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Physiological changes induced by either pre- or post-veraison deficit irrigation in 'Merlot' vines grafted on two different rootstocks

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Summary

Reduced summer precipitations and higher evapotranspiration due to elevated temperatures are expected to enhance the impact of water deficit in modern viticulture. We investigated the effect of the timing of deficit irrigation on vine growth, water relations, yield and grape composition in 'Merlot' vines grafted on 1103P or SO4. In both years we did not measure any differences between rootstocks in stem water potential (SWP). Vegetative growth was decreased by the restriction of irrigation between fruit set and veraison. Stomatal conductance (g_s) was affected by irrigation, but not by the rootstock. During the pre-veraison period there was a clear inverse relationship between g_s and SWP. The leaf non photochemical quenching readily responded to the stress imposed on 1103P rootstock. Vines subjected to water deficit between fruit set and veraison produced smaller berries than well irrigated ones, whereas deficit applied after veraison determined about 10 % differences in berry weight. The highest and lowest values of pH and TA were measured in berries from pre-veraison deficit irrigated vines grafted on both 1103P and SO4, respectively.

K e y w o r d s : berry quality; non-photochemical quenching; stem water potential; stomatal conductance; yield; *Vitis vinifera*. L.

Introduction

Almost all wine producing areas are located in temperate zones characterized by warm and dry summers. In these regions grapevines are regularly exposed to periods of drought unless irrigation is used, but currently most viticulture is still rainfed. In 2016 less than 10 % of European vineyards were irrigated (Costa *et al.* 2016). However, this percentage is steadily increasing due to a number of reasons including climate change and the removal of irrigation bans in many traditionally rainfed areas. Climate change can lead to adjustments in growth and grape quality of 'Merlot' due to the susceptibility of this cultivar to water deficit (Bucchetti *et al.* 2011) and managing deficit irrigation can be strategical to optimise the performance of this cultivar in warm and dry climates. Reduced summer precipitations and higher evapotranspiration due to elevated temperatures are expected to enhance the impact of water deficit in modern viticulture (Jones *et al.* 2005, IPCC 2019). In some years, even if annual rainfall remains stable, vineyards are subjected to prolonged droughts during the growing season because of changes in its distribution pattern (IPCC 2019).

The effect of water deficit on grapevine productivity and berry quality does not only depend on the severity of drought, but also on the phenological stage at which it occurs (Matthews and Anderson 1989). Previous studies reported that early-season stress applied to either field-grown or potted vines had stronger negative effects on vegetative growth, berry size, and yield than water stress applied after veraison (Ojeda *et al.* 1999, Girona *et al.* 2009, Chaves *et al.* 2010, Basile *et al.* 2011, Munitz *et al.* 2016). The greater effect of pre-veraison water deficit on berry growth can be explained by the fact that final berry mass is mainly determined by the number of cells divided between flowering and veraison which, in turn, is strongly affected by drought stress (Harris *et al.* 1968). The direct connection between roots and fruits through the xylem vessels before veraison increases the berry sensitivity to water stress (Greenspan *et al.* 1994, Rogiers *et al.* 2001, Chaves *et al.* 2010, Munitz *et al.* 2016).

Water stress between fruit set and veraison has been reported to decrease soluble solids content (SSC) and slightly increase the titratable acidity (TA) in the must of Tempranillo vines (Girona *et al.* 2009). The impact of water deficit on fruit composition appears to be mediated by a reduction in vigor and the consequent increase in light availability in the canopy zone where clusters are present (Castellarin *et al.* 2007, Chaves *et al.* 2007). On the other hand, yield and berry weight are less affected by post-veraison water deficit (Greenspan *et al.* 1994, Rogiers *et al.* 2001). In an experiment carried out on 'Merlot' vines cultivated in a semi-arid area Munitz *et al.* (2016) observed that vines receiving 56 % of irrigation volumes of control vines from bunch closure to harvest produced similar yields with only a slight reduction in berry mass (-7 %) than well irrigated ones. Other experiments reported that post-veraison water deficits (during fruit ripening) increased SSC, but they did not affect TA and pH (Basile *et al.* 2011, Intrigliolo *et al.* 2016). Deficit irrigation

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during ripening was effective to increase anthocyanins and tannins in 'Merlot' vines (BUCCHETTI *et al.* 2011).

Potential water scarcity in the near future may also be challenged by selecting appropriate rootstocks. It is well known that rootstocks allow to grow grapevines under different soil and cultural conditions (HOWELL 1987, BERDEJA *et al.* 2015, KELLER 2015). Moreover, rootstocks can change the duration of phenological stages, biomass accumulation, canopy structure, vine vigour, yield and berry quality (KOUNDOURAS *et al.* 2008, SOAR *et al.* 2006). A number of mechanisms have been proposed whereby the rootstock affects vine physiology (LOVEYS and KRIEDEMANN 1974, SOAR *et al.* 2006, ALSINA *et al.* 2011). Less gas exchange and canopy growth reduce transpiration needs when water becomes scarce (KOUNDOURAS *et al.* 2008). The anatomy of xylem vessels has been shown to influence scion water relations, water uptake and transport to the shoot (CARBONNEAU *et al.* 1985, LOVISOLO and SCHUBERT 1998, DE HERRALDE *et al.* 2006, KOUNDOURAS *et al.* 2008).

