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Impact of cherry, acacia and oak chips on red wine phenolic parameters and sensory profile

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Abstract

Aim: The evolution of global phenolic parameters and the sensory profile of a Portuguese red wine aged for 90 days in contact with wood chips from different species were evaluated.

Methods and results: For this purpose, wood chips from cherry (*Prunus avium*), acacia (*Robinia pseudoacacia*) and two oak species (*Quercus petraea* and *Quercus pyrenaica*) were added to a Portuguese red wine. Various global phenolic parameters of red wines were studied during the aging process (90 storage days). In addition, a sensory analysis was made after the 90-day aging period to determine the impact of the use of different wood chip species on red wine sensory profile. The results showed that during the aging period, only a few differences were detected between the wines. However, after 90 aging days, in general the wines aged in contact with cherry wood tended to have the lowest values for several phenolic parameters. For sensory parameters, the wine aged in contact with French oak chips showed significantly higher scores for several aroma descriptors, while for visual and taste descriptors no statistical significant differences were found between the wines.

Conclusion: At the concentration used (3 g wood chips/L wine), the different wood chip species studied had no clear influence on the evolution of the majority of the red wine phenolic parameters. However, from a sensory point of view, the use of different wood species induced greater differentiation, especially for aroma descriptors.

Significance and impact of the study: The outcomes of this study would be of practical interest to winemakers and regulatory institutions since they could improve the knowledge of the impact of alternative wood chip species, namely acacia and cherry, on red wine quality.

Keywords: acacia, cherry, oak, phenolic composition, red wine, sensory profile

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Introduction

The use of wood during the process of wine fermentation and/or aging is an ancient and common practice in most of the world's wine producing regions. The main objectives of this practice are to enrich the wine with substances released by the wood, promote reactions due to contact with air diffused through the wood pores and develop certain interactive chemical reactions that take place slowly in wines and consequently improve wine's quality and organoleptic characteristics (Cerdán and Ancín-Azpilicueta, 2006; Izquierdo-Cañas *et al.*, 2016).

Traditionally, there are three species of wood used in barrel making: *Q. petraea* Liebl. and *Q. robur* L., the most common oak species in French forests, and the American oak Q. *alba* L. (Jordão *et al.*, 2005; Jordão *et al.*, 2007; Cadahía *et al.*, 2009; Alañón *et al.*, 2011). Thus, the scientific literature contains a large number of data related to oak wood composition, namely volatile (Jordão *et al.*, 2005; Jordão *et al.*, 2006a; Chira and Teissedre, 2013) and phenolic profiles (Jordão *et al.*, 2007; Jordão *et al.*, 2012; Chira and Teissedre, 2013; 2015; Izquierdo-Cañas *et al.*, 2016), and its impact on wine chemical and sensory characteristics (De Coninck *et al.*, 2006; Gonçalves and Jordão, 2009; Oberholster *et al.*, 2015). However, it is important to note that the increasing demand for oak wood has caused a remarkable increase in production costs (due to the limited availability of materials) and environmental costs (due to intensive harvesting of oak trees in forest).

Thus, in recent years, heartwood from alternative species have been considered as possible sources of wood for the production of wine, such as acacia (*Robinia pseudoacacia*) and cherry (*Prunus avium*) (De Rosso *et al.*, 2009a; Chinnici *et al.*, 2011; Gortzi *et al.*, 2013). Acacia wood is generally characterized by a low content of extractable tannins and polyphenols (Citron, 2005; De Rosso *et al.*, 2009a). Its main extractive compounds are flavonoids (Sanz *et al.*, 2012a), and according to De Rosso *et al.* (2009a), it has a low content of non flavanoid compounds but a high content of aromatic aldehydes, particularly vanillin, syringaldehyde and dihydroxybenzaldehyde. On the other hand, cherry wood is characterized by high concentrations of flavonoid compounds, such as (+)-catechin, naringenin, isosakuranetin, aromadendrin, (+)-taxifolin, *p*-anisaldehyde and benzyl salicylate (Sanz *et al.*, 2010; 2012b; Fernández de Simón *et al.*, 2014). When compared to oak, this wood also has high levels of some volatile compounds such as methyl syringate and benzoic acid, and low levels of phenyl aldehydes and phenyl ketones, except vanillin and syringaldehyde (Sanz *et al.*, 2010; 2012b). Seasoned cherry wood is also characterized by high levels of condensed tannins and very low levels of hydrolysable tannins (De Rosso *et al.*, 2009a).

Some works reported the use of acacia and cherry barrels in wine aging (Kozlovic *et al.*, 2010; Chinnici *et al.*, 2011; Sanz *et al.*, 2012b; Fernández de Simón *et al.*, 2014; Chinnici *et al.*, 2015). However, despite the above-mentioned works demonstrating the value of cherry and acacia woods in cooperage, little information is available about the potential impact of the use of these wood species in the form of chips during the wine aging process. At the same time, there is scarce information about the comparison of the use of these alternative wood species with *Q. petraea* (from France) and *Q. pyrenaica* (from Iberian Peninsula) oak species.

Thus, this work intends to show the effect of acacia and cherry chips (using two different particle sizes for the latter wood species), in comparison with two oak chip species (*Q. petraea* and *Q. pyrenaica*), on time dependent changes in global phenolic composition and final sensory profile of a Portuguese red wine.

