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# Influence of Gibberellic Acid on *Beta vulgaris* L. Plants under Drought Conditions

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THIS INVESTIGATION was carried to assess whether gibberellic acid could be I improved suffering beet plants from water deficit under drought. two cultivars of Beta vulgaris L. (Farida and Sultan) and three concentrations of gibberellic acid (50-100-150 mg/l) were used to study their responses to three water regimes (100-75-50 % fc). The experiment was used to determine the tolerance indices, genetic parameters and anatomical and thermal images of various treatments for beet cultivars. The results showed an increase in mean values in studied traits like fresh and dry weight of root, sucrose, proline content, length of stoma and length of stoma pore related with increasing of gibberellic acid concentration up to 150mg/l. Tolerance indices like MP-TOL-YSI-GMP indicated that Farida cultivar was more drought tolerant than Sultan cultivar; and gave the highest yield under the three water regimes at 150 mg/l GA3. Moreover values of genetic parameters; PVC, GVC, GA and h<sup>2</sup> showed increasing in number of studied traits of shoot and root can used it for selection in successive breeding program. Recent developments in imaging technology such as thermal imaging, normalized difference vegetative index (NDVI), chlorophyll picture (SPAD reading) and water stress indices were used to agreement particular opportunities to develop robust high-throughput phenotyping. The surface temperature of crop canopies decreases with increasing transpiration as a result of evaporative cooling, therefore, sugar beet cultivars which had high leaves thermal temperature might be drought tolerance. The results, also, indicated that temporal ground-based NDVI was most current for studying the quantitative drought by a significant effect.

Keywords: Stomata, Proline, Infrared thermal images.

# INTRODUCTION

Beta vulgaris L. is considered an important sugar crop in temperate region. In Egypt (semi -arid region) sugar beet planted beside sugar cane crop to provide people with sugar needs consumption which increased with increasing number of populations. Sugar beet produced 1.255 million tons of sugar represented about 50% from the local production (Egyption Society of Sugar Technologies and Sugar Crops Research Institute, 2014). The shortage of water in Egypt in recent time is major limit to increase the planted area. Moreover, some cultivated area might be suffer from this shortage of water. Sugar beet crop is sensitive to drought especially in seedling stage (Caro & Cucci, 1986). Drought caused several morphological and metabolic changes in cells and whole plant as tolerant responses of it (Blum, 1996). Generally, water deficiency is one of the abiotic stresses that has restricts on cell division, elongation, growth development and plant yield (Pitman & Lauchli, 2002 and Shao et al., 2008).

Drought stress reduced root length, root fresh and dry weights (Rozita et al., 2012). The plant growth regulators can be considered as possible methods for improving the effects of abiotic environmental stresses (Kafi & Damghani, 2007). The pre seed treatment with different hormones *i.e.* gibberellic (GA3) improve the vegetative growth of two wheat cultivars under field condition but caused slight reduction in their grain yield and significant increase in photosynthesis activity in vegetative stage (Ashraf & Iram, 2002). Although, Poostchi & Schmehi (1970) found a decrease in sucrose content of tops and roots of sugar beet plants when treated with gibberellic acid, many tempts and experiments were carried to avoid this. Moreover, GA3 increased proline content in Maize plants (Shahram, 2015) and consequently increased the tolerance of drought. The information of genetic variation and association between agronomic characters is regarded to support considerable help to continue genetic improving in breeding programs. Estimation of genetic factors such as heritability, genetic variation and genetic

correlation are fundamental roles in to predict gains (Da Silva et al., 2008). Due to the variations in visible-near infrared spectral reflectance between healthy and unhealthy canopies, several researchers have recommended the application of visible-near infrared spectroscopy for identifying stress environments in different crops. The increasing availability of different imaging techniques has allowed real time image analysis of physiological changes in plants. Thermal imaging was used to measure and separate leaf surface temperatures to study plant water relations, and specifically for stomatal conductance. (Chaerle & Van Der Straeten, 2007). The aim of this work was; to study the effect of different pre-sowing seed treatments with gibberellic acid on some growth, physiological responses and yield production of sugar beet plants grown under drought stress conditions. Also, can gibberelic acid alleviate the influence of drought stress on sugar beet and how thermal images can detect the difference among the studied treatments as innovative technique.

# MATERIALS AND METHODS

Two Field experiments were performed on 15th November during the two growing seasons of 2013-2014 and 2014-2015 at the Experimental Farm of Faculty of Agriculture, Suez Canal University, Ismailia Governorate, to study the pre sowing treatments of three levels of gibberellic acid (50, 100 and 150 mg/L) in addition to control treatment on some botanical characters of two Beta vulgaris L. cultivars (Farida and Sultan) multigerme. These cultivars were obtained from Sugars Institute Research, Agricultural Center Research, Ministry of Agriculture, Giza, Egypt. The seeds were soaked for 10 hr in GA3 or distilled water. After the pre-sowing treatment, all seeds were washed with distilled water and dried. Drought stress treatments were applied by preventing irrigation to maintain soil moisture content at field capacity of 75% and 50%, in addition to control treatment, where, soil moisture was maintained to field capacity (100%) until harvest. Experimental sub plot consisted of 5 rides 5 m in length and 60 cm in width  $(5*3=15m^2)$ .

#### Root characters

At harvest (180 days after sowing) ten plants were taken randomly to recorded:

- 1- Root diameter (cm).
- 2- Root length (cm).
- 3- Root fresh weights (g /plant <sup>-1</sup>).
- 4- Root dry weights (g /plant <sup>-1</sup>).

# Chemical properties

### Sucrose content

Sucrose percentage in juice of beet root was determined by propol automatic polar meter on lead acetate extract of fresh macerated roots according to the method of Le Docte (1927).

#### Free proline content

It was determined in leaves (mg /g FW) according to Bates et al. (1973)

#### Physiological characters:

The normalized difference vegetation index (NDVI)

The NDVI of the total plant biomass in each plot was measured at 150 days after sowing by means of a portable Green Seeker TM spectroradiometer (Trimple, USA). The sensor head was placed 70 cm above the surface of the plot, covering the total area of the plants and about 10 NDVI measurements were taken at each plot. The averaged NDVI measurements were soil-adjusted by subtracting NDVI measurements taken in empty plot. The NDVI, in the range of -1 to 1, derived from red and near-infrared bands of remotely sensed images-(NIR-RED)/(NIR+RED).

#### Total chlorophyll content (SPAD)

Chlorophyll meter reading as a SPAD values (502 plus- Minolta, Japan) were repeatedly taken at fully expanded sugar beet leaves through the experiments three times and average was calculated.

#### Tolerance indices

Stress tolerance and susceptibility indices including mean productivity (MP), geometric mean productivity (GMP), tolerance (TOL), drought susceptibility index (DSI) and yield stability index (YSI) for water deficit environment were calculated based on seed yield under sever water stress (50% FC) and unstressed (100% FC). Stress tolerance attributes were calculated by the following formulae:

Mean productivity (MP) and Tolerance (TOL) was calculated according to Gupta et al. (2001).

Geometric Mean Productivity (GMP) = 1.

