Mean ± SD)

Variable	Abbreviation	Unit	A (Hawthorn vinegar)		B (Strawberry vinegar)		C (Mulberry vinegar)		D (Kiwi vinegar)		E (Grapes vinegar)		F (Rosehip vinegar)	
Total Phenolic Content	TPC	mg GAE L⁻¹	459.00±1.03	e	630.00±0.96	c	404.00±1.81	f	676.00±1.23	b	503.00±0.85	d	503.00±0.85	d
Total Flavonoid Content	TFC	mg QE L⁻¹	598.00±0.74	c	669.00±1.60	b	498.00±1.74	d	694.00±1.00	a	406.00±2.16	e	350.00±1.80	f
Antioxidant activity	DPPH	%	87.00±1.18	c	92.00±1.39	ab	92.00±1.86	ab	93.00±2.51	a	91.00±2.97	b	88.00±1.00	c
Protocatechic Acid	ProA	mg 100 g⁻¹	6.03±1.31	a	2.84±2.09	c	6.10±1.68	a	6.13±2.28	a	3.41±1.90	b	3.41±1.73	b
Gentisic Acid	GenA	mg 100 g⁻¹	42.43±0.69	c	45.40±0.90	b	52.10±2.52	a	40.43±2.68	d	32.63±0.97	e	32.34±1.85	e
Acetic Acid	AceA	mg 100 g⁻¹	254.67±9.02	d	188.33±4.93	e	127.00±4.58	f	308.33±3.51	b	287.33±8.50	c	325.67±12.58	a
Tartaric Acid	TarA	mg 100 g⁻¹	216.00±7.55	c	186.67±4.51	d	216.67±6.51	c	217.67±6.66	c	282.67±3.06	b	463.00±29.55	a
Ellagic Acid	EllA	mg 100 g⁻¹	8.60±0.78	b	8.63±0.30	b	9.40±2.15	a	8.77±0.23	b	8.76±0.28	b	8.70±0.14	b
Oxalic Acid	OxaA	mg 100 g⁻¹	185.67±3.51	a	179.67±4.04	a	179.33±4.16	a	119.67±4.51	b	178.33±4.51	a	185.67±1.53	a
Citric Acid	CitA	mg 100 g⁻¹	2.01±2.08	a	2.33±1.76	a	2.02±1.73	a	1.05±2.31	b	1.04±0.56	b	1.33±0.94	b

Values within the same column followed by different letters are significantly different at the $P < 0.05$ level.

Ousaaid *et al*. (2021), in a review of fruit vinegars, emphasized that fruit-based vinegars contain rich bioactive compositions, with polyphenolic and organic acids contributing to antioxidant properties and potential health benefits. In research on Cornelian cherry vinegars, Kawa-Rygielska *et al*. (2018) showed that total polyphenol content ranged between 327 and 757 mg GAE L^{-1}, and antioxidant activities (DPPH and FRAP) varied significantly depending on the fruit variety and fermentation method. Together, these studies support the high phytochemical diversity and antioxidant potential observed in the present work.

Compared with previous literature, the total phenolic content (TPC) and antioxidant activity values in the studied kiwi and strawberry vinegars are generally consistent with reported ranges. However, some variations observed among different fruit vinegars may be attributed to differences in raw materials, fermentation conditions, and analytical methods.

In this study, although comprehensive biochemical analyses were conducted, fundamental physicochemical parameters such as °Brix, pH, and total acidity were not assessed. Nevertheless, these parameters are critical for evaluating vinegar quality, fermentation efficiency, and stability. Specifically, low pH values (typically ranging from 2.0 to 3.5) have been reported to limit oxidative degradation of phenolic compounds, thereby enhancing their stability and preserving antioxidant capacity (Friedman and Jürgens 2000). Similarly, total acidity not only affects the sensory properties of the product but also influences the bioavailability and functional properties of organic acids formed during fermentation (Solieri and Giudici 2009). In this context, the high phenolic content and antioxidant activity observed in our samples may be associated with the acidic nature of the vinegar matrix. However, the lack of measurement of these parameters limits the direct assessment of this relationship. Therefore, future studies incorporating these fundamental physicochemical parameters are recommended to enable a more comprehensive and holistic evaluation of vinegar samples.

