We are IntechOpen,
the world's leading publisher of
Open Access books
Built by scientists, for scientists

6,900
Open access books available

186,000
International authors and editors

200M
Downloads

Our authors are among the

154
Countries delivered to

TOP 1%
most cited scientists

12.2%
Contributors from top 500 universities

WEB OF SCIENCE™

Selection of our books indexed in the Book Citation Index
in Web of Science™ Core Collection (BKCI)

Interested in publishing with us?
Contact book.department@intechopen.com

Numbers displayed above are based on latest data collected.
For more information visit www.intechopen.com

The Aroma of Rojal Red Wines from La Mancha Region – Determination of Key Odorants

Eva Sánchez-Palomo, Eva Gómez García-Carpintero,
Manuel Ángel Gómez Gallego and Miguel Ángel González Viñas
University of Castilla-La Mancha/Area of Food Technology,
Spain

1. Introduction

Wine aroma is one of the most influential properties when it comes to consumer preference, and is mainly determined by the volatile compounds. The flavour of young wines results from a series of different biochemical and technological processes. Formation of volatile compounds begins in the grape, while during juice production, fermentation, maturation, ageing and storage the chemical composition continues to change. The amount and type of chemicals that influence wine flavour therefore depend on many factors including the origin of the grapes, grape varieties and ripeness, soil and climate, yeast used during fermentation and a variety of other winemaking practices (Kotseridis & Baumes, 2000; Rapp, 1998; Spranger et al., 2004).

Grape aroma compounds mainly appear either in their free form, directly contributing to wine aroma, or as non-volatile sugar-bound conjugates (monoglucosides or disaccharide glucosides), these being the predominant form in aromatic varieties (Günata et al., 1985). To release the aglycones that enrich wine aroma, bound forms must be subjected to acid or enzyme hydrolysis, normally using commercial preparations with β-glucosidase activity (Marais & Rapp, 1988; Carballeira et al., 2001). Over the past ten years, many aroma compounds in monovarietal wines have been identified and quantified. Particular attention has been devoted recently to the analytical characterization and the quality improvement of the varietal aroma of wines. Several studies have focused on the identification of volatile components of different varieties including the volatile components originating from the nonvolatile precursors (Gunata et al., 1985; Sánchez-Palomo et al., 2006, 2007; Ugliano et al., 2006; Ugliano & Moio, 2008; Rocha et al., 2010; García-Carpintero et al., 2011a, 2011b). These precursors have been reported as glycosides having the aroma compounds as their aglycons. The knowledge of the varietal volatile composition offers a means of evaluating the potential aroma of a variety and to improve the wine aroma quality.

To understand the chemical compounds in wine that showed sensory characteristics, it is necessary to obtain some information regarding both volatile composition and sensory properties (Francis & Newton, 2005). Gas chromatography is an important analysis technique for volatile and non-volatile components to the aroma of the wine. However, the wine volatile fraction is extremely complex, mainly because of the great number of

compounds which form it. To date, more than 1000 compounds have been identified, which are from different chemical classes, covering a wide range of polarities, solubility and volatilities. The volatile compounds responsible for the varietal aroma are present in only trace amounts, which means that to carry on their identification and quantification, an effective method of enrichment is required prior to their analysis by gas chromatography–mass spectrometry (GC–MS).

The search of adequate extraction techniques allowing the identification and quantification of wine volatile compounds has attracted the attention of many scientists. This has resulted in the availability of a wide range of analytical tools for the extraction of these compounds from wine. These methodologies are mainly based on the solubility of the compounds in organic solvents (liquid–liquid extraction: LLE, simultaneous distillation liquid extraction: SDE), on their volatility (static and dynamic headspace techniques), or based on their sorptive/adsorptive capacity on polymeric phases (solid phase extraction: SPE, solid phase microextraction: SPME, stir bar sorptive extraction: SBSE). In addition, volatile compounds can be extracted by methods based on combinations of some of these properties (headspace solid phase microextraction HS-SPME, solid phase dynamic extraction: SPDE).

One of the most commonly used method for the analysis of volatile compounds in wine is SPE. The possibility of using different sorbent phases and eluents makes SPE a very selective technique, and the fact that only minor amounts of organic solvents are used compared to LLE, is why SPE has been extensively used for the analysis of volatile aroma compounds (Ferreira et al, 1998; Dominguez et al., 2002; Lopez et al., 2002; Ibarz et al., 2006; Campo et al., 2007; Loscos et al., 2009) and off-flavours (Dominguez et al., 2002; Insa et al., 2005) in wines. Solid-phase extraction (SPE) is widely used in analytical laboratories for either sample extraction or sample clean up procedures. This technique based on adsorbent materials where analytes are bound to active sites on a surface, allows the determination of a wide range of volatile compounds, requires smaller quantities of solvents and shorter time of analyses but is relatively tedious. Many benefits of SPE methods have been commonly cited including its robustness, potential for automation, capacity for providing clean extracts, selective isolations and even a fractionation of the different sample components. For these reasons, SPE is a powerful pre-concentration technique which can be easily adapted for routine analysis and, in fact, many studies based on SPE procedures for monitoring different compounds in wine samples have been published in the last years (Vianna & Ebeler, 2001; Rodriguez-Bencomo et al., 2002; Sala et al., 2002). However, as the SPE systems have a low number of chromatographic plates (Vianna & Ebeler, 2001) the selectivity (measured as the ratio between the chromatographic retention factors of analytes and interferences) must be high in order to get good separations. SPE has been successfully used to study the evolution of aromatic compounds of grapes during ripening (Lopez et al., 2002) and to determine the potential aroma in several varieties of Spanish grapes (Loscos et al., 2009).

Over the last few decades the introduction and spread of world renowned varieties has caused a massive loss of indigenous grapevine varieties traditionally grown in various grape-growing regions. *Vitis vinifera cv*. Rojal a minority grape variety is cultivated in La Mancha region in little areas with special climatologic conditions (warm summers, cold winters and low rain) that could influence on its aroma composition cultivated in a little restringed area. Only the knowledge of the chemical composition and sensory properties of

this variety can give opportunities for the adaptation of the characteristics of this minority grape variety to the winemaking procedures ruled by the consumer's preferences. As far as we know, the aroma of this grape variety has not yet been characterized.

The aim of this study was to characterize the free and bound volatile aroma compounds of Rojal red wines from La Mancha region (Spain) during four consecutive vintages by GC-MS, and determine the key odorants of the aroma of these wines.

2. Material and methods

2.1 Wine samples

Red, *cv.* Rojal grapes were obtained from the vineyards of La Mancha in the central-southeastern region of Spain. They were harvested at their optimal stage of ripeness and in health conditions, over four consecutive vintages (2007-2010).

Wines were elaborated from two batches of grapes (500 kg each) were elaborated in 250 l-Stainless steel tanks with skin maceration until the alcoholic fermentation. Winemaking conditions, included the addition of 100 ppm of SO_2, as $K_2S_2O_7$, after stemming and crushing, inoculation with *Saccharomyces cerevisiae* selected yeasts (UCLM S325, Fould-Springer), and fermentation temperature maintained at 24ºC. Manual punching down was done twice a day. Separation of wines from solids was performed when relative density reached a constant value. Subsequently, the malolactic fermentation was induced by inoculation with *Oenococcus oeni* lactic acid bacteria (Lactobacter SP1; Laffort). This second fermentation terminated in 2–3 weeks, as confirmed by TLC (Thin Layer Chromatography); the wines were then racked. After one month, the wines were racked again, filtered through 1.2 µm membranes (Millipore, Bedford, MA, USA), bottled, and stored in room with a constant temperature between 16 and 18 ºC.

2.2 Reagents and standards

Dichloromethane and methanol were purchased from Merck (Darmstadt, Germany). Ammonium sulfate and anhydrous sodium sulfate were from Panreac (Barcelona, Spain). Pure water was obtained from a Milli-Q purification system (Millipore, U.S.). LiChrolut EN resins were purchased from Merck (Darmstadt, Germany). The chemical standards were supplied by Sigma (St. Louis, MO, USA), Aldrich (Gillingham, UK), Firmenich (Geneva, Switzerland), Panreac (Barce lona, Spain), Merck (Darmstadt, Germany), Fluka (Buchs, Switzer land), and Lancaster (Strasbourg, France). An alkane solution (C8–C28) in dichloromethane was employed to calculate the linear retention index (LRI) of each analyte.

2.3 Analysis of major volatiles

Major volatile compounds were analyzed by direct injection (Sánchez-Palomo et al., 2006) of a HP-5890 GC with a FID detector, using a CP-Wax-57 capillary column (50 m×0.25mm i.d.; 0.25 µm film thickness). The oven temperature program was: 40 °C (5 min)–4 °C/129 min–120 °C. Injector and detector temperature were 250 and 280 °C, respectively. One microliter (1 µl) was injected in split mode, split ratio 1:15. Carrier gas was He (0.7 ml/min).

2.4 Extraction of minor volatiles

The aroma compounds were separated by adsorption/desorption on preconditioned polypropylene-divinylbenzene cartridges (Sánchez-Palomo et al., 2006) (LiChrolut EN, Merck, 0.5 g of phase). One hundred milliliters of wine added of 40 µl of 4-nonanol, as an internal standard, was passed through the LiChrolut EN column at a flow rate of 1 ml/min. The column was rinsed with 50 ml of pure water to eliminate sugars and other low-molecular-weight polar compounds. The free fraction was eluted with 10 ml of dichloromethane. All dichloromethane extracts were cooled to−20 °C to separate the frozenwater from the organic phase by decantation, and then dried over anhydrous sodiumsulfate using nitrogen stream, the organic phase was concentrated to a final volume of 200 µl. The bound fraction was eluted with 25ml of ethyl acetate. Ethyl acetate extracts were evaporated to dryness under vacuum, and then re-dissolved with 1 ml of methanol.

2.5 Enzymatic hydrolysis of bound fraction

A 500 µl methanol extract was evaporated to dryness under nitrogen stream. The dried glycosidic extract was dissolved in a 100 µl citrate–phosphate buffer (0.2 M, pH 5). Enzymatic treatment with AR2000 (Gist Brocades) was completed at 40 °C for 18 h according to optimum conditions described previously (Sánchez-Palomo et al., 2006). The mixture was then extracted five times with 2 ml of pentane–dichloromethane (2:1 v/v). After adding 4-nonanol (1 g/l) as the internal standard the extract was concentrated to a final volume of 200 µl under nitrogen stream.

