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IDENTIFICATION OF FACTORS INFLUENCING THE COMPOSITION AND ANTIOXIDANT ACTIVITY OF GRAPE POMACE AND ITS EXTRACTS

The object of research is the quality of grape berry components and the extracts obtained from them. The effect of grape variety and extraction method on the composition and antioxidant properties of berry components and their extracts has not been sufficiently studied.

It was found that, in terms of flavonoid content, the Madrasa and Merlot varieties are superior, while Merlot is also distinguished by higher tannin content. Among the white varieties, Rkatsiteli stands out with relatively higher indicators. In terms of antioxidant activity, the pomace of Rkatsiteli (79.2%) and Bayan Shirey (75.6%), as well as the seeds of Madrasa (87.1%) and Rkatsiteli (86.9%), showed the highest values. The CO₂ extract of grape skins demonstrated a higher radical scavenging capacity (128.1 mg/ml) and antioxidant activity (73.1%) compared to other samples.

This is explained by the biochemical characteristics of grape varieties, the fact that phenolic compounds are mainly concentrated in the skins and seeds, and the efficiency of extraction methods. Since CO₂ extraction is carried out at low temperatures in an inert gas environment, bioactive compounds are better preserved, resulting in higher antioxidant activity.

A comprehensive evaluation of the effect of grape variety and extraction method on the composition and antioxidant activity of berry components constitutes the features of this research.

The results can be applied in the food industry, winemaking, and pharmaceuticals. Their practical application ensures higher efficiency under optimal conditions – such as selecting suitable grape varieties, proper preparation of raw materials, and the use of more effective extraction methods like CO₂ technology.

Keywords: wine waste, grape skins, seed, pulp, grape pomace, antioxidant activity, radical scavenging activity phenolic compounds, anthocyanins.

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1. Introduction

During winemaking, approximately 17% of by-products known as grape pomace and stems are generated, representing a significant proportion of total production. These residues are rich in dry matter and its constituent components, including carbohydrates, phenolic compounds, nitrogenous substances, and other bioactive compounds. Therefore, these by-products are considered secondary raw material resources.

The main fraction of secondary resources formed during grape processing is seed-containing pomace. Specifically, stems account for 3–7% of the cluster weight, skins constitute 15–20% of the berry weight, and seeds represent 3–6% of the berry weight. In grape pomace, the seed content comprises approximately 20–25% of the total mass.

Efficient processing of these residues enables the recovery of polyphenols, vitamins, dietary fibers, and compositionally rich extracts. Moreover, dietary fibers and extracts obtained from these by-products are rich in biologically active compounds and can be used as fortifying ingredients in food products. As such, grape processing residues possess a rich chemical composition, and their recycling allows the production of a wide range of high-value products. However, due to the lack of cost-effective and accessible technologies adapted to local conditions, thousands of tons of these by-products are discarded annually without utilization.

In paper [1], the results of studies on the evaluation of grape stems, particularly their phenolic composition and therapeutic potential, are presented. It is shown that experimental results are provided on the health effects of polyphenols obtained from grape stems, their metabolism, and the role of intestinal microflora (microbiota) in these processes. However, these studies do not cover issues related to the composition of berry components.

In paper [2], updated information is presented on the composition of grape pomace obtained from winemaking, with particular emphasis on the sustainable extraction of phenolic compounds and their potential health benefits – antioxidant, antimicrobial, antidiabetic, cardioprotective, antiproliferative, anti-aging, and gut health-supporting properties. It is shown that the bioactive substances obtained from it can also be applied in the food, cosmetic, and pharmaceutical industries. However, these studies do not investigate the factors affecting the content of individual structural elements of grape pomace.

In paper [3], for the first time, the phenolic, antioxidant, macro- and micronutrient composition of grape pomace and seeds obtained from Lacrima di Morro d'Alba red and Verdicchio white grape varieties was evaluated, and results are presented indicating that they are rich sources of phenolic and nutritional compounds. However, the influence of variety and processing method on antioxidant properties is not examined in this research.

In paper [4], the results of studies conducted on grape pomace obtained from seven grape varieties over three consecutive vintages (2022–2024) are presented, including fatty acid methyl ester (FAME) profiles, total phenolic content (TPC), total flavonoid content (TFC), and antioxidant activity (DPPH, ABTS, ORAC analyses). However, the effect of grape variety on these processes is not investigated.

In paper [5], research results are presented on the microencapsulation of phenolic compounds obtained from grape pomace extract (GPE) from the Sonora region of Mexico. It is shown that microencapsulation was carried out using spray-drying, with maltodextrin and gum arabic as carrier agents. The main aim of the research was to determine the applicability of the bioactive extract in Greek-style yogurt products. However, the compositional indicators of individual berry components were not investigated.

In paper [6], gastrointestinal digestion models and factors affecting the bioavailability of phenolic compounds were analyzed, and results are presented on the behavior of grape-derived phenolic compounds during digestion under different food matrix conditions (presence or absence). However, clinical studies are not included in this research.

In paper [7], the results of a study on the optimization of pulsed electric field (PEF)-assisted extraction to sustainably increase the efficiency of phenolic compound extraction from white grape stems are presented. It is shown that under optimized conditions, PEF-treated samples exhibited 8% higher total phenolic content, 31% higher flavonoid content, and 36% higher antioxidant activity (FRAP). However, the extraction process of other solid parts of the cluster was not studied.

