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Evaluation of an aroma similar to that of sparkling wine: Sensory and gas chromatography analyses of fermented grape musts

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Abstract

The aim of our study was to verify the acceptability and volatile compound composition of grape musts fermented by yeasts commonly found in the natural grape microflora. A consumer panel determined the acceptability of the grape musts, comparing the aromas with that of a corresponding commercial sparkling wine. The volatile compounds were isolated by the purge and trap system. The highest means for acceptability were obtained by the Pinot Noir and Chardonnay musts fermented by *Pichia membranaefaciens* and by the Chardonnay must fermented by *Kloeckera apiculata*, with no significant difference amongst them ($p < 0.05$). Pinot Noir and Chardonnay musts fermented by *P. membranaefaciens* showed similar concentrations of the major fermentation aroma compounds (ethyl acetate, isoamyl acetate, acetaldehyde, ethanol, etc.) suggesting a correlation between the chemical and sensory data. The *P. membranaefaciens* isolate showed good potential as a producer of aroma similar to that of sparkling wine.

Keywords: Sparkling wine; Aroma production; Affective sensory tests; Purge and trap; Yeast

1. Introduction

Although a great number of volatile aroma compounds are found in grapes, most of those found in wine are formed during fermentation (Schreier, 1979), during which a vast number are formed by different metabolic routes, some yet to be clarified (Boulton, Singleton, Bisson, & Kunkee, 1996; Jackson, 1994). Mateo, Jimenez, Huerta, and Pastore (1991) studied the variation in volatile compounds produced in grape musts by yeasts such as *Hansenula*, *Kloeckera* and *Saccharomyces* and verified that the production was dependent on the species. In 1992 these same authors (Mateo, Jimenez, Huerta, & Pastore, 1992) observed differences in the quantities of the volatile compounds produced by the different species of *Saccharomyces cerevisiae*. *Kloeckera apiculata* produces great amounts of 1-propanol and isobutanol and small amounts of acetic acid as compared to *S. cerevisiae*, and this production is reflected in the sensory quality of the wine (Romano, Suzzi, Comi, & Zironi, 1992). Martinez, Valcarcel, Perez, and Benetiz (1998) investigated the production of aroma during the fermentation of Palomino grape must using species of *Saccharomyces cerevisise*. *S. cerevisise* (*montuliensis*) and *S. cerevisise* (*rouxii*) produced the highest concentrations of aroma compounds.

The volatile compounds determine the quality of each wine, especially the esters and higher alcohols produced during alcoholic fermentation (Valero, Moyano, Millan, Medina, & Ortega, 2002). The aroma is a result of the presence of volatile compounds, which make an impression on the olfactory system, which is extremely sensitive and can sense thousands of compounds. Sensory analysis allows us to study wine quality. It has been intensively used to classify wines based on their aromatic composition (Vannier, Brun, & Feinberg, 1999). The

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descriptive and affective sensory tests are very useful to classify and to evaluate the acceptability for beverages such as wine (Behrens & da Silva, 2000; Behrens, da Silva, & Wakeling, 1999). Many studies on the aromatic composition of wine use the chemical analysis of the aroma compounds together with the sensory analysis in order to establish a better quality control and to classify them (Iranzo, Magana, & Vinas, 2000; Lema, Gracia-Jares, Orriols, & Angulo, 1996; Noble, Flath, & Forrey, 1980).

The procedures for the isolation, concentration and quantification of volatile compounds from wine are very complicated due to the instability and low concentration levels of these components. Dynamic Headspace is an easily applied technique requiring little sample manipulation, detecting compounds present in low concentrations, and has been widely used for beverage and fruit analyses (Bertuccioli & Viani, 1976; Rosillo, Salinas, Garijo, & Alonso, 1999; Salinas, Alonso, & Esteben-Infantes, 1994; Stashenko, Macku, & Shibamato, 1992). The yeasts studied in this research are commonly found in grape microflora and some are rarely used in wine production, despite producing good aromas. Thus study verified the acceptability and purchasing intention of musts fermented by different yeast strains during seven days at 15 °C. The results are discussed on the basis of the data obtained in the affective sensory tests for acceptability and purchasing intention, together with the results of the analyses of the volatile compounds isolated using the Purge and Trap Concentrator/Dynamic Headspace system.