Materials and methods

1. Red wine

The wine used in this experiment was a blended red wine made from two Portuguese *Vitis vinifera* cv red grape varieties (Tinta Roriz, 80% and Touriga Nacional, 20%) harvested at the technological stage of ripeness in September of 2013. The red wine was made by the Casa da Passarella winery located in the Dão region, Portugal, following standard red winemaking technology, with a maceration time of 6 days. After alcoholic (sugar content below 2.5 g/L) and malolactic fermentation, the wine was kept under controlled conditions and the free SO_2 level was analyzed regularly. At the beginning of the wine aging process with wood chips, the main wine physico-chemical characteristics were the following: alcohol content 13.8% (v/v); pH 3.76; total acidity 5.59 g/L (expressed as tartaric acid); volatile acidity 0.46 g/L (expressed as acetic acid); free SO_2 29 mg/L; and total SO_2 54 mg/L. For global phenolic composition, the initial values were the following: total phenols 1981.3 mg/L (expressed as gallic acid); non flavonoid and flavonoid phenols 850.7 and 928.4 mg/L (expressed as gallic acid), respectively; total pigments 29.0 abs. units; total anthocyanins 518.9 mg/L (expressed as malvidin-3-glucoside); and color intensity 10.47 abs. units. After the aging process (90

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days), the main wine physico-chemical characteristics showed only slight differences: alcohol content 13.7% (v/v); pH 3.81; total acidity 5.48 g/L (expressed as tartaric acid); volatile acidity 0.51 g/L (expressed as acetic acid); free SO_2 23 mg/L; and total SO_2 48 mg/L.

2. Experimental conditions

The experimental aging conditions are listed in table 1. The wood materials used were acacia (*R. pseudoacacia*) chips from SAI (Paredes, Portugal), cherry (*P. avium*) and French oak (*Q. petraea*) chips from AEB Bioquímica (Viseu, Portugal), and Iberian oak (*Q. pyrenaica*) chips from J.M. Gonçalves (Palaçoulo, Portugal), with medium toasting (20 min at 160-170°C) and a particle size of 8 mm. For cherry wood, chips with a particle size of 2 mm (designated in the work as cherry powder) and medium toasting were also used. It was not possible to obtain Acacia chips with a particle size of 2 mm, as this size was not produced by the company that provided the samples. The red wine samples (10 litres each) were aged in contact with different wood chip species (3 g/L) in glass bottles for 90 storage days at cellar temperature (between 15-18°C) and stirred manually twice a week for 1 min. For each assay, only one repetition was made. A standard wine (without wood chips addition) was also considered. The wine samples were filtered before analysis.

Table 1 - Experimental conditions and respective red wine codes.

Experimental aging conditions	Wine codes
Wine + acacia wood chips	AWC
Wine + Portuguese oak wood chips	PWC
Wine + French oak wood chips	FWC
Wine + cherry wood chips	CWC
Wine + cherry wood powder	CWP
Wine (without any wood contact)	SW

3. General wine physico-chemical characterization

The general wine physico-chemical characterization (pH, total and volatile acidity, alcohol level, total and free SO_2) was made following the analytical methods recommended by the International Organisation of Vine and Wine (OIV, 2012).

4. Phenolic parameters

Total polyphenolic content was determined according to Ribéreau-Gayon *et al.* (2006), while non flavonoid and flavonoid phenols were determined using the improved method described by Kramling and Singleton (1969). In brief, the quantification of non flavonoid phenols is based on the determination of the phenolic content before and after the precipitation of flavonoids through the reaction with formaldehyde, under certain conditions (low pH, room temperature and darkness). After 24 hours, a dilution with distilled water (1:10) is carried out and the absorbance is read at 280 nm on a spectrophotometer. Flavonoid phenols come from subtracting non flavonoid phenols from total phenols. For these parameters, the results were expressed as gallic acid equivalents by means of calibration curves with standard gallic acid (Extrasynthese, Genay, France). Total pigments, total anthocyanins, colored anthocyanins, polymeric pigments, degree of ionization of anthocyanins, and degree of polymerization of pigments were quantified according to Somers and Evans (1977). For total and colored anthocyanins, the results were expressed as malvidin-3-monoglucoside equivalents by means of calibration curves with standard malvidin-3-monoglucoside (Extrasynthese, Genay, France). Color intensity at 420, 520 and 620 nm and color hue were evaluated following the methodology described by the OIV (2012).

Tanning power was quantified following the methodology developed by De Freitas and Mateus (2001). This method includes a 1:50 dilution with a hydroalcoholic solution (12% v/v, pH 3.2 at 20°C), followed by reading on a turbidimeter (d0). Then, 8 mL of the previous dilution and 300 µL of BSA (bovine serum albumin) are put in a tube and, after agitation and 45 min in the darkness, a second reading is carried out on the turbidimeter (d1). The final value (NTU/mL) was calculated from: Tanning power = (d1 - d0)/0.08.

All laboratory measurements were done immediately after wine samples were collected and were performed in triplicate (n = 3).

5. Sensory evaluation

The wine samples were immediately bottled after the 90-day aging period and evaluated two weeks after bottling by eleven expert judges with wine tasting experience (enologists and winemakers). Each red wine sample was stored for 24 hours at room temperature before sensory analysis, which was performed at 20-22°C in a sensory analysis room with individual booths for each expert and according to standardized procedures (ISO 3591, 1977). All evaluations were conducted in the morning from 10:00 am to 12:00 pm. Samples of 30 mL from each red wine sample were presented to the panel in

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tasting glasses marked with three digit numbers, in a randomized order.

The wines were evaluated using different attributes for color ("red" and "brown"), aroma ("fruity", "floral", "vanilla", "*boisé*", "toasted", "smoke", "spicy", "coconut", "coffee", "tobacco", "sawdust" and "balance") and taste ("bitterness", "astringency", "persistence" and "balance"). A final global appreciation of each wine was also evaluated. The experts scored each sensory attribute on a 1 to 5 point scale (1 = "absence"; 2 = "little intensity"; 3 = "moderate intensity"; 4 = "intense"; 5 = "high intensity") according to their sensory knowledge, training and experience; global appreciation was also scored on a 1 to 5 point scale (1 = "bad"; 2 = "pleasant"; 3 = "good"; 4 = "very good"; 5 = "excellent").

