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Impact of Drought Stress on Six Summer Squash (*Cucurbita pepo* L.) Genotypes in Sandy Reclaimed Land.

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Abstract

Drought stress is a major global challenge that threatens sustainable agriculture and food security. Drought stress is expected to become more severe with the rise of climate change and global warming. Summer squash consists of more than 90 % water; therefore, it is sensitive to drought stress. This study investigated the impact of three irrigation treatments, T1 (100 % RAW-full irrigation) control, T2 (75 % RAW- moderate stress), and T3 (50 % RAW-severe stress), on six summer squash genotypes grown under open field conditions during the seasons of 2018 and 2019. Our results showed that drought stress substantially affected all summer squash genotypes. Drought stress decreased the vegetative growth traits such as plant height and plant fresh weight. Also, it reduced the leaf relative water content, leaf chlorophyll content, and water use efficiency, but it slightly increased the leaf osmotic adjustment. The number of days to 50 % flowering slightly decreased with increasing drought stress. The total yield, number of fruits per plant and weight of fruits per plant decreased because of the negative effects of the drought on the traits studied above. These findings indicate that drought stress negatively affected all summer squash genotypes; however, they significantly differed in their response to the drought. All genotypes surpassed the check cultivar Iskandrany. Rivera, Azyad and Fadwa outperformed all summer squash genotypes in most studied traits, especially the yield and yield component traits under normal and stress conditions.

Keywords: full irrigation, moderate drought stress, severe drought stress, summer squash hybrids, desert soils.

INTRODUCTION

Drought stress is one of the most significant abiotic stresses affecting crop productivity and food security worldwide. In the context of climate change, the frequency and severity of drought are increasing in most regions of the world, posing a considerable threat to agriculture (Parkash *et al*, 2021 and Solankey *et al*, 2021). Summer squash (*Cucurbita pepo* L.), an economically valuable and widely cultivated vegetable, is highly sensitive to water deficit conditions due to its high-water demand, especially during the summer growing season (Refai *et al*, 2019 and Okasha *et al*, 2020). Drought stress can negatively impact summer squash growth, physiological processes, fruit quality, and overall yield, making the development of drought-tolerant genotypes a critical objective in breeding programs. Drought stress causes disturbances in all physiological, morphological, and biochemical processes in susceptible plants, which in turn limits their productivity (Du *et al*, 2020). These disturbances result in decreasing leaf chlorophyll content, relative water content, photosynthesis, and transpiration rate (Ors *et al*, 2016 and Lamaoui *et al*, 2018). Moreover, drought stress causes excessive production of reactive oxygen species ROS which causes oxidative stress such as lipid peroxidation in cell membranes, protein degradation and leads to cell death (Zia *et al*, 2021). On the other hand, the plants increase the accumulation of proline, total soluble carbohydrates, soluble proteins, and free amino acids (Laary *et al*, 2018 and Du *et al*, 2020). This accumulation is known as osmotic adjustment, which is considered a physiological mechanism of plants to cope with drought stress by maintaining turgor pressure and cell volume. (Nahar and Ullah, 2018). Also, the plants use another defensive system to cope with drought stress by increasing antioxidant enzymes, catalase CAT, peroxidase POD, and superoxide dismutase SOD, to scavenge reactive oxygen species ROS and reduce their damaging injuries (Lamaoui *et al*, 2018 and Batool *et al*, 2020). Where this damage depends on the balance between the production of ROS and antioxidant enzymes, therefore, tolerant plants produce

antioxidant enzymes more than ROS (Anjum *et al*, 2011). All physiological damages during drought stress appear in morphological symptoms in the susceptible plants in all growth stages. Where, during the vegetative stage, drought stress decreased root fresh and dry weight, plant height and plant fresh weight in many crops, such as in melon (Kava *et al*, 2013), in tomato (Shabbir *et al*, 2021), and in summer squash (Refai *et al*, 2019 and Okasha *et al*, 2020). Meanwhile, during the flowering stage, water deficit decreased the No. of days to 50 % flowering in rice, cowpea, and canola (Singh *et al*, 2018 and Kandil *et al*, 2017). Response of genotypes to drought stress involves physiological, biochemical, and molecular adaptations. These adaptations, such as changes in osmotic adjustment, stomatal conductance, accumulation of compatible solutes, and activation of antioxidant defense systems (Batool *et al*, 2020 and Du *et al*, 2020). The degree of these responses varies among genotypes, hence highlighting the importance of genetic diversity in breeding programs for improving drought tolerance. Developing drought-tolerant genotypes of summer squash can ensure stable yields under water-deficient environments, therefore, supporting sustainable agriculture in the face of climate change conditions. The objectives of this research are twofold: (1) to determine the best genotypes of summer squash resilient to drought stress. (2) to determine the best irrigation level suitable for cultivation in reclaimed land under Sohag conditions.

MATERIALS AND METHODS

1. Plant material and experimental design

This experiment was carried out at the Experimental Farm of the Faculty of Agriculture, Sohag University, New Campus, New Sohag City in 20th of February in 2018 and in 14th of February in 2019 in a reclaimed sandy soil. The soil texture of the experimental site was sandy loam. Physical properties of the soil of the experiment and chemical analysis of irrigation water are shown in Tables (1 and 2). The samples were analyzed in the soil science and water department labs according to (Lovedoy, 1974).

Table 1. Physical properties of the soil of the experiment.

Soil depth (cm)	Field capacity%	Wilting point%	Bulk density g/cm ³
0-15	13.4	8.9	1.23
15-30	7.04	3.5	1.35
30-50	4.3	2.15	1.48

Table 2. Chemical analysis of irrigation water.

pH	EC mg/l ⁻¹	Na mg/l ⁻¹	K mg/l ⁻¹	Ca mg/l ⁻¹	Mg mg/l ⁻¹	HCO ₃ mg/l ⁻¹	Cl mg/l ⁻¹	SO ₄ mg/l ⁻¹
7.64	971.2	82.90	134.92	80.42	40.62	191.06	80.26	354.72

Six genotypes of summer squash (*Cucurbita pepo* L.) were purchased from commercial Egyptian companies and used in this experiment. Three irrigation treatments were applied in this experiment and designated as: Treatment one, T1 (control): 100 % of readily available water (100 % RAW), Treatment two, T2: 75 % of readily available water (75 % RAW), and Treatment three, T3: 50 % of readily available water (50 % RAW). The water was delivered to the plants by the drip irrigation system. Each plot consisted of 4 drip tapes, one meter apart and each tape consisted of eight drippers 30 cm apart. The plot area was (9.6 m²). The dripper flow rate was approximately 4 L per h when measured in the field and according to the manufacturer. Thirty-two seeds were sown in the plot (4 drip tapes X 8 drippers). The quantity of water used in the irrigation and required to reach the 100 % readily available water RAW was six liters for one plant. The six liters were delivered to plants in 1.5 hours. The readily available water was calculated according to (Allen *et al*, 1998):

$$Aw = \frac{FC - PWP}{100} \times pb \times D$$

Where, AW is available water, F.C. is of field capacity, PWP is the permanent wilting point, pb is the soil bulk density, and D is the depth of roots for the plant.