2. Material and methods

2.1. Fermentation

Chardonnay and Pinot noir grape musts from the "Serra Gaúcha" region (RS-Brazil) were used in order to study aroma formation by the yeasts *Pichia membranaefaciens*, *K. apiculata*, *Candida valida* and *S. cerevisiae* in sparkling wine. Fermentation started after the inoculation of 10^7 cells/ml into 125 ml conical flasks containing 25 ml of must. The samples were incubated at 15 °C, with shaking at 50 rpm, for seven days. After 168 h of fermentation the musts were filtered through a Millipore membrane (0.22 μm pore). The cell free must was frozen at −10 °C until the chromatographic analyses were performed.

2.2. Sensory analyses

The acceptability of eight samples of fermented must were evaluated using sensory affective tests, comparing with the aroma of sparkling wine. The samples were served to the 25 consumers in codified tulip-shaped glasses covered with watch glasses, using a monadic presentation and a 9-cm non-structured hedonic scale (Stone & Sidel, 1993). The consumers also registered their purchasing intentions for each sample on the same score sheet, using a five-point attitude scale (Meilgaard, Civille, & Carr, 1991).

2.3. Statistical analyses

The results obtained in the affective sensory tests and the concentrations of the major compounds produced in the fermented musts were evaluated by the unvaried statistics analysis (analysis of variance – ANOVA) and Tukey test.

The histograms produced using the score distributions obtained from the hedonic scale and from the data for purchasing intention were also analyzed.

2.4. Volatile compound analysis

(a) The volatile compounds were isolated using the Purge and Trap Concentrator/Dynamic Headspace system, model HP-G1900-60500 (Hewlett–Packard, USA), equipped with a Tenax trap. The conditions for the isolation and concentration of the volatile compounds were: sample temperature: 30 °C, purge time: 15 min, flow-rate: 30 ml/min, desorption temperature: 180 °C, desorption time: 10 min. The temperature transfer line used was 180 °C.

(b) Gas chromatography was carried out using an HP-G 1908-60500 (Hewlett–Packard, USA – FID) gas chromatograph coupled to a QP-5000 – Shimadzu – EM mass spectrometer. An HP-INNOWax (Hewlett–Packard, USA) 30 m × 0.25 mm ID capillary column coated with 0.25 μm layer of cross-linked polyethylene glycol was used. The carrier gas was helium (1 ml/min), and the temperature was programmed as follows: 35 °C/5 min, ramp: 3 °C/min → 140 °C → 180 °C/5 min. The detector temperature was 250 °C and the injector temperature 200 °C, splitless. The ionization voltage applied was 70 eV and the mass spectra were obtained in a scan range from 35 to 350 m/z.

(c) Quantification was carried out by an external standard method using the calibration graphs of the corresponding standard volatile compound supplied by Sigma-Aldrich and Merck. The analyses were carried out in triplicate.

3. Results and discussion

According to the ANOVA, there was a significant difference in acceptability of the aroma of the fermented must as compared to that of the sparkling wine for at least one of the samples tested. However, Tukey's test showed no significant difference amongst samples (1)–(7)

Table 1
Acceptability tests for the aroma of Pinot Noir and Chardonnay fermented musts

Samples	Acceptance mean
(1) Pinot Noir must fermented by (*)	5.50[a]
(2) Chardonnay must fermented by (*)	5.43[a]
(3) Chardonnay must fermented by (**)	5.42[a]
(4) Pinot Noir must fermented by (***)	4.88[ab]
(5) Chardonnay must fermented by (***)	4.83[ab]
(6) Pinot Noir must fermented by (****)	4.58[ab]
(7) Chardonnay must fermented by (****)	4.00[ab]
(8) Pinot Noir must fermented by (**)	3.37[b]

Pure culture: (*) *Pichia membranaefaciens*; (**) *Kloeckera apiculata*; (***) *Candida valida* and (****) *Saccharomyces cerevisiae*. Means with the same letter do not differ significantly by Tukey's test ($p < 0.05$).

(Table 1). Although there was no statistical difference, the Pinot noir and Chardonnay musts fermented by *P. membraanefaciens* and the Chardonnay must fermented by *K. apiculata* presented higher acceptance means: 5.50, 5.43 and 5.42, respectively. These averages correspond to the concept of "liked moderately" (Table 1).

