



Controlled water deficit modifies the phenolic composition and sensory properties in Cabernet Sauvignon wines

Alejandro Cáceres-Mella^{a,*}, Camila Ribalta-Pizarro^b, Luis Villalobos-González^c, Italo F. Cuneo^a, Claudio Pastenes^c

^a Escuela de Agronomía, Pontificia Universidad Católica de Valparaíso, Av. San Francisco s/n, La Palma, Quillota, Chile

^b Centro Regional de Innovación Hortofrutícola de Valparaíso (Ceres), Pontificia Universidad Católica de Valparaíso, Av. San Francisco s/n, La Palma, Quillota, Chile

^c Universidad de Chile, Facultad de Ciencias Agronómicas, Santa Rosa 11315, La Pintana, Casilla 1004, Santiago, Chile



ARTICLE INFO

Keywords:

Controlled water deficit
Cabernet Sauvignon
Phenolic compounds
Anthocyanins
Sensory evaluation
Wine

ABSTRACT

This study investigates the chemical composition and sensory properties of wines from Cabernet Sauvignon vines grown under controlled water deficit in two consecutive seasons. The wines were made from fruit of grapevines that were maintained under three water status levels (i.e. T1, $\Psi_{\text{stem}} = -0.8$ MPa; T2, $\Psi_{\text{stem}} = -0.9$ MPa; T3, $\Psi_{\text{stem}} = -1.0$ MPa) from *veraison* until harvest. Our results suggest that wine phenolic composition was affected by controlled water deficit, where T3 wines exhibited a higher concentration of total phenols, total anthocyanins and chroma (C*) in both seasons. These results coincide with the principal component analysis that indicated a substantial separation between years and deficit irrigation. We found that irrigation treatments only produce differences in concentration, but not in anthocyanin composition in both years. Separation of proanthocyanidins fractions by solid phase extraction using Sep-Pak Plus tC_{18} cartridges showed only a change in the concentration of the monomeric fraction in 2014 season, but not in the proportion of the different proanthocyanidins fractions in both seasons. Finally, the sensory composition of wines showed differences that depend on the season and resulted in more red fruits, more fullness perception in mouth and more color intensity in wines from less irrigated treatments.

1. Introduction

Regulated-deficit irrigation is a common practice in many viticultural regions and in warmer geographical areas is essential during grape ripening (Santos et al., 2005; Chaves et al., 2010). This practice has a profound effect on grape berries, such as reductions in the size of berries, increasing the skin to pulp ratio, the improvement of the microclimate of the fruiting zone favoring the grape berry concentration, and directly affecting the secondary metabolism, at least in part as a consequence of the transient fastening of the grape berry ripening involving abscisic acid and sugar content at the time of *veraison*. These, in turn, are the necessary signals for gene expression and protein synthesis involved in the phenylpropanoid pathway in grapes, leading to the accumulation of flavanols, flavonols and anthocyanins (Kennedy et al., 2002; Castellarin et al., 2007; Deluc et al., 2009; Lacampagne et al., 2010; Villalobos-González et al., 2016). As part of the phenolic compounds, proanthocyanidins and anthocyanins are the most important qualitative factors in red wines due to their role in mouthfeel, astringency, bitterness and color (Gawel et al., 2001; Brossaud et al.,

2001; Boss et al., 1996). Although is recognized the relationship between the chemical composition of grapes and wines, the wine making process involve a complex series of activities that might affect that relationship. Big changes in the grape berry chemical compounds are known to occur during this process, making difficult the extrapolation from grape berry to wine chemical composition, not to mention the sensory attributes of the later. Since most studies of water stress have been performed on grape berries only, the knowledge of the impact of deficit irrigation on wines is, in many cases, less clear. Moreover, wine quality depends on several factors, such as concentration of phenolic compounds, aromatic composition, alcohol strength, acidity and consumer preferences. Regarding the chemical composition of the berries, the chemical properties of the different compounds, and not just their concentration, are important in the chemical and sensory attributes of wines. Proanthocyanidins for instance, depending on their composition and degree of polymerization, will result in variations in the body mouth feeling, bitterness and astringency (Vidal et al., 2004; Payne et al., 2009; Villamor et al., 2009). Therefore, even though the impact of regulated-deficit irrigation on grape berry composition may shed

* Corresponding author.

E-mail address: alejandro.caceres@pucv.cl (A. Cáceres-Mella).

light on the potential quality of the final wine, a precise prediction might be rather adventurous. For this reason, the aim of this study was to examine the phenolic composition and sensory attributes of Cabernet Sauvignon wines produced from vines subjected to different irrigation regimes during two consecutive seasons.

2. Materials and methods

2.1. Vineyard site and irrigation treatment

The experiment was conducted in two consecutive seasons (i.e. 2014 and 2015) in a commercial vineyard (Haras de Pirque Winery) located in the Maipo Valley in central Chile. Grapevines of 12-years-old own rooted *Vitis vinifera* plants cv. Cabernet Sauvignon were trained to a vertical trellising system (Guyot double pruning method) with vines planted 1.5 m by 2.5 m in N–S oriented rows and irrigated by conventional drip irrigation. The historical average yield of the vineyard was approximately 8 ton/ha. The site has deep colluvial soil with clay loam texture. The canopy management of the vineyard was the standard for the vineyards located in this geographical area. The climatic parameters during growing season (January to April) for the geographical region where the vineyard is located are presented in Table 1.

The experimental design consisted of a completely randomized blocks with five replicates. Replicates corresponded to seven consecutive plants. Three irrigation treatments were applied weekly during each season and were established by a combination of drip emitters of different flow rates, as follows; T1: 12 L/h; T2: 6 L/h and T3: 2 L/h. In both seasons, the irrigation in the field was suspended approximately 25 days before veraison until 10 days before veraison, to reduce the xylem water potential in grapevines. Thus, irrigation treatments started approximately 10 days before veraison resulting in the following average values of midday stem water potential: T1, $\Psi_{\text{stem}} = -0.8$ MPa; T2, $\Psi_{\text{stem}} = -0.9$ MPa and T3, $\Psi_{\text{stem}} = -1.0$ MPa. Irrigation treatments was applied during the same amount of time for each of the flow rates mentioned above. To check the correct differentiation of treatments in field, plant water status was monitored weekly by measuring midday stem water potential determined by means of a pressure chamber. For this, leaves were enclosed in aluminum plastic bags for 90 min at midday. Further details about midday stem water potential throughout ripening in both seasons are available in Talaverano et al. (2018). The commercial harvest date in each season was determined based on grape chemical analysis and degustation of the whole berries. The grapes from all the treatments were harvested the same day in each season (i.e. 2014, April, 4th; 2015, March 31th).

