



# Effect of alternative new pruning system and harvesting times on aroma compounds of young wines from Ecolly (*Vitis vinifera*) in a new grape growing region of the Weibei Plateau in China



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## ABSTRACT

Each training system affecting the volatile concentration of wine is closely related to the specific growing conditions of grape. The aim of this work is to evaluate the influence of different training systems, such as crawled cordon training (CCT) and independent long-stem pruning (ILSP), on the volatile composition of Ecolly wine in Weibei Plateau region in three periods (July 31, August 10 and 24). Total aroma concentration (271.15 mg L<sup>-1</sup>) and ethyl octanoate concentration (84.60 mg L<sup>-1</sup>) were the highest for ILSP in August 24 in aroma compounds. However, total aroma concentration of CCT exceeded the ILSP in July 31 and August 10, respectively. ILSP could lead to higher acetate esters compared with CCT except for August 10. Ethanol esters were also strongly influenced by training systems in July 31 (CCT > ILSP) and August 24 (ILSP > CCT), except for slight influence in August 10. But other esters were not strongly impacted by training system. Despite the highest higher alcohols in ILSP in August 24, CCT still displayed significant higher concentration in the first two harvesting times than ILSP. Organic acids showed the same trend as higher alcohols in the experiment. Odor activity values (OAVs) of the 16 aromas compounds showed similar results. Results have shown that CCT improved the accumulation of aroma compounds in early wine, if harvesting time need to be postponed, ILSP was also a suitable select. Whereas, a strongly suggestion of CCT was made because stabilization of yield and quality could be guaranteed by CCT annually.

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## 1. Introduction

The quality of product is an important consideration of wine. Compared with brewing technology, raw materials contribute more to product quality. Effect of trellising system on grape and wine composition is prominent, which helps the viticulturist to employ management techniques that improve production quality (Naylor, 2001; Reynolds and Heuvel, 2009). While tradition may have a bearing on the predominant training system used in a particular area, the final choice is often determined by the efficiency of the training system producing fruits of a desired quality.

As we all know, aroma is also an important index for evaluating the wine's quality (Henning and Villforth, 1942). Recent research has demonstrated that viticulture was one of the major factors determining the aromatic potential of grapes and wines (Song et al., 2012). It is worthy that effects of training systems (Reynolds et al.,

1996) and cover crops in vineyard (Xi et al., 2011) on grape and wine quality have been studied. It is also to be expected that the same wines from a same vineyard should share the typical aromatic characteristics of different pruning systems except for some similar features (Reynolds and Heuvel, 2009). Therefore, it is necessary to assess a wine's aroma from different trellises in order to improve wine quality.

Another, for cultivating grapes at the dormant area to thrive with the increase of the grapevine area and transfer of labor force to city, the following criteria need to be met: (a) new trellis provide possible manipulations to retard and avoid drought damage during short dry periods, regulate reproduction growth and vegetative growth and control disease; (b) a simple, low cost technique for modifying the microenvironment of fruit development and shortening the transportation distance of nutrient between root and berry to improve fruit quality, facilitate sufficient maturity and precocity of every berries under cooler climatic conditions; and (c) a trellis design and training techniques to improve harvest efficiency by maintaining fruiting zone to a accordant low position and be compatible with the winter protection strategy. Ideally, training systems should be labor-efficient and adapted to the local climate

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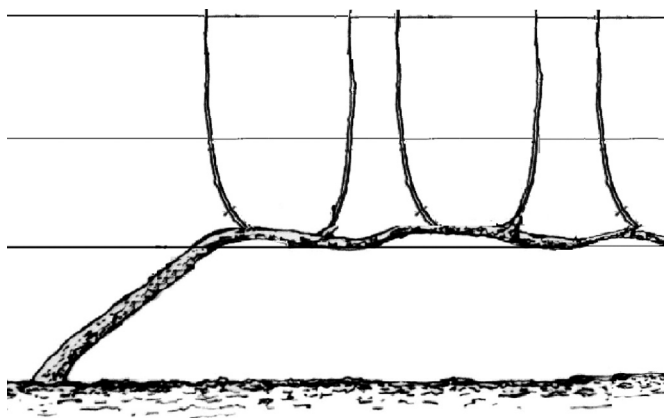


Fig. 1. Diagram of ILSP.

(Cavallo et al., 2001), to mitigate conditions that are unfavorable for growth and promote for the objectives of guaranteeing high quality of grapes, stable yield, longevity and beauty of the vineyards (Li and Fang, 2005). The final choice is often determined by the efficiency of the training system producing fruit of a desired quality.

A trellis system called the crawled cordon training involving covering wild crop (Li et al., 2010), which has the minimal pruning for lowering the cost of winegrape production and controlling moisture migration of the vines, was developed to aid mechanization operation of canopy management, fruit harvest, dormant pruning and overlaying soil in winter by the experimental studies. Although this system has been applied successfully in commercial grapevines more than 20 years ago, there have been few formal reports of experimental evaluation to sustain this trellis system, let alone its effects on the aroma of the wine made from its berries. Previous work focused mainly on effects of trellis on general indices of berry without controlling shoot and bunch counts per vine (Wolf et al., 2003). Hence, it is necessary that the traditional ILSP (Fig. 1) was taken as control to compare the effect of CCT (Fig. 2) on aroma quality under controlling shoot and bunch counts per vine among the treatments first.

In CCT, all vines planted were pruned to one cane along the surface of soil and five shoots per cane. Every shoot with a space of approximately 15–20 cm was trained to the vertical trellis during the growth period and a two-node spur in the late October (Fig. 1). While in ILSP, there was an acclivitous stem extending to the first wire positioned at 50 cm from the ground and continued to elongate horizontally and kept the five shoots trained to the “V” type trellis above the first wire in the growth period (Fig. 2). The other followed CCT.

