

Manuscript Number: FOODCHEM-D-18-04217R2

Title: Effect of the winemaking process on the volatile composition and aromatic profile of Tempranillo Blanco wines

Article Type: Research Article (max 7,500 words)

Keywords: carbonic maceration, Tempranillo Blanco wine, volatile compounds, sensory properties, PLS

Corresponding Author: Dr. Belén Ayestarán Iturbe,

Corresponding Author's Institution:

First Author: Belén Ayestarán Iturbe

Order of Authors: Belén Ayestarán Iturbe; Leticia Martínez-Lapuente; Zenaida Guadalupe; Clara Canals; Elena Adell; Mar Vilanova

Abstract: The effects of the carbonic maceration and conventional winemaking on the volatile composition and aromatic sensory characteristics of Tempranillo Blanco wines were studied for the first time, during three consecutive vintages. Relationships between instrumental (volatiles) and sensory variables were analyzed applying partial least squares regression (PLS). Carbonic macerated wines had higher contents of alcohols and carbonyl compounds, yet lower concentrations of C6 alcohols and volatile acids than wines conventionally produced. The Odor Activity Values (OAV) exhibited an increase in wines when carbonic maceration was applied. According to the geometric mean (% GM) obtained from aroma descriptors the effect of the winemaking process was significant for seed fruit, ripe fruit and floral notes. When subjected to PLS the data from the instrumental analysis yielded a satisfactory model for the prediction of aroma descriptors in this set of wines.

**Effect of the winemaking process on the volatile composition and aromatic profile  
of Tempranillo Blanco wines**

Belén Ayestarán<sup>a\*</sup>, Leticia Martínez-Lapuente<sup>a</sup>, Zenaida Guadalupe<sup>a</sup>, Clara Canals<sup>b</sup>,  
Elena Adell<sup>b</sup>, Mar Vilanova<sup>c</sup>

<sup>a</sup>Instituto de Ciencias de la Vid y del Vino (Universidad de la Rioja, Gobierno de La Rioja y CSIC), Finca La Grajera, Ctra. De Burgos Km 6, 26007 Logroño (La Rioja)

<sup>b</sup>Bodegas Campo Viejo (Grupo Pernod Ricard Bodegas), C/ Viejo Camino de Lapuebla 50, 26006 Logroño (La Rioja)

<sup>c</sup>Misión Biológica de Galicia (CSIC), C/ El Palacio-Salcedo, 36143 Pontevedra (Galicia)

\*Corresponding author: Phone: +34-941-299725. Fax: +34-941-299721. E-mail:  
belen.ayestaran@unirioja.es

1 **Abstract**

2 The effects of the carbonic maceration and conventional winemaking on the volatile  
3 composition and aromatic sensory characteristics of Tempranillo Blanco wines were  
4 studied for the first time, during three consecutive vintages. Relationships between  
5 instrumental (volatiles) and sensory variables were analyzed applying partial least  
6 squares regression (PLS). Carbonic macerated wines had higher contents of alcohols  
7 and carbonyl compounds, yet lower concentrations of C<sub>6</sub> alcohols and volatile acids  
8 than wines conventionally produced. The **Odor Activity Values** (OAV) exhibited an  
9 increase in wines when carbonic maceration was applied. According to the **geometric**  
10 **mean** (% GM) obtained from aroma descriptors the effect of the winemaking process  
11 was significant for seed fruit, ripe fruit and floral notes. When subjected to PLS the data  
12 from the instrumental analysis yielded a satisfactory model for the prediction of aroma  
13 descriptors in this set of wines.

14 **Keywords:** carbonic maceration, Tempranillo Blanco wine, volatile compounds,  
15 sensory properties, PLS.

## 16 **1. Introduction**

17 Aromas are an important parameter of wine quality, especially in white wines. Wine  
18 aroma is produced by the combination of hundreds of different compounds belonging to  
19 very heterogeneous chemical groups such as esters, aldehydes, terpenes, alcohols,  
20 ketones, acids, etc. The type, amount, and presence of chemicals that affect a wine's  
21 flavor also depends on many factors, such as grape origin, grape variety and ripeness,  
22 soil and climate, yeast used during fermentation along with other winemaking practices  
23 (Kotseridis & Baumes, 2000; Spranger et al., 2004).

24 White wines are commonly elaborated with mixtures of press fractions fermented  
25 separately. Blending of the fermented press fractions can produce a more balanced  
26 wine, wherein each fraction confers to greater overall aroma complexity in the wine.  
27 Maceration of whole crushed white grapes before must extraction by pressing has  
28 become a standard procedure in some wineries. In fact, skin contact has been shown to  
29 favor the release of aromatic compounds which are located mostly in the skins  
30 (Bayonove, Cordonnier, & Ratier, 1974). However, this practice could lead to a higher  
31 extraction of phenolic compounds; thus would produce white wines which are more  
32 bitter and astringent, with strange or undesirable colors and flavors (Olejar, Fedrizzi, &  
33 Kilmartin, 2016).

34 Carbonic maceration (CM) is another winemaking technique commonly used in red  
35 wine production. However, it has been only slightly studied as a white winemaking  
36 procedure (Olejar, Fedrizzi & Killmartin, 2015; Olejar et al., 2016; Ricardo-da-Silva,  
37 Cheynier, Samsom & Bourzeix, 1993). CM refers to putting the whole grapes in a tank  
38 filled with carbon dioxide (anaerobic atmosphere) during several days in order to allow  
39 some enzymatic reactions to take place without yeast participation: production of  
40 ethanol (up to 20–30 ml/L), malic acid consumption, extraction and formation of

41 secondary metabolites, such as volatile and phenolic compounds (Etaio, Meillon, Pérez-  
42 Elortondo & Schlich, 2016; Pace, Giacosa, Torchio, Río Segade, Cagnasso & Rolle,  
43 2014). After this phase the grapes break down due to the internal pressure and the yeast  
44 begins the fermentation process. Nowadays, the CM process is not isolated since some  
45 grapes break down when they are placed in the tank, thus permitting the beginning of  
46 fermentation by yeast; therefore, both phenomena occur simultaneously (Etaio et al.,  
47 2016).

48 The volatile compounds influencing the flavor of carbonic macerated grapes can be  
49 related to a concrete biochemical pathway regarding specific amino acids (Dourtoglou,  
50 Yannovits, Tychopoulos & Vamvakias, 1994). This pathway involves characteristic  
51 must aromas that remain after the fermentation of the carbonic macerated grapes. The  
52 scientific literature describes the odor of CM red wines as “distinctive” and “rich”, and  
53 sensorial attributes often used to describe their aromas are associated with fruits (Etaio  
54 et al., 2016; Tesniere & Flanzly, 2011). The combination of grape varietal aromas,  
55 volatile compounds originating in grape anaerobiosis, and fermentative aromas could  
56 increase the aromatic quality in carbonic macerated white wines in comparison with  
57 those white wines conventionally produced.

58 A new market strategy is emerging in the wine industry in order to diversify wine  
59 production and enhance the characteristics and particularities of different grape  
60 varieties. Tempranillo Blanco *Vitis vinifera* L. var. is a minority grape variety whose  
61 production has been recently authorized by the Regulatory Certification Council of La  
62 Rioja (D.O.Ca Rioja). This grape variety is nowadays used by some D.O.Ca Rioja  
63 certified wineries to elaborate fruity wines, particularly with intense citrus and tropical  
64 fruits characteristics (Balda & Martínez de Toda, 2017).

