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## The Effect of Pruning on the Polyphenols Content and Sensory Characteristics of Bobal Red Wines

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**Abstract** Research on viticultural techniques has gained attention for the impact they have in the composition and quality of grapes and wines. This work evaluates the effect of pruning on the physicochemical and sensory characteristics of Bobal red wines. Four pruning treatments which included winter pruning and green pruning activities were studied. Phenolic content of wines was obtained through analytical procedures and compared between them. Sensory analysis of the wines was made to evaluate the quality of the wines.

The treatments showed different results in physicochemical parameters. The control had better phenolic maturity when compared with the rest of the treatments, and thinning showed to improve maturity parameters. Sensory analyses results showed a preference for wines from smaller berries. Modification of berry and wine composition can be achieved by viticultural practices. A change of pruning system on the Bobal variety to a larger number of buds is possible to a certain extent. Thinning is a good practice to increase maturity levels for Bobal variety.

**Keywords** pruning, polyphenols, sensory analysis, Bobal red wine

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### 1. Introduction

It has long been known that high-quality wines are usually produced from vineyards having low to moderate yields based on variety and cultural practices [1]. Among these practices used to improve quality of the must and wines is the control of vigor through the vine pruning [2]. In viticulture, according to the varietal, place and year, different techniques are adjusted such as pruning to limit the production, which allows improving the characteristics of the fruit and, by consequence the quality of its wines [5]. Pruning is considered as the viticulture practice most decisive over the production and quality of the harvest [4]. It is also considered as one of the practices that tend to improve the organoleptic quality of the musts and of the wines [3].

When having smaller clusters with smaller grape size and greater leaf surface per volume unit, more aromatic wines and with more extract can be produced [5]. A reduction in compactness of the clusters and lighter berries increases the skin mass, providing more phenolic compounds found in the skin [6]. Having lighter berries can also increase the concentration of sugars and other compounds in the berry, helping have a better maturity. More mature grapes have shown to give wines with more polymerized tannins, lower gelatin indexes and more intense aromas [7].

Severe pruning has showed to increase brix, pH, tannins, anthocyanins, phenolic, color density, among other parameters [8], while minimal pruning has in effect shown reduction in color, pH, although sensory parameters show a better expression of fruitiness in wines coming from minimal pruned vines [9].



Another type of pruning is done during the active vegetative period of the vine and is generally known as green pruning or green operations. They contribute, along with the normal pruning to favor the production and quality of the fruit [4]. In most cases, cluster thinning induces faster grape ripening [10]. Furthermore, cluster thinning improves canopy sanitary conditions as it allows more enlightenment and fresh air penetration in the clusters and vegetation [11].

For some authors the enological practices are responsible for the quality of a wine [12], but for others one of the major factors affecting red wine quality is the real degree of phenolic maturity in the grapes at harvest time [13-14].

Relationships between berry composition, wine composition and wine sensory quality or attributes vary depending on the grape variety and the viticultural practices, indicating a high complexity among these relationships [15]. Phenolic compounds or polyphenols are known for being responsible for certain characteristics of wine such as bitterness, astringency, and color intensity of wine, making them play a major role in enology because of their contribution to wine sensory properties as well as to antioxidant activity [16].

Phenolic compounds are mainly located in the skin and seeds of the berries, and they are essential components of red wines. Their structure is complex and their evolution in the wines make their study difficult [17-18]. Most of the sensory attributes of wine such as color, body, and tactile and taste sensations like astringency and bitterness, are directly associated with the composition of anthocyanins and tannins [14, 16, 19]. Overall, polyphenols in wine have also attracted much interest due to their antioxidant properties and their potential health benefits in humans, especially resveratrol [18].

Although extensive research has been made for polyphenols and their contribution to wine quality, at present, the knowledge of the relationship between the quality of a particular wine and its phenolic composition remains one of the major challenges in enology research [20].

