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Influences of Genotype and Drought at Flowering and Grain Filling on Root Architecture Traits of Zea mays L.

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Authors' contributions

This work was carried out in collaboration between all authors. All authors read and approved the final manuscript.

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ABSTRACT

Root architecture traits are important for plant productivity under soil water deficit. The main objective of the present investigation was to assess the effects of deficit irrigation at flowering and grain filling and genotype on maize root traits and grain yield. Twenty two maize genotypes were evaluated in the field under three irrigation regimes; well watering (WW), water stress at flowering (WSF) and at grain filling (WSG) using a split-plot design with three replications. WSF and WSG caused significant reductions of 28.69 and 20.26% in grain yield/plant and 35.53 and 25.51% in grain yield/ha, respectively. WSF caused a significant reduction in four root traits, namely number of aboveground whorls occupied with brace roots (9.31%), number of brace roots (18.27%), number of crown roots (11.50%) and root dry weight (28.31%), but caused a significant increases in three root traits, namely number of brace roots (10.10%), number of crown roots (14.71%) and root dry weight (11.60%), but caused a significant reduction in branching density of crown roots

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(10.05%). Significant differences were observed among genotypes for all studied root traits and grain yield across all irrigation regimes. The best genotypes in grain yield under drought at either flowering or grain filling were characterized by more than one desirable root traits. The cultivars P-3444, Egaseed-77 and SC-128 were considered tolerant genotypes to drought at flowering and grain filling and would be recommended to future breeding programs for improving maize drought tolerance.

Keywords: Maize; water stress; brace roots; crown roots; branching density.

1. INTRODUCTION

Maize (Zea mays L.) is one of the most important cereal crops in the world as well as in Egypt. According to FAOSTAT [1], Egypt grew in 2014, 750,000 hectares and produced 5.8 million tons of maize grains, with an average yield of 7.73 tons ha⁻¹. According to the same report, Egypt ranks the fifth in the world with respect of average productivity of maize after USA, France, Germany and Italy. However, the local production of maize is not sufficient to satisfy the local consumption. Therefore, Egypt imports annually more than six million tons of maize grains. To reach self-sufficiency of maize production in Egypt, efforts are devoted to extend the acreage of maize; in the desert and to improve the maize productivity from unit area. Growing maize in the sandy soils of low waterholding capacity would expose maize plants to drought stress, which could result in obtaining low grain yields under such conditions. Nowadays maize breeders are paying great attention to develop drought tolerant maize cultivars that could give high grain yield under water-stress conditions. Maize is susceptible to drought particularly at the flowering stage [2]. Loss in grain yield is particularly severe when drought stress occurs at this stage [3-5].

Drought tolerance might be enhanced by improving the ability of the crop to extract water from the entire soil profile [6]. Since root is the principal plant organ for nutrient and water uptake, the ability of plant to grow deep roots is considered the most important trait to improve drought tolerance [7].

Deeper soil layers are predominantly reached by maize genotypes forming a sparsely branched axile root system [8]. Root architecture traits are difficult to evaluate directly in the field soil and several high-throughput methods to measure root traits have been reported [9]. At the flowering stage, roots have been measured in the field [10,11], in soil boxes [12] and in soil columns [13,14]. Growing plants in columns or boxes, filled with soil or artificial substrate, can help to reduce sampling efforts compared to field studies and allows growth under controlled conditions and the excavation of roots and measurement of root traits in these systems remains labor-intensive and does not allow for high throughput [9]. In the field, roots and exposed are very different shoots to environmental conditions, especially with regard to temperature, which is an important regulator of root development [15]. Trachsel et al. [9] presented a method to visually score 10 root architectural traits of the root crown of an adult maize plant in the field in a few minutes. According to them, visual measurement of the root crown required 2 min per sample irrespective of the environment. They reported that visual evaluation of root architecture would be a valuable tool in tailoring crop root systems to specific environments.

In general, information about effects of drought stress and maize genotype on root architecture in the field remains scarce. The objectives of the present investigation were: (i) to study the effects of drought stress at flowering and grain filling stages and genotype on the root architecture traits and grain yield of 22 maize genotypes and (ii) to identify high-yielding genotypes with desirable root traits for future use in plant breeding programs to improve drought tolerance.

2. MATERIALS AND METHODS

This study was carried out in the two successive growing seasons 2016 and 2017 at the Agricultural Experiment and Research Station of the Faculty of Agriculture, Cairo University, Giza, Egypt (30°02'N latitude and 31°13'E longitude with an altitude of 22.50 meters above sea level).