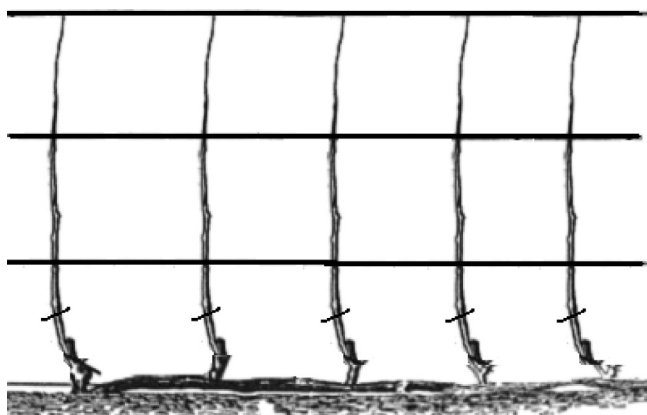


Fig. 2. Diagram of CCT.

To establish the aroma differences between wines exactly, the odor activity value (OAV) was introduced to quantify the levels of recognized odorant components (Peinado et al., 2004), and to estimate the contribution of a specific compound to the wine aroma. The principal objective of the work is to investigate the use of CCT with Ecolly winegrape variety and to analyze the influence of this new training system on wine aroma, and help winemakers to optimize viticulture managements.

## 2. Materials and methods

### 2.1. Experimental vineyard and layout

The study was carried out at the Heyang Chateau (109° W, 34° N, 780 m), Heyang County, China. The region characterized by a semiarid continental monsoon climate, with hot and dry summers and cold winters, having sunshine hours of approximately 2528.3 h per year and an average annual rainfall of 500–540 mm. The mean annual temperature is 11.5 °C, while frost-free season occurs on 208 d per year. Three test plots (180 plants per plot) were arranged in a completely randomized block design with three replicates (60 plants each). Each system was planned 30 plants at corresponding test plot. Vines were spaced 3.0 m × 1.0 m on a sandy loam soil, north-south orientated, trained to CCT and ILSP as mentioned above (Figs. 1 and 2).

### 2.2. Winemaking

Healthy grapes brewed, Ecolly (*Vitis vinifera* L.), were harvested manually on July 31, August 10 and 24, 2012, respectively, and microvinification was performed at the Heyang Chateau with 25 kg grape samples per replication and three replicates per trellis during the corresponding harvesting time, respectively. Technological processes of the six samples were same entirely. Musts were obtained using manual extrusion and the barrelling in glass fermentation vessels of 20 L. After the barrelling, 5 mL of sulphur dioxide (6%) per vessel was immediately added to the musts and the contents were mixed by hand, followed by adding 0.02 g L<sup>-1</sup> of pectinase (Lallzyme Ex) according to commercial specifications. After 12 h, 0.2 g L<sup>-1</sup> of dried active yeast (*Saccharomyces cerevisiae* RC 212, Lallemand, Danstar Ferment AG, Switzerland) were added to the supernatant extracted and fermentation temperature was controlled between 16 and 20 °C. Temperature and density controls were maintained during the whole fermentation period. After the alcoholic fermentation was finished (residual sugar <2 g L<sup>-1</sup>), all the wines were transferred to glass containers of 2–10 L differ in capacity to get rid of air in vessels by siphonage and then 25 mg L<sup>-1</sup> of SO<sub>2</sub> was added immediately into each container. Then some conventional parameters were assessed and the results were shown in Table 1. Subsequently, these wines were stored at 4–6 °C and avoided light for 5 months until final analyses.

### 2.3. General analysis

General properties of wines, including alcohol degree, residual sugar, volatile acid, free SO<sub>2</sub>, total SO<sub>2</sub>, total acids, and pH, were determined according to GB/T 15038-2006. All parameters were analyzed in triplicate.

### 2.4. Aroma components analysis

Aroma components were analyzed by GC–MS apparatus with a special configuration: one injector was connected to one capillary column with a flow splitter. Aromatic compounds were extracted by solid-phase microextraction technology. 16 mL of sample were extracted by shaking for 30 min with 16 μL of internal standard. The

**Table 1**  
The general parameters of Ecolly wines for different trellises in different picking times.

| Sample date | Trellis | Alcoholicity (v v <sup>-1</sup> , %) | Residual sugar (g L <sup>-1</sup> ) | Volatile acid (mg L <sup>-1</sup> ) <sup>a</sup> | Free SO <sub>2</sub> (mg L <sup>-1</sup> ) | Total SO <sub>2</sub> (mg L <sup>-1</sup> ) | TA (g L <sup>-1</sup> ) <sup>b</sup> | pH            |
|-------------|---------|--------------------------------------|-------------------------------------|--|--|---|--------------------------------------|---------------|
| July 31     | CCT     | 10.75 ± 0.13a                        | 1.03 ± 0.07a                        | 0.29 ± 0.09a                                     | 16.32 ± 0.52b                              | 29.5 ± 0.66b                                | 6.40 ± 0.29c                         | 3.29 ± 0.01a  |
|             | ILSP    | 10.55 ± 0.21a                        | 1.22 ± 0.04a                        | 0.34 ± 0.03a                                     | 15.32 ± 0.35a                              | 30.32 ± 0.19c                               | 6.51 ± 0.46c                         | 3.22 ± 0.01a  |
| August 10   | CCT     | 10.86 ± 0.30a                        | 1.08 ± 0.01a                        | 0.34 ± 0.01a                                     | 15.89 ± 0.29ab                             | 28.96 ± 0.57a                               | 4.29 ± 0.29a                         | 3.54 ± 0.02c  |
|             | ILSP    | 11.02 ± 0.16a                        | 1.23 ± 0.03a                        | 0.38 ± 0.02a                                     | 15.66 ± 0.34a                              | 28.33 ± 0.43ab                              | 4.71 ± 0.31a                         | 3.44 ± 0.03ab |
| August 24   | CCT     | 11.02 ± 0.29a                        | 1.06 ± 0.05a                        | 0.33 ± 0.03a                                     | 16.52 ± 0.34c                              | 29.91 ± 0.52b                               | 4.70 ± 0.35a                         | 3.52 ± 0.03c  |
|             | ILSP    | 11.00 ± 0.33a                        | 1.11 ± 0.03a                        | 0.31 ± 0.03a                                     | 16.66 ± 0.42c                              | 28.42 ± 0.64a                               | 5.06 ± 0.38b                         | 3.37 ± 0.01ab |

Different letters in the same column indicate significant differences ( $P < 0.05$ ). These data are from the means of 3 values ± standard deviation (SD).

