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Full Length Research Paper

Analysis of Non-Anthocyanin Phenolic Profiles in *Vitis Amurensis* Wines and Their Hybrids

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The non-anthocyanin phenolic compounds in wines from five grape cultivars (Zuo Shan Yi, Zuo Shan Er, Shuang Hong, Shuang You and Shuang Feng) of *VITIS AMURENSIS* and two hybrid cultivars (Zuo Hong Yi and Zuo You Hong) were examined by high-performance liquid chromatography/electrospray ionization tandem mass spectrometry (HPLC/ESI-MS/MS) technique in this study. The non-anthocyanin phenolic compounds detected from 7 grape cultivars included 5 benzoic acids, 7 cinnamic acids, 5 flavan-3-ols, 9 flavonols and 4 stilbenes. The detected benzoic acids were gallic acid, protocatechuic acid, syringic acid, ethyl gallate, and ellagic acid. The cinnamic acids were *TRANS*-cinnamic acid, *TRANS*-caftaric acid, *TRANS*-cutaric acid, ferulic acid, *TRANS*-fertaric acid, caffeic acid and ethyl caffeic acid. The flavan-3-ols were procyanidin B1, catechin, procyanidin B2, epicatechin, and trimeric procyanidin C1. The flavonols were dihydroquercetin, myricetin-3-glucoside, quercetin-3-galactoside, quercetin-3-glucuronide, quercetin-3-glucoside, myricetin, quercetin, naringenin and leutolin. The stilbenes were *TRANS*-piceid, *CIS*-piceid, *TRANS*-resveratrol and *CIS*-resveratrol. Principal components analysis showed that the non-anthocyanin phenolic compounds in wines were mainly composed of cinnamic acids, flavan-3-ols and benzoic acids. In addition, systematic cluster analysis suggested that the non-anthocyanin phenolic compound profiles were helpful for the classification of these cultivars of *V. AMURENSIS* and the hybrids.

Key words: Wine, non-anthocyanin phenolic compounds, high-performance liquid chromatography/electrospray ionization tandem mass spectrometry.

INTRODUCTION

The phenolic compounds in grape berries are responsible for organoleptic properties of grape berries and wine, such as color, bitterness, astringency, clarity, stability and aroma (Picinelli et al., 2000; Gawel, 2000; Minussi et al., 2003; Delgado et al., 2004; Pérez-Magariño and González-Sanjosé, 2006). In the processes of the red wine production, phenolic compounds from the skins of red grapes are transferred to the must during the fermentation of any maceration steps (Salas et al., 2003). Different types of phenolic compounds endow grape varieties and wines with specific quality characteristics (Jin et al., 2009).

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Non-anthocyanin phenolic compounds include benzoic and cinnamic acids, flavan-3-ols, flavonols, stilbenes except anthocyanins. These substances in the wine fermentation and aging process changes are extremely complex and have a pronounced influence on the quality of the wine (Hunter et al., 1991; Silvia and Luisa, 2005; Monagas et al., 2006). The grape berry includes the massive non-anthocyanin phenolic compounds, mainly distributed in the skin, the seed and the stem. In the winemaking process, these substances were dipped into wine. The wine's colour, flavour and mouth feel have important implications and constitute grape wine 'skeleton ingredient'. As hot spot, non-anthocyanin phe-nolic compounds have received great concern for many years in the study of domestic and foreign grapes and grape wine. Researches in the non-anthocyanin phenolic compounds of *Vitis vinifera* L. wine grapes of varieties

and contents have been reported (Jin et al., 2009; Mané et al., 2007; Iacopini et al., 2008; Yilmaz and Toledo, 2004).

Vitis amurensis is native to the North-eastern China, which is resistant to low temperature down to -40°C. These grapes wines have unusual color, aroma and taste, quite different from the wine made from grape of *V. vinifera*. We have reported the anthocyanins profile of grape berries from *V. amurensis* and so many wines (Zhao et al., 2010). However, studies on the non-anthocyanin phenolic compounds of *V. amurensis* and hybrid cultivars wines varieties are not available

The main aim of this study was to investigate the non-anthocyanin phenolic compounds in grape varieties and corresponding wines from five grape cultivars (Zuo Shan Yi, Zuo Shan Er, Shuang Hong, Shuang You and Shuang Feng) of *V. amurensis* and two hybrid cultivars (Zuo Hong Yi and Zuo You Hong). The non-anthocyanin phenolic compound profiles were detected by high-performance liquid chromatography/electrospray ioniza-tion tandem mass spectrometry (HPLC/ESI-MS/MS) technique in order to examine the differences of non-anthocyanin phenolic compounds in composition and content among the grape wines. The results of this study will provide the theoretical supports for grape wines processing and grape varieties breeding.

MATERIALS AND METHODS

Analytical standards and reagents

The standard samples of catechin, quercetin, gallic aid, cafteic acid, and resveratrol were all purchased from Sigma Company (Iowa, USA) . HPLC grade methanol, acetonitrile and glacial acetic acid were obtained from Fisher Company (Fairlawn, NJ, USA). Deionized water (<18 M resistance) was obtained from a Milli-Q element water purification system (Millipore, Bedford, MA, USA).

Sampling

The samples of grape berries of seven cultivars were collected from the Institute of Special Wild Economic Animal and Plant, Chinese Academy of Agricultural Sciences in 2008, including Shuang Feng, intraspecific hybrid of *V. amurensis*, hermaphroditic; Shuang Hong, intraspecific hybrid of *V. amurensis*, hermaphroditic; Shuang You, hybrid of *V. amurensis*, hermaphroditic; Zuo Shan Yi, hybrid of *V. amurensis*, female, selected from uncultivated resources; Zuo Shan Er, hybrid of *V. amurensis*, female, selected from uncultivated resources; Zuo Hong Yi, interspecific hybrid, *V. amurensis* × Мускат Розавый × *V. amurensis*, hermaphroditic; Zuo You Hong, inter-specific hybrid, *V. amurensis* × Мускат Розавый × *V. amurensis*, hermaphroditic, cultivated. All these cultivars were planted in the area with similar soil characteristics and climatic constraints. In addition, these cultivars were subjected to the same management practice, such as irrigation, fertilization, soil management, disease control and pruning. The grape berries were harvested at technological ripeness, determined on the basis of former years ripening dates and as judged from seed color change to dark brown without senescence of berry tissue.

Grape berries were processed in the 2008 vintage. For all seven wine samples, the maceration and fermentation were carried out in a small glass container (10 L). To each must, SO_2 (50 mg/L) was added before alcohol fermentation, and then the activated yeast was added in the next day. The temperature of alcohol fermentation ranged from 16 to 26°C. After alcohol fermentation, maceration and malolactic fermentation (MBR B1, Lallemand S. A., France, 0.02 g/L, temperature kept about 18°C) took place, and SO_2 (50 mg/L) was added at the end of malolactic fermentation. All the experiments were carried out in replicates, and each sample was independently analyzed twice. The wine samples were determined using HPLC-ESI-MS/MS with direct injection after filtration.