2.1 Plant Materials

Seeds of 22 maize (*Zea mays* L.) genotypes (10 single crosses, 5 three-way crosses and 7 open-

pollinated populations) were used in this study (Table 1); 13 genotypes of them were obtained from Agricultural Research Center (ARC), 3 genotypes from Hi-Tec Company, 3 genotypes from DuPont Pioneer Company, one genotype from Fine Seeds Company, one genotype from Egaseed Company, and one genotype from Wataniya Company.

2.2 Experimental Procedures

Sowing date was April 24^{th} in the 1^{st} season (2016) and April 30^{ht} in the 2^{nd} season (2017). Sowing was done in rows; each row was 4 m long and 0.7 m width. Seeds were over sown in hills 25 cm apart, thereafter (after 21 days from planting and before the 1^{st} irrigation) were thinned to one plant/hill to achieve a plant density of 24,000 plants/fed. Each experimental plot included two rows (plot size = 5.6 m²).

2.3 Experimental Design

A split-plot design in randomized complete block (RCB) arrangement with three replications was used. Main plots were allotted to three irrigation regimes, *i.e.* well watering (WW), water stress at flowering (WSF) and water stress at grain filling (WSG). Each main plot was surrounded with an alley (4 m width), to avoid water leaching between plots. Sub plots were devoted to twenty-two maize genotypes.

2.4 Water Regimes

1. Well watering (WW): Irrigation was applied by flooding, the second irrigation was given after three weeks and subsequent irrigations were applied every 12 days.

2. Water stress flowering (WSF): The irrigation regime was just like well watering, but the 4^{th} and 5^{th} irrigations were withheld, resulting in 24 days water stress just before and during flowering stage.

3. Water stress grain filling (WSG): The irrigation regime was just like well watering, but the 6th and 7th irrigations were withheld, resulting in 24 days water stress during grain filling stage.

| Genotype No. | Designation | Origin | Genetic nature | Grain colour |
|-----------------|---------------------|-------------------------|-----------------|-----------------|
| 1 | Hi-Tec-2031 | Hi-Tec, Egypt | Single cross | White |
| 2 | P-30K09 | DuPont Pioneer, Egypt | Single cross | White |
| 3 | Fine-1005 | Fine Seeds, Egypt | Single cross | White |
| 4 | Egaseed-77 | Egaseed Co., Egypt | Single cross | White |
| 5 | SC-10 | ARC, Egypt | Single cross | White |
| 6 | SC-128 | ARC, Egypt | Single cross | White |
| 7 | Hi Tec- 2066 | Hi-Tec, Egypt | Single cross | White |
| 8 | P-3444 | DuPont Pioneer, Egypt | Single cross | Yellow |
| 9 | SC-166 | ARC, Egypt | Single cross | Yellow |
| 10 | P-32D99 | DuPont Pioneer, Egypt | Single cross | Yellow |
| 11 | Hi Tec 1100 | Hi-Tec, Egypt | Three-way cross | White |
| 12 | Watania 11 | Watania Co., Egypt | Three-way cross | White |
| 13 | TWC-324 | ARC, Egypt | Three-way cross | White |
| 14 | TWC-360 | ARC, Egypt | Three-way cross | Yellow |
| 15 | TWC-352 | ARC, Egypt | Three-way cross | Yellow |
| 16 | Giza Baladi | ARC, Egypt | Population | White |
| 17 | Population-45 | ARC, Egypt | Population | Yellow |
| 18 | Nubaria | ARC, Egypt | Population | Yellow |
| 19 | Nebraska Midland | USA | Composite | Yellow |
| 20 | Midland Cunningham | Eldorado, Kansas, USA | Population | Yellow |
| 21 | Golden Republic | Beltsville, Kansas, USA | Population | Yellow |
| 22 | Sweepstakes 5303 Va | USA | Population | Yellow |

Table 1. Designation, origin and grain color of maize genotypes under investigation

2.5 Agricultural Practices

All other agricultural practices were followed according to the recommendations of ARC, Egypt. Nitrogen fertilization at the rate of 280 kg N/ha was added in two equal doses of Urea 46% before the first and second irrigation. Triple Superphosphate Fertilizer (46% P₂O₅) at the rate of 70 kg P₂O₅/ha, was added as soil application before sowing during preparation of the soil for planting. Weed control was performed chemically with Stomp herbicide just after sowing and before the planting irrigation and manually by hoeing twice, the first before the second irrigation (after 21 days from sowing) and the second before the third irrigation (after 33 days from sowing). Pest control was performed when required by spraving plants with Lannate (Methomyl) 90% (manufactured by DuPont, USA) against corn borers.

