



Effect of Leaf Removal and Shoot Topping on the Aromatic Composition of White Wines

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Abstract The aim of this study was to evaluate the effect of canopy treatments on the aromatic composition and sensory qualities of Muscatel wines produced in the Valencia Region (Spain). The evaluation was conducted during 2008 and 2009 in an experimental field on vines in a vertical trellis system. The canopy treatments were made in the form of leaf removal and shoot topping using three different rootstocks (Couderc 3309, Paulsen 1103 and Ruggeri 140). The vine canopies of the control plants were given no treatment. Ripening was monitored by evaluating the content of soluble solids, total acidity and pH from veraison to harvest. 50 Kg grape samples were microvinified, after which an evaluation was made of the aromatic composition and sensory attributes of the wines by gas chromatography. A sensory analysis was performed using a panel of wine expert tasters.

The results of sugar accumulation in the grapes showed that leaf removal was the best canopy treatment in every plant rootstock. Lower yield was observed with both leaf removal and shoot topping. The highest concentration of aromatic compounds (acetates, alcohols, ethyl esters, fatty acids and monoterpenes) was found in wines made from the vines on Couderc 3309 rootstock who underwent to leaf removal and shoot topping. The wines from Couderc 3309 also obtained the highest scores in the sensory analysis. The wines from vines with leaf removal were the highest rated, while those from plants whose shoots had been topped were the lowest rated, regardless of the rootstock used.

Keywords leaf removal, shoot topping, aromatic composition, rootstocks, Muscatel grapes

1. Introduction

Canopy management is designed to improve the distribution of shoots and leaves so as to encourage the uptake of solar radiation, decrease the amount of shade within the canopy and prevent the formation of a microclimate favorable to disease development. A balance between leaf area and yield benefits plant growth and grape composition [1].

There are numerous studies on the response of fruit set and other yield components to different treatments applied before and during the flowering period [2,3]. However, few studies have been published concerning the aromatic qualities of wine, which is one of its most important characteristics and is the result of a great number of substances affected by numerous factors related to viticultural and enological aspects [4-6].

Besides climate and soil characteristics, the canopy management of the grapevines is important for the viticultural aspects [7]. Removing leaves from the bunch zone can markedly improve fruit exposure, benefits fruit composition and reduces propensity to disease, especially to botrytis bunch rot [8]. Leaf removal was found



to reduce whole-vine photosynthesis more than shoot topping. However, both treatments significantly reduced whole-vine photosynthesis immediately after they were carried out [9-10].

Leaf removal of very dense canopies at veraison increased sugars, flavors, total and phenol-free glycosides and flavonoids and decreased acidity and gray mold attacks, as compared with untreated vines [11-15]. Shoot topping delayed crop maturity in vigorous vines from California but did not affect grape composition or performance and decreased the incidence of *Botrytis cinerea* rot [16-18].

The volatile content of varieties such as Muscat, Chardonnay, Riesling, Gewürztraminer, etc., have been widely investigated in recent decades [19]. Numerous studies have also been carried out in Spain on the volatile composition of grapes [20] and wines produced from varieties traditionally cultivated in this country [21,22], or in specific regions, such as Penedés [23], Valencia [24], La Rioja [25], Granada [26], Mallorca [27] or the Canary Islands [28].

There are several studies on the influence of rootstocks on the aromatic profile, productivity, composition, phenolic compounds and quality [29-33].

Muscatel is the most famous white grape variety typical of Valencia region in south-east of Spain, which produces well-known sweet and liqueur wines. These grapes are also used for making dry, fresh, light, fruity and highly aromatic wines. A well-structured Muscatel depends not only on terpenic compounds but also on the aromatic compounds formed during fermentation. However, the importance of attending to the quality of the grapes, and therefore of the wine produced, is an important aspect often overlooked when discussing the quality of Muscatel wine [34].

The present study deals with the volatile composition of monovarietal wines made with Muscatel grapes grafted on different rootstocks and subjected to leaf removal and shoot topping of the grapevines. The aim of the study was to evaluate the effects of canopy treatments on the aromatic composition of wines made from Muscatel grapes.

2. Materials and Methods

Field trials and experimental design

A two-year-trial (2008-2009) was carried out in an experimental field belonging to the Baronía de Turis cooperative winery (Valencia, Spain) located at 240 m asl. The viticultural area belongs to the Muscatel subzone of the Denomination of Origin Valencia (Spain). Muscatel is the most commonly cultivated *Vitis vinifera* L. variety in this region. The plants were about 22 years old and grown in a vertical trellis system. The soil was a fertile clay-calcareous flysch. The annual rainfall was 530 mm. The canopy treatments consisted of leaf removal and shoot topping using three different rootstocks: Couderc 3309, Paulsen 1103 and Ruggeri 140. Complete manual leaf removal around the cluster zone was performed at veraison; the percentage of canopy surface area removed was therefore 20%. Shoot topping was carried out at 30 cm from the top of the shoot. The control vines were left untreated. Three blocks of 10 plants each were considered in each rootstock and treatment, and eight plants per block were sampled at random, resulting in 24 plants per treatment. The vines did not show any symptoms of disease throughout the growing season or during harvest.

At harvest 24 vines were chosen per treatment. The complete clusters of each plant were counted and weighed. The following yield and fruit parameters were recorded: grape yield (Kg/plant), mean cluster weight (g), juice total soluble solids (°Bé), juice titratable acidity (g/L) by titration with NaOH 0.1N, juice pH by a pH meter. Ripening was monitored by evaluating the content of the soluble solids, total acidity and pH from veraison to harvest (ripening time). Figure 1 shows the experimental design. The whole experiment was performed in triplicate.