In paper [8], the results of researches on the composition of bioactive compounds, polyphenol profiles, and antioxidant activity of white and red grape pomace are presented. It is shown that the comparison of phenolic compounds characteristic of each pomace indicates differences in the chemical and functional properties of grape varieties. However, extracts obtained from the solid parts of the cluster are not presented in this research.

In paper [9], results are presented on the variability of polyphenol content in grape pomace, skin, and seed extracts obtained from white and red wine production of different Italian grape varieties. It is shown that total polyphenol content (GAE), main polyphenol classes, and antioxidant activity (DPPH) were determined. Significant differences in flavonol profiles were observed both among different grape varieties and between fermented and non-fermented stems. However, the factors influencing these indicators were not studied.

In paper [10], it is reported that Neuburger (NE), Radames (RA), and Regent (RE) varieties have the highest phenolic concentrations, particularly rich in catechin, epicatechin, and procyanidin dimers. It is shown that Neuburger (NE) and Fetească Regală (FR) exhibit the strongest radical scavenging and electron transfer activity in various antioxidant assays. However, the factors affecting antioxidant activity were not investigated.

In paper [11], results obtained using high-performance liquid chromatography (HPLC) analysis show that the pomace extract of Negru de Drăgășani contains high amounts of biologically active polyphenolic compounds such as catechin, myricetin, resveratrol, and vanillic acid.

In paper [4], research results are presented on the bioactive composition, polyphenol profile, and antioxidant activity of grape pomace obtained from various white and red grape varieties. It is shown that each type of grape pomace contains characteristic phenolic compounds. However, the factors affecting polyphenol profile and antioxidant activity were not investigated.

In paper [8], results are presented on the variability of polyphenol content in grape skin and seed extracts obtained from both white and red wine production of different Italian grape varieties. Data on total polyphenol content (GAE), main classes of polyphenols, and DPPH values are also provided. However, the influence of grape variety on antioxidant activity was not examined.

In paper [12], research results are presented indicating that white grape pomace extract has a cardioprotective effect against myocardial infarction by reducing inflammation and oxidative stress. However, clinical studies were not included.

In paper [13], it is shown that the skin of *Vitis labrusca*, particularly as a by-product of winemaking, can be a promising natural source of bioactive compounds for preventing and treating diseases related to oxidative damage and bacterial infections. However, clinical studies are lacking.

In paper [14], results are presented on the effects of grape seed and skin extract (GSSE) on human health. It is shown that GSSE contains high concentrations of biologically active polyphenols. Seed extract of *Vitis vinifera L.* reduces oxidative stress and improves overall lipid metabolism due to its anti-inflammatory, anti-apoptotic (reducing cell death), and proliferative (regenerative) effects. However, these findings are not supported by clinical studies.

In paper [15], it is shown that grape pomace has a wide diversity in chemical composition and functional properties and is beneficial for use in functional foods, nutraceuticals, and cosmetics. It is emphasized that this should be considered within the framework of circular economy principles. However, direct experimental studies on grape pomace are not presented.

In paper [9], results are presented on studies of various by-products of winemaking—grape pomace, lees, and grape seed meal—across three production methods (red, rosé, and white) and four grape varieties (Tempranillo, Syrah, Airén, Zalema) over two harvest seasons (2022 and 2023). It is shown that samples were differentiated by by-product type, production method, and grape variety using linear discriminant analysis (LDA) based on chemical and antioxidant parameters. However, studies on the composition and antioxidant properties of extracts were not included.

In paper [16], it is shown that red grape pomace is a sustainable source of phenolic compounds and has high application potential in pharmaceutical, cosmetic, and food industries. However, specific studies on its application in these fields are not provided.

In paper [17], it is reported that in fermented pomace, the amount of skin increased by 15% and the amount of seeds decreased by approximately 50%. However, the effect of fermentation on the antioxidant activity of skin and seeds was not investigated.

In paper [18], results are presented on the enrichment of low-extract juices and wines with extracts obtained from solid parts of the cluster for the production of functional beverages. However, studies on the extent to which these extracts enhance antioxidant properties are not included.

In paper [19], research results are presented on increasing the bioactivity of grape stems and seeds through solid-state fungal fermentation and enriching extractable phenolic content. It is shown that this approach creates new opportunities for the effective valorization of winemaking by-products. However, studies on other solid components of the berry are not included.

In paper [20], results are presented indicating that buckwheat extract powder can be used as an ingredient in beverages, as a standalone product, and as a recommended product for people with gluten intolerance. However, the factors affecting the antioxidant properties of extracts were not investigated.

In paper [21], research results are presented on the production of functional juices and wines with the addition of rosehip pomace extract. It is shown that pomace powder obtained during rosehip processing can be used in functional yogurt and bakery products (cakes), with optimal addition levels of 3.0% for yogurt and 2.5% for cakes. However, the influence of grape variety and processing method on extract composition was not studied.

Based on the analysis of the literature review, it can be concluded that in these studies the influence of grape variety on the composition

and antioxidant properties of berry components has not been sufficiently investigated, and the polyphenol content and antioxidant activity of extracts obtained from the solid parts of the cluster have not been evaluated in relation to grape variety and processing method. All this allows to conclude that it is advisable to conduct a research devoted to determining the factors affecting the composition and antioxidant activity of grape pomace and its extracts.

The object of research is the quality of grape berry components and the extracts obtained from them.

