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Differential Effects of Cornus officinalis-Derived Polysaccharides and Ursolic Acid on the Chemical Composition, Aroma Profile, and Bioactivity of Functional Fruit Wines

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Abstract Cornus fruit (Cornus officinalis) is a medicinal and edible fruit rich in bioactive compounds, including polysaccharides (PS) and ursolic acid (UA). However, the effects of these individual functional components on the quality and bioactivity of fruit wines remain poorly understood. This study aimed to investigate the distinct and matrix-dependent effects of *C. officinalis*-derived PS and UA on the physicochemical properties, chemical composition, aroma characteristics, and biological activities of grape wine and *C. officinalis* fermented wine. PS and UA were extracted from dried *C. officinalis* fruits and quantified using UV–Vis spectrophotometry. Their antioxidant and α-glucosidase inhibitory activities were evaluated in vitro. Subsequently, PS- and UA-fortified wines were prepared, and their physicochemical properties, phenolic profiles, volatile compounds, and bioactivities were analyzed using chemical assays, HPLC, GC–MS, and statistical analysis. The extraction yields of UA and PS were 4.17% and 2.88%, respectively. UA exhibited stronger bioactivity at low concentrations, whereas PS showed concentration-dependent antioxidant effects, particularly for hydroxyl radical scavenging. In wine matrices, UA promoted the retention of floral aroma compounds, increasing linalool and geraniol contents by 18.7% and 15.2%, respectively. PS markedly altered volatile profiles by enhancing higher alcohols and acetate esters. Moreover, PS increased ABTS scavenging activity, hydroxyl radical scavenging activity, and α-glucosidase inhibition by 16.4%, 19.1%, and 14.7%, respectively. UA and PS regulate fruit wine quality through different pathways. UA is more suitable for low-dose functional enhancement while preserving aroma characteristics, whereas PS contributes to bioactivity improvement and aroma modification. These findings provide a basis for the targeted application of *C. officinalis* functional components in fruit wine development.

Keywords: *Cornus officinalis, functional fruit wine, polysaccharides, ursolic acid, aroma characteristics, phenolic compounds, antioxidant activity*

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1. Introduction

Cornus fruit (*Cornus officinalis Sieb. et Zucc.*), a traditional medicinal and edible fruit, has attracted increasing attention due to its abundant phytochemical components and potential health-promoting properties [1]. Previous studies have demonstrated that *C. officinalis* contains various bioactive compounds, including iridoid glycosides, organic acids, phenolic compounds, triterpenoids, and polysaccharides, which contribute to its antioxidant, anti-inflammatory, and metabolic regulatory activities. Among these compounds, polysaccharides (PS) and ursolic acid (UA) are considered important functional constituents because of their reported biological activities, particularly antioxidant capacity and enzyme inhibition [2].

Despite its nutritional and medicinal value, direct utilization of *C. officinalis* in food products remains limited due to its strong sourness, astringency, and relatively restricted processing applications [3]. Fermentation into fruit wine provides an effective strategy to improve sensory acceptability while retaining or enhancing the functional properties of the fruit [4]. Beyond conventional physicochemical parameters, the quality and functional potential of fruit wines are also

influenced by their volatile and phenolic composition [5].

The incorporation of isolated functional components into fermented beverages has recently attracted interest because individual compounds may exhibit different behaviors compared with their original plant matrix [6]. Polysaccharides can interact with phenolic compounds, proteins, and volatile molecules through hydrogen bonding, hydrophobic interactions, and colloidal effects, thereby influencing aroma release, mouthfeel, and antioxidant properties. In contrast, ursolic acid, a pentacyclic triterpenoid with low polarity, possesses strong biological activity but its influence on wine composition and aroma characteristics remains insufficiently understood.

Previous studies have mainly focused on the extraction optimization and biological activities of PS and UA from *C. officinalis [7]*. However, the functional roles of these isolated components after incorporation into complex food matrices, particularly fermented beverages, remain unclear. Two important scientific questions require further investigation: (1) whether PS and UA exert similar or distinct effects when separated from the whole fruit matrix, and (2) whether their effects vary among different wine systems with different chemical backgrounds.

Therefore, this study aimed to investigate the matrix-dependent effects of *C. officinalis*-derived polysaccharides and ursolic acid in two different fruit wine systems: grape wine and *C. officinalis* fermented wine. The specific objectives were to: (1) extract and quantify PS and UA from dried *C. officinalis* fruits; (2) evaluate their antioxidant and α-glucosidase inhibitory activities; (3) determine their effects on physicochemical properties, phenolic profiles, and volatile aroma compounds of fruit wines; and (4) analyze the relationship between chemical composition and functional activities. The findings provide new insights into the targeted application of bioactive components in functional fruit wine production.

2. Materials and Methods

2.1. Materials and Reagents

Dried Cornus fruit (Cornus officinalis Sieb. et Zucc.) was provided by Shaanxi Changsheng Ecological Wine Industry Co., Ltd. (Shaanxi, China). Commercial dry red grape wine was purchased from the Mediterranean region of France and produced from Cabernet Sauvignon and Cabernet Franc grapes (vintage 2019). The basic physicochemical characteristics of the grape wine were as follows: reducing sugar content, 4.72 g/L; alcohol content, 10.21% (v/v); total acidity, 3.86 g/L; and pH value, 3.58.

The wine yeast strain CECA (Angel Yeast Co., Ltd., China) was used for fermentation. All chemical reagents and analytical standards were of analytical or chromatographic grade. Ursolic acid and glucose standards were purchased from commercial suppliers and used for quantitative analysis.

2.2. Extraction and Preparation of Functional Components from Cornus Fruit

2.2.1. Extraction of Ursolic Acid

Ursolic acid (UA) was extracted from dried *C. officinalis* fruit using ultrasound-assisted extraction according to a previously reported method with slight modifications. The optimized extraction conditions were as follows: ethanol concentration, 74%; liquid-to-solid ratio, 1:31 (g/mL); ultrasonic power, 200 W; extraction temperature, 70°C; and extraction time, 50 min.

After extraction, the mixture was centrifuged at 4000 rpm for 15 min, and the supernatant was collected. The extract was concentrated under reduced pressure using a centrifugal vacuum concentrator at 200 mbar, 90 °C, and 66% rotational speed [6]. The concentrated product was collected as UA extract for further analysis.

2.2.2. Extraction of Polysaccharides

Polysaccharides (PS) were extracted from dried *C. officinalis* fruit using ultrasound-assisted extraction with slight modifications. The optimized extraction conditions were as follows: liquid-to-solid ratio, 1:30 (g/mL); ultrasonic power, 400 W; extraction temperature, 70°C; and extraction time, 70 min.

After extraction, the suspension was centrifuged at 4000 rpm for 15 min at 4 °C. The supernatant was collected, and four volumes of absolute ethanol were added for polysaccharide precipitation. The mixture was stored at 4°C for 24 h and then centrifuged at 6000 rpm for 15 min [7]. The precipitate was collected, dried, and dissolved for subsequent analysis.

2.3. Determination of UA and PS Contents

2.3.1. Determination of Maximum Absorption Wavelength

UA: The UA content was determined using the vanillin–perchloric acid colorimetric method with minor modifications [8]. Briefly, 0.5 mL of UA standard solution or 0.2 mL of sample solution was transferred into a test tube and evaporated to dryness in a boiling water bath. Subsequently, 0.2 mL of 5% (w/v) vanillin in glacial acetic acid solution and 0.8 mL of perchloric acid were added sequentially. The mixture was incubated at 60°C for 15 min and rapidly cooled to room temperature. Then, 5 mL of glacial acetic acid was added and mixed thoroughly. A reagent blank containing only vanillin-glacial acetic acid solution, perchloric acid, and glacial acetic acid was prepared simultaneously. The absorbance spectrum was recorded from 450 to 700 nm using a UV-Vis spectrophotometer. The wavelength corresponding to the maximum absorption was selected for quantitative determination.

PS: The total polysaccharide content was determined using the phenol-sulfuric acid method [9]. Briefly, 1 mL of glucose standard solution was mixed with 1.2 mL of 5% phenol solution, followed by rapid addition of 7.5 mL concentrated sulfuric acid. After complete mixing, the reaction solution was allowed to stand at room temperature. The absorbance spectrum was recorded from 400 to 700 nm, and the wavelength showing the maximum absorbance was selected for polysaccharide determination.

2.3.2. Construction of Standard Curves

UA Standard Curve: Different volumes (0-1.0 mL) of

UA standard solution (0.1 mg/mL) were transferred into test tubes. After evaporation, the color development procedure described above was performed. The absorbance was measured at the selected wavelength. A calibration curve was established using UA concentration as the independent variable and absorbance as the dependent variable.

Total Sugar Standard Curve: Glucose solutions with different concentrations were prepared and treated according to the phenol-sulfuric acid method. Absorbance was measured at the selected wavelength. The calibration curve was generated using glucose concentration as the x-axis and absorbance as the y-axis.

Reducing Sugar Standard Curve: Reducing sugars were determined using the 3,5-dinitrosalicylic acid (DNS) method. Different concentrations of glucose standard solution were mixed with DNS reagent and heated in a boiling water bath for 5 min. After cooling, distilled water was added, and absorbance was measured at 562 nm. The standard curve was constructed using glucose concentration as the independent variable.

