

# Characteristic fruit wine production via reciprocal selection of juice and non-*Saccharomyces* species

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## ABSTRACT

This study examines the effect of juices and non-*Saccharomyces* yeasts on physicochemical properties and sensory quality for fruit wines. Here, fruit wines produced from six kinds of fruit juices and eight non-*Saccharomyces* yeasts were evaluated, and the results show significant phenotypic diversity within these yeasts for wine fermentation on a range of substrates. Substantial variations in indicators such as total acid, sugar content and organic acid content were observed, which were caused by selection of both yeasts and juice types. These differences in characteristics had an impact on consumer preference. Considering overall acceptance by the panelists, correspondence analysis showed that *Pichia kluyveri* X31-10 (Pk31) was the most suitable strain for apple juice fermentation, and Fuji apple juice was the best substrate for this fermentation. A partial least-squares regression model of aroma characteristics against aroma components revealed that acetate esters were the primary contributors to tropical fruit aroma notes of ciders from Pk31. Additionally, 1-butanol, 3-methyl-, acetate; acetic acid, hexyl ester; and acetic acid, pentyl ester have been implicated as characteristic volatiles associated with Pk31.

## 1. Introduction

Among fruit, grapes have seen the most technical and commercial use as substrates for winemaking. Today, a growing number of other fruits with different origins, cultivars, shapes, colors, tastes, and nutritive values, are available for the production of fruit wines (Jagtap and Bapat, 2015). As the world's most economically important and widely-consumed fruit, the apple is already famous for apple wine (also called cider) in many Western countries (Minnaar et al., 2017). For years, there has been both great interest and demand for apple wine production in China, which is a good use of surplus apple production (Fan et al., 2006). However, limitations of commercially active dry yeast and lack of yeasts tailored to cider fermentation have restrained the development of the cider industry.

The choice of the yeast for fermentation is crucial to obtaining a fermented product with the distinctive flavors appreciated by consumers. For an extended period, *Saccharomyces cerevisiae* strains are customarily used for wine-making due to their dominance and advantages in alcoholic wine fermentation. In recent years, non-*Saccharomyces* yeasts (otherwise called non-conventional yeasts) have been gaining attention due to their high intraspecific variability

(Romano et al., 1997), as well their roles in both ethanol yield reduction (Alonsodelreal et al., 2017) and flavor enhancement (Holt et al., 2018) in wines and beer. Several non-*Saccharomyces* species have been shown to contribute to aroma complexity and increase yields of desirable compounds depending on the modalities of their use (Canonico et al., 2018). Among these non-*Saccharomyces* yeasts, *Torulopsis delbrueckii*, *P. kluyveri*, and with some other species, are now available as dry or frozen active yeast (Chasseriaud et al., 2018). *T. delbrueckii* has received attention due to its positive and distinctive effect on the overall aroma of wines (Benito, 2018a; Canonico et al., 2017), and *Hanseniaspora vineae* and *Candida zemplinina* have also been used to increase flavor diversity (Englezos et al., 2016; Medina et al., 2013). Besides, different non-*Saccharomyces* species could be used to diversify taste and aroma profiles in wine, and strain variability also has effect on flavor production (Escribano-Viana et al., 2018).

However, depending on the type, variety, and pre-fermentation treatment of fruit, the application of non-*Saccharomyces* yeasts in the fruit wine fermentation may modify the flavor profile in unpredictable ways. Canonico et al. (2017) observed opposite trends in two fermentation studies with *T. delbrueckii* and suggested this variation could be explained by different initial substrates and nutrient availabilities (i.e.

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sugars, amino acid composition, flavor-active compounds precursors). Thus, careful study of substrates and initial culture conditions such as nutrient availability and their effect on flavor notes, especially in relation to different non-*Saccharomyces* yeasts, should be discreetly considered. To our knowledge, there have been no reports focused on this approach of reciprocal selection of fruit juice and non-*Saccharomyces* species.

Herein, this work studied apple, grape, kiwifruit, and pear juices as suitable substrates for fermentation by monocultures of non-*Saccharomyces* yeasts. The first goal was to explore the effect of each one of the yeast strains tested on the fruit matrix (and vice-versa). Afterward, considering the overall acceptability of fruit wines, a two-way selection of yeasts and fruit matrix was applied, and an optimal match was estimated. Moreover, the underlying relationship between volatiles and aroma descriptors was regressed using partial least-squares regression (PLSR).

## 2. Materials and methods

### 2.1. Fruit juices

Fresh, ripe, healthy fruits were purchased from the local market or vineyards in Shaanxi or Ningxia, China (Table 1). The fruits were washed to remove dirt and dust. After coring and chopping the apples and pears, peeling the kiwifruit, and de-stemming the grapes, the fruits were squeezed for juice using an electric juice squeezer. Sulfur dioxide (50 mg/L of K<sub>2</sub>S<sub>2</sub>O<sub>5</sub>) was added to inhibit bacterial growth and prevent browning. Pectinase ( $\geq 500$  U/mg) purchased from Shanghai Yuanye Bio-Technology Co., Ltd. (Shanghai, China) was also added, and the original juice was obtained after enzymolysis and filtration through double-layer filter paper to remove the solid particles. The acquired juices were frozen at  $-20$  °C until utilization.

### 2.2. Yeast strains

Eight strains of yeasts (Table 2) that had been isolated and screened in advance were chosen for fermentation. *S. cerevisiae* WLS21, used for cider fermentation and preserved in our laboratory, was used as a control. Yeasts were maintained at  $-80$  °C prior to use.

### 2.3. Fermentation trials

Each strain was independently grown in 100 mL YPD (yeast extract peptone dextrose) medium for 24 h on a rotary shaker (28 °C, 150 rpm), and then aliquots (100  $\mu$ L) of inocula were transferred into 100 mL juice in 250 mL Erlenmeyer flask at 28 °C on a rotary shaker (150 rpm). After the yeast had been activated and pre-cultured in separate fruit juice samples, approximately 10<sup>6</sup> CFU/mL of yeasts were transferred into a 500 mL Erlenmeyer flask containing 300 mL of the sterile fruit juice (pasteurized for 15 min at 104 °C). Fermentations were carried out at 25 °C for 15 days. Two independent experiments were performed for every combination of all strains and all juices. At the end of fermentation, all juices and wines (300 mL) were centrifuged for 10 min at 7000 rpm, and the cell-free supernatants were stored at  $-20$  °C for analysis.

**Table 1**  
Origins and cultivars of fruits used in the preparation of juices.

Type of fruit	Cultivar	Abbreviation	Region
Apple	Fuji	F	Shaanxi, China
Apple	Gala	G	Shaanxi, China
Apple	Red star	R	Shaanxi, China
Grape	Cabernet Sauvignon	C	Ningxia, China
Kiwifruit	Cuixiang	K	Shaanxi, China
Pear	Suli	P	Shaanxi, China

**Table 2**  
Yeast strains used in this study.

Name	Species	Abbreviation	Source
J16-7	<i>Candida zemplinina</i>	Cz16	Apple (Hebei, China)
X22-9	<i>Hanseniaspora uvarum</i>	Hu22	Apple (Shaanxi, China)
X11-10	<i>Hanseniaspora vineae</i>	Hv11	Apple (Shaanxi, China)
X26-8	<i>Lachancea thermotolerans</i>	Lt26	Apple (Shaanxi, China)
X31-10	<i>Pichia kluyveri</i>	Pk31	Apple (Shaanxi, China)
WLS21	<i>Saccharomyces cerevisiae</i>	Sc21	Lab <sup>a</sup> preservation
X28-10	<i>Torulasporea delbrueckii</i>	Td28	Apple (Shaanxi, China)
X24-4	<i>Torulasporea quercuum</i>	Tq24	Apple (Shaanxi, China)

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### 2.4. Physicochemical analysis

The SSC (soluble solid contents), pH values, total acid content, and total sugar content were quantified as described by Ye et al. (2014a). SSC and pH of samples were determined using a hand-held refractometer (WYT-4; Quanzhou Optical Instrument Co., Ltd., Quanzhou, China) and a pH meter (pH 211, Hanna, Italy), respectively. The total acid content was assayed by titration of appropriately diluted sample with 0.1 mol/L NaOH and was expressed as percentage of tartaric acid. The total sugar content was estimated by the reduction method using Fehling's reagent. The color measurements were made using the L\*a\*b\* system with an X-Rite Ci7600 benchtop spectrophotometer with Milli-Q water as a control. The CIELab color difference was calculated as:  $\Delta E^* = (\Delta L^{*2} + \Delta a^{*2} + \Delta b^{*2})^{1/2}$  (Torchio et al., 2011).