2.6 Gas Chromatography–Mass Spectrometry (GC–MS) analysis

An Agilent Gas Chromatograph model 6890N coupled to a Mass Selective Detector model 5973 inert equipped with a BP-21, Polyethylene glycol TPA treated, capillary column (60 m×0.25 mm i.d.; 0.25 µm film thickness) was used. Operating conditions were as follows. Oven temperature program was: 70 °C (5min)–1 °C/min–95 °C (10min)–2 °C/min–200 °C (40 min). Injector and transfer line temperatures were 250 °C and 280 °C, respectively. Mass detector conditions were: electron impact (EI) mode at 70 eV; source temperature: 178 °C; scanning rate: 1 scan/s; mass acquisition: 40–450 amu. One microlitre (1 µl) was injected in splitless mode. Carrier gas was helium (1 ml/min). Retention time, Wiley mass-spectral library, and pure volatile compounds were used for identification, confirmation and preparation of standard solutions of volatile compounds. The relative response areas for each of the volatile compounds to the internal standard were calculated and interpolated in the corresponding calibration graphs. For the calibration, standard solutions were prepared in 12% v/v ethanol with 5 g/l tartaric acid and the corresponding internal standard in the same concentration as in the samples. Calibration curves were drawn for each standard at eight different concentration levels. The measurements of all standards were performed in triplicate. When the authentic standard were not available the identification was based on the comparison with the spectral data of Wiley A library and the chromatographic dates of the literature, semi-quantitative analysis of these compounds were made assuming response factor equal to one.

2.7 Odor activity values

To evaluate the contribution of a chemical compound to the aroma of a wine the odor activity value (OAV) was determined. OAV is a measure of importance of a specific compound to the odor of a sample. It was calculated as the ratio between the concentration of an individual compound and the perception threshold found in literatures (Francis & Newton, 2005; Vilanova, et al., 2008).

2.8 Sensory analysis

Wines were evaluated in duplicate by a panel consisting in 10 experienced wine-testers (7 female and 3 male) ranging ages from 24 to 45 years. Assessment took place in a standard sensory-analysis chamber (ISO 8589, 1998) equipped with separate booths. Wines were sniffed. Three wines were presented in each session, in coded standard wine-testing glasses according to standard ISO 3591, 1997 and covered with a watch-glass to minimize the escape of volatile components. Testing temperature of wine was 18º C.

2.9 Statistical analysis

Analysis of variance (ANOVA) was performed using the general linear model procedure to determine significant differences in the concentration of volatile compounds of Rojal wines on different vintages. Student–Newman–Keuls test was conducted when the samples exhibited significance between them, with the level of significance set at P<0.05. Both ANOVA and Student–Newman–Keuls test were performed with SPSS 19.0 (2010) for Windows statistical package.

3. Results and discussion

3.1 Free volatile aroma composition of Rojal wines

The free volatile compounds of wines were extracted with dichloromethane. Representative wine aroma extracts for chemical and olfactory analysis were obtained using this solvent. Fig. 1 is the TIC of free volatile compounds of Rojal wines detected by SPE–GC–MS. Quantitative data of the volatile compounds found in free aroma fraction of the young red wines from Rojal grape variety are shown in Tables 1 and 2. The data are expressed as means (µg/l) of the GC–MS analyses of duplicate extractions and they correspond to the average of the analyzed wines. Improvement in the analytical method used to extract the volatile compounds from these wines has allowed us to identify and quantify 80 free volatile compounds in Rojal red wines including alcohols, esters, acids, terpenes, C_{13} norisoprenoids, C_6 compounds and benzenic compounds. They have been positively identified and quantitatively determined.

3.1.1 Varietal aroma compounds

C_6 compounds. All Rojal wines displayed higher concentration of C_6 alcohols, principally 1-hexanol. C6 compounds, which supply "vegetal" and "herbaceous" nuances to the wine, usually have a negative effect on wine quality when their concentration is above their odor threshold values (Ferreira et al., 1995). However, 1-hexanol and (Z)-3-hexen-1-ol were found
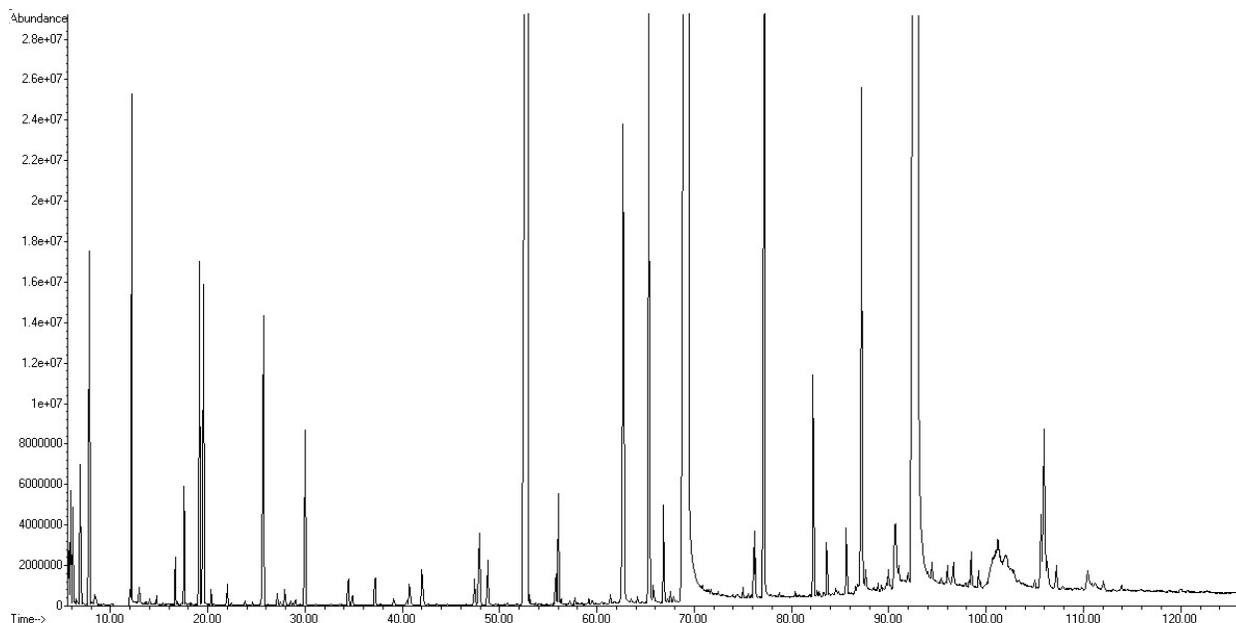
Fig. 1. TIC of free volatile compounds of Rojal wines detected by SPE–GC–MS.

at concentrations under their odor threshold values (8000 µg/l and 400 µg/l respectively) in the analyzed Rojal wines. Rojal wines from 2008 vintage showed the lower concentration of C_6 compounds, due to a lightly superior ripening stage than the rest of studied vintages. Among the compounds found, we give prominence to the ratio between *trans-* and *cis*-3-hexen-1-ol contents. As previously reported (Hatanaka, 1993), the composition of the C_6 compounds is strongly dependent on four enzymes, which catalyze the biosynthesis of these compounds, and among these four, lipoxygenase and hydroperoxide lyase are particularly important. Thus, the level and relationships between these compounds could be considered as characteristic of the *V. vinifera variety*. Specifically, for Rojal, *cis*-3-hexen-1-ol was higher than *trans* form in all of the samples analyzed from the four vintages considered; these results are in agreement with Boido et al., 2003 in wines from Tannat grape variety and by García-Carpintero et al., 2011a, in wines from Bobal grape variety and opposite result were found by García-Carpintero et al., 2011b in wines from Moravia Agria grape variety.

Terpenes and C_{13} norisoprenoids. Terpene compounds are characteristic of aromatic varieties such as Muscat, have a low olfactory threshold and are generally associated with floral and citric aromas (Etiévant, 1991; Guth, 1997). Linalool, β-citronellol and *trans*-geraniol were detected in wines from four vintages.

The concentration of linalool, β-citronellol and *trans*-geraniol was higher in the 2007 vintage. However, their contribution to Rojal wine seems negligible when their odor thresholds are considered (15 µg/l linalool, 100 µg/l β-citronellol and µg/l, 30 µg/l *trans*-geraniol Guth,1997). The first study on volatile composition of Rojal wines over four vintages from La Mancha region shows that the aroma of this cultivar is not terpene dependent.

	RI*	Aroma Compounds	Vintage			
			2007	2008	2009	2010
Fluka	1282	1-Hexanol	2187[c] (0.10)	1413[a] (0.12)	2013[b] (2.35)	2023[b] (1.56)
Sigma-Aldrich	1286	(E)-3-Hexen-1-ol	203[c] (4.50)	173[b] (0.26)	164[b] (2.14)	135[a] (1.05)
Sigma-Aldrich	1296	(Z)-3-Hexen-1-ol	208[b] (0.80)	185[b] (0.22)	198[b] (1.39)	149[a] (0.58)
Sigma-Aldrich	1300	(E)-2-Hexen-1-ol	15.7[a] (2.80)	9.01[a] (0.18)	11.9[a] (2.57)	15.5[a] (0.86)
		Total C$_6$ compounds	2614	1780	2387	2323
Fluka	1529	Linalool	13.9[b] (2.80)	11.8[b] (1.23)	12.2[b] (2.34)	9.11[a] (1.26)
Fluka	1755	β-Citronellol	14.5[b] (4.20)	8.93[a] (1.05)	12.3[b] (2.24)	13.9[b] (2.56)
Firmenich	1801	β-Damascenone	0.48[c] (2.80)	n.d	0.32[b] (0.87)	0.21[a] (0.85)
Fluka	1831	*Trans* geraniol	9.87[b] (2.00)	8.87[a] (1.36)	9.21[b] (1.23)	9.47[b] (0.59)
T.I.	2558	3-Hydroxy-β-damascone	29.5[c] (2.30)	7.52[a] (1.00)	15.6[b] (2.33)	19.3[b] (1.26)
T.I.	2582	3-Oxo-α-ionol	10.3[a] (1.23)	25.6[c] (1.08)	18.7[b] (3.14)	18.5[b] (0.24)
T.I.	2722	3-Hydroxy-7,8-dehydro-β-ionol	n.d	1.90[c] (2.05)	1.02[a] (1.87)	1.45[b] (0.94)
		Total Terpene + C$_{13}$ norisoprenoids	77.6	65.6	69.4	71.9
Sigma-Aldrich	1503	Benzaldehyde	2.54[a] (2.80)	2.81[c] (0.58)	2.64[b] (2.36)	2.88[c] (1.08)
T.I.	1505	3(2H)-2-methyldihydro-thiophenone	83.8[b] (5.30)	81.3[a] (3.21)	86.3[b] (1.87)	91.7[c] (1.98)
Sigma-Aldrich	1882	Guaiacol	41.7[a] (8.00)	62.5[b,c] (1.28)	58.3[b] (5.21)	66.9[c] (0.88)
Sigma-Aldrich	1895	Benzyl Alcohol	291[c] (1.40)	239[a] (0.59)	261[b] (4.26)	287[c] (2.19)
T.I.	1899	1,2-Benzothiazole	1.39[a] (5.10)	2.42[c] (1.21)	1.99[b] (2.87)	1.98[b] (1.69)
Sigma-Aldrich	1971	Phenol	3.04[a] (3.30)	11.5[c] (1.67)	5.64[b] (1.49)	9.58[c] (0.82)
Lancaster	2055	4-Ethylguaiacol	3.47[c] (2.70)	n.d.	0.68[a] (1.24)	1.97[b] (1.07)
Sigma-Aldrich	2193	Eugenol	5.01[a] (2.80)	6.18[b] (2.04)	5.21[a] (1.14)	6.51[b] (2.07)
Sigma-Aldrich	2208	4-Ethyl phenol	10.4[a] (1.90)	23.3[c] (0.74)	13.4[b] (1.23)	20.7[c] (1.92)
T.I.	2212	4-Hydroxy-2-methyl acethophenone	1.56[a] (0.85)	1.23[a] (1.59)	1.89[b] (2.14)	1.97[b] (1.48)
Sigma-Aldrich	2219	4-Vinylguaiacol	46.2[c] (2.10)	15.3[a] (0.66)	28.6[b] (3.29)	12.4[a] (0.88)
Sigma-Aldrich	2225	Syringol	156[b] (2.80)	107[a] (0.85)	172[b] (2.87)	194[b] (1.07)
Lancaster	2302	Isoeugenol	21.3[b] (1.20)	26.7[b] (1.09)	23.6[b] (1.82)	17.2[a] (1.45)