The aim of research is to determine the factors affecting the composition and antioxidant activity of grape pomace and the extracts obtained from it.

To achieve this aim, the following objectives were addressed:

- 1) to investigate the effect of grape variety on the composition and antioxidant properties of berry components;
- 2) to evaluate changes in polyphenol content and antioxidant activity of the extracts depending on grape variety and processing method.

2. Materials and Methods

The research aimed to investigate factors affecting the production of high-antioxidant and radical-scavenging extracts from residues of locally grown white and red technical grape varieties.

Grape seeds were obtained from mature grape pomace. The pomace was separated into seeds and skins for analysis. The "sweet pomace" fraction was used. Pulp was carefully separated from the seeds, and the seeds were washed and air-dried. Local varieties Madrasé and Bayan Shirey, as well as imported Rkatsiteli, Cabernet Sauvignon, and Merlot, were used. The pomace remaining after juice extraction was used for each variety. Seed fermentation was performed similarly to stems and pomace.

For extraction, 45–80% ethanol solution and liquid CO₂ were used at room temperature. The solid-to-solvent ratio was 1:3.

Samples were analyzed for pH (pH meter, Sartorius PB-II, Germany), total acidity, dry matter, ash, and reducing sugar content. Total monomeric anthocyanins in juice were determined by the pH differential method using malvidin-3-glucoside as a standard (mg/dm³).

Physicochemical and organoleptic characteristics of raw materials, semi-finished, and finished products were determined using standard enochemistry methods [22, 23]. High-performance liquid chromatography (HPLC), computer-assisted statistical analysis, and SPSS18 were applied [24, 25].

Total phenolic content was measured by the Folin-Ciocalteu method. Antioxidant activity was determined by the TEAC method (Trolox equivalent antioxidant capacity) based on the discoloration of the blue ABTS⁺ radical cation at 734 nm. Radical-scavenging activity was expressed in mol/kg relative to Trolox.

The antioxidant activity of samples was also evaluated in a linoleic acid system. The method models in vivo antioxidant activity under conditions of 40°C, pH 7.0, for 120 hours, followed by reaction with NH₄SCN and FeCl₂ to measure the degree of hydroperoxide formation. Antioxidant activity was expressed as the inhibition of linoleic acid oxidation (AOF, %).

3. Results and Discussion

3.1. Effect of grape variety on the composition and antioxidant properties of pomace components

Phenolic compounds, flavonoids, tannins, anthocyanins, and their antioxidant activity were analyzed in the pulp, skin, and seed fractions. Results showed that these parameters, especially antioxidant activity, significantly varied among grape varieties (Tables 1, 2).

Table 1

Composition of pomace components in the investigated red grape varieties

Parameters	By grape varieties								
	Madras			Isabella			Merlot		
	Pulp	Skin	Seed	Pulp	Skin	Seed	Pulp	Skin	Seed
Phenolic compounds, g gallic acid/100 g DM	0.59	3.7	3.5	0.31	2.9	2.6	1.07	3.8	4.9
Flavonoids, g catechin/100 g DM	0.18	0.91	2.6	0.09	1.18	1.7	0.78	1.0	2.2
Anthocyanins, mg cyanidin-3-glucoside equivalents/100 mg raw material	5.1	496.5	–	8.4	131.5	–	19.9	395.4	–
Tannins, mg catechin/100 g raw material	0.12	17.94	9.46	0.16	9.87	8.38	1.15	40.0	42.31
ARF EC ₅₀ , mg/mL	36.5	9.2	0.5	69.3	10.8	0.8	38.1	8.7	0.128
ARA, μmol Fe ²⁺ /1 kg raw material	2.11	26.25	30.01	1.95	6.96	27.8	3.31	7.44	58.01
BX, mmol Fe ²⁺ /1 kg raw material	1.09	7.83	14.58	1.17	11.07	21.96	1.50	1.05	25.32
Antioxidant activity, % ing.	–	–	87.1	13.7	11.3	15.5	15.03	13.96	5.10

Table 2

Composition of pomace components in the investigated white grape varieties

Parameters	By grape varieties					
	Rkatsiteli			Bayan Shirey		
	Pulp	Skin	Seed	Pulp	Skin	Seed
Total phenolic compounds, g gallic acid equivalents (GAE) per 100 g dry matter (DM)	0.31	0.7	4.3	0.25	0.16	2.9
Flavonoids, g catechin /100g DM	0.17	0.34	1.5	0.32	0.23	1.1
Total anthocyanins, mg cyanidin-3-glucoside equivalents (C3G) per 100 mg raw material	–	–	–	–	–	–
Total tannins, mg catechin equivalents (CE) per 100 g raw material	0.09	0.12	18.30	0.04	0.15	1.4
ARF EC ₅₀ , mg/mL	96.1	72.0	4.1	99.3	79.1	4.5
ARA, μmol Fe ²⁺ /1 kg raw material	2.21	4.66	15.51	1.86	3.97	18.24
BX, mmol Fe ²⁺ /1 kg raw material	0.41	2.46	14.55	0.15	1.36	13.31
Antioxidant activity, % ing.	79.2	71.3	86.9	75.6	64.1	85.7

Analysis of Tables 1, 2 indicates that phenolic compounds, flavonoids, tannins, and anthocyanins were significantly lower in the pulp compared to the skins and seeds. Regarding flavonoid content, the Madrasa and Merlot varieties exhibited the highest levels, while Merlot showed the highest tannin content. Among the white varieties, Rkatsiteli distinguished itself with the highest values in both phenolic content and radical scavenging activity.