The aim of this study was to explore the timing of deficit irrigation on vine growth, water relations, yield and grape composition in fully-productive 'Merlot' vines grafted on two rootstocks over two consecutive years.

Material and Methods

Plant material and irrigation regime: Six-year old grapevines (*Vitis vinifera* L.) of cultivar 'Merlot' grafted on two rootstocks [1103 Paulsen (*V. rupestris* x *V. berlandieri*) and SO4 (*V. riparia* x *V. berlandieri*)] were grown outdoor in 50 L containers at the Colignola experimental farm of the University of Pisa (43.73° N 10.47° E) over two consecutive growing seasons. All vines were pruned in winter to retain one spur with two count buds and one cane with 6-8 count buds according to a single Guyot training system. The containers were covered with plastic film to avoid evaporation from the soil. Temperature, radiation, precipitation, reference evapotranspiration were measured using a weather station (WatchDog, Spectrum Technologies Inc, Aurora IL, USA) installed on site. Annual precipitation was 932 and 970 mm in 2018 and 2019, respectively, almost equal to reference evapotranspiration (ET_0), calculated according to the Penman–Monteith equation, which was 927 and 900 mm in those respective years. The spring of 2019 was cooler than that of 2018 (mean air temperature was 17.7 °C and 16.0 °C in 2019 and 2018 respectively). During the summer (June 21 through September 21) the mean air temperature was similar (24.1 and 24.3 °C in 2018 and 2019, respectively, but it rained more in 2019 (157 mm) than in 2018 (92 mm).

Drip irrigation was used to supply water to the vines (two drippers of 2 $L \cdot h^{-1}$ each). The water volume was calculated based on effective evapotranspiration and adjusted to maintain the stem water potential (SWP) of 100 % irrigated vines above -0.5 MPa, unless otherwise stated. All vines received water to satisfy 100 % of their needs until day of the year (DOY) 154 (2018) and 164 (2019), when two regulated deficit irrigation (RDI) regimes were established.

Water deficit (34-49 % of full irrigation) was imposed from either fruit set through veraison (FS-V) or from veraison through harvest (V-H) (Table). Thus, fully-irrigated vines received 100 % of water need throughout the growing season, whereas both deficit treatments received full irrigation until fruit set and from harvest through leaf fall. In addition, RDI 2 vines were fully irrigated during the FS-V interval and RDI 1 ones during the V-H period in both years.

Phenology and vine development: Daily mean temperatures greater than 10 °C were summed to calculate growing degree days (GDD) from April 1 through October 31 (Fig. 2). Vine phenology was monitored to determine the dates of bud burst (BBCH 09), flowering (BBCH 65) and veraison (BBCH 87), using the modified Eichhorn-Lorenz (E-L) scale (COOMBE 1995). After

Table

Veraison and harvest dates, irrigation volumes of 'Merlot' vines grafted on either 1103P or SO4 rootstock in 2018 and 2019. In brackets the percentage of irrigation volume applied to deficit irrigated vines with respect to fully-irrigated ones (FI) of the same rootstock. Legend: DOY, day of the year; FS-V, fruit set – veraison; V-H, veraison-harvest; RDI 1, water deficit applied from fruit set through veraison; RDI 2, water deficit applied from veraison through harvest

Year	Rootstock	Irrigation	Veraison (DOY)	Harvest (DOY)	Irrigation volume ($L \cdot vine^{-1}$)		
					FS-V	V-H	FS-H
2018	1103P	FI	204	240	170	285	455
		RDI 1	204	239	59 (35)	289	349 (77)
		RDI 2	204	233	170	141 (49)	310 (68)
	SO4	FI	204	239	161	294	455
		RDI 1	200	241	55 (34)	307	362 (80)
		RDI 2	204	233	161	141 (48)	302 (66)
2019	1103P	FI	213	262	215	200	414
		RDI 1	217	246	77 (39)	184	262 (72)
		RDI 2	213	246	215	85 (49)	300 (81)
	SO4	FI	213	277	215	200	414
		RDI 1	217	276	77 (39)	184	262 (72)
		RDI 2	213	276	215	85 (49)	300 (81)

harvest we measured the number of nodes per shoot and internode length of the proximal, median and distal shoot on each vine.

The weight of pruned wood from five vines per treatment was measured to calculate the yield-to-pruning weight ratio (Ravaz Index). The rootstock trunk diameter was measured eight times in 2018 from budburst through harvest. In 2018 the canopy volume was measured manually at the beginning of irrigation regimes only. Maximum canopy height and canopy height from the ground were measured at regular intervals of 0.3 m along the row. Canopy width was measured at the same interval along the row and at 0.9, 1.3 and 1.8 m height from the ground. These data were used to create a 3-D canopy volume as reported in Caruso *et al.* (2017). In 2019 it was measured seven times from the beginning of irrigation differentiation through harvest using aerial images (Caruso *et al.* 2017). Images were obtained using a multispectral camera (Micasense RedEdge MX, MicaSense Inc., Seattle, WA) carried by an unmanned aerial vehicle (UAV) flying over a predetermined waypoint course at 50 m above ground level. The three-dimensional canopy volume was reconstructed from the digital surface model (DSM) using Agisoft Photo-Scan® (Agisoft LLC), and ArcGIS software® (ESRI, Redlands, CA, USA), as in Caruso *et al.* (2017).