65 However, Tempranillo Blanco wines are mostly produced by traditional winemaking;  
66 thus, there are no published studies on the chemical composition and sensory attributes  
67 of Tempranillo Blanco wines produced with CM.

68 Therefore, presented herein is the analysis of the effect of the fermentation process:  
69 carbonic maceration (CM) or conventional winemaking (CW) on the volatile  
70 composition and sensory properties of the Tempranillo Blanco white wines. The study  
71 was conducted over three consecutive vintages (2014, 2015 and 2016) in the winery  
72 Bodegas Campo Viejo (Grupo Pernod Ricard Bodegas), in the D.O.Ca Rioja region.

## 73 **2. Material and methods**

### 74 **2.1. Grapes and winemaking**

75 Grapes from Tempranillo Blanco variety were collected during three consecutive  
76 vintages (2014, 2015 and 2016) from the same vineyard of Bodegas Campo Viejo  
77 (Grupo Pernod Ricard Bodegas) from the D.O.Ca Rioja. Grapes were harvested by hand  
78 at adequate grape maturity and at optimum sanitary state; they were placed in 12 kg  
79 plastic boxes, and transported to the Bodegas Campo Viejo winery. Two winemaking  
80 procedures were then applied in triplicate: (i) conventional winemaking (CW), and (ii)  
81 carbonic maceration (CM).

82 For conventional winemaking (CW), grapes (2500 kg) were directly pressed in a  
83 pneumatic press (BucherVaslin XPro 8, France). The free run juice was quickly  
84 clarified via nitrogen flotation at room temperature and the clear juice was then moved  
85 to fermentation tank. The fermentation took place in 125 L stainless steel deposits at 16  
86 to 19 °C after inoculation with 20 g/hL of *S. cerevisiae* yeast (Zymaflore X16, Laffort,  
87 France). Once fermentation was completed (glucose plus fructose content lower than  
88 0.5 g/L), the wines were settled and sulphited (3 g/hL). Samples for analysis were taken

89 at the end of alcoholic fermentation, and were named as CW-14, CW-15 and CW-16  
90 according to the vintage they were produced, 2014, 2015 and 2016, respectively.

91 In carbonic maceration (CM), whole bunches of grapes (1000 kg) were placed into  
92 stainless steel deposits of 1500 liters, which were inerted with carbon dioxide before,  
93 during and after filling. The must from the bottom of the deposit was inoculated with 20  
94 g/hL of *Saccharomyces cerevisiae* yeast strain (Zymaflore X16, Laffort, France). The  
95 deposits were maintained at 16-19 °C and the must was pumped over 2 times a day.  
96 After 12 days of intracellular fermentation/maceration (density 1010 g/L), the mash was  
97 pressed. Free-run and press wines were combined, collected in a tank, and stored at 18  
98 °C to undergo extracellular fermentation. When alcoholic fermentation was finished  
99 (glucose plus fructose content lower than 0.5 g/L), the wine was sulphited (3 g/hL).  
100 Samples were taken at the end of alcoholic fermentation, and were named as CM-14,  
101 CM-15 and CM-16 according to the vintage they were produced, 2014, 2015 and 2016,  
102 respectively.

## 103 **2.2. Standard enological parameters**

104 Standard enological parameters were measured in the wines according to the official  
105 methods established by the International Organization of Vine and Wine (OIV, 2015).  
106 Malic acid and glucose plus fructose were analyzed by the autoanalyzer LISA 200  
107 (Biocode Hyad, Le Rhem, France).

## 108 **2.3. Wine volatile composition**

109 The volatile composition was analyzed according to Oliveira, Faria, Sá, Barros &  
110 Araújo (2006) and Moreno et al. (2017). A volume of 2.4 µg of internal standard (4-  
111 nonanol) was added to 8 mL of wine and 400 µL of dichloromethane. The extraction  
112 was done by stirring the sample with a magnetic stir bar during 15 min at room  
113 temperature. After cooling during 10 min at 0 °C, the magnetic stir bar was removed

114 and the sample was centrifugated during 5 min (RCF=5118, 4 °C). After centrifugation  
115 organic phase was recovered into a vial, using a Pasteur pipette. Then, the aromatic  
116 extract (200 µL) was picked up into a new vial after dried with anhydrous sodium  
117 sulphate. Extraction of volatiles were made in triplicate.

118 Gas chromatographic by Agilent Technologies (6890N) was used to analysis the  
119 volatile compounds. Chromatograph and an ion-trap mass spectrometer Agilent 5975C.  
120 1 µL injection was made into a capillary column CP-Wax 52 CB (50 m × 0.25 mm i.d.,  
121 0.2 µm film thickness, Chrompack). The temperature of the injector was programmed  
122 from 20 °C to 250 °C, at 180 °C min<sup>-1</sup>. The oven temperature was 40 °C, for 5 min, then  
123 programmed to rise from 40 °C to 250 °C, at 3 °C min<sup>-1</sup>, then held 20 min at 250 °C and  
124 finally programmed to go from 250 °C to 255 °C at 1 °C min<sup>-1</sup>. The carrier gas was  
125 helium N60 at 103 kPa, which corresponds to a linear speed of 180 cm s<sup>-1</sup> at 150 °C and  
126 the split vent was set to 13 mL/min. Sample of 3 µL was injected in the splitless mode  
127 (vent time 15 s). The detector was set to electronic impact mode (70 eV), with an  
128 acquisition range from 29 to 360 m/z, and an acquisition rate of 610 ms.

129 Peaks identification was performed by WSearch32 free software and by comparison  
130 with mass spectra and retention times with those of pure standard compounds and  
131 confirming these by GC-MS. All of the compounds were quantified as 4-nonanol  
132 equivalents.

#### 133 **2.4. Odor activity value**

134 To evaluate the contribution of a chemical compound to the aroma of a wine, the odor  
135 activity value (OAV) was determined. When OAV is higher than one a possible  
136 contribution to the wine aroma is considered. OAV was calculated as the ratio between  
137 the concentration and the perception threshold of the individual compound. The  
138 perception threshold used in this work were those found in the literature (Etiévant,



139 1991; Ferreira, Lopez & Cacho, 2000; Francis & Newton, 2005; Vilanova, Genisheva,  
140 Bescansa, Masa & Oliveira, 2009).

## 141 **2.5. Sensory analysis**

142 A sensory room ISO 8589 Standard (2010) was used to performed the sensory sessions.  
143 The sensory panel was formed by 19 expert tasters (11 male and 8 female between 35  
144 and 60 years old) from the D.O.Ca Rioja. In a first time, the wine tasters defined the  
145 descriptors used in the sensory analysis. One session was performed to train with  
146 descriptors using structured numerical scales according to UNE-87-020-93 Standard  
147 (ISO 4121:1987). In a second session, wine samples were analyzed with a structured  
148 numerical scale of six points where 0 represented no intensity and 5 the highest  
149 intensity). The wines were analyzed by duplicate. The olfactory attributes and defined in  
150 the first session were: aromatic intensity, seed fruit, citrus fruit, tropical fruit, ripe fruit,  
151 floral, herbaceous, mineral, and yeasty.