2.2. Winemaking procedure

For winemaking and due to grape volume limitation, adjacent vineyard replicates were combined so that triplicate wines ($n = 3$) were produced from each irrigation treatment. In brief, the grapes were handpicked and delivered immediately to the laboratory for winemaking. The vinification process started with the destemming and crushing of the grapes using a semiautomatic crusher machine (Eno 3, Enoitalia, Italy) and disposing the resulting crushed grapes in 25 L plastic alimentary vats (Plásticos Haddad S.A, Chile). Parameters such

Table 1
Climate data (January to April) for the period of grape ripening.^a

	Mean daily	Mean daily	Day degrees,	Rainfall	Etp
Vintage	max temp (°C)	min temp (°C)	base 10 °C	(mm)	(mm/day)
2014	28.9	7.1	1356.1	0	3.7
2015	29.7	8.4	1332.9	0.2	3.6

^a Data are from the Agroclimatic System FDF-INIA-DMC in Maipo Valley, Chile.

as pH, titratable acidity and yeast-assimilable nitrogen (YAN) were checked and adjusted in the juice as necessary before the fermentation process. LA Bayanus (Lamothe-Abiet, France) in a dose of 20 g/hL was used as yeast inoculum. The fermentation process was maintained at a temperature of 23–24 °C with punch down twice per day. After the fermentation process was completed, replicates were pressed using a single basket press (Model 50, Enoitalia, Italy). Free-run fractions of each deposit were racked, cold stabilized and the SO₂ free levels were adjusted to 30 mg/L and immediately bottled in dark green 750 mL glass bottle (Cristalchile, Santiago, Chile) and stored at 15 °C for further analysis. In this study, we employed the same winemaking process for all treatments in both vintages.

2.3. Spectrophotometric characterization

For chemical characterization, the methods recommended by OIV (2012) were used to determine the pH, sugar content (g glucose/L), titratable acidity (g tartaric acid/L) and ethanol content (% v/v). The total phenol content was determined by UV absorptiometry at 280 nm using gallic acid as a standard (Glories, 1984). The total tannin content was determined using methylcellulose (1500 cP, viscosity at 20 g/L, Sigma Chemical Co., St. Louis, MO, USA) as precipitant agent (Sarneckis et al., 2006). The total anthocyanins content was measured using the method described by Ribéreau-Gayon and Stonestreet (1965). The color intensity (CI) was estimated using the method described by Glories (1984). The color coordinated lightness (L*), chroma (C*) and hue (h*) were determined according to Pérez-Magariño et al. (2007). All reagents were of analytical grade or higher. Absorbance values were measured using a UV-1601 UV–vis spectrophotometer (Shimadzu, Kyoto, Japan).

2.4. Fractionation of proanthocyanidins using Sep-Pak Plus tC₁₈ cartridges

Wine samples (7 mL) were subjected to a solid phase extraction using two neutral Sep-Pak Plus tC₁₈ cartridges connected in series (top, Sep-Pak Plus Environmental tC₁₈ cartridge (900 mg); bottom, Sep-Pak Plus Short tC₁₈ cartridge (400 mg)) (Waters, MA, USA), according to the method described by Sun et al. (1998a). All the steps were as in previous work (Cáceres et al., 2012). For each fraction obtained previously, flavanols were quantified using the modified vanillin assay described by Sun et al. (1998b).

2.5. HPLC-DAD analysis of anthocyanin compounds

Anthocyanin composition in wine samples were performed using a 1200 Series HPLC system (Agilent Technologies, Santa Clara, CA, USA) consisting of a G1315B photodiode array detector (DAD), a G1311A quaternary pump, a G1313A autosampler, a G1322A degasser and a G1316A thermostatted column compartment. A 2-mL sample of wines were filtered through a 0.22- μ m pore size membrane (Millipore, Billerica, MA, USA), and an aliquot of 100- μ L of each sample were subjected to reverse-phase chromatographic separation at 20 °C using a LiChro Cart 100 RP-18 column (5 μ m, 4.0 mm i.d. x 250 mm; Agilent Technologies). The DAD detector was set from 210 to 600 nm. Two mobile phases were used: A, water/formic acid (90:10 v/v), and B, acetonitrile. Two gradients were used, first at a flow rate of 1.1 mL/min from 0 to 22 min and the second of 1.5 mL/min from 22 to 35 min. The mobile phases were applied as follows: 96–85% A from 0 to 22 min, 85–15% A from 12 to 22 min and 85–70% A from 22 to 35 min. The quantification was performed by peak area measurements at 520 nm. The calibration curves at 520 nm were obtained by injecting different volumes of standard solutions under the same conditions used for the samples (Fanzone et al., 2012).

2.6. Sensory evaluation of wines

The descriptive analysis was conducted on the wines one month after bottling in the Laboratory of Sensory Analysis from the Department of Agro-industry and Oenology of the University of Chile by means of a sensory panel consisting of 12 people (21–40 years old) with previous training and experience in descriptive methodology. Six attributes were evaluated, colour intensity, red fruits, bitterness, fullness, astringency and persistence on a 15-cm unstructured scale anchored from 0 (absence of sensation) to 15 (extremely high sensation). 20 mL of wine was served in technical glasses (Viticole, Arcoroc, France) at 18–19 °C using a completely randomised order. Between each sample there was 60-s break to decrease fatigue and the judges rinsed the mouth with water and unsalted crackers. Descriptive analysis took place in two sessions in order to obtain two replicates. The collection and interpretation of data was performed with FIZZ software version 2.47B (Biosystemes, Counternon, France).

