



Effects of regulated and sustained deficit irrigation on water use, physiology and yield of ‘Menara’ olive trees, in Morocco

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Abstract

The olive tree (*Olea europaea* L.) is culturally and economically vital in Morocco. However, its sustainability is threatened by aridity and water scarcity. Studying its response to different irrigation strategies is crucial for sustainable cultivation and improved water use efficiency in the face of future drought events. This work aimed to study the responses of sap flow rate, physiological, and agronomic parameters of the Moroccan olive cultivar ‘Menara’ to Regulated Deficit Irrigation (RDI) and Sustained Deficit Irrigation (SDI) strategies. Seven irrigation regimes were studied based on the sensitivity of phenological phases to water stress, distinguished as (SP) ‘Sensitive Period’ and (NP) ‘Normal Period’. SP involves flowering (SP1) and oil synthesis to harvest (SP2), while NP relates to pit hardening. ‘Menara’ olive trees were subjected to four RDI treatments: T1 (SP 100- NP 70% ETC), T2 (SP 100- NP 60% ETC), T3 (SP 80- NP 70% ETC), and T4 (SP 80- NP 60% ETC), and two SDI treatments: T5 (70% ETC) and T6 (60% ETC), compared with control (T0) trees under full irrigation (100% ETC). In comparison to the control T0, the deficit irrigation treatments exhibited lower sap flow rates. Specifically, T1 and T2 experienced reductions of 10% and 19% in sap flow rates, respectively, attributed to a decrease in water application of 11% and 14% compared to T0. Despite this decline, T1 and T2 demonstrated fruit yields comparable to T0. Conversely, T4, which received 28% less irrigation, displayed a yield reduction of approximately 23% compared to T0 in 2022. Moreover, adverse effects were observed in Menara olive trees treated with T4 after two consecutive seasons of deficit irrigation in 2023, indicating that prolonged stress effects could be detrimental in subsequent years. T3, under RDI, showed resilience with a 13% reduction in production despite a 37% decrease in sap flow rate and a 24% water restriction. Conversely, T5 and T6, employing SDI, experienced significant yield declines of 50%, with reductions in water application of 30% and 40% and sap flow rate of 51% and 80%, respectively, in 2022. The alternate bearing pattern significantly impacts Menara olive production, as evidenced by reduced sap flow and yield in the “off” year of 2023, regardless of irrigation strategies. A strong correlation ($R^2=0.84$) between sap flow and yield indicates that well-irrigated olive trees tend to transpire more, leading to higher yields. Stomatal conductance (gs) notably decreases with increased water deficit, with reductions of 8%, 12%, and 23% observed in T4, T5, and T6, respectively. Furthermore, a significant reduction in Fv/Fm, indicative of water stress, was observed with a 40% decrease in water supply in the T6 treatment group during both irrigation seasons in 2022 and 2023, with Fv/Fm reaching approximately 0.7. In general, Menara olive trees subjected to deficit irrigation, particularly under the T3 RDI treatment, showed the ability to adapt and cope with low water supply over time. However, the cumulative water shortage effect of the SDI treatment T6 resulted in a decline in both the agronomic and physiological performance of this cultivar.

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Introduction

The olive tree (*Olea europaea L.*) is the most iconic in the Mediterranean Basin, with Spain, Tunisia, Italy, Morocco, and Greece being the primary cultivators (Arenas-Castro et al. 2020; FAOSTAT 2018). However, agriculture in these regions faces challenges due to water scarcity and climate change, which are expected to worsen, impacting crop production (Feres and Soriano 2006; Tanasijevic et al. 2014; Fraga et al. 2020). The climate is typically arid or semiarid, with spatiotemporal unpredictability of precipitation and low rainfall, indicating the need for irrigation to guarantee production (Er-Raki et al. 2010; Tanasijevic et al. 2014). Moreover, increasing global food demand and socioeconomic factors are shifting towards more intensive cropping systems requiring irrigation and fertilization (Villalobos et al. 2006). From this perspective, irrigation stands out as one of the most crucial alternatives for climate change adaptation. Therefore, water-use efficiency is essential in irrigated agriculture due to the lower water demand from water resources.

Deficit irrigation is an important strategy for minimizing irrigation water use. According to Mairech et al. (2020), deficit irrigation has the potential to be a sustainable management solution under current conditions, as it reduces irrigation requirements while enhancing crop water use efficiency. For this purpose, improved water-use efficiency can be achieved by combining high-efficiency irrigation systems, such as trickle irrigation, with water-deficit irrigation strategies to match commercial objectives in the olive sector. Among these strategies, regulated deficit irrigation (RDI) and sustained deficit irrigation (SDI) stand out (Moriana et al. 2003). Sustained deficit irrigation involves applying a constant volume of water less than the evapotranspiration demand during the entire irrigation season. Optimizing the timing of irrigation is essential for minimizing crop losses and maximizing yields (Iniesta et al. 2009). Recently, growers have been exploring new techniques to optimize crop water use efficiency by adopting new irrigation strategies, such as regulated deficit irrigation (RDI), which has been accepted and popularized by the majority of horticultural crop growers (Yang et al. 2022). Iniesta et al. (2009) stated that this strategy is a viable alternative to full irrigation, and several authors have reported that the RDI strategy requires less water but has a similar or greater production than full irrigation, providing significant savings in irrigation water (Fernández et al. 2013). This strategy relies on knowledge of the crop responses to water stress at different phenological phases to help identify the periods when fruit trees are less sensitive (Fernández et al. 2013; Gucci et al. 2019; Moriana et al. 2003). Consequently, optimizing irrigation in olive orchards during sensitive periods such as

flowering and oil synthesis can mitigate the effects of water scarcity and sustain olive yield (Feres and Soriano 2006; Fernandes et al. 2018; Diaz-Espejo et al. 2018). For olive trees, water stress early in the growing season can significantly affect trees' growth and olive productivity, leading to economic losses due to interference with flowering and fruit setting (Hueso et al. 2021). During the spring season, spanning from budburst until fruit drop, numerous physiological processes, including the differentiation of inflorescences and flower structures, flowering, fruit set, and fruit drop, influence the ultimate fruit yield. Therefore, an irrigation deficit during this critical stage significantly reduces production (Pastor 2005). However, the pit hardening stage is the most resistant to water deficit (Goldhamer 1999; Gucci et al. 2019; Iba et al. 2023a). Moreover, several authors reported that RDI has been applied successfully from the pit hardening phase until the beginning of the oil accumulation phase in olive oil cultivars in the summer in the Mediterranean Basin (e.g., Tognetti et al. 2006; Lavee et al. 2007; Feres et al. 2012; Gómez del-Campo and García 2013; Gucci et al. 2019). Research on irrigation in olive cultivation has identified a second sensitive period, particularly during the oil synthesis phase, where water deficits notably reduce production and impact the quality of oil (García et al. 2020; Hueso et al. 2019). At the end of summer and beginning of autumn until harvest, active photosynthesis is required for the production and transport of the sugar alcohol mannitol to fruit for oil synthesis. A study by Tognetti et al. (2006) in southern Italy showed a linear relationship between irrigation amount and oil production during early autumn.

Previous investigations across various olive cultivars have consistently shown that restricting irrigation leads to water savings but also results in yield reductions (Lavee et al. 2007; Servili et al. 2007; Gómez del-Campo and García 2013; Caruso et al. 2017; Gucci et al. 2019; Serman et al. 2021). Within the context of our study, deficit irrigation (DI) strategies have been developed to align with commercial objectives in the olive sector, aiming to balance water savings with optimal yield. Optimizing sustained and regulated deficit irrigation involves finding treatments that strike the best balance between fruit yield, oil yield, and water conservation. Pierantozzi et al. (2013) suggest that providing full or elevated levels of water availability during the spring months can help protect photosynthetic pigments from oxidative degradation. They recommend maintaining either 100% or 75% of the estimated ETC to achieve this. Doing so can significantly improve CO₂ assimilation rates, leading to increased productivity. Hueso et al. (2021) reported significantly lower numbers of flower buds in rainfed and 40% ETC of 'Arbequina' trees than those in fully irrigated trees. Additionally, previous research (Moriana et al. 2003; Fernández et al. 2013; Gómez del-Campo and García 2013)

has shown that applying moderate RDI to olive trees, notably from fruit drop until the beginning of the oil synthesis stage when evapotranspiration is greatest (June and the first weeks of August in the Mediterranean area), leads to a balance between water savings, yield, and oil quality and can be applied with little effect on production. Studies conducted by Correa-Tedesco et al. (2010) demonstrate that applying 66% ETc over the growing season to a table olive cultivar ‘Manzanilla fina’ led to mild water stress, resulting in a 25% reduction in fruit yield compared to well-watered trees. Our study applies specific percentages of water reduction at specific phenological stages of olive trees development, such as 100% or 80% of estimated ETc during sensitive periods of flowering and oil synthesis and 70% or 60% of estimated ETc during the period from the beginning of pit hardening to the beginning of oil synthesis, to achieve optimal water conservation while maintaining acceptable yields.

Improving irrigation management requires a comprehensive understanding of transpiration (Oliveira Reis et al. 2009). Trunk sap flow serves as a reliable marker of tree transpiration, reflecting the physiological characteristics of trees and playing a crucial role in maintaining hydraulic transport between the soil and atmosphere (McDowell et al. 2008; Hernandez-Santana et al. 2016a). This approach has been frequently utilized by several investigations to determine transpiration rates in olive orchards across diverse conditions, particularly for evaluating water stress under deficit irrigation scenarios (Alcaras et al. 2016; Cammalleri et al. 2013; Charfi Masmoudi et al. 2011; Tognetti et al. 2004; Vandegehuchte et al. 2012; Kokkotos et al. 2021). The fluctuations in trunk sap flow and stem flow at a daily scale may reflect the plant’s water use response to changing environmental conditions (Zweifel and Hasler 2001). Transpiration rates, as measured by sap flow sensors, are strongly affected by the intensity of water stress in grapevines (Benyahia et al. 2023). According to Rousseaux et al. (2009), a study on sustained deficit irrigation in an olive orchard with the cultivar ‘Manzanilla’ revealed a 30% decrease in sap flow when applying 66% ETc. Additionally, regulated deficit irrigation (RDI) resulted in decreased sap flow in mango trees, as reported by Cotrim et al. (2019). Furthermore, during periods of water stress, olive trees typically experience reductions in transpiration, stomatal conductance, and net photosynthesis (Giorio et al. 1999; Bosabalidis and Kofidis 2002; Hernandez-Santana et al. 2016a; b; Yang et al. 2022; Zavadilová et al. 2023). In their study of the RDI strategy applied to the Manzanilla fina cultivar, Alcaras et al. (2016) observed that the decreases in sap flow appeared to largely follow the decreases found in leaf conductance. Li et al. (2017) and Bhusal et al. (2019) reported that moderate drought stress significantly decreased stomatal conductance, transpiration rate, and maximum photochemical

efficiency (Fv/Fm). For the majority of plants, normal healthy leaves in stress-free conditions have an Fv/Fm value of around 0.8 (Björkman and Demmig 1987). Boussadia et al. (2023) found, through chlorophyll fluorescence analysis, that Fv/Fm served as the strongest loading factor for screening drought stress among five olive tree cultivars. Exposure of leaves to increasing drought stress resulted in subsequent decreases in Fv/Fm. This decrease in Fv/Fm indicates a down-regulation of PSII, which is a protective or regulatory mechanism to prevent photodamage to the photosynthetic apparatus (Boussadia et al. 2008). In recent decades, novel experimental techniques utilizing sap flow measurements have enhanced the precision of quantifying tree water consumption. Sap flow sensors provide accurate estimates of daily transpiration rates, responding quickly to changes in both environmental and drought levels (Benyahia et al. 2023). Sap flow sensors are promising tools for detecting water stress in olives and optimizing irrigation management in olive groves, as outlined in prior agricultural literature (Granier 1985; Granier and Gross 1987a, b; Molina et al. 2019).

