

Volatile Compounds in Merlot Wine Pomace Extract Obtained by Supercritical Fluid Extraction

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Abstract

The valorisation of grape pomace, a major by-product of winemaking, presents both an environmental challenge and a resource opportunity. This study investigates the volatile compounds present in Merlot grape pomace extracts obtained by supercritical fluid extraction (SFE) using carbon dioxide as solvent. Extracts from whole grape pomace, seeds and skins were analysed. Volatile compounds were collected by headspace solid-phase microextraction (HS-SPME) and analysed using gas chromatography-mass spectrometry (GC-MS). A total of 29 different volatile compounds were identified across the three extracts, of which 11 were common to all fractions. The volatile profiles were dominated by alcohols, followed by esters, while carboxylic acids, ketones, and terpenoids were also detected. Butane-2,3-diol was the major constituent in terms of chromatogram peak area, followed by 2-phenylethanol and various esters. Many of the identified volatile compounds are recognised for their sensory and bioactive properties, possessing proven antioxidant, antimicrobial, and anti-inflammatory effects, making them of interest to the food, cosmetics, and pharmaceutical industries. The results of this study confirm the effectiveness of supercritical fluid extraction in recovering a broad range of volatile compounds from Merlot grape pomace, underscoring its value as a sustainable feedstock rather than agricultural waste. However, due to the limited efficiency of supercritical fluid extraction in mobilising polar compounds when carbon dioxide is used as the sole solvent, additional extraction strategies are required for more comprehensive pomace valorisation. This research supports the potential integration of grape pomace extracts into bio-based industry applications, contributing to the transformation of waste into value-added products.

Keywords

Grape wine pomace, volatile compounds, HS-SPME, GC-MS, supercritical fluid extraction

1 Introduction

The viticulture and winemaking sector represents one of the leading economic and agricultural sectors in the world and, in 2023 accounted for 7.2 Mha of cultivated land and 237 Mhl of wine produced globally, with France, Italy, and Spain ranking as the top three producing countries.¹ In the same year, according to the Croatian Ministry of Agriculture, Forestry and Fisheries, 19 826 ha were used for grape cultivation, yielding 94 905 t of grapes and 612 000 hl of wine. Of the total wine production, 9 585 t of grapes were of the Merlot variety.^{2,3} Approximately 80 % of grapes are used for wine production, and of this amount, as much as 20–25 % is converted into grape pomace, the most abundant by-product of the process. Grape pomace consists primarily of skins, seeds, pulp, and stalks, resulting in an estimated 9 Mt of pomace generated annually worldwide, or about 40 600 t in Croatia.⁴ Traditionally, grape pomace has been disposed of directly into the environment, leading to numerous environmental issues, including soil and groundwater contamination, unpleasant odour generation, attraction of disease vectors such as insects and rodents, and microbial

proliferation. Due to its high chemical oxygen demand and biodegradable organic content, improper disposal poses potential health risks to humans and aquatic organisms.⁵ However, studies have demonstrated that grape pomace remains a valuable source of bioactive compounds.⁶ Grape skin and seeds dominate the pomace, together accounting for up to 70 % of its total mass.⁵ Grape skins are rich in dietary fibres, predominantly insoluble,⁷ while seeds can account for as much as 50 % of the pomace dry matter and contain fibre, proteins, and lipids, as well as phenolic compounds, vitamin E, carotenoids, and phytosterols.^{8,9} In addition, grape pomace retains valuable volatile compounds shaped by winemaking techniques, making it a valuable source of functional ingredients in various applications.¹⁰ The volatile composition of grapes, and consequently of grape pomace, is influenced by many factors, including viticulture practices, regional and seasonal variations impacting the grapes' genetic expression and leading to distinct volatile profiles.¹¹ The volatile profile of the pomace is affected also by techniques applied during pre-fermentation and fermentation, as well as grape pressing, particularly in red wine production such as Merlot, significantly influencing modulation of grape aromas.¹¹ The complex chemical composition of grape pomace, including compounds like alcohols, organic acids, esters, aldehydes, polyphenols, pectins, minerals and sugars, underscores the potential of this agricultural waste for the recovery of functional ingredients and bioactive substances that can be applied in the food, cosmetics, and pharmaceutical industries.^{12,13} Bio-sourced