<sup>a</sup> Acetic acid.

<sup>b</sup> TA, total acidity.

**Table 2**  
Volatiles of Ecolly wines made from grapes with different trellises and harvesting times.

| Aroma components                             | Aroma concentration (mg L <sup>-1</sup> ) |                      |                     |                      |                     |                      |
|--|---|----------------------|---------------------|----------------------|---------------------|----------------------|
|  | CCT <sub>7,31</sub>                       | ILSP <sub>7,31</sub> | CCT <sub>8,10</sub> | ILSP <sub>8,10</sub> | CCT <sub>8,24</sub> | ILSP <sub>8,24</sub> |
| <i>Acetate esters</i>                        |   |                      |                     |                      |                     |                      |
| Ethyl acetate                                | 18.5 ± 0.01a                              | 19.5 ± 0.16a         | 18.91 ± 2.05a       | 10.9 ± 1.63a         | 10.96 ± 0.3a        | 6.14 ± 0.13a         |
| 2-Methylpropyl acetate                       | 0.97 ± 0.06a                              | 0.39 ± 0.01a         | ND                  | ND                   | ND                  | 0.44 ± 0.03a         |
| 3-Methyl-1-butyl acetate                     | 22.0 ± 1.55a                              | 9.50 ± 0.05a         | 2.13 ± 2.06a        | 1.34 ± 0.83a         | 1.29 ± 0.16a        | 27.80 ± 0.09a        |
| 2-Phenylethyl acetate                        | 12.8 ± 0.07a                              | 38.0 ± 0.09b         | 2.66 ± 0.65a        | 3.23 ± 0.57a         | 8.51 ± 0.03a        | 8.22 ± 0.03a         |
| Subtotal                                     | 54.37                                     | 67.51                | 23.7                | 15.5                 | 20.76               | 42.62                |
| Subtotal %                                   | 31.99                                     | 44.93                | 20.83               | 20.5                 | 21.97               | 13.13                |
| <i>Ethanol esters</i>                        |   |                      |                     |                      |                     |                      |
| Ethyl 2-methyl-propanoate                    | 0.39 ± 0.02a                              | 0.41 ± 0.04a         | ND                  | 0.36 ± 0.03ab        | ND                  | 0.84 ± 0.02b         |
| Ethyl butyrate                               | 0.69 ± 0.03a                              | 0.53 ± 0.02a         | 0.48 ± 0.04a        | 0.46 ± 0.03a         | 0.57 ± 0.02a        | 0.84 ± 0.01a         |
| Ethyl heptanoate                             | 0.53 ± 0.31a                              | ND                   | ND                  | ND                   | ND                  | 0.92 ± 0.18b         |
| Ethyl octanoate                              | 28.4 ± 0.12a                              | 17.3 ± 0.08a         | 11.46 ± 0.98a       | 11.5 ± 0.98a         | 14.2 ± 0.12a        | 84.60 ± 0.06b        |
| Ethyl decanoate                              | 6.60 ± 0.3a                               | 9.73 ± 0.24a         | 6.27 ± 0.84a        | 6.38 ± 0.88a         | 7.42 ± 0.44a        | 36.47 ± 0.03a        |
| Ethyl butanedioate                           | 2.08 ± 0.01a                              | 1.06 ± 0.14a         | ND                  | 0.25 ± 0.09a         | ND                  | 0.45 ± 0.02a         |
| Ethyl dodecanoate                            | 0.96 ± 0.04bc                             | 1.45 ± 0.05a         | 0.52 ± 0.02abc      | 0.43 ± 0.01ab        | 1.06 ± 0.02c        | 0.82 ± 0.03bc        |
| Ethyl 3-hydroxy-tridecanoate                 | 1.12 ± 0.04a                              | 0.55 ± 0.03a         | 0.618 ± 0.07a       | 0.47 ± 0.04a         | 0.37 ± 0.04a        | 1.01 ± 0.08a         |
| Ethyl hexadecanoate                          | ND  | 1.22 ± 0.02a         | ND                  | ND                   | 0.32 ± 0.10a        | ND                   |
| Ethyl 3-hydroxy-dodecanoate                  | ND  | 0.89 ± 0.04a         | ND                  | 0.47 ± 0.08a         | 0.37 ± 0.07a        | 1.01 ± 0.07a         |
| Ethyl hydrogen succinate                     | 1.39 ± 0.04a                              | 0.94 ± 0.02a         | 4.94 ± 0.09a        | 0.38 ± 0.01a         | 0.32 ± 0.03a        | 0.77 ± 0.03a         |
| Ethyl linoleic                               | 0.90 ± 0.04a                              | ND                   | ND                  | 0.63 ± 0.03a         | 0.40 ± 0.06a        | 0.64 ± 0.04a         |
| Ethyl 9-o-nonanoate                          | ND  | ND                   | 2.00 ± 0.18a        | 0.74 ± 0.04a         | 1.14 ± 0.26a        | ND                   |
| Subtotal                                     | 43.1                                      | 34.13                | 21.35               | 22.12                | 26.2                | 128.37               |
| Subtotal %                                   | 25.36                                     | 22.71                | 18.76               | 29.26                | 27.74               | 39.56                |
| <i>Other esters</i>                          |   |                      |                     |                      |                     |                      |
| 3,7-Dimethyl-6-octenyl formate               | 0.60 ± 0.04a                              | ND                   | ND                  | 0.49 ± 0.04a         | 0.52 ± 0.04a        | ND                   |
