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PHYSIOLOGICAL AND MORPHOLOGICAL RESPONSES OF GRAPEVINE (*V. vinifera* L. CV. ‘ITALIA’) LEAF TO WATER DEFICIT UNDER DIFFERENT ROOTSTOCK EFFECTS

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Abstract. Extreme weather conditions with prolonged dry periods and high temperature can severely influence grapevine physiology and morphology. Understanding the physiological and morphological responses of grapevines to water deficit is thus of utmost importance to modulate the appropriate plant development. The present study evaluates the effects of deficit irrigation (DI) on certain leaf characteristics of grapevine cv. ‘Italia’ cultivated on different rootstocks. DI had remarkable effects on the growth, morphology, tissue structure, water status and physiology of grapevine leaf. Response of the ‘Italia’ cultivar to DI depended on rootstock used. For example, leaf fresh weight of ‘Italia’/5 BB under DI decreased by 15.2% in comparison to full irrigation (FI), whereas fresh weight values for ‘Italia’/99 R and own-rooted vines under DI decreased 6.2 and 10.5%, respectively. Under FI treatment, stomatal conductance (g_s) reached values of 189.0 mmol m^{-2} s^{-1} in ‘Italia’/5 BB, and 178.8 mmol m^{-2} s^{-1} in ‘Italia’/99 R. The g_s values under DI condition were 178.1 and 164.0 mmol m^{-2} s^{-1} for the vines on 5 BB and 99 R respectively. Stomatal conductance decreased about 21.1, 13.8 and 10.2% in vines cultivated on 5 BB, 99 R and own root, respectively. In response to DI, leaf relative water content decreased about 9.4, 4.1 and 3.9% for ‘Italia’/5 BB vines, own roots, and ‘Italia’/99 R, respectively. Combined data over years revealed that the vines cultivated on 99 R displayed more tolerant leaf growth and physiology to drought in comparison to vines on 5 BB.

Key words: *Vitis*, drought, leaf physiology, stomatal conductance

INTRODUCTION

The majority of the grape acreage around the world is located in Mediterranean type climates, characterized by mild winters and dry summers with scarce water resources [Collins et al. 2009]. The climate scenarios projected for the region will exacerbate

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these impacts with more frequent and extreme high temperatures and drought events [Bates, 2008]. Traditionally, grapevine is cultivated without irrigation and thus yield and grape quality depend on the ability of vine to cope with periods of drought by adapting rootstock and scion to edaphic conditions [Lovisolo et al. 2010].

Plants have developed a wide range of strategies to ensure survival under environmental stress conditions. For example, plants close their stomata and accumulate compatible solutes to maintain a low water potential and avoid dehydration during drought [Skirycz and Inze 2010]. Drought also slows down photosynthesis since stomatal closure reduces transpiration and gaseous exchange [González-Fernández et al. 2015]. Studies revealed that grapevine regulates the water flow to the leaf and from the leaf to the atmosphere by aquaporins [Vandeleur et al. 2009], vessel anatomy [Lovisolo et al. 2010] and stomatal conductance [Soar et al. 2006]. Consequently, vines retard their vegetative growth gradually. In grapevine a reduced leaf and shoot growth, inhibition of leaf expansion and a decrease of average diameter of xylem vessels have been found [Lovisolo et al. 2002]. Such decreases in vegetative growth is result of the suppression of photosynthesis due to stomata closure and limitations to CO_2 diffusion within the leaf [Flexas et al. 2004]. When stomatal conductance drops below 50 mmol H_2O m^{-2} s^{-1} photosynthetic limitations become more dependent on nonstomatal processes, especially decreased mesophyll diffusion conductance to CO_2 and impaired photochemistry [Flexas and Medrano 2002].

For sustaining good quality yield, a high priority should be given to minimizing the detrimental effects of environmental stresses on crop production by (1) applying modern breeding techniques and biotechnological tools, (2) increasing physical and chemical fertility as well as maintaining productivity of cultivated soils by adequate and balanced supply of mineral nutrients, and (3) choosing concrete cultivars/rootstock combinations. The grapevine rootstocks vary in physiological response to water scarcity [Kounduras et al. 2008], since the rootstock genotypes were derived from various American *Vitis* species [Sabir et al. 2010]. A drought-tolerant rootstock would be welcomed as many vineyards around the world lack suitable or adequate quantities of irrigation water [Corso and Bonghi 2014]. Water stress has become a management target to secure high fruit quality and improve sustainability of water use by rewarding crop quality and quantity.

Due to increased global air temperature [Vicente-Serrano et al. 2014] and intensity of climatic anomalies, such as droughts and heat waves [Jones and Vaughan 2010], the optimization of water use for vineyards, by improving water use efficiency, is a core subject of interest to ensure sustainability in viticulture. Consequently, an important amount of applied and fundamental study has been focused on the exploration of the capacity to optimize grapevine water use. Majority of these researches are related to the evaluation of irrigation schedule by introducing new technologies to decrease water consumption. However, there is a limited knowledge on cultivar/rootstock interacting response of grapevines to water scarcity in spite of the fact that global climate change is expected to result in more frequent and intense drought in the Mediterranean Region. Particularly, shifts in precipitation patterns can affect most European regions, with increased risk of drought. Predicted changes in the climate of viticultural regions may alter both the spectrum and the distribution of grapevine varieties and rootstocks currently used. In vine regions with these climatic characteristics, it is fundamental to investigate the genotypic behaviour grapevines under the pressure of each of these factors as simple variable or in interaction, in order to adjust the best strategy to cultivate the vines without loss of the yield and quality. Therefore the present study was conducted (a) to evaluate leaf growth, morphology and physiology responses of 'Italia' grape, one of the most common worldwide table grape cultivar, (b) to compare worldwidely popular rootstocks characterized by different resistance aptitude to dry conditions, and (c) to analyze whether the rootstocks usage impair the leaf characteristics of 'Italia' grapes in comparison with own-rooted vines.