- 2.
- 3.
- 4.
- Mean productivity =  $Y_p + Y_s/2$ Tolerance index (TOL) =  $Y_p Y_s$ Yield stability index (Y SI) =  $Y_s/Y_p$ Drought susceptibility index =  $(1 Y_s/Y_p)/2$ 5. DII

According to Chaudhuri & Kanemasu (1982) where,

Ys = mean root yields of a given genotype in WS (50% FC) condition;

Yp = mean root yields of a given genotype in NS (100% FC) condition;

DII = Drought intensity index.

The drought intensity index (DII) for each water regime (FC%) was calculated as:

 $DII = 1 - X_s / X_n$ 

where, Xs= mean yield of all genotypes under stress and Xp= mean yield of all genotypes without stress

#### Thermal image acquisition

Thermal images of the plots were taken with infrared thermal camera Ti-32 (Fluke Thermography, Germany) equipped with a 320 x 240 pixel microbolometer sensor, sensitive in the spectral range of 7.5–13  $\mu$ m. The canopy height was about 1 m, Images were analyzed in Ti-32 Pro software (Infrared Solutions); Emissivity for measurements of leaves and plant canopies was set at 0.96 while transmission correction was 85%. For more accuracy, the span of auto adjusted thermal image is manually set, in addition to level of the displayed as an important camera feature in order to detect maximum and minimum temperature of the entire display (Wilcox & Makowski, 2014).

#### Thermal indices and stomatal conductance

Where individual leaves were imaged in 2013/2014, dry and wet references were used to mimic leaves with fully closed and fully open stomata, respectively (Jones et al., 2009). These references were determined for beet leaves, cut from the canopy prior to measurements and placed close to the leaves of interest. Wet reference leaves were sprayed with water on both sides, regularly, to maintain their moisture. Dry reference leaves were covered in petroleum jelly (Vaseline) on both sides. The temperatures of these references were obtained  $(T_{drv} \text{ and } T_{wet})$  and used in conjunction with leaf temperatures to obtain thermal indices. The index IG was proportional to the leaf conductance to water vapor transfer which was calculated from leaf temperatures as follow :

 $I_{G} = (T_{dry}-T_{leaf})/(T_{leaf}-T_{wet})$ . This index is theoretically proportional to stomatal conductance (gs) (Cohen *et al.*, 2005). An index analogous to Idso (1982) crop water stress index (CWSI) was

also calculated, where in this case:

$$CWSI = (T_{dry} - T_{leaf}) / (T_{dry} - T_{wet}).$$

Genetic variability

1- The genotypic variance  $\sigma^2 g = M_2 - M^1 / r$ 

2- The phenotypic variance  $\sigma^2 p = \sigma^2 g + \sigma^2 e$ 

 ${}^{\sigma 2}p$  denote to phenotypic variance ,  ${}^{\sigma 2}g$  denote to genotypic variance and  ${}^{\sigma 2}e$  denote to environmental variance.

M1 denote to expected mean squares of genotypes which calculated from ANOVA table. M2 denote to expected mean squares of error which calculated from ANOVA table.

- 3- The genotypic (G.C.V %) and phenotypic (P.C.V %) coefficients of variability were calculated as σ<sub>g</sub>/x<sup>-</sup> and σ<sub>p</sub>/x<sup>-</sup>, respectively.
- 4- Heritability in broad sense (h<sup>2</sup>): heritability in broad sense and coefficient of variation (CV) were calculated for each trait. The calculation of these summary statistics requires knowledge of the error variance for the trait. according to Hallauer & Miranda (1988).
- 5- The genetic advance (GA): calculated according to Allard (1964) was estimated from the following formula:

 $GA = I h^2 Vp.$ 

where I = 1.76 (A 10 % selection intensity ),  $h^2$  : heritability in broad sense.

#### Number and dimensions of stomata

Epidermal strip from upper leaf epidermis was made by eyepiece micrometer, then number (cm<sup>2</sup>), {length, width ( $\mu$ m)} of stomata and {length, width ( $\mu$ m)} of stomata pore were determined according to Willy (1971).

#### Statistical analysis

This study was analyzed by using appropriate analysis of variance (ANOVA) for three factorial experiments in split – split plot design with three replications. Whereas, water treatments were allocated in the main plot, while GA3 treatments In the sub plots and varieties in the sub- sub plots. To illustrate the interaction effects, combined analysis was carried out between the data in the two seasons. Statistical analysis was done using the COSTAT system for Window, version 6.311 (cohort software, Berkeley, CA, USA). Duncan, s test at P 0.05 was used to differentiate between the averages of each factor in this study (Steel *et al.*, 1997).

# **RESULTS AND DISCUSSION**

#### *Root characters*

Results in Table 1 and Fig. 1 cleared that varietal differences, concentrations of gibberellic acid and water stress regime have significant effect on the studied root characters (diameter, length, fresh and dry weights) of sugar beet plants at harvest in the two growing seasons.

Farida cultivar surpassed Sultan cultivar in the all studied characters in both 2013-2014 and 2014-2015 seasons. Farida cv. Recorded an increase in root diameter by 5.82% and 4.78%, in root length by 10.03% and 4.20%, in root fresh weight by 29.91% and 12.37 % and in root dry weight by 19.56% and 10.74% in the two seasons, respectively (Table 1). The differences between the two varieties in root characters might be due to their genetic makeup. Also, Fig. 1 and data in Table 1 showed that increasing concentration of gibberellic acid from 50 mg/l up to 150 mg/l in the soaking solution as pre sowing seed treatments significantly increased root diameter, root length, root fresh weight and root dry weight in both growing seasons. GA3 at 150 mg/l gave an increase in root diameter by 25.08%, 15.31% and 8.25% in first season and by 23.92%, 15.90% and 9.41% in the second season compared with control, 50 mg GA/l and 100 mg GA/l, respectively. Root length was increased by 57.75%, 37.97% and 20.41% in the first season and by 26.60%, 12.91% and 6.21% in the second season with increasing GA3 form 50 mg/l to 100 mg/l or 150 mg/l comparing with control treatment. Root fresh weight increased by 97.07%, 55.46 % and 21.10% in the first season and by 71.55%, 44.82% and 29.95% in the second season. While, root dry weight was increased by 125.97%, 54.98% and 37.98% in the first season and by 75.11%, 33.72% and 19.36% in the second season comparing 150 mg GA/l with control, 50 mg GA/l and 100 mg GA/l. Gibberllic acid as plant hormone is most favorable substance for promoting and improving plant growth expressed as root diameter and root length and consequently fresh and dry weight of root were increased. These results are in harmony with those reported by Poostchi & Schmehi (1970) and Jamil & Rha (2007).

Data in Table 1 as well as in Fig. 1 showed that more water regime stress expressed as 50% FC or 75% FC significantly decreased the studied root characters comparing with control treatment 100 FC% in 2013/ 2014 and 2014/ 2015 seasons. Root diameter decreased by 27.99% and 12.77% in first

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season and by 26.84% and 14.76% in the second one, root length decreased by 42.09% and 20.92% in the first season and by 24.48% and 12.62% in the second season when comparing 50% FC or 75% FC with control treatment (100% FC). Also, root fresh weight was decreased by 68.91% and 39.11% in the first season and by 50.88% and 35.17% in the second season, while root dry weight was decreased by 67.69% and 39.51% in the first season and by 49.63% and 33.78% in the second season. Interactions between (GA3\* FC%) and (Variety\*GA3 \*FC%) were significant for both seasons in root diameter and root length traits. About interactions of (GA3\*FC%), (Variety\* FC%) and (Variety\*GA3) in root fresh and dry weights were significant in both seasons. These results are confirmed with those reported by Caro & Cucci (1986), Blum (1996), Pitman & Lauchli (2002), Shao et al. (2008) and Rozita et al. (2012).

