



Pre-fermentative cold maceration, *saignée*, and various thermal treatments as options for modulating volatile aroma and phenol profiles of red wine



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ABSTRACT

The effects of six maceration treatments on volatile aroma and phenol composition of Teran red wine were studied: standard maceration (control C), cold pre-fermentation maceration (CPM), *saignée* (S), pre-fermentation heating with extended maceration (PHT) or juice fermentation (PHP), and post-fermentation heating (POH). PHP wine contained the highest amounts of esters, fatty acids and anthocyanins, and the lowest content of other phenols. Alternative treatments decreased higher alcohols in relation to control C. CPM treatment lowered the extraction of seed tannins, exhibited the highest acetaldehyde, ethyl acetate and C₆-compounds levels, and had increased ester levels in relation to control C. POH wine contained the highest concentration of total phenols, flavonoids, monomeric, oligomeric and polymeric flavanols, and color intensity and hue. S and PHT wines contained lower amount of total phenols, but higher than in C and CPM wines. The calculated Odor Activity Values were used to establish significant differences between the treatments.

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1. Introduction

Volatile aromas and phenols are two of the most important groups of chemical compounds that determine the sensory quality of red wine (Sáenz-Navajas et al., 2015; Álvarez, Aleixandre, García, & Lizama, 2006). Aroma compounds originate from grapes, and are formed in fermentation and during wine maturation. They pertain to different chemical classes, such as monoterpenes, norisoprenoids, higher alcohols, fatty acids, esters, aldehydes, ketones, etc. (Callejón, Margulies, Hirson, & Ebeler, 2012). Phenolic compounds are mainly extracted from grapes during maceration (skin and seed contact), and are responsible for several important sensory properties of red wine, such as taste, mouthfeel and color (González-Neves, Favre, Gil, Ferrer, & Charamelo, 2015; Hufnagel & Hofmann, 2008). Among them, phenolic acids, flavonols and tannins participate in astringency and bitterness (Garrido & Borges, 2013; Hufnagel & Hofmann, 2008), while red wine color is mostly

determined by anthocyanins (Garrido et al., 2013; González-Neves et al., 2015).

Maceration conditions, such as duration and temperature, are the most important factors that modify the diffusion of varietal aromas and phenols from solid grape parts and their solubilization into must and wine (Sacchi, Bisson, & Adams, 2005). Maceration parameters also affect the formation and levels of fermentation aromas, although to a lesser extent (Callejón et al., 2012). Various maceration procedures in the production of red wines have been studied to establish the selective effects of different parameters on the extraction of important grape components. Several treatments, such as cold-soak pre-fermentative maceration, carbonic maceration, and *delestage*, exhibited contradictory or mixed results, mainly depending on the variety (Sacchi et al., 2005). Other techniques, developed earlier but revisited today in a completely different context, such as extended maceration (Baiano, Terracone, Gambacorta, & La Notte, 2009), *saignée* (Harbertson, Mireles, Harwood, Weller, & Ross, 2009), and maceration with heating (Baiano et al., 2009), were not studied extensively, and their effects are practically unknown. For example, it is estimated that in France currently 500 millions of liters of red wine are produced by short pre-fermentative heat treatment and liquid juice

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fermentation (Geffroy et al., 2015), however, very few studies have investigated the effects of this technique.

In general, a small number of studies directly compared the effects of a larger number of maceration procedures on the same grape material (Baiano et al., 2009; Harbertson et al., 2009; Mihnea, González-San José, Ortega-Heras, & Pérez-Magariño, 2015), while the majority of further studies were often limited in that they either confronted a relatively small number or mostly standard techniques. Their relative effectiveness could often be assessed only on the basis of the comparison of results from different sources, which could lead to misleading conclusions. Finally, a rather limited number of investigations were comprehensive in a way they studied the effect of maceration on both aromas and phenols (Fischer, Strasser, & Gutzler, 2000; González-Neves et al., 2015; Álvarez et al., 2006), which are inseparably related, and complementary in the context of wine sensory quality.

For all the mentioned reasons, the aim of this work was to evaluate the effect of certain common, and particular emerging maceration techniques as alternatives to standard procedures, on the volatile aroma and phenol composition of red wine: cold pre-fermentative fermentation, *saignée*, pre-fermentative heating followed by both traditional fermentative maceration and fermentation of juice, and post-fermentative heating. Total duration of all treatments except juice fermentation was extended to 20 days, to annul the effect of maceration duration. Equating the duration of treatments was expected to more precisely determine the effects of low temperature in pre-fermentation cold maceration treatment in relation to previous research, where in most cases, it involved an additional skin contact time in relation to the control (Cai et al., 2014; González-Neves et al., 2015; Mihnea et al., 2015).