152 The Geometric Mean (GM %) was calculated as the square root of the product between  
153 relative intensity (I%) and relative frequency (F%). The wine descriptors were classified  
154 in basis to GM, according to the International Organization for Standardisation ISO  
155 11035 to make possible to eliminate the descriptors with low GM.

## 156 **2.6. Statistical analysis**

157 The statistical analyses were performed using XLstat-Pro (Addinsoft). One-way  
158 ANOVA analysis was made to determine the differences between treatments. Principal  
159 component analysis (PCA) was used to study the possible grouping of the wines  
160 according to the different winemaking procedures and volatile compounds. The GM (%)  
161 data obtained from the sensory analysis were submitted to a one way-ANOVA analysis.  
162 Analysis of partial least squares regression (PLS) were used to show the relationship

163 between significant aroma sensory variables with %GM > 10 and volatile compounds  
164 considered as an aroma-contributing substance (OAV > 0.2) (Belitz, & Grosch, 1999).

### 165 **3. Results and discussion**

#### 166 **3.1. Oenological parameters**

167 Alcohol content, sum of glucose plus fructose, pH, titratable acidity, volatile acidity and  
168 absorbance at 420 nm in wines after alcoholic fermentation are shown in Table 1. The  
169 values obtained after alcoholic fermentation for volatile acidity proved to be appropriate  
170 for winemaking without microbial alterations. All wines were fermented to dryness  
171 (combined glucose and fructose content < 0.5 g/L). The values of ethanol content, pH  
172 and titratable acidity were similar to those obtained in other mono varietal Spanish  
173 white wines (Vilanova et al., 2009). As expected, carbonic maceration produced wines  
174 with higher pH, volatile acidity and absorbance at 420 nm; and lower values of titratable  
175 acidity. The lower acidity and the higher pH and volatile acidity found in carbonic  
176 macerated wines was attributed to the partial metabolism of malic acid in the entire  
177 grape (Tesniere & Flanzy, 2011), which coincides with data reported by other authors  
178 (Etaio et al., 2016). As observed by Ricardo-da-Silva et al. (1993), carbonic macerated  
179 wines presented higher absorbance at 420 nm than wines conventionally produced,  
180 probably due to greater extraction of phenolic acids from the grape solids and higher  
181 browning susceptibility.

#### 182 **3.2. Effect of winemaking process on wine volatile composition**

183 Table 2 shows the individual volatile compounds and the volatile composition by  
184 families of Tempranillo Blanco wines made with conventional winemaking (CW) and  
185 carbonic maceration (CM) during the vintages 2014, 2015 and 2016. The data have  
186 been organized into six chemical families: alcohols, represented by 2-methyl-1-  
187 propanol, 3-methyl-1-butanol, 2-phenylethanol, 3-methyl-1-pentanol, and methionol;

188 C6 alcohols, represented by 1-hexanol, *E*-3-hexen-1-ol, and *Z*-3-hexen-1-ol; ethyl esters  
189 and acetates, represented by ethyl butyrate, isoamyl acetate, ethyl hexanoate, hexyl  
190 acetate, ethyl octanoate, ethyl decanoate, and 2-phenylacetate; volatile acids,  
191 represented by 2+3-methylbutanoic acid, hexanoic acid, octanoic acid, decanoic acid,  
192 geranic acid, dodecanoic acid, and hexadecanoic acid; phenol volatiles, represented by  
193 **4-vinylguaiacol**, and 4-vinylphenol; and carbonyl compounds, represented by acetoine.  
194 An ANOVA was applied to test the effect of the winemaking factor (W) on the average  
195 volatile composition of the three vintages by chemical family. Significant differences  
196 were detected between CW and CM wines for all volatile families except for phenol  
197 volatiles, ethyl esters and acetates. Some researchers also observed that carbonic  
198 maceration induced a modification of the wine volatile composition. Dourtoglou et al.  
199 (1994) concluded that red wines subjected to carbonic maceration were richer in volatile  
200 compounds than the controls after storage under the air. In the present study, carbonic  
201 maceration was found to affect various families of volatile compounds. Significantly  
202 higher concentrations of alcohols were produced in carbonic macerated wines than in  
203 wines traditionally elaborated; this was a reasonable finding being that other researchers  
204 came to the same conclusion (Bitteur, Tesniere, Sarris, Baumes, Bayonove & Flanzky,  
205 1992; Tesniere, Baumes, Bayonove & Flanzky, 1989). As in the case of alcohols, CM  
206 wines also contained higher contents of carbonyl compounds, indicating that carbonic  
207 anaerobiosis induced the augmentation when compared with traditional winemaking.  
208 On the contrary, carbonic macerated wines exhibited significantly lower concentrations  
209 of C6 alcohols, as shown by Bitteur et al., (1992) and Spranger et al., (2004) along with  
210 volatile acids. Finally, carbonic maceration did not affect the total content of volatile  
211 phenols, ethyl esters, and acetates. These results contrasted other studies which found  
212 carbonic maceration of red grapes to induce a significant increase of ester compounds

213 (Salinas, Alonso, Navarro, Pardo, Jimeno & Huerta, 1996; Tesniere et al., 1989) and  
214 volatile phenols (Ducruet, Flanzky, Bourzeix & Chambroy, 1983; Ducruet, 1984).  
215 However, a decrease in the total ester content in white wines has also been reported in  
216 macerations wherein the grape skin was involved (Aleixandre-Tudo, Weightman,  
217 Panzeri, Nieuwoudt & du Toi, 2015).

218 Results of the ANOVA on the average of the 2014, 2015 and 2016 vintages showed the  
219 effect of “winemaking” type on 56 % (14 out 25 compounds) of the volatiles identified  
220 and quantified.

221 Alcohols were the group with the highest content of volatile components both in CW  
222 and CM wines. Higher alcohols are synthesized by decarboxylation and reduction of  $\alpha$ -  
223 keto-acids produced as intermediates of amino acid synthesis and catabolism. Alcohols  
224 are related to herbaceous notes with strong and pungent tastes and smells. Contents of  
225 higher alcohols below 300 mg/L add desirable complexity to wine, whereas greater  
226 concentrations can be unfavorable to wine quality (Rapp & Versini, 1991). On the  
227 contrary, the alcohol 2-phenylethanol is generally a positive contributor to wine aroma,  
228 being characterized by a rose-like aroma (Francis & Newton, 2005). In the present  
229 study, the total content of higher alcohols in both winemaking procedures was below 40  
230 mg/L (mean total = 37.24 mg/L and 22.83 mg/L in CM and CW wines, respectively).

231 The compounds 2-methyl-1-propanol, 3-methyl-1-butanol and 2-phenylethanol were  
232 found in significantly higher concentrations when grapes were subjected to carbonic  
233 maceration. The winemaking process didn't affect the content of 3-methyl-1-pentanol  
234 and methionol. The increase of alcohols has been also reported in anaerobic metabolism  
235 of red grapes (Bitteur et al., 1992; Tesniere et al., 1989).