Regarding sensory attributes that characterize a wine, bitterness and astringency are two sensory terms of crucial importance for describing the sensory properties of a wine [21]. For instance, organic acids have been reported to impart astringency [22]. Organic acids along with their effect over the pH of the berry can have an influence over the aroma, the anthocyanin extraction and the color stability.

Many studies now involve sensory assessments, performed using different analysis to deliver sensory profiles for visual, olfactory or gustatory attributes, and information on perceived intensity of a given character [23-24]. The aim of this work was study the influence of pruning on the polyphenolic content and sensory characteristics of Bobal red wines.

## Materials and Methods

The study site is a parcel of 0.49 hectares of Bobal variety located at Requena (Valencia, Spain) at 700 m.a.s.l. with latitude 39° 31' and longitude 1° 7'. The type of soil that is found in this parcel is mainly limestone and the rootstock used is 110R.

The pruning was the double cordon royat with four spurs on each branch and 16 buds in total. The height of the vines is of 0.75 m and they are planted at 2850 vines per hectare (1.4 x 2.5 m). Treatments done to the soil include farming and application of 10,000 kilograms of manure per hectare every three years. The vines are also under a drip fed irrigation system. The climate is a Mediterranean-continental type with annual precipitations of 450 mm.

For the objectives of this project lighter pruning treatments were made, increasing the number of buds. In one treatment, one shoot with four extra buds was left during pruning on each branch giving a total of 24 buds, and in another treatment two shoots were left on each of the branches (32 buds). The green pruning activity of thinning was also made in a repetition of the pruning treatments previously described and included as another treatment. Thinning was made at a 30% level. A total of five type of pruning treatments were made and evaluated, including the control, which is the pruning normally made for these variety.

## Experimental Design

Three blocks of 10 rows each were destined for this experiment. The rows were put together into sets of two rows where a type of pruning was done. Each row was composed of 28 to 33 vines. The five different types of pruning treatments were done with two repetitions, one on each block.



The Experimental design on the parcel was:

- 2UT: Two shoots left (24 buds) with no thinning.
- 2T: Two shoots left (24 buds) + thinning.
- 4UT: Four shoots left (32 buds) with no thinning.
- 4T: Four shoots left (32 buds) + thinning.

#### Microvinification

Grapes were destemmed, crushed, and fermented into 25 liter plastic containers. Fermentation and maceration lasted for 10 days. During alcoholic fermentation, the cap was punched down twice a day. The wines were after removed from the skins and seeds. Malolactic fermentation was done in the same plastic containers and when it was over, the wine was corrected to 30 mg/L of SO<sub>2</sub>. The wines were moved into five liter containers and stored at 12°C. The wine was clarified by settling for three months, and after it was racked, bottled into 750 mL glass bottles with cork and stored at 12°C in a temperature-controlled room.

#### Analytical procedures

Acidity, pH, and alcohol degree analysis were done using the OenoFoss™ wine analyzer. The rest of the spectrophotometric procedures were made with a Spectrophotometer UV/VIS JASCO V-530. Color intensity, hue index, total polyphenol index and total anthocyanins following the method proposed by Glories [25]. Pigmented, polymerized and free anthocyanins following the methodology of Boulton [26]. Total polyphenols, total condensed tannins, ethanol index, ionization index, PVP index, gelatin index [27], and DMACH index [28]. A quantitative sensorial analysis was made regarding visual, olfactory and taste characteristics along with a global evaluation. The parameters were evaluated using a 7-point scale, being 1 the minimum value and 7 the maximum.

#### Statistical Analysis

The results obtained for the viticultural parameters, analytical procedures and sensory analysis were statistically analyzed by a variance analysis (ANOVA). The method used to discriminate among the means was Fisher's least significant difference (LSD) procedure. Pearson's product moment correlations were made between the variables to show the strength of the linear relationships between them. The software used was Statgraphics® Centurion XVI.

## Results and Discussion

#### Polyphenolic and standard parameters

According to results, the control wine presents the higher quality characteristics, having less astringent tannins, more stable color, and a higher color intensity when compared to the rest of the wines (Table 1).

