Advance Journal of Food Science and Technology 12(5): 271-280, 2016
DOI:10.19026/ajfst.12.2910
ISSN: 2042-4868; e-ISSN: 2042-4876

Submitted: November 3, 2015 Accepted: January 8, 2016 Published: October 15, 2016

Research Article

Correlations between Aroma Profiles and Sensory Characteristics of Red Wines by Using Partial Least Squares Regression Method

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Abstract: The aim of this study is to examine the effectiveness of Partial Least Squares Regression (PLSR) method in determing correlations between the aroma profiles and sensory characteristics of wines. A total of 45 volatile compounds in five different Chinese grape wines were identified and quantified by HS-SPME/GC-MS and 26 of them with OAV (odour activity value) >1. All aroma compounds with OAV>1 were selected for evaluating the correlations between the aroma profiles and 12 sensory descriptors using PLSR and their ROC (Relative Odour Contribution). The results showed that ethyl decanoate, ethyl hexanoate, acetaldehyde, isoamyl acetate, hexanoic acid, 4-vinylguaiacol and geraniol were the major contributors to the desirable balanced aroma of *muscat* wine. Ethyl hexanoate, ethyl butyrate, isoamyl acetate, acetaldehyde, hexanoic acid, 3-methyl-1-butanol and octanoic acid were mainly responsible for the aroma of *black beet* wine and *cabernet gernischt* wine whereas ethyl tetradecanoate, neryl acetate and nerol were the particular aroma compounds in *black beet* wine and γ-butyrolactone, nerolidol and β-ionone were special aroma compounds in *cabernet gernischt* wine. Both PLSR and ROC are effective methods to demonstrate the correlations between the sensory characteristics of the analyzed wines and their aroma compositions.

Keywords: Chinese grape wines, HS-SPME/GC-MS, odour activity value, partial least squares regression, relative odour contribution, sensory descriptors

INTRODUCTION

The aroma of wine is one of the most important factors contributing to its quality. The formation of wine aroma is mainly influenced by the grape variety, vine growing conditions and fermentation technology. HS-SPME/GC-MS has been widely applied to identify and quantify the volatile compounds in wines (Marquez *et al*., 2014; Pereira *et al*., 2010), because this approach can quickly, simply and relatively accurately assess the essential volatile compounds from wines (Sagratini *et al*., 2012). Moreover, only part of the volatile compounds make a contribution to wine aroma. Many researchers have proved that only those odorants with an Odour Activity Value (OVA) above 1 can contribute to the entire aroma of the wine (Allen *et al*., 1994). Odour Activity Value (OAV) and Relative Odour Contribution (ROC) are two conventional indicators for evaluation of contribution of volatile compounds to wine aroma (Wang *et al*., 2015). The OAV is calculated through dividing the concentration of an aroma compound by its odour threshold (Gómez-Míguez *et al*., 2007; Gil *et al*., 2006). ROC is defined as the ratio of the OAV percentage of each compound and the sum of the OAV of compounds which OAV>1 (Welke *et al*., 2014).

However, the sensory evaluation on the aroma by human subjects is very important and irreplaceable by any advanced instruments, because eventually the overall sensory sensation of the mouth-feel and aroma attributes are the most concern by the consumers.

Recently, several multivariate statistical methods such as Partial Least Squares Regression (PLSR) were used to predict the relationship between the aroma compounds and sensory properties (King *et al*., 2010). PLSR is soft modeling relating the variations in one or

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several response variables (Y-variables) to the variations of several predictors (X-variables), with explanatory or predictive purpose. For example, some researchers have evaluated the correlation between the aroma compounds and sensory properties in white and cherry wines using PLSR (Pereira *et al*., 2010; Sun *et al*., 2012; Xiao *et al*., 2014). However, using PLSR method to evaluate the correlation between the aroma compounds and sensory characteristics of Chinese wines has not been well documented. Therefore, in this study, five different red wines were used as a group of red wine examples to examine the effectiveness of PLSR method in determining correlations between the aroma profiles and sensory characteristics of red wines. ROC method was used to compare the effectiveness of PLSR method to demonstrate the correlations between the sensory characteristics and their aroma compositions.

MATERIALS AND METHODS

Chemicals:

Standard compounds: Ethyl acetate, ethyl lactate, ethyl octanoate, ethyl decanoate, diethyl succinate, γ-butyrolactone, 2-phenylethyl acetate, ethyl laurate, ethyl tetradecanoate, neryl acetate, ethyl valerate, ethyl hexanoate, ethyl butyrate, isoamyl acetate, isobutyl acetate, 1-propanol, 1-butanol, 3-methyl-1-butanol, 2, 3-butanediol, 2-phenylethanol, 1-hexanol, 2-propanol, acetaldehyde, furfural, benzaldehyde, 3-hydroxy-2-butanone, β-Ionone, 2-octanone, geranyl acetone, acetic acid, hexanoic acid, octanoic acid, decanoic acid, heptanoic acid, propionic acid, butyric acid, 4-ethylphenol, 4-ethylguaiacol, 4-vinylphenol, 4-vinylguaiacol, β-citronellol, geraniol, linalool, nerolidol and nerol were purchased from Sigma-Aldrich (Saint Luis, EUA). Standard chemicals of *n*-alkane standards (C7-C30) and 2-octanol (internal standard), were purchased from Sigma-Aldrich Chemical Co. Ltd (San Diego, USA).