	RI*	Aroma Compounds	Vintage			
			2007	2008	2009	2010
Sigma-Aldrich	2378	Benzoic acid	178[b] (2.84)	183[b] (2.04)	167[b] (0.99)	153[a] (0.08)
T.I.	2501	Benzeneacetic acid	27.2[a] (1.10)	46.2[c] (3.01)	37.2[b] (0.87)	33.6[b] (1.01)
Panreac	2511	Vanillin	6.76[b] (0.50)	4.55[a] (9.18)	5.12[a] (2.47)	5.81[a] (2.67)
Sigma-Aldrich	2543	Methyl vanillate	15.9[a] (2.20)	18.3[b] (1.69)	17.8[b] (2.07)	19.3[b] (0.27)
Lancaster	2676	Ethyl vanillate	195[b] (8.10)	98.2[a] (0.73)	174[b] (1.08)	182[b] (1.45)
Sigma-Aldrich	2685	Acetovanillone	114[b] (7.30)	79.2[a] (0.07)	91.5[b] (1.65)	75.8[a] (1.91)
Sigma-Aldrich	2936	Zingerone	6.35[b] (2.69)	2.88[a] (0.98)	3.64[a] (2.48)	15.9[c] (0.09)
Sigma-Aldrich	2755	3,4-Dimethoxy phenol	6.87[a] (0.20)	7.56[a] (0.41)	9.64[a] (1.05)	12.0[b] (1.37)
Sigma-Aldrich	3045	Cinamic acid	60.5[c] (0.29)	60.8[c] (0.99)	51.3[b] (5.32)	44.6[a] (0.61)
T.I.	3030	Methyl vanillil eter	15.4[a] (4.10)	19.3[b] (3.09)	18.1[b] (0.78)	16.5[a] (2.39)
		Total Bencenic compounds	**1292**	**1099**	**1237**	**1273**

*Linear retention index on a DB Wax column

nd: not detected.

[a, b, c, d] According to the result of the Student–Newman–Keuls test, values to that no share a common superscript are significantly different ($p < 0.05$).

T.I. Tentatively identified

Table 1. Mean concentration of free volatile compounds (µg/l) and relative standard deviations (n=2) of Rojal wines.

C_{13}-norisoprenoids are volatile compounds that could come from the direct degradation of carotenoid molecules such as β-carotene, lutein, neoxanthin and violaxanthin (Marais et al., 1992), and have an important role on the varietal character of wines because they have a very low odor threshold (Guth, 1997). These compounds were present in low concentration in all vintages. β-damascenone is normally considered a positive contributor to wine aroma (Escudero et al., 2007) and its odor threshold (0.05 µg/l) (Guth, 1997) was exceeded in 2007, 2009 and 2010 vintages. Floral and exotic fruit notes are attributed to β-damascenone, and due to its low odour threshold it can have a great sensorial impact on wines (Guth, 1997). However, other authors attribute its importance to a potentiating role of the fruit aromas of other compounds, rather than β-damascenone acting alone (Pineau et al., 2007).

Benzene compounds. Benzene compounds are an important group in varietal aroma, abundant in wines, including aromatic alcohols, aldehydes, volatile phenols and shikimic acid derivates. The volatile phenols in wines can come from grapes, both as free and bound aroma, or be generated during the alcoholic fermentation by chemical reactions such as phenolic acid degradation, or in the case of vinylphenols due to brettanomyces contamination (Suarez et al., 2007). Volatile phenols are considered characteristic components of wine aroma, although their influence on the final product may be positive or

negative depending on their concentrations. In our case volatile phenols did not attain levels sufficient to prompt off-flavours. Guaiacol, 4-ethylguaiacol and 4-vinylguaiacol were identified in wines. These compounds are principally formed during the fermentation process. Guaiacol is associated with medicinal flavours, the olfactory threshold of this compound 10 µg/l (Guth, 1997) was exceeded in all studied wines. Olfactory threshold of volatile phenols 4-ethylguaiacol, (110 µg/l), 4-vinylguaiacol (10000 µg/l) (Swiegers et al., 2005) were not exceeded in any case. Other remarkable benzenic compounds were the shikimic acid derivates, which point out by the elevated sensory impact. These compounds are formed by the aromatic aminoacid metabolic routes in plants or by yeasts, also can be extracting from wood (Swiegers et al., 2005). Benzaldehyde, benzyl alcohol and eugenol were other benzenic compounds identified in wines. Benzaldehyde and benzyl alcohol concentrations were not exceeded their olfactory threshold (350 and 10000 µg/l, respectively) (Etiévant, 1991), although these compounds could add a synergic effect to wine aroma with fruity and floral notes. The identification of eugenol in wines is related to sweet spice aroma, especially with clove aroma in wines, this compound exceeded in all wines studied their olfactory threshold (5 µg/l) (Guth, 1997).

3.1.2 Volatile compounds formed principally during the alcoholic fermentation

Although compounds from the grapes themselves are responsible for the varietal character of wines, the compounds formed during alcohol fermentation via yeast metabolism may have a positive or negative influence on wine sensory properties (Ferreira et al., 1995). Table 2 shows concentrations of volatile compounds formed principally during the alcoholic fermentation of Rojal red wines over four vintages expressed as mg/l as mean of two replicates. The major fermentation compounds such alcohols, ethyl esters, acetates and fatty acids were detected at similar total levels in both wines, some minor differences were observed.

Aldehydes. Acetaldehyde is the majority aldehyde in the wine. It is formed mainly by the metabolism of yeasts, and is associated with fruity aromas and notes to nuts or dried fruits. The amount of acetaldehyde found in the wines is closely related to enzymatic manning of the strain of yeast used but also, this concentration can be changed with the conditions of fermentation, especially with the amount of SO_2 added to the medium (Herraiz et al., 1989). In this study, the conditions of fermentation and the amount of SO_2 added to the musts were the same in all case, so that the observed differences can be attributed to the different composition of the initial must used in the elaboration that may be attributed to climatic variations.

Alcohols: Alcohols were one of the largest group of free volatile compounds in La Mancha Rojal wines. The most abundant compounds were the higher alcohols, in accordance with the literature (Baumes et al., 1986). These compounds can be recognized by their strong and pungent smell and taste and they are related to herbaceous notes. The total concentration of higher alcohols in Rojal wines was below 300 mg/l. This allowed them to contribute positively to the aroma of the Rojal wines, giving it complexity (Mateo et al., 2001; Selli et al., 2004). Among the aliphatic alcohol 3-methyl-1-butanol showed the highest concentration in the four vintages.

Methanol is derived from the demethylation of skin pectins. Since we have mentioned previously, the conditions of fermentation were the same for all wines, so in this case the concentration differences can be attributed to the decreased permeability of the skin of Rojal grapes.

	RI*	Aroma Compounds	Vintage			
			2007	2008	2009	2010
Sigma-Aldrich	800	Acetaldehyde	7.93[b] (3.60)	3.08[a] (0.99)	6.21[b] (1.26)	10.6[c] (1.25)
		Total aldehydes	**7.93**	**3.08**	**6.21**	**10.6**
Sigma-Aldrich	879	Metanol	81.6[c] (2.10)	57.6[b] (3.27)	51.3[b] (3.28)	40.1[a] (1.05)
Sigma-Aldrich	1060	1-Propanol	14.7[a] (2.60)	20.4[b] (1.29)	17.2[a] (1.08)	15.3[a] (0.36)
Merck	1214	Isobutanol	41.1[c] (2.30)	31.5[a] (3.64)	35.2[b] (1.68)	30.9[a] (2.36)
Sigma-Aldrich	1221	2-Methyl-1-butanol	58.0[a] (5.30)	60.2[b] (2.09)	58.7[a] (0.87)	57.6[a] (0.14)
Sigma-Aldrich	1221	3-Methyl-1-butanol	181[a] (1.00)	199[d] (1.18)	191[c] (3.08)	185[b] (2.58)
Fluka	1328	4-Methyl-1-pentanol	0.09[a] (5.66)	0.03[a] (6.17)	0.05[a] (2.87)	0.08[a] (3.69)
T.I.	1337	2-Penten-1-ol (Z)**	4.58[c] (1.28)	4.91[c] (3.17)	3.29[b] (0.84)	2.68[a] (1.29)
Fluka	1341	3-Methyl-1-pentanol	0.10[a] (1.00)	0.12[a] (5.65)	0.11[a] (1.69)	0.11[a] (4.69)
Fluka	1472	1-Heptanol	0.04[a] (3.97)	0.02[a] (5.29)	0.02[a] (0.34)	0.03[a] (5.18)
Fluka	1545	2,3-Butanediol (levo)**	0.06[a] (5.01)	0.08[a] (4.67)	0.06[a] (1.91)	0.07[a] (5.36)
Fluka	1585	2,3-Butanediol (meso)	0.02[a] (4.58)	0.03[a] (7.08)	0.02[a] (2.11)	0.04[a] (3.47)
Sigma-Aldrich	1725	3-Methylthio-1-propanol	2.51[a] (0.58)	2.68[a] (1.18)	2.63[a] (0.17)	2.57[a] (2.64)
Fluka	1892	Phenylethylalcohol	25.1[a] (2.20)	30.7[b] (0.28)	28.4[b] (3.27)	34.5[b] (2.36)
		Total alcohols	**409**	**407**	**388**	**369**
Sigma-Aldrich	834	Ethyl acetate	36.9[b] (2.60)	35.6[b] (0.25)	28.9[a] (1.29)	24.5[a] (2.01)
Fluka	1080	Ethyl butanoate	0.06[a] (5.90)	0.07[a] (1.36)	0.06[a] (2.17)	0.08[a] (3.78)
Sigma-Aldrich	1145	Isoamyl acetate	0.03[a] (2.90)	0.05[a] (1.48)	0.04[a] (2.64)	0.05[a] (5.87)
Fluka	1185	Ethyl hexanoate	0.43[b] (1.40)	0.38[a] (2.34)	0.41[b] (2.11)	0.35[a] (2.45)
Sigma-Aldrich	1294	Hexyl acetate**	1.99[a] (7.80)	2.65[b] (1.92)	2.08[a] (2.74)	3.07[b] (0.70)
Sigma-Aldrich	1326	Ethyl lactate	15.9[a] (1.80)	16.7[a] (0.58)	17.6[a] (2.98)	22.7[b] (1.64)
T.I.	1321	2-hydroxy 3-methylethyl butanoate**	6.95[b] (2.34)	5.64[b] (1.26)	4.12[a] (1.54)	3.24[a] (2.18)
Sigma-Aldrich	1436	Ethyl octanoate	0.45[a] (11.2)	0.37[a] (6.17)	0.39[a] (1.67)	0.41[a] (2.81)
T.I.	1461	2-Hydroxy 2-methylpropyl butanoate**	2.96[a] (1.23)	5.48[b] (2.35)	3.86[a] (1.78)	4.25[b] (2.45)
T.I.	1499	3-Hydroxy-ethyl butanoate	0.46[a] (0.80)	0.49[a] (4.07)	0.47[a] (1.24)	0.51[a] (4.11)
T.I.	1522	Ethyl, dI-2-hydroxycaproate**	25.4[b] (2.64)	16.3[a] (1.02)	23.1[b] (2.42)	38.4[c] (2.71)