The present study aims to 1) evaluate the impact of regulated and sustained deficit irrigation strategies on the ‘Menara’ cultivar in Morocco over two consecutive years (2022 and 2023) by assessing sap flow, physiological, and agronomical parameters and 2) identify the optimal water irrigation strategy for the local variety under water stress conditions, with the aim of improving its adaptation to water deficit and enhancing productivity.

Materials and methods

Experimental orchard and climate

This study was conducted in a 12-year-old Menara olive orchard located at the Saada Research Station of the National Institute of Agronomic Research in Marrakech, Morocco, throughout the years 2022 and 2023. The orchard is located near Marrakech city (31°37′33.6″ N, 8° 96′ 08.45.6″ W, 411 m a.s.l.). The trees were planted in a square-spaced scheme with an 8 m side length and a density of 156 trees ha⁻¹ and were irrigated through drip irrigation (Fig. 1). The irrigation method for the olive orchard changed from basin irrigation to drip irrigation in 2017. The soil at the experimental site has a clay texture, a pH of 7.82, and an organic matter content of 2.33%.

In this region, the climate is Mediterranean and defined by an arid to semiarid climate with hot summers, low rainfall, and irregular spatiotemporal precipitation throughout the year (Er-Raki et al. 2010). The annual precipitation was 110 mm in 2022 and 101 mm in 2023, with corresponding

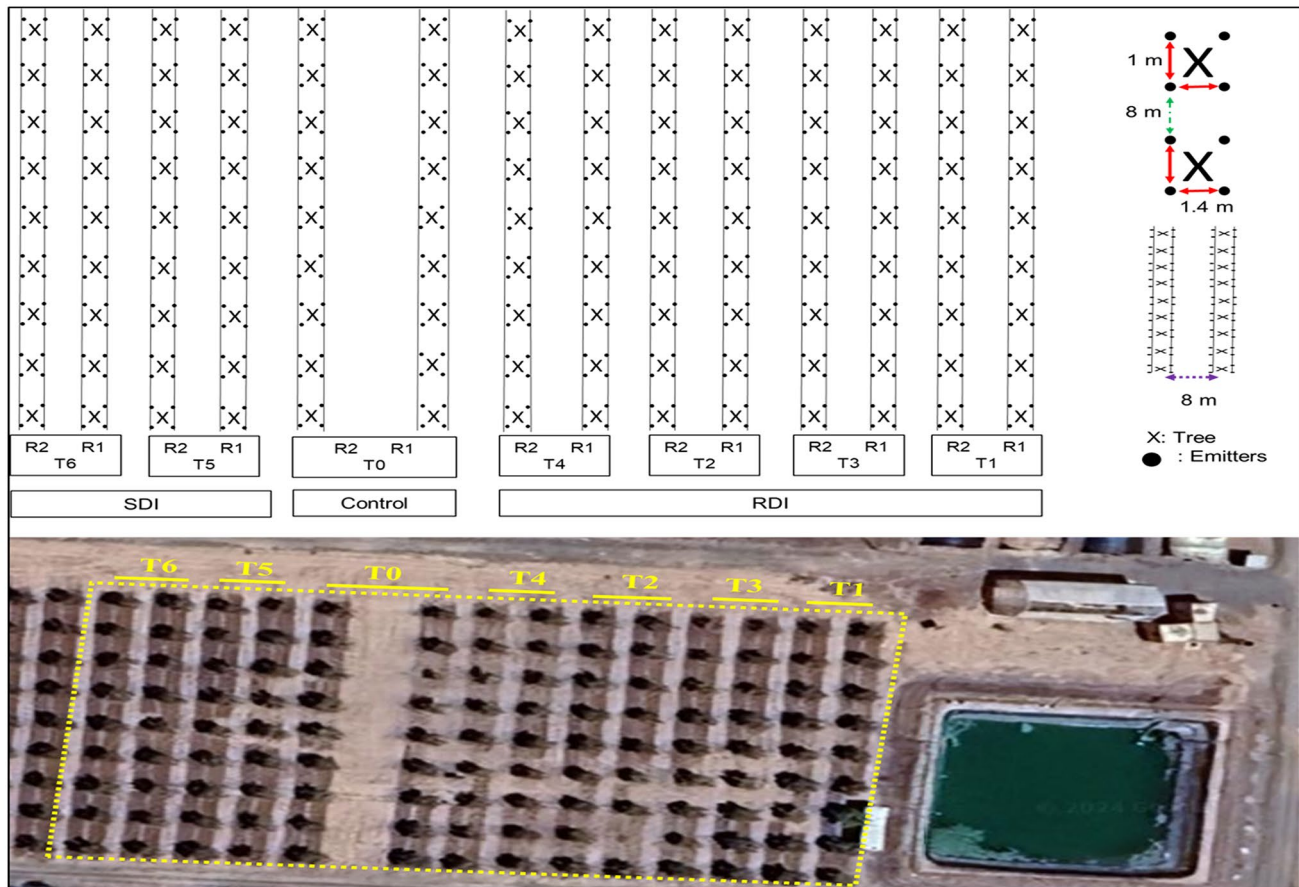


Fig. 1 A scheme depicting the experimental site layout for different deficit irrigation treatments, juxtaposed with an aerial view of the research station wherein the experimental trees are highlighted in yellow. Four emitters per tree were installed along two drip irrigation tubing lines (two emitters per side along the row, 1 m apart). The spatial

dimensions, including the emitter spacing, drip irrigation tube arrangement, and row and tree spacings, are described in the figure. Graphical representations in this and subsequent figures were generated using the Microsoft Office suite

reference evapotranspiration measurements of 1316 mm and 661 mm, respectively.

Irrigation treatments and experimental design

A standard meteorological station (Imetos model, Pessl instruments) was installed at the experimental site to measure weather variables. The station continuously recorded the hourly temperature, air relative humidity, wind speed, and global solar radiation. We used these measurements to calculate the E_{To} using the Penman–Monteith equation (Allen et al. 1998, 2006), as shown in Fig. 2. Average daily values were used to estimate daily crop water evapotranspiration (E_{Tc}) as $E_{Tc} = E_{To} \times K_c \times K_r$. The crop coefficient K_c was estimated to be 0.65 in April and October, 0.6 in May and June, 0.55 from July to September, and 0.7 in November and December (Orgaz et al. 2007). The coefficient reduction K_r , which is related to the degree of orchard floor plant cover and is applied to canopies that cover less

than 50% of the ground, was calculated as $K_r = 2C/100$, where C represents the percentage of canopy cover (Feres and Castel, 1981). The value of K_r goes from 1 to 0.5 as the ground cover goes from 50 to 30% (Fernandez and Moreno 1999). Because Menara olive trees were young and the resulting low ground cover (<30%), the coefficient K_r is set to 0.5 for areas with less than 30% coverage. This K_r value remained consistent across all treatments, as the trees in each treatment exhibited uniform characteristics in terms of canopy height and diameter, averaging 12% ground cover.

Seven different treatments were applied within two different rows (Fig. 1). Phenological growth phases, including full flowering, pit hardening, oil synthesis, and harvest, were recorded over 2022 and 2023 according to the BBCH scale outlined by Sanz-Cortés et al. (2002). The respective phases are presented in Table 1. The Regulated Deficit Irrigation (RDI) and Sustained Deficit Irrigation (SDI) strategies were first implemented in 2021. It is crucial to

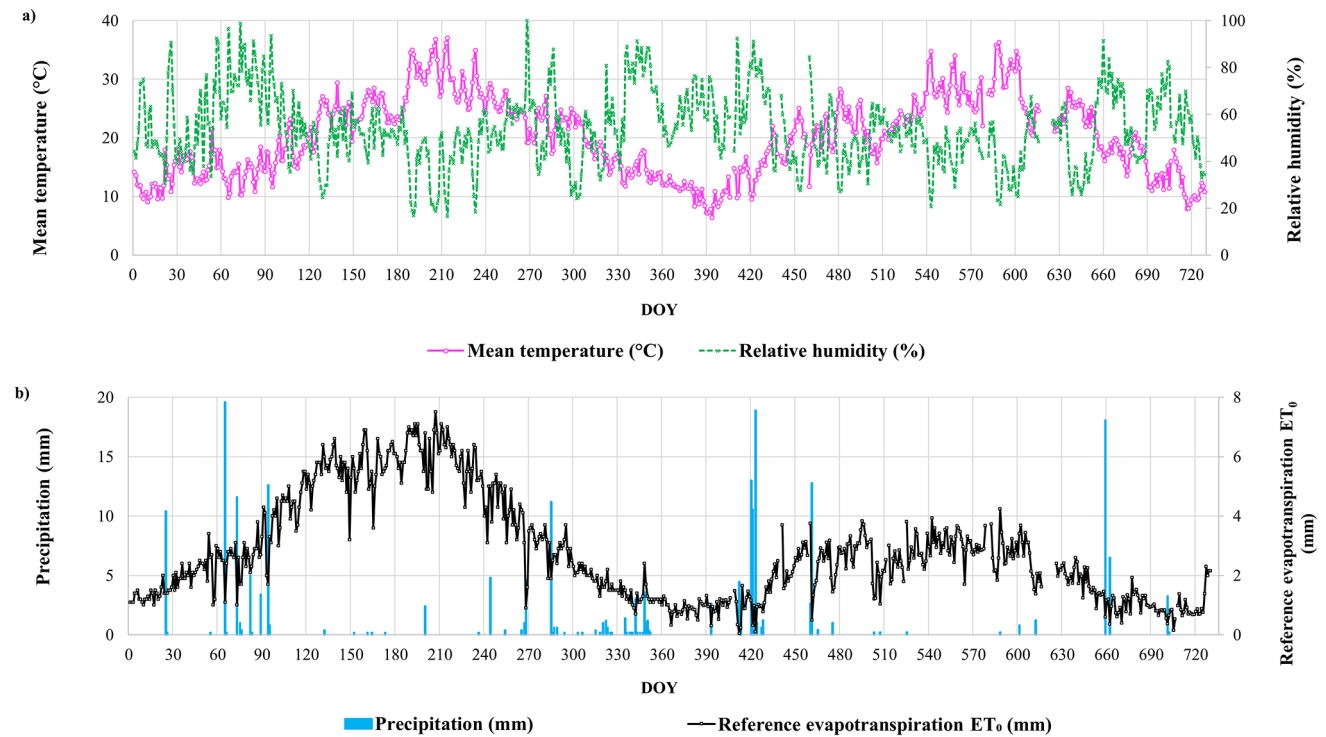


Fig. 2 Daily air temperature (°C), relative humidity (%), reference evapotranspiration ET_0 (mm) and precipitation (mm) at the experimental site in 2022 and 2023. DOY represents the day of the year, where 1 corresponds to January, 1

