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Application of fermented rose–mulberry fruit composite to enhance the antioxidant capacity, flavor, and sensory characteristics of mulberry wine

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Fruit wine made from a single fruit often lacks sufficient color, flavor complexity, and nutritional balance. In this study, we enhanced the sensory and functional qualities of mulberry wine through co-fermentation with *Rosa rugosa* petals. The rose fermentation wort was prepared by mixing rose petals and water at a 1 : 40 (w/w) ratio, and blended with mulberry pulp at a 1 : 1 (w/w) ratio prior to enzymatic treatment with pectinase. To comprehensively evaluate the effects of co-fermentation, a combination of analytical techniques was employed: the electronic nose (E-nose) and electronic tongue (E-tongue) were used to characterize overall aroma and taste profiles, while headspace solid-phase microextraction-gas chromatography-mass spectrometry (HS-SPME-GC-MS) was used to identify and quantify volatile aroma compounds. Nutritional composition and *in vitro* antioxidant activity were determined *via* standard biochemical assays, including DPPH and hydroxyl radical scavenging evaluation. This study aimed to elucidate how co-fermentation affects the antioxidant capacity, sensory characteristics, and flavor complexity of rose–mulberry wine compared to monovarietal mulberry and rose wines. The results showed that rose–mulberry wine exhibited significantly higher DPPH radical scavenging ability, hydroxyl radical scavenging activity (HRSA), hue angle (CH), and softness index (SI) compared to mulberry wine ($P < 0.05$). The E-tongue and E-nose indicated that rose–mulberry wine shared a closer flavor profile with rose wine, distinctly separating it from the more acidic and bitter profile of mulberry wine. GC-MS analysis identified 98 volatile compounds across the three wine varieties. Notably, rose–mulberry wine retained key volatile characteristics of mulberry wine, while co-fermentation significantly enhanced the presence of esters, alcohols, ketones, furans, pyrans, phenols, and others. Among these, newly identified esters such as isoamyl acetate, ethyl 9-decenoate, and ethyl undecanoate contributed distinct fruity and floral notes. In summary, rose–mulberry wine successfully integrates the unique aromatic traits of *R. rugosa* petals and mulberry fruit, producing a beverage with enhanced sensory complexity, functional antioxidant capacity, and improved overall acceptability.

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1 Introduction

Mulberry (*Morus alba* L.) fruit has been valued for thousands of years in Asia as both a nutritious food and a traditional medicinal ingredient. In 1993, it was officially approved by the Ministry of Health of China as a dual-purpose substance for both food and medicinal use.[1] Mulberries are rich in a wide array of nutrients, including fatty acids, amino acids, vitamins, minerals, and are particularly noted for their bioactive compounds such as anthocyanins, rutin, quercetin, chlorogenic acid, and polysaccharides, have been found in mulberry fruit.[2,3] These constituents have been associated with various biological activities, including antioxidant, neuroprotective, anti-atherosclerotic, and immunomodulatory effects.[4]

However, due to its high moisture content (ranging from 70.0% to 87.4%) and delicate epidermis, fresh mulberries are highly perishable and difficult to store for extended periods.[5] As a result, fermentation into wine has become an important method for preserving mulberries, reducing postharvest loss, and addressing issues related to overproduction.[6] Mulberry wine is one of the most popular fruit wines consumed worldwide due to its health benefits and unique flavor profile.[7] Current research on mulberry wine has primarily focused on optimizing the fermentation process;[8] analyzing the dynamic changes in volatile components, organic acids, volatile acids, and anthocyanins during fermentation;[9–11] screening *Saccharomyces cerevisiae* or non-*Saccharomyces cerevisiae* strains;[12] as well as exploring multi-strain co-fermentation techniques.[7,13]

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Similarly, *Rosa rugosa* cv. 'Plena' has been cultivated in China for millennia, with the Pingyin variety being especially prized for its rich, pure, and highly aromatic profile.[14] This rose cultivar is extensively utilized in the production of food, tea, and wine, among other applications. Furthermore, it was also officially recognized as both a food and a medicinal product by the Ministry of Health of China.[15] *R. rugosa* petals are a rich source of bioactive compounds, including polyphenols, amino acids, and vitamins, and are known for their antioxidant and anti-inflammatory properties.[16,17]

Aroma is a key quality attribute in wine evaluation and serves as a primary indicator of the stylistic differentiation among various products.[18] However, the majority of existing research has focused on the fermentation of single-source mulberry raw materials. This has typically resulted in mulberry wine with monotonous aroma profiles, dark coloration,[19] and excessive acetic acid levels,[13] which collectively reduce consumer acceptance. Recent studies have demonstrated that incorporating edible flower extracts into dealcoholized Merlot red wine can enhance its aromatic profile, showing stronger fruity and floral notes without altering critical chemical parameters such as sugar content, ethanol concentration, or total acidity.[20] Furthermore, mixed fermentation involving medicinal and edible homologous raw materials has also shown promising results in improving the overall flavor of fruit wine.[15,21,22] For example, compound wine produced through co-fermentation of *L. barbarum* and *P. cyrtonema* using *Saccharomyces cerevisiae* RW and *Debaryomyces hansenii* AS2.45 exhibited enhanced antioxidant properties and higher sensory acceptability scores.[21] Similarly, co-fermentation involving a *Cyclocarya paliurus*–kiwifruit composite not only enriched the flavor profile by introducing additional floral aromas but also significantly increased the levels of total flavonoids and polyphenols—compounds positively correlated with antioxidant capacity.[22] Therefore, the integration of fruits and flowers through compound fermentation represents a promising strategy to enhance both the nutritional value and sensory complexity of fruit wines. This approach has emerged as a significant research focus in the development of high-quality, functional fruit wines.

Our initial research optimized the co-fermentation process of mulberry juice and *R. rugosa* petals, resulting in a composite wine characterized by distinctive aromatic qualities and a smooth palate (the products of rose wine and rose–mulberry wine are commercially available and can be obtained from Sichuan Sangguaiguai Food Technology Co., Ltd). While both ingredients are individually known for their antioxidant properties and contributions to flavor, there is a notable lack of research investigating the synergistic effects of their co-fermentation on both antioxidant capacity and sensory characteristics. In particular, the extent to which co-fermentation can enhance these attributes beyond the additive effects of the individual components remains unclear. This study aims to fill this research gap by systematically evaluating the influence of co-fermentation on the antioxidant profile, sensory attributes, and aroma compounds of rose–mulberry wine. To address this, we employed an integrated analytical approach combining electronic nose (E-nose), electronic tongue (E-tongue), and headspace solid-phase microextraction-gas chromatography-mass spectrometry (HS-SPME-GC-MS), and *in vitro* antioxidant assays (DPPH and hydroxyl radical scavenging activity). These methods were complemented by multivariate statistical analyses, including principal component analysis (PCA), and partial least square discriminant analysis (PLS-DA), to systematically evaluate: (1) variations in nutritional components and antioxidant capacity, and (2) the differential flavor profiles of rose–mulberry wine compared to monovarietal mulberry and rose wine. The findings provide new insights into the synergistic effects of co-fermenting functional plant-based substrates and establish a theoretical foundation for the development of multifunctional rose–mulberry wine with enhanced market appeal.

2 Materials and methods

2.1 Materials

The experimental samples of mulberry wine, rose wine, and rose–mulberry wine were supplied by the Sericulture Research Institute, Sichuan Academy of Agricultural Sciences (Sichuan, China) and Sangguaiguai Food Technology Co., Ltd (Sichuan, China). Fresh mulberry fruits (*Morus alba* cv. 'Yueshen Dashi') were harvested from Xinmiao Township (Jialing District, Nanchong City, Sichuan Province, China), ensuring the selection of fully ripe fruits characterized by deep pigmentation and the absence of mechanical damage. Additionally, *R. rugosa* cv. 'Plena' was sourced from the Pingyin Rose Cultivation Base (Pingyin County, Jinan City, Shandong Province, China).