	RI*	Aroma Compounds	Vintage			
			2007	2008	2009	2010
Sigma-Aldrich	1605	Diethyl malonate**	0.14[a] (5.40)	0.18[a] (3.01)	2.23[b] (2.53)	1.81[b] (1.65)
Fluka	1655	Ethyl decanoate	0.08[a] (1.02)	0.07[a] (4.57)	0.07[a] (1.92)	0.07[a] (3.08)
Fluka	1702	Diethyl succinate	2.56[c] (2.64)	4.14[d] (2.36)	1.14[b] (1.05)	0.87[a] (2.11)
Fluka	1787	Methyl salicylate**	15.6[b] (3.24)	11.8[a] (0.54)	13.1[a] (1.25)	18.7[b] (2.31)
T.I.	1783	4-Hydroxy ethyl butanoate	1.71[a] (1.50)	1.67[a] (1.28)	1.62[a] (0.84)	1.57[a] (0.82)
Fluka	1936	2-phenylethyl acetate	0.03[a] (3.00)	0.09[a] (2.04)	0.05[a] (3.11)	0.13[a] (1.85)
Sigma-Aldrich	2070	Diethyl malate	1.13[b] (0.20)	0.22[a] (4.08)	0.84[b] (1.45)	1.07[b] (0.18)
T.I.	2331	Ethyl monosuccinate	3.30[a] (3.90)	3.59[a] (2.48)	3.52[a] (2.37)	3.91[a] (3.77)
		Total esters	**63.0**	**63.4**	**55.1**	**56.2**
Sigma-Aldrich	1426	Acetic acid	0.03[a] (5.20)	0.08[a] (4.25)	0.05[a] (6.14)	0.06[a] (1.12)
Sigma-Aldrich	1546	Propanoic acid**	1.45[a] (11.3)	1.81[b] (1.64)	1.63[a] (2.31)	1.89[b] (2.33)
Fluka	1583	Isobutanoic acid	1.34[a] (4.50)	1.49[b] (0.08)	1.43[b] (1.08)	1.52[b] (2.51)
Fluka	1600	Butanoic acid	1.40[c] (2.80)	1.25[b] (2.34)	1.21[b] (1.09)	0.92[a] (1.94)
T.I.	1642	3-Methyl butanoic acid	2.38[a] (1.00)	2.17[a] (1.18)	2.08[a] (2.31)	1.87[a] (0.74)
Fluka	1703	Pentanoic acid	0.01[a] (1.10)	n.d.	0.01[a] (2.47)	0.02[a] (1.23)
Fluka	1816	Hexanoic acid	2.62[b] (1.50)	1.85[a] (2.36)	2.32[b] (2.36)	1.58[a] (2.12)
Sigma-Aldrich	1917	Heptanoic acid**	6.59[b] (1.20)	2.69[a] (1.58)	6.24[b] (2.14)	11.4[c] (2.32)
T.I.	1929	(E)-2-Hexenoic acid**	12.6[b] (6.40)	8.81[a] (1.47)	11.4[b] (0.85)	13.6[b] (2.33)
Fluka	2024	Octanoic acid	2.48[a] (2.40)	2.31[a] (2.61)	2.24[a] (2.10)	1.98[a] (2.14)
Sigma-Aldrich	2289	Decanoic acid	0.48[a] (2.80)	0.53[a] (1.58)	0.59[a] (0.39)	0.62[a] (1.20)
Sigma-Aldrich	2439	Dodecanoic acid	0.03[a] (6.00)	0.04[a] (3.68)	0.03[a] (3.21)	0.03[a] (2.34)
		Total acids	**10.8**	**9.72**	**9.96**	**8.60**

n.d. not detected.

[a, b, c, d] According to the result of the Student–Newman–Keuls test, values to that no share a common superscript are significantly different (p<0.05).

** Units expressed as µg/l.

* Linear retention index on a DB Wax column

T.I. Tentatively identified.

Table 2. Mean concentrations (mg/l) and relative standard deviations (n=2) of volatile compounds formed during alcoholic fermentation of Rojal wine.

Esters: Table 2 shows the wine concentrations of the 19 esters identified. Most of them are ethyl esters of fatty acids produced during the alcoholic fermentation; broadly speaking, they played a positive role in the generation of the quality of the aroma of these wines, especially the fruity aromas (Etiévant, 1991). The wines from the four vintages contained between 55.1 and 63.4 mg/l of esters. High levels were observed for ethyl acetate, ethyl lactate, monoethyl succinate and diethyl succinate.

Acids: Fatty acids production is governed by the initial composition of the must and by fermentation conditions and have been described with fruity, cheese, fatty, and rancid notes (Rocha et al., 2004). The most abundant acids in Rojal wines were butyric acid, isobutyric acid, isovaleric acid, hexanoic acid, octanoic acid and decanoic acid. The contents of 6, 8, and 10-carbon atom fatty acids, were in agreement with those found by García-Carpintero et al., 2011a,b in wines made with others grape varieties and fermented in the same conditions.

3.2 Bound aroma compounds enzymatically released of the wines

The evaluation of glycosidic precursors in non-aromatic varieties has gain relevance during the last years (Sánchez-Palomo et al., 2007; Loscos et al., 2009; Pedroza et al., 2010; García-Carpintero, et al., 2011a, 2011b). The ability of the enzyme preparation used to release glycosidically-bound compounds from grapes, AR-2000, has been confirmed in numerous studies (Baek & Cadwallader, 1999; Sánchez-Palomo et al., 2006, 2007; García-Carpintero et al., 2011a, 2011b).

The results obtained by enzymatic hydrolysis are exactly the aglycones liberated by the aroma precursors (without further chemical transformations), while with acid hydrolysis other chemical transformations of the liberated compounds are possible. These transformations may be important since they are related with the evolution of the varietal aroma during wine storage (Rodriguez-Bencomo et al., 2011).

The volatile compounds released from the bound fraction by enzyme hydrolysis in Rojal wines are shown in Table 3. As can be observed, from the different chemical families considered in the analysis of the aglycones liberated by the aroma precursors, 60 compounds (three C_6-compounds, nine terpenes, five C_{13}-norisoprenoids, 22 benzenic compounds, four alcohols, five esters and 13 aliphatic acids) have been found in quantifiable amounts in studied wines.

In Rojal wines benzene and C_{13}-norisoprenoids compounds were the most abundant bound compounds, followed by terpene compounds. The C_6 compounds concentrations in all the wines studied were significant lower than the observed on free volatile aroma (Table 1). These results confirm the limited importance of C_6 compounds on the bound fraction, as occurs with other variety grapes (Cabaroglu et al., 2003; Sánchez-Palomo et al., 2006, 2007; García-Carpintero et al, 2011a, 2011b). In all wines studied the major component of this group of compounds was 1-hexanol.

It can be seen that the total concentration of terpenes + C_{13} norisoprenoid compounds in the bound forms was always higher than that of the free forms in all studied wines, as would correspond to a quality variety (Diéguez et al., 2003). All of the terpene compounds present in wines were found in low concentrations as expected for a neutral grape variety and some of these compounds α-terpineol, trans-linalool oxide (furanoid), cis-linalool oxide (furanoid), cis-linalool oxide (pyranoid), nerol and geranic acid were not present in the free fraction of wines. The bound fraction of others, such as linalool, geraniol and β-citronellol, was more abundant than the free fraction. Geranic acid, geraniol and linalool were the major components of this group in bound fraction of La Mancha Rojal red wines.

Source	RI*	Aroma Compounds	Vintage			
			2007	2008	2009	2010
Fluka	1282	1-Hexanol	95.5[c] (2.30)	85.6[b] (0.07)	80.1[b] (0.32)	42.8[a] (0.57)
Sigma-Aldrich	1296	(Z)-3-Hexen-1-ol	10.5[a] (7.80)	10.5[a] (2.14)	26.3[c] (1.28)	15.1[b] (2.04)
Sigma-Aldrich	1300	(E)-2-Hexen-1-ol	45.0[c] (10.4)	36.8[b] (1.08)	32.5[a] (1.34)	38.1[b] (1.27)
		Total C$_6$ compounds	151	133	139	96.0
T.I.	1455	Cis linalool oxyde furan	6.03[b] (1.02)	5.45[b] (0.25)	2.38[a] (1.32)	4.17[b] (2.64)
T.I.	1483	Trans linalool oxyde furan	3.03[a] (11.7)	2.89[a] (1.23)	2.31[a] (1.42)	3.01[a] (0.84)
Fluka	1529	Linalool	17.3[b] (3.70)	15.3[b] (0.15)	16.5[b] (1.01)	11.6[a] (2.15)
Fluka	1607	a –Terpineol	5.14[b] (0.90)	4.65[b] (1.64)	3.68[a] (1.37)	4.89[b] (1.36)
T.I.	1716	Cis linalool oxyde pyran	5.28[b] (3.90)	4.52[b] (1.09)	5.08[b] (1.34)	3.54[a] (2.03)
Fluka	1755	β-citronellol	4.28[b] (9.30)	4.14[b] (1.28)	3.21[a] (2.31)	3.96[a] (1.36)
Fluka	1819	Nerol	7.77[c] (7.50)	6.25[b] (2.01)	5.14[a] (1.09)	7.70[c] (1.54)
Fluka	1831	Geraniol	30.3[b] (9.90)	29.5[b] (2.48)	20.6[a] (2.30)	28.1[b] (1.77)
Sigma-Aldrich	2289	Geranic acid	60.1[a] (1.70)	68.4[b] (0.12)	64.2[b] (1.09)	67.2[b] (1.08)
		Total Terpene compounds	139	141	123	134
Sigma-Aldrich	1703	4-Oxo-isophorone	3.27[b] (1.00)	3.19[b] (1.58)	2.21[a] (1.31)	2.12[a] (1.85)
T.I.	2558	3-Hydroxy-β-damascone	401[a] (8.60)	427[b] (0.64)	441[b] (2.31)	395[a] (2.05)
T.I.	2582	3-Oxo-α-Ionol	134[b] (4.10)	128[b] (1.23)	130[b] (1.98)	106[a] (0.31)
T.I.	2722	3-Hydroxy-7,8-dehydro-β-ionol	202[b] (2.30)	185[b] (1.74)	153[a] (2.08)	191[b] (1.08)
Sigma-Aldrich	1873	α-Ionone	2.31[a] (11.7)	2.85[b] (1.99)	2.14[a] (3.01)	3.20[b] (2.30)
		Total C$_{13}$ norisoprenoids compounds	743	746	728	697
Sigma-Aldrich	1503	Benzaldehyde	3.93[c] (1.60)	3.58[c] (0.25)	2.54[b] (2.36)	1.84[a] (1.08)
Fluka	1667	Acetophenone	3.21[a] (1.21)	3.85[a] (1.35)	4.84[b] (1.34)	4.67[b] (2.18)
T.I.	1750	N-ethyl benzeamine	3.87[a] (0.25)	6.51[c] (1.47)	4.65[b] (0.98)	8.94[d] (0.59)
Sigma-Aldrich	1882	Guaiacol	62.1[c] (4.50)	58.4[b] (1.05)	50.7[a] (0.48)	60.3[c] (0.47)
Sigma-Aldrich	1895	Benzyl Alcohol	712[b] (0.40)	608[a] (2.36)	897[d] (0.96)	835[c] (0.85)
Fluka	1892	Phenylethylalcohol	567[a] (0.70)	905[d] (1.01)	706[c] (1.64)	684[b] (1.28)
T.I.	1899	1,2-Benzothiazole	6.84[a] (0.56)	7.12[a] (0.66)	10.2[b] (0.33)	9.58[b] (0.99)
Sigma-Aldrich	1971	Phenol	18.5[b] (6.90)	15.9[a] (0.65)	26.3[c] (0.65)	24.6[c] (1.28)
T.I.	2038	Benzenepropanol	5.32[c] (1.60)	6.45[c] (1.25)	2.81[a] (0.19)	3.64[b] (0.15)
Sigma-Aldrich	2193	Eugenol	23.4[a] (2.60)	20.7[a] (2.65)	29.4[b] (0.70)	32.6[b] (1.88)
Sigma-Aldrich	2219	4-Vinylguaiacol	89.1[c] (3.60)	73.6[b] (3.21)	80.6[c] (3.65)	67.8[a] (1.01)