Extraction and analysis of non-anthocaynins

The extraction of non-anthocyanins was performed following Garcia-Viguera and Bridle (1995) and modified. 100 ml wine samples and 100 ml distilled water were added into 100 ml volumetric flask, respectively with sequential extraction of 3 times using 80 ml ethyl acetate. Wine samples were separated from the separating funnel in the replicates of three times to collect the extracts. The extraction was concentrated under vacuum at 30°C using a rotary evaporator until dryness. The dry extraction was resolved in 5 ml solvent of methanol. About 1 ml of extracted solution was strained through a 0.45 m millipore filter for HPLC-ESI-MS/MS analysis.

An Agilent 1200 series HPLC-MSD trap VL instrument, equipped with a variable wavelength detector (VWD) and a reverse phase column (Zorbax SB-C18 column 250 × 4.6 mm, 5 m), was used for separation of non-anthocyanin phenolic compounds. Compounds were gradient eluted using two solutions as follows: (A) aqueous 1% acetic acid, and (B) Methanol containing 1% acetic acid. Elution program was as follows: 0 to 15 min, 10 to 26%B; 15 to 30 min, 26 to 40%B; 30 to 50 min, 40 to 65%B; 50 to 60 min, 65 to 95%B; 60 to 63 min, 95 to 10%B; 63 to 66 min, 10%B isocratic, at a flow rate of 1.0 ml/min. Injection volumes were 10 l, and the detection wavelength was 280 nm. Mass spectroscopy (MS) conditions were as follows: Electrospray ionization (ESI) interface, negative ion model, 30 psi nebulizer pressure, 10 L/min dry gas flow rate, 325°C dry gas temperature, and scans at *m/z* 100 to 1500. All analyses were replicated twice.

Statistical analysis

Benzoic acids, cinnamic acids, flavan-3- ols, flavonols and stilbenes were quantified and expressed as gallic acid, caffeic acid, catechin, quercetin and resveratrol content, respectively from the chromatographic results. Eight concentration level of 5 of the mixed mark was made. Each level was repeated three times. The average of each component peak area (area) on the concentration (amt, mg/L) created a standard curve in the workstation. Non-anthocyanin phenolic compounds standard samples, linear equation and correlation coefficient are shown Table 1. The results indicate that correlation coefficient of five standard non-anthocyanin phenolic compounds was above 0.998, and that linear goodness could satisfy the needs of the quantification. If any of these non-anthocyanins remained undetected in a sample, they were represented by zero in the data matrix for principal component analysis (PCA). PCA was performed with the statistical software SPSS 18.0 (USA).

RESULTS AND DISCUSSION

Qualitative analysis of non-anthocyanin phenolic compounds in grape wines

Currently, the non-anthocyanin phenolic compounds

Table 1. Linear equation and correlation coefficient of five non-anthocyanin phenolic compounds.

Standard sample	Linear equation	Correlation coefficient
Gallic acid	Area = 6.04767*Amt + 37.6951	0.99837
Caffeic acid	Area = 6.94488*Amt + 24.3721	0.99875
Catechin	Area = 1.15779*Amt + 4.85762	0.99878
Quercetin	Area = 3.230747*Amt + 9.9427	0.99885
Resveratrol	Area = 9.97568*Amt + 17.0442	0.99876

Table 2. The identification of non-anthocyanin phenolic compounds in wines by HPLC-ESI-MS/MS.

Peak number	RT	Molecular and product ion (*M/Z*)	Non-anthocyanin phenolic compound
1	0.72	169	Gallic acid
2	0.89	147	*trans*-Cinnamic acid
3	1.32	153	Protocatechuic acid
4	1.51	311(179,149)	*trans*-Caftaric acid
5	2.47	295(163)	*trans*-Cutaric acid
6	3.11	577(425,289)	Procyanidin B1
7	3.45	289(245)	Catechin
8	3.53	193	Ferulic acid
9	3.74	325(193)	*trans*-Fertaric acid
10	3.94	179(135)	Caffeic acid
11	4.81	197	Syringic acid
12	6.01	577(425.289)	B2 Procyanidin B2
13	6.28	197(169)	Ethyl gallate
14	7.03	289(245)	Epicatechin
15	9.78	865(577)	Trimeric Procyanidin C1
16	10.49	303	Dihydroquercetin
17	10.87	479(317)	Myricetin-3-glucoside
18	11.59	389(227)	*trans*-Piceid
19	12.83	301	Ellagic acid
20	13.46	463(301)	Quercetin-3-galactoside
21	13.63	477(301)	Quercetin-3-glucuronide
22	13.94	463(301)	Quercetin-3-glucoside
23	15.72	317	Myricetin
24	15.91	389(227)	*cis*-Piceid
25	17.52	207	Ethyl caffeic acid
26	19.32	301(179)	Quercetin
27	20.91	227	*trans*-Resveratrol
28	22.08	271	Naringenin
29	22.87	285	Leutolin
30	23.82	227	*cis*-Resveratrol

research in wines by HPLC-ESI-MS/MS relies on its molecular and product ions to complete, to retention time, elution order and spectral information in order to be able to accurately identify the polyphenols structure. The 30 non-anthocyanin phenolic compounds in these seven wines made from the *V. amurensis* and its hybrids are presented in Table 2 and Figure 1.

From Table 2 and Figure 1, 30 non-anthocyanin phenolic compounds were detected from 7 grape cultivars wines and significant differences were found among the cultivars. Zuo You Hong comprised 25 phenolic compounds detected in all wines; and 21 in the Shuang Hong and

