

## CHANGES IN THE PROFILE OF AROMA COMPOUNDS IN *VITIS VINIFERA* L. CV MERLOT FROM GRAPES TO WINE

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### Abstract

The volatile compounds that provide the aroma and flavor to wine are the products of several influences, namely grape cultivar, geographic location and associated environmental features, viticultural practices, and vinification techniques. Understanding those influences on the volatile aroma profile of wine is central to production protocols and quality evaluation. That knowledge is specially important for developing wine regions. Accordingly, we initiated the requisite studies in the emergent geographic wine region, Helan Mountain, in Ningxia, China, by determining the composition of volatile aroma compounds of dry red wine produced from *V. vinifera* cv Merlot after pre-fermentation maceration of the grapes and after fermentation. The volatile compounds of the test samples were obtained by solvent extraction and analyzed by GC-MS. Their relative contents were determined by area normalization. The detection rates accounted for 99.98 and 100.50% of the total peak areas, respectively. From the grape macerate, we separated 47 volatile compounds and identified 44. From the wine, we separated 69 compounds and identified 64. In the berry macerate, the volatile compounds with highest relative content, i.e. proportion of total peak area, included fatty acid esters, fatty alcohols, fatty acids, aldehydes and hydrocarbons. In the wine, fatty acids, fatty acid esters, fatty alcohols, aliphatic ketones and heterocyclic compounds, such as 3-methyl-1-butanol and ethyl acetate predominated. The relative content of major volatile aroma compounds in the berry macerate and the wine were similar, revealing a strong influence of the fruit. These results provide a benchmark for further comparative studies on the influence of geographic location, viticultural practices and fermentation technology on the flavor and aroma qualities of dry red Merlot wine.

### Introduction

The wine grape cultivar, Merlot, (*Vitis vinifera* L.) originally from France, was introduced to China in the 1980s, and is now widely cultivated there in the eastern base of the Helan mountains in Ningxia Hui Autonomous Region. The red wine produced from Merlot is becoming popular among consumers for its attractive ruby red color, fruity flavor, and appealing taste with a typically 'soft' tannin texture (Chira *et al.* 2011). The main aromas of Merlot dry red wine are reported as tropical and temperate fruit fragrances, such as those derived from bananas, pineapples, pears, green apples and strawberries, as well as butter and honey-like aromas derived from malolactic fermentation (Coldea *et al.* 2011).

Among fermented food products, the flavor of wine is one of the most complex; being dependent on volatile compounds originating from, or influenced by, one or more different grape cultivars (Hu *et al.* 2015), geographic location (Ziółkowska *et al.* 2016) and associated environmental features, viticultural practices, and vinification techniques (Martí *et al.* 2004, Setkova *et al.* 2007), including various strains of yeast and bacteria involved in fermentation, fermentation process variables, ageing and storage. Those compounds are perceived by smelling, as aroma, and retronasally, as flavor. Their type, proportion, content, sensory threshold and

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interaction greatly influence wine style and are important in the evaluation of wine quality (Griffin *et al.* 2006, Hale *et al.* 2006). However, there are a few reported comparative studies of aromatic components of cv. Merlot grapes and dry red wine produced from them (Faustino *et al.* 2003)

Since the late 1990s, the eastern part of the Helan Mountain in Ningxia has been developing as a wine region because of its favourable climate and irrigation capacity. In this study, Merlot, one of the main wine grape cultivars of the region, was chosen as the test material. By first identifying the volatile compounds in the grape macerate ('must'), and the subsequent contribution of those derived from primary fermentation, we aimed at establishing a sound foundation for the systematic examination of the relative effects of the many sources of variation in the quality of dry red Merlot wines.

### Materials and Methods

Merlot grapes were obtained from ten-year-old, non-grafted plants in the eastern Helan Mountains, Ningxia. The plants were spaced 0.5 m apart in rows 3 m wide and trained to a single wire trellis (Hu *et al.* 2015). Fully ripe grapes were harvested by hand from September 20 - 22, 2014, de-stemmed and crushed into 30,000 litre tanks. After 48 hrs aerobic maceration of pulp and skin, the juice was sampled from the tanks and centrifuged at 1000 to 1200 r/min to obtain 200 ml clarified samples (Ferreira *et al.* 2000). The physicochemical indices of the juice were: 22.9% total soluble solids, 210.3 g/l reducing sugar, 6.2 g/l titratable acid, 0.54 g/l total phenols and pH 3.10.