Wine samples: The five different kinds of wines fermentated using 5 different kinds of grapes of *summer black*, *black beet*, *cabernet franc*, *muscat* and *cabernet genischt* which were purchased from different vineyard located in China labeled as ‘A’, ‘B’, ‘C’, ‘D’ and ‘E’, respectively. The details of these wines and wine grapes were listed in Table 1. The winemaking procedures used were as reported in literature (King *et al*., 2008). Briefly, the grape juice involved triplicate fermentations in 20L stainless steel vessels and control of the temperature between 20-25°C. 30 mg/L of free SO_2 was added. The grape juice was sulfited and unfiltered to mimic commercial fermentations. The wine was bottled and kept in wine cabinet at 12°C for 6 months before analysis. 300 mg/L of wine yeast (*saccharomyces cerevisiae*, Angel yeast Co., LTD, Yichang, China) was added to ferment.

Extraction and headspace aroma compounds using SPME: Each wine (5 mL) was prepared by adding 1 g of NaCl and 5 µL of 2-octanol (262 mg/L in absolute ethanol as an internal standard) in 20 mL extraction bottle. The volatiles from the wines were balanced at 60°C for 10 min and then adsorbed by a 75 µL CAR/PDMS SPME fiber (Supelco, Bellefonte, USA) at 60°C for 30 min.

Identification and quantification of aroma compounds using GC-MS: The adsorbed wine volatiles on the SPME fiber were separated and identified using a 7890 Gas Chromatograph (GC) coupled with a 5973C Mass Spectrometer (MS) (Agilent Technologies, Palo Alto, USA) and an HP-INNOWAX fused-silica capillary column (60 m × 0.25 mm ID, 0.25 µm film thickness). Helium was used as the carrier gas at a flow rate of 1 mL/min. The injector temperature was set at 250°C. The oven temperature program was set as follows: 40°C for 2 min, 5°C/min ramp to 230°C and 230°C for 5 min. The temperatures of the transfer line, the ion-trap manifold and the quadruple mass filter were set at 250, 230 and 150°C, respectively. The energy for electron ionization was 70 eV. The chromatograms of the volatiles in the five wines were recorded by monitoring the total ion currents in a range of 30 to 450 m/z. The SPME fiber was introduced to the GC injector for 5 min desorption in a splitless mode.

Each of the volatile was identified through comparison of retention time and mass spectra data of corresponding authentic standard (Table 2 and 3). The identifications of the volatiles were further confirmed through comparison of the Kovats Retention Indices (RI) and fragmentation patterns reported in the Wiley 7.l Mass Spectra Database (Hewlett-Packard, Palo Alto, CA). The RIs of the volatiles were calculated using a homologous series of *n*-alkanes (C7-C30) under the same analysis conditions.

Table 1: The details of five wine samples and their wine grapes including *summer black*, *black beet*, *cabernet franc*, *muscat* and *cabernet gemischt*

Wines	Grape varieties	Areas of grape sources	Vintages	Vineyards	Climite of grape growing condition
A	Summer black	Li county, hunan	2013	Shenzhou	Humid subtropical monsoon climate
B	Black beet	Yantai, shandong	2013	Zhangyu	Temperate monsoon climate
C	Cabernet franc	Shihezi, xinjiang	2013	Zhangyu	Continental dry climate
D	Muscat	Hangu, tanjin	2013	Tea lake	Temperate monsoon climate
E	Cabernet gernischt	Yantai, shandong	2013	Zhangyu	Temperate monsoon climate

Table 2: Concentrations (mg/l), retention indices, odour descriptors and odour thresholds of aroma compounds in in five red wines

