Changes in Aromatic Characteristics of *Loureiro* and *Alvarinho* Wines during Maturation

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Abstract

Changes in volatiles during maturation in bottles of monovarietal *Vinhos Verdes* wines from *Loureiro* and *Alvarinho* grape varieties, were followed by chemical and sensory analyses. Young wines and wines matured for 8 and 20 months were studied. The volatiles were determined by GC-MS after extraction on XAD-2 resin. Straight chain fatty acid ethyl esters and acetates of fusel alcohols decreased quicker for *Loureiro* wine, while the increase in ethyl esters of branched fatty acids was similar for both varieties. Linalool, Ho-trienol, α-terpineol and β-damascenone could be used to differentiate between each variety. However, linalool decreased to negligible values after 20 months of maturation. β-damascenone decreased but remained high enough to be useful for differentiating each variety. Sensory analysis indicated a decrease of tropical fruit and tree fruit characters with conservation time for *Alvarinho* wine, and the opposite for *Loureiro*; moreover, citrus fruit character decreased in both varieties.

Keywords: wine; aroma; volatiles; *Loureiro*; *Alvarinho; Vinhos Verdes;* maturation conservation; ageing

1. Introduction

Wines designated "Appellation of Origin *Vinhos Verdes"* are produced in Northern Portugal composed of 9 sub-regions (Amarante, Ave, Baião, Basto, Cávado, Lima, Monção, Paiva and Sousa). There are seven recommended white grape varieties (*Alvarinho*, *Arinto*, *Avesso*, *Azal*, *Batoca*, *Loureiro* and *Trajadura*) and eight red grape varieties (*Amaral*, *Borraçal*, *Brancelho*, *Espadeiro*, *Padeiro de Basto*, *Pedral*, *Rabo de Ovelha* and *Vinhão*) to produce these wines. Among the white cultivars, *Alvarinho* and *Loureiro* are employed to produce quality monovarietal wines, which are characterized by freshness and floral and fruity flavours. In order to preserve these characteristics, traditional winemaking techniques are developed to encourage these notes and to avoid malolactic fermentation. Legislation stipulates ethanol concentrations of between 8.0 % and 11.5 %, but for *Alvarinho* wines it must be between 11.5 % and 14.0 %; other monovarietal wines may have concentrations below 14.0 %. Fix acidity, expressed as tartaric acid, must be at least 4.5 g L^{-1}. It is well known, however, that *Vinhos Verdes* loses quickly their aromatic characteristics during maturation. So, they are usually drunk during the first year; nevertheless, no systematic studies were conducted on this subject.

It is well known that during wine maturation and ageing there are many chemical changes in the volatile composition. These reactions depend on wine composition, pH, storage time and temperature (Marais, 1978; Marais et al., 1980; Ramey et al., 1980; Usseglio-Tomasset, 1983). The majority of fatty acid ethyl esters is hydrolysed during conservation and, ethyl esters of fatty acids related to yeast nitrogen metabolism and esters of organic acids increase during this period (Díaz-Maroto et al., 2005; Dubois, 1994; Shinohara et al., 1981). Also, the terpenic profile may change, with the disappearance or strong decline of the compounds initially present, with the simultaneous formation of other terpenic compounds with higher oxidation state; temperature and pH have a decisive influence (Di Stefano, 1986 and 1989; Di

Stefano and Castino, 1983; Marais et al., 1992). Some norisoprenoids may appear or increase their concentration during the ageing period, *e. g.* β-damascenone, TDN and vitispirane (Marais et al., 1992; Simpson, 1979; Simpson and Miller, 1983). The acidic medium also favours the hydrolysis of glycosidic precursors and the transformation of aglycon moieties (Dugelay, 1993; Günata et al., 1986; Sefton et al., 1993).

Since *Loureiro* and *Alvarinho* wines should be drunk as young wines (with about 8 months of conservation, in expert's opinion) and because their aromatic characteristics decline quickly during ageing, it is very important to study the volatile composition of these wines during storage in bottles. There are some published data on the volatile composition of *Loureiro* and *Alvarinho* wines (Guedes-de-Pinho, 1991; Oliveira, 1995; Oliveira et al., 1997; Rogerson and Silva, 1994) and the congeners *Loureira* and *Albariño* Galician wines of Northwest Spain (Falqué, 1998; García-Jares et al., 1994; Lema et al., 1996; Orriols and Camacho, 1991 and 1992; Orriols et al., 1993; Versini et al., 1994) but they do not refer to the changes occurring during maturation in the bottle. Nevertheless, Oliveira et al. (2008) conducted recently an exhaustive study on the volatile and glycosidically bound composition of *Loureiro* and *Alvarinho* wines.

The aim of the present work was to study the evolution of volatile composition of *Loureiro* and *Alvarinho* wines during maturation, *i. e*., at the end of alcoholic fermentation, and after stored in bottles, with 8 months and 20 months. Sensory evaluation was also undertaken for the last two stages.

2. Materials and Methods

2.1. Grape samples

About 40 kg of grapes were manually harvested in 1998, randomly, among the vines of 3 selected rows of the vineyard, at the recommended sub-region for each studied variety: *Loureiro* at Estação Vitivinícola Amândio Galhano –EVAG– (Lima sub-region), L_{AV}, and

Alvarinho at Solar de Serrade (Monção sub-region), A_{SS}. Both soils are from granitic origin and rows orientation is N-S. *Loureiro* and *Alvarinho* vineyards were 11 and 16 years old, respectively. Training systems and rootstocks are, respectively, for *Loureiro* and *Alvarinho*: single cordon and SO4; "cruzeta" and 1103 P.

2.2. Vinifications

Vinifications were made according to the traditional technology of the *Vinhos Verdes* region. The must obtained by crushing, pressing and static sedimentation was inoculated with *Saccharomyces cerevisiae bayanus* QA23. Fermentations took place at 18 ºC, in 10 L vessels, and were in duplicate. The produced wines were combined and the blend was treated with sodium bentonite –*Volclay KWK Food Grade*, 20-70 *mesh*, 10 % in aqueous solution– (0.4 g L^{-1}), the SO_2 content was corrected to 35 mg L^{-1}, and submitted to cold stabilization (between 0 ºC and 3 ºC) before bottling. The maturation of the wines occurred at cellar temperature and in the dark. The wines did not undergo malolactic fermentation. The evaluation of volatile composition was made in young wines –W_1– (subsequent to alcoholic fermentation), after 8 months –W_2– and 20 months –W_3– of maturation, which corresponds, respectively, to a period of 6 months and 18 months in bottle. General analyses of wines were performed at *Comissão de Viticultura da Região dos Vinhos Verdes*.

2.3. Solvents

All solvents were analytical grade and further purified. Diethyl ether (*Merck, ref. 1.00921*) was distilled on iron (II) sulphate (*Merck, ref. 1.03965*). Dichloromethane (*Merck, ref. 1.06050*) was washed with de-ionised water, and then distilled. Pentane (*Carlo Erba, ref. 468151*) was washed with H_2SO_4 (*Merck, ref. 1.00731*), $KMnO_4$ (*Carlo Erba, ref. 473387*) and ultrapure water, and next it was distilled on potassium hydroxide (*Merck, ref. 1.05033*). Azeotrope pentane-dichloromethane was distilled after combination of pentane and dichloromethane (2:1, v/v) and it was redistilled whenever necessary.

2.4. Extraction of volatile compounds

Wine samples result from the content of three bottles, by blend, and were extracted in triplicate. To 100 mL of wine, centrifuged (25 min, *RCF* = 12 225, 4 ºC) and diluted with ultrapure water to reduce the alcohol content to less than 5 %, were added 14.5 µg of 4-nonanol (*Merck*, ref. 818773). The solution was passed through an Amberlite XAD-2 resin (*20-60 mesh, Supelco, ref. 1-0357*) column according to the method of Günata et al. (1985). Volatile compounds were eluted with 50 mL of azeotrope pentane-dichloromethane. The eluate was dried over anhydrous sodium sulphate and concentrated to about 2 mL by solvent evaporation at 34 ºC through a Vigreux column, prior to analyses.

