Received: 26 July 2019 | Accepted: 3 December 2019 | Published: 18 March 2020
DOI:10.20870/oeno-one.2020.54.1.2547

OENO One
VINE AND WINE OPEN ACCESS JOURNAL

Effect of vine-shoot and oak extract foliar grapevine applications on the oenological parameters, phenolic acids and glutathione content of white musts and wines

Rosario Sánchez-Gómez[1], Eva P. Pérez-Álvarez[2], M. Rosario Salinas[1*], Ana Gonzalo-Diago[2], Amaya Zalacain[1] and Teresa Garde-Cerdán[2*]

[1]Universidad de Castilla-La Mancha, E.T.S.I. Agrónomos y Montes, Cátedra de Química Agrícola, Avda. de España s/n, 02071 Albacete, Spain
[2]Instituto de Ciencias de la Vid y del Vino (CSIC, Universidad de La Rioja, Gobierno de La Rioja), Ctra. de Burgos, Km. 6. 26007 Logroño, Spain

*corresponding authors: rosario.salinas@uclm.es, teresa.garde@icvv.es

ABSTRACT

Aim: An oxidation process frequently occurs during white winemaking, affecting its quality. The aim was to study, for two years (2013 and 2014), the effects that foliar applications of vine-shoot (Airén (AVS) and Moscatel (MVS)) and oak wood (OW) extracts on Airén grapevines have on wine color, and must and wine glutathione, *trans*-GRP, *trans*-caftaric acid, and *trans-p*-coutaric acid content.
Methods and results: These compounds were analyzed by HPLC. The results showed that, in general, foliar application of Airén vine-shoot extracts did not affect glutathione concentration, and the other treatments decreased it. AVS_{2013}-50 and AVS_{2014} samples were characterized by a high content of glutathione and *trans*-GRP, while MVS_{2014} samples retain high levels of *trans*-caftaric acid. *trans-p*-Coutaric acid concentration decreased after AVS_{2013} treatment, meanwhile in the 2014 season all applications increased its content. The content of these compounds in the wines was similar to those obtained in the musts. OW_{2013} showed the lowest value of Abs 420, likely because this treatment decreased glutathione concentration in musts.
Conclusions: The foliar applications of vine-shoots and oak extracts on Airén grapevines had a clear effect in reducing the wine alcohol degree and Baumé in case of grapes. This finding is interesting in the context of the current global warming change scenario. Although the two vintages studied were different, an improvement in the color quality was observed in both. The content of glutathione decreased from must to wine: the content in both matrices was lower in treated samples than in control samples, except for Airén extracts in both vintages. This means that glutathione is oxidized, avoiding the oxidation of other must and wine molecules.
Significance and impact of the study: These findings are important in relation to revalorizing waste from the vineyard, and thus being able to improve the quality of the white wine in relation to the oxidation processes that take place in the winemaking process.

KEYWORDS

vine-shoot extracts, foliar application, must, wine, glutathione, colour

 © 2020 International Viticulture and Enology Society - IVES

INTRODUCTION

It has become mandatory for the wine industry to apply waste products to grapevines, with the aim of reducing chemicals according to sustainable practices (European Parliament, 2008). In this sense, vine-shoots are one of the most important waste products and have been used as organic fertilizers and food additives, to produce paper pulp, or to create solid biofuels, etc. (Jiménez Gómez *et al.*, 1993; Jiménez *et al.*, 2006; Portilla *et al.*, 2008; Mendívil *et al.*, 2013). Sánchez-Gómez *et al.* (2014) studied the chemical composition of Airén vine-shoot waste aqueous extracts in terms of phenolic, volatile, and mineral compounds, and reported that the extracts showed an interesting composition that may be assimilated by plants. Thereby, Airén or Moscatel vine-shoot extracts applied foliarly to the grapevines improved wine amino acids, phenolic acids, and volatile composition, increased grape yield and decreased the alcohol degree (Sánchez-Gómez *et al.*, 2016a; Sánchez-Gómez *et al.*, 2016b; Sánchez-Gómez *et al.*, 2017).

Several residues are generated in the manufacturing process of oak barrels, which can be added in the form of chips or pellets as an alternative to wine aging. Pardo-García *et al.* (2014) observed that oak extracts, obtained from oak waste from the cooperages, are interesting applications for the sustainability of the grapevine and oak industry. Thereby, the foliar application of the oak extracts affected grape composition, producing less alcoholic and acidic red wines with higher color intensity, lower shade, a more stable color and a higher content of polyphenols such as gallic acid, hydroxycynnamoyltartaric acids, acylated anthocyanins, flavanols and stilbenes (Pardo-García *et al.*, 2014). In addition, after these applications, several compounds from the oak extracts were assimilated by vines and stored in grapes as glycosidic forms, affecting the wine volatile composition, mainly months after bottle storage (Martínez-Gil *et al.*, 2011; Martínez-Gil *et al.*, 2012; Martínez-Gil *et al.*, 2013).

Color is one of the most important properties of must and wines from white varieties. The oxidation process that occurs frequently in white wines is a well-known problem in the winemaking industry because the color of musts and wines can be modified (Cejudo-Bastante *et al.*, 2010). Sulfur dioxide is usually used to prevent must oxidation and browning development during bottle storage. However, there are strict regulations about sulfur dioxide use in the food industry, because of its toxicity and allergenic effects on human health (Lester, 1995). Hydroxycinnamic acids are prone to enzymatic oxidation reactions during the winemaking process, forming *o*-quinones and leading to color darkening. Thereby, glutathione, is an antioxidant tripeptide formed by three amino acids - cysteine, glutamate and glycine - and present in musts and wines. This tripeptide can regenerate the *o*-diphenol group of enzymatically oxidized *trans*-caftaric acid, giving rise to 2-S-glutathionyl-*trans*-caftaric acid (GRP) and thus inhibiting wine browning (Cheynier *et al.*, 1989; Cejudo-Bastante *et al.*, 2010; Kritzinger *et al.*, 2013). It has been recently reported that bisulfite competes as a nucleophile with glutathione for the *o*-quinone of *trans*-caftaric acid (Hayasaka *et al.*, 2017). Thus, the aim of this work was to study the effect that foliar applications of Airén and Moscatel vine-shoot (AVS and MVS) extracts and oak wood (OW) extracts from non-toasted and toasted wood have on Airén grapevines. The effect of these treatments on must and wine glutathione, *trans*-GRP, *trans*-caftaric acid, and *trans*-*p*-coutaric acid concentration, and on wine color was evaluated during two consecutive vintages.

MATERIALS AND METHODS

1. Vegetal raw material for extract preparation

1.1 Vine-shoot and oak samples

Vine-shoot waste from the Airén *Vitis vinifera* cultivar in 2013 and from Airén and Moscatel in 2014 were sampled in the Castilla-La Mancha Spanish region, four months after each harvest, by randomized selection. Each year, vine-shoots were dried at room temperature for three months until a final humidity of 6.5 % ($g_{water}/100g_{of\ sample}$). The 2013 oak sample was obtained from a commercial aqueous French oak extract (103 C), hereinafter OW_{2013}, supplied by Protea France S.A.S. (Gensac la Pallue, France). This extract was obtained by macerating French oak chips (*Quercus sessiliflora Salisb*) for natural seasoning with water at high temperatures, and this was later toasted at medium-intensity levels, as described by Martínez-Gil *et al.* (2011). In 2014, the French oak wood used was provided by Protea France S.A.S. in the form of non-toasted chips (*Quercus sessiliflora Salisb*), naturally seasoned for 24 months.