Figs. 1 and 2 show the purchasing intention of the eight samples analyzed by the ANOVA and Tukey tests. The samples showing higher acceptance means (Table 1) also showed higher purchasing intention. For the Pinot Noir must fermented by *P. membranaefaciens* 35.5% of the consumers showed a purchasing intention corresponding to the concept "probably would buy", as did 32.5% of the consumers for the Chardonnay musts fermented by *P. membranaefaciens* and by *K. apiculata*. For the samples (2) and (3), 16% of the consumers stated a purchasing intention of "definitely would buy".

For the Pinot Noir must fermented by *C. valida* (sample 4), 32% of the consumers gave values for purchasing intention corresponding to the concept "maybe/maybe not" and 23% corresponding to "probably would not buy". Very low values (≅20%) were obtained for the concept "probably would buy" by sample (4).

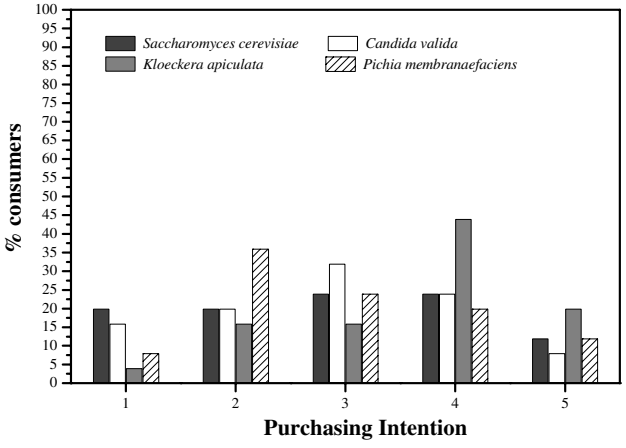

Fig. 1. Consumer purchasing intention for Pinot Noir fermented must. 1, definitely would buy; 2, probably would buy; 3, maybe/maybe not; 4, probably would not buy; 5, definitely would not buy.

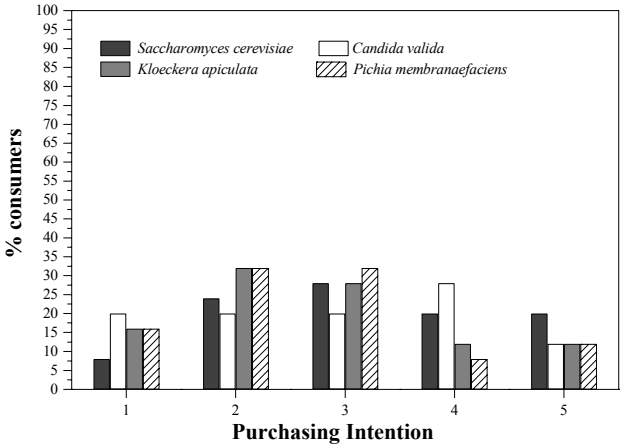

Fig. 2. Consumer purchasing intention for Chardonnay fermented must. 1, definitely would buy; 2, probably would buy; 3, maybe/maybe not; 4, probably would not buy; 5, definitely would not buy.

Although the Pinot Noir must fermented by *S. cerevisiae* (sample 6) and the Chardonnay must fermented by *S. cerevisiae* (sample 7) received scores for purchasing intention corresponding to the concept "probably would buy" from 24% to 20% of the consumers, respectively, these samples presented high percentages for purchasing intention corresponding to the concepts "maybe/maybe not", "probably would not buy" and "definitely would not buy" (Figs. 1 and 2).

The results for purchasing intention were in accordance with the means for acceptance, with samples showing higher means for acceptance also receiving more scores for purchasing intention corresponding to the concepts "definitely would buy" or "probably would buy".

The chemical compositions of the three samples showing greater acceptance means and purchasing intention were very similar, as shown in Table 2. Compounds like 2-propanol, butyl acetate, phenethyl acetate and 3-hydroxy-2-butanone were found in samples (1) and (2) occurred, whereas these compounds were not observed in sample (3). Nevertheless, the presence of propyl acetate and ethyl propionate was also detected in sample (3). Butyl isobutyrate was present in all of the fermented Chardonnay musts and is a constituent compound of pure Chardonnay must.