Source	RI*	Aroma Compounds	Vintage			
			2007	2008	2009	2010
Sigma-Aldrich	2225	Syringol	6.35[b] (3.60)	4.15[a] (0.41)	3.68[a] (1.85)	5.94[b] (2.31)
Lancaster	2302	Isoeugenol	13.5[b] (11.7)	10.6[a] (0.99)	13.2[b] (1.41)	11.8[a] (1.07)
Sigma-Aldrich	2378	Benzoic acid	96.2[c] (3.20)	95.3[c] (1.24)	78.4[a] (1.24)	84.1[b] (0.22)
Sigma-Aldrich	2424	(*E*)-4-Allylsyringol	16.4[a] (1.01)	20.3[b] (2.65)	15.4[a] (0.11)	19.6[b] (0.17)
T.I.	2501	Benzeneacetic acid	10.8[a] (2.30)	15.4[b] (1.48)	11.3[a] (0.74)	17.6[b] (1.09)
Panreac	2511	Vanillin	61.2[b] (2.20)	59.6[b] (2.31)	57.9[a] (1.32)	54.8[a] (10.80)
Sigma-Aldrich	2543	Methyl vanillate	205[c] (3.30)	199[c] (1.24)	153[a] (0.90)	178[b] (2.18)
Lancaster	2676	Ethyl vanillate	32.3[a] (1.01)	44.7[c] (0.54)	28.9[a] (1.25)	39.4[b] (5.21)
Sigma-Aldrich	2685	Acetovainillone	205[b] (1.70)	146[a] (1.44)	198[b] (0.81)	153[a] (1.65)
Sigma-Aldrich	2936	Zingerone	68.6[b] (3.30)	62.4[a] (0.86)	65.2[b] (2.10)	61.6[a] (4.14)
Sigma-Aldrich	2755	3,4-Dimethoxy phenol	79.5[c] (4.70)	72.3[b] (2.64)	67.3[a] (1.89)	71.3[b] (2.01)
		Total Bencenic compounds	2290	2439	2507	2430
Sigma-Aldrich	1221	3-Methyl-1-butanol	55.3[b] (2.36)	36.1[a] (1.05)	64.3[c] (2.31)	75.8[d] (1.62)
Sigma-Aldrich	1273	3-Methyl-2-buten 1-ol	3.62[a] (2.15)	6.57[b] (1.65)	5.21[b] (1.67)	7.98[b] (1.25)
		Total Alcohols	58.9	42.7	69.5	83.8
T.I.	1355	Octanoic acid methyl ester	3.25[a] (0.96)	2.65[a] (2.15)	1.36[a] (2.00)	4.28[b] (2.74)
Fluka	1655	Decanoic acid ethyl ester	1.23[a] (0.54)	0.67[a] (3.15)	0.95[a] (5.78)	1.14[a] (1.25)
T.I.	1704	Ethyl methyl succinate	6.68[c] (1.24)	11.3[d] (0.09)	3.14[b] (1.45)	1.05[a] (1.06)
Fluka	1787	Methyl Salicylate	2.67[a] (2.85)	2.31[a] (0.25)	2.55[a] (1.91)	2.53[a] (0.74)
T.I.	1827	Butanoic acid, 4-hydroxy-methyl ester	7.68[b] (1.26)	5.32[a] (1.97)	11.2[c] (1.62)	9.64[b] (1.63)
		Total Esters	21.5	22.3	19.2	18.6
Sigma-Aldrich	1426	Acetic acid	21.1[b] (6.60)	18.5[b] (1.54)	20.9[b] (2.07)	15.6[a] (1.65)
Sigma-Aldrich	1546	Propanoic acid	7.50[b] (4.20)	3.98[a] (1.65)	6.14[b] (1.25)	3.37[a] (1.26)
Fluka	1600	Butanoic acid	28.8[b] (6.70)	21.4[a] (1.06)	25.9[b] (0.32)	24.0[b] (2.31)
Sigma-Aldrich	1642	Isovaleric acid	30.5[b] (2.40)	28.4[b] (2.65)	24.6[a] (0.65)	30.2[b] (1.34)
Fluka	1703	Pentanoic acid	4.27[b] (9.60)	3.39[b] (1.99)	2.36[a] (1.99)	4.18[b] (1.27)
Fluka	1816	Hexanoic acid	154[c] (2.65)	139[b] (1.65)	152[c] (2.48)	112[a] (2.14)
T.I.	1857	2-Ethyl hexanoic acid	n.d.	n.d.	1.25[a] (1.28)	1.37[a] (1.48)
Sigma-Aldrich	1917	Heptanoic acid	11.5[b] (4.20)	9.64[b] (3.15)	7.51[a] (1.09)	11.4[b] (1.20)

Source	RI*	Aroma Compounds	Vintage			
			2007	2008	2009	2010
Fluka	2024	Octanoic acid	381[d] (3.80)	241[c] (1.65)	134[a] (1.39)	197[b] (0.64)
Sigma-Aldrich	2108	Nonanoic acid	65.9[c] (0.80)	61.3[c] (1.74)	48.7[a] (2.62)	56.1[b] (1.06)
Sigma-Aldrich	2289	Decanoic acid	140[d] (7.30)	121[c] (1.81)	112[b] (1.85)	69.4[a] (1.07)
Sigma-Aldrich	2439	Dodecanoic acid	43.1[b] (11.7)	45.0[b] (1.15)	33.4[a] (1.99)	49.0[b] (4.65)
Sigma-Aldrich	2653	Tetradecanoic acid	6.87[a] (2.20)	21.6[c] (0.97)	20.9[c] (2.36)	10.3[b] (2.61)
		Total Acids	895	714	590	584

*Linear retention index on a DB Wax column; nd: not detected.
[a, b, c, d] According to the result of the Student–Newman–Keuls test, values to that no share a common superscript are significantly different ($p < 0.05$)
T.I. Tentatively identified

Table 3. Mean concentration (µg/l) and relative standard deviations (n=2) of bound volatile compounds released by enzymatic hydrolysis of Rojal wines.

Norisoprenoids detected in negligible amounts or not found in the free fraction of studied wines, were relatively abundant in the bound fraction. The C_{13}-norisoprenoid pattern was composed by 3-hydroxy-β-damascone, 3-oxo-α-ionol, 3-hydroxy-7,8-dihydro-β-ionol and, in smaller concentrations, α-ionone and 4-oxo-isophorone. By contrast, β-damascenone – detected in the free fraction in wines– was not detected in bound form as this compound is formed principally from the precursors 3-hydroxy-β-damascone and 3-hydroxy-7,8-dehydro-β-ionol; the highest concentration of both compounds found in fraction of aroma of Rojal wines could be related to that in the free fraction Rojal wines presented lower concentration of β-damascenone.

The bound fraction of benzenic compounds was major quantitative than the free fraction in Rojal wines. *(E)*-4-Allylsyringol was not present in the free fraction of wines but was detected in the bound fraction. Benzyl alcohol and 2-phenylethanol were the compounds in higher concentration in this fraction in Rojal wines. Guaiacol, phenol, eugenol, isoeugenol, vanillin, methyl vanillate and ethylvanillate present higher concentrations in bound fraction of aroma of Rojal wines. García-Carpintero et al., 2011b founded higher concentrations in the total benzenic compound of bound fraction in Moravia Agria wines than in our studied wines.

The alcohols qualitative composition in bound aroma was lower than in free aroma, attributable to these compounds are principally formed by the yeast metabolism. The concentration of aliphatic acids in bound aroma were noteworthy lower than in free aroma, due to the principal formation pathway of this compounds is by the yeast metabolism.

During winemaking, some of these bound aroma compounds give rise to odorant compounds that play a role in certain aroma characteristics of wine; similar results were observed by Hernandez-Orte et al., 2009 studying the ability of glycosidase activity of

several lactic acid bacteria (LAB) to change the volatile fraction of wine by releasing aroma compounds. These authors observed that the studied LAB strains were able to release terpene compounds, C$_{13}$-norisoprenoid compounds, volatiles phenols and vanillin derivates. According to the result, the bound fraction of Rojal wines can be considered a potential aroma source which reveals the enrichment in varietal compounds of the must as a result of the transfer of these compounds from the skin (Cabaroglu et al., 2003; Sánchez-Palomo et al., 2006, 2007; García-Carpintero et al., 2011a, 2011b).

3.3 Odour activity values

The aroma of Rojal red wines from La Mancha region has been studied by sensory analysis. Relevant aroma sensory descriptors given by the expert panel are summarized in Table 4.

Aroma descriptors
Red fruit
Fresh
Clove
Pepper
Leather/Tobacco
Sweet
Fresh fruit

Table 4. Sensory Aroma Descriptors given by the expert panel to the La Mancha Rojal wines.

The table 5 shows the odour descriptors and odour threshold of the aroma compounds in La Mancha Rojal wines obtained by the bibliographic references (Kotseridis & Baumes, 2000; Lopez et al., 2003). With over 50 aroma components of wide-ranging intensities and no single character impact compounds, it is difficult to predict the overall aroma impact of these wines from the sheer size of the data. To estimate overall wine aroma, the odour descriptors were grouped in different aromatic series and every compound is assigned to one or several aromatic series based on similar odour descriptor used.

