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Mechanical yield regulation in winegrapes: comparison of early defoliation and crop thinning

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Abstract

Background and Aims: Winter pruning and manual bunch thinning are the primary methods for crop regulation in viticulture. Recently, innovative mechanical approaches have been proposed as cost-effective for yield management. The aim of this work was to compare the effectiveness of mechanical early defoliation and mechanical crop thinning on yield regulation, and on grape and wine composition.

Methods and Results: The impact of mechanical early defoliation and crop thinning, applied at different timings, was investigated in *Vitis vinifera* L. cv. Tempranillo vertically shoot-positioned-trained grapevines over two seasons. Effects on yield components, leaf area, botrytis incidence, grape and wine composition were determined. Yield per vine was drastically reduced by both techniques (35–40%). Bunch weight, number of berries per bunch, bunch compactness and botrytis were also reduced by most of the treatments. Total leaf area per vine was not affected, however, the total leaf area-to-yield ratio increased in most cases. Berry soluble solids, anthocyanins and total phenols increased in the grapes. Wines were higher in alcohol and more intensely coloured in mechanical early defoliation treatments in comparison with those of mechanical thinning.

Conclusions: Mechanical early defoliation and crop thinning may be suitable and efficient for regulating grape yield and improving grape and wine composition. Early defoliation, however, appeared to be more consistent.

Significance of the Study: Effective yield control in winegrapes may be accomplished by mechanical techniques implemented between pre-flowering and veraison. The choice of mechanical technique for yield management may influence the grape and wine composition.

Keywords: *grape composition, leaf removal, mechanisation, yield component, yield management*

Introduction

Yield management is becoming increasingly important in modern viticulture. The wine industry has been particularly interested in cost-effective yield regulation in order to minimise seasonal variation and to provide more consistent grape supply (Clingeleffer 2010) and to manage the large international wine surplus (IWSR Drinks Record 2006).

Traditionally, regulation of bud and therefore shoot number through winter pruning is used as the primary means to control yield, whereas finer tuning is achieved in both European (Bertamini et al. 1991, Ridomi et al. 1995, Guidoni et al. 2002) and New World countries (Chapman et al. 2004, Keller et al. 2005, Reynolds et al. 2007) through shoot and/or bunch thinning. Such operations are fairly expensive because of their high labour requirements (Martinez de Toda and Tardaguila 2003). On the other hand, Weyand and Schultz (2006) showed that regulating yield in winegrapes could be performed through the application of gibberellic acid; yet, May (2004) observed highly variable results and a negative effect on inflorescence induction. Recently, an innovative method using anti-transpirant applications, to reduce fruitset, was successfully used to control yield (Palliotti et al. 2010).

Mechanical crop regulation has the potential to be a cost-effective technique for both yield control and improved grape and wine composition (Intrieri et al. 2008, Tardaguila et al. 2008). Recently, two mechanical approaches based on totally different principles have been tested for yield control in the vineyard: mechanical crop thinning (MT) (Petrie and Clingeleffer 2006, Tardaguila et al. 2008) and early leaf removal (Intrieri et al. 2008, Tardaguila et al. 2010).

Mechanical thinning was originally developed in the USA to reduce yield and improve the sugar content of Concord grapes for juice production (Pool et al. 1993, Fendinger et al. 1996). Since then, different methods of mechanical thinning have been successfully trialled on minimally pruned vines in Australia (Clingeleffer et al. 2002, Petrie et al. 2003). Additionally, Petrie and Clingeleffer (2006) have further developed the mechanical thinning system on Cabernet Sauvignon minimally pruned vines, trained to two vertically divided cordons, with high yields. In this trial, a substantial yield reduction (25–45%) and advanced fruit ripeness were induced by mechanical thinning using a harvester machine. More recently, a 2-year study conducted on vertically shoot-positioned (VSP) Tempranillo and Grenache vines has shown that mechanical thinning applied by canopy shaking, without hitting the bunches, using an over-row harvester machine, significantly decreased yield and bunch compactness (Tardaguila et al. 2008). In a companion study, MT enhanced sugar and phenolic concentration and improved the wine's aroma, taste and mouthfeel (Diago et al. 2010a).

Early leaf removal, performed around flowering, is another method to manage yield. Crop regulation is achieved in early

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defoliated vines through reduced fruitset and/or berry size, leading to smaller and looser bunches that are less susceptible to botrytis rot (Poni et al. 2006, Intrieri et al. 2008, Tardaguila et al. 2010). Early leaf removal improved grape composition (Poni et al. 2006, Intrieri et al. 2008, Poni and Bernizzoni 2010) and wine chemical (Tardaguila et al. 2010) and sensory properties (Diago et al. 2010b). Feasibility of mechanical early defoliation has been recently demonstrated (Intrieri et al. 2008, Tardaguila et al. 2010). Intrieri et al. (2008) used a suction and cutting leaf plucking machine on COMBI-trained grapevines, whereas Tardaguila et al. (2010) used a pulsed air defoliator in VSP-trained grapevines.

The goal of this study was to compare the effectiveness of two mechanical approaches for yield regulation (early defoliation and crop thinning) applied at different timings on yield components, grape and wine composition of *Vitis vinifera* L. cv. Tempranillo VSP-trained grapevines as compared with an unthinned, non-defoliated control.

Material and methods

Plant material and experimental layout

This trial was conducted in a commercial cv. Tempranillo (*Vitis vinifera* L.) vineyard in Ollauri (lat: 42°31′N; long: 2°49′W, 527 m), La Rioja (Spain) in 2007 and 2008. Tempranillo vines were grafted onto 110 Richter rootstock and planted in 1996 in a clay-loam soil. Vines were spaced 2.70 m inter-row and 1.15 m intra-row. Row orientation was north-east–south-west. The vines were spur-pruned (12 nodes per vine) on a bilateral cordon and trained to a VSP trellis system. The trellis featured a supporting wire at 0.70 m, two wires at 1.00 m above-ground for protection against wind damage and a pair of movable shoot-positioned wires at 1.45 m. Vines were not irrigated during the growing season. Shoots were slightly trimmed before bunch closure in July and positioned twice between the catch wires. No fungicide sprays for botrytis control were applied.