2.5. Gas chromatography – mass spectrometry (GC-MS)

Gas chromatographic analysis of volatile compounds was performed using a GC-MS (Varian 3400 Chromatograph and an *ion-trap* mass spectrometer Varian Saturn II). Each 1 µL injection was made separately in two capillary columns, coated with CP-Wax 52 CB or CP-Wax 57 CB (50 m x 0.25 mm i.d., 0.2 µm film thickness, Chrompack). The temperature of the injector (SPI – septum-equipped programmable temperature) was programmed from 20 ºC to 250 ºC, at 180 ºC min^{-1}. The temperature of the oven was held at 60 ºC, for 5 min, then programmed from 60 ºC to 250 ºC (60 ºC to 220 ºC for the second column), at 3 ºC min^{-1}, then held 20 min at 250 ºC (30 min at 220 ºC) and finally programmed from 250 ºC to 255 ºC at 1 ºC min^{-1} (220 ºC to 225 ºC at 2 ºC min^{-1}). The carrier gas was helium N60 (Air Liquide), at 103 kPa. The detector was set to electronic impact mode (70 eV), with an acquisition range (*m/z*) from 29 to 360, and an acquisition frequency of 610 ms.

2.6. Identification and quantification of volatile compounds

Identification was performed using the software Saturn version 5.2 (Varian), by comparing mass spectra and retention index with those of pure standard compounds. In some cases, the identification was achieved by comparing our retention index and mass spectra with

published data. The quantification was performed using data obtained in CP-Wax 52 CB column, mainly. The second column, CP-Wax 57 CB, served essentially to confirm spectra of the co-eluted compounds and, in general, it was useful for alcohols. All the compounds were determined, semi-quantitatively, as 4-nonanol equivalents.

2.7. Sensory analysis

Wines with 8 months (W_2) and 20 months (W_3) of maturation were submitted to sensory evaluation at *Comissão de Viticultura da Região dos Vinhos Verdes* (CVRVV). Judges were chosen amongst wine experts and they had a full knowledge about the products. W_2 wines were evaluated by 7 tasters and W_3 wines by 8 (5 of them being common to both); *Loureiro* and *Alvarinho* wines were coded randomly and tasted independently in each session using the distribution prepared according to aleatory tables. Normalised glasses were used (ISO 3591) and the room was kept at 21 ºC and 65 % of relative humidity. The wine score card was that used by Tasting Room of CVRVV, evaluating several attributes (scale 0 to 5) relating to visual, olfactory and gustative characteristics. Tasters also classified global appreciation (scale 0 to 20).

2.8. Statistical analyses

Statistical differences between wines, with respect to chemical analysis, were evaluated by Analysis of Variance (ANOVA) or, by independent-samples T test, when comparing wines from the two varieties with the same age. Homogeneity of variances was checked with the Levene test and normality of the variables was checked by the Kolgomorov-Smirnov test with Lilliefors correction, both at a significance level of 5 %. Whenever one of these two conditions fails, the non-parametric Kruskall-Wallis test was applied. Also, global classification obtained in the sensory analysis was studied by means of Analysis of Variance in order to evaluate hypothetical differences between wines of the same variety. ANOVA was also used to assess the evolution of wines between W_2 and W_3 respecting global appreciation.

The behaviour of some compounds during conservation period was checked by Regression Analysis using linear, quadratic, cubic and exponential models, at a significance level of 5 %.

Similarities between wines, with respect to specific compounds, were analysed by Principal Component Analysis, being component extraction achieved by correlation matrix and their number fixed according to Kaiser criterion, *i.e.*, all the components with eigenvalues over 1.

The software used was SPSS 14.0 for Windows.

3. Results and Discussion

3.1. General analysis

The various characteristics of the wines matured for 8 months are summarised in table 1. Both monovarietal wines fulfil the criteria to obtain the Appellation of Origin *Vinho Verde* label.

3.2. Volatile composition of Loureiro and Alvarinho wines

The volatile extracts were obtained by solid phase extraction of diluted wines (lowering the alcoholic content below 5 %) using XAD-2 resin as report previously (Voirin et al., 1992; Aubert et al., 1997). GC-MS analysis allowed the identification and quantification of 120 volatile compounds including 5 C_6-compounds, 23 alcohols, 6 fatty acid ethyl esters related to lipid metabolism and 3 related to nitrogen metabolism, 10 esters of organic acids, 7 acetates, 8 monoterpenic alcohols, 15 monoterpenic oxides and diols, 13 C_{13}-norisoprenoids, 13 volatile phenols, 8 volatile fatty acids related to lipid metabolism and 3 related to nitrogen metabolism, 4 carbonyl compounds and also pantolactone and N-(2-phenylethyl)-acetamide (figure 1). This classification takes into account the chemical structure of the volatile compounds, the pathways that lead to their formation and the olfactory perception threshold. Only four compounds were identified by comparison of our retention index and mass spectra with published data and one was tentatively identified.

Table 2 shows the mean level obtained for each compound in the nine samples analysed. These levels were determined, semi-quantitatively, as 4-nonanol equivalents.

Monoterpenic compounds (alcohols, oxides and diols), C_{13}-norisoprenoids and some volatile phenols may be considered as varietal compounds because they were present in grape and/or arise from grape precursors. Unsaturated C_6-alcohols are related to varietal origin because they can be formed, via C_6-aldehydes, through enzymatic reactions from linolenic and linoleic acids present in grapes (Crouzet et al., 1998). However, because of their mainly fermentative origin, 1-hexanol, 4-ethylphenol, 4-vinylguaiacol and 4-vinylphenol were excluded from the varietal group (Chatonnet et al., 1992 and 1993; Joslin and Ough, 1978).

3.3. Evolution of volatile compounds during bottle conservation

Figure 2 represents the evolution of each group of volatile compounds during maturation of *Loureiro* and *Alvarinho* wines. Since the levels of esters of organic acids and volatile fatty acids related to yeast lipid metabolism are much higher than those of the other groups, they are not shown; furthermore, alcohols such as 2-methyl-1-butanol, 3-methyl-1-butanol and 2-phenylethanol, are not included for the same reason. However, the change in their levels can be observed in table 2.

The different groups of compounds generally behaved predictability. C_6-compounds, alcohols and volatile fatty acids related to yeast lipid metabolism are almost stable during the 20 months of maturation. The small fluctuations observed in table 2 and figure 2 were not statistically significant ($p>0.05$) except for C_6-compounds in *Alvarinho* wine which demonstrated a slight decline ($F=6.228$, $p=0.034$). Nevertheless, the analytical method used gave high concentration error for the more abundant alcohols (2-methyl-1-butanol, 3-methyl-1-butanol and 2-phenylethanol), probably attributed to the mechanism of adsorption/desorption on XAD-2 resin and/or to column and/or detector saturation. Excluding these compounds, the sum of the other alcohols decrease for *Alvarinho* wine, chiefly between

W_1 and W_2, whereas *Loureiro* wine present a minimum for the W_2 stage ($p<0.05$). However, benzyl alcohol increased significantly between W_1 and W_3 ($p<0.05$), predominantly for *Alvarinho* wine ($F=942.083$, $p=0.000$), probably because of precursor hydrolysis.

The sum of 2-methyl-1-butanol and 3-methyl-1-butanol concentrations were near to their perception threshold limits of 7 mg L^{-1}, contributing certainly to the olfactory characteristics of the 6 wines (Rapp and Mandery, 1986; Rapp and Versini, 1995; Salo, 1970). Also 3-(methylthio)-1-propanol, with *Odour Activity Values –OAV– (concentration/perception threshold*) of approximately 0.1, may contribute, although marginally, since its odour threshold is about 1 mg L^{-1} (Escudero et al., 2004; Meilgaard, 1975). On the other hand, 2-phenylethanol may contribute decisively to the aroma of these wines, mainly *Loureiro* ones, as the concentration was at least twofold the perception threshold of 7.5 mg L^{-1} (Salo, 1970); nevertheless, the floral descriptor found by sensory analysis was not significantly related with its concentration, as it was expected from the rose-like aroma (Escudero et al., 2004; Meilgaard, 1975).

The (*E*)/(*Z*) isomer ratio of 3-hexen-1-ol was almost constant during the storage period, with mean values of 6.33 ± 0.22 ($n=9$) and 0.66 ± 0.02 ($n=9$) for *Loureiro* and *Alvarinho*, respectively (95 % confidence level). These results indicate the possibility to discriminate wines from these two varieties.