2.3.3. Method Validation

Stability Test: Aliquots of 1 mL of 0.1 mg/mL ursolic acid reference solution, glucose reference solution, and *C. officinalis* test solution from the same batch were transferred into test tubes. After evaporation to dryness in a boiling water bath to remove the solvent, the samples were treated according to the color development procedure described in Section 2.3.1. The absorbance (A) was measured at the selected wavelength every 10 min over a period of 2 h. The relative standard deviation (RSD) of the stabilized absorbance values was calculated to determine the time range over which the color development of the sample solution remained stable.

Precision Test: Aliquots of 1 mL of 0.1 mg/mL ursolic acid reference solution, glucose standard solution, and *C. officinalis* test solution from the same batch were transferred into test tubes. After solvent removal by evaporation in a boiling water bath, the samples were treated according to the procedure. The absorbance (A) was measured six consecutive times at the selected wavelength, and the relative standard deviation (RSD) of the absorbance values was calculated [10].

Repeatability Test: Six portions of *C. officinalis* dried fruit powder with identical mass were weighed and independently prepared into test solutions. Each sample was subjected to the color development procedure described in Section 2.3.1, and the absorbance (A) was measured at the selected wavelength. The relative standard deviation (RSD) of the absorbance values was calculated to evaluate repeatability [11].

2.3.4 Calculation of Extraction Yields

The active material extracted following the procedure in Section 2.2 was subsequently weighed. The extraction yields of UA (1) and PS (2, 3) from *C. officinalis* were calculated according to the following equation.

$$T_u(\%) = \frac{m_1 \times n \times V_2}{V_1 \times m_2 \times 10^6} \times 100\%\# \quad (1)$$

In Equation (1): T_u: extraction yield of UA from *C. officinalis*, %; m_1: mass of UA calculated from the standard curve, μg; n: dilution factor of the sample; V_2: total volume of the sample solution, mL; V_1: volume of sample solution used for determination, mL; m_2: total mass of the sample, g.

$$C_P(\%) = \left(\frac{c_1 \times n_1}{10^3} - c_2 \times n_2 \right) \times \frac{V_1}{V_2} \times 100\%\# \quad (2)$$

$$T_p(\%) = \frac{C_p \times V}{m \times 10^3} \times 100\%\# \quad (3)$$

In Equation (2) and (3):

C_p: concentration of PS from *C. officinalis*, g/L; c_1: total sugar concentration calculated from the total sugar standard curve, μg/mL; n_1: dilution factor for total sugar determination; c_2: reducing sugar concentration calculated from the reducing sugar standard curve, g/L; n_2: dilution factor for reducing sugar determination; V_1: volume of distilled water added to dissolve the polysaccharide precipitate, mL; V_2: volume of polysaccharide supernatant before precipitation, mL; T_p: extraction yield of PS from *C. officinalis*, %; V: total volume of the sample solution, mL; m: total mass of the sample, g.

2.4. Determination of In Vitro Antioxidant Activity

According to previously described methods [12], the antioxidant capacities (hydroxyl radical, DPPH, and ABTS scavenging) of UA and PS were evaluated using commercial assay kits (Solarbio, Beijing, China) with vitamin C as the positive control. Briefly, gradient dilutions of each sample were prepared and mixed with the respective kit reagents. Following incubation under the conditions specified in assay kits (temperature, time, and wavelength), the absorbance was recorded using a UV-Vis spectrophotometer. All tests were conducted in triplicate, and scavenging/inhibition rates were calculated according to the kit-specific equations.

2.5. Determination of α-Glucosidase Inhibitory Activity

A defined amount of *C. officinalis* functional components was dissolved in PBS buffer to prepare a series of gradient concentrations (0-0.10 mg/mL). The functional component solutions at different concentrations, together with the extract solution, were centrifuged at 10,000 r/min for 10 min at 4°C [13]. The resulting supernatants were collected as test samples. The corresponding reagents were added sequentially and mixed thoroughly, after which the absorbance was measured at 405 nm.The α-glucosidase inhibitory activity was calculated according to Equation (4).

$$G(\%) = \left(1 - \frac{A_a - A_b}{A_c - A_d} \right) \times 100\%\# \quad (4)$$

In Equation (4): G: α-glucosidase inhibitory activity; A_a: Absorbance value of the test tube; A_c: Absorbance value of the positive control tube; A_d: Absorbance value of the blank tube; A_b: Absorbance value of the control tube.

2.6. Preparation of Fruit Wines

2.6.1. *C. officinalis* Fermented Wine

The production process of *C. officinalis* fermented wine is shown in Figure 1. Dried fruits were washed, deseeded, destemmed, oven-dried (60°C, 24h), and rehydrated with hot water (85–95°C, 1:10, g). After cooling, pectinase (20mg/L) and SO_2 (60mg/L) were added. The mash sugar content was adjusted to 187 g/L (11% potential alcohol) and homogenized. Activated CECA yeast (0.3%, v/v; activated in water, 1:10, g, at 37°C for 30min) was inoculated, and fermentation proceeded at room temperature under an airlock. CO_2-related weight loss was monitored daily until a plateau indicated fermentation completion. The wine was then supplemented with SO_2 (60 mg/L), filtered through sterile gauze, and clarified at 4°C for 4 days.

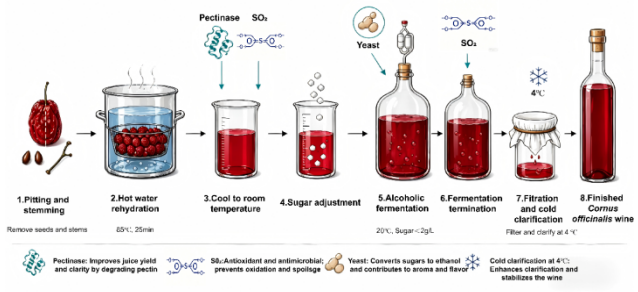

Figure 1. Process Flow for the Production of *C. officinalis* Fermented Wine

2.6.2. C. officinalis-Infused Grape Wine

Dried *C. officinalis* fruits were washed, cleaned, and dried at 60°C for 24h. Commercial grape wine was added to the dried fruit material at a solid-to-liquid ratio of 1:10 (g:mL). The mixture was sealed and macerated at room temperature in darkness for 15 days.

After maceration, the mixture was filtered through sterile gauze to obtain *C. officinalis*-infused grape wine.

2.6.3. Functional Component-Fortified Wines

Based on the optimal concentrations determined from in vitro antioxidant and α-glucosidase inhibitory assays, UA and PS were separately incorporated into two wine matrices.

For *C. officinalis* fermented wine: UA was added to obtain a final concentration of 1.5 mg/mL; PS was added to obtain a final concentration of 3.0 mg/mL. The resulting samples were designated as UA-fortified fermented wine and PS-fortified fermented wine.

For grape wine: UA was added to obtain a final concentration of 1.5 mg/mL; PS was added to obtain a final concentration of 3.0 mg/mL. The resulting samples were designated as UA-fortified grape wine and PS-fortified grape wine.

All wine samples were stored under identical conditions before analysis.

2.6.4. Experimental Design and Sample Coding

Seven wine samples were included in this study:
grape wine (W);
C. officinalis-infused grape wine (CW);
UA-fortified grape wine (WTU);
PS-fortified grape wine (WTP);
C. officinalis fermented wine (CB);
UA-fortified *C. officinalis* fermented wine (CTU);
PS-fortified *C. officinalis* fermented wine (CTP).

2.7. Determination of Physicochemical Properties of Fruit Wines

The main physicochemical parameters of fruit wines, including residual sugar, alcohol content, total acidity, pH value, and volatile acidity, were determined according to the Chinese national standard method GB/T 15038-2006 (General Analytical Methods for Wine and Fruit Wines). All measurements were performed in triplicate.

2.8. Determination of Individual Phenolic Compounds

Monomeric phenolic compounds in *C. officinalis* fruit wine were determined using ethyl acetate extraction combined with reverse-phase high-performance liquid chromatography (RP-HPLC) [14].

Chromatographic Conditions: An XTerra MS C18 reverse-phase column (250 mm × 4.6 mm, 5 μm) was employed. For sample preparation, 100 mL of ethyl acetate was added to 50 mL of the wine sample, and the extraction was performed three times. The combined extracts were concentrated to dryness by rotary evaporation under reduced pressure at 35°C, then reconstituted with methanol to a final volume of 25 mL. The solution was filtered through a 0.45 μm organic microporous membrane prior to analysis.

Mobile Phase: Solvent A was 2% aqueous acetic acid, and solvent B was a mixture of 0.5% aqueous acetic acid and acetonitrile (50:50, V/V). The volume ratio of mobile phase A to B was 50:50.

Gradient Elution Program: 0-50 min, 10%-55% A and 90%-45% B; 50-60 min, 55%-100% A and 45%-0% B; 60-65 min, 100%-10% A and 0%-90% B; 65-75 min, 10% A and 90% B. The column temperature was maintained at 30 °C, and the flow rate was 0.8 mL/min.