The experimental design compared the following treatments: (i) control or non-defoliated, non-thinned vines; (ii) mechanical defoliation (MD) at pre-flowering (stage 19); (iii) MD at fruitset (stage 27); (iv) mechanical thinning at bunch closure (stage 32); and (v) mechanical thinning at the beginning of veraison, with 10–15% coloured berries (stage 35); all growth stages as per Coombe (1995). Pre-flowering leaf removal treatments were performed on 10 June in 2007 and on 13 June in 2008. Fruitset defoliation was carried out on 27 June in 2007 and on 1 July in 2008. Bunch closure thinning treatments were conducted on 18 July 2007 and 20 July 2008. Veraison thinning treatments were performed on 12 August 2007 and 17 August 2008.

MD was conducted with a tractor-mounted pulsed air leaf remover (Collard, Bouzy, France), which operates by blowing compressed air with enough force to tear a whole leaf or sections of leaf blades off. The machine was driven at approximately 0.5 km/h and removed the leaves around the basal 0.6 m of foliage in the fruiting zone. The air shear system was positioned close to the canopy. The leaf remover made two passes, one on each side of the canopy.

MT was performed by an over-row grape harvester (New Holland VL610, Coex, France). The harvester was fitted with two pairs of bow rods operating at 470 beats/min and driven at approximately 3.0 km/h based on the results of previous work (Tardaguila et al. 2008). The height of the harvester and the position of the bow rods were adjusted so that the fruiting zone of the canopy was not hit by rods. The bunches were mostly located between 0 and 0.25 m above the cordon, and bow rods hit the vine trunks below the cordon, causing the thinning to occur because of canopy vibration. After mechanical thinning, some damaged bunches and berries were not completely removed, but stayed on the vines and desiccated over the following 5–7 days.

In each experiment, treatments were arranged in a completely randomised design consisting of five replicates of 20 vine plots for each treatment. Several 25 test vines (five test vines × five replicates) were tagged per treatment. The treatments were applied to the same vines in 2007 and 2008.

Yield components, leaf area and Botrytis assessment

In MD, fruitset was estimated in the 25 tagged basal bunches (one bunch per each test vine) per treatment using the method proposed by Poni et al. (2006). Each bunch was photographed against a dark background with a digital camera the day before defoliation. An initial flower number on tagged inflorescences was estimated using a linear regression between actual flower number and the number of flowers counted on photo prints, established for 30 inflorescences taken from extra vines (regression equation: $y = 1.9335x^{0.9684}$, $R^2 = 0.89$).

Just before harvest, total leaf area was assessed using the method proposed by Smart and Robinson (1991). In this way, for two representative shoots per vine, all main and lateral leaves were separately removed and weighed. One-hundred discs (diameter: 2 cm) were cut from these leaves and weighed, allowing the estimation of the weight-to-area ratio, which was further used for the calculation of the main and lateral leaf area per shoot. Total leaf area per shoot was computed by the sum of main and lateral leaf areas. The number of shoots per vine was also counted and the total leaf area per vine (TLA) calculated.

All treatments were harvested on the same date (8 October 2007 and 15 October 2008), and yield components were assessed. In the defoliation treatments, yield and bunch number per vine were recorded. In MT, bunches were classified by visual inspection in three classes: 'undamaged' (less than 5% damaged berries), 'partially-dried' (between 5% and 80% of damaged berries) and 'dried' (more than 80% damaged berries). The bunches were counted and weighed per each class. For the estimation of the average bunch weight, only the 'undamaged' bunches were considered in mechanical thinning treatments. The leaf-to-fruit ratio, denoted as total leaf area per yield (TLA/Y) was computed per each vine.

Berry number and weight, bunch compactness and visual presence or absence of botrytis bunch rot were assessed on three bunches per vine. Bunch compactness was visually assessed following the International Organization of Vine and Wine (OIV), code 204 (OIV 2009b) which ranks 'berries in grouped formation with many visible pedicels' as 1 and 'misshapen berries' as 9. Botrytis incidence was then estimated as the proportion of affected bunches per treatment, these showing a minimum of 2% of infected berries. Only 'undamaged' bunches (less than 5% damaged berries), were considered for mechanical thinning treatments for the appraisal of yield components, compactness and botrytis incidence.

Grape composition

After yield components and botrytis incidence appraisal, all bunches from each test vine (five vines per replicate) were manually destemmed, and the berries were mixed. Within this pool of berries, a representative subsample of 50 berries per vine was randomly taken for subsequent analysis of grape composition. The total soluble solid concentration (°Brix) was determined using a temperature-compensating digital refractometer (Atago, Tokyo, Japan), and titratable acidity, pH, tartaric and malic acid concentration was determined according to the OIV methods (OIV 2009a). Furthermore, another 50-berry subsample per vine was taken for the analysis of anthocyanins and total phenols and weighed. These subsamples were frozen and stored at −18°C until phenolic analysis. Berries were allowed to partially thaw prior to homogenisation, and temperature was kept under 5°C at all times. Each subsample of 50 berries was homogenised using an Ultra Turrax grind mixer (IKA, Staufen, Germany) at high speed (1425 *g* for 1 min). Anthocyanin and phenolic concentration was determined according to the method of Iland et al. (2004). Total anthocyanins were expressed as mg per berry and mg per gram of berry mass, whereas total phenols were expressed as absorbance units at 280 nm per berry and per gram of berry mass.