The levels of esters of organic acids underwent a significant increase during the storage period, because of chemical esterification. This was most pronounced for monoethyl succinate, diethyl succinate and diethyl malate, in agreement with previous observations (Shinohara, 1984).

On the other hand, while the levels of acetates decreased sharply with ageing in the wines of both varieties, the ethyl esters of straight chain fatty acids related to yeast lipid metabolism decreased slowly and progressively during maturation of *Loureiro* wines, and did not

significantly decreased in *Alvarinho* wines (p>0.05). Thus, W_3 *Loureiro* wine had 80 % of the total level of the straight chain fatty acid ethyl esters present at the end of alcoholic fermentation (W_1) but it contained 5 % only of acetates; W_2 stage presented 97 % and 30 %, respectively. In *Alvarinho* wines, acetates also decreased sharply from W_1 to W_2 and W_3 stages, being about 52 % and 11 % respectively of the initial level, but this decrease was slower than in *Loureiro* wines. The faster ester hydrolysis in *Loureiro* wines could be due to their lower pH (Ramey and Ough, 1980). Contrarily to these esters, the ethyl esters of fatty acids related to yeast nitrogen metabolism, *i. e.* ethyl 2-methylbutyrate, ethyl 3-methylbutyrate and ethyl benzeneacetate, increased in the wines of both varieties during the conservation period, as their esterification ratios were very low in W_1 (Díaz-Maroto et al., 2005). The first two esters may contribute marginally to the aroma of *Loureiro* and *Alvarinho* wines as they present *OAV* values above 0.1, since their odour thresholds are 18 μg L^{-1} and 3 μg L^{-1}, respectively (Escudero et al., 2004); additionally, for W_2 and W_3 wines of both varieties, ethyl 3-methylbutyrate has *OAV* values much higher than 1.0.

With the notable exception of linalool in *Loureiro* wines, the monoterpenic alcohols in the wines of both varieties had similar behaviours, showing a sharp increase between W_1 and W_2. As their levels were much lower than the levels of their bound forms (Oliveira et al., 2008), these variations were mainly due to the acid-catalyzed transformations of these monoterpenols during ageing, particularly that of linalool into α-terpineol, linalool hydrate and furan linalool oxides, explaining its sharp decrease in *Loureiro* wines, but the hydrolysis of the bound forms could also be involved at a lesser extent, explaining the increase of linalool in *Alvarinho* wines (Di Stefano and Castino, 1983; Dugelay, 1993; Günata et al., 1986; Marais et al., 1992; Simpson and Miller, 1983; Usseglio-Tomasset and Di Stefano 1980; Williams et al., 1980 and 1982). Then, between W_2 and W_3, these effects continue, but due to the lowering of the levels of the starting materials of the above primary

transformations, they were no longer able to match the decrease of the products formed, due to their own transformations into even more polar forms or more complex ones, leading to the beginning of their decrease.

Thus, monoterpenic alcohols present a maximum concentration in W_2 wines for both varieties. The levels of linalool in *Loureiro* wine decreased almost linearly during the conservation period, being present in W_3 only at trace amounts, whereas α-terpineol and Ho-trienol remained at these last stages the most abundant monoterpenols. It must be noted that after alcoholic fermentation (W_1), the level of linalool was approximately 3.5 times higher than in *Alvarinho* wine, but it decreased to the level in *Alvarinho* wine after 8 months of maturation (W_2), then kept on decreasing to very low levels for both varieties after 20 months of maturation (W_3) (2.0 μg L^{-1} and 12.0 μg L^{-1}, respectively). On the other hand, the levels of Ho-trienol and α-terpineol in the wines of both varieties became increasingly similar with ageing. Thus, ageing appeared to decrease differentiation of the wines of each variety based on these compounds. Myrcenol was characteristic of *Loureiro* wines, but it was not detected in grapes and musts of this variety (Oliveira, 2000; Oliveira et al., 2000).

The total levels of monoterpenic oxides and diols increased during the storage of the wines of both varieties. However, 2 groups of compounds could be differentiated. The first included furan linalool oxides, neroloxide and the hydrates of linalool, citronellol and terpin and demonstrated a sharp increase in their levels during the 20 months of maturation. The second group, included 3 oxides (pyran linalool oxides and *exo*-2-hydroxy-1,8-cineole) and the diendiols, did not present a significant evolution during the same period, but generally reached a maximum of concentration in the W_2 wine. The behaviours of these 2 groups could be explained by the mechanisms detailed above for the monoterpenic alcohols. As reported previously (Oliveira et al., 2008), (*Z*)-8-hydroxylinalool may distinguish *Alvarinho* and *Loureiro* wines, increasing the difference with the storage time.

For both varieties, the concentration of all the C_{13}-norioprenoids increased during wine maturation, with the exception of β-damascenone and 3-hydroxy-β-damascone. These compounds constantly decreased, but β-damascenone was always present above its human perception threshold which is very low, 45 ng L^{-1} (Ribéreau-Gayon et al., 2000). Indeed, C_{13}-norisoprenoids were found in musts and young wines almost exclusively as glycosidic precursors, with levels much higher than those of the free forms in the W_1 wines. This explained their increase through hydrolysis of these bound forms during winemaking and the relatively short storage time of the study (Winterhalter, 1992 and 1996; Winterhalter and Schreier 1994). Thus, the C_{13}-norisoprenoids listed in table 2 from 3-hydroxy-7,8-dihydro-β-ionone to vomifoliol were unchanged aglycons from these glycosides. On the other hand, the compounds listed from vitispirane I to 3-hydroxy-β-damascone arise from norisoprenoidic precursor transformations during wine conservation. The referred precursors of β-damascenone and 3-hydroxy-β-damascone are 3,6,9-trihydroxymegastigma-6,7-diene and 3-hydroxy-7,8-dehydro-β-ionol (Puglisi et al., 2005; Winterhalter and Schreier, 1994); however, 3,6,9-trihydroxymegastigma-6,7-diene could not be identified under our GC-MS conditions. The sharp decrease of β-damascenone and 3-hydroxy-β-damascone, in contrast to the increase of other C_{13}-norisoprenoids, would be explained by their rapid release from 3,6,9-trihydroxymegastigma-6,7-diene and their interaction with sulfur dioxide, as demonstrated previously (Daniel et al., 2004). Indeed, β-damascenone and 3-hydroxy-β-damascone contained two reactive cross-conjugated enones moieties, absent in the ionone derivatives. On the other hand, TDN could derive from different precursors, namely 3-hydroxy-β-ionone, 3,4-dihydroxy-β-ionol, 3,4-dihydroxy-7,8-dihydro-β-ionol, 3,9-dihydroxytheaspirane and 3,4-dihydroxy-7,8-dihydro-α-ionone, and as well as vitispiranes from 3,4-dihydroxy-7,8-dihydro-β-ionol, megastigma-4-ene-3,6,9-triol and 3,4-dihydroxy-6,9-epoxymegastigmane

(Winterhalter, 1993; Winterhalter and Skouroumounis, 1997; Winterhalter and Schreier, 1994; Winterhalter et al., 1998).

The most abundant volatile phenols, 4-vinylguaiacol and 4-vinylphenol, were mainly generated by yeasts during alcoholic fermentation, and decreased significantly during the storage time. That was consistent with their conversion during wine storage, into derivatives not amenable to our GC-MS conditions, such as 4-(ethoxyethyl)-phenol, 4-(ethoxyethyl)-guaiacol and pyroanthocyanins (Dugelay et al., 1995; Hayasaka and Asenstorfer, 2002; Mateus et al., 2004). The other volatile phenols had a varietal origin, occurring mainly from hydrolysis of their glycoconjugates (Oliveira et al., 2000). As in the W_1 wines, the levels of these bound forms were as low as those of their free forms (Oliveira et al., 2008). Variations were generally not significant, despite an upward trend.

Finally, volatile compounds supposed to influence the aromatic characteristics of *Alvarinho* and *Loureiro* wines (from their levels and perception thresholds) which exhibited statistically significant ($p<0.05$) variations during bottle conservation were grouped according to their behaviour, *i. e.*, if their level decreased, increased or if a maximum at W_2 stage was observed (table 3). These results corroborate the discussion above as demonstrated in table 2 and figure 2. However, most variations observed in the wines of the two varieties were small, particularly those between the stages W_2 and W_3.