Monomeric phenolic compounds were identified based on their retention times, and their concentrations were quantified using the external standard method.

2.9. Determination of Aroma Components

Volatile aroma compounds were qualitatively and quantitatively analyzed using gas chromatography-mass spectrometry (GC-MS) [15].

Headspace Solid-Phase Microextraction (HS-SPME): A 5.0 mL aliquot of the sample wine was transferred into a 15 mL vial, followed by the addition of 1.0 g NaCl, 10 μL of internal standard solution (2,000 mg/L 4-methyl-2-pentanol), and a magnetic stirring bar. The vial was placed on a magnetic stirrer and equilibrated at 40 °C with stirring for 30 min. Subsequently, the SPME fiber was inserted into the headspace and extracted at 40 °C under continuous stirring for 30 min. After extraction, the fiber was immediately inserted into the GC injection port and thermally desorbed at 250°C for 8 min.

GC-MS Analysis: Gas chromatographic analysis was performed using an Agilent 7890B GC coupled with an Agilent 5975B MS, equipped with an HP-INNOWAX capillary column (60 m×0.25 mm×0.25 μm). Samples were introduced via splitless injection. High-purity helium was used as the carrier gas at a constant flow rate of 1 mL/min. The injector temperature was set at 250°C, the MS transfer line at 280°C, and the ion source at 230°C. The oven temperature program was as follows: initial temperature of 50°C held for 1 min, increased to 220°C at a rate of 3°C/min, and held for 5 min. Electron ionization (EI) was employed at 70 eV, with a mass scanning range of 25-350 amu.

Qualitative and Quantitative Analysis: Compounds were identified by comparison with the NIST14 mass spectral library and by matching retention indices (RI) with those reported in the NIST Chemistry WebBook. Quantification was performed using the external standard calibration method, with correlation coefficients (R^2) exceeding 0.99.

2.10. Statistical Analysis

All experiments were conducted with three independent biological replicates, and results were expressed as mean ± standard deviation. Statistical significance was evaluated using one-way analysis of variance (ANOVA), followed by Duncan's multiple range test using IBM SPSS Statistics 26.0. Response surface methodology (RSM), regression analysis, and response surface plots were performed using Design-Expert 8.0.6. Principal component analysis (PCA), hierarchical cluster analysis (HCA), and Pearson correlation analysis were performed using Origin 2021. Statistical significance was defined as: $p < 0.05$: significant difference; $p < 0.01$: highly significant difference.

3. Results

3.1. Establishment and Validation of Analytical Methods for Functional Components

3.1.1. Determination of Maximum Absorption Wavelength and Calibration Curves

The maximum absorption wavelengths of ursolic acid (UA) and polysaccharides (PS) extracted from Cornus officinalis were determined using UV-Vis spectrophotometry [16].

The UA standard solution and *C. officinalis* extract exhibited identical absorption maxima at 548 nm (Figure 2A), while the reagent blank showed no obvious interference at this wavelength. Therefore, 548 nm was selected as the detection wavelength for UA quantification.

For PS determination, the glucose standard solution and polysaccharide extract showed maximum absorbance at approximately 485 nm and 490 nm (Figure 2B), respectively. Considering the stronger signal intensity and lower background interference, 490 nm was selected as the analytical wavelength.

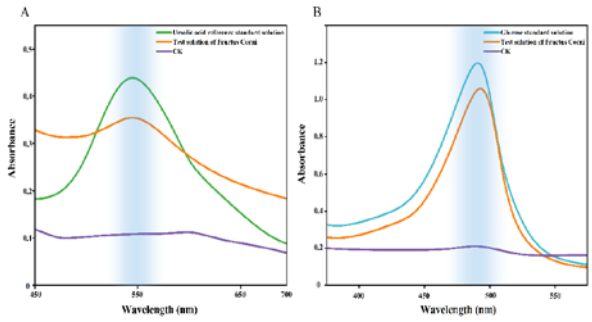

Figure 2. Determination of detection wavelengths for UA(A) and PS(B) in *C. officinalis*

The standard curves for each analyte were established at the selected wavelengths:

UA standard curve:

$$y = 0.0107x + 0.0155 \left(R^2 = 0.9994 \right)$$

Total sugar standard curve:

$$y = 0.0077x + 0.0791 \left(R^2 = 0.9993 \right)$$

Reducing sugar standard curve:

$$y = 0.9235x - 0.0105 \left(R^2 = 0.9993 \right)$$

3.1.2. Validation of Analytical Methods

Stability: For UA determination, the absorbance values remained relatively stable after color development, with RSD values of 2.29% for the standard solution and 0.93% for the sample solution. For PS determination, both the standard and sample solutions exhibited excellent stability, with RSD values of 0.24% and 0.22%, respectively.

Precision: The precision tests showed that the RSD values of repeated measurements were below 0.20%, demonstrating good instrumental precision.

Repeatability: As shown in Table 1. The repeatability test using six independently prepared samples showed RSD values of 0.93% for both UA and PS determination, confirming good reproducibility.

Recovery: As shown in Table 2. The recovery experiment showed an average recovery rate of 98.14% with an RSD of 1.76%, indicating that the established methods were accurate and reliable for determining UA and PS contents in *C. officinalis* samples.

Table 1. Repeatability test results

Sample	Absorbance（n=6）						Absorbance	
	1	2	3	4	5	6	Average	*RSD*(%)
UA test sample	0.2290	0.2281	0.2281	0.2321	0.2325	0.2322	0.2303	0.93
PS test sample	0.4076	0.4025	0.4084	0.4050	0.4153	0.4127	0.4086	1.16

Table 2. Sample recovery test results

Sample	No.	Background Value (μg)	Spiked Amount (μg)	Measured Value (μg)	Sample Recovery (%)	Average Recovery (%)	*RSD* (%)
UA test sample	1	18.56	10.64	28.93	97.42	98.14	1.76
	2	20.24	10.64	30.51	96.54		
	3	19.95	10.64	30.33	97.51		
	4	20.28	10.64	31.08	101.50		
	5	20.25	10.64	30.67	97.95		
	6	19.87	10.64	30.29	97.90		
PS test sample	a	45.52	29.51	68.09	99.62	100.01	0.64
	b	46.07	29.51	68.96	100.96		
	c	42.66	29.51	65.74	99.91		
	d	45.66	29.51	68.11	99.27		
	e	43.89	29.51	67.00	100.61		
	f	42.32	29.51	65.39	99.68		

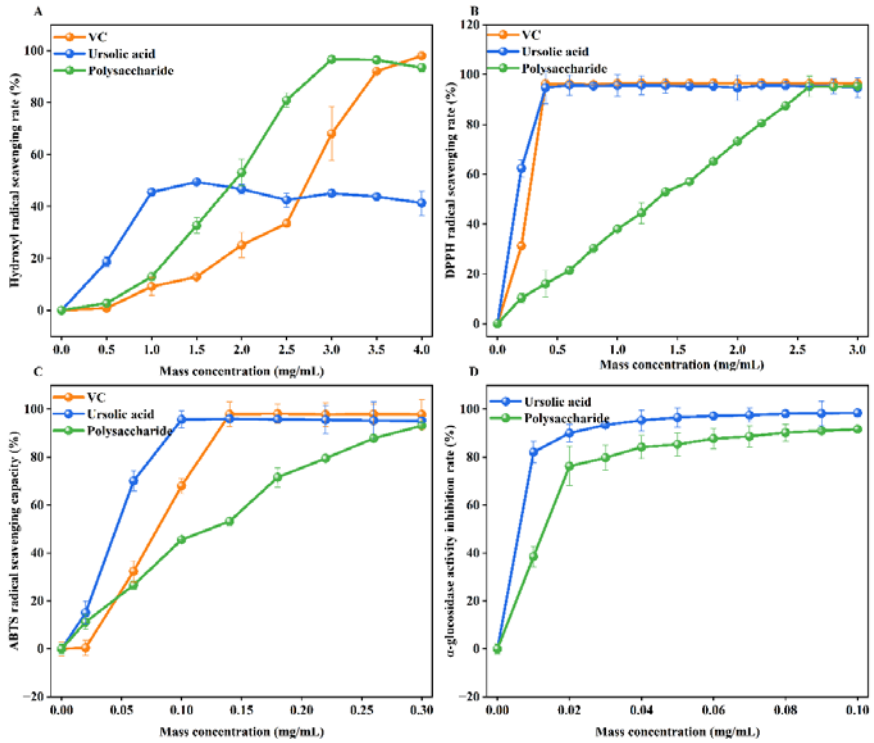

Figure 3. A. Hydroxyl radical scavenging capacity; B. DPPH radical scavenging capacity;; C. ABTS radical scavenging capacity; D. α-glucosidase activity inhibition rate

3.1.3. Extraction Yield of UA and PS from Cornus officinalis

The extraction yield of UA was 4.17 ± 0.12%, whereas the yield of PS was 2.88 ± 0.14%.

The results demonstrated that ultrasonic-assisted extraction effectively enriched both functional components from *C. officinalis*. The obtained yields provided sufficient quantities for subsequent biological evaluation and wine fortification experiments.