Readily available water RAW was calculated as follows: **RAW = AW X MAD**

Where, MAD is maximum allowable depletion, which is 0.50 in squash (Allen *et al*, 1998). Plants were irrigated every two days in the

first 25 days of plant growth for all treatments. The T1, T2 and T3 irrigation treatments were applied after 25 days from the planting date. The soil moisture content was measured daily to determine the irrigation time in each treatment by using the Delta SM150 portable soil moisture kit Figure 2 (Delta-T Devices Ltd, Burwell, Cambridge, U.K). The kit includes the SM150 soil moisture sensor and the HH 150 meter (readout unit). The soil moisture content was measured by removing the upper 15 cm from the surface of the soil and the sensor was inserted into the soil 15 cm away from the drippers. Five to seven readings were taken from different areas of each irrigation treatment to determine the soil moisture content daily. Readings of the soil moisture kit were shown as volumetric values of the soil moisture content. Therefore, irrigation was done in each treatment when soil moisture content reached 16.5 % vol in T1 (100 % of RAW) (control), 12.4 % vol in T2 (75 % of RAW), and 8.4 % vol in T3 (50 % of RAW). The Delta SM150 portable soil moisture kit was calibrated according to the manufacturer's instructions.



Figure 1. Delta MS 150 moisture meter kit. **A.** The kit used to measure the soil moisture. **B.** The SM 150 moisture sensor. **C.** The HH150 moisture meter (readout unit).

2. Measurements

The following three traits were determined 41-51 days from irrigation treatments and after irrigation of plants in all treatments. Three plants from each plot were pulled out and their roots were removed:

a. Plant Height (cm):

Plant height was determined by measuring the length from the base to the tip of the plant using a ruler.

b. Plant Fresh Weight PFW (g):

Plant fresh weight was immediately weighed after cutting their roots. The following measurements were taken from the average of five labeled plants per plot.

c. Relative Water Content RWC (%):

Relative water content was determined 27-29 days from irrigation treatments at the end of each treatment and right before the irrigation. Five-second fully expanded leaves were weighed to determine fresh weight FW and then immersed in deionized water overnight. The next day, the leaves were taken away from deionized water on tissue paper and carefully removed excess water and weighed to obtain turgid weight TW. The turgid leaves were oven-dried at 70 °C for 24 h to obtain dry weight DW. Relative water content RWC was calculated according to the following formula:

$$RWC \% = \frac{FW - DW}{TW - DW} \times 100$$

d. Osmotic Adjustment OA (MPa):

Osmotic adjustment was determined 54-57 days from irrigation treatments at the end of each treatment and right before the irrigation. This trait was measured according to (Wilson *et al*, 1979 and Ludlow *et al*, 1983). Calculated from three fully expanded leaves. Three discs of known area were cut out from the leaf and well ground well in 1.5 ml of deionized water using a mortar and pestle. Then the suspension was transferred to Eppendorf tubes and centrifuged at 13000 rpm for three minutes. The supernatant was transferred to a new tube. Samples were stored at -3 °C until the next day for measuring. A sample of 50 µ was taken and measured by (osmomat 300).

e. Chlorophyll Content:

Chlorophyll Content was measured 41-49 days from irrigation treatments at the end of each treatment and right before the irrigation. Readings were taken by the chlorophyll meter SPAD-502 (KONICA MINOLTA, INC., JAPAN) from three different spots of the second fully expanded leaf from three plants per plot.

f. Water Use Efficiency WUE (kg/m³):

Water use efficiency was calculated according to (El-Gindy *et al*, 2009) by the following formula:

$$\text{Water Use Efficiency (kg/m}^3\text{)} = \frac{\text{Average yield (kg/fed)}}{\text{Amount of applied water (m}^3\text{/fed)}}$$

g. No. of Days to 50 % Flowering:

No. of days to 50 % flowering was counted when 50 % of the plants flowered.

h. Total Yield per fed. (ton):

Total yield was determined by summing the weight of fruits from the whole plot picked throughout the entire season and the total yield per feddan was calculated as follows:

$$\text{Total Yield (ton/fed)} = \frac{\text{Weight of fruits per plot (ton)} \times \text{Feddan area (m}^2\text{)}}{\text{plot area (m}^2\text{)}}$$

The following measurements were taken from the average of five labeled plants per plot:

i. No. of Fruits per Plant:

No. of fruits per plant was calculated as the average No. of fruits picked throughout the entire season.

j. Weight of Fruits per Plant (g):

The weight of fruits per plant was calculated by adding up the weight of fruits picked throughout the entire season from the five labeled plants. Fruits were picked when the fruit length reached 12-16 cm in all irrigation treatments.

3. Statistical analysis:

A split-plot layout was used with three replicates. The main plot was assigned to the irrigation treatments and the subplot was assigned to the genotypes. Data were statistically analyzed

using the MSTAT package program. The mean for all the treatments was calculated and analyses of variance of all the characters were performed by the F-variance test. Data obtained during the two seasons of the study were statistically analyzed and treatment means were compared using Duncan's multiple range tests (Gomez and Gomez, 1984). Regression was calculated and figures were created using Excel software (Microsoft Office software package 2016). Simple regression analysis was done between the traits and the irrigation treatments.

RESULT



Figure 2. Visual representation of plants of varying sizes under different irrigation treatments of summer squash genotypes in the second season. The photos were taken during the measurement of plant height and plant fresh weight (41-51) days after irrigation treatments application. The ruler height used is 30 cm.