**Table 1:** Pruning effect over physicochemical parameters

| Parameter                      | Control      | 2 UT          | 2 T           | 4 UT          | 4 T            |
|--------------------------------|--------------|---------------|---------------|---------------|----------------|
| <i>Standard parameters</i>     |              |               |               |               |                |
| pH                             | 3.56±0.09d*  | 3.18±0.05a    | 3.40±0.11c    | 3.23±0.60a    | 3.31±0.05b     |
| Acidity (g/L)                  | 5.87±0.30a   | 7.16±0.27d    | 6.66±0.15c    | 7.11±0.12d    | 6.47±0.30b     |
| Alcohol degree                 | 11.81±0.24d  | 8.59±0.53a    | 10.52±0.40c   | 9.70±0.11b    | 9.60±0.43b     |
| <i>Polyphenolic parameters</i> |              |               |               |               |                |
| CI                             | 10.35±0.11d  | 7.11±0.12a    | 7.63±0.06b    | 7.63±0.02b    | 8.43±0.02c     |
| Total Anthocyanins (mg/L)      | 325.30±2.54c | 267.99± 6.16a | 296.59±10.35b | 294.27±13.17b | 283.19±10.11ab |
| Copigmented Ant. (%)           | 47.16±2.70b  | 34.93±1.95a   | 50.79±0.89c   | 51.47± 0.93c  | 50.90±0.54c    |
| Free Anthocyanins (%)          | 6.83±0.88a   | 39.65±1.78d   | 17.20±0.55b   | 19.85±1.65bc  | 21.86±3.52c    |
| Polymerized Ant. (%)           | 46.01±2.51c  | 25.42±0.46a   | 32.01±0.34b   | 28.69±1.25ab  | 27.24±3.13a    |



|                         |              |             |              |              |              |
|-------------------------|--------------|-------------|--------------|--------------|--------------|
| Hue Index               | 31.98±0.36c  | 30.29±0.50a | 31.86±0.14c  | 30.84±0.24b  | 30.36±0.20ab |
| TPI                     | 38.53±1.06d  | 26.17±1.57a | 29.90±1.95b  | 30.50±1.36b  | 32.63±1.47c  |
| Total Polyphenols (g/L) | 1.09±0.03d   | 0.77±0.02a  | 0.95±0.03c   | 0.89±0.04b   | 0.87±0.03b   |
| Tannins (g/L)           | 1.46±0.13c   | 1.03±0.07a  | 1.35±0.09bc  | 1.07±0.05ab  | 1.21±0.02b   |
| Ethanol Index           | 92.27±1.90c  | 79.86±1.32a | 89.93±2.65bc | 87.96±2.12b  | 86.93±1.06b  |
| Ionization Index        | 32.14±2.49a  | 45.91±0.88c | 38.73±2.51b  | 41.37±1.36bc | 44.61±1.61c  |
| PVP Index               | 62.97±6.38ab | 57.47±4.56a | 64.51±1.59b  | 57.80±1.29a  | 59.86±1.49ab |
| Gelatin Index           | 54.04±6.98a  | 84.81±5.22c | 59.64±1.51a  | 75.92±4.05b  | 69.16±2.05b  |
| DMACH Index             | 25.62±2.19ab | 27.76±1.17b | 23.54±0.63a  | 22.97±3.72a  | 23.24±1.42a  |

\* Numbers followed by the same letter in the row do not differ significantly at  $p \leq 0.05$

The Gelatin index for the two-shoot thinned treatment presented no significant difference from the control, and it passed from having a lot of astringent tannins to being in the superior limit of convenient tannins [29]. A decrease between the two-shoot thinned and unthinned treatment was observed.

Both pruning and thinning factors were analyzed separately due to interactions seen between them (Table 2). Thinning had larger effects over almost all of the parameters, as seen in the larger F-ratios for thinning when compared to those of pruning.

