

## PHYSIOLOGICAL ADAPTATIONS OF HENNA PLANT (*LAWSONIA INERMIS* L.) TO DIFFERENT IRRIGATION CONDITIONS IN TUNISIAN ARID REGION

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### ABSTRACT

In this study, we aim to investigate the photosynthetic adaptation of henna plants to various irrigation regimes and describe the effects of water stress on their growth performance and chlorophyll contents. For this reason, an experiment of four months was conducted. Henna plants were first grown in a greenhouse and then, exposed to three different irrigation regimes, whereby, they were irrigated up to the field capacity (FC) of 0% (control, T0), 50% of the control (moderate stress, T1) and 25% of the control (severe stress, T2). Results showed that exposure to drought induced an important decrease in gas exchange and photosynthetic pigment contents as compared to control. These shifts became more pronounced under severe stress (T2). Furthermore, after two harvests made, stomatal conductance and photosynthetic assimilation rates were significantly reduced under T2 conditions. In addition, a decline in total Chlorophyll amount and shoot dry weight was noticed, whereas, increases in the intrinsic water use efficiency and in the root-shoot ratio were discerned, especially, under the severe drought. These results revealed a close relationship between henna plants and irrigation dose and showed that for photosynthesis process and growth performance, the most appropriate water regime was the moderate stress (T1) rather than the severe stress (T2).

**Key words:** gas exchange, growth, *Lawsonia inermis*, photosynthesis, intrinsic water use efficiency, water stress.

### INTRODUCTION

In Tunisia, henna has been known since 1400 to 500 BCE. It has been used extensively as a traditional remedy, its leaves; flowers, seeds, stem bark and roots are used to treat rheumatoid arthritis, headache, ulcers, diarrhea, fever, diabetes, and cardiac diseases (Subbaiah and Savithramma, 2012). Besides, henna leaves are used in cosmetics for staining hands and hair. Henna plants grew best in tropical savannah and tropical arid zones that produced highest dye content in temperatures between 35 and 45 °C (Chaudhary, 2010).

On the other hand, in the arid and semi-arid Mediterranean climates, rainfall is limited and unreliable resulting in various abiotic stresses, generally, drought and salinity (El-Beltagy and Madkour, 2012). Drought is the most complex and devastating on a global scale and its frequency is expected to increase as a consequence of climate changes (Ceccarelli *et al.*, 2010). Consequently, the fact that Mediterranean agrosystem must face up to the need to cope with water scarcity cannot be questioned because any strategy of constant expansion of the supply is invalid. Because of these restrictions, it is important to consider the use of drought tolerant species for revegetation and to preserve soils with little plant cover (Morales *et al.*, 1998).

Studies showed that water stress affects various physiological and biochemical parameters and these

effects depend on genotype, degree of water limitation and duration of treatment (Petridis *et al.*, 2012; Husnain, 2014). Moreover, the drought impacts on plants differ both with endogenous factors, e.g. plant height, stomatal size and density and root structures, and with environmental conditions such as soil and air temperature, Photosynthetically active radiation (PAR) and air humidity (Bollig and Feller, 2014). Besides, plants have employed a variety of physiological and morphological strategies that allow them to cope with drought stress (Auge *et al.*, 2003). Indeed, under drought stress, the high relative apoplastic water content (42–58%), commonly seen in xeromorphic plants would contribute to the retention of water at low leaf water potentials (Rodriguez *et al.*, 2012). Furthermore, Ünayyar *et al.* (2004) have also found that resistant genotypes of sunflower leaves had higher RWC in water-stressed plants.

