

Change in photosynthetic pigments of Date palm offshoots under abiotic stress factors

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Abstract

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Increasing world temperatures are bringing about climate changes creating abiotic stress in plants. Date palm offshoot leaves (Khadrawi cv.) were analyzed for chlorophyll Chl *a*, Chl *b*, Total Chl, Chl *a/b* ratio, anthocyanin and carotenoid subject to salinity, drought and temperature stress under field conditions. Results demonstrated that drought and salinity stress accompanied by high temperatures in July and August significantly reduced the Chl *a*, Chl *b*, and Total Chl relative to the control. Anthocyanins, carotenoids, hydrogen peroxide, and malondialdehyde were markedly higher in July and August (45 °C), whereas September showed lower values in these substances. Temperature reduction to 35 °C accompanied by drought or salinity stress, brought about a critical increment in relative water content and a decrease in electrolyte leakage. Although the impact of drought and salinity stress continued, the reduced temperatures in September resulted in a reduction of abscisic acid and proline concentration. Cluster analysis showed the two groups. In this first group, the significant similarity between the treatments is illustrated by the influence of the high temperature of 43–45 °C. Recovery of photosynthesis following low-temperature, for the most part, determines plant flexibility to water deficiencies and salinity. Thermal stress, associated with salinity or drought stress is more damaging to the photosynthetic pigments than any single factor.

Keywords

antioxidative mechanism, electrolyte leakage, lipid peroxidation, *Phoenix dactylifera* L.

Introduction

The increase in world temperatures is bringing about climate change resulting in significantly higher abiotic stresses in crop plants (MATHUR et al., 2014)ribulose-1,5-bisphosphate carboxylase/oxygenase (Rubisco). The recurrence and duration of such abiotic stresses will soon be intensified with the warming of the global climate and its impact on annual crop plants in critical periods of growth (LAMAOUI et al., 2018).

Together, salinity, drought, and heat stress commonly account for a decline in photosynthetic pigment substances, especially chlorophyll. This reduction occurs because of stress-initiated alterations in pigment synthesis or pigment degradation. Further, the degree of those phenomena depends on the species, type of stress, duration of the stress period, and resilience to the stresses (WATKINS et al., 2017). Anthocyanin assumes an essential function in protecting plants through scavenging receptive ROS (URBAN et al., 2017). Likewise, carotenoids are critical for the

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photoprotection of photosynthesis and plants experiencing abiotic/biotic stress (PANDEY et al., 2017). Moreover, understanding the biosynthesis/degradation of photosynthetic pigments is, in fact, at the center of the plant stress tolerance mechanism underlying abiotic stress factors (AL OMRON et al., 2012).

Under field conditions, abiotic stress conditions like drought and salinity infrequently happen in separation, and the course of many stress burdens is usually not predictable through single-factor examinations on account of synergistic, threatening, or, on the other hand, masking impacts which can occur (RASTOGI et al., 2019) which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited. *Ocimum tenuiflorum* is a widely used medicinal plant since ancient times and still continues to be irreplaceable due to its properties. The plant has been explored chemically and pharmacologically, however, the molecular studies have been started lately. In an attempt to get a comprehensive overview of the abiotic stress response in *O. tenuiflorum*, *de novo* transcriptome sequencing of plant leaves under the cold, drought, flood and salinity stresses was carried out. A comparative differential gene expression (DGE). An instance of an opposite reaction to stresses is the perception that the constant temperature of photosynthesis can capture water, focusing on the plant leaves, in contrast with wetter conditions, indicating that one factor may shape another (CHAVES et al., 2012). The high temperature disrupts the synthesis of photosynthetic pigments, damages the chloroplast membranes, and lowers chlorophyll and other pigments. As well as increase the abscisic acid in controlling closure of stomata, compatible osmolyte, and the evolution of genes of an adaptation response (ZANDALINAS et al., 2016). In this case, the stomata prepare the CO₂ pathway to stabilize the carbon photosynthesis. And stomata prevent further water loss through the transpiration process to maintain the leaf temperature (REYNOLDS-HENN et al., 2010) bean (*Phaseolus vulgaris* L..