The wine was made by the standard wine process using the following techniques: De-stemming and crushing, addition of 50 mg/l SO<sub>2</sub> and RC212 yeast, fermentation at 25°C for 8 - 9 days followed by gentle pressing the pomace. Fermentation was completed at the end of September. No fining agents were used. Malolactic fermentation did not occur and the wines were stored in the tanks until analyses began in March 2016. The general wine components were: 12.0% (v/v) alcohol, 2.06 g/l residual sugar, 5.2 g/l titratable acid, 3.22 pH, 23.5 g/l extract, 24.7 mg/l free SO<sub>2</sub> and 825 mg/l total phenols.

Analysis of the volatile components was conducted in the Testing and Analysis Centre of Yangzhou University in China. The volatile components were obtained by continuous liquid-liquid extraction. Pre-ferment samples of 200 ml grape juice and post-ferment samples of 350 ml wine were transferred to 500 ml spherical flasks and extracted with 100, 80 and 60 ml of freshly redistilled dichloromethane three times separately. The three organic phases were combined and concentrated to 1 ml for GC-MS using a rotary evaporator (0 - 5°C) (Ferreira *et al.* 1996). The extracts were then analyzed, in duplicate, with a GC-MS system.

The Hewlett-Packard 6890 gas chromatograph was used in the analysis consisted of a detector (model HP 5972) and a HP WAX: 30 m × 0.25 μm × 0.25 mm, column). Samples were injected in split mode (50 : 1), and the components were separated using GC-grade helium as the carrier gas at a constant flow rate of 1 ml/min. Temperature-programming was conducted as follows. When the column temperature reached 60°C the temperature was maintained for 5 min then the temperature of the oven was raised to 110°C for 2 min. The injector temperature was set at 250°C (mass analyzer) and held for 30 min and then it was gradually increased to 240°C at 5°C/min. The detector temperature was set at 280°C and maintained for 30 min and then it was raised to 270°C at 10°C/min.

The different parameters involved in the operation of the mass spectrometer, were also standardized as follows: Inlet line temperature and source temperature were both 200°C. Mass spectrometry energy was 50 eV and electric impact mode was set at 1800 V.

By comparing the retention time and percentage of average peak area to total peak area constituent compounds were identified and ascertained by reference to the NBS/WILEY standard library of HP MSD Chem-Station and related literature. GC-MS total ion chromatograms of aroma components in Merlot grape and dry red wine were plotted using HP MSD Chem-Station.

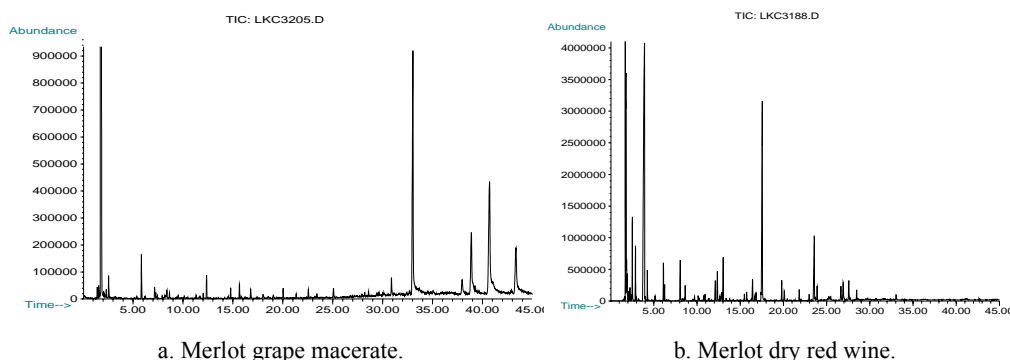


Fig. 1. GC-MS total ion chromatograms of volatile compounds in Merlot grape macerate and wine.