Table 1 Phenological growth phases during 2022 and 2023 used for characterizing the sensitive periods (SP1 and SP2) and the normal period (NP) for regulated RDI deficit irrigation treatments

Phenological growth stage	Dates	Duration	DOY	Periods	References
Full flowering to the beginning of pit hardening	05 April 2022-01 June 2022	58	95–152	SP1	(Rapoport et al. 2012; Pierantozzi et al. 2013; Hueso et al. 2021; Tadayon and Hosseini 2023).
	07 April 2023-06 June 2023	61	97–157		
From pit hardening to the beginning of oil synthesis	02 June 2022-03 August 2022	63	153–215	NP	(Goldhamer 1999; Moriana et al. 2003; Lavee et al. 2007; Gómez-del Campo and García 2013; Ibba et al. 2023a).
	07 June 2023-05 August 2023	60	158–217		
From the beginning of oil synthesis to harvest	04 August 2022-03 November 2023	92	216–307	SP2	(Tognetti et al. 2006; Martin-Vertedor et al. 2011; Gucci et al. 2019; García et al. 2020).
	06 August 2023-09 November 2023	96	218–313		

underscore that retrospective confirmation of the impact of deficit irrigation strategies on the agronomic and productive parameters of the Menara olive cultivar occurred in 2021. The studied irrigation treatments, percentage of crop water requirements (ET_c), and total water delivered based on ET_c and phenological growth are summarized in Table 2.

Sap flow measurements

The sap flow measurements were conducted using the thermal dissipation probe (TDP) method based on Granier's approach (1985, 1987a, b). Two olive trees per treatment were selected for measurements, with thermal sensors installed in the sapwood trunks. The sensors used were cylindrical probes (Dynamax) equipped with heating resistance,

thermocouples, and electronic modules for control. Probes were inserted radially, with one probe positioned 10 cm above the other. A Campbell Scientific CR1000 data logger recorded sap flow data at thirty-minute intervals. Sap flow sensors were not initially installed at the beginning of SP1 on DOY 95 (April 5th). The installation of sap flow sensors occurred in the last days of this first sensitive period, precisely on DOY 136 (May 16th) in 2022. In the second year of experimentation in 2023, the sap flow sensors for both the control treatment (T0) and the T2 treatment of the RDI strategy malfunctioned, leading to a loss of sap flow data for these treatments.

The Granier method is based on the liquid velocity heat dissipation theory and not on a specific model of heat transport in plant stems or tree trunks. The Granier method

Table 2 Treatments and irrigation water (in mm) applied during the growing seasons in 2022 and 2023

Year	Irrigation strategy	Treatments	Irrigation water by phase (mm)			Total water delivered (mm)
			SP1	NP	SP2	
2022	Control	T0- Irrigation with 100%ET _c in all phases	88	110	106	304
2023			48	52	59	159
2022	RDI	T1- Irrigation with 70%ET _c in phase NP and full in phases SP 1 and SP2	88	77	106	271
2023			48	36	59	143
2022	RDI	T2- Irrigation with 60%ET _c in phase NP and full in phases SP 1 and SP2	88	66	106	260
2023			48	31	59	138
2022	RDI	T3- Irrigation with 70%ET _c in phase NP and 80%ET _c in phases SP 1 and SP2	70	77	84	231
2023			38	36	47	122
2022	RDI	T4- Irrigation with 60%ET _c in phase NP and 80ET _c in phases SP 1 and SP2	70	66	84	220
2023			38	31	47	117
2022	SDI	T5- Irrigation with 70%ET _c in all phases	62	77	74	213
2023			33	36	41	111
2022	SDI	T6- Irrigation with 60%ET _c in all phases	53	66	63	182
2023			29	31	35	95

requires knowledge of the physical dimensions of the sapwood to convert velocity to the sap flow rate. Granier defined a dimensional parameter K as shown in Eq. (1):

$$K = (dT_M - dT)/dT$$

where dT is the difference in temperature measured between the heated needle and the lower nonheated needle and placed at a fixed distance below the heated needle. The value of dT is found from the differential voltage measured between the upper and lower thermocouples. The parameter dT_M is the dT when there is no sap flow (zero set value). Clearly, when $dT = 0$, K equals infinity, and if $dT = dT_M$, $K = 0$ (zero flow).

Granier found empirically that the average sap flow velocity V (cm s^{-1}) could be related to K by the exponential expression given in Eq. (2):

$$V = 0.0119 * K^{1.231} \text{cm s}^{-1}$$

To convert the velocity to the sap flow rate, one uses Eq. (3):

$$F_s = A_s * V * 3600 (\text{sh}^{-1}) \text{cm}^{-3} \text{h}^{-1}$$

where F_s ($\text{cm}^3 \text{h}^{-1}$) is the sap flow ($\text{cm}^3 \text{h}^{-1}$) and A_s is the cross-sectional area of the sap conducting wood (cm^2).

Physiological and agronomical measurements: stomatal conductance, chlorophyll fluorescence and fruit yield

Leaf stomatal conductance was periodically measured on different dates during the SP1, NP, and SP2 periods on fully expanded leaves from the mid-canopy (current-year shoot) on twelve leaves from individual trees for each

treatment, with a portable SC-1 leaf porometer from the Decagon Devices from 10:00 to 13:00 h when observations showed that at this time, gas exchange was at its maximum. Chlorophyll fluorescence was measured between 10:00 and 13:00 after leaf adaptation to darkness (30 min) with a portable OS30P + chlorophyll fluorometer from the Opti-Sciences Company to estimate the effect of water stress on a biophysical parameter that describes the PSII maximal photochemical efficiency (FV/FM). The potential quantum yield of PS II photochemistry was calculated by Eq. (4): $[F_v/F_m = (F_m - F_0)/F_m]$, where F_0 is the background fluorescence signal, F_m represents the maximal fluorescence intensity, and F_v corresponds to the difference between F_m and F_0 (Maxwell and Johnson 2000). Fruit yield was assessed in 2022 and 2023 for each treatment. Measurements were taken on twelve trees, and manual harvesting was carried out. The weight of the harvested fruit was recorded.

Statistical analysis

To evaluate the different deficit irrigation treatment effects, an analysis of variance (ANOVA) was carried out. Descriptive statistical analyses were used to calculate the mean and standard deviation and determine the variability of the measured variables across the different treatments. Tukey's multiple range test was used to determine significant differences among the treatments at a significance level of $P < 0.05$. The ANOVA with Repeated Measures analysis was employed to understand how variables are influenced by repeated measurements taken at different phenological stages (SP1, NP, and SP2). Moreover, Pearson's correlation coefficient and the principal component analysis method (PCA) were used to determine the relationships between the

studied agronomical and physiological parameters and the consecutive years 2022 and 2023 of deficit irrigation among the irrigation treatments. This approach allowed to effectively demonstrate the variability of functional responses of Menara cultivar to deficit irrigation strategies across various treatments and years. To investigate the correlation between fruit yield and cumulative sap flow volume, a regression analysis was performed. To test for differences between the studied years, we conducted a t-test between the years and for each variable.

Results

Characteristics of changes in sap flow over time during the entire irrigation season

Diurnal changes in sap flow patterns under different deficit irrigation treatments

Figure 3 compares the diurnal average sap flow patterns of the ‘Menara’ olive cultivar during the irrigation seasons of 2022 (a) and 2023 (b), demonstrating the observed response throughout these periods. The reference evapotranspiration (ET_0), representing climatic demand, is also

plotted in the figure. As shown, sap flow exhibited a bell-shaped diurnal pattern, with significant increases in the morning and fluctuating peaks in the early afternoon in response to climatic demand (ET_0), followed by a gradual decrease stabilizing around zero flow at midnight. The diurnal average sap flow, measured for each deficit irrigation treatment across distinct irrigation periods (SP1, NP, and SP2), demonstrated a significant influence from both the RDI and SDI treatments. ANOVA, with a significance level (α) of 0.05, revealed an extremely significant variation in the average diurnal sap flow among the irrigation treatments, with a p-value < 0.001. This approach provides a holistic view of sap flow dynamics throughout the growth seasons of 2022 and 2023 and enables comparisons of how these treatments influence the observed patterns. The diurnal average patterns of sap flow served as an indicator of the water status of the trees within each treatment. During the sensitive periods SP1 and SP2, the control T0 and the RDI treatments T1 and T2 received 100% of ET_c . However, during the succeeding water restriction period (NP), irrigation reductions of 30% and 40% resulted in a decrease in sap flow, significantly altering the patterns. Consequently, in 2022, the shape and temporal variations observed in deficit treatments T1 and T2 were closest to the control T0 compared to other

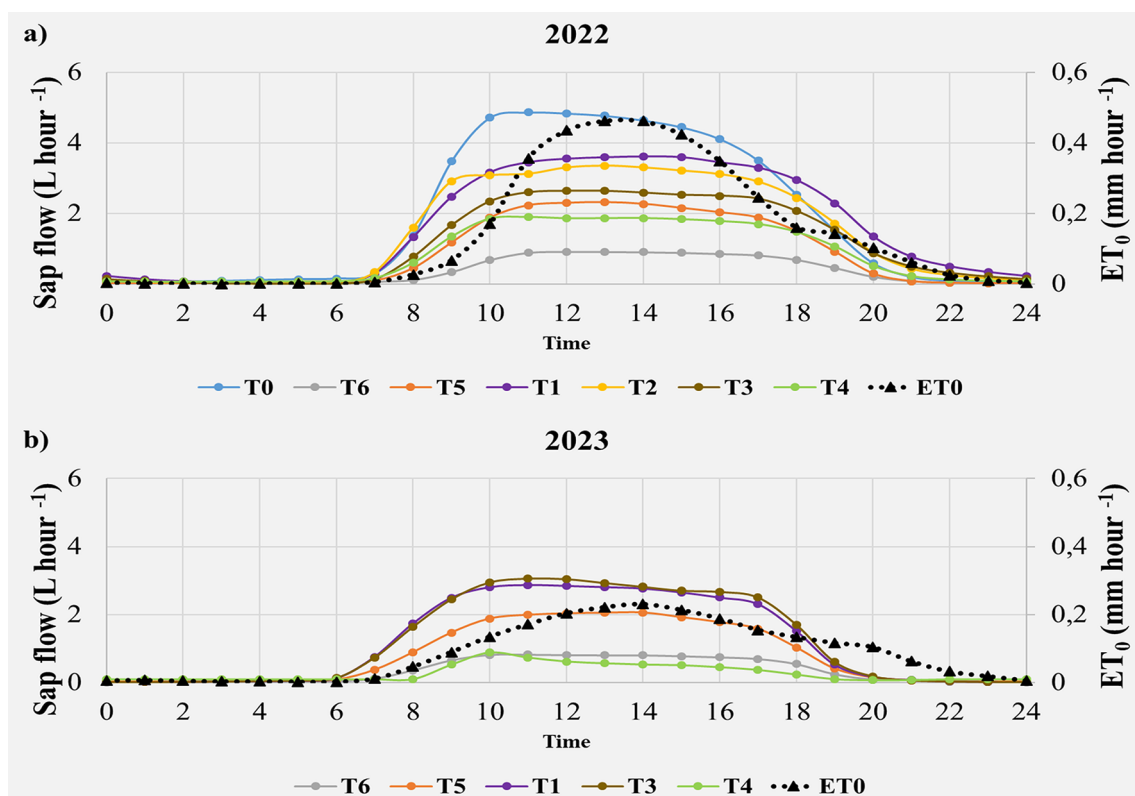


Fig. 3 Average diurnal patterns of sap flow and reference evapotranspiration (ET_0) in the ‘Menara’ olive plantation throughout the irrigation seasons of 2022 (a) and 2023 (b). * The ANOVA test, assuming