236 The C6 compounds, hexanols and hexenols, add herbaceous and vegetal notes to grapes  
237 and wines. This group includes aldehydes and alcohols, which are derived from

238 membrane lipids via the lipoxygenase pathway (Oliveira et al., 2006). Carbonic  
239 maceration of red wines has been related to lower levels of C6 compounds (Bitteur et  
240 al., 1992; Ducruet, 1984; Salinas et al., 1996; Spranger et al., 2004). This is not  
241 surprising since carbonic anaerobiosis limits the synthesis of these compounds by  
242 enzymatic oxidation of linolenic and linoleic acids. In the present study, 1-hexanol and  
243 *Z*-3-hexen-1-ol were affected by the winemaking process, resulting in lower  
244 concentrations in CM wines than in CW wines. No significant differences were detected  
245 in the content of *E*-3-hexen-1-ol.

246 Ethyl esters have a strong influence on wine flavor because they are normally found in  
247 high concentrations and have low detection thresholds; thus they play an essential role  
248 in the fruity aromas of wines. Ethyl esters content depends on different factors, such as  
249 sugar content, fermentation temperature, aeration, and yeast strain (Perestrelo,  
250 Fernandes, Albuquerque, Marques & Câmara, 2006). In the present study, CW and CM  
251 wines showed the same profile of ethyl esters of fatty acids. Ethyl hexanoate and  
252 octanoate were the major ethyl esters of fatty acids, whereas ethyl decanoate was found  
253 in the lowest concentration in both wines. Carbonic maceration induced a significant  
254 increase of ethyl butyrate, but a significant decrease in the content of ethyl hexanoate  
255 and ethyl decanoate.

256 The formation of acetates depends on must nutrient concentration (Gambetta, Bastian,  
257 Cozzolino & Jeffery, 2014), the content of unsaturated fatty acids available in the  
258 medium, and carbon-to-nitrogen ratio (Saerens, Delvaux, Verstrepen, Van Dijck,  
259 Thevelein & Delvaux, 2008). Volatile acetates are among the key compounds in the  
260 fruity flavor of wines (Vilanova et al., 2009). Three acetates were identified in the  
261 present study: hexyl acetate, isoamyl acetate and 2-phenylethylacetate. Only hexyl  
262 acetate, which supplies pleasant “apple” nuances to the wines (Etiévant, 1991), varied

263 with the winemaking treatment; the highest value was produced in the conventional  
264 winemaking process.

265 Fatty acids are produced in the lipid metabolism of yeast; thus, have been related with  
266 fatty, cheese and rancid attributes (Rocha, Rodrigues, Coutinho, Delgadillo & Coimbra,  
267 2004). Among the fatty acids are decanoic acid, octanoic acid and hexanoic acid which  
268 were present at high concentrations in both wines. 2+3-methylbutanoic acid, octanoic  
269 acid, decanoic acid and geranic acid concentrations were significantly lower when  
270 carbonic maceration was applied.

271 Among the group of vinylphenols, only 4-vinylguaiacol and 4-vinylphenol were  
272 detected. This result was logical being that vinylphenols are the main phenols in white  
273 wines, while ethylphenols are more abundant in red wines (Boidron, Chatonnet & Pons,  
274 1988). These compounds are related with heavy pharmaceutical odors (Castro Mejías,  
275 Natera Marín, García Moreno & García Barroso, 2003). The winemaking procedure  
276 didn't affect the content of volatile phenols in wines.

277 Regarding carbonyl compounds, carbonic maceration induced an increase in the acetoin  
278 concentration.

### 279 **3.3. Principal component analysis (PCA) applied on wine volatile composition**

280 Principal component analysis (PCA) was applied on Tempranillo Blanco wine volatile  
281 compounds with significant differences among winemaking treatments (Figure 2).

282 The first two principal components (PC1 and PC2) explained 74.76 % of the total  
283 variance (50.91 % and 23.85 %, respectively). PC1 was characterized by the major  
284 contribution of 2-methyl-1-propanol, 3-methyl-1-butanol, 2-phenylethanol and ethyl  
285 butyrate on the positive loading, and hexyl acetate, 2+3-methylbutanoic acid, octanoic  
286 acid, and decanoic acid on the negative loading. PC2 was characterized by ethyl

287 hexanoate and ethyl decanoate in the positive side and 1-hexanol, geranic acid and Z-3-  
288 hexen-1-ol in the negative side.

289 PCA differentiated two groups. The first group, formed by carbonic macerated wines  
290 (CM) from 2014, 2015 and 2016 vintages, was located in the positive side of PC1. The  
291 second group, formed by wines made by conventional winemaking (CW) from 2014,  
292 2015 and 2016 vintages, was positioned in the negative side of PC1. CM wines from  
293 2014 and 2015 vintages were related to high contents of ethyl butyrate and 2-  
294 phenylethanol. However, CM wines from 2016 vintage were strongly associated with  
295 alcohols (2-methyl-1-propanol and 3-methyl-1-butanol). CW wines produced from the  
296 2014 vintage were strongly related to high ethyl esters (ethyl hexanoate and decanoate)  
297 and acetate (hexyl acetate) contents. Finally, the presence of high contents of volatile  
298 acids (2+3-methylbutanoic acid, octanoic acid, decanoic acid and geranic acid) and C6  
299 compounds (1-hexanol and Z-3-hexen-1-ol) contributed to the distinction of CM wines  
300 from 2015 and 2016 vintages.

### 301 **3.4. Odor activity values of Tempranillo Blanco wines**

302 The results in Table 3 illustrate how that 12 out of 25 quantified volatile compounds (48  
303 % of total compounds) were found at average concentrations higher than their  
304 corresponding odor thresholds (OAV >1). These volatiles are considered to be potential  
305 contributors to the global bouquet of the wines analyzed; although their sensory impact  
306 is likely to be affected by the actual wine matrix (Pineau, Barbe, Van Leeuwen &  
307 Dubourdieu, 2007).

308 Tempranillo Blanco wines from carbonic maceration were the most aromatic wines,  
309 with a total OAV of 822, versus an OAV of 770 from CW wines. Four ethyl esters  
310 (fruity aroma), which contribute favorably to white wine aroma as fruity characteristics  
311 (Vilanova et al., 2009), were found in levels over the perception threshold in both the

312 CM and CW wines. Isoamyl acetate and ethyl octanoate, related to banana and apple  
313 aroma respectively, were the most powerful odorants in wines regardless of the  
314 vinification method. Carbonic macerated wines contained the highest OAV of isoamyl  
315 acetate (367), while ethyl octanoate was highest in wines elaborated by traditional  
316 winemaking (353). 2-phenylacetate, with floral aroma, was also detected above its odor  
317 threshold for all wines, with similar values from both winemaking procedures. Among  
318 alcohols, 3-methyl-1-butanol (alcohol and banana notes) was the only alcohol with  
319 OAV > 1, and made a major contribution in wines from carbonic maceration.

320 The presence of C6–C10 fatty acids, generally associated with the occurrence of  
321 negative odors, has been associated with notes described as fruity, cheese-like and  
322 rancid (Rocha et al., 2004). However, they could have a great impact on the aromatic  
323 balance of wines because they oppose the hydrolysis of the corresponding esters  
324 (Edwards, Beelman, Bartley & McConnell, 1990). In the present study, 2+3  
325 methylbutanoic acid, hexanoic, octanoic and decanoic acids showed a high contribution  
326 to wine aroma in traditional white wines (CW), although their influence to the wine  
327 aroma was low in comparison with other compounds. Only the volatile phenol 4-  
328 vinylguaiacol, associated with clove and curry notes, presented OAV > 1 in all wines.  
329 This compound showed a maximum OAV value in wines subjected to conventional  
330 winemaking (CW). The OAV of ethyl esters and acetates illustrated an increase in fruity  
331 odors in Tempranillo Blanco wines when carbonic maceration was applied. An increase  
332 in fruity notes together with a decrease in fatty acids with rancid aromas would  
333 contribute to the improvement of the overall aroma in carbonic macerated wines.