On the other hand, it has been previously reported that oasis plants confronted drought by developing stress avoidance and stress tolerance mechanisms (Sun *et al.*, 2013). Besides, from the time of deficit irrigation began to be applied, investigations have shown a decrease in the stomatal conductance in order to control water loss via transpiration and to avoid leaf turgor loss (stress avoidance mechanism) (Connor, 2005). However, other plants like *G. hirsutum*, have responded to water deficit by decreasing stomatal conductance and by increasing the photorespiration, and the ratio of dark

respiration, thereby limiting net photosynthesis rate and decreasing lint yield (Chastain *et al.*, 2014). Besides, in the reduced soil water content, it has also been demonstrated that chlorophyll concentration and fluorescence of some cultivars have decreased (Sun *et al.*, 2013). In a previous work, we showed that moderate drought (T1) did not damage henna morphology (leaf area, leaf number and stem length); however, at severe deficit irrigation stress (T2), plants seemed to be more sensible to drought (Enneb *et al.*, 2015). The aim of the present study was to investigate, if the irrigation with the three different water regimes will positively influence the growth and photosynthetic characteristics of *L. inermis*, originated from Gabes, in southern Tunisia.

## MATERIALS AND METHODS

**Plant growth conditions:** Seeds of *Lawsonia inermis* L. were obtained from plants that were collected from the oasis of Chenini-Gabes, one of the most important oases of the southern Tunisia (Latitude 33° 53' N, Longitude 10° 12'E). The climate is Mediterranean and dry, characterized by hot summers and mild winters. Rainfall is low and erratic with an average of 186 mm. The annual potential evapotranspiration is estimated at 1417 mm year<sup>-1</sup> (Kouki and Bouhaouach, 2009). Henna seeds were then surface sterilized with sodium hypochlorite (NaOCl) solution for 2 min and germinated at room temperature for two weeks on moistened filter paper in Petri dishes wrapped with parafilm. The maximum of seed germination rate was observed after 15 d. Thus, at this stage, henna seedlings were transferred to 4 L plastic pots filled with a mixture of sand and soil (1:2). Each pot was irrigated 3 times per week. An experiment of 4 months was carried out at the Institute of Arid Region, Medenine-Tunisia and plants were then either subjected to water deficit or continually well-watered (control: T0). Control pots were irrigated several times each week to maintain soil moisture near field capacity (FC), while stress pots experienced soil drying by withholding irrigation until they reached 50% of FC for moderate stress (T1) and 25% FC for severe stress (T2). The glasshouse conditions were as follows: 25 ± 1°C temperature, 50% day / 75% night relative humidity and 16 h light / 8 h dark regime. At 2-month-old, henna plants were submitted to three irrigation treatments: (T0, T1, T2), the irrigations were applied from August and maintained until November. Two harvests were made, 3-month-old plants: First harvest and 4-month-old plants: second harvest. The experiment was arranged with 3 water regime treatments × 4 replicates. Henna plants were removed from pots and dipped in a water filled bucket. The plant roots were washed with distilled water carefully to remove the adhering soil particles.

**Growth and hydration measurements:** After the two harvests, 3- and 4-month-old plants were divided into shoots and roots. Their respective fresh weights (FW) were measured immediately while the dry weights (DW) were determined after placing plants in an oven at 70°C for 48 h, till the weight became constant. FW of roots and shoots were taken by digital single pan balance. Turgid weights (TW) were obtained after soaking leaves in distilled water in test tubes for 12 h at room temperature (~20°C), under low light laboratory conditions. After that, leaves were quickly and carefully blotted and dried with tissue paper for determining the TW. RWC was measured in the third youngest fully expanded leaves that were harvested from three different plants in the morning. RWC was determined using the following equation:  $RWC (\%) = 100 \times [(FW - DW) / (TW - DW)]$ .