Rosehip is rich in phenolic compounds and certain organic acids, whereas kiwi stands out with its high TFC and strong antioxidant capacity. The antioxidant properties of these vinegars are important because of their potential beneficial effects on health. In this context, studies investigating biochemical variations among different vinegar samples have attracted attention in the literature. Marangoz (2016) examined the bioactive compounds and antioxidant capacity of black mulberry fruit. The study observed some variations in phenolic content, although these changes were reported as not statistically significant. However, increases in catechin, caffeic acid, and chlorogenic acid levels were recorded. The high TPC and antioxidant activity values observed in the present study are consistent with Marangoz's findings and align with studies suggesting that certain phenolic compounds can increase naturally over time. In a study conducted by Budak *et al*. (2014), gallic acid, chlorogenic acid, syringic acid, p-coumaric acid, and catechin were detected in white mulberry samples, with gallic and chlorogenic acids being the dominant phenolic compounds. In the same study, the TPC of fig samples was reported as 767 mg GAE L^{-1} and mulberry samples as 558 mg GAE L^{-1}. In another measurement, the TPC of mulberry samples was reported as 973 mg GAE L^{-1}. The phenolic compounds identified in the present study were similar to those reported by these researchers, confirming that these compounds are among the major phenolics in vinegar samples (Zengin 2021). Şengün and Kılıç (2020) reported that fig samples had higher total phenolic content than mulberry samples. Additionally, an earlier study by Şengün (2013) found that mulberry samples had higher total acidity than fig samples.

The results obtained in the present study also demonstrated differences in phenolic content and acidity levels among the vinegar samples, consistent with the existing literature. In their study, Xiong *et al*. (2025) reported TPC values of 523 to 558 mg GAE L^{-1} in mulberry samples and noted a positive correlation between DPPH activity and TPC. These findings are in parallel with the phenolic and antioxidant differences observed among vinegar samples in the present study. Öztürk (2021) reported that homemade and commercial vinegar samples had total acidity values of 3.89 mg 100 g^{-1} and 10.52 mg 100 g^{-1}, respectively, with dry matter contents of 40.7% and 26.6%. Öztürk *et al*. (2015) investigated the microbiological characteristics of vinegar samples prepared by different methods. In homemade grape vinegars, the highest TPC (2230 mg GAE L^{-1}) and DPPH values were observed. The TPC and antioxidant activity reported for grape samples in Öztürk *et al*. (2015) were higher than those obtained in the present study, which is likely due to differences in grape variety, preparation duration, methods and analytical procedures. Bıldır *et al*. (2023) determined the TPC, TFC and antioxidant capacity of hawthorn vinegar samples as 56.8 mg GAE L^{-1}, 290 mg QE L^{-1} and 53.4 %, respectively. In another study, Tomar *et al*. (2020) reported the total antioxidant activity of hawthorn (*Crataegus tanacetifolia*) vinegar samples as 86.2 µg trolox equivalent (TE)/mL and total phenolic content as 751 mg GAE L^{-1}.

The TPC and antioxidant capacity of hawthorn vinegar samples in this study were in agreement with these findings, indicating that the biochemical properties of hawthorn were preserved. Bozdemir *et al*. (2021) reported that hawthorn vinegar samples had the lowest total acidity, whereas pomegranate vinegar samples had the highest total acidity. Volatile acidity was highest in pomegranate vinegar samples and lowest in hawthorn vinegar samples, while non-volatile acidity was highest in pomegranate and lowest in grape vinegar samples. Additionally, the highest TPC and DPPH antioxidant capacity were observed in pomegranate and grape vinegar samples. Kan (2021) analyzed antioxidant activity, phenolic and flavonoid contents in naturally and industrially produced vinegar samples. The TPC ranged from 210 to 1540 mg 100 g^{-1} in natural vinegar. The DPPH-based antioxidant activity analysis showed that the vinegar sample prepared from hawthorn exhibited the highest antioxidant activity in both natural and industrial groups, while the sample prepared from pomegranate showed the lowest.