Figure 3 exhibits the impact of three irrigation treatments on plant height in six genotypes of summer squash grown in 2018 and 2019. Under full irrigation conditions (T1, 100 % RAW) (control), New Iskandrany and Azyad gave the highest plant height with 8.37 % and 3.34 % increase over the check cultivar Iskandrany in 2018. While, in 2019, New Iskandrany showed the highest plant height with 8.29 % increase over Iskandrany. Meanwhile, under moderate drought stress (T2, 75 % RAW), Azyad showed the highest plant height with 5.80 % and 5.73 % increase over the check cultivar in

1- The impact of three irrigation treatments on plant height and plant fresh weight of summer squash genotypes grown under open field conditions in the seasons of 2018 and 2019

Our data showed that drought stress decreased plant height and plant fresh weight PFW of all summer squash genotypes in both seasons (Figures 2, 3 and 4). There were significant differences among genotypes, irrigation treatments, and their interaction with genotypes in the studied traits in both seasons.

both seasons. Under severe drought stress (50 % RAW), Mabrouka and Azyad gave the highest plant height with 6.13 % and 5.81 % increase over Iskandrany in 2018 and 5.97 % and 5.77 %. The least reduction in plant height due to water stress occurred in Azyad under moderate drought stress (T2, 75 % RAW), with 6.88 % and 6.91 % decrease compared to the same hybrid under the conditions of full irrigation in both seasons. Meanwhile, under severe drought stress (T3, 50 % RAW), the least reduction occurred in Mabrouka and Azyad, with 20.86 % and 20.25 % decrease compared to the same hybrids under the

conditions of full irrigation in 2018 and 20.88 % and 20.22 % in 2019.

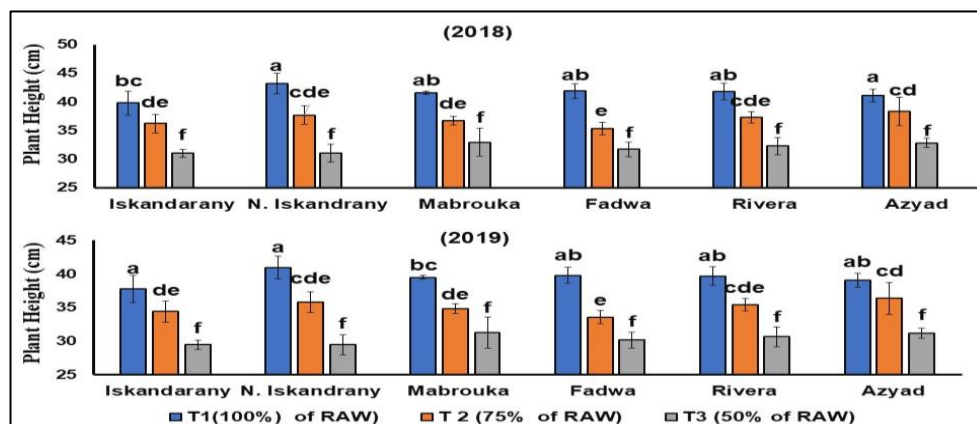


Figure 3. Impact of three irrigation treatments on plant height of summer squash genotypes during the seasons of 2018 and 2019. Means followed by the same letter (s) are not significantly different at the 5 % level. Error bars are the \pm SD of three biological replicates.

The data illustrated in Figure 4 presents the impact of three irrigation treatments on plant fresh weight PFW in six genotypes of summer squash grown in both 2018 and 2019. Under full irrigation conditions (T1, 100 % RAW), Rivera consistently recorded the highest PFW in both 2018 and 2019, with a 13.65 % and 13.64 % increase over the check cultivar Iskandarany in each season. Under moderate drought stress (T2, 75 % RAW), Rivera also performed best, with increases of 25.18 % in 2018 and 25.16 % in 2019. Under severe drought stress (T3, 50 %

RAW), Mabrouka and Rivera showed the highest PFW, outperforming Iskandarany by 64.13 and 55.44 % in 2018 and 64.10 and 55.43 % in 2019, respectively. In terms of reduction in PFW due to water stress compared to the control, the least reduction at T2 (75 % of RAW) occurred in Azyad in 2018, with only a 17.16 % decrease compared to the same hybrid at the control in both seasons. Under T3 (50 % RAW), Mabrouka had the least reduction, with a 36.19 % decrease in 2018 and 2019.

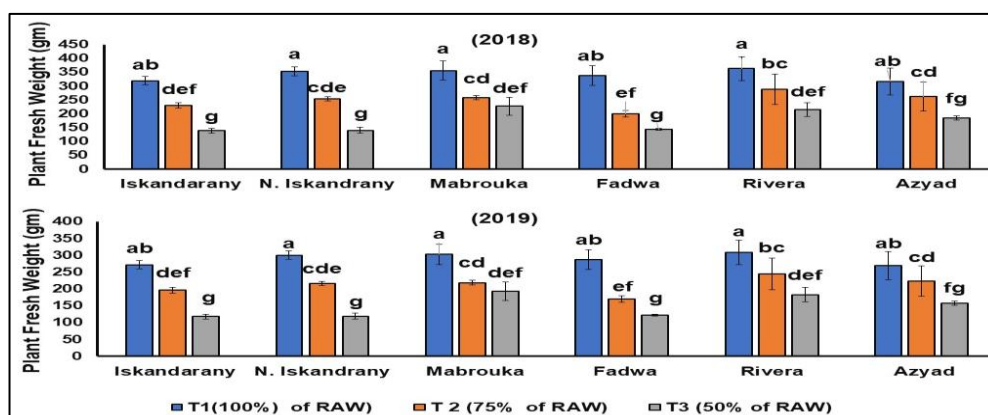


Figure 4. Impact of three irrigation treatments on plant fresh weight of summer squash genotypes during the seasons of 2018 and 2019. Means followed by the same letter (s) are not significantly different at the 5 % level. Error bars are the \pm SD of three biological replicates.

2- The impact of three irrigation treatments on relative water content, chlorophyll content, water use efficiency, and osmotic adjustment of summer squash genotypes grown under open field conditions in the seasons of 2018 and 2019.

Data presented in Figures (5, 6, 7 and 8) illustrated that drought stress decreased relative water content, chlorophyll content and water use efficiency, but slightly increased osmotic adjustment of all summer squash genotypes. Significant differences were observed among

irrigation treatments, genotypes, and their interaction in the chlorophyll content and water use efficiency in both seasons. In the relative water content, there were significant differences among genotypes, their interaction, and between T1 (control) and T3, but there were no significant differences between T1 and T2 in both seasons. For osmotic adjustment OA, there were no significant differences among the genotypes and their interaction with irrigation treatments; however, there were significant differences among the irrigation treatments.