Compound [a]	Rical [b]	Riref [c]	Id [d]	A	B	C	D	E	Odour descriptor [e]	Odour threshold (mg/l) [f]
Esters										
Ethyl acetate	873	907	R	16.90±0.24	25.05±0.13	17.03±1.28	20.99±0.01	14.79±0.18	Pineapple [1]	12.27 [6]
Ethyl lactate	1350	1358	R	0.93±0.02	1.40±0.01	0.93±0.02	1.24±0.01	0.91±0.02	Fruit, lactic [1]	154 [7]
Ethyl octanoate	1434	1436	R	1.18±1.02	2.32±1.23	0.97±0.02	1.58±1.01	0.95±0.78	Pineapple, pear, sweet [3]	0.6 [8]
Ethyl decanoate	1630	1636	R	0.18±0.13	0.03±0.01	0.04±0.03	0.18±0.08	0.30±0.25	Grape [1]	0.023 [9]
Diethyl succinate	1693	1689	R	0.12±0.02	0.12±0.07	0.15±0.12	0.12±0.17	0.32±0.12	Wine, fruit [1]	200 [7]
2-phenylethyl acetate	1832	1829	R	0.26±0.05	Nd	0.63±0.98	0.26±0.12	0.39±0.17	Rose, honey, floral [1]	0.25 [10]
Ethyl laurate	1845	1842	R	0.06±0.02	0.41±0.04	Nd	0.04±0.01	0.07±0.01	Leaf [1]	0.4 [9]
Ethyl tetradecanoate	2031	2042	R	Nd	1.26±0.09	Nd	Nd	Nd	Fruit, ether [1]	0.18 [9]
Neryl acetate	1718	1742	R	Nd	1.03±0.16	Nd	Nd	Nd	Fruit [1]	0.88−905.4 [11]
Ethyl valerate	1152	1133	R	Nd	Nd	Nd	Nd	0.02±0.01	Yeast, fruit [1]	0.0015-0.005 [9]
Ethyl hexanoate	1232	1220	R	0.38±0.12	0.48±0.14	0.29±0.08	0.31±0.14	0.44±0.15	Fruity, green, apple [4]	0.014 [12]
Ethyl butyrate	1039	1028	R	0.02±0.02	0.17±0.10	0.08±0.02	0.06±0.01	0.21±0.12	Strawberry [3], apple [1]	0.02 [12]
Isoamyl acetate	1204	1117	R	0.13±0.02	0.22±0.03	0.16±0.06	0.14±0.07	0.21±0.08	Banana [1]	0.03 [13]
Isobutyl acetate	1021	1015	R	0.04±0.01	0.06±0.02	Nd	0.04±0.04	0.05±0.03	Fruit, apple, banana [1]	1.6 [6]
Total esters (14)				20.20±1.67	32.55±2.03	20.8±2.02	24.96±1.67	18.45±2.07		
Alcohols										
1-propanol	1052	1037	R	4.68±0.01	9.87±0.04	1.20±0.01	4.68±0.06	4.75±0.01	Alcohol, ripe fruit [1]	9 [14]
1-butanol	1134	1158	R	0.18±0.07	0.38±0.09	0.13±0.09	0.18±0.07	0.31±0.08	Medicine, fruit [1]	4.33 [9]
3-methyl-1-butanol	1198	1205	R	79.03±10.87	114.18±12.98	74.18±10.47	79.03±7.67	108.54±13.72	Whiskey, malt, burnt [1]	30 [13]
2, 3-butanediol	1530	1583	R	2.50±0.02	2.68±0.04	2.34±0.03	2.50±0.04	1.12±0.01	Fruit, onion [1]	>100 [9]
2-phenylethanol	1921	1925	R	16.81±2.12	18.63±3.14	16.81±0.02	32.60±3.08	26.15±5.30	Rose [1]	14 [13]
Leaf alcohol	1368	1392	R	Nd	Nd	Nd	Nd	0.91±0.42	Grass [1]	0.91 [9]
2-propanol	1550		M	Nd	Nd	1.05±0.12	Nd	Nd	Alcohol, musty, woody [2]	40~78 [9]
Total alcohols (7)				103.20±13.09	145.74±16.19	95.71±12.74	118.99±10.92	141.78±19.54		
Aldehydes										
Acetaldehyde	717	724	R	1.65±0.08	0.65±0.06	1.83±0.12	0.79±0.01	0.56±0.07	Pungent, ethereal, fruity [2]	0.01 [9]
Furfural	1463	1455	R	0.08±0.04	0.06±0.01	Nd	0.08±0.04	0.03±0.01	Bread, almond, sweet [1]	0.77 [9]
Benzaldehyde	1501	1495	R	0.06±0.01	0.15±0.12	0.13±0.25	0.06±0.01	0.06±0.02	Almond, burnt sugar [1]	2 [13]
Total aldehydes (3)				1.79±0.13	0.86±0.19	1.96±0.37	0.93±0.06	0.65±0.10		
Ketones										
3-hydroxy-2-butanone	1275	1287	R	2.04±0.96	0.73±0.02	3.40±0.28	2.04±0.20	0.09±0.01	Butter, cream [1]	0.8 [9]
Γ-butyrolactone	1643	1647	R	Nd	Nd	Nd	Nd	5.03±0.15	Caramel, sweet [1]	1 [9]
Β- ionone	1971	1912	R	Nd	Nd	Nd	Nd	0.05±0.02	Violet flower, raspberry [1]	0.0009 [13]
2-octanone	1283	1285	R	Nd	Nd	Nd	Nd	0.03±0.01	Earthy, weedy, natural woody, herbal [2]	0.05 [9]
Geranyl acetone	1838	1840	R	0.04±0.01	0.13±0.01	Nd	0.22±0.06	Nd	Magnolia, green [1]	0.186 [9]
Total ketones (5)				2.08±0.97	0.86±0.03	3.40±0.28	2.26±0.26	5.20±0.14		
Acids										
Acetic acid	1458	1450	R	1.92±0.95	1.14±0.68	17.18±1.20	1.92±0.82	0.47±0.04	Sour [1]	25.59-26 [9]
Hexanoic acid	1863	1864	R	2.04±0.01	1.83±0.02	1.02±0.02	2.04±0.01	2.07±0.02	Green [2]	0.42 [13]
Octanoic acid	2079	2083	R	1.16±0.01	2.88±0.04	1.11±0.03	1.16±0.01	3.12±0.01	Sweat, cheese [1]	0.5 [13]
Decanoic acid	2368	2361	R	0.18±0.03	Nd	Nd	0.18±0.03	0.10±0.02	Rancid, fat [1]	1 [13]
Heptanoic acid	1836		M	Nd	Nd	Nd	0.04±0.01	Nd	Rancid, sour cheesy, sweat [2]	0.64-0.91 [9]
Propionic acid	1520	1523	R	Nd	Nd	0.07±0.02	Nd	Nd	Pungent, rancid [1]	8.1 [7]
Butyric acid	1623	1619	R	0.40±0.04	0.80±0.04	4.80±0.08	0.44±0.01	0.50±0.02	Fatty-rancid, cheesy, sweaty [4]	0.4 [12]
Total acids (7)				5.70±1.04	6.65±0.78	24.18±1.35	5.78±0.88	6.26±0.11		
Phenols										
4-ethylphenol	2194	2195	R	1.50±0.14	Nd	0.81±0.14	0.07±0.02	Nd	Phenol, spice [1]	0.44 [15]
4-ethylguaiacol	2018	2031	R	0.58±0.04	Nd	0.64±0.12	0.04±0.01	Nd	Spice, clove [1]	0.033 [13]
4-vinylphenol	2425	2427	R	0.03±0.01	0.08±0.02	0.17±0.04	0.13±0.02	Nd	Almond shell [1]	0.18 [15]
4-vinylguaiacol	2202	2198	R	1.00±0.12	Nd	1.01±0.09	0.18±0.04	0.04±0.01	Clove, curry [1]	0.04 [10]
Total phenols (4)				3.11±0.31	0.08±0.02	2.63±0.39	0.42±0.09	0.04±0.01		
Terpenes										
β-Citronellol	1759	1762	R	0.04±0.01	0.20±0.12	0.05±0.03	0.04±0.01	0.03±0.01	Rose [1]	0.1 [10]
Geraniol	1854	1847	R	Nd	0.03±0.02	Nd	0.17±0.12	0.04±0.03	Rose [1]	0.03 [13]