### 334 **3.5. Aromatic sensory properties**

335 Table 4 shows the geometric mean (GM %) of the aromatic sensory descriptors of  
336 Tempranillo Blanco wines, together with the ANOVA results for the winemaking factor



337 (W). The aromatic sensory characterization of the Tempranillo Blanco wine was  
338 obtained by an experienced panel using a total of nine aromatic descriptors (Table 4).  
339 According to the ANOVA analysis, the effect of the winemaking procedure was  
340 significant ( $p < 0.05$ ) for the seed fruit, ripe fruit and floral notes. Ripe fruit presented  
341 maxima GM (%) for carbonic macerated wines (CM) in all vintages studied, whereas  
342 seed fruit and floral notes exhibited the highest GM (%) for wines made with  
343 conventional winemaking (CW). This indicated that, from the nine descriptors  
344 evaluated, these three notes were useful to differentiate between Tempranillo Blanco  
345 wines. Apart from the aromatic intensity, the maxima GM (%) values for both  
346 winemaking processes were given to the seed fruit, citrus, tropical, floral and ripe fruit  
347 notes (GM >30%). All sensory attributes evaluated showed GM >10 % in both wines  
348 and therefore, could be used to define the aroma of Tempranillo Blanco wines, as  
349 defined by Vilanova, Genisheva, Masa & Oliveira (2010).

### 350 **3.6. PLS modeling relationship between aromatic sensory descriptors and volatile** 351 **compounds of wines**

352 Partial least squares (PLS) regression was used to show the relationships between the  
353 sensory attributes and volatile compounds of the wines. Figure 3 contains the PLS  
354 carried out between aroma descriptors with GM > 10 % (aromatic intensity, seed fruit,  
355 citrus, tropical, ripe fruit, floral, herbaceous, mineral, and yeasty notes), and volatiles  
356 considered as an aroma-contributing substance with OAV > 0.2 (V1-V20).

357 The data were standardized before to PLS analysis for easier identification of the  
358 relationships between volatiles and sensory variables. The plot (explaining 62.57 % of  
359 the total variance) suggested a correlation of volatile compounds and sensory  
360 descriptors and their association with the wines analyzed. The PLS discriminated  
361 between the winemaking processes. CM wines were positioned in the positive side of

362 PC1 and CW wines in the negative side of the same axe. In both elaborations wines  
363 from 2015 were different than the other vintages.

364 Aromatic intensity and ripe fruit descriptors were highly correlated and were associated  
365 to CM-14 and CM-16 wines. According to the loading weight, these attributes were  
366 mainly predicted by 3-methyl-1-butanol (V1), 2-phenylethanol (V2), and ethyl butyrate  
367 (V6). The perception of wine fruitiness is highly dependent on some esters (Ferreira,  
368 Fernandez, Pena, Escudero & Cacho, 1995). Some authors have suggested that higher  
369 alcohols give wine a pungent aroma (Rapp & Versini, 1991) or a solvent aroma nuance  
370 (Cameleyre, Lytra, Tempere & Barbe, 2015). On the other hand, some authors suggest  
371 that 2-phenylethanol present a positive correlation with aromatic intensity in red wines  
372 (Vilanova, Campo, Escudero, Graña, Masa & Cacho, 2012).

373 Figure 3 showed positive correlations between floral descriptor and hexanoic (V14) and  
374 decanoic acids (V15). The citrus, tropical and seed fruit attributes were highly  
375 correlated with hexyl acetate (V9), ethyl decanoate (V11) and hexanoic acid (V14).  
376 Floral, citrus, tropical and seed fruit notes were mainly associated to CW wines from  
377 2014 and 2016 vintages. The herbaceous attribute was highly correlated to E-3-hexen-1-  
378 ol (V4), ethyl butyrate (V6) and 2-phenylacetate (V12). CM wines had strong  
379 herbaceous notes; probably due to the presence of stems during the fermentation  
380 process. CW wines from 2015 vintage showed the highest mineral aroma. This  
381 descriptor was satisfactorily predicted by 4-vinylguaiacol (V19) and 4-vinylphenol  
382 (V20). These results were in agreement with other authors who reported volatile  
383 phenols to be related to wine aromatic minerality (Zaldivar, 2017). Finally, CM wines  
384 from 2014 and 2016 vintages presented the highest correlation with a yeasty aroma and  
385 4-vinylphenol (V20).

#### 386 **4. Conclusions**

387 The effect of the winemaking process (carbonic maceration *vs* conventional  
388 winemaking) on the volatile composition and aromatic sensory properties of  
389 Tempranillo Blanco wines was studied during three consecutive vintages. Carbonic  
390 maceration led to wines of higher pH, volatile acidity and lower titratable acidity,  
391 whereas ethanol content remained unaffected. A significant change in the volatile  
392 profile of wines was also observed when carbonic maceration was applied. Carbonic  
393 maceration produced wines with significantly higher contents of alcohols and carbonyl  
394 compounds, and lower concentrations of C<sub>6</sub> alcohols and volatile acids. However, total  
395 content of ethyl esters and acetates and volatile phenols in wines were not affected. The  
396 effect of the winemaking process also had an influence on the odor activity value  
397 (OAV), where twelve volatile compounds reached values > 1; accounting for the  
398 highest total OAV from carbonic macerated wines. The OAV analysis from ethyl esters  
399 and acetates showed an increase of fruity odor on Tempranillo Blanco wines when  
400 carbonic maceration was applied. In the sensory analysis, the ripe fruit descriptor  
401 exhibited maxima geometric mean (GM %) for carbonic macerated wines, whereas the  
402 seed fruit and floral notes were exhibited to be higher in conventionally produced  
403 wines. Partial least square (PLS) regression applied to aroma active compounds and  
404 aroma sensory descriptors showed how carbonic macerated wines presented higher  
405 aromatic intensity and ripe fruit descriptors. Citrus, tropical and seed fruit attributes  
406 were associated to wines conventionally made.

407 The results obtained suggest carbonic maceration in Tempranillo Blanco wines to be an  
408 alternative to the traditional winemaking to obtain wines with intense ripe fruit features.  
409 Therefore, it would increase the diversification and differentiation of white wines.  
410 However, further studies need to be developed aiming at the evaluation of the effect of  
411 carbonic maceration on the other wine chemical compounds.

