OENO One VINE AND WINE OPEN ACCESS JOURNAL | By IVES International Viticulture & Enology Society

DOI: https://doi.org/10.20870/oeno-one.2023.57.1.7226

OPEN ACCESS

ORIGINAL RESEARCH ARTICLE

Use of unripe grape wine as a tool for reducing alcohol content and improving the quality and oenological characteristics of red wines

Alejandro Martínez-Moreno[1,2*], Pilar Martínez-Pérez[1], Ana Belén Bautista-Ortín[1], and Encarna Gómez-Plaza[1]

[1] Department of Food Science and Technology, Faculty of Veterinary Sciences, University of Murcia, Campus de Espinardo, 30100, Spain
[2] Department of Plant Nutrition, CEBAS-CSIC, Campus Universitario de Espinardo, P.O. Box 164, 30100, Murcia, Spain

*correspondence: martinezamoreno@gmail.com

Associate editor: Jorge M. Ricardo-da-Silva

Received: 24 October 2022
Accepted: 21 December 2022
Published: 24 January 2023

ABSTRACT

In Mediterranean viticulture, climatic conditions are the main factors that determine grape composition and wine quality. Global warming is causing an increase in the accumulation of soluble solids in grapes, leading to early harvests that result in wines with high alcohol and low phenolic content and colour intensity. The aim of this research was to determine the effectiveness of mixing wines vinified from grapes with different maturation degrees from two consecutive vintages (2018 and 2019) in order to obtain wines with lower pH, lower alcohol content and higher phenolic compound concentration. The wine obtained after mixing wine from unripe grapes (URG) with second harvest (H2) wines significantly improved the physicochemical, chromatic and phenolic characteristics compared to the first harvest (H1) wine. On the other hand, the URGH2 wine had significantly lower alcohol content and pH and higher titratable acidity than the H2 wine. URGH2 maintained its colour intensity in both years and similar values of anthocyanin and tannin in 2018 compared to H2. All the wines were subjected to sensory analysis and the panelists were able to differentiate the wines in a triangle sensory test; the URGH2 wine was preferred in both years. These results show that the blend of unripe grape with wines from technologically mature grapes to reduce alcohol content and improve wine colour could be a useful tool for mitigating the problems caused by global warming in a warm and semiarid Mediterranean climate.

KEYWORDS: ripening, alcohol content, sensory analysis, phenolic compounds, chromatic composition

INTRODUCTION

Climate plays an important role in the wine regions, as it strongly determines grapevine growth, yield and berry composition, which together have an effect on wine composition and its quality. The reality of climate change has been admitted by the majority of the scientific community (IPCC, 2019). The main measurable effects are the increase in temperatures, the change in rainfall distribution and heatwaves (Mirás-Avalos and Intrigliolo, 2017). As detailed in the report of the International Panel on Climate Change (IPCC) the future scenarios predict a worsening of these extreme climate events. In this context, climate change is a challenge that must be faced by winegrowers and the wine industry, particularly in arid and semi-arid regions (Martínez-Moreno *et al*., 2022).

In recent years, global warming has shifted vine and grape physiological events, shortening the period between veraison and harvest, and producing a decoupling between the technological and phenolic maturity of grapes. This acceleration in the dynamics of grape ripening entails considerable compositional changes (Salazar Parra *et al*., 2010). As a consequence of the increase in temperatures during the vegetative cycle of the grapevine, the grape pulp matures faster, resulting in a reduction of total acidity and a high concentration of sugar content and pH in grape must (Kontoudakis *et al*., 2011). Moreover, the maturity of grape skin and seeds is known to strongly influence the phenolic composition of red wines (Marques *et al*., 2005) and to have different behavior to pulp maturity. The phenolic maturity of grapes is a parameter that is linked to the concentration of phenolic compounds (mainly anthocyanins and tannins) and the ease of their extraction from the grape skin and seeds during vinification (Saint-Cricq de Gaulejac *et al*., 1998). It follows that phenolic immature grapes have a low concentration of anthocyanin, and the extractability of proanthocyanins and anthocyanins from their skins to must during the winemaking process is low (Canals *et al.*, 2005). Consequently, it is generally thought that insufficient phenolic ripened grapes have a lower concentration of polyphenols, which are important for wine quality due to their chromatic, health-related and sensory characteristics (Fulcrand *et al*., 2006).

Wine colour is one of the most important parameters of red wines, as it is the first attribute to be perceived by the tasters and is directly associated with its quality. Usually, wine colour is associated with the concentration of phenolic compounds in the grapes; therefore, harvesting fruit at their optimal levels of phenolic and technology maturity is the first step to producing high-quality wines (Bautista-Ortín *et al.*, 2006).

Structured and intensity coloured red wines are known to be highly considered by consumers; for this reason, it is of interest to the wine industry to harvest the grape when it is as ripe as possible, with the objective of maximising the phenolic extraction of the grapes. However, due to climate change and rising temperatures, winemakers are faced with a dilemma: either they harvest the grapes with adequate sugar concentration and acidity, but with inadequate skin and seed phenolic maturity (which could result in lower colour intensity and high astringent wines due to the low extractability of skin phenolic compounds and the high extraction of seed tannins in unripe grapes during vinification), or they wait for optimum phenolic maturity and accept that they will obtain wines with high alcohol content and pH, and low total acidity (Jones *et al*., 2005).

In this context, the wine sector and scientific community have proposed several possible techniques or processes to mitigate the negative effects of global warming on grapes and wines. According to Varela *et al*. (2015), these can be grouped as viticultural or winemaking practices. The simplest viticultural technique is to harvest fruit at an early stage of maturity. Nevertheless, if the goal is to obtain premium quality wines, this is not a good solution, because the grapes might not have reached an adequate phenolic composition, which could result in the production of unbalanced wines with low colour intensity (Kontoudakis *et al*., 2011). Another viticultural technique is double-pruning, which forces bud growth during the spring and summer, shifting berry ripening to cooler periods of the season, and thus improving the phenolic composition and chromatic parameters of the berries and ensuring a more balanced sugar/phenolic content (Martínez-Moreno *et al*., 2019). It is also possible to introduce new cultivars with delayed grape sugar accumulation and acid combustion (Schultz, 2000), as well as to implement and modify cultural techniques, such as post-veraison leaf removal, post bud-break pruning and severe trimming (Martínez-Moreno *et al*., 2019).

In the wine industry, several oenological techniques have been proposed for making wines with a reduced alcohol content; for example, i) the addition of water and mineral acids to the must before fermentation begins; this reduces sugar concentration, but generally has a negative effect on the resulting wines, because it can reduce must acidity, lower the concentrations of phenolics compounds and have a negative impact on wine appearance and sensory attributes (Gardner *et al*., 2020); furthermore, this practice is illegal in Spain and in most wine producing countries (Sam *et al*., 2021); ii) the blending of high alcohol wines (16–17 % ABV) with low-alcohol wines (12–13 % ABV); this usually requires very high volumes of low-alcohol wines to significantly reduce alcohol content; iii) the addition of glucose oxidase; this enzyme has two drawbacks: it can generate a gluconic acid in the wines and consuming oxygen as a substrate, which can reduce the quality of the wines (Pickering *et al*., 1998); iv) the use of yeasts (*Saccharomyces* or non-*Saccharomyces* yeast) with a lower ethanol production during the winemaking process (Ciani and Ferraro, 1996); and v) in finished wines, physical methods are the most commonly used, such as membrane techniques, vacuum distillation and supercritical fluid extraction (Gil *et al*., 2013); of all the physical methods used in the wine industry, the most common are the spinning cone column and the reverse osmosis system (Pickering, 2000), which are effective, but involve expensive and bulky equipment, and their effects on wine quality are still not very clear (Kontoudakis *et al*., 2011).