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flavours represent a rapidly expanding market segment, driven by increasing consumer demand for sustainable, natural and clean-label alternatives to synthetic ingredients.¹⁰ In response, innovative green technologies, such as supercritical fluid extraction (SFE), have been applied to recover high-value bioactive compounds, technologies that can be optimised for maximum yield. Avenues for efficient recovery of bioactive compounds from agricultural waste, in this case Merlot grape pomace, align with efforts to promote sustainability within the winemaking sector through waste valorisation based on circular economy principles. In this study, the volatile compound profiles of extracts obtained by supercritical fluid extraction using carbon dioxide (CO₂) as solvent applied to pomace generated during Merlot (*Vitis vinifera* L.) wine production were determined using headspace solid-phase microextraction (HS-SPME) combined with gas chromatography-mass spectrometry (GC-MS), to identify potentially valuable volatile compounds. The use of supercritical fluid extraction with carbon dioxide as the sole solvent ensured the production of highly pure extracts free from solvent residues. Moreover, the relatively low operating temperatures and pressures minimised thermal degradation, thereby preserving compound integrity and enhancing their commercialisation potential.^{14,15}

2 Experimental

2.1 Samples

Grape pomace derived from Merlot (*Vitis vinifera* L.) wine production was kindly provided by the Sarač family agricultural holding (Čaporice, Split-Dalmatia County, Croatia), following the 2023 harvest. The pomace was collected after alcoholic fermentation and subsequent pressing, as per conventional red wine production practices. The material, consisting of grape skins and seeds, was dried in a chamber dryer (KSVP 20R, Euclid Ltd., Vinkovci, Croatia). A portion of the pomace was separated into skins and seeds using a fan, resulting in 3 fractions – whole pomace, skins, and seeds. Each fraction was ground using a mill (Retsch SM 100, Retsch GmbH, Haan, Germany, equipped with a 4 mm sieve). The resulting fine material was immediately subjected to extraction to minimise compound loss.

2.2 Supercritical fluid extraction

Samples of the ground pomace fractions were processed in a pilot-scale supercritical fluid extraction system (Extratx SFI, Point-Saint-Vincent, France), using CO₂ as solvent (99.98 % purity, supplied by Messer Croatia plin Ltd., Zaprešić, Croatia). Extraction time was 120 min, at 30 MPa (300 bar) pressure, and temperature set to 45 °C with CO₂ flow rate of 150 kg h⁻¹. The extracts were collected in a separator heated at 50 °C with pressure at 5 MPa (50 bar), and refrigerated at 6 °C until analysis.

2.3 Volatile compound screening

Volatile organic compounds were analysed using headspace solid-phase microextraction (HS-SPME) coupled

with gas chromatography-mass spectrometry (GC-MS). The DVB/CAR/PDMS fibre (Agilent Technologies Inc., Santa Clara, California, USA) was thermally conditioned prior to analysis following the manufacturer's recommendations. Each sample of the three grape pomace extract fractions (whole pomace, skins, and seeds) was pipetted into 20 ml headspace vials and septum was sealed using aluminium caps. The vials were placed in a thermostatic block preheated to 60 °C and equilibrated for 15 min. HS-SPME extraction and desorption were carried out in automated mode using a PAL Auto Sampler System (model PAL RSI 85, CTC Analytics AG, Zwingen, Switzerland), which controlled vial incubation, fibre exposure to the headspace, and subsequent desorption of analytes in the GC injector. The analytical set-up was completed with an Agilent gas chromatograph (model 8890, Agilent Technologies Inc., Santa Clara, California, USA) coupled with an Agilent mass spectrometer (model 5977E MSD, Agilent Technologies Inc., Santa Clara, California, USA). Separation of the volatile organic compounds was achieved using an HP-5MS capillary column (30 m × 0.25 mm, 0.25 μm film thickness, Agilent Technologies Inc., Santa Clara, California, USA). Helium was used as carrier gas (Helium 5.0, supplied by Messer Croatia plin Ltd., Zaprešić, Croatia) with a flow rate of 1 ml min⁻¹. Inlet temperature was 250 °C, the GC oven temperature started at 60 °C (isothermal for 2 min), followed by an increase to 246 °C at 3 °C min⁻¹ (isothermal for 25 min), resulting in an 89 min run time. The injection mode was split (ratio 5 : 1) and transfer line temperature was 280 °C. The MS system operated on electron impact (EI) at 70 eV, source temperature at 230 °C, and quadrupole detector at 150 °C. The mass range acquired ranged from 40 to 450 m/z. Chromatographic peaks for all samples were identified by searching against the Wiley 9 (Wiley Inc., Hoboken, New York, USA) and NIST 20 (National Institute of Standards and Technology, Gaithersburg, Maryland, USA) mass spectral libraries using Dana Analysis software (Agilent Technologies Inc., Santa Clara, California, USA). Compound identifications were verified by comparing their retention indices (RI), calculated from retention times of *n*-alkanes (alkane standard solution C8-C20, Supelco, Merck KGaA, Darmstadt, Germany; C22 standard, Sigma-Aldrich, Aldrich brand, Merck KGaA, Darmstadt, Germany; C23 standard and C24 standards, Fluka Analytical, Honeywell, Seelze, Germany), with reference values reported in literature (NIST), as well as by matching their mass spectra to those in spectral libraries (Wiley 9 and NIST 20).