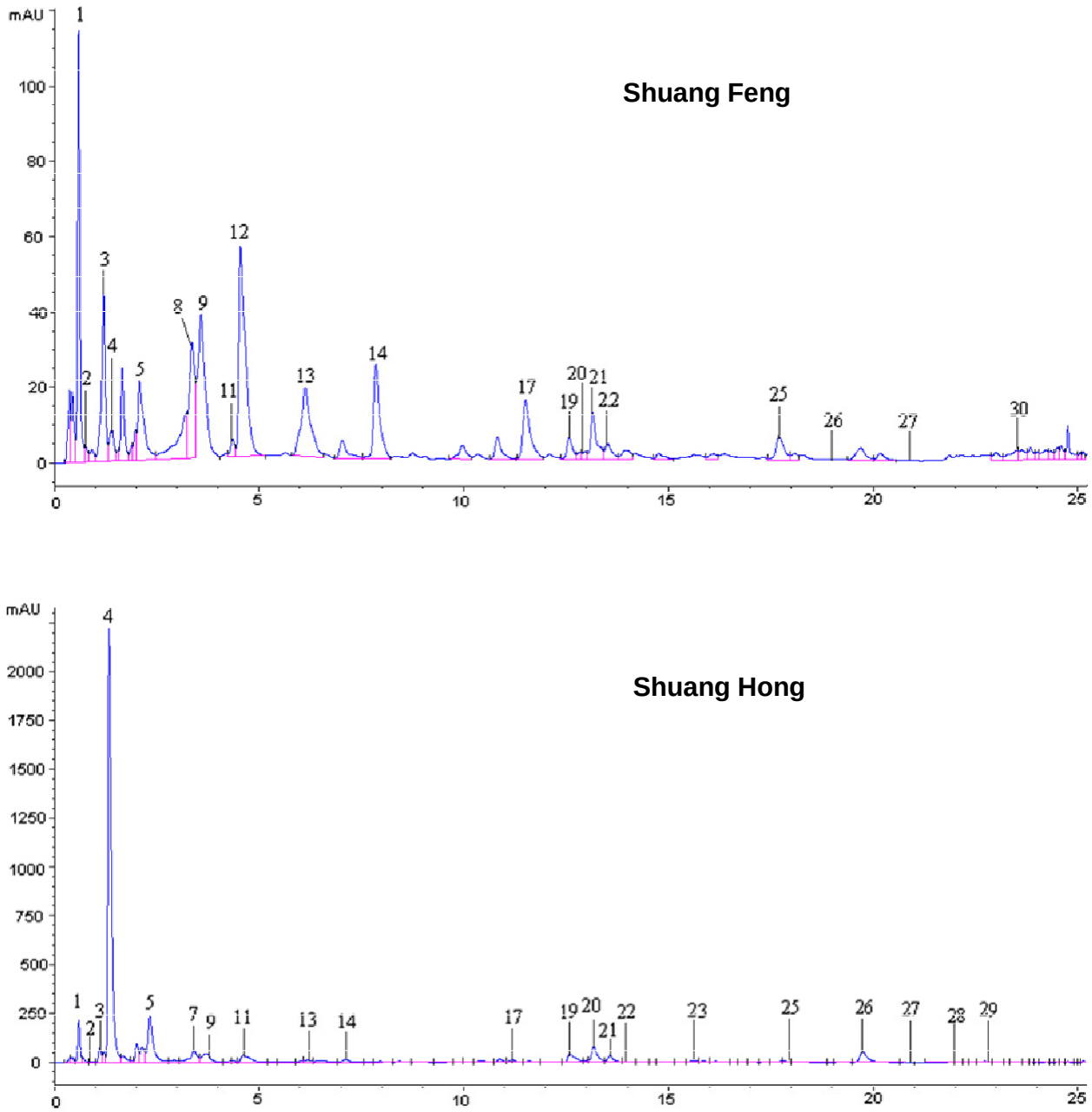


Figure 1. The chromatograms of non-anthocyanin phenolic compounds in wines.

Zuo Shan Yi wine, 20 in the Shuang Feng and Zuo Hong Yi wine, 19 and 17 in the Zuo Shan Er and Shuang You wine, respectively.

Quantitative analysis of non-anthocyanin phenolic compounds in grape wines

The content of total non-anthocyanin phenolic compounds

As shown in Figure 2, the content of total non-anthocyanin phenolic compounds in these wines ranged from 23.85 to 240.91 mg/L, decreasing in the order: Zuo You Hong (240.91 mg/L) > Shuang Hong (221.97 mg/L) > Zuo Hong Yi (156.97 mg/L) > Zuo Shan Er (87.00 mg/L) > Shuang Feng (61.00 mg/L) > Zuo Shan Yi (43.76 mg/L) > Shuang You (23.85 mg/L).

The content of benzoic acids

Five benzoic acids were examined in 7 grape wines, that is, gallic acid, protocatechuic acid, syringic acid, ethyl

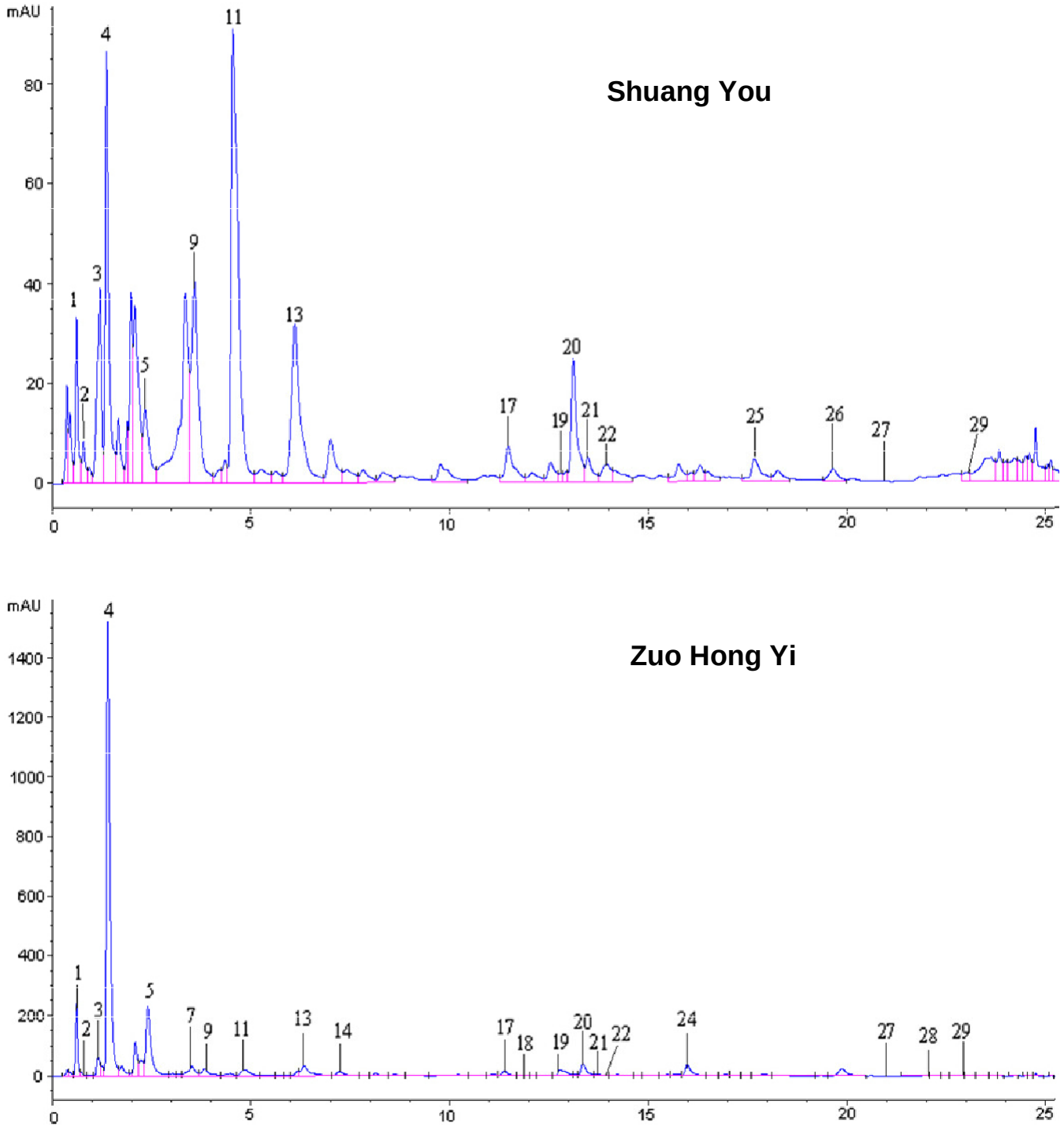
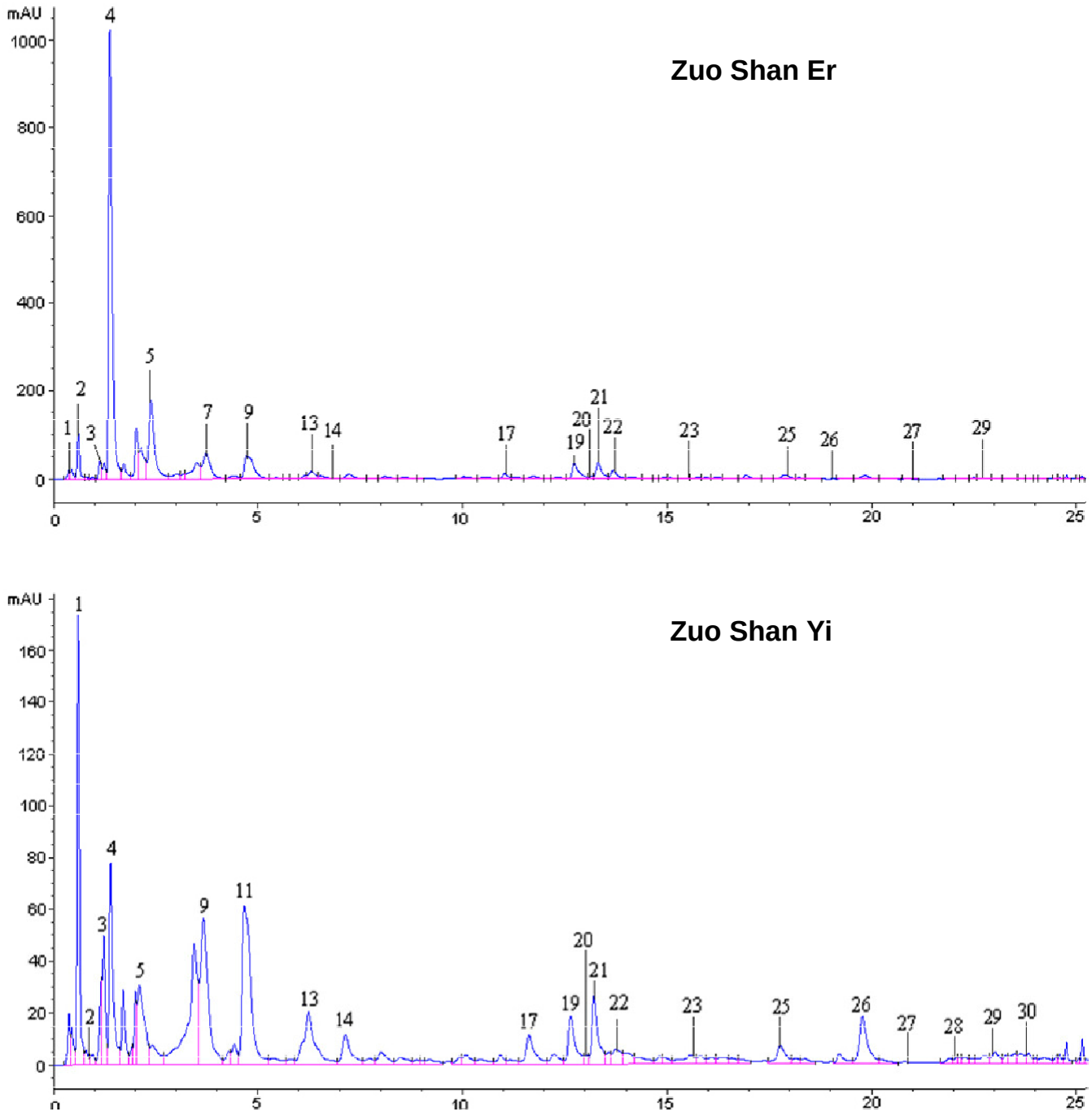