MATERIALS AND METHODS

Plant materials and growth condition. Experiments were carried out in the controlled glasshouse of Selcuk University, Konya, Turkey during two consecutive years 2011 and 2012. The 'Italia' table grapevine cultivar was cultivated on two different rootstocks or on its own root. The experimental layout was a randomized complete block design with two irrigation [full irrigation (FI) and deficit irrigation (DI)] and different grafting combinations of 'Italia' on Kober 5 BB (5 BB), Richter 99 (99 R) or grown as own roots. At the beginning of first experimental year, two years old vines grown in such equal-sized pots were selected on the basis of homogeneity in growth. Treatments were replicated three times in randomized blocks, with four vines per replicate. The first group of this cultivar was cultivated on 5 BB (*Vitis berlandieri* × *V. riparia* hybrid which is commonly known as less adapted to limited water conditions), while the second group was on 99 R (*V. berlandieri* × *V. rupestris* qualified as drought tolerant) [Galet 1979]. The final third one was own-rooted 'Italia' vines. The vines were individually cultivated in 40 l (solid volume) pots (35 cm diameter, 35 m height) filled with sterile peat (1.034% N, 0.94% P_2O_5, 0.64% K_2O pH 5.88, Klassman®) and perlite (0–3 mm in diameter) mixture in equal volume. The pots were isolated from the ground with plastic sheets. The vines were spur pruned to leave only the single bud (one main shoot) per plant. In the second year, while the vines in the fourth year life, they pruned to three buds (subsequently three shoots) per vine. The shoots were tied with thread to wires 2.5 m above the pots to let plants grow on a perpendicular position to ensure equally benefiting from the sunlight [Sabir 2013]. All the vines received the same annual amount of fertilizer (approx. 20 g N, 12 g P, 20 kg K, and 1.5 g Fe chelate per vine) from April to July.

Scheduling and achieving the irrigation regimes. Two types of irrigation regimes, full irrigation (FI) and continual deficit irrigation (DI), were applied to the vines. Irrigations were regulated according to soil water matric potential (Ψm) levels using tensiometers (The Irrometer Company, Riverside, CA) placed at a depth of 20 cm and approximately 12 cm from the trunk and were applied from bud break (early March) to the end of vegetation period (beginning of October) for two consecutive years. Field capacity levels were calculated in order to verify the accuracy of tensiometers for measuring soil moisture. For this, the randomly taken two pots filled with known volume of oven-dried growth media for each group of vines were irrigated up to field capacity before imposing different levels of soil moisture. To calculate the field capacity, the pots were placed in the large plastic buckets and irrigated with known quantity of water and kept for 6 h to attain the field capacity. After six hours, the amount of the drained water in the plastic bucket was measured and was subtracted from total amount of water applied initially [Satisha et al. 2006]. The obtained value was considered as the volume of the irrigation water that has to be applied to attain 100% field capacity (FI). Forty percentage amount of FI was considered as DI [Sabir and Kara 2010]. In these conditions, tensiometers were employed for a more realistic expression of soil water depletion in terms of Ψm following the slightly modified procedure described by Myburgh and van der Walt [2005]. Changes in Ψm were continuously monitored with daily readings at around 13:00 pm as well as before and after irrigations [Okamoto et al. 2004]. To ensure the uniformity of irrigation, the water was transported directly into the pots by microirrigation systems consisted of individual spaghetti tubes. Repeated readings during several days showed that the tensiometers readings at midday (13.00 pm) were constantly around 0.8–12 kPa (centibars) and 36–44 kPa for FI and DI conditions, respectively. For DI, irrigation was started when Ψm reached 44 kPa and was terminated when the calculated amount of water was applied to ensure 40% amount of field capacity. The start value of irrigation system for FI group vines were adjusted to 12 kPa to ensure that the full water amount of field capacity is given. Relatively higher air temperature in the glasshouse was also performed to simulate the typical semi-arid Mediterranean climate. During vegetation periods (15 March – 30 October) for two experimental years, daily air temperature and relative humidity, recorded using data logger (Ebro EBI 20 TH1) inside the glasshouse, were 25–38°C and 33–55%, respectively. During the hot and dry summers for both years (from the beginning of June to September), excessive heat accumulation in glasshouse was avoided by opening the roof and sidewall windows as well as slight whitewash painting (providing approx. 30% light reflection) to keep clusters and young leaves from burning. Under this condition, the instantaneous light intensity inside and outside the glasshouse was 37330 and 53330 lux (Lutron LX-105) at 13:00 p.m.

Leaf investigations. Leaf area (cm^2), leaf fresh weight (g), leaf dry weight (g), leaf chlorophyll content, and leaf relative water content (RWC) were investigated on fully expanded leaves of representative grapevines of each treatment [Tramontini et al. 2013]. Fifteen transpiring leaves per treatment were sampled early in the morning and immediately weighed to determine fresh weight. To determine single leaf area (LA, cm^2), one fully-expanded mature leaf from the mid-shoot area of each sampled plant were used in order to minimize age effects. Leaf area was estimated using WinFolia computer software program. Specific leaf area (SLA, cm^2 g^{-1}) was determined as the ratio of LA to DM. Leaf thickness (LT) was calculated as the ratio of FM to LA [de Pinheiro and Marcelis 2000] and leaf tissue density (D) as (DM/FM) * 1000 [Bacelar et al. 2006]. Chlorophyll contents of leaves (the 3rd and 4th leaf at the shoot tips) were estimated by using portable chlorophyll meter (Minolta SPAD-502, Japan). To investigate the relative water content (RWC) of the mature leaves, fifteen freshly sampled leaves were rehydrated by submerging in deionized water for 24 h in dark to obtain turgid weight [Yamasaki and Dillenburg 1999]. During the rehydration period, leaf samples were weighed periodically up to a constant value to ensure full rehydration. Measurements were performed after gently wiping the water from the leaf surface with tissue paper. At the end of rehydrationperiod, leaf samples were weighed to obtain final turgid mass (TM) and placed in an oven at 70°C for 48 h in order to obtain the dry mass (DM). All mass measurements were made using an analytical scale, with precision of 0.0001 g. Values of FM, TM, and DM were used to calculate RWC, using the equation suggested by Weatherley [1950]: RWC (%) = [(FM – DM)/(TM – DM)] * 100.