Therefore, in recent years, other winemaking techniques have been proposed to mitigate the elevated alcohol content in red wines resulting from global warming. Concretely, the blending of grapes, must or wines from unripe and fully mature grapes has generated much interest in the wine sector. Several studies with different Vitis vinifera varieties (Shiraz, Malbec, Cabernet-Sauvignon, Grenache, Pinot noir and Tannat) have been conducted with the aim of investigating the effect of blending unripe grapes low-alcohol wine with musts from well-ripened grapes (Kontoudakis *et al*., 2011; Schelezki *et al*., 2020; Piccardo *et al*., 2019; Fanzone *et al*., 2020). These authors obtained mixed wines with higher total acidity and lower alcohol and pH content. Moreover, blended unripe grape must with well-ripened grape must had a positive impact on phenolic composition, especially in terms of anthocyanins and tannins, which had great repercussions on the final colour and mouthfeel sensations. These results indicate the potential benefits of the consecutive harvest to reduce alcohol and pH and increase phenolic content in wines. Although similar studies have been conducted in the past (Kontoudakis *et al*., 2011; Piccardo *et al*., 2019; Schelezki *et al*., 2018), none of these studies have involved the blending of low-alcohol wines with wines from very ripe grapes; however, all the authors removed a volume of juices of very ripe grapes and replaced the same volume with low-alcohol wine and carryied out the alcoholic fermentation jointly (low-alcohol wine and full-mature must). Moreover, these studies were not conducted on Monastrell wines, whose grapes are genetically characterised by a more rigid cell-wall structure which makes the extractability of phenolic compounds during vinification more difficult (Ortega-Regules *et al*., 2006). To our knowledge, there is to date no published information on the effect of mixed low-alcohol wine with fully mature grape wines of the red variety Monastrell.

The present study focuses on developing a winemaking strategy for reducing the alcohol content in wines without altering or even improving their phenolic composition and chromatic parameters. This process involves the use of fermented unripe grape berries to obtain a highly acidic "wine" with low alcohol content. This wine will be used later to blend with the wine obtained from grapes that have reached the optimum phenolic maturity in the vineyard. Therefore, the objective of this research was to obtain wines with better phenolic and anthocyanins concentrations (more intense colour), high acidity, and lower alcohol content.

MATERIALS AND METHODS

1. Harvesting and winemaking

This study was carried out during two consecutive vintages (2018–2019) in a commercial vineyard located in Jumilla, Spain. *Vitis vinifera* L. cv Monastrell grapes from the same vineyard were used to obtain three different wines. To obtain the unripe grape wine (URG), 150 kg of green clusters were thinned at the grape phenological stage number 33 in the BBCH-scale (Lorenz *et al*., 1995), and taken to an experimental winery at the University of Murcia (UMU), where the grapes were crushed and destemmed and pressed in a hydraulic wine press to obtain around 50 L of grape must. The must was sulphited ($K_2S_2O_5$) with 70 mg/L of SO_2 and placed in a 50-L steel tank. The URG (approximately 5 °Be) was left to settle for 24 hours, racked in another tank and inoculated (40 g/hL) with selected yeast (Enartis Ferm Perlage, Ciudad Real, Spain). Alcoholic fermentation (AF) was carried out at 21 ± 1°C. When the AF had finished, the wine was racked and sulphited (70 mg/L SO2) and stored at 2 °C. The URG comprised the following analytical parameters for 2018 and 2019: 3.9 % and 3.4 % (v/v) of ethanol, a total acidity of 17.5 and 19.6 g of tartaric acid per liter and a pH of 2.98 and 2.85.

Grapes from the same vineyard were harvested at two subsequent moments of the ripening stage: the first harvest (H1) and second harvest (H2) were carried out when the potential degree of alcohol was 13° and 14.5° respectively. 150 kg of H1 grapes and 300 kg of H2 were hand-picked and taken to an experimental winery at the UMU where the grapes were crushed, destemmed and randomly distributed into 50 L stainless steel tanks: three tanks (H1) and six tanks (H2). All the tanks were sulphited (70 mg/L of SO_2) and inoculated with 30 g/hL of selected yeast (EnartisFerm Q7, Ciudad Real, Spain). All the vinifications were conducted at 24 ± 1°C until the end of fermentation and monitored daily for temperature and density of the must-wine. A manual punch down was carried out daily for seven days during maceration. At the end of maceration, the free-run and pressed wines were combined and returned to clean tanks at room temperature until the end of fermentation (residual sugar < 2 g/L). After fermentation, three 50-L tanks from H1 and H2 were used as the Control wine. The wine of the other three resulting tanks was mixed with elaborated unripe grape wine to obtain the URGH2 wine: specifically, the URG wine was added to H2 wine tanks, until it had reached the same alcoholic degree obtained in H1 (13 % v/v).

2. Analytical determinations

The wines were characterised by measuring the alcohol content, pH, total acidity and acetic acid according to European Community methods. Spectrophotometric parameters were determined using a Shimadzu UV-1603 spectrophotometer (Shimadzu, Deutschland), and colour intensity (CI) was calculated as the sum of absorbance at 620, 520, and 420 nm. Total and polymeric anthocyanins were determined following the method of Ho *et al*. (2001). Total phenol index (TPI) was calculated by measuring wine absorbance at 280 nm, according to Ribéreau-Gayon *et al*. (2006). Total tannins were determined following the methylcellulose precipitation method described by Smith (2005).

Determination of tannins by the phloroglucinolysis method was carried out according to Busse-Valverde *et al*. (2010) from an optimisation of the method proposed by Pastor del Rio and Kennedy (2006). For the wine sample preparation, 5 mL of wine was evaporated in a centrivap concentrator (Labconco, Kansas City, MO, United States), dissolved in 3 mL of water, and then passed through a C18-SPE column (1 g, Waters, Milford, MA, United States). The compounds of interest were eluted with 10 mL of methanol, evaporated and then dissolved in 1 mL of methanol. The analyses of tannins were done by depolymerisation of the molecule using the phloroglucinol reagent. The depolymerised samples (10 µL injection volume) were analysed by HPLC in order to determine total tannin content, apparent mean degree of polymerisation (mDP) and percentage of galloylation (% Gal).

The chromatic parameters and phenolic compounds were determined at bottling and after twelve months of aging in the bottle.

3. Sensory analysis

Monastrell red wines (elaborated in 2018-2019) were subjected to a triangular and descriptive sensory test at two different moments: i) at bottling, and ii) twelve months after bottling. These were carried out by ten staff members with experience in wine sensory analysis and interest in the project. Prior to the sensory analysis, the wine from the three different replications for each treatment was pooled to obtain a representative sample and to avoid differences among the replications. Regarding the triangular test, three samples were tested, two of which were identical and one different. Panelists selected the sample that they considered different and liked the best.

Forty mL of each wine was poured into coded glasses 5 min before evaluation. The glasses were presented to the panel in a sensory room that was kept at 21 °C and free of unusual odours. Each panelist sat in an individual isolated booth illuminated with white light. The intensity of each attribute was rated on a scale of zero to ten, with a score of zero indicating that the descriptor was not perceived. Data from all the panel for all samples were used in the analysis.