## 5. References

- Aleixandre-Tudo, J.L., Weightman, C., Panzeri, V., Nieuwoudt, H.H., & du Toi, W.J. (2015). Effect of skin contact before and during alcoholic fermentation on the chemical and sensory profile of South African Chenin Blanc white wines. *South African Journal of Enology and Viticulture*, 36, 366–377.
- Balda, P., & Martínez de Toda, F. (2017). *Varietades minoritarias de vid en La Rioja*. (1th ed.). Logroño: Consejería de Agricultura, Ganadería y Medio Ambiente.
- Bayonove, C., Cordonnier, R., & Ratier, R. (1974). Localisation de l'arome dans la baie de raisin: variétés Muscat d'Alexandrie et Cabernet-Sauvignon. *Comptes rendus de l'Académie d'Agriculture de France*, 60, 1321-1328.
- Belitz, H. D., & Grosch, W. (1999). *Food chemistry*. (2nd ed.). Berlin: Springer-Verlag.
- Bitteur, S., Tesniere, C., Sarris, J., Baumes, R., Bayonove, C., & Flanzly, C. (1992). Carbonic anaerobiosis of muscat grapes. I. Changes in the profiles of free and bound volatiles. *American Journal of Enology and Viticulture*, 43, 41-48.
- Boidron, J. N., Chatonnet, P., & Pons, M. (1988). Influence du bois sur certaines substances odorantes des vins. *Connaissance Vigne Vin*, 22, 275–293.
- Cameleyre, M., Lytra, G., Tempere, S., & Barbe, J. C. (2015). Olfactory impact of higher alcohols on red wine fruity ester aroma expression in model solution. *Journal of Agricultural and Food Chemistry*, 63, 9777–978.
- Castro Mejías, R., Natera Marín, R., García Moreno, M. V., & García Barroso, C. (2003). Optimisation of headspace solid-phase microextraction for the analysis of volatile phenols in wine. *Journal of Chromatography A*, 995, 11–20.
- Dourtoglou, V.G., Yannovits, N. G., Tychopoulos, V. G., & Vamvakias, M. M. (1994). Effect of storage under CO<sub>2</sub> atmosphere on the volatile, amino acid, and pigment

- constituents in red grape (*Vitis vinifera* L. Var. Agiorgitiko). *Journal of Agricultural and Food Chemistry*, 42, 338-344.
- Ducruet, V. (1984). Comparison of the headspace volatiles of carbonic maceration and traditional wine. *LWT - Food Science and Technology*, 17, 217-221.
- Ducruet, V., Flanzky, C., Bourzeix, M., & Chambroy, Y. (1983). Les constituants volatils des vins jeunes de macération carbonique. *Sciences des Aliments*, 3, 413-426.
- Edwards, C.G., Beelman, R.B., Bartley, C.E., & McConnell, A.L. (1990). Production of decanoic acid and other volatile compounds and the growth of yeast and malolactic bacteria during vinification. *American Journal of Enology and Viticulture*, 41, 48-56.
- Etaio, I., Meillon, S., Pérez-Elortondo, F. J., & Schlich, P. (2016). Dynamic sensory description of Rioja Alavesa red wines made by different winemaking practices by using Temporal Dominance of Sensations. *Journal of the Science of Food and Agriculture*, 96, 3492-3499.
- Etiévant, P. X. (1991). Wine. In H. Maarse (Ed.), *Volatile compounds in food and beverages* (pp. 483-546). New York: Marcel Dekker Inc.
- Ferreira, V., Fernandez, P., Pena, 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*, 67, 381-392.
- Ferreira, V., Lopez, R., & Cacho, J.F. (2000). Quantitative determination of the odorants of young red wines from different grape varieties. *Journal of the Science of Food and Agriculture*, 80, 1659-1667.

- Francis, I.L., & Newton, L. (2005). Determining wine aroma from compositional data. *Australian Journal of Grape and Wine Research*, 11, 114–126.
- Gambetta, J.M., Bastian, S.E.P., Cozzolino, D., & Jeffery, D.W. (2014). Factors influencing the aroma composition of Chardonnay wines. *Journal of Agricultural and Food Chemistry*, 62, 6512–6534.
- 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*, 48, 400–406.
- Moreno, D., Valdés, E., Uriarte, D., Gamero, E., Talaverano, I., & Vilanova, M. (2017). Early leaf removal applied in warm climatic conditions: Impact on Tempranillo wine volatiles. *Food Research International*, 98, 50–58.
- OIV (2015). *Compendium of international methods of wine and must analysis*. Paris: International Organisation of Vine and Wine.
- Olejar, K. J., Fedrizzi, B., & Kilmartin, P. A. (2016) Enhancement of Chardonnay antioxidant activity and sensory perception through maceration technique. *LWT Food Science and Technology*, 65, 152-157.
- Olejar, K.J., Fedrizzi, B., & Killmartin, P.A. (2015). Antioxidant activity and phenolic profiles of Sauvignon Blanc wines made by various maceration techniques. *Australian Journal of Grape and Wine Research*, 21, 57-68.
- Oliveira, J.M., Faria, M., Sá, F., Barros, F., & Araújo, I.M. (2006). C6-alcohols as varietal markers for assessment of wine origin. *Analytica Chimica Acta*, 563, 300–309.
- Pace, C., Giacosa, S., Torchio, F., Río Segade, S., Cagnasso, E., & Rolle, L. (2014). Extraction kinetics of anthocyanins from skin to pulp during carbonic

- maceration of wine grape berries with different ripeness levels. *Food Chemistry*, *165*, 77–84.
- Perestrelo, R., Fernandes, A., Albuquerque, F.F., Marques, J.C., & Câmara, J.S. (2006). Analytical characterization of the aroma of Tinta Negra Mole red wine: identification of the main odorants compounds, *Analytica Chimica Acta*, *563*, 154–164.
- 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*, *55*, 4103–4108.
- Rapp, A., & Versini, G. (1991). Influence of nitrogen compounds in grapes on aroma compounds of wine. In *International Symposium on Nitrogen in Grapes and Wine* (pp. 156–164). Davis: American Society for Enology and Viticulture.
- Ricardo-da-Silva, J. M., Cheynier, V., Samsom, A., & Bourzeix, M. (1993). Effect of pomace contact, carbonic maceration, and hyperoxidation on the procyanidin composition of Grenache blanc wines. *American Journal of Enology and Viticulture*, *44*, 168-172.
- 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 odorants. *Analytica Chimica Acta*, *513*, 257–262.
- Saerens, S. M. G., Delvaux, F., Verstrepen, K. J., Van Dijck, P., Thevelein, J. M., & Delvaux, F. R. (2008). Parameters affecting ethyl ester production by *Saccharomyces cerevisiae* during fermentation, *Applied and Environmental Microbiology*, *74*, 454–561.
- Salinas, M. R., Alonso, G. L., Navarro, G., Pardo, F., Jimeno, J., & Huerta, M. D. (1996). Evolution of the aromatic composition of wines undergoing carbonic