1.2 Extracts preparation

All non-toasted vine-shoot waste samples (2013 and 2014) and the oak chips from 2014, were ground in a hammer miller (RETSCH, SM100 Comfort, Haan, Germany), sieved in a 10-mesh

 © 2020 International Viticulture and Enology Society - IVES

to get a homogeneous sawdust, and kept under vacuum at room temperature (25-27ºC) until use. To obtain the toasted samples for the 2014 experiment, dried Airén and Moscatel vine-shoots and oak chip materials were heated at 180 °C for 45 min according to the methods in Sánchez-Gómez *et al.* (2016c) and Sánchez-Gómez *et al.* (2016d) and the extraction procedure was as described in Sánchez-Gómez *et al.* (2014). The resulting Airén vine-shoot wastes extracts were as follows: Airén aqueous extract from non-toasted vine-shoots in 2013 (AVS_{2013}), Airén aqueous extract from non-toasted vine-shoots in 2014 (AVS_{2014}), and Airén aqueous extract from toasted vine-shoots in 2014 ($AVS_{Toasted}$). For Moscatel vine-shoot waste, the resulting extracts were Moscatel aqueous extract from non-toasted vine-shoots in 2014 (MVS_{2014}) and Moscatel aqueous extract from toasted vine-shoots in 2014 ($MVS_{Toasted}$). For oak wood, the extracts obtained were French oak wood extract from non-toasted chips in 2014 (OW_{2014}) and French oak wood extract from toasted chips in 2014 ($OW_{Toasted}$). For each extract, 1.8 L was prepared.

2. Vines treatments

The white grapevines used in this study were from Airén *Vitis vinifera* grown at O.D. Jumilla in southwest Spain (38º 35' 17" N and 1º 16' 20" W; 511 m above sea level) during the 2013 and 2014 vintages. The grapevines were trained on the traditional bush vines Gobelet system with a row distribution north to south.

The formulations from 2013 (AVS_{2013} and OW_{2013}) and 2014 (AVS_{2014}, $AVS_{Toasted}$, MVS_{2014}, $MVS_{Toasted}$, OW_{2014} and $OW_{Toasted}$) were prepared using 0.05 % (v/v) of the Agral (Syngenta, Switzerland) adjuvant, which is a superficial wetting agent typically used for treatments with foliar applications, constituted by an inert mixture of polymers. The chemical composition of the extracts from 2013 and 2014, used in these applications, has been discussed in detail in previous works (Sánchez-Gómez *et al.*, 2014; Sánchez-Gómez *et al.*, 2016c; Sánchez-Gómez *et al.*, 2016d).

In 2013, the formulation effect of vine-shoot extracts was tested, and the previous dilutions carried out with oak extracts (Martínez-Gil *et al.*, 2011, 2012) were used as a reference. The AVS_{2013} extract was applied once on day 7 post-veraison (AVS_{2013}-100) and the second application was carried out with AVS_{2013} diluted with water at 50 % and applied twice on days 7 and 14 post-veraison (AVS_{2013}-50). The commercial aqueous oak extract (OW_{2013}) was applied once on day 7 post-veraison for comparing these results the previous ones from the same oak extract (Martínez-Gil *et al.*, 2011, 2012).

In 2014, the toasted procedure effect and possible vine-shoot varieties effect were tested. Because of the results obtained in 2013, all treatments were applied once. Thus, Airén grapevines were treated with four different vine-shoot extracts (AVS_{2014}, $AVS_{Toasted}$, MVS_{2014}, $MVS_{Toasted}$) and two oak extracts (OW_{2014}, $OW_{Toasted}$), which were applied once on day 7 post-veraison. Figure 1 summarizes the experimental design for the two years.

Each year, the treatments were applied on plants arranged in rows and in an edaphologically homogeneous portion of the plot, avoiding its edges. Each treatment was carried out on six plants randomly selected in the same row, distributed on three replicates with two plants each (n = 3), leaving other rows with untreated plants between the different applications to avoid contamination. Moreover, six additional plants were selected each year as controls, which were treated with water and the adjuvant following the same treatment protocol. The treatments were carried out when the environmental temperature was below 20 °C, between approximately 7 a.m. and 9 a.m. The grapevines were treated with 300 mL of each formulation per plant by spraying locally over the leaves.

3. Winemaking

Grapes were manually harvested on 26 September 2013 and 9 September 2014 at the optimum technological maturation moment for the grapevines used as the control sample. In both years, the grapes (approximately 20 kg for each treatment) were separately processed. These grapes were destemmed and the whole grape mass was mixed and homogenized (must and solid parts combined). Then, the grape mass from each treatment was placed into 30 L stainless steel tanks and ammonium bisulfite (150 mg/mL) was added to the resulting musts. Maceration was carried out at 11 °C ± 1 °C for 17 h.

Skins and seeds from each treatment were removed with a traditional vertical hand-press and the free-run musts were put into stainless steel tanks to finish the alcoholic fermentation. Musts were inoculated with 20 g/hL of UCLM S377 active dry yeast strain (Springer Oenologie, Maisons-Alfort Cedex, France) of *Saccharomyces bayanus,* previously rehydrated and activated according

© 2020 International Viticulture and Enology Society - IVES

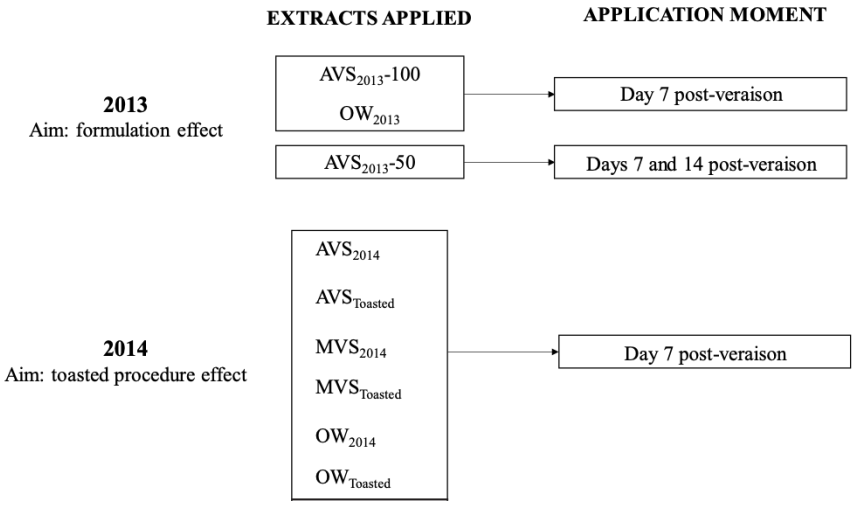


FIGURE 1. Experimental design for 2013 and 2014 vintages.

AVS: grapevines treated with Airén aqueous extract from non-toasted vine-shoots: from 2013 (AVS_{2013}) and 2014 (AVS_{2014}). $AVS_{Toasted}$, grapevines treated with Airén aqueous extract from toasted vine-shoots; MVS_{2014}, grapevines treated with Moscatel aqueous extract from non-toasted vine-shoots; $MVS_{Toasted}$, grapevines treated with Moscatel aqueous extract from toasted vine-shoots; OW_{2013}, commercial aqueous oak extract; OW_{2014}, French oak wood extract from non-toasted chips; $OW_{Toasted}$, French oak wood extract from toasted chips.

to the supplier. The alcoholic fermentation took place at 16 °C ± 1 °C. The density was measured each day and alcoholic fermentation was considered complete after 16 days in 2013 and 17 days in 2014, when it was constant at 0.990 g/L and the residual sugars value were below 5 g/L. As no malolactic fermentation was carried out, at this final point three wines from each treatment (33 wines in total: 12 from 2013 and 21 from 2014) were obtained, bottled and stored at 22 °C ± 2 °C.