Compounds	Sensory description	Odorant series*	Odour Threshold (µg/L)
Acetaldehyde	pungent, ripe apple	1,6	500[a]
Ethyl acetate	fruity, solvent	1,6	7500[a]
Ethyl butyrate	fruity	1	20[a]
Isoamyl acetate	banana	1	30[c]
Methanol	chemical , medicinal	6	668000[b]
1-propanol	ripe fruit, alcohol	1,6	830[b]
Isobutanol	oily, bitter, green	3,6	40000[b]
3-Methyl-1-butanol	burnt, alcohol	4,6	30000[a]
1-Butanol	medicinal, phenolic	6	150000[b]
Ethyl caproate	green apple	1	14[b]
1-Pentanol	almond, syntetic, balsamic	6	64000[b]
Hexyl acetate	green, floral	2,3	1500[c]
Ethyl pyruvate	vegetable, caramel	4,7	100000[b]
Ethyl lactate	acid, medicine	6	154636[c]

Compounds	Sensory description	Odorant series*	Odour Threshold (µg/L)
1-Hexanol	flower, green, cut grass	2,3	8000[a]
4-Methyl-1-pentanol	almond, toasted	4	50000[c]
3-Methyl-1-pentanol	vinous, herbaceous, cacao	1,3,7	50000[c]
(Z)-3-Hexen-1-ol	green, cut grass	3	400[a]
Ethyl caprilate	sweet, fruity	1,4	5[b]
Acetic acid	sour, pungent, vinegar	6	200000[a]
1-Heptanol	oily	6	2500[b]
3-Hydroxy, ethyl butyrate	caramel, toasted	4	20000[b]
Benzaldehyde	sweet, fruity	1,4	350[c]
Propanoic acid	pungent, racid, soy	6	8100[c]
2, 3-Butanediol (levo)	fruity	1	150000[b]
Linalool	floral	2	15[a]
Isobutyric acid	rancid, butter, cheese	6	2300b
2,3-Butanediol (meso)	fruity	1	150000[b]
γ-Butyrolactone	sweet, toast, caramel	4	35[c]
Butyric acid	rancid, cheese, sweat	6	173[b]
Ethyl caprate	sweet/fruity	1,4	200[c]
Isovaleric acid	sweet, acid, rancid	4,6	33[c]
Diethyl succinate	vinous	7	200000[b]
3-(Methylthio)-1-propanol	cooked vegetable	7	1000[a]
2-Phenylethyl acetate	floral	2	250[a]
β-damascenone	sweet, fruity	1,4	0,05[a]
Hexanoic acid	sweat	6	420[b]
Geraniol	roses, geranium	2	30[a]
2-Methoxyphenol,	medicine, sweet, smoke	4,6	10[c]
Benzyl Alcohol	sweet, fruity	1,4	200000[b]
2-Phenyethyl alcohol	floral, roses	2	10000[a]
Diethyl malate	over-ripe, peach, cut grass	1	760000[b]
Octanoic acid	sweat, cheese	6	500[c]
Eugenol	spices, clove, honey	4,5	6[c]
4-Vinylguaiacol	spices/curry	5	40[a]
Decanoic acid	rancid fat	6	1000[b]
Ethyl cinnamate	fruity, honey, cinnamon	1,4,5	1,1[a]
Isoeugenol	clove	5	6[b]
Ethyl monosuccinate	caramel, coffee	4	1000000[c]
Benzoic acid	chemical	6	1000[b]
Vainillin	vanillin	5	60[b]
Methyl vanillate	honey, vanillin	4,5	3000[b]
Ethyl vanillate	sweet, honey, vanillin	4,5	990[b]
Acetovanillone	sweet spices	5	1000[b]

*1 = fruity; 2 = floral; 3 = green, fresh; 4 = sweet; 5 = spicy; 6 = fatty; 7 = others.
[a] Guth 1997; [b] Etiévant, 1991 ; [c] Ferreira et al., 2000

Table 5. Odour descriptors, odorant series and odour threshold (µg/L) of the aroma compounds in monovarietal and co-winemaking wines.

Table 6 lists the OAV for all vintages studied and average OAVs values for the 31 aroma compounds with OAV>0.1 studied in Rojal wines during four consecutive years the odorant series of these compounds. The method based in the OAV has been used in the latter years in studies on wine aroma, such as in the discrimination of wines obtained from different grapes varieties (Guth, 1997), in works on accelerated ageing wines (Muñoz et al., 2007), in works on wines subjected to biological ageing (Moyano et al., 2002; Zea et al., 2007) and in studies of characterization of impact compounds of monovarietal wines (Sánchez-Palomo et al., 2010; García-Carpintero et al., 2011a, 2011b).

Compounds	Vintage			
	2007	2008	2009	2010
Ethyl caprilate	90.0	74.0	78.0	82.0
Isovaleric acid	72.1	65.8	63.0	56.7
Ethyl caproate	30.7	27.1	29.3	25.0
1-propanol	17.7	24.6	20.7	18.4
Acetaldehyde	35.8	6.2	12.4	21.2
Butyric acid	8.09	7.23	6.99	5.32
3-Methyl-1-butanol	6.03	6.63	6.37	6.17
Guaiacol	4.17	6.25	5.83	6.69
Hexanoic acid	6.24	4.40	5.52	3.76
beta-damascenone	9.60	0.00	6.40	4.20
Octanoic acid	4.96	4.62	4.48	3.96
Ethyl acetate	4.92	4.75	3.85	3.27
Isoeugenol	3.55	4.45	3.93	2.87
Ethyl butyrate	3.00	3.50	3.00	4.00
2-phenyethyl alcohol	2.51	3.07	2.84	3.45
3-Methylthio-1-propanol	2.51	2.68	2.63	2.57
Isoamyl acetate	1.00	1.67	1.33	1.67
Eugenol	0.84	1.03	0.87	1.09
Isobutanol	1.03	0.79	0.88	0.77
Linalool	0.93	0.79	0.81	0.61
4-vinylguaiacol	1.16	0.38	0.72	0.31
Isobutyric acid	0.58	0.65	0.62	0.66
Decanoic acid	0.48	0.53	0.59	0.62
(Z)-3-Hexen-1-ol	0.52	0.46	0.50	0.37
Ethyl caprate	0.40	0.35	0.35	0.35
Geraniol	0.33	0.30	0.31	0.32
1-Hexanol	0.27	0.18	0.25	0.25
2-Phenylethyl acetate	0.12	0.36	0.20	0.52
Propanoic acid	0.18	0.22	0.20	0.23
Benzoic acid	0.18	0.18	0.17	0.15
Ethyl vanillate	0.20	0.10	0.18	0.18
Ethyl lactate	0.10	0.11	0.11	0.15
Methanol	0.12	0.09	0.08	0.06
Vainillin	0.11	0.08	0.09	0.10
Acetovanillone	0.11	0.08	0.09	0.08

Table 6. Odor activity values of free aroma compounds in Rojal wines.

The aromatic series used in this work group volatile compounds with similar odour descriptors: fruity, floral, green/fresh, sweet, spice, fatty and other odours taking into account their use in previous papers (Sánchez-Palomo et al., 2010; Gómez García-Carpintero el al., 2011a, 2011b). Because of the high complexity of olfactive perceptions, some aroma compounds were included in two or more odorant series according to the finding of some authors (Zea et al., 2007; Charles et al., 2000).

The total intensities for every aromatic series were calculated as sum of the OAV of each one of the compounds assigned to this series and the results were graphed in Figure 2. This procedure makes it possible to relate quantitative information obtained by chemical analysis, to sensory perception, providing a single aroma profile based on an objective. It has recently been used some authors (Peinado et al., 2004, 2006; López de Lerma & Peinado 2011; García-Carpintero el al., 2011a, 2011b).

Intensity patterns in the category suggest that the major aroma characteristic of these wines would consist of fruity, sweet and fatty. Fruity was one of the aromatic series with major intensity (Figure 2). This series is formed principally by 7 esters, 1 alcohol and 1 C_{13}-norisoprenoid compound (beta-damascenone), identified and quantified by GC-MS. According to the results showed in Figure 2 can be observed that the aromatic series 4 (Sweet) showed the greatest intensity.
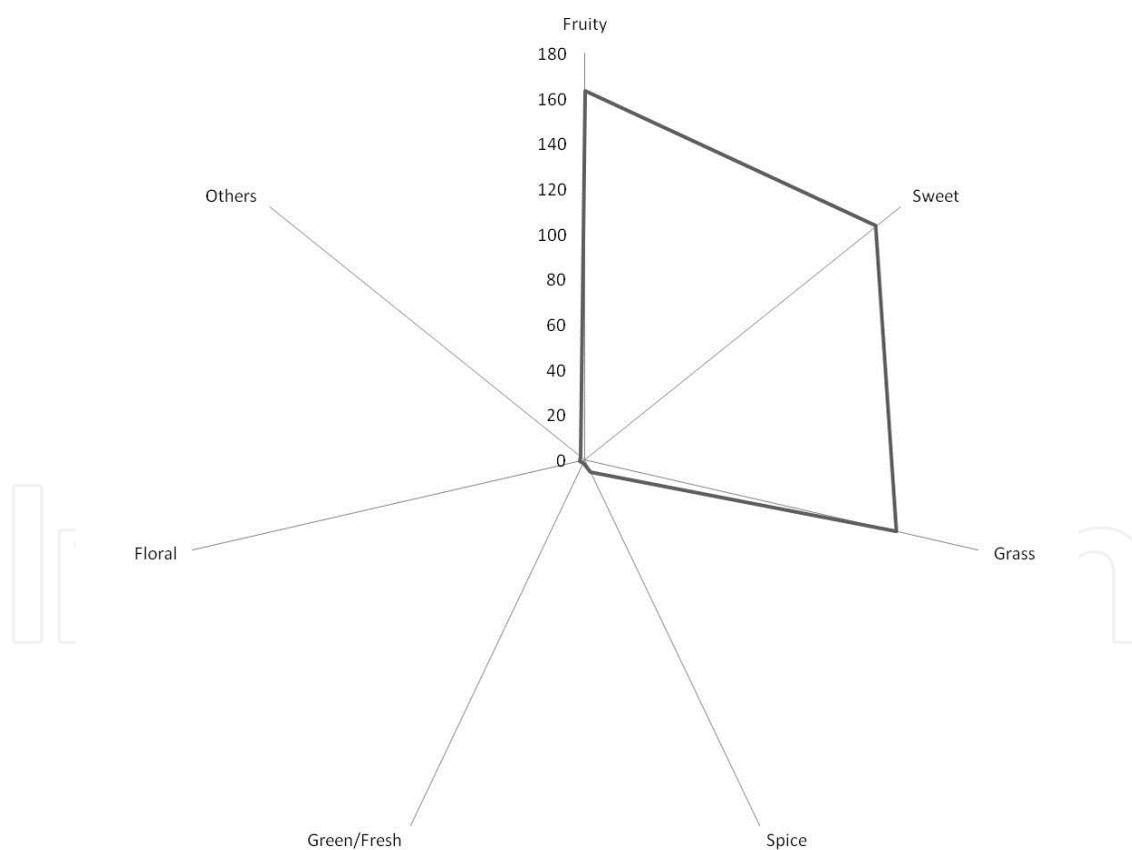
Fig. 2. Aromatic series in La Mancha Rojal wines (ΣOAV_{medium} over four vintages).