2.7. Statistical analysis

For the chemical parameters, analysis of variance (ANOVA) and Tukey's honestly significant difference (HSD) test were used for mean separation with a significance level of 95% ($p < 0.05$). For descriptive analysis, ANOVA and LSD test were used with a significance level of 90% ($p < 0.1$). The descriptive and chemical results were analyzed by principal component analysis (PCA). Statistical analysis was performed using R statistical software version 3.03 (R Foundation for Statistical Computing, Austria) and Statgraphics Centurion version 15.2 (Statpoint Technologies, USA)

3. Results and discussion

3.1. Wine chemical composition

Water scarcity is already affecting many of the most productive grape growing regions and it has been reported that water availability will become the limiting factor in wine production in the future (IPCC, 2007; Jones et al., 2005). In our study, the contrasting water regimes did not result in differences in the general chemical properties of wines, such as pH, titratable acidity, sugar content and ethanol content in both seasons (Table 2). On the contrary, in the overall phenolic composition, differences were seen in the concentration of total phenols and total anthocyanins. Total phenols ranged from 1452.0 to 1557.7 mg/L in 2014 and from 1439.7 to 1523.7 mg/L in 2015. Total anthocyanins varied from 466.0 to 533.7 mg/L in 2014 and from 620.3 to 696.4 mg/L in 2015. In general, we observed in both seasons, higher values on the concentration of total phenols, total anthocyanins and chroma (C^*) in the most restricted treatment (T3) (Table 2). Even though, the water status in grapevines could cause changes in fruit composition by producing a reduction in the size of the berries, resulting in a higher skin to pulp relationship, i.e. concentrating compounds (Kennedy et al., 2002), in our study the application of different water regimes did not produce differences in berry weight in none of the two season, and only slight differences in the skin/berry weight relationship only in the 2014 season (data not shown).

Previous studies showed that water stress induce changes in the expression level of certain genes and transcription factors involved in the phenylpropanoid pathway producing an increase in concentration of certain phenolic compounds, such as anthocyanins and proanthocyanidins in grapes (Castellarin et al., 2007; Deluc et al., 2009; Genebra et al., 2014; Cáceres-Mella et al., 2017). It is clear from our results that the eventual impact of the more severe water stress on the grape berry total phenols, total tannins and total anthocyanins (Cáceres-Mella et al., 2017), likely achieved by an increased metabolic synthesis, was translated to the final wines. These findings also confirm previous results, demonstrating that water deficit could modify the chemical

composition of wines (Acevedo-Opazo et al., 2010; Casassa et al., 2015; Zarrouk et al., 2012; Bonada et al., 2015).

The anthocyanins composition in wines from both seasons indicates that irrigation treatments induce increases in concentration of glucosylated anthocyanins, such as delphinidin-3-glucoside, petunidin-3-glucoside, peonidin-3-glucoside and malvidin-3-glucoside, similar to the acetylated anthocyanins, but in a much lower concentration than the previous (Table 3). Regarding the coumaroylated anthocyanins, in 2014 only petunidin-3-*p*-coumaroylglucoside was increased by the more restrictive irrigation regime. In 2015, however, several differences between irrigation treatments were observed for delphinidin-3-*p*-coumaroylglucoside, cianidin-3-*p*-coumaroylglucoside, petunidin-3-*p*-coumaroylglucoside and malvidin-3-*p*-coumaroylglucoside. In general, the higher concentration of several anthocyanins were observed in T3 in both seasons, but with lower differences compared to T1 in 2014. These results clearly suggest that irrigation treatments only produce differences in anthocyanin concentration, but not in anthocyanin composition.

Even though wines elaborated by the industry use different wine-making techniques, which results in different extracting rates and consequent highly variable phenolic compound concentrations (Cáceres et al., 2012), in our study, the vinification process was the same for all vats. The fact that controlled deficit irrigation improves wine concentration is of importance not only for the wine quality but also for the implications in terms of water savings, especially in warm Mediterranean areas facing Climate change (Montes et al., 2012; Jones et al., 2005).

Even though many studies have shown an increase in phenolic compounds content and concentration in grape berries upon water stress (Roby et al., 2004; Casassa et al., 2015), few studies have explored the different proanthocyanidin fractions in wines and how water stress could modify these fractions. Fig. 1 displays the monomeric, oligomeric and polymeric flavan-3-ol proportions in the wine samples in both seasons. In the case of monomeric fraction, values ranged from 39.7 to 66.6 mg/L in 2014 and from 6.5 to 14.1 mg/L in 2015. Regarding oligomeric fraction, values varied in 2014 from 111.4 to 214.4 mg/L and from 23.41 to 94.3 mg/L in 2015. Finally, polymeric fraction showed values ranging from 689.2 to 1122.4 mg/L in 2014 season. In 2015 season, the values ranged from 429.4 to 835.1 mg/L. In 2014 season, only the monomeric fraction presented differences between treatments where T1 had the highest concentration of flavan-3-ol monomers, while in 2015 there were no differences in concentration between treatments for each fraction. In general, for each flavan-3-ol fraction in 2014 there was a higher concentration compared to 2015. Also, the water stress did not produce differences in flavan-3-ol proportion in any of the seasons. Recently, Cáceres-Mella et al. (2017) showed that water stress produced differences in terms of concentration between the different fractions of proanthocyanidins in grape skins, observing that the most restricted treatment (T3) had a higher polymerized fraction and a higher mean degree of polymerization (mDP) at harvest. The grapes of that trial were used to elaborate the wines in this assay revealing a discrepancy between the concentration of some fractions in grape skins and the final wine. The wine making process consist of a complex series of procedures affecting the chemical composition of the grape berries, making difficult the extrapolation from the grape berry to the wine chemical composition (Busse-Valverde et al., 2010). For instance, previous investigations demonstrated that several cell wall components limit the extraction of anthocyanins from grape skins, especially polysaccharides (Bindon et al., 2014; Bindon et al., 2016; Bautista-Ortín et al., 2016) and skin tannins from red grapes are more readily extracted during winemaking than seed tannins (Sacchi et al., 2005; Busse-Valverde et al., 2012). For this reason, the impact of deficit irrigation on proanthocyanidin fraction in wines is not easily extrapolated from the grape berry composition.