4. Analytical methods

4.1 Musts and wines oenological parameters

Must parameters such as °Baumé (ºBé), pH, total acidity (g/L of tartaric acid) and malic acid (g/L), along with wines parameters such as alcoholic degree (°A), pH, total acidity (g/L of tartaric acid), volatile acidity (g/L), malic acid (g/L), lactic acid (g/L) and color intensity (Abs 420 nm) were analyzed using equipment based on Fourier Transform-infrared spectroscopy (FT-IR Multispec; TDI, Barcelona, Spain) using the methods of ECC (1990) as reference.

4.2 Determination of glutathione by HPLC

The glutathione analysis was performed using the method described by Garde-Cerdán *et al.* (2014). This compound was analyzed by reverse-phase HPLC using a liquid chromatograph Agilent 1100 Series (Palo Alto, USA). Each sample (5 mL) was mixed with 100 µL of norvaline (internal standard). The mixture was submitted to an automatic precolumn derivatization with *o*-phthaldialdehyde (OPA Reagent, Agilent). The injected amount from the derived sample was 10 µL, with a constant temperature of 40 ºC. All separations were performed on a Hypersil ODS (250 × 4.0 mm, I.D. 5 µm) column (Agilent).

Two eluents were used as mobile phases: eluent A was 75 mM sodium acetate, 0.018 % triethylamine (pH 6.9) + 0.3 % tetrahydrofuran; eluent B was water, methanol, and acetonitrile (10:45:45, v/v/v). Detection was performed using a fluorescence detector (FLD). The identification of glutathione was performed by comparison of its retention time with that of pure reference standard. The pure reference compound and internal standard were from Sigma-Aldrich (Steinheim, Germany). The results for glutathione correspond to the average of three analyses (n = 3).

4.3 Determination of trans-GRP, *trans*-caftaric and *trans*-p-coutaric acids by HPLC-DAD-MS

The analysis was based on methods from Pardo-García *et al.* (2014) and Sánchez-Gómez *et al.* (2014). Briefly, the HPLC grade solvents used were water/formic acid/acetonitrile (97.5:1.5:1 v/v/v) as solvent A and acetonitrile/formic acid/solvent A (78.5:1.5:20 v/v/v) as solvent B. The elution gradient was set up for solvent B as follows: 0 min, 5 %; 2 min, 10 %; 4 min, 14 %; 9 min, 14 %; 37 min, 18.5 %; 35 min, 20 %;

50 min, 25 %; 55 min, 50 %; 60 min, 5 %; and 65 min, 5 %. The loop volume was 20 µL.

Identification of *trans*-GRP (2-S-glutathionyl-*trans*-caftaric acid or grape reaction product) and *trans*-caftaric and *trans*-*p*-coutaric acids was carried out using MS and DAD detectors in comparison with the corresponding mass fragmentation and UV-Vis spectra. As they were not available, they were quantified at the same wavelength as their analogous hydroxycinnamic acids (*trans*-caffeic and *trans*-*p*-coumaric acids, respectively): *trans*-GRP and *trans*-caftaric acid at 324 nm, and *trans*-*p*-coutaric acid at 308 nm. Quantification was based on *trans*-caffeic and *trans*-*p*-coumaric acids calibration curves of their respective standards (Sigma-Aldrich) at five different concentrations achieved by UV-Vis signal (0.70-175 mg/L) (R^2 = 0.92-0.99).

5. Statistical analysis

Data statistical analysis was performed using the SPSS Version 22.0 statistical package for Windows (SPSS, Chicago, USA). Oenological parameters, glutathione, *trans*-GRP, *trans*-caftaric and *trans*-*p*-coutaric acids data concentration from musts and wines were processed using variance analysis (ANOVA). Differences between means were compared using the Tukey test at 99.95 % probability level. Canonical discriminant analyses were performed in both years for all treatments with the glutathione, *trans*-GRP, *trans*-caftaric and *trans*-*p*-coutaric acids concentration in musts and wines.

RESULTS AND DISCUSSION

1. Must and wine oenological parameters

The oenological parameters for musts in both years of study are shown in Table 1. In 2013, control must presented significant differences in all parameters studied in relation to the treatments. Control musts showed the highest ºBaumé content and the lowest pH, although in this last case pH range values were very close among control and musts from treatments. On the contrary, OW_{2013} presented the lowest value of ºBaumé and the highest total acidity and malic acid content followed in these two last parameters for control must. Musts from grapevines treated with Airén vine-shoot extracts (AVS_{2013}-50 and AVS_{2013}-100) showed intermediate content of alcohol and the lowest values of total acidity and malic acid, and therefore, the highest pH values.

In the 2014 vintage, ºBaumé in the control must showed a similar behavior than in the 2013 must, presenting the highest content and significant differences regarding the treatments. Thus, in general, the treatments tested seem to affect the ºBaumé content, providing in

TABLE 1. Oenological parameters of musts at harvest day after the different grapevine treatments applied in 2013 and 2014.

		ºBaumé	pH	Total acidity (g/L tartaric acid)	Malic acid (g/L)
Must 2013	Control $_{2013}$	10.20 ± 0.00 d	3.49 ± 0.00 a	4.81 ± 0.01 c	4.06 ± 0.05 c
	AVS_{2013}-50	9.90 ± 0.00 b	3.51 ± 0.00 c	3.96 ± 0.01 a	2.94 ± 0.05 a
	AVS_{2013}-100	10.10 ± 0.00 c	3.54 ± 0.00 d	4.65 ± 0.01 b	3.90 ± 0.05 b
	OW_{2013}	9.30 ± 0.00 a	3.50 ± 0.00 b	4.99 ± 0.01 d	4.68 ± 0.05 d
Must 2014	Control $_{2014}$	11.25 ± 0.07 e	3.57 ± 0.01 b	2.22 ± 0.01 ab	0.57 ± 0.04 ab
	AVS_{2014}	10.70 ± 0.00 d	3.60 ± 0.00 d	2.25 ± 0.04 b	0.75 ± 0.05 b
	$AVS_{Toasted}$	9.55 ± 0.07 a	3.56 ± 0.00 a	2.40 ± 0.01 c	0.55 ± 0.02 ab
	MVS_{2014}	9.80 ± 0.00 b	3.57 ± 0.01 b	2.44 ± 0.01 c	0.56 ± 0.06 ab
	$MVS_{Toasted}$	10.30 ± 0.00 c	3.55 ± 0.00 a	2.23 ± 0.03 b	0.39 ± 0.01 a
	OW_{2014}	10.30 ± 0.00 c	3.58 ± 0.00 c	2.15 ± 0.01 a	0.65 ± 0.09 b
	$OW_{Toasted}$	10.40 ± 0.00 c	3.55 ± 0.01 a	2.40 ± 0.01 c	0.70 ± 0.05 b

The mean values are shown with their standard deviation (n = 3). For each year, different letters in the same column indicate significant differences between treatments ($p \leq 0.05$).

Control: untreated grapevines, from 2013 ($Control_{2013}$) and 2014 ($Control_{2014}$). AVS, grapevines treated with Airén aqueous extract from non-toasted vine-shoots. From 2013, applied once on day 7 post-veraison (AVS_{2013}-100) and applied twice, at days 7 and 14 post-veraison, diluted with water at 50 % (AVS_{2013}-50); from 2014, applied once on day 7 post-veraison (AVS_{2014}). $AVS_{Toasted}$: grapevines treated with Airén aqueous extract from toasted vine-shoots, applied once on day 7 post-veraison in 2014. MVS_{2014}, grapevines treated with Moscatel aqueous extract from non-toasted vine-shoots, applied once on day 7 post-veraison in 2014. $MVS_{Toasted}$, grapevines treated with Moscatel aqueous extract from toasted vine-shoots, applied once on day 7 post-veraison in 2014. OW, commercial aqueous oak extract (OW_{2013}) applied once on day 7 post-veraison; French oak wood extract from non-toasted chips in 2014 (OW_{2014}) and French oak wood extract from toasted chips in 2014 ($OW_{Toasted}$).