Figure 1. Contd.

gallate and ellagic acid. Shuang Hong wine had the highest benzoic acids content (17.94 mg/L), while Shuang You wine had the lowest (9.92 mg/L) (Figure 3). The contents of Zuo Shan Yi, Zuo You Hong, Zuo Shan Er, Shuang Feng and Zuo Hong Yi wines between both cultivars and their contents were 17.63, 13.87, 11.26, 10.98 and 10.84 mg/L, respectively.

The content of cinnamic acids

Seven kinds of cinnamic acids were examined in 7 grape cultivars including *trans*-cinnamic acid, *trans*-caftaric acid, *trans*-cutaric acid, ferulic acid, *trans*-fertaric acid, caffeic acid and ethyl caffeic acid. The highest cinnamic acids contents were observed in Shuang Hong (106.72 mg/L)



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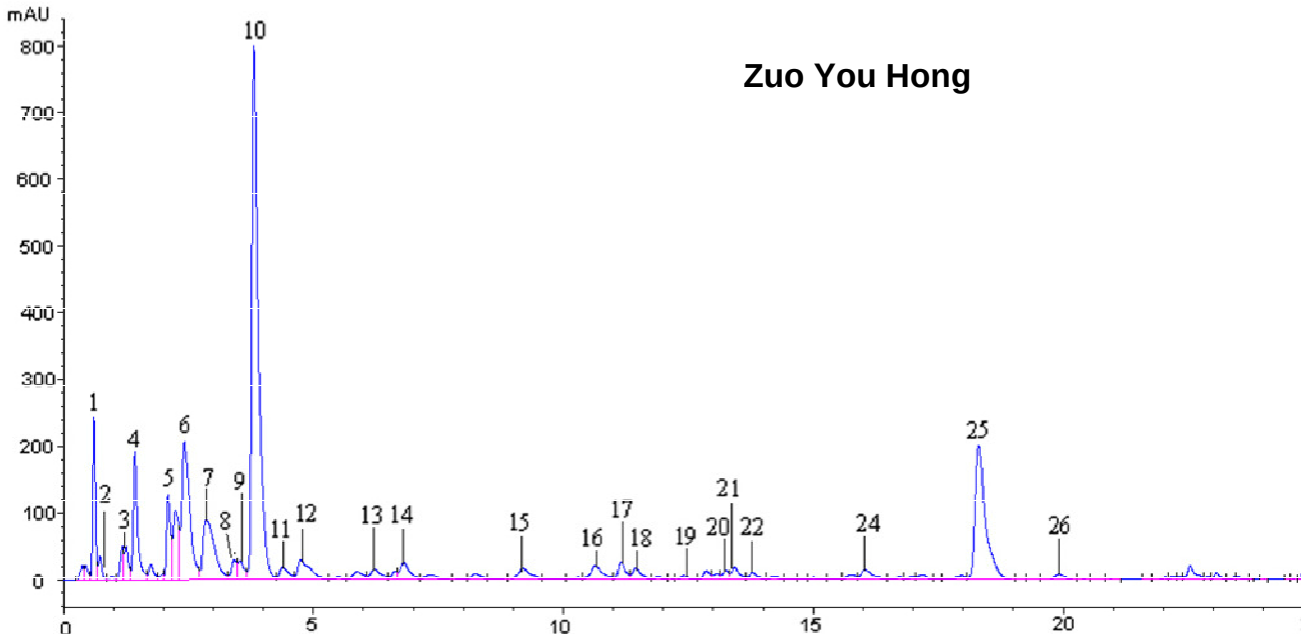
(Figure 4). The contents of Zuo You Hong, Zuo Hong Yi, Zuo Shan Er, Shuang Feng, Zuo Shan Yi and Shuang You were 94.26, 75.75, 54.71, 42.38, 14.13 and 6.88 mg/L, respectively.