**Table 1. Aroma compounds identified in Merlot grape macerate and dry red wine.**

No.	Aroma compound	Molecular formula	Grape macerate	Dry red wine
			Relative content (%)	Relative content (%)
1	2-butenal	C <sub>4</sub> H <sub>6</sub> O	0.09	
2	3-buten-2-one, 3-methyl-	C <sub>5</sub> H <sub>8</sub> O	0.19	
3	3-buten-2-ol, 2-methyl-	C <sub>5</sub> H <sub>10</sub> O	0.08	
4	2-butenal*	C <sub>4</sub> H <sub>6</sub> O	0.51	
5	Hexanal	C <sub>6</sub> H <sub>12</sub> O	1.33	
6	3-penten-2-ol	C <sub>5</sub> H <sub>10</sub> O	0.10	
7	Butanol, 3-hydroxy-	C <sub>4</sub> H <sub>8</sub> O <sub>2</sub>	0.09	
8	1-butanol, 3-methyl-	C <sub>5</sub> H <sub>12</sub> O	0.09	36.81
9	1-butanol, 3-methyl- *	C <sub>5</sub> H <sub>12</sub> O	0.04	
10	Cyclohexanone	C <sub>6</sub> H <sub>12</sub> O	0.05	
11	Cyclopentane, 1-ethyl-1-methyl-	C <sub>8</sub> H <sub>16</sub>	0.19	
12	2-heptenal, (E)-	C <sub>7</sub> H <sub>12</sub> O	2.79	
13	1-hexanol	C <sub>6</sub> H <sub>14</sub> O	0.14	0.67
14	Nonanal	C <sub>9</sub> H <sub>18</sub> O	0.81	
15	2-hexen-1-ol, (E)	C <sub>6</sub> H <sub>12</sub> O	0.40	
16	2-octenal, (e)-	C <sub>8</sub> H <sub>14</sub> O	0.19	
17	1-octen-3-ol	C <sub>8</sub> H <sub>15</sub> O	0.14	
18	Butane, 2,3-dimethyl-2-nitro-	C <sub>6</sub> H <sub>13</sub> O <sub>2</sub> N	0.58	
19	2,4-heptadienal, (E,E)-	C <sub>7</sub> H <sub>10</sub> O	0.32	
20	2,4-heptadienal, (E,E)- *	C <sub>7</sub> H <sub>10</sub> O	0.04	
21	2-nonenal, (E)-	C <sub>9</sub> H <sub>16</sub> O	0.23	

(Contd.)

(Table 1 contd.)