Temperatures of the 5th to the 7th basal leaf (T_{leaf}) were recorded daily at around 12 a.m. using an infrared thermometer (Ebro TFI-220) [Okamoto et al. 2004]. For this, the infrared thermometer was held perpendicularly to the center of the leaf at a distance of ≈25 cm to measure the temperature of a 25 cm^2 circular area of leaf surface. Measurements were performed between 12:00 and 13:30 h from six from south-facing, sun-exposed leaves for the 4 treatment vines in each replicate (i.e.12 vines) [Stavrinides et al. 2010]. Stomatal conductance (g_s) measurements were carried out on adult, non-senescent and well exposed to direct sunlight leaves using portable porometer (SC-1 Leaf Porometer) [Zufferey et al. 2011].

Statistical analysis. As the differences between the years for all parameter collected over two experimental years were insignificant, they were averaged before statistical analyses to avoid redundancy. Data were subjected to the variance analyses using SPSS 13.0 for Windows (SPSS Inc., Chicago, IL, USA) at $P < 0.05$ level. The mean values of three replications (with four leaves per replication) of each treatment were compared using the least significant difference (LSD) test. Significant differences among rootstock × irrigation interactions were presented in figures while rootstock or treatment effects were evaluated in tables when the interactions showed no significant differences.

RESULTS

Leaf growth. Leaf attributes of all the DI plants decreased (except for leaf dry weight) in comparison to the FI. LA (fig. 1) and SLA (fig. 2) were significantly affected by treatment x rootstock interaction. Combined data over years revealed that negative effects of water limitation were more pronounced in grapes cultivated on 5 BB rootstock, in comparison with those on 99 R or own roots. To illustrate, LA of 'Italia'/99 R vines subjected to DI decreased by 21.2%, such rates were 23.3 and 26.4% for vines on own roots and 5 BB, respectively. DI also resulted in significant decreases in SLA, quite similar to those of LA. Leaf fresh and dry weight characteristics as well as leaf number significantly affected by different irrigation regimes (tab. 1). Percent decrease in leaf fresh weight resulted from DI in 'Italia'/5 BB was 15.2% while the negative effect of DI were 6.2 and 10.5% among 'Italia'/99 R and own-rooted plants, respectively. Similar decreases were also investigated with respect to leaf dry weight, with the values 6.7, 11.9 and 14.9% for vines cultivated on 99 R, own-root and 5 BB, respectively. DI had significantly negative effects on leaf number per vines, regardless of the rootstock usage with similar decrease values 13.3, 13.7 and 13.8% for vines on 5 BB, 99 R and own-roots, respectively. According to the rootstock means, leaf fresh weight and leaf number values of 'Italia'/99 R vines were higher than others while dry weight was slightly higher in 'Italia'/5 BB vines.

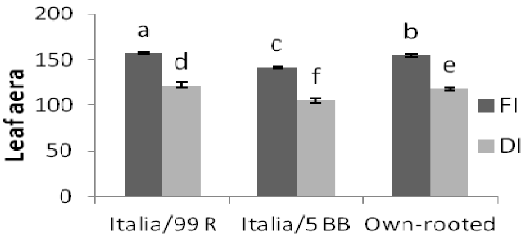

Fig. 1. Changes in leaf area (cm^2) as affected by irrigation level (FI: full irrigation, DI: deficit irrigation). Values of bars indicated by different letters identify significantly different groups ($P < 0.05$, LSD test)

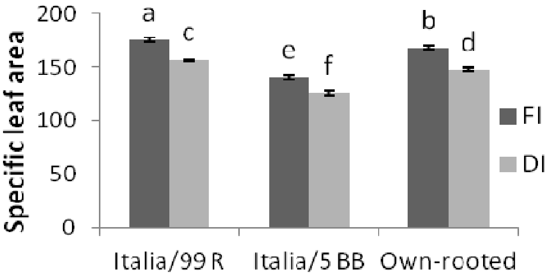

Fig. 2. Changes in specific leaf area (cm^2) as affected by irrigation level (FI: full irrigation, DI: deficit irrigation). Values of bars indicated by different letters identify significantly different groups (*P* < 0.05, LSD test)

T_{leaf}, chlorophyll concentrations and leaf thickness values across the 'Italia' vines significantly increased correspondingly with DI (tab. 2). However, the tissue density response of the vines to irrigation regimes were insignificant. Rootstocks significantly affected the increment levels of such leaf investigations. For example, the highest mean leaf temperature was determined in vines on 5 BB, followed by those on 99 R and the least value of own rooted ones (32.8, 28.2 and 26.5°C, respectively). Under DI irrigation condition, T_{leaf} values for vines on own-root, 99 R and 5 BB were 1.7, 2.9 and 4.0°C higher than those of FI groups.

