



PHYSIOLOGICAL RESPONSE TO EARLY LEAF REMOVAL OF CV. VERDEJO IN RAINFED CONDITIONS, THROUGHOUT THE DAY, IN THE D.O. RUEDA (SPAIN)

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Abstract

Aim: Early leaf removal, generally applied before flowering, is mostly conceived as a technique to control grape yield and improve the health of grapes and focused on the final objective of increasing wine quality. New knowledge of its possible physiological effects in the cv. Verdejo, in rainfed conditions, should facilitate the understanding of agronomic, vegetative and qualitative behaviour of the vineyard, thus generating more possibilities of adaptation to optimize the grape ripening process.

Methods and Results: Leaf removal was carried out by removing the first eight adult leaves, from the base, on all shoots. The trial was carried out with cv. Verdejo, grafted onto 110R, planted in 2006 and trained on a vertical trellis, in rainfed conditions, in the D.O. Rueda. Throughout the period of 2016-2018, the physiological response of the vines to early leaf removal (before flowering) was studied through measurements of water potential at 7, 9, 11 and 12 hours (solar time) and stomatal conductance, transpiration and net photosynthesis at 9 and 12 hs. The water potential measured at different times of the day showed no differences between treatments. The values were slightly higher in the control vines sometimes and higher in the leaf plucked vines other times, but more frequently favourable to the control vines in 2017 and 2018, especially in the measurements at 9 and 11 hs. Gas exchange (G_s , E , A_n) also did not show statistically significant differences between treatments. Some values were slightly favourable to the leaf removal treatment, such as at 9 hs in 2016, and other values were slightly favourable to the control treatment, such as at 12 hs in 2017 and at 9 hs in 2018.

Conclusions: The results observed in the water potential and in the gas exchange at different times of the day have not generally discriminated between the leaf removal applied at the beginning of flowering and the control treatments.

Significance and Impact of the Study: The agronomic benefit intended with the early leaf removal, generally to lighten the compactness and weight of the bunch, as well as its aeration and luminosity, does not have to be questioned from the water or physiological point of view in the cv. Verdejo on rainfed conditions.

Keywords: Grapevine, photosynthesis, stomatal conductance, transpiration, water potential

Introduction

Early leaf removal, generally applied before flowering, has been conceived mainly as a technique to control yield and improve the health of the grapes, with the final objective of increasing wine quality. The immediate effect of early leaf removal is essentially based on the functional relationship between the productive potential and the availability of sugars at the time of flowering (Diago, 2010).

Studies on early defoliation were initially carried out in red varieties, such as Sangiovese (Poni *et al.*, 2006), and they showed that the elimination of four to six leaves from the basal part of the main shoot at flowering limits fruit set or the initial development of the berry, and therefore its final size, giving rise to bunches of lower weight and less compact, of higher quality, thanks to the increase in the concentration of sugars, anthocyanins and total polyphenols (Poni *et al.*, 2006). In addition, this green intervention reduces the incidence of *Botrytis cinerea* and other fungal diseases, generating better sanitary status of the grapes (Smith and Centinari, 2019). This practice has been the subject of various studies in different red varieties and wine-growing areas, having achieved variable results depending on the conditions in which the trials were carried out. Diago (2010) observed that early defoliation in the varieties Tempranillo, Graciano and Mazuelo significantly reduced fruit set, bunch weight and grape yield per vine, obtaining looser bunches, with fewer berries of less or equal weight but with a higher skin / pulp ratio. The vines showed a compensatory vegetative growth, maintaining and even increasing the leaf / fruit ratio, but with higher porosity of the canopy and better exposure of the bunches. The grape improved in terms of technological and phenolic maturity and organoleptic quality.

The magnitude of the effects in the vineyard can vary according to the variety and the conditions of leaf removal application (phenological stage, intensity) and cultivation, as various studies have shown (Arrillaga, 2017; López *et al.*, 2018; Poni *et al.*, 2009; Rodríguez *et al.*, 2015; VanderWeide *et al.*, 2020).

The application of early defoliation in white varieties has been studied less frequently, focusing mainly on the impact on the aromatic potential of the grape (Vilanova *et al.*, 2018). In La Rioja, the works have been developed with White Tempranillo (García-Escudero *et al.*, 2011), as well as Viura and Alarije (Martínez *et al.*, 2016), observing that this practice allows the control of the productive potential through the reduction of the berry number per bunch and berry weight, that is, limiting bunch weight. Likewise, an increase in the concentration of sugars, pH and potassium was found, without significantly affecting the level of total acidity. However, these authors draw attention to the inconvenience that the possible reduction in acidity may entail in varieties with low acidity potential, especially through malic acid, and the increase in pH.