Table 2: Continue

Linalool	1543	1537	R	Nd	0.01±0.01	Nd	Nd	0.02±0.02	Flower, lavender [1]	0.025 [13]
Nerolidol	2014	2009	R	Nd	Nd	Nd	Nd	2.56±0.02	Floral [1]	2.25~10 [9]
Nerol	1779	1770	R	Nd	0.70±0.18	Nd	Nd	Nd	Flower, grass [5]; sweet [1]	0.04 [10]
Total terpenes (5)				0.04±0.01	0.94±0.23	0.05±0.03	0.21±0.11	2.65±0.06		

nd: not found; a. Quantification of volatiles in five red grape wines was carried out by standard curves of each standard compound from six different concentrations in model wines; b. Linear retention index of unknown aroma compound on a hp-innowax fused-silica capillary column (60×0.25×mm×0.25µm) with a homologous series of n-alkanes(c7-c30); c. The referenced ri from the database (http://webbook.nwast.gov/chemwastry/); d. Identification method is indicated as follows: m, mass spectrum and ri agree with of aroma compound conducted under similar gc-ms condition; r, identification of retention index with literature data; e. Odour descriptor from literature: [1]http://www.flavornet.org/flavornet.html; [2] http://www.thegoodscentscompany.com/index.html; [3] Moyano *et al*. (2002); [4] Capone *et al*. (2013); [5] Van Gemert (2003); f. Odour threshold from literature: [6] Amoore and Hautala (1983); [7] Maarse (1991); [8] Moyano *et al*. (2002); [9] Van Gemert (2003); [10] Guth (1997); [11] Liu and Pilone (2000); [12] Capone *et al*. (2013); [13] Ferreira *et al*. (2000); [14] Fazzalari (1978); [15] López *et al*. (2002)

Table 3: The scores and definitions of sensory descriptors in five red wines. Mean scores in the same row followed by different letters are significantly different ($p<0.05$)

Descriptor	A	B	C	D	E	Specific aroma nuances
Odour intensity	6.51±0.46c	8.53±0.39a	6.97±0.33bc	7.38±0.28b	7.87±0.65ab	Overall odour strength
Citrus fruit	0.00±0.00b	0.00±0.00b	0.00±0.00b	0.00±0.00b	4.62±0.15a	Lemon, orange, grapefruit
Red fruit	5.84±0.19d	6.25±0.03c	5.35±0.03e	6.86±0.09b	7.17±0.09a	Strawberry, redcurrant, red cherry
Green/vegetal	0.00±0.00d	5.09±0.03b	0.76±0.21c	0.00±0.00d	6.03±0.02a	Asparagus, cauliflower, green beans, hay, herbaceous/cut grass
Dried fruit	5.60±0.24d	7.46±0.05a	4.36±0.07e	5.97±0.05c	7.05±0.03b	Raisins, fig, prune, fermented fruit
Lactic	3.53±1.00c	6.45±0.93a	5.52±0.98ab	3.06±0.55c	4.29±0.79bc	Sour milk, cream, butter
Nuts	3.82±0.29b	5.83±0.51a	1.08±0.04c	4.05±0.45b	4.25±0.28b	Almond, walnut, hazelnut
Spice	4.61±1.00a	1.25±0.51c	5.05±0.59a	3.26±0.93b	1.02±0.11c	Vanilla, clove, anis/fennel, liquorice
Floral	4.32±0.45cd	4.85±0.38c	4.04±0.37d	8.85±0.09a	8.03±0.22b	Jasmine, lilac, violet, rose
Orchard fruit	2.25±0.60c	5.97±0.72a	2.16±0.60c	4.41±0.47b	3.96±0.07b	Pear, apple, pineapple, peach, nectarine
Jammy/lolly	5.02±1.00cd	7.14±0.47a	5.53±0.50bc	4.24±0.22d	6.46±0.41ab	Fruit jam, cooked fruit, fruity lolly
Undesirable aroma	5.27±0.26a	0.00±0.00c	4.36±0.34b	0.00±0.00c	0.00±0.00c	Smoke, plastic, burnt plastic, cow dung

Table 4: Odour activity values of the aroma compounds (oav>1) in the five wine and their relative odour contributions (roc) to the wines