In the seeds, the levels of flavanols followed the sequence Madrasa > Merlot > Isabella. Anthocyanins were predominantly present in the skins of Madrasé, whereas in Merlot they were found in both the skins and, to a lesser extent, in the pulp. The highest restorative properties were observed in the seeds of the Merlot variety, followed by Isabella. Conversely, the pulp of all varieties exhibited lower restorative activity.

Radical scavenging activity, expressed as EC_{50} (the extract concentration required to neutralize 50% of free radicals), was lower in the pulp compared to seeds and skins in both red and white grape varieties. The lowest antiradical activity was observed in the seeds of Bayan Shirey ($EC_{50} = 4.5$), whereas the highest was in the seeds of Madrasa ($EC_{50} = 0.5$). Skin antiradical activity was highest in Merlot and lowest in Bayan Shirey.

In terms of radical capture capacity, Merlot and Madrasa seeds were superior (58.01 and 30.01 mmol Trolox/100 g DW, respectively), whereas in the skins, Madrasa showed the highest capacity (26.25 mmol Trolox/100 g DW). The radical capture activity of the pulp was relatively uniform across all varieties. Antioxidant activity was highest in the pulp of Rkatsiteli (79.2%) and Bayan Shirey (75.6%), and in the seeds of Madrasa (87.1%) and Rkatsiteli (86.9%).

These results suggest that the Merlot and Madrasa varieties contain the highest levels of bioactive compounds and antioxidants. Bioactive compounds are primarily localized in the skins and seeds of grapes, indicating that pomace, as a secondary raw material, possesses significant potential as a source for high-value products. Utilization of this secondary raw material not only minimizes production waste but also increases the yield of valuable compounds from the raw material.

Furthermore, the analysis of seeds from different grape varieties revealed that they are rich in proteins and lipids (Fig. 1), confirming their potential as a nutritionally valuable component of winemaking by-products.

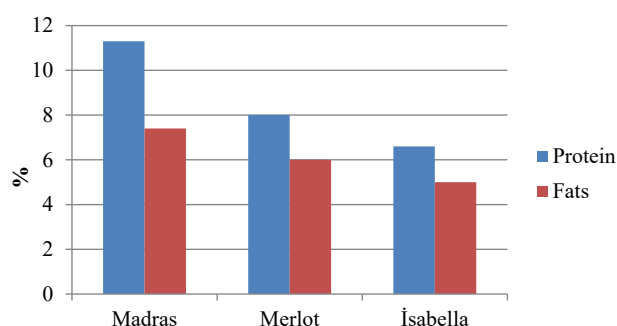


Fig. 1. Protein and fat content in grape seed samples by variety

As observed, the protein content varied significantly among the grape varieties. In the Madrasa variety, seed protein levels were approximately 3% higher compared to Merlot and nearly 4% higher than in Isabella seeds. At the same time, the examined seed samples proved to be rich in fat. Specifically, fat content ranged from 5.0% to 7.4% across the samples. The highest fat content was observed in Madrasa seeds, followed by Merlot and Isabella varieties. Grape seeds are characterized by a high lipid content. The seeds separated from the pomace contain an endosperm, which is encased by a delicate, lignified cuticle with tannins.

The investigated seed samples exhibited significant lipid richness, with lipid content ranging from 5.0 to 7.4%, depending on the variety. This characteristic should be considered an important factor in their valorization and further processing.

However, studies have shown that the beneficial properties of the seeds are not limited to their lipid content. During the investigations, it was also found that the seed samples are a rich source of phenolic compounds (Fig. 2–4).

The total phenolic content in the seed samples ranged from 2950 to 4560 mg/kg^{-1} , with the highest value observed in the Madrasa variety.

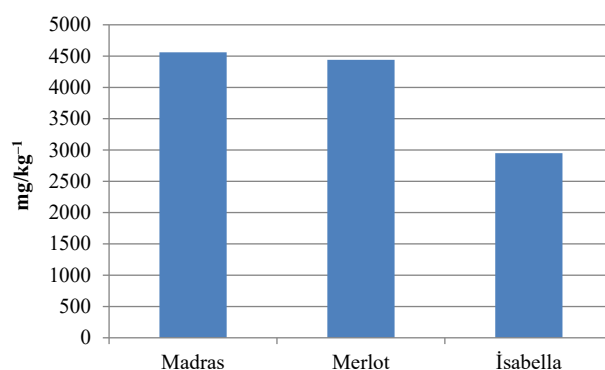


Fig. 2. Total phenolic content (mg/kg) in Seed Samples of different grape varieties

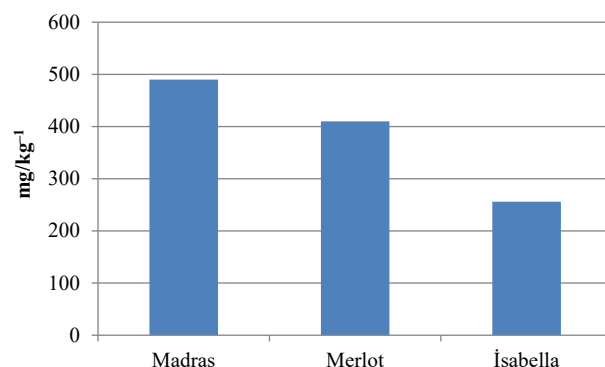


Fig. 3. Amount of flavonoids in seed samples (mg/kg^{-1})