Water potential and gas exchange: The stem water potential (SWP) of one fully-expanded leaf per vine was measured from three vines per treatment using a pressure chamber (PMS Instruments, Albany OR, USA) every week starting from DOY 154 and DOY 164 in 2018 and 2019, respectively. Transpiration was stopped by enclosing the leaf in a dark plastic bag for about 60 min, then the leaf was excised with a sharp razor blade, immediately put in the chamber cylinder and pressurized. The water stress integral (WSI) was calculated using a standard procedure (Myers 1988).

Gas exchange parameters were measured on fully-expanded leaves of three plants per treatment using a portable open system CIRAS-1 (PP Systems, Hitchin Herts, UK). Measurements were taken on cloudless days between 9:00 a.m. and 11:00 a.m. at photosynthetic photon flux density greater than 900 $\mu mol \cdot m^{-2} \cdot s^{-1}$), ambient CO_2 ranging from 390 to 410 $\mu L \cdot L^{-1}$, and air temperature of 29.8 ± 1.3 °C (mean ± standard deviation). In 2019 nonphotochemical theoretical quenching (NPQ_t) was measured concomitantly on the same leaves used for leaf gas exchange using a MultispeQ v2.0 (Photosinq Inc., Lansing, MI, USA) following standard procedures (Tietz *et al.* 2017, Kuhlgert *et al.* 2016).

Yield and must composition: Clusters were harvested when berry SSC had reached 22 ± 0.5 °Brix: between DOY 233 and 241 in 2018 and between DOY 246 and 277 in 2019. The crop was harvested from individual vines, weighed, the berry fresh weight immediately determined on samples of 20 berries for each rootstock-irrigation treatment (three 20-berry replicates per treatment), then berries were dried in an oven at 70 °C for dry weight. Total SSC, TA, and pH were determined on the same berry samples. The juice was extracted from each sample of berries, the SSC measured with a hand refractometer, and a 10 mL aliquot titrated with 0.1 N NaOH to an endpoint pH of 8.2 to determine TA ($g \cdot mL^{-1}$). The pH was measured with a pH meter (Hanna Instruments, Woonsocket, RI, USA) calibrated at pH 7.0 and 4.0.

Experimental design and statistical analysis: The experimental design was a split-plot with rootstock (R) as the main plot and irrigation (I) as the subplot. Forty-eight vines received water according to three irrigation regimes (24 vines per rootstock). All measurements were carried out on at least three vines per treatment. Stem water potential, leaf gas exchange and berry weight were measured on three vines per treatment, whereas trunk diameter, canopy volume, pruning weight, vine yield, SSC, TA, and pH were measured on five vines per treatment. Where applicable, data were analyzed by regression using Costat (CoHort Software, Monterey, USA).

Results

Budbreak occurred for both rootstocks on DOY 99 and 100-101 in 2018 and 2019, respectively. Flowering, veraison, and harvest were delayed in 2019 with respect to 2018 due to the cooler spring of 2019. In 2019 flowering occurred on DOY 162 corresponding to 480 GDD, veraison on DOY 214 (GDD equal to 1250), and harvest on DOY 264 (Table, annual average of all treatments). There were no differences in the dates of flowering and veraison between rootstocks, while grapes were harvested about 25 d earlier in 2018 than in 2019. In 2018 flowering (DOY 148 equal to 482 GDD), veraison and harvest occurred at similar dates for both rootstocks (Table). The progression of GDD showed the delay in the accumulation of heat units in the spring of 2019 with a recovery in the second part of the growing season, which ended in values similar to the 1998-2017 average (data not shown). On the other hand, the 2018 growing season was evidently warmer than the 20-year average.

Because of the differences in GDD between the two years, treatments were started at different dates. As a result, in 2018 RDI 1 vines received more water than RDI 2 (77-80 % vs. 68-66 % of well irrigated vines over the entire irrigation period), whereas in 2019 the opposite was true (RDI 1 received 72 % of well irrigated vines and RDI 2 81 %; Table). Water deficit imposed between veraison and harvest resulted in early harvest dates of seven (1103P) and six (SO4) days with respect to fully-irrigated vines in 2018 and 16 and one for those respective rootstocks in 2019 (Table).

The SWP of fully-irrigated vines ranged between -0.3 and -0.6 MPa with the exception of two dates (DOY 218 in 2018 and DOY 175 in 2019) when a pump failure caused SWP to drop to -1.0 MPa (2018) and -0.7 MPa (2019). No differences between rootstocks in SWP of control vines were measured in both years (Fig. 1). The water deficit imposed between fruit set and veraison caused a significant reduction of SWP in RDI 1 vines after 8 and 18 d from the beginning of the irrigation differentiation in 2018 and 2019, respectively (Tab. 1, suppl. material). In both rootstocks the vine water status readily responded to the irriga-

