Response of two grapevine (Vitis vinifera L.) Portuguese varieties Tinta Roriz and Touriga Nacional to different irrigation regimes in the Douro Region, Portugal

António Castro Ribeiro, David Santos Barreales and João Verdial Andrade

Centro de Investigação de Montanha (CIMO), Escola Superior Agrária, Instituto Politécnico de Bragança, Campus Sta Apolónia, 5300-253 Bragança, Portugal.

Abstract. The effect of several deficit irrigation regimes on vine water status, grape yield and quality parameters were studied in two Portuguese cultivars, Tinta Roriz (2007 and 2008 growing seasons) and Touriga Nacional (2014 and 2015 growing seasons) (Vitis vinifera L.) grown in a commercial vineyards located in the Douro region, Portugal. Treatments consisted of non-irrigated vines and three deficit irrigation regimes with a constant fraction of reference evapotranspiration (ETo): 0.2, 0.4 and 0.6. The reference evapotranspiration was calculated using modified FAO Penman-Monteith equation and water was applied three times a week, from pre-veraison until one week before harvest, through a drip irrigation system. The results showed that moderate water supplies during ripening period, for the region where the study was conducted (severe water deficits), improved significantly the grapevine water status, leaf photosynthesis and transpiration in both cultivars. Yield components and pruning weights showed a significant increase in irrigated treatments with more water supplied. There were no significant differences in the majority of fruit quality parameters. However, the total phenols and the colour intensity showed a tendency to decrease in irrigated treatments with more water supplied.

1 Introduction

The vineyards located in regions with seasonal drought (e.g. climate of the Mediterranean type), where soil and atmospheric water deficits, together with high temperatures, have significant constraints in yield and quality [1]. In the hot and dry Douro Region, limitations in water supply have a great impact on grape production as the annual rainfall is not adequate to provide grapevines with their water requirements, and water deficits usually develop gradually during summer causing important crop losses [2].

Irrigation is commonly used to stabilize yield and maintain or improve grape quality in many wineproducing regions in the world that experience seasonal drought [3]. Regulated Deficit Irrigation (RDI) is one of the most frequently used irrigation strategies in vineyards with the aim to balance grapevine vegetative and reproductive growth by applying less than the full vineyard water use at specific periods of the growing season [4,5]. However, successful strategies may vary among regions with different climates and can even be site specific, depending on the interactions within the grapevine variety, soil type and vineyard management practices. The objective of this study was to determine the effect of different irrigation amounts in physiology, yield and grape composition of two Portuguese grapevine varieties (Tinta Roriz and Touriga Nacional), growing in Douro region, NE of Portugal.

2 Materials and methods

This study was conducted over two consecutive years for each variety. The experiment in 'Tinta Roriz' vineyard was carried out in 2007 and 2008 growing seasons and the 'Touriga Nacional'experiment was carried out in 2014 and 2015 growing seasons. The trials were located in a commercial vineyards, located in Douro region, Portugal (Tinta Roriz' vineyard: lat. 41°11' N; long., 7°6' W, elevation 116 m; 'Touriga Nacional': lat. 41°31' N; long., 07°05' W, elevation 326 m).

In both experiments the water was supplied (I) according to the reference evapotranspiration (ETo) using the following equation: I= (K ETo - P), where P represents effective rainfall and K a constant coefficient. Three irrigation treatments were established for the 'Tinta Roriz' experiment: T0 was rain-fed control; T1 was irrigated with a constant fraction of the ETo (K=0.2) and T2 was irrigated with a constant fraction of the ETo (K=0.4). In the experiment with 'Touriga Nacional' was imposed a third treatment with a 0.6 fraction of ETo. The reference evapotranspiration was calculated using modified FAO Penman-Monteith equation [6].

Each treatment had four replicates in a randomized complete block design. Each plot consisted of four rows with six vines per row and the surrounding perimeter vines were used as buffers.