2.6 Soil Analysis

Physical and chemical soil analyses of the field experiments were performed at laboratories of Soil and Water Research Institute of ARC, Egypt. Across the two seasons, soil type was clay loam: Silt (36.4%), clay (35.3%), fine sand (22.8%) and coarse sand (5.5%), pH (7.92), EC (1.66 dSm⁻¹), SP (62.5), CaCO₃(7.7%), Soil bulk density (1.2 g cm⁻³), HCO₃ (0.71 mEqu/l), CI (13.37 mEqu/l), SO₄ (0.92 mEqu/l), Ca⁺⁺ (4.7 mEqu/l), Mg⁺⁺(2.2 mEqu/l), Na⁺ (8.0 mEqu/l), K⁺ (0.1 mEqu/l), N, P, K, Zn, Mn and Fe (371, 0.4, 398, 4.34, 9.08 and 10.14 mg/kg, respectively).

2.7 Data Recorded

- Grain yield plant⁻¹ (GYPP) (g): It was estimated by dividing the grain yield plot⁻¹ (adjusted at 15.5% grain moisture) on number of plants plot⁻¹ at harvest.
- Grain yield ha⁻¹ (GYPH) (ton): It was estimated by adjusting grain yield plot⁻¹ at 15.5% grain moisture to grain yield ha⁻¹.

At the end of each water stress treatment (80 and 100 days from emergence for WSF and WSG, respectively) and just after re-irrigation, three plant roots from each experimental plot were excavated by removing a soil cylinder of 40 cm diameter and a depth of 40 cm with plant base as the horizontal centre of the soil cylinder. Excavation was carried out using standard shovels. The excavated root crowns were shaken briefly to remove a large fraction of the soil adhering to the root crown. Most of the remaining soil was then removed by soaking the root crown in running water. In a third step remaining soil particles were removed from the root crown by vigorous rinsing at low pressure. The clean roots were measured or visually scored (Fig. 1) for the following traits:

- 3. Number of aboveground whorls occupied with brace roots (BW).
- 4. Number of brace roots (BN).
- 5. Angle of 1st arm of the brace roots originating from whorl 1 (BA) (score).
- 6. Branching density of brace roots (BB) (score).
- 7. Number of crown roots (CN) (score).
- 8. Crown roots angle (CA) (score).
- 9. Branching density of crown roots (CB) (score).

Traits from No. 5 to No. 9 were assigned values from one to nine according to Trachsel et al. [9], where one indicates shallow root angles (10°), low root numbers and a low branching density and nine indicates steep root angles (90°), high numbers and a high branching density (Fig. 1).

- 10. Crown root length (CRL) (cm). The root length, measured as the distance between the last nodes to the end tip of the root.
- 11. Root circumference (RC) (cm). RC was measured from maximum root system width.
- 12. Roots (crown and brace) dry weight (RDW) (g).

The measured root was first spread out in the sun for partial drying and then put in an oven for total drying at 40°C for 24 hours. After drying the roots were weighed using an electronic scale.

2.8 Biometrical Analyses

Analysis of variance of the split-split plot design in RCB arrangement was performed on the basis of individual plot observation using the MIXED procedure of MSTAT ®. Combined analysis of variance across the two growing seasons was also performed if the homogeneity test was nonsignificant. Moreover, combined analysis for each environment separately across seasons was performed as randomized complete block design. Least significant difference (LSD) values were calculated to test the significance of differences between means according to Steel et al. [16].

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Fig. 1. Images of brace roots angle (BA), brace roots branching density (BB), crown roots number (CN), crown roots angle (CA) and crown roots branching (CB) displayed were scored with 1, 3, 5, 7 and 9

3. RESULTS

3.1 Analysis of Variance

Combined analysis of variance across seasons (S) of the split-split plot design (Table 2) indicated that mean squares due to seasons were significant ($P \le 0.05$ or $P \le 0.01$) for six out of studied 12 traits, namely brace root whorls (BW), brace root angle (BA), crown root angle

(CA), crown root branching (CB), grain yield/plant and grain yield/ha. Mean squares due to irrigation regime were significant ($P \le 0.05$ or $P \le 0.01$) for six out of studied 12 traits, namely crown root number (CN), CB, root circumference (RC) and root dry weight (RDW), GYPP and GYPH. Mean squares due to genotype were significant ($P \le 0.01$) for all studied root and grain yield traits. Mean squares due to the 1st order interaction were significant ($P \le 0.05$ or 0.01) for four traits (BN, RC, RDW and GYPH) due to I×S, for six traits (BB, CN, CB, RDW, GYPP and GYPH) due to G×S and two traits (GYPP and GYPH) due to G× I. Mean squares due to the 2nd order interaction, *i.e.* G×S× I, were significant ($P \le$ 0.01) for three traits, namely BB, GYPP and GYPH (Table 2).