6. Statistical analysis

The data are presented as mean ± standard deviation. Phenolic and sensory parameters data were statistically tested by analysis of variance (ANOVA, one-way). Tukey test ($p < 0.05$) was applied to the

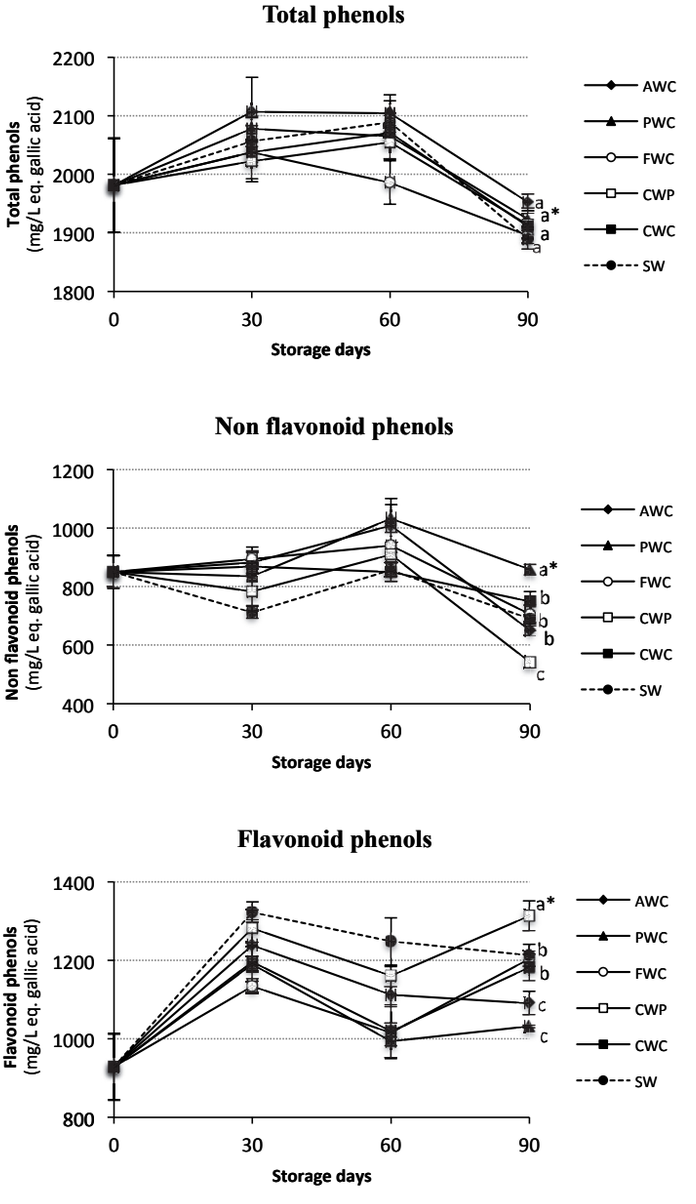


Figure 1 - Evolution of total, non flavonoid and flavonoid phenols from red wines aged in contact with different wood chip species for 90 storage days.

Wine codes in table 1; * Data points at 90 storage days with the same letter are not significantly different ($p < 0.05$); the error bars represent the standard deviation of the measurements within each observation.

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data to determine significant differences between wines. Principal component analysis (PCA) was used to analyze the data and study the relations between the red wines aged in contact with different wood chip species and their phenolic composition and sensory characteristics after 90 storage days. All analyses were performed using SPSS software version 24 (SPSS Inc., Chicago, IL, US).

Results and discussion

1. Evolution of global phenolic parameters

1.1. Total, non flavonoid and flavonoid phenols

Figure 1 shows the evolution of total, non flavonoid and flavonoid phenols during red wine aging in contact with different wood chip species for 90 storage days.

In general, for all red wines, total and non flavonoid phenols showed a slight increase in the first 60 storage days, followed by a decrease. Specifically, for total phenols after 30 storage days, wine samples aged in contact with acacia chips (2106 mg/L gallic acid equivalents) and Portuguese oak chips (2077 mg/L gallic acid equivalents) showed slightly higher values than the other red wine samples. For non flavonoid phenols, a tendency for a slight increase of the values was detected up to 60 storage days, followed also by a decrease in all wines.

The decrease in total and non flavonoid phenols between 60 and 90 storage days could be due to the precipitation of phenolic compounds and oxidation and polymerization phenomena, where phenolic leakage from wood plays an important role. It is also important to note that the wood chips concentration used in our study (3 g/L), although commonly used by winemakers, is much lower than that used in previous published works. In fact, the majority of the published works used oak chips at concentrations from 4 to 40 g/L (Arapitsas *et al.*, 2004; De Coninck *et al.*, 2006; Gonçalves and Jordão, 2009; Rudnitskaya *et al.*, 2009), while for acacia and cherry chips, the few published works until now do not give us a real perception of the wood chips concentration usually used. Thus, this fact could explain the low differentiation among the wines aged with different wood chip species and the control wine for the majority of the phenolic parameters evaluated.

After 90 storage days, total phenols content did not show significantly different values among all wines aged in contact with the different wood chip species (including the standard wine). However, for non flavonoid phenols, the values were significantly higher for the wine sample aged in contact with Portuguese oak chips (857.7 mg/L gallic acid equivalents) and significantly lower for the wine sample aged in contact with cherry powder (541.7 mg/L gallic acid equivalents). The remaining wines presented intermediate values, with no significant differences between them. The highest values of non flavonoid compounds in the wine aged in contact with Portuguese oak chips could correspond to a higher potential extraction of individual non flavonoid compounds, such as gallic, protocatechuic, vanillic, caffeic, syringic and *p*-coumaric acids, ellagitannins and ellagic acid from this oak wood species to the wine. According to previous work (Jordão *et al.*, 2007), oak wood, and in particular *Q. pyrenaica* species, is an important source of hydrolysable tannins, while for cherry heartwood an absence of hydrolysable tannins was already noticed (Sanz *et al.*, 2010), which confirms the significantly lower values of non flavonoid phenols quantified in our study. In addition, according to Chinnici *et al.* (2015), when compared to oak, cherry wood promotes a faster evolution of wine constitutive phenols, inducing a greater reduction of flavanol and flavonol phenols.