4. Statistics

The significance of the treatments was assessed using a one-way analysis of variance (ANOVA). The means were separated by Tukey's Multiple Range Test ($p < 0.05$) using IBM SPSS Statistics 26 (Armonk, New York, USA).

RESULTS AND DISCUSSION

1. General composition of wines

Table 1 shows the physicochemical parameters of Monastrell wines from grapes harvested at 22.6 ºBrix (H1), technologically well-matured grapes at 24.4 ºBrix (H2) and the blending of URG and H2 wines (URGH2). As expected, the URGH2 wine had a lower ethanol content and pH than its corresponding control (H2). In fact, URGH2 obtained the lowest values for ethanol content and pH of all the wines produced, but showed higher titratable acidity. The results are also in accordance with those of Piccardo *et al*. (2019), who substituted grape must from fully mature grapes by must from unripe grapes from two red varieties (Pinot noir and Tannat). Other red varieties such as Cabernet-Sauvignon, Merlot and Bobal showed similar results when the musts were mixed with green musts obtained from cluster thinning (Kontoudakis *et al*., 2011). Therefore, the results obtained in this study are quite conclusive and show that adding some "green" wine to fully mature grape wines may be a useful tool for obtaining reduced alcohol wines with lower pH and higher total acidity.

2. Chromatic Characteristics

The phenolic and chromatic characteristics of the wines are shown in Table 2. In both years at bottling, the URGH2 wine showed a significant improvement in terms of chromatic characteristics compared to H1, with higher colour intensity (CI), total phenol index (TPI), total anthocyanin (TAnt), polymeric anthocyanins (PolAnt) and total tannin content (TT); however, it had the same alcohol content. Moreover, these significant differences between the wines (H1 and URGH2) were maintained after twelve months of bottle-aging in terms of all the chromatic parameters. When the URGH2 wine was compared with the H2 wine, as expected the addition of a small volume of "green" wine to the wine obtained from fully mature grapes barely modified the chromatic parameters, since, in general, the URGH2 wine showed chromatic characteristics more similar to H2 than to H1 wines. The results obtained in terms of chromatic characteristics agree with those obtained in the study carried out by Piccardo *et al*. (2019), who substituted ripe grape must with grape must with a lower level of maturity in red varieties (Tannat and Pinot noir).

In both years of the study, the colour intensity of H2 and URGH2 wines was significantly higher than that of H1 wines, probably because the lower pH of these wines increased the proportion of the anthocyanins in the flavylium cation form (Glories, 1984). In 2019, all the wines (H1, H2 and URGH2) showed an large decrease in IC values after 12 months in the bottle, possibly due to a greater loss of phenolic compounds by precipitation, as reflected by the larger decrease observed in the TPI values.

Regarding anthocyanin concentrations, in 2018, H2 and URGH2 wines obtained similar concentrations (489 and 500 mg/L). Moreover, these concentrations were significantly higher than that of H1 wine (325 mg/L). In the second year of the trial, the same trend was observed, with a higher anthocyanin concentration in the URGH2 and H2 wines compared to the H1 wine. This higher concentration of anthocyanins correlates with the high level of grape maturity used to vinify H2 wine (Jordão *et al*., 1998). After 12 months of aging, the total anthocyanin concentration in all the wines was reduced significantly in both years of study; this may be due to a constant decline in the concentration of monomeric anthocyanin during the aging process, which is related to different mechanisms, such as their precipitation with proteins, polysaccharides or condensed tannins, their degradation and oxidation, and the progressive and irreversible formation of more complex and stable anthocyanin derived pigments (Costa *et al*., 2014). In both years, the polymeric anthocyanin content in H2 and

TABLE 1. Physico-chemical characteristics of Monastrell musts (°Brix, Tac and pH) and wines at bottling.

	Sample	°Brix*	Tac* (g/L)	pH*	Alcohol (%v/v)	pH	Tac (g/L)	Ace (g/L)
2018	H1	22.5	4.63	3.11	13.0 ± 0.0 a	3.57 ± 0.05 c	5.7 ± 0.02 b	0.25 ± 0.01 a
	URGH2	–	–	–	12.9 ± 0.0 a	3.19 ± 0.02 a	6.2 ± 0.09 c	0.32 ± 0.01 b
	H2	24.3	4.21	3.09	14.1 ± 0.0 b	3.37 ± 0.01 b	5.3 ± 0.06 a	0.37 ± 0.01 c
2019	H1	22.7	4.59	3.62	13.1 ± 0.01 a	4.01 ± 0.04 c	5.7 ± 0.17 a	0.39 ± 0.01 c
	URGH2	–	–	–	12.9 ± 0.02 a	3.53 ± 0.01 a	6.7 ± 0.32 b	0.37 ± 0.01 b
	H2	24.5	5.26	3.72	14.2 ± 0.05 b	3.78 ± 0.03 b	5.89 ± 0.14 a	0.32 ± 0.01 a

*Parameter measured in musts. Tac: Total acidity. Ace: Acetic acid. H1: First harvest. H2: second harvest. URGH2: a mixture of "green" and H2 wines. Different letters within the same column and for each year indicate significant differences ($p < 0.05$).

TABLE 2. Chromatic and phenolic characteristics of Monastrell wines at bottling and after 12 months in bottle.

		Sample	CI	TPI	Tant (mg/L)	PolAnt (mg/L)	TT (MC) (mg/L)
At bottling	2018	H1	7.3 ± 0.09 a	46.7 ± 0.24 a	325 ± 11.61 a	25.4 ± 0.14 a	959 ± 40.33 a
		URGH2	14.9 ± 0.41 b	54.9 ± 1.09 b	484 ± 15.99 b	32.9 ± 0.07 b	1567 ± 43.62 b
		H2	15.4 ± 0.54 b	56.7 ± 0.08 c	500 ± 2.39 b	34.3 ± 0.24 c	1514 ± 25.35 b
	2019	H1	8.3 ± 0.02 a	50.5 ± 0.81 a	440 ± 5.68 a	22.8 ± 0.21 a	1246 ± 82.37 a
		URGH2	12.2 ± 0.42 b	60.0 ± 0.58 b	496 ± 19.85 b	27.8 ± 1.12 b	1625 ± 87.87 b
		H2	12.1 ± 0.54 b	62.8 ± 1.54 c	539 ± 11.73 c	31.9 ± 4.56 b	1414 ± 130.98 a
12 months in bottle	2018	H1	6.8 ± 0.02 a	43.6 ± 2.14 a	273 ± 13.30 a	31.2 ± 0.11 a	1180 ± 30.31 a
		URGH2	14.6 ± 0.35 b	51.8 ± 0.48 b	418 ± 4.24 b	38.4 ± 0.15 b	1751 ± 58.14 c
		H2	14.9 ± 0.15 c	53.4 ± 0.69 c	457 ± 5.37 c	40.3 ± 1.02 c	1600 ± 20.00 b
	2019	H1	6.1 ± 0.04 a	42.1 ± 0.33 a	225 ± 2.41 a	36.1 ± 0.21 a	1298 ± 60.43 a
		URGH2	10.1 ± 0.05 b	55.8 ± 0.21 b	335 ± 4.85 b	41.1 ± 1.12 b	1777 ± 39.37 c
		H2	10.3 ± 0.03 b	57.7 ± 0.30 c	349 ± 4.51 c	42.2 ± 1.10 b	1616 ± 60.26 b

CI: color intensity. TPI: total phenol index. TAnt: total anthocyanins. PolAnt: polymeric anthocyanins. TT (MC): total tannins (determined by the methylcellulose method). H1: First harvest. H2: second harvest. URGH2: mixture of "green" and H2 wines. Different letters within the same column and for each year and analysis time indicate significant differences ($p < 0.05$).