#### Chemical properties

Data in Table 2 as well as Fig. 2 illustrated that varietal differences, concentrations of gibberellic acid and water regimes had significant effect on root sucrose content (%) and leaf proline content (mg/ g FW) in the two growing seasons.

Farida cv. significantly superior Sultan cv. in sucrose % by 4.51% and 3.44% and in leaf proline content by 8.69% and 11.11% in the two growing seasons. Increasing GA3 level in the seed soaking solution significantly increased sucrose percentage in root juice and leaf proline content. 150 mg GA3/l produced the highest value of these chemical compounds, while control treatment gave the lowest values. These results are in harmony with those obtained by Kafi & Damghani (2007), Ashraf & Iram (2002) and Shahram (2015). On the other hand, Poostchi & Schmehi (1970) found a decrease in sucrose content accompanied with GA3 treatments.

Significant decreases were found in sucrose % accompanied with increasing water stress (*i.e.* decreasing water field capacity from 100% FC to 75% or 50% FC). On the other hand, the values of leaf proline content raised by decreasing soil moisture content. The lowest water regime (50% FC) produced the highest values of proline content (8.6 and 10.8 mg/g) in the two growing seasons, respectively. In proline content traitthe interactions between (GA3\*FC%), (Variety \*GA3) and (Variety \*GA3 \*FC%) were significant in both seasons. These results are in agreement with those found by Blum (1996).

|               | Root di   | ameter    | Root l    | ength     | Root free | sh weight | Root dr   | y weight  |
|---------------|-----------|-----------|-----------|-----------|-----------|-----------|-----------|-----------|
| Treat.        | (ст       | n)        | (er       | n)        | (g/plant) |           | (g/p      | lant)     |
|               | 2013/2014 | 2014/2015 | 2013/2014 | 2014/2015 | 2013/2014 | 2014/2015 | 2013/2014 | 2014/2015 |
| Varieties (V) |           |           | -         |           |           |           |           |           |
| Farida        | 32.7 a    | 39.4 a    | 34.0 a    | 34.7 a    | 800.0 a   | 1259.0 a  | 222.5 a   | 330.8 a   |
| Sultan        | 30.9 b    | 37.6 b    | 30.9 b    | 33.3 b    | 615.8 b   | 1120.4 b  | 186.1 b   | 298.8 b   |
| GA3 (G)       |           |           |           |           |           |           |           |           |
| Control       | 28.3 d    | 34.7 d    | 25.8 d    | 29.7 d    | 482.8 d   | 910.8 d   | 128.6 d   | 227.8 d   |
| 50mg/l        | 30.7 c    | 37.1 c    | 29.5 c    | 33.3 c    | 612.0 c   | 1078.9 c  | 187.5 c   | 298.3 c   |
| 100mg/l       | 32.7 b    | 39.3 b    | 33.8 b    | 35.4 b    | 785.6 b   | 1206.7 b  | 210.6 b   | 334.2 b   |
| 150mg/l       | 35.4 a    | 43.0 a    | 40.7 a    | 37.6 a    | 951.4 a   | 1562.5 a  | 290.6 a   | 398.9 a   |
| FC%           |           | •         |           |           | •         |           |           |           |
| 100%          | 36.8 a    | 44.8 a    | 41.1 a    | 38.8 a    | 1106.3 a  | 1668.3 a  | 317.9 a   | 436 a     |
| 75%           | 32.1 b    | 38.1 b    | 32.5 b    | 34.0 b    | 673.5 b   | 1081.5 b  | 192.3 b   | 288.8b    |
| 50%           | 26.5 c    | 32.7 c    | 23.8 c    | 29.3 c    | 344.0 c   | 819.4 c   | 102.7 c   | 219.6 c   |
| Interactions  |           | •         |           |           | •         |           |           |           |
| G * FC        | ***       | ***       | ***       | ***       | ***       | ***       | ***       | ***       |
| V * FC        | ns        | Ns        | *         | *         | ***       | ***       | ***       | ***       |
| V*G           | ns        | Ns        | **        | **        | ***       | ***       | ***       | ***       |
| V*G*FC        | **        | **        | ns        | Ns        | ***       | ***       | ***       | ***       |

TABLE 1. Effect of GA3 treatments and water stress on some root characters of two sugar beet (*Beta vulgaris* L.)cultivars (Farida and Sultan) during 2013/2014 and 2014/2015 seasons.

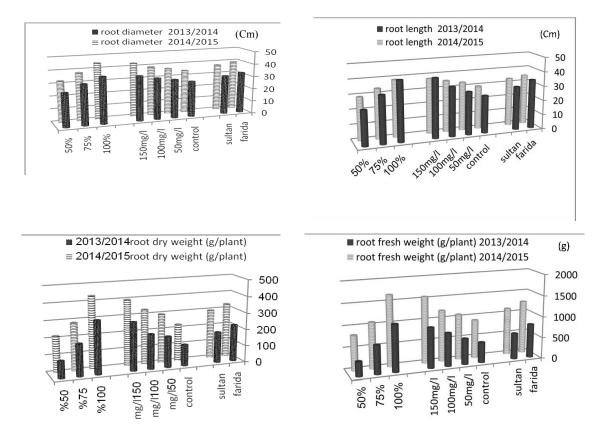
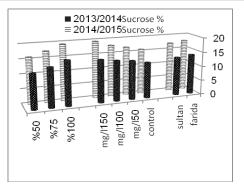


Fig. 1. Effect of GA3 treatments and water stress on root characters of two sugar beet (*Beta vulgaris* L.) cultivars (Farida and Sultan) during 2013/2014 and 2014/2015 seasons.

| Treat.        | Sucro  | se (%)    | Proline con                           | tent (mg/g FW) |
|---------------|--------|-----------|---------------------------------------|----------------|
| 2013/2014     |        | 2014/2015 | 2013/2014                             | 2014/2015      |
| Varieties (V) |        |           |                                       |                |
| Farida        | 16.9 a | 18.0 a    | 7.5 a                                 | 10.0 a         |
| Sultan        | 16.3 b | 17.4 b    | 6.9 b                                 | 9.0 b          |
| GA3(G)        |        |           |                                       |                |
| Control       | 15.4 d | 15.9 d    | 6.0 d                                 | 7.9 d          |
| 50 mg/l       | 16.4 c | 17.1 c    | 6.9 c                                 | 8.7 c          |
| 100 mg/l      | 16.9 b | 18.1 b    | 7.4 b                                 | 9.6 b          |
| 150 mg/l      | 17.6 a | 19.7 a    | 8.5 a                                 | 10.7 a         |
| FC%           |        | 1         |                                       |                |
| 100 %         | 18.3 a | 19.5 a    | 6.0 c                                 | 7.8 c          |
| 75 %          | 16.5 b | 17.6 b    | 7.0 b                                 | 9.2 b          |
| 50 %          | 15.0 c | 16.0 c    | 8.6 a                                 | 10.8 a         |
| Interactions  | •      |           | · · · · · · · · · · · · · · · · · · · |                |
| G * FC        | Ns     | Ns        | *                                     | *              |
| V * FC        | Ns     | Ns        | Ns                                    | Ns             |
| V * G         | Ns     | Ns        | **                                    | **             |
| V*G*FC        | Ns     | Ns        | **                                    | **             |

| TABLE 2. | Effect | of    | GA3     | treatments     | and    | water    | stress or | physiological | characters of two s | ugar beet ( <i>Beta</i> |
|----------|--------|-------|---------|----------------|--------|----------|-----------|---------------|---------------------|-------------------------|
|          | vulga  | ris L | .) cult | tivars ( Fario | la and | l Sultan | ) during  | 2013/2014 and | 2014/2015 seasons.  |                         |



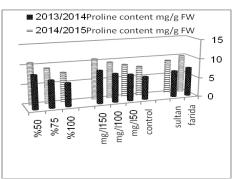


Fig. 2. Effect of GA3 treatments and water stress on physiological characters of two sugar beet (*Beta vulgaris* L.) cultivars (Farida and Sultan) during 2013/2014 and 2014/2015 seasons.