	Oav					Roc%				
Compounds	A	B	C	D	E	A	B	C	D	E
Ethyl acetate	1.38	2.04	1.39	1.71	1.21	1.12	1.85	1.12	2.12	0.75
Ethyl octanoate	1.97	3.87	1.61	2.64	1.59	1.60	3.51	1.30	3.27	0.98
Ethyl decanoate	7.87	1.26	1.91	7.87	13.22	6.39	1.14	1.55	9.77	8.17
Γ-butyrolactone	<1	<1	<1	<1	5.03	0.00	0.00	0.00	0.00	3.11
2-phenylethyl acetate	1.05	<1	2.53	1.05	1.57	0.85	0.00	2.05	1.30	0.97
Ethyl laurate	<1	1.02	<1	<1	<1	0.12	0.93	0.00	0.12	0.11
Ethyl tetradecanoate	<1	6.99	<1	<1	<1	0.00	6.34	0.00	0.00	0.00
Ethyl valerate	<1	<1	<1	<1	4.62	0.00	0.00	0.00	0.00	2.86
Ethyl hexanoate	27.14	34.29	20.71	22.29	31.07	22.04	31.10	16.73	27.67	19.21
Ethyl butyrate	1.20	8.50	4.15	3.20	10.60	0.97	7.71	3.35	3.97	6.55
Isoamyl acetate	4.40	7.33	5.20	4.70	7.00	3.57	6.65	4.20	5.84	4.33
1-propanol	<1	1.10	<1	<1	<1	0.42	0.99	0.11	0.65	0.33
3-methyl-1-butanol	2.63	3.81	2.47	2.63	3.62	2.14	3.45	2.00	3.27	2.24
2-phenylethanol	1.20	1.33	1.20	2.33	1.87	0.98	1.21	0.97	2.89	1.16
Acetaldehyde	16.53	6.54	18.31	7.86	5.63	13.42	5.93	14.79	9.76	3.48
3-hydroxy-2-butanone	2.55	<1	4.25	2.55	<1	2.07	0.83	3.43	3.16	0.07
Β-ionone	<1	<1	<1	<1	58.89	0.00	0.00	0.00	0.00	36.41
Geranyl acetone	<1	<1	<1	1.17	<1	0.17	0.62	0.00	1.46	0.00
Hexanoic acid	4.86	4.35	2.43	4.86	4.93	3.94	3.95	1.96	6.03	3.05
Octanoic acid	2.32	5.75	2.22	2.32	6.23	1.89	5.21	1.79	2.88	3.85
Butyric acid	<1	2.01	12.01	1.10	1.26	0.80	1.82	9.70	1.36	0.78
4-ethylphenol	3.40	<1	1.84	<1	<1	2.76	0.00	1.48	0.18	0.00
4-ethylguaiacol	17.70	<1	16.21	1.21	<1	14.37	0.00	13.10	1.50	0.00
4-vinylguaiacol	25.10	<1	25.20	4.60	<1	20.38	0.00	20.36	5.71	0.59
Geraniol	<1	1.07	<1	5.70	1.20	0.00	0.97	0.00	7.08	0.74
Nerol	<1	17.40	<1	<1	<1	0.00	15.78	0.00	0.00	0.00

Standards solutions at six different concentrations were prepared in a model wine. The model wines consisted of ethanol 12%, tartaric acid 0.6% in MilliQ water (purification system Millipore, Bedford, MA, USA) and their pH was adjusted to 3.5 using sodium hydroxide. 2-Octanol was used as an internal standard. 5 µL of the internal standard at a concentration 262 mg/L in ethanol was added to each standards solution. The SPME extraction and GC-MS analysis experiment for each standards solution were performed in triplicates to prepare their standard curves. The concentrations of the forty-five volatiles in five wines were calculated by the standard curves and expressed by mg/L.

Sensory analysis: The sensory analysis of the five wines was performed in a sensory laboratory set in accordance with the International Standard Organization ISO8589 (2007). A sensory panel consisted of ten members with a sharp sense of smell (5 men and 5 women, 23-35 years old) and was trained and done according to literature (Niu *et al*., 2011). Firstly, panelists generated descriptive terms for wines. Secondly, different aroma standards were discussed and distinguished by panelists. 12 sensory descriptors were analyzed for aroma quality and prepared according to a previous sensory study for red wines (Table 4) (Rutan *et al*., 2014). Thirdly, the wine were evaluated in duplicate using a nine-centimeter scale (0 = not perceived, 9 = extremely strong). An aliquot of 30 ml of each wine was poured in a 215 ml wine tasting glasses (Jackson, 2009). The panelists smelled the five wines in a random order, noted the specific sensory descriptors and rated the intensity of each sensory attribute on a nine-centimeter scale. The sensory analysis experiment was repeated in triplicate to calculate the average scores of the descriptors for each wine.

Statistical analysis: The general chemical data and sensory analysis results of the red wines were examined using the software of SAS version 8 (SAS Institute Inc, Cary, NC, USA) with ANOVA. Duncan's multiple range tests were applied to ascertain a significant difference at $p<0.05$ between the same parameter and sensory attribute. To explore the correlations between the sensory characteristics of the wines and their aroma profiles, PLSR was performed by using Unscrambler version 9.7 (CAMO ASA, Olso, Norway).

RESULTS AND DISCUSSION

Aroma compounds in the red wines: A total of 45 volatile compounds in the five wines were identified and classified into 7 groups, namely esters, alcohols, acids, aldehydes, ketones, volatile phenols and terpenes (Table 2). Esters are an important group of volatile compounds in red wine and generally formed by the esterification of alcohols and acids and their formation in red wine was mainly influenced by the composition of musts and conditions during fermentation (Perestrelo *et al*., 2006). Fourteen esters were identified in the five wines, with B and D having higher total amount of esters compared to the other wines (Table 2). Ethyl acetate, which is associated with a pleasant pineapple aroma, was dominant in this class of volatiles. The highest content was in B (25.05 mg/L), followed by D (20.99 mg/L), C (17.03 mg/L), A (16.90 mg/L) and E (14.79 mg/L). Ethyl lactate, a fruit and lactic aroma, was higher in B and D (>1 mg/L) than in A, C and E (<1 mg/L). B also had the highest concentration of ethyl octanoate (2.32 mg/L) associated with pineapple aroma, ethyl laurate (0.41 mg/L) associated with leaf aroma, ethyl hexanoate (0.48 mg/L) associated with apple aroma, isoamyl acetate (0.22 mg/L) associated with banana aroma and isobutyl acetate (0.06 mg/L) associated with apple or banana aroma. Moreover, it was reported that diethyl succinate associated with a pleasant fruit aroma was the main volatile compound presenting in Portugieser and Kekfrankos red wines (Ivanova *et al*., 2013). However, in this study, diethyl succinate had a lower concentration which was between 0.12 and 0.32 mg/L in the five wines compared with other ester compounds. Furthermore, ethyl valerate (0.02 mg/L) associated with yeast and apple aroma was only found in E, while fruity aroma ethyl tetradecanoate (1.26 mg/L) and neryl acetate (1.03 mg/L) were only detected in B.