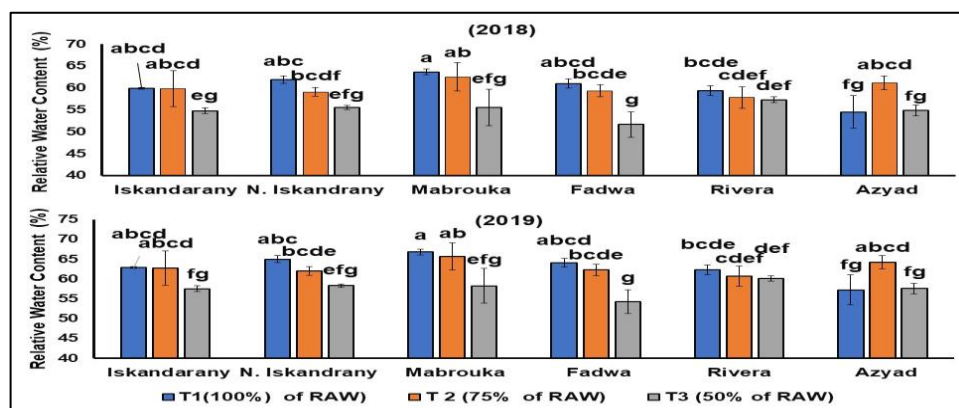


Figure 5. Impact of three irrigation treatments on the relative water content of summer squash genotypes during the seasons of 2018 and 2019. Means followed by the same letter (s) are not significantly different at the 5 % level. Error bars are the \pm SD of three biological replicates.

Figure 5 illustrates the impact of three irrigation treatments on relative water content % RWC in six genotypes of summer squash grown in 2018 and 2019. Under full irrigation conditions (T1, 100 % RAW), Mabrouka exhibited the highest relative water content (RWC) in both 2018 and 2019, with increases of 6.18 % and 6.24 % over the check cultivar Iskandarany, respectively. Under moderate drought stress (T2, 75 % RAW), Mabrouka maintained the highest RWC with 4.62 % and 4.56 % increase over Iskandarany in both seasons. Under severe drought stress (T3, 50 % RAW), Rivera showed the highest RWC in both seasons, surpassing Iskandarany by 4.54 % in 2018 and 4.57 % in 2019. Regarding the reduction in RWC due to water stress compared to the control, Azyad showed an increase at T2 (75 % RAW), indicating a relative improvement in performance under moderate drought. Excluding Azyad, Iskandarany had the least reduction at T2, with

only 0.22 % and 0.16 % decreases in 2018 and 2019, respectively. At severe drought stress (T3, 50 % RAW), Azyad recorded the least reduction in RWC, with only 0.61 % and 0.58 % decreases in 2018 and 2019. Figure 6 shows the impact of three irrigation treatments on leaf chlorophyll content in six genotypes of summer squash grown in 2018 and 2019. Under full irrigation conditions (T1, 100 % RAW), the hybrid Fadwa exhibited the highest chlorophyll content in both 2018 and 2019, showing increases of 15.6 % over the check cultivar Iskandarany in both seasons. Under moderate drought stress (T2, 75 % of RAW), New Iskandarany recorded the highest values of chlorophyll content with increases of 14.83 % in 2018 and 14.88 % in 2019 compared to Iskandarany. Under severe drought stress (T3, 50 % RAW), Fadwa again showed the highest chlorophyll content, surpassing Iskandarany by 14.60 % and 14.47 % in 2018 and 2019, respectively. Regarding the least reduction in

chlorophyll content due to drought stress, Mabrouka showed the least decrease at T2, with only 1.33 % and 1.36 % reduction in 2018 and

2019, respectively. At T3, Azyad exhibited the lowest reduction, with 4.70 % and 4.40 % decreases in 2018 and 2019, respectively.

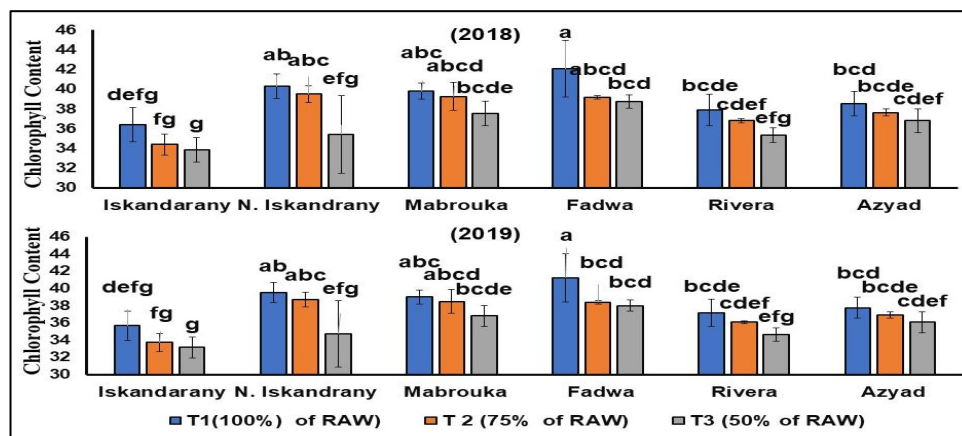


Figure 6. Impact of three irrigation treatments on chlorophyll content of summer squash genotypes during the seasons of 2018 and 2019. Means followed by the same letter (s) are not significantly different at the 5 % level. Error bars are the \pm SD of three biological replicates.

Figure 7 shows the impact of three irrigation treatments on water use efficiency WUE in six genotypes of summer squash grown in 2018 and 2019. Under full irrigation conditions (T1, 100 % RAW), the hybrids Azyad and Rivera recorded the highest water use efficiency (WUE) in both seasons, with increases of 63.16% and 59.1 in 2018 and 62.26 % and 59.12 in 2019 over the check cultivar Iskandarany. Under moderate drought stress (T2, 75 % RAW), Rivera showed the highest WUE values, exceeding Iskandarany

by 59.87 % in 2018 and 60.27 % in 2019. Under severe drought stress (T3, 50 % RAW), Rivera again outperformed all hybrids, with remarkable increases of 168.52 % and 165.08 % over Iskandarany in 2018 and 2019, respectively. The least reductions in WUE due to water stress occurred in Rivera at T2 with only 7.72 % and 7.51 % reductions in 2018 and 2019, respectively. At T3, Rivera also exhibited the lowest reductions, with 46.69 % and 33.99 % decreases in 2018 and 2019, respectively.