- maceration under different aging conditions. *American Journal of Enology and Viticulture*, 47, 134-144.
- Spranger, M. I., Clímaco, M. C., Sun, B., Eiriz, N., Fortunato, C., Nunes, A., et al. (2004). Differentiation of red winemaking technologies by phenolic and volatile composition. *Analytica Chimica Acta*, 513, 151–161.
- Tesniere, C., & Flanzy, C. (2011). Carbonic maceration wines: characteristics and winemaking process. *Advances in Food and Nutrition Research*, 63, 1–15.
- Tesniere, C., Baumes, R., Bayonove, C., & Flanzy, C. (1989). Effect of simulated alcoholic fermentation on aroma components of grape berries during anaerobic metabolism. *American Journal of Enology and Viticulture*, 40, 183-188.
- Vilanova, M., Campo, E., Escudero, A., Graña, M. Masa, A., & Cacho, J. (2012). Volatile composition and sensory properties of *Vitis vinifera* red cultivars from North West Spain: Correlation between sensory and instrumental analysis. *Analytica Chimica Acta*, 720, 104–111.
- Vilanova, M., Genisheva, Z., Bescansa, L., Masa, A., & Oliveira, J. M. (2009). Volatile composition of wines from cvs. Blanco lexítimo, Agudelo and Serradelo (*Vitis vinifera*) grown in Betanzos (NW Spain). *Journal of the Institute of Brewing*, 115, 35-40.
- Vilanova, M., Genisheva, Z., Masa, A., & Oliveira, J.M. (2010). Correlation between volatile composition and sensory properties in Spanish Albariño wine. *Microchemical Journal*, 95, 240-246
- Zaldivar, E. (2017). *Caracterización químico-sensorial en vinos blancos y tintos del atributo mineralidad*. Tesis doctoral. Logroño: Servicio de Publicaciones, Universidad de La Rioja.



## Figure captions

**Figure 1.** Principal components analysis (PCA) on volatile compounds with significant differences among conventional winemaking (CW) and carbonic maceration (CM) during 2014, 2015 and 2016 vintages.

**Figure 2.** Partial least squares regression (PLS) product space volatile compounds with OAV > 0.2 and aroma descriptors on Tempranillo Blanco wines produced by conventional winemaking (CW) and carbonic maceration (CM) during 2014, 2015 and 2016 vintages. Aroma descriptors: aromatic intensity, seed fruit, citrus fruit, tropical fruit, ripe fruit, floral, herbaceous, mineral, and yeasty. Volatile compounds: V1, 3-methyl-1-butanol; V2, 2-phenylethanol; V3, 3-methyl-1-pentanol; V4, E-3-hexen-1-ol; V5, Z-3-hexen-1-ol; V6, ethyl butyrate; V7, isoamyl acetate; V8, ethyl hexanoate; V9, hexyl acetate; V10, ethyl octanoate; V11, ethyl decanoate; V12, 2-phenylacetate; V13, 2+3-methylbutanoic acid; V14, hexanoic acid; V15, octanoic acid; V16, decanoic acid; V17, dodecanoic acid; V18, hexadecanoic acid; V19, 4-vinylguaiacol; V20, 4-vinylphenol.

## Tables

**Table 1.** Standard enological parameters of Tempranillo Blanco wines produced by conventional winemaking (CW) and carbonic maceration (CM) during 2014, 2015 and 2016 vintages and significance of winemaking (W) factor according to one-way ANOVA.

	CW-14	CM-14	CW-15	CM-15	CW-16	CM-16	Mean		Anova
							CW	CM	W
Ethanol <sup>1</sup>	13.10	12.90	13.50	13.90	13.40	13.10	13.33	13.30	ns
G+F <sup>2</sup>	0.13	0.01	0.04	0.01	0.04	0.04	0.07	0.02	**
pH	3.25	3.32	3.01	3.15	3.20	3.39	3.16	3.29	*
TA <sup>3</sup>	6.45	6.12	8.20	7.15	7.30	6.54	7.32	6.60	*
VA <sup>4</sup>	0.14	0.18	0.22	0.37	0.21	0.28	0.19	0.28	*
A420 <sup>5</sup>	0.14	0.28	0.12	0.21	0.15	0.26	0.14	0.25	***

<sup>1</sup>Ethanol: mL ethanol for 100 mL of wine at 20°C; <sup>2</sup>G+F: sum of glucose plus fructose as g/L; <sup>3</sup>VA: volatile acidity as g acetic acid/L; <sup>4</sup>A420: absorbance at 420 nm.

Level of significance: \*, \*\* and \*\*\* indicates significance at  $p < 0.05$ ,  $p < 0.01$  and  $p < 0.001$  respectively; ns indicates no significant difference.

**Table 2.** Concentration ( $\mu\text{g/L}$ ) of volatile compounds and volatile families of Tempranillo Blanco wines produced by conventional winemaking (CW) and carbonic maceration (CM) during 2014, 2015 and 2016 vintages and significance of winemaking (W) factor according to one-way ANOVA.

Volatile compounds	CW-14	CM-14	CW-15	CM-15	CW-16	CM-16	Mean		Anova
							CW	CM	W
2-Methyl-1-propanol	360	888	581	1170	603	1117	515	1058	***
3-Methyl-1-butanol	11864	18735	11200	29720	23103	27718	15389	25391	***
2-Phenylethanol	5359	9664	7304	14096	7442	8355	6702	10705	**
3-Methyl-1-pentanol	69.1	21.5	248	46.8	nd	nd	159	34.2	ns
Methionol	28.9	14.7	99.5	66.3	37.5	70.7	55.3	50.6	ns
<i>Total alcohols</i>	17681	29323	19432	45099	31186	37260	22819	37239	***
1-Hexanol	280	177	293	148	651	375	408	233	***
<i>E</i> -3-Hexen-1-ol	11.5	7.7	284	nd	nd	nd	148	7.7	ns
<i>Z</i> -3-Hexen-1-ol	68.1	44.2	156	47.6	nd	nd	112	45.9	*
<i>Total C6 alcohols</i>	360	229	733	195	651	375	668	287	**
Ethyl butyrate	483	293	nd	2428	440	664	461	1128	**
Isoamyl acetate	9449	6004	4337	19306	11769	7731	8519	11014	ns
Ethyl hexanoate	1483	799	722	1027	836	581	1014	802	*
Hexyl acetate	619	52.5	nd	85.9	333	110	476	82.9	***
Ethyl octanoate	2792	1816	475	1990	2029	1010	1766	1605	ns
Ethyl decanoate	796	276	377	301	606	117	593	232	***
2-Phenylacetate	721	409	591	1142	704	352	672	634	ns
<i>Total ethyl esters and acetates</i>	16343	9650	6502	26280	16717	10565	13499	15498	ns
2+3-Methylbutanoic acid	nd	nd	220	59.9	nd	nd	200	59.9	**
Hexanoic acid	1798	846	2122	1298	1941	1143	1954	1096	ns
Octanoic acid	5308	2235	6840	2931	7739	3457	6629	2874	*
Decanoic acid	1820	1248	2885	806	2468	797	2391	950	**
Geranic acid	109	148	nd	286	761	597	435	343	**
Dodecanoic acid	171	114	366	94.2	102	74.9	213	94.2	ns
Hexadecanoic acid	123	166	318	225	117	156	186	183	ns
<i>Total volatile acids</i>	9328	4757	12751	5701	13128	6224	12027	5600	*
4-Vinylguaiacol	23.6	55.3	235	48.4	26.0	71.5	94.7	58.4	ns
4-Vinylphenol	13.2	210	368	77.9	78.8	128	153	139	ns
<i>Total phenol volatiles</i>	36.8	265	602	126	105	200	248	197	ns
Acetoine	26.2	198	nd	84.5	nd	nd	26.2	160	***

Level of significance: \*, \*\* and \*\*\* indicates significance at  $p < 0.05$ ,  $p < 0.01$  and  $p < 0.001$  respectively; ns indicates no significant difference. nd: non detected

**Table 3.** Odor activity values of compounds reaching a concentration above the odor threshold (OAV > 1) in Tempranillo Blanco wines produced by conventional winemaking (CW) and carbonic maceration (CM).