The flavonoid content in the seeds of the grape varieties was 490 mg/kg in Madrasa, 410 mg/kg in Merlot, and 256 mg/kg^{-1} in Isabella, which corresponded to the sequence observed among the varieties in terms of total phenolic compounds

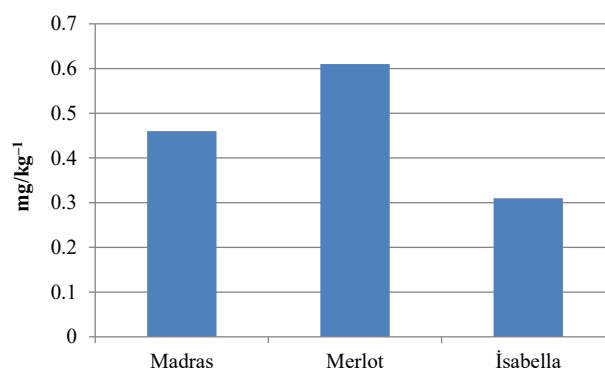


Fig. 4. Anthocyanin content in seed samples (mg/kg^{-1})

The anthocyanin content exhibited a different pattern compared to total phenolic compounds and flavonoids. In particular,

the anthocyanin content was 0.61 mg/kg⁻¹ in the Merlot variety, 0.46 mg/kg in Madrasa, and 0.31 mg/kg⁻¹ in Isabella. Notably, the seeds obtained from Isabella also ranked lowest in terms of other phenolic compounds. Grape seeds contain tannins and leucoanthocyanins. Depending on the grape variety, seeds in the berry account for 22–56% of total polyphenols, 28–56% of leucoanthocyanins, 67–86% of catechins, as well as malic and caffeic acids.

These observations allow to conclude that, similar to their content of oils and proteins, grape seeds are rich in phenolic compounds. Considering the significant technological role of these compounds in food products, it is crucial to organize their efficient extraction from seeds.

Grapes used in extraction technology should be fully ripened and rich in nutrients. The primary composition of seeds in the grape berry includes up to 40% water, up to 35% carbohydrates, up to 29% lipids, 4–6% tannins, 4.5–6.0% nitrogenous compounds, and 2–4% mineral substances.

The oil content of berry seeds in various grape varieties was determined using gas-liquid chromatography (Table 3).

Table 3

Fatty acid composition of oils obtained from grape seed samples

Fatty acids	Mass fraction of fatty acids by grape varieties, %			
	Rkatsiteli	Bayan Shirey	Madrasa	Isabella
Myristic acid	0.09	0.08	0.13	0.07
Palmitic acid	14.25	11.21	15.03	10.1
Palmitoleic acid	0.14	0.15	0.16	0.19
Stearic acid	5.41	4.46	10.36	3.25
Oleic acid	34.15	36.03	28.10	21.51
Linoleic acid	42.05	39.15	45.12	62.30
Linolenic acid	1.72	1.81	0.55	0.61
Arachidic acid	0.32	0.24	0.46	0.40
Eicosenoic acid	0.45	0.32	0.25	0.18
Behenic acid	0.36	0.41	0.35	0.07
Lignoceric acid	0.52	0.27	1.22	0.11
Erucic acid	–	0.01	0.20	0.02
Total	99.46	94.14	101.93	98.81

As seen from the table, the oil obtained from grape seeds contains the full range of both saturated and unsaturated fatty acids. The main representatives of saturated fatty acids in the studied samples are myristic, palmitic, and stearic acids. Among them, the highest content is observed for palmitic acid, while the lowest is for myristic acid.

Other fatty acids shown in the table are unsaturated. This group dominates the total fatty acid content. The composition of unsaturated fatty acids includes monoenoic acids such as palmitoleic and oleic acids, as well as dienic linoleic and trienoic linolenic acids.

Grape oil belongs to the linoleic acid group. Its fatty acid composition is as follows: saturated (3.8–7.2%), monounsaturated (27.0–30.0%), and polyunsaturated (58.0%). Oleic acid accounts for 30.0% and linoleic acid for 57.0%.

The total tocopherol content is 94 mg%, with 76% (of total tocopherols) being α -tocopherol, and 24% being β + γ -tocopherols. The total carotenoid content is 13.88%, providing good oxidation stability during storage and high biological activity of the oil.

Phytosterols and squalene (0.35%), which have pharmacological properties, were detected in the oil. Additionally, grape oil contains sterols, including campesterol (9.5%), stigmasterol (9.2%), β -stigmasterol (7.2%), and β -sitosterol (9.2%), bioflavonoids, phospholipids, vitamins A, D, B₁, B₂, and PP; as well as free amino acids (leucine, isoleucine, methionine, valine, glutamic acid). The oil is distinguished by a high content of biologically active substances. It contains 53 micro- and

macroelements (Mg, Fe, Co, Zn, Se), with iron at 13–15 mg%, magnesium at 3–4 mg%, zinc at 8–10 mg%, and selenium at 5–6 mg%. Due to its high biological value, grape oil is included in many medicinal preparations.

Based on the above, it can be concluded that grape oil is a suitable raw material for the preparation of functional foods, and it is time to take significant steps in this direction.