### 2.5. Ethanol quantification

Wine alcohol levels were measured in accordance with the instructions provided with the BioVision ethanol assay kit (Catalog #K620-100).

### 2.6. HPLC analyses

The organic acid content was determined using a HPLC (Shimadzu, Kyoto, Japan) with a Shim-pack VP-DOS C<sub>18</sub> analytical column (250 mm  $\times$  4.6 mm i.d., particle size 4.6  $\mu$ m, Shimadzu, Kyoto, Japan). The column chromatography conditions (Ye et al., 2014b) were as follows: flow rate 0.7 mL/min, temperature 40 °C, and mobile phase 0.01 mol/L ammonium phosphate (pH adjusted to 2.7 by phosphoric acid). Organic acids were detected using a PDA detector.

Glucose, fructose, and glycerol were analyzed using HPLC (Shimadzu, Kyoto, Japan) with a Zorbax NH<sub>2</sub> column (4.6 mm  $\times$  250 mm, 5  $\mu$ m) linked with refractive index detector with a modified version of the method reported by Ye et al. (2014a). The column was maintained at 40 °C. The eluent was a mixture of acetonitrile and water (85:15 v/v) at a flow rate of 1.5 mL/min. Each sample was measured in duplicate.

### 2.7. Volatile compound analysis by GC/MS

Volatiles of all samples were analyzed by headspace solid-phase micro-extraction gas chromatography-mass spectrometry (SPME-GC-MS) using a modified version of the method described by Lan et al. (2017). Regarding sample preparation, 5 mL of sample were placed in a 20 mL vial, followed by addition of 1.5 g NaCl and 40  $\mu$ L internal standard (4-methyl-1-pentanol, final conc. 394.08  $\mu$ g/L). The following determination was performed by a GC-MS QP2010 Ultra system (Shimadzu, Kyoto, Japan) and all samples were determined at a semi-quantitative level. Volatile compounds were extracted by HS-SPME with a carboxen/divinylbenzene/polydimethylsiloxane (CAR/DVB/

PDMS) fiber (Supelco Ltd., US). After incubation at 40 °C for 15 min, the extraction was performed at the same temperature over the course of 20 min, under continuous stirring (250 rpm). After extraction, the analytes were thermally desorbed into the GC injector at 250 °C with a Shimadzu AOC-6000 automatic sampler under the splitless injection mode. DB-1MS chromatographic column (60 m × 0.25 mm × 0.25 μm) was used to separate volatile components. The GC and MS conditions were similar to those used by Lan et al. (2017), and all samples were analyzed in duplicate. The component assignment was based on computer matching with data in the NIST14 library. The amounts of the volatile compounds were calculated using the concentration of the internal standard multiplied the ratio of the peak area of the volatiles to the peak area of the internal standard.

Odor activity value (OAV), defined as the ratio between the concentration of the individual chemical compound and its sensory detection threshold in literature data (supplementary material 1), was calculated for all the identified volatiles to evaluate the contribution of volatiles to wine aroma.

## 2.8. Sensory analysis

Sensory analysis was conducted by a trained panel of 6 students (3 males and 3 females aged 20 to 30) in our lab. As described in a previous work (Wang et al., 2017), sensory analysis panel was trained with Le Nez du Vin aroma kit, and every panelist was asked to describe the wine aroma profile using several Le Nez du Vin terms or other descriptive terms about wine-relevant aroma when necessary in actual evaluation. In addition to the selection of terms, scores needed to be obtained for each term by five-point scaling. Here, 0 was used for undetectable; 1 for very weak; 2 for weak; 3 for medium; 4 for strong; and 5 for very strong. Finally, modified frequency (MF%) was used as a characteristic parameter for sensory analysis, which was calculated by squaring the product of the detection frequency (%F) of an aromatic attribute and the average intensity (%I). The wine was evaluated for overall acceptability (considering appearance, taste, and aroma) according to the hedonic scale by 32 tasters of 18–40 of age (staff and students of Northwest A&F University). Scores were logged on a scale of 1–5: 1 for disgusting; 2 for unacceptable; 3 for neutral; 4 for good, and 5 for excellent. The mean score of wine was analyzed to assess overall quality. Similarly, leading feature of each wine was also recorded based on tasters' subjective perception.

## 2.9. Statistical analysis

Two determinations were performed on every index for all samples and the mean values of results were calculated. One-way analysis of variance (ANOVA) and Duncan's multiple-range test ( $P < 0.05$ ) was carried out with SPSS 22.0 for Windows (SPSS Inc., Chicago, IL, US). Pearson correlation analysis was used to determine the existence of correlations between individual wine indicators and the overall acceptability for all wines. A  $P$ -value of  $< 0.05$  was here considered significant. Correspondence analysis (CA) was used to seek associations among juices, yeasts, and acceptability. A partial least-squares regression (PLSR) model was established to produce a linear regression model from the data using Unscrambler 10.4 (Camo, Trondheim, Norway).

## 3. Results and discussion

### 3.1. Physicochemical analysis

Table S1 shows the pH values of different fruit juice controls (CK) and different wines obtained through the fermentation process (supplementary material 2). The pH values of pear juice and pear wines ranged from 3.53 to 4.93, which were relatively high, while those of kiwi juice and kiwi wines ranged somewhat lower from 3.10 to 3.31. When different juices were fermented by different yeasts, the trends in

the changes in pH were also different. The pH of pear wine was significantly lower than that of the corresponding fresh pear juices, and the pH values of Fuji apple wine and kiwi wine either increased or did not change significantly. The pH values of different wines fermented by Pk31 were substantially lower, closely followed by Hu22, whereas the pH values of different wines fermented by Tq24 were noticeably higher. Changes in pH caused by fermentation of other yeasts in different juices were showed less discernible trends.

The SSC (soluble solid contents), total acid and total sugar results of all samples are listed in Tables S2, S3, and S4, respectively. The SSC and total sugar of all the fermentations with SC21, Td28, and Tq24 notably decreased, indicating the fermentations went smoothly and thoroughly. However, strains, where the opposite is true, are ones like Pk31 and Hu22. Concerning total acid, a significant increase can be seen in grape, pear and Red Star apple wines. Total acid varied drastically influenced by both juice type and strains, and as a result, contributed accordingly to pH values.

Regarding chromatic characteristics, significant differences were observed in CIELab parameters among different wines (data not shown). The fermentation process resulted in significantly lower values of  $L^*$  coordinates after grape wine fermentation, indicating that grape wines were darker than grape juice. Except for the fluctuation of  $a^*$  coordinates in grape wines compared to grape juices, those in other juices decreased compared to corresponding juices, suggesting these wines have less pronounced red color components. Similarly, less pronounced yellow color components were observed in all wines except for grape and pear. These changes may be explained by the variation of pH in wines (Englezos et al., 2018). A  $\Delta E^*$  value of 3 was assessed as the general color tolerance perceptible by the human eye (Martínez et al., 2001). In our study, all wines evidenced a perceptible color difference ( $\Delta E^*$  higher than 3) when compared with corresponding CK, apart from some pear wines. Although color directly affects appearance, the tasters focused more on taste and aroma of the wines than on the color in subsequent analyses.