Finally, with respect to the evolution of flavonoid phenols, there was an evident increase of the values after 30 storage days for all wines. After, an evident decrease was detected for the wine aged in contact with acacia chips and the standard wine. The remaining wines showed a tendency for an oscillation of the values over time. At the end of the storage time, the wine aged in contact with cherry powder had a significantly higher value (1313.7 mg/L gallic acid equivalents), while the wines aged in contact with acacia and Portuguese oak chips showed significantly lower values (1031.4 and 1091.2 mg/L, respectively). The use of wood chips from cherry species with low particle size (2 mm) probably influenced the potential extraction of flavonoid phenols compounds from wood to the wine. This higher potential extraction was probably caused by the higher ratio between solvent (wine) and particle size when the smallest wood piece samples (2.0 mm of particle size) are used. Jordão *et al.* (2012) studied the phenolic content of different commercial oak chip samples and reported that for the same oak species and toasting level, a higher concentration of total phenols was found in the extracts produced from the oak samples with the smaller particle size.

Thus, the results concerning the effect of the different wood species used on the evolution of red wine global phenolic parameters (figure 1) are not evident.

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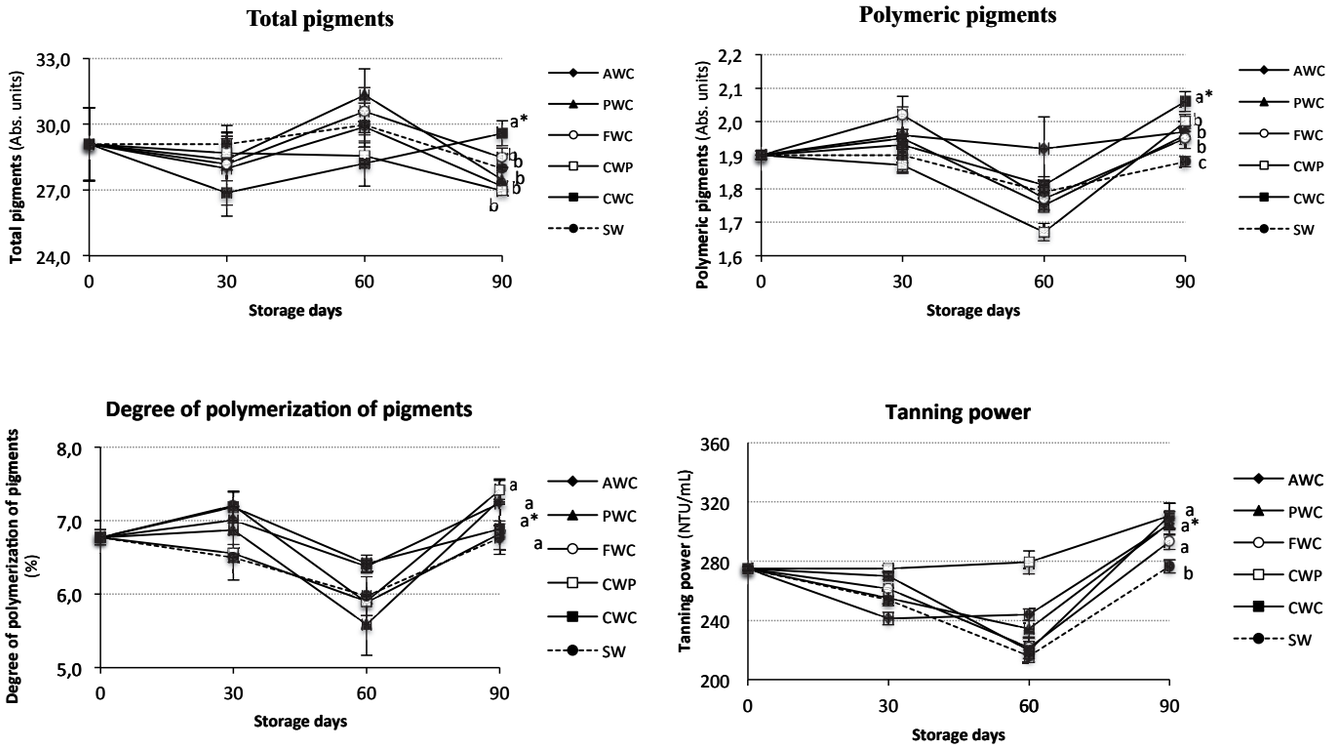


Figure 2 - Evolution of total and polymeric pigments, degree of polymerization of pigments and tanning power from red wines aged in contact with different wood chip species for 90 storage days.

Wine codes in table 1; * Data points at 90 storage days with the same letter are not significantly different ($p < 0.05$); the error bars represent the standard deviation of the measurements within each observation.

A similar tendency has previously been reported by other authors (Pomar and Gonzalez-Mendoza, 2001; De Coninck *et al.*, 2006). The storage time and/or wood chips concentration used was probably not enough to detect remarkable differences between the wines aged in contact with the different wood chip species used in this study. Recently, Chinnici *et al.* (2015) analyzed the changes in phenolic composition of red wines aged in cherry barrels and only detected significant changes in wine phenolic compounds after 4 months of storage, especially for flavanol compounds (procyanidin B1 and B2, (+)-catechin and (-)-epicatechin).

1.2. Total, polymeric and degree of polymerization of pigments

Concerning the evolution of total, polymeric and degree of polymerization of pigments (figure 2), it was possible to detect that in general values oscillated over time, with no clear differentiation among the wines. However, an exception occurred for the wine aged in contact with cherry chips, where there was a tendency for an increase of total pigments values. After 90 storage days, the wine aged in contact with cherry chips showed significantly higher values for total and polymeric pigments (29.59 and 2.06 abs. units, respectively), while the standard wine showed a significantly lower value for polymeric pigments (1.88 abs. units). For the remaining wines, there was no clear differentiation of the values. These results allow us to consider that the use of cherry chips could induce a faster evolution of phenolic compounds and a fast increase in the formation of derived and polymeric compounds. According to Chinnici *et al.* (2011), the use of cherry barrels resulted in a faster evolution of wine pigments with a fast increase in the formation of derived and polymeric compounds. Furthermore, cherry barrels proved to provide a favorable environment for oxidative reactions, thus making it less suitable for longer aging periods (De Rosso *et al.*, 2009b). In addition, it is important to note that the majority of the research works published analyzed the impact of different wood species, especially for the potential alternative wood species like acacia and cherry, by the use of barrels and not chips. This is an important factor because the use of barrels allows, in addition to the extraction of wood compounds, for a slow oxidation of certain compounds by atmospheric oxygen (which passes through wood pores), resulting