Table 1. Leaf development response of own-rooted or grafted 'Italia' grape to different irrigation levels and rootstock

			Leaf Freshweight (g)	Leaf dryweight (g)	Leaf (node) number
Grafts or own root	'Italia'	FI	3.56 a	0.90 a	43.6 a
	99 R	DI	3.34 b	0.84 b	37.6 b
	LSD ($P \leq 0.05$)		0.26	0.08	1.37
	'Italia'	FI	3.57 a	1.01	37.5 a
	5 BB	DI	2.92 b	0.86	32.5 b
	LSD ($P \leq 0.05$)		0.14	ns	4.12
	'Italia'	FI	3.19	0.92	41.2 a
	Own root	DI	2.83	0.81	35.5 b
	LSD ($P \leq 0.05$)		ns	ns	3.97
Rootstocks	99 R		3.34 a	0.87 ab	40.6 a
	5 BB		3.25 a	0.93 a	35.0 b
	Own root		3.03 b	0.84 b	38.3 a
	LSD ($P \leq 0.05$)		0.18	0.08	2.39

FI – full irrigation, DI – deficit irrigation, ns – not significant. Means were combined over two sampling years (n = 12). Mean values indicated by different letters identify significantly different groups (*P* < 0.05, LSD test)

Table 2. Leaf feature response of own-rooted or grafted 'Italia' grape to different irrigation levels and rootstock

Grafts or own root		Leaf temperature (°C)	Chl in SPAD unit (mg kg^{-1})	Leaf thickness (mg cm^{2})	Tissue density (mg g^{-1})
'Italia'	FI	26.7 b	29.8 b	22.6 b	251.3
99 R	DI	29.6 a	32.4 a	28.3 a	252.0
LSD ($P \leq 0.05$)		1.02	0.85	4.12	ns
'Italia'	FI	30.5 b	30.4 b	25.3	282.2
5 BB	DI	34.5 a	33.8 a	27.3	293.6
LSD ($P \leq 0.05$)		2.51	1.56	ns	ns
'Italia'	FI	25.6 b	29.6	20.9	288.1
Own root	DI	27.3 a	31.5	24.2	286.1
LSD ($P \leq 0.05$)		1.09	ns	ns	ns
99 R		28.2 b	31.1 ab	25.5 a	251.6 b
5 BB		32.8 a	32.1 a	26.3 a	287.9 a
Own root		26.5 c	30.5 b	22.5 b	287.1 a
LSD ($P \leq 0.05$)		1.21	1.34	1.42	22.56

FI – full irrigation, DI – deficit irrigation, ns: not significant, Chl – chlorophyl. Means were combined over two sampling years (n = 12). Mean values indicated by different letters identify significantly different groups ($P < 0.05$, LSD test)

Stomatal conductance. g_s showed significant variation among the water treatments for all the grapevines (fig. 3). In fact, the g_s difference followed the T_{leaf} changes such that increases in leaf temperature were accompanied by decreases in g_s. DI regime resulted in significant decreases in g_s values across the vines with significant interaction of rootstock × treatment. To illustrate, under FI treatment, g_s values reached up to 189.0 mmol m^{-2} s^{-1} for vines on 5 BB and up to 178.8 mmol m^{-2} s^{-1} for those on 99 R. Whereas, the g_s values uder DI condition decreased to 149.1 and 154.0 mmol m^{-2} s^{-1} for the vines on 5 BB and 99 R, respectively. The decrease percentages were 21.1, 13.8 and 10.2% for vines on 5 BB, 99 R and own root, respectively.

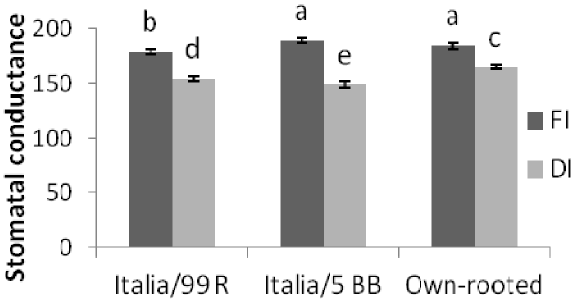

Fig. 3. Changes in stomatal conductance (mmol m^{-2} s^{-1}) as affected by irrigation level (FI – full irrigation, DI – deficit irrigation). Values of bars indicated by different letters identify significantly different groups ($P < 0.05$, LSD test)

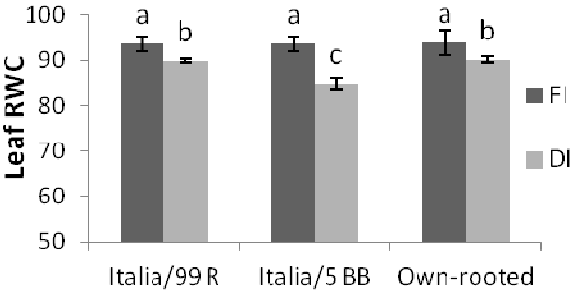

Fig. 4. Changes in leaf relative water content (RWC, %) as affected by irrigation level (FI – full irrigation, DI – deficit irrigation). Values of bars indicated by different letters identify significantly different groups ($P < 0.05$, LSD test)

Leaf relative water content. RWC was significantly affected by irrigation regimes (fig. 4). DI resulted in decreases in RWC across the vines and magnitudes of such decreases were rootstock-dependent. The leaf RWC values were 84.9, 89.9 and 90.1% for DI, and 93.7, 93.6 and 93.9% for FI for vines on 5 BB, 99 R and own roots. Decrease percentages in response to DI were 9.4, 4.1 and 3.9% for 'Italia'/5 BB vines, own roots, and 'Italia'/99 R, respectively. The highest negative effect of DI on leaf RWC was found in vines on 5 BB, while the lowest change was observed in those on 99 R.