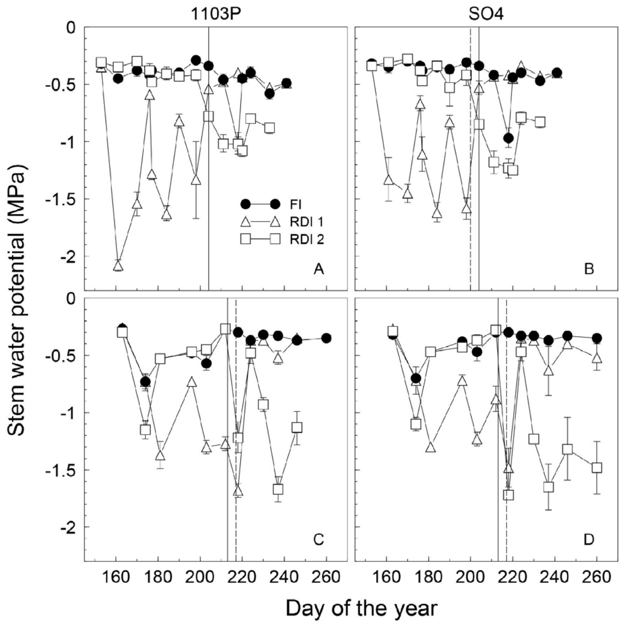

Fig. 1: Stem water potential (SWP) measured in 2018 (**A**, **B**) and 2019 (**C**, **D**) of *Vitis vinifera* L. vines ('Merlot') grafted on either 1103P or SO4 rootstock and subjected to different irrigation regimes (FI, full irrigation from budburst through harvest; RDI 1 and RDI 2, water deficit applied from fruit set through veraison and from veraison through harvest, respectively). Values are means ± standard error of three vines per treatment. Solid (SO4) and dotted (1103P) vertical lines indicate the dates of veraison.

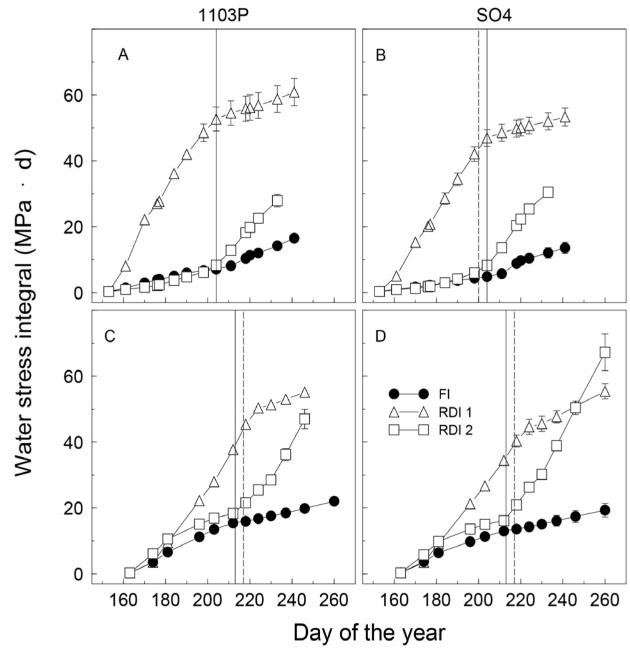

Fig. 2: Water stress integral measured in 2018 (**A**, **B**) and 2019 (**C**, **D**) in *Vitis vinifera* L. vines ('Merlot') grafted on either 1103P or SO4 rootstock and subjected to different irrigation regimes (FI, full irrigation from budburst through harvest; RDI 1 and RDI 2, water deficit applied from fruit set through veraison and from veraison through harvest, respectively). Values are means ± standard error of three vines per treatment. Solid (SO4) and dotted (1103P) vertical lines indicate the dates of veraison.

tion regime (Fig. 1). The water stress integral (WSI) well represented the progression of stress and its subsequent relief upon abundantly watering the vines. At harvest 2018 the WSI of FI, RDI 1 and RDI 2 vines reached 17, 61 and 28 MPa x d (1103P), and 14, 53, 31 MPa x d (SO4), respectively (Fig. 2). In 2019, the WSI was 22, 55, 47 (1103P) and 19, 55 and 67 (SO4). In 2018 higher values of WSI were measured in RDI 1 vines than in RDI 2 ones, whereas similar values were measured for both RDI treatments in 2019 (Fig. 2). The higher level of stress experienced by RDI 1 vines in 2018 was mainly due to the severe water deficit imposed during the first week of the experiment and to the shorter period of the post-veraison water deficit (RDI 2) with respect to the RDI 1 one (Figs. 1 and 2). The different duration of the V-H period in 2018 and 2019 was also responsible for the differences in WSI values reached by the RDI 2 treatments in those two years (Table).