 © 2020 International Viticulture and Enology Society - IVES

some cases musts with minor ºBaumé values: OW_{2013} in 2013 and $AVS_{Toasted}$ in 2014 (Table 1). However, in general, the differences were very low, so the effect was minimal. This grape ripening delay could be due to the application of these vegetal extracts, so this should be taken into account in the current global warming/climate change scenario (Jones *et al.*, 2005; Hannah *et al.*, 2013; Webb *et al.*, 2012). Regarding pH, AVS_{2014} must showed the highest value, with the same behavior in 2013 for AVS_{2013}-100 treatment whereas $AVS_{Toasted}$, MVS_{2014} and $OW_{Toasted}$ musts had the highest total acidity content and OW_{2014} the lowest. In relation to malic acid content, must from AVS_{2014} treatment and both from oak extracts applications showed the highest content, whereas the $MVS_{Toasted}$ value was the lowest.

The oenological parameters of the wines at the end of alcoholic fermentation after different grapevine treatments for both vintages, 2013 and 2014, are shown in Table 2. All parameters measured related to their technological quality have values according to the bibliography for Airén high-quality wines (Bueno *et al.*, 2006; Cejudo-Bastante *et al.*, 2012; Benito *et al.*, 2016). The same behavior observed in control musts for both vintages in relation to ºBaumé was evidenced in the case of wines. Control musts presented the highest alcohol degree, being statistically different from the other treatments, except for AVS_{2013}-50 treatment. This decrease in wine alcoholic degree was previously reported by Pardo-García *et al.* (2014), when an oak extract was applied on Monastrell grapevines.

In 2013, no significant differences were observed among treatments and control wines in total acidity. In relation to other parameters, control and OW_{2013} wines presented lower pH values than wines from vine-shoot treatments (AVS_{2013}-50 and AVS_{2013}-100), which may be due to the high malic acid content of OW_{2013} wine and the high lactic acid content of control wine, as total acidity, quantified as tartaric acid, was similar in all wines. The values of malic acid found in all the wines revealed that no malolactic fermentation occurred. Wines from AVS treatments showed lower values of malic and lactic acids, exhibiting the highest value of volatile acidity with AVS_{2013}-50. Notwithstanding, this was below 0.6 g/L, indicating that all wines were well conserved, which means that there were no microbiological problems (Ribéreau-Gayon *et al.*, 2006). OW_{2013} wine was the only one that presented significant differences in Abs 420 nm with respect to control and AVS treatments (AVS_{2013}-50 and AVS_{2013}-100), as the OW_{2013} value the lowest. In 2014, the control wines had the highest pH, whereas in the case of total and volatile acidity, AVS_{2014} presented the lowest value for both parameters, but only in the first one; these differences were statistically significant with respect to the control and the rest of the wines.

TABLE 2. Oenological parameters of wines at the end of alcoholic fermentation after the different grapevine treatments applied in 2013 and 2014.

		Alcohol degree (%, v/v)	pH	Total acidity (g/L tartaric acid)	Volatile acidity (g/L)	Malic acid (g/L)	Lactic acid (g/L)	Abs 420
Wines 2013	Control $_{2013}$	9.70 ± 0.13 c	3.25 ± 0.01 a	6.81 ± 0.11 a	0.15 ± 0.02 ab	3.65 ± 0.03 b	0.29 ± 0.03 c	0.52 ± 0.04 b
	AVS_{2013}-50	9.95 ± 0.30 c	3.28 ± 0.01 b	6.83 ± 0.08 a	0.26 ± 0.03 c	3.40 ± 0.09 a	0.14 ± 0.02 ab	0.52 ± 0.03 b
	AVS_{2013}-100	8.96 ± 0.14 b	3.33 ± 0.01 c	6.71 ± 0.09 a	0.11 ± 0.03 a	3.60 ± 0.09 b	0.10 ± 0.03 a	0.58 ± 0.04 b
	OW_{2013}	8.13 ± 0.36 a	3.23 ± 0.01 a	6.83 ± 0.08 a	0.20 ± 0.04 bc	3.86 ± 0.07 c	0.18 ± 0.03 b	0.45 ± 0.02 a
Wines 2014	Control $_{2014}$	12.03 ± 0.05 f	3.34 ± 0.01 e	6.08 ± 0.04 b	0.14 ± 0.02 ab	1.86 ± 0.02 bc	0.40 ± 0.02 b	0.53 ± 0.04 ab
	AVS_{2014}	10.86 ± 0.03 e	3.31 ± 0.01 d	5.67 ± 0.03 a	0.09 ± 0.01 a	1.70 ± 0.03 a	0.39 ± 0.03 ab	0.47 ± 0.02 a
	$AVS_{Toasted}$	9.70 ± 0.03 a	3.20 ± 0.01 a	6.13 ± 0.02 bc	0.10 ± 0.01 ab	1.78 ± 0.02 ab	0.31 ± 0.03 ab	0.50 ± 0.03 ab
	MVS_{2014}	9.79 ± 0.02 a	3.23 ± 0.02 b	5.95 ± 0.22 b	0.12 ± 0.08 ab	1.86 ± 0.13 bc	0.40 ± 0.12 b	0.49 ± 0.02 a
	$MVS_{Toasted}$	10.48 ± 0.04 c	3.28 ± 0.00 c	6.32 ± 0.11 c	0.15 ± 0.01 b	2.25 ± 0.10 d	0.58 ± 0.02 c	0.54 ± 0.02 b
	OW_{2014}	10.67 ± 0.07 d	3.28 ± 0.01 d	6.13 ± 0.02 bc	0.12 ± 0.01 ab	1.92 ± 0.04 c	0.36 ± 0.02 ab	0.55 ± 0.02 b
	$OW_{Toasted}$	10.00 ± 0.17 b	3.25 ± 0.00 b	6.08 ± 0.14 b	0.09 ± 0.01 a	1.98 ± 0.07 c	0.29 ± 0.06 a	0.53 ± 0.02 ab

The mean values are shown with their standard deviation (n = 3). For each year, different letters in the same column indicate significant differences between treatments ($p \leq 0.05$). Control: untreated grapevines, from 2013 ($Control_{2013}$) and 2014 ($Control_{2014}$). AVS, grapevines treated with Airén aqueous extract from non-toasted vine-shoots, from 2013, applied once on day 7 post-veraison (AVS_{2013}-100) and applied twice, on days 7 and 14 post-veraison, diluted with water at 50 % (AVS_{2013}-50); from 2014, applied once on day 7 post-veraison (AVS_{2014}). $AVS_{Toasted}$, grapevines treated with Airén aqueous extract from toasted vine-shoots, applied once on day 7 post-veraison in 2014. MVS_{2014}, grapevines treated with Moscatel aqueous extract from non-toasted vine-shoots, applied once on day 7 post-veraison in 2014. $MVS_{Toasted}$, grapevines treated with Moscatel aqueous extract from toasted vine-shoots, applied once on day 7 post-veraison in 2014. OW, commercial aqueous oak extract (OW_{2013}) applied once on day 7 post-veraison; French oak wood extract from non-toasted chips in 2014 (OW_{2014}) and French oak wood extract from toasted chips in 2014 ($OW_{Toasted}$).