| Methyl salicylate                            | 0.73 ± 0.04a                              | 0.69 ± 0.04a         | 2.28 ± 0.04b        | 2.49 ± 0.06b         | 0.58 ± 0.16a        | 0.86 ± 0.18a         |
| Benzyl benzoate                              | ND  | 0.45 ± 0.05a         | 0.93 ± 0.04a        | 0.49 ± 0.06a         | ND                  | 0.68 ± 0.04a         |
| 2-Methylpropyl hexadecanoate                 | 0.70 ± 0.05a                              | 0.51 ± 0.04a         | 0.65 ± 0.04a        | 0.43 ± 0.04a         | ND                  | ND                   |
| Subtotal                                     | 2.02                                      | 1.66                 | 3.87                | 3.91                 | 1.1                 | 1.53                 |
| Subtotal %                                   | 1.19                                      | 1.1                  | 3.4                 | 5.17                 | 1.16                | 0.47                 |
| <i>Higher alcohols</i>                       |   |                      |                     |                      |                     |                      |
| 2-Methyl-1-propanol                          | 1.77 ± 0.05a                              | 0.43 ± 0.04a         | 0.90 ± 0.12a        | 0.65 ± 0.03a         | 0.40 ± 0.1a         | 0.55 ± 0.08a         |
| 3-Methyl-1-butanol                           | 22.00 ± 0.9a                              | 13.7 ± 0.51a         | 8.36 ± 1.33a        | 5.72 ± 1.02a         | 5.02 ± 0.78a        | 18.77 ± 0.49a        |
| 3,7-Dimethyl-1,6-octadien-3-ol               | ND  | 0.58 ± 0.04a         | 0.82 ± 0.05a        | 0.75 ± 0.05a         | 0.60 ± 0.06a        | ND                   |
| 2,3-Butanediol                               | 1.67 ± 0.04a                              | 3.52 ± 0.11a         | 2.35 ± 0.02a        | ND                   | ND                  | 0.83 ± 0.15a         |
| Phenyl ethanol                               | 15.11 ± 0.7a                              | 13.3 ± 0.48a         | 14.63 ± 0.22a       | 11.1 ± 1.08a         | 13.00 ± 0.6a        | 30.29 ± 0.2b         |
| (E)-3,7,11-Trimethyl-1,6,10-dodecatrien-3-ol | 2.96 ± 0.08a                              | 2.32 ± 0.07a         | 2.62 ± 0.09a        | 1.36 ± 0.25a         | 2.01 ± 0.2a         | 1.02 ± 0.12a         |
| 3,7,11-Trimethyl-2,6,10-dodecatrien-1-ol     | 2.92 ± 0.16a                              | 2.62 ± 0.53a         | 8.33 ± 0.28a        | 1.84 ± 0.34a         | 4.36 ± 0.52a        | 1.48 ± 0.25a         |
| 1-Octadecanol                                | ND  | 0.67 ± 0.04a         | 1.85 ± 0.03a        | 0.39 ± 0.05a         | 0.37 ± 0.11a        | 0.59 ± 0.02b         |
| Subtotal                                     | 46.43                                     | 37.19                | 39.85               | 21.9                 | 25.76               | 53.53                |
| Subtotal %                                   | 27.32                                     | 24.75                | 35.02               | 28.96                | 27.26               | 16.49                |
| <i>Organic acids</i>                         |   |                      |                     |                      |                     |                      |
| Octanoic acid                                | 4.26 ± 0.19a                              | 2.10 ± 0.18a         | 1.87 ± 0.14a        | 1.59 ± 0.2a          | 2.16 ± 0.29a        | 10.40 ± 0.13a        |
| n-Decanoic acid                              | 11.10 ± 0.67a                             | 4.31 ± 0.46a         | 10.46 ± 0.68a       | 5.72 ± 0.26a         | 10.98 ± 0.5a        | 20.00 ± 0.56a        |
| 9-Decenoic acid                              | 3.55 ± 0.74a                              | 2.55 ± 0.26a         | 4.35 ± 0.23a        | 1.62 ± 0.2a          | 3.62 ± 0.27a        | 10.72 ± 0.27a        |
| Dodecanoic acid                              | 2.19 ± 0.16a                              | ND                   | 2.30 ± 0.18a        | 0.75 ± 0.08a         | 1.95 ± 0.11a        | 1.02 ± 0.06a         |
| Tetradecanoic acid                           | 0.69 ± 0.61a                              | 0.71 ± 0.13a         | 1.34 ± 0.21a        | 0.78 ± 0.23a         | 0.38 ± 0.12a        | 1.14 ± 0.11a         |
| n-Hexadecanoic acid                          | 2.16 ± 1.46a                              | 1.56 ± 0.85a         | 4.71 ± 0.53a        | 1.72 ± 0.48a         | 1.58 ± 0.49a        | 1.61 ± 0.44a         |
| Subtotal                                     | 24.01                                     | 11.23                | 25.02               | 12.18                | 20.66               | 45.1                 |
| Subtotal %                                   | 14.13                                     | 7.47                 | 21.99               | 16.11                | 21.87               | 13.89                |
| Total  | 169.94                                    | 151.72               | 113.78              | 75.6                 | 94.48               | 271.15               |

ND means not detected. Different letters in the same column indicate significant differences ( $P < 0.05$ ). 7.31, 8.10 and 8.24 means July 31, August 10 and August 24, respectively, sampling time.

mixture was filtered by 0.4  $\mu\text{m}$  of filter membrane, and the filtrate was injected on the GC. All samples were analyzed in triplicate and average results are presented in [Table 2](#).