3 Results and discussion

To explore potential valorisation options for grape pomace extracts obtained by supercritical fluid extraction, considering they could contain bioactive compounds with valuable properties, the volatile fingerprints of the three extracts were established to identify potential applications in the food, cosmetics, and pharmaceutical industries. The volatile compounds identified in each grape pomace extract obtained by supercritical fluid extraction are presented in Tables 1–3.

Table 1 – Volatile compounds identified in Merlot grape pomace extract obtained by supercritical fluid extraction (RI – retention index, RI (database) – retention index according to database)

Tablica 1 – Hlapljivi spojevi identificirani u ekstraktu komine grožđa Merlot dobivenom superkričnom ekstrakcijom (RI – retencijski indeks, RI (database) – retencijski indeks sukladno bazi podataka)

No. Br.	Compound name (common and IUPAC) Naziv spoja (uobičajen i po IUPAC-u)	RI	RI (database)	Area/% Površina/%
1	Ethanoic acid Octena kiselina	<800	645	7.20
2	Acetoin (3-Hydroxybutan-2-one) Acetoin (3-hidorksibutan-2-on)	<800	743	9.26
3	2,3-Butanediol (Butane-2,3-diol) 2,3-Butandiol (butan-2,3-diol)	817	819	44.78
4	Butyrolactone (Oxolan-2-one) Butirolakton (oksloan-2-on)	926	920	1.26
5	α -Thujene ((1R,5R)-2-methyl-5-propan-2-ylbicyclo[3.1.0]hex-2-ene) α -Thujen ((1R,5R)-2-methyl-5-propan-2-ilbiciklo[3.1.0]heks-2-en)	939	930	0.49
6	α -Pinene ((1R,5R)-2,6,6-trimethylbicyclo[3.1.1]hept-2-ene) α -Pinen ((1R,5R)-2,6,6-trimetilbiciklo[3.1.1]hept-2-en)	945	939	0.40
7	Hexyl acetate (Hexyl ethanoate) Heksil-acetat (heksil-etanoat)	1004	1014	2.13
8	Benzyl alcohol (Phenylmethanol) Benzilni alkohol (fenilmetanol)	1045	1061	0.36
9	Linalool (3,7-Dimethylocta-1,6-dien-3-ol) Linalool (3,7-dimethilokta-1,6-dien-3-ol)	1105	1104	0.65
10	2-Phenylethanol 2-Feniletanol	1123	1117	12.08
11	Terpinen-4-ol (4-(propan-2-yl)-1-methylcyclohex-3-en-1-ol) Terpinen-4-ol (4-(propan-2-il)-1-methylcicloheks-3-en-1-ol)	1185	1178	0.62
12	Octanoic acid Oktanska kisleina	1190	1180	1.73
13	Hexyl butyrate (Hexyl butanoate) Heksil-butirat (heksil-butanoat)	1196	1190	1.57
14	Ethyl octanoate (Octanoic acid ethyl ester) Etil-oktanoat (etilni ester oktanske kiseline)	1199	1196	3.57
15	Linalyl acetate (3,7-Dimethylocta-1,6-dien-3-yl acetate) Linalil-acetat (3,7-dimethilokta-1,6-dien-3-il acetat)	1260	1257	0.30
16	2-Phenylethyl acetate (2-Phenylethyl ethanoate) 2-Feniletil-acetat (2-feniletil-etanoat)	1262	1260	0.43
17	Decanoic acid Dekanska kiselina	1382	1377	1.27
18	Ethyl decanoate Etil-dekanoat	1397	1397	7.47
19	Ethyl dodecanoate Etil-dodekanoat	1595	1596	2.10
20	Hexadecanoic acid Heksadekanska kiselina	1994	1997	0.55
Total peak area identified Ukupna identificirana površina ispod vrhova				97.68