**Table 2:** Pruning and thinning interactions

| Parameters               | P-value | F-ratio Pruning | F-ratio Thinning |
|--------------------------|---------|-----------------|------------------|
| pH                       | 0.0003* | 3.00            | 168.75           |
| Acidity                  | 0.0346  | 17.63           | 411.36           |
| Alcohol degree           | 0.0000  | 4.20            | 360.93           |
| CI                       | 0.0068  | 279.87          | 277.06           |
| Total Anthocyanins       | 0.0101  | 1.18            | 2.19             |
| Copigmented Anthocyanins | 0.0000  | 143.96          | 121.44           |
| Free Anthocyanins        | 0.0000  | 36.88           | 67.13            |
| Polymerized Anthocyanins | 0.0036  | 0.58            | 6.79             |
| Hue Index                | 0.0002  | 8.36            | 10.78            |
| TPI                      | 0.0049  | 4.48            | 163.11           |
| Total Polyphenols        | 0.0009  | 1.25            | 19.8             |
| Tannins                  | 0.0369  | 1.96            | 38.97            |
| Ethanol Index            | 0.0010  | 5.41            | 17.04            |
| Ionization Index         | 0.0007  | 0.47            | 4.06             |
| PVPP Index               | 0.1229  | 2.23            | 9.91             |
| Gelatin Index            | 0.0020  | 0.02            | 60.95            |
| DMACH Index              | 0.1004  | 4.43            | 2.68             |

\* Significant interactions between pruning and thinning were found at  $p\text{-value} \leq 0.05$

#### Effect of pruning

Table 3 shows the results obtained when comparing the two-shoot and four-shoot pruning for thinned and unthinned treatments.

**Table 3:** Pruning effect over physicochemical parameters

| Parameter                  | Unthinned   |            | Thinned    |            |
|----------------------------|-------------|------------|------------|------------|
|                            | 2 shoots    | 4 shoots   | 2 shoots   | 4 shoots   |
| <i>Standard parameters</i> |             |            |            |            |
| pH                         | 3.18±0.05a* | 3.23±0.60b | 3.40±0.11b | 3.31±0.05a |
| Acidity (g/L)              | 7.16±0.27a  | 7.11±0.12a | 6.66±0.15b | 6.47±0.3a  |



|                                |               |               |              |               |
|--------------------------------|---------------|---------------|--------------|---------------|
| Alcohol degree                 | 8.59±0.53a    | 9.70±0.11b    | 10.52±0.40b  | 9.60±0.43a    |
| <i>Polyphenolic parameters</i> |               |               |              |               |
| IC                             | 7.11±0.12a    | 7.63±0.02b    | 7.63±0.06a   | 8.43±0.02b    |
| Total Anthocyanins (mg/L)      | 267.99± 6.16a | 294.27±13.17b | 296.59±10.3a | 283.19±10.11a |
| Copigmented Ant (%)            | 34.93±1.95a   | 51.47± 0.93b  | 50.79±0.89a  | 50.90±0.54a   |
| Free Ant (%)                   | 39.65±1.78b   | 19.85±1.65a   | 17.20±0.55a  | 21.86±3.52a   |
| Polymerized Ant (%)            | 25.42±0.46a   | 28.69±1.25b   | 32.01±0.34a  | 27.24±3.13a   |
| Hue Index                      | 30.29±0.50a   | 30.84±0.24a   | 31.86±0.14b  | 30.36±0.20a   |
| TPI                            | 26.17±1.57a   | 30.50±1.36b   | 29.90±1.95a  | 32.63±1.47a   |
| Total Polyphenols (g/L)        | 0.77±0.016a   | 0.89±0.043b   | 0.95±0.028b  | 0.87±0.032a   |
| Tannins (g/L)                  | 1.03±0.07a    | 1.07±0.05a    | 1.35±0.09b   | 1.21±0.02a    |
| Ethanol Index                  | 79.86±1.32a   | 87.96±2.12b   | 89.93±2.65a  | 86.93±1.06a   |
| Ionization Index               | 45.91±0.88b   | 41.37±1.36a   | 38.73±2.51a  | 44.61±1.61b   |
| PVPP Index                     | 57.47±4.56a   | 57.80±1.29a   | 64.51±1.59b  | 59.86±1.49a   |
| Gelatin Index                  | 84.81±5.22a   | 75.92±4.05a   | 59.64±1.51a  | 69.16±2.05b   |
| DMACH Index                    | 27.76±1.17a   | 22.97±3.72a   | 23.54±0.63a  | 23.24±1.42a   |

Numbers followed by the same letter in the row do not differ significantly at  $p \leq 0.05$

Acidity results were consistent with those of Archer and Schalkwyk [9] and Walteros et al. [3] who reported a decrease in acidity with larger pruning (Table 4). The result was only significant for the thinned wines.