A Principal Component Analysis (PCA) was performed for providing a visual representation of the relationship between irrigation

Table 2
Wine analytical parameters from different irrigation treatments in cv. Cabernet Sauvignon.

	T1 ($\Psi_{\text{stem}} = -0.8$ Mpa)	T2 ($\Psi_{\text{stem}} = -0.9$ Mpa)	T3 ($\Psi_{\text{stem}} = -1.0$ Mpa)
2014			
pH	3.84 ± 0.1	3.86 ± 0.2	3.79 ± 0.1
Titrateable acidity (g tartaric acid/L)	3.2 ± 0.1	3.1 ± 0.2	3.1 ± 0.1
Sugar content (g glucose/L)	1.9 ± 0.1	2.2 ± 0.1	2.1 ± 0.2
Ethanol content (% v/v)	13.4 ± 0.2	13.5 ± 0.1	13.0 ± 0.3
Total phenols (mg GAE/L)	1452.0 ± 10.3 b	1505.2 ± 13.1 a	1557.7 ± 9.2 a
Total tannins (mg catechin/L)	829.5 ± 76.3 ab	781.2 ± 10.6 b	1037.2 ± 12.6 a
Total anthocyanins (mg malvidin/L)	466.0 ± 5.8 c	490.2 ± 7.2 b	533.7 ± 5.1 a
Color intensity (a.u)	6.0 ± 0.1 c	6.8 ± 0.1 b	7.3 ± 0.0 a
L*	70.7 ± 0.6 a	67.5 ± 0.3 b	65.6 ± 0.1 c
C*	32.1 ± 0.6 c	34.1 ± 0.2 b	36.1 ± 0.2 a
h*	21.8 ± 0.2	23.0 ± 0.5	23.4 ± 0.5
2015			
pH	3.79 ± 0.1	3.43 ± 0.2	3.75 ± 0.1
Titrateable acidity (g tartaric acid/L)	3.8 ± 0.1	3.6 ± 0.2	3.7 ± 0.1
Sugar content (g glucose/L)	1.4 ± 0.1	1.3 ± 0.1	1.6 ± 0.2
Ethanol content (% v/v)	13.5 ± 0.2	13.6 ± 0.1	14.5 ± 0.3
Total phenols (mg GAE/L)	1509.6 ± 10.3 ab	1439.7 ± 30.4 b	1523.7 ± 14.0 a
Total tannins (mg catechin/L)	1057.0 ± 53.0	1150.3 ± 53.2	1149.3 ± 24.7
Total anthocyanins (mg malvidin/L)	645.4 ± 6.6 b	620.3 ± 8.8 b	696.4 ± 8.8 a
Color intensity (a.u)	14.9 ± 0.3	13.6 ± 0.4	15.6 ± 0.4
L*	42.0 ± 0.6	44.4 ± 1.0	40.8 ± 0.6
C*	61.8 ± 0.1 a	59.9 ± 0.1 b	61.9 ± 0.2 a
h*	4.8 ± 0.6	3.8 ± 0.4	5.9 ± 0.9

All data are expressed as the mean ± standard error. Different letters indicate significant differences ($p < 0.05$) between treatments according Tukey's HSD test. GAE: gallic acid equivalent. a.u: absorbance unit.

Table 3
Wine anthocyanin concentrations from different irrigation treatments in cv. Cabernet Sauvignon.

Compound	T1 ($\Psi_{\text{stem}} = -0.8$ Mpa)	T2 ($\Psi_{\text{stem}} = -0.9$ Mpa)	T3 ($\Psi_{\text{stem}} = -1.0$ Mpa)
2014			
Delphinidin-3-glucoside	7.68 ± 0.08 b	3.60 ± 0.03 c	8.98 ± 0.09 a
Cianidin-3-glucoside	13.88 ± 2.48	14.45 ± 2.15	18.53 ± 0.05
Petunidin-3-glucoside	19.67 ± 0.21 c	20.83 ± 0.45 b	22.96 ± 0.08 a
Peonidin-3-glucoside	9.26 ± 0.03 b	9.45 ± 0.25 ab	10.18 ± 0.09 a
Malvidin-3-glucoside	153.16 ± 1.15 b	169.08 ± 6.41 ab	175.91 ± 0.45 a
Glucosylated anthocyanins	203.65 ± 3.95	217.41 ± 9.29	236.56 ± 0.76
Delphinidin-3-acetylglucoside	3.37 ± 0.09 b	3.52 ± 0.03 ab	3.83 ± 0.02 a
Cianidin-3-acetylglucoside	6.45 ± 0.10 b	6.78 ± 0.07 ab	7.48 ± 0.31 a
Petunidin-3-acetylglucoside	4.29 ± 0.05 b	4.49 ± 0.12 ab	4.82 ± 0.03 a
Peonidin-3-acetylglucoside	3.76 ± 0.04 b	4.11 ± 0.08 a	4.17 ± 0.04 a
Malvidin-3-acetylglucoside	35.54 ± 0.89	37.40 ± 1.47	38.60 ± 0.09
Acetylated anthocyanins	53.41 ± 1.17	56.3 ± 1.77	58.9 ± 0.49
Delphinidin-3-p-coumaroylglucoside	1.26 ± 0.02	1.86 ± 0.14	1.83 ± 0.24
Cianidin-3-p-coumaroylglucoside	1.44 ± 0.05	1.37 ± 0.02	1.45 ± 0.05
Petunidin-3-p-coumaroylglucoside	1.16 ± 0.01 b	1.19 ± 0.02 b	1.29 ± 0.02 a
Peonidin-3-p-coumaroylglucoside	1.17 ± 0.01	1.10 ± 0.04	1.20 ± 0.01
Malvidin-3-p-coumaroylglucoside	6.15 ± 0.11	6.28 ± 0.41	6.75 ± 0.06
Coumaroylated anthocyanins	11.18 ± 0.20	11.8 ± 0.63	12.52 ± 0.38
2015			
Delphinidin-3-glucoside	15.76 ± 1.22 ab	11.63 ± 0.69 b	19.63 ± 1.35 a
Cianidin-3-glucoside	5.12 ± 0.23	4.58 ± 0.18	5.60 ± 0.24
Petunidin-3-glucoside	20.82 ± 0.60 ab	16.50 ± 0.13 b	25.15 ± 2.26 a
Peonidin-3-glucoside	11.57 ± 0.58 a	9.29 ± 0.07 b	12.38 ± 0.74 a
Malvidin-3-glucoside	261.10 ± 5.69 ab	234.79 ± 4.41 b	269.28 ± 3.60 a
Glucosylated anthocyanins	314.37 ± 8.32	276.79 ± 5.48	332.04 ± 8.19
Delphinidin-3-acetylglucoside	11.03 ± 0.63 ab	9.60 ± 0.40 b	14.08 ± 0.48 a
Cianidin-3-acetylglucoside	5.91 ± 0.47	5.65 ± 0.30	6.78 ± 0.36
Petunidin-3-acetylglucoside	4.23 ± 0.06 ab	3.46 ± 0.03 b	4.84 ± 0.28 a
Peonidin-3-acetylglucoside	3.98 ± 0.10 ab	3.22 ± 0.03 b	4.42 ± 0.29 a
Malvidin-3-acetylglucoside	48.14 ± 1.52 b	44.18 ± 0.21 b	53.23 ± 0.28 a
Acetylated anthocyanins	73.29 ± 2.78	66.11 ± 0.97	83.35 ± 1.69
Delphinidin-3-p-coumaroylglucoside	1.16 ± 0.02 a	0.96 ± 0.01 b	1.10 ± 0.04 a
Cianidin-3-p-coumaroylglucoside	1.30 ± 0.05 ab	1.07 ± 0.03 b	1.34 ± 0.06 a
Petunidin-3-p-coumaroylglucoside	1.63 ± 0.04 a	1.43 ± 0.01 b	1.54 ± 0.01 ab
Peonidin-3-p-coumaroylglucoside	1.85 ± 0.13	1.43 ± 0.05	1.91 ± 0.16
Malvidin-3-p-coumaroylglucoside	13.07 ± 0.23 a	11.13 ± 0.23 b	13.60 ± 0.06 a
Coumaroylated anthocyanins	19.01 ± 0.47	16.02 ± 0.33	19.49 ± 0.33