**Gas exchange parameters:** Photosynthetic gas exchange parameters were measured between 10:00 and 12:00 h using a portable photosynthetic meter model (LCi, IRGA; ADC Bioscientific Ltd.). The leaf was irradiated with PAR of 1.500 μmol m<sup>-2</sup> s<sup>-1</sup> of internal light source. The CO<sub>2</sub> concentration in the leaf chamber was set at 360 mmol mol<sup>-1</sup> and temperature was kept at 30° C. The third youngest fully expanded leaves were used for these measurements. Readings were logged every 30 s until stable values for photosynthetic assimilation rate (Pn), stomatal conductance (gs) and transpiration rate (E) were reached. Intrinsic water use efficiency (IWUE) was obtained as the ratio of photosynthetic assimilation rate (Pn) to stomatal conductance for water vapor (gs). The results were expressed as follows: photosynthetic assimilation rate (μmol CO<sub>2</sub> m<sup>-2</sup> s<sup>-1</sup>), stomatal conductance for water vapor (mol H<sub>2</sub>O m<sup>-2</sup>s<sup>-1</sup>) and intrinsic water use efficiency (μmol CO<sub>2</sub> mol<sup>-1</sup> H<sub>2</sub>O).

**Chlorophyll content analysis:** Chlorophylls (Chl) a and b were determined following the method described by Arnon (1949). The absorbance of each extract was measured at 663 and 645 nm. Chl content (mg g<sup>-1</sup> DW) was calculated using the following equations:

$$\text{Chl a} = 12 (\text{DO } 663) - 2.67 (\text{DO } 645)$$

$$\text{Chl b} = 22.5 (\text{DO } 645) - 4.68 (\text{DO } 663)$$

**Statistical analysis:** Statistical analysis was performed using SPSS version (15.0). Data collection was carried out regarding four replications of the same henna cultivar for each measurement. A couple of factors (3 irrigation levels vs 2 harvests) have been subject of statistical analysis. Analysis of variance (ANOVA) at =0.05, 0.01 and 0.001 showed differences between and within factor combinations. Duncan's new multiple range test (MRT) was used to compare means and to distinguish between henna responses to each irrigation regime and for each harvest.

## RESULTS

**Leaf water status:** Our results showed that *L. inermis* leaves maintained higher water content, under both stress conditions (T1 and T2) in three-months-old plants (first Harvest) but not in four-months-old plants (second Harvest). Moreover, RWC retained high values, especially, after the first harvest with a significant difference between treatments. In fact, in T2-stressed plants, RWC decreased by 5.8% and 13.05%, after first harvest, as compared to control (Fig.1). In table 2, the ANOVA test showed interaction between RWC, treatment and harvest. These results suggested that *L. inermis* is significantly tolerant to water shortage up to 25% FC.

**Growth performance:** As shown in Figure 2, water deficit caused a significant reduction ( $P < 0.05$ ) in the shoot DW of the T2-stressed henna, compared to the optimal irrigation (T0) after second harvest. Indeed, in response to severe stress (T2), *L. inermis* reduced the shoot DW by 58.24% (second harvest), as compared to T0-plants. Similarly, four-month-old plants were significantly affected by both treatments T1 and T2. Growth inhibition was significantly more pronounced for plants cultivated under severe stress (T2; - 55.64%) than those cultivated under moderate stress (T1; -15.88%). But this reduction is less pronounced compared to treated plants of the first harvest (Fig. 2a). While root DW increased significantly with treatments for the second harvest, it showed a slight increase for the first harvest compared to their control (Fig. 2b). After the first harvest, henna plants exhibited an increase of 49.27 and 92.82% for T1 and T2 respectively. This root growth improvement was more pronounced (101.79%) in four-month-old plants subjected to severe drought (Fig. 2b). Root to shoot ratio increased with treatments and not with time of harvest. This increase was significant only for T2 in the first harvest but was significant for both treatments in the second harvest (Fig. 2c). ANOVA test indicated significant interaction between root-shoot ratio and water deficit but not time of harvest (Table 1).

**Gas- exchange and Chlorophyll content:** Amount of photosynthetic pigments, chlorophyll a (Chla), chlorophyll b (Chlb), and total chlorophyll (chla + chl b) declined in the leaf tissues for both time of harvest, especially at severe stress (Table 2). In three-month-old plants, degradation of chla, Chlb and total chlorophyll in the plants subjected to moderate stress (50% FC) was 30.27%, 23.62% and 27.7%, respectively, whereas at severe stress (25% FC) it was 62.28%, 80.31% and 69.25%, respectively. Under extreme water stress (25% FC), Chla, Chlb and total chlorophyll in four-month-old plants were significantly declined (by 54.1%, 66.01% and 58.93%) compared to control plants. However, this decrease was very low compared with that recorded in

three-month-old plants (Table 2). Repeated measures ANOVA, treatment; time of harvest and treatment  $\times$  time of harvest were very highly significant ( $P < 0.001$ ), highly significant ( $P < 0.01$ ) and significant ( $P < 0.05$ ) respectively (Table 1).