that $\alpha = 0.05$, revealed an extremely significant variation in the average diurnal sap flow among irrigation treatments, with a p value < 0.001. The T0 and T2 treatments are not represented in 2023

deficit irrigation treatments, as indicated by the curve correspondence in Fig. 3a. This finding demonstrates that trees subjected to this specific irrigation regime, with a 100% ETc rate during SP1 and SP2 and a 70% and 60% ETc rate during NP, had consistent responses in terms of their sap flow dynamics, indicating a uniform response to this volume of water delivered. During SP1 and SP2, the T3 and T4 treatments received 80% of the ETc, while the T5 and T6 treatments received 70% and 60% of the ETc. As a result, the diurnal curves indicating sap flow in these treatments showed lower patterns compared to the fully watered treatments that received 100% of the ETc. In the case of the T3 treatment, there was a relatively modest 10% reduction in applied water during the water restriction period (NP) compared to the sensitive periods (SP1 and SP2). In 2022, these variations in water supply resulted in reductions in sap flow compared to the T1 and T2 treatments. In 2023, T3 closely matched T1 in sap flow dynamics, as indicated by the curve correspondence in Fig. 3b. Compared with those in the T3 treatment, the T4 and T5 treatments showed a decrease in sap flow. Interestingly, both the T4 and T5 treatments, which reduced ETc by 20% and 30%, respectively, during SP1 and SP2, exhibited similar reductions in sap flow in 2022. T4 experienced a decrease in applied water from 80% during SP1 to 60% during NP, resulting in a decrease in sap flow compared to that in T5, which received 70% of the total water throughout the irrigation season. This explains the observed decrease in sap flow in T4, as shown in Fig. 3a and b. In 2023, the T4 treatment had the most significant reduction in diurnal sap flow patterns, along with the T6 treatment. The T6 treatment, which received 60% of the total ETc, showed the most significant reduction in diurnal sap flow patterns in both years 2022 and 2023 (Fig. 3a, b). This notable decline in sap flow, compared to that in the T0 treatment, can be chiefly attributed to the substantial 40% reduction in the volume of water applied. The second irrigation season in 2023 saw reduced climatic demand (ET0) compared to 2022, resulting in lower sap flow rates across all treatments during this period.

Figure 3 also depicts the phases outlined by Herzog et al. (1995) when comparing the main characteristics of diurnal patterns of sap flow. Phase I is the nightly period of resaturation when there is no or little measurable flow, marked by the end of the peak time. This phase occurs between night and 06:00 for all irrigation treatments during the two irrigation seasons 2022 and 2023. Phase II indicates an increase in sap flow, characterized by the initial diurnal activity of sap flow across various irrigation treatments. Subsequently, phase III represents the interval during which the sap flow reaches its peak within the

day. In 2022, these phases were also observed in the control treatment (T0) between 06:00 and 10:00. In contrast, generally within the deficit irrigation treatments, these phases extended over an extended duration, ranging from 06:00 to 12:00. Therefore, compared to deficit irrigation, full irrigation led to greater morning sap flow. Additionally, the plateau in sap flow during the central hours of the day was notably greater in the well-irrigated treatment than in the control treatment, reflecting the physiological behavior of Menara olive trees. Phase IV and the final phase V describe the delay after the maximum flow, characterized by a subsequent decrease in sap flow rates.

Changes in cumulative sap flow volume under different deficit irrigation treatments

Figure 4 summarizes the cumulative sap flow volume throughout the irrigation seasons of 2022 and 2023. Because sap flow sensors were not initially installed when sensitive period 1 (SP1) began, the irrigation season did not appear within the results of the SP1 period, as delineated in Table 3. This temporal gap between the beginning of the SP1 and the installation of sap flow sensors necessitates a procedure to estimate sap flow for the duration of the SP1. To address this, we computed the average daily sap flow using available data from the last days of this first sensitive period, specifically on May 16th, which extended over 18 days. Subsequently, we extrapolated this average by multiplying it by the total number of days encompassing SP1, which started at the flowering phenological stage in early April and spanned 58 days.

The results showed that deficit irrigation had a significant effect on the periodic cumulative sap flow rate across all the treatments during the consecutive years 2022 and 2023. Notably, it is worth mentioning that the cumulative sap flow volume for the control T0 treatment was the highest, totaling 9892 L. However, treatment T6 exhibited lower transpiration rates, indicating minimal water intake, with total cumulative sap flow amounts of 1961 and 1760 L occurring in 2022 and 2023, respectively. Interestingly, compared with the T0 treatment, the T1 treatment transpired almost the same amount of water as the control T0, with 8889 L, in 2022, exhibiting a 30% reduction in water consumption during the NP treatment. Additionally, compared to the T1 treatment, the T3 treatment transpired nearly the same amount of water, with 6091 and 6460 L, respectively, in 2023. However, the T4 treatment under the RDI strategy showed adverse effects after two consecutive seasons of deficit irrigation, resulting in a cumulative sap flow of 1646 L in the second year of the experiment. In 2022, the sap flow decreased

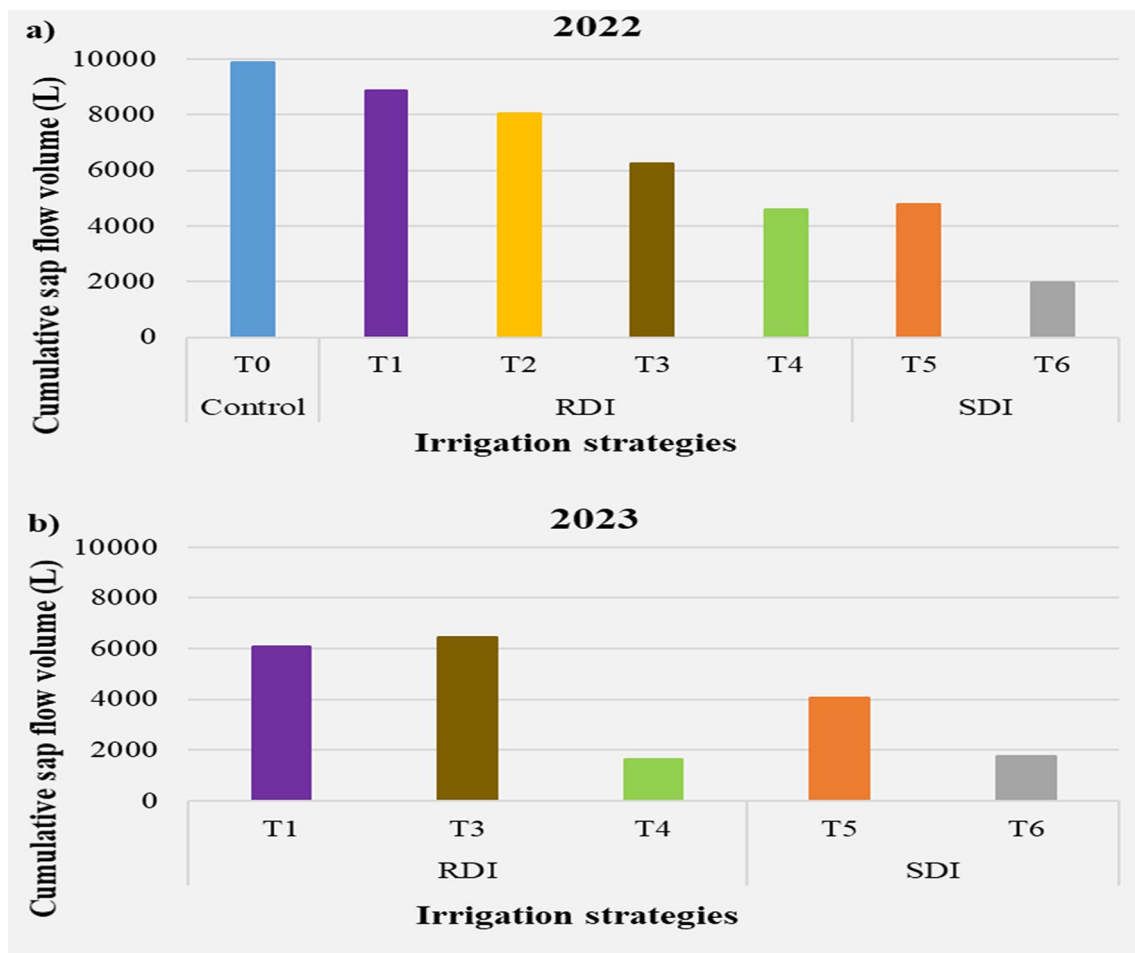


Fig. 4 Cumulative sap flow volume (L) variation throughout the irrigation season for the control, regulated (RDI) and sustained (SDI) deficit irrigation treatments. Figure 4a and b correspond to the growing irriga-

tion season 2022 and 2023, respectively. The T0 and T2 treatments are not represented in 2023

significantly in the range of 10–54%, from T1 to T4 for the RDI strategy and 51–80% from T5 to T6 for the SDI strategy. Notably, when total water savings of 11% and 14% were implemented throughout the irrigation season, compared to those in the fully irrigated treatment T0, the total cumulative sap flow volume was not severely affected by the T1 and T2 treatments.