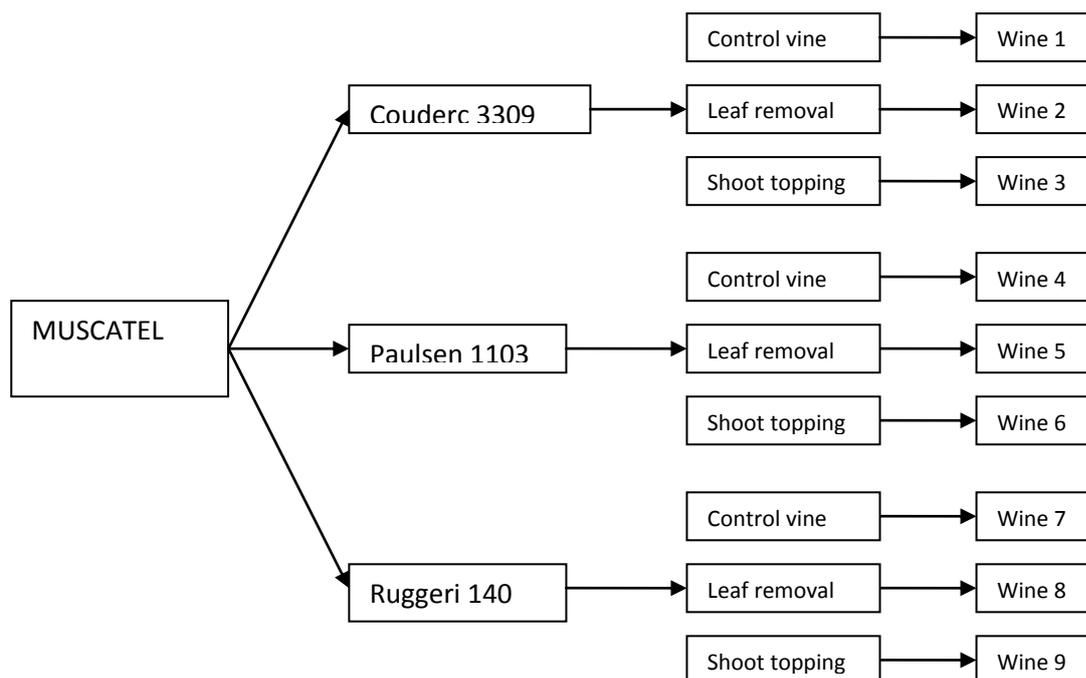


Figure 1: Experimental Design

Winemaking procedure

The grapes were hand-harvested in plastic crates from vines grafted on different rootstocks. In the vinification process the grapes were pressed in a hydraulic press and the must was separated into nine different tanks each containing 50 L by volume adding 5 g/hL of sulfur dioxide to protect against oxidation.

The must was treated with fining agents immediately after pressing with bentonite at 15 g/hL and gelatine at 4 g/hL. The fining agents reacted with the compounds in the must for 48 hours in a cold chamber at a constant temperature of 7 °C. After 48 h the “débouillage” process was carried out and fermentation took place at 18°C using the traditional winemaking method of the inoculation of commercial yeast (*Saccharomyces cerevisiae*) at a concentration of 25 g/hL.

After two weeks of fermentation the wine was racked and preserved by adding 30 mg/L of sulfur dioxide. Before bottling it was stored in a cold room at 5°C for one month, after which chemical analyses were carried out on the samples.

Analytical methods

Enological parameters such as alcohol (% vol), volatile acidity, total acidity, pH, sugars, and tartaric, malic and citric acids were determined according to official EU methods [35].

Volatile compounds were quantified by chromatography with a HP-5890 (Hewlett Packard Corp., USA) chromatograph equipped with a flame ionization detector (FID) using nitrogen as a carrier gas. Isobutyl and isoamyl alcohols, ethyl and methyl acetates, methanol and 1-propanol were determined by the direct injection of 1 µL of wine containing 4-methyl-2-pentanol as an internal standard, in a Carbowax 1500 capillary column (length 4 m, i.d. 0.32 cm) over Chromosorb to 15%, with 80–100 meshes [36]. Minor wine components were determined by making a prior extraction. 2-phenylethanol, isoamyl acetate, isobutyl acetate, ethyl butyrate, ethyl lactate, ethyl octanoate, diethylsuccinate, and n-amyl alcohol were extracted using organic solvents (diethyl ether and n-pentane 2:1). As an internal standard 1 mL of 2-octanol was added to 100 mL of wine. The extraction procedure was optimized by means of ultrasound. The combined extracts were dried with anhydrous sodium sulphate, reduced in volume to 20 µL in a vacuum rotary evaporator, and then evaporated with a gentle stream of nitrogen. One microlitre of extract was injected in a HP-INNOWax (crosslinked polyethylene glycol) capillary column 60 m long with 0.25 mm internal diameter. Compound quantification was based on the internal



standard method. The efficacy of the method was verified from the analysis performed on standard solutions of the components, with the aid of an HP-5979 mass spectrophotometer linked to the chromatograph. The variance of the method was determined by the analysis of three replicates of each sample.

Fatty acids and their ethyl esters (ethylhexanoate, ethyldecanoate, ethylpropionate and ethyllaurate), acetates (hexylacetate and phenylacetate), and 1-hexanol were extracted and quantified by the Bertrand method (1993). Two milliliters of 3-octanol (50 mg L⁻¹) as internal standard and 2 mL of sulphuric acid (1/3) were added to 50 mL of wine. This was extracted three times (4, 2 and 2 mL) with diethyl ether-hexane (1:1, v/v). The organic extract (1 µL) was injected into a Varian 3400 chromatograph under the same capillary column and chromatographic conditions, indicated for the monoterpenes.

Monoterpene alcohols were extracted as follows: a sample of 100 mL wine was adjusted to pH seven by the addition of NaOH, and 1 mL of 3-octanol (10 mg L⁻¹) was added as an internal standard. The sample was extracted three times (10, 5 and 5 mL) with diethyl ether-pentane (1:1, v/v). The organic extract was concentrated to 0.5 mL under nitrogen. A Hewlett-Packard HP 5890 gas chromatograph, with flame ionisation detector and equipped with a Carbowax-20M capillary column was used. A 1 mL sample of the extract was injected in splitless mode (30 s). Temperature program: held 1 min at 45 °C, raised at 3 °C min⁻¹ to 230 °C, and held for 25 min. Hydrogen was used as carrier gas (18 psi). Temperature for both the injector and detector was 230 °C.

