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# Evaluation of the potential of high pressure technology as an enological practice for red wines



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#### ABSTRACT

The impact of high hydrostatic pressure (HHP) treatments on the phenolic compounds composition of a red wine was studied after storage in order to evaluate the potential of this technology as an enological practice. Red wines pressurized at 500 and 600 MPa at 20 °C for 5 and 20 min, respectively, showed a lower content of monomeric anthocyanins (13 to 14%), phenolic acids (8 to 11%), and flavonols (14 to 19%) after 5 months of storage, when compared to the unpressurized wine. These results, together with the different degree of tannin polymerization and flavan-3-ol content in the pressurized wines led to propose an effect of HHP in the increase of polymerization and cleavage reactions of proanthocyanidins. The sensorial analysis of pressurized wines showed lower astringency, a higher intensity of cooked fruit aroma and lower intensity of fruity notes, when compared with the unpressurized wine. These effects are associated to those observed during wine aging.

Industrial relevance: During the last decade, the use of high hydrostatic pressure (HHP) as a non-thermal technology for food preservation and modification has increased substantially. Recently, pressure treatments have shown to influence long term red wine physicochemical and sensorial characteristics, leading to aged wine-like characteristics. Therefore, the use of HHP technology to modify wine composition could benefit the wine industry, especially to improve wines with low aging potential. This work shows that HHP can be potentially used as enological practice, modulating the organoleptic properties of wine by decreasing astringency and increasing pleasant aromas. It seems possible to exploit commercially the production of young red wines with distinct characteristics using this technology, addressing the market and consumer demand.

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## 1. Introduction

During the last decade, the use of high hydrostatic pressure (HHP) for food preservation and processing has increased substantially. Foods commercially processed by HHP are subjected to pressures around 400–600 MPa to destroy microorganisms and inactivate enzymes (Ramirez, Saraiva, Pérez Lamela, & Torres, 2009).

In the oenological sector, the use of high pressure treatments has already been tested for preservation of grape juice (Daoudi et al., 2002), must (Talcott, Brenes, Pires, & Del Pozo-Insfran, 2003), and wine (Morata et al., 2012). Some studies report that pressures between 200 and 500 MPa are able to inactivate bacteria and yeasts in red and white wines without causing significant sensorial changes (Buzrul, 2012; Chen et al., 2012; Mok et al., 2006; Puig, Olmos, Quevedo, Guamis, & Minguez, 2008; Puig, Vilavella, Daoudi, Guamis, & Mínguez, 2003), suggesting that HHP might be an alternative process to SO<sub>2</sub>

addition. Furthermore, the use of more severe high pressure treatments (650 MPa for 1 and 2 h) appeared to promote undesirable changes on the physicochemical characteristics of red wine, namely the decrease of color intensity and the content of phenolic compounds. In terms of sensorial properties, the sour and fruity aroma of the wine became weaker after 2 h of pressurization whereas the intensities of several gustatory attributes, including astringency and alcoholic and bitter taste, were enhanced slightly (Tao et al., 2012). Moreover, Corrales, Butz, & Tauscher (2008) revealed that combined temperature/pressure treatments (70 °C, 600 MPa) for 1 h affected the condensation reactions of anthocyanins. Higher pressure (≥600 MPa) and/or longer pressure holding time (≥1 h) can influence, immediately after the pressure treatment, the phenolic composition of the wine (Buzrul, 2012; Tao et al., 2012, 2013). Also, pressure treatments of 400-500 MPa for 5 min have shown to influence long term (above 6 months) red wine physicochemical and sensorial characteristics, namely more orangered color, and lower antioxidant activity, total phenolic content, and anthocyanins content due to an increase of phenolic compounds condensation reactions along storage. These changes led to aged-like wine

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characteristics, reflected on wines physicochemical properties, and also on sensorial appreciation (Santos, Nunes, Cappelle, et al., 2013). These results show that, in addition to pressure and processing time, aging time of the pressurized wines is also important for the final quality of the wines (Santos, Nunes, Cappelle, et al., 2013; Santos, Nunes, Rocha, et al., 2013).

During the aging of wine, phenolic compounds participate in several reactions, namely polymerizations and oxidations (Chen et al., 2012; Oliveira, Ferreira, De Freitas, & Silva, 2011; Ribéreau-Gayon, Glories, Maujean, & Dubourdieu, 2006; Soto Vázquez, Río Segade, & Orriols Fernández, 2010). HHP treatments promote reactions giving rise to sensorial characteristics of an aged-like wine (Santos, Nunes, Cappelle, et al., 2013; Santos, Nunes, Rocha, et al., 2013). It seems possible to exploit this technology for red wines production with lower storage time (<1 year) achieving novel pleasant and distinct characteristics to address particular market and consumer demands. Therefore, the use of HHP technology to modify wine composition could benefit the wine industry, especially to improve wines with low aging potential (Tao et al., 2014). The aim of this work was to study the effect of HHP treatments on the phenolic composition of a red wine after storage, since these compounds play an important role in wine color and taste that are important wine quality parameters. In this study two different high pressure treatments were selected, one mild (500 MPa/5 min) and other more severe (600 MPa/20 min). The treatment at 500 MPa/ 5 min was chosen because, in previous works (Santos, Nunes, Cappelle, et al., 2013), this treatment has already showed to influence long term (above 6 months) physicochemical and sensorial characteristics of sulfur-dioxide free red wine. The severe treatment with higher pressure (600 MPa) and longer time (20 min) was selected in order to enhance the changes in wine characteristics and to confirm the influence of HHP treatment on the wine characteristics. This study allows evaluating the feasibility of HHP use as an enological treatment to enhance its sensorial characteristics.