The injector temperature was set at 250 °C. The column temperature was programmed according to the following: 40 °C for 3 min, increasing at 5 °C  $\text{min}^{-1}$  to 120 °C, then at 8 °C  $\text{min}^{-1}$  to 230 °C and maintained for 10 min. The temperatures for the transfer line and ionic source were set at 230 °C, respectively. Mass range ( $m/z^{-1}$ ) was 33–450 Da. The whole system was operated at a flow rate of 1.00  $\text{mL min}^{-1}$ , helium was used as the gas carrier. Using 2-octanol (99% optical purity) as the internal standard, which were obtained from Aldrich (Milwaukee, WI, USA).

Identification of the components was taken from the retention indices obtained from one column and compared with those of reference components, meanwhile their aroma contents were determined by overall quantitative method. The mass spectra were obtained from the NIST2.0 MS library database, or in literature.

### 2.5. Odor activity values (OAV)

To evaluate the contribution of aroma compound to wine, the odor activity value (OAV) was introduced. Odor activity values ( $\text{OAV} = x/\text{OTH}$ ), where  $x$  is the concentration mean value of each volatile compound and odor threshold (OTH) is its odor threshold (Ferreira et al., 2002; López et al., 2002; Peinado et al., 2004; Gil et al., 2006).

### 2.6. Statistical analysis

A one-way analysis of variance (ANOVA) was used to determine the significance of the difference of each chemical or volatile factor with three replications of the same sample. All statistical analyses were performed using Excel 2003 and SPSS 16.0.

## 3. Results

In all experiments, the effect of the trellises on the aromas of the Ecolly white wines was highly significant during the different harvesting time ([Table 2](#)). To compare differences of the CCT and ILSP to aroma compound, the chemical constituents of wine were extracted by solvent micro extraction and analyzed by GC–MS, meanwhile their aroma contents were determined by overall quantitative method. Finally, 35 components were preliminarily identified and quantified. The total aroma concentration of six samples was between 75.60  $\text{mg L}^{-1}$  and 271.15  $\text{mg L}^{-1}$ . These volatiles were classified into three major groups, namely esters (acetate esters, ethanol esters and other esters), higher alcohols and organic acids, according to their similarity of chemical properties ([Table 2](#)).

### 3.1. Acetate esters

Acetate esters were isolated in every wine sample except 2-methylpropyl acetate ([Table 2](#)). By comparing the means of acetate esters in CCT and ILSP across different harvesting times, there were higher mean values for ILSP wine in July 31. The subtotals concentration of acetate esters varied from 20.76 to 54.37  $\text{mg L}^{-1}$ , being 20.83–31.99% of the total aroma detected in CCT. However, in ILSP, it varied from 15.5 to 67.51  $\text{mg L}^{-1}$ , being 3.13–44.93% of the total compounds detected.

### 3.2. Ethanol esters

The concentration and ratio of ethanol esters for each sample was calculated in [Table 2](#). 13 ethanol esters detected represented the largest group in terms of aroma compounds identified in all wines. The subtotal concentrations of ethanol esters from CCT

and ILSP samples were in the range of 21.35–43.1  $\text{mg L}^{-1}$  and 22.12–128.37  $\text{mg L}^{-1}$ , being 18.76–27.74% and 22.71–39.56% of the total volatile compounds detected, respectively. These esters were mainly composed by ethyl octanoate, ethyl decanoate, ethyl hydrogen succinate, ethyl 3-hydroxy-tridecanoate, ethyl dodecanoate, and ethyl butyrate. However, other aroma compounds like ethyl 2-methyl-propanoate, ethyl heptanoate, ethyl butanedioate, ethyl hexadecanoate, ethyl 3-hydroxy-dodecanoate, ethyl linoleic, and ethyl 9-o-nonanoate, were in minute concentration.

At the same time, the accumulation of ethanol esters was correlated with harvesting times as for the same trellis in the trial. Ethyl octanoate, ethyl decanoate, ethyl 3-hydroxy-tridecanoate, ethyl butyrate, ethyl dodecanoate and ethyl hydrogen succinate which were fundamental constituents of wine, were detected in all harvesting times. And it was obvious that the earlier the harvest did, the more in accumulation of ethanol esters in CCT. However, in ILSP, the aroma concentration increased with the harvesting time delayed ([Table 2](#)). Ethyl 2-methyl-propanoate and ethyl butanedioate were accumulated only in early period in CCT, while accumulated in all three ILSP periods. It was interesting that ethyl heptanoate could be only detected in CCT wines in July 31 and ILSP wines in August 24, and ethyl hexadecanoate was only detected in CCT wine samples in August 24 while ILSP in July 31.

### 3.3. Other esters

Like acetate esters, the third group esters (other esters) also occupied four compounds, namely 3,7-dimethyl-6-octenyl formate, methyl salicylate, benzyl benzoate and 2-methylpropyl hexadecanoate, and there is no obvious subtotal concentration difference between trellises ([Table 2](#)). In CCT wines, a total of 2 compounds were detected in August 24 samples, 3 compounds were detected in July 31 and August 10 samples. However, in August 10's ILSP wines, there were 4 compounds in all were detected. But, there was no obvious differences of these compounds in both CCT and ILSP ([Table 2](#)). The subtotal aroma concentrations were in the 1.10–3.87  $\text{mg L}^{-1}$  in CCT samples, while 1.53–3.91  $\text{mg L}^{-1}$  in ILSP wines detected. The percentages of the concentrations ranged from 1.16 to 3.4% in CCT, and ILSP from 0.47 to 5.17%. CCT increased the percentages, whereas ILSP increased concentration in most situations.

In general, the subtotal concentration of three esters in six samples was between 1.10 and 128.37  $\text{mg L}^{-1}$ , which occupied 1.10–44.93% of the total volatile compounds detected, respectively.