2.2 Chemicals and reagents

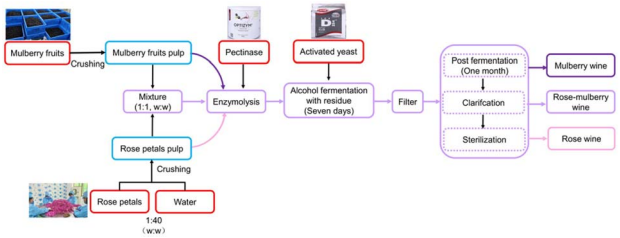
The Folin-Ciocalteu reagent was purchased from Xiamen Haibiao Technology Co., Ltd (Xiamen, China). A gallic acid standard solution (5 mg mL^{-1}) was purchased from Xiamen Haibiao Technology Co., Ltd (Xiamen, China). Rutin standard (≥98% purity) was purchased from Hefei Bomei Biotechnology Co., Ltd (Hefei, China). 2-Methyl-1-butanol (chromatographic purity, 99%) was purchased from Sigma-Aldrich (Shanghai, China). HPLC-grade methanol (99.9% purity) was purchased from Merck KGaA (Germany).

2.3 Instrumentation

BSA224S precision analytical balance (Sartorius AG, Germany); ZDJ-4B automatic potentiometric titrator (Shanghai Yidian Scientific Instrument Co., Ltd, China); UV-2550 ultraviolet-visible spectrophotometer (Shimadzu Corp., Japan); PEN3 electronic nose system equipped with 10-metal oxide sensors (Airsense Analytics GmbH, Germany); SA402B PLUS electronic tongue system equipped with 7 taste sensors (INSENT Inc., Japan); 7890A-5975C gas chromatography-mass spectrometer (GC-MS) equipped with a HP-5MS capillary column (30 m × 250 μm × 0.25 μm; Agilent Technologies, USA).

2.4 Methods

2.4.1 Preparation of experimental samples. The main fermentation was conducted for seven days, after which the



Scheme 1 The production process of rose–mulberry wine.

mixture was filtered. Subsequently, the wine was transferred to a new stainless-steel tank to initiate post-fermentation, which lasted for one month (Scheme 1).

Fresh mulberry fruits were first crushed to obtain mulberry pulp. Rose petals were mixed with pure water at a ratio of 1 : 40 (w/w) and subsequently crushed to form rose pulp. The resulting rose pulp was then blended with mulberry pulp at a 1 : 1 (w/w) ratio. The mixture's sugar content was adjusted to a reducing sugar content of 220 g L^{-1} using white granulated sugar, and the pH was regulated to 4.5 by adding citric acid. To facilitate the breakdown of plant cell walls and improve juice extraction, pectinase (≥500 U mg^{-1}) was added at a concentration of 20 mg kg^{-1}. The enzymatic treatment was conducted in a constant-temperature water bath at 20 °C for 4 h. Subsequently, *S. cerevisiae* was inoculated at a concentration of 200 mg kg^{-1}. The yeast was pre-activated by hydration in a 5% glucose solution at 35 °C for 20 minutes. Alcoholic fermentation was carried out in the dark at 24 °C for 7 days without stirring. After fermentation, the crude rose–mulberry wine was filtered through membrane filtration (using a 0.45 μm filter membrane) and transferred to a new sterilized stainless-steel tank to rest at 15 °C for 30 days and clarify (Scheme 1). The mulberry wine and rose wine were prepared using the same procedure, with fresh mulberry fruit and *R. rugosa* as the sole fermentation substrates, respectively. All other materials and fermentation conditions were identical to those used for rose–mulberry wine.

2.4.2 Analytical procedures

2.4.2.1 Physicochemical analysis. Total alcohol content (TAC), total sugar (TS), reducing sugar (RS), total acidity (TTA), and total volatile acidity (TVA) were analyzed in accordance with the Chinese National Standard GB/T 15038–2006 (General Analysis Method for Wine and Fruit Wine). The assessment of total flavonoids (TFC) utilized the aluminum chloride method as described by Magalhães *et al.*,[23] resulting in a spike recovery rate of 89.56% ± 1.77%. The determination of total phenolics (TPC) was performed following the Folin-Ciocalteu method as detailed by Bajčan *et al.*,[24] resulting in a spike recovery of 110.54% ± 2.37%. The quantification of tannins adhered to the Chinese National Standard NY/T 1600-2008, resulting in a recovery rate of 98.80% ± 0.33%. Vitamin C analysis was conducted in accordance with the Chinese National Standard GB/T 15038–2006, achieving a recovery rate of 108.02% ± 0.01%. The softness index (SI) was evaluated using the methodology proposed by Li *et al.*,[25] the samples were computed in accordance with eqn (1):

$$\text{Softness index (SI)} = \text{Total alcohol content} - (\text{Total acidity} + \text{Tannins}) \quad (1)$$

2.4.2.2 Determination of color. Color detection was performed using the method of Zhang *et al.*[26] All samples were prepared to measure the absorbance at wavelengths of 420, 520, and 620 nm. The color intensity (CI) and hue angle (CH) were calculated in accordance with eqn (2) and (3):

$$\text{Color intensity(CI)} = A420 + A520 + A620 \quad (2)$$

$$\text{Hue angle(CH)} = A420/A520 \quad (3)$$

2.4.2.3 Antioxidant assays. The assessment of 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical-scavenging activity was conducted following the methodology established by Kwaw *et al.*,[27] who employed a modified approach to evaluate DPPH radical scavenging capabilities. Additionally, the hydroxyl radical scavenging ability (HRSA) was measured using the technique outlined by Çelik *et al.*[28]

2.4.2.4 Volatile flavor analysis by electronic nose. The sample odor was analyzed using a PEN3 electronic nose (E-nose) following a standardized procedure. Initially, a 5 mL sample was placed in a sealed sampling bottle and allowed to equilibrate at room temperature for 30 min to facilitate the stabilization of volatile compounds. Following this equilibration period, the probe of the E-nose was inserted into the headspace of the bottle to detect volatile substances. The E-nose operated under predetermined conditions, which included a sample interval time of 1 s, a cleaning time of 100 s, a zeroing time of 10 s, a sample preparation time of 5 s, a determination time of 100 s, and an injection flow rate of 400 mL min^{-1}. Signal stability was achieved after 110 s, with signal acquisition occurring between 115 and 120 s. Each sample underwent five consecutive measurements. The collected data were processed using Winmuster software. The PEN3 E-nose detector consists of ten distinct sensor elements, each responsive to different sensitive substances, as detailed in Table 1.[29]

2.4.2.5 Taste analysis by electronic tongue. Before the analysis, the sensor array was subjected to immersion in a reference solution consisting of 30 mmol L^{-1} potassium chloride and 0.3 mmol L^{-1} tartaric acid for a duration of 30 seconds. Subsequently, the sensor array was immersed in the sample solution for 120 seconds, after which the equilibrium response value was recorded for subsequent statistical analysis. After each measurement, a 5% ethanol solution was used to clean the sensor until a stable sensor reading was obtained. Each sample was read three times in parallel, and the average was recorded for further analysis. Prior to analysis, the sensor array was immersed in a reference solution (30 mmol L^{-1} potassium chloride and 0.3 mmol L^{-1} tartaric acid) for 30 s, then immersed in the sample solution for 120 s. The response value at equilibrium was recorded for subsequent statistical analysis.