Overall results, concerning the total levels by groups of the varietal compounds in the 6 wines were analyzed by principal component analysis. Figure 3 represents the two first principal components, which accounted for 82.8 % of samples initial variability. Component 1 accounted for 42.6 % of total variance and showed the potential to discriminate between *Loureiro* and *Alvarinho* wines. Component 2, which explained 40.2 %, allowed differentiation according to the ageing period. *Loureiro* wines were characterized by higher levels of C_6-compounds and monoterpenic compounds, including alcohols, oxides and diols,

whereas higher levels of varietal volatile phenols and C_{13}-norisoprenoids were characteristic of W_3 wines. On the other hand, the fermentative compounds listed in table 2 did not permit discrimination the wines of each cultivar, which was consistent with their usual classification as non-varietal compounds.

3.4. Sensory analysis

Sensorial descriptive analyses of *Loureiro* and *Alvarinho* wines with 8 months and 20 months of maturation were made (table 4).

Loureiro wines were clear. W_2 wines revealed a pale citrus colour while W_3 wine present a citrus colour, more appreciated by tasters. Wines were also classified as medium quality from overall sensations, including olfactory and gustative ones. Statistically, reporting on global appreciation, there were no differences between the two wines (F=0.008, p>0.05), *i.e.*, there was not any change of organoleptic characteristics. *Alvarinho* wines were clear and demonstrated an open straw colour. Concerning olfactory overall impression, the tasters considered that wines lost quality between W_2 and W_3 stages, having classified as "good" the wine after 8 months and as "medium" quality the wine after 20 months. This was caused by the lost of aromatic intensity on floral, citrus fruit and tropical fruit characters, and by the appearance of a slightly vegetal character. Gustative analysis revealed the same tendency. These considerations were also confirmed by statistic analysis considering global classification (F=16.603, p<0.01).

Alvarinho variety are characterised by a more intense tropical fruit and tree fruit character (figure 4), while in *Loureiro* wines the floral and citrus fruit aromas are more pronounced. *Alvarinho* wines were also characterised by dried fruit flavour. These considerations are in agreement with Guedes-de-Pinho et al. (1998) and Cerdeira et al. (1998 and 1999).

The main aromatic descriptors for *Alvarinho* wine were tropical fruit, tree fruit and dried fruit whereas for *Loureiro* they were floral and citrus fruit. These descriptors may be

associated to some flavour compounds; among them, (*Z*)-3-hexen-1-ol (green leaves), 3-methylbutyl acetate (banana, apple), β-damascenone (tropical fruit, stewed apple), 4-vinylguaiacol (phenolic, clove) and 4-vinylphenol (stramonium) may contribute chiefly for *Alvarinho* wines and 2-phenylethanol (rose), linalool (rose, floral, lemon) and Ho-trienol (linden) for *Loureiro* ones (Boidron et al., 1988; Escudero et al., 2004; Meilgaard, 1975; Ribéreau-Gayon et al., 2000).

During the maturation period (from 8 months to 20 months), *Alvarinho* wine lose overall aromatic intensity mainly related to tropical fruit, tree fruit and citrus fruit characters, while for *Loureiro*, only citrus fruit character decreased its intensity and the other two descriptors increased.

From the presented results it is clear that not all aroma sensations could be explained by the studied compounds. These monovarietal wines may contain some other aromatic contributors which were not identified by the methodology. In this context, varietal volatile thiols like 4-methyl-4-mercaptopentan-2-one, 3-mercaptohexanol and 3-mercaptohexyl acetate were not considered because analytical methodology was not available, although they may have an important role.

Future work may consider a large number of analyses between initial and end points, *i. e*. between young wine and matured wine for several months. Maturation influence on flavor characteristics of wines may be only discussed on the basis of some key volatile compounds, which should be quantitatively determined.

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Table 1. General analysis of wines with 8 months (L_{AV}-W_2 and A_{SS}-W_2)

	Loureiro	***Alvarinho***
Ethanol/(% vol.)	10.2	13.5
Reducing sugars/(g L^{-1})	1.1	3.4
Total acidity */(g L^{-1})	10.6	7.6
Volatile acidity **/(g L^{-1})	0.33	0.40
pH	2.81	3.03

*, as tartaric acid

**, as acetic acid

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Table 2. Mean levels* (*C*) with 95 % confidence limits for the volatile compounds found in *Loureiro* (L_{AV}) and *Alvarinho* (A_{SS}) wines, after alcoholic fermentation (W_1) and after 8 months (W_2) and 20 months (W_3) of maturation

				Loureiro						*Alvarinho*					
				W_1		W_2		W_3		W_1		W_2		W_3	
	#	*roi*	*RI*	*C*/(μg L^{-1})	±	*C*/(μg L^{-1})	±	*C*/(μg L^{-1})	±	*C*/(μg L^{-1})	±	*C*/(μg L^{-1})	±	*C*/(μg L^{-1})	±
C_6-compounds *(5)*															
1-hexanol	1	*a*	*1348*	972.2	145.9	976.8	109.4	1195.6	689.5	869.5	137.1	739.5	152.0	765.5	91.7
(*E*)-3-hexen-1-ol	2	*a*	*1358*	206.2	56.9	182.7	11.6	233.0	139.9	57.6	11.9	46.6	4.9	49.8	9.3
(*Z*)-3-hexen-1-ol	3	*a*	*1379*	33.6	11.3	29.6	3.9	35.0	22.4	90.4	17.8	71.3	4.8	72.7	10.7
(*E*)-2-hexen-1-ol	4	*a*	*1400*	tr		0.2	0.7	tr		—		tr		0.2	0.2
(*Z*)-2-hexen-1-ol	5	*a*	*1410*	2.3	0.8	2.0	0.5	2.4	1.4	0.9	0.2	0.8	0.1	0.7	0.2
total				**1214.3**		**1191.3**		**1466.0**		**1018.4**		**858.2**		**888.9**	
Alcohols *(23)*															
2-methyl-3-buten-2-ol	6	*a*	*1068*	2.6	0.0	6.7	2.8	7.8	2.7	?		3.7	0.9	2.8	0.7
2-methyl-1-propanol	7	*a*	*1082*	1233.1	535.3	775.1	100.2	932.7	689.9	1435.0	141.1	1067.2	344.2	986.6	143.8
1-butanol	8	*a*	*1140*	22.7	14.0	18.3	1.8	20.9	13.7	68.6	9.5	50.0	15.0	47.2	7.0
4-methyl-2-pentanol	9	*a*	*1164*	45.5	18.6	41.6	10.6	55.6	31.6	53.9	15.3	52.4	11.7	49.8	3.3
2-methyl-1-butanol + 3-methyl-1-butanol	10 11	*a*	*1204*	58535.5	28686.6	54741.1	12317.2	64583.2	3058.8	74488.5	25717.8	71637.3	29248.7	65672.3	23675.9
3-methyl-3-buten-1-ol	12	*a*	*1243*	4.3	1.9	3.6	0.9	2.4	1.1	5.5	0.9	4.1	1.6	3.5	1.1
1-pentanol	13	*a*	*1244*	6.5	3.2	6.5	2.9	13.4	16.7	11.6	2.7	10.0	4.5	10.7	1.8
2-methyl-1-pentanol	14	*a*	*1298*	?		0.6	0.1	0.3	0.6	—		—		—	
4-methyl-1-pentanol	15	*a*	*1309*	26.8	6.8	24.3	3.7	29.6	14.6	37.5	6.5	32.5	4.9	32.2	3.5
(*Z*)-2-penten-1-ol	16	*a*	*1313*	0.7	0.3	0.3	0.1	0.4	0.3	0.7	1.4	0.4	0.3	0.6	0.1
3-methyl-2-buten-1-ol + 2-heptanol	17 18	*a* *a*	*1316*	2.5	0.9	2.8	0.7	3.8	2.9	2.3	1.4	1.3	0.3	1.3	0.5
3-methyl-1-pentanol	19	*a*	*1322*	65.5	16.5	54.5	29.1	75.1	38.8	140.3	32.7	113.7	8.7	118.6	19.3
3-ethoxy-1-propanol	20	*a*	*1369*	66.5	41.6	58.2	7.4	56.8	41.7	75.5	16.6	54.2	11.5	48.6	6.1
1-octen-3-ol	21	*a*	*1445*	1.1	0.1	1.0	0.4	1.1	0.3	0.7	0.4	0.6	0.3	0.7	0.2
1-heptanol	22	*a*	*1449*	16.4	3.3	14.7	0.9	16.6	3.4	13.7	2.4	12.3	2.8	12.0	0.3
2-nonanol	23	*a*	*1541*	1.0	1.3	1.0	0.3	1.1	2.7	1.5	0.7	2.7	5.1	1.5	1.1
1-octanol	24	*a*	*1552*	9.5	0.6	13.5	3.1	10.8	2.0	6.2	1.6	8.3	0.6	7.7	1.2
3-(methylthio)-1-propanol	25	*a*	*1709*	103.4	67.2	79.8	11.8	84.5	55.5	135.2	25.3	98.5	20.6	87.1	8.3
benzyl alcohol	26	*a*	*1869*	5.2	1.2	18.2	8.3	16.2	9.6	5.9	1.2	13.7	1.8	36.4	3.2
2-phenylethanol	27	*a*	*1908*	28196.3	21202.6	23561.6	3908.8	31281.4	14343.7	20507.9	4319.5	15894.8	6174.5	16464.7	8149.2
tyrosol	28	*a*	*3008*	127.1	65.8	152.1	30.8	222.6	157.7	61.7	25.4	123.0	30.0	142.9	31.9
total				**88472.2**		**79575.5**		**97416.3**		**97052.2**		**89180.7**		**83727.2**	
total*				**1740.4**		**1272.8**		**1551.7**		**2055.8**		**1648.6**		**1590.2**	
Fatty acid ethyl esters – lipid metabolism *(6)*															
ethyl butyrate	29	*a*	*1032*	167.7	15.9	141.7	4.7	146.3	55.3	221.4	19.4	211.2	28.6	196.2	14.9
ethyl hexanoate	30	*a*	*1234*	465.1	46.0	422.8	51.1	336.7	44.1	513.6	75.5	488.5	46.4	462.5	49.1
ethyl octanoate	31	*a*	*1434*	482.1	30.3	545.5	48.3	437.1	153.5	577.8	91.0	672.7	170.3	664.2	71.2
ethyl decanoate	32	*a*	*1636*	151.0	25.3	107.1	16.8	76.4	38.1	169.9	17.9	240.1	104.9	171.2	10.4
ethyl 9-decenoate	33	*b,c*	*1688*	55.6	11.4	52.7	8.5	36.1	18.2	42.9	6.0	67.5	28.2	48.5	5.7
ethyl dodecanoate	34	*a*	*1855*	7.1	3.3	3.8	1.8	?		9.7	1.0	6.3	4.6	5.7	1.6
total				**1328.6**		**1273.6**		**1032.6**		**1535.3**		**1686.3**		**1548.3**	
Fatty acid ethyl esters – nitrogen metabolism *(3)*															
ethyl 2-methylbutyrate	35	*a*	*1049*	—		3.2	1.4	8.8	1.4	tr		5.1	2.5	12.9	0.9
ethyl 3-methylbutyrate	36	*a*	*1066*	2.4	0.3	8.5	2.2	22.2	1.1	1.8	0.5	11.3	0.8	28.2	1.5
ethyl benzeneacetate	37	*a*	*1782*	1.3	0.4	2.0	0.6	4.0	1.3	0.5	0.4	2.8	0.8	5.9	0.1
total				**3.7**		**13.7**		**35.0**		**2.3**		**19.2**		**47.0**	