The highest flavonoid content was observed in the vinegar sample prepared from pomegranate. Although pomegranate vinegar was not included in that study, similar diversity in phenolic content and acidity was observed among different vinegar samples, with lower acidity detected in hawthorn and grape vinegars, consistent with literature reports. Xiong *et al*. (2025) reported TPC values of 523 to 558 mg GAE L^{-1} in mulberry vinegar samples. Selvanathan and Masngut (2020) compared the phytochemical properties of pineapple vinegar samples with commercial apple and date vinegars. Pineapple peel vinegar had an acidity of 3.03% and vitamin C content of 0.61%, with phenolic content and DPPH antioxidant capacity comparable to commercial samples. No significant differences were observed in TPC and TFC among vinegar samples. In contrast, this study observed notable differences in these parameters among vinegar samples obtained from different vinegar types, indicating that the bioactive compound composition is influenced not only by production method but also by raw material. Öztürk (2022) examined vinegar samples from ‘Cardinal grape’, ‘Napoleon cherry’, plum, kiwi and peach, reporting acidity values ranging from 0.53 to 3.23. The highest acidity was found in ‘Cardinal grape’ vinegar and the lowest in ‘Napoleon cherry’ vinegar. The measured acidity values in this study were generally higher compared to some vinegars, particularly highlighting rosehip and kiwi vinegars. Kahraman *et al*. (2022) reported that homemade grape vinegar exhibited higher total acidity, organic acid content, phenolic compound levels, and DPPH antioxidant activity compared to apple vinegar. Astringent and tartaric acids were dominant in grape vinegar, while acetic and succinic acids were predominated in apple vinegar. Proteggente *et al*. (2002) also reported that the vinegar sample prepared from strawberry is rich in phenolic compounds and antioxidant content, supporting its value as a source of bioactive compounds. Türkol *et al*. (2024) observed increased levels of biologically active compounds, including malic, lactic, and oxalic acids, in vinegar samples prepared from strawberry. Similarly, the high phenolic content and antioxidant capacity found in the strawberry vinegar sample confirmed its significance as a source of bioactive compounds. Gokirmakli *et al*. (2019) reported a total acidity of 4.59 in the vinegar sample prepared from strawberry, with moderate antioxidant activity compared to other vinegar samples. In contrast, the present study observed high phenolic content and antioxidant capacity in the strawberry vinegar sample, indicating some deviations from the literature likely due to differences in analytical methods or sample sources. Özdemir *et al*. (2022) reported that the vinegar sample prepared from rosehip exhibits high TPC, TFC, and strong antioxidant activity. Key phenolic compounds identified in the vinegar samples include catechin, ellagic acid, quercetin, taxifolin, 4-hydroxybenzoic acid, and salicylic acid, with an increase in vitamin C observed during the fermentation process. Similarly, the results showed that the vinegar sample prepared from rosehip is rich in phenolic compounds. Pashazadeh *et al*. (2021) observed high antioxidant activity in the vinegar sample prepared from black rosehip (*Rosa pimpinellifolia* L.), with increases in gallic acid, catechin, ellagic acid, protocatechuic acid, epicatechin, and vitamin C content, supporting the biochemical potential of rosehip.