For the unthinned treatments, the larger pruning treatment was significantly higher in alcohol degree, being consistent with Walteros et al. [3]. The opposite results in the thinned wines might be because of a thinning effect. Considering only the unthinned treatments, it can be said that pruning had no significant effect on pH or on total acidity.

Color intensity increased in the larger pruning treatments. Total anthocyanins, TPI, and total polyphenols showed a significant increase in the four-shoot unthinned treatment. No significant differences were found in these parameters for thinned treatments.

Different studies have shown opposite results regarding pruning levels and polyphenolic concentration. Some show that as vigor increases TPI and anthocyanins decrease [30]. Ortega-Farias et al. [2] concluded that, for Cabernet Sauvignon, by pruning more severely the anthocyanin content was increased and the total polyphenols showed no significant differences. The difference in the content of anthocyanins, polyphenols and tannins between the unthinned treatments could be ascribed to berry size, since larger pruning resulted in having smaller berries. Holt et al. [31] found that anthocyanin, total polyphenols and tannin levels increased for their pruning treatment with more buds and concluded that it was due to a decrease in berry size. Anthocyanin concentration has been positively related to total skin surface per kilogram of grape, resulting in the smaller berries being characterized by the highest quantities of these components [32].

Berry sizes were very similar for the thinned treatments reflecting no significant differences in TPI and anthocyanins. Differences in polyphenolic content and composition have generally been associated with berry size, although studies have shown that composition can be influenced more by the treatment and by all factors that influence the composition and berry size than by berry size per se [33]. This argument might be the reason why tannins and total polyphenols were significantly higher in the two-shoot thinned wines when compared with the four-shoot thinned wines, and the reason why values are inconsistent between thinned and unthinned treatments.

Significant differences were found for the Ethanol (EI) and Ionization indexes (II) on the unthinned treatments. A larger pruning showed higher tannins polymerized with polysaccharides (EI), and less anthocyanins contributing to color (II). Better quality parameters were obtained for the larger pruning treatment, which showed lower Gelatin index and DMACH although no significant difference was found.



The results for the thinned treatments were inconsistent with those of the unthinned treatments. Ionization and Gelatin indexes resulted higher in the four-shoot treatment and PVP was significantly higher for the two-shoot treatment. So, for the thinned treatments, the more severe pruning had more stable color and less astringent tannins. Finding more significant differences in these indexes in the thinned treatments than in the unthinned indicates that other factors besides the number of buds left at winter pruning have significant effects over the phenolic quality of the berries, reflected in the wines.

These results for the pruning effect on the wines partly confirm the second hypothesis that states that different pruning levels will cause differences in the polyphenolic parameters of wines. In the unthinned treatments, no significant differences were found in the quality indexes suggesting that a larger pruning increases the polyphenolic content but not its quality.

#### Effect of thinning

Results showed that, for the more severe pruning, thinning presented significantly higher pH and alcohol levels, and lower acidity (Table 4). For the larger pruning treatments, only acidity decreased significantly in the thinned wines.