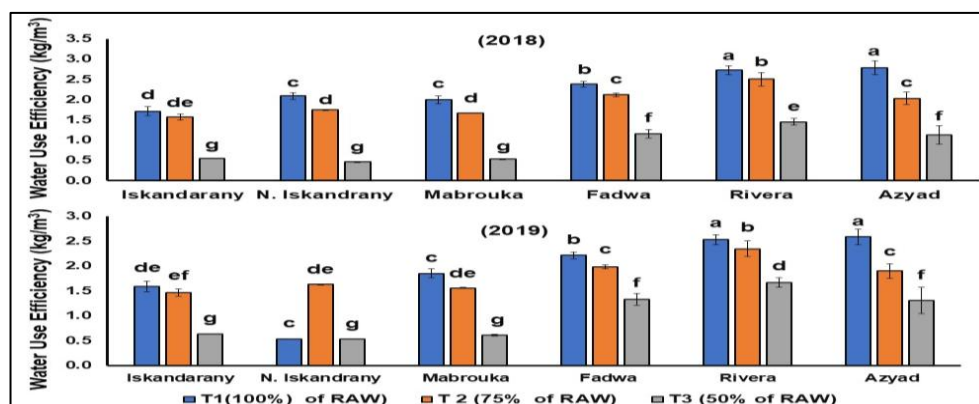


Figure 7. Impact of three irrigation treatments on water use efficiency of summer squash genotypes during the seasons of 2018 and 2019. Means followed by the same letter (s) are not significantly different at the 5 % level. Error bars are the \pm SD of three biological replicates.

Figure 8 exhibits the impact of three irrigation treatments of osmotic adjustment OA in

six genotypes of summer squash grown in the season of 2019. Under full irrigation conditions

(T1, 100 % RAW), Mabrouka recorded the highest osmotic adjustment values, showing a 23.81% increase over the check cultivar Iskandrany. Under moderate and severe drought stress (T2, 75 % RAW and T3, 50 % RAW), Mabrouka also exhibited the highest values with a 12.5 % and 6.67 % increase over Iskandrany. The highest reduction in osmotic adjustment due

to water stress occurred in Azyad under moderate drought stress (T2, 75 % RAW), with 23.81 % increase compared to the same hybrid in the control. Under severe drought stress (T3, 50 % RAW), the least reduction due to drought stress occurred in Iskandrany and Azyad with a decrease of 33.33 % compared to the same hybrids at the control.

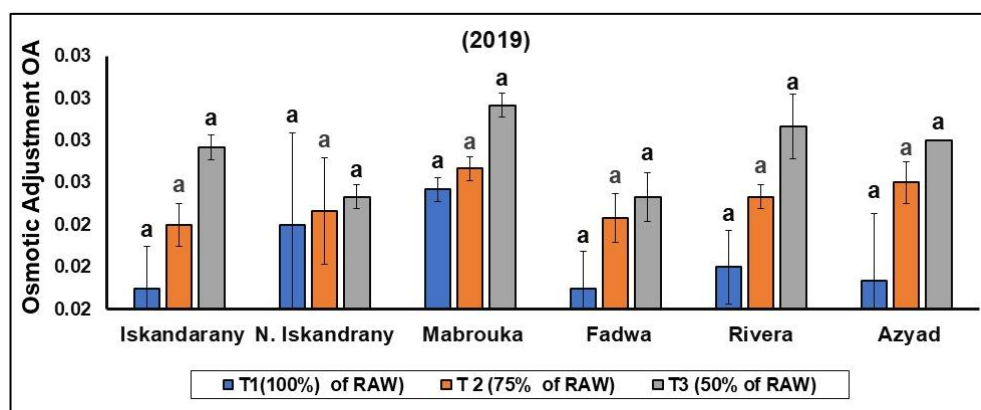


Figure 8. Impact of three irrigation treatments on osmotic adjustment of summer squash genotypes during the season of 2019. Means followed by the same letter (s) are not significantly different at the 5 % level. Error bars are the \pm SD of three biological replicates.

3- The impact of three irrigation treatments on No. of days to 50 % Flowering of summer squash genotypes grown under open field conditions in the seasons of 2018 and 2019.

Figure 9 shows the impact of three irrigation treatments on the number of days to 50 % in six genotypes of summer squash grown in 2018 and 2019. Under full irrigation (T1, 100 % RAW), the earliest flowering genotypes were New Iskandrany, Mabrouka, and Azyad with 3.37 % and 3.34 reduction compared to the check cultivar Iskandrany in both seasons. Under moderate drought stress (T2, 75 % RAW), the earliest flowering genotypes in 2018 were New

Iskandrany and Azyad with a 5.50 % and 4.82 % decrease compared to Iskandrany in 2018. In 2019, New Iskandrany showed the earliest flowering genotype with 5.59 % decrease in 2019. Under severe drought stress (T3, 50 % RAW), Mabrouka had a 6.29 % and 6.24 % decrease compared to Iskandrany in both seasons. The highest reductions in the number of days to 50 % flowering due to water stress occurred under moderate and severe drought stress (T2, 75 % RAW and T3, 50 % RAW) occurred also in New Iskandrany, with a 4.20 % decrease compared to the same hybrid at the control in 2018 and 4.19 % in 2019.

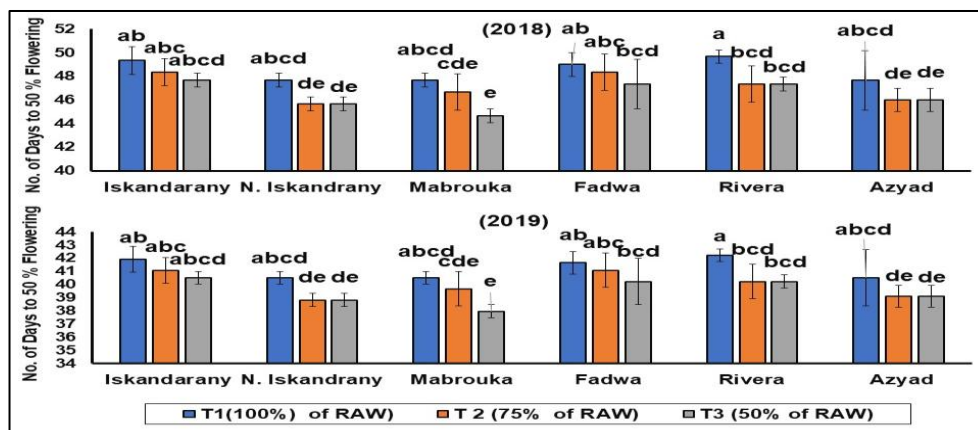


Figure 9. Impact of three irrigation treatments on No. of days to 50 % flowering of summer squash genotypes during the season of 2019. Means followed by the same letter (s) are not significantly different at 5 % level. Error bars are the \pm SD of three biological replicates.