### 3.4. Higher alcohols

In this study, 8 main higher alcohols from three harvesting times were identified ([Table 2](#)). Their subtotal concentrations were from 25.76 to 46.43  $\text{mg L}^{-1}$ , which was 27.26–35.02% of the total aroma components detected in CCT, while 21.9–53.53  $\text{mg L}^{-1}$  and 16.49–28.96% in ILSP. The CCT wines during the first two experiments had higher content of higher alcohols than these of ILSP ([Table 2](#)). The four higher alcohols, 3-methyl-1-butanol, phenyl ethanol, (E)-3,7, 11-trimethyl-1,6,10-dodecatrien-3-ol, 3,7,11-trimethyl-2,6,10-dodecatrien-1-ol, occupied the highest values in these six wines.

### 3.5. Organic acids

Six main organic acids were simultaneously detected, and effect of trellises on the content of organic acids was also obvious during different picking times ([Table 2](#)). The first and second harvest times were optimal occasion of organic acids accumulation in CCT. While the third harvest time was optimal for accumulation of organic acids in ILSP ([Table 2](#)).



The subtotal concentrations of organic acids detected in the sample wines were 20.66–25.02 mg L<sup>-1</sup>, occupying 14.13–21.99% of total aroma components in CCT, 11.23–45.10 mg L<sup>-1</sup> and 7.47–13.89% of total aroma components in ILSP. n-Decanoic acid, 9-decenoic acid, octanoic acid, n-hexadecanoic acid, dodecanoic acid and tetradecanoic acid were found in every sample wines however dodecanoic acid was absent in ILSP samples in July 31. Tetradecanoic acid could only be identified as a trace component in the wines (Table 2).

### 3.6. Odor activity values (OAVs)

In order to assess the influence of the compounds on overall wine aroma, OAV was introduced. Sixteen volatile compounds, odor threshold values (OTH) for every compound and their odor activity values (OAVs), were shown in Table 3. The mean value for every compound, 0.0012–16919.1, could be acceptable for a typical Ecolly wine.

The highest OAVs (>30) were those for ethyl octanoate, ethyl decanoate and ethyl butyrate, followed by those for n-decanoic acid, octanoic acid and 2-phenylethyl acetate. Ethyl dodecanoate, ethyl acetate from July 31 and scattered phenyl ethanol and dodecanoic acid also reached concentrations over the perception threshold (OAVs > 1). OAVs were higher in CCT wines than in ILSP wines if harvested early (Table 3). The OAVs in July 31 showed that aroma properties of some components, including dodecanoic acid (three harvesting times), ethyl butyrate, n-decanoic acid, octanoic acid, phenyl ethanol and ethyl octanoate, were modified easier by CCT than by ILSP. While other properties of aroma components, such as ethyl decanoate, ethyl acetate, ethyl dodecanoate and 2-phenylethyl acetate, were modified easier by ILSP compared with CCT. Only aroma property of ethyl hexadecanoate in CCT and ILSP was displayed in August 24 and July 31, respectively.

## 4. Discussion

Training system plays an essential role in determining wine aroma (Reynolds and Heuvel, 2009). Among them, higher alcohols are thought to contribute positively to the overall wine quality when presented in small quantities, while esters are the main contributors to the bouquet of young wines (Morakul et al., 2013). With regard to the compound concentration, a single and violent aroma in same specific group may be more important than the total group concentration (Nykänen, 1986). Therefore, every aroma compound in wines could not be neglected. Moreover, Pretorius and Bauer (2002) thought that specific ratios of many compounds contributed perceived flavor.

### 4.1. Acetate esters

The wines in the experiment showed higher concentrations of acetate esters in July 31 than in August 10 and August 24, which was the same as Marić and Firšt-Bača (2003). Although small in quantity, acetate esters represented the largest group in terms of the contents and proportions of aroma compounds identified in the six sample wines. Ethyl acetate and 2-phenylethyl acetate (Lilly et al., 2006), 2-methylpropyl acetate (Swiegers et al., 2009) and 3-methyl-1-butyl acetate (González-Rodríguez et al., 2011) are considered as the major components of a “fruity” flavor. Concentration of 3-methyl-1-butyl acetate has something to do with the quality of top wine (Wondra and Berovič, 2001) in the trial. According to Table 2, it seemed that ILSP was more conducive to accumulation of acetate esters than CCT.

### 4.2. Ethanol esters

Esters are formed when alcohol and carboxylic acid functional groups react, while a water molecule is eliminated. The ethanol esters are comprised of an acid group (medium-chain fatty acid) and an alcohol group (ethanol). Whereas the acetate esters are comprised by an acid group (acetate) and an alcohol group which is either ethanol or a complex alcohol derived from amino acid metabolism (Saerens et al., 2008). Therefore, ethanol esters and acetate esters were formed by the same pathway but different precursors. Although in minute concentration, ethanol esters were thought to contribute the same function as acetate esters (Swiegers et al., 2005; Saerens et al., 2008). From Table 2, CCT could produce more ethanol esters than ILSP on July 31, whereas ILSP was the best trellis for the accumulation of ethanol esters in August compared with CCT.

### 4.3. Other esters

A total of 4 other esters in the group were detected in wine samples and there were no obvious concentration differences between trellises in Table 2. Among these esters, benzyl benzoate and methyl salicylate were identified as flavor stabilizer and enhancer (Ebeler et al., 2001) and aglycones in the bound fraction of the aroma of grape berries (Sarry and Günata, 2004), respectively. Thus, these components, although the minimum concentration, were more important than the total group concentration (Nykänen, 1986), which shared the similar contribution with acetate esters and ethanol esters (Swiegers et al., 2005). Hence, these esters cannot be also ignored (Jiang and Zhang, 2010).

In general, esters are an important facet of wine flavor (Marić and Firšt-Bača, 2003). The esters (acetate esters, ethanol esters and other esters) were the most abundant, and showed significant differences between CCT and ILSP wines (Table 2) in the trial. The acetate esters analyzed were considered as factors contributing to young wine's quality, as they gave consumers a pleasant flavor (Ferreira et al., 2000). In July, it seemed that CCT are more applicable for the accumulation of ethanol esters and other esters than ILSP, while in August, especially late August, ILSP played a leading role in the accumulation of ethanol esters and other esters. Accumulation of acetate esters was just opposite.