3.2. In Vitro Antioxidant Activity of UA and PS

The antioxidant activities of UA and PS were evaluated using DPPH, ABTS, and hydroxyl radical scavenging assays [17]. Both UA and PS exhibited concentration-dependent antioxidant activities, although their response

patterns differed significantly.

UA showed strong antioxidant capacity at relatively low concentrations. At 0.4 mg/mL, UA achieved more than 95% DPPH radical scavenging activity (Figure 3B), while ABTS radical scavenging activity exceeded 96% at 0.14 mg/mL(Figure 3C).

PS exhibited a gradual concentration-dependent increase in antioxidant activity. Compared with UA, PS required higher concentrations to achieve comparable DPPH and ABTS scavenging effects. However, PS showed excellent hydroxyl radical scavenging ability, reaching 96.7% inhibition at 3.0 mg/mL(Figure 3A).

The antioxidant activity profiles indicated that UA and PS exhibited different functional characteristics.

3.3. In Vitro α-Glucosidase Inhibitory Activity

As shown in Figure. 3D, at a concentration of 0.01 mg/mL, UA from *C. officinalis* exhibited a significant increase in α-glucosidase inhibitory activity, reaching 82.13 ± 0.47%. This result indicates strong α-glucosidase inhibitory activity of the UA-containing sample at a low test concentration, corresponding to 2.13 times the inhibition observed for PS at the same concentration. At 0.10 mg/mL, the inhibitory activity of UA reached a maximum value of 98.50 ± 0.52%.

For *C. officinalis* PS, the inhibitory effect increased significantly at a concentration of 0.02 mg/mL, achieving an inhibition rate of 76.25 ± 8.17%. Previous studies have demonstrated that higher contents of galactose, galacturonic acid, and glucuronic acid, lower molecular weight, and a greater abundance of free carboxyl and hydroxyl groups enhance the α-glucosidase inhibitory activity of PS. At 0.10 mg/mL, the inhibitory rate of PS reached a peak value of 91.70 ± 1.08%, indicating effective inhibition of α-glucosidase activity. These findings suggest that the PS from *C. officinalis* may possess relatively low molecular weight and be rich in galactose and galacturonic acid. Previous studies have identified polysaccharide fractions such as PFC and FCPC in *C. officinalis*, both of which contain glucuronic acid and relatively high levels of xylose, contributing positively to their hypoglycemic activity [18].

Both UA and PS exhibited strong α-glucosidase inhibitory activity at 0.10 mg/mL; however, UA showed greater inhibition under the tested conditions.

3.4. Effects of UA and PS on Physicochemical Properties of Fruit Wines

The effects of UA and PS supplementation on the physicochemical properties of different wine matrices are summarized in Table 3.

The addition of PS showed no significant influence on residual sugar, total acidity, volatile acidity, alcohol content, or pH values in either grape wine or *C. officinalis* fermented wine. In contrast, UA supplementation resulted in significant increases in total acidity contents in both wine systems. No significant changes in volatile acidity were observed among different treatments, indicating that the addition of UA and PS did not promote microbial spoilage during storage.

3.5. Effects of UA and PS on Phenolic Composition of Fruit Wines

The phenolic profiles of different wine samples are presented in Table 4. A total of 15 individual phenolic compounds were detected in grape wine, whereas 10 compounds were identified in *C. officinalis* fermented wine. The addition of UA and PS produced different effects depending on the wine matrix.

In grape wine, UA supplementation increased the contents of several phenolic compounds, including chlorogenic acid, catechin, resveratrol, and total flavonoids. PS addition mainly affected specific compounds, increasing phloridzin content while reducing several phenolic acids and flavonoids.

In *C. officinalis* fermented wine, UA increased the contents of several phenolic compounds, including epicatechin, rutin, quercetin, and gentisic acid. PS supplementation resulted in increases in ferulic acid, p-coumaric acid, and syringic acid.

These results demonstrated that UA and PS affected phenolic composition differently depending on the wine matrix.

Table 3. Basic physicochemical indexes of different *C. officinalis* fruit wines

Sample	Total Acid (g/L)	Volatile Acid (g/L)	pH	Alcohol (%vol)	Reducing Sugar (g/L)
W	3.86±0.02e	0.39±0.02a	3.58±0.01a	10.32±0.19a	4.72±0.08d
CW	6.71±0.09b	0.12±0.06b	3.34±0.02b	10.38±0.06a	19.01±2.12a
WTP	3.89±0.02e	0.37±0.02a	3.56±0.01a	10.32±0.18a	4.68±0.19d
WTU	4.65±0.14d	0.41±0.01a	3.33±0.01b	10.29±0.18a	10.47±0.44b
CB	5.60±0.07c	0.12±0.01b	3.29±0.01c	8.97±0.06b	2.70±0.07e
CTP	5.59±0.02c	0.12±0.01b	3.30±0.02c	8.97±0.06b	2.68±0.24e
CTU	7.02±0.14a	0.15±0.01b	3.22±0.01d	8.95±0.13b	7.60±0.32c

Note: Values marked with different lowercase letters a–e in the same column are significantly different (p<0.05).

Table 4. Functional substances in different *C. officinalis* fruit wines(μg/L)

Functional substances	Wine samples						
	W	CW	WTP	WTU	CB	CTP	CTU
Polysaccharide	1973.10±54.07 c	6368.50±510.88a	3022.65±36.92 b	1988.70±47.71 c	2875.49±43.44b	3047.66±43.66 b	3033.94±4.99b
Ursolic acid	1065.58±3.74c	2141.74±11.69 a	1105.66±18.35 c	1498.43±15.06 b	988.08±32.95 d	909.55±59.84e	1503.27±7.97b
Total Phenols	3524.04±35.58 cd	6214.42±235.58a	3374.36±556.00d	5101.28±702.45b	3159.23±37.69d	3003.84±131.81d	4066.15±113.49c
Total flavonoids	3182.5±118.00 b	3689.50±22.50 a	2750.67±101.36c	3591.67±44.84 a	1097.50±12.25e	1080.67±17.82 e	1503.92±20.60 d
Gallic acid	85.29±3.52c	159.44±3.01a	41.34±3.89e	80.20±4.20c	109.90±0.55b	77.73±20.81c	121.48±1.92b
3,4-Dihydroxybenzoic acid	61.96±8.05e	105.73±0.02b	33.31±7.13f	66.51±0.37e	102.22±1.08bc	87.76±4.18d	120.73±4.36a
Chlorogenic acid	22.63±0.83c	37.80±0.47a	18.88±3.10d	32.61±1.45b	ND	ND	ND
Vanillic acid	6.4±0.03bc	28.15±3.34a	8.06±2.31bc	19.42±3.94ab	ND	23.04±17.55a	ND
Caffeic acid	8.10±0.22c	17.90±1.07ab	6.70±0.91c	15.18±0.48bc	13.39±0.05bc	11.29±0.92bc	14.87±0.45bc
Syringic acid	8.16±0.20b	ND	8.38±1.41b	ND	19.88±0.04a	20.27±2.05a	ND
Epicatechin	14.64±1.85g	85.90±3.47d	23.67±4.84f	66.67±3.00e	136.75±1.10b	127.65±8.28c	148.91±5.26a
Catechin	2.53±0.46e	15.62±1.01b	6.79±1.07d	21.42±0.65a	21.10±0.01a	ND	13.52±0.78c
Coumaric acid	3.33±2.76ab	1.79±0.11bc	1.32±0.25bc	2.30±0.13bc	1.48±0.09bc	5.64±2.49a	1.73±0.18bc
Ferulaic acid	ND	ND	ND	ND	ND	34.69±5.64	ND
Melim	26.18±1.73e	88.77±0.81b	27.24±4.69e	61.12±3.63d	82.81±2.13c	ND	110.20±3.49a
Coumarin	ND	7.80±0.25a	3.31±0.68c	5.04±0.69b	ND	ND	ND
Phloridzin	8.54±0.29b	5.09±0.21cd	11.94±3.25a	7.43±0.90bc	2.69±0.01de	4.50±0.46d	4.60±0.25d
Myricetin	17.31±0.46	ND	ND	ND	ND	ND	ND
Resveratrol	6.28±0.02ab	5.17±0.3bc	5.03±0.62c	7.42±1.52a	ND	ND	ND
Meletin	ND	ND	ND	ND	ND	ND	10.35±0.39a
Campherol	1.28±0.15	ND	ND	ND	ND	ND	ND
Gentianic acid	19.63±8.18e	172.46±0.65c	8.86±2.82e	75.37±2.39d	218.74±1.60b	181.82±38.27c	266.10±11.89a

Note: "ND" indicates the substance was not detected or below the detection limit; Values with different lowercase letters a-e in the same row are significantly different (p<0.05).