The beginning of water supplied was determined by the threshold value (-0.4 MPa) of pre-dawn leaf water potential [2] and the frequency of water applications was the same for all treatments and varied from 2 to 3 days per week applied continuously until harvest. The dates of first and last irrigation and total water applied for the three treatments are shown in Table 1.

Climatic data were automatically collected from a weather station located near the vineyard. Fig. 1 shows the monthly rainfall and the mean air temperature at the experimental sites during the growing seasons.

Vine water status was monitored using a pressure chamber (Model 1000, PMS Instrument Company, Albany, USA) according to the method of Scholander et al. [7]. Stem water potential was measured in four fully expanded leaves, per plot (16 per treatment) of four representative plants.

Leaf gas-exchange rates were measured using a portable gas exchange system (LCA-4, Analytical Development Co., Hoddesdon, England). Measurements were performed in eight fully expanded leaves per treatment.

At harvest, yield components were assessed, following manual harvesting and weighing the production on-site. Number of clusters and yield per vine were recorded for 12 vines in each plot. Three 100-berry samples per treatment were previously collected. Samples were put into plastic bags, placed in a portable cooler and taken to the laboratory. They were weighed immediately and processed to determine berry composition following the procedures of OIV 1990[8] for each parameter.

At winter, pruning weight per vine was recorded and crop load (yield/pruning weight) was calculated.

Statistical data analysis was performed by analysis of variance. Tukey HSD tests were carried out to determine the significance of differences between treatments means, using JMP®11.0.0 2013 (SAS Institute Inc.).

 Table 1. Dates of first and last irrigation and total water applied for the three treatments..

Growing season	Dates		W	ater app (mm)	lied	Rainfall (mm)
	First irrigatior	Last irrigation	T1	T2	T3	
2007	19/Jul	04/Sep	59.6	119.2	-	265.0
2008	14/Jul	08/Sep	76.7	153.4	-	163.0
2014	25/Jun	05/Sep	70.3	140.6	211,0	192.8
2015	15/Jul	11/Sep	65.2	130.3	195,5	172.2

3 Results and discussion

3.1. Climate conditions and vine water status

Precipitation varied considerably during the four growing seasons studied. The rainfall in May and June of 2008

was very low and the summer exceptionally dry. The year of 2015 was very dry during winter and spring months and the summer extremely dry. Seasonal temperatures and reference evapotranspiration were within 10% of the 30-year sites average in each study year.

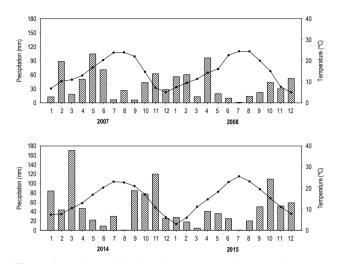


Figure 1. Total precipitation (bars) and monthly mean air temperature (line) at the experimental vineyards during 2007, 2008, 2014 and 2015 growing seasons.

Seasonal course of stem water potential, for the different treatments and growing seasons, is shown in Table 2 Stem water potential of non-irrigated vines (T0) showed a progressive decline during the ripening period. The lower values ($\Psi_{stem} < -1.4$ MPa) are indicative of a relatively severe water [9,10]. Irrigated vines showed a slightly decrease of Ψ_{stem} throughout the ripening. The values of Tinta Roriz vines, in the irrigation treatments, were indicative of moderated to weak water stress.

Table 2. Seasonal evolution of stem water potential for the three treatments during 2007 and 2008 growing seasons.