Stomata characters

Table 3 and Plate 1 and 2 illustrate the effect of main factors (varieties, GA3 concentrations and water regimes) on stomata characters in 2014/2015 season.

The varietal differences between Farida cv. and Sultan cv. did not reach to the level of significance 0.5% concerning the all studied characters of stomata, except stoma width and stoma pore length which Farida cv. surpassed Sultan cv. for both traits and that held true in 2013/2014 and 2014/2015 seasons (Table 3 and Plate 1, 2). Increasing GA3 concentration in pre sowing seed treatment significantly increased stoma number, stoma length, stoma width, stoma pore length and stoma pore width in the two

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growing seasons. GA3 at 150 mg/l produced the highest values of these traits.

Data in Table 3 and Fig. 1, 2 showed that reducing the available water in the soil increased numbers of stomata per unit leaf area, but produced smaller sizes of stomata. The ability of plants to be able to regulate the size of the stomatal opening is a very significant mechanism to control water loss. This ability is important during water stress, when loss of water can have serious consequences for the plants. According to McCrea & Richardson (1987) and Sade *et al.* (2012) *Beta* species appear to react with water stress by osmotic adjustment with help of the closing stomata. Osmotic adjustment refers to reduction of osmotic potential in cell due to accumulation of soluble substances under water stress and is the main mechanism used by plants to avoid water stress in dry conditions (Bajji *et al.*, 2001). Generally, water deficit reduces plant growth as a result of inhibition of leaf growth and development due to the lower water availability and turgor pressure (Smirnoff, 1995 and Clover 1997).

 TABLE 3. Effect of GA3 treatments and water stress on some stomata characters of two sugar beet (*Beta vulgaris* L.) cultivars (Farida and Sultan) during 2014/2015 seasons.

| Treat.        | Stoma number      | Stoma Length | Stoma Width | Stoma Pore | Stoma Pore |
|---------------|-------------------|--------------|-------------|------------|------------|
|               | / cm <sup>2</sup> | μm           | μm          | Length µm  | Width µm   |
| Varieties (V) |                   |              |             |            |            |
| Farida        | 244.1 a           | 28.4 a       | 19.9 a      | 16.6 a     | 6.3 a      |
| Sultan        | 244.4 a           | 28.1 a       | 19.0 b      | 15.7 b     | 6.3 a      |
| GA3(G)        |                   |              |             |            |            |
| Control       | 275.0 a           | 27.0 b       | 18.4 b      | 14.8 c     | 6.0 b      |
| 50mg/l        | 247.2 ab          | 27.7 b       | 18.9 b      | 16.2 b     | 6.1 ab     |
| 100mg/l       | 238.9 ab          | 28.9 a       | 20.0 a      | 16.3 b     | 6.5 ab     |
| 150mg/l       | 213.9 b           | 29.4 a       | 20.4 a      | 17.6 a     | 6.7 a      |
| FC%           |                   |              |             |            |            |
| 100%          | 237.5 b           | 28.6 a       | 19.5 a      | 16.6 a     | 7.0 a      |
| 75%           | 237.5 b           | 28.1 b       | 19.5 a      | 16.1 a     | 6.1 b      |
| 50%           | 256.3 a           | 28.0 b       | 19.3 a      | 15.9 a     | 5.9 b      |
| Interactions  |                   |              |             |            |            |
| G * FC        | ns                | ***          | **          | ***        | *          |
| V * FC        | *                 | ***          | ns          | ***        | ns         |
| V * G         | ***               | ***          | ***         | ***        | ***        |
| V*G*FC        | ns                | ***          | ***         | *          | ***        |

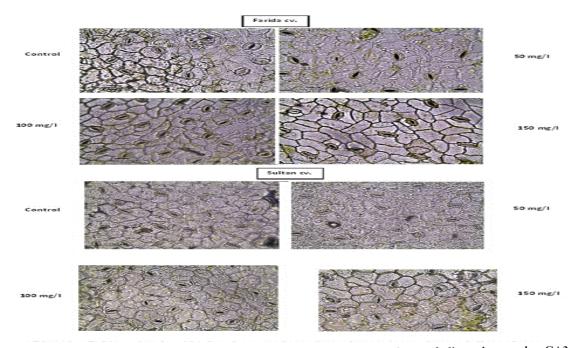


PLATE 1. Epidermal strip of leaflets in sugar beet showed stomata number and dimensions under GA3 treatments. (bar=100 μm).

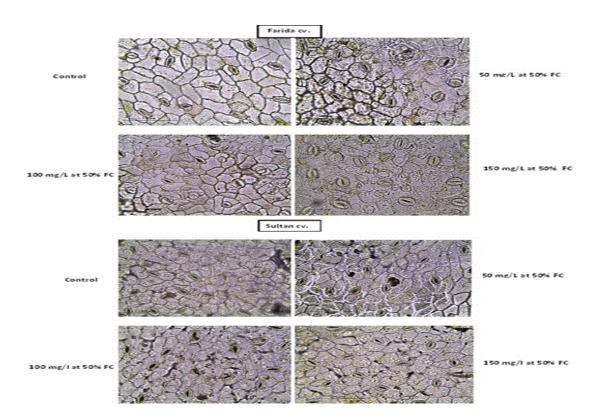


PLATE 2. Epidermal strip of leaflets in sugar beet showed stomata number and dimensions under GA3 treatments at 50% FC. (bar=100 μm).