Variations in sap flow and physiological and agronomical parameters

Variations in sap flow and stomatal conductance (gs)

The stomatal conductance (gs) was significantly affected by water deficit during the irrigation seasons of 2022 and 2023. ANOVA, assuming that $\alpha=0.05$, showed an extremely significant variation in the average (gs) between irrigation treatments, with $p<0.001$ (Fig. 5). Figure 5a shows that water deficit decreased the stomatal conductance (gs) of the

leaves of the Menara cultivar studied in 2022. The leaves of the control and T1, T2, and T3 treatments of the RDI strategy were characterized by almost the same stomatal conductance between 146 and 138 $\text{mmol H}_2\text{O m}^{-2} \text{s}^{-1}$ from T1 to T3. However, the mean stomatal conductance in the T4 treatment of the RDI strategy and the SDI treatments (T5 and T6) demonstrated a significant reduction, with values ranging between 131, 123, and 110 $\text{mmol H}_2\text{O m}^{-2} \text{s}^{-1}$ for T4, T5, and T6, respectively, representing decreases of approximately 8%, 13%, and 23%, respectively, compared to those of the control (T0). These reductions were attributed to the implementation of water-saving practices through the RDI and SDI strategies, which reflect the significant impact of reducing the applied total water by 28%, 30%, and 40%, respectively. In 2023, stomatal conductance was significantly affected by the T5 and T6 treatments, with values ranging between 148 and 126 $\text{mmol H}_2\text{O m}^{-2} \text{s}^{-1}$ and reductions of approximately 10% and 23%, respectively, due to 30% and 40% water conservation during the whole

Table 3 Percentage of variation in irrigation water applied, cumulative sap flow volume and fruit yield compared to those in T0 during the growing season in 2022

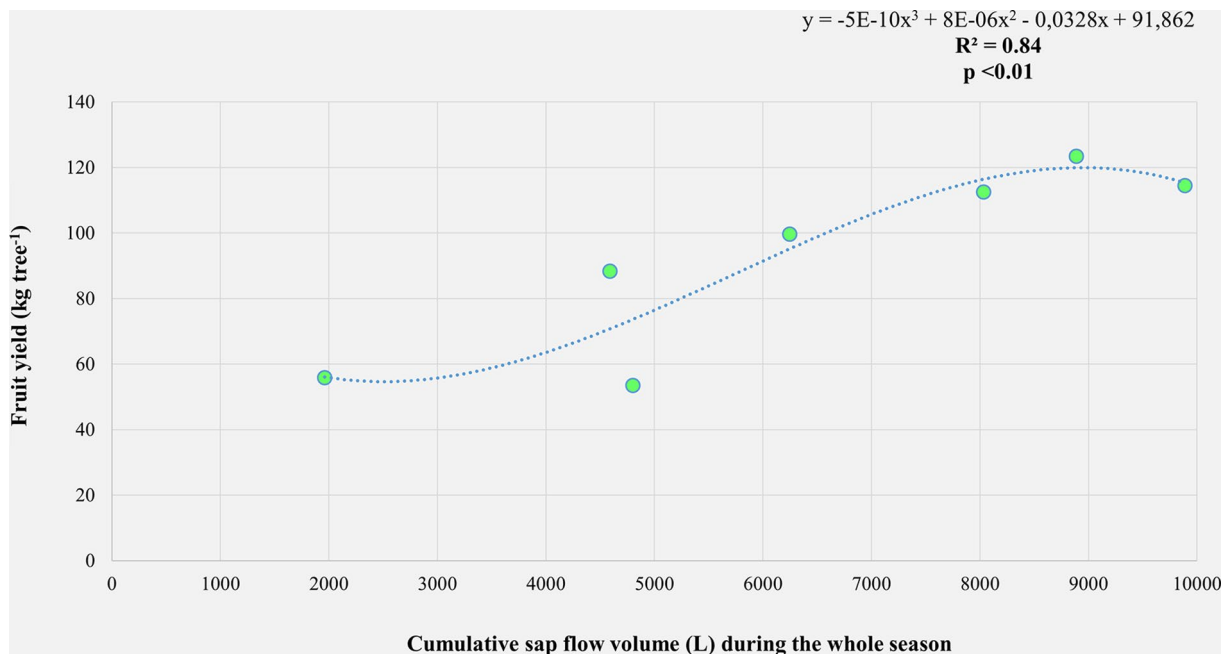
Year	Irrigation strategies	Treatments	Total water (%) T0	Cumulative sap flow volume (%) T0	Fruit yield (%) T0	
2022	Control	T0	100	100	100	
2023			100	-	100	
2022	RDI	T1	(-) ^b 89	(-) 90	(+) ^a 108	
2023			(-) 90	-	(-) 88	
2022		T2		(-) 86	(-) 81	(-) 98
2023				(-) 87	-	(-) 94
2022		T3		(-) 76	(-) 63	(-) 87
2023				(-) 77	-	(-) 89
2022	T4		(-) 72	(-) 46	(-) 77	
2023			(-) 73	-	(-) 93	
2022	SDI	T5	(-) 70	(-) 49	(-) 47	
2023			(-) 70	-	(-) 89	
2022		T6		(-) 60	(-) 20	(-) 49
2023				(-) 60	-	(-) 86

^a Positive (+) or ^b negative (-) signs indicate whether there is an increase or decrease relative to T0

irrigation season treatment compared to the control T0 treatment, as shown in Fig. 5b. The ANOVA with repeated measures analysis was employed to understand how stomatal conductance is influenced by repeated measurements taken at different phenological stages (SP1, NP, and SP2). This analysis showed the high significance of different phenological stages with $p < 0.001$ and $p < 0.01$ in 2022 and 2023, respectively, as shown in Supplementary Table 1.

Therefore, a 30% reduction in applied water during NP and 20% during SP2 did not significantly affect stomatal conductance in the T1, T2, and T3 treatments. Therefore, olive trees subjected to these treatments potentially exhibited a degree of tolerance, enabling the preservation of stomatal conductance despite the imposed water supply limitation. Notably, our analysis revealed no statistically significant difference between the RDI treatments and the control T0, which is explained by their irrigation levels closely aligning with evapotranspiration demands of 100% and 80% ETc. It was also observed that the stomata did not close or regulate the aperture in response to a 20% reduction in water availability under the RDI strategy, which may be considered a nonstressful condition.

The behavior of sap flow closely followed that of stomatal conductance, as shown by the graphical representation in Fig. 5. ANOVA analysis, assuming $\alpha = 0.05$, revealed a significant variation in this parameter between irrigation treatments, with $p < 0.001$ in both the years 2022 and 2023. Furthermore, the ANOVA with repeated measures analysis indicated that sap flow rates are influenced by different phenological stages (SP1, NP, and SP2) with $p < 0.001$. Sap flow rates were consistently greater in fully irrigated trees (T0, T1, and T2) than in stressed plants subjected to T3, T4, T5, and T6 treatments. In 2023, sap flow rates were consistently lower in the T4 and T6 treatments. However, there were no significant differences in sap flow rates between the trees subjected to treatments T3, T4, and T5 in 2022. This is due to the similar trends observed in these treatments, where water restriction was

**Fig. 8** Correlations between fruit yield and cumulative sap flow volume in the ‘Menara’ olive cultivar for the control, regulated and sustained deficit irrigation treatments

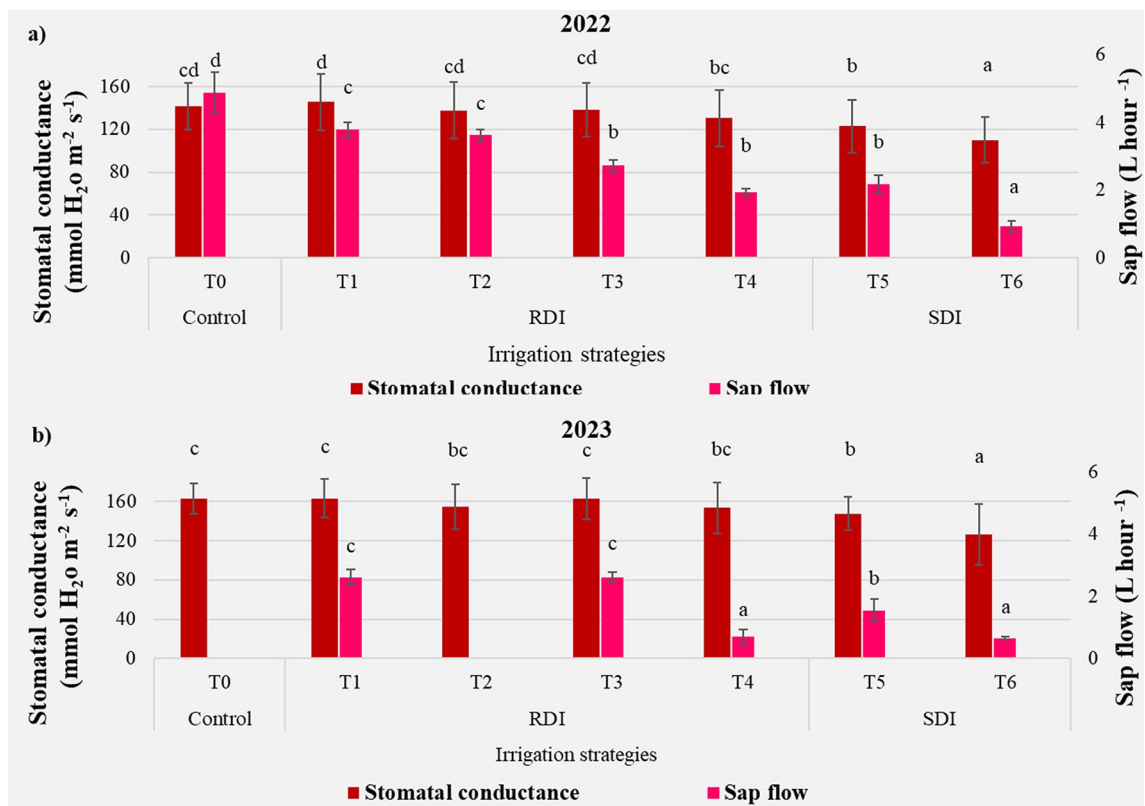


Fig. 5 Stomatal conductance and sap flow measurements for each irrigation treatment. Figure 5a and b correspond to the growing irrigation seasons 2022 and 2023, respectively, with error bars representing the standard deviation

maintained at around 20–30%. Figure 5a and b illustrates these trends.

Variations in sap flow and chlorophyll fluorescence (Fv/Fm)

Figure 6 shows the variation in chlorophyll fluorescence (Fv/Fm) during the irrigation seasons of 2022 and 2023. This biophysical parameter was significantly affected by the water deficit. ANOVA, assuming that $\alpha=0.05$, revealed a significant variation in the average (Fv/Fm) between the irrigation treatments, with $p<0.05$ in 2022 and $p<0.01$ in 2023. A significant reduction in Fv/Fm started with a 40% decrease in water supply in the T6 treatment group during both irrigation seasons in 2022 and 2023, resulting in reductions of approximately 10% and 7%, respectively, compared to those in the control T0 treatment, which was well-watered and maintained a Fv/Fm value of approximately 0.80 during each year. As a result, olive trees subjected to this amount of water stress exhibited lower Fv/Fm values by about 0.70. Notably, these values dip below the critical sensitivity threshold of 0.75 (Fv/Fm < 0.75), indicating that the critical levels are associated with water deficiency. Notably, leaves subjected to the control fully irrigated treatment exhibited nearly the same chlorophyll fluorescence intensity across treatments T1 to T5 (Fig. 6a,

b). This observation suggested that the induced water stress conditions from these treatments did not impact the Fv/Fm values in a manner distinct from that of well-watered olive trees. The ANOVA with repeated measures analysis showed a high significance of different phenological stages on this parameter with $p<0.001$ in 2022, as shown in Supplementary Table 1. Therefore, the Fv/Fm was notably impacted in T6 treatments of the SDI strategy compared with the control T0 because water savings were reduced by 40%, respectively, during the whole irrigation season, as illustrated in Fig. 6.