Date palm (*Phoenix dactylifera* L.) is the third most essential palm species in world agricultural industries, after coconut and African oil palm. Date palms are subjected to various abiotic stress factors in southern Iraq, such as salinity, drought, and heat; the palms try to regulate their growth and developmental processes to reduce the magnitude of injury caused by the environmental burden (SHAREEF, 2019). These changes are typically transient and reversible, and their induction level is determined by the exposure time to a particular ecological factor (ABBAS et al., 2015). Furthermore, Date palm offshoot growth decreases from May to September as a result of the abnormal state of salinity, water scarcity, and high temperatures (JASIM et al., 2016).

Most abiotic stress studies are performed under controlled conditions inside a research facility and do not duplicate the specific requirements found in the field. An impressive gap exists between the data generated from laboratory and greenhouse experiments, and the data required to create plants with expanded resilience of yields

under field conditions (PEREIRA, 2016). In particular, the chance to measure the responses of the plant stress *in vivo* is becoming increasingly important. In this study, we investigated photosynthetic pigment parameters as indicators in plants subjected to abiotic stress to determine the critical period in the summer season, plant responses and recovery ability to changes of temperature, which accompanies the salinity and drought stress factors.

Materials and methods

Research site and development conditions

Field analysis was performed at a private date plantation in the Alhartha region, Basrah, Iraq (30°37'52.68"N and 47°45'8.15"E), two growing seasons. Twenty uniform date palms, girth ±10 cm, vigorous 3–4-year-old 'Khadrawi' offshoots were used in the experiment. The selected offshoots were planted at 5 × 5 m spacing in silty clay loam soil. Regular soil EC was 4 dS m⁻¹, just as the normal EC water 5 dS m⁻¹, then drip irrigation followed for all treatments. In the first season, 15 May 2017 to 15 May 2018, each treatment was replicated five times. Each plant was treated with Control irrigated 75% field capacity (120 ml depth of water) without saltwater 0 dS m⁻¹, for the drought treatment, the offshoots irrigated without saltwater 0 dS m⁻¹ with 25% field capacity (40 ml depth of water), for salinity treatment irrigated 75% field capacity with salt water 5 dS m⁻¹, all these treatments subjected to the field temperature. The average temperature of the field area was in May 37 °C, June 40 °C, July 43 °C, August 45 °C, and September 35 °C. These averages were measured by hygro-thermometer at 1,100 h daily, to record the correct air temperature in circulation. There was no rainfall in the months of the experiments.

After one year of treatments, the same procedures were continued in the second season (15 May to 15 September 2018); the following data were recorded on day 15 of each month:

Parameters of the experiment

Pigment contents were extracted according to LICHTENTHALER and WELLBURN (1983). Hydrogen peroxide (H₂O₂) accumulation in date palm leaves determined following SERGIEV et al. (1997). Lipid peroxidation constrained by assessing the proportion of Malondialdehyde (MDA) arrangement using the thiobarbituric acid system after STEWART and BEWLEY (1980). Film strength was evaluated by estimating the conductivity of leachates contained in the harmed plasma layer following the strategy of SHANAHAN et al. (1990). In the proline focus, 0.5 g of dry leaves was homogenized with 5 mL of 95% ethanol, according to IRIGOYEN et al. (1992).

Extraction, purification, and quantitative assurance of free and bound abscisic acid were done, with minor changes, following the techniques of TANG et al. (2011). Leaf samples were weighed (fresh biomass) promptly after col-

lection and soaked in distilled water at 25 °C for 24 h, to estimate the RWC, according to JASIM et al. (2016).

Statistical analyses

Randomized completely block design of abiotic stress factors replicated five times were used in the experiment. Test information on all factors exposed to the investigation of fluctuation (ANOVA) systems utilized a factual bundle, SPSS (ver. 21.0), a Tukey's correction for multiple comparisons treatments considered at the $P \leq 0.05$ levels. The cluster analysis was performed using IBM SPSS (Ver. 23.0), targeting the attributes measured in the leaves with distances as similarity measures were applied to analyze the effect of treatments on photosynthetic pigments and biochemical parameters.