#### Tolerance indices

The cultivar Farida with treatment 150mg/l gave the highest yield under the three water regimes in the two seasons (Table 4 and Table 5). Yield decreased about 29.6% to 54.7% in season 2013-2014 and 26.2% to 39.7% in season 2014-2015 when plants exposed to 75 and 50% FC, respectively. The values of geometric mean productivity (GMP) ranged from 296.9 g<sup>-1</sup> plant to 1688.2 g<sup>-1</sup> plant in season 2013-2014 and 707.5 g-1 plant to 2466.2 g-1 plant in season 2014-2015 and the cultivar Farida with treatment 150 mg/ IGA3 was the most productive. Mean productivity (MP) ranged between 426.1 g<sup>-1</sup> plant to 1700 g<sup>-1</sup> plant in season 2013-2014 and 805.9 g<sup>-1</sup> plant to 2485 g<sup>-1</sup> plant in season 2014-2015. Tolerance (TOL) index ranged between 400-726.7 in season 2013-2014 and 610-771.7 in season 2014-2015, the lower value of TOL, the mild is the stress tolerance. Yield Stability index (YSI) ranged from 0.16 -0.79 in season 2013-2014 and 0.35 -0.78 in seasons 2014-2015 the higher values indicate high stress tolerance. Besides the yield stability, mean productivity (MP) and geometric mean productivity (GMP) presented similar

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ranking pattern as in drought susceptibility index (DSI). Drought susceptibility index has been used to characterize relative drought tolerance of treatments. Low drought susceptibility index (S < 1) is synonymous with higher stress tolerance. The DSI ranged from 0.31-1.21 in season 2013-2014 and 0.32-0.94 in season 2014-2015. The cultivar Farida with treatment 150mg/l gave low value (0.31, 0.32) for drought susceptibility index (DSI) under the three water regimes in the two seasons The determination of classical vield indices, e.g. DSI, TOL, YSI, MP and GMP measurements, is helpful in the screening of drought tolerance (Jones 2007). Generally, the previous indices indicated that treating cultivar Farida with 150mg/lGA3 gave the most tolerant treatments. Table 6 showed the indices GMP and MP were very similar to the selection based on Yis and Ys. This was confirmed by positive and highly correlations between Ys and GMP (r = 0.99), MP (r = 0.99) and YSI (r = 0.93) and the correlation between YIS and GMP (r = 0.98), MP (r = 0.99) and YSI (r = 0.89). MP is the mean production under both stress and non-stress conditions, and it was highly correlated with yield under both conditions. Thus, MP can be used to identify treatments in the tolerant group. While, there was a high negative correlation between drought susceptibility index (DSI) and the other tolerance indices except TOL (r = 0.79). Therefore, these indices were able to identify superior treatments under drought stress.

DSI, YSI, GMP and MP were correlated with yield under stress conditions, suggesting that these factors are suitable for screening drought tolerant and high yielding treatments in drought stress conditions (Bayoumi *et al.*, 2015).

| TABLE 4 | . Mean harvest root fresh weight per plant for various water regimes, mean productivity (MP),        |
|---------|------------------------------------------------------------------------------------------------------|
|         | geometric mean of productivity (GMP), tolerance index (TOL), yield stability index (YSI) and drought |
|         | susceptibility index (DSI) for season 2013-2014.                                                     |

| Treatmen | ts      |                          | ot fresh weig<br>per plant (g) | ht      | MP     | GMP    | TOL   | YSI  | DSI  |  |
|----------|---------|--------------------------|--------------------------------|---------|--------|--------|-------|------|------|--|
|          |         | 100 % FC 75 % FC 50 % FC |                                | 50 % FC |        |        |       |      |      |  |
|          | Control | 836.7                    | 485                            | 246.7   | 541.7  | 454.3  | 590   | 0.29 | 1.02 |  |
|          | 50mg/l  | 1041.7                   | 666.7                          | 315     | 678.4  | 572.8  | 726.7 | 0.30 | 1.01 |  |
| Farida   | 100mg/l | 1466.7                   | 900                            | 763.5   | 1115.1 | 1058.2 | 703.2 | 0.52 | 0.70 |  |
|          | 150mg/l | 1900                     | 1700                           | 1500    | 1700   | 1688.2 | 400   | 0.79 | 0.31 |  |
|          | Control | 731.7                    | 358.3                          | 120.5   | 426.1  | 296.9  | 611.2 | 0.16 | 1.21 |  |
| Sultan   | 50mg/l  | 843.3                    | 555                            | 250     | 546.7  | 459.2  | 593.3 | 0.30 | 1.02 |  |
| Sultan   | 100mg/l | 1000                     | 693.3                          | 383.3   | 691.7  | 619.1  | 616.7 | 0.38 | 0.89 |  |
|          | 150mg/l | 1030                     | 875                            | 431.7   | 730.9  | 666.8  | 598.3 | 0.42 | 0.84 |  |
| L. S. D  |         | 73.94                    | 81.83                          | 32.85   |        |        |       |      |      |  |
| R. D %   |         |                          | 29.6                           | 54.7    |        |        |       |      |      |  |

TABLE 5. Mean harvest root fresh weight per plant for various water regimes, mean productivity (MP),<br/>geometric mean of productivity (GMP), tolerance index (TOL), yield stability index (YSI) and<br/>drought susceptibility index (DSI) for season2014-2015.

|         |         | Root fres | sh weight per | plant (g) |        |        |       |      |      |
|---------|---------|-----------|---------------|-----------|--------|--------|-------|------|------|
| Treat   | ments   | 100 % FC  | 75 % FC       | 50 % FC   | MP     | GMP    | TOL   | YSI  | DSI  |
| Farida  | Control | 1278.3    | 983.3         | 635       | 956.7  | 901.0  | 643.3 | 0.50 | 0.73 |
|         | 50mg/l  | 1490      | 1073.3        | 820       | 1155.0 | 1105.4 | 670   | 0.55 | 0.65 |
|         | 100mg/l | 1650      | 1123.3        | 1000      | 1325   | 1284.5 | 650   | 0.61 | 0.57 |
|         | 150mg/l | 2790      | 2200          | 2180      | 2485   | 2466.2 | 610   | 0.78 | 0.32 |
|         | Control | 1191.7    | 850           | 420       | 805.9  | 707.5  | 771.7 | 0.35 | 0.94 |
|         | 50mg/l  | 1340      | 1033.3        | 716.7     | 1028.4 | 980.0  | 623.3 | 0.53 | 0.68 |
| Sultan  | 100mg/l | 1570      | 1090          | 873.3     | 1221.7 | 1170.9 | 696.7 | 0.56 | 0.64 |
|         | 150mg/l | 2036.7    | 1500          | 1410      | 1723.4 | 1694.6 | 626.7 | 0.69 | 0.45 |
| L. S. D | ~       | 130.8     | 25.3          | 53.5      |        |        |       |      |      |
| R. D %  |         |           | 26.2          | 39.7      |        |        |       |      |      |

| TABLE 6. Simple correlation of harvest root fresh weight per plant in 100% FC (Yp), 75% FC (YIS) and stressed |
|---------------------------------------------------------------------------------------------------------------|
| 50% FC (Ys) conditions with mean productivity (MP), geometric mean productivity (GMP), tolerance              |
| index (TOL), yield stability index (YSI) and drought susceptibility index (DSI), in treatments.               |

| Variable | Ур   | Yis    | Ys     | МР     | GMP    | TOL     | YSI      | DSI     |
|----------|------|--------|--------|--------|--------|---------|----------|---------|
| Yp       | 1.00 | 0.98** | 0.99** | 0.99** | 0.99** | -0.56** | 0.91**   | -0.91** |
| Yis      |      | 1.00   | 0.99** | 0.99** | 0.98** | -0.59** | 0.89**   | -0.89** |
| Ys       |      |        | 1.00   | 0.99** | 0.99** | -0.63** | 0.93**   | -0.94** |
| MP       |      |        |        | 1.00   | 1.00** | -0.60** | 0.92**   | -0.93** |
| GMP      |      |        |        |        | 1.00   | -0.61** | 0.93**   | -0.93** |
| TOL      |      |        |        |        |        | 1.00    | - 0.79** | 0.79**  |
| YSI      |      |        |        |        |        |         | 1.00     | -1.0**  |
| DSI      |      |        |        |        |        |         |          | 1.00    |