The large decrease in sap flow rates observed in the T4 and T6 treatments resulted in a significant decrease in this parameter throughout the irrigation seasons 2022 and 2023 with ($p<0.001$). It is important to highlight that the utilization of Fv/Fm as an indicator for assessing the level of water stress in Menara olive trees initiates a 40% reduction in applied water. This reduction is indicated by the decrease in Fv/Fm values below 0.75, which is a threshold that marks the onset of water stress.

Relationship between tree sap flow and fruit yield

ANOVA, assuming that $\alpha=0.05$, revealed significant variation in the average fruit yield between the irrigation

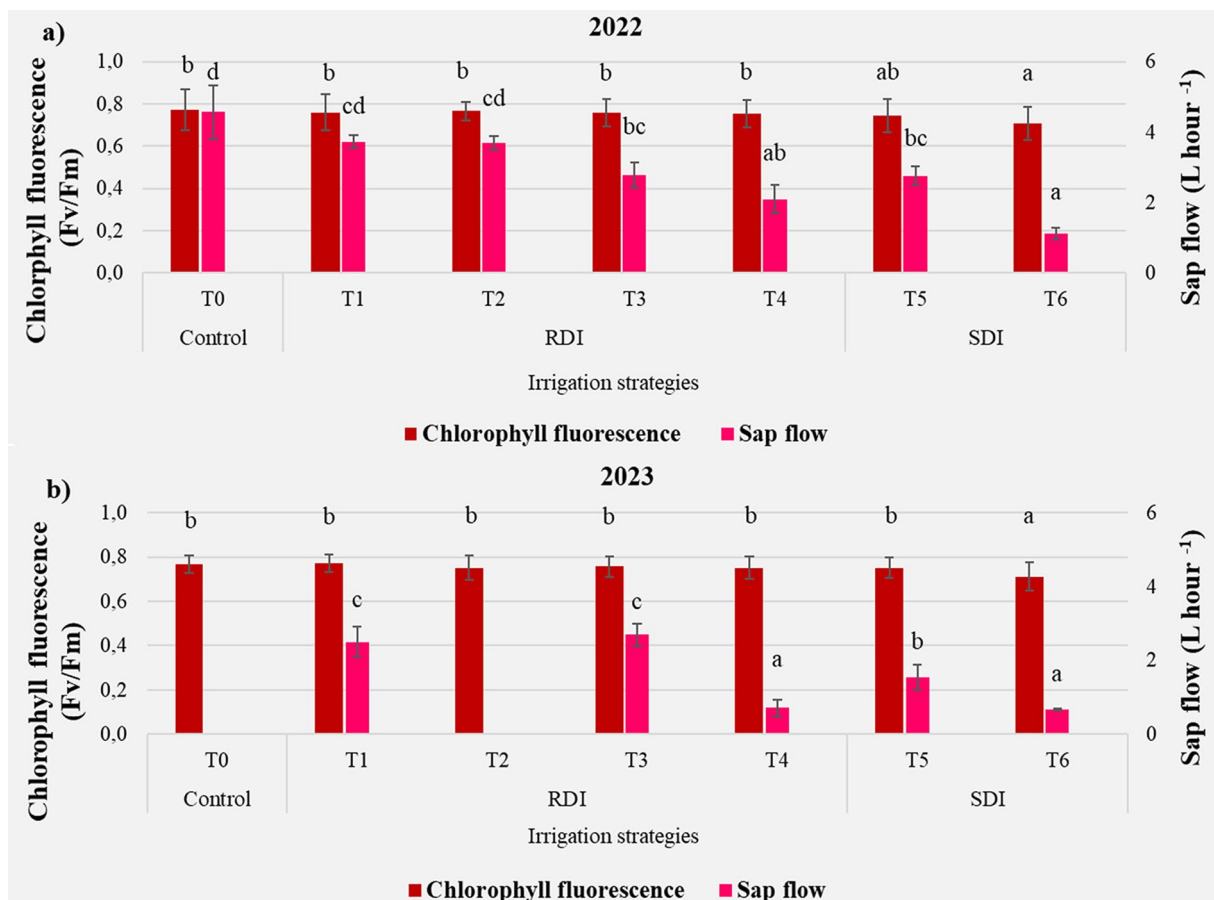


Fig. 6 Chlorophyll fluorescence and sap flow measurements for each irrigation treatment. Figure 6a and b correspond to the growing irrigation seasons 2022 and 2023, respectively, with error bars representing the standard deviation

treatments in 2022 ($p < 0.001$) and in 2023 ($p < 0.05$) (Fig. 7a, b). In 2022, in comparison to those in the deficit irrigation treatment, more fruits in the control treatment (T0) developed fruit yield, except for in the T1 treatment, in which approximately 123.3% (kg/tree) of the fruits were produced, even with a 10% reduction in cumulative sap flow. This was observed despite a 30% reduction in water applied during the NP period or an 11% reduction throughout the full irrigation season. Furthermore, T2 produced approximately 112.5 kg/tree, comparable to that of the T0 control, despite a 40% reduction in water delivered throughout the NP period. These results can be attributed to the fact that deploying full irrigation water during sensitive periods did not adversely affect fruit yield production. Despite receiving almost 24% less irrigation, the fruit yield of the T3 treatment (99.6 kg/tree) was not severely affected compared to that of the T0 treatment (114.4 kg/tree) for the growing season 2022. Furthermore, compared with those in the control, the production in the T4 treatment under the RDI strategy demonstrated resilience, exhibiting a modest reduction of approximately 23% in yield despite a substantial 28% reduction in applied water and 54% in cumulative sap flow volume. In contrast,

the T6 treatment within the SDI strategy experienced a significant decline in production, with a 50% reduction when subjected to a 40% reduction in water applied and 80% in cumulative sap flow volume throughout the entire irrigation season, confirming the sensitivity of the SP1 and SP2 periods to water stress imposed by the sustained deficit irrigation strategy, as illustrated in Fig. 7 and detailed in Table 3. In 2023, the average fruit yield ranged between 32 and 37 in the different irrigation treatments from T0 to T6. There was a noticeable decrease in the amount of fruit produced by Menara olive trees as compared to the previous year of 2022 (see Fig. 7 and Supplementary Table 5). This decrease in olive production was due to the alternation of fruit loads from one year to another. The Menara cultivar had an “off” year in 2023 after having an “on” year in 2022.

In 2022, a polynomial regression analysis was used to assess the correlation between average fruit yield and cumulative sap flow volume across various irrigation regimes. The results showed a strong polynomial correlation with an R^2 value of 0.84, suggesting a significant correlation with a p -value of < 0.01 , as shown in Fig. 8. The correlation between average fruit yield and cumulative sap flow

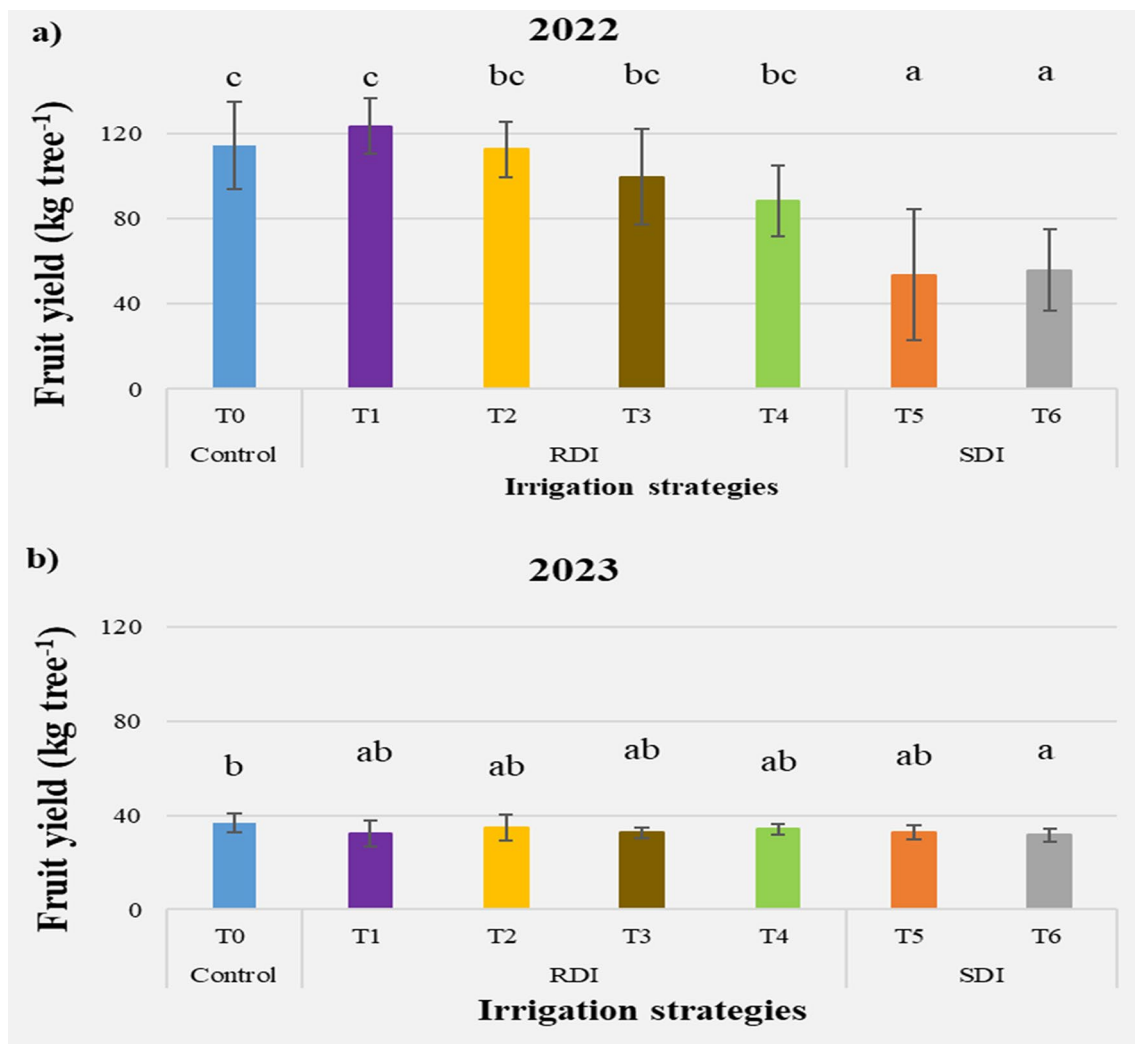


Fig. 7 Fruit yield measured for each irrigation treatment in 2022 and 2023, with error bars representing the standard deviation

volume across various irrigation treatments in 2023 was not analyzed due to the unavailability of sap flow data for the T0 and T2 treatments, coupled with the fact that the Menara cultivar experienced an alternation of fruit load.