**Table 4:** Thinning effect over physicochemical parameters in thinned and unthinned wines

| Parameter                      | 2-shoot       |               | 4-shoot       |               |
|--------------------------------|---------------|---------------|---------------|---------------|
|                                | Unthinned     | Thinned       | Unthinned     | Thinned       |
| <i>Standard parameters</i>     |               |               |               |               |
| pH                             | 3.18±0.05a    | 3.40±0.11b    | 3.23±0.60a    | 3.31±0.05a    |
| Acidity (g/L tart. ac.)        | 7.16±0.27b    | 6.66±0.15a    | 7.11±0.12b    | 6.47±0.30a    |
| Alcohol degree                 | 8.59±0.53a    | 10.52±0.40b   | 9.70±0.11a    | 9.60±0.43a    |
| <i>Polyphenolic parameters</i> |               |               |               |               |
| CI                             | 7.11±0.12a    | 7.63±0.06b    | 7.63±0.02a    | 8.43±0.02b    |
| Total Anthocyanins (mg/L)      | 267.99± 6.16a | 296.59±10.35b | 294.27±13.17a | 283.19±10.11a |
| Copigmented Ant (%)            | 34.93±1.95a   | 50.79±0.89b   | 51.47± 0.93a  | 50.90±0.54a   |
| Free Ant (%)                   | 39.65±1.78b   | 17.20±0.55a   | 19.85±1.65a   | 21.86±3.52a   |
| Polymerized Anth (%)           | 25.42±0.46a   | 32.01±0.34b   | 28.69±1.25a   | 27.24±3.13a   |
| Hue Index                      | 30.29±0.50a   | 31.86±0.14b   | 30.84±0.24b   | 30.36±0.20a   |
| TPI                            | 26.17±1.57a   | 29.90±1.95b   | 30.50±1.36a   | 32.63±1.47b   |
| Total Polyphenols (g/L)        | 0.77±0.016a   | 0.95±0.028b   | 0.89±0.043a   | 0.87±0.032a   |
| Tannins (g/L)                  | 1.03±0.07a    | 1.35±0.09b    | 1.07±0.05a    | 1.21±0.02a    |
| Ethanol Index                  | 79.86±1.32a   | 89.93±2.65b   | 87.96±2.12a   | 86.93±1.06a   |
| Ionization Index               | 45.91±0.88a   | 38.73±2.51b   | 41.37±1.36a   | 44.61±1.61a   |
| PVPP Index                     | 57.47±4.56a   | 64.51±1.59a   | 57.80±1.29a   | 59.86±1.49a   |
| Gelatin Index                  | 84.81±5.22b   | 59.64±1.51a   | 75.92±4.05a   | 69.16±2.05a   |
| DMACH Index                    | 27.76±1.17b   | 23.54±0.63a   | 22.97±3.72a   | 23.24±1.42a   |

\* Numbers followed by the same letter in the row do not differ significantly at  $p \leq 0.05$ .

The differences in the two-shoot treatment are consistent with results found in other grapes like Grenache that went under cluster thinning treatments increasing their pH and alcohol levels, and lowering their acidity [10].

The effect of thinning in the polyphenolic parameters was significant for the two-shoot treatment. Color intensity (CI), TPI, total polyphenols, tannins, and total anthocyanins values increased with thinning. More polymerized and less free anthocyanins were also found in the thinned wines. Regarding the four-shoot treatment, thinning showed significantly higher values only for CI and TPI.



Results regarding color intensity are consistent with the results of previous studies [34] where wines made from cluster-thinned vines presented significant better chromatic characteristics when compared to the non-thinned control. Both the two-shoot and four-shoot treatments presented higher TPI and CI values for the thinned wines, which confirm the results obtained by Avizcuri-Inac et al. [10] for Tempranillo and Grenache wines. The increase in TPI and CI could be ascribed to a higher total leaf area/fruit ratio, which usually present higher anthocyanin and phenol values and is achieved by thinning [35].

Thinning also increased tannins in bot pruning levels. Fanzzone et al. [36] found that thinning can increase the content of flavanols in skins and seeds of the grape, which can be later reflected in the wine. Also, there is a better exposure of the clusters to light, which may cause polyphenolic levels such as those of tannins to increase [37].

Significant differences in quality indexes were found for the two-shoot treatment. The Ethanol Index was significantly higher for the thinned treatment and the gelatin index and DMACH test were significantly lower, indicating less astringency and more polymerized tannins.