Water deficit was followed by a gradual decline in net photosynthesis rate (Pn), stomatal conductance (gs) and transpiration rate (E) for both time of harvest as compared to their control respectively ( $P < 0.001$ ) as shown in Fig. 4. The values of Pn of well-watered plants ranged from 5.97 to 9.15  $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$  at the first and second time of harvest respectively; however, there is a significant decrease in the Pn of stressed plants, by 21.4 and 50.57% for moderate and severe stress respectively at the first harvest. This depressive effect was more pronounced for the second harvest. Differences of Pn increased with time of harvest for both treatments (Fig. 3a). Similarly, E reached the highest values in control plants (4.28 and 4.33  $\text{mmol H}_2\text{O m}^{-2}$ ), and was to some extent inhibited by 30.84 % and 63.24 % for moderate stress (50% FC), and by 28.62 % and 57.15 % for severe stress (25% FC) at the first and second harvest respectively (Fig. 3b). Repeated measures of E and gs data were significantly affected by the treatment ( $P < 0.001$ ), but showed no differences between time of harvest and interaction between treatment and time of harvest ( $P > 0.05$ ) (Table 1). Plants, cultivated under moderate and severe stress, closed their stomata considerably compared to control plants for both harvests. Stomatal closure of four-months-old plants increased by 44.71% of the control values for moderate stress, and by 75.29% for severe stress (Fig. 3c). Intrinsic WUE ( $\text{WUE}_i = \text{ACO}_2 \text{ gs}^{-1}$ ) showed a significant increased divergence between drought-stressed and well-watered plants, especially for severe stress. For the first harvest, stressed plants displayed higher  $\text{WUE}_i$  values than in control plants with a raise of about 48.63% and 134.84% under moderate and severe stress respectively. Similar pattern of increasing intrinsic WUE was observed for the second harvest. The values of intrinsic WUE of stressed plants reached 42.34 and 55.64  $\mu\text{mol CO}_2 \text{ mol}^{-1} \text{ H}_2\text{O}$  showing an increase of about 30.17 and 71.08%, under moderate and severe stress respectively, compared to control plants (Fig. 3d). Repeated measures ANOVA, treatment; time of harvest and treatment  $\times$  time of harvest were very highly significant ( $P < 0.001$ ), highly significant ( $P < 0.01$ ) and significant ( $P < 0.05$ ) respectively (Table 1).

## DISCUSSION

**Effects of water stress on henna leaf hydration:** Relative water content (RWC) was significantly reduced in severe stress conditions, for the second harvest as compared to control. Similarly, several studies have reported a decrease in RWC under severe water deficit (Gorai *et al.*, 2010). The significant decline of RWC was

reported in several species such as *Oudneya africana* (Talbi *et al.*, 2014) and chickpeas (Khanna *et al.*, 2014). Despite the decline in RWC values, stressed plants kept their tissues well hydrated (around 80%) compared to controls. These results corroborate with other works showing that the ability of the plant to cope with drought may depend on different mechanisms, including its capacity to maintain a high RWC which can be considered as an index for drought tolerance (Oukarrouma *et al.*, 2007; Khanna *et al.*, 2014). The high RWC of henna during water deficit could be an import strategy for acquiring tolerance to growth in extreme drought conditions.