Year	Т	Stem water potential (MPa				
		09/08	23/08	06/09		
2007	T0	-1.25 ^b	-1.59 ^b	-1.41 ^b		
	T1	-1.01 ^a	-1.07 ^a	-1.10 ^a		
	T2	-0.95 ^a	-0.89 ^a	-0.95 ^a		
	Sig.	***	***	***		
		31/07	21/08	10/09		
2008	T0	-1.58 ^a	-1.64 ^b	-1.80 ^b		
	T1	-1.20 ^a	-1.03 ^a	-1.03 ^a		
	T2	-1.02 ^b	-0.84 ^a	-0.80 ^a		
	Sig.	***	***	***		

Means within a column, for each season, flanked by se same letter are not significantly different at $P \le 0.05$ (Tukey HSD test). Significance of difference between treatments: ns – not significant; * 0.01< P \le 0.05; ** 0.001< P \le 0.01; *** P \le 0.001

The non-irrigated 'Touriga Nacional' vines showed, in both growing seasons, a severe water stress during the ripening period. The Ψ_{stem} values of T2 and T3 irrigated treatments stabilize during ripening period with a thresholds indicating a moderate to weak water stress. In both varieties we observed that T1 vines maintained, during ripening period, Ψ_{stem} values above the severe water stress threshold (Table 3).

Year	Т	Stem water potential (MPa)						
		15/07	22/07	25/08	08/09			
2014	T0	-1.18 ^c	-1.20 ^c	-1.65 ^c	-1.73 °			
	T1	-1.11 ^{bc}	-1.11 ^{bc}	-1.43 ^b	-1.37 ^{ab}			
	T2	-1.01 ^{ab}	-1.00 ^{ab}	-1.11 ^a	-1.21 ^b			
	T3	-0.96 ^{ac}	-0.93 ^a	-1.03 ^a	-1.11°			
	Sig.	***	***	***	***			
		28/07	11/08	25/08	08/09			
2015	T0	-1.27°	-1.52 ^b	-1.52 ^b	-1.49 ^a			
	T1	-0.99 ^b	-1.18 ^a	-1.16 ^a	-1.21 ^b			
	T2	-0.81 ^a	-1.10 ^a	-1.09 ^a	-1.21 ^b			
	Т3	-0.82 ^a	-1.06 ^a	-1.06 ^a	-1.06 ^c			
	Sig.	***	***	***	***			

Table 3. Seasonal evolution of stem water potential for thethree treatments during 2014 and 2015 growing seasons.

Means within a column, for each season, flanked by se same letter are not significantly different at $P \le 0.05$ (Tukey HSD test). Significance of difference between treatments: ns – not significant; * 0.01< P \le 0.05; ** 0.001< P \le 0.01; *** P \le 0.001

3.2. Net CO₂ assimilation and transpiration rates

The results showed that the water availability affected significantly the transpiration rate and net CO2 assimilation rate. Irrigation treatments induced highly significant differences in these physiological parameters in both varieties (Tables 4 to 7).

Table 4. Transpiration rate (E) measured during hot and cleardays in the ripening period for the different water treatments in2007 and 2008 growing seasons.

Year	Treatment	E (mmol m ⁻² s ⁻¹)		
		23 Aug	06 Sep	
2007	Т0	1.21ª	1.23ª	
	T1	2.28 ^b	2.46 ^b	
	T2	4.30 ^c	4.32 ^c	
	Sig.	***	***	
		21 Aug	10 Sep	
2008	TO	1.47 ^a	1.40 ^a	
	T1	4.46 ^b	4.16 ^b	
	T2	7.62 ^c	5.57 ^b	
	Sig.	***	***	

Means within a column, for each season, flanked by se same letter are not significantly different at $P \le 0.05$ (Tukey HSD test). Significance of difference between treatments: ns – not significant; * 0.01< P \le 0.05; ** 0.001< P \le 0.01; *** P \le 0.001

Table 5. Net CO2 assimilation rate (A) measured during hot and clear days in the ripening period for the different water treatments in 2007 and 2008 growing seasons.