 © 2020 International Viticulture and Enology Society - IVES

TABLE 3. Glutathione, *trans*-GRP and trans-caftaric and *trans*-p-coutaric acids content (mg/L) in musts and wines after the different grapevine treatments applied in 2013 and 2014.

		Glutathione		*trans* -GRP		*trans* -Caftaric acid		*trans* -*p*- Coutaric acid	
		Musts	Wines	Musts	Wines	Musts	Wines	Musts	Wines
2013	Control $_{2013}$	44.69 ± 1.99 bc, β	1.20 ± 0.46 b, α	10.79 ± 0.06 a, β	7.05 ± 0.06 a, α	57.46 ± 0.17 d, β	29.16 ± 0.40 a, α	6.98 ± 0.09 b, α	10.84 ± 0.16 b, β
	AVS_{2013}-50	45.95 ± 1.97 c, β	1.26 ± 0.40 b, α	12.35 ± 0.11 c, β	9.17 ± 0.04 c, α	47.49 ± 0.47 a, β	31.41 ± 0.31 b, α	6.05 ± 0.03 a, α	11.43 ± 0.17 c, β
	AVS_{2013}-100	40.86 ± 0.94 b, β	0.29 ± 0.14 a, α	12.70 ± 0.07 d, β	7.71 ± 0.06 b, α	53.41 ± 0.68 b, β	28.08 ± 1.04 a, α	6.92 ± 0.08 b, α	10.27 ± 0.12 a, β
	OW_{2013}	29.31± 0.91 a, β	0.83 ± 0.35 ab, α	11.31 ± 0.10 b, β	7.03 ± 0.02 a, α	55.05 ± 0.86 c, β	30.74 ± 0.31 b, α	7.02 ± 0.13 b, α	13.10 ± 0.08 d, β
2014	Control $_{2014}$	20.18 ± 1.31 c, β	1.06 ± 0.24 b, α	6.52 ± 0.02 d, β	4.97 ± 0.02 d, α	18.87 ± 0.58 a, α	28.82 ± 0.09 a, β	6.63 ± 0.45 a, α	8.60 ± 0.03 a, β
	AVS_{2014}	21.01 ± 0.18 c, β	1.15 ± 0.02 b, α	7.76 ± 0.15 f, β	5.38 ± 0.14 e, α	26.67 ± 0.87 c, α	29.16 ± 0.01 ab, β	9.59 ± 0.30 c, α	9.62 ± 0.49 b, α
	$AVS_{Toasted}$	17.80 ± 0.23 bc, β	0.32 ± 0.03 a, α	4.93 ± 0.08 b, β	4.20 ± 0.01 b, α	25.55 ± 0.77 c, α	32.02 ± 0.04 c, β	10.39 ± 0.30 c, α	12.02 ± 0.03 c, β
	MVS_{2014}	15.23 ± 0.02 ab, β	0.39 ± 0.06 a, α	7.19 ± 0.02 e, β	4.54 ± 0.26 c, α	40.60 ± 0.78 e, β	38.89 ± 1.00 d, α	16.42 ± 0.55 e, β	14.10 ± 0.13 d, α
	$MVS_{Toasted}$	15.20 ± 0.41 ab, β	0.35 ± 0.00 a, α	7.19 ± 0.06 e, β	3.91 ± 0.02 b, α	32.21 ± 0.37 d, α	32.96 ± 0.18 c, α	12.64 ± 0.38 d, β	11.26 ± 0.27 c, α
	OW_{2014}	12.95 ± 0.32 a, β	0.47 ± 0.06 a, α	6.12 ± 0.02 c, β	4.07 ± 0.06 b, α	22.69 ± 0.63 b, α	28.36 ± 0.29 a, β	7.91 ± 0.13 b, α	9.54 ± 0.03 b, β
	$OW_{Toasted}$	13.50 ± 2.04 b, β	0.22 ± 0.02 a, α	4.66 ± 0.10 a, β	3.09 ± 0.01 a, α	18.13 ± 0.33 a, α	30.07 ± 0.01 b, β	7.81 ± 0.25 b, α	11.16 ± 0.65 c, β

For each year, letters in the same column indicate significant differences between treatments and Greek letters indicate, for each compound, differences between musts and wines according to the Tukey test ($p \leq 0.05$). The mean values are shown with their standard deviation ($n = 3$).

Control: untreated grapevines, from 2013 ($Control_{2013}$) and 2014 ($Control_{2014}$). AVS, grapevines treated with Airén aqueous extract from non-toasted vine-shoots, from 2013, applied once on day 7 post-veraison (AVS_{2013}-100) and applied twice, on days 7 and 14 post-veraison, diluted with water at 50 % (AVS_{2013}-50); from 2014, applied once on day 7 post-veraison (AVS_{2014}). $AVS_{Toasted}$, grapevines treated with Airén aqueous extract from toasted vine-shoots, applied once on day 7 post-veraison in 2014. MVS_{2014}, grapevines treated with Moscatel aqueous extract from non-toasted vine-shoots, applied once on day 7 post-veraison in 2014. $MVS_{Toasted}$, grapevines treated with Moscatel aqueous extract from toasted vine-shoots, applied once on day 7 post-veraison in 2014. OW, commercial aqueous oak extract (OW_{2013}) applied once on day 7 post-veraison; French oak wood extract from non-toasted chips in 2014 (OW_{2014}) and French oak wood extract from toasted chips in 2014 ($OW_{Toasted}$).

© 2020 International Viticulture and Enology Society - IVES

As in 2013 vintage, malolactic fermentation was not carried out, which was confirmed by the high values of malic acid (Table 2): the wines with the $MVS_{Toasted}$ treatment had a significantly higher value of malic and lactic acid. Although the vintage variability is observed, foliar applications seem to have a clear effect on the alcohol degree, as in both vintages the treated wines were less alcoholic than control ones. Additionally, the formulation effect along with the wood toasting procedure or wood type, does not seem to have an influence on the alcohol degree.