The content of flavan-3-ols

The monomeric, oligomeric, or polymeric forms of flavan-3-ols which are found in both the seed and skin of the berry are responsible for the important wine astringency (Monagas et al., 2005). Five flavan-3-ols extracted from 7 grape cultivars included procyanidin B1, catechin, procyanidin B2, epicatechin and trimeric procyanidin C1. The highest contents of flavan-3-ols occurred in hybrid cultivars Zuo You Hong (116.09 mg/L), but the least in Shuang You (Figure 5). Shuang Hong, Zuo Hong Yi, Zuo Shan Er, Zuo Shan Yi and Shuang Feng had the



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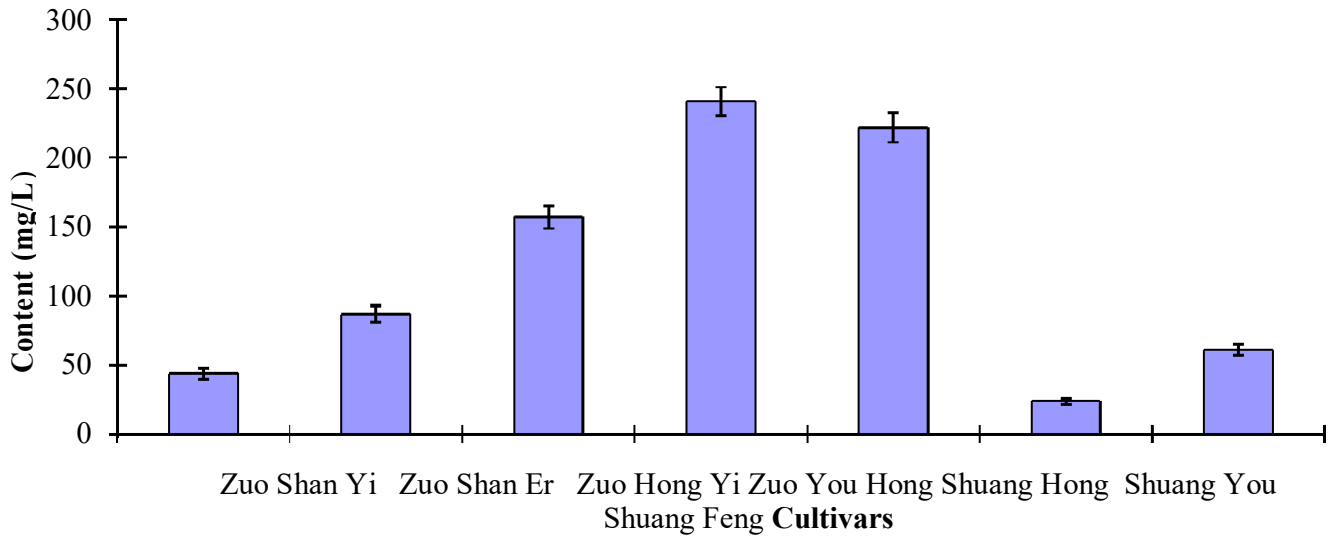

Figure 2. The content of total non-anthocyanin phenolic compounds.

contents of these compounds as 46.74, 43.21, 5.81, 3.32 and 2.38 mg/L, respectively. Trimeric procyanidin C1 was only found in Zuo You Hong. The content was 3.78 mg/L and occupied 1.57% of the non-anthocyanin phenolic compounds.

The content of flavonols

Flavonols generally act as UV protectors (Smith and Markham, 1998), and co-pigments of anthocyanins in flowers and fruits (Scheffeldt and Hrazdina, 1978). Eight flavonol aglycones, quercetin, kaempferol, isorhamnetin, myricetin, laricitrin, syringetin, dihydroquercetin, dihydro-kaempferol, were detected in the skins of Syrah, Cabernet Gernischt, Cabernet Sauvignon, Merlot and Garmay (Jin et al., 2009). Nine flavonols detected in 7 wine samples included dihydroquercetin, *myricetin-3-glucoside*, quercetin-3-galactoside, quercetin-3-glucuro-nide, quercetin-3-glucoside, myricetin, quercetin, naringenin and leutolin. It can be seen from Figure 6 that the contents of flavonols were the highest in Shuang

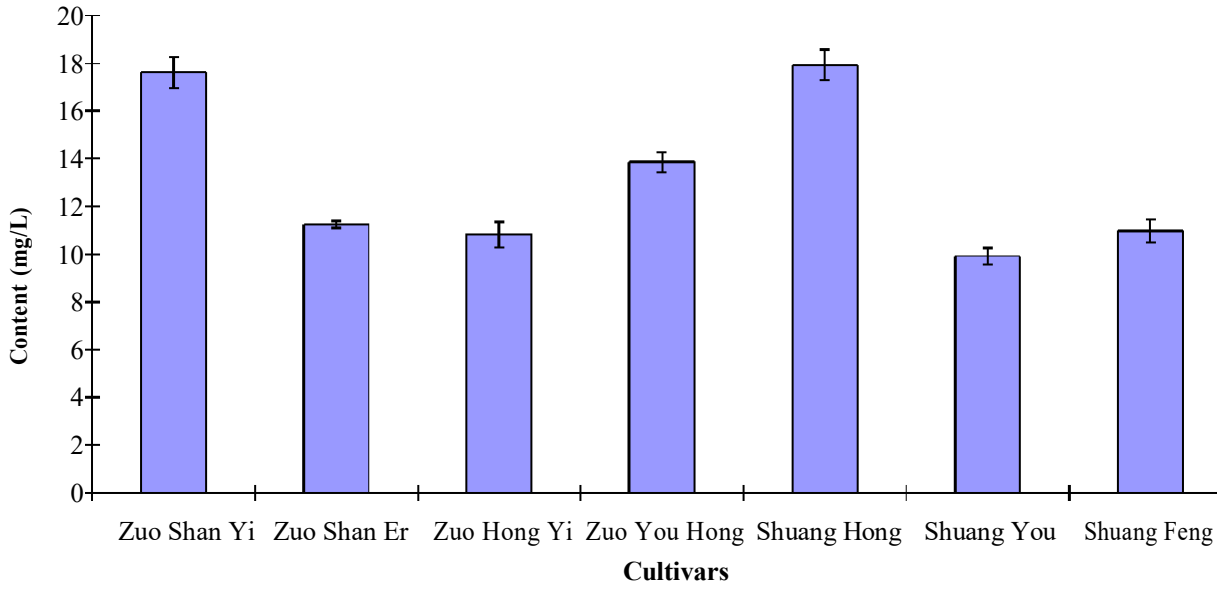

Figure 3. The content of benzoic acids.