The absence of significant differences in the four-shoot treatment might be due to poor fruit set that was caused by an excess in clusters or production, which caused that even with thinning, the polyphenols could not be better concentrated, and a better quality between the thinned and unthinned treatments could not be achieved.

It must also be considered that there might be a vintage effect in green pruning activities. Some authors have confirmed better results with green pruning activities in years less favorable for ripening or in rainy and cool seasons [6, 38-39]. Cluster thinning has also been found to be eventually void by the plant, and after a few years its effects are no longer visible [11]. Repetitions of this study are essential to confirm the reasons of the results obtained.

#### Anthocyanidin content

HPLC resulted in similar anthocyanidin concentrations for almost every treatment, with no significant differences in the total anthocyanidin content between treatments (Table 5).

**Table 5:** Effect of pruning on anthocyanidin content in wines

| Anthocyanidin        | Control     | 2 shoots UT | 2 shoots T  | 4 shoots UT | 4 shoots T  |
|----------------------|-------------|-------------|-------------|-------------|-------------|
| Delfinidin (mg/L)    | nd          | Nd          | Nd          | nd          | nd          |
| Cyanidin (mg/L)      | 2.21±0.18a* | 2.66±0.09a  | 2.67±0.23a  | 2.62±0.11a  | 2.59±0.05a  |
| Petunidin (mg/L)     | 3.17±0.07ab | 3.31±0.05b  | 3.34±0.10b  | 3.35±0.06b  | 3.05±0.12a  |
| Peonidin (mg/L)      | 1.92±0.30a  | 2.43±0.18a  | 2.68±0.33a  | 2.68±0.13a  | 2.48±0.28a  |
| Malvidin (mg/L)      | 19.33±0.20a | 20.75±0.15a | 20.26±0.27a | 20.26±0.19a | 19.69±0.09a |
| Total content (mg/L) | 26.63±0.14a | 29.15±0.11a | 28.96±0.25a | 28.92±0.67a | 27.80±0.16a |

\*Numbers followed by the same letter in the row do not differ significantly at  $p \leq 0.05$ . nd: not detected

Total content for all the treatments was similar to values found in other studies for varieties like Cabernet sauvignon which were of 26.1 mg/L [40].

#### Sensory analysis

The sensory analysis showed very few differences between treatments (Table 6).

**Table 6:** Influence of different treatments on sensory attributes of wines

| Parameters      | Control     | 2 shoots UT | 2 shoots T  | 4 shoots UT | 4 shoots T  |
|-----------------|-------------|-------------|-------------|-------------|-------------|
| Color           | 6.17±0.06b* | 5.50±0.94a  | 5.83±0.83ab | 6.00±0.62ab | 5.75±0.61ab |
| Aroma Intensity | 5.75±1.15b  | 4.83±0.89a  | 5.33±1.42ab | 4.67±0.85a  | 5.00±0.95ab |
| Aroma Quality   | 3.5±0.84a   | 3.50±0.76a  | 3.75±1.02a  | 4.17±0.92a  | 3.67±0.74a  |
| Tannin Quality  | 4.25±0.77a  | 4.17±0.73a  | 3.75±0.76a  | 4.33±1.03a  | 3.92±0.77a  |



|                   |            |            |             |            |             |
|-------------------|------------|------------|-------------|------------|-------------|
| Structure         | 4.17±1.11a | 4.08±0.83a | 4.08±1.03a  | 4.42±0.91a | 4.42±0.89a  |
| Equilibrium       | 3.67±0.65a | 3.67±0.51a | 3.75±0.76ab | 4.25±0.77b | 4.00±0.97ab |
| Global Evaluation | 3.67±0.51a | 3.67±0.50a | 3.58±0.65a  | 4.33±0.95b | 3.92±0.83ab |

\* Numbers followed by the same letter in the row do not differ significantly at  $p \leq 0.05$ .

The control treatment had significantly higher color quality when compared to the two-shoot treatment. This is consistent with the CI and anthocyanin levels found during the polyphenolic analyses, where the control showed the highest values, and the two-shoot treatment the lowest.