Sensory analysis

A sensory analysis was performed in a standardized wine tasting room using a panel of nine expert tasters. The sensory profile was determined using 6 descriptors (color, aroma intensity, aroma quality, taste intensity, taste quality and a final assessment of each wine). The judges were asked to evaluate each descriptor on a 0-10 point scale, in which zero (0) indicated very bad while a score of ten (10) was excellent.

Statistical analysis

All data were statistically analyzed using Statgraphic Plus 5.1. ANOVA statistical methods were employed. The statistical significance of each factor under consideration was calculated at $\alpha = 0.05$ using the Student's t-test.

3. Results and Discussion

Yield components and fruit composition

Table 1 shows the grape parameters recorded. The weight of berries and in general the yield of the vine plant are high in the control vines without any treatment, which shows that leaf removal and shoot topping reduce yield [12]. The grapevines from the three rootstocks produced the same quantity of grapes.

Table 1: Mean values of weight of berries, bunch and vine plant

	Berries		Bunch of grapes		Vine plant	
	Weight (g)	Diameter (cm)	Weight (g)	Nº Berries /Bunch	Weight (kg)	nº Bunch/Vine
Wine 1	6.53 ± 0.39	2.03 ± 0.12	283.4 ± 14.35	43.30 ± 2.26	4.75 ± 0.21	16.80 ± 1.55
Wine 2	5.39 ± 0.43	1.95 ± 0.10	233.4 ± 15.56	34.30 ± 1.85	2.64 ± 0.21	14.30 ± 1.19
Wine 3	5.47 ± 0.41	2.00 ± 0.11	249.1 ± 15.37	36.30 ± 1.98	2.77 ± 0.11	13.70 ± 0.70
Wine 4	6.85 ± 0.25	2.00 ± 0.12	289.0 ± 15.91	43.30 ± 1.91	4.76 ± 0.24	16.05 ± 1.07
Wine 5	5.23 ± 0.35	2.08 ± 0.13	194.8 ± 13.93	33.00 ± 2.40	2.74 ± 0.17	15.85 ± 1.23
Wine 6	5.25 ± 0.41	2.05 ± 0.12	259.2 ± 19.55	39.30 ± 2.91	2.75 ± 0.17	13.30 ± 0.57
Wine 7	7.45 ± 0.49	1.95 ± 0.12	311.5 ± 12.66	40.00 ± 1.84	4.56 ± 0.24	15.30 ± 1.12
Wine 8	6.90 ± 0.49	2.00 ± 0.14	241.5 ± 14.92	35.00 ± 1.91	3.04 ± 0.17	14.70 ± 1.16
Wine 9	5.49 ± 0.41	1.99 ± 0.12	225.0 ± 16.69	31.00 ± 2.77	2.50 ± 0.14	14.70 ± 1.26

Standard wine parameters

The mean values of the standard wine parameters are shown in Table 2. In general the wines from grapevines with leaf removal presented the highest alcohol content regardless of the rootstock used. These wines had the lowest total acidity and lowest concentration of tartaric, malic and citric acids, as well the lowest pH values [18].



Significant differences were found in the alcohol content between different rootstocks, but only in total acidity and malic acid for the 3309 rootstock, and tartaric and citric acids for the 140 rootstock. Concerning the canopy treatments, only the plants with leaf removal showed significant differences in alcohol content, total acidity and tartaric, malic and citric acids (Table 3).

Table 2: Mean values of some standard wine parameters

Parameters	Wine 1	Wine 2	Wine 3	Wine 4	Wine 5	Wine 6	Wine 7	Wine 8	Wine 9
Alc (%vol.)	13.7 ± 1.21	14.7 ± 1.41	12.7 ± 1.01	13.40 ± 1.10	13.65 ± 1.11	12.30 ± 1.01	13.4 ± 1.15	14.65 ± 1.29	12.6 ± 1.09
Volatile acidity (g/L acetic acid)	0.14 ± 0.01	0.14 ± 0.01	0.13 ± 0.01	0.16 ± 0.01	0.15 ± 0.01	0.16 ± 0.01	0.14 ± 0.01	0.15 ± 0.01	0.17 ± 0.01
Total acidity (g/L tartaric acid)	4.75 ± 0.31	4.35 ± 0.21	4.43 ± 0.21	4.90 ± 0.21	4.700 ± 0.28	4.75 ± 0.29	4.75 ± 0.20	4.65 ± 0.19	4.70 ± 0.29
pH	3.36 ± 0.33	3.38 ± 0.13	3.38 ± 0.22	3.32 ± 0.17	3.38 ± 0.26	3.41 ± 0.26	3.38 ± 0.21	3.42 ± 0.13	3.41 ± 0.23
Sugars (g/L)	1.55 ± 0.08	1.45 ± 0.07	1.65 ± 0.14	1.70 ± 0.21	1.55 ± 0.14	1.75 ± 0.12	1.70 ± 0.11	1.50 ± 0.14	1.70 ± 0.12
Tartaric acid (g/L)	2.30 ± 0.13	2.10 ± 0.13	2.15 ± 0.20	2.45 ± 0.20	2.10 ± 0.17	2.17 ± 0.17	2.45 ± 0.21	2.00 ± 0.16	2.10 ± 0.16
Malic acid (g/L)	2.00 ± 0.11	1.45 ± 0.11	1.85 ± 0.15	1.90 ± 0.09	1.80 ± 0.08	1.85 ± 0.08	1.85 ± 0.10	1.80 ± 0.15	2.00 ± 0.15
Citric acid (g/L)	0.43 ± 0.03	0.39 ± 0.02	0.44 ± 0.03	0.41 ± 0.03	0.40 ± 0.03	0.41 ± 0.03	0.41 ± 0.02	0.38 ± 0.03	0.43 ± 0.03

The influence of canopy treatments on the standard parameters of the wines (Table 3) shows that wines from vines from leaf removal had a higher alcohol content, since the grapes get more sunlight and have lower contents of tartaric, malic and citric acids as well as reduced total acidity [37].

Table 3: Influence of rootstocks and canopy treatments on some standard wine parameters.