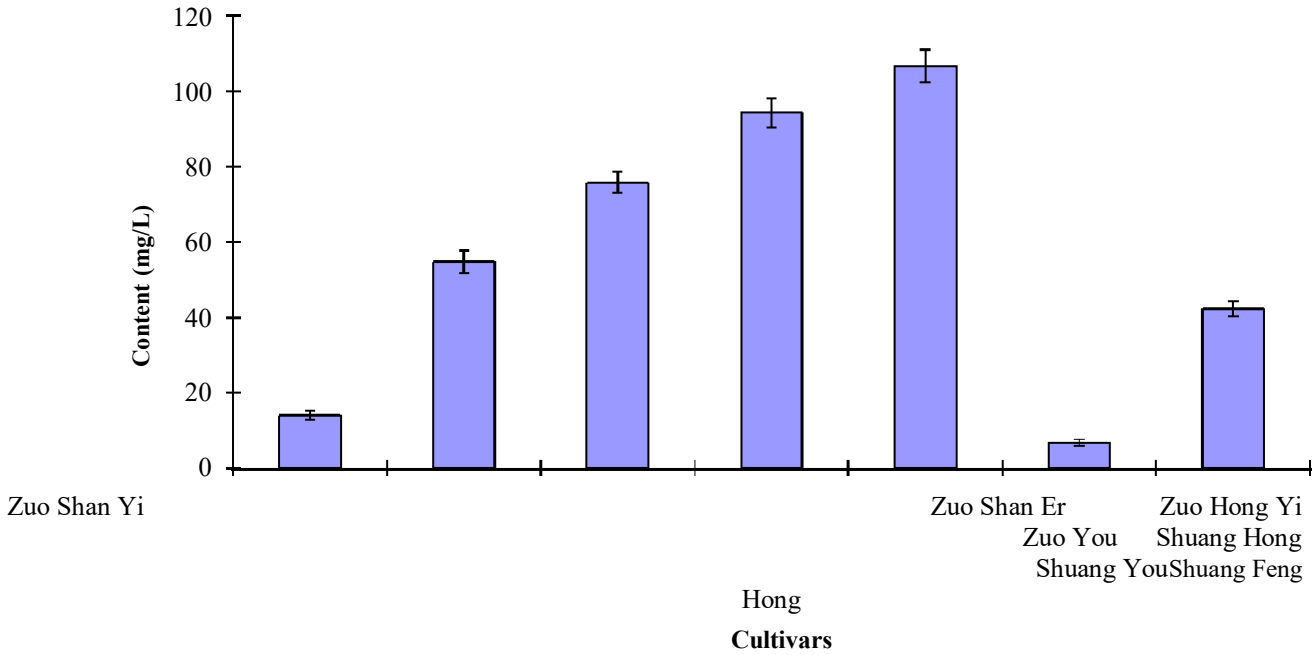

Figure 4. The content of cinnamic acids.

Hong (49.71 mg/L), and the contents in Zuo Hong Yi, Zuo Shan Er, Zuo You Hong, Zuo Shan Yi, Shuang You and Shuang Feng were 23.93, 14.80, 14.32, 11.11, 6.21 and 4.75 mg/L, respectively. The composition of flavonols varied widely between *V. amurensis* and two hybrid cultivars.

The content of stilbenes

Sato et al. (1997) studied contents of resveratrol, piceid, and their isomers from 42 kinds of red and white wines in Japan different region. They found the average content of total stilbene compounds was 4.37 mg/L in red wines (Sato et al., 1997). Four kinds of stilbenes were found in 7 wine samples including *trans*-Piceid, *cis*-Piceid, *trans*-Resveratrol, *and cis*-Resveratrol. Figure 7 shows that the contents of resveratrols were the highest in Zuo Hong Yi (3.23 mg/L), the intermediate in Zuo You Hong (2.38 mg/L) and the lowest (0.418 mg/L) in Zuo Shan Er. The content of two hybrid cultivars (Zuo Hong Yi and Zuo You Hong) was higher than *V. amurensis*.

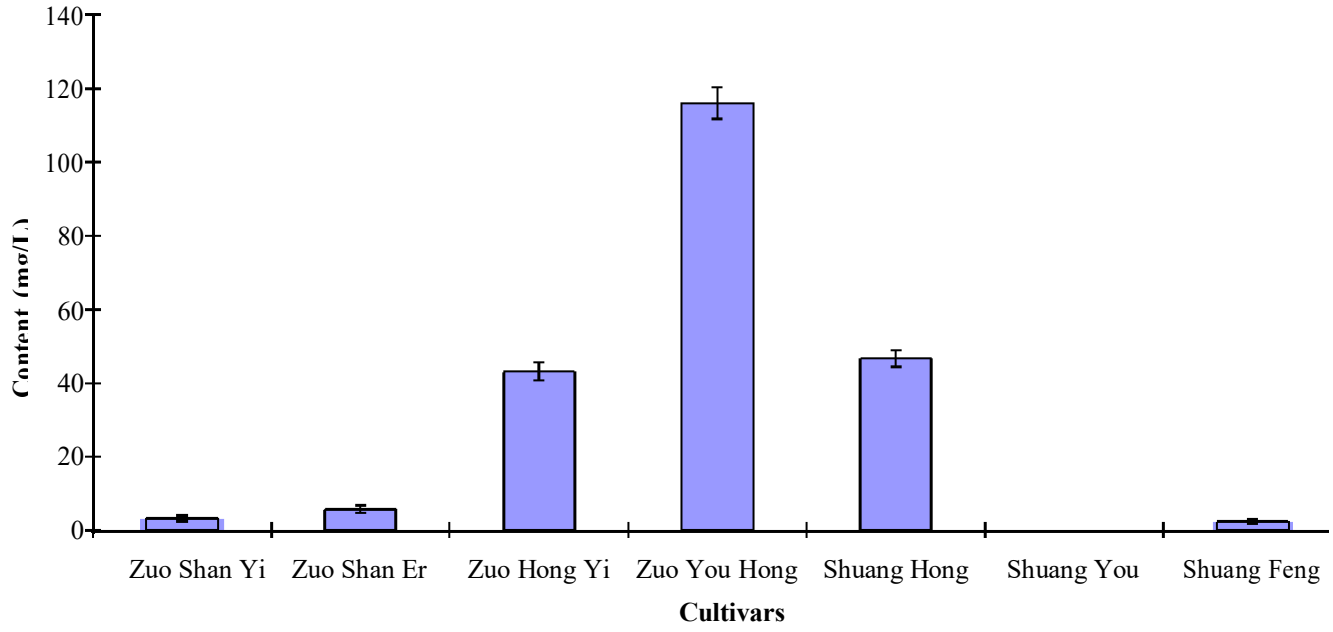

Figure 5. The content of flavan-3-ols.