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Esters of organic acids *(10)*														
ethyl pyruvate	38	*a*	*1267*	—		7.1	1.6	37.9	30.1	—		9.6	0.0	39.6	4.9
ethyl lactate	39	*a*	*1338*	266.3	145.9	472.3	103.5	463.8	246.8	188.4	33.8	437.7	72.9	478.6	73.0
ethyl 3-hydroxybutyrate	40	*a*	*1512*	40.3	22.7	36.2	4.2	43.1	28.7	77.1	14.7	58.4	7.9	59.3	5.7
diethyl malonate	41	*a*	*1574*	—		1.7	0.2	5.3	1.9	tr		3.1	0.2	11.6	0.8
ethyl 2-furoate	42	*a*	*1618*	0.4	0.3	2.2	0.5	6.7	2.2	1.6	1.3	4.4	1.3	11.9	0.4
diethyl succinate	43	*a*	*1672*	41.0	5.6	896.4	7.1	3760.0	1227.6	27.5	9.1	966.3	101.5	4569.4	426.1
diethyl glutarate	44	*a*	*1774*	0.5	0.2	6.2	0.2	13.1	1.4	tr		8.2	1.1	24.5	0.6
diethyl malate	45	*a*	*2037*	164.7	52.6	2477.7	292.6	11363.6	5670.8	39.1	13.0	1248.8	108.7	6162.3	479.2
diethyl tartrate	46	*a*	*2351*	—		43.2	10.4	590.3	1537.0	—		10.6	5.2	186.8	16.3
monoethyl succinate	47	*a*	*2377*	452.8	131.7	3473.8	525.2	8905.7	7612.2	105.8	81.4	3305.1	604.4	8940.2	1321.3
total				**966.0**		**7416.8**		**25189.5**		**439.5**		**6052.2**		**20484.2**	
Acetates *(7)*														
2-methylpropyl acetate	48	*a*	*1009*	32.0	17.5	11.3	6.6	2.6	1.8	40.8	13.3	16.9	12.0	4.2	3.1
butyl acetate	49	*a*	*1071*	2.3	0.5	tr		—		3.0	1.3	1.5	1.0	—	
3-methylbutyl acetate	50	*a*	*1125*	1041.4	89.3	331.3	16.4	63.5	4.6	1567.5	150.2	823.5	36.8	168.4	4.1
hexyl acetate	51	*a*	*1272*	181.4	7.3	47.1	1.4	2.4	1.9	151.5	13.9	64.5	8.8	8.6	1.3
(*Z*)-3-hexenyl acetate	52	*a*	*1307*	8.9	0.8	2.7	0.7	—		3.0	0.5	1.3	0.2	tr	
2-phenylethyl acetate	53	*a*	*1810*	249.9	7.8	93.2	8.5	16.1	0.3	325.1	7.7	189.4	10.1	41.8	2.6
tryptophyl acetate	54	*b,c*	*3369*	75.1	8.0	6.1	1.4	—		20.5	2.0	3.9	1.7	0.3	0.1
total				**1591.0**		**491.7**		**84.6**		**2111.4**		**1101.0**		**223.3**	
Monoterpenic alcohols *(8)*														
myrcenol	55	*a*	*1533*	3.9	2.1	8.0	5.6	17.9	1.9	—		—		—	
linalool	56	*a*	*1541*	143.2	11.6	68.6	5.3	2.0	0.8	39.9	3.7	78.4	5.5	12.0	1.5
4-terpineol	57	*a*	*1597*	1.0	0.4	1.0	0.1	1.4	0.5	0.9	0.2	0.8	0.4	0.9	0.6
Ho-trienol	58	*a*	*1605*	31.0	8.0	102.0	24.7	80.2	43.7	25.5	2.3	60.8	15.9	54.0	4.0
α-terpineol	59	*a*	*1691*	21.2	3.7	111.6	11.5	66.0	18.3	11.8	2.1	67.8	8.3	72.7	8.9
citronellol	60	*a*	*1760*	7.2	1.7	2.6	0.4	tr		7.3	1.4	4.0	0.8	0.7	0.2
nerol	61	*a*	*1793*	2.6	1.4	3.1	1.3	tr		1.2	0.4	5.7	2.1	?	
geraniol	62	*a*	*1847*	3.2	1.8	?		?		8.3	1.8	?		3.3	0.5
total				**213.3**		**269.9**		**167.5**		**94.9**		**217.5**		**143.6**	
Monoterpenic oxides and diols *(15)*														
trans- furan linalool oxide	63	*a*	*1436*	13.7	2.4	29.1	3.3	81.6	18.4	1.2	0.5	13.6	5.8	36.8	4.7
cis- furan linalool oxide	64	*a*	*1464*	3.2	0.9	11.5	0.1	33.1	9.6	0.6	0.0	3.6	0.9	12.7	1.4
neroloxide	65	*b,c*	*1467*	9.1	2.7	16.3	0.6	38.0	9.9	6.5	0.8	11.6	1.7	26.8	0.3
trans- pyran linalool oxide	66	*a*	*1732*	92.5	29.0	73.3	7.8	93.3	48.9	7.0	3.5	7.0	1.1	12.2	2.1
cis- pyran linalool oxide	67	*a*	*1756*	16.6	3.2	17.9	2.8	15.6	9.7	tr		0.5	0.2	tr	
exo-2-hydroxy-1,8-cineole	68	*a*	*1857*	2.0	0.9	0.9	0.5	2.1	2.2	—		—		—	
3,7-dimethylocta-1,5-dien-3,7-diol	69	*a*	*1935*	223.4	48.4	297.7	32.4	210.1	135.1	64.9	31.7	217.1	17.3	189.7	32.0
linalool hydrate	70	*a*	*1967*	3.0	1.6	47.6	5.3	63.9	21.7	0.6	1.0	15.1	5.9	41.7	0.9
terpin hydrate	71	*a*	*2087*	—		11.0	3.5	53.6	41.8	—		3.2	1.1	13.3	1.6
3,7-dimethylocta-1,7-dien-3,6-diol	72	*a*	*2121*	58.7	12.9	64.4	7.2	55.1	25.0	3.9	1.1	12.3	6.0	14.3	3.9
citronellol hydrate	73	*a*	*2196*	—		1.0	0.2	3.1	1.5	—		0.7	0.4	2.4	1.0
8-hydroxy-6,7-dihydro-linalool	74	*a*	*2197*	0.7	0.8	1.1	0.6	1.0	0.7	0.6	0.5	1.9	0.9	1.2	0.7
(*E*)-8-hydroxy-linalool	75	*a*	*2265*	?		?		tr		?		?		3.4	2.5
(*Z*)-8- hydroxy-linalool	76	*a*	*2302*	0.5	1.3	2.0	0.4	2.0	2.0	2.0	1.0	15.8	4.2	30.0	3.7
p-1-menthen-7,8-diol	77	*a*	*2517*	tr		1.4	0.6	tr		—		—		—	
total				**423.4**		**575.2**		**652.5**		**87.3**		**302.4**		**384.5**	
C_{13}-norisoprenoids *(13)*														
vitispirane I	78	*a*	*1524*	—		1.4	0.7	4.0	0.2	—		2.1	0.5	9.0	0.5
vitispirane II	79	*a*	*1527*	—		0.8	0.3	2.5	0.4	—		1.8	1.1	7.3	0.9
1,1,6-trimethyl-1,2-dihydronaphtalene	80	*b,c*	*1741*	tr		tr		0.8	0.2	—		—		0.6	0.3