All data are expressed as the mean ± standard error (n = 3). Different letters indicate significant differences ($p < 0.05$) between treatments according to Tukey's HSD test.

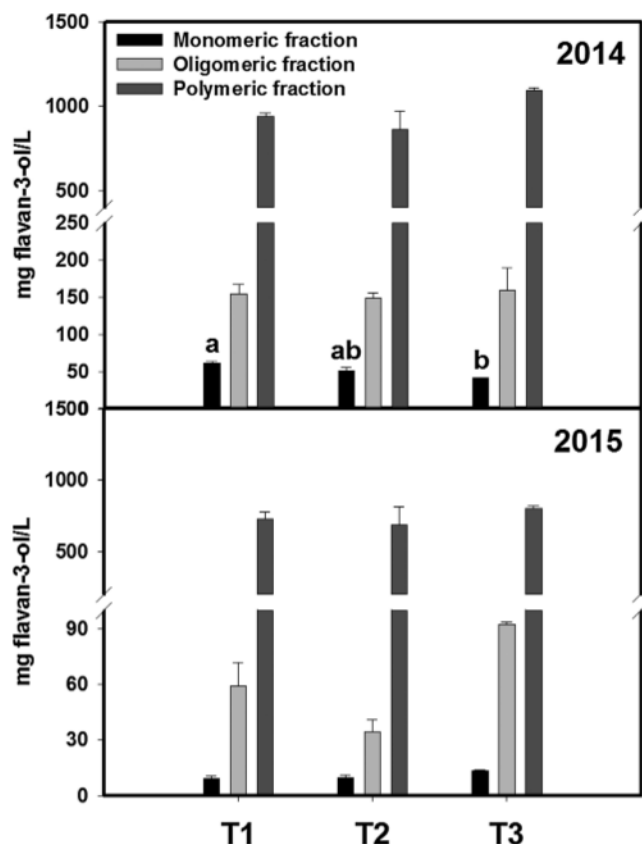


Fig. 1. Wine proanthocyanidin fractions from different irrigation treatments in cv. Cabernet Sauvignon. Different letters denote significant differences among treatments ($p < 0.05$, Tukey's HSD test). T1, $\Psi_{stem} = -0.8$ MPa; T2, $\Psi_{stem} = -0.9$ MPa; T3, $\Psi_{stem} = -1.0$ MPa.

Table 4

Wine descriptive analysis from different irrigation treatments in cv. Cabernet Sauvignon.

	T1 ($\Psi_{stem} = -0.8$ Mpa)	T2 ($\Psi_{stem} = -0.9$ Mpa)	T3 ($\Psi_{stem} = -1.0$ Mpa)
2014			
Color intensity	7.8 ± 0.7	7.9 ± 0.8	8.7 ± 0.8
Red fruits	6.9 ± 0.8 b	8.4 ± 0.6 a	8.6 ± 0.7 a
Bitterness	8.7 ± 0.7	8.6 ± 0.8	9.7 ± 0.9
Fullness	5.9 ± 0.4 c	8.3 ± 0.5 a	7.1 ± 0.5 b
Astringency	8.5 ± 0.9	7.8 ± 0.5	8.9 ± 0.6
Persistence	7.8 ± 0.3	8.4 ± 0.5	8.4 ± 0.8
2015			
Color intensity	7.8 ± 0.6 b	10.8 ± 0.6 a	11.0 ± 0.6 a
Red fruits	8.4 ± 0.6	7.2 ± 0.6	8.4 ± 0.8
Bitterness	5.9 ± 0.9	5.8 ± 0.9	6.2 ± 1.0
Fullness	8.8 ± 0.6	7.2 ± 0.5	8.0 ± 0.5
Astringency	8.6 ± 0.7	7.7 ± 0.8	8.4 ± 0.7
Persistence	9.5 ± 0.9	8.0 ± 0.6	9.5 ± 0.7