Compounds such as butyl acetate and phenethyl acetate that are of great sensory importance in sparkling wines (Garofolo, Morassut, & Ciolfi, 1990), were only detected in the musts fermented with *P. membranaefaciens* and *C. valida* (Table 2).

The usual description for isoamyl acetate is sweet, fruity and banana-like (Simpson, 1979). This compound was detected in all the samples analyzed.

Acetate ester compounds such as ethyl acetate and isoamyl acetate have a profound effect on the quality of the aroma of sparkling wine (Lepe & Leal, 1990) and

Table 2
General composition of the volatile compounds from the fermented musts

Volatile compounds	Samples							
	(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)
Ethanol	×	×	×	×	×	×	×	×
1-Propanol	×	×	×	×	×	×	×	×
2-Propanol	×	×						
1-Butanol						×	×	
1-Hexenol	×	×	×	×	×	×	×	×
2-Methyl propanol	×	×	×	×	×	×	×	×
3-Methyl butanol	×	×	×	×	×	×	×	×
Methyl acetate	×	×	×	×	×			×
Ethyl acetate	×	×	×	×	×	×	×	×
Propyl acetate			×					×
Butyl acetate	×	×		×	×			
Ethyl butyrate		×	×		×		×	
Isopropyl acetate	×	×	×	×	×			×
Isoamyl acetate	×	×	×	×	×	×	×	×
Butyl Isobutyrate		×	×		×		×	
Ethyl propionate			×					×
Phenethyl acetate	×	×		×	×			
3-Hydroxy-2-butanone	×	×				×	×	
Acetaldehyde	×	×	×	×	×	×	×	×
Butanal						×	×	
Isobutyraldehyde						×	×	
3-Methyl butanal						×	×	
Propanal						×	×	
Acetic acid	×	×	×	×	×	×	×	×
2-Ethyl hexanoic acid	×	×	×	×	×	×	×	×

Samples: (1) P. Noir must fermented by *P. membranaefaciens*. (2) Chardonnnay must fermented by *P. membranaefaciens*. (3) Chardonnay must fermented by *K. apiculata*. (4) P. Noir must fermented by *C. valida*. (5) Chardonnay must fermented by *C. valida*. (6) P. Noir must fermented by *S. cerevisiae*. (7) Chardonnay must fermented by *S. cerevisiae*. (8) P. Noir must fermented by *K. apiculata*.

these compounds were detected in all the fermented must samples (Table 2).

Esters are mainly produced by yeast during alcoholic fermentation, in reactions between alcohols and acetyl-CoA catalyzed by alcohol acetyltransferase and other enzymes. Ethanol is the main alcohol in wine, therefore ethyl acetate produced by ethanol and acetyl-CoA is the major ester formed during fermentation. Other acyl-CoA compounds also show similar behaviors in the production of other esters.

Higher alcohols are produced from amino acids and sugar metabolism. 3-Methyl butanol, 2-methyl propanol and 1-propanol are the principal higher alcohol constituents (Boulton et al., 1996).

The concentrations of the major volatile compounds produced in the alcoholic fermentations are shown in Table 3.

The data for the concentrations of the major fermentation compounds in the eight samples were analyzed together, as were the data for acceptability, in order to establish correlations between the sensory and chemical data.

According to Tukey's test there were no significant differences between the productions of 1-propanol by the different yeasts. The mean production of this compound was in the range from 8 to 10.7 mg/l. Garofolo et al. (1990) showed that good sparkling wines presented 1-propanol concentrations in the range from 5 to 27 mg/l, thus the results obtained in this study were consistent with previous reports.

In wines, ethyl acetate exhibits a fruity aroma but in concentrations exceeding 200 mg/l it produces a solvent-like odor. However, the concentrations reported here for all the samples were within the established limits for wine.

Samples (1) and (2), which showed the highest scores for the acceptability, showed no significant differences between the concentrations for the following volatile compounds: ethyl acetate, isoamyl acetate, acetaldehyde, ethanol, 1-propanol and 2-methyl butanol (Table 3).