Combined analysis of variance of a randomized complete blocks design (RCBD) (data not presented) under four environments, *i.e.* well watering at flowering (WWF), well watering at grain filling (WWG), water stress at flowering (WSF) and water stress at grain filling (WSG) across two seasons indicated that mean squares due to genotypes under all environments were significant ($P \le 0.05$ or 0.01) for 35 out of 46 studied cases (76.1%).

3.2 Effect of Water Stress

Water stress conditions imposed during flowering and grain filling stages caused a significant reduction, of 28.69 and 20.26% in grain yield/plant and 35.53 and 25.51% in grain yield/ha, respectively (Table 3 and Fig. 2).

The changes due to water stress and plant age of the ten measured or scored root traits are presented in Table (3). Water stress at flowering stage caused a significant ($p \le 0.05$ or 0.01) reduction in four root traits, namely number of above-ground whorls occupied with brace roots; BW (9.31%), number of brace roots; BN (18.27%), number of crown roots; CN (11.50%) and roots dry weight; RDW (28.31%), but caused a significant increase (elongation) in crown root length; CRL (9.90%).

On the contrary, water stress at grain filling caused a significant increase in three root traits, namely number of brace roots; BN (10.10%), number of crown roots; CN (14.71%) and roots dry weight; RDW (11.60%), but caused a significant reduction only in branching density of crown roots; CB (10.05%).

Ageing of corn plant from WWF (80 days age) to WWG (104 days age) caused a reduction in 8 out of 10 root traits; such reduction reached significance ($p \le 0.05$ or $p \le 0.01$) in five traits, namely branching density of brace roots (12.62%), number of crown roots (30.45%), branching of crown roots (10.81%), root circumference (10.98%) and root dry weight (20.16%).

| Variance source | | Mean squares | | | | | | | |
|--------------------------|--------|--------------|---------|----------|-----------|----------|--|--|--|
| _ | BW | BN | BA | BB | CN | СА | | | |
| Season (S) | 5.32* | 487.8 | 33.5** | 5.5 | 0.4 | 103.2** | | | |
| Irrigation regime (I) | 2.78 | 2139.6** | 3.2 | 12.9 | 32.5* | 5.4 | | | |
| ÎxS | 4.9* | 615.6 | 3.3 | 15.1 | 4.3 | 10.4 | | | |
| Genotype (G) | 2.91** | 1014.5** | 6.1** | 16.6** | 12.3** | 9** | | | |
| GxS | 0.218 | 85.9 | 2.2 | 10.8** | 4* | 1.7 | | | |
| GxI | 0.449 | 146.8 | 1.5 | 3.7 | 2.5 | 1.6 | | | |
| GxSxI | 0.362 | 122.6 | 1.2 | 5.2* | 2.3 | 1.1 | | | |
| | СВ | CRL | RC | RDW | GYPP | GYPH | | | |
| Season (S) | 28.2** | 243.5 | 107.5 | 94.5 | 26041.5* | 124.7** | | | |
| Irrigation regime (I) | 26** | 115.7 | 618.1** | 1336.5** | 47158.4** | 2041.1** | | | |
| ÎxS | 3.8 | 201.9 | 232.9* | 1278.1** | 3864.3 | 225.5** | | | |
| Genotype (G) | 13.1** | 59.4** | 263.2** | 955.5** | 12428.3** | 707.3** | | | |
| GxS | 4.7** | 13.6 | 26.9 | 234.1** | 3439.6** | 46.4** | | | |
| GxI | 2.5 | 17.2 | 26.7 | 132.9 | 1335.8** | 34.8** | | | |
| GxSxI | 1.8 | 23.1 | 32.2 | 142.4 | 1383.5** | 19.6** | | | |

 Table 2. Mean squares from combined analysis of variance across 2016 and 2017 years for studied root traits of 22 maize genotypes under four irrigation regimes

