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Impact of pre-fermentative mash cooling and heating on anthocyanin concentration and color of Teran wines

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Abstract

Six vinification treatments, including control wine (TK7) were carried out to investigate: the impact of 48-h pre-fermentative mash cooling (8 °C) followed by maceration of 13 days (TC15), 28 days (TC30) and *saignée* technique (TCS15); and the impact of 48-h heating (50 °C) followed by maceration of 13 days (TH15) and 28 days (TH30) on anthocyanin concentration and color in Teran wines. Anthocyanins, intensity, hue and composition of color were analyzed by UV/Vis spectrometry. The results showed that total anthocyanins and color hue values in all treatments were higher compared to control wine. The highest concentrations of free anthocyanins were found in wines with pre-fermentative heating treatment. Color intensity exposed the best results in 15-day macerations regardless of pre-fermentative treatment.

Key words: Teran wines, pre-fermentation treatments, anthocyanins, color intensity, color hue

Introduction

The color of red wines is one of the most important sensory attributes and is related to the extraction of anthocyanins from grape skins during winemaking. There are several hundred known anthocyanins and anthocyanin-derived pigments with diverse structures and properties (Sáenz-Navajas et al., 2011), responsible for color in red grapes and wines. They are accumulated in berry skins during the grape ripening (Perez-Lamela et al., 2007). Sacchi et al. (2005) report that the following winemaking variables and techniques increase phenolic composition: fermentation temperature, thermovinification, must freezing, *saignée*, pectolytic enzyme treatments, and extended macerations. In wines, monomeric anthocyanins can also undergo a wide variety of reactions and associations and various anthocyanin-derived new pigments are formed, which are extremely crucial for color stability (He et al., 2012). Many winemaking techniques have been developed to improve color extraction to obtain products with the market demanding characteristic (Puertas et al., 2008). Basic thermovinification entails short heating of the skins from 50 to 80 °C extracting them with the juice, pressing, and then cooling before fermentation. If heating at the same temperature before fermentation is extended for a longer period (for instance, up to 24 h), the process is called pre-fermentative heat treatment (Escudier et al., 2008; Rossi et al., 2022). The heat damages the hypodermal cell membranes, releasing anthocyanins, and it also denatures polyphenol oxidase, preventing browning. Since there is no alcohol present at the time of heating, it would not be expected to increase tannin extraction (Sacchi et al., 2005). Pre-fermentative cold maceration, also known as cold soaking or cryomaceration, is being increasingly used by enologists worldwide in order to improve some important quality characteristics of wines such as color and aroma. This technique consists in maintaining the crushed grapes at low temperatures (5–10 °C) (Heredia et al., 2010). In pre-fermentation

juice runoff which is also known as *saignée*, the juice is removed before fermentation, thus increasing the skin to juice ratio (Sacchi et al., 2005). Teran (*Vitis vinifera* L.) is the most widespread red autochthonous cultivar on the Istrian peninsula, Croatia (Plavša et al., 2012; Rossi et al., 2022). The aim of this study was to investigate how pre-fermentation cooling or heating treatments, *saignée* technique, and various maceration durations affect the anthocyanin concentrations and parameters of color in Teran red wine.

Material and methods

The grapes of cv. Teran (*Vitis vinifera* L.) were grown in Western Istria, the town of Poreč, in a typical Istrian terroir. The harvest was held in 2020 when the sugar content was measured at 18.9 °Brix, 8.0 g L⁻¹ of total acidity expressed as tartaric acid, and pH 3.2. On the same day, manually harvested grapes were destemmed and crushed with standard equipment and homogenized. Red grape mash was equally divided according to the plan of the experiment (Table 1). Five vinification treatments were submitted to pre-fermentative mash cooling at 8 °C (cryomaceration) or mash heating at 50 °C for 48 hours, proceeded with fermentation at 24 °C and followed by prolonged maceration in two periods of duration: 13 days, respectively 15 days in total including pre-fermentative process (TC15; TH15) and 28 days, in total 30 days (TC30; TH30). In one of the treatments with pre-fermentative mash cooling, the *saignée* technique was performed before fermentation. A proportion (33 %) of the total juice quantity was racked and concentrated mash proceeded with fermentation (24 °C) and prolonged maceration of 13 days, 15 days in total (TCS15). This experiment has also included a control treatment (TK7), with a standard technique of a 7-day maceration and a maceration/fermentation temperature of 24 °C. Every treatment was performed in three replications in 220 L-stainless steel tanks. All grape mashes were inoculated with 30 g hl⁻¹ of selected *Saccharomyces cerevisiae* dry yeast (Fermol Mediterranee, AEB), and chaptalized with 3 kg hl⁻¹ of saccharose. After the end of the maceration process, fermented mashes were pressed and wine was racked in clean tanks. After six months the wine was bottled and stored prior to analysis.