URGH2 was higher than in H1. Moreover, in 2019 there were no significant differences in the concentration of polymeric anthocyanins in the H2 and URGH2 wines. The concentration of polymeric anthocyanins in wines is closely related to a lower astringency and higher colour stability of the wine, and thus its quality. Therefore, the URGH2 wine was able to produce a long-lasting and stable colour similar to that of the H2 wine, with lower alcohol content and pH. Previous research has suggested that the A:T ratio plays an important role in polymeric pigment synthesis, because both tannin and anthocyanin are needed for the development of polymeric pigments (Fulcrand *et al*., 2004). In contrast, a polymeric pigment trial carried out by Merrell *et al*. (2017) to study the impact of grape maturity on these pigments showed the A:T ratio to be a very poor predictor of polymeric pigment concentration. Being the best predictor to determine polymeric pigment, the initial wine anthocyanin content, which usually increases with the degree of ripening of grapes. These results are in agreement with those obtained in our trial, the concentration of anthocyanin being highest in the URGH2 and H2 wines compared to the H1 wines. Finally, the total concentration of tannins was measured by precipitation with metyl-cellulose (Table 2). In both years, tannin concentrations in the URGH2 and H2 wines were significantly higher than in the H1 wines. In 2019, URGH2 also showed the highest total tannin concentration (1625 mg/L). These data confirm that the riper grapes produced wines with a higher concentration of tannins. While ripening has a straightforward impact on anthocyanin development, the same is not necessarily true for tannin development (Rousserie *et al*., 2020). Concerning the concentration of tannins in berries, skin tannin remains relatively stable throughout ripening, while seed tannins decline slightly (Harbertson *et al*., 2002). Moreover, the extraction of these compounds cannot easily be predicted from berry ripeness, since winemaking techniques can affect the concentration of extracted phenolics in the wine (Sacchi *et al*., 2005). the Monastrell variety used in this trial is characterized by the fact that it is difficult to extract phenolic compounds from the berry skin to the must-wine; i.e., it has low extractability, as numerous studies have already shown (Bautista-Ortín *et al*., 2012; Romero-Cascales *et al*., 2005; Ortega-Regules *et al*., 2006; Ortega-Regules *et al*., 2008). Tannin extraction is complex, but this generally rises with the increase of maceration time, fermentation temperature and alcohol content (Casassa *et al*., 2013). In our study all the wines were vinified in the same way; therefore, the only variable that can differ between the wines is the alcohol content. The higher tannin concentration in the H2 and URGH2 wines can thus be explained by the higher alcohol content present during fermentative maceration compared to the H1 wine.

3. Wine tannin content and composition

The tannins were measured using the phloroglucinolysis method (Table 3). At the time of bottling, the data confirms that in both years (2018–2019), the URGH2 wines had higher tannin content than the H2 wines, the lowest concentration being found in the H1 wines. The results agree with those obtained by spectrophotometry. After 12 months of bottling the concentration of tannins decreased and, although the URGH2 wine achieved the highest values of these compounds, non-significant differences were observed between the H1 and H2 wines. This decrease in tannin concentration was not observed when these compounds were measured using the metyl-cellulose method, which is mainly due to the evolution of tannin molecules towards complex and oxidized forms that are more difficult to depolymerize by phloroglucinol reagent. With respect to the tannin composition, the results indicate that in the two years of study the mean degree of polymerisation (mDP) was similar in the H2 and URGH2 wines, which was significantly higher than in the H1 wine; this is probably due to an increase in the mDP of skin tannins during the grape ripening process (Kennedy *et al*., 2002), as well as to a better extractability of these compounds during maceration due to a higher degradation of the berry cell walls and higher ethanol content, which favours the extraction of larger molecules (Kontoudakis *et al*., 2011). The mDP values obtained in the wines were maintained after 12 months of bottling. The percentage of galoylation (% Gal) was significantly higher in URGH2 wines for both years at the time of bottling. It is known that insufficiently ripened grapes can release a large amount of seed proanthocyanidins, which are highly galloylated (Romeyer *et al*., 1985); this explains how mixing H2 wine with a wine from unripe grapes increased the galloylated tannin content in the URGH2 wine. These values were maintained after 12 months of bottling in 2018. However, when the 2019 wines were analysed after 12 months of bottling, % Gal was significantly higher in the H2 wines. The concentrations of epigallocatechin (EGC) were higher than that of epicatechin gallate (ECG) in all wines, as was to be expected. The URGH2 treatment obtained the highest values of EGC and ECG in both years of the study and at both time points (at bottling and after 12 months of aging), probably as a consequence of the addition of "green" wine with a high concentration of tannins (data not shown). It is known that grapes contain the highest concentrations of tannin in early fruit development (Downey *et al*., 2003), with high concentrations of epigallocatechin and epicatechin gallate subunits (Bautista-Ortín *et al*., 2013). In unripe grapes, the lignification of seeds is low, and it is easy for the solvent (alcohol) to access the inner integument and thus extract a large number of tannins (Bautista-Ortín *et al*., 2012). The wines from the H2 treatment obtained a higher concentration of EGC and ECG compared to H1 at bottling due to the higher alcoholic content of the H2 wine favoring the higher extraction of these compounds. This trend was maintained for the EGC values after 12 months of aging. In contrast, the ECG values of the H2 and H1 wines were similar after twelve months of bottling.

4. Sensory Analysis

To determine whether the results observed in the different wines (H1, H2 and URGH2) could be detected at the sensory level, the wines were tested at two different times (bottling and 12 months of aging in bottle) using descriptive sensory analysis (Figures 1 and 2) and the triangular test (Table 4).

TABLE 3. Concentration and composition of tannins determined by the phloroglucinolysis method in Monastrell wines at bottling and after 12 months in bottle.

		Sample	TT (mg/L)	mDP	%Gal	EGC (µM)	ECG (µM)
At bottling	2018	H1	700 ± 28 a	4.53 ± 0.13 a	2.65 ± 0.07 a	328 ± 11.14 a	62 ± 3.42 a
		URGH2	993 ± 25 c	4.90 ± 0.16 b	3.71 ± 0.21 c	438 ± 15.30 c	123 ± 9.40 c
		H2	817 ± 28 b	4.85 ± 0.11 b	2.92 ± 0.06 b	394 ± 17.07 b	80 ± 2.67 b
	2019	H1	522 ± 17 a	3.82 ± 0.19 a	2.37 ± 0.03 a	176 ± 12.46 a	42 ± 0.92 a
		URGH2	997 ± 26 c	4.80 ± 0.07 b	3.80 ± 0.24 c	426 ± 14.31 c	127 ± 10.79 c
		H2	640 ± 8 b	4.80 ± 0.13 b	3.46 ± 0.04 b	333 ± 10.96 b	74 ± 0.75 b
12 months in bottle	2018	H1	625 ± 35 a	4.14 ± 0.02 a	2.66 ± 0.06 a	227 ± 12.32 a	61 ± 4.74 a
		URGH2	840 ± 28 b	4.63 ± 0.18 b	3.07 ± 0.04 b	397 ± 11.45 c	97 ± 3.70 b
		H2	695 ± 31 a	4.43 ± 0.12 b	2.63 ± 0.09 a	327 ± 20.39 b	65 ± 4.67 a
	2019	H1	425 ± 5 a	4.44 ± 0.05 a	4.80 ± 0.04 a	127 ± 18.40 a	84 ± 0.15 a
		URGH2	837 ± 68 b	4.82 ± 0.06 b	5.23 ± 0.04 b	335 ± 26.58 c	163 ± 13.04 c
		H2	509 ± 29 a	4.89 ± 0.17 b	6.29 ± 0.10 c	260 ± 10.25 b	106 ± 4.48 b

TT: total tannins. mDP: mean degree of polymerization. %Gal: percentage of galloylation. EGC: epigallocatechin. ECG: epicatechin gallate. H1: First harvest. H2: second harvest. URGH2: mixture of "green" and H2 wines. Different letters within the same column and for each year and analysis time indicate significant differences ($p < 0.05$).