DISCUSSION

Leaf parameters serve simple indicators of water stress since the leaves accurately respond to mild or moderate water deficit [Pellegrino et al. 2005], as was investigated in the present study in which DI caused reductions in leaf mass. In addition, plants have developed long- and short-term acclimation strategies to cope with water stress, such as reducing the leaf surface area, modifying the leaf anatomy, and conducting the essential nutrient elements [Schultz and Matthews 1988]. Decreases in LA and SLA of 'Italia' scion in response to DI verify the mentioned acclimation strategy, suggesting that water limitation led to a modification in canopy architecture as previously stated by Zsófi et al. [2011]. On the other hand, magnitude of the leaf growth restricting effect of DI varied among the vines on different root systems. Similar to growth features, the leaf number also decreased with DI treatment, in varying levels according to the rootstock use. Such differences in response to water shortage is not a surprise but vary significantly among the genotypes according to their adaptive strategies to water deficit [Baert et al. 2013, Corso and Bonghi 2014].

When the water shortage proceed, normally hydraulic and chemical signals (such as ABA) are sent from drying roots to the shoots and subsequently plant reduce water use by decreasing stomatal conductance which causes growth inhibition. Stomatal regulation of leaf gas exchange amongst regulation strategies, plays a pivotal role in plant response to water stress [Zufferey et al. 2011], although stomatal behavior is a complex phenomenon involving feedback controls which interact with a wide range of environmental conditions [Zweifel et al. 2007]. Leaf water potential, plant nutritional level and certain additional factors seem to modulate the sensitivity of stomata [Socías et al. 1997]. Decreases in leaf stomatal conductance along with long term exposure to DI in the present study might most likely related to lower photosynthetic area which will eventually become too low to support continuing optimal growth. Chlorophyll degradation has been frequently reported among different plants suffering water stress [Verhoeven et al. 1997]. In contrast, Alaei [2011] reported that drought stress increased the leaf chlorophyll content in wheat genotypes. This is most likely because the exact effect of deficit irrigation may vary according to the intensity of the water stress imposed. The chloroplast has been reported to have several adaptive mechanisms against drought [Biswal and Biswal 1999]. The chlorophyll increment found in the present study may indicate that DI at 40% field capacity led to better chlorophyll synthesis in the chloroplasts of the studied grapevines. Also, increase of tissue density in response to DI may result in escalation of leaf greenness.

As known, grapevine rootstocks can modify the leaf gas-exchange of the scion [Padgett-Johnson et al. 2003]. In the present study, DI caused 21.1% reduction in g_s of vines on 5 BB, while such decrease was 13.8% for vines on 99 R and 10.2% for own root vines. Considering the lower decrease in g_s of vines on 99 R in spite of the similar decrease rate response of all the vines in both shoot length and leaf area under DI effect, 99 R may more positively modulates the photosynthesis physiology of scion cultivar than 5 BB. This may be particularly important, as reported by Romero et al. [2013], in semiarid regions (like Mediterranean areas) in order to control excessive vegetative development without affecting photoassimilation during early shoot growth stage. However the present findings partly contradict with the results of Koundouras et al. [2008] who reported that the g_s values of 'Cabernet Sauvignon' wine grape were not affected by rootstocks 1103 P and S04 under DI. Nonetheless, grapevines are generally considered a drought-avoiding species, with an efficient stomatal control over transpiration [Chaves et al. 2010], although some genotypes show lower control [Soar et al. 2006]. Relatively lower decrease rates in g_s values compared to those occurred in vegetative development as response to DI imply that stomatal control and photosynthesis are more resistant to the drought stress than vegetative growth. Nonetheless, bearing in mind the available literature data including the present findings, it seems plausible that stomatal responses to water deficits in a specific variety will vary according to the particular combination of the rootstock [Padgett-Johnson et al. 2003], the climatic variables [Sabir and Yazar 2015], and the intensity and duration of water deficits [Costa et al. 2012]. The explanation of these differences may be found from the intrinsic characteristics of these two different rootstocks which might had direct influence on water absorption and root hydraulic conductivity. It has been well-known that reduction in root hydraulic conductivity after soil water depletion [Serra et al. 2013] lead to lower stomatal conductance [Domec et al. 2009, Tsegay et al. 2014]. Furthermore, leaf area development which decreased in response to DI, also lowered the stomatal conductance in this study.

Relative water content (RWC) of the plant leaf has long been used as an appropriate measure of plant water status in terms of the physiological consequence of cellular water deficit as it is a satisfactory basis for relating cellular water status to metabolism. In the present study, DI resulted in remarkable decreases in RWC of the scion *cv*. 'Italia' and the changes were depending on the rootstocks. Higher decrease rate in RWC of DI-subjected 'Italia'/5 BB vines is in concordance with many other low tolerance response of 5 BB vines investigated in the present study. Generally, decreasing RWC below *ca.* 75–85% progressively impairs photosynthetic metabolism in plants [Peňuelas et al. 1993]. In a field study, the response of grape leaves to drought shown that their photosynthetic metabolism was impaired below *c.* 75% RWC [Escalona et al. 1999]. Similarly, Bongi and Palliotti [1994] reported that a reduction in RWC from 96 to 80% caused a conspicuous reduction in photosynthetic CO_2 assimilation in olives. Considering the mentioned decrease percentages above, in the present study the leaf RWC values of overall vines were not at critical levels that impair the photosynthesis.