### 3.2. Glucose and fructose

As shown in Table 3, the glucose in pear juice was fully utilized by various yeasts. Strains Sc21, Td28, Tq24, and Hv11 consumed a significant amount of glucose in each fruit juice (all consumption rates were over 92%, and the majority were more than 98%). The residual contents of glucose after fermentation with Cz16 and Hu22 were comparatively high. However, Cz16 nearly depletes the fructose in all fruit juices (except pear juice) (Table 4). In contrast, the residual fructose levels of Pk31 and Hu22 fermentation were generally much higher. In addition, Sc21, Td28, Tq24, and Hv11 consumed a great deal of fructose, with small differences among them. As shown, the difference in utilization of fructose and glucose between different yeasts varied strongly. The fructophilic character of the yeasts was determined according to the glucose-fructose consumption ratio (Mestre Furlani et al., 2017). The fructophilic characteristic was found to be robust in Cz16, especially in apple juice (all three apple juices), and this strain was also reported to show similar behavior in other studies (Magyar and Tóth, 2011). Conversely, Pk31 displayed an intense glucophilic nature (particularly in grape and Gala apple juice), followed by Hu22 and Lt26, while the remaining four strains did not show any apparent preference for glucose. The levels of glucose and fructose varied depending on fruit type, variety, and maturity. Thus, suitable yeasts can be selected for the fermentation of different fruit wines according to this characteristic.

The intersections of rows and columns represent juice or wine fermented with the corresponding yeast and fruit juice. Results represent the mean  $\pm$  SD for two independent experiments. Mean values in the same column with the same letter are not significantly different at the 5% confidence level (Duncan's test). ND, not detected. CK, respective juice. Cz16, *Candida zemplinina* J16-7; Hu22, *Hanseniaspora uvarum*

**Table 3**  
Concentrations of glucose (g/L) of different juices and wines.

Strain	C	F	G	K	P	R
CK	82.200 ± 7.623 a	16.900 ± 9.461 a	11.495 ± 0.106 a	33.110 ± 8.952 a	14.755 ± 0.191 a	11.645 ± 3.274 a
Cz16	15.105 ± 0.064 c	9.575 ± 0.615 ab	4.135 ± 1.039 b	1.690 ± 0.467 b	0.695 ± 0.134 b	2.705 ± 0.106 bc
Hu22	42.265 ± 1.549 b	3.835 ± 0.361 bc	2.085 ± 0.106 c	4.745 ± 1.054 b	0.485 ± 0.064 b	1.900 ± 0.693 bc
Hv11	6.335 ± 0.092 d	0.290 ± 0.410 c	ND	0.050 ± 0.071 b	0.030 ± 0.042 c	0.755 ± 0.629 c
Lt26	18.525 ± 2.666 c	1.485 ± 0.884 c	1.650 ± 0.721 c	6.670 ± 2.220 b	0.030 ± 0.042 c	1.235 ± 0.021 c
Pk31	36.695 ± 1.393 b	6.205 ± 2.595 bc	1.485 ± 0.884 c	1.650 ± 0.552 b	ND	4.475 ± 0.672 b
Sc21	0.680 ± 0.226 d	0.405 ± 0.573 c	0.150 ± 0.212 d	0.245 ± 0.163 b	0.010 ± 0.014 c	0.390 ± 0.269 c
Td28	0.665 ± 0.205 d	ND	0.215 ± 0.304 d	0.015 ± 0.021 b	ND	0.090 ± 0.014 c
Tq24	0.490 ± 0.085 d	ND	0.095 ± 0.134 d	ND	0.150 ± 0.156 c	0.035 ± 0.050 c

The intersections of rows and columns represent juice or wine fermented with the corresponding yeast and fruit juice. Results represent the mean ± SD for two independent experiments. Mean values in the same column with the same letter are not significantly different at the 5% confidence level (Duncan's test). ND, not detected. CK, respective juice. Cz16, *Candida zemplinina* J16-7; Hu22, *Hanseniaspora uvarum* X22-9; Hv11, *Hanseniaspora vineae* X11-10; Lt26, *Lachancea thermotolerans* X26-8; Pk31, *Pichia kluyveri* X31-10; Sc21, *Saccharomyces cerevisiae* WLS21; Td28, *Torulasporea delbrueckii* X28-10; Tq24, *Torulasporea quercuum* X24-4. F, Fuji apple; G, Gala apple; R, Red star apple; C, Cabernet Sauvignon grape; K, Cuixiang kiwifruit; P, Suli pear.

X22-9; Hv11, *Hanseniaspora vineae* X11-10; Lt26, *Lachancea thermotolerans* X26-8; Pk31, *Pichia kluyveri* X31-10; Sc21, *Saccharomyces cerevisiae* WLS21; Td28, *Torulasporea delbrueckii* X28-10; Tq24, *Torulasporea quercuum* X24-4. F, Fuji apple; G, Gala apple; R, Red star apple; C, Cabernet Sauvignon grape; K, Cuixiang kiwifruit; P, Suli pear.

### 3.3. Ethanol, glycerol, and acetic acid

Glycerol is an intriguing metabolite in wines because of its positive contribution to the mouthfeel sensations such as sweetness, softness, silkiness, and body (Jolly et al., 2006). The taste threshold of glycerol is 5.2 g/L (Noble and Bursick, 1984), whereas the maximum acceptable level is 25 g/L (Scanes and Prior, 1998). The glycerol contents of all wines (Table 5) were here within an acceptable range. Glycerol could be perceived in all grape wine samples, while it may not have been tasted in most other wine samples. In addition, Cz16 produced notable amounts of glycerol ranging from 3.650 to 11.975 g/L, in accordance with the findings reported by Tofalo et al. (2012). *C. zemplinina* is a high glycerol producer, like *Candida stellata* with glycerol concentrations reported in wine up to 14 g/L (Jolly et al., 2014). The highest ethanol producers of all yeasts were Sc21, Td28, and Tq24, which produced 113.756, 118.724, and 137.979 g/L, respectively. However, Pk31 can produce, at most, 34.365 g/L ethanol (Table 6). Pk31 produces significantly lower content of acetic acid, followed by Lt26 (Table S9). According to the “Instituto Nacional de Vitivinicultura” (INV; National Viticulture Institute, San Juan, Argentina), the maximum acceptable level of volatile acidity for commercialization of wines is 0.7 g/L of acetic acid. It is interesting to emphasize the high acetic acid production of Hu22 (934.050–2383.750 mg/L), which could contribute to negative evaluation.

A simple correlation analysis was performed to determine the correlation between the metabolites glycerol-acetic acid, glycerol-ethanol, and acetic acid-ethanol after 15 days of fermentation because these three metabolites are simultaneously produced during glycerol-pyruvate and alcoholic fermentation processes (Ribereau-Gayon et al.,

2006). A very weak positive correlation was found (Pearson's coefficients of 0.108, 0.294 and 0.170, respectively).

### 3.4. Organic acid analysis

Because both the type and the quantity of organic acids can make a difference in sensory and chemical characteristics such as pH, total and volatile acidity, and microbiological stability, it is of great importance to analyze organic acids (Loira et al., 2015). Determination of eight organic acids in wines was performed by HPLC (Tables S5–S12). Only three of them were detected in every wine: oxalic acid, malic acid and citric acid. Levels of oxalic acid in fermentation with Pk31 were substantially high (i.e. the closest to that of CK), while those with other yeasts degraded, especially significantly with Cz16 in apple and grape juices and with Hv11 in apple and pear juices. All the wines produced by Lt26, exhibited relatively low amount of malic acid. Since *Lachancea thermotolerans* can limit the concentration of malic acid, it has been specially used to modify the wine acidity (Benito, 2018b; Chen et al., 2018). Similar results were also observed in apple and grape wines with Cz16. As Tofalo et al. (2012) reported, *C. zemplinina* has the capacity to metabolize malic acid. It is interesting to emphasize the high production of malic acid and citric acid observed with Td28 in apple wines. Conversely, the level of citric acid in Hu22 and Tq24 fermentation samples was noticeably lower. Unlike in other samples, quinic acid was not found in grape juice or grape wines. Moreover, quinic acid contents were relatively high in wines fermented by Tq24. Interestingly, substantially augmentation of succinic acid can be seen in fermentations of Cz16. Succinic acid has an unusual salty, bitter taste (Coulter et al., 2004) and high levels might have a negative influence on the mouthfeel of the wines. In contrast, Hu22 only produced a small quantity of succinic acid in apple wines, followed by Td28. Low production of succinic acid in fermentation byproducts formed by *H. uvarum* or *T. delbrueckii* has also been reported by Ciani and Maccarelli (1998). Highly unlike aforementioned organic acids, lactic acid can only be generated in fermentation processes. Pk31 showed a