2.4.2.6 Determination of volatile aroma components. The HS-SPME combined with GC-MS was used. Specifically, 5 mL of the sample was added to a 20 mL headspace vial. The SPME syringe

Table 1 The main compounds detected by different chemical sensors

Sensor number	Sensor name	Performance description (sensitivity to)
R1	W1C	Aromatic compounds
R2	W5S	Nitrogen oxides, very sensitive to negative signal
R3	W3C	Aromatic amines
R4	W6S	Hydride, mainly selective for hydrogen
R5	W5C	Short-chain alkanes
R6	W1S	Methyl compounds
R7	W1W	Inorganic sulfides
R8	W2S	Alcohol
R9	W2W	Aromatic ingredients, sensitive to organic sulfides
R10	W3S	Long-chain alkanes

was inserted into the vial for the extraction process. The extraction temperature was set at 50 °C, and the extraction time was 30 min. Then, the extraction head was removed, and the chromatograph inlet was promptly inserted. Thermal desorption was conducted at 250 °C for 3 min, followed by chromatography and mass spectrometry.

Chromatographic conditions: an HP-5MS capillary column (30 m × 250 μm × 0.25 μm) was utilized, with the interface temperature set at 230 °C. The temperature program commenced at 60 °C, maintained for 2 min, then increased to 110 °C at a rate of 5 °C min^{-1} for 2 min, followed by a rise to 220 °C at a rate of 10 °C min^{-1} for 10 min. The carrier gas flow rate was maintained at 1.0 mL min^{-1} without any diversion.

Mass spectrometry conditions: 5975C quadrupole mass spectrometer, electron bombardment (EI) ion source, electron energy bombardment ionization was 70 eV; ion source temperature was set to 230 °C, quadrupole temperature was set to 150 °C, mass scan range was from 45 to 450 amu.

The volatile compounds identified were compared with the data available in the NIST 17 (National Institute of Standards and Technology Mass Spectrometry Library). Furthermore, qualitative analysis was conducted using the material retention index and the SH-Rxi-5Sil MS column as referenced in the literature.[30] The quantitative analysis was performed using the area normalization method to express the relative percentage content of each volatile compound.

2.4.3 Statistical analysis. The measurement and analysis results were processed using SPSS 23.0 and Excel 2007 for data processing. The results were presented as mean ± standard deviation, with a significance threshold set at $P < 0.05$. PCA and cluster analysis were performed using Origin 2023, and PLS-DA was performed using SIMCA 14.1.

3 Results and discussions

3.1 Quality attributes of different flavor types of wines

As shown in Table 2, all three wine samples exhibited alcohol concentrations of approximately 12% vol, and their total sugar (TS) level complied with the Chinese National Standard GB/T 15037-2006, which classifies wines with residual sugar content ≤4.0 g L^{-1} as dry wines. Total titratable acidity (TTA) is a critical parameter that influences the balance, taste, and overall quality of fermented alcoholic beverages. The TTA of mulberry wine exceeded 8 g L^{-1}, significantly surpassing that of rose wine. In contrast, the rose–mulberry wine, produced through the co-fermentation of mulberry fruit and rose petals, exhibited a notably lower TTA of 4.23 g L^{-1}, suggesting that co-fermentation can effectively moderate acidity and improve sensory balance. This reduction could be attributed to the metabolic activities of yeast and enzymatic interactions during fermentation, where acids may be consumed or converted into esters and alcohols, thereby improving the sensory balance and roundness of the wine.[31]

The total volatile acidity (TVA) is an indicator of acetic acid and potential spoilage, which significantly influences the quality of fermented beverages. In alcoholic fermentation, heightened concentrations of TVA are considered unfavorable, as they not only modify the sensory attributes of flavor and aroma but also indicate potential contamination by acetic bacteria.[32] The current study recorded TVA values for all treatments that were below 1 g L^{-1} (Table 2), suggesting that the fermentation process was well-controlled, minimizing the risk of acetic acid bacteria contamination and maintaining the microbial stability and quality of the final product.

Color characteristics were analyzed using color intensity (CI) and hue angle (CH), which are important visual quality indicators in fruit wines. Mulberry wine exhibited the highest CI due to its rich anthocyanin and polyphenol content. In contrast, rose wine, derived primarily from rose petals, showed minimal pigmentation. The rose–mulberry wine displayed intermediate CI values, reflecting the dilution and potential partial degradation of color compounds due to the presence of rose petals and possible oxidative interactions during co-fermentation. Interestingly, the rose–mulberry wine had the highest hue angle (CH = 0.796), indicating a shift toward a more orange or brown hue, possibly due to the formation of polymeric pigments or oxidation products of anthocyanins during fermentation and storage.[33]

Together, these physicochemical results suggest that co-fermentation not only altered sugar and acid dynamics but also contributed to color modulation through both chemical transformation and matrix effects. These changes improve both the sensory and nutritional quality of the final rose–mulberry wine.

3.2 Nutritional and antioxidant functionality of different types of wines

The softness index (SI) serves as a critical parameter for evaluating wine quality. According to Li *et al.*[25] and Lu *et al.*,[34] wines

Table 2 Physicochemical characterization of different wines[a]

Physicochemical parameter	Mulberry wine	Rose wine	Rose–mulberry wine
TAC/% vol	11.73 ± 0.46[b]	13.87 ± 0.98[a]	13.57 ± 0.55[a]
TS g^{-1} L^{-1}	3.92 ± 0.66[a]	2.74 ± 0.56[b]	2.88 ± 0.25 [ab]
RS g^{-1} L^{-1}	1.26 ± 0.65[a]	0.75 ± 0.12[a]	1.53 ± 0.79[a]
TTA g^{-1} L^{-1}	8.74 ± 0.07[a]	3.58 ± 0.07[c]	4.23 ± 0.15[b]
TVA g^{-1} L^{-1}	0.29 ± 0.08[a]	0.23 ± 0.03[a]	0.30 ± 0.02[a]
CI	1.965 ± 0.007[a]	0.079 ± 0.001[c]	0.630 ± 0.004[b]
CH	0.709 ± 0.015[b]	0.468 ± 0.040[c]	0.796 ± 0.005[a]

[a] Note: TAC, total alcohol content; TS, total sugar; RS, reducing sugar; TTA, total acidity; TVA, total volatile acidity; CI, color intensity; CH, hue angle. Each value is the mean ± standard deviation of triplicate measurements ($n = 3$). Values within the same row that have different letters are significantly different ($P < 0.05$).

with an SI greater than 6 are generally considered to have more favorable flavor profiles and improved mouthfeel. In this study, the SI of mulberry wine was notably low at 0.69, well below the acceptable threshold of 5, indicating an overly sharp or unbalanced taste. Traditionally, enhancing SI typically involves chemical, physical, or biological deacidification techniques, but these methods often increase production costs and may raise food safety concerns.[35] Notably, the rose–mulberry wine achieved a value of 8.16 ± 0.035, comparable to rose wine and significantly higher than mulberry wine ($P < 0.05$). This suggests that co-fermentation with *R. rugosa* petals effectively moderated acidity while enhancing mouthfeel, possibly due to the buffering effects of rose-derived compounds and their interactions with mulberry phenolics. This natural fermentation strategy not only enhances flavor complexity but also offers a cost-effective and clean-label solution to improve wine quality.