#### *Genetic variability*

Results of genetic variation of ecophysiological traits and yield are illustrated in Table 7. The magnitude of phenotypic coefficient of variation (PCV) values for all the characters were higher than the corresponding (GCV) values indicating that these characters may impacted by environmental factors. Phenotypic coefficients of variability ranged from 9.18 to 63.72% in season 2013-2014 and 7.81 % to 28.89% in season 2014-2015 and the highest PCV attained from harvest root fresh weight and the lowest from osmotic pressure in the two seasons. Genotypic coefficient of variation also had similar trend as phenotypic coefficient of variation. The broad sense heritability was highest for all the recorded traits in the two seasons. For efficient selection we cannot only believe on heritability the combination of high heritability with high genetic advance will provide a clear base on the reliability of that particular trait in the selection of variable entrances. The genetic advance as percentage of means (GA) for traits ranged from 18.65 to 95.7% in season 2013-2014 and 15.04% to 59.5% in season 2014-2015 and the highest GA3 obtained from harvest root fresh weight and the lowest from osmotic pressure in the two seasons. In the breeding program, high range of variability, heritability, genetic improvement and positive correlation coefficient among traits could be an excellent tool for improving or selection genotype (Akbar et al. 2003).

| TABLE 7. Estimation of variance components phenotypic (PCV) (%) and genotypic (GCV) (%) coefficients of |
|---------------------------------------------------------------------------------------------------------|
| variation, broad sense heritability (h <sup>2</sup> ) (%) and genetic advance (GA) (%) for each trait.  |

|                       |       | 2013 / | 2014  |                |       | 2014 / 2 | 2015  |                |
|-----------------------|-------|--------|-------|----------------|-------|----------|-------|----------------|
| Traits                | PCV   | GCV    | GA    | h <sup>2</sup> | PCV   | GCV      | GA    | h <sup>2</sup> |
|                       | (%)   | (%)    | (%)   | (%)            | (%)   | (%)      | (%)   | (%)            |
| Root diameter (cm)    | 13.7  | 12.64  | 27.89 | 99.12          | 11.13 | 10.03    | 22.52 | 98.21          |
| Root length (cm)      | 23.69 | 22.39  | 47.57 | 97.49          | 9.80  | 8.73     | 19.88 | 98.46          |
| Root fresh weight (g) | 63.72 | 61.58  | 95.7  | 99.54          | 28.89 | 26.36    | 59.5  | 98.74          |
| Root dry weight (g)   | 43.63 | 41.56  | 89.59 | 99.69          | 24.97 | 22.88    | 51.07 | 99.31          |
| Sucrose               | 9.23  | 8.16   | 18.8  | 98.44          | 9.75  | 8.61     | 19.5  | 97.04          |
| Proline content       | 20.27 | 19.11  | 41.09 | 98.41          | 15.89 | 14.79    | 3233  | 98.77          |
| Osmotic Pressure      | 9.18  | 8.12   | 18.65 | 98.57          | 7.81  | 6.51     | 15.04 | 93.5           |

#### Physiological characters

Normalized difference vegetation index (NDVI) and total chlorophyll content (SPAD) Results in Figs.3 and 4 illustrated the values of SPAD and NDVI as affected by the three studied factors in 2014-2015 season. It is evident that Farida cv. surpassed Sultan cv. concerning both of SPAD and NDVI values and that held true under the three water regimes. Increasing GA3 concentration up to 150 mg/l increased SPAD

values, but 50 mg/l gave the higher value of NDVI surpassed 150 mg/l. on the other hand, water stress (50% FC) produced smaller SPAD and NDVI values. The healthy vegetation have a good absorption in visible region due to the presence of leaf pigments like xanthophyll, chlorophylls and carotenoids, which strongly absorb the visible area. The NDVI and SPAD reflects vegetation

density and greenness of the land cover and that can be viewed as a holistic indicator for status of plants (healthy or unhealthy) as well as for environmental conditions. Therefore, NDVI and SPAD values can be used as tools for selection of drought tolerance under water stress (Ya Liu *et al.*, 2011).

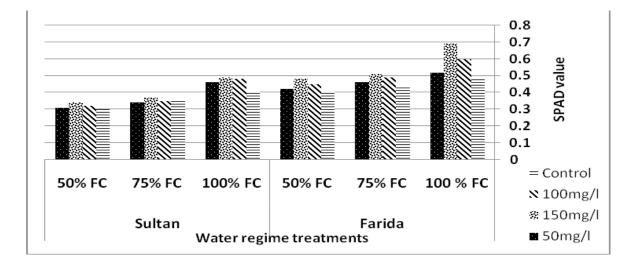


Fig. 3 . Effect of Gibberellic acid on SPAD values for sugar beet cultivars under three field capacitie.

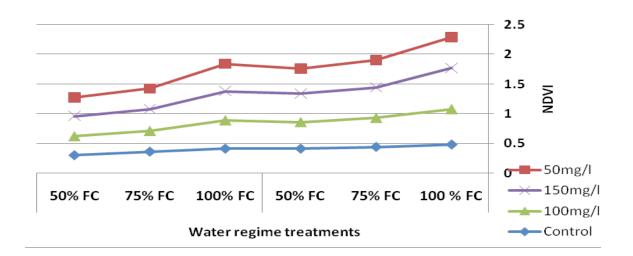


Fig. 4. Effect of Gibberellic acid on NDVI values for sugar beet cultivars under three field capasities.

#### Thermal images acquisition

The plant stress detection using thermal imaging is potentially possible due to the stomatal opening or other physiological changes that happen as a result of plant response/resistance to water deficit. As stomata close under water stress, leaf temperatures rise. Thus leaf or canopy temperatures can be used as an indicator of plant stress and stomatal closure. Thermal imaging systems allow rapid and non-invasive collection

of data, integrated over the area of individual leaves or areas of canopies. Canopy temperature has long been recognized as an indicator of plant water status; therefore, a high-resolution thermal imaging system was used to map crop water status. Potential methods for estimating crop water status from digital infrared images of the canopy were estimated in Fig. 5, 6, 7, 8, 9 and 10. Leaf temperature has been shown to vary when plants are subjected to water stress condition (Fig. 5). Using thermal camera to obtain canopy temperature in sugar beet for two cultivars; Farida cv. was more vigor's than Sultan cv. and appeared less temperature under different environmental stress. Thermal temperature ranged from 27°C to 28.9°C for Farida cv. but it ranged from 27.3 to 30.7°C for Sultan cv. under 150 mg/l. GA3 with 100 mg/l enhanced the plants behavior under the various stress treatments and reduced the thermal temperature. It was ranged from 24.9°C to 26.7°C for Farida cv. and ranged from 26.9°C to 30.7°C for Sultan cv. These results indicated that GA3 with 100 mg/l was more efficient for reducing temperature

of sugar beet leaves In order to make these thermal images suitable for supporting variable-rate irrigation in space and time for sugar beet, identification and assignment of temperature and the derived indices are required. The process of transformation of raw thermal imagery to irrigation and other plant hormone treatments involves three main phases, each related to a wide research area: (1) Definition of crop water status according to temperature; (2) Characterization of the spatial variability pattern of water status in the field; and (3) Determination of thermal indices and comparing with other indices. Measuring stomatal conductance has an advantage overmeasuring photosynthesis, as the former is often more sensitive to water stress on a per unit area basis (James et al., 2002). The canopy stomatal conductance (IG) can be interpreted as the stomatal conductance of the 'big leaf' that characterizes the canopy and is composed of the stomatal conductance of all individual leaves and comparing with anatomical results of stomatal number per unit leaf area and dimensions of stomata.