Results

Changes in photosynthesis pigments under abiotic stress

Data regarding Chl *a*, *b*, total Chl, Chl *a/b* ratio, anthocyanin, and carotenoid are presented in Figs 1–2, a–b. It is demonstrated that the drought and salinity stress accompanied by high field temperature in July (43 °C) and August (45 °C) significantly reduced the Chl *a*, *b*, and total Chl compared with the control, whereas anthocyanins and carotenoids increased considerably. In September (37 °C) Chl *a*, *b*, total Chl, Chl *a/b* ratio increased whereas anthocyanin and carotenoids significantly decreased. The higher temperature of the field in August (45 °C) recorded a considerably higher Chl *a/b* ratio, whether under stress (drought and salinity) or without stress. In September, the Chl *a/b* ratio decreased except under drought stress (Fig. 1d). The decrease in Chl *b* was very sharp in August, which brought about a higher Chl *a/b* proportion as the field temperatures peaked. The reduction of chlorophyll pigments was higher under salt stress than under drought stress (Fig. 1c).

Changes in the chemical response under abiotic stress

The salinity and drought treatments significantly increased electrolyte leakage, proline concentration, hydrogen peroxide, MDA, and ABA (Figs 2–3) in July and August, during both months; the average temperature remained between 43–45 °C, whereas the RWC decreased (Fig. 3d). These results are striking as a result of high air temperature causing a significant decrease in membrane stability using electrolyte leakage as index and RWC of all treatments to include freshwater, drought and salinity-treated plants (Fig. 3a–d). In contrast, reducing the temperature to (37 °C) in September, accompanied by both drought and salinity stress, resulting in a significant increase in membrane stability and RWC. Although the impact of drought and salinity stress continued, the reduction in temperature

in September resulted in a decrease of ABA and proline concentration (Fig. 2d). In contrast, ABA content in September was higher than May under drought stress (Fig. 3c). The production of H₂O₂ was lower in control in comparison to salt and drought treatments (Fig. 2c).

Cluster analysis of abiotic stress treatments

Hierarchical cluster analysis was carried out of abiotic factors represented by all 15 treatments and their effects on the photosynthesis pigments (Chl *a*, *b*, total Chl, Chl *a/b* ratio, anthocyanin, and carotenoid) of the leaf noted (Fig. 4). The cluster analysis showed two distinct clusters. The first group included drought, salinity, and control treatments in July and August (July-S, Aug-D, Aug-S, July-D, Aug-C). In this first group, the significant similarity between the treatments is illustrated by the influence of the high temperature of 43–45 °C while the second group included drought, salinity, and control treatments in May, June, and September (May-C, Sept-C, June-D, June-S, May-D, June-C, May-S, Sept-S, Sept-D), which showed the effect of low temperatures (37–40–35 °C) on improved plant growth, except for the control in July (July-C).