Table 2 – Volatile compounds identified in Merlot grape seed (from pomace) extract obtained by supercritical fluid extraction (RI – retention index, RI (database) – retention index according to database)

Tablica 2 – Hlapljivi spojevi identificirani u ekstraktu sjemenki grožđa (iz komine) Merlot dobivenom superkričnom ekstrakcijom (RI – retencijski indeks, RI (database) – retencijski indeks sukladno bazi podataka)

No. Br.	Compound name (common and IUPAC) Naziv spoja (uobičajen i po IUPAC-u)	RI	RI (database)	Area % Površina %
1	Ethanoic acid Octena kiselina	<800	645	4.03
2	Acetoin (3-Hydroxybutan-2-one) Acetoin (3-hidorksibutan-2-on)	<800	743	4.09
3	2,3-Butanediol (Butane-2,3-diol) 2,3-Butandiol (butan-2,3-diol)	813	819	43.98
4	Butyrolactone (Oxolan-2-one) Butirolakton (oksloan-2-on)	926	920	1.81
5	Hexanoic acid Heksanska kiselina	992	990	0.47
6	Ethyl hexanoate (Hexanoic acid ethyl ester) Etil-heksanoat (etil ester heksanoične kiseline)	1003	1002	0.96
7	1,8-Cineole (2,2,4-Trimethyl-3-oxabicyclo[2.2.2]octane) 1,8-Cineol (2,2,4-trimetil-3-oksabiciklo[2.2.2]oktan)	1036	1035	0.95
8	(E)- β -Ocimene ((E)-3,7-Dimethylocta-1,3,6-triene) (E)- β -Ocimen ((E)-3,7-dimetilokta-1,3,6-trien)	1045	1040	0.41
9	(Z)- β -Ocimene ((Z)-3,7-Dimethylocta-1,3,6-triene) (Z)- β -Ocimen ((Z)-3,7-dimetilokta-1,3,6-trien)	1062	1051	0.43
10	Hexanal diethyl acetal Heksanal-dietilacetal	1096	1092	0.24
11	Linalool (3,7-Dimethylocta-1,6-dien-3-ol) Linalool (3,7-dimetilokta-1,6-dien-3-ol)	1105	1104	2.81
12	2-Phenylethanol 2-Feniletanol	1123	1117	14.12
13	Camphor (1,7,7-trimethylbicyclo[2.2.1]heptan-2-one) Kamfor (1,7,7-trimetilbiciklo[2.2.1]heptan-2-on)	1151	1154	0.25
14	Borneol (1,7,7-trimethylbicyclo[2.2.1]heptan-2-ol) Borneol (1,7,7-trimetilbiciklo[2.2.1]heptan-2-ol)	1173	1173	0.39
15	Terpinen-4-ol (4-(propan-2-yl)-1-methylcyclohex-3-en-1-ol) Terpinen-4-ol (4-(propan-2-il)-1-metilcicloheks-3-en-1-ol)	1183	1178	0.36
16	Octanoic acid Oktanska kiselina	1189	1180	1.20
17	Hexyl butyrate (Hexyl butanoate) Heksil-butirat (heksil-butanoat)	1193	1190	1.11
18	Ethyl octanoate (Octanoic acid ethyl ester) Etil-oktanoat (etilni ester oktanske kiseline)	1199	1196	4.50
19	Linalyl acetate (3,7-Dimethylocta-1,6-dien-3-yl acetate) Linalil-acetat (3,7-dimetilokta-1,6-dien-3-il acetat)	1260	1257	1.02
20	Decanoic acid Dekanska kiselina	1382	1377	0.99
21	Ethyl decanoate Etil-dekanoat	1397	1397	10.45
22	Ethyl dodecanoate Etil-dodekanoat	1595	1596	2.53
23	Hexadecanoic acid Heksadekanska kiselina	1994	1997	0.55
Total peak area identified Ukupna identificirana površina ispod vrhova				96.47