3.2. Evaluation of polyphenol content and antioxidant activity of extracts depending on grape variety and processing method

The content of polyphenols in extracts obtained from the pomace and seeds of white grape varieties (Rkatsiteli and Bayan Shirey) was studied (Table 4).

Table 4

Composition of monomeric and oligomeric polyphenols in pomace and seed extracts of white grape varieties (Rkatsiteli + Bayan Shirey)

Polyphenols	Mass fraction in the extract, mg/dm ³	
	Pulp	Seed
Flavonols		
Quercetin	9	0
Flavan-3-ols		
(+) – D – Catechin	203	205
(–) – Epicatechin	156	147
(–) – Epicatechin gallate (ECG)	17	16
Oxidized cinnamic acids		
Caffeic acid	57	0
p-Coumaric acid	41	0
Oxybenzoic acids		
Gallic acid	225	237
Syringic acid	66	70
Oligomeric procyanidins		
Procyanidin B ₁	221	236
Procyanidin B ₂	52	121
Procyanidin B ₃	15	27
Procyanidin B ₅	26	40
Procyanidin B ₇	9	17
Total monomeric and oligomeric polyphenols	1097	1116

It was observed that the extraction of polyphenols from pomace and seeds is largely completed within 60 days. Naturally, this process is influenced by the type of extractant, the solid fraction of the berries, and the hydromodule. In this research, following the methodology, the raw material and extractant were combined at 1:3 ratio.

Similar to the white grape blend, the pomace and seed extracts obtained from the red grape blend (Madrese + Cabernet Sauvignon) were also investigated. The results concerning the total polyphenol content in these extracts are presented in Table 5

The total polyphenol content in the pomace extract was 2162 mg/dm³, while in the seed extract it was 100 mg/dm³ lower, i. e., 2060 mg/dm³. However, the individual polyphenol composition differed between the pomace and seed extracts. As observed, anthocyanins were present only in the pomace extract, with a content of 201 mg/dm³, and were not detected in the seed extract.

When comparing the white grape blend with the red grape blend, it can be seen that in the seed extract of the white blend (unlike the red blend), the polyphenol content was slightly higher than in the pomace extract. At the same time, compared to the white blend, the total polyphenol content in both pomace and seed extracts of the red blend was almost twice as high.

Table 5

Polyphenol content of water-alcohol extracts from pomace and seeds of the red grape blend

Polyphenols	Mass fraction of polyphenols mg/dm ³	
	Pulp extract	Seed extract
Anthocyanins		
Delphinidin-3-0-glucoside	21	0
Cyanidin-3-0-glucoside	7	0
Peonidin-3-0-glucoside	10	0
Petunidin-3-0-glucoside	3	0
Malvidin-3-0-glucoside	136	0
Delphinidin-3-0-(6 ¹ -acetyl-glucoside)	11	0
Cyanidin-3-0-(6 ¹ -acetyl-glucoside)	2	0
Peonidin-3-0-(6 ¹ -acetyl-glucoside)	3	0
Petunidin-3-0-(6 ¹ -acetyl-glucoside)	2	0
Malvidin-3-0-(6 ¹ -acetyl-glucoside)	1	0
Delphinidin-3-0-(6 ¹ -n-coumaroyl-glucoside)	2	0
Petunidin (6 ¹ -n-coumaroyl-glucoside)	1	0
Malvidin (6 ¹ -n-coumaroyl-glucoside)	2	0
Total:	201	0
Flavan-3-ols		
Quercetin	50	0
Flavan-3-ols		
(+) - D - Catechin	596	441
(-) - Epicatechin	322	289
(-) - Epicatechin gallate (ECG)	11	40
Oxidized cinnamic acids		
Gallic acid	53	0
Tartaric acid	16	0
p-Coumaric acid	12	119
Oxybenzoic acids		
Gallic acid	331	293
Syringic acid	244	107
Oligomeric procyanidins		
Procyanidin B ₁	226	327
Procyanidin B ₂	154	223
Procyanidin B ₃	85	112
Procyanidin B ₅	34	48
Procyanidin B ₇	27	61
Total monomeric and oligomeric polyphenols	2162	2060

During the research, it was found that the antioxidant activity of the extracts was several times higher than that of the wine material. Moreover, the antioxidant activity of the extracts obtained from pomace and seeds differed from each other. For various grape varieties, these values varied as shown in Table 6.

Table 6 shows that the seed extract exhibited higher antioxidant activity compared to the pomace extract. When compared to individual white grape varieties, the antioxidant activity of seed extracts from various red grape varieties was slightly higher. For example, if the polyphenol content in the pomace extract of the Bayan Shirey variety was 5.0 g/dm³ with an antioxidant activity of 1.06% (ing.), then in the seed extract from the same variety, these values were 32 g/dm³ and 12.8% (ing.), respectively. A similar trend was observed in red grape varieties as well.