BW= Number of above-ground whorls occupied with brace roots, BN= Number of brace roots, BA= Brace root angle, BB= Branching density of brace roots, CN= Number of crown roots, CA= Crown roots angle, CB= Branching density of crown roots, CRL= Crown root length, RC= Root circumference, RDW= Roots dry weight, GYPP= Grain yield/plant, GYPH= grain yield/ha, * and ** indicate significance at 0.05 and 0.01 probability levels, respectively

| Trait | Parameter | WWF | WSF | WWG | WSG |
|---------------------------------------|-----------|-------|---------|---------|---------|
| BW | Mean | 2.52 | 2.29 | 2.48 | 2.64 |
| | Change % | | 9.31* | 1.82 | -6.42 |
| BN | Mean | 38.58 | 31.53 | 37.08 | 40.83 |
| | Change % | | 18.27** | 3.88 | -10.10* |
| BA (score) | Mean | 6.70 | 6.91 | 6.71 | 6.53 |
| , , , , , , , , , , , , , , , , , , , | Change % | | -3.17 | -0.12 | 2.61 |
| BB (score) | Mean | 5.34 | 4.94 | 4.67 | 4.69 |
| | Change % | | 7.53 | 12.62* | -0.47 |
| CN (score) | Mean | 3.82 | 3.38 | 2.65 | 3.04 |
| . , | Change % | | 11.50* | 30.54** | -14.71* |
| CA (score) | Mean | 6.77 | 6.89 | 6.49 | 6.49 |
| , , , , , , , , , , , , , , , , , , , | Change % | | -1.67 | 4.25 | -0.06 |
| CB (score) | Mean | 4.56 | 4.59 | 4.07 | 3.66 |
| | Change % | | -0.66 | 10.81* | 10.05* |
| RL (cm) | Mean | 21.74 | 23.90 | 22.99 | 21.76 |
| | Change % | | -9.90* | -5.74 | 5.33 |
| RC (cm) | Mean | 34.51 | 34.36 | 30.72 | 30.87 |
| | Change % | | 0.45 | 10.98** | -0.49 |
| RDW (g) | Mean | 26.17 | 18.76 | 20.89 | 23.32 |
| | Change % | | | 20.16** | -11.60* |
| | - | | WW | WSF | WSG |
| GYPP (g) | Mean | | 128.17 | 91.39 | 102.20 |
| | Change % | | | 28.69** | 20.26** |
| GYPH (ton) | Mean | | 9.02 | 5.82 | 6.72 |
| | Change % | | | 35.53** | 25.51* |

Table 3. Means of root and grain yield traits under well watering (WW), at flowering (WWF) and at grain filling (WWG), water stress at flowering (WSF) and at grain filling (WSG), and change % from WW, WWF or WWG to WSF or WSG, respectively across 2016 and 2017 seasons

BW= Number of above-ground whorls occupied with brace roots, BN= Number of brace roots, BA= Brace root angle, BB= Branching density of brace roots, CN= Number of crown roots, CA= Crown roots angle, CB= Branching density of crown roots, CRL= Crown root length, RC= Root circumference, RDW= Roots dry weight, GYPP= Grain yield/plant, GYPH= Grain yield/ha, * and ** indicate significance at 0.05 and 0.01 probability levels, respectively

| Table 4. Average, minimum (Min) and maximum (Max) values of all studied traits of ea | ach |
|--|-----|
| genotype combined across all irrigation regimes and across 2016 and 2017 season | S |

| Parameter | meter Traits | | | | | |
|--------------------|----------------|-----------|-----------|-----------|----------|----------|
| | BW | BN | BA | BB | CN | СА |
| | (No.) | (No.) | (score) | (score) | (score) | (score) |
| Average | 2.5 | 37.1 | 6.7 | 4.9 | 3.2 | 6.7 |
| Min | 1.9 (8) | 25.6 (21) | 5.5 (1) | 3.4 (18) | 1.9 (21) | 5.6 (7) |
| Max | 3.0 (10,11,17) | 49.0(10) | 7.7(19) | 6.2(9) | 4.5(6) | 8.1(10) |
| LSD _{.05} | 0.36 | 6.8 | 0.74 | 1.09 | 0.86 | 0.76 |
| | СВ | CRL | RC | RDW | GYPP | GYPH |
| | (score) | (cm) | (cm) | (g) | (g) | (ton) |
| Average | 4.2 | 22.8 | 32.7 | 22.3 | 107.3 | 7.18 |
| Min | 3.0 (21) | 20.4 (18) | 25.9 (21) | 11.2 (20) | 62.5(22) | 2.69(22) |
| Max | 6.5 (8) | 26.1 (5) | 38.1 (8) | 36.8(8) | 158.5(6) | 13.03(8) |
| LSD.05 | 0.91 | 2.57 | 2.85 | 6.05 | 9.72 | 0.39 |

Means of minimum and maximum are followed by genotype No. (Between brackets). BW= Number of aboveground whorls occupied with brace roots, BN= Number of brace roots, BA= Brace root angle, BB= Branching density of brace roots, CN= Number of crown roots, CA= Crown roots angle, CB= Branching density of crown roots, CRL= Crown root length, RC= Root circumference, RDW= Roots dry weight, GYPP= Grain yield/plant, GYPH= grain yield/ha



Fig. 2. Means of root traits across genotypes under well watering (WW) at flowering (WWF) and at grain filling (WWG), water stress at flowering (WSF) and at grain filling (WSG) across two seasons

3.3 The Effect of Genotype

Average, minimum and maximum values of all studied traits of 22 genotypes across all irrigation treatments combined across two seasons are presented in Table 4.