22	2-nonenal, (e)- *	C <sub>9</sub> H <sub>16</sub> O	0.20	
23	1-octanol	C <sub>8</sub> H <sub>18</sub> O	0.07	
24	2-cyclohexen-1-one,3,5,5-trimethyl-	C <sub>9</sub> H <sub>14</sub> O	0.21	0.15
25	2-octen-1-ol	C <sub>8</sub> H <sub>16</sub> O	0.07	
26	2(3H)-furanone, dihydro-	C <sub>4</sub> H <sub>6</sub> O <sub>2</sub>	0.34	1.19
27	2-decenal, (e)-	C <sub>10</sub> H <sub>18</sub> O	1.45	
28	2,4-decadienal, (e,e)-	C <sub>10</sub> H <sub>16</sub> O	0.62	
29	Hexanoic acid	C <sub>6</sub> H <sub>12</sub> O <sub>2</sub>	0.11	
30	Benzenemethanol	C <sub>7</sub> H <sub>8</sub> O	0.72	0.57
31	Benzeneethanol	C <sub>8</sub> H <sub>10</sub> O	0.12	16.21
32	1,10-decanediol	C <sub>10</sub> H <sub>22</sub> O <sub>2</sub>	0.53	
33	1,13-tetradecadiene	C <sub>14</sub> H <sub>26</sub>	0.42	
34	Octanoic acid	C <sub>8</sub> H <sub>16</sub> O <sub>2</sub>	0.62	
35	3-octyn-1-ol	C <sub>8</sub> H <sub>14</sub> O	0.73	
36	Hexadecanoic acid, ethyl ester	C <sub>18</sub> H <sub>36</sub> O <sub>2</sub>	0.37	5.60
37	5.beta., 8.beta.H, 9.beta.H, 10.alpha. -Labd-14-ene, 8,13-epoxy-	C <sub>20</sub> H <sub>32</sub> O	0.88	
38	Benzaldehyde, 4-hydroxy-3-methoxy-	C <sub>8</sub> H <sub>8</sub> O <sub>3</sub>	0.25	
39	Phytol	C <sub>20</sub> H <sub>37</sub> O	0.38	
40	Hexadecanoic acid	C <sub>16</sub> H <sub>32</sub> O <sub>2</sub>	32.24	
41	Octadecanoic acid	C <sub>18</sub> H <sub>36</sub> O <sub>2</sub>	2.59	
42	9-octadecenoic acid (Z)-	C <sub>18</sub> H <sub>34</sub> O <sub>2</sub>	11.22	
43	9,12-octadecenoic acid (Z,Z)-	C <sub>18</sub> H <sub>34</sub> O <sub>2</sub>	24.34	
44	11,14,17-eicosatrienoic acid, methyl ester	C <sub>21</sub> H <sub>36</sub> O <sub>2</sub>	10.20	
45	Unidentified		2.89	2.94
46	Acetic acid, ethyl ester	C <sub>4</sub> H <sub>8</sub> O <sub>2</sub>		6.93
47	Propanoic acid, 2-methyl-,ethyl ester	C <sub>6</sub> H <sub>12</sub> O <sub>2</sub>		0.44
48	Acetic acid, 2-methylpropyl ester	C <sub>6</sub> H <sub>12</sub> O <sub>2</sub>		0.24
49	1-Propanol	C <sub>3</sub> H <sub>8</sub> O		0.12
50	Butanoic acid, ethyl ester	C <sub>6</sub> H <sub>12</sub> O <sub>2</sub>		0.38
51	Butanoic acid, 2-methyl-, ethyl ester	C <sub>7</sub> H <sub>14</sub> O <sub>2</sub>		0.17
52	Butanoic acid, 3-methyl-, ethyl ester	C <sub>7</sub> H <sub>14</sub> O <sub>2</sub>		0.23
53	1-propanol, 2-methyl-	C <sub>4</sub> H <sub>10</sub> O		2.72
54	1-butanol, 3-methyl-, acetate	C <sub>7</sub> H <sub>14</sub> O <sub>2</sub>		2.63
55	1-butanol	C <sub>4</sub> H <sub>10</sub> O		0.15
56	Hexanoic acid, ethyl ester	C <sub>8</sub> H <sub>16</sub> O <sub>2</sub>		1.14
57	Acetic acid, propyl ester	C <sub>5</sub> H <sub>10</sub> O <sub>2</sub>		0.05
58	Acetic acid, hexyl ester	C <sub>8</sub> H <sub>16</sub> O <sub>2</sub>		0.04
59	Butanoic acid, 3-methyl-, 3-methylbutyl ester	C <sub>10</sub> H <sub>20</sub> O <sub>2</sub>		0.03
60	2-butanone, 3-hydroxy-	C <sub>4</sub> H <sub>8</sub> O <sub>2</sub>		0.21
61	Butanoic acid, 3-methyl-, 3-methylbutyl ester*	C <sub>10</sub> H <sub>20</sub> O <sub>2</sub>		0.53
62	1-pentanol, 3-methyl-	C <sub>6</sub> H <sub>14</sub> O		0.03

(Contd.)

(Table 1 contd.)