2. Materials and methods

2.1. Grapes

The experiment was performed in harvest 2015 with grapes from Teran, native and the most widespread red grapevine variety (*Vitis vinifera* L.) grown in the Istria region of Croatia. The phenolic and volatile aroma profiles of Teran wine have been scarcely investigated to date. Teran grapes usually have difficulties reaching sugars levels higher than that equivalent to 12–12.5 vol% of ethanol, and for this reason, when wines with higher alcohol, acidity, and secondary metabolites content are produced (for example, when destined for long maturation), are partially dehydrated prior to harvest. The degree of ripeness was monitored by standard chemical analyses (sugars, total acidity, and pH), and average data at harvest were 108 °Oe (according to the Salleron's table), 10.7 g/L titratable acidity (as tartaric acid), and pH 3.03. In this work, the grapes were harvested manually on September 27, 2015, and collected in plastic cases of 22 kg capacity each.

2.2. Winemaking and maceration techniques

All the vinifications were done with randomly chosen 5 cases of grapes (approximately 100 L of grape mash) in 130 L stainless steel tanks. All treatments were conducted in duplicates after destemming, crushing and mashing of the grapes, and sulphiting of the mash (10 g/hL of potassium metabisulfite). Six maceration treatments were investigated:

- (i) C – control vinification: the mash was fermented on skins, and kept on the skins after fermentation, all at 23 ± 1 °C;
- (ii) CPM – cold pre-fermentative fermentation: the mash was kept at 5 °C for 5 days, then fermented and kept during post-fermentation at 23 ± 1 °C;

- (iii) S – *saignée*: the quantity of juice equal to 10% of the total volume was raked from the mash, and the following vinification was performed as for C treatment;
- (iv) PHT – pre-fermentative heating followed by traditional maceration: heating of mash was performed by submerging the tank into heated water in a water bath. The temperature of the mash was kept at 50 ± 2 °C for 6 h. The mash was stirred every 15 min to homogenize the temperature. After heating, the vinification was performed as for C;
- (v) PHP – pre-fermentative heating followed by pressing and fermentation of juice: the heating was the same as for (iv) PHT. After heating, mash was pressed while at high temperature, and transferred into another tank. The juice was fermented at 23 ± 1 °C until completion;
- (vi) POH – post-fermentative heating: the vinification was performed as for C treatment, and the last 5 days the fermented mash was heated at 37 ± 1 °C.

Mashes were inoculated with selected wine yeasts *Saccharomyces cerevisiae* Premium Zinfandel (Enologica Vason S. p.A., Verona, Italy) at 20 g/hL, and *Saccharomyces bayanus* Lalvin EC 1118 (Lallemand SA, Montreal, Canada) at 5 g/hL. The C, S, and POH treatment tanks were supplemented with inactivated yeast just after sulphiting, while for C, PHT and PHP treatments the addition of yeasts was done after heating or cooling, when temperature reached 17 °C. Yeast supplements (Active Red, Leafood Group S.r.L., Bari, Italy) were added in two portions of 20 g/L, the first on the third, and the second on the 10th day after inoculation. Two punch-downs per day during the first 10, and a single punch-down per day during the last 10 days of vinification were performed for all treatments except PHP. Total duration of all maceration treatments, except for PHP, was 20 days.

The pressing of all treatments was performed by the same pressing regime with a hydraulic basket press of 120 L capacity, Lancman VSPIX 120 (Gomark d.o.o., Vransko, Slovenia). After the completion of the treatments, the free-run wines were raked off, and then blended with the first press wines (0.8 bars). Wines were stored for 3 months at temperatures not exceeding 20 °C, raked off, filtered, and samples in 0.75-L bottles were taken for analyses. The level of free SO₂ was monitored throughout the whole process and was corrected to 20 mg/L after each critical step.

Standard physico-chemical wine parameters were determined according to the OIV methods, and the average values were: alcoholic strength $15.53 \pm 0.46\%$ vol., titratable acidity 9.71 ± 1.12 g/L (as tartaric acid), volatile acidity 0.64 ± 0.09 g/L (as acetic acid), and pH 3.21 ± 0.06 .

2.3. Chemical standards and standard solutions of volatile aroma and phenolic compounds

Methanol, formic acid, water and acetonitrile were of HPLC grade purity (Sigma-Aldrich, St. Louis, MO, USA).

Pure standards of phenols were purchased from Sigma-Aldrich, Acros Organics (Geel, Belgium), and Extrasynthese (Genay, France). Qualitative standards of *trans*-coutaric acid, *trans*-ferric acid, and *cis*-piceid were kindly donated by Dr. Urska Vrhovsek from Fondazione Edmund Mach (FEM), San Michele all'Adige, TN, Italy. The *cis*-isomers of hydroxycinnamic acids were obtained by UV illumination of a methanol solution of the *trans*-isomers for 4 h (Vrhovsek, 1998). Pure standards of aroma compounds were purchased from Merck (Darmstadt, Germany), Sigma-Aldrich, and Fluka (Buchs, Switzerland). Working standard solutions were prepared in synthetic wine containing 12 vol% of ethanol and 5 g/L of tartaric acid, adjusted to pH 3.2.

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