Parameters	Rootstocks			Canopy treatments		
	3309	1103	140	Control	Leaf removal	Topping
Alc (%vol.)	*	*	*	ns	*	ns
Volatile acidity (g/L acetic acid)	ns	ns	ns	ns	ns	ns
Total acidity (g/L tartaric acid)	*	ns	ns	ns	*	*
pH	ns	ns	ns	ns	ns	ns
Sugars (g/L)	ns	ns	ns	ns	ns	ns
Tartaric acid (g/L)	ns	ns	*	ns	*	ns
Malic acid (g/L)	*	ns	ns	ns	*	ns
Citric acid (g/L)	ns	ns	*	ns	*	ns

In the same row for each rootstocks and canopy treatments: * indicates significant differences at 0.05

Volatile compounds

Table 4 shows the mean values of wine volatile compounds. The wines made with Muscatel grapes grafted onto 140 rootstock had the highest values of alcohols, ethyl esters, fatty acids and monoterpenes and the lowest values of acetates. The wines from 3309 rootstock had the lowest values of alcohols, ethyl esters, fatty acids and monoterpenes, but the highest values of acetates. The wines from 1103 rootstock had values midway between

the other two rootstocks studied in the experience. The trend for the total compounds measured was the same and wines with a high concentration of volatile compounds were obtained from the 140 rootstock.

As regards the influence of the rootstocks and canopy treatments on the wines' volatile compounds, significant differences were found between the concentrations of certain volatile compounds (Table 5). For the 3309 and 1103 rootstocks, significant effects were observed in all the volatile compounds, but not in the 140 rootstock, in which only concentrations of acetates, alcohols and monoterpenes had significant effects. For acetates, significant differences were found between the wines from control vines and those made from vines subjected to leaf removal and topping treatments on 1103 and 140 rootstocks, with the highest values found in wines made from the control vines.

The total amount of all volatile compounds also showed significant differences within the three rootstocks used in the experience, but these were found between wines from the control vines and those from the vines subjected to leaf removal and topping, but not between the wines from the leaf removal and topping treatments.

With respect to canopy treatments, the control, leaf removal and topping effects varies for each group of volatile compounds according to the different rootstocks used. Significant differences were observed between the wines from the 3309 and 1103 rootstocks and those from the 140 rootstock. For alcohols, significant differences were observed between wine from the 3309 rootstock and those from 1103 and 140 rootstocks, but not between the two latter wines.

Significant differences were found between the wines from vines subjected to leaf removal for acetates, fatty acids and monoterpenes but not for alcohols and ethyl esters. Significant differences for the two latter compounds were found between wines from 3309 and those from 1103 and 140 rootstocks, but were not found between the wines from the two latter rootstocks.

Topping treatment was found to have significant differences between the wines for fatty acids and monoterpenes, but not for alcohols and ethyl esters, which was found to have significant differences between the wines from 3309 rootstock and those from 1103 and 140 rootstocks. Leaf removal did not show significant differences between the wines from the two latter rootstocks.

Table 4: Means values (mg L⁻¹) of the volatile compounds of wines

Parameters	Wine 1	Wine 2	Wine 3	Wine 4	Wine 5	Wine 6	Wine 7	Wine 8	Wine 9
ACETATES									
Ethylacetate	0.127 ± 0.02	0.122 ± 0.01	0.141 ± 0.02	0.110 ± 0.01	0.132 ± 0.02	0.101 ± 0.01	0.096 ± 0.01	0.099 ± 0.01	0.0125 ± 0.02
Isoamylacetate	0.624 ± 0.05	0.724 ± 0.07	0.398 ± 0.04	0.820 ± 0.09	0.578 ± 0.07	0.588 ± 0.06	0.516 ± 0.07	0.716 ± 0.06	0.825 ± 0.07
Hexylacetate	0.532 ± 0.03	0.878 ± 0.07	0.987 ± 0.08	0.657 ± 0.05	0.665 ± 0.04	0.867 ± 0.05	0.733 ± 0.05	0.435 ± 0.03	0.525 ± 0.04
Methylacetate	6.18 ± 0.39	5.98 ± 0.56	6.12 ± 0.60	5.98 ± 0.41	5.45 ± 0.55	5.50 ± 0.55	4.89 ± 0.51	4.43 ± 0.43	4.34 ± 0.39
Phenylacetate	0.526 ± 0.03	0.730 ± 0.07	0.865 ± 0.04	0.590 ± 0.06	0.468 ± 0.05	0.427 ± 0.03	0.425 ± 0.02	0.449 ± 0.05	0.626 ± 0.05
Isobuthylacetate	0.103 ± 0.01	0.114 ± 0.01	0.147 ± 0.02	0.121 ± 0.01	0.119 ± 0.01	0.154 ± 0.02	0.111 ± 0.01	0.122 ± 0.01	0.144 ± 0.02
Σ Acetates	8.092 ± 0.71	8.548 ± 0.56	8.658 ± 0.44	8.278 ± 0.56	7.412 ± 0.56	7.637 ± 0.56	6.771 ± 0.34	6.251 ± 0.55	6.473 ± 0.63
ALCOHOLS									
Methanol	26.8 ± 1.22	19.9 ± 1.06	23.4 ± 1.56	30.5 ± 1.34	24.6 ± 1.88	31.1 ± 1.44	29.8 ± 1.28	32.3 ± 1.87	27.9 ± 1.36
1-propanol	34.8 ± 2.06	36.90 ± 2.66	47.43 ± 3.01	40.88 ± 2.45	43.67 ± 2.78	41.67 ± 2.89	44.78 ± 2.88	45.68 ± 2.05	47.89 ± 2.45
Isoamilic alcohols	105.8 ± 5.23	113.1 ± 5.12	109.0 ± 3.77	97.89 ± 4.22	107.67 ± 6.01	106.1 ± 5.18	100.5 ± 3.25	109.9 ± 4.28	112.34 ± 4.89
Isobutanol	34.56 ± 1.27	39.67 ± 2.01	38.54 ± 1.66	39.89 ± 1.78	42.09 ± 1.50	43.06 ± 1.22	32.66 ± 1.90	38.77 ± 2.11	37.89 ± 1.90
n-amylalcohol	0.120 ± 0.02	0.098 ± 0.02	0.088 ± 0.02	0.109 ± 0.02	0.106 ± 0.03	0.105 ± 0.02	0.093 ± 0.01	0.095 ± 0.01	0.094 ± 0.01
1-hexanol	2.66 ± 0.02	2.89 ± 0.02	2.65 ± 0.02	2.90 ± 0.02	3.21 ± 0.03	3.24 ± 0.23	3.22 ± 0.22	3.69 ± 0.23	3.78 ± 0.01