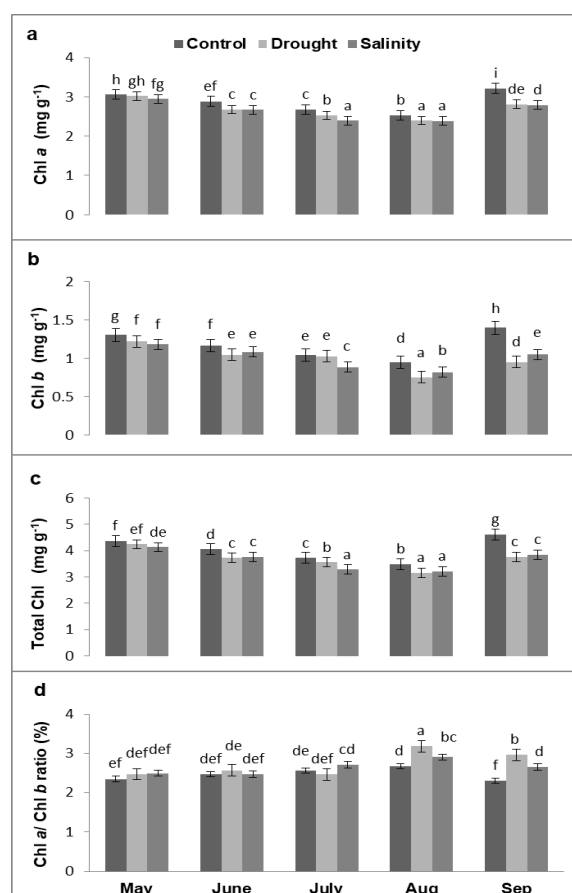


Fig. 1. Changes in Chl *a* (a), Chl *b* (b), total Chl (c), and Chl *a*/Chl *b* ratio (d) of Date palm offshoots leaves under different abiotic stresses. Results are means \pm SD ($n = 5$). The same letters are not significantly different $p \leq 0.05$ after Tukey's correction for multiple comparisons.

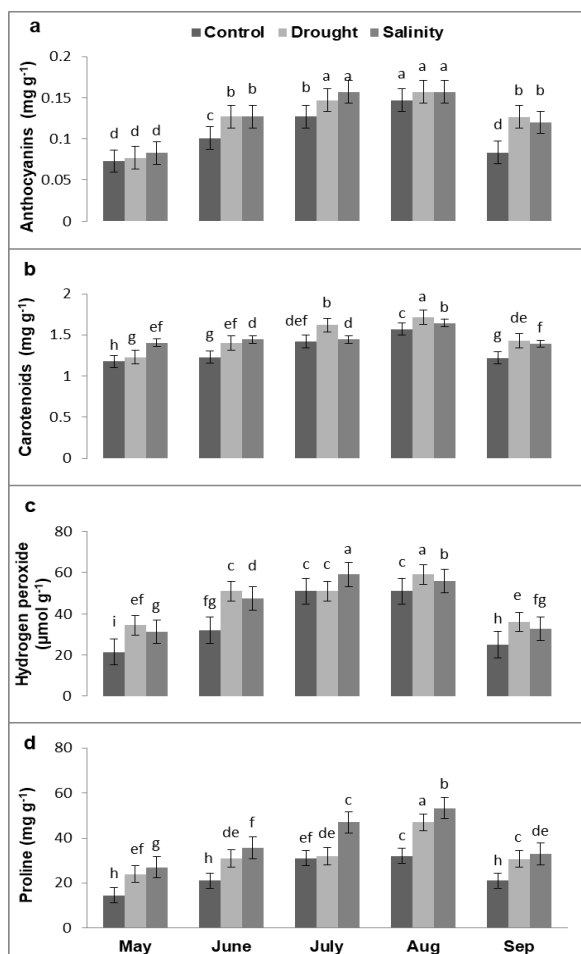


Fig. 2. Changes in anthocyanins (a), carotenoids (b), hydrogen peroxide (c), and proline concentration (d) of Date palm offshoots leaves under different abiotic stresses. Results are means \pm SD (n = 5). The same letters are not significantly different $p \leq 0.05$ after Tukey's correction for multiple comparisons.

Hierarchical cluster analysis of all parameters (pigments contents, hydrogen peroxide, malondialdehyde, electrolyte leakage, proline, ABA, and leaf RWC) affected by abiotic stress (Fig. 5) showed two distinct clusters. The second group included drought, salinity, and control treatments in May, June, and September (May-C, Sept-C, May-D, June-C, May-S), the significant similarity between the treatments is illustrated by the influence of the low temperature of (37–40–35 °C) on improved plant growth while the first group included drought, salinity, and control treatments in May, June, and September (June-D, July-C, Sept-S, June-S, July-D, Aug-C, Aug-D, Aug-S, July-S, Sept-D), which showed the interaction of high temperatures (43–45 °C) with drought and salinity on decreased plant growth.