The studies carried out on early leaf removal have barely emphasized the physiological effects, and the water relations associated with them, which can be derived from its application. Poni *et al.* (2008) made an evaluation of the gas exchange in the Sangiovese variety, showing a global reduction in the net photosynthesis rate in the leaf plucked vines compared to the non-leaf plucked vines, but an increase in rates per unit of leaf area in the vines subjected to leaf removal, thus justifying the higher concentration of carbohydrates available for the ripening of the grapes in these vines. The knowledge of the physiological foliar response can facilitate the understanding of the agronomic functioning of the vineyard and its vegetative and qualitative repercussion, thus generating more possibilities for adaptation in order to optimize the ripening process of the grape.

Taking into account the possibilities that early defoliation can offer as a tool for controlling yield, as well as for the sanitary improvement of the vineyard, in the absence of previous specific works, its application in the white Verdejo variety was considered, under rainfed cultivation. The work was proposed in the D.O. Rueda, located in the center of the Duero river valley (Castilla y Leon, Spain), with the aim of evaluating its possible physiological effects, through measures of water potential and gas exchange (G_s , E and A_n) at different times of the day, as a basic aspect to deepen the study of the global response of the vineyard.

Materials and Methods

The trial was conducted over the period 2016-2018 in Medina del Campo (Valladolid, Spain), in a vineyard belonging to the Grupo Yllera S.L. winery, located within the D.O. Rueda, in the center of the Castilla y Leon region (41°21'02"N and 4°56'16"O). The vines, planted in 2006, are from cv. Verdejo, grafted onto 110R. The vineyard cultivation was under rainfed conditions. The vine distances were 2.60 m x 1.25 m (3,077 vines/ha). The vines were trellis trained, with bilateral Royat cordon and vertical positioning of the vegetation (VSP). Row orientation was NNO (N-25°). The pruning load was 16 buds per vine, with spurs of two buds. A green pruning operation was applied every year, after the period of risk of spring frost, for the adjustment of the shoot load

per vine. The soil of the trial is deep, with a clay upper horizon, and has a slight slope in direction East to West and good drainage in general.

An early basal leaf removal or defoliation treatment (D) was applied at the beginning of the flowering phase, suppressing the first eight adult leaves, from the base, of all shoots, for comparison with a control treatment (T). The experimental design of the trial was in random blocks, with four repetitions of 20 control vines per plot.

Measurements of water potential were carried out with a Scholander pressure chamber at least once every two weeks, at 7 hs (solar time) on two adult leaves in the shadow, 9 and 11 hs, on two adult leaves exposed to the sun, in two different vines per replication, also at 12 hs for stem water potential. Similarly, gas exchange measurements (Gs, E, An) were done through an IRGA portable instrument (Li-COR 6400, USA) at 9 and 12 hs. The average seasonal data of temperature, ETo and rainfall, for the period 2016-2018, is detailed in table 1. The statistical analysis was carried out at each date and time of measurement by ANOVA ($p < 5\%$).

Table 1: Data of temperature, ETo and rainfall of seasons 2106, 2017 and 2018, corresponding to the annual period October (previous year) – September (season year), registered in Medina del Campo (Valladolid). **Tm:** average temperature ($^{\circ}\text{C}$), **Tmax:** maximum temperature ($^{\circ}\text{C}$), **Tmin:** minimum temperature ($^{\circ}\text{C}$), **ETo** 1-apr/30-sep (mm), **P:** precipitation (mm), **Pa:** Oct 1 / Sep 30, **Pc:** Apr 1/ Sep 30, **Pv:** Jul 1/ Sep 30.

	Tm	Tmax	Tmin	ETo	Pa	Pc	Pv
2016	12.8	19.1	6.9	830	318	93	0.2
2017	13.2	20.2	6.5	970	167	89	43.0
2018	12.4	19.3	6.0	843	323	139	3.4

Results and Discussion

Leaf Water Potential in 2016

The water potential in 2016, at 9, 11 and 12 hs, showed similar values between treatments. It could only be mentioned that the stem water potential values, at 12 hs, were somewhat less negative in the leaf removal treatment than in the control one in the 2nd week of August and the 2nd of September, as well as the values of leaf water potential at 11 hs in the 2nd week of August. On the contrary, the values were somewhat less negative in the control than in the leafless treatment in the stem water potential at 12 hs in the 3rd week of July and in the leaf water potential at 9 hs in the 3rd week of July and the 2nd of September, without the differences being notable in practically any case.