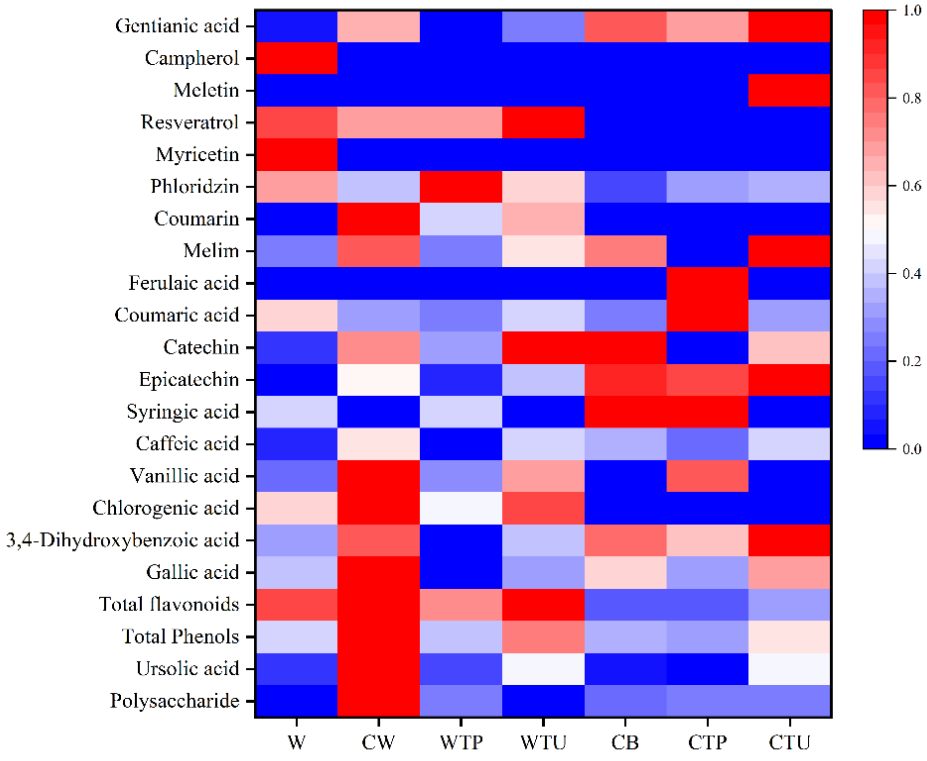

Figure 4. Effects of functional factors on functional substances of *C. officinalis* fruit wine

3.6. Effects of UA and PS on Bioactivities of Fruit Wines

The antioxidant activities and α-glucosidase inhibitory activities of different fruit wines are presented in Figure 5.

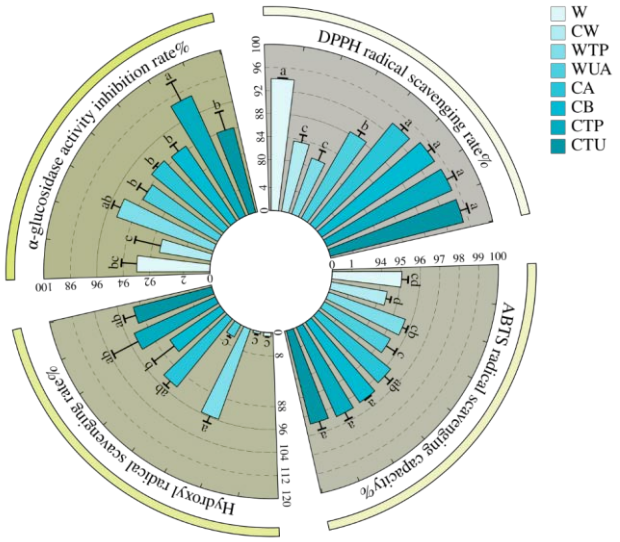

Figure 5. Effects of functional factors on biological activities of *C. officinalis* fruit wine

Overall, Cornus officinalis fermented wines exhibited stronger biological activities than grape wine-based samples. This difference was mainly attributed to the naturally abundant bioactive compounds present in *C. officinalis*, including polysaccharides, phenolic compounds, and triterpenoids.

Compared with untreated grape wine, the addition of PS produced different effects on antioxidant activities. PS significantly increased ABTS radical scavenging activity and hydroxyl radical scavenging activity, while DPPH radical scavenging activity showed a slight decrease.

UA supplementation showed a relatively limited effect on radical scavenging activities in grape wine. However, UA addition improved α-glucosidase inhibitory activity, suggesting that UA may contribute more strongly to enzyme-related functional properties rather than general antioxidant enhancement.

In *C. officinalis* fermented wine, PS supplementation further improved α-glucosidase inhibitory activity, whereas UA showed relatively minor effects.

These results indicate that the functional effects of UA and PS are strongly dependent on the chemical environment of the wine matrix.

3.7. Correlation between Chemical Components and Functional Activities

Pearson correlation analysis was performed to investigate relationships between chemical components and biological activities of fruit wines (Figure 6).

The correlation results showed that total phenolic content and total flavonoid content were not always positively correlated with antioxidant activities. Although phenolic compounds are generally considered important antioxidant contributors, the present results indicated that the functional activity of fruit wines was influenced not only by the total amount of bioactive compounds but also by the composition and interactions among individual molecules.

Several individual phenolic compounds exhibited strong positive correlations with antioxidant activity. Chlorogenic acid showed a strong correlation with DPPH radical scavenging activity (r = 0.86), while quercetin derivatives were positively associated with antioxidant performance (r = 0.82). Epicatechin showed a positive relationship with ABTS scavenging activity, suggesting that individual phenolic compounds may contribute more significantly to antioxidant activity than total phenolic concentration.

For α-glucosidase inhibitory activity, ferulic acid and syringic acid showed positive correlations, with correlation coefficients of 0.63 and 0.52, respectively.

Interestingly, UA, PS, total phenolics, and total flavonoids showed negative correlations with several antioxidant indicators. These results suggest that interactions among different functional components may influence bioactivity through matrix-dependent effects.

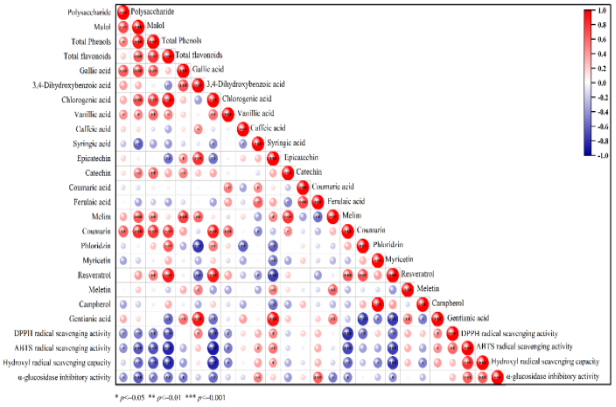

Figure 6. Correlation analysis between functional substances and biological activities of *C. officinalis* fruit wine

3.8. Effects of UA and PS on Aroma Characteristics of Fruit Wines

3.8.1. Changes in Volatile Aroma Compounds

The volatile aroma profiles of different fruit wines were analyzed using GC–MS, and the identified compounds are summarized in Table 5. A total of 50 volatile compounds were detected, including higher alcohols, esters, terpenes, acids, and aldehydes.

Table 5. Aroma composition of different dogwood fruit wines(μg/L)