In Table 3, 7 alcohols were detected in the five wines with concentrations ranging from 95.71 mg/L in C to 145.74 mg/L in B. The total concentration below 300 mg/L in red grape wines had an positive impact on the wines aroma and flavor and these alcohols are mainly produced during the yeast metabolism (Rapp and Versini, 1996). 1-Propanol, a ripe fruity alcohol aroma, had the highest concentration in B (9.87 mg/L) and the lowest in C (1.20 mg/L). The highest concentration of 3-methyl-1-butanol associated with banana fragrance was in B (114.18 mg/L) and followed by E (108.54 mg/L). In addition, the concentration of 2-phenylethanol (rose aroma), which was a fusel alcohol resulted from yeast metabolism during the alcoholic fermentation, in D and E was higher than that in A, B and C. Pungent smell alcohol of 2-propanol was only found in C. In general, the alcohols were the largest group of aroma compounds and accounted for more than a half of the total volatile constituents of the wines.

Three aldehydes, which can have a significant impact on wine aroma, namely acetaldehyde, furfural and benzaldehyde, were detected in the wines (Table 2). Acetaldehyde was higher in A (1.65 mg/L) and C (1.83 mg/L). Acetaldehyde at a low concentration provided a pleasant fruity aroma to wine, but turned to a pungent irritating odor reminiscent of green grass or apples at higher levels (Liu and Pilone, 2000). In addition, five ketones were also identified in the five wines. The ketone 3-hydroxy-2-butanone responsible for butter and cream notes was the most ubiquitous ketone in all the five wines. In particular, gamma-butyrolactone (5.03 mg/L) associated with sweety aroma was detected in E. Furthermore, seven volatile acids in the wines have been reported to be responsible for wine flavors. However, only hexanoic acid (green), octanoic acid (sweat, cheese) and butyric acid (sweaty) have significant influence on the wine aroma and their OAV were above 1. Acetic acid (sour) was particularly high in C and reached 17.18 mg/L and the content of it in other samples was all lower than 2 mg/L. Propionic acid (pungent, rancid) was detected in E. Nevertheless, the OAV of both acetic acid and propionic acid was below 1.

The highest phenol content was found in A and followed by C (Table 2). 4-Vinylguaiacol has been reported to have clove and spicy aroma and originates from the decarboxylation of the non-flavonoid compound ferulic acid during fermentation (Chatonnet *et al*., 1993). 4-Vinylguaiacol was present in the highest amount in C and A wines (1.01 mg/L), but not detected in B. 4-Ethylphenol and 4-ethylguaiacol are believed to contribute to smoke, plastic, burnt plastic, cow dung and barnyard aromas (Galafassi *et al*., 2011; López *et al*., 2002). Suárez *et al*. (2007) concluded that the decarboxylation of a number of phenolic acids in grape such as p-coumaric acid and ferulic acid could form 4-vinyl phenol and 4-vinylguaiacol respectively by hydroxy cinnamic acid enzyme. The deoxidation reaction of those hydroxyl styrene substances produced 4-ethylphenol and 4-ethylguaiacol by vinyl phenol reductase. Furthermore, five terpenes were found in the wines: Geraniol (rose aroma), β-citronellol (rose aroma), linalool (lavender aroma), nerolidol (floral aroma) and nerol (sweet aroma) (Van Gemert, 2003). The aromatic monoterpenes were formed from the precursor mevalonate, a metabolite derived from acetyl-CoA (Styger *et al*., 2011). These monoterpenes in red wines were odorless and found in glycoside bound forms in grape berries associated with their maturity (Fenoll *et al*., 2009; Palomo *et al*., 2007).

Sensory characteristics of the red wines: There were significant differences for all of the 12 sensory descriptors ($p<0.05$) used to describe the aroma perception (Table 3). In Table 4, B had the highest intensities for odor intensity (8.53), orchard fruity (5.97), dried fruity (7.46), nuts (5.83) and jammy/lolly (7.14). E had the highest intensities for red fruity (7.17), green/vegetal (6.03) and citrus fruity (4.62) and the higher intensities for dried fruity (7.05), odour intensity (7.87) and floral aroma (8.03). Styger *et al*. (2011) found that odor intensity was dependent on a high concentration of alcohols and their types. B and E wine had high concentration alcohols of 145.74 and 141.78 mg/L, respectively, which may contribute to their high value of odour intensity. D had the highest score in the floral descriptor (8.85). Escudero *et al*. (2007) reported that the fruity esters conferred fruity aroma to wines and norisoprenoids compounds enhanced the fruity notes of premium red wine. González Álvarez *et al*. (2011) found that fruity and floral aromas (floral, apple and citrus) and herbaceous notes had the highest intensity in Godello wines.