**Table 4**  
Concentrations of fructose (g/L) of different juices and wines.

Strain	C	F	G	K	P	R
CK	114.870 ± 7.778 a	67.755 ± 8.436 a	42.790 ± 0.990 a	39.595 ± 10.189 a	44.995 ± 6.088 a	33.310 ± 6.194 a
Cz16	ND	0.265 ± 0.149 d	0.370 ± 0.085 d	ND	2.790 ± 0.198 cd	0.295 ± 0.035 c
Hu22	54.900 ± 1.315 c	21.625 ± 5.070 c	15.875 ± 2.793 c	11.710 ± 1.160 b	5.735 ± 1.662 bc	9.130 ± 0.453 b
Hv11	35.695 ± 0.007 d	0.325 ± 0.106 d	0.665 ± 0.134 d	0.365 ± 0.120 c	0.300 ± 0.085 d	1.160 ± 0.297 c
Lt26	48.900 ± 4.936 c	15.280 ± 2.998 c	24.020 ± 4.059 b	14.455 ± 5.919 b	0.935 ± 0.290 cd	11.000 ± 4.045 b
Pk31	85.080 ± 2.885 b	41.535 ± 8.549 b	26.375 ± 3.415 b	7.230 ± 1.089 bc	9.065 ± 0.219 b	12.515 ± 0.926 b
Sc21	0.425 ± 0.050 f	3.350 ± 4.200 d	1.350 ± 1.556 d	0.755 ± 0.064 c	0.035 ± 0.007 d	2.745 ± 0.898 c
Td28	9.220 ± 3.366 e	2.370 ± 0.467 d	2.270 ± 0.877 d	0.255 ± 0.134 c	0.330 ± 0.269 d	0.535 ± 0.021 c
Tq24	10.890 ± 3.536 e	0.920 ± 0.735 d	0.760 ± 0.552 d	0.285 ± 0.021 c	0.375 ± 0.191 d	0.535 ± 0.106 c

**Table 5**  
Concentrations of glycerol (g/L) of different juices and wines.

Strain	C	F	G	K	P	R
CK	2.900 ± 0.014 e	0.440 ± 0.127 d	0.230 ± 0.014 f	0.285 ± 0.092 e	0.180 ± 0.042 c	0.290 ± 0.071 c
Cz16	11.975 ± 1.379 a	5.665 ± 0.700 ab	5.825 ± 0.587 a	6.020 ± 0.735 a	3.650 ± 0.156 b	4.380 ± 0.820 a
Hu22	6.295 ± 0.233 d	2.985 ± 0.290 cd	2.910 ± 0.622 e	4.615 ± 0.403 ab	2.555 ± 0.516 b	2.395 ± 0.389 b
Hv11	6.705 ± 0.134 cd	4.060 ± 0.778 bc	5.040 ± 0.198 abcd	3.210 ± 0.325 bcd	3.220 ± 1.174 b	3.685 ± 1.096 ab
Lt26	8.705 ± 0.460 bc	2.390 ± 0.014 cd	3.735 ± 0.743 bcde	4.020 ± 1.103 bc	2.525 ± 0.601 b	3.650 ± 0.735 ab
Pk31	10.510 ± 0.778 ab	6.420 ± 1.881 ab	3.595 ± 0.530 cde	3.715 ± 0.347 bc	7.300 ± 0.339 a	3.510 ± 0.566 ab
Sc21	8.855 ± 0.573 bc	8.225 ± 2.256 a	5.405 ± 1.025 ab	2.540 ± 0.622 cd	0.095 ± 0.092 c	3.060 ± 0.339 ab
Td28	7.145 ± 1.450 cd	4.660 ± 0.849 bc	5.290 ± 1.301 abc	3.950 ± 1.160 bc	2.845 ± 0.856 b	3.770 ± 1.047 ab
Tq24	7.140 ± 1.414 cd	4.255 ± 0.361 bc	3.400 ± 0.509 de	1.910 ± 0.014 d	2.380 ± 0.453 b	3.535 ± 0.629 ab

The intersections of rows and columns represent juice or wine fermented with the corresponding yeast and fruit juice. Results represent the mean ± SD for two independent experiments. Mean values in the same column with the same letter are not significantly different at the 5% confidence level (Duncan's test). CK, respective juice. Cz16, *Candida zemplinina* J16-7; Hu22, *Hanseniaspora uvarum* X22-9; Hv11, *Hanseniaspora vineae* X11-10; Lt26, *Lachancea thermotolerans* X26-8; Pk31, *Pichia kluyveri* X31-10; Sc21, *Saccharomyces cerevisiae* WLS21; Td28, *Torulaspora delbrueckii* X28-10; Tq24, *Torulaspora quercuum* X24-4. F, Fuji apple; G, Gala apple; R, Red star apple; C, Cabernet Sauvignon grape; K, Cuixiang kiwifruit; P, Suli pear.

pronounced ability to generate lactic acid in all wines, whereas this was true of Td28 alone in apple wines.

### 3.5. Volatile compound analysis

#### 3.5.1. Concentrations and categories

The data from SPME-GC-MS of the juices and wines were analyzed (Supplementary material 3). In total, 11 categories of aroma compounds and 195 individual aroma components were detected. The numbers of categories and individual aroma components for each treatment type detected ranged from 9 to 11 and from 42 to 79, respectively. The total concentrations of detected volatile aroma components fell in between 7.20 and 90.87 mg/L. All wines obtained from Pk31 consisted of the highest total concentration of volatiles, followed by those from Sc21 and Lt26, while Hu22 and Cz16 produced the lowest levels of total volatile compounds. Among the detected volatile compounds in different wines, the most abundant were alcohols, acetate esters, and ethyl esters, especially alcohols, which made up 30–80% of the aroma profiles of most of wine samples. It should be noted that acetate esters contributed at least 75% of the aroma profiles to wines fermented by Pk31. Benito et al. (2015) have also reported high production of acetate esters by *Pichia kluyveri*. 1-butanol, 3-methyl-, acetate; acetic acid, 2-phenylethyl ester; octanoic acid, ethyl ester; and decanoic acid, ethyl ester can be detected in all wines.

#### 3.5.2. Odor activity values

Volatile compounds that are present in wine and have an odor activity value (OAV) > 1 are commonly considered likely contributors to characteristic aromas (Song et al., 2015). Results of OAVs are shown in Supplementary material 4. In our study, the concentrations of 57 volatiles, including aldehydes (n = 14), ethyl esters (n = 10), terpenes

(n = 9), acetate esters (n = 8), acids (n = 5), alcohols (n = 5), ketones (n = 2), phenols (n = 2), hydrocarbons (n = 1), and other esters (n = 1) were found to be higher than their respective thresholds in at least one sample. It was observed that the most common aromatic categories were aldehydes and terpenes for fruit juices, and esters, aldehydes, and terpenes for fruit wines. Important acids and alcohols were found only in wines. These findings suggested fermentation with non-*Saccharomyces* adjusted aroma characteristics through the formation of impact compounds within esters, acids and alcohols. They were classified as fermentative volatiles (Robinson et al., 2014), and the others were categorized as non-fermentative aroma compounds.

Among fermentative aromas, some of them were only produced with an OAV > 1 (or increased significantly) after the fermentation of specific yeasts and could be regarded as characteristic aromas. Octanoic acid, ethyl ester showed the highest OAV of 3549.9 (for sample C-Sc21) and showed significant production in all wines. Specifically, the highest increase was observed in wines obtained from Sc21 and the lowest from Cz16. Correspondingly, it gives the wine aromas with varying degrees of sweetness, fruitiness and pear aroma (Welke et al., 2014). 1-Butanol, 3-methyl-, acetate; and acetic acid, hexyl ester, reminiscent of banana (Ruiz et al., 2014), were multiplied in wines from all strains except Cz16, but extremely significant for Pk31. On the contrary, 1-butanol, 2-methyl-, acetate, also having a banana-like smell (Lee et al., 2012), cannot be detected in wines from Pk31, but increased significantly in wines from Sc21, Hu22, and Lt26. In addition, acetic acid, 2-phenylethyl ester (rose, honey) for wines from Pk31 and Hv11, heptanoic acid, ethyl ester (sweet, strawberry, banana) for Lt26, Sc21 and Tq24, decanoic acid, ethyl ester (fruity, grape) for Sc21 and Tq24, propanoic acid, 2-methyl-, ethyl ester (sweet, fruity) for Cz16, 1-propanol, 3-ethoxy-(fruity) for Td28, propanoic acid, 2-methyl- for Hu22, Hv11, Sc21, and Td28, and acetic acid, pentyl ester (floral, fruity, banana) and