Vegetative growth was markedly affected by the restriction of irrigation between fruit set and veraison. In 2018 differences in trunk diameter increment between well irrigated (FI and RDI 2 treatments) and RDI 1 appeared from DOY 169 for both rootstocks (Fig. 3). During the FS-V period trunk shrinkage was more pronounced in vines grafted on 1103P than in SO4 (Fig. 3A, B). In general, the trunk growth of 1103P rootstock was higher than that of SO4 under non limiting water supply (Fig. 3A, B; Tab. 2, suppl. material). Vines grafted on 1103P had larger canopies (0.42 ± 0.04 m^3) than those grafted on SO4 (0.29 ± 0.03 m^3) at the beginning of irrigation differentiation in 2018 (data not shown, values are means ± standard deviations of nine vines). In 2019, irrigation significantly

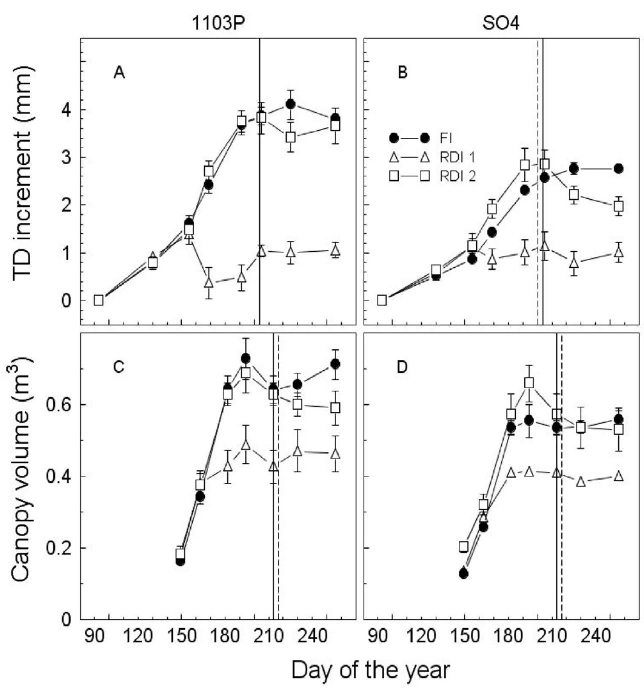

Fig. 3: Seasonal courses of trunk diameter increment in 2018 (**A**, **B**) and canopy volume measured in 2019 (**C**, **D**) of *Vitis vinifera* L. vines ('Merlot') grafted on either 1103P or SO4 rootstock and subjected to different irrigation regimes (FI, full irrigation; RDI 1, water deficit applied from fruit set through veraison; RDI 2, water deficit from veraison through harvest). Values are means ± standard error of five vines per treatment. Solid (SO4) and dotted (1103P) vertical lines indicate the dates of veraison. Trunk diameter increments were normalized against values at the beginning of the experiment (trunk diameter = 0).

affected canopy growth of both rootstocks, even though differences between irrigation treatments were less evident for SO4 vines. Significant differences between irrigation treatments were evident about 20 d after the beginning of differentiation and lasted until harvest (Fig. 3C, D; Tab. 2, suppl. material). At veraison, the canopy volume of control vines was 51 % (1103) and 31 % (SO4) greater than that of RDI 1 ones (Fig. 3). Significant differences in canopy volume between rootstocks were measured only for well irrigated vines. Water stress imposed after veraison on canopy volume only slightly contributed to final canopy volume (Fig. 3). There were no significant differences in pruned wood between irrigation treatments in 2018, whereas in 2019 FI vines showed higher values (545 g, average of SO4 and 1103P) than those of RDI 1 and RDI 2 (435 and 441 g, respectively). Pruned wood weight was unaffected by the rootstock in both years (404 and 413 g for 1103P and SO4 vines, respectively, in 2018 and 477 and 471 g in 2019).

Stomatal conductance was affected by irrigation, but not by the rootstock (Fig. 4; Tab. 3, suppl. material). The lowest values of g_s were measured for RDI 1 vines, which experienced water stress before veraison (40 % and 50 % of FI in 2018 and 2019; average of both rootstocks). In 2019 g_s values of RDI 1 leaves did not return to those of control vines after veraison when RDI 1 vines were also irrigated (Fig. 4). In 2018 stomatal conductance of RDI 2 leaves showed smaller reductions during the V-H period than in 2019. In our experiment g_s values lower than 50 $mmol \cdot m^2 \cdot s^{-1}$, which is considered a threshold for severe water stress (Cifre *et al.* 2005, Lovisolo *et al.* 2010), were measured only for RDI 1 vines on four (2018) and one (2019) date of measurement. During the pre-veraison period there was a clear inverse relationship between g_s and SWP without differences due to the rootstock, but this relationship was not evident during the post-veraison period, when data appeared more scattered (Fig. 5). The leaf non photochemical quenching readily responded to the stress imposed both before and after veraison on 1103P rootstock. As for the SO4 rootstock the effect was evident during pre-veraison deficit, but not during post-veraison (Fig. 6). NPQ_t values of FI-1103P vines were quite stable during the entire irrigation period (with the exception of the last measurement in 1103P vines), whereas in FI-SO4 vines they were slightly higher during the V-H period than in the FS-V one.