Discussion

One of the pressing issues arising in some areas of Date palm offshoot cultivation is different levels of injuries

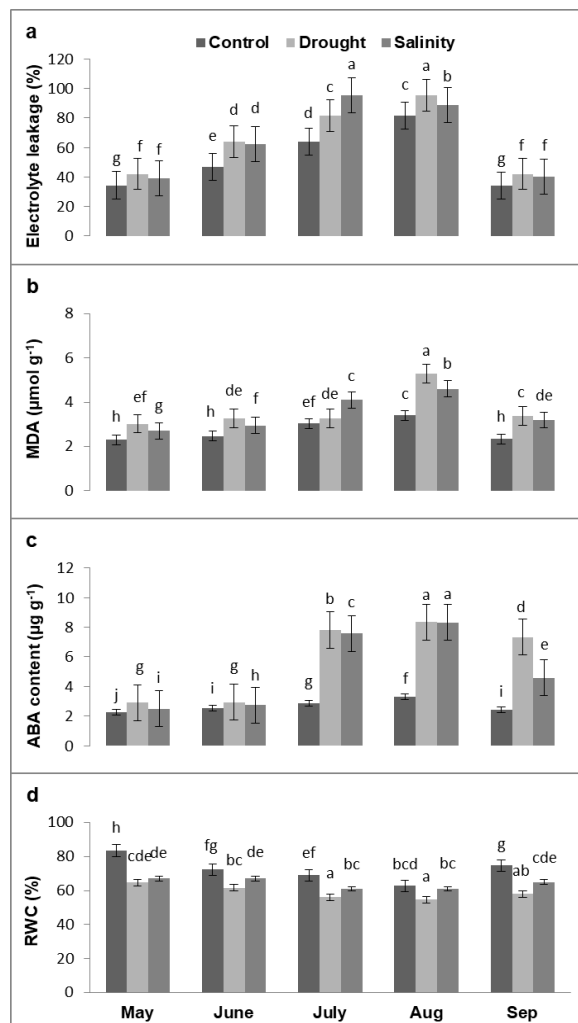


Fig. 3. Changes in Electrolyte leakage (a), MDA (b), ABA content (c), and RWC (d) of Date palm offshoots leaves under different abiotic stresses. Results are means \pm SD (n = 5). The same letters are not significantly different $p \leq 0.05$ after Tukey's correction for multiple comparisons.

caused by salinity, drought and high temperatures in the summer season, which is a combination of these factors of abiotic stresses. Hence, we hypothesize that date palms with high-stress tolerance have a high capacity to change photosynthesis pigments because of abiotic stress, such as high temperature. By targeting chlorophyll, anthocyanin and carotenoid to identify abiotic stress responses in Date palm offshoots, we focused on different factors of abiotic stress such as salinity and drought under the various temperature conditions of May, June, July, August, and September.

Reduction in pigment content may be a direct result of the prohibitive consequence of the aggregated particles of various salts on the biogenesis of the different pigment portions (SHAH et al., 2017). Chlorophyll pigments depending on the factor of abiotic treatments, and the high temperature in the field reduced. Concerning drought stress treatment and water scarcity, they cause pigment degradation and membrane damage (Figs 1–3a). Salt stress causes decreased leaf water potential, and the RWC was thereby