Vinification and wine analysis

For each treatment, the remaining grapes from the five labelled vines per replicate were blended and stored for 12 h at 4.5°C at the winery of the University of La Rioja. Five wine fermentations were conducted for each treatment using 5 kg of grapes, after Sampaio et al. (2007). Grapes (manually destemmed) were slightly crushed using a crusher machine (Eno-50, Enomundi, Zaragoza, Spain). Sulfur dioxide was added at a rate of 60 mg/kg and musts inoculated with yeast (*Saccharomyces cerevisiae*, Uvaferm 71B, Lallemand, Montréal, Québec, Canada) at a rate of 20 g/hL. The fermentation temperature was kept between 27–30°C. The acidity was not adjusted during winemaking. Alcoholic fermentations were completed after 7 days (glucose + fructose <1 g/L), but extended maceration was allowed for 8 more days in all cases. After fermentation, wines were racked off and pressed manually. For each microfermentor, the free-run and pressed wine fractions were blended and bottled as described by Diago et al. (2010b). The alcohol content, titratable acidity, pH and malic acid concentration were determined after OIV (2009a). Colour density was calculated by adding the absorbance readings at 420, 520 and 620 nm, whereas hue was measured as the ratio of absorbance readings at 420–520 nm (Glories 1984). Total polyphenol index was calculated by the absorbance reading at 280 nm as described by the EEC method (1990). All analyses were run in triplicate.

Statistical analysis

Analysis of variance was performed using the InfoStat statistical package (Professional 2007 edition, Universidad Nacional de Cordoba, Cordoba, Argentina). Year was considered as a random variable. Mean separation between defoliation levels was performed with the Student–Newman–Keuls test ($P = 0.05$).

Effects of berry size and total soluble solids (as indicator of different ripening stages) on the acidity parameters (pH and titratable acidity), anthocyanins and total phenol content were examined by covariate analysis using berry weight and soluble solids as covariate variables. Similarly, the alcohol content and berry weight were used as covariate variables for the analysis of wine colour and phenolic traits.

Results

Yield components

Overall, all yield components were significantly affected by both yield management techniques (Table 1). Yield per vine was drastically reduced (35–42%) on average by MD treatments, whereas MT led to a significant reduction in the yield per vine in 2007 (−52% as compared with control) and a trend towards a reduced yield was observed only in 2008. No significant differences due to timing of MT were observed on yield per vine.

Table 1. Influence of mechanical early defoliation (MD) at pre-flowering and fruitset, and mechanical thinning (MT), at bunch closure and veraison on Tempranillo yield components in 2007 and 2008 ($n = 25$).

Treatment	Yield/vine (kg)		Bunches/vine		Bunch weight (g)		Berries/bunch		Fruitset (%)†		Berry weight (g)		Bunch compactness (rating)‡	
Year	**2007**	**2008**	**2007**	**2008**	**2007**	**2008**	**2007**	**2008**	**2007**	**2008**	**2007**	**2008**	**2007**	**2008**
Control	4.59^{a}	4.11^{a}	15.0^{a}	17.4^{a}	307.2^{a}	229.8^{a}	187.8^{a}	155.2^{a}	54.8^{a}	44.1^{a}	1.67^{ab}	1.58^{c}	6.4^{a}	4.4^{a}
MD at pre-flowering	3.13^{b}	2.18^{c}	14.1^{b}	13.8^{b}	222.4^{b}	149.7^{c}	146.2^{b}	91.2^{c}	46.3^{b}	33.4^{b}	1.57^{bc}	1.63^{bc}	4.7^{b}	3.1^{b}
MD at fruitset	2.18^{c}	3.18^{b}	13.4^{b}	15.8^{b}	167.4^{c}	198.4^{b}	137.3^{b}	124.5^{b}	23.8^{c}	38.6^{ab}	1.37^{c}	1.56^{c}	2.9^{c}	3.4^{b}
MT at bunch closure	2.32^{c}	3.58^{ab}	13.2^{b}	14.9^{b}	287.6^{a}	256.9^{a}	142.9^{b}	140.7^{ab}	—	—	1.96^{a}	1.88^{a}	4.7^{b}	4.9^{a}
MT at veraison	2.05^{c}	3.40^{ab}	13.1^{b}	14.8^{b}	207.0^{bc}	240.5^{a}	98.5^{c}	138.2^{ab}	—	—	1.75^{ab}	1.76^{ab}	3.9b^{c}	4.5^{a}
Analysis of variance *P*-values														
Treatment	<0.001	<0.001	0.021	0.024	<0.001	<0.001	0.001	<0.001	0.013	0.011	0.001	0.001	0.001	0.001
Treatment†year	<0.001		0.028		0.020		0.004		n.s.		n.s.		<0.001	

Means values ($n = 25$) within columns designated by different superscript letters are significantly different by the Student–Newman–Keuls test ($P < 0.05$). †Fruitset was estimated using the method proposed by Poni et al. (2006). ‡Bunch compactness was visually assessed using the code 204 of the International Organization of Vine and Wine (OIV 2009b), which ranks 'berries in grouped formation with many visible pedicels' as 1 and 'misshapen berries' as 9. —, data not measured; n.s., not significant.

On the other hand, the effectiveness of MD at pre-flowering versus fruitset varied between seasons.

The number of bunches per vine was decreased in MD and MT vines in both years as compared with that of the control (Table 1). No significant differences in number of bunches per vine were observed between MD and MT vines. No timing effect was observed with either crop regulation techniques.

Fruitset was reduced by the MD treatments in both seasons, and the number of berries per bunch was reduced by an average of 27% compared with that of control vines over the two seasons (Table 1). Interseasonal discrepancies in the effect of the timing of MD in fruitset were observed. Bunch weight was significantly diminished in MD vines in both years, whereas it did not vary in MT vines as compared with control (except for MT at veraison in 2007). Bunch compactness was significantly reduced in MD vines in both seasons, whilst berry weight remained unaffected as compared with that of the control with the exception of MD at fruitset in 2007, which led to reduced berry weight. On the other hand, berry number per bunch, berry weight and bunch compactness responded differently to MT treatments in the two seasons. In 2007, looser bunches containing less berries (–28% for MT-bunch closure and –47% for MT at veraison compared with control vines) were obtained.