63	Propanoic acid, 2-hydroxy-, ethyl ester	C <sub>5</sub> H <sub>10</sub> O <sub>3</sub>	1.60
64	1,3-oxathiane	C <sub>4</sub> H <sub>8</sub> OS	0.05
65	1-propanol, 3ethoxy-	C <sub>5</sub> H <sub>12</sub> O <sub>2</sub>	0.04
66	Octanoic acid, ethyl ester	C <sub>10</sub> H <sub>20</sub> O <sub>2</sub>	2.21
67	Acetic acid	C <sub>2</sub> H <sub>4</sub> O <sub>2</sub>	0.14
68	Hexanoic acid, 2-methylbutyl este	C <sub>11</sub> H <sub>22</sub> O <sub>2</sub>	0.13
69	2-furancarboxaldehyde	C <sub>5</sub> H <sub>4</sub> O <sub>2</sub>	0.72
70	Ethanone, 1-(2-furanyl)-	C <sub>6</sub> H <sub>6</sub> O <sub>2</sub>	0.09
71	Butanoic acid, 3-hydroxy-, ethyl ester	C <sub>6</sub> H <sub>12</sub> O <sub>3</sub>	0.22
72	2,3-butanediol	C <sub>4</sub> H <sub>10</sub> O <sub>2</sub>	0.31
73	2-(3H) furanone	C <sub>8</sub> H <sub>16</sub> O <sub>3</sub>	0.15
74	2-pentanol	C <sub>5</sub> H <sub>12</sub> O	0.31
75	2-furancarboxaldehyde, 5-methy-	C <sub>6</sub> H <sub>6</sub> O <sub>2</sub>	0.39
76	Pentanoic acid, 4-oxo-, ethyl ester	C <sub>7</sub> H <sub>12</sub> O <sub>3</sub>	0.04
77	Decanoic acid, ethyl ester	C <sub>12</sub> H <sub>24</sub> O <sub>2</sub>	1.77
78	Octanoic acid, 3-methylbutyl ester	C <sub>13</sub> H <sub>26</sub> O <sub>2</sub>	0.30
79	Butanoic acid, 3-methyl-	C <sub>5</sub> H <sub>10</sub> O <sub>2</sub>	0.46
80	Butanoic acid, diethyl ester	C <sub>8</sub> H <sub>14</sub> O <sub>4</sub>	2.14
81	1-propanol, 3-(methylthio)-	C <sub>4</sub> H <sub>10</sub> OS	0.12
82	Formic acid, 2-phenylethyl ester	C <sub>9</sub> H <sub>10</sub> O <sub>2</sub>	0.04
83	Pentanoic acid, ethyl ester	C <sub>7</sub> H <sub>14</sub> O <sub>2</sub>	0.33
84	Acetic acid, 2-phenylethyl ester	C <sub>10</sub> H <sub>12</sub> O <sub>2</sub>	0.43
85	Hexanoic acid	C <sub>6</sub> H <sub>18</sub> O <sub>2</sub>	0.08
86	Dodecanoic acid, ethyl ester	C <sub>14</sub> H <sub>28</sub> O <sub>2</sub>	1.37
87	1-(2-Furyl)-2-hydroxyethanone	C <sub>6</sub> H <sub>6</sub> O <sub>3</sub>	0.08
88	Butanedioic acid, hydroxy-, diethyl ester	C <sub>8</sub> H <sub>14</sub> O <sub>5</sub>	0.96
89	Tetradecanoic acid, ethyl ester	C <sub>16</sub> H <sub>32</sub> O <sub>2</sub>	0.70
90	Octanoic acid, 3-methylbutyl ester	C <sub>13</sub> H <sub>26</sub> O <sub>2</sub>	0.18
91	Thiazole	C <sub>3</sub> H <sub>3</sub> SN	0.52
92	2-Butenoic acid,(Z)-	C <sub>4</sub> H <sub>6</sub> O <sub>2</sub>	0.32
93	Benzeneacetic acid, 2-propenyl ester	C <sub>11</sub> H <sub>12</sub> O <sub>2</sub>	0.18
94	Octadecanoic acid, ethyl ester	C <sub>20</sub> H <sub>40</sub> O <sub>2</sub>	1.20
95	9-octadecenoic acid(Z)-,ethyl ester	C <sub>20</sub> H <sub>38</sub> O <sub>2</sub>	1.58
96	9,12,15-octadecatrienoic acid, methyl ester,(Z,Z,Z)-	C <sub>19</sub> H <sub>32</sub> O <sub>2</sub>	0.71
97	Hexadecanoic acid, ethyl ester	C <sub>16</sub> H <sub>36</sub> O <sub>2</sub>	0.44

## Results and Discussion

The volatile compounds that appear after grape berries are crushed include compounds that occur naturally in the fruit, and others that result from the breakdown of internal compartmentation of the berry (Hardie *et al.* 1988), the latter group include compounds those are released on hydrolysis of glyconjugated forms. Subsequently, during the fermentation process, the yeast and possibly other microorganisms convert sugar, organic acids, flavor precursors and other ingredients into alcohols, esters and other volatile products. Hydrolysis of glycoconjugated aroma

volatiles also continues to contribute to wine aroma and flavour development. Present authors examined the volatiles formed during 48 hrs of aerobic maceration of crushed Merlot berries and those formed during fermentation and in the seven months thereafter.