2. Glutathione, trans-GRP and *trans*-caftaric and *trans*-p-coutaric acids content in musts and wines

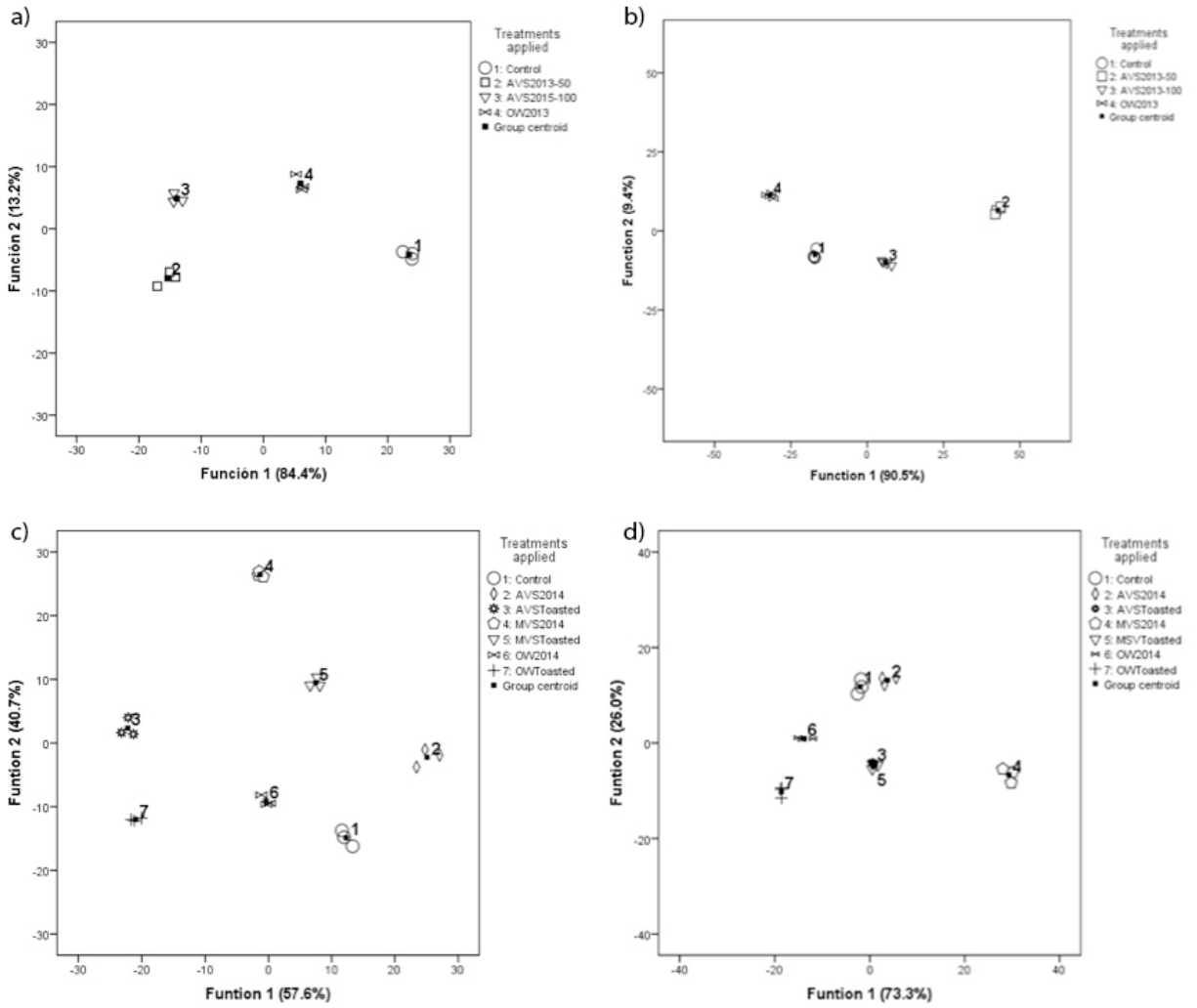
Glutathione, *trans*-GRP and *trans*-caftaric and *trans-p*-coutaric acids content in musts and wines for the two years studied after different grapevines treatments are summarized in Table 3. In both studied vintages, the application of Airén vine-shoot extracts (AVS and $AVS_{Toasted}$) did not affect glutathione concentration in musts with respect to the control vines. However, in both cases, the treatment carried out with the oak wood extracts (OW and $OW_{Toasted}$) significantly decreased the glutathione content in the musts compared to control samples. In relation to the Moscatel vine-shoot extracts (MVS and $MVS_{Toasted}$), their effect was similar to those obtained by the OW extracts, as glutathione concentration in musts decreased compared to control samples. The concentration of all these extracts decreased considerably after alcoholic fermentation, even below 2 mg/L (Table 3). In the 2013 vintage, wines from grapevines treated with $AVS_{2013\text{-}100}$ showed the lowest glutathione content with respect to the control wines. However, in the 2014 vintage, wines elaborated from the grapevines treated with different treatments presented lower glutathione concentration than control samples, except for AVS_{2014} wines. The different behavior in the glutathione concentration could be due to the climatic conditions, among other reasons, as those affect the vines development and by the pathways that modify or activate vine secondary mechanisms due to the different extract applications.

All treatments applied during the 2013 vintage increased significantly the *trans*-GRP content in musts, with respect to controls. In this regard, the most effective treatments were both vine-shoot extracts (AVS_{2013}-50 and AVS_{2013}-100) compared to the oak wood extract application (OW_{2013}) (Table 3). In the 2014 vintage, the content of *trans*GRP in musts was increased by the application of non-toasted Airén vine-shoot extract (AVS_{2014}) and from both Moscatel vine-shoot extracts, non-toasted and toasted. The rest of the treatments, such as application of oak wood extracts (OW_{2014} and $OW_{Toasted}$) and $AVS_{Toasted}$ produced a decrease in *trans*-GRP content respect to control musts. *trans*-GRP concentration also decreased after alcoholic fermentation, but to a lesser extent than glutathione (Table 3). In the 2013 vintage, wines from grapevines treated with AVS_{2013}-50 and AVS_{2013}-100 had higher *trans*-GRP content than the control and OW_{2013} wines. In 2014, the highest *trans*-GRP concentration was found in the AVS_{2014} wines, while the lowest content was found in $OW_{Toasted}$ wines.

In the first year of study, the concentration of *trans*-caftaric acid decreased in the musts from grapevines treated with vine-shoot and OW extracts (Table 3). However, in the 2014 vintage, all treatments except $OW_{Toasted}$ increased *trans*-caftaric acid content in the musts compared to control samples. In this way, the most effective treatments were the vine-shoot extracts from Moscatel (MVS_{2014} and $MVS_{Toasted}$). *trans*-caftaric acid concentration during the 2013 vintage decreased after alcoholic fermentation, but it increased or remained similar in the 2014 season. It is possible that during the 2013 winemaking this compound was involved in enzymatic oxidation reactions. *trans*-Caftaric acid forms *o*-quinone leading to color darkening (Ferreira-Lima *et al.*, 2016). So, wines from 2013 vintage elaborated from grapevines treated with AVS_{2013}-50 and $OW_{2013,}$ had higher *trans*-caftaric acid content than the control and AVS_{2013}-100 wines. In the 2014 vintage, wines from untreated grapevines AVS_{2014} and OW_{2014} showed lower *trans*-caftaric acid content than $AVS_{Toasted}$, MVS_{2014} and $MVS_{Toasted}$ wines.

The treatments did not affect *trans-p*-coutaric acid concentration in the 2013 vintage musts, except AVS_{2013}-50 (Table 3), which showed lower content compared to the control and the rest of the treatments. However, in the 2014 vintage all treatments increased *trans-p*-coutaric acid content compared to untreated grapevines: MVS_{2014} and $MVS_{Toasted}$ were the most effective treatments. This was similar for *trans*-caftaric acid, as after alcoholic fermentation, its concentration increased in all samples and in both vintages except for MVS_{2014} and $MVS_{Toasted}$ (Table 3). The increase in the *trans-p*-coutaric acid content from must to wine was also reported by Portu *et al.* (2015) when jasmonate foliar application was carried out on

 © 2020 International Viticulture and Enology Society - IVES



	Musts 2013		Wines 2013		Musts 2014		Wines 2014	
	F1	F2	F1	F2	F1	F2	F1	F2
Glutatione	0.736	-1.036	-0.858	0.525	0.343	0.251	0.044	0.579
trans-GPR	-1.475	0.429	1.399	0.240	1.262	-0.150	1.282	0.757
trans-Caftaric acid	0.409	0.387	1.380	-0.394	-0.409	0.813	1.440	-0.078
trans-p-Coutaric acid	0.628	0.414	-1.161	1.256	-0.332	0.613	0.005	-0.584

FIGURE 2. Canonical discriminant analysis with glutathione, *trans*-GRP and *trans*-caftaric and *trans*-p-coutaric acids concentration in musts and wines after the different grapevine treatments of a) musts in 2013, b) wines in 2013, c) musts in 2014 and d) wines in 2014.