**Table 6**  
Concentrations of ethanol (g/L) of different wines.

Strain	C	F	G	K	P	R
Cz16	94.983 ± 5.111 c	42.615 ± 1.767 d	46.315 ± 1.680 c	31.487 ± 0.602 cd	24.040 ± 0.915 e	40.717 ± 1.089 c
Hu22	52.812 ± 0.956 e	32.406 ± 0.831 e	18.806 ± 0.446 f	22.965 ± 0.291 e	18.375 ± 0.220 f	17.726 ± 0.467 e
Hv11	87.844 ± 1.812 c	42.010 ± 1.802 d	44.243 ± 1.025 c	34.746 ± 1.452 c	7.781 ± 0.078 h	30.311 ± 1.104 d
Lt26	70.855 ± 1.923 d	47.056 ± 1.846 d	29.786 ± 1.166 e	28.923 ± 0.812 d	30.925 ± 1.363 d	38.489 ± 1.103 c
Pk31	33.664 ± 1.053 f	15.372 ± 0.146 f	7.744 ± 0.120 g	34.365 ± 1.177 c	13.573 ± 0.224 g	29.760 ± 1.183 d
Sc21	113.756 ± 5.740 b	74.932 ± 1.581 b	38.345 ± 1.655 d	33.491 ± 1.384 c	42.001 ± 0.893 c	69.832 ± 3.363 b
Td28	118.724 ± 3.749 b	66.231 ± 1.802 c	58.349 ± 1.473 b	63.463 ± 2.481 b	50.287 ± 2.436 b	73.820 ± 2.252 b
Tq24	137.979 ± 5.161 a	107.809 ± 5.482 a	93.417 ± 1.789 a	82.822 ± 2.134 a	90.241 ± 2.792 a	95.088 ± 2.213 a

The intersections of rows and columns represent juice or wine fermented with the corresponding yeast and fruit juice. Results represent the mean ± SD for two independent experiments. Mean values in the same column with the same letter are not significantly different at the 5% confidence level (Duncan's test). Cz16, *Candida zemplinina* J16-7; Hu22, *Hanseniaspora uvarum* X22-9; Hv11, *Hanseniaspora vineae* X11-10; Lt26, *Lachancea thermotolerans* X26-8; Pk31, *Pichia kluyveri* X31-10; Sc21, *Saccharomyces cerevisiae* WLS21; Td28, *Torulaspora delbrueckii* X28-10; Tq24, *Torulaspora quercuum* X24-4. F, Fuji apple; G, Gala apple; R, Red star apple; C, Cabernet Sauvignon grape; K, Cuixiang kiwifruit; P, Suli pear.

2-furanmethanol, acetate (toasted) for Pk31, were observed for their specific emergence or significant increase of OAVs. These aroma descriptors were from several different papers (Arcari et al., 2017; Ruiz et al., 2014; Welke et al., 2014). Analogously, some aromas were shown to be juice/matrix-dependent. Butanoic acid, 2-methyl-, ethyl ester (fruity (pineapple, banana, apple)) could only be found in apple wines, while benzoic acid, methyl ester in kiwi juice and kiwi wines. It was interesting to mention that butanoic acid, 3-methyl-, ethyl ester could only be determined in wines with Pk31 and non-apple juices.

Among non-fermentative aromas, the compounds beta-damascenone; decanal; butanal, 3-methyl-; methional; octanal, and other aromas, even at low concentrations, make positive contributions to the flavor of wines and may work synergistically with other volatile compounds by modifying the perception of flavor notes. Similarly, aromas that were strain- or juice/matrix-dependent were also identified. Beta-damascenone (honey, sweet) were not detected in any wines from Hv11 or Sc21. 3-Furaldehyde vanished after fermentation except in some kiwi wines. Butanal, 3-methyl- (green, malt) for Lt26 and Sc21, eugenol (spices, clove, honey) for Pk31 and Td28, butanal, 2-methyl- for Cz16 and Sc21, propanal, 2-methyl- (chocolate) for Cz16 and Sc21, 2-nonanone and 2-methoxy-4-vinylphenol for Sc21 were only detected in their fermented wines. Meanwhile, 2-pentanone (fruity); trans-linalool oxide (leafy, sweet, floral, creamy, earthy); and alpha-terpineol were only found in kiwi wines, while OAVs of nerolidol (mild rose, convallaria, apple flower) were significantly higher in Fuji and Gala apple wines. These aroma descriptive terms were used as described in previous works (Brunschwig et al., 2012; Fariña et al., 2015; Gu et al., 2013; Meng et al., 2015; Song et al., 2015; Wang et al., 2015; Welke et al., 2014).

Considering that synergic effects can be elicited by compounds with similar structures and odors (Tosi et al., 2012), potential aroma-causing components with OAVs between 0.2 and 1 were also noted during this analysis. There were 12 substances in 195 volatiles that satisfied this criterion, and most of them were esters.

### 3.6. Sensory analysis

To evaluate the effect of each different culture on organoleptic quality of fermented products, the sensory analysis of different fruit wines was carried out. The average score was between 1.625 (K-Hv11) and 4.125 (F-Pk31). Wines fermented with kiwi juice with low scores were not found acceptable overall.

#### 3.6.1. Correlation between wine indicators and acceptability

To extract some conclusions about the impact of specific wine indicators on the wine acceptability during the inmouth passage of wine, a Pearson correlation analysis was performed. The wine indicators involved excluded color parameters and volatiles. According to the results, ethanol, fructose, glycerol, tartaric acid, lactic acid and total sugar did not seem to correlate with acceptability of wine. However, total acid showed a significant negative correlation with acceptability ( $r = -0.752$ ). More specifically, the acceptability was negatively related with quinic acid ( $r = -0.732$ ), oxalic acid ( $r = -0.683$ ), acetic acid ( $r = -0.462$ ), and citric acid ( $r = -0.749$ ). Weaker negative associations between acceptability SSC, and glucose were also found. Exhibiting the opposite trend, several indicators showed a relatively weak positive correlation, such as pH value ( $r = 0.457$ ), or the presence of malic acid ( $r = 0.527$ ) or succinic acid ( $r = 0.451$ ).

#### 3.6.2. Correspondence analysis

Correspondence analysis (CA) has been applied in wine research on many previous occasions (Ferreira et al., 2016; Rodrigues et al., 2017). In our study, CA was performed on a juice (6)  $\times$  strain (9) contingency table in which each cell in the table represents the average score of the wine. Results (Fig. 1) showed that both juices and strains influenced the acceptability of the wine. Three apple juice fermentations were

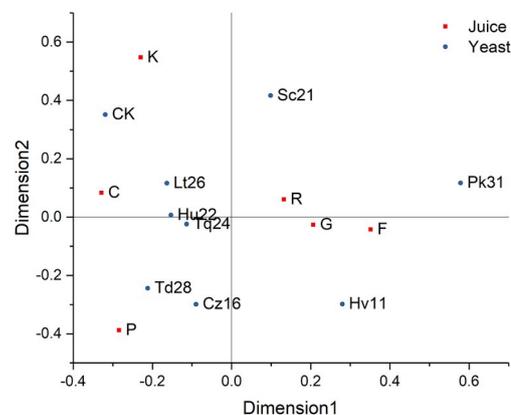


Fig. 1. Projection of the non-*Saccharomyces* yeasts and fruit juices on the first two dimensions of the correspondence analysis space. CK, respective juice. Cz16, *Candida zemplinina* J16-7; Hu22, *Hanseniaspora uvarum* X22-9; Hv11, *Hanseniaspora vineae* X11-10; Lt26, *Lachancea thermotolerans* X26-8; Pk31, *Pichia kluyveri* X31-10; Sc21, *Saccharomyces cerevisiae* WLS21; Td28, *Torulaspora delbrueckii* X28-10; Tq24, *Torulaspora quercuum* X24-4. F, Fuji apple; G, Gala apple; R, Red star apple; C, Cabernet Sauvignon grape; K, Cuixiang kiwifruit; P, Suli pear. (For interpretation of the references to color in this figure legend, the reader is referred to the Web version of this article.)