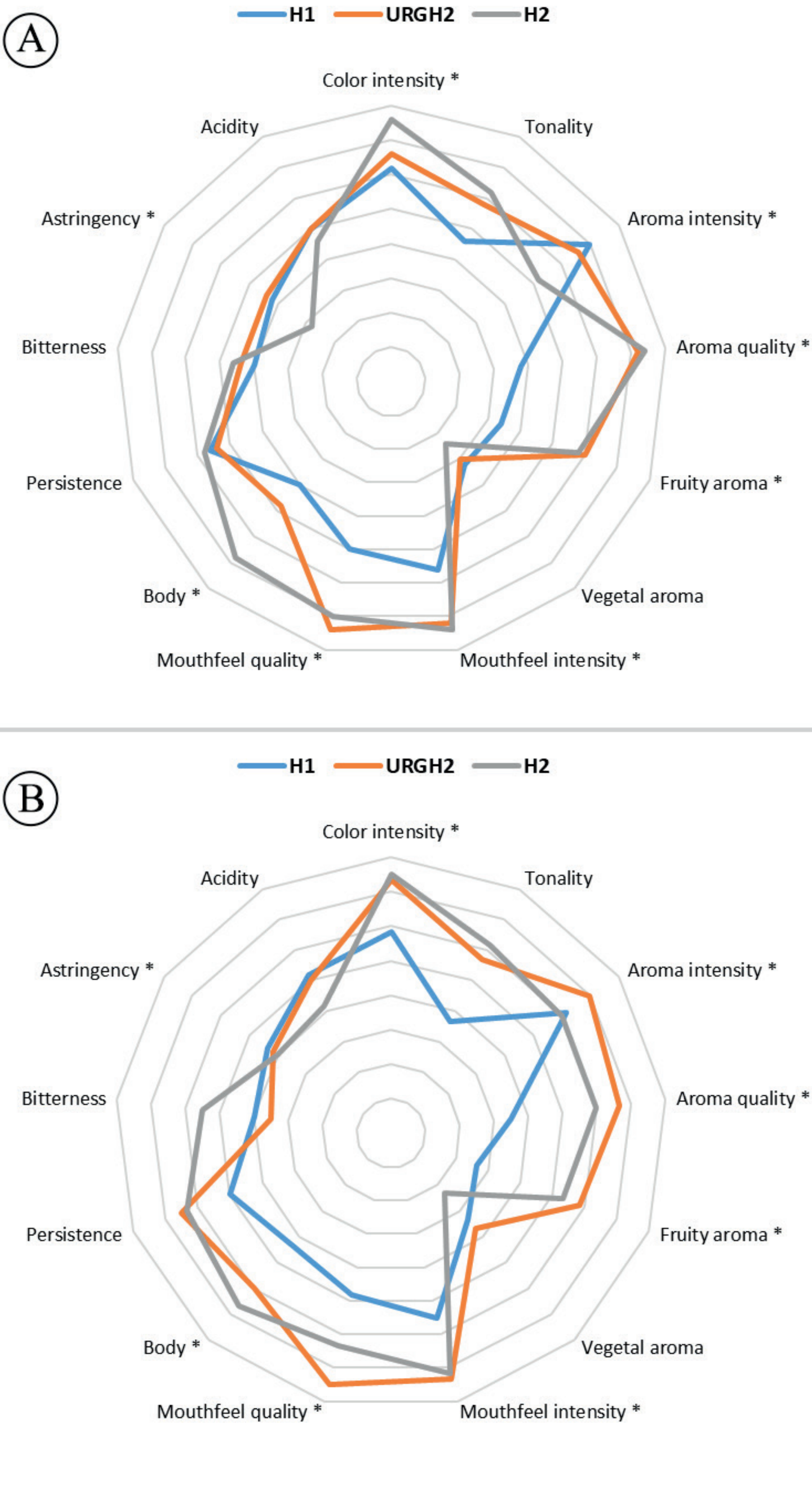

FIGURE 1. Sensory analysis of three different wines (2018) at bottling (A) and after 12 months of bottling (B) (* denotes significant differences $p < 0.05$).