	0.22	0.21	0.21	0.23	0.22				0.22
2-phenylethanol	12.45 ± 0.81	21.14 ± 1.02	14.99 ± 1.01	24.78 ± 1.92	32.24 ± 2.03	26.76 ± 2.02	29.77 ± 2.03	30.05 ± 3.02	28.23 ± 3.03
Cis-3-hexenol	0.285 ± 0.02	0.340 ± 0.03	0.540 ± 0.04	0.326 ± 0.02	0.459 ± 0.03	0.506 ± 0.04	0.480 ± 0.03	0.612 ± 0.04	0.670 ± 0.05
Σ Alcohols	217.5 ± 1.66	234.0 ± 1.98	236.6 ± 1.85	237.3 ± 2.08	254.1 ± 3.22	252.5 ± 2.34	241.3 ± 2.88	261.1 ± 3.01	258.79 ± 2.56
ETHYL ESTERS									
Ethylbutyrate	0.365 ± 0.01	1.280 ± 0.06	0.648 ± 0.02	0.890 ± 0.04	2.230 ± 0.11	1.988 ± 0.09	1.220 ± 0.09	2.228 ± 0.13	2.068 ± 0.11
Ethylhexanoate	0.890 ± 0.03	1.056 ± 0.04	0.905 ± 0.02	1.220 ± 0.05	1.899 ± 0.09	1.767 ± 0.06	2.011 ± 0.08	2.022 ± 0.11	1.989 ± 0.09
Ethyl octanoate	0.522 ± 0.02	0.787 ± 0.04	0.723 ± 0.03	0.976 ± 0.04	1.568 ± 0.06	0.987 ± 0.04	2.034 ± 0.07	1.889 ± 0.08	1.654 ± 0.06
Ethyldecanoate	0.202 ± 0.01	0.389 ± 0.02	0.250 ± 0.01	0.738 ± 0.02	0.679 ± 0.02	0.389 ± 0.01	1.005 ± 0.03	0.972 ± 0.04	0.786 ± 0.03
Ethyllactate	9.22 ± 0.45	9.01 ± 0.43	9.67 ± 0.33	9.99 ± 0.78	10.11 ± 0.59	10.87 ± 0.46	10.97 ± 0.88	11.06 ± 0.65	11.02 ± 0.67
Ethylpropionate	0.595 ± 0.04	0.598 ± 0.04	0.603 ± 0.05	0.600 ± 0.05	0.599 ± 0.04	0.607 ± 0.05	0.615 ± 0.05	0.622 ± 0.05	0.619 ± 0.05
Ethyllaurate	0.230 ± 0.02	0.210 ± 0.01	0.245 ± 0.01	0.256 ± 0.02	0.275 ± 0.03	0.278 ± 0.02	0.287 ± 0.02	0.302 ± 0.03	0.290 ± 0.02
Diethylsuccinate	0.433 ± 0.01	0.887 ± 0.06	0.567 ± 0.02	0.990 ± 0.03	0.989 ± 0.06	0.830 ± 0.03	0.565 ± 0.11	0.349 ± 0.09	0.477 ± 0.10
Σ Ethyl esters	12.46 ± 0.93	14.22 ± 0.87	13.61 ± 0.78	15.66 ± 0.91	18.35 ± 1.09	17.72 ± 1.12	18.71 ± 1.45	19.44 ± 1.77	18.90 ± 1.43
FATTY ACIDS									
Isobutyric acid*	1.23 ± 0.09	0.95 ± 0.03	1.01 ± 0.05	2.02 ± 0.11	1.12 ± 0.05	2.05 ± 0.34	2.97 ± 0.12	2.36 ± 0.11	3.10 ± 0.48
Butyric acid*	2.34 ± 0.13	1.25 ± 0.04	3.56 ± 0.13	3.13 ± 0.12	2.22 ± 0.10	3.45 ± 0.22	4.15 ± 0.26	2.86 ± 0.13	3.90 ± 0.023
Isopentanoic acid	3.45 ± 0.12	4.87 ± 0.26	4.59 ± 0.25	4.47 ± 0.22	7.58 ± 0.37	6.66 ± 0.42	6.56 ± 0.35	8.69 ± 0.43	8.24 ± 0.51
Hexanoic acid	2.59 ± 0.09	3.25 ± 0.13	2.66 ± 0.10	3.33 ± 0.14	4.10 ± 0.16	3.75 ± 0.16	4.78 ± 0.19	4.89 ± 0.20	4.45 ± 0.17
Octanoic acid	2.07 ± 0.14	2.67 ± 0.18	2.95 ± 0.19	3.79 ± 0.15	3.85 ± 0.16	3.50 ± 0.14	4.47 ± 0.25	4.30 ± 0.18	4.22 ± 0.16
Decanoic acid	0.556 ± 0.01	0.770 ± 0.02	0.789 ± 0.01	0.892 ± 0.02	1.123 ± 0.07	1.023 ± 0.08	1.478 ± 0.11	1.564 ± 0.13	1.345 ± 0.11
Σ Fatty acids	12.24 ± 0.75	13.76 ± 0.69	15.56 ± 0.98	17.63 ± 0.88	19.99 ± 1.11	20.43 ± 0.98	24.41 ± 1.66	24.66 ± 1.86	25.26 ± 1.89
MONOTERPENES									
Linalol	0.109 ± 0.006	0.123 ± 0.007	0.121 ± 0.003	0.112 ± 0.007	0.156 ± 0.007	0.134 ± 0.008	0.138 ± 0.009	0.179 ± 0.006	0.169 ± 0.005
α-terpinol	0.055 ± 0.004	0.067 ± 0.005	0.065 ± 0.003	0.087 ± 0.005	0.089 ± 0.005	0.096 ± 0.006	0.105 ± 0.006	0.123 ± 0.007	0.125 ± 0.007
Citronellol	0.029 ± 0.002	0.037 ± 0.003	0.036 ± 0.002	0.056 ± 0.003	0.069 ± 0.003	0.088 ± 0.004	0.095 ± 0.004	0.104 ± 0.005	0.112 ± 0.006
Nerol	0.008 ± 0.001	0.011 ± 0.001	0.010 ± 0.001	0.037 ± 0.002	0.056 ± 0.003	0.048 ± 0.003	0.089 ± 0.003	0.090 ± 0.005	0.095 ± 0.005
Geraniol	0.046 ± 0.002	0.034 ± 0.002	0.030 ± 0.001	0.076 ± 0.005	0.078 ± 0.005	0.066 ± 0.003	0.093 ± 0.006	0.091 ± 0.006	0.089 ± 0.005
Σ Monoterpenes	0.247 ± 0.022	0.272 ± 0.01	0.262 ± 0.02	0.368 ± 0.03	0.448 ± 0.03	0.432 ± 0.02	0.520 ± 0.04	0.587 ± 0.04	0.580 ± 0.05
Σ TOTAL	250.56 ± 2.29	270.82 ± 2.79	274.71 ± 2.98	279.22 ± 3.78	300.25 ± 3.45	298.76 ± 3.67	291.71 ± 3.36	312.03 ± 3.25	310.00 ± 3.17