The type of must did not influence the amounts of the major compounds or the general composition of the aroma compounds formed during fermentation by *P. membranaefaciens*. Moreover, the amounts of the major volatile compounds were consistent with the established composition for sparkling wine.

The Chardonnay must fermented by *K. apiculata* was the third sample in order of acceptability (sample (3),

Table 3
Analysis of variance of major compound production in fermented musts

Sample	Mean production of major compounds						
	Ethyl acetate (mg/l)	Isoamyl acetate (mg/l)	Acetaldehyde (mg/l)	Ethanol (g/l)	1-Propanol (mg/l)	2-Methyl propanol (mg/l)	3-Methyl butanol (mg/l)
1	131.3^{bc}	27.3^{a}	32.3^{c}	24.7^{ba}	8.00^{a}	25.7^{dc}	97.3^{a}
2	147.0^{bc}	28.0^{a}	35.3^{c}	25.0^{ba}	8.33^{a}	27.3^{bc}	79.7^{b}
3	200.0^{a}	19.7^{ba}	15.0^{d}	12.0^{bc}	9.67^{a}	18.0^{d}	98.3^{a}
4	151.0^{bc}	26.7^{a}	38.0^{c}	22.7^{ba}	10.7^{a}	38.3^{a}	95.3^{ba}
5	165.0^{ba}	27.0^{a}	43.3^{c}	25.7^{a}	8.00^{a}	40.0^{a}	85.0^{ba}
6	120.0^{cd}	14.7^{b}	80.3^{b}	27.3^{a}	6.00^{a}	36.0^{ba}	5.00^{c}
7	110.0^{d}	15.0^{b}	99.0^{a}	28.0^{a}	8.00^{a}	37.3^{a}	5.00^{c}
8	197.0^{a}	19.3^{ba}	14.3^{d}	6.3^{c}	9.33^{a}	20.0^{dc}	98.0^{a}

Samples: (1) P. Noir must fermented by *P. membranaefaciens*. (2) Chardonnnay must fermented by *P. membranaefaciens*. (3) Chardonnay must fermented by *K. apiculata*. (4) P. Noir must fermented by *C. valida*. (5) Chardonnay must fermented by *C. valida*. (6) P. Noir must fermented by *S. cerevisiae*. (7) Chardonnay must fermented by *S. cerevisiae*. (8) P. Noir must fermented by *K. apiculata*. Means with the same letter do not differ significantly by Tukey's test ($p < 0.05$).

Table 1). This sample presented slightly lower mean values for the concentrations of the following major fermentation compounds as compared to sample (2) (Chardonnay must fermented by *P. membranaefaciens*): ethanol, 2-methyl butanol and isoamyl acetate (Table 3), but these differences were not significant when compared to the sample with higher acceptance means. The sensory and chemical data for samples (1)–(3) suggest a possible correlation between the two sets of data. The acceptance means for these samples correspond to a concept of ''liked moderately''. This is a reasonably good concept considering that these samples did not show a concept giving a doubt of choice, such as ''neither like nor dislike''.

Samples (4) and (5) (Pinot Noir and Chardonnay grape musts fermented by *C. valida*) showed very similar concentrations for their major fermentation volatile compounds, which did not differ significantly (Table 3). The acceptation means were practically the same, 4.88 and 4.83 for samples (4) and (5), respectively, corresponding to the concept ''neither like nor dislike'' (Table 1).

Samples (6) (Pinot Noir must fermented by *S. cerevisiae*) and (7) (Chardonnay must fermented by *S. cerevisiae*) differed from all the other samples with respect to the amounts of ethyl acetate, isoamyl acetate and 3-methyl butanol (Table 3). These volatile compounds show an important contribution to wine aroma, as mentioned above. Thus the low acceptance of these samples, as shown in Table 1, was probably due to the low production of the compounds ethyl acetate, isoamyl acetate and 3-methyl butanol. Another curious fact is the presence of some low boiling point aldehydes such as propanal and butanal (Table 2). In general, the aldehydes are reduced to their respective alcohols, and therefore at the end of fermentation their quantities are reduced. In these samples the complete reduction of the aldehydes to alcohols did not occur. Although these samples showed low acceptance values, probably because of the low concentrations of ethyl acetate, isoamyl acetate and 3-methyl butanol, they presented acetaldehyde concentrations within the limits established for sparkling wine (Garofolo et al., 1990).