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β-damascenone	81	*a*	*1816*	4.3	0.7	1.3	0.3	0.7	0.1	5.3	0.8	3.4	0.2	1.1	0.3
3-hydroxy-β-damascone	82	*a*	*2529*	1.0	0.8	tr		tr		0.9	0.1	0.7	0.2	—	
3-hydroxy-7,8-dihydro-β-ionone	83	*a*	*2533*	—		tr		—		—		—		—	
megastigm-7-ene-3,9-diol	84	*d*	*2568*	—		4.4	0.9	4.7	1.4	—		4.8	0.5	10.4	3.1
3-oxo-α-ionol	85	*a*	*2628*	4.4	2.1	7.6	1.5	7.2	3.7	3.0	1.8	7.0	0.7	10.6	2.5
3-hydroxy-7,8-dihydro-β-ionol	86	*a*	*2654*	0.3	0.2	0.6	0.3	—		—		0.8	0.5	tr	
3-oxo-7,8-dihydro-α-ionol	87	*a*	*2702*	0.7	0.5	0.5	0.0	1.9	1.4	tr		0.9	0.3	0.6	0.5
3-hydroxy-5,6-epoxy-β-ionone	88	*a*	*2721*	0.7	0.4	—		tr		—		tr		tr	
3-hydroxy-7,8-dehydro-β-ionol	89	*a*	*2742*	—		tr		—		—		0.7	0.1	0.8	0.2
vomifoliol	90	*a*	*3139*	2.0	1.1	2.4	0.3	2.4	1.0	—		tr		0.6	0.2
total				**13.4**		**19.0**		**24.2**		**9.2**		**22.2**		**41.0**	
Volatile phenols *(13)*															
methyl salicylate	91	*a*	*1770*	tr		tr		tr		tr		tr		tr	
guaiacol	92	*a*	*1852*	tr		1.2	1.5	2.8	2.4	tr		2.7	2.0	3.3	0.4
phenol	93	*a*	*2006*	1.1	0.3	1.6	0.5	1.4	1.4	1.2	0.4	1.1	0.2	2.2	1.2
4-ethylphenol	94	*a*	*2172*	1.5	0.6	0.5	0.3	1.5	1.6	1.4	0.1	1.2	0.5	0.8	0.2
4-vinylguaiacol	95	*a*	*2192*	89.3	12.0	24.5	1.3	21.1	9.3	192.7	31.7	62.9	12.9	49.4	13.0
4-vinylphenol	96	*a*	*2409*	50.3	7.4	tr		?		144.9	31.4	20.9	5.9	?	
vanillin	97	*a*	*2560*	—		—		2.9	0.7	—		tr		5.6	0.9
methyl vanillate	98	*a*	*2601*	tr		tr		tr		10.7	1.5	7.9	1.6	7.0	1.7
acetovanillone	99	*a*	*2635*	8.9	2.3	10.8	1.7	14.4	7.5	9.8	4.2	11.6	0.7	11.4	2.9
3,4-dimethoxyphenol	100	*a*	*2759*	—		—		tr		tr		0.7	0.1	1.1	0.4
2-(4'-guaiacyl)-ethanol	101	*a*	*2844*	6.1	1.9	6.7	2.5	8.2	6.1	0.8	0.3	2.5	0.2	2.1	1.0
3,4,5-trimethoxybenzyl alcohol	102	*a*	*2879*	4.3	0.5	5.1	1.8	6.4	3.0	4.3	2.2	8.0	0.8	9.8	2.2
3,4,5-trimethoxyphenol	103	*a*	*3060*	1.2	0.3	2.6	1.2	1.6	1.7	tr		1.9	0.8	2.2	0.6
total				**162.7**		**53.0**		**60.3**		**365.8**		**121.4**		**94.9**	
Volatile fatty acids – lipid metabolism *(8)*															
acetic acid	104	*a*	*1453*	16.0	7.6	9.4	4.2	19.3	13.8	16.1	4.9	16.1	6.1	18.2	7.5
butanoic acid	105	*a*	*1626*	133.1	71.0	125.3	20.1	138.3	97.4	156.7	42.5	139.2	33.5	131.5	13.5
hexanoic acid	106	*a*	*1841*	2995.1	561.1	3045.0	497.4	3716.8	1630.1	3461.6	972.9	3643.4	326.3	3331.7	179.4
(*E*)-2-hexenoic acid	107	*a*	*1964*	21.2	2.7	23.0	5.9	24.0	8.6	14.9	3.2	16.9	3.4	10.4	2.0
octanoic acid	108	*a*	*2057*	3501.4	259.4	3565.4	264.1	2902.4	298.9	3235.2	347.8	3419.2	218.5	2844.8	148.6
decanoic acid	109	*a*	*2269*	1451.7	218.8	1418.4	111.3	1080.9	339.8	1739.8	84.9	1542.7	311.6	1222.2	71.9
dodecanoic acid	110	*a*	*2481*	17.6	2.8	12.6	3.9	4.8	3.5	27.4	5.2	13.7	4.9	4.1	1.5
hexadecanoic acid	111	*a*	*2903*	tr		tr		—		1.9	2.2	tr		tr	
total				**8136.1**		**8199.1**		**7886.5**		**8653.6**		**8791.2**		**7562.9**	
Volatile fatty acids – nitrogen metabolism *(3)*															
2-methylpropanoic acid	112	*a*	*1567*	59.2	27.7	52.1	7.6	54.4	39.0	71.4	20.8	56.7	10.8	52.1	6.7
3-methylbutyric acid + 2-methylbutyric acid	113 114	*a a*	*1667*	241.2	115.0	206.9	16.7	234.3	138.7	361.1	88.6	311.7	50.4	269.7	24.5
total				**300.4**		**259.0**		**288.7**		**432.5**		**368.4**		**321.8**	
Carbonyl compounds *(4)*															
2-nonanone	115	*a*	*1386*	1.3	0.2	1.5	0.1	1.0	0.4	2.4	0.2	2.7	0.5	2.7	0.2
2-furaldehyde	116	*a*	*1460*	—		0.3	0.3	5.2	12.2	—		0.6	0.1	6.8	0.9
benzaldehyde	117	*a*	*1517*	7.4	1.9	7.1	0.7	8.9	5.8	8.3	2.0	7.9	0.8	7.4	0.1
4-ethylbenzaldehyde	118	*a*	*1703*	25.5	4.6	25.7	8.7	28.7	17.7	27.6	8.1	27.2	5.1	22.4	1.9
total				**34.2**		**34.6**		**43.8**		**38.3**		**38.4**		**39.3**	
Other *(2)*															
pantolactone	119	*a*	*2026*	3.0	0.8	6.9	2.0	10.5	9.5	2.2	1.1	4.8	1.2	7.8	2.4
N-(2-phenylethyl)-acetamide	120	*a*	*2575*	91.4	24.1	89.5	6.5	92.4	46.3	46.7	13.3	53.5	8.2	41.9	3.3
total				**94.4**		**96.4**		**102.9**		**48.9**		**58.3**		**49.7**	