#### 2. Materials and methods

#### 2.1 Chemicals

Milli-Q water (Millipore, Bedford, MA) was used in all this work. HPLC-grade methanol, acetonitrile and formic acid were purchased from Merck (Darmstadt, Germany). Delphinidin-3-O-glucoside, cyanidin-3-O-glucoside, petunidin-3-O-glucoside, peonidin-3-O-glucoside, malvidin-3-O-glucoside, procyanidins B1, B2, B4 and C1, and phloroglucinol were purchased from Extrasynthese (Lyon, Genay — France). Gallic acid, protocatechuic acid, ferulic acid, caftaric acid, vanilic acid, caffeic acid, syringic acid, p-hydroxybenzoic acid, coutaric acid, p-coumaric acid, sinapic acid, chlorogenic acid, myricetin, quercetin, kaempferol, (+)-catechin, and (—)-epicatechin were purchased from Sigma-Aldrich Co. (St. Louis, MO, USA).

#### 2.2 Wine samples and high pressure treatments

Red wine samples were produced by Dão Sul SA (Carregal do Sal, Portugal) using Tinta Roriz (50%) and Touriga Nacional (50%) red grape varieties from Dão Appellation from 2013 harvest. An industrial batch fermenter of 16,000 L was used. After malolactic fermentation, the wine was transferred to 250 mL polyethylene bottles (commercially used to pasteurize fruit juices by high pressure, kindly supplied by Frubaça and produced by Espaçoplás — Indústria e comercialização de plásticos Lda, Portugal), stoppered with a polyethylene screw cap, and pressurized at 600 MPa during 20 min or 500 MPa during 5 min, at 20 °C (ten bottles for each treatment), in a hydrostatic press (Hiperbaric 55, Hiperbaric, Burgos, Spain), giving origin to samples 600 MPa and 500 MPa, respectively. The HHP equipment has a pressure vessel of 200 mm inner diameter and 2000 mm length and a maximum operation pressure of 600 MPa. It was connected to a refrigeration unit

(RMA KH 40 LT, Ferroli, San Bonifacio, Italy) that allowed to control the temperature of the input water used as a pressurizing fluid. Pressurizing water had a controlled temperature of 15 °C. Pressure build-up took place at a compression rate of about 300 MPa/min and adiabatic heating caused an increase in temperature of only 2.0 °C, while decompression was nearly instantaneous. A lot of the same wine was not submitted to high pressure treatment (*unpressurized*) and was also bottled in the polyethylene bottles. As polyethylene bottles were shown to have a little impact on the sensorial properties of the wine (Ghidossi et al., 2012), all wine samples (pressurized and unpressurized) were bottled in polyethylene bottles, to avoid this to be a factor affecting only the pressurized wines. All wines were stored at 80% relative humidity in the absence of light at room temperature ranging between 20 and 25 °C.

### 2.3 Oenological parameter determination

The ethanol content, titratable acidity, pH, volatile acidity, reducing sugars, free and bound SO<sub>2</sub>, HCl and gelatine index were determined for each wine according to the methods described by the International Organisation of Vine and Wine (OIV, 1990; Ough & Amerine, 1988; Ribéreau-Gayon et al., 2006). All analyses were carried out in triplicate.

#### 2.4 Color determination

The color intensity (CI) was calculated as the sum of the absorbance values at 420 nm, 520 nm, and 620 nm and the color tonality was determined by dividing the absorbance at 420 nm by the absorbance at 520 nm. Absorbance measurements were recorded on an Uvikon 922 spectrophotometer (Kontron Instruments, Saint Quentin en Yvelines, France). The contribution of each coloration (yellow, red, and blue) to the overall color of wine was calculated by dividing the absorbance at 420 nm (Ye%), 520 nm (Rd%), and 620 nm (Bl%) by the color intensity (CI). The proportion of red coloration produced by free and bound anthocyanins under their flavylium cations form (dA%) was calculated using the following formula, as describe by Kelebek, Canbas, Jourdes, & Teissedre (2010):

$$dA\% = \left[1 - \frac{\textit{Abs}420 + \textit{Abs}620}{2 \times \textit{Abs}520}\right] \times 100.$$

#### 2.5 Phenolic composition by spectrophotometric methods

The total phenolic (TP) content of the samples was determined by the Folin–Ciocalteu method (Singleton, 1985). The samples were appropriately diluted in a solution of 10% ethanol. The calibration curve was performed using gallic acid as standard in a concentration range between 50 and 500 mg/L. The results were expressed as gallic acid equivalents.

Total proanthocyanidins were estimated according to Chira, Jourdes, & Teissedre (2011). This method is based on the Bate–Smith reaction, in which the proanthocyanidins in acid medium release anthocyanidins by heating. The wines were diluted to 1:50 in a 10% ethanol solution. One milliliter of the samples was added to 0.5 mL of water and 1.5 mL of 12 M HCl and the mixture was homogenized. Two tubes for each sample were prepared: one was heated for 30 min in boiled water (sample A), while the other one was maintained at room temperature (sample B). To each tube, 0.25 mL of 95% ethanol was added. The absorbance at 550 nm was then read through a 10 mm optical path. Total proanthocyanidins (g/L) were calculated as  $19.33 \times (abs_{550~nm}A - abs_{550~nm}B)$ .

Anthocyanins were determined using the  $SO_2$  bleaching method (Chira, Pacella, Jourdes, & Teissedre, 2011). A solution"A" was prepared as follows: 1 mL of wine, 1 mL of 0.1% HCl in ethanol and 20 mL of 2% aqueous HCl. Blank (B) was prepared as follows: 2 mL of solution "A" and 0.8 mL of water. Sample (S) was prepared as follows: 2 mL of

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