Year	Treatment	A (µmo	l m ⁻² s ⁻¹)
		23 Aug	06 Sep
2007	Т0	2.66 ^a	2.34 ^a
	T1	5.51 ^b	4.80 ^b
	T2	9.68 ^c	8.19 ^c
	Sig.	***	***
		21 Aug	10 Sep
2008	Т0	0.52 ^a	0.95ª
	T1	1.48 ^b	1.80 ^b
	T2	2.38 ^b	2.35 ^b
	Sig.	**	***

Means within a column, for each season, flanked by se same letter are not significantly different at $P \le 0.05$ (Tukey HSD test). Significance of difference between treatments: ns – not significant; * 0.01< P \le 0.05; ** 0.001< P \le 0.01; *** P \le 0.001

Table 6. Transpiration rate (E) measured during hot and clear
days in the ripening period for the different water treatments in
2014 growing season.

Year	Treatment	E (mmol m ⁻² s ⁻¹)			
		05 Aug	02 Sep		
2014	Т0	1.26ª	0.82 ^{a c}		
	T1	2.25 ^a	3.23 ^b		
	T2	4.20 ^b	3.41 ^{ab}		
	T3	4.30 ^b	4.03 ^{a c}		
	Sig.	***	***		

Table 7. Net CO2 assimilation rate (A) measured during hotand clear days in the ripening period for the different watertreatments in 2014growing season.

Year	Treatment	A (µmol m ⁻² s ⁻¹)		
		05 Aug	02 Sep	
2014	т0	7.09 ^a	2.63ª	
	T1	5.63 ^a	6.80 ^b	
	T2	10.32 ^b	8.65 ^b	
	T3	10.13 ^b	9.01 ^b	
-	Sig.	***	***	

3.3 Yield components and berry composition

Yield and yield components were not significantly affected by irrigation treatments in 2007. However, in 2008 the yield (kg vine⁻¹) and the mean weight per cluster were significantly higher in T2 treatment (Table 8).

Table 8. Yield components at harvest, pruning weight andyield/pruning weight ratio for the different water treatments in2007 and 2008 growing seasons.

Year	Т	Yield	Clus	ster	Pruning	Y/Pw
		(Y)	Number	Weight	weight	
		(kg/	per vine	(g)	(P w)	
		vine)			(kg/vine)	
	T0	2.1	12.0	166.2	0.71	4.7
2007	T1	2.1	11.7	179.6	0.72	3.6
2007	T2	2.1	12.4	175.4	0.81	3.6
	Sig.	ns	ns	ns	ns	ns
	T0	1.9 ^a	13.3	139.1ª	0.50 ^a	3.9 ^a
2008	T1	2.7 ^a	12.4	207.0 ^b	0.64 ^a	4.5 ^a
	T2	4.3 ^b	15.2	271.0 ^c	0.66 ^b	6.8 ^b
	Sig.	***	ns	***	*	*

Means within a column, for each season, flanked by se same letter are not significantly different at $P \le 0.05$ (Tukey HSD test). Significance of difference between treatments: ns – not significant; * 0.01< P \le 0.05; ** 0.001< P \le 0.01; *** P \le 0.001

The number of clusters per vine was similar among years and among irrigation treatments. Pruning weight per vine was significantly lower in non-irrigated vines. The effect of irrigation in the increase of pruning weight was more pronounced in Touriga Nacional (Table 9). The balance between vine supply capacity and crop demand expressed in terms of yield/pruning weight was not impaired by the irrigation applied in 2007 and 2014, which is in agreement with other authors [11]. However, in 2008 and 2015 this ratio was higher in irrigated vines confirmed the considerable differences among years, in this values [11,12]

Year	Т	Yield	Clu	ster	Pruning	Y/Pw
		(Y)	Number	Weight	weight	
		(kg/	per vine	(g)	(Pw)	
		vine)			(kg/vine)	
	T0	1.48	16.79	90.76	0.60 ^a	2.67
	T1	1.64	18.09	92.91	0.69 ab	2.76
2014	T2	1.75	17.17	93.56	0.76 ^b	2.23
	T3	1.76	17.85	96.80	0.90 ^b	2.21
	Sig.	ns	ns	ns	***	ns
	T0	1.82 ^b	15.3	122.30 ^b	0.43 ^b	4.64 ^b
	T1	2.45 ^a	16.2	152.00 ^a	0.72 ^a	3.70 ^a
2015	T2	2.37 ^a	16.3	148.49 ^a	0.73 ^a	3.70 ^a
	T3	2.53 ^a	15.2	167.99 ^a	0.85 ^a	3.36 ^a
	Sig.	***	ns	***	***	***