#### Effects of water stress on the growth performance:

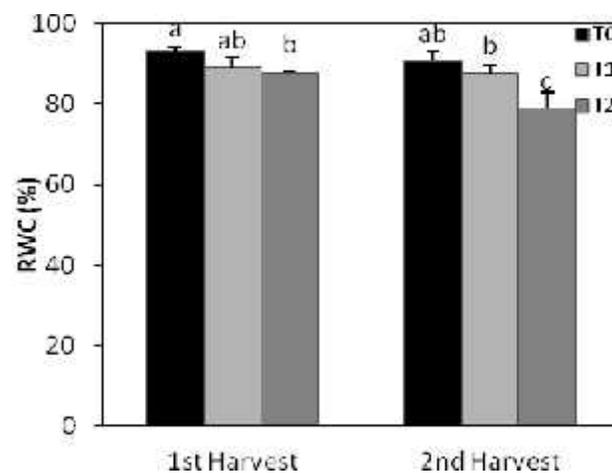
Water deficit caused a significant decrease in shoot DW and a noticeable increase in root biomass of henna plants. The reduction in shoot DW (41.53–61.45%) for the first and (15.88–58.24%) for the second harvest under moderate and severe stress respectively was similar to that reported by Van Den Boogaard *et al.* (1996). As was shown in *A. gombiformis* (Boughalleb *et al.*, 2014), this slowdown in the growth is a function of adaptation for the survival of plants under stress, reorienting redirect cells resources (energy and metabolic precursors) in the direction of defensive reactions against stress. Under drought *Lawsonia inermis* L. increased their root DW (Fig.2b). Root growth improvement was also reported for *A. gombiformis* (Gorai *et al.*, 2010). This tendency indicated greater ability of *L. inermis* to adapt to moisture stress (Montero *et al.*, 2001) due to greater root biomass allocation.

Root to shoot ratio increased with treatments and not with time of harvest. This increase is one of the mechanisms involved in the acclimation of plants to drought (Turner, 1997). Stressed plants divert assimilates to root growth under water stress, resulting in a high root to shoot ratio (Lucero *et al.*, 1999). In fact, the increase in root to shoot ratio in *Lawsonia inermis* was due to root dry matter accumulating and not just a decrease in shoot dry matter. Similarly, Lambers *et al.* (1998) expected larger root to shoot ratio under reduced water availability suggesting large biomass allocation to roots relative to shoots facilitating water and nutrient uptake.

#### Effects of water stress on the photosynthesis apparatus:

Drought-induced decreases in photosynthetic pigments contents for both time of harvest especially at severe stress (T2). Leaf Chl content declined, to a greater extent for Chlb than Chla for both harvests. Indeed, Chla and Chlb decreased by 62.28 and 80.31% for the first harvest. However, when prolonging water deficit, the diminution in Chla (54.1%) and Chlb (66.01%) was less marked. These results show that henna succeeded to acclimate with severe water deficit. The chl content is known to decrease as the relative water content and leaf water potential decreases (Lawlor and Cornic, 2002).

Drought-induced decreases in chlorophyll contents have been reported previously in various species (Anjum *et al.*, 2011; Oraki *et al.*, 2012).



**Fig. 1. Changes in relative water content (RWC) of henna leaves exposed to T0 (control), T1 (moderate deficit irrigation (50%FC)) and T2 (severe deficit irrigation (25%FC)) treatments after two harvests (3- and 4-month-old plants). Means  $\pm$  S.E. ( $n = 4$ ). Means with different letters (a, b, c) are significantly different at  $P < 0.05$  and values sharing a common letter are not significantly different at  $p < 0.05$ .**

Water deficit was followed by a gradual decline in net photosynthesis rate (Pn), stomatal conductance (gs) and transpiration rate (E) for both time of harvest as compared to their control respectively (Fig. 4). Compared to control plants, severe stress led to a rapid inhibition of Pn and gs (Fig. 3a–c) during drought periods (first and second harvests), consistent with previous studies (Vyas *et al.*, 2001; Zhang *et al.*, 2004). This reduction is related especially with RWC decline in plants subjected to severe stress for the second harvest. These observations are in conformity with what Yayong *et al.* (2011) remarked for physiological acclimation of two psammophytes to repeated soil drought. Also, Vyas *et al.* (2001) showed that, in cluster bean, Pn was lower under drought-stress conditions. Plants, cultivated under moderate and severe stress, closed their stomata considerably compared to control plants for both harvests. Stomatal closure of four-months-old plants increased by 44.71% of the control values for moderate stress, and by 75.29% for severe stress (Fig. 3c). Galle *et al.* (2007) have reported that stomatal closure protects against further water loss and irreversible cell dehydration under progressing soil drought conditions, consistent with the present study. Generally, physiological responses to water stress were associated with carbon fixation and can be conveniently