Previously, drought-dependent variability in T_{leaf} has been mentioned by Durigon and van Lier [2013]. In the present study, DI caused increments in T_{leaf} in varying levels according to the use of different root systems. To illustrate, difference of T_{leaf} was lower between FI and DI in own root or ‘Italia’/99 R compared to larger difference in those of ‘Italia’/5 BB. Marguerti et al. [2012] demonstrated that scion leaf temperature, transpiration rate and its acclimation to water deficit are controlled genetically by the rootstock. Therefore, differences in T_{leaf} of ‘Italia’ vines on different root systems may possible be related with different effects of rootstocks on transpiration rates and cooling of the scion. In spite of the mentioned differences, the range of T_{leaf} was between the threshold values for optimum photosynthesis (25–30°C) suggested by Greer [2012] for grapevine. This may indicate that DI at 40% field capacity did not significantly affect photosynthesis. In contrast to the highest decrease rates in g_s investigated in the grapevines cultivated on the 5 BB in response to DI, the highest increase rate in chlorophyll values and T_{leaf} was recorded when 5 BB was used as root system. The g_s and chlorophyll responses of own-rooted and ‘Italia’/99 R vines were smaller than that of 5 BB. These findings may indicate the higher sensitivity of 5 BB to water depletion in the soil in comparison to 99 R. Although the chlorophyll contents increased in water stress, the overall vegetative growth depressed. It has been assumed that under stress conditions the internal concentration of CO_2 may rise which results in turn decrease the photsystem II activity (decrease in electron transport) [Ramanjulu et al. 1998]. According to the detailed physiological investigations performed by Sun et al. [2006], water deficit may affect the balance between CO_2 fixation and light absorption within chloroplasts during photosynthesis, resulting in further accumulation of excessive light energy and intensifying photoinhibition.

CONCLUSION

Considering the overall pooled data, (a) water deficit remarkably induced the dehydration of the leaves and decreased the stomatal conductance which caused leaf growth inhibition. (b) The ‘Italia’ cultivar leaves differently responded to water depletion in relation to rootstock use. (c) Negative effects of water limitation were more pronounced in ‘Italia’ vines cultivated on 5 BB rootstock, whereas the vines on 99 R displayed more tolerant growth and physiology to drought in comparison with 5 BB. Therefore the use of 99 R seems more appropriate than 5 BB to cope with negative effects of drought. It is proposed that studies on responses of vines to drought in new scion/rootstock combinations will gain importance over time as producers try to increase yield and quality despite the ever-increasing water shortage on the faces of climate change.

REFERENCES

Alaei, Y. (2011). The Effect of Amino acids on leaf chlorophyll content in bread wheat genotypes under drought stress conditions. Middle-East J. Sci. Res., 10, 99–101.

Bacelar, E.A., Santos, D.L., Moutihno-Pereira, J.M., Goncalves, B.C., Ferreira, H.F., Correia, C.M. (2006). Immediate responses and adaptive strategies of three olive cultivars under contrasting water availability regimes: changes on structure and chemical composition of foliage and oxidative damage. Plant Sci., 170, 596–605.

Baert, A., Villez, K., Steppe, K. (2013). Automatic drought stress detection in grapevines without using conventional threshold values. Plant Soil, 369, 439–452.

Bates, B.C., Kundzewicz, Z.W., Wu, S., Palutikof, J.P. (2008). Climate Change and Water. Technical Paper of the Intergovernmental Panel on Climate Change, IPCC Secretariat, Geneva, 210 pp.

Biswal, B., Biswal, U.C. (1999). Photosynthesis under stress: stress signals and adaptive response of chloroplasts. In: Handbook of plant and crop stress, Pessarakli M, (ed.). New York, Marcel Dekker, Inc., 315–336.

Bongi, G., Palliotti, A. (1994). Olive. In: Handbook of environmental physiology of fruit crops: Temperate crops, vol. I, Shaffer, B., Anderson, P.C. (eds). CRC Press, Boca Raton, pp. 165–187.

Chaves, M.M., Zarrouk, O., Francisco, R., Costa, J.M., Santos, T., Regalado, A.P., Rodrigues, M.L., Lopes, C.M. (2010). Grapevine under deficit irrigation: hints from physiological and molecular data. Ann. Bot-London, 105, 661–676.

Collins, R., Kristensen, P., Thyssen, N. (2009). Water resources across Europe – confronting water scarcity and drought. European Environmental Agency (EEA) Report series. N. 2/2009. ISSN 1725–9177, 55p.

Corso, M., Bonghi, C. (2014). Mini review: Grapevine rootstock effects on abiotic stress tolerance. Plant Sci. Tod., 1, 108–113.

Costa, J.M., Ortuño, M.F., Lopes, C.M., Chaves, M.M. (2012). Grapevine varieties exhibiting differences in stomatal response to water deficit. Funct. Plant Biol., 39, 179–189.

de Pinheiro-Henriques, A.R., Marcelis, L.F.M. (2000). Regulation of growth at steady state nitrogen nutrition in lettuce (*Lactuca sativa* L.): Interactive effects of nitrogen and irradiance. Ann. Bot., 86, 1073–1080.

Domec, J., Noormets, A., King, J., Sun, G., McNulty, S., Gavazzi, M., Boggs, J., Treasure E. (2009). Decoupling the influence of leaf and root hydraulic conductances on stomatal conductance and its sensitivity to vapour pressure deficit as soil dries in a drained loblolly pine plantation. Plant Cell Environ., 32, 980–991.

Durigon, A., van Lier, Q.J. (2013). Canopy temperature versus soil water pressure head for the prediction of crop water stress. Agric. Water Manag., 127, 1–6.

Escalona, J.M., Flexas, J., Medrano, H. (1999). Stomatal and non-stomatal limitations of photosynthesis under water stress in field-grown grapevines. Aust. J. Plant Physiol., 26, 421–433.

Flexas, J., Bota, J., Loreto, F., Cornic, G., Sharkey, T.D. (2004). Diffusive and metabolic limitations to photosynthesis under drought and salinity in C(3) plants. Plant Biol. (Stuttg), 6, 269–279.

Flexas, J., Medrano, H. (2002). Drought-inhibition of photosynthesis in C-3 plants: stomatal and non-stomatal limitations revisited. Ann. Bot., 89, 183–189.

Galet, P.A. (1979). A practical ampelography (translated by L.T. Morton). Cornell University Press, Ithaca, NY, USA.

González-Fernández, A.B., Rodríguez-Pérez, J.R., Marabel, M., Álvarez-Taboada, F. (2015). Spectroscopic estimation of leaf water content in commercial vineyards using continuum removal and partial least squares regression Sci. Hortic., 188, 15–22.