Aroma compounds	Wine samples							Threshold[a,b]	OAV	Aroma Characteristics
	W	CW	WTP	WTU	CB	CTP	CTU			
(E)-3-hexene-1-alcohol	70.66±0.39b	37.96±3.01bc	ND	122.79±65.19a	ND	ND	ND	400	<1	Green grass
(Z)-3-hexene-1-alcohol	30.96±5.98a	16.14±0.07b	ND	26.63±4.89a	19.01±0.57b	ND	14.88±1.05b	400	<0.1	Green grass
(Z)-2-hexenol	1826.82±0.00a	ND	ND	576.91±166.75b	18.34±2.51c	ND	ND	400		NF
C6 alcohols	1928.44	54.10	ND	726.33	37.35	ND	14.88			
D-limonene	2.28±0.07b	2.15±0.03b	3.03±0.54a	2.34±0.13b	2.35±0.06b	2.43±0.15b	2.24±0.08b	10	>0.1	Citrus, Lemon
linalool	1.63±0.10e	8.71±0.36a	ND	1.63±0.14e	3.61±0.05c	2.47±0.18d	4.22±0.35b	25	<1	Floral
(E)-β-terpenol	203.58±54.46b	74.37±0.30c	ND	121.85±26.13c	66.11±0.75c	520.15±59.16a	78.09±1.52c	NF		Rose, lily, floral
α-Alphalinene	328.66±58.56a	ND	ND	261.43±11.92b	68.04±0.58c	ND	ND	> 100		Rose, lily, floral
4-terpenol	9.37±0.35a	7.42±0.06d	ND	7.48±0.07cd	7.54±0.02c	ND	7.68±0.06b	NF		NF
citronellol	21.33±0.20a	21.36±0.04a	ND	21.32±0.03ab	ND	21.33±0.04a	21.28±0.01b	100	<1	Floral, green note
nerol	61.12±0.03b	ND	ND	61.66±0.20b	61.02±0.35b	61.46±0.47b	62.63±0.99a	300	<1	Floral, sweet woody
α-terpineol	6.09±0.08d	6.02±0.04d	31.8±0.12a	ND	7.33±0.33c	7.23±0.01c	8.27±0.05b	330	<0.1	Camphor, pungent
geraniol	75.85±0.45ab	71.75±5.33ab	57.24±0.44b	81.83±26.09a	56.69±0.17b	58.68±2.40b	58.41±0.93b	20	>1	Lemon, geranium
farnesol	99.07±0	107.43±0	697.46±0	97.04±0	ND	100.32±0	86.3±0	20		Lily of the valley, green grass
Terpenes	808.98	299.20	789.53	656.57	272.69	774.07	329.12			
1-propanol	20505.49933.49b	20623.01±1202.91b	287725.11±119993.15a	27346±8222.62b	3505.63±1453.41b	7621.98±2403.61b	2940.73±229.29b	50000		Fruity, alcoholic
isobutanol	35960.29±186.84c	32888.04±132.30c	167080.2±58973.70b	46080.46±13633.98c	182473.78±29496.28b	256598.84±36935.92a	162419.7±11949.14b	4000	>1	Fusel, green note
1-butanol	2523.72±32.61a	2466.15±12.35a	2307.2±97.69b	2531.31±42.51a	2541.85±35.43a	2533.93±13.86a	2541.54±14.11a	150000	<0.1	Medicinal, phenolic
1-pentanol	ND	249.82±11.09b	335.97±0.02a	224.22±0.01c	ND	ND	220.34±10.72c	1000	<1	Fusel odor, Bread aroma
4-methyl-1-pentanol	18.52±1.94b	5.88±0.88c	ND	33.19±7.74a	2.44±1.81c	ND	1.38±2.20c	50000	<0.1	Almond, roasted
3-methyl-1-pentanol	57.28±2.67b	47.91±1.50b	ND	100.99±21.29a	ND	ND	ND	500	<1	Pungent, Bitter almond
1-hexanol	926.97±42.73b	433.41±16.46c	ND	1259.74±276.60a	22.81±3.49d	ND	37.17±21.47d	8000	<1	
1-heptanol	133.71±2.76b	137.6±5.47b	1707.41±518.42a	196.45±43.35b	137.59±31.99b	329.71±32.42b	142.83±17.19b	250	>0.1	Green grass, fruity
1-octen-3-ol	ND	ND	ND	ND	10.47±1.24a	2.56±0.34b	ND	20	<1	Fatty
1-octanol	13.35±0.43a	8.24±0.14c	6.33±0.05c	13.69±2.50a	11.19±0.20b	2.70±0.68d	8.32±0.76c	900	<0.1	Mushroom
1-decanol	25.82±0.18a	25.7±0.08ab	ND	25.66±0.06bc	ND	25.83±0.03a	25.52±0.01c	400	<0.1	Citrus, lemon, Jasmine
Higher alcohols	60165.15	56885.76	459162.22	77811.72	188705.75	267115.55	168337.53			
Ethyl acetate	36860.17±729.72b	33192.35±520.79b	180912.51±56086.35	38204.34±23243.28b	29084.54±4521.41b	43185.7±13167.98b	22255.19±10864.52b	7500	>	Pineapple, banana, Fruity,

Aroma compounds	Wine samples							Threshold[a, b]	OAV	Aroma Characteristics
	W	CW	WTP	WTU	CB	CTP	CTU			
			a						1	balsamic
Isobutyl acetate	20.00±1.02b	11.1±0.60b	ND	14.46±10.14b	82.66±9.45a	70.56±13.07a	59.34±36.79a	NF		NF
Isoamyl acetate	126.97±2.41	71.83±2.87	106.77±17.01	123.36±98.67	140.12±3.42	93.69±7.65	86.12±55.36	30	> 1	Banana, fruity
Acetate esters	37007.14	33275.28	181019.28	38342.16	29307.32	43349.95	22400.64			
Ethyl isobutyrate	1.94±0.07b	1.67±0.01b	929.08±115.37a	0.97±0.69b	1.59±0.26b	14.04±18.98b	1.29±0.93b	15		Fruity, vanilla
Ethyl butyrate	164.34±6.69ab	110.57±6.71bc	235.53±71.01a	152.77±106.97ab	22.81±2.11c	21.24±6.61c	16.19±7.65c	20	> 0.1	Strawberry, apple Banana
Ethyl 2-methylbutyrate	1.37±0.68bc	ND	11.25±2.18a	3.80±3.46b	ND	ND	ND	18	< 1	Apple peel Pineapple peel
Ethyl isovalerate	3.62±0.25ab	2.28±0.05bc	6.16±2.20a	3.26±2.41bc	1.14±0.14bc	1.56±0.89bc	0.88±0.48c	3	> 0.1	Banana, sweet fruit
Ethyl propionate	0.44±0.02ab	0.24±0.02c	ND	0.52±0.16a	0.50±0.02a	0.30±0.12bc	0.47±0.12ab	2100	< 0.1	Green apple
Ethyl 3-hydroxybutyrate	619.53±35.9b	625.02±18.97ab	ND	933.83±407.04a	ND	ND	919.86±0.04ab	20000	< 0.1	Grape
Ethyl nonanoate	5.22±0.35b	5.21±0.01b	ND	5.24±0.02b	5.39±0.04a	5.42±0.25a	5.41±0.04a	1300	< 0.1	Waxy, fruity
Ethyl decanoate	8.70±0.45b	13.58±0.03a	8.59±0.22b	8.19±0.36c	ND	ND	ND	200	< 0.1	Floral, rose, Fatty
Ethyl laurate	13.63±0.01b	ND	ND	13.69±0.04a	13.54±0.69d	13.57±0.23cd	13.58±0.01b	1500	< 0.1	Candy, floral Fruity
Fatty acid ethyl esters	818.79	758.57	1190.61	1122.28	44.97	56.13	957.69			
Isobutyl isovalerate	104014.13±539.9cd	88776.55±1036.84d	141897.79±24612.03ab	135712.73±32063.69abc	129819.2±332.35bc	167122.51±16115.78a	145953.45±12501.1ab	NF		NF
Ethyl lactate	63748.4±2360.52b	57832.08±2478.39b	ND	103827.1±20976.54a	1235.36±681.57c	609.36±25.68c	2236.41±645.23c	14000		Frankincense, raspberry
Isoamyl hexanoate	1.63±0.02de	1.77±0.21b	1.68±0.01c	1.62±0.11e	1.95±0.04a	1.67±0.05cd	1.77±0.02b	900	< 0.1	Apple, banana
Diethyl succinate	5889.21±313.30b	3024.24±198.81c	ND	10183.9±2038.04a	ND	ND	31.52±5.95d	120000	< 0.1	Grape
Other esters	173653.37	149634.64	141899.47	249725.35	131056.51	167733.54	148223.15			
Acetic acid	54468.43±1564.99c	56676.54±3099.93c	94656.21±29387.86b	90034.74±24573.43bc	56667.04±18132.03c	165575.78±18375.72a	59636.98±9742.55bc	200000	> 0.1	Vinegary
Propionic acid	159034.52±112206.49c	331388.58±17406.18b	ND	248401.51±49347.68bc	ND	ND	499262.79±0.86a	8100		NF
Isobutyric acid	941.70±77.72c	888.73±53.70c	4946.41±1988.27b	1789.86±743.11c	7457.78±1294.28a	9505.83±861.98a	8048.67±1488.40a	2300	> 0.1	Fatty, earthy, caramel
Isovaleric acid	938.60±0.85b	ND	ND	1317.14±588.64b	ND	3426.32±1052.74a	ND	1500		Rancid, Cheese
Octanoic acid	879.79±187.94ab	557.24±68.88bc	ND	1144.89±365.97a	522.00±10.45c	415.52±84.40c	535.57±48.90c	500		Butter, almond
Volatile fatty acids	216263.04	389511.09	99602.62	342688.15	64646.82	178923.45	567484.02			
3-Hydroxy-2-butanone	17573.97±440.94b	16175.18±1212.79b	ND	31229.02±9249.71a	ND	ND	ND	NF		NF
Furfural	97.18±1.28b	80.18±0.26c	ND	120.18±19.57a	ND	72.96±0.68c	70.39±2.43c	15000	< 0.1	Roasted, caramel
Decanal	1.84±0.03b	1.62±0.08	ND	1.69±0.21b	ND	2.31±0.17	1.46±0.02	10	<	Orange peel