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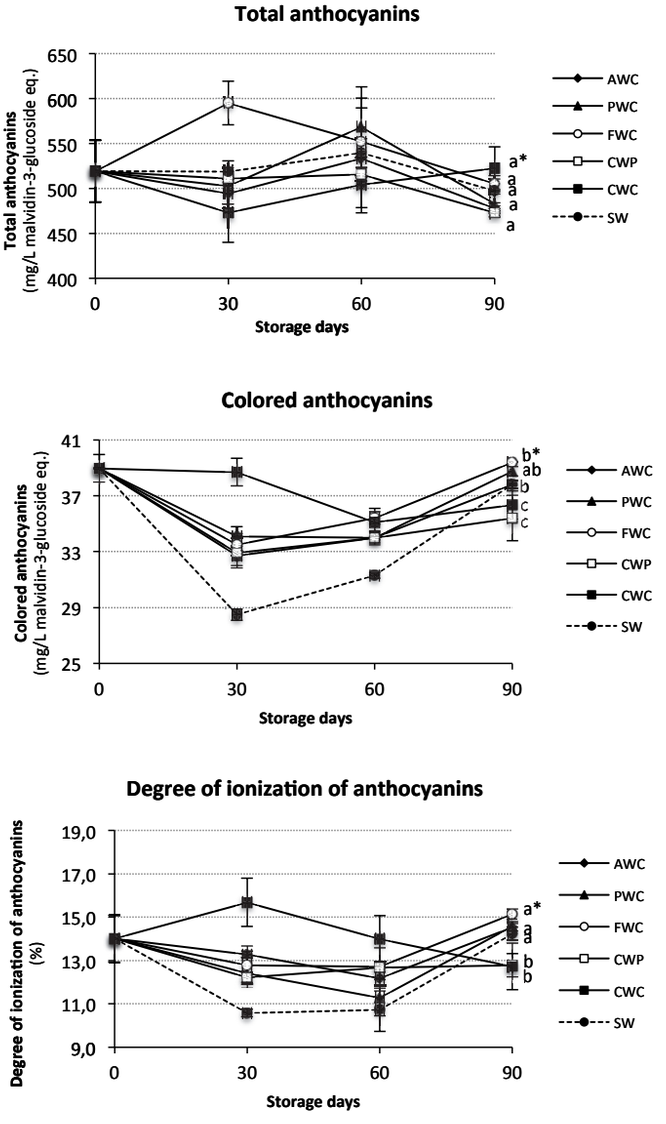


Figure 3. Evolution of total and colored anthocyanins and degree of ionization of anthocyanins from red wines aged in contact with different wood chip species for 90 storage days.

Wine codes in table 1; * Data points at 90 storage days with the same letter are not significantly different ($p < 0.05$); the error bars represent the standard deviation of the measurements within each observation.

in major changes in wine phenolic composition (Bozalongo *et al.*, 2007).

In figure 2, it is also possible to analyze the wine tanning power evolution during the 90 storage days. The tanning power stands for the expression of the "potential tannins" of a wine, namely the capacity of some tannins (such as proanthocyanidins with particular polymerization degree) to interact with proteins, influencing the astringent character of the wine at taste. In general, except for the wine aged with cherry powder, all wines showed a tendency for a slight decrease of tanning power during the first 60 storage days, followed by an increase. For the wine aged with cherry powder, a small but steady increase of the values was detected during all storage time. After 90 storage days, all wines aged in contact with different wood species showed significantly higher values than the standard wine. However, there were no significant differences among the wines aged with the different wood species. The final tanning power of the wines was (in descending order) the wines aged with cherry powder and chips (310.6 and 309.3 NTU/mL, respectively), the wine aged with Portuguese oak chips (305.5 NTU/mL), the wine aged with acacia chips (304.9 NTU/mL), the wine aged with French oak chips (293.1 NTU/mL) and the standard wine (276.4 NTU/mL).

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In fact, the phenolic composition that characterizes the woods (Jordão *et al.*, 2007; 2012; Sanz *et al.*, 2012b; Chira and Teissedre, 2015) and the extraction of these phenolic compounds from wood to wine, in particular ellagitannins which have a high reactivity with proteins, may explain the significantly high tanning power quantified in red wines aged in contact with the different wood species after 90 storage days and consequently the high level of interactions between these phenolic compounds and saliva proteins inducing a potentially higher level of wine astringency.

1.3. Anthocyanins and color parameters

The results obtained for total, colored and degree of ionization of anthocyanins during the red wine aging process are shown in figure 3. In general, total anthocyanins evolution was characterized by a very slight decrease of the values during the storage time considered (except for the wine aged with cherry chips, where a slight increase in total anthocyanins occurred after 30 storage days). The total anthocyanin decrease detected was probably due to anthocyanin condensation and polymerization reactions, and the precipitation of these compounds during wine aging (Pomar and Gonzalez-Mendoza, 2001; De Coninck *et al.*, 2006).