At harvest, no damaged bunches were found in any MD treatment, and in the MT treatments, most of the fruit was not damaged by the machine (data not shown). Seventy per cent of the bunches from MT vines, representing 83% of the final yield, corresponded to 'undamaged' fruit, averaged over the two seasons. The number of 'dried' and 'partially-dried' bunches per vine varied between 5–13% and 11–19%, respectively. The incidence of the damage to the bunches caused by MT was not affected by the timing of thinning.

Botrytis incidence was substantially lowered by both mechanical thinning techniques in 2008 (Table 2), whereas no symptoms of this fungal infection were detected in the fruit of any treatment in 2007 (data not shown). Regarding the effects of the mechanical treatment and its timing, no differences in botrytis incidence were found.

At harvest, TLA was not affected by either of the two yield regulation techniques as compared with that of the control in both years (Table 2). The total leaf area-to-yield ratio (TLA/Y), however, was often increased for both MD and MT treatments, although statistical significance was only attained in some cases (Table 2). A significant treatment × season interaction was observed. Interseasonal discrepancies in the effectiveness of MD at pre-flowering versus fruitset were observed for TLA/Y.

Grape composition

MD largely improved grape soluble solids regardless of year and timing of intervention as compared with that of control and MT treatments, whereas MT induced higher °Brix values than control only in 2007 (Table 3). Similarly, must pH increased due to MD with respect to control in both seasons, and MT led to enhanced pH only in 2007. Must titratable acidity values were generally lower than those of control for MD (except for MD at fruitset) and MT treatments in 2007, but no differences were encountered in 2008. Overall, both malic and tartaric acid fractions were less affected by crop regulation treatments in the two seasons. In general, little difference was found for grape acidity parameters between MD and MT treatments, and these variations were not consistent over the two seasons. Grape composition did not differ largely between pre-flowering and fruitset MD treatments, and only in 2008, total soluble solids and pH values were higher in the berries of pre-flowering defoliated vines. Little to no difference in grape composition was found between the two timings for the MT treatments.

Both mechanical yield management techniques led to a significant enhancement in grape anthocyanins, either per berry or per gram of berry basis, as compared with that of the control, with the exception of MT at veraison in 2008 (Table 3). Between the two techniques, MD led to a larger increase in the anthocyanins per berry than MT in season 2008, but this was not reflected in the values expressed per gram of berry. In terms of total phenols, the effects of both techniques as compared with control and between them were less consistent over the 2 years, and a trend towards a larger concentration of total phenols in MD and MT treatments with respect to control was observed. The timing of MD did not influence the anthocyanin and total phenol concentration in either season, whereas for MT, lower values of both anthocyanins and total phenols were found in berries of mechanically thinned vines at veraison in 2008. A significant treatment × season interaction was observed for all berry composition parameters except for malic acid.

Table 2. Influence of mechanical early defoliation (MD) at pre-flowering and fruitset, and mechanical thinning (MT), at bunch closure and veraison on Tempranillo total leaf area per vine, total leaf area per yield and botrytis incidence in 2007 and 2008 of the trial ($n = 25$).

Treatment	Total leaf area (m^2/vine)		Total leaf area/yield (m^2/kg)		Botrytis incidence (%)	
Year	2007	2008	2007	2008	2007	2008
Control	5.1	4.2	1.22^c	0.75^b	0	14.6^a
MD at pre-flowering	4.7	4.1	1.46^c	1.85^a	0	1.9^b
MD at fruitset	5.3	4.3	2.50^b	1.15^b	0	1.6^b
MT at bunch closure	5.7	3.9	2.60^b	0.92^b	0	1.2^b
MT at veraison	5.8	4.4	4.33^a	1.37^a	0	1.4^b
Analysis of variance *P*-values						
Treatment	n.s.	n.s.	0.002	0.004	n.s.	<0.001
Treatment year	n.s.		0.024		<0.001	

Mean values ($n = 25$) within columns designated by different superscript letters are significantly different by the Student–Newman–Keuls (SNK) test ($P < 0.05$). Values within columns without any superscript letter are not significantly different by the SNK test. n.s., not significant.

Table 3. Influence of mechanical early defoliation (MD) at pre-flowering and fruitset, and mechanical thinning (MT), at bunch closure and veraison, on Tempranillo grape composition at harvest in 2007 and 2008 of the trial ($n = 25$).