All data are expressed as the mean of 12 judges ± standard error. Different letters indicate significant differences ($p < 0.1$) between treatments according to LSD test.

treatments in both years and the global chemical analysis (Fig. 2). PC1 and PC2 accounted for 91.2% of the total variation (78.4% and 12.8% respectively). PC1 was mainly linked to total tannins, total anthocyanins, flavan-3-ol monomers, glucosylated anthocyanins, acetylated anthocyanins and coumaroylated anthocyanins and color variables such as color intensity (CI), L^* , C^* and h^* , whereas PC2 was related to total phenols, tannins, flavan-3-ol oligomers and flavan-3-ol polymers. A contrasting wine score was observed based on the year, for any given irrigation regime. The 2014 wines were characterized mainly by total phenols, L^* , h^* and flavanol fractions (monomers, oligomers and polymers), whereas the 2015 wines were described mainly by total tannins, total anthocyanins, color intensity, C^* , glucosylated

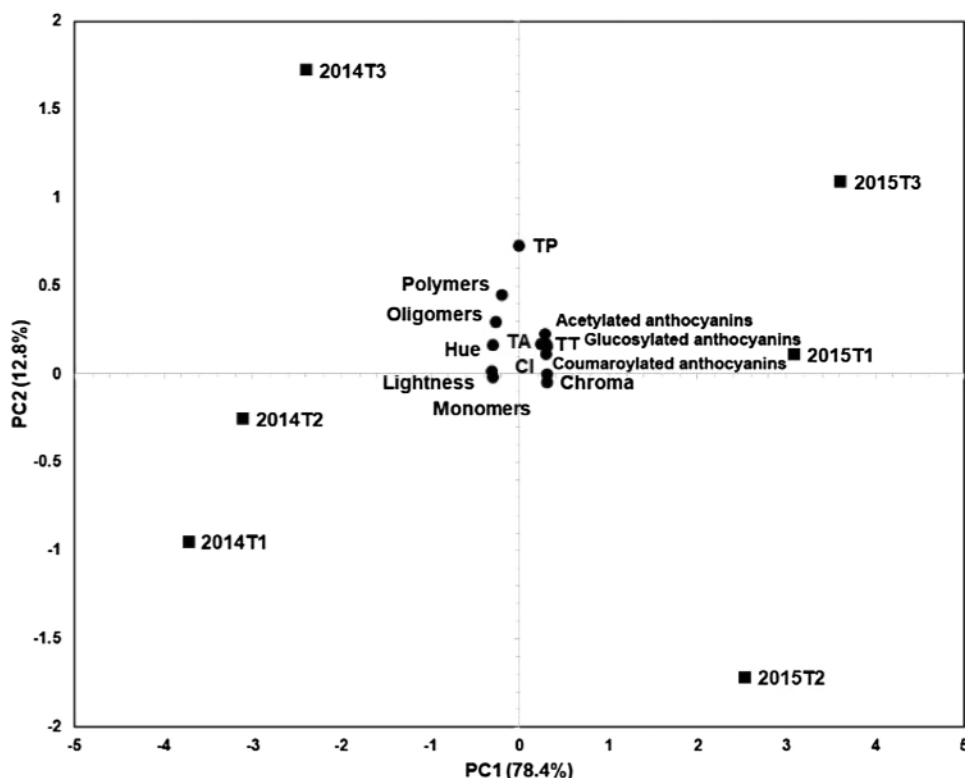


Fig. 2. Principal component analysis (PCA) comparing chemical parameters of Cabernet Sauvignon wines. TP, total phenols; TT, total tannins; TA, total anthocyanins; CI, color intensity.

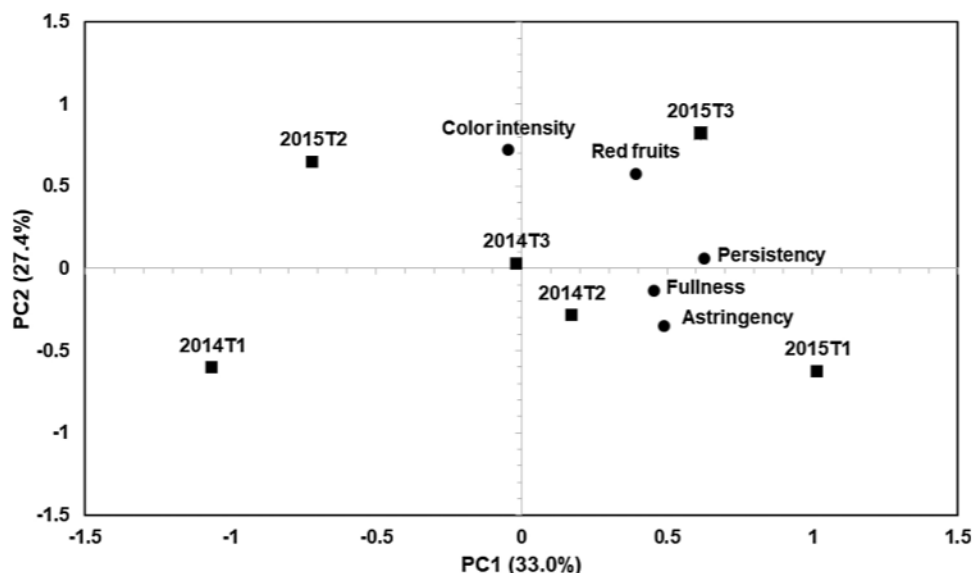


Fig. 3. Principal component analysis (PCA) showing descriptive analysis scores for Cabernet Sauvignon wine samples.

anthocyanins, acetylated anthocyanins and coumaroylated anthocyanins. In general, most variables were related to T3, rather than T1 and T2 in both years, supporting the importance of controlled deficit irrigation on the chemical composition of wines.