The results for the aroma descriptors in Table 3 were further analyzed using a PLSR method. Figure 1 shows the relationships between the sensory aroma descriptors and the five wines. The sensory characteristics of A and C were close and similar in the spice and undesirable aromas, i.e., phenol-like flavor. Escudero *et al*. (2007) observed that these aroma were related to the presence of phenol compounds in red wines. B was prominent in the jammy/lolly, odour intensity, dried fruity, nuts and orchard fruity (Fig. 1 and Table 2). The aroma of citrus fruity and red fruity was mainly found in E. It also had green/vegetal and floral aroma. D showed a strong correlation with floral aroma (Fig. 1). However, the five wines of this study did not present toasted aroma.

Correlations of sensory characteristics and aroma profiles in the red wines: To evaluate the effects of the specific aroma compounds in the five wines on their overall wine sensory characteristics, OAV values of the aroma compounds were calculated and are listed in Table 4. Among the 26 esters, there were 11 esters whose OAV>1. Particularly, ethyl hexanoate (OAV = 34.29) detected in B, had the highest OAV among the 11 esters. Ethyl decanoate (OAV = 13.2) and ethyl butyrate (OAV = 10.6) had the highest OAV value in E compared with others. Acetaldehyde (OAV = 18.3), butyric acid (OAV = 12.0) and 4-vinylguaiacol (OAV = 25.2) in C were the highest OAV among the aldehydes, acids and phenols. For terpenes, only nerol was identified in B at a very high OAV of 17.4. Beekwilder *et al*. (2014) found that beta-ionone was produced from the cleavage reaction of precursor beta-carotene which is catalyzed by carotenoid cleavage dioxygenase enzyme. It had fruity odour and was found in arabidopsis, rose, raspberry and other plant species. beta-Ionone only occurred in E and had the highest OAV value (OAV = 58.9) among the 26 aroma compounds of OAV>1.

The PLSR was performed to examine correlations between the aroma descriptors and the aroma profiles. The aroma descriptors in Table 4 and the 26 aroma compounds (OAV>1) in Table 4 served as the X and Y variables, respectively. In Fig. 2, the PLSR method provided a two-factor model, which had 84% of the variance in X (sensory descriptors) and 93% of that in Y (aroma compounds of OAV >1). It was suggested that the spice descriptor located in the leftmost of PC1 in Fig. 2 was positively correlated with 4-vinylguaiacol (N25), 4-ethylguaiacol (N24) and geranyl acetone (N17). The undesirable aroma positively correlated with 4-ethylphenol (N23), which is responsible for off-odor such as plastic smell of some wines (Amoore and Hautala, 1983). In the rightmost of PC1 in Fig. 2, the red fruity aroma was contributed by β-ionone (N18) and γ-butyrolactone (N4) which occurred only in E. The citrus aroma had a positive correlation with ethyl valerate (N8) which was only detected in E as well (Table 2 and Fig. 2). In Fig. 1, B was characterized by the aromas close to orchard fruity, nuts, odour intensity, dried fruity and jammy/lolly which were contributed by the following aroma compounds: ethyl tetradecanoate (N7), ethyl hexanoate (N9), isoamyl acetate (N11), ethyl acetate (N1), ethyl octanoate (N2), 1-propanol (N12), 3-methyl-1-butanol (N13) and nerol (N15) in Fig. 2. Floral aroma was also a typical aroma in several red wines (Snitkjær *et al*., 2011) and strongly correlated with 2-phenylethanol (N14) and geraniol (N26). Hence, the correlations between the sensory characteristics and aroma profiles of red wine were visually revealed by the PLSR method.

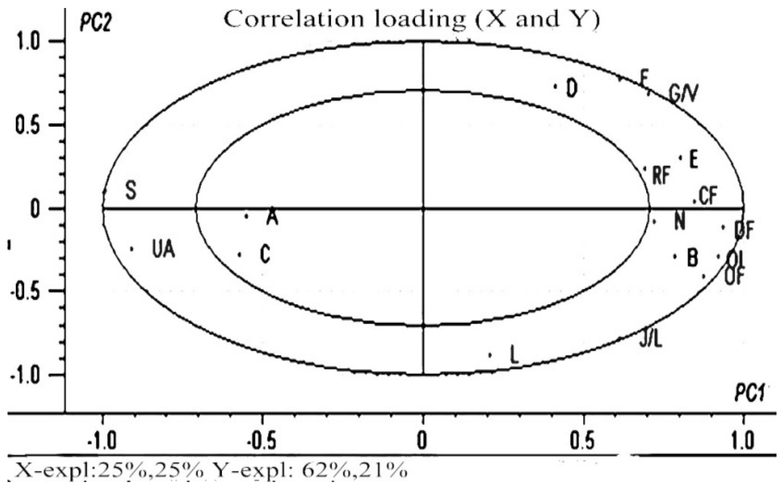

Fig. 1: Two-dimensional partial least squares regression (PLSR) of A, B, C, D and E wines and sensory descriptors OI: Odour Intensity; CF: Citrus Fruit; RF: Red Fruit; G/V: Green/Vegetal; DF: Dried Fruit; L: Lactic; N: Nuts; S: Spice; F: Floral; OF: Orchard fruit; J/L: Jammy/Lolly; UA: Undesirable aroma