After 90 storage days, total anthocyanins content was not significantly different between the wines. However, the wine aged in contact with cherry chips showed the highest total anthocyanins content (522.1 mg/L of malvidin-3-monoglucoside equivalents), while the wine aged with cherry powder showed the lowest content (472.6 mg/L of malvidin-3-monoglucoside equivalents). Several authors reported that wines aged without oak wood contact showed less pronounced changes in anthocyanins content than wines aged in contact with chips or in oak barrels as a consequence of the lower level of tannin condensation (Del Alamo-Sanza *et al.*, 2004; Gonçalves and Jordão, 2009; Cristino *et al.*, 2013). Jordão *et al.* (2006b) reported in model wine solutions that individual anthocyanins, namely malvidin-3-monoglucoside, declined more rapidly in the presence of oak wood extracts than in their absence, with a subsequent decrease in red color. However, in our work it was not possible to detect this tendency. The use of a longer aging period or a higher wood chips concentration could be a factor to

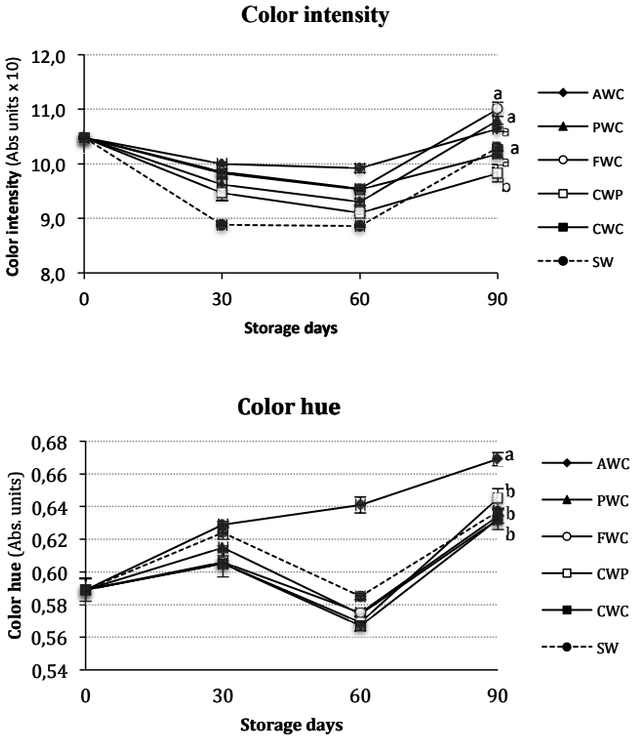


Figure 4. Evolution of color intensity and color hue from red wines aged in contact with different wood chip species for 90 storage days.

Wine codes in table 1; * Data points at 90 storage days with the same letter are not significantly different ($p < 0.05$); the error bars represent the standard deviation of the measurements within each observation.

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consider in the future to highlight eventual differences.

For colored anthocyanins and degree of ionization of anthocyanins, a similar evolution was detected (figure 3). Thus, in general, after a decrease in colored anthocyanins and degree of ionization of anthocyanins in the first 60 storage days (except for the standard wine, where the decrease was only evident for the first 30 storage days), a slight increase was detected for the majority of the wines. The standard wine showed the lowest values for colored anthocyanins and degree of ionization of anthocyanins during most of the storage time studied; however, after 90 storage days they were similar to those of the other wines. At the end of the storage time considered, the wines aged in contact with cherry wood (chips and powder) showed significantly lower values for colored anthocyanins (36.33 and 35.40 mg/L of malvidin-3-monoglucoside equivalents, respectively). The remaining wines showed no significant differences (39.40, 38.75, 37.82 and 37.80 mg/L of malvidin-3-monoglucoside equivalents for the wines aged in contact with French and Portuguese oak chips, acacia chips and standard wine, respectively).

The ionization of anthocyanins represents the percentage of anthocyanins in the red carbonium ion form I.e., anthocyanins in the colored form. Thus, for the degree of ionization of anthocyanins, the wines aged in contact with cherry wood also showed a tendency for a slight decrease of the values during the storage time considered. After 90 storage days, these wines showed significantly lower values (12.72 and 12.77% for wines aged in contact with cherry chips and powder, respectively). The highest degree of ionization of anthocyanins was detected in the wine aged in contact with French oak chips (15.12%).

The color intensity and hue results are shown in figure 4. Color intensity evolution followed the same trend observed for colored anthocyanins I.e., the standard wine showed the lowest values for most of the storage time, whereas after 90 storage days the lowest value was found in the wine aged in contact with cherry powder (9.83 abs. units). The remaining wines showed no significant differences (11.0, 10.8, 10.64, 10.3 and 10.18 abs. units for the wines aged in contact with French and Portuguese oak chips, acacia chips, standard wine and wine aged with cherry chips).

Several research works (Chinnici *et al.*, 2011; Gallego *et al.*, 2012) reported that the use of wood promotes pigment stabilization, namely anthocyanin pigments, while maintaining the highest color intensity and the best chromatic attributes of wines. However, in our study, the wine aging process in contact with the different wood species seems to have only a slight positive effect on the levels of color parameters (colored anthocyanins, degree of ionization of anthocyanins and color intensity) during the first 60 storage days in relation to the standard wine. However, after 90 storage days this effect was no longer observed.

Finally, for color hue, a tendency for an increase of the values, with some fluctuations, was observed during the storage time considered. The increase in color hue was more evident for the wine aged in contact with acacia chips than for the other wines. After 90 storage days, this wine showed the highest color hue values (0.66), while among the remaining wines the values were not significantly different. According to Psarra *et al.* (2015), polyphenols extracted from oak chips yield almost 6.4 times more reducing power compared to acacia chips. Therefore, the antioxidant effects exerted by polyphenols do not depend exclusively on their total amount, but also on the different polyphenolic structures (Aoun and Makris, 2013). This fact could help us to explain the increased browning of the wine aged in contact with acacia chips.

2. Sensory evaluation

A sensory evaluation of the wines aged in contact with different wood species was made after 90 storage days. The most significant differences were related to the majority of the wine aroma descriptors ("fruity", "floral", "vanilla", "*boisé*", "coconut" and "sawdust"). For visual and taste descriptors, no statistically significant differences were found (data not shown). A previous study of Pérez-Prieto *et al.* (2003) using a red wine made from Monastrell grapes showed that wine aging in oak barrel produced differences in all sensory descriptors, especially aroma descriptors.