Principal Component Analysis (PCA) in this study revealed clear differentiation among vinegar samples based on phenolic compounds and organic acid contents (Fig. 1). Rosehip, kiwi, hawthorn, strawberry, mulberry, and grape vinegars were distinctly separated according to their phenolic and organic acid profiles. Rosehip vinegar samples were separated along the positive direction of PC1, influenced by acetic acid, TPC, and tartaric acid. Similarly, kiwi vinegar samples were differentiated along the PC2 axis, associated with protocatechuic acid, TFC, and caffeic acid. These findings demonstrate the critical role of phenolic and organic acid profiles in the chemical diversity of vinegar samples. Similarly, Yıldız (2023) reported that PCA of phenolic and organic acid components in vinegars revealed distinct separations among products, which is consistent with these findings.

The PCA plot explained 60.6% of the total variance, which is considered a moderate level of variance explanation in the literature and is often preferred for exploratory analyses (El Abdali *et al*. 2023). These results were supported by one-way ANOVA and post-hoc tests to ensure the statistical validity of differences among groups. The combined use of these methods provides more reliable outcomes in multidisciplinary analyses (Ghosh and Chattopadhyay 2012). The PCA results in this study indicate that phenolic and organic acid compounds play a discriminative role in the phytochemical profiles of vinegars. Examination of the loading scores presented in Table 2 reveals that gentisic acid (GenA) contributed the most to PC1 with 20.6%, followed by acetic acid (AceA) at 18.8% and tartaric acid (TarA) at 14.7%. These compounds stand out as the primary variables underlying the major differences among the vinegar samples. On the other hand, citric acid (CitA) showed the highest contribution to PC2 at 10.10%, which supports the observed group separations along the PC2 axis.

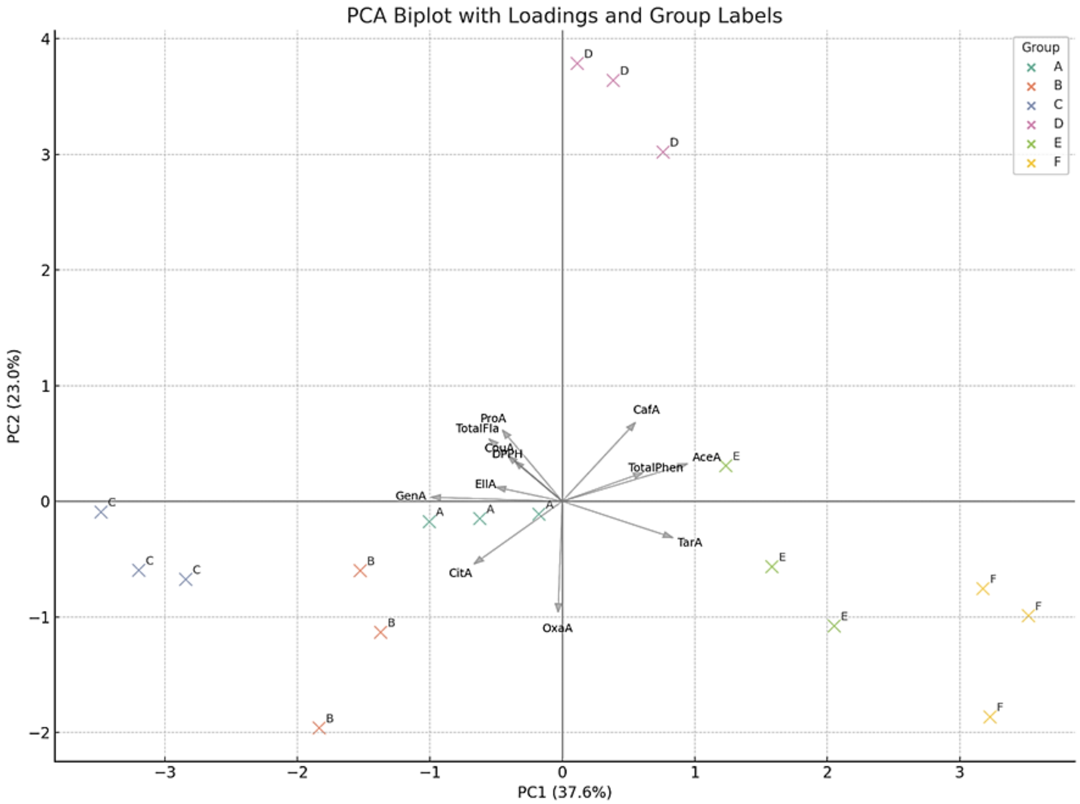

Fig. 1. The PCA biplot illustrating the distribution of vinegar samples based on their phytochemical composition and organic acid profiles