Fig. 1 presents the total ion chromatograms of the volatile compounds obtained from the Merlot grape macerate and the wine. A total of 96 known compounds were obtained. From the grape macerate, 47 compounds were detected and 44 of them were identified. The detection rate was 99.98%. From the dry red wine, 69 compounds were detected and 64 of them were identified. Overall the identified constituents of the wine represented 100.50% of the total peak area.

The compounds identified in Merlot grape macerate and dry red wine are shown in Table 1. The main volatile components of the grape must were hexadecanoic acid (32.24%), 9,12 - octadecenoic acid (Z,Z)- (23.34%); 9-octadecenoic acid (Z)- (11.22%); 11,14,17-eicosatrienoic acid, methyl ester (10.20%), 2-heptenal (E) (2.79%), octadecanoic acid (2.59%), 2-decenal,(E)- (1.45%), hexanal (1.33%). These compounds were carboxylic acids, aldehydes, and esters. However, unlike other wine grape cultivars from same, e.g. Cabernet Gernischt and Chardonnay, (Jiang *et al.* 2010, Wondra *et al.* 2001), there were no heterocyclic compounds found in the Merlot grape macerate, and the reason might lie in the difference of maceration time. It was reported that the contents of 2-furancarboxaldehyde and 5-methyl-2-furaldehyde were very high in Merlot must (Cerdán *et al.* 2004), but they were not detected in our study. Since both compounds are mainly derived from oak (Cerdán *et al.* 2006), authors would not expect to find them in large amounts, if at all, except in oak-treated musts.

During fermentation and storage, the number and type of aroma compounds in the wine increased significantly. The newly formed compounds included esters, alcohols, carboxylic acids, ketones, aldehydes, hydrocarbons, heterocyclic, sulfur-containing compounds, nitrogen-containing compounds, ethers and phenols. Their formation is largely attributable to the action of yeasts under acidic and anaerobic fermentation conditions (Cliff *et al.* 2002). Specifically, the main compounds detected in the wine were: 3-methyl-1-butanol, (36.81%); benzeneethanol (16.21%), acetic ether (6.93%); hexadecanoic acid, ethyl ester (5.60%); 2-methyl-1-propanol (2.72%); acetic acid 3-methylbutyl ester (2.63%), ethyl caprylate (2.21%), diethyl succinate (2.14%), ethyl caprate (1.78%), ethyl lactate (1.60%). Following fermentation and seven months storage, esters and alcohols were the main aroma compounds. Aldehydes, ethers and hydrocarbons were not detected in the wine.

Table 1 shows that Merlot macerate and dry red wine had seven volatile compounds in common. They were 1-butanol, 3-methyl-, 1-hexanol, 2-cyclohexen-1-one,3,5,5-trimethyl-, 2(3H)-furanone: dihydro-, benzenemethanol, benzeneethanol, hexadecanoic acid and ethyl ester. 0

Table 2 compares the number of volatile compounds of each major class in the grape macerate with that of the wine. It can be clearly seen from Table 2 that alcohols and esters increased during the fermentation and storage while carboxylic acids decreased. Changes in the relative contents of the volatile compounds in each stage are displayed in Fig. 2.

As expected, in present study, during fermentation, the number and amount of esters relative to the total volatiles, increased greatly, especially aliphatic esters, their number increased from 2 to 30 and their relative content rose from 10.57 to 33.94%. Moreover, three aromatic esters, viz. prop-2-enyl 2-phenylacetate, 2-phenylethyl ester acetate and 2-phenylethyl formate which were not found in the grape macerate, were formed during fermentation and storage. In addition, at sampling after fermentation, the relative content of ethyl acetate had increased to 6.93%. Recent research found that ethyl acetate was also the predominant aromatic component of Merlot dry red wine, which had the aroma of green apples and bananas (Lilly *et al.* 2000). The aroma of ethyl caprylate is perceived as floral and fruity, while ethyl caprylate smells like brandies and grapes

(Peinado *et al.* 2004). The esters constituted the main aroma constituents of the Merlot wine. Lactone - cyclic esters of hydroxycarboxylic acids which usually have fruity flavors - was found before and after fermentation.