COMPOUNDS

Significant effects were observed in the total amount of the volatile compounds within the three rootstocks used for each canopy treatment of the experience.

Table 5: Influence of the rootstocks and canopy treatments on the volatile compounds of wines

Parameters	Rootstocks			Canopy treatments		
	3309	1103	140	Control	Leaf removal	Topping
ACETATES						
Ethylacetate	Ns	ns	ns	ns	*	*
Isoamylacetate	*	*	*	*	*	*
Hexylacetate	*	*	*	*	*	*
Methylacetate	Ns	ns	ns	*	*	*
Phenylacetate	*	*	*	*	*	*
Isobuthylacetate	*	*	*	ns	ns	ns
∑ Acetates	*	*	*	*	*	*
ALCOHOLS						
Methanol	*	*	ns	*	*	*
1-propanol	Ns	ns	*	*	ns	*
Isoamyl alcohols	*	*	*	*	ns	ns
Isobutanol	*	*	*	*	*	*
n-amylalcohol	*	ns	ns	ns	ns	*
1-hexanol	*	*	*	*	*	*
2-phenylethanol	*	*	ns	*	*	*
Cis-3-hexenol	*	*	*	*	*	*
∑ Alcohols	*	*	*	*	*	*
ETHYL ESTERS						
Ethylbutyrate	*	*	*	*	*	*
Ethylhexanoate	Ns	*	ns	*	*	*
Ethyl octanoate	*	*	*	*	*	*
Ethyldecanoate	*	*	*	*	*	*
Ethyl lactate	*	ns	ns	*	*	*
Ethylpropionate	Ns	ns	ns	*	*	*
Ethyl laurate	*	*	ns	*	*	*
Diethylsuccinate	*	*	*	*	*	*
∑ Ethyl esters	*	*	ns	*	*	*
FATTY ACIDS						
Isobutyric acid*	Ns	ns	ns	*	*	*
Butyric acid*	*	*	*	*	*	*
Isopentanoic acid	*	*	*	*	*	*
Hexanoic acid	*	*	*	*	*	*
Octanoic acid	*	*	ns	*	*	*
Decanoic acid	*	ns	*	*	*	*
∑ Fatty acids	*	*	ns	*	*	*
MONOTERPENES						
Linalol	Ns	*	*	*	*	*
α-terpinol	Ns	ns	*	*	*	*
Citronellol	*	*	ns	*	*	*
Nerol	Ns	*	ns	*	*	*
Geraniol	*	*	ns	*	*	*
∑ Monoterpenes	*	*	*	*	*	*
TOTAL COMPOUNDS	*	*	*	*	*	*

In the same row for each rootstock and canopy treatment: * indicates significant differences at 0.05.

The results of the wine sensory analysis (Table 6) show that there were significant differences in color among the wines from 1103 and 140 rootstocks, with the highest values in those from the 140 rootstock. The intensity and quality of the aroma were found to be significantly different in all the wines from each of the rootstocks, with the highest values in the wines from the 140 rootstock. There were also significant differences for the intensity and quality of the taste between the wines from the three rootstocks, with the highest values in the wines from the 3309 rootstock.

As regards the influence of canopy treatments on the wines' sensory attributes, significant differences were found between the wines from vines subjected to leaf removal in relation to color, intensity and quality of aroma, with the highest values in the wines from the Ruggeri 140. Significant differences were found in the intensity and quality of taste between the wines from control, leaf removal and topping. For the control and topping treatments, differences were found between the wines from the 3309 rootstock and the wines from the 1103 and 140 rootstocks for taste intensity and quality, but there were no significant differences between the 1103 and 140 rootstocks. The highest values were given to the wines from 3309 rootstock for taste intensity and those from 1103 rootstock for taste quality.

In the final assessment of the wines, significant differences were found for control, leaf removal and topping treatment, with the values of the wines from the 3309 rootstock obtaining the highest values. There were significant differences between the wines from the 3309 and those from the 1103 and 140 rootstocks, but not between the two latter wines.