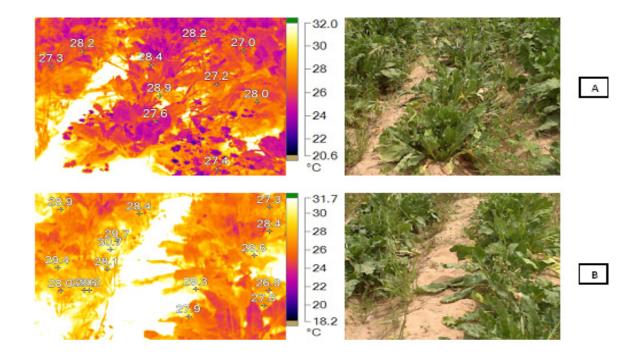


Fig.5. Infrared thermal and visible images under 100% FC and 150 GA level, Farida cultivar (A) and Sultan (B).

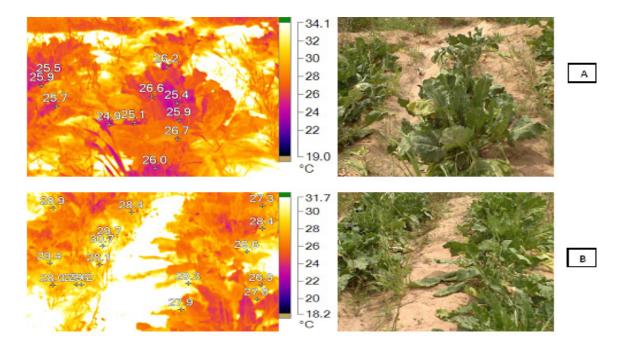


Fig.6. Infrared thermal and visible images under 100% FC and 100 GA level, Farida cultivar (A) and Sultan (B).

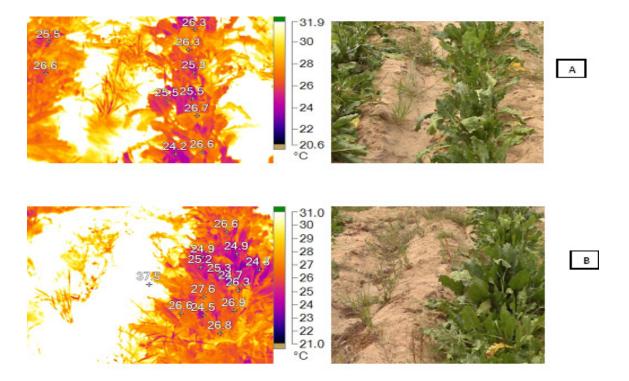


Fig.7. Infrared thermal and visible images under 100% FC and 50 GA level, Farida cultivar (A) and Sultan (B).

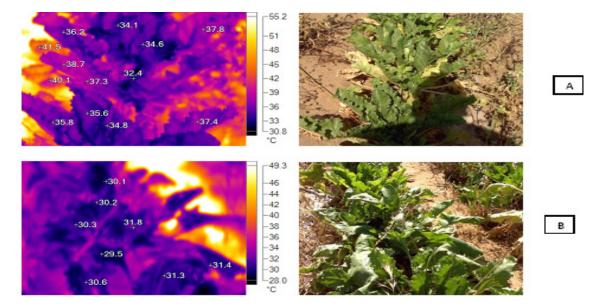


Fig.8. Infrared thermal and visible images under 50% FC and 150 GA level, Farida cultivar (A) and Sultan (B).

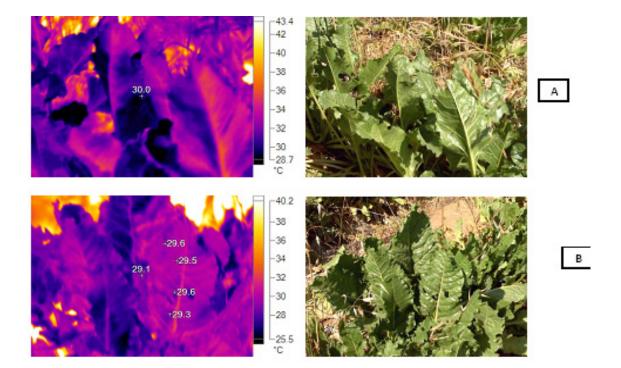


Fig.9. Infrared thermal and visible images under 50% FC and 100 GA level, Farida cultivar (A) and Sultan (B).

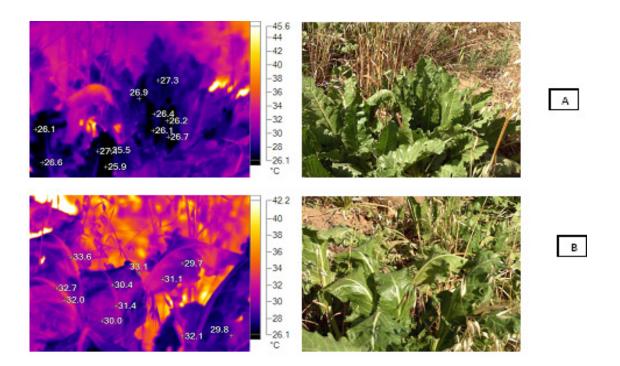


Fig.10. Infrared thermal and visible images under 50% FC and 50 GA level, Farida cultivar (A) and Sultan (B).

# *Effect of water stress on stomatal conductance (IG) and crop water stress index (CWSI)*

The development of thermal imaging can be provide new and practical screening methods for identifying genetic variation among genotypes in the depiction of CWSI and stomatal response (IG) to water stress, in addition to , NDVI as indicator for photosynthesis. Many spectral indices have been suggested to estimate remotely water content of tissues as a measure of water stress. Another powerful application of thermal imaging is to provide a rapid means for screening stomatal conductance or for the identification of genotypes with particular stomatal responses to imposed drought.Treatment of Gibberellic acid with at various levels decreased the leaf temperature (T leaf) and stomatal conductance. The various water deficit treatments created a wide range of water statuses as mirrored by vegetative leaves temperature ( $T_{leaf}$ ), wet leaf temperature ( $T_{wet}$ ), dry leaf temperature ( $T_{dry}$ ), stomatal conductance (IG) and crop water stress index (CWSI) values which were calculated from leaf temperature (Table 8). Stomatal conductance can be used as a indicator of growth rate response to stress, and thermal imaging is a possible screening method for both the laboratory and field. Genetic variation

in response to stress can be exploited in annual crops if irrigation water is available. Tolerant (small stomatal response) selections could be useful for irrigation in arid conditions. Sensitive (large stomatal response) selections could be useful for long-term drought. Another modified index (CWSI) was calculated by thermography to give indication about water relation among treatments.