**Table 2. Classification of aromatic compounds in Merlot grape macerate and dry red wine.**

Aroma compound category		Grape macerate		Dry red wine	
		Kinds	Relative content (%)	Kinds	Relative content (%)
Esters	Aliphatic esters	2	10.57	30	33.94
	Aromatic carboxylic esters	nd	nd	3	0.65
	Lactones	1	0.34	1	1.19
	<b>Total</b>	<b>3</b>	<b>10.91</b>	<b>34</b>	<b>35.78</b>
Alcohols	Aliphatic alcohol	12	2.77	10	41.27
	Aromatic alcohol	2	0.84	2	16.77
	<b>Total</b>	<b>14</b>	<b>3.61</b>	<b>12</b>	<b>58.04</b>
Carboxylic acids	Aliphatic carboxylic acids	6	71.12	5	1.44
	<b>Total</b>	<b>6</b>	<b>71.12</b>	<b>5</b>	<b>1.44</b>
Ketones	Aliphatic ketone	3	0.45	4	0.53
	<b>Total</b>	<b>3</b>	<b>0.45</b>	<b>4</b>	<b>0.53</b>
Aldehydes	Aliphatic aldehyde	14	8.93	nd	nd
	aromatic aldehyde				
	<b>Total</b>	<b>14</b>	<b>8.93</b>	<b>nd</b>	<b>nd</b>
Hydrocarbons		3	1.49	nd	nd
	<b>Total</b>	<b>3</b>	<b>1.49</b>	<b>nd</b>	<b>nd</b>
Heterocyclic compounds	Furan	nd	nd	2	1.15
	Thiazole	nd	nd	1	0.52
	<b>Total</b>	<b>nd</b>	<b>nd</b>	<b>3</b>	<b>1.67</b>
Sulfur or nitrogen containing compounds	Sulfur containing compound	nd	nd	1	0.05
	Nitrogen containing compound	1	0.58	nd	nd
	<b>Total</b>	<b>1</b>	<b>0.58</b>	<b>1</b>	<b>0.05</b>
Unidentified	<b>Total</b>	<b>3</b>	<b>2.89</b>	<b>5</b>	<b>2.94</b>
<b>Total</b>		<b>47</b>	<b>99.98</b>	<b>69</b>	<b>100.50</b>

nd = Not detected.

Compared with macerate of Merlot grape, the relative content of alcohols in the wine was increased during fermentation and storage (Table 2). Low saturated fatty alcohols, aliphatic alcohols and aromatic alcohols increased in this period. By the end of fermentation, of all the alcohols, the relative content of aliphatic alcohols (including ethyl alcohol) was 41.27%; nearly half that of all the volatile aroma compounds. During the same period, lower unsaturated fatty alcohols that were present in the macerate transformed and disappeared. In wine production, alcohols are synthesized by yeast action via two mechanisms, viz. the anabolic pathway of glucose,

and catabolic pathway of corresponding amino acids, such as valine, iso-leucine, leucine and phenylalanine (Jiang *et al.* 2013). Alcohols contribute greatly to the primary aroma of wine (Rodríguez *et al.* 2008). Notably, the relative content of the aromatic alcohol, phenethyl alcohol - which as a rose and honey-like aroma (Chen *et al.* 2010) was 16.21%.

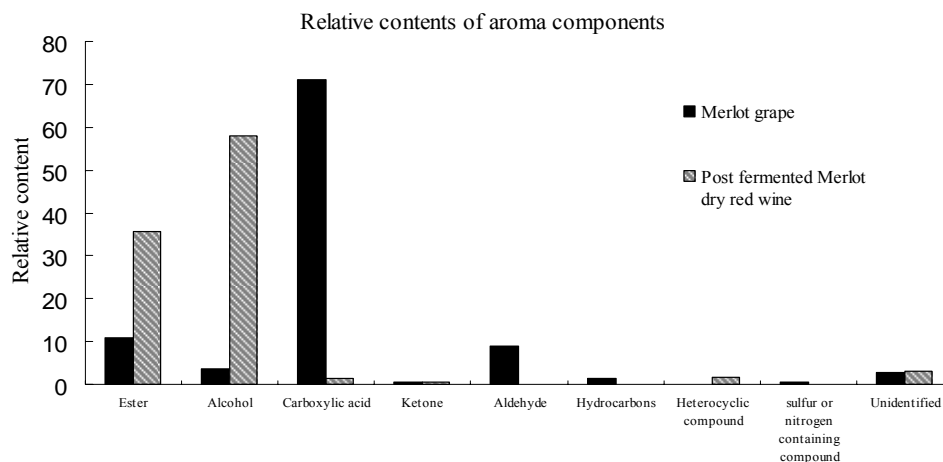


Fig. 2. Relative contents of aroma components in Merlot grape macerate and wine.