Greer, D.H. (2012). Modelling leaf photosynthetic and transpiration temperature-dependent responses in *Vitis vinifera* cv. Semillon grapevines growing in hot, irrigated vineyard conditions. AoB Plants, doi:10.1093/aobpla/pls009.

Jones, H.G., Vaughan, R.A. (2010). Remote sensing of vegetation: principles, techniques, and applications. Oxford University Press, Oxford.

Kounduras, S., Tsialtas, I.T., Zioziou, E., Nikolaou, N. (2008). Rootstock effects on the adaptive strategies of grapevine (*Vitis vinifera* L. *cv*. Cabernet-Sauvignon) under contrasting water status: Leaf physiological and structural responses. Agr. Ecosyst. Environ., 128, 86–96.

Lovisolo, C., Perrone, I., Carra, A., Ferrandino, A., Flexas, J., Medrano, H., Schubert, A. (2010). Drought-induced changes in development and function of grapevine (Vitis spp.) organs and in their hydraulic and non hydraulic interactions at the whole plant level: a physiological and molecular update. Func. Plant Biol., 37, 98–116.

Lovisolo, C., Schubert, A., Sorce, C. (2002). Are xylem radial development and hydraulic conductivity in downwardly-growing grapevine shoots influenced by perturbed auxin metabolism? New Phytol., 156, 65–74.

Marguerti, E., Brendel, O., Lebon, E., van Leewen, C., Ollat, N. (2012). Rootstock control of scion transpiration and its acclimation to water deficit are controlled by different genes. New Phytol., 194, 416–429.

Myburgh, P.A., van der Walt, L.D. (2005). Cane water content and yield responses of *Vitis vinifera* L. cv. Sultanina to overhead irrigation during the dormant period. S. Afr. J. Enol. Vitic., 26, 1–5.

Okamoto, G., Kuwamura, T., Hirano, K. (2004). Effects of water deficit stress on leaf and berry ABA and berry ripening in Chardonnay grapevines. Vitis, 43, 15–17.

Padgett-Johnson, M., Williams, L.E., Walker, M.A. (2003). Vine water relations, gas exchange, and vegetative growth of seventeen *Vitis* species grown under irrigated and nonirrigated conditions in California. J. Am. Soc. Hort. Sci., 128, 269–276.

Pellegrino, A., Lebon, E., Simonneau, T., Wery, J. (2005). Towards a simple indicator of water stress in grapevine (*Vitis vinifera* L.) based on the differential sensitivities of vegetative growth components. Aust. J. Grape Wine Res., 11, 306–315.

Peňuelas, J., Filella, I., Biel, C., Serrano, L., Save, R. (1993). The reflectance at the 950–970 nm region as an indicator of plant water status. Int. J. Remote Sens., 14, 1887–1905.

Ramanjulu, S., Sreenivasulu, N., Sudhakar, C. (1998). Effect of water stress on photosynthesis in two mulberry genotypes with different drought tolerance. Photosynthetica, 35, 279–283.

Romero, P., Gil-Muňoz, R., del Amor, F.M., Valdés, E., Fernández, J.I., Martinez-Cutillas, A. (2013). Regulated deficit irrigation based upon optimum water status improves phenolic composition in Monastrell grapes and wines. Agric. Water Manag., 121, 85–101.

Sabir, A. (2013). Improvement of grafting efficiency in hard grafting grape *Berlandieri* hybrid rootstocks by plant growth-promoting rhizobacteria (PGPR). Sci. Hortic., 164, 24–29.

Sabir, A., Dogan, Y., Tangolar, S., Kafkas, S. (2010). Analysis of genetic relatedness among grapevine rootstocks by AFLP (Amplified Fragment Length Polymorphism) markers. J. Food Agric. Environ., 8, 210–213.

Sabir, A., Kara, Z. (2010). Silica gel application to control water runoff from rootzone microenvironment's climate of grapevine rootstocks grown under drought condition. International Sustainable Water and Wastewater Management Symposium 2, 1365–1372, 26–28 Oct. Konya, Turkey.

Sabir, A., Yazar, K. (2015). Diurnal dynamics of stomatal conductance and leaf temperature of grapevines (*Vitis vinifera* L.) in response to daily climatic variables. Acta Sci. Pol. Hortorum Cultus, 14, 3–15.

Satisha, J., Prakash, G.S., Venugopalan, R. (2006). Statistical modeling of the effect of physio-biochemical parameters on water use efficiency of grape varieties, rootstocks and their stionic combinations under moisture stress conditions. Turk. J. Agric. For., 30, 261–271.

Schultz, H.R., Matthews, M.A. (1988). Resistance to water transport in shoots of *Vitis vinifera* L. Relation to growth at low water potential. Plant Physiol., 88, 718–724.

Serra, I., Strever, A., Myburg, P.A., Deloire, A. (2013) Review: the interaction between rootstocks and cultivars (*Vitis vinifera* L.) to enhance drought tolerance in grapevine. Aust. J. Grape Wine Res., doi:10.1111/ajgw.l2054.

Skirycz, A., Inze, D. (2010). More from less: plant growth under limited water. Curr. Opin. Biotechnol., 21, 197–203.

Soar, C.J., Speirs, J., Maffei, S.M., Penrose, A.B., McCarthy, M.G., Loveys, B.R. (2006). Grape vine varieties Shiraz and Grenache differ in their stomatal response to VPD: apparent links with ABA physiology and gene expression in leaf tissue. Aust. J. Grape Wine. Res., 12, 2–12.

Socías, F.X., Correia, M.J., Chaves, M.M., Medrano, H. (1997). The role of abscisic acid and water relations in drought responses of subterranean clover. J. Exp. Bot., 48, 1281–1288.