Variability in physiological and agronomical parameters of Menara under deficit irrigation across consecutive years 2022 and 2023

The principal component analysis (PCA) was conducted to explore the relationships among all the examined agronomical and physiological parameters. The results of this analysis are illustrated in Fig. 9a. The PCA axes collectively explain 95% of the total variance, as detailed in Table 4. The primary PCA axis alone accounts for 63% of the total variance and exhibits a positive correlation with all variables, indicating consistent responses to deficit irrigation treatments affecting the studied parameters. Conversely,

the second axis (contributing 21.1% of the total variance) demonstrates a positive correlation with cumulative sap flow (CSFV) and fruit yield (FY), while showing a negative correlation with stomatal conductance (g_s) and chlorophyll fluorescence (F_v/F_m) (refer to Table 4; Fig. 9a). Tree-related variables (CSFV, F_v/F_m , and FY) are highly and positively correlated with each other, while stomatal conductance (g_s) shows a strong correlation with chlorophyll fluorescence, as indicated in Table 5. This implies that maintaining optimal sap flow levels in olive trees is critical for supporting their photosynthetic activity and overall production. Figure 9b illustrates the projection of various deficit irrigation strategies over two consecutive years, 2022 (A) and 2023 (B), using the first two principal components. The response of different irrigation treatments exhibited notable variations between years, leading to the identification of three distinct response groups. The first group (1) comprised treatments T1B, T3B, T4B, and

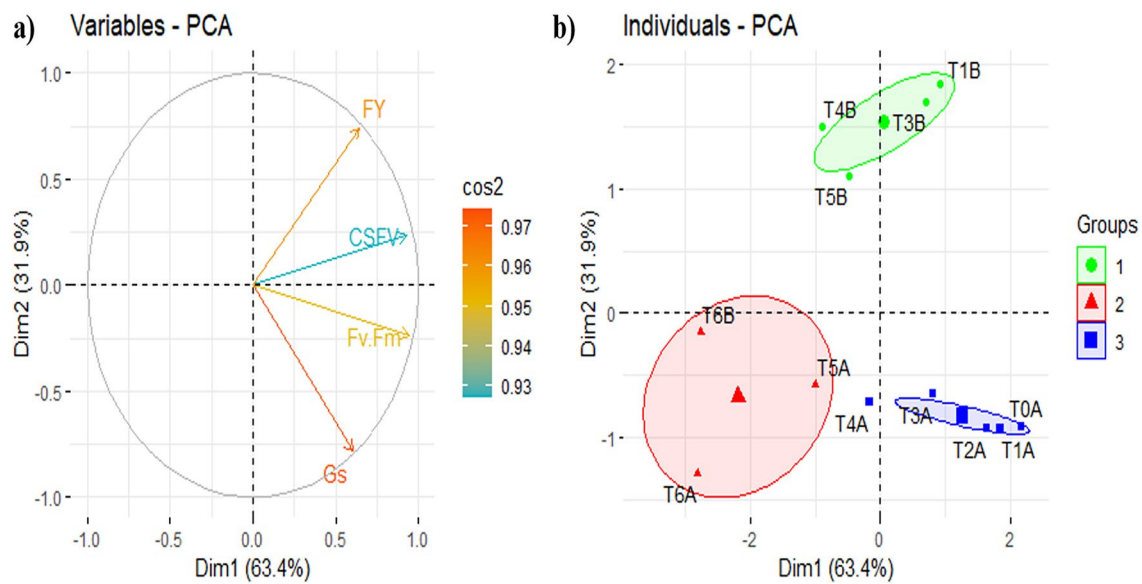


Fig. 9 Plot illustrating the contribution of agronomic and physiological parameters studied to the PCA axes (a) and graphical representation of the PCA showing deficit irrigation treatments over two consecutive years (b). (A) and (B) stand for the years 2022 and 2023, respectively

Table 4 Correlations between agronomical and physiological traits and the two first PCA axes identified

Variables	Dim1	Dim2
Cumulative sap flow volume (CSFV)	0.93	0.24
Stomatal conductance (gs)	0.60	-0.78
Chlorophyll fluorescence (Fv/Fm)	0.95	-0.24
Fruit yield (FY)	0.64	0.74
Variance (%)	0.63	0.32
Cumulative variance %	0.63	0.95

Table 5 Correlation coefficients between the studied variables

Variables	CSFV	gs	Fv/Fm	FY
CSFV	1.00			
gs	0.37 ^{n.s}	1.00		
Fv/Fm	0.79 ^{**}	0.73 ^{**}	1.00	
FY	0.73 ^{**}	-0.17 ^{n.s}	0.43 ^{n.s}	1.00

Computed correlation used the Pearson-method with listwise-deletion. Within the table, asterisks represent statistical significance levels, with *** $p < 0.001$, ** $p < 0.01$ and * $p < 0.1$, n.s., non-significant

T5B; the second group (2) included T5A, T6A, and T6B; and the third group (3) consisted of T0A, T1A, T2A, T3A, and T4A. In 2022, notable differences were observed, particularly between groups 2 and 3. Group 2 exclusively represented treatments utilizing sustained deficit irrigation (SDI) (T5A and T6A), whereas group 3 included all regulated deficit irrigation (RDI) treatments (T1A, T2A, T3A, T4A) along with the control (T0A). These differences were highlighted by distinct response patterns to water stress observed between the SDI treatment group and the RDI and control treatment groups. In 2023, noticeable differences emerged between groups 1 and 2. Notably, treatment

T5B showed a response similar to RDI treatments in the second year of the experiment, suggesting adaptation to water stress. Both RDI and control treatments exhibited generally similar response patterns. The impact of each year was markedly different between 2022 (A) and 2023 (B), evident in the response differences between group 1 (B) and group 3 (A) irrigation treatments. These findings underscore the adaptive response of olive trees to water stress over time and demonstrate the sustainable effects of the RDI strategy on Menara olive tree performance across consecutive years.

The Student's t-test analysis for sap flow, stomatal conductance, and fruit yield showed significant differences between the years 2022 and 2023 for all RDI and SDI treatments, as presented in Supplementary Tables S2, S3, and S5. In 2023, there was a significant decrease in both sap flow and fruit yield for the regulated deficit irrigation (RDI) and sustained deficit irrigation (SDI) treatments compared to the first year of 2022 (see Supplementary Tables 2 and 5). This decrease in olive production was due to the Menara cultivar having an “off” year in 2023 following an “on” year in 2022. The Fv/Fm of Menara trees was not significantly influenced by the year, with $p > 0.05$ in all irrigation treatments (Supplementary Table 4). The gs of Menara trees were also significantly influenced by the year, with $p < 0.001$ in T0, T1, T3, T4, and T5 and with $p < 0.01$ in T2, T6 (Supplementary Table 3). This parameter showed significant increases for all irrigation treatments compared to the first year of 2022 and demonstrated increased stomatal conductance with a lower fruit load.

Discussion

The diurnal sap flow patterns observed in this study closely resemble patterns documented in prior research on various plant species (Edwards and Warwick 1984; López-Bernal et al. 2010; Sun et al. 2021, 2022; Tognetti et al. 2004). Specifically, we observed a rapid morning increase in sap flow rates, reaching a peak around midday, followed by a sharp decline towards late afternoon (López-Bernal et al. 2010). Our findings align with those of Tognetti et al. (2004), who reported higher sap flow rates in control (T0) compared to deficit irrigation treatments (Fig. 3). In T1 and T2 treatments, we observed a slight decrease in diurnal sap flow patterns compared to T0. The impact of the phenological stage on sap flow rates was significant (Supplementary Table 1), indicating that water restriction by 30% and 40% during the normal period (NP) influenced this parameter. However, full irrigation during sensitive periods (SP1 and SP2) led to recovery, consistent with rapid stress recovery observed by Iniesta et al. (2009) in the ‘Arbequina’ cultivar. Many authors recommend reducing irrigation during pit hardening, as this phase is less sensitive to water deficit (Gómez Del Campo and García 2013; Ibba et al. 2023a). Ballester et al. (2012) observed expected decreases in sap flow during water restriction periods, with recovery upon reestablishing normal irrigation in citrus trees. Based on our observations in 2022 and 2023, it was found that trees subjected to RDI treatments (T1, T2, and T3) experienced less water stress. They exhibited patterns that were close to the control treatment by applying 100% or 80% of estimated crop evapotranspiration (ETc) during critical growth phases. Diurnal sap flow patterns under T4 treatment showed more pronounced decreases in 2023 compared to 2022 (Supplementary Table 2). Therefore, the negative effects of T4 RDI treatment were observed in Menara olive trees after two consecutive seasons of deficit irrigation. This indicates that prolonged stress effects can be detrimental in subsequent years (Gucci et al. 2019). In the case of the SDI strategy, the T6 treatment was consistently subject to permanent reductions of 40% in applied water throughout the whole irrigation season. As a result, the diurnal sap flow patterns observed under this treatment exhibited more pronounced decreases than those in the control T0 (Fig. 3).

Shabani et al. (2013) suggested that transpiration rates change due to water deficit, thus improving transpiration efficiency. López-Bernal et al. (2010) reported that, in the ‘Arbequina’ olive cultivar, the control treatment presented greater maximum values of sap flow than did the SDI treatment. Furthermore, the PCA analysis revealed that the first PCA axis explains 63% of the total variance and shows a positive correlation with all variables, suggesting consistent responses to deficit irrigation treatments across the studied

parameters. Water stress signals may be transmitted to leaf blades through the abscisic acid (ABA) pathway, influencing stomatal conductance, reducing transpiration, and modulating photosynthetic intensity (Yang et al. 2022). During the second irrigation season, all deficit irrigation treatments showed the lowest transpiration rates compared to those observed in 2022, which can be attributed to reduced evapotranspiration demand and fruit yield load. A study by Fernández et al. (2001) revealed a strong correlation between the daily sap flow of olive trees and potential evapotranspiration. On the other hand, variations in sap flow dynamics, as shown in Fig. 3, align with the analysis of the five phases outlined by Herzog et al. (1995) across various irrigation treatments. With increasing sunlight, sap flow and physiological activity increase gradually, followed by a decrease around noon when leaf stomata close. This dynamic reflects the physiological processes of trees throughout the day (Chen et al. 2020; Klein et al. 2018). Notably, full irrigation in the T0 treatment resulted in greater morning sap flow. Furthermore, the plateau in sap flow during central daytime hours was greater in the well-irrigated treatments. However, Fernández et al. (2011) reported the limited utility of the five-phase sap flow dynamics study outlined by Herzog et al. (1995) for identifying water stress in olive trees.