**Esters:** As a group, esters are the next major constituents in wine after water, ethanol and fusel alcohols (Etiévant 1991, Sumby *et al.* 2010), and are the primary source of fruity aromas (Gurbuz *et al.* 2006). They are formed when alcohols and carboxylic acid functional groups react, and a water molecule is eliminated. Relatively a few volatile esters have been found in grape juice. They are predominantly formed in wine where, individually, they are frequently in trace amounts, such that they are often at or below aroma threshold concentrations, and collectively they generally do not exceed concentrations of 100 mg/l.

The relative content of carboxylic acids in the grape macerate was 71.12%, four higher fatty acids accounting for 70.39% of the total and two lower fatty acids, accounting for the balance. During fermentation and storage, the carboxylic acid content of the wine declined to 1.44% of the total volatiles and comprised of only lower carboxylic acids. This confirmed a previously reported major change the content of carboxylic acids in the transformation of grape juice to wine (Ardö 2006).

The relative content of ketones changed very little during fermentation and storage but several types increased slightly. Aldehydes in the grape macerate were transformed rapidly and disappeared after fermentation. However, it did not mean that there were more aldehydes in Merlot berries for the reason that aldehydes could be formed as well after crushing of grapes. Moreover, acetaldehyde is a very common constituent, and the major aldehyde of wine. Sulphur dioxide, resulting from the usual addition of metabisulphite to prevent oxidation and microbial spoilage, chemically bonds to acetaldehyde so this almost certainly accounts for the disappearance of the aldehydes among the volatile compounds.

Three species of volatile hydrocarbons were detected in the grape macerate and they accounted for 1.49% of the total peak area. However, the hydrocarbons disappeared altogether after fermentation.

The grape macerate did not contain any heterocyclic compounds (e.g. furans, thiazoles and thiophenes), but two types of furans and one type of thiazole with relative contents of 1.15 and



0.52%, respectively were found in the wine. A close relationship between volatile heterocyclic compounds and the characteristic aroma components of different varietal wine has been reported (Burin *et al.* 2013). It is promising concerning the availability of the aroma patterns as fingerprints of different wines.

Volatile sulfur-containing compounds were not found in the grape macerate, but some were generated during fermentation. At the same time, volatile nitrogen-containing compound (2, 3-dimethyl-2-nitro-butane, 0.58%) that was present in the macerate disappeared completely. Three compounds in the grape macerate and five compounds in the wine were unidentified; respectively accounting for 2.89 and 2.94% of the total peak area.

Following aerobic maceration after crushing the fruit, Merlot grape must for the production of dry red wine contains many volatile compounds. Primary fermentation and seven months storage generates a major increase in the number and relative content of volatile compounds. Many of the compounds are known to contribute to wine aroma, depending on their sensory thresholds. Authors conclude that the content of major volatile components of Merlot grape macerate and dry red wine, produced in the geographic origin of Ningxia Helan Mountain Eastern region, were not very different to those found in other regions. However, the content of trace compounds, such as heterocyclic and sulfur and nitrogen containing compounds, was markedly different to similar studies in other regions. These slight distinctions are likely to depend mainly on differences in the geographical location and growth conditions like solar irradiance, and precipitation, and irrigation and plant nutrition.

Present results provide a benchmark for further comparative studies on the influence of geographic location, viticultural practices and fermentation technology on the flavor and aroma qualities of dry red Merlot wine. Specifically, they provide a baseline for future studies of the influence of those factors on the aroma and flavor components of Merlot dry red wine in the Ningxia Helan Mountain Eastern geographic region. Key aroma compounds identified in this study may also be used to establish a sensory evaluation system specifically for wines of that cultivar.

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