# **CONCLUSIONS**

It was concluded that drought stress can seriously hamper sugar beet growth but Gibberellic acid might be alleviate this side effect of this stress. There are many physiological, agronomical and anatomical responses are reflected the effect of both drought and Gibberellic acid (GA3) and illustrate how GA3 can be used to enhance the growth and productivity of sugar beet. In our experiments, Application of GA3 with 150 mg/l during presoaking seeds treatments has improved growth performance and production parameters and this reflected on tolerance indices. This paper also provided extensive details about the application of remote sensing technology for detecting the effect of water stress on sugar beet

plants in picture of NDVI and infrared thermal images. This paper proved a concept of using these promising tools as innovation techniques for screening genotypes under water stress. incorporating thermal camera technology with physiological traits was sufficiently to predict clear image for drought tolerance in sugar beet.

| TABLE 8 . Leaf temperatures and thermal indices of stomatal conductance (IG) and crop water stress index |
|----------------------------------------------------------------------------------------------------------|
| (CWSI) for sugar beet under various Gibberellic acid levels for drought treatments.                      |

|        | WS       | GA 3     | T <sub>leaf</sub> | T <sub>wet</sub> | $\mathbf{T}_{dry}$ | $\mathbf{I}_{\mathbf{G}}$ | CWSI   |
|--------|----------|----------|-------------------|------------------|--------------------|---------------------------|--------|
|        |          | Control  | 15.36             | 12.9             | 19.45              | 0.33                      | 0.31   |
|        | 100 % FC | 50 mg/l  | 15.9              | 11.05            | 19.15              | 0.29                      | 0.2    |
|        |          | 100 mg/l | 15.6              | 12.05            | 19.7               | 0.305                     | 0.315  |
|        |          | 150 mg/l | 13.9              | 9.35             | 15.85              | 0.295                     | 0.15   |
| Farida | 75% FC   | Control  | 23.04             | 19.35            | 29.175             | 0.495                     | 0.465  |
|        |          | 50 mg/l  | 23.85             | 16.575           | 28.725             | 0.435                     | 0.3    |
|        |          | 100 mg/l | 23.4              | 18.075           | 29.55              | 0.4575                    | 0.4725 |
|        |          | 150 mg/l | 20.85             | 14.025           | 23.775             | 0.4425                    | 0.225  |
|        | 50 % FC  | Control  | 30.72             | 25.8             | 38.9               | 0.66                      | 0.62   |
|        |          | 50 mg/l  | 31.8              | 22.1             | 38.3               | 0.58                      | 0.4    |
|        |          | 100 mg/l | 31.2              | 24.1             | 39.4               | 0.61                      | 0.63   |
|        |          | 150 mg/l | 27.8              | 18.7             | 31.7               | 0.59                      | 0.3    |
| Sultan |          | Control  | 19.758            | 15.836           | 22.274             | 0.5254                    | 0.2886 |
|        | 100 % FC | 50 mg/l  | 22.875            | 18.9             | 25.05              | 0.5625                    | 0.2625 |
|        |          | 100 mg/l | 22.48             | 15.36            | 23.92              | 0.512                     | 0.136  |
|        |          | 150 mg/l | 20.1              | 14.925           | 22.05              | 0.51                      | 0.2025 |
|        | 75% FC   | Control  | 26.7              | 21.4             | 30.1               | 0.71                      | 0.39   |
|        |          | 50 mg/l  | 30.5              | 25.2             | 33.4               | 0.75                      | 0.35   |
|        |          | 100 mg/l | 28.1              | 19.2             | 29.9               | 0.64                      | 0.17   |
|        |          | 150 mg/l | 26.8              | 19.9             | 29.4               | 0.68                      | 0.27   |
|        | 50 % FC  | Control  | 32.2              | 22.3             | 38.2               | 0.58                      | 0.38   |
|        |          | 50 mg/l  | 28.98             | 20.07            | 34.38              | 0.522                     | 0.342  |
|        |          | 100 mg/l | 29.946            | 20.739           | 35.526             | 0.5394                    | 0.3534 |
|        |          | 150 mg/l | 28.98             | 20.07            | 34.38              | 0.522                     | 0.342  |

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# تأثير حمض الجبريلليك على نباتات بنجر السكر تحت ظروف الجفاف

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اجريت هذه التجربة لتقييم حمض الجبريلاليك في تحسين نباتات بنجر السكر التى تعانى من نقص الماء تحت ظروف الجفاف. استخدم صنفين من اصناف بنجر السكر (Sultan و Farida) مع تركيزات من حمض الجبريلليك (٥٠- ١٠٠ - ١٠ ملليجرام/لتر) لدر اسة استجابتها لثلاث مستويات من الاجهاد المائى هى (٥٠- ٢٥- ١٠٠ ٪) من السعة الحقلية . استخدمت التجربة ادلة التحمل (GMP - ISL - VO - ۷۰ ٪) من السعة الحقلية . استخدمت التجربة ادلة والجمل (QM - - TOL - YSL - ۷۵ - ۷۰ ٪) من السعة الحقلية . استخدمت التجربة ادلة الحرارى مع كل المعاملات لكلا الصنفين . واوضحت النتائج ار تبلط بين زيادة الوزن الغض والجاف للجذر والنسبة المؤية للسكروز ومحتوى البرولين وطول الثغر وطول فتحة الثغر بزيادة تركيزات حمض الجبريلايك واخرص عاملات لكلا الصنفين . واوضحت النتائج ار تبلط بين زيادة الوزن الغض والجاف للجذر والنسبة المؤية للسكروز ومحتوى البرولين وطول الثغر وطول فتحة الثغر بزيادة تركيزات حمض الجبريلايك و محتوى البرولين وطول الثغر وطول فتحة الثغر بزيادة الحرز العض الجبر الحمن الحبين الحوار ومحتوى البرولين وطول الثغر وطول فتحة الثغر بزيادة الحرين وي ار الحمن الجبريلايك حتى ١٥٠ ماليجرام/لتر . اظهرت ادلة التحمل على ان الصنف الاجهاد المائي عند تركيز ١٥٠ ماليجرام/لتر . اظهرت ادلة التحمل على ان الصنف الاجهاد المائي عند تركيز ١٥٠ مالينون والول الثير وطول الثغر ووف المانف العنف العن المانف على ان الصنف والور اثية مثل (درجة التوريث و AG) التباين الور اثي ومعامل التباين المظهري ) زيادة في عدد الصفات المدروسة للمجموع الخضري والجزري التي يمكن استخدامها في برامج التربية. الور اثيبة مثل (درجة التوريث و AG) التباين الور اثي ومعامل التباين المظهري ) زيادة في الور اثيبة مثل (درجة التوريث و AG) التباين الور اثي ومعامل التباين المظهري ) زيادة في الور اثيبة مثل (درجة المو) والغول التوبية ومعامل التباين المول التوبية مثل والور التي ومعامل التباين المو التوبية مثل التربية. الور اثيبة ومعامل التباين المظهري ) زيادة في عدد الصفات المدروسة للمجموع الخضري والجزري التي يمكن المون الخوس الخصري استخدمت تكنولوجيا التصوير الحديثة مثل التصوير الحرارى ودليل الاختلاف في النمو الخمري . المتخري واليمان مو والغول وي الخمري . والما ماليموري الخمري المو مي مانمو الخو وي الخمري . والغول والغو مي مانمو الخي . والمام م