Aroma compounds	Wine samples							Threshold[a, b]	OAV	Aroma Characteristics
	W	CW	WTP	WTU	CB	CTP	CTU			
		cd		c		a	d		1	
4-Methyl-5-butyldihydro-2-(3H)-furanone	ND	ND	339.86±0.42a	313.32±1.70b	304.63±0.23d	309.37±1.08c	ND	NF		NF
Aldehydes and ketones	17672.99	16256.98	339.86	31664.21	304.63	384.64	71.85			
Benzaldehyde	29.58±0.37b	20.52±1.02b	375.08±235.08a	36.50±7.39b	1.44±0.02b	1.21±0.05b	1.5±0.07b	500	< 1	Pungent, ripe apple, bitter almond
Methyl salicylate	1949.58±1637.45b	ND	ND	4955.36±1399.87a	ND	ND	ND	40		Mint
Ethyl salicylate	9.92±0.01a	9.8±0.01c	ND	9.89±0.11b	9.78±0.25d	ND	ND	NF		Wintergreen, Mint
Phenethyl acetate	13.51±.62a	7.23±0.26b	ND	9.98±2.99b	ND	ND	7.38±0.31b	250	< 0.1	Peach, strawberry, Rose, candy
Benzyl alcohol	1041.43±5.57b	861.58±75.84b	ND	1621.33±335.14a	152.48±5.80c	169.53±7.49c	157.36±2.13c	200000	< 0.1	Roasted, almond flavor
Phenethyl alcohol	18683.74±687.62b	12510.14±236.00c	ND	27914.26±3750.80a	13991.48±2173.57c	15076.14±221.68bc	16635.98±3572bc	10000		Rose, floral, honey
Phenol	4.50±0.02c	64.05±2.17b	ND	19.59±10.66c	65.69±11.34b	129.27±25.97a	88.26±21.00b	25000	< 0.1	Shoe polish
4-Ethyl-2-methoxyphenol	61.68±2.01b	57.99±0.44c	ND	66.72±1.50a	ND	ND	ND	25		Woody, smoky, spicy
4-Ethylphenol	36.31±0.02b	28.87±1.65b	ND	57.63±12.61a	11.86±0.07c	12.11±0.21c	11.70±0.06c	440	< 1	Animalic
Benzoic acid	20032.37±0.87b	ND	54205.78±28860.58a	ND	ND	20521.37±1.95b	ND	NF		NF
Aromatic compounds	41862.63	13560.18	54580.86	34691.24	14232.74	35909.63	16902.17			
2-methoxy-3-isobutylpyrazine	2.38±0.01a	ND	ND	2.38±0.02a	ND	ND	ND	NF		NF
3-(methylthio)-1-propanol	58342.33±21981.89a	ND	ND	31295.23±8686.78b	23078.95±3662.86b	35654.43±399.41b	30635.41±1676.86b	NF		Onion, meaty
β-ionone	2.58±0.34b	ND	4.12±0.50a	ND	ND	ND	ND	0.09		Violet
Whisky lacton	312.97±5.39a	ND	ND	ND	ND	ND	ND	74		NF
Other compounds	58660.27	ND	4.12	31297.60	23078.95	35654.43	30635.41			

Note: "ND" indicates the compound was not detected or below the detection limit; OAV = detected concentration / threshold concentration; values with different lowercase letters a–e in the same row are significantly different (p<0.05).

Grape wine exhibited a more complex volatile profile than *C. officinalis* fermented wine, with higher levels of varietal aroma compounds such as geraniol, citronellol, and nerol. The addition of PS produced significant changes in volatile composition, particularly in grape wine.

Compared with untreated grape wine, PS supplementation reduced several varietal aroma compounds, including C6 alcohols and terpenes, while increasing fermentation-derived compounds such as higher alcohols and acetate esters. The contents of higher alcohols, acetate esters, and fatty acid ethyl esters increased by approximately 7.63-, 4.89-, and 1.45-fold, respectively.

In contrast, UA addition had a relatively mild influence on volatile composition. UA supplementation maintained the main varietal aroma compounds of grape wine while increasing some fatty acid esters associated with creamy and fruity aromas.

These results demonstrate that PS and UA regulate aroma profiles through different pathways.

3.8.2. Hierarchical Cluster Analysis of Aroma Profiles

Hierarchical cluster analysis was performed based on volatile compound composition. The seven wine samples were classified into three major groups(Figure 7A).

The first group consisted of grape wine, *C. officinalis*-infused grape wine, and UA-fortified grape wine. These samples shared similar aroma characteristics dominated by floral and fruity volatile compounds. The second group included *C. officinalis* fermented wine and its UA- and PS-fortified derivatives. These samples showed relatively similar aroma profiles, characterized by higher alcohols

and fatty acid-related compounds. The third group consisted solely of PS-fortified grape wine, indicating that PS supplementation caused substantial modification of the original grape wine aroma profile.

3.8.3. Principal Component Analysis of Aroma Compounds

Principal component analysis was conducted using volatile compounds with odor activity values (OAV) greater than 0.1. The first principal component clearly separated PS-fortified grape wine from other samples (Figure 7B).

PS-fortified grape wine showed strong associations with fermentation-derived aroma compounds, including isoamyl acetate, ethyl acetate, and n-heptanol, which contribute fruity and fatty aroma characteristics.

Grape wine, *C. officinalis*-infused grape wine, and UA-fortified grape wine were mainly associated with geraniol and other terpene compounds responsible for floral and citrus-like aromas.

C. officinalis fermented wine and its derivatives were associated with compounds such as isobutanol and isobutyric acid, indicating stronger fermentation-derived and fatty aroma characteristics.

Overall, PS mainly enhanced fermentation-related fruity aromas but altered the original varietal aroma characteristics of grape wine. UA showed a better ability to preserve the inherent aroma structure while providing moderate aroma enhancement.

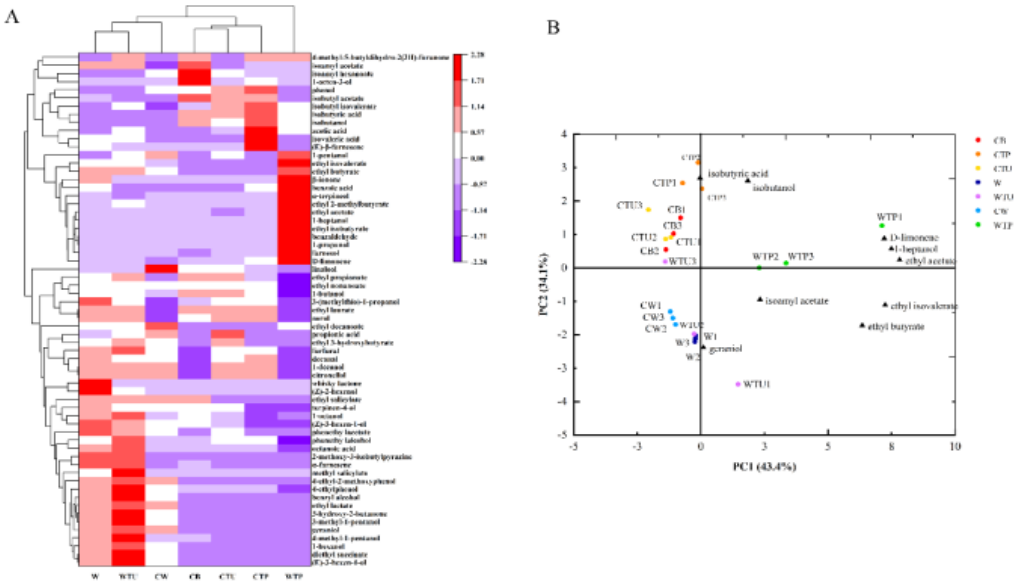

Figure 7. A. Heatmap of aroma cluster analysis; B. PCA principal component analysis

4. Discussion

4.1. Structure-related differences in the bioactivities of UA and PS

UA showed relatively high antioxidant and α-glucosidase inhibitory activities at lower concentrations than PS. This behavior is consistent with the structural characteristics of UA, a pentacyclic triterpenoid containing a hydrophobic carbon skeleton together with hydroxyl and carboxyl groups [19]. The hydrophobic region may facilitate association with nonpolar regions of proteins, whereas the polar groups can participate in hydrogen bonding. Previous kinetic, fluorescence, and molecular-docking analyses demonstrated that UA can inhibit α-glucosidase through noncompetitive binding and that hydrogen bonding contributes substantially to the formation of the UA–enzyme complex [20]. Therefore, the high inhibitory activity observed in the present study may be related to the capacity of UA to interact with sites outside the catalytic center and induce changes in enzyme conformation rather than simply competing with the substrate.

In contrast, the bioactivity of PS increased more gradually with concentration, and PS displayed particularly strong hydroxyl radical scavenging activity. Polysaccharide antioxidant activity is generally influenced by molecular weight, monosaccharide composition, branching degree, uronic acid content, and the availability of hydroxyl and carboxyl groups. These groups may contribute to radical scavenging through hydrogen or electron donation and may also chelate transition metal ions involved in hydroxyl radical generation. However, the present study did not characterize the molecular weight distribution, glycosidic linkage pattern, or monosaccharide composition of the extracted PS [21]. Consequently, the proposed contribution of uronic acids or specific sugar residues should be regarded as a mechanistic hypothesis rather than a confirmed structural explanation.

The different concentration–response patterns of UA and PS also indicate that activity measured per unit mass should not be equated directly with their practical value in food systems. UA may provide strong enzyme inhibition at a comparatively low dose, but its low aqueous solubility and hydrophobicity may restrict homogeneous dispersion in wine. PS is more compatible with aqueous systems but can interact extensively with other macromolecules, which may either expose or mask active groups [18]. Thus, the apparent activities of both components are likely to reflect a balance between intrinsic chemical reactivity and matrix-mediated accessibility.

4.2. Matrix-dependent modulation of phenolic composition and biological activity

The responses to UA and PS differed between grape wine and *C. officinalis* fermented wine, demonstrating a clear matrix effect. Grape wine is a complex colloidal system containing phenolic acids, flavan-3-ols, anthocyanins, condensed tannins, proteins, grape-derived polysaccharides, and yeast mannoproteins [22]. In comparison, *C. officinalis* fermented wine contains a different spectrum and proportion of iridoids, organic acids, triterpenoids, phenolics, and fruit-derived polysaccharides. These differences can alter the solubility, binding behavior, oxidation stability, and analytical recovery of exogenously added compounds [23].