Beyond sensory improvements, the nutritional and functional indices presented in Table 3 demonstrate notable changes in the antioxidant properties of the wines. While the total polyphenol content (TPC) and total flavonoids (TFC) in rose–mulberry wine were slightly lower than in mulberry wine, its DPPH radical scavenging capacity and hydroxyl radical scavenging activity (HRSA) were significantly higher ($P < 0.05$). These findings suggest that co-fermentation enhances antioxidant efficiency through mechanisms beyond simple polyphenol content—possibly due to the generation of more bioactive compounds or improved synergistic interactions between phenolic subclasses, tannins, and vitamin C. This effect is consistent with previous reports. For instance, Zhang *et al.*[36] observed that co-fermentation of kiwi wine with *Saccharomyces cerevisiae* and non-*Saccharomyces* strains improved DPPH activity due to enhanced phenolic metabolism and aroma complexity. Similarly, Gui *et al.*[37] reported that *Jerusalem* artichoke fermented with lactic acid bacteria and yeast exhibited increased DPPH and ABTS radical scavenging activities compared to single-strain fermentation, attributing the effect to functional synergism. From a functional perspective, these elevated antioxidant capacities are of nutritional and health significance. Antioxidants such as those present in rose–mulberry wine help neutralize free radicals, reduce oxidative stress, and may mitigate the risk of chronic conditions, including cardiovascular disease, neurodegenerative disorders, and age-related decline.[38] As such, rose–mulberry wine not only exhibits superior flavor and mouthfeel but also shows promise as a functional beverage with potential health benefits, aligning with the growing consumer demand for both sensory enjoyment and wellness support in food and drink products.

3.3 Volatile flavor composition and identification of sensory attributes

3.3.1 Aroma characteristics of different types of wines based on E-nose. The E-nose is a multidimensional sensing technology designed to detect and discriminate volatile compounds based on sensor response patterns. As illustrated in Table 4 and Fig. 1A, rose–mulberry wine exhibited markedly higher response intensities at the W1S, W3C, and W5C sensors compared to both mulberry and rose wines. These sensors are known to respond primarily to aromatic compounds, aromatic

Table 3 Nutritional and functional indices of different wines[a]

Physicochemical parameter	Mulberry wine	Rose wine	Rose–mulberry wine
TFC g^{-1} L^{-1}	0.63 ± 0.04[a]	—	0.12 ± 0.03[b]
TPC g^{-1} L^{-1}	1.38 ± 0.22[a]	0.90 ± 0.18[b]	1.08 ± 0.06 [ab]
Tannins g^{-1} L^{-1}	2.24 ± 0.21[a]	1.49 ± 0.02 [ab]	1.27 ± 0.04[b]
Vitamin C mg^{-1} L^{-1}	2.74 ± 0.08[a]	1.77 ± 0.03[b]	1.60 ± 0.03[b]
SI	0.69 ± 0.19[b]	8.79 ± 0.93[a]	8.16 ± 0.35[a]
DPPH scavenging/μmol Trolox·mL^{-1}	0.575 ± 0.064[c]	0.844 ± 0.001[a]	0.801 ± 0.001 [ab]
HRSA/%	29.91 ± 1.31[c]	78.21 ± 0.24[a]	79.05 ± 0.32 [ab]

[a] Note: TFC, total flavonoids; TPC, total phenolics; SI, softness index; HRSA, hydroxyl radical scavenging activity. Each value is the mean ± standard deviation of triplicate measurements ($n = 3$). The symbol "—" indicates values that were not detected. Values within the same row that have different letters are significantly different ($P < 0.05$).

amines and alkanes, suggesting that co-fermentation increased the production of aroma-contributing volatiles such as esters and terpenoids. Conversely, W5S, W1W, and W2W, which are sensitive to nitrogen oxides, sulfur compounds, and methane, respectively, showed significantly lower response values in rose–mulberry wine than in mulberry wine ($P < 0.05$). The elevated W5S response in mulberry wine aligns with literature indicating that this sensor is particularly sensitive to nitrogen-containing volatiles such as amines and nitroso compounds.[39,40] During fermentation, amino acid degradation and pectin hydrolysis can elevate soluble nitrogen levels, especially in single-fruit fermentations like mulberry wine.[41] The W1W sensor's sensitivity to sulfur-containing volatiles may reflect the catabolism of sulfur-rich amino acids like cysteine, which are more prominent in unmodified mulberry fermentation.[42] It is important to note that sulfide volatiles such as hydrogen sulfide or thiols are often associated with off-odors (*e.g.*, rotten egg), and their reduction in rose–mulberry wine implies a potential sensory improvement through co-fermentation. This compositional shift supports the hypothesis that rose petals either dilute or biochemically modulate precursors responsible for off-flavor formation.

PCA (Fig. 1B) further confirmed the distinct aroma profiles among the wine types. The first two principal components (PC1 and PC2) explained a cumulative variance of 98.5%, suggesting a robust model for differentiating samples.[43] Rose–mulberry wine clustered closely with rose wine, while mulberry wine was clearly separated, indicating a substantial aromatic shift induced by co-fermentation. This suggests that the integration of rose petals reorients the aroma profile of mulberry wine toward a more floral and fruity spectrum, likely due to increased ester and terpene formation. PLS-DA is a statistical method with supervised discriminative patterns, which can effectively help in the visualization of high-dimensional data and the discriminant analysis of potential metabolites related to metabolic changes.[44] As illustrated in Fig. 1C, rose wine and rose–mulberry wine cluster together, showing a significant difference in distribution compared to mulberry wine. This indicates that the introduction of rose resulted in a significant change in the aroma characteristics of mulberry wine. The variable importance in the projection (VIP) scores (Fig. 1D) highlighted W1W,

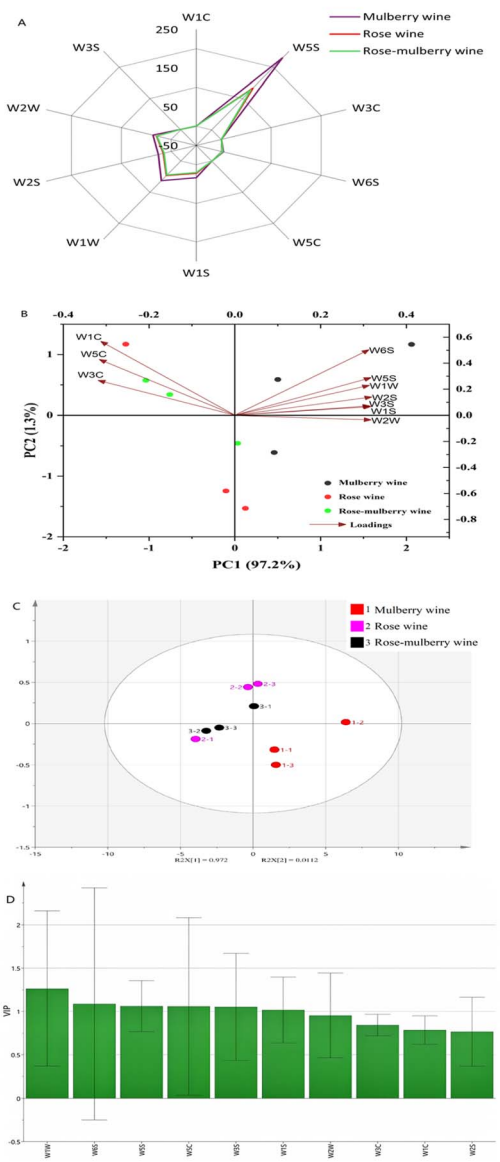


Fig. 1 E-nose analysis of different flavor types of wine. Note: (A), radar fingerprint chart; (B), PCA plots; (C), PLS-DA results; (D), VIP scores, error bars correspond to the standard error ($n = 3$).