divided into stomatal and non-stomatal responses (Flexas and Medrano, 2002). stomatal closure was one of the earliest responses of the plants to drought resulted in reducing CO<sub>2</sub> uptake, photosynthetic activity and transpiration (E), (De Souza *et al.*, 2013). E declined by 30.84 % and 63.24 % for moderate stress (T1), and by 28.62 % and 57.15 % for severe stress (T2) at the first and second harvest respectively, as compared to control henna plants (Fig. 3b). Furthermore, stomatal control in response to varying moisture stress was the most important step contributing to the photosynthesis process. However, non-stomatal response, such as mesophyll resistance or photosynthetic mechanisms may be important aspects of stress tolerance (Pinheiro and Chaves, 2011). Many studies have used stomatal conductance as a reference parameter which explain the gradual response to water stress which first acts as a partial closure leading to a metabolic adjustment through limited generation of ribulose-1, 5-bisphosphate (RuBP) and thereafter as the drought stress progressed, the closure of stomatal leads to reduced photochemistry and then carboxylation efficiency (Rennenberg *et al.*, 2006). Moreover, an increase in WUEi was occurred in henna stressed plants; this behavior suggests that *L. inermis* presents a greater capacity to protect its photosynthetic apparatus against water stress. Similar results were reported by Passos *et al.* (2005) and Aasamaa *et al.* (2010).

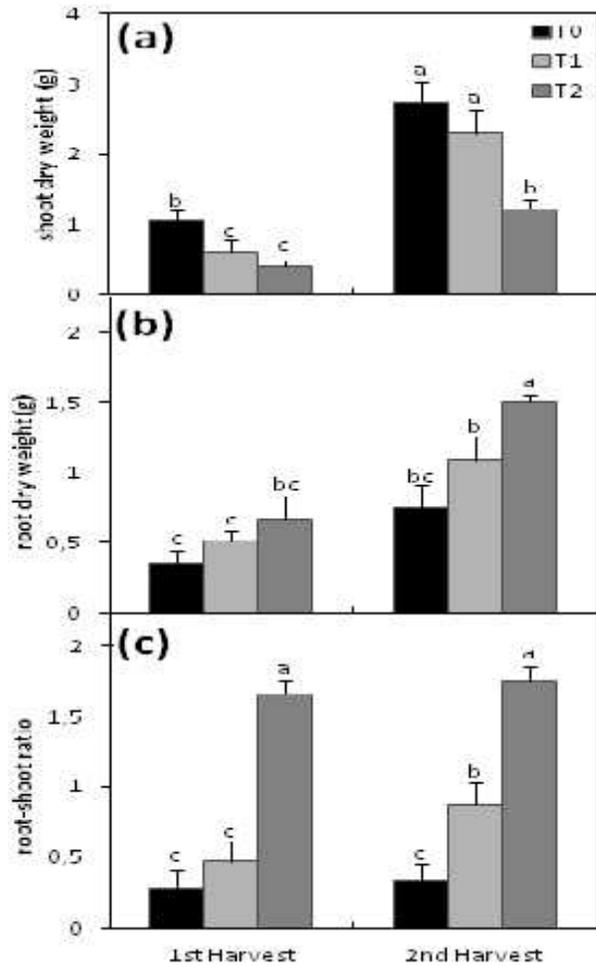


Fig. 2. Changes in (a) Shoot DW, (b) Root DW and (c) Root-Shoot DW ratio for henna plants in T0 (control), T1 (moderate deficit irrigation (50%FC)) and T2 (severe deficit irrigation (25%FC)) treatments at both harvest during the experimental period. Means  $\pm$  S.E. ( $n = 4$ ). Means with different letters (a, b, c) are significantly different at  $P < 0.05$  and values sharing a common letter are not significantly different at  $p < 0.05$ .