Genotypes varied for grain yield/fed from 13.03 ton (genotype No. 8) to 2.69 ton (genotype No. 22), grain yield/plant from 158.5 g (genotype No. 6) to 62.5 g (genotype No. 22), number of aboveground whorls occupied with brace roots from 3.0 from (genotype No. 17) to 1.9 (genotype No. 8), number of brace roots from 49.0 (genotype No. 10) to 25.6 (genotype No. 21), angle of 1st arm of the brace roots originating from whorl 1 from 7.7 (genotype No. 19) to 5.5 (genotype No. 1), branching density of brace roots from 6.2 (genotype No. 9) to 3.4 (genotype No. 18), number of crown roots from 4.5 (genotype No. 6) to 1.9 (genotype No. 21), crown roots angle from 8.1 (genotype No. 10) to 5.6 (genotype No. 7), branching density of crown roots from 6.5 (genotype No. 8) to 3.0 (genotype No. 21), crown root length from 26.1 cm (genotype No. 5) to 20.4 cm (genotype No. 18), root circumference from 38.1 cm (genotype No. 7) to 25.9 cm (genotype No. 21) and roots dry weight from 36.8 g (genotype No. 8) to 11.2 g (genotype No. 20).

The genotype No. 8 (Pioneer-3444) exhibited the highest mean values for four traits [GYPH, root circumference (RC), crown root branching (CB) and roots dry weight (RDW)] and second highest for GYPP, brace root branching (BB), number of crown roots (CN), crown root length (CRL), i.e. most important yield and root traits. The genotype No. 6 (SC-128) developed by ARC-Egypt was the highest in GYPP and number of crown roots and second highest in crown root branching. The genotype No. 4 (Egaseed 77) developed by Fine Seed Co. showed the third highest in grain yield and the highest in brace root angle (BA). The genotype No. 5 (SC-10) developed by ARC-Egypt showed the highest means for one trait (crown root length; CRL); it gave the fourth highest grain yield per plant and per hectare.

On the contrary, the genotype No. 22 (Pop. Sweepstakes 5303) exhibited the lowest means for two traits, namely GYPP, GYPH. The genotype No. 21 (Pop. Golden Republic) exhibited the lowest means for two traits, namely BN and CN. The genotype No. 18 (Pop. Nubaria) showed the lowest means for two traits (BB and CRL).

3.4 Genotype × Water Stress Interaction

Average, minimum and maximum values under each irrigation treatment for all studied root traits and grain yield across two seasons are presented in Table 5.

For root traits (Table 5), data were measured under WWF, WWG, WSF and WSG. Under WWF, WWG, WSF and WSG, for BW the lowest mean was exhibited by genotypes No. 2, 13, 17 and 21 and the highest mean was shown by genotypes No. 17, 19, 4 and 10, for BN the lowest mean by genotypes No. 21, 12, 4 and 21 and the highest mean by genotypes No. 11, 11, 10 and 10, for BA the lowest by genotypes No. 1, 9, 14 and 1 and the highest mean was shown by genotypes No. 19, 21, 21 and 19, for BB the lowest by genotypes No. 18, 18, 13 and 20 and the highest mean was shown by genotypes No. 5, 15, 6 and 9, for CN the lowest by genotypes No. 18, 19, 13 and 13 and the highest mean was shown by genotypes No. 12, 8, 6 and 3, for CA the lowest by genotypes No. 2, 5, 7 and 1 and the highest mean was shown by genotypes No. 10, 10, 21 and 10, for CB the lowest by genotypes No. 21, 17, 19 and 19 and the highest by genotypes No. 8, 8, 6 and 8, for CRL the lowest by genotypes No. 14, 18, 22 and 22 and the highest mean by genotypes No. 8, 5, 9 and 4, for RC the lowest by genotypes No. 18, 19, 19 and 21 and the highest by genotypes No. 7, 8, 7 and 8 and for RDW the lowest by genotypes No. 20, 18, 19 and 21 and the highest by genotypes No. 8, 8, 5 and 8, respectively.