Leaf Water Potential in 2017

The measures of water potential in 2017 did not show statistical differences between treatments either. However, some values slightly favourable to the control treatment with respect to the leaf removal treatment were observed at 7 and 12 hs from the 3rd week of July to the last measurement date, in the 2nd week of September. At 9 and 11 hs, the leaf water potential hardly showed any slightly less negative value in the control treatment between the 3rd week of July and the 2nd of August, while the leaf removal treatment only presented a slightly less negative value at 12 hs in the 1st measurement, in the 1st week of July.

Leaf Water Potential in 2018

The water potential in 2018 did not show significant differences between treatments. Only in the 2nd week of September and in the 1st of October slightly higher values were observed in the control than in the leaf removal treatment from 9 hs, as well as, exceptionally, at 12 hs in the 1st week of July. The only measurement that showed a slightly higher value in the leaf removal treatment than in the control one was at 12 hs in the 1st week of August. In all cases, the differences between water potential values were small.

Gas Exchange in 2016

Gas exchange (Gs, E, An) measured at both 9 and 12 hs in 2016 did not show different values between the leaf removal and control treatments, except for the measurement at 9 hs in the 2nd fortnight of August, in which the Gs and E rates were statistically favourable to leaf removal treatment. At 9 hs, the values of the leaf removal treatment were generally slightly higher than those of the control one, with the opposite exception of the measurement in the 1st fortnight of August. However, at 12 hs, the physiological rates were only slightly higher in the leaf removal treatment than in the control one in the 2nd half of August, contrary to the two previous measurements, the differences between values being of little magnitude.

Gas Exchange in 2017

The gas exchange measurements (Gs, E, An) carried out at both 9 and 12 hs in 2017 did not show differences between treatments, even though at 9 hs the leaf removal treatment showed slightly higher values than the control in the 2nd fortnight of August and slightly lower in the 2nd fortnight of July. At 12 hs somewhat higher values were found, especially of Gs and An, in the control than in the leaf removal treatment in the two measurements carried out in August, being the differences between values of little magnitude.

Gas Exchange in 2018

The gas exchange rates (Gs, E, An) measured at both 9 and 12 hs in 2018 did not show differences between treatments either. At 9 hs the control showed a higher frequency of slightly higher values than the leaf removal treatment, while at 12 hs a definite trend was not observed, since the Gs rate more frequently presented slightly higher values in the leaf removal treatment than in the control one, while the An rate presented the opposite response. In any case, the differences observed between values were small.

Conclusions

The water potential, measured at different times of the day with different measurement modalities, did not show significant differences between treatments in the rainfed conditions of cultivation. The application of early leaf removal caused some values slightly higher or lower than the control vines changing sign between both treatments throughout the dates, with more frequency of slightly less negative values in the control treatment in 2017 and 2018, especially in the measurements made at 9 and 11 hs.

The effect of early leaf removal on gas exchange rates, both at 9 and 12 hs, also did not show differences with respect to control vines, although some slightly different values were observed between treatments, which sometimes were mostly favourable to the leaf removal treatment, as at 9 hs in 2016, and other times to the control one, as at 12 hs in 2017 and at 9 hs, especially in terms of Gs, in 2018. In any case, the differences in gas exchange values were of little magnitude, not being able to discriminate in general leaf plucked vines from the control vines.

Therefore, the possible agronomic utility pursued with leaf removal, generally to lighten the compactness and weight of the bunch, as well as its aeration and luminosity, does not have to be questioned from the hydric or physiological aspects in the cv. Verdejo grown in rainfed conditions.

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Appendix

Table 2: Water potential (MPa) throughout the period 2016-2018, of Control (T) and Leaf removal (D) treatments, according to the following types and hours (solar time) at each date (week of the month): 7 h in the shadow (Ψ_s); 9 h (Ψ_f) and 11 h (Ψ_f) to the sun; 12 h on the stem (Ψ_s). Statistical significance: $p < 0.05 = *$.