 Table 9. Yield components at harvest, pruning weight and yield/pruning weight ratio for the different water treatments in 2014 and 2015 growing seasons..

Means within a column, for each season, flanked by se same letter are not significantly different at $P \le 0.05$ (Tukey HSD test). Significance of difference between treatments: ns – not significant; * 0.01< P \le 0.05; ** 0.001< P \le 0.01: *** P \le 0.001

At the time of harvest no significant differences in must composition were found among treatments for Tinta Roriz variety (Table 10). Similar results were obtained by Centeno et al. [13], in Spain, for the same variety (Tempranillo) and the same irrigation treatments. The exception was for total phenols that showed significantly lower values for irrigation treatments in 2008.

 Table 10. Berry composition at harvest for the different water treatments in 2007 and 2008 growing seasons.

Year	Т	Total soluble solids (°Brix)	Titratable acidity (g L ⁻¹ tartaric acid)	Colour intensity	Total phenols
2007	T0	20.57	3.21	5.33	76.53
	T1	21.10	3.68	3.43	63.07
	T2	21.53	4.05	3.33	65.23
	Sig.	ns	ns	ns	ns
2008	T0	21.55	3.18	4.57	55.97 ^a
	T1	21.87	2.86	4.17	44.17 ^b
	T2	21.75	3.40	4.07	43.50 ^b
	Sig.	ns	ns	ns	*

Means within a column, for each season, flanked by se same letter are not significantly different at $P \le 0.05$ (Tukey HSD test). Significance of difference between treatments: ns – not significant; * 0.01< P \le 0.05; ** 0.001< P \le 0.01; *** P \le 0.001

The total soluble solids (TSS) were not affected by irrigation treatments in Touriga National (Table 11). In 2014 titratable acidity was significantly affected by irrigation treatments with more water supplied (T3). This could be due to a slower degradation of malic acid influenced by microclimate effects through high vigour [14]. Colour intensity and total phenols were the quality parameters most affected by irrigation treatments in the Touriga Nacional variety. As the amount of water applied increase, the colour intensity and the total phenols decrease significantly. These results showed that high irrigation treatments could affect wine structure, colour stability and wine ageing.

Table 11. Berry composition at harvest for the different water	
treatments in 2014 and 2015 growing seasons.	

Year	Т	Total soluble solids (°Brix)	Titratable acidity (g L ⁻¹ tartaric acid)	Colour intensity	Total phenols
2014	T0	18.6	4.5 ^b	3.27 ^a	77.9 ^a
	T1	19.6	4.4 ^b	2.63 ^{ab}	67.2 ^{ab}
	T2	20.2	5.4 ^{ab}	2.62 ^{ab}	60.1 ^{bc}
	Т3	19.8	6.0 ^a	2.09 ^b	49.3°
	Sig.	ns	***	**	***
2015	T0	24.1	4.5	3.69 ^a	65.3ª
	T1	24.8	5.0	3.01 ^{ab}	50.4 ^{ab}
	T2	25.4	5.2	2.61 ^b	52.0 ^{ab}
	T3	25.8	5.3	2.61 ^b	35.4 ^b
	Sig.	ns	ns	*	*

Means within a column, for each season, flanked by se same letter are not significantly different at $P \le 0.05$ (Tukey HSD test). Significance of difference between treatments: ns – not significant; * $0.01 < P \le 0.05$; ** $0.001 < P \le 0.01$; *** $P \le 0.001$

4 Conclusions

The results showed that moderate water supplies during ripening period, for the region where the study was conducted (severe water deficits), benefit yield of Tinta Roriz and Touriga Nacional varieties. The main differences in yield between moderate water supplies and rainfed vines occurred in the growing season with the driest summers (2008 and 2015).