### 4.4. Higher alcohols

Alcohols are formed by the degradation of corresponding amino acids, carbohydrates and lipids of berries (Antonelli et al., 1999), through the Ehrlich and Ribereau-Gayon metabolic pathways. Together with homologous esters, these higher alcohols contribute positively to the fine fruity character of wine (Swiegers et al., 2005). From Table 2, for the six samples, the effects of CCT on 3,7,11-trimethyl-2,6,10-dodecatrien-1-ol and (E)-3,7,11-trimethyl-1,6,10-dodecatrien-3-ol were far greater than ILSP. During the first two harvest time, CCT has greater effect on 2-methyl-1-propanol, 3-methyl-1-butanol and phenyl ethanol than ILSP does, while in the third harvest time, the effects of ILSP on three higher alcohols are greater than CCT, and the highest concentration of 3-methyl-1-butanol corroborates the conclusion reported by Rebière et al. (2010). In addition, it seemed as if the effects of pruning system on 1-octadecanol and 3,7-dimethyl-1,6-octadien-3-ol were not prominent in three picking times (Table 2).

### 4.5. Organic acids

It is reported that the accumulation of organic acids depends on the components and fermentation conditions of the must (Schreier and Jennings, 1979). Juice and wine compositions were impacted

**Table 3**  
Odor thresholds (OTH) and odor activity values (OAV) of the aroma compounds of different trellises during the three harvesting times.

| Aroma component              | OTH (mg L <sup>-1</sup> ) | OAV                 |                      |                     |                      |                     |                      |
|------------------------------|---------------------------|---------------------|----------------------|---------------------|----------------------|---------------------|----------------------|
|                              |                           | CCT <sub>7,31</sub> | ILSP <sub>7,31</sub> | CCT <sub>8,10</sub> | ILSP <sub>8,10</sub> | CCT <sub>8,24</sub> | ILSP <sub>8,24</sub> |
| Ethyl acetate                | 12.26 <sup>a</sup>        | 1.51 ± 0.01a        | 1.59 ± 0.16a         | 1.54 ± 2.05a        | 0.890 ± 1.63a        | 0.893 ± 0.3a        | 0.501 ± 0.13a        |
| 2-Methylpropyl acetate       | 1.6 <sup>b</sup>          | 0.603 ± 0.06a       | 0.241 ± 0.01a        | ND                  | ND                   | ND                  | 0.274 ± 0.03a        |
| 2-Phenylethyl acetate        | 1.8 <sup>c</sup>          | 7.135 ± 0.07a       | 21.15 ± 0.09b        | 1.475 ± 0.65a       | 1.796 ± 0.57a        | 4.730 ± 0.03a       | 4.565 ± 0.03a        |
| Ethyl butyrate               | 0.02 <sup>a</sup>         | 34.29 ± 0.03a       | 26.54 ± 0.02a        | 23.82 ± 0.04a       | 22.92 ± 0.03a        | 28.41 ± 0.02a       | 41.77 ± 0.01a        |
| Ethyl octanoate              | 0.005 <sup>a</sup>        | 5691.6 ± 0.12a      | 3471.9 ± 0.08a       | 2291.6 ± 0.98a      | 2310.8 ± 0.98a       | 2842.5 ± 0.12a      | 16,919.1 ± 0.06b     |
| Ethyl decanoate              | 0.2 <sup>a</sup>          | 32.98 ± 0.3a        | 48.63 ± 0.24a        | 31.34 ± 0.84a       | 31.88 ± 0.88a        | 37.10 ± 0.44a       | 182.3 ± 0.03a        |
| Ethyl butanedioate           | 1.9 <sup>d</sup>          | 0.0103 ± 0.01a      | 0.0052 ± 0.14a       | ND                  | 0.0012 ± 0.09a       | ND                  | 0.0022 ± 0.02a       |
| Ethyl dodecanoate            | 1.5 <sup>b</sup>          | 1.914 ± 0.04bc      | 2.900 ± 0.05a        | 1.044 ± 0.02abc     | 0.864 ± 0.01ab       | 2.118 ± 0.02c       | 1.630 ± 0.03bc       |
| Ethyl hexadecanoate          | 1.5 <sup>b</sup>          | ND                  | 1.221 ± 0.02a        | ND                  | ND                   | 0.324 ± 0.10a       | ND                   |
| 2-Methylpropyl hexadecanoate | 1.5 <sup>e</sup>          | 0.464 ± 0.05a       | 0.340 ± 0.04a        | 0.433 ± 0.04a       | 0.288 ± 0.04a        | ND                  | ND                   |
| 2-Methyl-1-propanol          | 40 <sup>b</sup>           | 0.044 ± 0.05a       | 0.011 ± 0.04a        | 0.023 ± 0.12a       | 0.016 ± 0.03a        | 0.010 ± 0.1a        | 0.014 ± 0.08a        |
| 2,3-Butanediol               | 120 <sup>b</sup>          | 0.014 ± 0.04a       | 0.029 ± 0.11a        | 0.020 ± 0.02a       | ND                   | ND                  | 0.007 ± 0.15a        |
| Phenyl ethanol               | 14.0 <sup>a</sup>         | 1.079 ± 0.7a        | 0.953 ± 0.48a        | 1.045 ± 0.22a       | 0.799 ± 1.08a        | 0.928 ± 0.6a        | 2.164 ± 0.2b         |
| Octanoic acid                | 0.5 <sup>a</sup>          | 8.513 ± 0.19a       | 4.210 ± 0.18a        | 3.729 ± 0.14a       | 3.170 ± 0.2a         | 4.311 ± 0.29a       | 20.80 ± 0.13a        |
| n-Decanoic acid              | 1.0 <sup>b</sup>          | 11.17 ± 0.67a       | 4.306 ± 0.46a        | 10.45 ± 0.68a       | 5.721 ± 0.26a        | 10.98 ± 0.5a        | 20.22 ± 0.56a        |
| Dodecanoic acid              | 1.5 <sup>b</sup>          | 1.989 ± 0.16a       | ND                   | 2.090 ± 0.18a       | 0.679 ± 0.08a        | 1.773 ± 0.11a       | 0.925 ± 0.06a        |

Note: ND means not detected. Different letters in the same column indicate significant differences ( $P < 0.05$ ). 7.31, 8.10 and 8.24 mean July 31, August 10 and 24, respectively.