The aromatic series 6 (pungent, chemical, fatty, dry) was also major aroma categories in the current study. These attributes were not detected in the sensory flavour profile studies of wines. The aromatic series 3 (green, fresh) was one of the minor aroma categories,

nevertheless this attribute was ones of the most characteristics in the sensory profile of Rojal wines. This can be due to that the values of total intensity in the different aromatic series were obtained as sum of the individual OAVs of each one of the components without bearing in mind the rest of present compounds in the matrix of wine. Nevertheless when combined, synergy, suppression and matrix effects may alter the intensity of the descriptors, masking the descriptors of some aromatic series (series 4 and 6) and increasing the intensity of others odour descriptors (series 2 and 3). These results are in agreement with the results obtained in red wines made from Merlot and Cabernet Sauvignon grape varieties (Gürbüz et al., 2006), in wines made from Moravia Agria grape variety (García-Carpintero et al., 2011b) and in wines made from Bobal grape variety (García-Carpintero et al., 2011a).

4. Conclusion

This work provide a better knowledge of the aroma composition of Rojal wines elaborated with grapes cultivated in La Mancha region, also this study presents results from the first experiment performed on the free and bound aroma compounds from this minority grape variety from Castilla La Mancha region. Rojal wines present a complex chemical profile with a high richness in their aromatic composition. The free aroma of La Mancha Rojal wines is characterized by large amounts of C_6 and benzene compounds. The most abundant glycosilated fraction was the benzene compounds followed by C_{13}-norisoprenoids compounds. By other hand, the sensory aroma profile of Rojal wines was characterized by red fruit, fresh, clove, leather, tobacco, sweet and fresh fruit aroma descriptors. This study showed that this grape variety present a great aroma potential providing a viable alternative to traditional grape varieties cultivated in La Mancha region, increasing the offer to the consumer, which favors the differentiation of La Mancha wines on the national and international market.

5. Acknowledgment

6. References

Baek, H. H. & Cadwallader, K. R. (1999). Contribution of free and bound volatile compounds to the aroma of muscadine grape juice. *Journal of food Science*, Vol.64, No. 3, (July 2006), pp.441-444, ISSN: 1750-3841

Baumes, R.; Cordonnier, R.; Nitz, S. & Drawert, F. (1986). Identification and determination of volatile constituents in wines from different vine cultivars. *Journal of the Science Food and Agriculture*, Vol. 37, No.1, (September 2006), pp. 927–943, ISSN: 1097-0010

Boido, E.; Lloret, A.; Medina, K.; Faria, L.; Carrau F.; Versini, G. & Dellacasa E. (2003). Aroma Composition of Vitis vinifera Cv. Tannat: the typical red wine from Uruguay. *Journal of Agricultural and Food Chemistry,* Vol.51, No. 18 , (August 2003), pp.5408-5413, ISSN: 0021-8561

Cabaroglu, T.; Selli, S.; Canbas, A.; Lepoutre, J. P. & Günata, Z. (2003). Wine flavor enhancement through the use of exogenous fungal glycosidades. *Enzyme and Microbiology Technology*, Vol.33, No.5, (October 2003), pp.581-587, ISSN: 0141-0229

Campo, E.; Cacho, J. & Ferreira V. (2007). Solid phase extraction, multidimensional gas chromatography mass spectrometry determination of four novel aroma powerful ethyl esters: Assessment of their occurrence and importance in wine and other alcoholic beverage. *Journal of Chromatography A* Vol. 1140, No.1-2 (January 2007), pp. 180-188, ISSN: 0021-9673

Carballeira, L.; Cortes, S.; Gil, M.L. & Fernández, E. (2001). Determination of aromatic compounds, during ripening, in two white grape varieties, by SPE-GC. Chromatographia, Vol.53, Suppl.1, (January 2001), pp S350-S355, ISSN: 0009-5893

Charles, M.; Martin, B.; Ginies, C.; Etievant, P.; Coste, G. & Guichard, E. (2000). Potent aroma compounds of two red wine vinegars. *Journal of Agricultural and Food Chemistry,* Vol.48, No.1, (December 1999), pp.70–77, ISSN: 0021-8561

Diéguez, S.C.; Lois, L.C.; Gómez, E.F. & de la Peña, M.L.G. (2003). Aromatic composition ofthe Vitis vinifera grape Albariño. *Lebensmittel-Wissenschaft und-Technologie*, Vol.36, No.6, (September 2003), pp.585–590, ISSN: 0023-6438

Dominguez, C.; Guillen, D.A. & Barroso, C.G. (2002). Determination of volatile phenols in fino sherry wines. *Analytical Chimica Acta*, Vol.458, No.1, (April 2002), pp.95-102, ISSN: 0003-2670

Escudero A.; Campo E.; Farina L., Cacho J. & Ferreira V. (2007). Analytical Characterization of the Aroma of Five Premium Red Wines. Insights into the Role of Odor Families and the Concept of Fruitiness of Wines. *Journal of Agricultural and Food Chemistry*, Vol. 55, No. 11, (May 2007), pp.4501-4510, ISSN: 0021-8561

Etiévant, P.X. (1991). Wine. In: *Volatile compounds of food and beverages,* Maarse H (ed) Marcel Dekker, 483–546, ISBN 0-8247-8390-5, New York

Ferreira, V.; Fernández, P.; Peña, C.; Escudero, A. & Cacho, J. F. (1995). Investigation on the role played by fermentation esters in the aroma of young Spanish wines by multivariate analysis. *Journal of the Science of Food and Agriculture.* Vol.67, No.3, (September 2006), pp.381-392, ISSN: 1097-0010

Ferreira, V.; López, R. & Cacho, J. (2000). Quantitative determination of the odorants of young red wines from different grape varieties. *Journal of the Science of Food and Agriculture,* Vol.80, No.11, (July 2000), pp.1659–1667, ISSN: 1097-0010

Ferreira, V.; Lopez, R.; Escudero, A. & Cacho, J.F. (1998). Quantitative determination of trace and ultratrace flavour active compounds in red wines through gas chromatographic–ion trap mass spectrometric analysis of microextract. *Journal of Chromatography A,* Vol. 806, No.2, (May 1998), pp.349-354, ISSN: 0021-9673

Francis, I.L. & Newton, J.L. (2005). Determining wine aroma from compositional data. *Australian Journal of Grape and Wine Research.* Vol.11, No. 2, (Jun 2008), pp. 114–126. ISSN: 1755-0238

García-Carpintero, E.G.; Sánchez-Palomo, E. & González-Viñas, M.A. (2011a). Aroma characterization of red wines from cv. Bobal grape variety grown in La Mancha region. *Food Research International*, Vol. 44, No. 1, (January 2011), pp.61-70, ISSN: 0963-9969

García-Carpintero, E.G.; Sánchez-Palomo, E.; Gómez Gallego, M.A. & González-Viñas, M.A. (2011b). Volatile and sensory characterization of red wines from cv. Moravia Agria minority grape variety cultivated in La Mancha region over five consecutive vintages. *Food Research International*, 44, No. 5, (June 2011), pp. 1549-1560, ISSN: 0963-9969

Günata, Y. Z.; Bayonove, C.; Baumes, R. & Cordonnier, R. (1985). The aroma of grapes. I. Extraction and determination of free and glycosidically bound fraction of some

grape aroma components. *Journal of Chromatography*, Vol.331, (November 2001), pp. 83–90, ISSN: 0021-9673

Gürbüz, O.; Rouseff J.M. & Rouseff, R.L. (2006). Comparison of aroma volatiles in commercial Merlot and Cabernet Sauvignon wines using gas chromatography-olfactometry and gas chromatography-mass spectrometry. *Journal of Agricultural and Food Chemistry*, Vol.54, No.11, (May 2006), pp.3990-3996, ISSN: 0021-8561

Guth, H. (1997). Identification of character impact odorants of different white wine varieties. *Journal of Agricultural and Food Chemistry.* Vol.45, No.8, (August 1997), pp.3027-3032, ISSN: 0021-8561

Hatanaka, A. (1993). The biogeneration of green odour by green leaves. *Phytochemistry.* Vol.34, No.5, (November 1993), pp.1201–1218, ISSN: 0031-9422

Hernandez-Orte, P.; Cersosimo, M.; Loscos, N.; Cacho, J.; Garcia-Moruno, E. & Ferreira, V. (2009). Aroma development from non-floral grape precursors by wine lactic acid bacteria. *Food Research International,* Vol.42, No. 7, (August 2009), pp.773–781, ISSN: 0963-9969

Herraiz, T.; Martin-Alvarez, P.J.; Reglero, G.; Herraiz, M. & Cabezudo, M.D. (1989). Differences between wines fermented with and without sulphur dioxide using various selected yeast. *Journal of the Science of Food and Agriculture*, Vol.49, No.2, (September 2006), pp.249-258, ISSN: 1097-0010

Ibarz, M.J.; Ferreira, V.; Hernandez-Orte, P.; Loscos, N. & Cacho, J. (2006). Optimization and evaluation of a procedure for the gas chromatographic–mass spectrometric analysis of the aromas generated by fast acid hydrolysis of flavor precursors extracted from grapes. *Journal of Chromatography A.* Vol. 1116, No.1-2, (May 2006), pp.217-229, ISSN: 0021-9673

Insa, S.; Antico, E. & Ferreira, V. (2005). Highly selective solid-phase extraction and large volume injection for the robust gas chromatography–mass spectrometric analysis of TCA and TBA in wines. *Journal of Chromatography A* Vol. 1089, No.1-2, (September 2005), pp.235-242, ISSN: 0021-9673

Kotseridis, Y. & Baumes, R. (2000). Identification of impact odorants in Bordeaux red grape juice, in the commercial yeast used for its fermentation, and in the produced wine. *Journal of Agricultural and Food Chemistry*, Vol.48, No. 2, (January, 2000), pp. 400-406, ISSN 0021-8561

López de Lerma, N. & Peinado, R.A. (2011). Use of two osmoethanol tolerant yeast strain to ferment must from Tempranillo dried grapes: Effect on wine composition. *International Journal of Food Microbiology*, Vol.145, No.1, (January 2011), pp.342-348, ISSN: 0168-1605

Lopez, R.; Ortin, N.; Perez-Trujillo, J.P.; Cacho, J. & Ferreira, V. (2003). Impact odorants of different white wines from the Canary Islands. *Journal of Agricultural and Food Chemistry*, Vol.51, No.11, (April 2003), pp.3419–3425, ISSN: 0021-8561

Lopez, R.; Aznar, M.; Cacho, J. & Ferreira, V. (2002). Determination of minor and trace volatile compounds in wine by solid-phase extraction and gas chromatography with mass spectrometric detection. *Journal of Chromatography A* Vol. 966, No.1-2, (August 2002), pp.167- 177, ISSN: 0021-9673