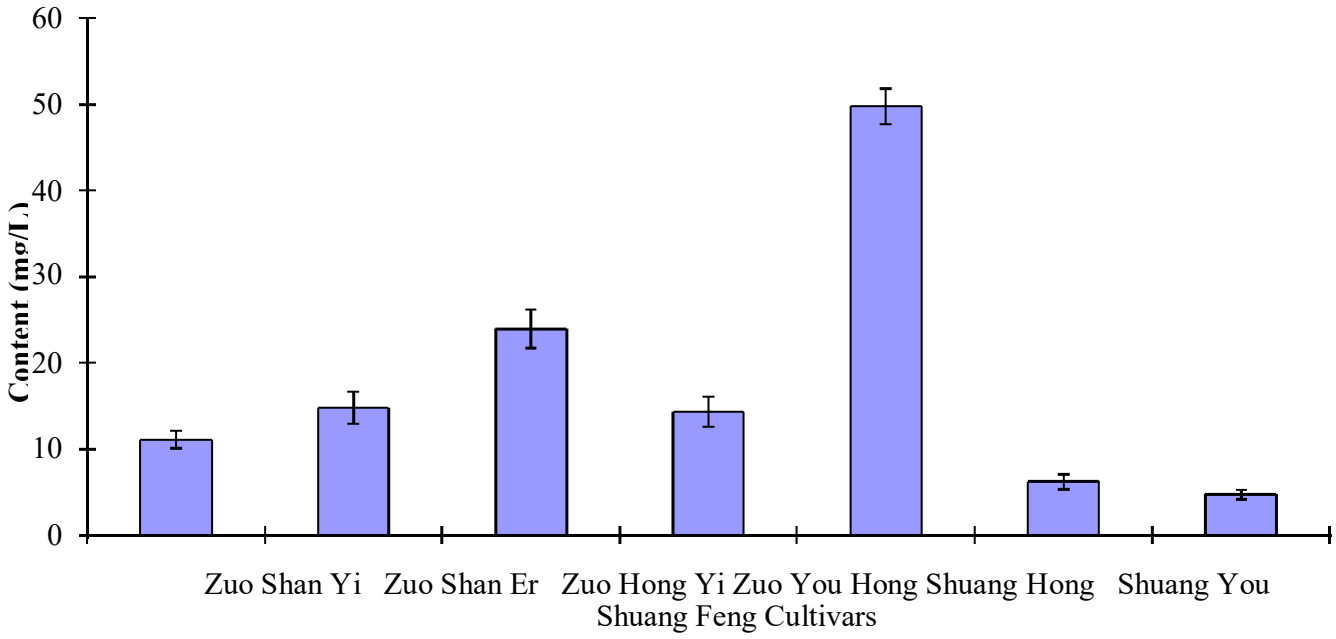

Figure 6. The content of flavonols.

Principal components analysis

To better understand the characteristics of non-anthocyanin phenolic compounds from 7 wine samples, principal components analysis was performed in terms of HPLC-MS/MS quantitative analysis. Five types of non-anthocyanin phenolic compounds were taken as analytical aim including benzoic acids (X_1), cinnamic acids (X_2), flavan-3-ols (X_3), flavonols (X_4) and stilbenes (X_5). SPSS data processing system was used to carry out principal components analysis. The principal components of non-anthocyanin phenolic compounds in wines from 7 grape cultivars were obtained. Eigenvectors and percentage of accumulated contribution of principal component are presented in Table 3. Two previous principal component proportions were 54.622 and 27.722%, respectively. The percentage of accumulated contribution reached 82.344%. Others that contribute less were

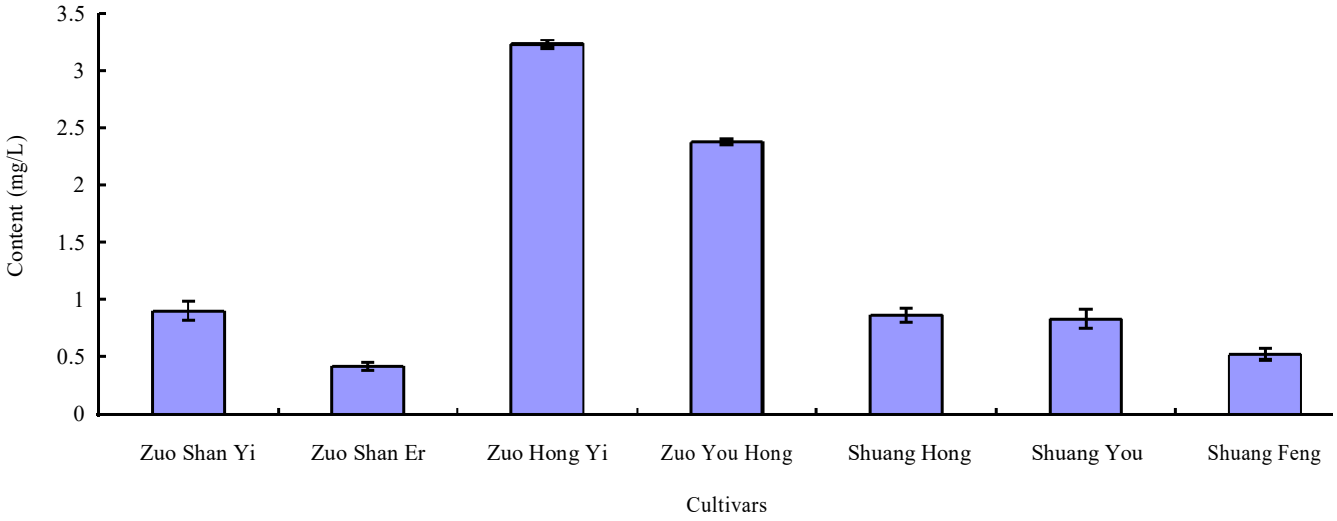

Figure 7. The content of stilbenes.

Table 3. Eigenvectors and percentage of accumulated contribution of principal component.

Item	Principal component				
	F_1	F_2	F_3	F_4	F_5
X_1	0.285	0.626	0.680	0.119	0.224
X_2	0.565	-0.009	-0.386	-0.329	0.651
X_3	0.507	-0.314	0.336	-0.523	-0.509
X_4	0.465	0.315	-0.480	0.373	-0.493
X_5	0.356	-0.582	0.215	0.682	0.154
Eigenvector	2.731	1.386	0.492	0.347	0.044
Proportion (%)	54.622	27.722	9.836	6.933	0.887
Cumulative (%)	54.622	82.344	92.180	99.113	100.000

ignored, but when they worth more than 1 principle according to the Eigenvector, it is suitable to distill 2 from numerous Eigenvector.

As it is shown in Table 3, the first principal component contributed 54.622%, mainly decided by cinnamic acids (X_2) and flavan-3-ols (X_3). Among them, cinnamic acids and flavan-3-ols had relatively bigger positive coefficient (r = 0.565 and r = 0.507). The second principal com-ponent contributed 27.722%; it is mainly decided by benzoic acids (X_1). The cinnamic acids, flavan-3-ols and benzoic acids played crucial roles in the composition of non-anthocyanin phenolic compounds in wines, accor-ding to principal components analysis.

Systematic cluster analysis

Clustering on non-anthocyanin phenolic compounds in 7 grape cultivars by Euclidean distance are given in Figure 8. Assemble firstly appeared in Shuang You and Zuo Shan Yi with the shortest distance of 0.03979 indicated that kinds and composition of this two cultivars were similar. The similar kinds and composition were also found in Shuang Hong and Zuo Hong Yi, Shuang Feng and Zuo Shan Er, but the distance between Zuo Shan Yi and Zuo Shan Er were longest (0.41727) showing that max-difference existed in 2 kinds.