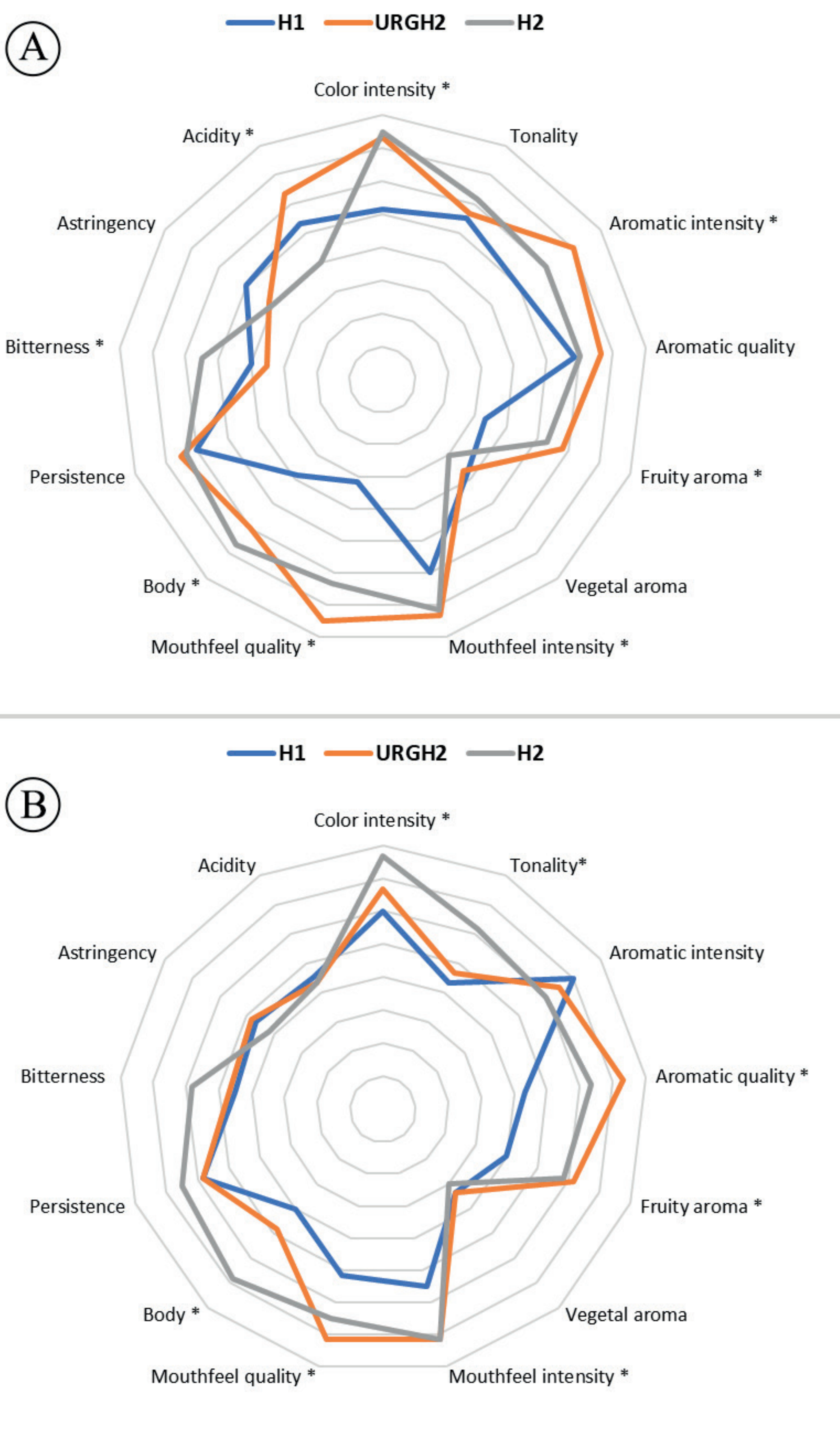

FIGURE 2. Sensory analysis of three different wines (2019) at bottling (A) and after 12 months of bottling (B) (* denotes significant differences $p < 0.05$).

TABLE 4. Sensory analysis (triangular discriminative and preference test) for different Monastrell wines produced in this study at bottling and after 12 months in bottle.

		Triangular test	Positive identifications	Preferences
At bottling	2018	H1 vs. URGH2	10/10 ***	URGH2 (9)
		H2 vs. URGH2	4/10	—
	2019	H1 vs. URGH2	8/10 **	URGH2 (6)
		H2 vs. URGH2	7/10 *	URGH2 (4)
12 months in bottle	2018	H1 vs. URGH2	10/10 ***	URGH2 (6)
		H2 vs. URGH2	4/10	—
	2019	H1 vs. URGH2	9/10 **	URGH2 (7)
		H2 vs. URGH2	8/10 *	URGH2 (4)

H1: First harvest. H2: second harvest. URGH2: mixture of "green" and H2 wines. Asterisks within the same column indicate significant differences * $p < 0.05$ **, $p < 0.01$ and *** $p < 0.001$.

Figure 1 shows the sensory attributes of different wines (H1, H2 and URGH2) produced in 2018 at bottling (Figure 1a) and after 12 months in the bottle (Figure 1b). For most of the analysed attributes, both the H2 and URGH2 wines received significantly higher scores than the H1 wine. However, as regards aroma intensity and astringency, URGH2 wine received similar scores to the H1 wine, with both obtaining significantly higher scores than the H2 wine. Moreover, the H2 wine achieved the highest colour intensity, followed by URGH2. After 12 months in the bottle, the URGH2 wine score improved significantly, obtaining the highest rates for the attributes of aroma intensity, aroma quality, fruity aroma and mouthfeel quality. Moreover, the panelists gave similar scores for the colour intensity of the URGH2 and H2 wines. The results obtained for descriptive sensory analysis of the wines in 2019 are shown in Figure 2. At the time of bottling, the panelists found that the colour and mouthfeel intensity of the H2 and URGH2 wines were significantly higher than that of the H1 wine. In addition, the URGH2 wine obtained the highest scores for aromatic intensity, aromatic quality, fruity aroma, mouthfeel quality and acidity. It is necessary to highlight that the panelists were not able to find significant differences in astringency and herbaceous notes among the wines. After 12 months of bottling, the URGH2 and H2 wines showed higher colour intensity, body, aromatic quality, fruity aroma and mouthfeel intensity than the H1 wine ($p < 0.05$). Moreover, the panelists were not able to find any differences in the vegetal notes among the wines.

In the triangular test at the time of bottling in 2018, and as expected, all ten panelists were able to distinguish the URGH2 wine from the H1 wine, because the H1 grapes were less ripe than the URGH2 grapes. Furthermore, nine out of ten panelists preferred the URGH2 wine. However, when the URGH2 wine was compared with the H2 wine, the panelists were not able to significantly differentiate between them; only four out of ten tasters were able to positively identify the samples. These results are in accordance with those obtained in the descriptive test, which showed the similarity of the H2 and URGH2 wines in terms of parameters such as aroma and mouthfeel quality, fruity aroma and mouthfeel intensity. Given the different analytical results obtained for each of the two wines, this was unexpected; nevertheless, colour intensity was similar in both wines and it is well known that colour has a significant effect on the sensory appreciation of flavour and texture (Stillman, 1993). The results of the triangular analysis performed after 12 months of aging were consistent with these results (URGH2-H1 and URGH2-H2), although the number of preferences decreased from 9 to 6 when H1 and URGH2 were evaluated. In 2019, at bottling, the URGH2 wine was clearly differentiated from H1 wine and was preferred by panelists. After 12 months in the bottle, the panelists were able to significantly distinguish between the wines in both triangular tests and they preferred the URGH2 wine. The tasters were able to distinguish the wines much better when H1 vs URGH2 were compared than when H2 vs URGH2 were compared, as was expected due to the large differences in the analysed physical-chemical and chromatic parameters. Our results are in agreement with the results obtained by Kontoudakis *et al*. (2011) for red wines of different varieties (Bobal, Merlot and Cabernet-Sauvignon), which the tasters distinguished just as well or better when they were tested after 12 months of bottling. Therefore, the differences found by the panelists at the time of bottling between the different wines seem to be not only maintained over time but also accentuated.

CONCLUSIONS

The results obtained in this study show that the URGH2 wine (made by blending wine from unripe grapes and wine from fully mature grapes), had a reduced alcohol content and improved physicochemical parameters compared to the H2 wine; particularly noteworthy was the significant increase in total acidity. The chromatic characteristics of the URGH2 wine significantly improved compared to the H1 wine, obtaining higher values for colour intensity, total phenolic index, anthocyanin and tannin content; moreover, these parameters were very similar to those of the H2 wine. The blending of wines from grapes harvested at different ripening stages was found to improve the sensorial quality of the wines, enhancing wine colour, fruity aroma and mouthfeel characteristics. Therefore, this procedure could be a useful tool for mitigating the negative impacts of climate change on semi-arid Mediterranean viticulture, as it reduces alcohol content and pH simultaneously without impairing the chromatic characteristics of the wines. Moreover, the use of this technique is easy to apply in most of wineries and does not involve any additional costs or equipment.