4- The impact of three irrigation treatments on total yield, number of fruits per plant, and weight of fruits/plant of summer squash genotypes grown under open field conditions in the seasons of 2018 and 2019.

Data presented in Figures (10, 11, and 12) show that drought stress decreased the total

yield, No. of fruits per plant, and the weight of fruits per plant of all summer squash genotypes in both seasons. Significant differences were observed among irrigation treatments, genotypes, and their interaction in the studied traits in both seasons.

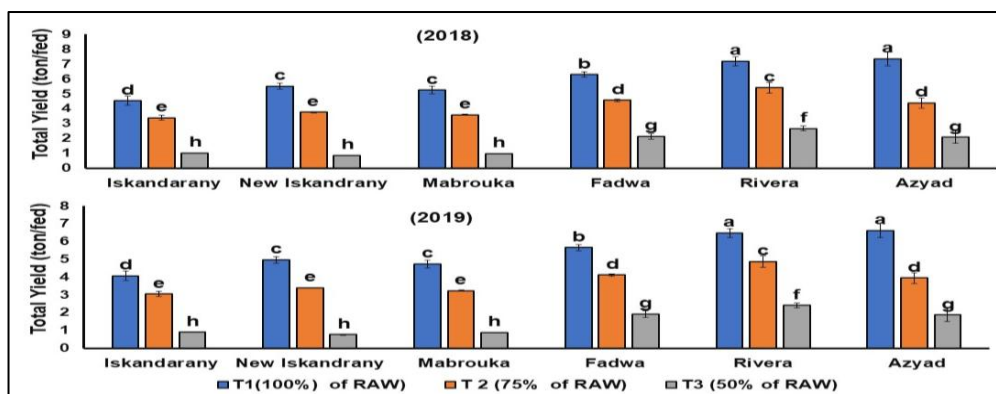


Figure 10. Impact of three irrigation treatments on the total yield of summer squash genotypes during the season of 2019. Means followed by the same letter (s) are not significantly different at the 5 % level. Error bars are the \pm SD of three biological replicates.

Figure 10 illustrates the impact of three irrigation treatments on the total yield in six summer squash genotypes grown in 2018 and 2019. Under full irrigation conditions (T1, 100 % RAW), the hybrids Azyad and Rivera recorded the highest total yield in both seasons, with 62.25 % and 58.71 increase over the check cultivar Iskandarany in 2018 and 62.56 % and 58.97 % in 2019. Under moderate drought stress (T2, 75% RAW), Rivera showed superior performance,

representing 60.06 % and 59.67 % increases over Iskandarany in both seasons. Under severe drought stress (T3, 50 % RAW), Rivera again outperformed all hybrids, with 164.36 % and 166.67 % increase over the check cultivar Iskandarany. Regarding yield stability under water deficit, Rivera exhibited the least reduction compared to its performance in the control. At T2, its yield decreased by only 24.76 % in 2018 and 24.73 % in 2019, while at T3, the reductions

were 62.87 % and 62.91 %, respectively. These findings indicate that Rivera is the most drought-tolerant and stable hybrid in terms of total yield under both moderate and severe water stress. Figure 11 shows the impact of three irrigation treatments on the No. of fruits per plant in six genotypes of summer squash grown in 2018 and 2019. Under full irrigation conditions (T1, 100 % RAW), the hybrids Azyad and Rivera recorded the highest No. of fruits per plant in both seasons, with 36.96 % and 34.96 % increases over Iskandrany in 2018 and 36.51 % and 34.60 % in 2019. Under moderate drought stress (T2, 75 % RAW), Rivera showed superior performance, representing 45.89 % and 45.63% increases over

Iskandrany in both seasons. Under severe drought stress (T3, 50 % RAW), Rivera again outperformed all hybrids, with 93.75 % and 96.00 % increase over the check cultivar Iskandrany. Regarding fruit number stability under water deficit, Rivera exhibited the least reduction compared to its performance at the control. At T2, No. of fruits per plant decreased by only 9.55 % in 2018 and 9.67 % in 2019, while at T3 the reductions were 53.93 % and 53.77 %, respectively. These results indicate that Rivera is the most drought-tolerant hybrid for maintaining No. of fruits per plant under both moderate and severe water stress.

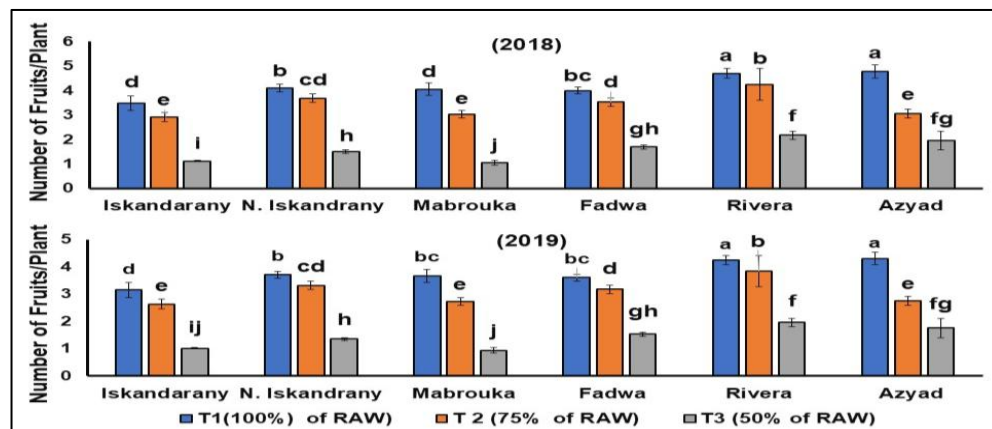


Figure 11. Impact of three irrigation treatments on No. of fruits per plant of summer squash genotypes during the season of 2019. Means followed by the same letter (s) are not significantly different at the 5 % level. Error bars are the \pm SD of three biological replicates.