Treatment	Soluble solids (ºBrix)		pH†		Titratable acidity† (g/L)		Malic acid (g/L)		Tartaric acid (g/L)		Anthocyanins (mg/g berry)		Anthocyanins (mg/berry)		Total phenols (AU/g berry)		Total phenols (AU/berry)	
Year	**2007**	**2008**	**2007**	**2008**	**2007**	**2008**	**2007**	**2008**	**2007**	**2008**	**2007**	**2008**	**2007**	**2008**	**2007**	**2008**	**2007**	**2008**
Control	21.8^{c}	20.1^{c}	3.49^{c} *(3.58^{b})*	3.46^{c}	6.36^{a} *(6.09^{a})*	5.48	5.71^{a}	5.46	3.67^{ab}	3.25	1.51^{b}	1.20^{c}	2.47^{b}	2.34^{b}	1.60^{c}	1.48^{b}	2.62^{b}	2.90^{ab}
MD at pre-flowering	25.6^{a}	25.8^{a}	3.65^{b} *(3.60^{b})*	3.76^{a}	5.08^{bc} *(5.25^{bc})*	4.83	4.83^{b}	5.37	3.46^{bc}	3.18	1.91^{a}	1.75^{a}	3.09^{a}	2.88^{a}	1.71^{c}	1.83^{a}	2.77^{b}	3.00^{ab}
MD at fruitset	26.8^{a}	23.7^{b}	3.77^{ab} *(3.67^{b})*	3.63^{b}	5.77^{ab} *(6.08^{a})*	5.06	4.98^{ab}	5.64	3.28^{c}	3.06	1.93^{a}	1.73^{a}	3.22^{a}	2.76^{a}	1.77^{bc}	1.94^{a}	2.95^{ab}	3.13^{ab}
MT at bunch closure	24.1^{b}	20.9^{c}	3.64^{b} *(3.65^{b})*	3.57^{bc}	5.59^{b} *(5.59^{ab})*	5.27	5.48^{ab}	5.29	3.92^{a}	3.27	1.85^{a}	1.47^{b}	3.41^{a}	2.85^{a}	1.91^{ab}	1.82^{a}	3.52^{a}	3.25^{a}
MT at veraison	23.7^{b}	21.9^{c}	3.87^{a} *(3.89^{a})*	3.54^{c}	4.82^{c} *(4.77^{c})*	5.18	5.25^{ab}	5.42	3.90^{a}	3.11	1.72^{a}	1.33^{bc}	3.01^{a}	2.20^{b}	2.02^{a}	1.54^{b}	3.43^{a}	2.71^{b}
Analysis of variance *P*-values																		
Treatment	<0.001	<0.001	<0.001	<0.001	<0.001	n.s.	0.011	n.s.	<0.001	n.s.	<0.001	<0.001	0.004	<0.001	<0.001	<0.001	<0.001	0.014
Treatment year	0.010		<0.001		0.047		n.s.		<0.001		<0.001		<0.001		<0.001		<0.001	

Mean values ($n = 25$) within columns designated by different superscript letters are significantly different by the Student–Newman–Keuls (SNK) test ($P < 0.05$). Values within columns without any superscript letter are not significantly different by the SNK test. †Mean values from covariate analyses using soluble solids as covariate are shown in italics and brackets. AU, absorbance units; n.s., not significant.

The analysis of covariance on grape acidity data using the total soluble solids as covariate revealed significant differences in titratable acidity and pH (Table 3). The covariate analysis, however, on grape anthocyanins and phenols using the total soluble solids and berry weight as covariates did not exhibit significant differences (data not shown).

Figure 1 shows the relationship between total soluble solids of Tempranillo berry juice at harvest and the total leaf area-to-yield ratio (TLA/Y) for MD and MT treatments. For both techniques, the total soluble solids were positively correlated with the TLA/Y, with Pearson correlation coefficients, r = 0.74*** (for MD) and 0.75*** (for MT). When the two techniques were compared, higher total soluble solid values were observed at any TLA/*Y*-value in MD vines. Likewise, for TLA/*Y*-values ranging from 1 to 2 m^2/kg, enhancements of 1–2°Brix could be observed in MD berries with respect to MT fruit, reaching 2.5–3°Brix from TLA/Y of 2 m^2/kg onwards. The Student *t*-test confirmed that MD and MT regressions with total soluble solids were significantly different ($P < 0.001$).

Furthermore, significant relationships between the anthocyanin concentration in berries (mg/g berry) at harvest, and the total leaf area-to-yield ratio (TLA/Y) for MD (r = 0.53*) and MT (r = 0.60*) treatments in both seasons were found (Figure 2). For values of TLA/Y larger than 1.5 m^2/kg, the anthocyanin concentration in berries corresponding to MD treatments was always significantly higher (0.10–0.15 mg/g berry) than those of MT (1.75–1.90 mg/g berry). No relationship between total phenols and TLA/Y was found for any of the two mechanical techniques (data not shown). The Student *t*-test confirmed that MD and MT regressions with anthocyanins were significantly different ($P = 0.021$).

Wine composition

For wines, the alcohol content was significantly increased by MD in both seasons (regardless of the timing of defoliation) and for MT treatments only in 2007 (Table 4). Small differences in the acidity parameters were caused by both regulation techniques. MD led to wines more intensely coloured (41–54% enhancement of colour density) and of higher total phenol index (30% on average) than that of control wines in both seasons. In contrast, colour density and total phenol index were substantially improved by MT only in 2007. Comparing both techniques, the main difference in wine composition was alcohol content, which was significantly enhanced by MD treatments as compared with that of MT wines. Furthermore, colour and phenolic traits were superior in MD wines as compared with that of MT wines in 2008. The influence of timing of intervention on the wine composition was negligible and inconsistent over the two seasons. For MD, differences between preflowering and fruitset treatments were recorded in pH and total phenols in 2007. For the MT, higher pH and hue values, as well as lower colour density, were observed in wines of mechanically thinned vines at veraison with respect to MT at bunch closure, only in 2007. The covariate analyses on wine colour density and phenols data using alcohol content and berry weight as covariates did not reveal significant differences (data not shown).

Discussion

Yield components

Yield regulation was achieved by both mechanical techniques, although the effectiveness of MD appeared to be more consistent over the seasons. In previous studies (Poni et al. 2006, Intrieri et al. 2008, Tardaguila et al. 2010), early leaf removal either manually or mechanically performed was able to reduce