Loscos, N.; Hernandez-Orte, P.; Cacho, J. & Ferreira, V. (2009). Fate of Grape Flavor Precursors during Storage on Yeast Lees. *Journal of Agricultural and Food Chemistry*, Vol.57, No. 12, (May 2009), pp.5468-5479, ISSN 0021-8561

Marais, J. & Rapp, A. (1988). Effects of skin-contact time and temperature on juice and wine composition and wine quality. *South African Journal of Enology and Viticulture*, Vol.9, No. 1, (May 1988), pp. 22–30, ISSN: 0253-939X

Marais, J.; Van Wyk, C.J. & Rapp, A. (1992).Effect of sunlight and shade on norisoprenoids levels in maturing weisser Riesling and Chenin blane grapes and weisser Riesling wines. *South African Journal of Enology and Viticulture*. Vol. 13, No. 1, (Mach 1992), pp. 23-32, ISSN: 0253-939X

Mateo, J. J.; Jiménez, M.; Pastor, A. & Huerta, T. (2001). Yeast starter cultures affecting wine fermentation and volatiles. *Food Research International*, Vol.34, No.4, (April 2001), pp.307–314, ISSN: 0963-9969

Moyano, L.; Zea, L.; Moreno, J. & Medina, M. (2002).Analytical study of aromatic series in sherry wines subjected to biological ageing. *Journal of Agricultural and Food Chemistry*. Vol.50, No.25, (November 2002), pp.7356–7361, ISSN: 0021-8561

Muñoz, D.; Peinado, R.A.; Medina, M. & Moreno, J. (2007). Biological aging of sherry wine under periodic and controlled microaerations with *Saccharomyces cerevisiae* var. *Capensis*: Effect on odorant series. *Food Chemistry*,Vol.100, No.3, (January 2003), pp.1188-1195, ISSN: 0308-8146

Pedroza, M. A.; Zalacain, A.; Lara, J.F. & Salinas, M.R. (2010). Global grape aroma potential and its individual analysis by SBSE–GC–MS. *Food Research International*, Vol.43, No.4, (May 2010), pp.1003–1008, ISSN: 0963-9969

Peinado, R. A.; Mauricio, J. C. & Moreno, J. (2006). Aromatic series in sherry wines with gluconic acid subjected to different biological aging conditions by Saccharomyces cerevisiae var. Capensis. *Food Chemistry*, Vol.94, No.2, (January 2006), pp. 232–239, ISSN: 0308-8146

Peinado, R.A.; Moreno, J.; Bueno, J.E.; Moreno, J.A. & Mauricio, J.C. (2004). Comparative study of aromatic compounds in two young white wines subjected to pre-fermentative cryomaceration. *Food Chemistry*, Vol.84, No.4, (March 2004), pp.585-590, ISSN: 0308-8146

Pineau, B.; Barbe, J. C.; Van Leeuwen, C. & Dubourdieu, D. (2007). Which impact for b-damascenone on red wines aroma. *Journal of Agricultural and Food Chemistry*., Vol.55, No.10, (April 2007), pp.4103–4108, ISSN: 0021-8561

Rapp, A. (1998). Volatile flavour of wine: Correlation between instrumental analysis and sensory perception. *Food/Nahrung*, Vol. 42, No. 6, (December 1998), pp. 351–363, 1521-3803

Rocha, S. M.; Coutinho, P.; Coelho, E.; Barros, A. S.; Delgadillo, I. & Coimbra, M. A. (2010). Relationships between the varietal volatile composition of the musts and white wine aroma quality. A four year feasibility study. *LWT - Food Science and Technology*. Vol. 43, No. 10, (December 2010), pp. 1508-1516, ISSN: 0023-6438

Rocha, S.M.; Rodrigues, F.; Coutinho, P.; Delgadillo, I. & Coimbra, M.A. (2004). Volatile composition of Baga red wine: Assessment of the identification of the would-be impact odourants. *Analytical Chimica Acta*, Vol. 513, No.1, (June 2004), pp. 257-262, ISSN: 0003-2670

Rodríguez-Bencomo, J.J.; Cabrera-Valido, H.M.; Pérez-Trujillo, J.P. & Cacho, J. (2011). Bound aroma compounds of Gual and Listán blanco grape varieties and their influence in the elaborated wines. *Food Chemistry*, Vol.127, No.3, (August 2011), pp.1153-1162, ISSN: 0308-8146

Rodríguez-Bencomo, J.J.; Conde, J.E.; Rodríguez-Delgado, M.A.; García-Montelongo, F. & Perez-Trujillo J.P. (2002). Determination of esters in dry and sweet white wines by headspace solid-phase microextraction and gas chromatography. *Journal of Chromatography*, Vol.963, No.1-2, (July 2002) pp.213-223, ISSN: 0021-9673

Sala, C.; Mestres, M.; Marti, M.P.; Busto, O. & Guasch, J. (2002). Headspace solid-phase microextraction analysis of 3-alkyl-2-methoxypyrazines in wines. *Journal of Chromatography A* Vol. 953, No.1-2, (April 2002), pp.1-6, ISSN: 0021-9673

Sánchez Palomo, E.; González-Viñas, M.A.; Díaz-Maroto, M.C.; Soriano-Pérez, A. & Pérez-Coello, M.S. (2007). Aroma potential of Albillo wines and effect of skin-contact treatment. *Food Chemistry* Vol.103, No. 2, (March 2007), pp.631–640, ISSN: 0308-8146

Sánchez-Palomo, E.; Pérez-Coello, M. S.; Díaz-Maroto, M. C.; González Viñas, M. A., & Cabezudo, M. D. (2006). Contribution of free and glicosidically bound volatile compounds to the aroma of Muscat "a petit grains"wines and effect of skin contact. *Food Chemistry*, Vol.95, No. 2, (October 2006), pp.279-289, ISSN: 0308-8146

Sánchez-Palomo, E.; Gómez García-Carpintero, E.; Alonso-Villegas, R. & González-Viñas, M. A. (2010). Characterization of aroma compounds of Verdejo white wines from the La Mancha region by odour activity values. *Flavour and Fragrance Journal*, Vol.25, No.6, (July 2010), pp.456–462, ISSN: 1099-1026

Selli, S.; Cabaroglu, T.; Canbas, A.; Erten, H.; Nurgel, C.; Lepoutre, J.P. & Gunata, Z. (2004). Volatile composition of red wine from cv. Kalecik Karasi grown in central Anatolia. *Food Chemistry*, Vol.85, No.2, (April 2004), pp.207-213, ISSN: 0308-8146

Spranger, M. I.; Clímaco, M. C.; Sun, B.; Eiriz, N.; Fortunato, C.; Nunes, A.; Leandro, M. C.; Avelar, M. L. & Belchoior, P. (2004). Differentiation of red winemaking technologies by phenolic and volatile composition. *Analytical Chimica Acta* Vol.513, No. 1, (June 2004), pp. 151-161, ISSN: 0003-2670

Suárez, R.; Suárez-Lepe, J.A.; Morata, A. & Calderón, F. (2007). The production of ethylphenols in wine by yeast of the genera Brettanomyces and Dekkera: A review. *Food Chemistry*, Vol. 102, No.1, (March 2006), pp.10-21, ISSN: 0308-8146

Swiegers, J.H.; Bartowsky, E.J.; Henschke, P.A. & Pretorius, I.S. (2005). Yeast and bacterial modulation of wine aroma and flavour. *Australian Journal of grape and wine research*, Vol. 11, No.2, (June 2008), pp.139-173, ISSN: 1755-0238

Ugliano, M. & Moio, L. (2008). Free and hydrolytically released volatile compounds of Vitis vinifera L. cv. Fiano grapes as odour-active constituents of Fiano wine. *Analytica Chimica Acta*, Vol.621, No.1, (July 2008), pp.79-85, ISSN: 0003-2670

Ugliano, M.; Bartowsky, E.J.; McCarthy, J.; Moio, L. & Henschke, P. A. (2006). Hydrolysis and transformation of grape glycosidically bound volatile compounds during fermentation with three Saccharomyces yeast strains. *Journal of Agricultural and Food Chemistry*, Vol. 54, No.17, (August 2006), pp. 6322-6331, ISSN: 0021-8561

Vianna, E. & Ebeler, S.E. (2001). Monitoring Ester Formation in Grape Juice Fermentations Using Solid Phase Microextraction Coupled with Gas Chromatography−Mass Spectrometry. *Journal of Agricultural and Food Chemistry*, Vol.49, No.2, (January 2001), pp.589-595, ISSN: 0021-8561

Vilanova, M.; Cortés, S.; Santiago, J. L.; Martínez, C. & Fernández, E. (2008). Contribution of some grape-derived aromatic compounds to the primary aroma in red wines from cv. Caiño Tinto, cv. Caiño Bravo and cv. Caiño Longo grapes. *Journal of Agricultural Science*, Vol.146, No.3, (November 2007), pp.325–332, ISSN: 1916-9752

Vilanova, M.; Masa, A. & Tardaguila, J. (2009). Evaluation of the aromatic variability of Spanish grape by quantitative descriptive analysis. *Euphytica*, Vol.165, No.2, (January 2009) pp.383–389, ISSN: 1573-5060

Zea, L.; Moyano, L.; Moreno, J. & Medina, M. (2007). Aroma series as fingerprints for biological ageing in fino sherry-type wines. *Journal of the Science of Food and Agriculture*, Vol.87, No.12, (July 2007), pp.2319–2326, ISSN: 1097-0010

ISO, (1997). Sensory Analysis. Apparatus wine-tasting glass. ISO 3591-1997, Group B, 3 pp.

ISO, (1998). Guide for the installation of a chamber for sensory analysis. ISO 8589-1998, Group E, 9 pp.

**Gas Chromatography in Plant Science, Wine Technology,
Toxicology and Some Specific Applications**
Edited by Dr. Bekir Salih

ISBN 978-953-51-0127-7

Hard cover, 346 pages

Publisher InTech

Published online 29, February, 2012

Published in print edition February, 2012

The aim of this book is to describe the fundamental aspects and details of certain gas chromatography applications in Plant Science, Wine technology, Toxicology and the other specific disciplines that are currently being researched. The very best gas chromatography experts have been chosen as authors in each area. The individual chapter has been written to be self-contained so that readers may peruse particular topics but can pursue the other chapters in the each section to gain more insight about different gas chromatography applications in the same research field. This book will surely be useful to gas chromatography users who are desirous of perfecting themselves in one of the important branch of analytical chemistry.

How to reference

In order to correctly reference this scholarly work, feel free to copy and paste the following:

Eva Sánchez-Palomo, Eva Gómez García-Carpintero, Manuel Ángel Gómez Gallego and Miguel Ángel González Viñas (2012). The Aroma of Rojal Red Wines from La Mancha Region – Determination of Key Odorants, Gas Chromatography in Plant Science, Wine Technology, Toxicology and Some Specific Applications, Dr. Bekir Salih (Ed.), ISBN: 978-953-51-0127-7, InTech, Available from: http://www.intechopen.com/books/gas-chromatography-in-plant-science-wine-technology-toxicology-and-some-specific-applications/the-aroma-of-rojal-red-wines-from-la-mancha-region-determination-of-key-odorants

INTECH
open science | open minds