Table 4 Aroma characteristics of different types of wine based on E-nose[a]

Sensor name	Mulberry wine	Rose wine	Rose–mulberry wine
W1C	0.175 ± 0.041[a]	0.259 ± 0.066[a]	0.271 ± 0.047[a]
W5S	229.689 ± 50.916[a]	133.586 ± 34.361[b]	127.747 ± 31.872[b]
W3C	0.726 ± 0.035[a]	0.780 ± 0.036[a]	0.788 ± 0.025[a]
W6S	4.757 ± 1.026[a]	3.088 ± 0.509[b]	3.164 ± 0.603[b]
W5C	0.754 ± 0.024[a]	0.791 ± 0.029[a]	0.802 ± 0.019[a]
W1S	33.668 ± 10.867[a]	22.120 ± 6.944[a]	20.160 ± 4.814[a]
W1W	63.257 ± 7.954[a]	46.693 ± 5.889[b]	44.915 ± 3.247[b]
W2S	25.454 ± 7.548[a]	15.391 ± 4.877[a]	14.238 ± 4.348[a]
W2W	36.725 ± 3.644[a]	29.310 ± 3.515[b]	28.507 ± 2.285[b]
W3S	1.272 ± 0.121[a]	1.152 ± 0.072[a]	1.133 ± 0.049[a]

[a] Note: Each value is the mean ± standard deviation of triplicate measurements ($n = 3$). Values within the same row that have different letters are significantly different ($P < 0.05$).

W6S, W5S, W5C, and W3S as key contributors to group discrimination (VIP > 1). These sensors are predominantly responsive to small-molecule volatiles like sulfides, alcohols, and nitrogenous compounds, which reinforces the idea that co-fermentation shifts the chemical equilibrium of mulberry wine away from undesirable sulfur/nitrogen volatiles and toward more favorable aromatic constituents.

In summary, the co-fermentation of mulberry juice with *R. rugosa* petals led to measurable shifts in volatile compound profiles, particularly through the reduction of sulfur- and nitrogen-based compounds and an enhancement of aromatic volatiles. These results provide a mechanistic basis for the improved aroma quality of rose–mulberry wine, emphasizing the role of biochemical modulation and synergistic fermentation pathways in shaping sensory attributes.

3.3.2 Taste characteristics of different types of wine based on E-tongue. The E-tongue employs sensor arrays designed to mimic human gustatory perception, offering objective quantification of basic taste modalities and aftertastes. As shown in Table 5 and Fig. 2A, the co-fermentation of mulberry juice and *R. rugosa* petals significantly altered several taste-related sensory parameter outputs, indicating compositional changes in the wine matrix. Specifically, there was a significant reduction in sweetness and saltiness response values in the rose–mulberry wine compared to monovarietal mulberry wine ($P < 0.05$), suggesting dilution or modification of sugar and ionic content through co-fermentation. Conversely, aftertaste-A (associated with pleasant lingering taste) and astringency increased significantly ($P < 0.05$), while bitterness, aftertaste-B (often related to lingering bitterness), and richness remained below the neutral point, indicating limited perceptual impact. These shifts in taste attributes have practical implications for consumer acceptance. The moderate increase in astringency and aftertaste-A may contribute positively to wine body and complexity, especially for consumers who prefer fuller and more structured wines. However, excessive astringency can be perceived as unpleasant by some drinkers, particularly those unfamiliar with polyphenol-rich wines.[45] The reduction in bitterness and aftertaste-B, which are often associated with negative sensory perceptions, is likely to enhance overall palatability.[46,47] Therefore, the co-fermentation process appears to strike a balance—enhancing sensory depth without introducing strong bitterness, which is generally considered undesirable in fruit wines. Additionally, the saltiness in mulberry wine was significantly higher than that of the other samples. This difference may be attributed to the elevated protein content, which undergoes oxidation and decomposition during the fermentation of mulberry wine, yielding flavor-enhancing amino acids.[48] Although the radar plots show overlapping taste profiles between rose wine and rose–mulberry wine, the latter displays higher values for sourness, saltiness, and aftertaste-A, suggesting a slightly more intense and lingering taste experience. The considerable sensory distance between rose wine and mulberry wine further highlights the moderating effect of co-fermentation on taste profile, potentially appealing to a broader consumer base.

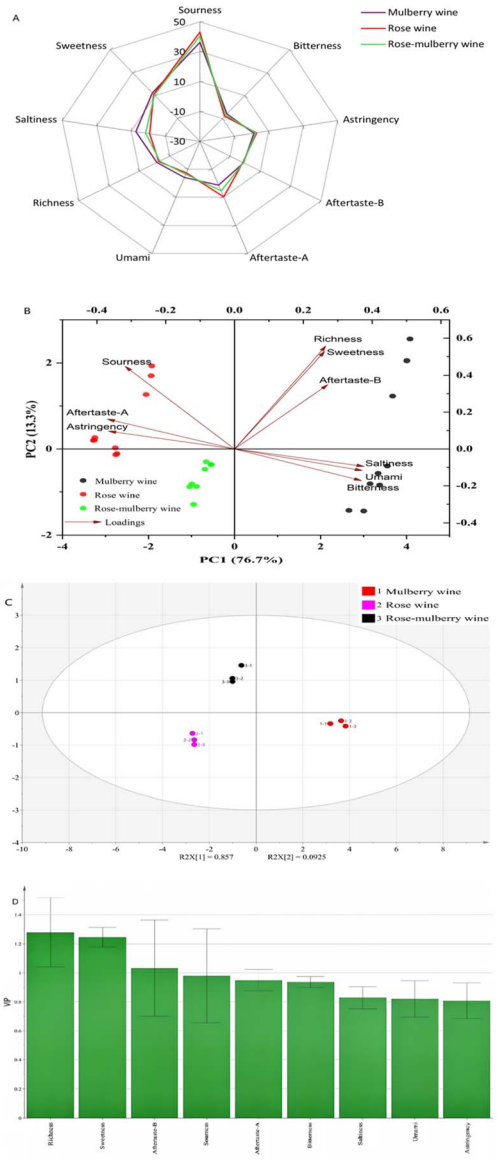


Fig. 2 E-tongue analysis of different flavor types of wine. Note: (A), radar fingerprint chart; (B), PCA plots; (C), PLS-DA results; (D), VIP scores, error bars correspond to the standard error ($n = 3$).

Table 5 Taste characteristics of different types of wine based on E-tongue[a]

Flavor types	Mulberry wine	Rose wine	Rose–mulberry wine
Sourness	36.30 ± 2.24^{c}	43.02 ± 1.33^{a}	40.65 ± 0.46^{b}
Bitterness	-5.96 ± 0.05^{a}	-8.11 ± 0.10^{c}	-7.04 ± 0.05^{b}
Astringency	1.39 ± 0.14^{c}	3.02 ± 0.37^{a}	2.48 ± 0.11^{b}
Aftertaste-B	-1.12 ± 0.27^{a}	-1.51 ± 0.10^{b}	-1.27 ± 0.15^{a}
Aftertaste-A	1.27 ± 0.07^{c}	9.67 ± 0.81^{a}	5.49 ± 0.09^{b}
Umami	-4.05 ± 0.10^{a}	-7.44 ± 0.19^{c}	-6.33 ± 0.05^{b}
Richness	-1.49 ± 1.09^{a}	-2.81 ± 0.40^{b}	-3.43 ± 0.12^{b}
Saltiness	7.29 ± 0.19^{a}	-0.67 ± 0.16^{c}	1.68 ± 0.07^{b}
Sweetness	12.37 ± 0.73^{a}	11.11 ± 0.95^{b}	10.53 ± 0.24^{b}

[a] Note: Each value is the mean ± standard deviation of triplicate measurements ($n = 3$). Values within the same row that have different letters are significantly different ($P < 0.05$).