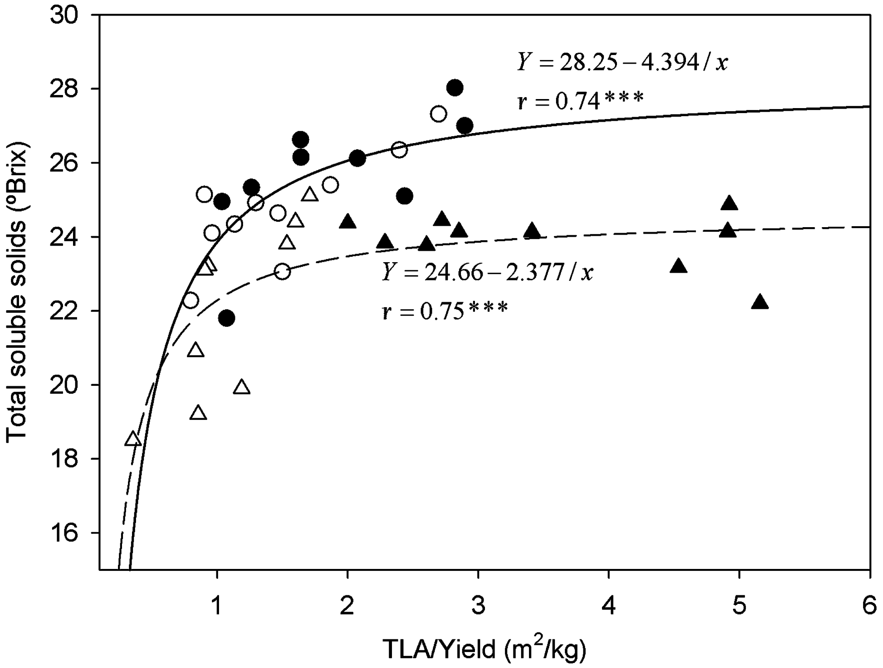

Figure 1. Regressions of total soluble solids (°Brix) of Tempranillo berry juice at harvest on total leaf area per yield ratio (TLA/Y, m^2/kg) for mechanical defoliation (MD, solid line) and mechanical thinning (MT, dashed line) in seasons 2007 and 2008. (●) MD in 2007; (○) MD in 2008; (▲) MT in 2007; (△) MT in 2008. Significance: *** $0.01 \leq P \leq 0.001$. MD and MT regressions were significantly different ($P <$ 0.001) using the Student *t*-test.

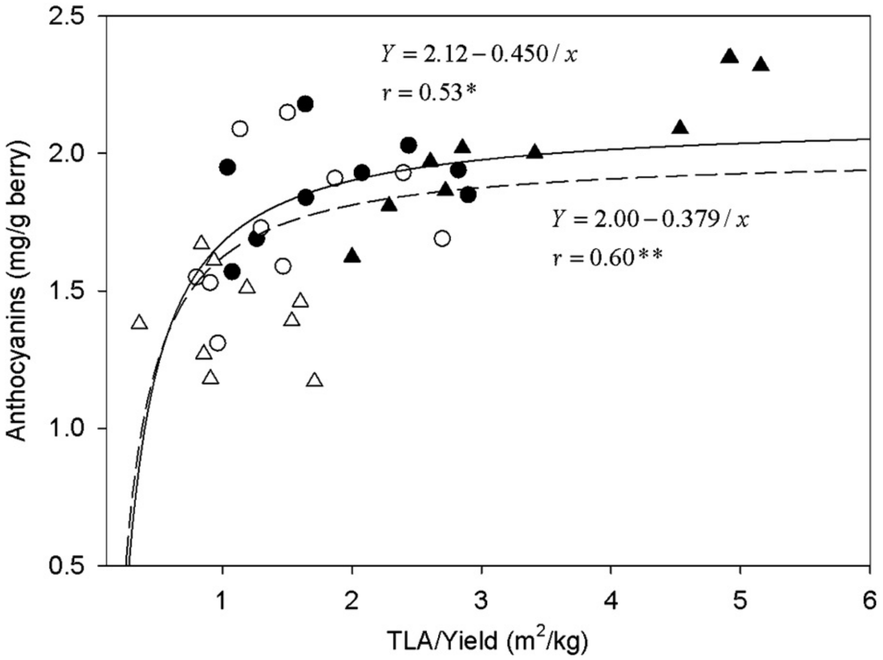

Figure 2. Regressions of anthocyanin concentration (mg/g berry) of Tempranillo berries at harvest on total leaf area per yield (TLA/Y) ratio (m^2/kg) for mechanical defoliation (MD, solid line) and mechanical thinning (MT, dashed line) in seasons 2007 and 2008. (●) MD in 2007; (○) MD in 2008; (▲) MT in 2007; (△) MT in 2008. Significance: * $P \leq 0.05$; ** $0.05 \leq P \leq 0.01$. MD and MT regressions were significantly different ($P =$ 0.021) using the Student *t*-test.

yield by approximately 30 and 50%, respectively. As for mechanical thinning, Petrie and Clingeleffer (2006) showed a substantial reduction in yield in the range of 25–45% in high-yielding Cabernet Sauvignon vineyards in a warm climate in Australia, whereas Tardaguila et al. (2008) were able to remove 35% of the fruit using a conventional harvester which caused canopy vibration in VSP Tempranillo and Grenache vineyards in Spain. Moreover, the potential risk associated with MD treatments prior to fruitset as compared with MT, where fruitset has been completed, should be considered. In this sense, MT seems to be a 'more conservative' practice than MD, as a preliminary estimation of the final yield is available prior to MT but not to MD, and therefore the viticulturist may decide whether to thin or not (i.e. crop reduction may not be required in years of poor fruitset, as well as the extent of the reduction). MT provides an opportunity to regulate the crop once the final crop potential is established after fruitset.

Overall, the number of bunches per vine was reduced by both crop management techniques, although different mechanisms may have occurred. MT led to the detachment of entire bunches by canopy vibration caused by the harvester, while in MD, the mechanical effect caused by the pressurised air jet of the leaf remover on the flowers, and possibly, the direct rubbing of the defoliator might have removed the majority of flowers of some bunches. The low speed of the leaf remover (0.5 km/h) together with two passages per row may have contributed to the detachment of all flowers or berries of some inflorescences, or bunches, during the pre-flowering or fruitset leaf removal treatments, respectively.

Both mechanical techniques modified bunch morphology, albeit through different means. A diminution in fruitset because of indirect effects of leaf removal could have caused a reduction in bunch size in MD treatments. Previous studies have suggested that carbohydrate supply at flowering can be a major determi-

Table 4. Influence of mechanical early defoliation (MD) at pre-flowering and fruitset, and mechanical thinning (MT), at bunch closure and veraison on Tempranillo wine composition in 2007 and 2008 of the trial ($n = 5$).