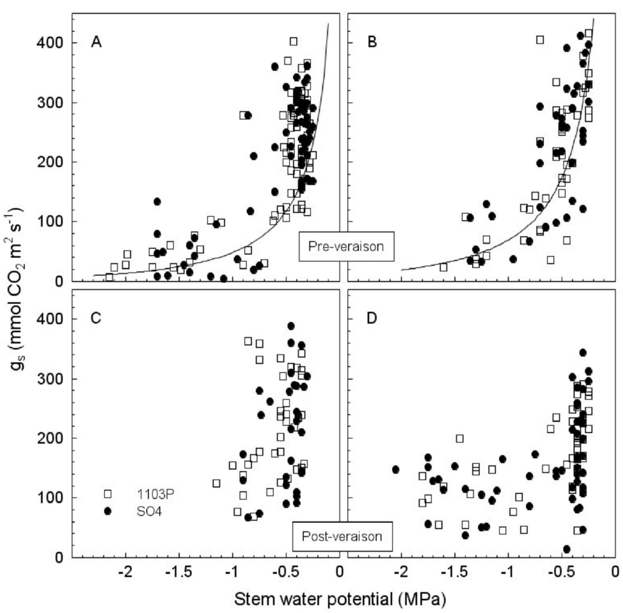

Fig. 5: The relationship between stomatal conductance (g_s) and stem water potential in *Vitis vinifera* L. vines ('Merlot') grafted on either 1103P or SO4 rootstock and subjected to different irrigation regimes in 2018 (**A**, **C**) and 2019 (**B**, **D**). Each symbol represents one vine. Regression equations: g_s = 1/(-0.0103+0.0115 e^{-SWP}) (**A**); g_s = 1/(-0.077+0.008 e^{-SWP}) (**B**).

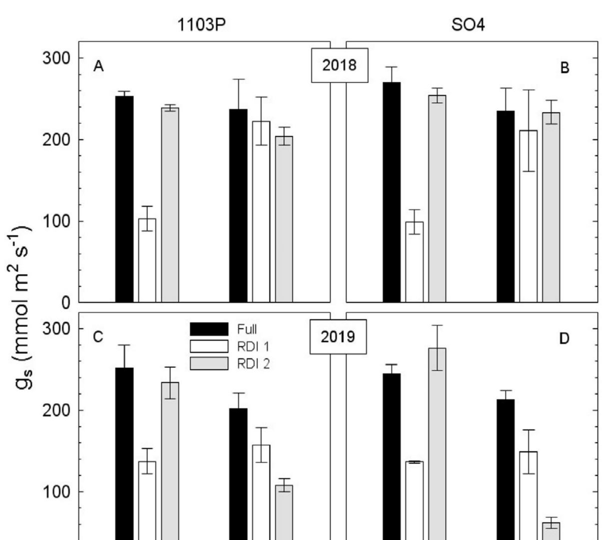

Fig. 4: Stomatal conductance (g_s) measured in 2018 (**A**, **C**) and 2019 (**B**, **D**) in *Vitis vinifera* L. vines ('Merlot') grafted on either 1103P or SO4 rootstock and subjected to different irrigation regimes (FI, full irrigation from budburst through harvest; RDI 1 and RDI 2, water deficit applied from fruit set through veraison and from veraison through harvest, respectively). Values are means ± standard error of three vines per treatment. Legend: FS, fruit set; V, veraison; H, harvest.

Yields ranged between 1116 and 1852 $g \cdot vine^{-1}$ or 1047 and 2173 $g \cdot vine^{-1}$ in 2018 and 2019, respectively. Higher yields were obtained in vines grafted on 1103P in 2019, but not in 2018 (data not shown). In both years the lowest values of yield, berry fresh and dry weight were measured for RDI 1 treated vines regardless of rootstock (Fig. 7). In particular, berry fresh and dry weight of RDI 1 vines were 68 and 71 % of that of FI vines (average of both rootstocks and two years), respectively, whereas RDI 2 vines showed smaller differences with control ones (90 and 89 % for FW and DW, respectively). The highest and lowest values of pH and TA were measured in berries from RDI 1 vines grafted on both 1103P and SO4, respectively (Fig. 7). The TA of RDI 1 berries was 68 % (2018) and 71 % (2019) than that of FI ones (average of both rootstocks). In 2019 the same parameters showed a significant interaction between irrigation and rootstock (Fig. 7). Irrigation significantly affected the yield to pruning weight ratio in both years. The lowest values were measured in SO4-RDI 1 vines in both years.

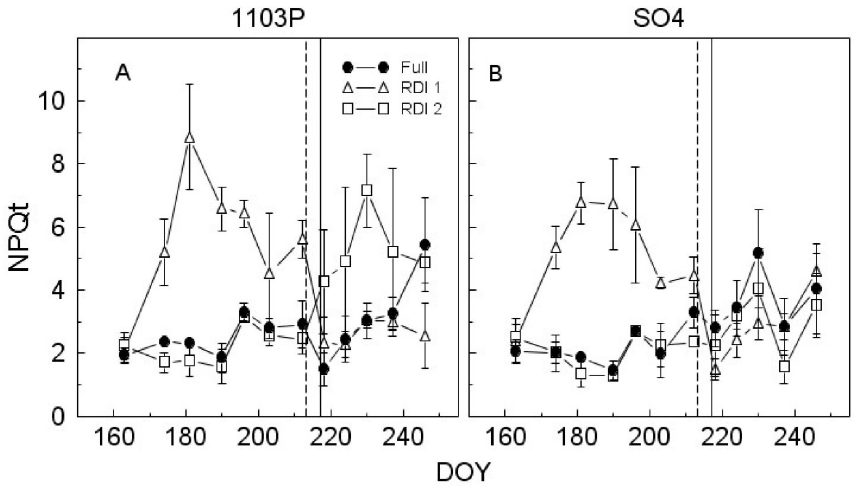

Fig. 6: Nonphotochemical quenching (NPQ_t) measured in leaves of *Vitis vinifera* L. ('Merlot') grafted on either 1103P (**A**) or SO4 (**B**) rootstock and subjected to different irrigation regimes (FI, full irrigation; RDI 1, water deficit applied from fruit set through veraison and RDI 2, water deficit from veraison through harvest) in 2019. Values are means ± standard error of three vines per treatment. Solid (RDI 1) and dotted (FI and RDI 2) vertical lines indicate the date of veraison within each irrigation treatment.