Table 1. Overview of the experiment: pre-fermentative treatments, winemaking techniques and maceration duration in Teran wine treatments

Treatment*	Pre-fermentative treatment	Fermentation and maceration			Maceration duration including pre-fermentative treatment
	Duration – 48 hours	Winemaking technique	Maceration/fermentation temperature	Maceration duration	
TCS15	Cooling - at 8 °C	<i>Saignée</i> technique	24 °C	13 days	15 days
TC15				13 days	15 days
TC30		Prolonged maceration		28 days	30 days
TH15	Heating - at 50 °C			15 days	15 days
TH30				28 days	30 days
TK7	/	Standard technique			7 days

*T-Teran; C-cooling; H-heating; S- *saignée*; K-control; 7,15,30 – days of maceration duration in total

The color intensity and composition i.e. partial contribution to the full color were determined according to Glories (1984) using a Cary 50 UV/Vis spectrophotometer (Varian Inc., Harbour City, CA, USA). The method is based on the determination of absorbances values on three different wavelengths; 420 nm, 520 nm, and 620 nm. Glories (1984) modified the definition of color intensity (CI') as a sum of absorbances (A) on those three wavelengths: CI' = A₄₂₀ + A₅₂₀ + A₆₂₀. Partial contribution to the full color was calculated using this

formulas: for % of red color = $(A_{520} \cdot 100) / 100$; % of yellow = $(A_{420} \cdot 100) / 100$; % of blue = $(A_{620} \cdot 100) / 100$. Color hue was determined using the method by Sudraud (1958) defined as a ratio of absorbances A_{420} and A_{520} . Total and free anthocyanin content was analyzed according to the methods described by Di Stefano (1989). Briefly, methods are based on the reading of absorbances at wavelengths between 536 and 540 nm using the solutions of wine and other chemicals previously prepared according to the described instructions. The highest absorbance is calculated and given results are expressed as mg l^{-1} . One-way analysis of variance (ANOVA) and Fisher's least significance difference (LSD) test was used to compare mean values ($p \leq 0.05$). Statistical analyses were performed using Statistica 10.0. software (Sta-Soft Inc. Tulsa, OK).

Results and discussion

Total anthocyanin concentration in all treatments differed significantly in comparison to the control wine ($p \leq 0.05$) (Figure 1). The highest value was found in treatment subjected to the pre-fermentative heating process, TH15. Treatments submitted to the diverse duration of prolonged maceration TCS15, TC15 and, TH30 showed no significant difference. Boulton (2001) reports that many studies showed additional contact time between skins and wine cannot provide additional anthocyanin content or color. Regarding free anthocyanins, it was found that treatments that underwent pre-fermentative heating process (TH15 and TH30) were statistically the highest and different compared to others (Figure 2). There was no difference between pre-fermentative cooling processes (TCS15, TC15 and, TC30) regardless of maceration duration. Heredia et al. (2010) exhibit that low temperatures can influence phenolic co-pigmentation power which means they can reduce free anthocyanin content as in our case in comparison to heating treatment.

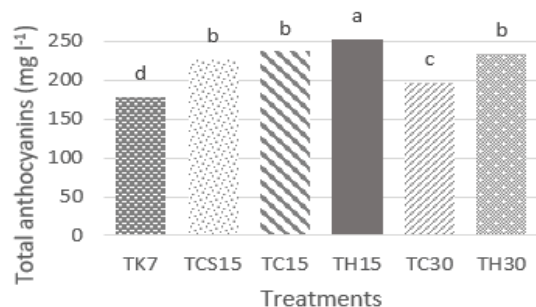


Figure 1. Total anthocyanin concentrations (mg L^{-1}) in Teran wines

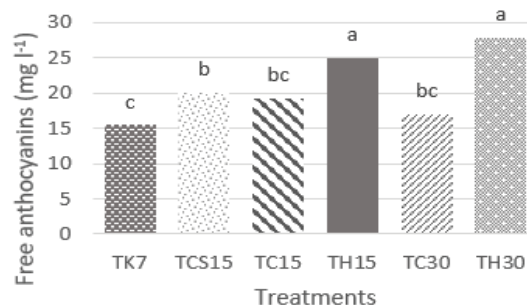


Figure 2. Free anthocyanin concentrations (mg L^{-1}) in Teran wines

Lower-case letters above column represent significant differences at $p \leq 0.05$ level according to the LSD test