Table 3 – Volatile compounds identified in Merlot grape skins (from pomace) extract obtained by supercritical fluid extraction (RI – retention index, RI (database) – retention index according to database)

Tablica 3 – Hlapljivi spojevi identificirani u ekstraktu kožiće grožđa (iz komine) Merlot dobivenom superkričnom ekstrakcijom (RI – retencijski indeks, RI (database) – retencijski indeks sukladno bazi podataka)

No. Br.	Compound name (common and IUPAC) Naziv spoja (uobičajen i po IUPAC-u)	RI	RI (database)	Area % Površina %
1	Ethanoic acid Octena kiselina	<800	645	7.00
2	Acetoin (3-Hydroxybutan-2-one) Acetoin (3-hidoksibutan-2-on)	<800	743	8.94
3	2,3-Butanediol (Butane-2,3-diol) 2,3-Butandiol (butan-2,3-diol)	813	819	25.45
4	Butyrolactone (Oxolan-2-one) Butirolakton (oksolan-2-on)	926	920	0.75
5	Ethyl hexanoate (Hexanoic acid ethyl ester) Etil-heksanoat (etil ester heksanoične kiseline)	1003	1002	2.83
6	1,8-Cineole (2,2,4-Trimethyl-3-oxabicyclo[2.2.2]octane) 1,8-Cineol (2,2,4-trimetil-3-oksabiciklo[2.2.2]oktan)	1040	1035	1.15
7	(E)- β -Ocimene ((E)-3,7-Dimethylocta-1,3,6-triene) (E)- β -Ocimen ((E)-3,7-dimetilokta-1,3,6-trien)	1045	1040	0.68
8	(Z)- β -Ocimene ((Z)-3,7-Dimethylocta-1,3,6-triene) (Z)- β -Ocimen ((Z)-3,7-dimetilokta-1,3,6-trien)	1062	1051	1.41
9	Hexanal diethyl acetal Heksanal-dietilacetal	1096	1092	0.60
10	Linalool (3,7-dimethylocta-1,6-dien-3-ol) Linalool (3,7-dimetilokta-1,6-dien-3-ol)	1105	1104	2.30
11	2-Phenylethanol 2-Feniletanol	1122	1117	17.47
12	Ethyl octanoate (octanoic acid ethyl ester) Etil-oktanoat (etilni ester oktanske kiseline)	1199	1196	12.48
13	Linalyl acetate (3,7-dimethylocta-1,6-dien-3-yl acetate) Linalil-acetat (3,7-dimetilokta-1,6-dien-3-il acetat)	1260	1257	1.04
14	Ethyl decanoate Etil-dekanoat	1397	1397	13.09
15	Ethyl dodecanoate Etil-dodekanoat	1595	1596	2.10
16	Hexadecanoic acid Heksadekanska kiselina	1994	1997	0.59
Total peak area identified Ukupna identificirana površina ispod vrhova				97.85

A total of 29 volatile compounds were identified across the three pomace extract samples obtained by supercritical fluid extraction, of which 11 were common to all analysed samples: 4 to samples from whole pomace and pomace seeds, 5 to samples from pomace seeds and pomace skins, while 5 compounds were unique to the whole pomace sample, and 3 to the pomace seed sample (Fig. 1). These overlaps indicate a core set of volatile compounds that probably reflect shared metabolic origins and similar extraction efficiency across the three different matrices. The identified volatile compounds belong to several chemical families: alcohols (6), esters (8), carboxylic acids (5), alde-

hyde (1), ketones (3), terpenoids (monoterpenes) (7). Linalool as well as terpinene-4-ol can be classified as both alcohols and terpenoids (monoterpenes) and have been counted in both categories. In terms of total chromatogram peak area, alcohols are the most abundant, accounting for 58.49 % of the peak area identified in the grape pomace extract, 61.66 % identified in the grape seed extract, and 45.22 % identified in grape skin extract. This was followed by esters, which represented 17.57 % of peak area identified in the grape pomace extract across 7 compounds. Carboxylic acids accounted for 10.75 % of the total peak area, while ketones represented 10.52 %.