Table 6

Antioxidant activity of pomace and seed extracts from different grape varieties

Grape variety	Mass fraction of phenolic compounds in the pomace extract, g/dm ³	Antioxidant activity, % ing.
Bayan Shirey	5.0	1.06
Rkatsiteli	4.5	0.95
Aligote	4.0	0.85
Madras	15.5	2.30
Merlot	18.0	5.08
Cabernet Sauvignon	16.0	2.42
Grape variety	Phenolic compounds in seed extracts, g/dm ³	Antioxidant activity, % ing.
Bayan Shirey	32	12.8
Rkatsiteli	36	8.89
Aligote	31	12.15
Madras	22.5	14.0
Merlot	32.5	31.60
Cabernet Sauvignon	23.5	14.98

Grape pomace contains high amounts of leucoanthocyanins, catechins, anthocyanins, and other polyphenolic compounds in both monomeric and polymeric forms. These compounds exhibit antioxidant activity and P-vitamin activity. Polyphenols belong to antioxidants and demonstrate antimicrobial effects against a wide spectrum of microorganisms. Anthocyanins are distributed in different parts of the berry depending on the grape variety and cultivation conditions. They show particularly high reactivity depending on their structure and the pH of the environment.

To determine the effect of the extraction method on the composition, antioxidant, and antiradical activity of the extracts, two methods were employed in the research: extraction with alcohol and with supercritical carbon dioxide (Table 7).

Table 7

Antioxidant and antiradical properties of seed and skin extracts depending on the extraction method

Experimental variants	Flavonoids, mg/100 g	β -carotene, mg/100 g	Antiradical activity, mg/ml	Antioxidant activity, % ing.
CO ₂ extract of grape seeds	82.5	4.8	96.1	65.6
CO ₂ extract of cranberry peel	86.4	1.3	128.1	73.1
Alcohol extract of cranberry peel	94.3	1.1	88.1	51.8

As seen from Table 7, some compositional parameters as well as the antiradical and antioxidant properties of grape pomace skin extracts obtained with carbon dioxide and alcohol, as well as seed extracts obtained with carbon dioxide, are presented. It was found that the highest β -carotene content (4.8 mg/100 g) was observed in the carbon dioxide extract of grape seeds, while the lowest content (1.1 mg/100 g) was in the alcohol extract of the pomace skin. The carbon dioxide extract of the pomace skin contained slightly more β -carotene (1.3 mg/100 g) compared to the alcohol extract.

The highest flavonoid content was found in the alcohol extract of the pomace skin (94.3 mg/100 g), followed by the carbon dioxide extract of the pomace skin (86.4 mg/100 g), and the relatively lower content in the carbon dioxide extract of grape seeds (82.5 mg/100 g). Nevertheless, the carbon dioxide extract of the pomace skin exhibited the

highest antiradical (128.1 mg/ml) and antioxidant activity (73.1% ing.). The carbon dioxide extract of grape seeds was slightly lower than the pomace skin carbon dioxide extract, with an antiradical activity of 96.1 mg/ml and antioxidant activity of 65.6% ing. The lowest values were characteristic of the alcohol extract of the pomace skin, with an antiradical activity of 88.1 mg/ml and antioxidant activity of 51.8% ing.

Thus, compared to alcohol extracts, carbon dioxide extracts demonstrated higher antiradical and antioxidant activity. A similar trend can also be observed in the polyphenol content across the samples (Fig. 5).

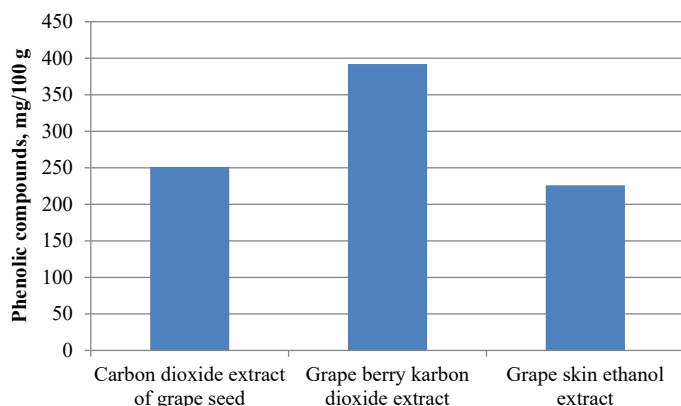


Fig. 5. Total polyphenol content in extracts obtained from pomace and seeds using different methods

As seen in Fig. 5, the total polyphenol content in the carbon dioxide extract of grape seeds was 251.2 mg/100 g, whereas the carbon dioxide extract obtained from the pomace significantly exceeded this value, with a total polyphenol content of 392 mg/100 g. The alcohol extract of the pomace was comparatively lower in total polyphenol content, with a value of 226.1 mg/100 g.

From these observations, it is evident that CO₂ extracts are considerably richer in biologically active compounds compared to alcohol extracts. Due to the content of valuable unsubstituted compounds, carbon dioxide extracts can be considered as natural antioxidants.

The research of fermented grape marc showed that it contains various chemical groups, including carbohydrates, pectins, vitamins, organic acids, nitrogenous compounds, and minerals. During grape processing, the resulting marc is rich in biologically active compounds, some of which function as natural antioxidants. This is further confirmed by the determination of antioxidant activity in marc extracts obtained from various grape varieties (Fig. 6).