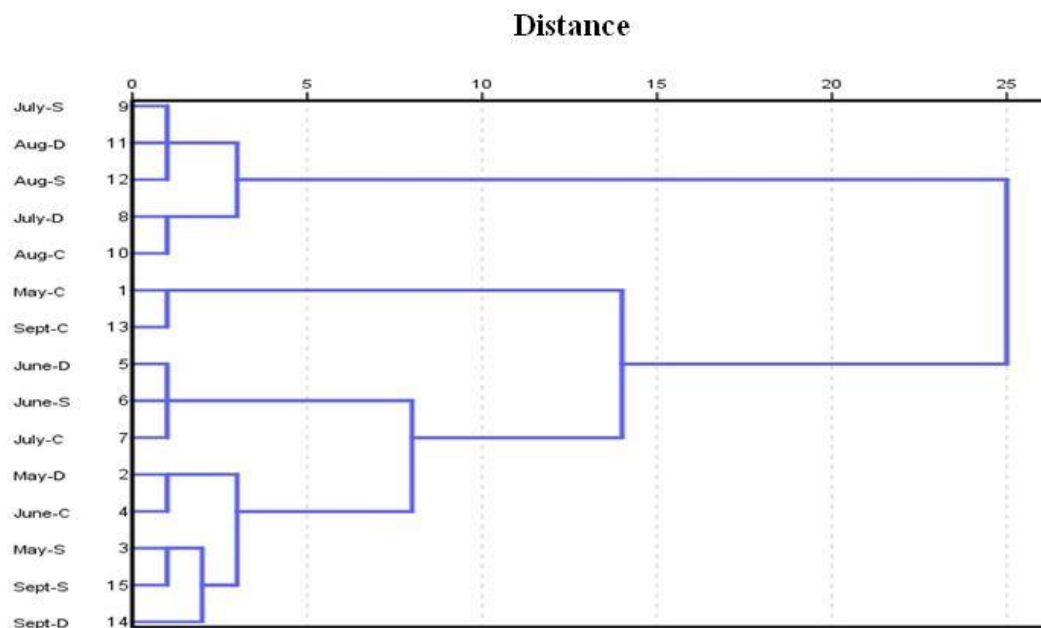


Fig. 4. Dendrogram of hierarchical clustering to leaves of Date palm offshoot under different abiotic stress by using photosynthesis pigments as an indicator.

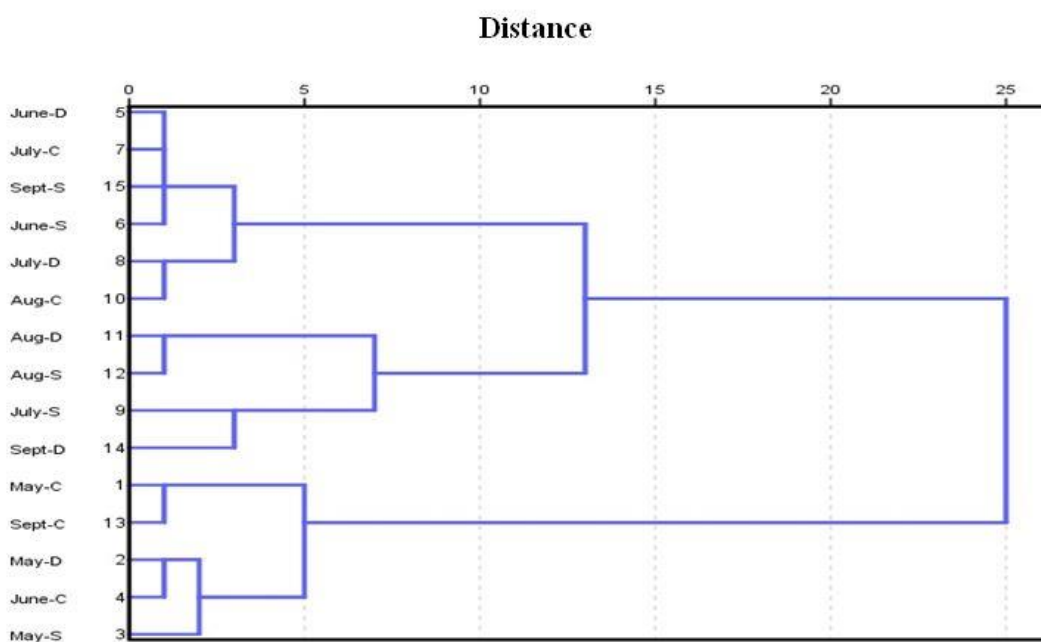


Fig. 5. Dendrogram of hierarchical clustering to leaves of date palm offshoot under different abiotic stress by use (pigments contents, hydrogen peroxide, malondialdehyde, electrolyte leakage, proline, ABA, and leaf RWC) as an indicator.

affected by overall photosynthetic capacity (HNILČKOVÁ et al., 2017). However, anthocyanin or carotenoid accumulation may facilitate the protection of chemical process mechanisms by screening them from the hazardous impacts of pressure producing superoxide radicals, while not constraining photosynthesis. Anthocyanins are thought to limit oxidative harm and act as antioxidants by neutralizing ROS directly (FONDOM et al., 2009). However, the high air temperature reveals significant impacts on plant metabolism — that affect the generation and aggrega-

tion of auxiliary plant mixes, for example, a carotenoid which is layer bound exacerbates fundamental capacities in photosynthesis (XU et al., 2015). Heat stress arises from high temperatures that lead to damage plant tissue; temperatures in the degree of 35–45 °C in the tropics affect the different plant species (NIEVOLA et al., 2017).