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TOTAL	**102953.3**	**99468.8**	**134450.4**	**111889.6**	**108817.4**	**115556.6**
TOTAL**	**16221.9**	**21166.1**	**38585.8**	**16893.2**	**21285.3**	**33419.6**

roi, reliability of identification

RI, linear retention index on column CP-Wax 52 CB

a, identified by comparing retention time and mass spectra with those of a pure standard

b, identified by comparing retention index with published data

c, identified by comparing mass spectra with published data

d, tentative identification: molecular weight = 212 g/mol; m/z (%) = 43 (100.0), 41 (42.8), 39 (30.9), 29 (25.2), 79 (20.4), 55 (18.2), 97 (18.0), 120 (16.6), 94 (15.4), 77 (14.4); other characteristic ions, m/z (%) = 212 (int, M^+), 179 (int, M^+-H_2O-CH_3), 161 (int, M^+-$2H_2O$-CH_3)

*, levels were determined as 4-nonanol equivalents

**, without 2-methyl-1-butanol, 3-methyl-1-butanol and 2-phenylethanol

?, quantification not possible

–, not detected

tr, traces

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Table 3. Behavior during maturation, of volatile compounds found in *Loureiro* and *Alvarinho* wines

Behavior	Compound	*Loureiro*		*Alvarinho*	
		Regression equation	**R^2**	**Regression equation**	**R^2**
Increase	ethyl 2-methylbutyrate	$y=-a+bx$	0.988	$y=-a+bx$	0.991
	ethyl 3-methylbutyrate	$y=a+bx+cx^2$	0.997	$y=a+bx+cx^2$	0.999
	diethyl succinate	$y=a+bx+cx^2$	0.979	$y=a+bx+cx^2$	0.998
	neroloxide	$y=a+bx$	0.975	$y=a+bx+cx^2$	0.998
	guaiacol	$y=a+bx$	0.791	$y=-a+bx-cx^2$	0.933
Decrease	(*Z*)-3-hexen-1-ol	*(lack of correlation)*	---	$y=a-bx+cx^2$	0.822
	3-methylthio-1-propanol	*(lack of correlation)*	---	$y=a-bx$	0.912
	ethyl butyrate	*(lack of correlation)*	---	$y=a-bx$	0.677
	ethyl hexanoate	$y=a-bx-cx^2$	0.922	$y=a-bx+cx^2$	0.541
	ethyl decanoate	$y=a-bx+cx^2$	0.916	*(maximum at W_2)*	---
	3-methylbutyl acetate	$y=a-bx+cx^2$	0.998	$y=a-bx+cx^2$	0.997
	hexyl acetate	$y=a-bx+cx^2$	1.000	$y=a-bx$	0.997
	2-phenylethyl acetate	$y=a-bx+cx^2$	0.999	$y=a-bx$	1.000
	linalool	$y=a-bx+cx^2$	0.998	*(maximum at W_2)*	---
	citronellol	$y=a-bx+cx^2$	0.988	$y=a-bx$	0.987
	β-damascenone	$y=a-bx+cx^2$	0.992	$y=a-bx+cx^2$	0.991
	4-vinylguaiacol	$y=a-bx+cx^2$	0.992	$y=a-bx+cx^2$	0.989
	decanoic acid	$y=a+bx-cx^2$	0.816	$y=a-bx$	0.920
	3-methylbutanoic acid + 3-methylbutanoic acid	*(lack of correlation)*	---	$y=a-bx+cx^2$	0.779
Maximum at W_2	ethyl octanoate	$y=a+bx-cx^2$	0.671	*(lack of correlation)*	---
	ethyl decanoate	*(decreasing behaviour)*	---	$y=a+bx-cx^2$	0.723
	linalool	*(decreasing behaviour)*	---	$y=a+bx-cx^2$	0.998
	Ho-trienol	$y=a+bx-cx^2$	0.905	$y=a+bx-cx^2$	0.959
	α-terpineol	$y=a+bx-cx^2$	0.987	$y=a+bx-cx^2$	0.993
	octanoic acid	$y=a+bx-cx^2$	0.916	$y=a+bx-cx^2$	0.893

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Table 4. Sensory characterisation of *Loureiro* and *Alvarinho* wines with 8 months (W_2) and 20 months (W_3), represented by the median of each attribute and the average of the final classification (attributes ranging from 0 to 5 in order of increasing intensity)

Attribute		*Loureiro*		*Alvarinho*	
		W_2	W_3	W_2	W_3
Visual Assessment	clarity	limpid	limpid	limpid	limpid
	colour	pale citrus	citrus	open straw	open straw
	colour quality	2	3	3	3
Olfactory Assessment	intensity	3	3	4	3.5
	floral	2	2	2	1.5
	citrus fruit	3	2	1.5	1
	tree fruit	1	2	3	3
	tropical fruit	1	1.5	3	2
	dried fruit	0	0	0.5	1
	vegetal	0	1	0	1
	overall sensation	**3**	**3**	**4**	**3**
Gustative Assessment	sweetness	1	1	2	2
	acidity	3	3.5	2	2.5
	heat	2	2	4	3
	bitterness	1	2	0.5	1
	structure	3	3	3	3
	balance	2.5	3	4	3
	persistence	3	3	4	3
	overall sensation	**3**	**3**	**4**	**3.5**
Final Classification	(median)	**13**	**13**	**15.5**	**15**
	(mean)	**13.5**	**12.9**	**15.6**	**14.9**