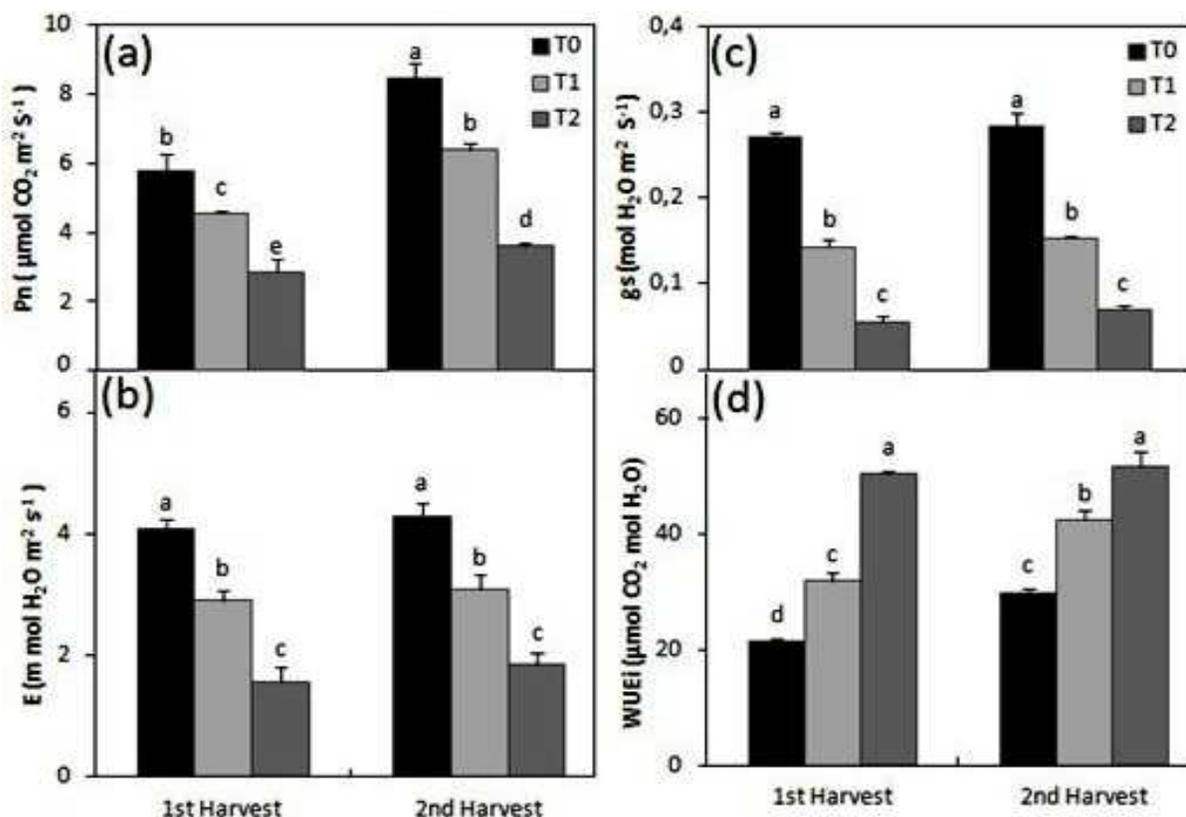


Fig. 3. Variation in (a) Net photosynthetic rate (Pn), (b) transpiration rate (E), (c) stomatal conductance (gs) and (d) intrinsic water use efficiency (WUEi) for henna plants in T0 (control), T1 (moderate deficit irrigation (50%FC)) and T2 (severe deficit irrigation (25%FC)) treatments at both harvest during the experimental period. Means  $\pm$  S.E. ( $n = 4$ ). Means with different letters (a, b, c, d and e) are significantly different at  $P < 0.05$ .