Table 6: Mean values of the wines sensory attributes

WINE	COLOUR	AROMA		TASTE		FINAL ASSESSMENT
		INTENSITY	QUALITY	INTENSITY	QUALITY	
Wine 1	5.80 ± 0.40	5.10 ± 0.36	5.30 ± 0.26	6.00 ± 0.24	6.00 ± 0.44	6.00 ± 0.33
Wine 2	6.20 ± 0.30	5.60 ± 0.46	5.60 ± 0.29	6.80 ± 0.24	6.50 ± 0.51	6.70 ± 0.34
Wine 3	6.20 ± 0.40	5.10 ± 0.38	5.10 ± 0.28	5.70 ± 0.26	5.80 ± 0.34	5.50 ± 0.25
Wine 4	5.60 ± 0.42	5.00 ± 0.37	5.20 ± 0.24	5.00 ± 0.59	5.40 ± 0.39	5.50 ± 0.40
Wine 5	6.60 ± 0.32	6.00 ± 0.43	5.80 ± 0.29	6.10 ± 0.59	6.20 ± 0.31	6.20 ± 0.35
Wine 6	5.90 ± 0.32	5.30 ± 0.27	5.00 ± 0.20	5.30 ± 0.59	5.20 ± 0.30	5.30 ± 0.34
Wine 7	5.90 ± 0.34	5.40 ± 0.33	5.20 ± 0.28	5.30 ± 0.22	5.10 ± 0.27	5.50 ± 0.24
Wine 8	6.80 ± 0.29	6.30 ± 0.44	6.40 ± 0.49	6.40 ± 0.22	6.30 ± 0.47	6.40 ± 0.56
Wine 9	6.00 ± 0.24	5.30 ± 0.30	5.00 ± 0.28	5.10 ± 0.22	5.40 ± 0.29	5.20 ± 0.22

Table 7 shows the influence of rootstock and canopy treatment on the wines' sensory attributes. There were no significant differences in color for the same canopy treatment, and all have similar values. The aroma of wines from 1103 and 140 was found to be significantly different from those from 3309, with the highest values found in these and other wines from control and topping. In the 3309 rootstock, wines were given higher taste values and there were significant differences with those obtained from the other two rootstocks, for all canopy treatments. In the final assessment of the wines, those from the 3309 rootstock were given the highest values, with wines from the leaf removal vines obtaining the highest scores. The wines from vines whose leaves had been topped had the lowest scores.

Table 7: Influence of rootstock and canopy treatment on the wines sensory attributes

Attributes	Rootstocks			Canopy treatments		
	3309	1103	140	Control	Leaf removal	Topping
Color	Ns	*	*	ns	*	ns
Aroma intensity	*	*	*	ns	*	ns
Aroma quality	*	*	*	ns	*	ns
Taste intensity	*	*	*	*	*	*
Taste quality	*	*	*	*	*	*
Final assessment	*	*	*	*	*	*

In the same row for each rootstock and canopy treatment: * indicates significant differences at 0.05

4. Conclusions

For the three rootstocks used in the present study, yield was seen to decrease after both leaf removal and shoot topping canopy treatments. The Beaumé degree of must, and therefore the alcohol content, of wine increases when leaf removal treatment is used, regardless of the rootstock used.

The Ruggeri 140 rootstock obtained the wines with the highest concentration of aromatic compounds when using both leaf removal and shoot topping canopy treatments. The wines obtained from Couderc 3309 had the lowest concentration of aromatic compounds, regardless of the canopy treatment used. The wines from vines subjected to leaf removal and topping had higher concentrations of aromatic compounds than those from the control vines.

In the sensory analysis, the wines from Couderc 3309 generally obtained the highest scores in taste and in the final assessment, while those obtained from Ruggeri 140 were given the highest score for aroma. The wines from vines subjected to leaf removal were the highest rated for color, aroma, and taste and also in the final assessment in the three rootstocks used, while the wines from the control vines were given the lowest ratings.

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References

- [1]. Borghezán, M., Pit, F.A., Gavioli, O., Malinovski, L.I., & Da Silva, A.L. (2011). Effect of leaf area on the grape composition and sensory quality of wines from cultivar Merlot (*Vitis vinifera*) in Sao Joaquim, SC, Brazil. *Ciencia e Tec. Vitivini.*, 26:1-10.
- [2]. Guerra, B. (2006). Tipping Malbec shoots to improve set and yield. *Practical Winery and Vineyard* January/February, 47-53.
- [3]. Collins, C., & Dry, P.R. (2009). Response of fruitset and other yield components to shoot topping and 2-chlorethyltrimethyl-ammonium chloride. *Aus. J. Grape Wine Res.*, 15:256-267.
- [4]. Jackson, D. I., & Lombard, P.B. (1993). Environmental and Management practises affecting grape composition and wine quality. A review. *Am. J. Enol. Vitic.*, 22:409-429.
- [5]. Falqué-López, E., & Fernández Gómez, E. (1996). Effects of different skin contact times on Treixadura wine composition. *Am. J. Enol. Vitic.*, 47:309-312.
- [6]. De Revel, G., Matín, N., Pripis-Nicolau, L., Lonvaud-Funel, A., & Bertrand, A. (1999). Contribution to the knowledge of malolactic fermentation influence on wine aroma. *J. Agr. Food Chem.*, 47:4003-4008.
- [7]. Smart, R. E., Dick, J. K., Gravett, I.M., & Fisher, B. (1990). Canopy management to improve grape yield and wine quality. Principles and practices. *S. Afr. J. Enol. Vitic.*, 11:3-17.
- [8]. Smith, S.M., Codrington, I. C., Robertson, M., & Smart R. E. (1988). Viticultural and oenological implications of leaf removal for New Zealand vineyards. Pro. Second Int. Symp. Cool Climate Viticulture and Oenology, January, Auckland, New Zealand. *NZ Soc. Vitic. And Oenol.*, 127-133.
- [9]. Petrie, P.R., Trought, M.C.T., & Howell, G.S. (2000a). Influence of leaf ageing, leaf area and crop load on photosynthesis, stomatal conductance and senescence of grapevines (*Vitis Vinifera* L. cv. Pinot noir) leaves. *Vitis*, 39:31-36.
- [10]. Petrie, P.R., Trought, M.C.T., Howell, G.S., & Buchan, G.D. (2003). The effect of leaf removal and canopy weight on whole-vine gas Exchange and fruit development of *Vitis Vinifera* L. Sauvignon Blanc. *Functional Plant Biology*, 30:711-717.
- [11]. Percival, D.C., Fisher, K.H., & Sullivan, J.A. (1994). Use of fruit zone leaf removal with *Vitis vinifera* L. cv. Riesling grapevines. II. Effect on fruit composition, yield, and occurrence of bunch rot (*Botrytis cinerea* Pers.: Fr). *Am. J. Enol. Vitic.*, 45:133-140
- [12]. Poni, S., Casalini, L., Bernizzoni, F., Civardi, S., & Intrieri, C. (2006). Effects of early defoliation on shoot photosynthesis, yields components and grape composition. *Am. J. Enol. Vitic.*, 57:397-407.