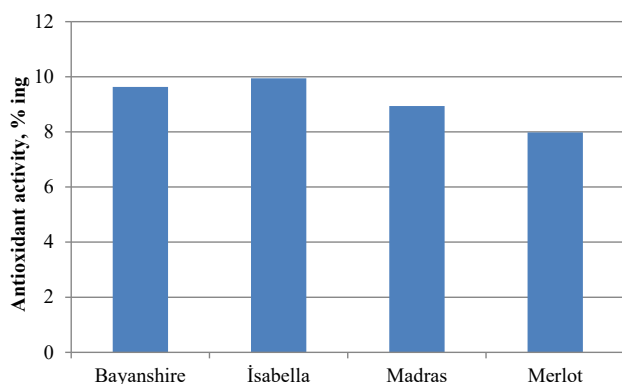


Fig. 6. Antioxidant activity of marc from different grape varieties

As shown in Fig. 6, the marc extract of the Isabella variety exhibited the highest antioxidant activity at 9.945% ing., followed by Bayan Shirey at 9.630% ing., Madrasa at 8.940% ing., and finally Merlot at 7.980% ing.

The moisture content of the marc samples from the studied varieties ranged from 10.760% to 12.730%, with the highest moisture in Bayan Shirey (12.730%), followed by Madrasa (11.280%), Isabella (10.760%), and Merlot (10.410%). The rutin content per 100 g of dry matter was lowest in Merlot (7.980 g) and highest in Isabella (9.945 g). The value of grapes for functional beverage production is determined by the amount of physiologically active compounds, including vitamins, microelements, and other constituents, which in turn influence their organoleptic and pharmacological properties.

3.3. Limitations and future research directions

The results obtained in the course of this research have certain limitations. In particular, the findings are constrained by the grape varieties used, their degree of ripeness, and cultivation conditions. Grapes grown under different climatic and soil conditions may have varying chemical compositions, which directly affect the content of phenolic compounds and antioxidant activity. In addition, the methods applied during the extraction process (type of solvent, temperature, duration, etc.) may lead to variability in the results. Therefore, the direct application of the obtained results to other conditions should be approached with caution.

Another limitation of research is that it was conducted under laboratory conditions. Since the technologies and scales used in real industrial settings (especially in food or pharmaceutical production) differ, additional optimization may be required when applying laboratory results in practice.

The reproducibility of the results largely depends on the standardization of the methodology used. Precise control of extraction parameters (temperature, time, solvent concentration), as well as the correct application of analytical methods (such as DPPH and ABTS assays for measuring antioxidant activity), are key factors ensuring reproducibility. In addition, the use of raw materials with consistent properties is essential for obtaining repeatable results.

The future development of this research offers broad opportunities. Further studies may focus on a more comprehensive comparative analysis of different grape varieties, as well as the application of new and more efficient extraction techniques (such as ultrasound-assisted or supercritical fluid extraction). Moreover, investigating the application of the obtained extracts in food products, cosmetic formulations, and dietary supplements represents a promising direction. At the same time, studying the effects of antioxidant activity on human health under in vivo conditions would contribute to a deeper understanding in this field.

Thus, although the present research provides important findings, further research is necessary to expand its scope and enhance its practical applicability.

4. Conclusions

1. Depending on the variety, the composition of berry components and their antioxidant properties showed different values. The content of phenolic compounds and tannins was higher in the peel of Medrese and Isabella varieties; in the seeds of Merlot, Rkatsiteli and Bayan Shirey varieties; while antioxidant activity was higher in the pulp of Merlot and in the seeds of the other varieties. The total amount of phenolic compounds in the seeds was 4560 mg/kg⁻¹ for the Medrese variety, 4230 mg/kg⁻¹ for Merlot, and 2950 mg/kg⁻¹ for Isabella; accordingly, the flavonoid content was 490, 410, and 256 mg/kg⁻¹, respectively. Antiradical activity was lowest in Bayan Shirey, relatively higher in the seeds of Medrese; while in the peel it was highest in Merlot and lowest in Bayan Shirey.

2. In extracts obtained from pomace of mixed white grape varieties, the total amount of monomeric and oligomeric polyphenols was 1097 mg/dm³, and from seeds 1116 mg/dm³; for mixed red grape

varieties, these values were 2162 and 2060 mg/dm³, respectively. In pomace extracts, the highest content of phenolic compounds was found in Merlot (18 g/dm³), and the lowest in Aligote (4 g/dm³), with antioxidant activity showing corresponding values (5.08% and 0.85% inhibition). In seed extracts, the highest phenolic content was observed in Rkatsiteli (36 g/dm³), the lowest in Medrese (22.5 g/dm³), while antioxidant activity was highest in Merlot (31.60% inhibition) and lowest in Rkatsiteli (8.89% inhibition). A comparative analysis of peel and seed extracts obtained using carbon dioxide and ethanol extraction showed that the peel extract had the highest antioxidant activity (73.1% inhibition). Although the highest flavonoid content was found in the ethanol extract of the peel (94.3 mg/100 g), this sample exhibited the lowest antioxidant activity (51.8% inhibition).

Conflict of interest

The authors declares that they have no conflict of interest in relation to this research, including financial, personal, authorship or other, which could affect the research and its results presented in this article.

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Data availability

The manuscript has no linked data.

Use of artificial intelligence

The authors confirm that they did not use artificial intelligence technologies in creating the submitted work.

Authors' contributions

Hasil Fataliyev: Writing – original draft, Writing – review and editing, Project administration; **Gunay Hajiyeva:** Investigation, Validation, Writing – review and editing; **Natavan Gadimova:** Conceptualization, Investigation, Writing – original draft; **Konul Baloghlanova:** Conceptualization, Methodology, Investigation; **Shabnam Fataliyeva:** Conceptualization, Methodology, Investigation.

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