Table 2. Loading Scores of the Measured Phenolic Compounds, Organic Acids, and Antioxidant Parameters Contributing to Principal Component Separation in Fruit Vinegar Samples

Variable	Abbreviation	Unit	PC1 Contribution (%)	PC2 Contribution (%)
Total Phenolic Content	TPC	mg GAE L⁻¹	7.85	2.12
Total Flavonoid Content	TFC	mg QE L⁻¹	5.88	13.51
Antioxidant Activity	DPPH	%	2.50	10.45
Phenolic Compounds				
Protocatechuic Acid	ProA	mg 100 g⁻¹	2.93	20.24
Gentisic Acid	GenA	mg 100 g⁻¹	20.63	0.04
Caffeic Acid	CafA	mg 100 g⁻¹	4.40	8.89
Coumaric Acid	CouA	mg 100 g⁻¹	3.90	12.83
Ellagic Acid	EllA	mg 100 g⁻¹	2.04	4.33
Succinic Acid	SucA	mg 100 g⁻¹	1.36	0.20
Carboxylic Acid	CarbA	mg 100 g⁻¹	1.52	0.34
Gallic Acid	GalA	mg 100 g⁻¹	1.24	0.29
Organic Acids				
Oxalic Acid	OxaA	mg 100 g⁻¹	2.33	6.12
Tartaric Acid	TarA	mg 100 g⁻¹	14.66	3.41
Ascorbic Acid	AscA	mg 100 g⁻¹	1.89	2.19
Acetic Acid	AceA	mg 100 g⁻¹	18.80	3.63
Citric Acid	CitA	mg 100 g⁻¹	9.32	10.10

Similarly, Stoenescu and Stănică (2025) emphasized that phenolic contents and organic acid profiles play a crucial role in determining the chemical profiles of different vinegars, with gentisic acid and acetic acid being particularly influential in the distribution of compounds. Moreover, the richness of rosehip and kiwi vinegars in phenolic and organic acid compounds enhances their biological activities. Yıldız (2023) reported that tartaric and citric acids are important organic acids affecting the quality and chemical properties of vinegars and that PCA analysis of phenolic and organic acid components revealed distinct separations among the products. These findings are consistent with the results. Similarly, Zhang *et al*. (2019), in their study investigating the effects of phenolic acids on antioxidant properties and aroma compounds, highlighted gentisic and tartaric acids as key determinants of the chemical profile in vinegar samples. Additionally, Hata *et al.* (2023) reported that acetic acid, as the primary organic acid in vinegar fermentation, directly influences the product's biological activities and acidity.

In a study conducted by Sorathiya *et al*. (2025), the presence of various organic acids in vinegars, particularly citric acid, was emphasized as important factors affecting the sensory profiles and microbial stability. This finding supports the contribution of citric acid to PC2 observed in the vinegar samples analyzed in the present study. Furthermore, Ye *et al*. (2024) reported that the phenolic compound and organic acid profiles of vinegars vary according to seasonal and regional conditions, which in turn shape their functional food properties and health effects. This finding helps explain the influence of these components on variance observed among the vinegar samples in this study.

Additionally, the dendrogram obtained from hierarchical cluster analysis, conducted to independently assess sample similarity based on standardized compound concentrations (Fig. 2), reveals clustering patterns largely consistent with the PCA results. Vinegar samples from hawthorn (A), strawberry (B), and mulberry (C) cluster closely together, indicating the compositional similarity. In contrast, the rosehip vinegar group forms a distinct branch, reflecting its unique chemical profile.