For grain yield (Table 5), data were measured under WW, WSF and WSG. The lowest mean GYPP was shown by genotypes No. 19, 22 and 15 and the highest by genotypes No. 1, 6 and 8 under WW, WSF and WSG, respectively. For GYPH, the lowest mean was exhibited by Genotypes No. 22, 22 and 22 and the highest mean was shown by Genotypes No. 8, 4 and 8 under WW, WSF and WSG, respectively.

On the contrary, the worst genotypes were No. 22 (Sweepstakes) in 3 traits (GYPP, GYPH, CRL) under WSG, 3 traits (GYPP, GYPH, CRL) under WSF and one trait (GYPH) under WW, the genotype No. 21 (Golden Republic) in 4 traits (BW, BN, RC, RDW) under WSG, two traits (BN,CB) under WWF, the genotype No. 19 (Nebraska) in one trait (CB) under WSG, and 3 traits (CB, RC, RDW) under WWG and the genotype No. 18 (Nubaria) in two traits (CN, RC) under WWG and one trait (GYPP) under WW.

| Parameter | WWF | WWG | WSF | WSG | WWF | WWG | WSF | WSG | |
|---------------------------|-----------------------|-------------|-------------|----------|------------------------|----------------|---------------|---------------|--|
| | Brace root whorls no. | | | | | Brace root no. | | | |
| Aver. | 2.52 | 2.48 | 2.29 | 2.64 | 39 | 37.1 | 31.5 | 40.8 | |
| Min | 2 (2) | 1.66 (13) | 1.8 (17) | 1.5 (21) | 27.3 (21) | 22.7 (12) | 23 (4) | 25.2 (21) | |
| Max | 3.1(17) | 3.33(19) | 2.9 (4) | 3.3(10) | 47(11) | 54.7(11) | 43.3(10) | 59(10) | |
| LSD.05 | 0.7 | 0.81 | 0.57 | 0.81 | 16.58 | 14.5 | 7.3 | 14.76 | |
| | Br | race root a | angle (Sco | re) | | Brace root bra | anching (Sco | ore) | |
| Aver. | 6.7 | 6.7 | 6.9 | 6.5 | 5.3 | 4.7 | 4.9 | 4.7 | |
| Min | 5 (1) | 5 (9) | 5.8 (14) | 4.7 (1) | 3.3 (18) | 2 (18) | 3 (13) | 2.3 (20) | |
| Max | 8.3 (19) | 7.3 (21) | 7.5 (21) | 7.5 (19) | 7 (5) | 7 (15) | 6.8 (6) | 6.2 (9) | |
| LSD.05 | 1.62 | 1.88 | 1.02 | 1.25 | 2.38 | 2.66 | 1.66 | 2.02 | |
| | Cro | wn root n | umber (Sc | ore) | | Crown root | angle (Score | e) | |
| Aver. | 3.82 | 2.66 | 3.38 | 3.05 | 6.8 | 6.5 | 6.9 | 6.5 | |
| Min | 1.7 (18) | 1(19) | 1.8 (13) | 1.8 (13) | 5.7 (2) | 5.3 (5) | 5 (7) | 5.2 (1) | |
| Max | 6 (12) | 4 (8) | 5.3 (6) | 5 (3) | 8 (10) | 8 (10) | 8 (21) | 8.5 (10) | |
| LSD.05 | 2.2 | 1.8 | 1.3 | 1.47 | 1.6 | 1.92 | 1.2 | 1.25 | |
| Crown root branching (Sco | | | | icore) | Crown root length (cm) | | | | |
| Aver. | 4.6 | 4.1 | 4.6 | 3.7 | 22.4 | 23.2 | 23.9 | 21.76 | |
| Min | 3 (2) | 2 (17) | 3.2 (19) | 2.2 (19) | 18.6 (14) | 18.8 (18) | 21.2 (22) | 16.9 (22) | |
| Max | 6 (8) | 7.3 (8) | 6.3 (6) | 6.5 (8) | 25.9 (8) | 28.1(5) | 26.2 (9) | 26 (4) | |
| LSD.05 | 1.95 | 2.35 | 1.49 | 1.54 | 6.67 | 5.1 | 4.1 | 4.4 | |
| | Ro | oot circun | nference (c | :m) | | Root dry | weight (g) | | |
| Aver. | 34.7 | 30.7 | 34.4 | 30.9 | 26.2 | 21 | 18.8 | 23.3 | |
| Min | 28.1(18) | 23.3 (19) | 26.5(19) | 23.3(21) | 8.2 (20) | 8.2 (18) | 9.8 (19) | 9.9 (21) | |
| Max | 40.4(7) | 41(8) | 42.5(7) | 36.6(8) | 40.7 (8) | 44.9 (8) | 33.6 (5) | 40.1(8) | |
| LSD.05 | 6.48 | 6.5 | 4.97 | 4.95 | 14.36 | 12.96 | 9.53 | 11.53 | |
| Grain YIELD/PLANT (g) | | | | | Grai | n YIELD/ha (t | ton) | | |
| | WW | WSF | WSG | | | WW | WSF | WSG | |
| Aver. | 128.2 | 91.4 | 102.2 | | | 9.03 | 5.8 | 6.72 | |
| Min. | 82.9 (19) | 31.8 (22) | 58.9 (15) | | | 3.91 (22) | 1.39 (22) | 2.77 (22) | |
| Max. | 168.1 | 156.4 | 179.7 | | | 15.25 (8,5,6) | 10.55 (4,8.6) |) 13.45 (8,6) | |
| | (1,5) | (6,4) | (8,6,4) | | | | | | |
| LSD.05 | 23 | 13.3 | 12.7 | | | 0.75 | 0.63 | 0.71 | |