The moderate irrigation applied did not affected, significantly, berry sugar accumulation and titratable acidity. The total phenols were significantly lower in musts from irrigated vines and the colour intensity of Touriga Nacional musts was significantly reduced for the high irrigations treatments.

References

- Chaves M. M., Zarrouk O., Francisco R., Costa J. M., Santos T., Regalado A. P., Rodrigues M. L. and Lopes C. M., Grapevine under deficit irrigation: hints from physiological and molecular data. Annals of Botany **105**(5): 661-676 (2010).
- 2. Malheiro A.N.C., Microclimate, yield and water-use of vineyards in the Douro Region, Portugal. PhD Thesis. Cranfield University, Silsoe (2005).
- 3. Shellie, K., Water productivity, yield, and berry composition in sustained versus regulated deficit irrigation of Merlot grapevines. American Journal of Enology and Viticulture. **65**:197-205 (2014).
- 4. Dry P.R., Loveys B.R., McCarthy M.G., Stoll M., Strategic irrigation management in Australian vineyards. Journal International des Sciences de la Vigne et du Vin, **35**, 129-139 (2001).
- McCarthy M.G., Loveys B.R., Dry P.R., Stoll M., Regulated deficit irrigation and partial rootzone drying as irrigation management techniques for grapevines. In: 'Deficit Irrigation practices, Water Reports No 22, FAO-Food and Agriculture

Organization of the United Nations, Rome, pp 79-87 (2002).

- 6. Allen RG, Pereira LS, Raes D, Smith M (1998) Crop evapotranspiration. Guidelines for computing crop water requirements. FAO Irrigation and Drainage Paper 56, Rome (1998).
- 7. Scholander, P.F., Hammel, H.T., Brandtreet, E.T. and Hemmingsen, E.A., Sap pressure in vascular plants: negative hidrostatic pressure can be measured in plants. Science, **148**: 339-346 (1965).
- OIV. (1990). Récueil des Méthodes Internationales d'Analyse des Vins et des Moûts. Bull.OIV (Off.Int. Vigne Vin), París, (1990).
- Deloire A., Carbonneau A., Wang Z.P., Ojeda H., 2004. Vine and water a short review. Journal International des Sciences de la Vigne et du Vin, 38:1-13.
- Van Leeuwen, C., Tregoat, O., Chone, X., Bois, B., Pernet, D., Gaudillere, J.-P., Vine water status is a key factor in grape ripening and vintage quality for red bordeaux wine. How can it be assessed for vineyard management purposes?. Journal International des Sciences de la Vigne et du Vin, 43 (3), 121-134. (2009).
- Intrigliolo, D.S., Castel, J.R., Response of grapevine cv. 'Tempranillo' to timing and amount of irrigation: water relations, vine growth, yield and berry and wine composition. Irrigation Science, 28: 113-125 (2010)
- 12. Bravdo B., Hepner Y., Loinger C., Cohen S., Tabacman H., Effect of irrigation and crop level on growth, yield and wine quality of Cabernet Sauvignon. American Journal of Enology and Viticulture, **36**, 132–139 (1985).
- Centeno A., Sánchez-de-Miguel P., Linares R., Lissarrague J.R., Changes in must composition during ripening of "Tempranillo" grapevines as a function of two irrigation treatments. Acta Horticulturae, 689, 391-395 (2005).
- Baeza, P., Sánchez-de-Miguel, P., Centeno A., Junquera P., Linares R., Lissarrague J.R., 2007. Water relations between leaf water potential, photosynthesis and agronomic vine response as a tool for establishing thresholds in irrigation scheduling. Sci. Hortic. 114:151–158.