Thus, figure 5 shows the aroma profile (considering only the descriptors where statistical differences were obtained) of the wines aged in contact with different wood species. According to the test panel, the wine aged in contact with French oak chips showed significantly higher scores for the "vanilla", "*boisé*" and "coconut" aroma descriptors, while the wine aged in contact with Portuguese oak chips showed significantly higher scores for "sawdust" but lower scores for the "fruity" and "floral" aroma descriptors. In addition, these wines showed intermediary scores for the "*boisé*" and "vanilla" aroma descriptors.

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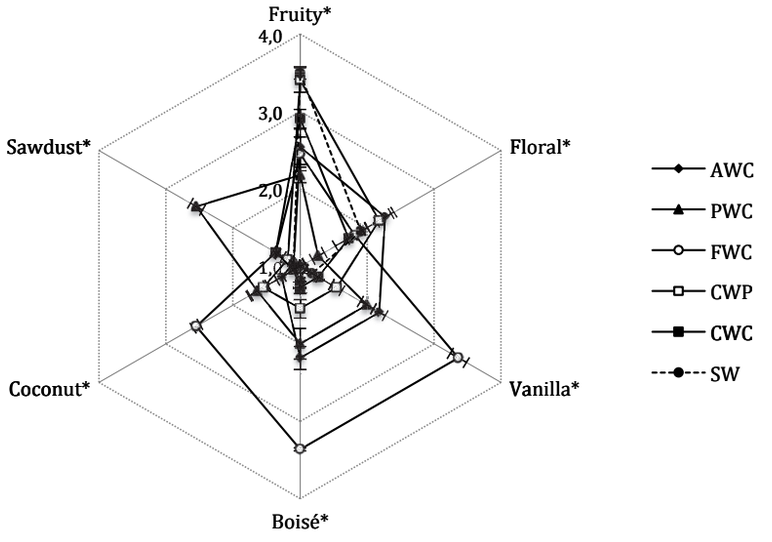


Figure 5. Sensory aroma profile from red wines aged in contact with different wood chip species after 90 storage days.
Wine codes in table 1; the error bars represent the standard deviation of the measurements within each sensory descriptor.

According to several research works (Gutiérrez-Afonso, 2002; De Coninck *et al.*, 2006), these differences could be explained as a result of higher levels of β-methyl-γ-octalactone (especially *cis*-β-methyl-γ-octalactone), vanillin, furfural, 5-methylfurfural and other volatile compounds in wines aged in contact with oak chips that had an important role in several wine aroma descriptors such as "coconut", "*boisé*" and "vanilla". In addition, according to Kozlovic *et al.* (2010), acacia barrels are less "aggressive" compared to oak and add less wood character to the wines.

3. Principal components analysis applied to wine phenolic and sensory characterization

To better understand the relationship between the different wood species used in wine aging, phenolic composition and sensory evaluation, a principal component analysis (PCA) was performed after 90 storage days. The corresponding loading plots that establish the relative importance of each variable are shown in figure 6. Thus, figures 6A and 6B show the relationship between the different wood species used in the wine aging process and the most relevant independent global phenolic parameters evaluated (total anthocyanins, total phenols, non flavonoid phenols, color intensity, degree of ionization of anthocyanins, total pigments, polymeric pigments and tanning power). The PCA (figure 6A) showed that the first two PCs explained 76.8% of the total variance. The first PC (PC1, 43.8% of the variance) was positively correlated with color intensity, degree of ionization of anthocyanins, non flavonoid phenols and total phenols. The second PC (PC2, 33.0% of the variance) was positively correlated with total anthocyanins, total pigments, polymeric pigments and tanning power and negatively correlated with degree of ionization of anthocyanins. In addition, it can be observed that the majority of the phenolic parameters are mainly located in the first quadrant.

In figure 6B, it is possible to visualize the spatial distribution of the samples evaluated concerning the global phenolic parameters considered. Thus, after a cluster analysis, one group is formed by the wine aged in contact with cherry chips; this wine is positioned on the positive side of PC1. The wine aged in contact with cherry chips was characterized by higher values of tanning power and polymeric pigments, while the wine aged in contact with French oak chips was characterized by higher values of degree of ionization of anthocyanins and color intensity. The wine aged in contact with Portuguese oak chips was characterized by higher values of total pigments, total anthocyanins and non flavonoid phenols, while the wine aged in contact acacia chips was characterized by higher values of total phenols.

Concerning the sensory evaluation, figures 6C and 6D show the relationship between the different wood species used in the wine aging process and the sensory descriptors. After 90 storage days, the PCA (figure 6C) showed that the first two PCs explained 72.30% of the total variance. The first PC (PC1, 42.0% of the variance) was positively correlated with "vanilla", "*boisé*", "toasted", "smoke", "coconut", "coffee", "tobacco" and "sawdust", and negatively