The PCA results (Fig. 2B) revealed that PC1 and PC2 explained 76.7% and 13.3% of the variance, respectively, totaling 90.0%.

The spatial separation of the three wine types demonstrated effective discrimination based on taste. Mulberry wine was positioned separately, driven by its stronger umami, saltiness, and bitterness signals, while rose–mulberry wine clustered closer to rose wine, supporting the moderating effect of co-fermentation. The PLS-DA results (Fig. 2C) further confirmed that the inclusion of rose petals led to a distinct flavor transformation of mulberry wine. The rose–mulberry wine cluster shifted away from the mulberry wine group, illustrating a significant modification in its gustatory fingerprint. VIP analysis (Fig. 2D) highlighted that richness, sweetness, and aftertaste-A had VIP scores > 1, identifying them as key discriminators among the wines. This suggests these parameters were most influenced by the co-fermentation process and contributed significantly to the sensory differentiation of rose–mulberry wine.

Taken together, these results underscore that the co-fermentation process not only adjusted individual taste components but also orchestrated a multi-faceted shift in the sensory profile, leading to improved balance, complexity, and potentially broader consumer acceptability.

3.3.3 Combination of E-nose and E-tongue. Flavor perception arises from the integrated simulation of both olfactory (aroma) and gustatory (taste) receptors. Therefore, the combined use of an E-nose and E-tongue provides a comprehensive profile of wine flavor, capturing both volatile compounds and non-volatile taste components. As shown in Fig. 3, the PCA combining both E-nose and E-tongue sensor outputs explained 79.5% (PC1) and 13.5% (PC2) of the total variance, for a cumulative explanation of 93.0%. This high explanatory power indicates robust differentiation among the three wine types based on their flavor characteristics. Specifically, mulberry wine was strongly associated with bitterness, saltiness, and umami, aligning with its higher levels of total acidity and tannins, as discussed in Section 3.1. Rose wine showed strong associations with sourness, astringency, and aftertaste-A, reflecting the influence of rose petal-derived phenolics and organic acids. Rose–mulberry wine was positioned closer to volatile-sensitive sensors W1C, W3C, and W5C, which are responsive to aromatic hydrocarbons, esters, and aldehydes, respectively. This indicates that co-fermentation introduced or enhanced specific aroma compounds that are not prominent in either monovarietal wine alone.

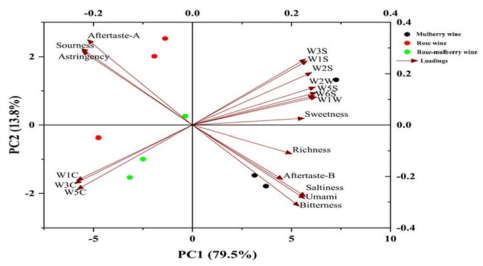


Fig. 3 Results of PCA of different types of wine. Note: the length of the arrow represents the intensity of the impact on the flavor and taste of the wine. The longer arrow signifies the greater impact. The angle between the arrows indicates a positive or negative correlation: an acute angle represents a positive correlation, while an obtuse angle represents a negative correlation.

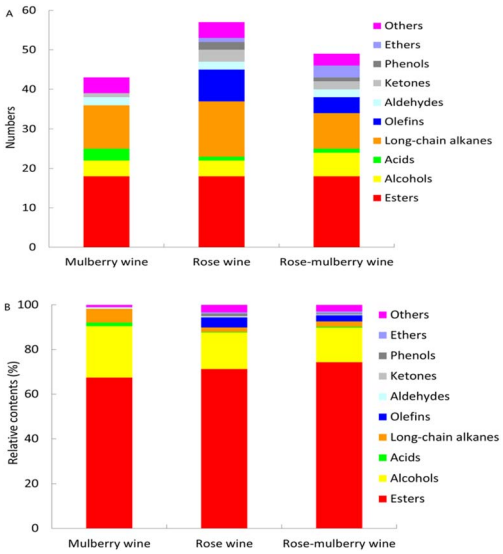


Fig. 4 Changes in quantities (A) and relative contents (B) of volatile compounds in different types of wine.

3.4 The analysis of volatile compounds in three wines

Volatile flavor compounds are essential parameters for assessing the quality of fruit wine and critical factors in determining consumer acceptance. Utilizing HS-SPME-GS-MS analysis, all the volatile compounds in wine were detected and classified. A total of 98 volatile compounds were detected and identified in the wine samples, comprising 30 esters, 9 alcohols, 4 acids, 21 long-chain alkanes, 8 olefins, 6 aldehydes, 6 ketones, 2 phenols, 3 ethers, and 9 others (Fig. 4A). The aroma profile of single-ingredient mulberry wine was determined to be less complex than that of rose–mulberry wine and rose wine. The diversity of volatile compounds was ranked in the following order: rose wine > rose–mulberry wine > mulberry wine, corresponding to 57, 49, and 43, respectively. There were 17 flavor substances common to the three wines, accounting for 17.35% of the total volatile compounds. Notably, 31 volatile compounds were common to both rose–mulberry wine and rose wine, mainly concentrated in esters, alcohols, and hydrocarbons (Fig. 5A). The number and relative content in the three wines (Fig. 4B and 5B) exhibited no significant differences. This observation is consistent with the E-nose, as esters are known to be key contributors to fruity aromas in wine. Their relatively stable concentrations across samples likely explain the consistent response observed from the W1C sensor, which is sensitive to aromatic compounds such as esters. In contrast, the decreased levels of acids and alcohols, which have been previously reported to be associated with off-flavors in citrus-based products, may explain the lower responses recorded by the W5S and W1W sensors in the rose–mulberry wine.[49]

3.5 Multivariate statistical analysis of volatile flavor compounds

The PCA results, illustrated in Fig. 6A, revealed close associations among 98 aroma-active compounds. The contribution rate of PC1 was 61.1%, and that of PC2 was 38.9%. Together, these rates represent the characteristics of flavor substances. PC1

exhibited positive correlations with isoamyl acetate (A1), ethyl heptanoate (A7), ethyl octanoate (A10), and ethyl decanoate (A22), while demonstrating negative correlations with eicosamethylcyclododecasiloxane (D21), 2,6,10,14-tetramethylhexadecane (D17), and octadecamethylcycloonasiloxane (D19). Conversely, PC2 showed positive correlations with 2,6,10,14-tetramethylpentadecane (D15), ethyl tetradecanoate (A28), ethyl hexadecanoate (A30), and ethyl dodecanoate (A25), showing negative correlations with (S)-(−)-citronellic acid, methyl ester (A16), and ethoxydi(*tert*-butyl)silane (D4).

In the PCA plot, mulberry wine, rose wine, and rose–mulberry wine were located in the second, first, and fourth quadrants, respectively, indicating clear differentiation based on their aromatic compositions. Both rose wine and rose–mulberry wine displayed strong positive correlations with PC1, while mulberry wine exhibited a strong negative correlation. Conversely, mulberry and rose wines were positively associated with PC2, whereas rose–mulberry wine showed a negative correlation along this axis.