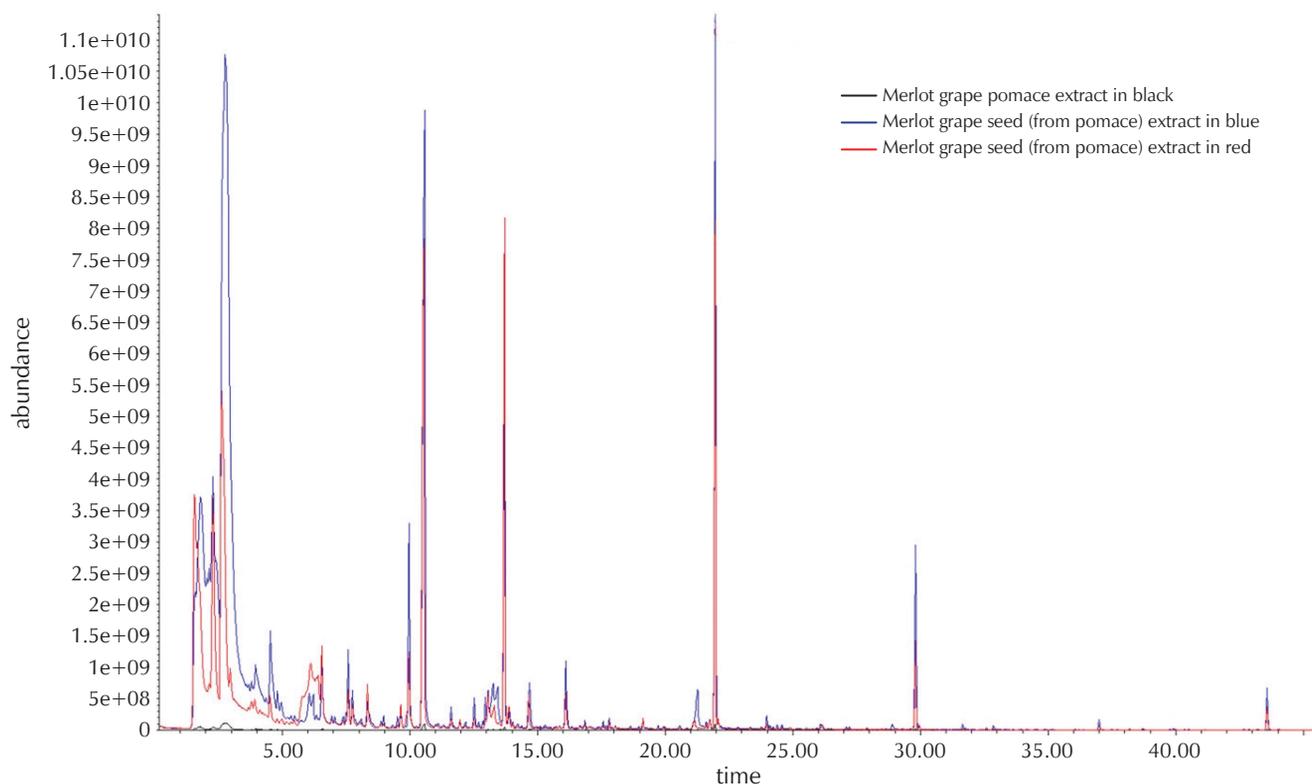


Fig. 1 – Overlaid GC-MS chromatograms of volatile compounds in Merlot pomace extracts obtained by supercritical fluid extraction (whole pomace in black, pomace seed in blue, pomace skins in red).

Slika 1 – Preklapljeni GC-MS kromatogrami hlapljivih spojeva u ekstraktima iz komine grožđa sorte Merlot dobivenima superkričnom ekstrakcijom (cijela komina u crnoj, sjemenke iz komine u plavoj i koži iz komine u crvenoj boji).