	Date	2016 - 7 h		2017 - 7 h		2018 - 7 h		2016 - 12 h		2017 - 12 h		2018 - 12 h	
		T	D	T	D	T	D	T	D	T	D	T	D
Ψ_s	w1. Jul			-0.88	-0.92	-0.16	-0.16			-0.91	-0.86	-0.30	-0.37
	w3. Jul			-0.56	-0.64	-0.34	-0.34	-0.80	-0.86	-1.00	-1.04	-0.64	-0.64
	w4. Jul									-0.97	-1.02		
	w1. Aug			-0.86	-0.92	-0.45	-0.45					-0.74	-0.71
	w2. Aug			-0.87	-0.93	-0.47	-0.48	-1.05	-1.00	-1.33	-1.33	-0.89	-0.88
	w3. Aug			-1.15	-1.19								
	w4. Aug			-1.14	-1.22	-0.55	-0.55	-1.22	-1.22	-1.53	-1.58	-1.19	-1.17
	w5. Aug					-0.80	-0.82					-1.22	-1.29
	w2. Sep			-0.90	-0.94	-0.63	-0.64	-1.47	-1.39	-1.35	-1.42	-1.33	-1.33
	w3. Sep							-1.54	-1.55				
w1. Oct					-0.81	-0.80	-1.64	-1.66			-1.28	-1.32	
	Date	2016 - 9 h		2017 - 9 h		2018 - 9 h		2016 - 11 h		2017 - 11 h		2018 - 11 h	
		T	D	T	D	T	D	T	D	T	D	T	D
Ψ_f	w3. Jul	-1.24	-1.27	-1.05	-1.16	-0.68	-0.68	-1.24	-1.24			-0.92	-0.90
	w4. Jul									-1.19	-1.26		
	w2. Aug	-1.30	-1.30	-1.43	-1.46	-1.03	-1.03	-1.36	-1.32	-1.50	-1.55	-1.16	-1.16
	w4. Aug	-1.62	-1.60	-1.65	-1.65			-1.48	-1.47	-1.74	-1.72		
	w5. Aug					-1.42	-1.40					-1.39	-1.38
	w2. Sep	-1.41	-1.44	-1.47	-1.43	-1.41	-1.46			-1.59	-1.58	-1.54	-1.58

Table 3: Stomatal conductance, G_s ($\text{mmol H}_2\text{O.m}^{-2}.\text{s}^{-1}$), Transpiration, E ($\text{mmol H}_2\text{O.m}^{-2}.\text{s}^{-1}$) and Net photosynthesis, A_n ($\mu\text{mol CO}_2.\text{m}^{-2}.\text{s}^{-1}$), at 9 and 12 hs (solar time), throughout the period 2016-2018, of control (T) and leaf removal (D) treatments. Statistical significance: $p < 0.05 = *$.

	Date	2016 - 9 h		2017 - 9 h		2018 - 9 h		2016 - 12 h		2017 - 12 h		2018 - 12 h	
		T	D	T	D	T	D	T	D	T	D	T	D
Gs	w2. Jul	104.6	107.0	87.9	74.5	194.4	182.0	50.1	47.2	21.7	21.4	112.5	105.8
Gs	w1. Aug	80.1	69.0	45.9	46.9	126.0	109.8	46.3	40.3	41.4	34.7	72.4	62.0
Gs	w2. Aug	2.1	12.6*	28.9	33.6	71.1	58.6	5.4	6.4	19.4	18.2	41.2	38.1
Gs	w1. Sep					58.7	63.2					36.7	42.3
Gs	w2. Sep	15.1	31.2										
E	w2. Jul	3.80	4.04	2.60	2.44	5.06	5.56	2.87	2.83	1.16	1.20	4.50	4.85
E	w1. Aug	2.06	1.78	1.22	1.26	3.35	3.08	1.72	1.61	1.31	1.21	2.70	2.52
E	w2. Aug	0.08	0.49*	1.16	1.47	1.94	1.69	0.35	0.44	0.89	0.90	1.54	1.53
E	w1. Sep					1.70	2.01					1.46	1.81
E	w2. Sep	0.34	0.67										
An	w2. Jul	13.6	14.7	14.4	13.3	17.6	17.2	9.3	8.0	5.2	4.7	11.9	12.6
An	w1. Aug	10.8	9.5	11.7	12.5	15.1	14.6	8.3	7.8	11.6	9.8	9.4	8.5
An	w2. Aug	5.2	4.4	7.6	9.4	13.0	11.7	2.6	3.0	6.3	5.4	8.5	10.0
An	w1. Sep					12.6	12.3					7.9	9.2
An	w2. Sep	6.3	8.6										