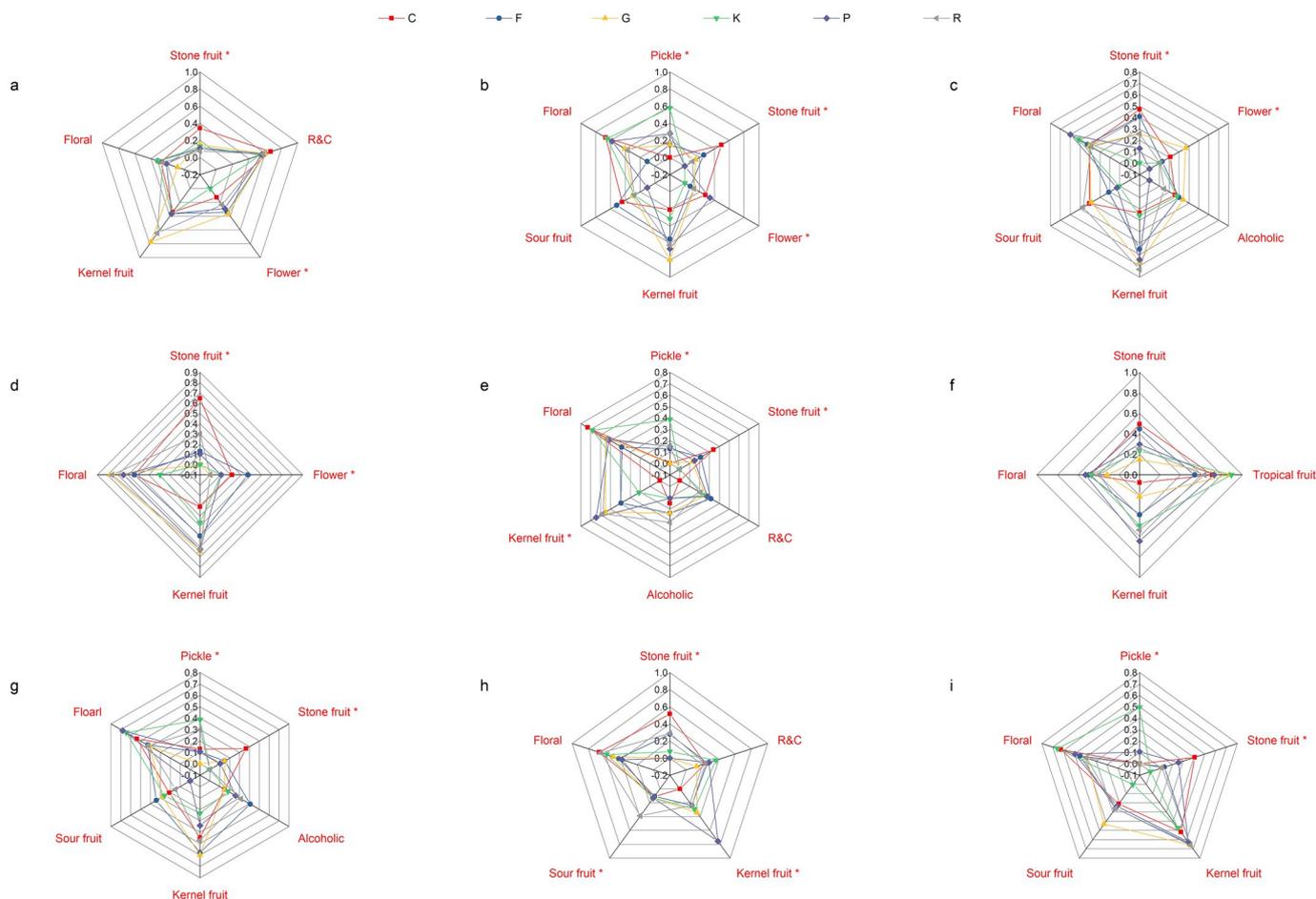
observed to occur on the right side of the figure, while the other juice fermentations appeared on more to the left side, indicating inconsistent sensory scores for different fruit wines, while apple fermentations were more consistent. After fermentation, differences in sensory preference between wines and corresponding juices were large for apples and pears, medium for grapes, and small for kiwis. Different yeasts may be suitable for fermentation of different types of juice. The proximity of the strain to the juice reflected appropriateness between them. It can be concluded from this figure that Td28 and Cz16 were more suitable for pear juice, Lt26 for grape juice, and Pk31, Hv11 and Sc21 for apple juices. Unfortunately, none of the strains were appropriate for fermentation of kiwi juice, since unfermented kiwi juice was obtained a better evaluation than fermented. In addition, Pk31 showed a special preference for apple juice, while Hu22 and Tq24 with little characteristic were obtained a good evaluation in all kinds of juices. However, Fuji apple juice offered the best matrix for Pk31, followed by Gala and Red Star apple juice. The same conclusion can be drawn for Hv11.

#### 3.6.3. Aroma characteristics

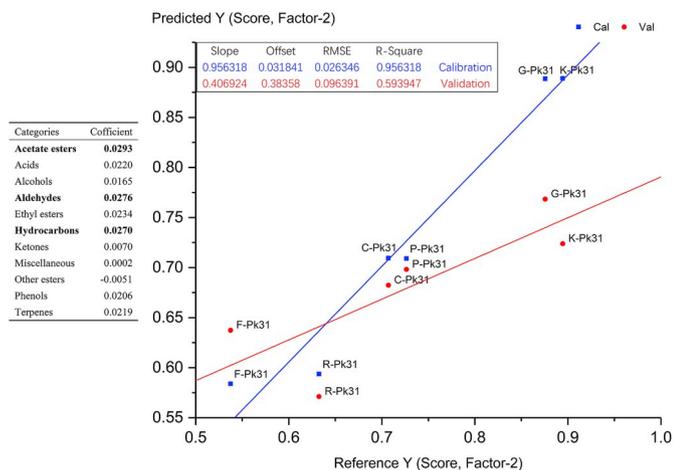
Fig. 2 shows the aroma characteristics of the wine samples that had MF values  $> 0.3$ . For each strain, aroma characteristics were significantly different across the different types of fruit ( $RSD > 0.05$ ). With respect to CK, roasted and cooked was important and differed slightly among all juices, while Gala and Red star apple juice were reminiscent of kernel fruit. Despite the aroma diversity in different wines caused by Cz16, K-Cz16 had a smell of fermented vegetable, which led to a low score. Regarding Hu22, Lt26, Tq24, and Hv11, kernel fruit and floral were detectable aroma characteristics, whereas stone fruit had an impact on the organoleptic property of C-Hv11. Floral notes were easily found in wines from Sc21 and Td28. Meanwhile, the dominant aroma was kernel fruit for G-Sc21 and P-Td28, and stone fruit for C-Td28, respectively. Remarkably, tropical fruit contributed most to every kind of wine from Pk31. Our results suggested that non-*Saccharomyces* yeast strains with substantial interspecies diversity greatly contributed to the fruity aroma quality of wines, which was in line with other reports (Liu et al., 2016; Rossouw and Bauer, 2016).

#### 3.6.4. PLSR

Since Pk31 and Hv11 displayed a predilection for fermentation of apple juice, they were selected for future analysis. To determine the complex correlation between the aroma characteristics and identified



**Fig. 2.** Modified frequency (MF) values of aroma characteristics in different wines fermented with different fruit juices and non-*Saccharomyces* yeasts. (a–h) represent CK and the wines from Cz16, Hu22, Hv11, Lt26, Pk31, Sc21, Td28, Tq24, respectively. They share the legend at the top of the figure. R&C, roasted and cooked. CK, respective juice. Cz16, *Candida zemplinina* J16-7; Hu22, *Hanseniaspora uvarum* X22-9; Hv11, *Hanseniaspora vineae* X11-10; Lt26, *Lachancea thermotolerans* X26-8; Pk31, *Pichia kluyveri* X31-10; Sc21, *Saccharomyces cerevisiae* WLS21; Td28, *Torulasporea delbrueckii* X28-10; Tq24, *Torulasporea quercuum* X24-4. F, Fuji apple; G, Gala apple; R, Red star apple; C, Cabernet Sauvignon grape; K, Cuixiang kiwifruit; P, Suli pear. \*: the aroma characteristics that the relative standard deviation (RSD) of MF values is over the average level of RSD (RSD > 0.59). (For interpretation of the references to color in this figure legend, the reader is referred to the Web version of this article.)