3.2. Wine descriptive analysis

In 2014, the wines showed significant differences in red fruits and fullness, with higher scores for T2 and T3. Instead, in 2015 only differences in color intensity were evident, with higher scores for T2 and T3 (Table 4). The differences in fullness especially in T2 and T3 may be related to their higher tannin concentration (Table 2). The higher color intensity scored by the sensory panel, on the other hand, matches the higher anthocyanins concentration in T3 (Table 2). In general, the wines from the less irrigated treatments, such as T2 and T3, were associated with positive sensory properties, including more pronounced red fruits aromas, greater fullness perception in mouth and higher color intensity. This results support the positive effect of water stress in the sensory properties of wine that was corroborated in the study of Talaverano et al. (2018) that demonstrate the link between irrigation treatment and wine aroma compounds, although their composition was strongly influenced by the season. This last find, visually, can be explained in a better way by observing the principal component analysis (PCA) relating irrigation treatments and sensory parameters in both years (Fig. 3), in which PC1 and PC2 accounted for 60.4% of the total variation (33.0% and 27.4% respectively), PC1 being mainly linked to fullness, astringency and persistency while PC2 was associated to color intensity and red fruits. PC3 explained another 16% of the variation and was mainly linked to Bitterness (data not shown). In contrast to the results from Fig. 2, by observing the PCA (Fig. 3) there is no apparent contrast between the wines of 2014 and 2015 but, in general, in 2015, T3 was correlated with red fruits and persistency and much less with color intensity, instead astringency and fullness was correlated with T2 in 2014 and T1 in 2015 corroborating the differences between the chemical composition and the sensory composition of the wines that could be influenced by differences in climatic parameters between season (Table 1).

4. Conclusions

The results of 2-years experiment showed that controlled water deficit modify the chemical composition in wines, especially in terms of total phenols, total anthocyanins and chroma (C*). Regarding the

fractionation of proanthocyanidins, water deficit did not result in differences in concentration or composition of the different flavanol fractions in wines. Also, wines from the more restrictive irrigation practice results highly appreciated sensory properties like more pronounced red fruits aromas, more fullness perception in mouth and more color intensity. Still, some sensory characteristics in wines were strongly influenced by the year. Further studies in this area that take into account other compounds that may influence wine composition (i.e polysaccharides) and the effect of irrigation treatments on the wine chemical composition during aging are necessary to determine how the water deficit could improve the phenolic and sensory composition of the wines, especially in a climate change context, where the water supply is thought to decline worldwide. Finally, it is important to remark that the impact of irrigation regimes on grape berry composition cannot be simply extrapolated to the resulting wines.

Acknowledgements

This work was supported through funding from the Comisión Nacional de Investigación Científica y Tecnológica (CONICYT), Fondecyt Postdoctoral Fund (grant no. 3140269). The authors thanks Haras de Pirque vineyards for their collaboration with the plant material and field support.

References

- Acevedo-Opazo, C., Ortega-Farías, S., Fuentes, S., 2010. Effects on grapevine (*Vitis vinifera* L.) Water status on Water consumption, vegetative growth and grape quality: an irrigation scheduling application to achieve regulated deficit irrigation. *Agric. Water Manag.* 97, 956–964.
- Bautista-Ortín, A., Martínez-Hernández, A., Ruiz-García, Y., Rocío Gil-Muñoz, R., Gómez-Plaza, E., 2016. Anthocyanins influence tannin-cell wall interactions. *Food Chem.* 206, 239–248.
- Bindon, K., Madani, S., Pendleton, P., Smith, P., Kennedy, J., 2014. Factors affecting skin tannin extractability in ripening grapes. *J. Agric. Food Chem.* 62, 1130–1141.
- Bindon, K., Li, S., Kassara, S., Smith, P., 2016. Retention of proanthocyanidin in wine-like solution is conferred by a dynamic interaction between soluble and insoluble grape cell wall components. *J. Agric. Food Chem.* 64, 8406–8419.
- Bonada, M., Jeffery, D., Petrie, P., Moran, M., Sadras, V., 2015. Impact of elevated temperature and water deficit on the chemical and sensory profiles of Barossa Shiraz grapes and wines. *Aust. J. Grape Wine Res.* 21, 240–253.
- Boss, P.K., Davies, C., Robinson, S., 1996. Analysis of the expression of anthocyanin pathway genes in developing *Vitis vinifera* cv. Shiraz grape berries and the implications for pathway regulation. *Plant Physiol.* 111, 1059–1066.
- Brossaud, F., Cheynier, V., Noble, A., 2001. Bitterness and astringency of grape and wine polyphenols. *Aust. J. Grape Wine Res.* 7, 33–39.
- Busse-Valverde, N., Bautista-Ortín, A., Gómez-Plaza, E., Fernández-Fernández, J., Gil-Muñoz, R., 2012. Influence of skin maceration time on the proanthocyanidin content