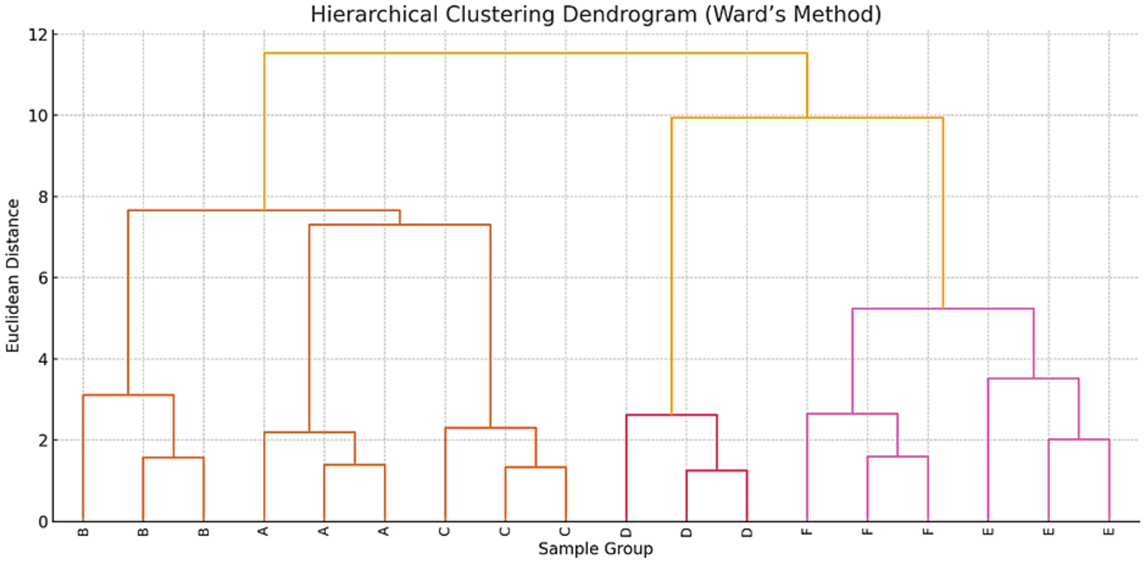

Fig. 2. Hierarchical clustering dendrogram of vinegar samples using Ward's method (A: Hawthorn, B: Strawberry, C: Mulberry, D: Kiwi, E: Grapes, F: Rosehip)

The findings presented in Fig. 2 are consistent with reports by Yıldız (2023) and Ye *et al*. (2024), which emphasized the significant role of phenolic compounds and organic acid profiles in the classification of vinegar samples. Similarly, Hata *et al*. (2023) highlighted that the combined use of hierarchical clustering and PCA analyses is a robust and reliable approach for discriminating against vinegar samples based on their chemical compositions. When these multivariate analyses are considered together, it is concluded that phenolic content and specific organic acid profiles are the main determinants of differences among groups and can be effectively used to distinguish and classify vinegar samples.

CONCLUSIONS

1. This study demonstrated significant differences in the phytochemical contents and antioxidant capacities of vinegars obtained from various fruit species.
2. Statistical analyses, including one-way ANOVA, PCA, and hierarchical clustering, identified key variables such as acetic acid, total phenolic compounds and specific phenolic acids including gentisic, protocatechuic, and tartaric acids as decisive factors in differentiating vinegar sample groups.
3. Rosehip and kiwi vinegar samples stood out with their high total phenolic content and antioxidant capacities, indicating that these vinegars contain biochemically valuable components.
4. Standardization of production processes and the establishment of quality criteria for raw material selection are essential to ensure consistency and reliability of the vinegar samples.
5. This study contributes to the scientific elucidation of the potential of traditionally underexplored fruit vinegars and offers new perspectives for understanding biochemical diversity and the sustainable utilization of natural resources.

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