<sup>a</sup> (Gil et al., 2006).

<sup>b</sup> (Howard et al., 2005).

<sup>c</sup> (Peinado et al., 2004).

<sup>d</sup> (Ferreira et al., 2000).

<sup>e</sup> (Moyano et al., 2002).

by pruning (Main and Morris, 2008). Therefore, the concentration accumulation of organic acids was strongly influenced by trellises. Except in dodecanoic acid of ILSP produced on July 31, n-decanoic acid, 9-decenoic acid, octanoic acid, n-hexadecanoic acid, dodecanoic acid and tetradecanoic acid were found in every other sample wines, which also had a great influence on the complexity of the aroma (Shinohara, 1985).

Appropriate content of organic acids in wines was necessary for higher contents of aroma esters. Shinohara (1985) found that C<sub>6</sub>–C<sub>10</sub> organic acids with optimal concentrations of 4–10 mg L<sup>-1</sup> but not more than 20 mg L<sup>-1</sup>, produces mild and pleasant aroma for wines. Therefore, the test data of octanoic acid and n-decanoic acid in the study had positive effect on the global aroma quality. In addition, the research of Bardi et al. (1998) has shown that organic acids with a chain length of C<sub>8</sub>–C<sub>14</sub> exhibit strong antimicrobial activity and that the effect is intensified under unsaturated fatty acid. Both conclusions proved that octanoic acid and n-decanoic acid also played an important role in antimicrobial activity and 9-decenoic acid could strengthen the role.

In experiments, earlier harvesting could control appropriate content of octanoic acid, and the effect of CCT were prominent. From Table 2, the concentrations of n-decanoic acid in CCT were stable during three picking times, while in ILSP the concentrations increased with the season. Owe to antimicrobial function of C<sub>8</sub>–C<sub>14</sub> organic acids, antimicrobial effect of 9-decenoic acid in CCT surpassed ILSP when berry was harvested early, while the function of dodecanoic acid in CCT exceeded ILSP during three picking times. Tetradecanoic acid, although the minimum values, could not be ignored in all the sample wines. The n-hexadecanoic acid, a sub-essential composition in wines, was also reported by Liberatore et al. (2010).

Wine quality, especially aroma concentration, originated from berry compounds and content as aroma precursors improved by training systems (Fragasso et al., 2012), which can potentially be released during processing or storage, and consequently improve the complexity of wine aroma (Winterhalter et al., 1990). Owe to large crop size and high crop loads, berry and wine composition (Brix, titratable acidity, and pH) were not strongly impacted by training system (Reynolds et al., 2004). However, different character of wine composition in the trial was achieved by optimized branch numbers of cluster and fruit to balance the quantity of

production. Of course, minimized path of element transportation decided by trellis should be considered compared with Reynolds et al. (2004). What is more, optimal conditions are demanded to produce the aroma characteristic of wine compounds sufficiently. In the study, aroma compounds not detected in Table 2 showed the improper harvesting time along with different pruning systems.

#### 4.6. Odor activity values (OAVs)

Not all of the compounds detected in the wine samples have the same impact on the overall aroma character of a wine (Jiang and Zhang, 2010). Study about the odor activity values (OAVs) by Guth (1997) noted considerable different contents in white wines. These variations can be ascribed to the inevitable dispersion resulting from slight differences among grapes and the fermentation and aging conditions used.

Volatile compounds, with OAVs >1, make an active contribution to the odor of the wine (Louw et al., 2009). However, a compound with OAV <1 might also contribute to the wine aroma because of the additive effect of similar compounds (Francis and Newton, 2005), and compounds with similar OAVs can enhance the existing contribution through synergy with other compounds (López et al., 2003). Therefore, every compound in the trial could not be ignored because of its direct or indirect contribution to wine aroma.

OAVs, such as ethyl octanoate, ethyl butyrate, n-decanoic acid and octanoic acid, were higher in CCT wine than that of ILSP wine if harvested early (Table 3). The result showed that CCT could modify the final aroma properties of wines brewed by berries picked early, which agreed with the result of Diago et al. (2010) about Tempranillo wines. With postponement of picking time, OAVs of wine from ILSP, such as ethyl octanoate, ethyl butyrate, n-decanoic acid and octanoic acid, could reproduce the result of Diago et al. (2010). Only ILSP controlled OAVs of ethyl decanoate in wine during the whole trial.

## 5. Conclusion

The results of the experiment clearly demonstrate that aroma character of Ecolly wines in the Weibei Plateau imposed by trellises has much to do with harvesting times. The total volatile compounds and the total and subtotal aroma values were identified

and quantified in different picking times and trellises, respectively. CCT wines produced from grapes of advancing harvest resulted in higher concentrations of two esters group, namely ethanol esters (ethyl octanoate) and other esters, higher alcohols (3-methyl-1-butanol) and organic acids (n-decanoic acid), while ILSP wine showed another result. Although acetate esters did not show a clear trend with harvesting date, they exhibited higher concentrations in ILSP than in CCT during the trial. Analysis of OAVs demonstrated further that effect of CCT was more prominent than that of ILSP on wine aroma compounds before August 10. Therefore, CCT could promote accumulation of aroma compounds early, if the harvesting time need to be postponed, ILSP was also a fine choice.

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