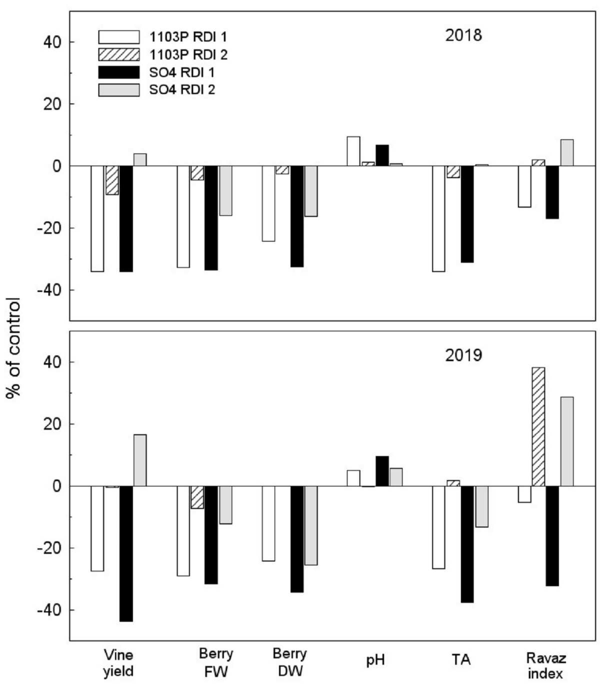

Fig. 7: Changes in yield, berry fresh weight (FW), berry dry weight (DW), pH, titratable acidity (TA), Ravaz index measured in 2018 and 2019 in grapevines ('Merlot') grafted on either 1103P or SO4 rootstock and subjected to different irrigation regimes (RDI 1, water deficit applied from fruit set through veraison and RDI 2, water deficit from veraison through harvest). Values are expressed as percentage of those measured in fully-irrigated (FI).

Discussion

The water deficits imposed were severe as the SWP reached -1.5 MPa in both years, comparable to those measured in field-grown vines cultivated in semi-arid regions (Intrigliolo and Castel 2010, Romero *et al.* 2010). The lowest SWP (-2.0 MPa) was measured for RDI 1 vines on 1103P rootstock in 2018, when differences between rootstocks were evident before veraison; on the other hand, in 2019 SWP was similar for both rootstocks throughout the experimental period. The dramatic drop in SWP in grapevines grafted on 1103P in 2018 might have been enhanced by the larger canopy and more vigorous growth of this rootstock compared with SO4. Leaf area was greater in 1103P-grafted vines than in SO4 ones, which received the same volumes of water by irrigation. In 2019, when canopy size at the beginning of the irrigation differentiation was similar, we measured similar values of SWP in 1103P and SO4 grafted vines. In general, the 1103P rootstock induced greater vigour to the scion than SO4 in well irrigated vines, which included FI and RDI 2 treatments during the pre-veraison period. Similar findings were also reported in previous studies carried out on both field-grown and potted grapevines (Koundouras *et al.* 2008, De Herralde *et al.* 2006).

Water stress applied at the beginning of the berry growing season (from fruit-set till veraison) had a greater effect on vegetative growth than that applied during berry maturation (from veraison to harvest). These results are in agreement with previous findings indicating that in grapevines the main vegetative growth period occurs before bunch closure (Munitz *et al.* 2016, Intrigliolo and Castel 2008, Romero *et al.* 2010). Under well irrigated conditions both canopy and trunk growth of the more vigorous 1103P were higher than those of vines on SO4. The trunk growth of 1103P rootstock was more sensitive to changes in soil water availability than SO4. The canopy reduction we observed in RDI 2 vines was primarily due to the higher pre-harvest defoliation rather than internode length (data not shown) and pruned material. Other authors measured a cessation of canopy development when water stress was imposed before veraison (Picón-Toro *et al.* 2012, Intrigliolo and Castel 2008, Romero *et al.* 2010). Many studies have shown that grapevine responses to water deficit involve a decrease in leaf expansion and internode elongation (Schultz and Mathews 1988, Lovisolo *et al.* 2010). The seasonal pattern of canopy volume in 2019 showed differences only between 1103P and SO4 fully-irrigated 'Merlot' vines, whereas no differences where measured between rootstocks under water deficit conditions both before and after veraison. Koundouras *et al.* (2008) observed a significantly higher vegetative growth in 1103P-grafted 'Cabernet' vines compared to SO4 and attributed this result to the larger root system of 1103P. Similar findings on the effect of root system on canopy growth were also reported in previous studies (Winkel and Rambal 1993, De Herralde *et al.* 2006). The general lack of differences in canopy volume between rootstocks under water deficit conditions observed in our study might have been caused by the fact that root systems of all vines were similar because of restrictions imposed by the container. Roots confined within a pot cannot explore deeper soil layers and this amplifies the effect of water shortage. In field-grown grapevines there is a balance between canopy and root system, so that bigger canopies are sustained by larger root systems. On the contrary, the higher leaf area-to-root ratio of potted vines can lead to partial tissue dehydration to satisfy the water requirements of the canopy when water stress occurs. Previous studies indicated that internal water redistribution might play an important role in drought resistance in woody perennial plants (Smart *et al.* 2005, Bauerle *et al.* 2008). The results obtained in our experiment confirm the crucial role of root system in conferring the drought-tolerant behavior to 1103P (Alsina *et al.* 2011). Differences in spring climatic conditions were found between the two years. The lowest air temperature in April and May in 2019 led to a delay in flowering of about two weeks with respect to the previous year. In general, grapevines in 2019 showed a slower vegetative growth and berry maturation rates than in 2018. Previous studies confirmed the strong relationship between the air temperature and the earlier occurrence of phenological phases, which may also affect the final quality of products (Jones *et al.* 2005, Dalla Marta *et al.* 2010). In our experiment, this behavior may have been responsible for the lack of differences in canopy volume between vine grafted on 1103P and SO4 at the beginning of the irrigation differentiation in 2019, which, on the contrary, were measured in 2018.