The changes in individual phenolic concentrations after PS addition may arise from noncovalent interactions between PS and phenolic molecules. Hydrogen bonding can occur between the hydroxyl groups of polysaccharides and phenolics, whereas hydrophobic association may involve less polar regions of phenolic structures. Such interactions can modify phenolic solubility, aggregation, precipitation, and extractability during HPLC sample preparation [24]. More broadly, studies of complex food systems have shown that phenolics interact with polysaccharides and other macromolecules, thereby altering their accessibility and measured biological performance.

Accordingly, an increase or decrease in the measured concentration of a phenolic compound should not automatically be interpreted as chemical synthesis or degradation [25]. Because UA and PS were added after fermentation, the observed differences may partly reflect changes in phenolic stabilization, complex formation, extraction recovery, or oxidation during storage and analysis. Direct evidence for these possibilities would require complementary techniques such as size-exclusion chromatography, fluorescence spectroscopy, Fourier-transform infrared spectroscopy, nuclear magnetic resonance spectroscopy, or isothermal titration calorimetry [26].

The weak or negative relationships between total phenolic or flavonoid contents and some antioxidant indices further support the importance of compositional quality rather than total concentration alone. Individual phenolics differ markedly in the number and position of hydroxyl groups, conjugation patterns, redox potentials, and steric accessibility. Therefore, chlorogenic acid, epicatechin, quercetin derivatives, ferulic acid, and syringic acid may contribute differently to DPPH, ABTS, and hydroxyl radical scavenging assays. In addition, interactions among phenolics, PS, UA, organic acids, ethanol, and metal ions may produce additive, synergistic, or antagonistic responses [27].

However, correlations calculated from a small number of wine treatments should be interpreted cautiously. A high Pearson correlation coefficient does not demonstrate that a particular compound directly caused the observed biological activity. Some variables may change simultaneously because of treatment effects, and multiple compounds may contribute to the same assay response. Thus, the present correlation analysis is more appropriately viewed as a hypothesis-generating approach that identifies potential contributors for subsequent validation using purified compounds, reconstructed model wines, or multivariate regression.

The apparent increase in total acidity and reducing sugar after UA supplementation also requires cautious interpretation. Given the low UA addition level, a direct quantitative contribution of UA to total acidity or reducing sugar is unlikely to fully explain the observed changes. Possible explanations include matrix interference with the analytical assays, altered equilibrium among wine components, variation during sample handling, or treatment-associated analytical uncertainty [28]. This result should therefore not be presented as evidence that UA generated sugars or organic acids. Additional recovery experiments using UA-spiked model wine would help determine whether UA interferes with the relevant analytical methods.

4.3. Possible mechanisms underlying changes in volatile profiles

The most pronounced change in volatile composition was observed in PS-fortified grape wine. Because PS was added after alcoholic fermentation, the higher HS-SPME-GC–MS responses of higher alcohols and esters cannot reasonably be attributed to increased microbial production. Instead, PS may have altered the partitioning of pre-existing volatile compounds among the liquid phase, colloidal structures, and headspace, thereby affecting their availability for SPME extraction.

Wine polysaccharides can interact with aroma compounds and other matrix components through hydrogen bonding, hydrophobic interactions, and colloidal effects [29]. These interactions are highly dependent on polysaccharide structure, concentration, and the physicochemical properties of individual volatile compounds. Previous studies have shown that polysaccharides can either enhance or suppress the headspace availability of aroma compounds and that these effects may be further modified by phenolics and other macromolecules [30]. Therefore, the higher apparent abundances of isoamyl acetate, ethyl acetate, higher alcohols, and fatty acid ethyl esters observed after PS addition may reflect altered liquid–headspace partitioning or HS-SPME extraction efficiency rather than true increases in their total concentrations. Conversely, the lower measured levels of some C6 alcohols and terpenes may result from preferential retention within PS-associated colloidal structures or competitive extraction effects.

UA caused substantially smaller changes in the volatile profile and was associated with a terpene pattern more similar to that of untreated grape wine. Its lower addition level and smaller molecular size may have caused less disruption of the wine matrix than PS. However, the present results do not demonstrate that UA directly protects or retains linalool, geraniol, or other terpenes. Rather, UA may exert a comparatively limited effect on volatile partitioning under the tested conditions.

Overall, the present HS-SPME-GC–MS data cannot distinguish true changes in total volatile concentration from matrix-dependent changes in headspace partitioning and extraction response. Future studies using reconstructed model wines, matrix-matched standard addition, stable-isotope dilution analysis, or complementary exhaustive extraction methods are required to clarify these mechanisms

and determine whether the observed chemical differences translate into perceptible sensory changes.

4.4. Implications for the Targeted Formulation of Functional Fruit Wines

The results indicate that UA and PS serve different formulation objectives. UA appears more suitable when strong α-glucosidase inhibition is required at a relatively low addition level and when substantial modification of the original volatile profile is undesirable. PS may be more appropriate when enhancement of selected antioxidant properties and deliberate modulation of aroma release or mouthfeel are desired. Nevertheless, the optimal dose cannot be determined from in vitro activity alone, because increasing PS concentration may also alter clarity, viscosity, colloidal stability, and aroma perception [30].

The practical interpretation of α-glucosidase inhibition should also remain conservative. Enzyme inhibition measured in vitro indicates potential bioactivity but does not demonstrate an antidiabetic effect in humans. Wine ethanol content, gastrointestinal digestion, compound bioaccessibility, metabolism, and realistic serving size will influence the physiological relevance of UA and Ps [31]. Therefore, terms such as "hypoglycemic activity" should preferably be replaced with "in vitro α-glucosidase inhibitory activity" unless animal or clinical evidence is available.

Several limitations should be acknowledged. First, the PS extract was quantified as total polysaccharide but was not structurally characterized; therefore, the molecular features responsible for its activity remain unknown. Second, UA solubility and possible precipitation in the wine matrices were not reported. Third, the study relied on chemical antioxidant assays that do not fully reproduce biological oxidative conditions. Fourth, sensory analysis was not performed, making it impossible to determine whether the GC-MS differences were perceptible or desirable. Finally, direct molecular interactions among UA, PS, phenolics, proteins, and volatile compounds were inferred rather than experimentally demonstrated.

Future studies should characterize PS using monosaccharide profiling, molecular-weight determination, methylation analysis, and spectroscopic methods [32]. The binding of PS and UA to representative phenolics and aroma compounds should be evaluated in reconstructed model wines, followed by validation in real wine matrices. Sensory analysis, gastrointestinal digestion models, cellular assays, and storage-stability studies would further establish whether the observed chemical changes provide meaningful technological and physiological benefits.

Overall, the present study supports a component-specific rather than extract-wide strategy for the formulation of functional fruit wines. UA and PS should not be regarded as interchangeable sources of bioactivity: UA primarily provides low-dose enzyme inhibition with comparatively limited alteration of the aroma matrix, whereas PS exerts broader matrix-mediated effects on radical scavenging, phenolic behavior, and volatile partitioning. Rational formulation should therefore consider both the intrinsic activity of each component and its interactions with the chemical environment of the target beverage.

5. Conclusion

This study systematically compared the effects of Cornus officinalis-derived polysaccharides (PS) and ursolic acid (UA) on the chemical composition, volatile profile, and in vitro functional activities of two wine matrices. UA exhibited stronger radical-scavenging and α-glucosidase inhibitory activities at relatively low test concentrations, whereas PS showed a more gradual concentration-dependent antioxidant response and pronounced hydroxyl radical scavenging activity. In the wine systems, PS supplementation enhanced selected antioxidant and α-glucosidase inhibitory activities and was associated with substantial changes in the HS-SPME-determined volatile profile. In contrast, UA caused comparatively smaller changes in the original volatile pattern of grape wine and was associated with higher measured levels of the floral terpenes linalool and geraniol.

More importantly, the effects of UA and PS differed between grape wine and *C. officinalis*fermented wine, demonstrating that the functional behavior of isolated phytochemicals cannot be predicted solely from their intrinsic in vitro activities. The surrounding wine matrix appeared to influence phenolic composition, bioactivity, and volatile partitioning, resulting in component- and matrix-dependent responses. PS exerted broader matrix-mediated effects on radical scavenging and volatile distribution, whereas UA provided relatively strong α-glucosidase inhibition with less extensive alteration of the volatile profile. These findings support a component-specific formulation strategy rather than treating UA and PS as interchangeable functional ingredients.

Overall, this study provides a scientific basis for the targeted application of *C. officinalis*-derived UA and PS in functional fruit wine formulation and emphasizes the importance of matrix effects when incorporating isolated phytochemicals into fermented beverages. Further studies combining polysaccharide structural characterization, matrix-matched analytical validation, sensory evaluation, and physiologically relevant in vitro or cellular models are needed to confirm the mechanisms underlying these responses and to support the development of fruit wines with targeted chemical, sensory, and in vitro functional properties.

Conflicts of Interest

There are no conflicts of interest to declare.

Data Availability Statement

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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