**Fig. 3.** PLS regression of tropical fruit aroma characteristics by aroma categories. Pk31, *Pichia kluyveri* X31-10. F, Fuji apple; G, Gala apple; R, Red star apple; C, Cabernet Sauvignon grape; K, Cuixiang kiwifruit; P, Suli pear. (For interpretation of the references to color in this figure legend, the reader is referred to the Web version of this article.)

aroma components, PLSR was performed based on eleven aroma component groups and aroma characteristics of apple wines from Pk31 or Hv11 fermentation. The results, which were presented in Fig. 3, suggested that only tropical fruit produced PLSR models with calibration  $R^2$  values greater than 0.90, and validation  $R^2$  values of greater than 0.50. This indicated that there might not be definite associations between a volatile compound and an odor characteristic; rather, this association changed according to the product matrix and composition (López-López et al., 2018). However, it was clear from the PLSR results that all component groups except other esters were positively correlated with tropical fruit aroma characteristics. Acetate esters, aldehydes, and hydrocarbons which had regression coefficient values higher than 0.025 contributed most to this correlation. Combined with results above, it may conclude that 1-butanol, 3-methyl-, acetate; acetic acid, hexyl ester; and acetic acid, pentyl ester were characteristic aromas of *Pichia kluyveri* species.

#### 4. Conclusion

In conclusion, under these conditions, these findings highlight that both yeasts and juices shape the characteristics and aroma profiles of wines. Furthermore, non-*Saccharomyces* yeasts have interesting and discrepant oenological properties, in terms of consumption of sugars

and acids, and production of important metabolites. On the other hand, contribution to characteristic wines of non-*Saccharomyces* yeasts was affected by the type of fruit juices. In addition, Pk31 was the most suitable strain for apple juice fermentation, and Fuji apple juice was its best substrate. The biggest contributor to tropical fruit aroma characteristics of wine from Pk31 was acetate esters, especially 1-butanol, 3-methyl-, acetate; acetic acid, hexyl ester; and acetic acid, pentyl ester.

### Conflicts of interest

The authors have no competing financial interests to declare.

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### Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.fm.2018.11.008>.