The fermentation of Pinot Noir must by *K. apiculata* (sample 8) resulted in the lowest mean values for acceptance: 3.37, corresponding to the concept of ''dislike slightly'', and consequently this sample also showed a low purchasing intention, about 43% of the consumers opting for the concept ''probably would not buy'' and only 4% opting for the concept ''definitely would buy''. The concentrations of the major compounds in this sample were as follows: ethyl acetate 197.0 mg/l; isoamyl acetate 19.3 mg/l; acetaldehyde 14.3 mg/l; ethanol 6.3 g/l; 1-propanol 9.3 mg/l; 2-methyl propanol 20.0 mg/l and 3-methyl butanol 98.0 mg/l. The concentration of volatile compounds in the Chardonnay and Pinot Noir musts fermented by *K. apiculata* were statistically similar, with the exception of ethanol (Table 3).

The increase or decrease in aroma volatility is correlated with the nature and interactions of the non-volatile components (Lubbers, Voilley, Charpentier, & Feuillat, 1994) with the volatile compounds (Willians & Rosser, 1981), greatly influencing the overall aroma of the wine.

Probably the liberation of these compounds from the matrix to the headspace suffers from the influence of the interactions between the non-volatile and volatile compounds, but the statistical difference in acceptability of the Pinot Noir must fermented by *K. apiculata* (sample 8) cannot be based exclusively on these interactions. It is recognized that ethanol plays a major role in the volatility of the flavors and the sensory quality of the wine (Voilley & Lubbers, 1998). Thus the amount of volatile compounds perceived by the olfactory system is greatly dependent on the ethanol concentration (Rothe & Schrodter, 1996).

Consequently, a low mean for sample acceptance (8) is correlated with a low production of ethanol.

4. Conclusions

From the affective sensory testes for acceptability and purchasing intention, little difference in aroma was shown between that of the fermented musts and that of sparkling wine. The results of the affective sensory tests showed good correlation with the data from the chemical analyses of the major compounds of the two samples with the highest acceptance and purchasing intention. *P. membranaefaciens* was shown to be suitable for the production of sparkling wine aroma. *S. cerevisiae* has been widely used for the production of sparkling wine, but this study showed that the aroma produced by this yeast presented low scores for the acceptability and purchasing intention.

Ethanol production by *K. apiculata* in Pinot Noir must was even lower than in Chardonnay must, this fact possibly being the reason for its low acceptance mean, i.e. the low ethanol levels could lead to higher volatilization of the aroma compounds resulting in a negative response and therefore low sample (8) acceptance.

A discriminative sensory analysis of the flavor and aroma of the musts produced in industrial fermentations should be carried out.

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References

Behrens, J. H., & da Silva, M. A. A. (2000). Perfil sensorial de vinhos brancos varietais brasileiros através de análise descritiva quantitativa. *Ciência e Tecnologia de Alimentos, 20*(1), 60–67.

Behrens, J. H., da Silva, M. A. A., & Wakeling, I. N. (1999). Avaliação da aceitação de vinhos brancos varietais brasileiros através de testes sensoriais afetivos e técnica multivariada de mapa de preferência interno. *Ciência e Tecnologia de Alimentos, 19*(2), 214–220.

Bertuccioli, M., & Viani, R. (1976). Red wine: Identification of headspace constituents. *Journal Science Food Agricultural, 27*, 1035–1038.

Boulton, R. B., Singleton, V. I., Bisson, I. F., & Kunkee, R. E. (1996). *Principles and practices of winemaking*. Davis: Chapman & Hall, pp. 102–181.

Garofolo, A., Morassut, M., & Ciolfi, G. (1990). Composizione della base spumante in relazione al metabolismo di stipiti di specie diverse. *Industrie delle Bevande, XIX*, 388–393.

Iranzo, J. F. U., Magana, F. G., & Vinas, M. A. G. (2000). Evaluation of the formation of volatiles and sensory characteristics in the industrial production of white wines using different comercial strains of the genus *Saccharomyces*. *Food Control, 11*, 143–147.

Jackson, R. S. (1994). *Wine science: Principles and applications*. London: Academic Press, pp. 220–276.