Terpenoids (monoterpenes) accounted for 2.16 % of the total peak area in the grape pomace extract. In the grape seed extract, following alcohols, esters represented 20.57 % of the total peak area across 6 compounds, carboxylic acids accounted for 7.24 %, ketones for 6.15 %, and terpenoids (monoterpenes) for 4.96 %. In the grape skin extract, alcohols were followed by esters at 31.54 % (across 5 compounds), ketones at 9.69 %, carboxylic acids at 7.59 %, and terpenoids (monoterpenes) at 5.53 %. The alcohols identified in all Merlot grape pomace extracts comprised both aliphatic (butane-2,3-diol, linalool, borneol and terpinene-4-ol) and aromatic (2-phenylethanol and benzyl alcohol) volatiles. The aliphatic butane-2,3-diol was the most abundant by peak area across all samples, ranging from 25.45 % in the grape skin extract to 44.78 % in the pomace extract. This compound has been highlighted for its potential application in the manufacturing of printing inks, perfumes, synthetic rubber, antifreeze agents, fuel additives, foods, and pharmaceuticals.¹⁶ The other dominant alcohol, 2-phenylethanol, ranged in peak area from 12.08 % in grape pomace extract to as much as 17.47 % in grape skin extract. *Abreu et al.* also found 2-phenylethanol to be among the major volatile organic compounds in grape pomace, associated with floral and honey-like odours.¹⁷ Other studies similarly identified 2-phenylethanol at relatively high concentrations in grape pomace, a compound known for imparting a characteristic rose-like aroma.¹⁸ The second most abundant compound

group by peak area were esters, encompassing the highest number of individual compounds. Literature suggests that esters contribute positively to overall aroma at lower concentrations,¹⁹ producing fruity notes reminiscent of bananas, pears, roses, honey or apples. Literature also confirms that alcohols and esters are therefore predominantly responsible for the total volatile profile of grape pomace, regardless of grape variety.¹⁷ The variations in the number, composition, and peak areas of the identified volatile compounds across the three Merlot pomace extracts can be attributed to the inherent anatomical and compositional differences between the matrices (whole pomace, seeds, and skins), combined with varying efficiency of the extraction method on each material. The seeds typically contain higher lipid content and different cell wall structures compared to skins, resulting in higher concentrations of terpenoids and aromatic compounds.⁷ Whole pomace represents a composite mixture of seeds and skins, resulting in volatile compounds from both components. The analysis revealed composition variations, with the seed extract containing unique compounds not detected in other fractions (camphor, borneol, hexanoic acid, hexanal diethyl acetal), while the skin extract, although containing fewer total compounds, exhibited higher concentrations of certain esters, especially ethyl octanoate. The whole pomace fraction contained unique terpenoids (α -thujene, α -pinene) and additional esters not detected in the separate fractions. Differences were also observed across peak

areas, with compound concentration variations in different matrices: 2-phenylethanol increased in peak area from whole pomace, to seeds, to skins, whereas, butane-2,3-diol decreased from whole pomace to seeds to skins, similar to ethyl octanoate. The variation in extraction efficiency, despite identical parameters applied, may be attributed to the varying structures of the matrices. The oil-rich seeds may facilitate the release of volatile compounds more efficiently than the fibre-rich skins. This may also be impacted by different cell wall compositions influencing compound accessibility during the extraction process.^{7,20} The distribution pattern of 2-phenylethanol reflects the localisation of 2-phenylethanol, as its biosynthesis occurs predominantly in grape skins, where aromatic alcohols and phenolic biosynthesis pathways are most active.²⁰ 2-Phenylethanol in the seed fraction is likely due to skin tissue contamination during the mechanical separation process, as complete separation of seeds from skin is difficult to achieve at a pilot-scale level, and the whole pomace fraction reflects the impact of both tissue concentrations in the initial material. The overall distribution and diversity of volatile compounds identified in the three samples highlight the effectiveness of supercritical CO₂ extraction as a method for recovering a broad spectrum of volatiles from grape pomace. Several of the identified compounds have been reported in foods and beverages with many biological properties, including antioxidant, antimicrobial, anti-inflammatory, anti-proliferative, antidiabetic, antitumor activities, showcasing their potential use in food, but also in the cosmetics and pharmaceutical industries. Key compounds include alcohols (2-phenylethanol and linalool), esters (ethyl hexanoate, ethyl octanoate, ethyl decanoate, hexyl acetate, linalyl acetate, 2-phenylethyl acetate and hexyl butanoate), and terpenoids (monoterpenes: linalool, terpinene-4-ol, 1,8-cineole).^{22–24}