The color intensity results showed that treatments TCS15, TC15, and TH15 were significantly different in comparison to other samples ($p \leq 0.05$) (Table 2). Also was found that intensity in 30-day maceration treatment (TC30) was statistically lower than in control wine (TK7). A decrease in color intensity in TC30 treatment could be related to a lower concentration of total anthocyanins (Figure 2). Gonzalez-Neves et al. (2010) suggest that the decrease of anthocyanins in prolonged macerations is a result of post-fermentation fixation by yeasts and solids. Color hue results presented that control wine (TK7) was significantly the lowest in comparison to other treatments. The highest value was obtained in TC30 treatment, and compared to the other 30-day maceration treatment (TH30) was different. There was no statistical difference in color hue values between cooling and heating pre-fermentative processes in treatments with 15-day maceration (TCS15, TC15, TH15) (Table

2). There was a significant difference in the distribution of yellow color between cooling and heating processes but only in 30-days maceration treatments (TC30 and TH30) (Table 2). The highest value was obtained in TC30 treatment, which might be related to color hue results, because several authors noted that tone (hue) and proportion of yellow increase during winemaking and aging due to oxidation processes (Puertas et al., 2008). Treatments TCS15, TC15 and, TH15 did not differ significantly in distribution of yellow color. Regarding blue color distribution, it increased in all treatments, except TC15, in comparison to control wine (TK7). The highest distribution value of blue color was found in treatment TH30. Other treatments (TCS15, TH15 and, TC30) were identical regardless of pre-fermentative process and maceration duration. Boulton (2001) report that blue color increases with winemaking and also with aging due to anthocyanins condensation and polymerization with other polyphenols, which might be the case in wines from our experiment. The distribution of red color was significantly the highest in control wine (TK7). On the contrary Puertas et al. (2008) obtain that percentage of red color is the lowest in control wine in comparison to other investigated treatments. Red color values obtained in treatments with 15-day maceration were equal, and higher than in 30-day maceration treatments despite to used pre-fermentative process (Table 2).

Table 2. Color intensity, color hue values and percentage of red, yellow and blue color in Teran red wines

	Color intensity	Color hue	Red color (%)	Yellow color (%)	Blue color (%)
TCS15	0.94 ± 0.0058a	0.73 ± 0.0038c	49.40 ± 0.0364b	35.87 ± 0.1631c	14.73 ± 0.1268b
TC15	0.94 ± 0.0222a	0.72 ± 0.0068c	49.66 ± 0.1491b	35.89 ± 0.2293c	14.45 ± 0.0813c
TH15	0.95 ± 0.0288a	0.72 ± 0.0075c	49.56 ± 0.2094b	35.82 ± 0.223cd	14.63 ± 0.0267b
TC30	0.73 ± 0.0014d	0.76 ± 0.0047a	48.55 ± 0.1875c	36.81 ± 0.0854a	14.64 ± 0.1026b
TH30	0.85 ± 0.0022b	0.74 ± 0.0044b	48.68 ± 0.1098c	36.26 ± 0.1308b	15.06 ± 0.0235a
TK7	0.8 ± 0.0038c	0.1 ± 0.0069d	50.20 ± 0.2616a	35.51 ± 0.1751d	14.28 ± 0.1379c

Each value is the mean ± standard deviation, n=3. Lower-case letters represent significant differences at $p \leq 0.05$ level (LSD test).

Conclusions

Obtained results demonstrated the notable impact of pre-fermentative cooling and heating, together with prolonged macerations of Teran mash on free and total anthocyanins, color intensity, color hue, and percentage of red, yellow and blue color. Both pre-fermentative cooling and heating treatments positively affected the color intensity and hue, while pre-fermentative heating treatment significantly enhanced free anthocyanin concentration in Teran wines. On the basis of obtained results, wine producers can choose the appropriate winemaking technology for improving the quality of their products.

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Utjecaj predfermentacijskog hlađenja i zagrijavanja masulja na koncentraciju antocijana i boju vina sorte Teran (*Vitis vinifera* L.)

Sažetak

Provedeno je šest vinifikacijskih tretmana, uključujući kontrolu (TK7) kako bi se istražio: utjecaj 48 satnog predfermentacijskog tretmana hlađenja masulja (8 °C) koji se nastavio maceracijom od 13 dana (TC15), 28 dana (TC30), te tretmanom *saignée* tehnologije (TCS15); i utjecaj 48 satnog tretmana grijanja (50 °C), koji se nastavio maceracijom od 13 (TH15) i 28 dana (TH30) na koncentraciju antocijana i boje u vinima sorte Teran. Antocijani i parametri boje analizirani su spektrofotometrijski. Utvrđeno je da su ukupni antocijani i nijansa boje u svim tretmanima viši u odnosu na kontrolu. Najveće koncentracije slobodnih antocijana pronađene su u vinima s predfermentacijskim grijanjem. Najbolji rezultati intenziteta boje dobiveni su u tretmanima s 15 dana maceracije bez obzira na predfermentacijski tretman.

Ključne riječi: Teran, predfermentacijski tretmani, antocijani, intenzitet boje, nijansa boje