Aromatic intensity was significantly higher for the control when compared with the non-thinned treatments. According to Di Profio et al. [41], viticulture treatments such as cluster thinning enhance intensities of several aroma characteristics in wines.

The tannin quality showed no significant differences between treatments, even though gelatin indexes showed the opposite. A better equilibrium was found in the four-shoot wine, and showed significant differences with the two-shoot and control pruning wines.

Finally, the wine that was better evaluated was the four-shoot wine, followed by the four-shoot thinned wine. This result shows that lower gelatin indexes, or less astringent tannins, did not represent a perception of better quality in the wines, as was expected.

The differences found in the sensory analysis are inconsistent with the differences found in the polyphenolic analyses. There may be a variety of influences affecting the overall quality score of the wines, but the judges agreed in giving the highest global scores to the four shoot treatments. This result confirms the widely accepted belief that smaller berries produce better wine [42]. A graphic representation of the results is shown in Figure 1.

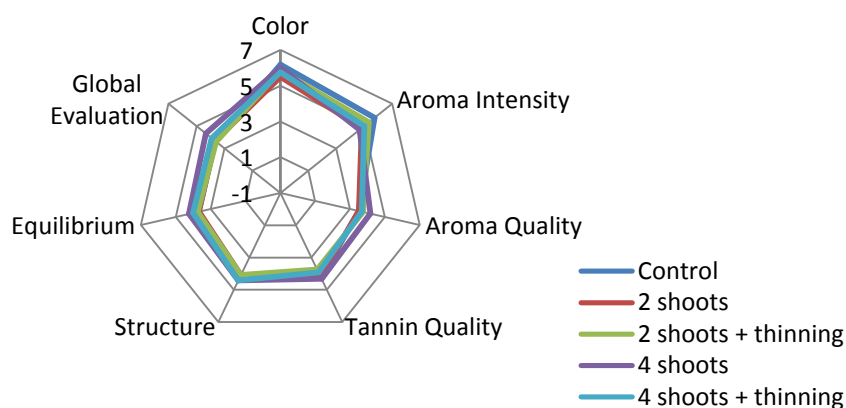


Figure 1: Radar diagram of sensory analysis for different pruning treatments

### Correlations between parameters

The parameters measured in this study have to be correlated in order to know if pruning had an effect in the physicochemical and sensory parameters of the wine. Improving quality with the actual characteristics of the Bobal variety planted suggested leaving a larger pruning to modify its viticultural parameters (vigor, vine balance, berry size), which could help improve polyphenolic and sensorial parameters.

Sensory results did not help very much to differentiate between wines, and this tendency was also reflected when making the correlations. When comparing the standard and polyphenolic parameters with those of the sensory analysis, no significant correlation was found. However, it can be said that higher tannin quality was correlated with lower gelatin indexes and DMACH indexes, although not significantly. Aroma intensity showed significant positive correlations with pH and alcohol degree, and a negative correlation with acidity. Global evaluation of





the wines was positively correlated only with the equilibrium parameter, which was positively correlated to aroma quality and structure.

## 5. Conclusion

In this study polyphenolic composition of the wines was achieved through viticultural practices. Different pruning levels modified basic analyses and polyphenolic analyses, although thinning showed to have a larger effect in the modification of these parameters. It helped achieve better maturity indexes in the wine. The control showed to have better maturity parameters, followed by the two-shoot thinned treatment. The four-shoot treatments resulted in being over cropped and showing symptoms of acrotony and inhibition.

Results observed in the physicochemical analyses suggest that a change of pruning to an increased number of buds to 24 could give favorable results in quality parameters, as long as thinning is included. An increase to the largest number of buds to 32 is not favorable for the Bobal variety in this vineyard.

Sensory analysis did not show significant differences between treatments, although the treatments with smaller berries resulted in better global evaluations, confirming the common belief that smaller berries give better quality wines. Polyphenolic content relationships with sensory analysis continue to be complex and difficult to explain.

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