Control: untreated grapevines, from 2013 ($Control_{2013}$) and 2014 ($Control_{2014}$). AVS, grapevines treated with Airén aqueous extract from non-toasted vine-shoots. From 2013, applied once on day 7 post-veraison (AVS_{2013}-100) and applied twice, at days 7 and 14 post-veraison, diluted with water at 50 % (AVS_{2013}-50); from 2014, applied once on day 7 post-veraison (AVS_{2014}). $AVS_{Toasted}$, grapevines treated with Airén aqueous extract from toasted vine-shoots, applied once on day 7 post-veraison in 2014. MVS_{2014}, grapevines treated with Moscatel aqueous extract from non-toasted vine-shoots, applied once on day 7 post-veraison in 2014. $MVS_{Toasted}$: grapevines treated with Moscatel aqueous extract from toasted vine-shoots, applied once on day 7 post-veraison in 2014. OW, commercial aqueous oak extract (OW_{2013}) applied once on day 7 post-veraison; French oak wood extract from non-toasted chips in 2014 (OW_{2014}) and French oak wood extract from toasted chips in 2014 ($OW_{Toasted}$).

Tempranillo grapevines. In 2013, all wines showed different *trans-p*-coutaric acid concentration, where OW_{2013} wines had the highest content and AVS_{2013}-100 the lowest. In the 2014 vintage, control and MVS_{2014} wines presented the lowest and the highest *trans-p*-coutaric acid content, respectively. Except for *trans-p*-coutaric acid, whose content was similar during both vintages, the concentration of the rest of the compounds studied in the Airén musts was higher in 2013 than in 2014 vintage. *trans-p*-Coutaric acid is an ester formed from *trans-p*-coumaric acid and tartaric acid. Acid esters are formed in simple hydrogen-ion-catalyzed esterification (Margalit, 2004)

 © 2020 International Viticulture and Enology Society - IVES

and it has been shown that their formation and the hydroxycinnamic acid production depends on pH (Edwards *et al.*, 1985; Hall and De Luca, 2007). As for the glutathione concentration, the differences found in the phenolic acid composition could be attributed to the physiological impact of the use of the extracts on biosynthesis of the compounds analyzed in must and wines, related to the secondary mechanism.

In an attempt to differentiate the treatments of both vintages in musts and wines according to glutathione, *trans*-GRP and *trans*-caftaric and *trans*-*p*-coutaric acids compounds, complementary discriminants analyses were also carried out (Figure 2). Figure 2a shows the graphical representation of the must samples in the 2013 vintage for each treatment. The two canonic discriminant functions explained 97.60 % of the total variance, reaching 84.40 % with the first. In case of function 1, *trans*-GRP was the compound that contributed more to the differentiation with a negative correlation. For function 2, glutathione had the highest weight, also with a negative correlation. Figure 2b shows the discriminant analysis for wines from 2013 vintage, which presented a wide distance between control, OW_{2013} and AVS_{2013}-100, with respect to AVS_{2013}-50 treatment. Function 1 and 2 explained 90.50 % and 9.40 % of the total variance, respectively. *trans*-GRP and *trans*-caftaric acid were the compounds that contributed more to the differentiation with a positive correlation in function 1, whereas for function 2 *trans*-*p*-coutaric acid had a higher weight and a positive correlation. According to the information provided by the variables related to functions 1 and 2, the must from AVS_{2013}-50, and AVS_{2013}-100 keep a higher level of *trans*-GRP; along with this, AVS_{2013}-50 was also describedas having a high glutathione content. In contrast, the control kept a lower level of *trans*-GRP, while OW_{2013} had a lower content of glutathione. With respect to wines, as occurred in musts, AVS_{2013}-50 was described by *trans*-GPR, while OW_{2013} was described by glutathione and *trans*-*p*-coutaric acid, contrary to those that occurred in the musts. Figure 2c shows a graphical representation of the must samples in the 2014 vintage for each treatment: 98.30 % of the total variance was explained by the two canonic discriminant functions. In the case of function 1, which explained the 57.60 % of the total variance, *trans*-GRP was the compound that contributed more to the differentiation with a positive correlation. For function 2, which showed 40.70 % of the total variance, the *trans*-caftaric acid had the highest weight, also with a positive correlation. Figure 2d shows the graphical representation for wines from 2014: 73.30 % and 26.00 % of the total variance were explained by function 1 and 2, respectively. *trans*-GRP and *trans*-caftaric acid were the compounds that contributed more to the differentiation with a positive correlation in function 1, whereas for function 2 *trans*-GRP had a higher weight and a positive correlation. According to the information provided by the variables related to functions 1 and 2, it could be concluded that musts from AVS_{2014} were described by *trans*-GRP, while musts from MVS_{2014} retain higher levels of *trans*-caftaric acid. With respect to the wines, AVS_{2014} and control samples were described by *trans*-caftaric acid and *trans*-GRP, while MVS_{2014} samples were characterized by *trans*-GRP. $AVS_{Toasted}$, $MVS_{Toasted}$, OW_{2014} and $OW_{Toasted}$ wine samples presented similar behavior.

Acknowledgments: Many thanks for the financial support given by the Ministerio de Economía y Competitividad-FEDER of the Spanish Government to the Project AGL2012-33132. T.G.-C. wishes to thank the MINECO for her Ramón y Cajal contract. Also, R.S.-G. is grateful for her predoctoral contract (PREDUCLM17) from Universidad de Castilla-La Mancha. A.G.-D. and E.P.P.-Á. thank the Gobierno de La Rioja for the research personal formation grant. We wish to thank the Bodega San Isidro (Winery in Murcia, southwest Spain) for allowing us to use its vineyards and Francisco Pardo, Diego Cutillas and Juan Miguel Baños for their technical assistance during winemaking and Manuel Márquez (Viticultural Service Company) for selecting and suppling the vine-shoots

REFERENCES

Benito A., Calderón F., Palomero F. and Benito S., 2016. Quality and composition of Airén wines fermented by sequential inoculation of *Lachancea thermotolerans* and *Saccharomyces cerevisiae*. *Food Technology and Biotechnology*, 54, 135-144. doi:10.17113/ftb.54.02.16.4220

Bueno J.E., Peinado R.A., Medina M. and Moreno J., 2006. Effect of a short contact time with lees on volatile composition of Airen and Macabeo wines. *Biotechnology Letters*, 28, 1007-1011. doi:10.1007/s10529-006-9038-2

Cheynier V., Souquet J.M. and Moutounet M., 1989. Glutathione content and glutathione to hydroxycinnamic acid ratio in *Vitis vinifera* grapes and musts. *American Journal of Enology and Viticulture*, 40, 320-324.

Cejudo-Bastante M.J., Pérez-Coello M.S. and Hermosín-Gutiérrez I., 2010. Identification of new derivatives of 2-S-glutathionylcaftaric acid in aged white wines by HPLC-DAD-ESI-MS^n. *Journal of Agricultural and Food Chemistry*, 58, 11483-11492.

 © 2020 International Viticulture and Enology Society - IVES

Cejudo-Bastante M.J., Pérez-Coello M.S., Pérez-Juan P.M. and Hermosín-Gutiérrez I., 2012. Effects of hyper-oxygenation and storage of Macabeo and Airén white wines on their phenolic and volatile composition. *European Food Research and Technology*, 234, 87-99. doi:10.1007/s00217-011-1619-1

ECC, 1990. Commission regulation VO 2676/90 concerning the establishment of common analytical methods in the sector of wine. *Official Journal of the European Community*, 272, 1-192.

Edwards T., Singleton V. and Boulton R., 1985. Formation of ethyl acids of tartaric acid during wine aging: Chemical and sensory effects. *American Journal of Enology and Viticulture*, 36, 118-124.

European Parliament, 2008. Directive 2008/98/EC of the European Parliament and of the Council on waste and repealing certain directives. *Official Journal of the European Union*, 98, 3-4.