Rose–mulberry wine was located in the positive region of PC1 and the negative region of PC2, aligning with volatile compounds such as isoamyl acetate (A1), ethyl 9-decenoate (A21), ethyl octanoate (A10), and ethyl decanoate (A22). These results indicate that esters and alcohols dominate the aromatic profile of rose–mulberry wine, highlighting the enhancing effect of rose petal incorporation on the fragrance complexity of mulberry wine. Notably, ethyl hexanoate (A2), known for its distinctive green apple and brandy aroma notes, was strongly loaded on both PC1 and PC2, further emphasizing its contribution to overall aroma perception.[20]

A hierarchical cluster heatmap (Fig. 6B) was generated to visualize the differences in the relative contents of volatile components among the three wine types. Color intensity differences in the heatmap reflect variations in compound abundance across the samples. The clustering results revealed two major groups: the first comprising mulberry wine, characterized by long-chain alkanes and a distinct ester profile; and the second group including rose wine and rose–mulberry wine, both dominated by a broader spectrum of esters. These findings

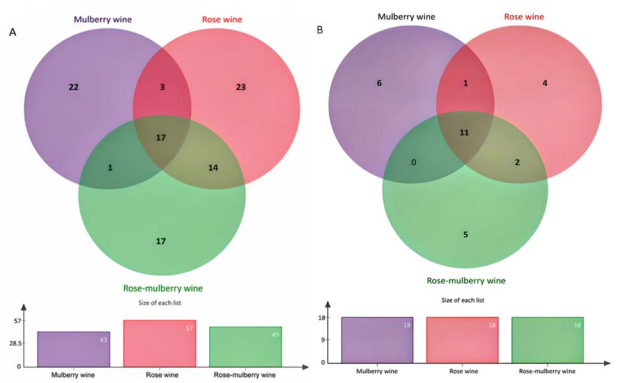


Fig. 5 Venn diagrams of the number of volatile compounds in different types of wine. Note: (A) represents the shared characteristics of all volatile compounds in the samples, while (B) represents the shared characteristics of all ester compounds in the samples.

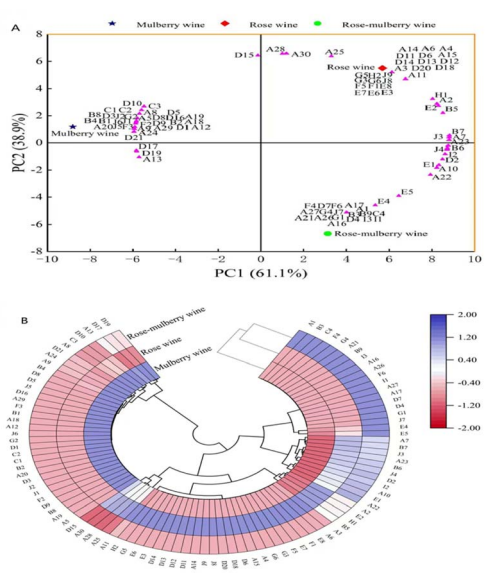


Fig. 6 (A) PCA model based on volatile compounds in different types of wine. (B) Cluster heatmap of volatile compounds in different types of wine.

suggest that rose–mulberry wine possesses a richer and more diverse volatile profile than mulberry wine alone, with an aroma profile more closely aligned with that of rose wine. The enhanced abundance and variety of esters in the co-fermented wine underscore the synergistic effect of rose petal addition in enriching the aromatic complexity of mulberry wine.

The VIP value serves as a quantitative indicator that reflects the relative contribution of each volatile compound to the overall flavor profile of wine. Compounds with higher VIP values represent greater disparities between sample groups and are considered potential marker compounds for distinguishing different wine types. Using a threshold of VIP values > 1, a total of 18 differential compounds were identified, as shown in Table 6. These compounds contributed most significantly to the aromatic differentiation among the three types of fruit wines. Notably, esters comprised the most abundant class of these discriminant compounds. Among them, isoamyl acetate (A1) and ethoxydi(*tert*-butyl)silane (D4) were only detected in rose–mulberry wine. 3,7-dimethyl-oct-6-enoic acid, ethyl ester (A14) was only detected in rose wine. Ethyl 3-phenylpropionate (A18), 2,7-dimethyl-4,5-octanediol (B2), phosphonoacetic acid, 3TMS derivative (C1), and 1-[(2-hydroxyphenyl)thioxomethyl] pyrrolidine (D1) were only detected in mulberry wine.

The high content of esters is one of the most important characteristics of the compounds in rose–mulberry wine. Specially, isoamyl acetate is primarily responsible for banana- and apple-like aromas and plays a key role in enhancing fruity notes. However, isoamyl acetate was not detected in mulberry wine and rose wine. This is consistent with the findings of Ding *et al.*[7]

The relative contents of flavor compounds in the three wines, ranked from highest to lowest, were as follows: esters, alcohols, long-chain alkanes, olefins, acids, aldehydes, and ketones. The

Table 6 Analysis of aroma compounds distinctive to different types of wine[a]

No.	Compound	Odor description	VIP	Relative contents/%		
				Mulberry wine	Rose wine	Rose–mulberry wine
A25	Ethyl dodecanoate	Leaf	1.448	1.127	1.501	0.852
A14	3,7-Dimethyl-oct-6-enoic acid, ethyl ester	—	1.445	0	0.527	0
A30	Ethyl hexadecanoate	Wax	1.411	0.410	0.542	0.137
A28	Ethyl tetradecanoate	Ether	1.404	0.375	0.538	0
D15	2,6,10,14-Tetramethylpentadecane	—	1.348	0.072	0.085	0
A11	Ethyl phenylacetate	Fruit, sweet	1.297	0.811	2.756	1.062
E4	1-Methyl-4-(1-methylethylidene)cyclohexene	Pine, plastic	1.163	0	0.301	1.422
H1	Methyl eugenol	Clove, spice	1.125	0	1.037	0.421
A2	Ethyl hexanoate	Apple peel, fruit	1.083	6.437	7.814	7.076
D4	Ethoxydi(*tert*-butyl)silane	—	1.080	0	0	0.809
E2	1-Methyl-3-(1-methylethenyl)cyclohexene(±)	—	1.066	0	0.969	0.472
A1	Isoamyl acetate	Banana, apple	1.056	0	0	0.325
A18	Ethyl 3-phenylpropionate	Flower	1.056	0.536	0	0
B2	2,7-Dimethyl-4,5-octanediol	—	1.056	0.474	0	0
C1	Phosphonoacetic acid, 3TMS derivative	—	1.056	0.364	0	0
D1	1-[(2-Hydroxyphenyl)thioxomethyl]pyrrolidine	—	1.056	0.326	0	0
E5	2,6-Dimethylocta-2,6-diene	—	1.044	0	0.255	0.664
B5	Terpineol	—	1.009	0	0.840	0.475

[a] Note: Odor descriptions are sourced from the flavornet database (http://www.flavornet.org), "—" means that the odor description of the substance was not found.

composition and relative proportions of these compounds play a critical role in defining the distinctive character of each wine.