- [13]. Reynold, A.G., Edwards, D.A., Wardle, D.R., Webster, D.R., Dever, M. (1994). Shoot density affects “Riesling” grapevines. I. Vine performance. *J. Am. Soc. Hortic. Sci.*, 119:874-880.
- [14]. Reynolds, A.G., Wardle, D.A., & Dever, M. (1996). Vine performance, fruit composition, and wine sensory attributes of Gewürtztraminer in response to vineyard location and canopy manipulation. *Am. J. Enol. Vitic.*, 47:77-92.
- [15]. Zoecklein, B.W., Wolf, T.K., Duncan, S.E., Marcy, J.E., & Jasinski, Y. (1998). Effect of fruit zone leaf removal on total glycoconjugates and conjugates fraction concentration of Riesling and Chardonnay (*Vitis vinifera* L.) grapes. *Am. J. Enol. Vitic.*, 49:259-265.
- [16]. Kliewer, W.M., & Bledsoe, A. (1987). Influence of hedging and leaf removal on canopy microclimate, grape composition, and wine quality under California conditions. *Acta Hort.*, 206:157-168.
- [17]. Kliewer, W.M., & Dokoozlian, N.K. (2005). Leaf area/crop weight ratios of grapevines influence on fruit composition and wine quality. *Am. J. Enol. Vitic.*, 56:170-181.
- [18]. Gil, G.F., & Pszczolkowski, P. (2007). *Viticultura. Fundamentos para optimizar producción y calidad*. Ed. Universidad Católica de Chile, p. 301-302.
- [19]. Ribereau-Gayon, P., Glories, Y., Maujean, A., & Ddubourdieu, D. (2000). *Handbook of Enology*. pp 205-207. Chichester: Wiley.
- [20]. López-Tamames, E., Carro-Mariño, N., Gunatas, Y.Z., Sapis, C., Baumes, R., & Bayonove, C. (1997). Potential aroma in several varieties of spanish grapes. *J. Agr. Food Chem.*, 45:1729-1735.
- [21]. Pérez-Coello, M. S., González-Viñas, M. A., Cabezudo, M. D. (2000). Chemical characteristics of young White wines of the varieties Airén, Viura and Macabeo. *Alimentación, Equipos y Tecnología*, 19:125-129.
- [22]. Reyero, J. R., Garijo, J., Díaz-Plaza, E., Cuartero, H., Salinas, M. R., & Pardo F. (2000). Comparison of the aromatic compositions of monovarietal red wines. *Alimentación, Equipos y Tecnología*, 19:101-110.
- [23]. De La Presa-Owens, C., Lamuela-Reventos, R. M., Buxaderas, S., & De La Torre- Boronat, M.C. (1995). Differentiation and grouping characteristics of varietal grape musts from Penedés region. *Am. J. Enol. Vitic.*, 46:283-291.
- [24]. Aleixandre, J. L., Lizama, V., Alvarez, I., & García, M. J. (2002). Varietal differentiation of red wines in the Valencian region (Spain). *J. Agr. Food Chem.*, 50:751-755.
- [25]. Aznar, M., López, R., Cacho, J.F., & Ferreira, V. (2001). Identification and quantification of impact odorants of aged red wines from Rioja. GC-Olfactometry, quantitative GC-MS, and odor evaluation of HPLC fractions. *J. Agr. Food Chem.*, 49:2924-2929.
- [26]. Olalla, M., López, M. C., López, H., & Villalón, M. (1994). Quelques composants volatils des vins de la region de l’Alpujarra –Contraviesa (ESpagne). *J. Int. Sci. Vigne Vin*, 28:277-281.
- [27]. Mulet, A., Bernat, A., & Forcén, M. (1992). Differentiation and grouping characteristics of varietal grape musts and wines from Majorcan Origin. *Am. J. Enol. Vitic.*, 43:221-226.
- [28]. Pomar-García, M., González-Mendoza, L. A., & Díaz-Rodríguez, F. (1994). Analytical characteristics of red wine from the Canary Islands (Spain). *J. Int. Sci. Vigne Vin*, 28:173-179.
- [29]. Koblet, W. (1987) Effectiveness of shoot topping and leaf removal as a means of improving quality. *Acta Horticulturae, Wageningen*, 206:141-155.
- [30]. Reynolds, A.G., & Wardle, D.A. (2001). Rootstocks impact vine performance and fruit composition of grapes in Britis Columbia. *HortTechnology, Stanford* 11, 419-427.
- [31]. Main, G., Morris, J., & Striegler, K. (2002). Rootstocks effects on Chardonnay productivity, fruit and wine composition. *Am. J. Enol. Vitic.*, 53:37-40.
- [32]. Mandelli, F., Miele, A., Rizzon, L.A., & Zanus, M.C. (2008). Efeito da poda verde na composição fisico-química do mosto da uva Merlot. *Revista Brasileira de Fruticultura, Jaboticabal*, 30:667-674.
- [33]. Brighenti, A.F., Rufato, L., Kretschmar, A.A., & Madeira, F.C. (2010). Shoot topping on Merlot grapevines grafted on Paulsen 1103 and Couderc 3309 and its effects on the quality of grapes. *Revista Brasileira de Fruticultura, Jaboticabal*, 23:1-6.

- [34]. Aleixandre, J. L., Padilla, A. I., Navarro, L.L., Suria, A., García, M. J., & Alvarez, I. (2003). Optimization of making barrel fermented dry Muscatel wines. *J. Agr. Food Chem.*, 51:1889-1893.
- [35]. UE, 2003: Official methods to wine analyses. Reglamento 440/2003
- [36]. O.I.V. (1990). Recopilación de los métodos internacionales de análisis de vinos 16.
- [37]. Bavaresco, L., Gatti, M., Pezzutto, S., Fregoni, M., & Mattivi, F. (2008). Effect of leaf removal on grape yield, berry composition, and stilbene concentration. *Am. J. Enol. Vitic.*, 59:292-298.