Chemical responses to abiotic stress may lead to conditions of a superior level of assurance against oxidative harm in the control treatment by quick evacuation of H₂O₂ or by other scavenging frameworks. H₂O₂ was lower under

drought than salt in the case of the high temperature of the field, which implies that the generation of H₂O₂ decreased by the antioxidative actions of the plant. Under salinity, H₂O₂ content increased at higher temperatures (Fig. 2c). The electrolyte leakage through plasmalemma is associated with the activity of photosynthetic and mitochondrial reactions (SHALABY, 2018). The increase in H₂O₂ content in the present analysis initiated malondialdehyde (MDA) (Fig. 3b). The harmful impact of ROS is the lipid peroxidation anticipated by the formation of MDA (AYALA et al., 2014). MDA significantly increased upon water deprivation, as the field temperature increased (Fig. 2b). Generally, under typical conditions, the generation and expulsion of ROS are in a balanced state (SCHOOTS et al., 2018) "ISSN" : "15323102", "abstract" : "The most important function of the placenta is the exchange of nutrients and oxygen between a mother and her fetus. To establish a healthy functioning placenta, placentation needs to occur with adequate remodelling of spiral arteries by extravillous trophoblasts. When this process is impaired, the resulting suboptimal and inadequate placenta function results in the manifestation of pregnancy complications. Impaired placenta function can cause preeclampsia and leads to fetal growth restriction due to hypoxia. Presence of hypoxia leads to oxidative stress due to an imbalance between reactive oxygen species and antioxidants, thereby causing damage to proteins, lipids and DNA. In the placenta, signs of morphological adaptation in response to hypoxia can be found. Different placental lesions like maternal or fetal vascular malperfusion or chronic villitis lead to a decreased exchange of oxygen between the mother and the fetus. Clinically, several biomarkers indicative for oxidative stress, e.g. malondialdehyde and reduced levels of free thiols are found. This review aims to give an overview of the causes and (potential).

Proline accumulation in response to stress supplies vitality for advancement and survival, and in this manner, allows the plant to endure stress (JASIM et al., 2016). The ABA has an impact on some pathways unmistakable from the acceptance of HSPs (HSP qualities deciphered in all eukaryotes through a similar procedure). Specific saved interpretation factors, called HSFs, perceive and tie to rationed *cis* components (the HSE) in the advertiser of HSP and different qualities (NOVER et al., 2001). ABA content in September was higher than May under drought stress (Fig. 2c). ABA-prompted stomata closure mostly dependent on NADPH oxidase action (WATKINS et al., 2017). Our outcomes revealed that RWC decreased more with drought stress than salt stress conditions (Fig. 3d), indicating that plants were under stress at the time of sampling. Among every one of the treatments examined, the imposition of salt and drought treatments resulted in a critical decline in RWC of those offshoots which developed under field conditions.

Cluster analysis showed the convergence of the effect of salt, drought stress, and control in the impact on the date palms under low temperature while distinguishing the effect of low and high temperature in the photosynthetic pigments and biochemical parameters. Similarities in the results were obtained with a study of HAIDER et al. (2015)

on the effect of environmental differences on Date palm varieties. The difference in temperature affecting the content of photosynthesis pigments in the leaves indicates that high temperature is the determining factor for plant growth and the most influential in plant development.

Conclusions

Photosynthesis pigments are a fundamental indicator of the effect of various abiotic stress factors on plants. Periods of stress that occur commonly during the two hottest months of the summer season indicated that high temperature is the primary factor that determines the growth of plants. Recovery of photosynthesis following low-temperature alleviation, for the most part, determines the plant flexibility to water deficiencies and salinity. Consequently, our study represents an initial contribution to understanding the physiological events occurring during the critical period of date palm offshoot growth throughout the summer season.

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