Lema, C., Gracia-Jares, C., Orriols, I., & Angulo, L. (1996). Contribution of *Saccharomyces* and non-*Saccharomyces* populations to the production of some components of Albariño wine aroma. *American Journal Enological and Viticulture, 47*(2), 206–216.

Lepe, J. A. S., & Leal, B. I. (1990). Microbiologia Enologica – Fundamentos de vinificación. Madrid: Ediciones Mundi-Prensa. pp. 475–482.

Lubbers, S., Voilley, A., Charpentier, C., & Feuillat, M. (1994). Influence of yeast walls on the behaviour of aroma compounds in a model wine. *American Journal Enological and Viticulture, 45*, 29–33.

Martinez, P., Valcarcel, M. J., Perez, L., & Benetiz, T. (1998). Metabolism of *Saccharomyces cerevisiae* flor yeast during fermentation and biological aging of fino sherry: By-products and aroma compounds. *American Journal Enology and Viticulture, 49*(3), 240–250.

Mateo, J. J., Jimenez, M., Huerta, T., & Pastore, A. (1991). Contribution of different yeast isolated from musts of monastrell grapes to the aroma of wine. *International Journal of Food Microbiology, 14*, 153–160.

Mateo, J. J., Jimenez, M., Huerta, T., & Pastore, A. (1992). Comparison of volatiles produced by four *Saccharomyces cerevisiae* strains isolated from Monastrell Musts. *American Journal Enological and Viticulture, 43*(2), 206–209.

Meilgaard, G. K., Civille, G. V., & Carr, B. I. (1991). *Sensory evaluation techniques: Marketing and R&D approaches*. Westport: Food & Nutrition Press.

Noble, A. C., Flath, R. A., & Forrey, R. R. (1980). Wine headspace analysis. Reproducibility and application to varietal classification. *Journal Agricultural Food Chemistry, 28*, 346–353.

Romano, P., Suzzi, G., Comi, G., & Zironi, R. (1992). Higher alcohol and acetic acid production by apiculate wine yeasts. *Journal of Applied Bacteriology, 73*, 126–130.

Rosillo, L., Salinas, M. R., Garijo, J., & Alonso, G. L. (1999). Study of volatiles in grapes by dynamic headspace analyses. Application to the differentiation of some *Vitis vinifera* varieties. *Journal of Chromatography A, 847*, 155–159.

Rothe, M., & Schrodter, R. (1996). Flavour contribution of ethanol, a neglected aroma compound. In A. J. Taylor & D. S. Mottram (Eds.), *Flavour science* (pp. 348–349). Cambridge, UK: The Royal Society of Chemistry.

Salinas, M. R., Alonso, G. L., & Esteben-Infantes, F. J. (1994). Adsorption-thermal desorption-gas chromatography applied to the determination of wine aromas. *Journal Agricultural and Food Chemistry, 42*, 1328–1331.

Schreier, P. (1979). Flavor composition of wines: A review. *Critical Reviews in Food Science and Nutrition*, 59–111.

Simpson, R. F. (1979). Some important aroma components of white wines. *Food Technological Australian*, 516–522.

Stashenko, H., Macku, C., & Shibamato, T. (1992). Monitoring volatile chemicals formed from must during yeast fermentation. *Journal Agricultural and Food Chemistry, 40*, 2257–2259.

Stone, H. J., & Sidel, J. L. (1993). *Sensory evaluation practices*. London: Academic Press.

Valero, E., Moyano, L., Millan, M. C., Medina, M., & Ortega, J. M. (2002). Higher alcohols and esters production by *Saccharomyces cervisiae*. Influence of the inicial oxygenation of the grape must. *Food Chemistry, 78*, 57–61.

Vannier, A., Brun, O. X., & Feinberg, M. H. (1999). Application of sensory analysis to champagne wine characterization and discrimination. *Food Quality and Preference, 10*, 101–107.

Voilley, A., & Lubbers, S. (1998). Flavor–matrix interactions in wine. In A. L. Waterhouse & S. E. Ebeler (Eds.), *Chemistry of wine flavor* (pp. 217–229). Washington, DC: American Chemical Society.

Willians, A. A., & Rosser, P. R. (1981). Aroma enhancing effects of ethanol. *Chemical Senses, 6*(2), 149–153.