Stavrinides, M.C., Daane, K.M., Lampinen, B.D., Mills, N.J. (2010). Plant water stress, leaf temperature, and spider mite (Acari: *Tatranychidae*) outbreaks in California vineyards. Environ. Entomol., 39, 1232–1241.

Sun, Y., Xu, W.J., Fan, A.L. (2006). Effects of salicycacidon chlorophyll fluorescence and xanthophylls cycle in cucumber leaves under high temperature and strong light. Chin. J. Appl. Ecol., 17, 399–402 [in Chinese].

Tramontini, S., van Leuwen, C., Domec J. C., Irvine, A. D., Basteau, C., Vitali, M., Schulz, O. M., Lovisolo, C. (2013). Impact of soil texture and water availability on the hydraulic control of plant and grape-berry development. Plant Soil, 368, 215–230.

Tsegay, D., Amsalem, D., Almeida, M., Crandles, M. (2014) Review: Responses of grapevine rootstocks to drought stress. Inter. J. Plant Physiol., 6, 1–6.

Vandeleur, R.K., Mayo, G., Shelden, M.C., Gilliham, M., Kaiser, B.N., Tyerman, S.D. (2009). The role of plasma membrane intrinsic protein aquaporins in water transport through roots: diurnal and drought stress responses reveal different strategies between isohydric and anisohydric cultivars of grapevine. Plant Physiol., 149, 445–460.

Verhoeven, A.S., Demmig-Adams, B., Adams, W.W. (1997). Enhanced employment of xanthophyll cycle and thermal energy dissipation in spinach exposed to high light and N-stress. Plant Physiol., 113, 817–824.

Vicente-Serrano, S.M., Lopez-Moreno, J.I., Beguería, S., Lorenzo-Lacruz, J., Sanchez-Lorenzo, A., García-Ruiz, J.M., Azorin-Molina, C., MoránTejeda, E., Revuelto, J., Trigo, R., Coelho, F., Espejo, F. (2014). Evidence of increasing drought severity caused by temperature rise in Southern Europe. Environ. Res. Lett., 9:044001. doi:10.1088/1748-9326/9/4/044001.

Weatherley, P.E. (1950). Studies in the water relations of the cotton plant. I. The field measurement of water deficits in leaves. New Phytolog., 49, 81–87.

Yamasaki, S., Dillenburg, L.R. (1999). Measurements of leaf relative water content in *Araucaria angustifolia*. Revis. Brasil. de Fisiol. Veget., 11, 69–75.

Zsófi, Z.S., Tóth, E., Rusjan, D., Bálo, B. (2011). Terroir aspects of grape quality in a cool climate wine region: Relationship between water deficit, vegetative growth and berry sugar concentration. Sci. Hortic., 127, 494–499.

Zufferey, V., Cochard, H., Ameglio, T., Spring, J.L., Viret, O. (2011). Diurnal cycles of embolism formation and repair in petioles of grapevine (*Vitis vinifera* cv. Chasselas). J. Exp. Bot., doi:10.1093/jxb/err081).

Zweifel, R., Steppe, K., Sterck, F.J. (2007). Stomatal regulation by microclimate and tree water relations: interpreting eco-physiological field data with a hydraulic plant model. J. Exp. Bot., 58, 2113–2131.

FIZJOLOGICZNE I MORFOLOGICZNE REAKCJE LIŚCI WINOROŚLI (*V. vinifera* L. CV. 'ITALIA') NA DEFICYT WODY POD WPŁYWEM RÓŻNYCH PODKŁADEK

Streszczenie. Ekstremalne warunki pogodowe z przedłużającymi się suchymi okresami i wysoką temperaturą mogą znacznie wpłynąć na fizjologię i morfologię winorośli. Zrozumienie fizjologicznych i morfologicznych reakcji winorośli na deficyt wody ma więc ogromne znaczenie dla kształtowania odpowiedniego rozwoju roślin. Niniejsze badanie ocenia wpływ niedostatecznego nawadniania (DI) na pewne cechy liście winorosli odmiany 'Italia' uprawianej na różnych podkładkach. DI miał istotny wpływ na rozwój, morfologię, strukturę tkanek, status wodny i fizjologię liści winorosli. Reakcja odmiany 'Italia' na DI zależała od stosowanej podkładki, np. świeża masa liści odmiany 'Italia'/5 BB pod wpływem DI zmienijszyła się o 15,2% w porównaniu z pełnym nawadnianiem (FI), natomiast wartości świeżej masy dla odmiany 'Italia'/99R i winorośli zakorzenionych na własnych korzeniach pod wpływem DI zminiejszyły się, odpowiednio, o 6,2 i 10,5%. Pod wpływem FI, przewodność szparkowa (g_s) osiągnęła wartości 189,0 mmol m^{-2} s^{-1} dla 'Italia'/5 BB i 178.8 mmol m^{-2} s^{-1} dla 'Italia'/99 R. Wartości g_s w warunkach DI wynosiły odpowiednio 178,1 i 164,0 mmol m^{-2} s^{-1} dla winorośli na 5 BB i 99 R. Przewodność szparkowa zmniejszyła się odpowiednio o ok. 21,1, 13,8 i 10,2% na winorośli uprawianej na 5 BB, 99 R oraz na własnych korzeniach. W reakcji na DI względna zawartość wody w liściach zminiejszyła się odpowiednio o 9,4, 4.1 i 3,9% dla winorośli 'Italia'/5 BB, winorośli na własnych korzeniach oraz 'Italia'/99 R. Wieloletnie dane wykazały, że winorośle uprawiane na 99 R miały bardziej tolerancyjny wzrost liści i fizjologię w reakcji na suszę w porównaniu z winoroślami na 5 BB.

Słowa kluczowe: *Vitis*, susza, fizjologia liści, przewodność szparkowa

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