Treatment	Alcohol (% v/v)		pH		Titratable acidity (g/L tartaric acid)		Malic acid (g/L)		Colour density (AU)		Hue		Total phenol index (AU)	
Year	**2007**	**2008**	**2007**	**2008**	**2007**	**2008**	**2007**	**2008**	**2007**	**2008**	**2007**	**2008**	**2007**	**2008**
Control	13.1^{c}	12.5^{b}	3.75^{b}	3.82	6.87	5.23	3.73	3.04	17.4^{b}	10.1^{b}	0.46^{c}	0.68^{ab}	60.3^{c}	46.0^{b}
MD at pre-flowering	14.6^{a}	14.8^{a}	3.85^{b}	3.83	6.48	5.70	3.37	2.83	24.1^{a}	15.3^{a}	0.49^{bc}	0.76^{a}	71.7^{b}	60.5^{a}
MD at fruitset	14.8^{a}	13.6^{a}	4.08^{a}	3.91	6.36	5.22	3.71	3.03	24.9^{a}	15.9^{a}	0.52^{ab}	0.74^{a}	87.7^{a}	57.6^{a}
MT at bunch closure	13.9^{b}	12.8^{b}	3.72^{b}	3.94	6.72	4.96	3.31	2.83	23.9^{a}	9.3^{b}	0.44^{c}	0.61^{b}	77.1^{ab}	43.9^{b}
MT at veraison	13.7^{b}	13.0^{b}	4.10^{a}	3.88	6.19	5.29	3.67	2.76	20.2^{b}	9.8^{b}	0.53^{a}	0.57^{b}	78.1^{ab}	38.1^{b}
Analysis of variance *P*-values														
Treatment	0.005	0.013	<0.001	n.s.	n.s.	n.s.	n.s.	n.s.	<0.001	0.001	0.002	0.002	<0.001	0.022
Treatment year	0.010		0.001		n.s.		n.s.		0.007		0.002		0.039	

Mean values ($n = 25$) within columns designated by different superscript letters are significantly different by the Student–Newman–Keuls (SNK) test ($P < 0.05$). Values within columns without any superscript letter are not significantly different by the SNK test. AU, absorbance units; n.s., not significant.

nant of fruitset (Caspari et al. 1998, Poni et al. 2006). In a prior mechanical early defoliation trial on Sangiovese (Intrieri et al. 2008), calculations based on flower counts before and after machine runs indicated that about 50% of the decrease in set was a result of a physiological effect of leaf removal, whereas the other 50% of the decrease depended on a reduction in the number of flowers, which were usually cut off at the distal part of the rachis. In this trial, Intrieri et al. (2008) used a different type of leaf removal system (suction and cutting leaf plucking machine), and it was likely that this model of leaf remover may have had a larger impact on removal of inflorescences (or parts of them) than the type of machine used in our trial (pulsed air leaf remover). In the present study, flowers were not counted before and after machine passage, so the mechanical and physiological effect could not be separated. No carry-over effects on fruitset because of MD were observed in a companion study performed in Tempranillo and other varieties, as the number of flowers per inflorescence was not different between non-defoliated and defoliated vines in the previous season (Diago 2010). In contrast, it is likely that the reduced berry number per bunch in MT was because of a direct mechanical effect caused by bunch shaking, leading to some berry dropping and/or causing abscission of part of a bunch. Tardaguila et al. (2008) also observed this phenomenon in a mechanical thinning trial based on vine vibration (using a conventional harvester) in Tempranillo and Grenache VSP vineyards. In contrast, Petrie and Clingeleffer (2006) adjusted the machine to remove fruit from the upper and lower parts of the canopy of minimal pruned vines and did not hit all the fruit zone, hence, retained bunches were not affected by the treatment.

In general, reduced berry number per bunch in MD and MT led to 'looser' bunches. This was an important outcome as Tempranillo is prone to yield large and tight bunches even in dry Spanish areas (Tardaguila and Diago, unpublished data). The benefits on botrytis incidence were evident in a wet season (2008), with abundant rain throughout berry formation (102 mm in June and July, data not shown). Similar findings on the reduction in botrytis incidence were observed in mechanically thinned vines (Tardaguila et al. 2008) and defoliated vines (Tardaguila et al. 2010) under Mediterranean climate. It is important to highlight that botrytis incidence in MT vines was significantly lower than that of control, despite the partial damage of some berries of the bunches in these treatments and the fact that no fungicide sprays for botrytis control were applied in either treatment. The reduction of botrytis in MT vines could be explained by a trend towards reduced berry number per bunch despite the lack of variation in compactness (the bunch compactness, determined at harvest only, may have been affected by the increase in berry weight in 2008).

Berry size tended to increase following MT operations, whereas MD had little effect on berry development. These differences reflect changes of source availability as early leaf area limitation overlapping with cell division during stage I of berry growth is quite often conducive to smaller final berry size (Petrie et al. 2000, Poni et al. 2009). Conversely, despite being performed later in the season, MT did not affect leaf area availability while still allowing post-veraison berry growth compensation.

In the present MT trial, bunch and/or berry detachment were caused by vibration, not by direct hitting of the fruiting area, as applied by Petrie and Clingeleffer (2006) in Australia. In our study, most of the final harvest yield was represented by undamaged bunches. The proportion of completely dry bunches per vine was low, and during mechanical harvesting, the dry berries did not detach because of their low weight (data not shown).