4 Conclusion

This study confirms the efficiency of supercritical fluid extraction using CO₂ as solvent in recovering a broad spectrum of volatile compounds from Merlot grape pomace. The volatile profiles of the three explored fractions, extracts obtained from whole pomace, seeds, and skins, demonstrated consistent recovery of the major compound classes (especially alcohols, but also esters and ketones), as well as contributions from carboxylic acids and terpenoids (monoterpenes). Several of the identified compounds, including 2-phenylethanol, possess documented bioactive properties and are of relevance to key industries, supporting the ongoing valorisation of grape pomace as a resource for the food, cosmetics, and pharmaceutical industries. It should be mentioned that supercritical fluid extraction using CO₂ as the sole solvent inherently mobilises nonpolar compounds. Polar compounds require complementary extraction techniques to fully exploit grape pomace as a potential resource. This research confirms the potential of grape pomace and emphasises the need for further studies to expand its utilisation as a valuable resource, and to integrate it into products thereby transforming it from a by-product and waste into a sustainable feedstock for bio-based industries.

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List of abbreviations and symbols Popis kratica i simbola

SFE	– supercritical fluid extraction – ekstrakcija superkričnim fluidom
HS-SPME	– headspace – solid-phase microextraction, mikroekstrakcija vršnih para na krutoj fazi
GC-MS	– gas chromatography – mass spectrometry – plinska kromatografija – masena spektrometrija
DVB/CAR/PDMS	– Divinylbenzene/Carboxen/Polydimethylsiloxane – divinilbenzen/karboksen/polidimetilsiloksan
HP-5MS	– Hewlett-Packard – (5%-phenyl)-methylpolysiloxane phase – for Mass Spectrometry – Hewlett-Packard – (5%-fenil)-metilpolisiloksan faza – za masenu spektrometriju
EI	– electron impact – elektronski impakt
NIST	– National Institute of Standards and Technology – Nacionalni institut standarda i tehnologije
RI	– retention index – retencijski indeks

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SAŽETAK

Isparljivi spojevi u ekstraktu komine vina sorte Merlot dobiveni ekstrakcijom superkritičnim fluidom

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Valorizacija komine grožđa, glavnog nusproizvoda proizvodnje vina, istodobno predstavlja izazov za okoliš i priliku za iskorištavanje resursa. Ovim istraživanjem ispituju se isparljivi spojevi ekstrakata komine grožđa sorte Merlot dobiveni ekstrakcijom superkritičnim fluidom koristeći se ugljikovim dioksidom kao otapalom. Analizirani su ekstrakti dobiveni od cijele komine, sjemenki iz komine i kože iz komine grožđa. Isparljivi spojevi izdvojeni su mikroekstrakcijom vršnih para na krutoj fazi (HS-SPME), a potom analizirani plinskom kromatografijom spregnutom s masenom spektrometrijom (GC-MS). Ukupno je identificirano 29 isparljivih spojeva u trima ekstraktima, od kojih je 11 zajedničko svim uzorcima. U profilima isparljivih spojeva dominiraju alkoholi, zatim razni esteri te karboksilne kiseline, ketoni i terpenoidi. Najzastupljeniji spoj, kad je u pitanju površina ispod kromatografskog vrha, je butan-2,3-diol te nakon njega 2-feniletanol i različiti esteri. Mnogi od identificiranih spojeva poznati su po senzorskim i bioaktivnim svojstvima te posjeduju dokazane antioksidativne, antimikrobne i protuupalne učinke, od interesa u prehrambenoj, kozmetičkoj i farmaceutskoj industriji. Rezultati istraživanja potvrđuju učinkovitost superkritične ekstrakcije u izdvajanju širokog raspona isparljivih spojeva iz komine grožđa sorte Merlot, naglašavajući vrijednost tog materijala kao održive sirovine umjesto poljoprivrednog otpada. Uslijed slabijeg učinka superkritične ekstrakcije s isključivo ugljikovim dioksidom kao otapalom, kad je u pitanju mobilizacija polarnih spojeva, za potpuniju valorizaciju komine grožđa potrebno je primijeniti dodatne metode ekstrakcije. Ovo istraživanje podržava moguću integraciju ekstrakata komine grožđa u tzv. *bio-based* industrije, pretvarajući otpad u proizvode s dodanom vrijednosti.

Ključne riječi

Komina grožđa, isparljivi spojevi, HS-SPME, GC-MS, ekstrakcija superkritičnim fluidima

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