The data presented in Figure 12 shows the impact of three irrigation treatments on the weight of fruits per plant in six summer squash genotypes grown in 2018 and 2019. Under full irrigation conditions (T1, 100 % RAW), the hybrids Azyad and Rivera recorded the highest weight of fruits per plant in both 2018 and 2019, with 62.46 % and 58.99 % increases over the check cultivar Iskandrany in both seasons. Under moderate drought stress (T2, 75 % RAW), Rivera showed superior performance, representing increases of 59.81 % and 59.83 % over Iskandrany in both seasons. Under severe drought

stress (T3, 50 % RAW), Rivera again outperformed all hybrids, with a 165.30 % increase over the check cultivar in both seasons. Regarding stability under water deficit, Rivera exhibited the least reduction in weight of fruits per plant compared to its performance at the control. At T2, it decreased by only 24.76 % in 2018 and 24.78 % in 2019, while at T3, the reduction was 62.93 % in 2018 and 62.93 % in 2019. These findings indicate that Rivera is the most drought-tolerant hybrid for maintaining fruit biomass under both moderate and severe water stress.

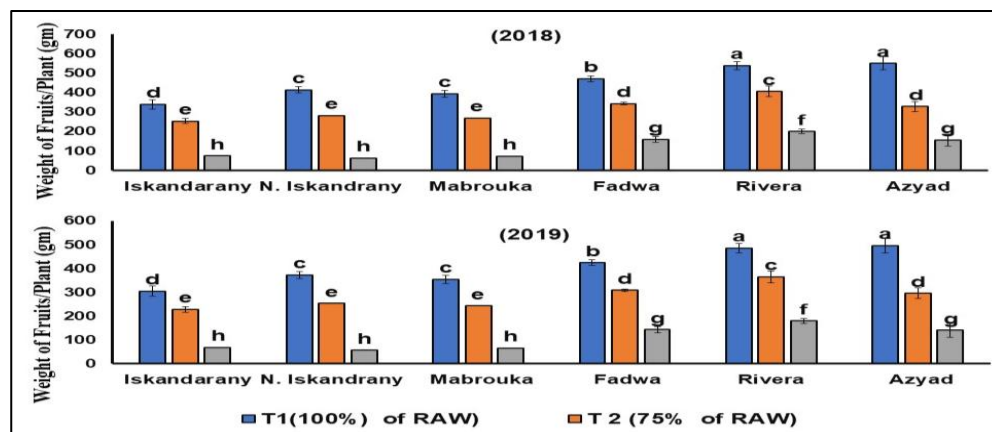


Figure 12. Impact of three irrigation treatments on the weight of fruits per plant of summer squash genotypes during the season of 2019. Means followed by the same letter (s) are not significantly different at the 5 % level. Error bars are the \pm SD of three biological replicates.

DISCUSSION

The importance of finding genotypes tolerant to drought stress is more urgent than any time ever. It is predicted that the dry regions of the planet may become drier in the future due to climate change and global warming. The Sahara and Arabian deserts are among these regions. It is clear that drought stress severely affects plant performance and growth. In this experiment, we tested the effect of different irrigation treatments, T1 (100 % RAW) control, T2 (75 % RAW) (moderate stress), and T3 (50 % RAW) (severe stress) on six summer squash genotypes. From our results, it is evident that drought stress affected all the studied traits of summer squash genotypes grown under open field conditions during the seasons of 2018 and 2019. Drought stress decreased the plant height and plant fresh weight PFW in all summer squash genotypes in both seasons. The reduction in plant height in T2 and T3 is not very high and genotypes did not show a big difference among them in all treatments. It seems that this trait in summer squash is not affected substantially by different levels of water stress. Plant fresh weight PFW, on the other hand, is more affected by the water stress level. The genotypes also showed no big differences among them in each treatment. It is noticeable that PFW showed almost the same behavior under normal and water stress conditions. Decrease in plant height and PFW may be due to the shortage of water in the plant

cells which decrease the cell division and elongation, which leads to a decrease in each cell turgor, cell volume and eventually cell growth (Anjum *et al*, 2011; Mirabad *et al*, 2014). Drought stress decreases the flow rates of nutrients in the soil, their absorption by stressed root cells, and translocation through the different organs and tissues. Drought stress decreases photosynthesis due to the reduction of CO₂ uptake and stomatal conductance (Efeoglu *et al*, 2009). The reduction of photosynthesis leads to a reduction of dry matter accumulation and biomass (Vashi *et al*, 2020; Kim *et al*, 2020 and Parkash *et al*, 2021). Similar results have been reported by researchers on different crops. Drought stress decreased plant height in many crop plants, such as in tomato (Pervez *et al*, 2009), and in summer squash, (Refai *et al*, 2019; Okasha *et al*, 2020). Plant fresh weight, also, was decreased by drought stress in crops such as in tomato (Khan *et al*, 2015), in melon (Kava *et al*, 2013), in squash (Ors *et al*, 2016), and in pepper (Krishna *et al*, 2018). Drought stress slightly decreased chlorophyll content in leaves of all summer squash genotypes in both seasons. The genotypes did not show a big difference among them. There is no definite pattern to identify the behavior of these genotypes regarding this trait. The chlorophyll pigment is positively correlated with photosynthetic rate and the decrease of chlorophyll content is associated with photosynthesis declining (Anjum *et al*, 2011 and Chen *et al*, 2016). The decrease in the chlorophyll