Total leaf area per vine was not changed by MT, indicating that the harvester did not remove a significant portion of leaves and that bunch microclimate was not significantly altered in MT vines. Because of leaf growth compensation responses (Poni et al. 2006), MD did not affect final TLA, yet the bunch exposure and canopy porosity may have substantially improved during berry formation and the ripening period. The leaf recovery capacity of the vine, triggered by MD, was quite significant in spite of the severe defoliation conducted by the mechanical leaf remover. In fact, in a companion study, the proportion of total leaf area removed by the same model of leaf remover in VSP vineyards was 95% and 60% (estimated at the time of MD) at pre-flowering and fruitset, respectively, with respect to that of the control (Diago 2010).

The final leaf-to-fruit ratio expressed as total leaf area/yield increased or did not change in both MD and MT vines as a combined effect of no major changes in final total leaf area and a significant yield reduction. Similar findings were reported by Tardaguila et al. (2008) in mechanical thinning trials on Tempranillo and Grenache, and by several other authors in early defoliation studies on different varieties (Poni et al. 2006, 2009, Intrieri et al. 2008).

Yield interseasonal discrepancies in both MD and MT treatments could be explained by differences in machinery adjustments rather than the variation in climatic conditions in both seasons. Concerning MD, the significant year × treatment interaction revealed opposite results between pre-flowering and fruitset. Probably, the effect of MD at fruitset in 2007 was overexpressed (high mechanical abscission by the blowing machine on the recently set berries). The results in previous early defoliation studies have shown larger yield reduction in pre-flowering than in fruitset defoliated vines (Poni et al. 2006, Tardaguila et al. 2010). Regarding MT, large differences in yield reduction between both seasons were observed because of the ineffectiveness of the treatment in 2008. These results suggest that more research is needed on machinery set-up for effective crop regulation.

Grape and wine composition

The yield decrease and the subsequent changes in the leaf-to-fruit ratio (TLA/Y) caused by MD and MT seem to have played an important role in the enhancement of the total soluble solid content of berries, hence, in wine alcohol concentration. Likewise, the larger total soluble solid values observed at any TLA/*Y*-value in MD as compared with MT suggest the influence of additional mechanisms on the berry sugar accumulation in early defoliated vines. In this regard, Poni et al. (2006) and Palliotti et al. (2011) described an increase of the photosynthetic capacity of the 'younger' (main leaves above defoliated nodes and lateral leaves) canopies, as well as the hastening of the translocation of assimilates towards the bunch in early defoliated vines. As a result, berry ripening rates were enhanced from veraison onwards as compared with that of the control (non-defoliated) vines of Sangiovese (Palliotti et al. 2011). In MT, an advanced ripeness rather than the increase in the fruit sugar accumulation rate was postulated by Petrie and Clingeleffer (2006). The lack of impact of the MT treatment on yield in 2008 seems to have driven the lack of effect on the total soluble solids and alcohol concentration in must and wine, respectively, as compared with that of control and MD treatments.

A larger anthocyanin concentration in berries was observed for MD than for MT for a given TLA/*Y*-value. This outcome seems to highlight the importance of other factors, not affected by MT treatments, in the berry accumulation of these pigments. Basal defoliation alters bunch microclimate, whereas no changes are caused by mechanical thinning. While no seasonal monitoring of bunch microclimate was conducted in this study, enhanced bunch exposure and canopy porosity were recorded 1 week prior to harvest in early defoliated vines as compared with un-defoliated vines (Tardaguila et al. 2010) in a companion study. Larger and improved sun exposure favours the accumulation of phenolic compounds in the berries, mainly anthocyanins and flavonols (Price et al. 1995). Moreover, the increase in anthocyanin and phenol concentration in the berries of MD or MT vines as compared with that of control berries seems to occur irrespective of berry size and ripeness level (total soluble solids), as confirmed by covariate analysis. These results indicate that the increase in grape anthocyanins and phenols was mostly explained by physiological factors rather than advanced ripeness. Similar findings were observed in previous mechanical thinning (Diago et al. 2010a) and defoliation studies (Poni et al. 2009, Tardaguila et al. 2010). Regardless of berry size, a consistent enhancement of berry anthocyanins was observed, as well as the increase of the relative skin mass in berries of vines manually defoliated at pre-flowering involving Barbera (Poni et al. 2009) and Sangiovese varieties (Palliotti et al. 2011). From these results, it can be inferred that the method conducive to yield control seems to have a stronger influence in the berry compositional response than yield change per se.

Wine colour and phenolic composition are intrinsically related to anthocyanins and other phenols in grapes. In this way, the wine colour density and total phenol index enhancements observed for MD and MT treatments were reflecting the increase in anthocyanins and total phenols in the berries. The removal of leaves in the basal area around the bunches, applied at different timings, also brought about more intensely coloured wines (Staff et al. 1997, Tardaguila et al. 2010, Palliotti et al. 2011) in diverse varieties. Price et al. (1995) also observed a higher total phenol concentration in Shiraz wines made from defoliated vines, because of the better fruit exposure. In addition to this environmental effect, it has also been reported that ethanol facilitates the extraction of anthocyanins and other phenolic compounds, such as proanthocyanides, from the grapes into the fermenting must (Canals et al. 2005).

Between the two mechanical techniques, their effect on berry composition was largely reflected in the wines. In contrast, the significant improvement of berry and wine composition of MT treatments suggests that the partial damage of some bunches (partially dried bunches) did not play a negative role in grape and wine composition.

Conclusions

Mechanical early defoliation and mechanical crop thinning proved to be effective to regulate grape yield in grapevine, albeit through different mechanisms. Mechanical early defoliation appeared to be more consistent than MT.

Grape and wine composition improved in vines following both mechanical yield management techniques, however, mechanical early defoliation appeared to give better results than mechanical thinning at any given crop load. A broad time window, from pre-flowering to veraison, is open to viticulturists aiming to regulate grape yield by mechanical operations.

Field conditions, machinery adjustments and the skills and expertise of the machine operator can affect the effectiveness of mechanised yield management. Differences in any of these factors may lead to either insufficient or overexpressed effects on the vines, resulting in differential responses for a given mechanical practice.

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