Conclusion

In this experiment, 30 kinds of non-anthocyanin phenolic compounds in wines were identified in 7 grape cultivars including 5 benzoic acids, 7 cinnamic acids, 5 flavan-3-ols, 9 flavonols and 4 stilbenes. Also were found 25, 21, 21, 20, 20, 19 and 17 kinds in Zuo You Hong, Shuang Hong, Zuo Shan Yi, Shuang Feng, Zuo Hong Yi, Zuo Shan Er and Shuang You, respectively. Benzoic acids included gallic acid, protocatechuic acid, syringic acid, ethyl gallate and ellagic acid. Cinnamic acids were *trans*-cinnamic acid, *trans*-caftaric acid, *trans*-cutaric acid, ferulic acid, *trans*-fertaric acid, caffeic acid and ethyl caffeic acid. Flavan-3-ols were procyanidin B1, catechin, procyanidin B2, epicatechin, and trimeric procyanidin C1.

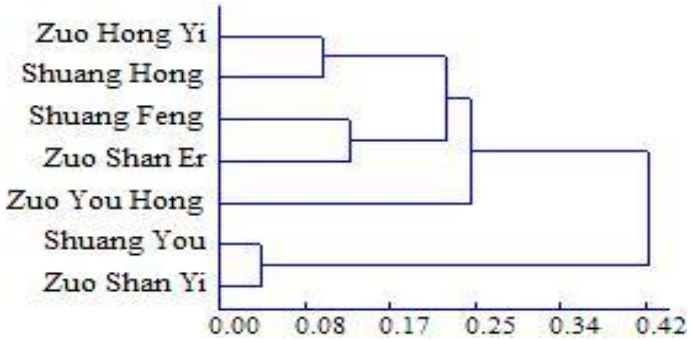

Figure 8. Systematic cluster analysis of non-anthocyanin phenolic compounds.

Flavonols were dihydroquercetin, *Myricetin-3-glucoside*, quercetin-3-galactoside, quercetin-3-glucuronide, quercetin-3-glucoside, myricetin, quercetin, naringenin and leutolin. Stilbenes were *trans*-piceid, *cis*-piceid, *trans*-resveratrol and *cis*-resveratrol. Systematic cluster analysis suggested that the non-anthocyanin phenolic compounds profiles were helpful to classify these cultivars of *V. amurensis* and the corresponding hybrids. In addition, this study shows that *V. amurensis* and its hybrids grape wines the non-anthocyanin phenolic com-pounds differed in varieties and contents. *V. amurensis* hybrids grape wines had better quality than *V. amurensis.* Therefore, the hybridization between *V. amurensis* and the Eurasian species help improve the quality of grape wines. Research on the varieties and contents and composition ratio characteristics of non-anthocyanin phenolic compounds in *V. amurensis* grape and its hybrids will enhance the identification of the grape wine materials and nutrition health.

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REFERENCES

Delgado R, Martín P, del Álamo M, González MR (2004). Changes in the phenolic composition of grape berries during ripening in relation to vineyard nitrogen and potassium fertilisation rates. J. Sci. Food Agric. 84: 623-630.

Garcia-Viguera C, Bridle P (1995). Analysis of non-coloured phenolic compounds in red wines, a comparison of hige-performance liquid chromatography and capillary zone electrophresis. Food Chem. 54: 349-352.

Gawel R (1998). Red wine astringency: A review. Aust. J. Agr. Res. 4: 74-95.

Sato M, Suzuki Y, Okuda T, Yokotsuka K (1997). Contents of resveratrol, piceid, and their isomers in commercially available wines made from grapes cultivated in Japan. Biosci. Biotech. Biochem. 61: 1800-1805.

Hunter J, De Villier O, Watts J (1991). The effect of partial defoliation on quality characteristics of *Vitis vinifera* L. cv. Cabernet Sauvignon grapes. II. Skin color, skin sugar, and wine quality. Am. J. Enol. Vitic. 42: 13-18.

Iacopini P, Baldi M, Storchi P, Sebastiani L (2008). Catechin, epicatechin, quercetin, rutin and resveratrol in red grape: Content, in vitro antioxidant activity and interactions. J. Food Compost. Anal. 21: 589-598.

Jin ZM, He JJ, Bi HQ, Cui XY, Duan CQ (2009). Phenolic compound profiles in berry skins from nine red wine grape cultivars in northwest China. Molecules, 14: 4922-4935.

Mané C, Souquet JM, Ollé D,Verriés C, Véran F, Mazerolles G, Cheynier V, Fulcrand H (2007). Optimization of simultaneous flavanol, phenolic acid, and anthocyanin extraction from grapes using an experimental design: application to the characterization of champagne grape varieties. J. Agric. Food Chem. 55: 7224-7233.

Minussi RC, Rossi M, Bologna L, Cordi L, Rotilio D, Pastore GM, Durán N (2003). Phenolic compounds and total antioxidant potential of commercial wines. Food Chem. 82: 409-416.

Monagas M, Bartolomé B, Gómez-Cordovés C (2005) . Updated knowledge about the presence of phenolic compounds in wine. Crit. Rev. Food Sci. Nutr. 45: 85-118.

Monagas M, Martín-Álvarez PJ, Bartolomé B, Gómez-Cordovés C (2006). Statistical interpretationof the color parameters of red wines in function of their phenolic composition during aging in bottle. Eur. Food Res. Technol. 222: 702-709.

Pérez-Magariño S, González-Sanjosé ML (2006). Polyphenols and colour variability of red wines made from grapes harvested at different ripeness grade. Food Chem. 96: 197-208.

Picinelli A, Suárez B, García L, Mangas JJ (2000). Changes in phenolic contents during sparkling apple winemaking. Am. J. Enol. Vitic. 51: 144-149.

Salas E, Fulcrand H, Meudec E, Cheynier V (2003). Reactions of anthocyanins and tannins in model solutions. J. Agr. Food Chem. 51: 7951-7961.

Scheffeldt P, Hrazdina G (1978). Copigmentation of antho-cyanins under physiological conditions. J. Food Sci. 43: 517-520.

Silvia P, Luisa M (2005). Effect of ripening stage of grapes on the low molecular weight phenolic compounds of red wines. Eur. Food Res. Technol. 220: 579-607.

Smith GJ, Markham KR (1998). Tautomerism of flavonol glucosides: Relevance to plant UV protection and flower colour. J. Photochem. Photobiol. Chem. 118: 99-105.

Sun B, Leandro C, Ricardo da Silva JM (1998). Spranger I. Separation of grape and wine proanthocyanidins according to their degree of polymerization. J. Agric. Food Chem. 46 (4): 1390-1396.

Yilmaz Y, Toledo RT (2004). Major flavonoids in grape seeds and skins: Antioxidant capacity of catechin, epicatechin, and gallic acid. J. Agric. Food Chem. 52: 255-260.

Zhao Q, Duan CQ, Wang J (2010). Anthocyanins profile of grape berries of Vitis amurensis, its hybrids and their wines. Int. J. Mol. Sci. 11: 2212-2228.