### References

- Alonsodelreal, J., Contrerasruiz, A., Castiglioni, G.L., Barrio, E., Querol, A., 2017. The use of mixed populations of *Saccharomyces cerevisiae* and *S. kudriavzevii* to reduce ethanol content in wine: limited aeration, inoculum proportions, and sequential inoculation. *Front. Microbiol.* 8, 2087.
- Arcari, S.G., Caliarì, V., Sganzerla, M., Godoy, H.T., 2017. Volatile composition of Merlot red wine and its contribution to the aroma: optimization and validation of analytical method. *Talanta* 174, 752–766.
- Benito, S., 2018a. The impact of *Torulaspora delbrueckii* yeast in winemaking. *Appl. Microbiol. Biotechnol.* 102, 3081–3094.
- Benito, S., 2018b. The impacts of *Lachancea thermotolerans* yeast strains on winemaking. *Appl. Microbiol. Biotechnol.* 102, 6775–6790.
- Benito, S., Hofmann, T., Laier, M., Lochbühler, B., Schuttler, A., Ebert, K., Fritsch, S., Rocker, J., Rauhut, D., 2015. Effect on quality and composition of Riesling wines fermented by sequential inoculation with non-*Saccharomyces* and *Saccharomyces cerevisiae*. *Eur. Food Res. Technol.* 241, 707–717.
- Brunschwig, C., Senger-Emonnot, P., Aubanel, M.L., Pierrat, A., George, G., Rochard, S., Raharivelomanana, P., 2012. Odor-active compounds of Tahitian vanilla flavor. *Food Res. Int.* 46, 148–157.
- Canonica, L., Comitini, F., Ciani, M., 2017. *Torulaspora delbrueckii* contribution in mixed brewing fermentations with different *Saccharomyces cerevisiae* strains. *Int. J. Food Microbiol.* 259, 7–13.
- Canonica, L., Comitini, F., Ciani, M., 2018. *Torulaspora delbrueckii* for secondary fermentation in sparkling wine production. *Food Microbiol.* 74, 100–106.
- Chasseriaud, L., Coulon, J., Marullo, P., Albertin, W., Bely, M., 2018. New oenological practice to promote non-*Saccharomyces* species of interest: saturating grape juice with carbon dioxide. *Appl. Microbiol. Biotechnol.* 102, 3779–3791.
- Chen, K., Escott, C., Loira, I., del Fresno, J.M., Morata, A., Tesfaye, W., Calderon, F., Suárez-Lepe, J.A., Han, S., Benito, S., 2018. Use of non-*Saccharomyces* yeasts and oenological tannin in red winemaking: influence on colour, aroma and sensorial properties of young wines. *Food Microbiol.* 69, 51–63.
- Ciani, M., Maccarelli, F., 1998. Oenological properties of non-*Saccharomyces* yeasts associated with wine-making. *World J. Microbiol. Biotechnol.* 14, 199–203.
- Coulter, A., Godden, P.W., Pretorius, I., 2004. Succinic acid-how is it formed, what is its effect on titratable acidity, and what factors influence its concentration in wine? *Aust. N. Z. Wine Ind. J.* 19, 16–25.
- Englezos, V., Rantsiou, K., Francesco, C., Torchio, F., Giacosa, S., Ortiz-Julien, A., Gerbi, V., Rolle, L., Cocolin, L., 2018. Volatile profiles and chromatic characteristics of red wines produced with *Starmarella bacillaris* and *Saccharomyces cerevisiae*. *Food Res. Int.* 109, 298–309.
- Englezos, V., Torchio, F., Craverio, F., Marengo, F., Giacosa, S., Gerbi, V., Rantsiou, K., Rolle, L., Cocolin, L., 2016. Aroma profile and composition of Barbera wines obtained by mixed fermentations of *Starmarella bacillaris* (synonym *Candida zemplinina*) and *Saccharomyces cerevisiae*. *LWT - Food Sci. Technol. (Lebensmittel-Wissenschaft -Technol.)* 73, 567–575.
- Escribano-Viana, R., Gonzalez-Arenzana, L., Portu, J., Garijo, P., Lopez-Alfaro, I., Lopez, R., Santamaria, P., Gutierrez, A.R., 2018. Wine aroma evolution throughout alcoholic fermentation sequentially inoculated with non-*Saccharomyces/Saccharomyces* yeasts. *Food Res. Int.* 112, 17–24.
- Fan, W., Xu, Y., Yu, A., 2006. Influence of oak chips geographical origin, toast level, dosage and aging time on volatile compounds of apple cider. *J. Inst. Brew.* 112, 255–263.
- Fariña, L., Villar, V., Ares, G., Carrau, F., Dellacassa, E., Boido, E., 2015. Volatile composition and aroma profile of Uruguayan Tannat wines. *Food Res. Int.* 69, 244–255.
- Ferreira, V., Sáenz-Navajas, M.P., Campo, E., Herrero, P., de la Fuente, A., Fernández-Zurbano, P., 2016. Sensory interactions between six common aroma vectors explain four main red wine aroma nuances. *Food Chem.* 199, 447–456.
- Gu, S.Q., Wang, X.C., Tao, N.P., Wu, N., 2013. Characterization of volatile compounds in different edible parts of steamed Chinese mitten crab (*Eriocheir sinensis*). *Food Res. Int.* 54, 81–92.
- Holt, S., Mukherjee, V., Lievens, B., Verstrepen, K.J., Thevelein, J.M., 2018. Bioflavoring by non-conventional yeasts in sequential beer fermentations. *Food Microbiol.* 72, 55–66.
- Jagtap, U.B., Bapat, V.A., 2015. Wines from fruits other than grapes: current status and future prospectus. *Food Biosci.* 9, 80–96.
- Jolly, N.P., Augustyn, O.P.H., Pretorius, I.S., 2006. The role and use of non-*Saccharomyces* yeasts in wine production. *S. Afr. J. Enol. Vitic.* 27, 15–39.
- Jolly, N.P., Varela, C., Pretorius, I.S., 2014. Not your ordinary yeast: non-*Saccharomyces* yeasts in wine production uncovered. *FEMS Yeast Res.* 14, 215–237.
- Lan, Y., Jin, W., Wang, X., Sun, X., Hackman, R.M., Li, Z., Feng, X., 2017. Evaluation of antioxidant capacity and flavor profile change of pomegranate wine during fermentation and aging process. *Food Chem.* 232, 777–787.
- Lee, P.R., Chong, I.S.M., Yu, B., Curran, P., Liu, S.Q., 2012. Effects of sequentially inoculated  *Williopsis saturnus* and *Saccharomyces cerevisiae* on volatile profiles of papaya wine. *Food Res. Int.* 45, 177–183.
- Liu, P.T., Lu, L., Duan, C.Q., Yan, G.L., 2016. The contribution of indigenous non-*Saccharomyces* wine yeast to improved aromatic quality of Cabernet Sauvignon wines by spontaneous fermentation. *LWT - Food Sci. Technol. (Lebensmittel-Wissenschaft -Technol.)* 71, 356–363.
- Loira, I., Morata, A., Comuzzo, P., Callejo, M.J., González, C., Calderón, F., Suárez-Lepe, J.A., 2015. Use of *Schizosaccharomyces pombe* and *Torulaspora delbrueckii* strains in mixed and sequential fermentations to improve red wine sensory quality. *Food Res. Int.* 76, 325–333.
- Lópezlopez, A., Sánchez, A.H., Cortésdelgado, A., Castro, A.D., Montaña, A., 2018. Relating sensory analysis with SPME-GC-MS data for Spanish-style green table olive aroma profiling. *LWT - Food Sci. Technol. (Lebensmittel-Wissenschaft -Technol.)* 89, 725–734.
- Magyar, I., Tóth, T., 2011. Comparative evaluation of some oenological properties in wine strains of *Candida stellata*, *Candida zemplinina*, *Saccharomyces uvarum* and *Saccharomyces cerevisiae*. *Food Microbiol.* 28, 94–100.
- Martínez, J.A., Melgosa, M., Pérez, M.M., Hita, E., Negueruela, A.I., 2001. Note. Visual and instrumental color evaluation in red wines. *Food Sci. Technol. Int.* 7, 439–444.
- Medina, K., Boido, E., Fariña, L., Gioia, O., Gomez, M.E., Barquet, M., Gaggero, C., Dellacassa, E., Carrau, F., 2013. Increased flavour diversity of Chardonnay wines by spontaneous fermentation and co-fermentation with *Hanseniaspora vineae*. *Food Chem.* 141, 2513–2521.
- Meng, J.F., Xu, T.F., Song, C.Z., Yu, Y., Hu, F., Zhang, L., Zhang, Z.W., Xi, Z.M., 2015. Melatonin treatment of pre-veraison grape berries to increase size and synchronicity of berries and modify wine aroma components. *Food Chem.* 185, 127–134.
- Mestre Furlani, M.V., Maturano, Y.P., Combina, M., Mercado, L.A., Toro, M.E., Vazquez, F., 2017. Selection of non-*Saccharomyces* yeasts to be used in grape musts with high alcoholic potential: a strategy to obtain wines with reduced ethanol content. *FEMS Yeast Res.* 17, fox010.
- Minnaar, P.P., Jolly, N.P., Paulsen, V., Du Plessis, H.W., Van Der Rijst, M., 2017. *Schizosaccharomyces pombe* and *Saccharomyces cerevisiae* yeasts in sequential fermentations: effect on phenolic acids of fermented Kei-apple (*Dovyalis caffra* L.) juice. *Int. J. Food Microbiol.* 257, 232–237.
- Noble, A.C., Bursick, G.F., 1984. The contribution of glycerol to perceived viscosity and sweetness in white wine. *Am. J. Enol. Vitic.* 35, 110–112.
- Cytology, taxonomy and ecology of grape and wine yeast. In: In: Ribereau-Gayon, P., Dubourdieu, D., Doney, B., Lonvaud, A. (Eds.), *Handbook of Enology—the Microbiology of Wine and Vinifications*, vol. 1. Wiley, Chichester, England, pp. 1–49.
- Robinson, A.L., Boss, P.K., Solomon, P.S., Trengove, R.D., Heymann, H., Ebeler, S.E., 2014. Origins of grape and wine aroma. Part 1. Chemical components and viticultural impacts. *Am. J. Enol. Vitic.* 65, 1–24.
- Rodrigues, H., Sáenz-Navajas, M.-P., Franco-Luesma, E., Valentin, D., Fernández-Zurbano, P., Ferreira, V., De La Fuente Blanco, A., Ballester, J., 2017. Sensory and chemical drivers of wine minerality aroma: an application to Chablis wines. *Food Chem.* 230, 553–562.
- Romano, P., Suzzi, G., Domizio, P., Fatichenti, F., 1997. Secondary products formation as a tool for discriminating non-*Saccharomyces* wine strains. *Antonie Leeuwenhoek* 71, 239–242.
- Rossouw, D., Bauer, F.F., 2016. Exploring the phenotypic space of non-*Saccharomyces* wine yeast biodiversity. *Food Microbiol.* 55, 32–46.
- Ruiz, M.J., Moyano, L., Zea, L., 2014. Changes in aroma profile of musts from grapes cv. *Pedro Ximenez* chamber-dried at controlled conditions destined to the production of sweet Sherry wine. *LWT - Food Sci. Technol. (Lebensmittel-Wissenschaft -Technol.)* 59, 560–565.
- Scanes, K.T., Prior, B.A., 1998. Glycerol production by the yeast *Saccharomyces cerevisiae* and its relevance to wine: a review. *South Afr. J. Enol. Vitic.* 19, 17–24.
- Song, C., Zuo, L., Shi, P., Meng, J., Wang, Y., Zhang, Z., Xi, Z., 2015. Aroma characterization of Chinese hutai-8 wines: comparing with Merlot and Cabernet Sauvignon wines. *Sci. Hortic.* 194, 237–245.
- Tofalo, R., Schirone, M., Torriani, S., Rantsiou, K., Cocolin, L., Perpetuini, G., Suzzi, G., 2012. Diversity of *Candida zemplinina* strains from grapes and Italian wines. *Food Microbiol.* 29, 18–26.

- Torchio, F., Segade, S.R., Gerbi, V., Cagnasso, E., Rolle, L., 2011. Changes in chromatic characteristics and phenolic composition during winemaking and shelf-life of two types of red sweet sparkling wines. *Food Res. Int.* 44, 729–738.
- Tosi, E., Fedrizzi, B., Finato, F., Simonato, B., Zapparoli, G., 2012. Effects of noble rot on must composition and aroma profile of Amarone wine produced by the traditional grape withering protocol. *Food Chem.* 130, 370–375.
- Wang, P.P., Li, Z., Qi, T.T., Li, X.J., Pan, S.Y., 2015. Development of a method for identification and accurate quantitation of aroma compounds in Chinese Daohuaxiang liquors based on SPME using a sol-gel fibre. *Food Chem.* 169, 230–240.
- Wang, X.J., Tao, Y.S., Wu, Y., An, R.Y., Yue, Z.Y., 2017. Aroma compounds and characteristics of noble-rot wines of Chardonnay grapes artificially botrytized in the vineyard. *Food Chem.* 226, 41–50.
- Welke, J.E., Zanus, M., Lazzarotto, M., Zini, C.A., 2014. Quantitative analysis of head-space volatile compounds using comprehensive two-dimensional gas chromatography and their contribution to the aroma of Chardonnay wine. *Food Res. Int.* 59, 85–99.
- Ye, M.Q., Yue, T.L., Yuan, Y.H., 2014a. Effects of sequential mixed cultures of *Wickerhamomyces anomalus* and *Saccharomyces cerevisiae* on apple cider fermentation. *FEMS Yeast Res.* 14, 873–882.
- Ye, M.Q., Yue, T.L., Yuan, Y.H., 2014b. Evolution of polyphenols and organic acids during the fermentation of apple cider. *J. Sci. Food Agric.* 94, 2951–2957.