Table 1. Analyses of variance for relative water content RWC (%), photosynthetic assimilation rate (Pn) ( $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ ), transpiration E ( $\text{m mol H}_2\text{O m}^{-2} \text{ s}^{-1}$ ), stomatal conductance gs ( $\text{mol H}_2\text{O m}^{-2} \text{ s}^{-1}$ ), intrinsic water use efficiency WUEi ( $\mu\text{mol CO}_2 \text{ mol H}_2\text{O}^{-1}$ ), total Chlorophyll (Chl) content ( $\text{mg g}^{-1} \text{ FW}$ ) and dry root-shoot ratio in henna plant (*L. inermis*) ( $n=4$ ).

Source of variation	RWC	Pn	E	gs	WUEi	Chla +Chb	Root-shoot ratio
Treatment	***	***	***	***	***	***	*
Harvest	**	***	ns	ns	***	***	ns
Treatment*Harvest	*	*	ns	ns	*	**	ns

Within each column, different letters indicate significant differences at  $P < 0.05$  (Duncan's test). n.s., \*, \*\* and \*\*\* indicate non-significant or significant differences at  $P < 0.05$ , 0.01 or 0.001, respectively.

Table 2. Variation in chlorophyll a, chlorophyll b and total chlorophyll (a+b) ( $\text{mg g}^{-1} \text{ FW}$ ) of henna plants exposed to T0 (control), T1 (moderate deficit irrigation (50%FC)) and T2 (severe deficit irrigation (25%FC)) treatments after two harvests (3- and 4-month-old plants). Means  $\pm$  S.E. ( $n = 4$ ).

	1st Harvest			2nd Harvest		
	T0	T1	T2	T0	T1	T2
Chl a	0,403 $\pm$ 0,023 <sup>c</sup>	0,281 $\pm$ 0,029 <sup>d</sup>	0,152 $\pm$ 0,035 <sup>e</sup>	0,682 $\pm$ 0,029 <sup>a</sup>	0,510 $\pm$ 0,0243 <sup>b</sup>	0,313 $\pm$ 0,017 <sup>d</sup>
Chl b	0,254 $\pm$ 0,022 <sup>b</sup>	0,194 $\pm$ 0,021 <sup>c</sup>	0,050 $\pm$ 0,017 <sup>e</sup>	0,459 $\pm$ 0,015 <sup>a</sup>	0,266 $\pm$ 0,009 <sup>b</sup>	0,156 $\pm$ 0,018 <sup>d</sup>
Chl a+Chl b	0,657 $\pm$ 0,045 <sup>c</sup>	0,475 $\pm$ 0,065 <sup>d</sup>	0,202 $\pm$ 0,019 <sup>e</sup>	1,142 $\pm$ 0,015 <sup>a</sup>	0,777 $\pm$ 0,032 <sup>b</sup>	0,469 $\pm$ 0,027 <sup>d</sup>

Values with different letters (a, b, c, d and e) in a row are significantly different at  $P < 0.05$ .

**Conclusion:** As a conclusion, water stress seems to be the main limitation factor to primary photosynthetic process in henna. Under the mild stress (T1), *L. inermis* plants showed higher photosynthetic performance, with smaller decreases in the rates of Pn, E and gs, because the water availability in the soil was higher than under severe stress (T2). Furthermore, *L. inermis* responded to water stress by increasing root biomass accumulation, intrinsic water use efficiency and by reducing shoot biomass, Pn, E and gs. These results suggest that henna plants adopt the strategy of avoidance (stomatal closure) as a mechanism of tolerance to drought that may probably prevent stressed plants from physiological and biochemical damages occurred during drought periods. In addition, applying an appropriate irrigation regime is recommended to improve the photosynthetic efficiency and to alleviate the rapid photodamage to PSII, which would increase the commercial value of *L. inermis* in Tunisia.

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