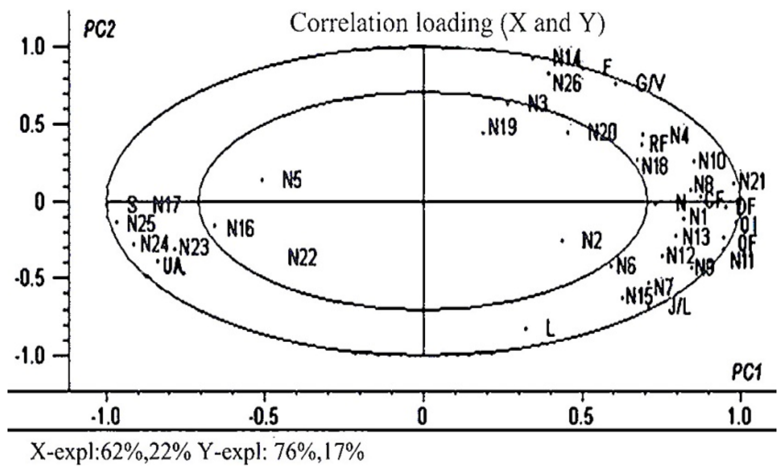

Fig. 2: Two-dimensional partial least squares regression (PLSR) of sensory descriptors and aroma compounds whose OAV > 1 OI: Odour intensity; CF: Citrus fruit; RF: Red fruit; G/V: Green/Vegetal; DF: Dried fruit; L: Lactic; N: Nuts; S: Spice; F: Floral; OF: Orchard fruit; J/L: Jammy/Lolly; UA: Undesirable aroma. N1: Ethyl acetate; N2: Ethyl octanoate; N3: Ethyl decanoate; N4: γ-Butyrolactone; N5: 2-Phenylethyl acetate; N6: Ethyl laurate; N7: Ethyl tetradecanoate; N8: Ethyl valerate; N9: Ethyl hexanoate; N10: Ethyl butyrate; N11: Isoamyl acetate; N12: 1-Propanol; N13: 3-Methyl-1-butanol; N14: 2-Phenylethanol; N15: Acetaldehyde; N16: 3-Hydroxy-2-butanone; N17: Geranyl acetone; N18: β- Ionone; N19: Hexanoic acid; N20: Octanoic acid; N21: Butyric acid; N22: 4-Ethylphenol; N23: 4-Ethylguaiacol; N24: 4-Vinylguaiacol; N25: Geraniol; N26: Nerol

To compare with the PLSR method, a ROC method was used to further identify the contribution of each individual compound to the overall aroma of the wines (Table 4). For A, also 4-vinylguaiacol has high ROC, higher than 4-ethylguaiacol. Although only 19 volatiles (OAV>1) occurred in B, the fruity aroma of ethyl hexanoate displayed the greatest contribution to B and reached to 31.1% of ROC. Nerol (ROC = 15.8%) can be linked to agreeable floral aroma in B. 4-ethylguaiacol (ROC = 20.4%) occupied the highest percentage and ethyl hexanoate (ROC = 16.7%), acetaldehyde (ROC = 14.8%) and butyric acid (ROC = 9.7%) accounted for a relatively high percentage of ROC in C. Ethyl hexanoate (ROC = 27.7%), ethyl decanoate (ROC = 9.8%), acetaldehyde (ROC = 9.8%) and geraniol (ROC = 7.1%) were present prominently in D. It was noted that β-ionone (ROC = 36.4%) was a particular aroma compound only found in E and offered the highest contribution to the E aroma, whereas ethyl decanoate, ethyl hexanoate, ethyl butyrate and isoamyl acetate also made contributions to this wine. The ROC method successfully identified the contribution percentage of a particular aroma compound to the overall aroma of wines. Compared with the PLRS method, the ROC method was not able to straightforwardly demonstrate the correlations between the sensory characteristics and aroma profiles of the red wines and the relationships between the sensory descriptors and their associated aroma compounds (Wang *et al.*, 2015).

CONCLUSION

The purpose of this study was to find the aroma compounds in five different Chinese grape wines and determine the relationship between the aroma profiles and sensory characteristics of these wines using PLSR method. Firstly, a total of 45 volatile compounds in these wines were identified and quantified by HS-SPME/GC-MS. Furthermore, OAV of each detected volatile compound was calculated and found 26 kinds of the aroma compounds whose OAV > 1 in part of the samples. All these aroma compounds with OAV > 1 were selected for evaluating the correlations between the aroma profiles and sensory characteristics of the wines using PLSR and their ROC.

Based on the results of PLSR and ROC methods, it was found that ethyl decanoate, ethyl hexanoate, acetaldehyde, isoamyl acetate, hexanoic acid, 4-vinylguaiacol and geraniol were the major contributors to the desirable balanced aroma of D (*muscat* wine). Ethyl hexanoate, ethyl butyrate, isoamyl acetate, acetaldehyde, hexanoic acid, 3-methyl-1-butanol and octanoic acid were mainly responsible for the aroma of B (*black beet* wine) or E (*cabernet genischt* wine), whereas ethyl tetradecanoate, neryl acetate and nerol were the particular aroma compounds in *black beet* wine; and γ-butyrolactone, nerolidol and β-ionone were special aroma compounds in *cabernet genischt* wine. Both the PLSR method and the ROC method are effective methods to demonstrate the correlations between the sensory characteristics of red wines and their aroma compositions visually and integrally but the PLSR method were more straightforwardly than the ROC method.

The results obtained in this study demonstrated that there are close correlations between aroma profiles and sensory characteristics of red wines in different grape varieties and the correlations are significant in different grape varieties. This conclusion will make fix a standard to divide different red wines more easily and both PLSR method and ROC are good ways to build the correlations between aroma profiles and sensory characteristics of red wines in different grape varieties.

ACKNOWLEDGMENT

This research was financially supported by the Key Projects of Technology Innovation of Shanghai Education Committee (12ZZ187).

Conflict of interest: The author reports no conflicts of interest in this study.

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