content occurs as a result of the damage to the chloroplasts by the ROS (Mafakheri *et al*, 2010). Under drought stress conditions, excessive production of ROS causes photo-oxidation in the membrane of the chloroplasts and causes chlorophyll degradation (Fathi and Tari, 2016). Many researchers pointed out results similar to results reported in this experiment. Drought stress decreases leaf chlorophyll content in vegetables such as tomato (Pervez *et al*, 2009), in cantaloupes (Mirabad *et al*, 2014), in squash (Ors *et al*, 2016), in okra (Deveci *et al*, 2017), and in potato (Batool *et al*, 2020). The water deficit decreased the leaf relative water content RWC% in leaves in all summer squash genotypes in both seasons. RWC of leaves is considered an indication of plant water status, reflecting the metabolic activity in the plant (Abd El-Mageed and Semida, 2015). Genotypes with high RWC have been considered to be drought resistant than genotypes with low RWC (Atteya, 2003). Drought stress decreases leaf RWC in vegetables such as tomato, (Khan *et al*, 2015), in squash (Ors *et al*, 2016), in okra (Deveci *et al*, 2017), in melon (Lamaoui *et al*, 2018), in potato (Batool *et al*, 2020). Drought stress increased osmotic adjustment in all summer squash genotypes in both seasons. Increase of osmotic adjustment under drought stress has been considered a physiological mechanism of plants to cope with the drought stress by maintaining turgor pressure and cell volume (Ozturk *et al*, 2020). Osmotic adjustment occurs by lowering the osmotic potential by increasing the accumulation of compatible solutes such as free amino acids, sugars, and proline (Abid *et al*, 2018; Shokat *et al*, 2020). These solutes may be enhancing the root water uptake by the regulation of leaf water potential (Wu *et al*, 2014; Sivakumar *et al*, 2014). In addition, osmotic adjustment enhances the plant capacity to recover its metabolic activities by maintaining the turgor of cells during drought stress (Blum, 2017; Abid *et al*, 2018). Sensitive genotypes showed lower osmotic adjustment than tolerant genotypes under drought stress conditions (Wu *et al*, 2014; Sivakumar *et al*, 2014; Rajarajan *et al*, 2021). Therefore, the osmotic adjustment has been considered an indication of genotypes tolerance to drought stress (Atteya, 2003; Shokat *et al*, 2020). Water

deficit increased leaf osmotic adjustment in tropical and sub-tropical plants, such as in tomato (Sivakumar *et al*, 2014), in cotton (Wu *et al*, 2015), in wheat (Shokat *et al*, 2020), and in Sorghum (Rajarajan *et al*, 2021). Water use efficiency WUE decreased due to increasing drought stress in all summer squash genotypes in both seasons. Genotypes that produced higher yields had higher WUE under normal and stress conditions. WUE under drought stress has been considered as an important physiological parameter reflecting plant tolerance for drought stress (Shabbir *et al*, 2021; Huang *et al*, 2021). Tolerant plants to drought stress have higher values of WUE than sensitive plants (Fandika *et al*, 2011 and Omidian *et al*, 2021). Consequently, WUE is used for determining crop yields in the agricultural production system under drought stress conditions (Omidian *et al*, 2021; Shabbir *et al*, 2021). There has been a discrepancy in research regarding water use efficiency WUE in drought stress. Some researchers found that WUE decreased in moderate and severe drought stress (Zhao *et al*, 2004) in rice, (Ors *et al*, 2016) in summer squash (Gholinezhad, 2020) ,and in wheat. Meanwhile, some researchers found that WUE did not affect moderate drought stress and decreased in severe drought stress (Nazarli *et al*, 2010) in sunflower and (Ahmed and Suliman, 2010) in cowpea. On the other hand, some researchers found that WUE increased in moderate drought stress, while it decreased in severe drought stress (Xu *et al*, 2019; Huang *et al*, 2021). For instance, in cantaloupe (Mirabad *et al*, 2014), in melon (Lamaoui *et al*, 2018), in summer squash (Ati *et al*, 2017). The decrease in WUE might be related to the reduction in the activities of photosynthetic enzymes and the reduction of the yield (Ors *et al*, 2016). Meanwhile, the increase of WUE in moderate drought stress may be due to plants reducing the water consumption and decreasing the water loss through transpiration (Cai *et al*, 2017). Results indicated that drought stress slightly decreased the number of days to 50 % flowering in all summer squash genotypes in both seasons. Under drought stress conditions, plants respond to escape from drought stress by early flowering (Pingping *et al*, 2017). Singh *et al*, (2018) reported that genotypes of rice flowered earlier

when exposed to drought stress and susceptible plants to drought stress flowered earlier than tolerant plants. (Blum and Tuberosa, 2018; Aliarab *et al*, 2020). Water deficit decreased the No. of days to 50 % flowering in tropical and sub-tropical plants such as rice (Singh *et al*, 2018), canola (Kandil *et al*, 2017). Susceptible plants flowered earlier than tolerant plants under drought stress (Singh *et al*, 2018). Also, drought stress decreased the No. of days to heading in wheat (Kiliç and Yagbasanlar, 2010). Our results revealed that drought stress affected yield and yield components. Drought stress decreased total yield, number of fruits/plant and weight of fruits per plant in all summer squash genotypes. A decrease in the yield of summer squash genotypes may be due to the cumulative impacts of drought stress on different growth stages and physiological processes of summer squash genotypes. Water deficit during early growth stages decreased plant height, plant fresh weight, and relative water content, indicating the reduction of photosynthesis. Therefore, all these effects cause a reduction in the yield and yield components of summer squash genotypes. Drought stress decreased the yield, No. of fruits per plant and weight of fruits per plant in many crops, in tomato (Wahb-Allah *et al*, 2011), in summer squash (Amer, 2011; Sadik and Abd El-Aziz, 2018), and in cucumber (Parkash *et al*, 2021). Our results showed that there are differences among genotypes of summer squash under drought stress conditions. Some genotypes performed better than others under drought stress conditions, such as Rivera, Azyad, and Fadwa. These differences in performance might be related to the genotypic variations among these genotypes (Chen *et al*, 2016 and Kandil *et al*, 2017). Also, in tropical and sub-tropical crops, there were differences in genotypes response to drought stress such as in tomato (Wahb-Allah *et al*, 2011), in pepper (Penella *et al*, 2014), in peanut (Pereira *et al*, 2016), in melon (Lamaoui *et al*, 2018), in soybean (Du *et al*, 2020; Basal *et al*, 2020), in potato (Batoool *et al*, 2020).

CONCLUSION

From our results, it could be concluded that drought stress substantially affected all

summer squash genotypes. Decreasing the amount of water applied decreased all studied traits except the osmotic adjustment. In general, all the genotypes surpassed the check cultivar Iskandrany in most of the studied traits. Furthermore, Rivera and Azyad outperformed all summer squash genotypes in most studied traits, especially the yield and yield component traits under normal and stress conditions. Thus, Rivera and Azyad can be considered relatively drought stress-tolerant. Genotypes that performed well under water stress conditions gave high values for traits that indicate water tolerance, such as osmotic adjustment and water use efficiency WUE except for Mabrouka, which was high in osmotic adjustment.

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