REFERENCES

Bautista-Ortín, A. B., Fernández-Fernández, J. I., López-Roca, J. M., & Gómez-Plaza, E. (2006). The effect of grape ripening stage on red wine color. *OENO One*, *40*(1), 15-24. https://doi.org/10.20870/oeno-one.2006.40.1.879

Bautista-Ortín, A., Rodríguez-Rodríguez, P., Gil-Muñoz, R., Jiménez-Pascual, E., Busse-Valverde, N., Martínez-Cutillas, A., López-Roca, J., & Gómez-Plaza, E. (2012). Influence of berry ripeness on concentration, qualitative composition and extractability of grape seed tannins. *Australian Journal of Grape and Wine Research*, *18*(2), 123–130. https://doi.org/10.1111/j.1755-0238.2012.00178.x

Bautista-Ortín, A. B., Busse-Valverde, N., Rodriguez-Rodriguez, P., Jimenez-Pascual, E., Gil-Muñoz, R., & Gómez-Plaza, E. (2013). Qualitative composition and extractability of grape skin tannins during the ripening period. Role of the extraction solvent. *OENO One*, *47*(2), 137-143. https://doi.org/10.20870/oeno-one.2013.47.2.1539

Busse-Valverde, N., Gómez-Plaza, E., López-Roca, J. M., Gil-Muñoz, R., Fernández-Fernández, J. I., & Bautista-Ortín, A. B. (2010). Effect of Different Enological Practices on Skin and Seed Proanthocyanidins in Three Varietal Wines. *Journal of Agricultural and Food Chemistry*, *58*(21), 11333–11339. https://doi.org/10.1021/jf102265c

Canals, R., Llaudy, M. C., Valls, J., Canals, J. M., & Zamora, F. (2005). Influence of Ethanol Concentration on the Extraction of Color and Phenolic Compounds from the Skin and Seeds of Tempranillo Grapes at Different Stages of Ripening. *Journal of Agricultural and Food Chemistry*, *53*(10), 40194025. https://doi.org/10.1021/jf047872v

Casassa, L. F., Larsen, R. C., Beaver, C. W., Mireles, M. S., Keller, M., Riley, W. R., Smithyman, R., & Harbertson, J. F. (2013). Impact of Extended Maceration and Regulated Deficit Irrigation (RDI) in Cabernet Sauvignon Wines : Characterization of Proanthocyanidin Distribution, Anthocyanin Extraction, and Chromatic Properties. *Journal of Agricultural and Food Chemistry*, *61*(26), 64466457. https://doi.org/10.1021/jf400733u

Ciani, M., & Ferraro, L. (1996). Enhanced Glycerol Content in Wines Made with Immobilized Candida stellata Cells. *Applied and Environmental Microbiology*, *62*(1), 128132. https://doi.org/10.1128/aem.62.1.128-132.1996

Costa, E., Cosme, F., Jordão, A. M., & Mendes-Faia, A. (2014). Anthocyanin profile and antioxidant activity from 24 grape varieties cultivated in two Portuguese wine regions. *OENO One*, *48*(1), 51. https://doi.org/10.20870/oeno-one.2014.48.1.1661

Downey, M. O., Harvey, J. S., & Robinson, S. P. (2003). Analysis of tannins in seeds and skins of Shiraz grapes throughout berry development. *Australian Journal of Grape and Wine Research*, *9*(1), 1527. https://doi.org/10.1111/j.1755-0238.2003.tb00228.x

Fanzone, M., Sari, S. E., Mestre, M. V., Catania, A. A., Catelén, M. J., Jofré, V. P., González-Miret, M. L., Combina, M., Vazquez, F., & Maturano, Y. P. (2020). Combination of pre-fermentative and fermentative strategies to produce Malbec wines of lower alcohol and pH, with high chemical and sensory quality. *OENO One*, *54*(4). https://doi.org/10.20870/oeno-one.2020.54.4.4018

Fulcrand, H., Atanasova, V., Salas, E., & Cheynier, V. (2004). The Fate of Anthocyanins in Wine : Are There Determining Factors ? *ACS Symposium Series*, 6888. https://doi.org/10.1021/bk-2004-0886.ch006

Fulcrand, H., Dueñas, M., Salas, E., & Cheynier, V. (2006). Phenolic Reactions during Winemaking and Aging. *American Journal of Enology and Viticulture*, *57*(3), 289297. https://doi.org/10.5344/ajev.2006.57.3.289

Gardner, J.M., Walker, M.E., Boss, P.K., & Jiranek, V. (2020). *The Effect of Grape Juice Dilution on Oenological Fermentation*. bioRxiv. https://doi.org/10.1101/2020.07.29.226142

Gil, M., Estevez, S., Kontoudakis, N., Fort, F., Canals, J.M., & Zamora, F. (2013) *Influence of partial dealcoholization by reverse osmosis on red wine composition and sensory characteristics*. European Food Research and Technology. 237:481-488. https://doi.org/10.1007/s00217-013-2018-6

Glories, Y. (1984). La couleur des vins rouges. 1^{e} Partie. Les équilibres des anthocyanes et des tanins. *Journal International des Sciences de la Vigne et du Vin, 18*, 253-271. https://doi.org/10.20870/oeno-one.1984.18.4.1744

Harbertson, J.F., Kennedy, J.A., & Adams, D.O. (2002). Tannin in skins and seeds of Cabernet-Sauvignon, Syrah, and Pinot noir berries during ripening. *American Journal of Enology and Viticulture. 53*:54-59. https://doi.org/10.5344/ajev.2002.53.1.54

Ho, P., Da Silva, M., & Hogg, T. A. (2001). Changes in colour and phenolic composition during the early stages of maturation of port in wood, stainless steel and glass. *Journal of the Science of Food and Agriculture, 81*(13), 1269-1280. https://doi.org/10.1002/jsfa.938

IPCC (2019). International Panel on Climate Change. *Climate Change 2019: Impacts, Adaptation, and Vulnerability*. http://ipcc-wg2.gov/AR5/report/final-drafts

Jones, G. V., White, M. A., Cooper, O. R., & Storchmann, K. (2005). Climate Change and Global Wine Quality. *Climatic Change*, *73*(3), 319–343. https://doi.org/10.1007/s10584-005-4704-2

Jordão, A.M., Ricardo-da-Silva, J.M., & Laureano, O. (1998). Evolution of anthocyanins during grape maturation of two varieties (*Vitis vinifera* L.), Castelão Francês and Touriga Francesa. *Vitis 37*, 93-94.

Kennedy, J. A., Matthews, M. A., & Waterhouse, A. L. (2002). Effect of maturity and vine water status on grape skin and wine flavonoids. *American Journal of Enology and Viticulture, 53*(4), 268-274. https://doi.org/10.5344/ajev.2002.53.4.268

Kontoudakis, N., Esteruelas, M., Fort, F., Canals, J., & Zamora, F. (2011). Use of unripe grapes harvested during cluster thinning as a method for reducing alcohol content and pH of wine. *Australian Journal of Grape and Wine Research*, *17*(2), 230–238. https://doi.org/10.1111/j.1755-0238.2011.00142.x

Lorenz, D., Eichhorn, K., Bleiholder, H., Klose, R., Meier, U., & Weber, E. (1995). Growth Stages of the Grapevine : Phenological growth stages of the grapevine (*Vitis vinifera* L. ssp. vinifera)—Codes and descriptions according to the extended BBCH scale. *Australian Journal of Grape and Wine Research*, *1*(2), 100103. https://doi.org/10.1111/j.1755-0238.1995.tb00085.x

Marques, J., Reguinga, R., Laureano, O., & Ricardo-da-Silva, J.M. (2005) Changes in grape seed, skins and pulp condensed tannins during berry ripening: effect of fruit pruning. *Ciência e técnica vitivinícola. 20*, 35-52.