Esters play a significant role as aromatic constituents in wine, predominantly responsible for floral and fruity notes that significantly shape the overall sensory profile. These compounds are mainly produced during yeast-mediated alcoholic fermentation, as well as through the enzymatic esterification involving higher alcohols and fatty acids. The aroma of mulberry wine is not influenced by the concentration of individual esters, but also by the combined synergistic effects of multiple ester compounds. As illustrated in Fig. 3 and Table 6, esters were the dominant class of volatiles across all three wine samples, both in terms of compound number and relative abundance. The key esters identified included diethyl butanedioate, ethyl octanoate, ethyl decanoate, and ethyl hexanoate. Notably, rose–mulberry wine exhibited the highest total ester content, accounting for 74.346% of its volatile profile (Table S1). Compared to the monovarietal wines, rose–mulberry wine showed a marked increase in the relative concentrations of ethyl octanoate (38.75%) and ethyl decanoate (13.90%), both of which contribute significantly to fatty, fruity notes.[50] Conversely, the relative level of diethyl butanedioate, while still present, was reduced in rose–mulberry wine. These shifts in ester composition reflect the impact of co-fermentation on modulating key aroma contributors, thereby enriching the wine's aromatic complexity. In terms of diversity, 11 esters were common across all three wines. Rose–mulberry wine shared 11 esters with mulberry wine and 13 with rose wine, indicating that co-fermentation not only preserved core aromatic constituents but also introduced additional floral and fruity esters likely derived from the *R. rugosa* petals. Six new esters were identified in the rose–mulberry wine, including isoamyl acetate, ethyl 9-decenoate, ethyl undecanoate, and others. These compounds are known to play important roles in shaping the fruity and floral aroma profiles of fermented beverages. For instance, isoamyl acetate is commonly associated with a strong banana-like aroma and contributes to the perception of sweetness and fruitiness in wine.[51] Ethyl 9-decenoate imparts citrus and waxy notes, while ethyl undecanoate is linked to coconut- and woody-like nuances.[52] Collectively, these esters enhance the aromatic complexity and perceived freshness of the wine. Typically, esters are formed through enzymatic or acid-catalyzed esterification reactions between carboxylic acids and alcohols during fermentation.[40] Moreover, non-enzymatic acid-catalyzed esterification may also occur under the low pH conditions of wine fermentation. The co-fermentation of mulberry juice with *R. rugosa* petals likely introduces additional phenolic acids, fatty acids, or higher alcohols, which may serve as precursors or modulators of enzymatic activity.[53] The presence of these newly identified esters suggests that the co-fermentation of mulberry juice with *R. rugosa* petals not only introduces new precursors but may also influence enzymatic activity, leading to the formation of unique volatile compounds. However, the E-nose results did not fully capture the changes in aroma attributes, which may be attributed to alterations in the relative proportions of other volatile compounds that mask or counterbalance the impact of the newly formed esters.

Alcohols are secondary products generated by yeast metabolism during the wine fermentation process, and they are also crucial components of volatile aroma. The relative contents of alcohols in the three wines were 23.635%, 16.375%, and 15.384%, respectively. Although 2-phenylethyl alcohol, which had the highest relative content in mulberry wine (21.61%) and was characterized by rose and honey notes, its presence

remained substantial in rose–mulberry wine (11.80%). Notably, the quantities of alcohol substances in rose–mulberry wine were higher than those in mulberry wine and rose wine. Terpenoid alcohols such as citronellol, terpineol, and L-α-terpineol, which were not detected in mulberry wine, were introduced into the rose–mulberry wine through co-fermentation. These compounds are associated with citrus, floral, and woody notes, and are well-documented contributors to wine aroma complexity and perceived freshness.[49]

Additionally, a total of 20 long-chain alkanes and 8 olefins were identified. Rose wine contained 14 long-chain alkanes and 8 olefins, whereas mulberry wine exhibited the lowest diversity of hydrocarbons, with no olefins detected. Although the relative contents of these hydrocarbons were generally low, with only a few compounds such as decamethylcyclopentasiloxane, octadecamethylcycloonasiloxane, (+)-2-carene, and 1-methyl-4-(1-methylethylene)cyclohexene exceeding 1%, their presence reflects contributions from fatty acid degradation and floral precursors. The long-chain alkanes are primarily derived from the cleavage of long-chain fatty acid alkoxy radicals, but due to their high odor thresholds, their direct impact on wine aroma is minimal.[54] In contrast, olefins such as D-limonene and 1-methyl-4-(1-methylethylidene) cyclohexene, known for their citrus and pine-like aromas, were identified exclusively in rose and rose–mulberry wines. Their formation is likely attributed to terpenoid precursors from *R. rugosa* petals introduced during co-fermentation. These compounds contribute to the bright, refreshing top notes of the wine, enhancing aromatic lift and freshness.[55]

Although phenols, ketones, ethers, furans, and pyrans were present in relatively low concentrations, co-fermentation clearly increased the diversity of these minor volatile classes in rose–mulberry wine. Compared to mulberry wine, rose–mulberry wine contained 2 additional ketones, 1 phenol, 2 ethers, 1 furan, and 2 pyrans, highlighting the complementary biochemical interactions between the two substrates. While these compounds are individually subtle, their synergistic interactions can enhance the depth, roundness, and persistence of aroma.

Taken together, the distinctive flavor profile of rose–mulberry wine can be attributed to the integration of floral-derived terpenoids and olefins, the modulation of key esters and alcohols, and the enrichment of minor aroma-active compounds through co-fermentation. These compositional changes not only differentiate rose–mulberry wine from its monovarietal counterparts, but also reflect the aromatic synergy achieved by combining mulberry fruit and rose petals. This supports growing evidence that botanical co-fermentation is an effective strategy to diversify flavor and enhance consumer appeal in functional fruit wines.[20,22]

4 Conclusions

This study systematically evaluated the effects of co-fermentation of mulberry fruit and rose petals on the nutritional quality, antioxidant capacity, and volatile flavor profile of mulberry wine using integrated analytical tools, including E-nose, E-tongue, HS-SPME-GS-MS, and *in vitro* antioxidant assays. The results demonstrated that rose–mulberry wine exhibited significantly higher hydroxyl radical scavenging activity (HRSA) and hue angle (CH) compared to both mulberry and rose wines, indicating improved antioxidant potential and color characteristics. Flavor and aroma analysis showed that co-fermentation with rose petals enriched the sensory complexity of mulberry wine. E-nose and E-tongue data confirmed a reduction in undesirable odor compounds (*e.g.*, nitrogen oxides, sulfides, methane) and an increase in favorable sensory attributes such as astringency and aftertaste-A, alongside reduced sweetness and saltiness—contributing to a more balanced and structured flavor profile. Multivariate analysis (PCA and PLS-DA) confirmed distinct separations among wine types and highlighted the substantial modification of volatile compound composition due to co-fermentation. Rose–mulberry wine contained six additional aroma-active compounds and significantly higher levels of esters (6.88% more than mulberry wine and 3.13% more than rose wine), including floral and fruity esters such as ethyl decanoate and ethyl hexanoate, and unique volatiles like isoamyl acetate and ethyl undecanoate that contributed banana, apple, lemon, woody, and frankincense-like notes.

Despite these promising findings, several limitations should be acknowledged. First, this study focused solely on *in vitro* antioxidant activity, which may not fully reflect *in vivo* bioavailability or health effects. Second, sensory data were obtained *via* intelligent sensing instruments, which, while objective and reproducible, may not entirely capture human sensory perceptions and preferences. Third, only one ratio of mulberry-to-rose substrate was tested; variations in formulation could yield different outcomes. Therefore, future research should explore: (1) *in vivo* studies to validate the functional health benefits of rose–mulberry wine; (2) consumer sensory evaluations with human preferences. Overall, this study demonstrates that natural co-fermentation with *R. rugosa* petals is a viable, clean-label strategy to enhance the antioxidant potential and sensory appeal of mulberry wine. The approach holds substantial promise for developing novel, functional, fruit-flower-based fermented beverages with differentiated market value and consumer appeal.

Author contributions

Conceptualization, H. Y. and X. W.; methodology, H. Y. and X. W.; investigation, J. Y. and J. P.; resources, Q. Y. and W. X.; data curation, J. Y.; writing – original draft preparation, X. W.; writing – review and editing, X. W., Q. Y., J. P., W. X., J. W., J. Y., M. H., D. R., and H. Y.; funding acquisition, H. Y. and X. W. All authors have read and agreed to the published version of the manuscript.

Conflicts of interest

The authors declare no conflicts of interest in this work.

Data availability

The data supporting this article have been included as part of the supplementary information (SI). Supplementary information: Table S1. See DOI: https://doi.org/10.1039/d5ra05062f.

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