Ferreira-Lima N., Vallverdú-Queralt A., Meudec E., Mazauric J.P., Sommerer N., Bodignon-Luiz M.T., Cheynier V. and Le Guernevé C., 2016. Synthesis, identification, and structure elucidation of adducts formed by reactions of hydroxycinnamic acids with glutathione or cysteinylglycine. *Journal of Natural Products*, 79, 2211-2222. doi:10.1021/acs.jnatprod.6b00279

Garde-Cerdán T., López R., Portu J., González-Arenzana L., López-Alfaro I. and Santamaría P., 2014. Study of the effects of proline, phenylalanine, and urea foliar application to Tempranillo vineyards on grape amino acid content. Comparison with commercial nitrogen fertilizers. *Food Chemistry*, 163, 136-141. doi:10.1016/j.foodchem.2014.04.101

Hannah L., Roehrdanz P.R., Ikegami M., Shepard A.V., Shaw M.R., Tabor G., Zhi L., Marquet P.A. and Hijmans R.J., 2013. Climate change, wine, and conservation. *Proceedings of the National Academy of Sciences of the United States of America*, 110, 6907-6912. doi:10.1073/pnas.1210127110

Hall D. and De Luca V., 2007. Mesocarp localization of a bifunctional resveratrol/hydroxycinnamic acid glucosyltransferase of Concord grape (*Vitis labrusca*). *The Plant Journal*, 49, 579-591. doi:10.1111/j.1365-313X.2006.02987.x

Hayasaka Y., Black C.A., Hack J. and Smith P., 2017. Structural characterization of reaction products of caftaric acid and bisulfite present in a commercial wine using high resolution mass spectrometric and nuclear magnetic resonance techniques. *Food Chemistry*, 230, 99-107. doi:10.1016/j.foodchem.2017.03.005

Jiménez L., Angulo V., Ramos E., De la Torre J.L. and Ferrer A., 2006. Comparison of various pulping processes for producing pulp from vine shoots. *Industrial Crops and Products*, 23, 122-130. doi:10.1016/j.indcrop.2005.05.001

Jiménez Gómez S., Cartagena Causapé M.C. and Arce Martínez A., 1993. Distribution of nutrients in anaerobic digestion of vine shoots. *Bioresource Technology*, 45, 93-97. doi:10.1016/0960-8524(93)90096-T

Jones G.V., White M.A., Cooper O.R. and Storchmann K., 2005. Climate change and global wine quality. *Climatic Change*, 76, 319-343. doi:10.1007/s10584-005-4704-2

Kritzinger E.C., Bauer F.F. and du Toit W.J., 2013. Role of glutathione in winemaking: A review. *Journal of Agricultural and Food Chemistry*, 61, 269-277. doi:10.1021/jf303665z

Lester M.R., 1995. Sulfite sensitivity: significance in human health. *The Journal of the American Collage of Nutrition*, 14, 229-232. doi:10.1080/07315724.1995.10718500

Margalit Y., 2004. Concepts in Wine Chemistry. The Wine Appreciation Guild, San Francisco.

Martínez-Gil A.M., Angenieux M., Pardo-García A.I., Alonso G.L., Ojeda H. and Salinas M.R., 2013. Glycosidic aroma precursors of Syrah and Chardonnay grapes after an oak extract application to the grapevines. *Food Chemistry*, 138, 956-965. doi:10.1016/j.foodchem.2012.11.032

Martínez-Gil A.M., Garde-Cerdán T., Zalacain A., Pardo-García A.I. and Salinas M.R., 2012. Applications of an oak extract on Petit Verdot grapevines. Influence on grape and wine volatile compounds. *Food Chemistry*, 132, 1836-1845. doi:10.1016/j.foodchem.2011.12.016

Martínez-Gil A.M., Garde-Cerdán T., Martínez L., Alonso G.L. and Salinas M.R., 2011. Effect of oak extract application to verdejo grapevines on grape and wine aroma. *Journal of Agricultural and Food Chemistry*, 59, 3253-3263. doi:10.1021/jf104178c

Mendívil M.A., Muñoz P., Morales M.P., Juarez M.C. and García-Escudero E., 2013. Chemical characterization of pruned vine shoots from la Rioja (Spain) for obtaining solid bio-fuels. *Journal of Renewable and Sustainable Energy*, 5, 033113. doi:10.1063/1.4808043

Pardo-García A.I., Martínez-Gil A.M., Cadahía E., Pardo F., Alonso G.L. and Salinas, M.R., 2014. Oak extract application to grapevines as a plant biostimulant to increase wine polyphenols. *Food Research International*, 55, 150-160. doi:10.1016/j.foodres.2013.11.004

Portilla O.M., Rivas B., Torrado A., Moldes A.B. and Domínguez J.M., 2008. Revalorisation of vine trimming wastes using *Lactobacillus acidophilus* and *Debaryomyces hansenii*. *Journal of the Science of Food and Agriculture*, 88, 2298-2308. doi:10.1002/jsfa.3351

Portu J., Santamaría P., López-Alfaro I., López R. and Garde-Cerdán T., 2015. Methyl jasmonate foliar application to Tempranillo vineyard improved grape and wine phenolic content. *Journal of Agricultural and Food Chemistry*, 63, 2328-2337. doi:10.1021/jf5060672

Ribéreau-Gayon P., Glories Y., Maujean A. and Dubourdieu D., 2006. *Handbook of Enology: The Chemistry of Wine Stabilization and Treatments*. John Wiley & Sons, Chichester, England. doi:10.1002/0470010398

Sánchez-Gómez R., Garde-Cerdán T., Zalacain A., Garcia R., Cabrita M.J. and Salinas M.R., 2016a. Vine-shoot waste aqueous extract applied as foliar fertilizer to grapevines: Effect on amino acids and fermentative volatile content. *Food Chemistry*, 197, 132-140. doi:10.1016/j.foodchem.2015.10.034

Sánchez-Gómez R., Zalacain A., Pardo F., Alonso G.L. and Salinas M.R., 2016b. An innovative use of vine-shoots residues and their *"feedback"* effect on wine quality. *Innovative Food Science & Emerging Technologies*, 37, 18-26. doi:10.1016/j.ifset.2016.07.021

Sánchez-Gómez R., Zalacain A., Alonso G.L. and Salinas M.R., 2016c. Effect of vine-shoots toasting on the generation of high added value volatiles. *Flavour and Fragance Journal*, 31, 293-301. doi:10.1002/ffj.3313

Sánchez-Gómez R., Zalacain A., Alonso G.L. and Salinas M.R., 2016d. Effect of toasting on non-volatile

© 2020 International Viticulture and Enology Society - IVES

and volatile vine-shoots low molecular weight phenolic compounds. *Food Chemistry*, 204, 499-505. doi:10.1016/j.foodchem.2016.02.137

Sánchez-Gómez R., Zalacain A., Pardo F., Alonso G.L. and Salinas M.R., 2017. Moscatel vine-shoot extracts as a grapevine biostimulant to enhance wine quality. *Food Research International*, 98, 40-49. doi:10.1016/j.foodres.2017.01.004

Sánchez-Gómez R., Zalacain A., Alonso G.L. and Salinas M.R., 2014. Vine-shoot waste aqueous extracts for re-use in agriculture obtained by different extraction techniques: Phenolic, volatile, and mineral compounds. *Journal of Agricultural and Food Chemistry*, 62, 10861-10872. doi:10.1021/jf503929v

Webb L.B., Whetton P.H., Bhend J., Darbyshire R., Briggs P.R. and Barlow E.W.R., 2012. Earlier wine-grape ripening driven by climatic warming and drying and management practices. *Nature Climate Change*, 2, 259-264. doi:10.1038/nclimate1417

 © 2020 International Viticulture and Enology Society - IVES