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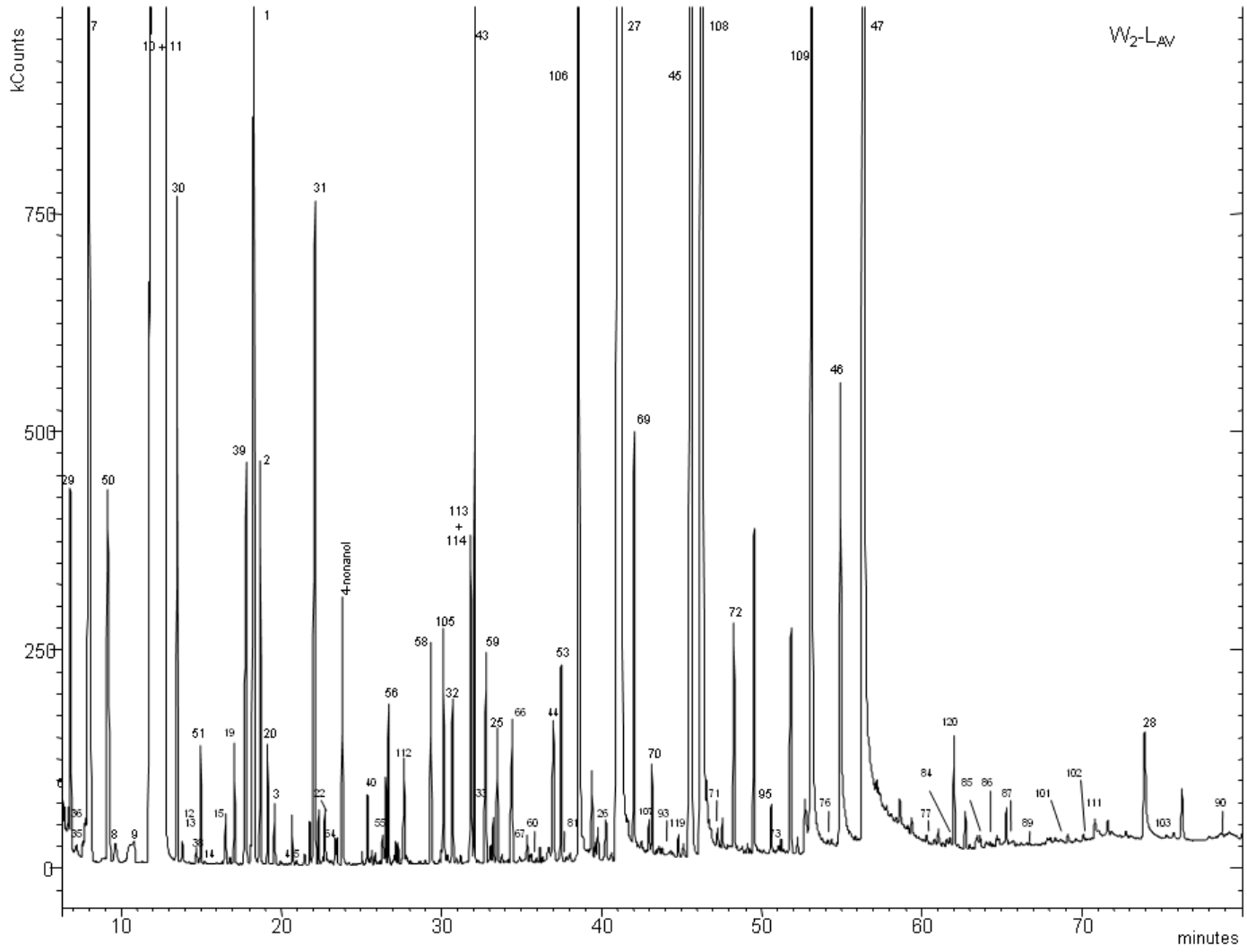

Figure 1. Representative section of a GC-MS chromatogram respecting W_2 *Loureiro* wine (peak identification may be assessed on table 2).

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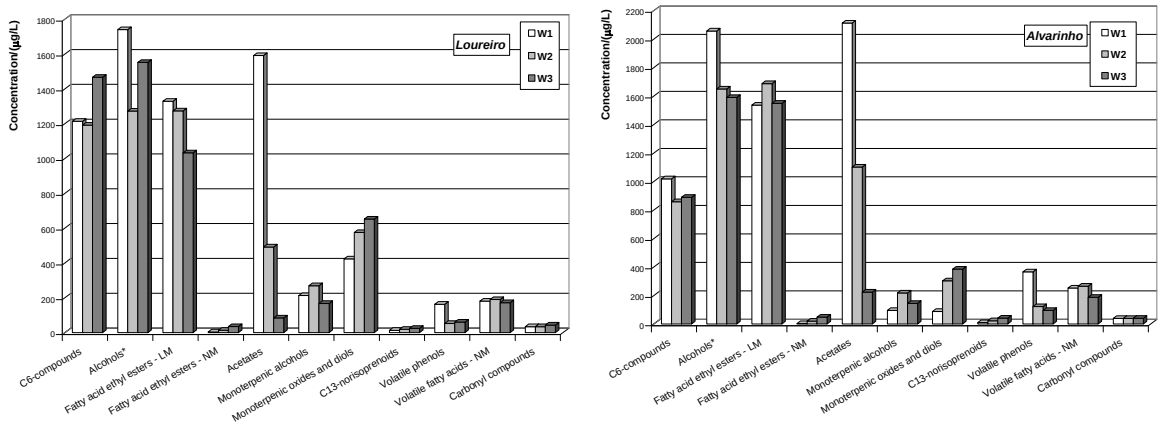

Figure 2. Evolution of each group of volatile compounds in the *Loureiro* and *Alvarinho* wines, after alcoholic fermentation (W_1) and with 8 months (W_2) and 20 months (W_3) (* without 2-methyl-1-butanol, 3-methyl-1-butanol and 2-phenylethanol; LM – lipid metabolism, NM – nitrogen metabolism).

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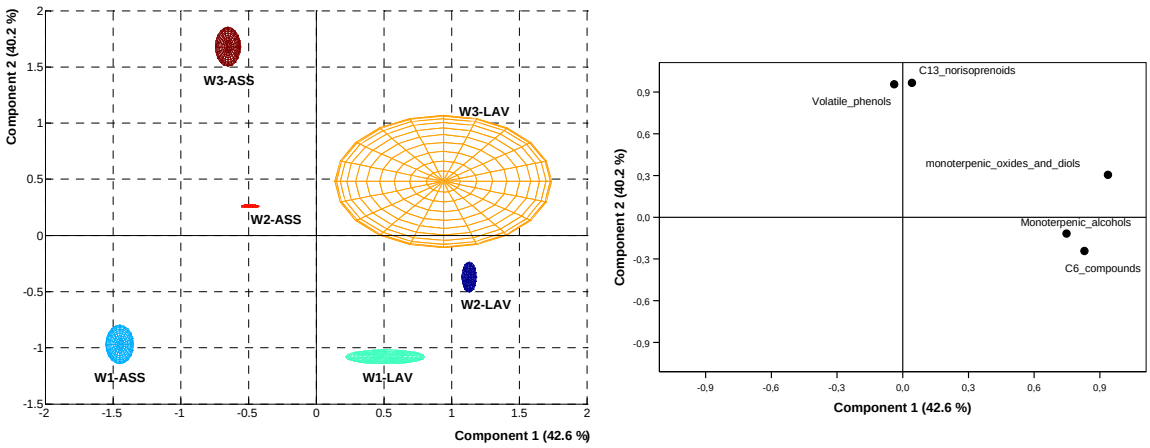

Figure 3. Graphic representation of the two principal components for the volatile varietal compounds (total levels by groups) of W_1, W_2 and W_3 wines of *Alvarinho* and *Loureiro* varieties. The ellipsoids represent the 95 % confidence level.

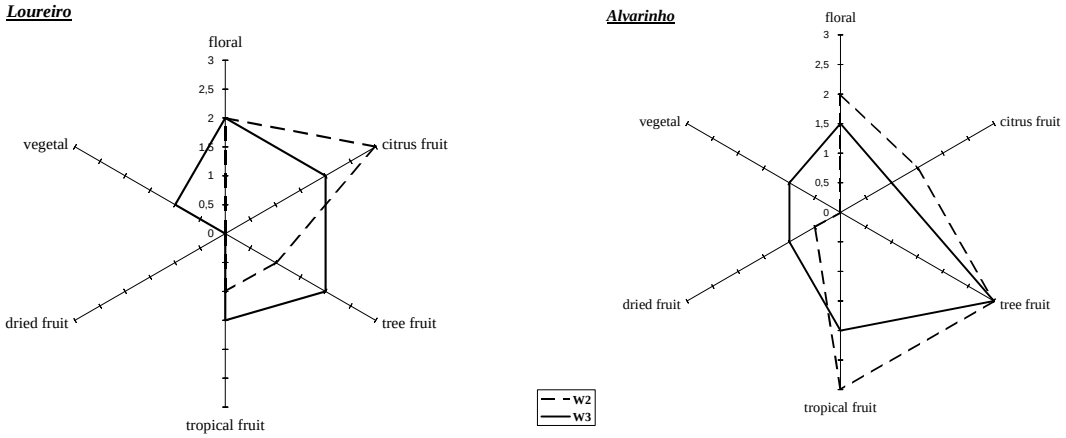

Figure 4. Aromatic descriptor intensity (median) for *Loureiro* and *Alvarinho* wines at the W_2 and W_3 stages.

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