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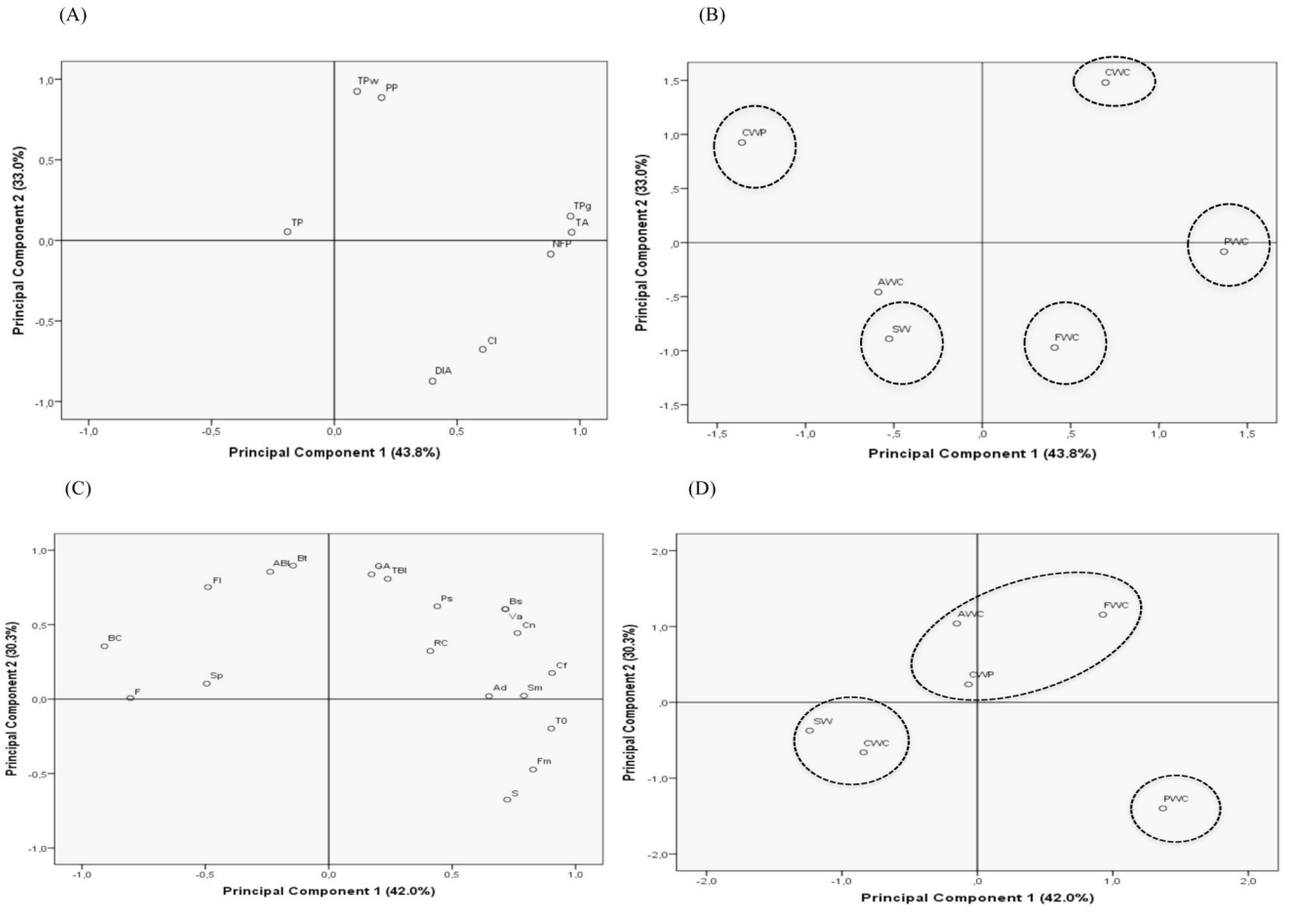


Figure 6 - Principal component analysis (PC1 and PC2) for phenolic parameters (A - loading plots and B - score plots) and sensory analysis (C - loading plots and D - score plots) in red wines aged in contact with different wood chip species after 90 storage days.

Wine codes in table 1; Phenolic parameters: TA - total anthocyanins; TP - total phenols; NFP - non flavonoid phenols; CI - color intensity; DIA - degree of ionization of anthocyanins; TPg - total pigments; PP - polymeric pigments; TPw - tanning power.
Sensory parameters: *Visual descriptors:* RC - red color; BC - brown color; *Aroma descriptors:* F - fruity; FI - floral; Va - vanilla; Bs - *boisé*; TO - toasted; Fm - smoke; Sp - spicy; Cn - coconut; Cf - coffee; Sm - tobacco; S - sawdust; ABL - balance; *Taste descriptors:* Bt - bitterness; Ad - astringency; Ps - persistence; TBL - balance; *Global evaluation:* GA - global appreciation.

correlated with "brown color" and "fruity". The second PC (PC2, 30.3% of the variance) was positively correlated with "floral", "vanilla", "*boisé*", "bitterness", "aroma balance", "persistence", "taste balance" and "global appreciation", and negatively correlated with "sawdust". It can also be observed that the majority of the sensory descriptors are mainly located in the first and second quadrants.

The spatial distribution of the wines evaluated concerning the sensory evaluation made by the use of different sensory descriptors is shown in figure 6D. Thus, one group is formed by the wines aged in contact with French oak and acacia chips and cherry powder; the wine aged in contact with French oak chips is positioned on the positive side of PC1 and PC2, while the wines aged in contact with acacia chips and cherry powder are positioned on the negative side of PC1 and on the positive side of PC2. The wine aged in contact with French oak chips was characterized by higher "*boisé*", "vanilla" and "coconut" descriptors. In addition, the wine aged with Portuguese oak chips is positioned on the positive side of PC1 and on the negative side of PC2 and was characterized by higher "sawdust" descriptor. Finally, another group formed by the wine aged in contact with cherry chips and standard wine positioned on the negative side of PC1 and PC2 was characterized by higher "fruity" descriptor.

Conclusion

In this work, it was not possible to detect a clear influence of the use of different wood chip species on

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the evolution of the global phenolic parameters evaluated. However, after the aging time considered (90 storage days), the wines aged in contact with cherry wood showed a slight tendency for lower values associated to red wine color parameters, such as color intensity, colored anthocyanins and degree of ionization of anthocyanins. From a sensory point of view, the wine aged in contact with French oak chips showed a tendency for higher aroma scores than the wines aged in contact with Portuguese oak, cherry and acacia chips. However, although acacia and cherry chips showed lower scores for aroma parameters, no statistical differences in visual and taste descriptors were found between wines aged with oak wood species and wines aged with alternative wood species (acacia and cherry). Thus, this result could be interesting to increase the knowledge of the potential use of these alternative wood species since the increasing demand for oak wood could induce a remarkable increase in production costs due to the limited availability of oak wood in the future. In addition, the outcomes of our study would be of practical interest to winemakers since they could improve the control of the wood chips extraction process, contributing to a better optimization of the use of wood chips from different species.

Further research, including a more detailed chemical analysis, an extended aging time and different wood chips concentrations, will be necessary to improve our understanding of the potential impact of the use of other non-oak wood chip species on wine quality. Finally, our work also provides valuable inputs for future approval by regulatory institutions (namely the OIV and the European Union) of the use of cherry and acacia wood chips in the winemaking process.

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