Martínez-Moreno, A., Sanz, F., Yeves, A., Gil-Muñoz, R., Martínez, V., Intrigliolo, D., & Buesa, I. (2019). Forcing bud growth by double-pruning as a technique to improve grape composition of Vitis vinifera L. cv. Tempranillo in a semi-arid Mediterranean climate. *Scientia Horticulturae*, *256*, 108614. https://doi.org/10.1016/j.scienta.2019.108614

Martínez-Moreno, A., Pérez-Álvarez, E. P., Intrigliolo, D. S., Mirás-Avalos, J. M., López-Urrea, R., Gil-Muñoz, R., Lizama, V., García-Esparza, M. J., Álvarez, M. I., & Buesa, I. (2022). Effects of deficit irrigation with saline water on yield and grape composition of *Vitis vinifera* L. cv. Monastrell. *Irrigation Science*. https://doi.org/10.1007/s00271-022-00795-x

Merrell, C. P., Larsen, R. C., & Harbertson, J. F. (2017). Effects of Berry Maturity and Wine Alcohol on Phenolic Content during Winemaking and Aging. *American Journal of Enology and Viticulture*, *69*(1), 111. https://doi.org/10.5344/ajev.2017.17035

Mirás-Avalos, J. M., & Intrigliolo, D. S. (2017). Grape Composition under Abiotic Constrains : Water Stress and Salinity. *Frontiers in Plant Science*, *8*. https://doi.org/10.3389/fpls.2017.00851

Ortega-Regules, A., Romero-Cascales, I., Ros-García, J., López-Roca, J., & Gómez-Plaza, E. (2006). A first approach towards the relationship between grape skin cell-wall composition and anthocyanin extractability. *Analytica Chimica Acta*, *563*(1–2), 26–32. https://doi.org/10.1016/j.aca.2005.12.024

Ortega-Regules, A., Romero-Cascales, I., Ros García, J. M., Bautista-Ortín, A. B., López-Roca, J. M., Fernández-Fernández, J. I., & Gómez-Plaza, E. (2008). Anthocyanins and tannins in four grape varieties (*Vitis vinifera* L.). Evolution of their content and extractability. *OENO One*, *42*(3), 147. https://doi.org/10.20870/oeno-one.2008.42.3.818

del Rio, J. L. P., & Kennedy, J. A. (2006). Development of Proanthocyanidins in *Vitis vinifera* L. cv. Pinot noir Grapes and Extraction into Wine. *American Journal of Enology and Viticulture*, *57*(2), 125132. https://doi.org/10.5344/ajev.2006.57.2.125

Piccardo, D., Favre, G., Pascual, O., Canals, J. M., Zamora, F., & González-Neves, G. (2019). Influence of the use of unripe grapes to reduce ethanol content and pH on the color, polyphenol and polysaccharide composition of conventional and hot macerated Pinot Noir and Tannat wines. *European Food Research and Technology*, *245*(6), 13211335. https://doi.org/10.1007/s00217-019-03258-4

Pickering, G., Heatherbell, D., & Barnes, M. (1998). Optimising glucose conversion in the production of reduced alcohol wine using glucose oxidase. *Food Research International*, *31*(10), 685692. https://doi.org/10.1016/s0963-9969(99)00046-0

Pickering, G. J. (2000). Low- and Reduced-alcohol Wine : A Review. *Journal of Wine Research*, *11*(2), 129144. https://doi.org/10.1080/09571260020001575

Ribéreau-Gayon, P., Dubourdieu, D., Donèche, B., & Lonvaud, A. (2006). *Handbook of Enology: The Microbiology of Wine and Vinifications, Volume 1, 2nd Edition*. John Wiley & Sons, West Sussex, UK, 44. https://doi.org/10.1002/0470010363

Romero-Cascales, I., Ortega-Regules, A., López-Roca, J. M., Fernández-Fernández, J. I., & Gómez-Plaza, E. (2005). Differences in Anthocyanin Extractability from Grapes to Wines According to Variety. *American Journal of Enology and Viticulture*, *56*(3), 212–219. https://doi.org/10.5344/ajev.2005.56.3.212

Romeyer, F. M., Macheix, J. J., & Sapis, J. C. (1985). Changes and importance of oligomeric procyanidins during maturation of grape seeds. *Phytochemistry*, *25*(1), 219221. https://doi.org/10.1016/s0031-9422(00)94532-1

Rousserie, P., Lacampagne, S., Vanbrabant, S., Rabot, A., & Geny-Denis, L. (2020). Influence of berry ripeness on seed tannins extraction in wine. *Food Chemistry*, *315*, 126307. https://doi.org/10.1016/j.foodchem.2020.126307

Sacchi, K. L., Bisson, L. F., & Adams, D. O. (2005). A Review of the Effect of Winemaking Techniques on Phenolic Extraction in Red Wines. *American Journal of Enology and Viticulture*, *56*(3), 197206. https://doi.org/10.5344/ajev.2005.56.3.197

Saint-Cricq de Gaulejac, N., Vivas, N., & Glories, Y. (1998). Maturité phénolique: définition et contrôle. *Revue française d'oenologie*, (173), 22-25.

Salazar Parra, C., Aguirreolea, J., Sánchez-Díaz, M., Irigoyen, J. J., & Morales, F. (2010). Effects of climate change scenarios on Tempranillo grapevine (Vitis vinifera L.) ripening : response to a combination of elevated CO2 and temperature, and moderate drought. *Plant and Soil*, *337*(12), 179191. https://doi.org/10.1007/s11104-010-0514-z

Sam, F. E., Ma, T. Z., Salifu, R., Wang, J., Jiang, Y. M., Zhang, B., & Han, S. Y. (2021). Techniques for Dealcoholization of Wines : Their Impact on Wine Phenolic Composition, Volatile Composition, and Sensory Characteristics. *Foods*, *10*(10), 2498. https://doi.org/10.3390/foods10102498

Schelezki, O. J., Smith, P. A., Hranilovic, A., Bindon, K. A., & Jeffery, D. W. (2018). Comparison of consecutive harvests versus blending treatments to produce lower alcohol wines from Cabernet Sauvignon grapes : Impact on polysaccharide and tannin content and composition. *Food Chemistry*, *244*, 50–59. https://doi.org/10.1016/j.foodchem.2017.10.024

Schelezki, O. J., Antalick, G., Šuklje, K., & Jeffery, D. W. (2020). Pre-fermentation approaches to producing lower alcohol wines from Cabernet Sauvignon and Shiraz : Implications for wine quality based on chemical and sensory analysis. *Food Chemistry*, *309*, 125698. https://doi.org/10.1016/j.foodchem.2019.125698

Schultz, H. (2000). Climate change and viticulture : A European perspective on climatology, carbon dioxide and UV-B effects. *Australian Journal of Grape and Wine Research*, *6*(1), 212. https://doi.org/10.1111/j.1755-0238.2000.tb00156.x

Smith, P.A. (2005). Precipitation of tannin with methyl cellulose allows tannin quantification in grape and wine samples. *Technical Review. AWRI. 158*, 3-7.

Stillman, J. A. (1993). Color Influences Flavor Identification in Fruit-flavored Beverages. *Journal of Food Science*, *58*(4), 810812. https://doi.org/10.1111/j.1365-2621.1993.tb09364.x

Varela, C., Dry, P., Kutyna, D., Francis, I., Henschke, P., Curtin, C., & Chambers, P. (2015). Strategies for reducing alcohol concentration in wine. *Australian Journal of Grape and Wine Research*, *21*, 670679. https://doi.org/10.1111/ajgw.12187