Stomatal regulation is controlled to optimize water vapor outward diffusion and CO₂ diffusion into the leaf during photosynthesis (Hetherington and Woodward 2003). In the present study, monitoring stomatal conductance decreased when the level of water stress increased with deficit irrigation strategies, particularly with the SDI strategy (Fig. 5). In 2022, the stomatal conductance exhibited a significant decrease in response to the T4 treatment, with decreases of approximately 8%. These changes were attributed to the implementation of water-saving measures, specifically a 40% reduction during the NP compared to the control T0. During both irrigation seasons, the Menara olive trees exhibited a significant decrease in stomatal leaf conductance in response to T5 and T6 treatments. These treatments involved conserving 30% and 40% of water throughout the entire irrigation season, resulting in average decreases of approximately 12% and 23%, respectively. The phenological phases significantly affect this parameter (Supplementary Table 1). Therefore, these changes were attributed to water conservation during the entire irrigation season. The RDI treatments confirmed that the olive trees’ response to water deficit varies depending on the stage at which the water deficit is imposed. The intensity of the water deficit, the growth stage of the plant, and the duration of the deficit are all factors that influence stomatal leaf conductance in Menara olive trees. One of the main responses to water scarcity is a decrease in stomatal conductance, leading to stomatal closure, which restricts photosynthesis (Flexas

et al. 2013). This is consistent with the correlated analysis between stomatal conductance (g_s) and the maximum quantum yield of photosystem II (Fv/Fm), which was significantly correlated (Table 5). This has significant implications for plant function, growth, and yield limitation in fruit trees (Brito et al. 2019; Hernandez-Santana et al. 2016b). On the other hand, the stomata of olive leaves are small, and they become even smaller and denser in water shortage situations, enabling better control of water loss by transpiration (Boughalleb and Hajlaoui 2011; Ennajeh et al. 2010). The control of stomatal opening is a well-documented mechanism for plants to restrict water loss through transpiration (Lovisollo et al. 2002). Several studies have suggested that higher stomatal conductance values indicate high transpirational water losses, which require high sap flow rates to maintain an adequate water status in plants (Kokkotos et al. 2021). Moreover, Hernandez-Santana et al. (2016a) revealed a strong correlation between sap flow and stomatal conductance in olive trees. Their findings showed that the diurnal dynamics of sap flow in the olive trunk directly reflect stomatal conductance dynamics at leaves in the canopy.

The Fv/Fm ratio is a widely used index for evaluating stress intensity in plant leaves and is used to estimate the maximal quantum yield of PSII. Under the SDI strategy, the ‘Menara’ cultivar was most affected by water stress and exhibited the greatest decrease in Fv/Fm in the T6 treatment. This indicates a significant reduction in the maximum quantum efficiency of photosystem II (PSII), with values of around 0.70, suggesting that this critical threshold in Menara olive trees fell below the threshold of 0.75 as reported by Woo et al. (2008), leading to damage to the photosynthetic apparatus of the ‘Menara’ cultivar (Fig. 6). The plants experiencing critical levels of water deficit demonstrated significantly lower Fv/Fm values ranging from 0.45 to 0.75 (Woo et al. 2008). Increasing levels of water stress led to decreasing values of chlorophyll fluorescence compared to those in control T0 trees (Wang et al. 2023). As reported for various Mediterranean species, such as olive trees, stress imposition negatively impacts chlorophyll across all studied cultivars (Boughalleb and Hajlaoui 2011; Boussadia et al. 2008, 2023; Vasques et al. 2016). However, the maximal photochemical efficiency of PSII (Fv/Fm) remained stable under the RDI strategy, indicating that olive trees maintained their photosynthetic efficiency despite the imposed water stress, highlighting the resilience of the studied species under the RDI strategy. The findings suggest that photosynthetic processes were either slightly damaged or not damaged at all, as the values of Fv/Fm were within the expected ranges for healthy plants (0.80 H; Lapa et al. 2017; Maxwell and Johnson 2000). A more significant reduction in the SDI strategy was due to a 40% reduction in applied water during the entire irrigation season, indicating the phenomenon of photoinhibition and the downregulation of photosystem II (PSII), which reflects the

protective or regulatory mechanism to avoid photodamage to the photosynthetic apparatus, as elucidated in previous studies (Boussadia et al. 2008, 2023; Maxwell and Johnson 2000).

Regarding the fruit yield, the well-irrigated treatments (T0 and T1) demonstrated the highest fruit yield (114.4 and 123.3 kg/tree) in 2022 (Fig. 7a). However, in 2023, a notable decline in fruit yield was observed across all irrigation treatments compared to 2022, with T0 exhibiting the highest yield at 37 kg/tree (Fig. 7b). This decrease in production during 2023 can be attributed to the Menara cultivar experiencing an “off” year (Fig. 7 and Supplementary 5). Specifically, the variation in irrigation volume between regulated deficit irrigation (RDI) and standard deficit irrigation (SDI) significantly influenced this parameter during 2022. Ibba et al. (2023b) provided compelling evidence that the implementation of the regulated deficit irrigation (RDI) strategy can result in significant water savings while maintaining or even enhancing agriculture, indicating that water availability during the sensitive period (SP1 and SP2) of the season has a good impact on production, aligning with findings from Correa-Tedesco et al. (2010) and Iniesta et al. (2009). During the first irrigation season, the study reaffirms these observations by demonstrating that reducing applied water by 30% or 40% during a non-sensitive period (pit hardening) resulted in a 10–19% reduction in cumulative sap flow volume without adversely impacting plant production in T1 or T2 treatments (Fig. 4; Table 3). Notably, full irrigation during sensitive phases (SP1 and SP2) is essential for maximizing fruit yield, as evidenced by treatments T1 and T2. T3 and T4 treatments exhibited slight decreases in fruit production (approximately 13% and 23%, respectively), correlating with significant reductions in cumulative sap flow volume (37% and 54%, respectively) due to reduced irrigation (Table 3). Despite a 54% reduction in cumulative sap flow volume in 2022, the T4 treatment demonstrated efficacy in fruit yield production. However, this treatment experienced a notable decline in cumulative sap flow (1646 L) in 2023 (Fig. 4). The T3 treatment, despite a 37% reduction in cumulative sap flow volume during the first irrigation season, displayed commendable fruit yield performance, suggesting adaptation to consecutive seasons of water deficit conditions. Several studies suggest that RDI may lead to slight reductions in fruit yield (Ibba et al. 2023a, b; Lu et al. 2019). However, plants subjected to moderate RDI can quickly recover upon returning to normal irrigation levels, minimizing the impact on yield (Kang et al. 2000). A strong polynomial correlation between fruit yield and cumulative sap flow in olive trees (Fig. 8). This relationship was observed across different irrigation treatments and supported by a significant correlation coefficient in Pearson’s analysis (Table 5). Specifically, the findings suggest that well-irrigated olive trees tend to transpire more and produce higher yields.

The variations observed in irrigation treatments during two consecutive growing seasons were due to the cumulative effect

of deficit irrigation strategies. As the water deficit accumulates over two years, the impact becomes more noticeable, resulting in different responses across irrigation regimes, as illustrated in Fig. 9. In general, Menara olive trees exposed to deficit irrigation, particularly the RDI treatments, showed the ability to adapt and cope with low water supply over time. However, the SDI treatment (T6) had a cumulative water shortage effect, resulting in a decrease in the agronomical and physiological performance of Menara olive trees. It is crucial to highlight that the effect of drought on plants is mainly determined by cumulative responses over time (Zhao et al. 2020). The variation in fruit yield and sap flow observed across consecutive years indicates the alternate bearing pattern commonly seen in olive trees, where fruit load fluctuates between high (2022) and low (2023) from year to year (Supplementary Table S2 and S5, Figs. 3 and 8). This phenomenon significantly affects olive production, with the “off” year in 2023 resulting in reduced sap flow and yield despite irrigation strategies. Studies by Bustan et al. (2016) have shown a linear increase in whole-tree transpiration with fruit load in olive trees. According to Çiğdem et al. (2016), the first year is typically an “off year” for the ‘Gemlik’ olive variety, with significantly higher yields observed in the second year (on year), representing a nearly 72% increase compared to the first year. Previous studies by Miserere et al. (2019) have suggested that olive trees experience decreased sap flow when fruit load is low, consistent with the findings of reduced production in the Menara cultivar in 2023. Additionally, the year itself emerged as a significant factor impacting stomatal conductance across all irrigation treatments (Supplementary Table S3, Fig. 5). This parameter increased with a lower fruit load. While some studies (Martín-Vertedor et al. 2011; Naor et al. 2013) have shown increased stomatal conductance with higher fruit load, others (Proietti et al. 2006; Bustan et al. 2016) have reported contrasting results, consistent with the findings of the Menara cultivar. These findings emphasize the complex interactions influencing olive tree physiology and productivity under varying fruiting conditions.

Conclusions and perspectives

In this study, we investigated the performance of two deficit irrigation strategies: regulated deficit irrigation (RDI) and sustained deficit irrigation (SDI), differing in irrigation timing and water application, on ‘Menara’ olive trees. Our findings revealed that implementing the RDI strategy can result in significant water savings, while preserving or even increasing olive production. The desired level of productivity in our study refers to achieving olive production levels similar to or approaching those of the control treatment (T0), while also maximizing water savings to enhance orchard profitability and benefit olive growers. Among the RDI and SDI treatments evaluated,

T1, T2, and T3 demonstrated the ability to achieve the desired productivity level defined as similar or approaching that of the control treatment (T0) while conserving water resources. T1, T2 treatments achieved this desired productivity level with water savings of 11–14% in total applied water. Notably, the T3 treatment, involving reduced irrigation during sensitive periods (SP 80 - NP 70% ETc), aimed at a 24% reduction in water requirements for the ‘Menara’ variety, resulting in significant water savings without compromising productivity. Despite experiencing a substantial 37% reduction in cumulative sap flow volume, the T3 treatment only exhibited a slight decrease in fruit production, amounting to approximately 13%. Importantly, this reduction did not significantly affect key physiological parameters such as stomatal conductance (gs) and chlorophyll fluorescence (Fv/Fm), indicating the plants’ ability to adapt to modest water deficit conditions. Moreover, this treatment contributes to the overall sustainability of agricultural systems, ensuring their continued productivity and economic viability, particularly under changing climatic conditions over consecutive years. Conversely, the T6 treatment (60% ETc) within the SDI strategy experienced a substantial decline in production, exhibiting a 50% reduction when subjected to a 40% reduction in applied water and an 80% decrease in cumulative sap flow volume in 2022. On the other hand, the RDI strategy could induce trees to make appropriate physiological adjustments and enhance their resistance to stress and production decline. This approach conserves water and contributes to the sustainability of olive trees, ensuring continuous productivity over the years 2022 and 2023.

Extending observation durations and expanding the range of olive cultivars used in research investigations are recommended actions. This addition allows for a more in-depth investigation of long-term performance dynamics, providing vital insights into the adaptation processes of olive groves to climatic change over time. Furthermore, assessing the adaptability and efficiency of deficit irrigation strategies across various geographical locations and climate zones is critical. Tailoring these regulated and sustained strategies to varied environmental conditions ensures their global applicability.

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Declarations

Competing of interest The authors declare no competing interests.

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