Volatile compounds	Odor descriptor	Odor threshold	Reference	CW	CM
3-Methyl-1-butanol	Alcohol, Banana	7000	3	2	4
Ethyl butyrate	Papaya, butter, sweetish, apple	20	4	23	56
Isoamyl acetate	Banana	30	2	284	367
Ethyl hexanoate	Apple, fruity, sweetish	14	4	72	57
Ethyl octanoate	Apple, fruity, sweetish	5	4	353	321
Ethyl decanoate	Fruity, Strawberry	200	3	3	1
2-Phenylacetate	Floral	250	2	3	3
2+3-Methylbutanoic	Cheese, sweet	33	3	7	2
Hexanoic acid	Goaty, fatty acid, vegetable oil	420	1	5	3
Octanoic acid	Goaty, fatty acid, vegetable oil	500	1	13	6
Decanoic acid	Waxy, tallowy, rancid, soapy	1000	1	2	1
4-Vinylguaiacol	Clove, Curry	40	1	2	1

[1]: Ferreira et al. (2000); [2]: Francis, & Newton (2005); [3]: Etiévant (1991); [4] Vilanova et al. (2009)

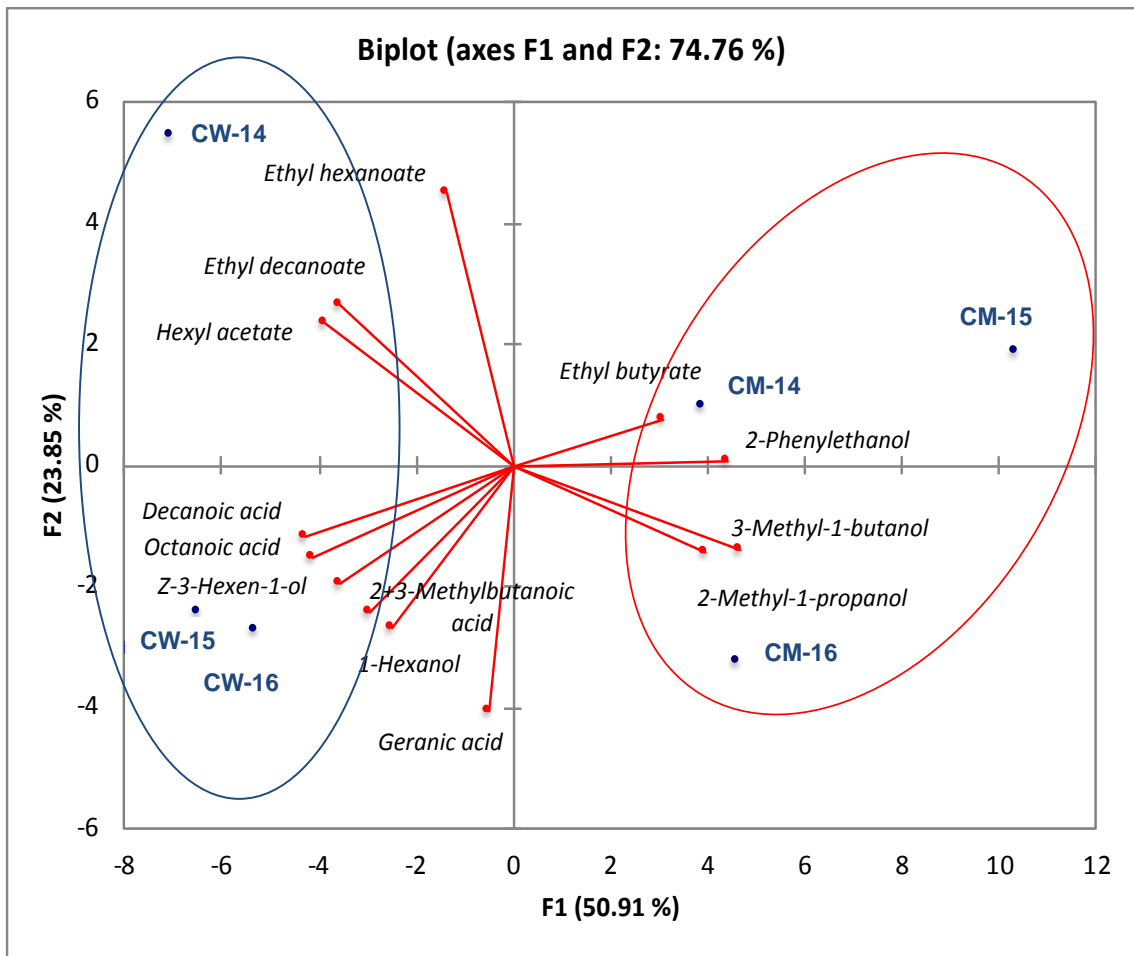
**Table 4.** Geometric mean (% GM) of aromatic sensory descriptors of Tempranillo Blanco wines produced by conventional winemaking (CW) and carbonic maceration (CM) during 2014, 2015 and 2016 vintages and significance of winemaking (W) factor according to one-way ANOVA.

	CW-14	CM-14	CW-15	CM-15	CW-16	CM-16	Mean		Anova
							CW	CM	W
Aromatic intensity	<b>83</b>	<b>83</b>	<b>73</b>	<b>88</b>	<b>63</b>	<b>100</b>	<b>76</b>	<b>90</b>	ns
Seed fruit	<b>88</b>	<b>35</b>	<b>71</b>	<b>64</b>	<b>77</b>	<b>36</b>	<b>79</b>	<b>45</b>	*
Citrus	<b>57</b>	<b>31</b>	<b>55</b>	<b>44</b>	<b>42</b>	<b>18</b>	<b>51</b>	<b>31</b>	ns
Tropical	<b>74</b>	<b>57</b>	<b>60</b>	<b>52</b>	<b>79</b>	<b>32</b>	<b>71</b>	<b>47</b>	ns
Ripe fruit	<b>52</b>	<b>83</b>	<b>62</b>	<b>83</b>	<b>45</b>	<b>75</b>	<b>53</b>	<b>80</b>	**
Floral	<b>50</b>	<b>29</b>	<b>63</b>	<b>31</b>	<b>70</b>	<b>11</b>	<b>61</b>	<b>24</b>	*
Herbaceous	<b>15</b>	<b>14</b>	<b>27</b>	<b>34</b>	<b>18</b>	<b>14</b>	<b>20</b>	<b>21</b>	ns
Mineral	7	<b>26</b>	<b>31</b>	0	<b>12</b>	<b>12</b>	<b>17</b>	<b>13</b>	ns
Yeasty	<b>19</b>	<b>48</b>	<b>45</b>	<b>30</b>	<b>46</b>	<b>42</b>	<b>37</b>	<b>40</b>	ns

Descriptors with GM > 10% are highlighted in bold letters.

Level of significance: \*, \*\* and \*\*\* indicates significance at  $p < 0.05$ ,  $p < 0.01$  and  $p < 0.001$  respectively; ns indicates no significant difference.

**Figure 1.**



**Figure 2.**

**Correlations on axes t1 and t2**