Stomata were relatively insensitive to water stress above an SWP threshold of about -0.7 MPa, whereas below that threshold stomata rapidly closed, driven by the decreasing SWP. This is a common response in grapevines subjected to drought since many previous studies showed that stomatal conductance and photosynthesis of grapevine declined when water deficit increased (Romero *et al.* 2010, Iacono *et al.* 1998, Koundouras *et al.* 2008). The g_s-SWP relationship was tight when the deficit was imposed before veraison, but more erratic when water deficit was applied after veraison. In fact, differences between RDI 2 and FI vines during the V-H period were evident only in 2019. Our results are coherent with those reported by Koundouras *et al.* (2008), who showed that scion ('Cabernet Sauvignon') stomatal conductance response to water conditions was not altered by the rootstock (1103P and SO4). Other experiments showed that the rootstock effect on scion gas exchange and water status observed in field-grown 'Shiraz' vines was likely induced by differences in the rootstocks ability to uptake and provide water to the scion (Soar *et al.* 2006). Yields were affected by the irrigation regime being lowest for RDI 1-treated vines (-34.2 -34.0 % for 1103P and -27.7 -43.9 % for SO4). On the other hand, the yield of RDI 2-vines was similar to that of controls probably due to the later stage of fruit development at which deficit occurred. Some studies showed that yields decreased linearly with decreases in leaf water potential at a reduction rate comprised between 41 % and 66 % per MPa (Grimes and Williams 1990, Williams 2010, Miras-Avalos and Intrigliolo 2017). The timing of water deficit also alters berry weight. An early water stress (RDI 1) decreased berry weight, in agreement with previous studies reporting the high sensitivity of berry growth to soil water deficit during the post-bloom cell division period (Basile *et al.* 2011, Palliotti *et al.* 2014, Merli *et al.* 2016, Matthews and Anderson 1988, Girona *et al.* 2009). In an experiment carried out on field-grown 'Merlot' vines, Munitz *et al.* (2016) measured an increase in berry size and yield if the water supply was not limited during early fruit development, whereas restricting water during berry maturation did not alter yield or berry maturation.

Irrigation significantly affected pH and TA of berry juice, whereas the rootstock did not. Pre-veraison water deficit consistently increased pH and decreased TA in both years, whereas post-veraison deficit had negligible effects on both parameters. In RDI 1 vines titratable acidity content was in general depressed as compared to FI and RDI 2 ones. The lowest values of TA were measured in RDI 1 grapevines that, in turn, showed the lowest pruning weight (in 2018 and 2019) and canopy volumes (in 2019). In an experiment carried out on 'Cabernet Sauvignon', Keller *et al.* (2016) observed a negative correlation between pruning weight and the light intercepted by the fruit zone and measured a higher cluster light interception in deficit irrigated vines than in well irrigated ones. In another experiment carried out on potted grapevines, the titratable acidity decreased as the leaf water potential measured from fruit-set to veraison declined, whereas the grapevine water status did not affect this parameter from flowering to full fruit-set and from 60 % of veraison to harvest (Basile *et al.* 2011).

'Merlot' is an international cultivar grown on about 266.000 ha (OIV 2017) and often used for top quality wines. We showed the different effect of pre- or post-veraison water deficits on the physiology, growth and berry quality of 'Merlot' grapevines grown under Mediterranean climate conditions. With the increasing threats posed by global warming, such as the occurrence of early soil water shortage events, managing deficit irrigation can be strategical to maintain and improve the performance of this cultivar in warm and dry climates at a reasonable cost in terms of water spending. Water restrictions from fruit set till veraison appear to affect mainly gas exchange parameters, canopy growth, berry weight, Ravaz index and titratable acidity, whereas changes due to post-veraison deficit were smaller or minor.

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