- of red wines. *Eur. Food Res. Technol.* 235, 1117–1123.
- Busse-Valverde, N., Gómez-Plaza, E., López-Roca, J.N., 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. *J. Agric. Food Chem.* 58, 11333–11339.
- Cáceres, A., Peña-Neira, A., Gálvez, A., Obreque-Slier, E., López-Solís, R., Canals, J.M., 2012. Phenolic compositions of grapes and wines from cultivar Cabernet Sauvignon produced in Chile and their relationship to commercial value. *J. Agric. Food Chem.* 60, 8694–8702.
- Cáceres-Mella, A., Talaverano, M.I., Villalobos-González, L., Ribalta-Pizarro, C., Pastenes, C., 2017. Controlled water deficit during ripening affects proanthocyanidin synthesis, concentration and composition in Cabernet Sauvignon grape skins. *Plant Physiol. Biochem.* 117, 34–41.
- Casassa, L., Keller, M., Harbertson, J., 2015. Regulated deficit irrigation alters anthocyanins, tannins and sensory properties of Cabernet Sauvignon grapes and wines. *Molecules* 20, 7820–7844.
- Castellarin, S., Matthews, M., Gaspero, C., Gambetta, G., 2007. Water deficits accelerate ripening and induce changes in gene expression regulating flavonoid biosynthesis in grape berries. *Planta* 227, 101–112.
- Chaves, M., Zarrouk, O., Francisco, R., Costa, J., Santos, T., Regalado, A., Rodrigues, M., Lopes, C., 2010. Grapevine under deficit irrigation: hints from physiological and molecular data. *Ann. Bot.* 105, 661–676.
- Deluc, L., Quilici, D., Decendit, A., Grimplet, J., Wheatley, M., Schlauch, K., Méridon, J.M., Cushman, J., Cramer, G., 2009. Water deficit alters differentially metabolic pathways affecting important flavor and quality traits in grape berries of Cabernet Sauvignon and Chardonnay. *BMC Genomics* 10, 212.
- Fanzone, M., Peña-Neira, A., Gil, M., Jofré, V., Assof, M., Zamora, F., 2012. Impact of phenolic and polysaccharidic composition on commercial value of Argentinean Malbec and Cabernet Sauvignon wines. *Food Res. Int.* 45, 402–414.
- Gawel, R., Iland, P.G., Francis, L.L., 2001. Characterizing the astringency of red wine: a case study. *Food Qual. Prefer.* 12, 83–94.
- Genebra, T., Santos, R., Francisco, R., Pinto-Marijuan, M., Brossa, R., Serra, A., Duarte, C., Chaves, M., Zarrouk, O., 2014. Proanthocyanidin accumulation and biosynthesis are modulated by the irrigation regime in Tempranillo seeds. *Int. J. Mol. Sci.* 15, 11862–11877.
- Glories, Y., 1984. La couleur des vins rouges, 2eme Partier. Mesure, origine et interpretation. *Connaiss. Vigne Vin.* 18, 253–271.
- IPCC, 2007. *Climate Change 2007: the Physical Basis Summary for Policy Makers*. Cambridge University Press, Cambridge.
- Jones, G., White, M., Cooper, O., Storchmann, K., 2005. Climate change and global wine quality. *Climatic Change* 73, 319–343.
- Kennedy, J., Matthews, M., Waterhouse, A., 2002. Effect of maturity and vine water status on grape skin and wine flavonoids. *Am. J. Enol. Vitic.* 53, 268–274.
- Lacampagne, S., Gagné, S., Gény, L., 2010. Involvement of abscisic acid in controlling the proanthocyanidin biosynthesis pathway in grape skin: New elements regarding the regulation of tannin composition and leucoanthocyanidin reductase (LAR) and anthocyanidins reductase (ANR) activities and expression. *J. Plant Growth Regul.* 29, 81–90.
- Montes, C., Pérez-Quezada, J., Peña-Neira, A., Tonietto, J., 2012. Climatic potential for viticulture in Central Chile. *Aust. J. Grape Wine Res.* 18, 20–28.
- OIV, 2012. *Compendium of International Methods of Wine and Must Analysis*, vol I-II Sect 3. OIVEd., Paris.
- Payne, C., Bowyer, P., Herderich, M., Bastian, S., 2009. Interaction of astringent grape seed procyanidins with oral epithelial cells. *Food Chem.* 115, 551–557.
- Pérez-Magariño, S., Sánchez-Iglesias, M., Ortega-Heras, M., González-Huerta, C., González-Sanjosé, M., 2007. Colour stabilization of red wines by microoxygenation treatment before malolactic fermentation. *Food Chem.* 101, 881–893.
- Ribéreau-Gayon, J., Stonestreet, E., 1965. Le dosage des anthocyanes dans le vin rouge. *Bull. Soc. Chim. Fr.* 9, 2649–2652.
- Roby, G., Bisson, L., Adams, D., Matthews, M., 2004. Berry size and vine water deficits as factors in winegrape composition: anthocyanins and tannins. *Aust. J. Grape Wine Res.* 10, 100–107.
- Sacchi, K., Bisson, L., Adams, D., 2005. A review of the effect of winemaking techniques on phenolic extraction in red wines. *Am. J. Enol. Vitic.* 56, 197–206.
- Santos, T., Lopes, C., Rodrigues, M., De Souza, C., Ricardo-Da-Silva, J., Maroco, J., Pereira, J., Chaves, M., 2005. Effects of partial root-zone drying irrigation on cluster microclimate and fruit composition of field-grown Castela grapevines. *Vitis* 44, 117–125.
- Sameckis, C., Damberg, R., Jones, P., Mercurio, M., Herderich, M., Smith, P., 2006. Quantification of condensed tannins by precipitation with methylcellulose: development and validation of an optimized tool for grape and wine analysis. *Aust. J. Grape Wine Res.* 12, 39–49.
- Sun, B., Leandro, C., Ricardo da Silva, J., Spranger, I., 1998a. Separation of grape and wine proanthocyanidins according to their degree of polymerization. *J. Agric. Food Chem.* 46, 1390–1396.
- Sun, B., Ricardo da Silva, J., Spranger, I., 1998b. Critical factors of the vanillin assay from catechins and proanthocyanidins. *J. Agric. Food Chem.* 46, 4267–4274.
- Talaverano, I., Ubeda, C., Cáceres-Mella, A., Valdés, M.E., Pastenes, C., Peña-Neira, A., 2018. Water stress and ripeness effects on the volatile composition of Cabernet Sauvignon wines. *J. Sci. Food Agric.* 98, 1140–1152.
- Vidal, S., Francis, L., Williams, P., Kwiatkowski, M., Gawel, R., Cheynier, V., Waters, E., 2004. The mouth-feel properties of polysaccharides and anthocyanins in a wine like medium. *Food Chem.* 85, 519–525.
- Villamor, R., Harbertson, J., Ross, C., 2009. Influence of tannin concentration, storage temperature, and time on chemical and sensory properties of Cabernet Sauvignon and Merlot wines. *Am. J. Enol. Vitic.* 60, 442–449.
- Villalobos-González, L., Peña-Neira, A., Ibáñez, F., Pastenes, C., 2016. Long-term effects of abscisic acid (ABA) on the grape berry phenylpropanoid pathway: gene expression and metabolite content. *Plant Physiol. Biochem.* 105, 213–223.
- Zarrouk, O., Francisco, R., Pinto-Marijuan, M., Brossa, R., Santos, R., Pinheiro, C., Chaves, M., 2012. Impact of irrigation regime on berry development and flavonoids composition in Aragonez (syn. Tempranillo) grapevine. *Agric. Water Manag.* 114, 18–29.