 Table 5. Average, minimum (Min) and maximum (Max) values under each irrigation treatment

 for all studied root traits and grain yield across two seasons

Means of minimum and maximum are followed by genotype No. (Between brackets)

The four highest and the four lowest performing genotypes under water stress at flowering (WSF) and grain filling (WSG) across seasons are presented in Table 6. Under WSF conditions, the highest mean grain yield/ha was achieved by the single cross Egaseed-77 (developed by Egaseed Co.), followed by P-3444 (developed by Pioneer Co.), SC 128 (developed by ARC, Egypt) and HT-2066 (developed by Hi Tec Co.) in a descending order. The single cross Egaseed-77 was amongst the four highest genotypes under WSF for GYPH, GYPP, BA and CRL. The single cross P-3444 was amongst the four highest genotypes under WSF for GYPH, GYPP, CN, CB and CRL. The single cross SC-128 was amongst the four highest genotypes under WSF for GYPH, GYPP, BB, CN, CB, RC, and RDW. The single cross HT-2066 was amongst the four

highest genotypes under WSF for GYPH, GYPP, CN and RC.

Under WSG conditions, the highest mean grain yield/ha was achieved by the single cross P-3444 (developed by Pioneer) followed by SC-128 (developed by ARC), TWC-324 (developed by ARC) and SC-166 (developed by ARC) in a descending order. The single cross P-3444 was amongst the four highest genotypes in GYPH, GYPP, BB, CB, CRL, RC and RDW, i.e. most important grain yield and root architecture traits. The single cross SC-128 was amongst the four highest genotypes in GYPH, GYPP, BB, CN, CB and RDW (the most important grain yield and root architecture traits). The single cross SC-166 was amongst the four highest genotypes in GYPH and BB.

| WSF | WSG | WSF | WSG | WSF | WSG | |
|--|--|---|--|--|---|--|
| Brace root whorls No. | | Brace root No. | | Brace root angle (score) | | |
| Highest | | | | | | |
| Pop-45 | 32D99 | 32D99 | 32D99 | Nebraska | Nebraska | |
| HT-1100 | HT-1100 | TWC-352 | TWC-352 | Golden | SC-10 | |
| 32D99 | TWC-360 | Pop-45 | HT-1100 | Fine 1005 | Golden | |
| TWC-360 | Pop-45 | HT-1100 | TWC-360 | Eg-77 | Sweep | |
| Lowest | | | | | | |
| Fine 1005 | Eg-77 | Fine 1005 | P-3444 | SC-128 | TWC-352 | |
| SC-128 | P-3444 | Midland | Eg-77 | HT-2066 | Giza | |
| Eg-77 | 30K09 | Golden | 30K09 | SC-166 | TWC-324 | |
| P-3444 | Golden | Eg-77 | Golden | TWC-360 | HT-2031 | |
| Brace root bi | ranching (score) | Crown root r | number (score) | Crown root a | angle (score) | |
| Highest | | | | | | |
| SC-128 | SC-166 | SC-128 | Fine 1005 | Golden | 32D99 | |
| TWC-352 | SC-128 | P-3444 | HT-2031 | 32D99 | Nebraska | |
| SC-166 | P-3444 | HT-2066 | SC-128 | Midland | Midland | |
| 32D99 | SC-10 | TWC-352 | HT-1100 | TWC-324 | Golden | |
| Lowest | | | | | | |
| Golden | Nubaria | Eg-77 | SC-166 | TWC-360 | P-3444 | |
| Giza | Wat- 11 | Sweep | Midland | P-3444 | HT-1100 | |
| Nebraska | Golden | TWC-324 | TWC-324 | HT-2031 | HT-2031 | |
| TWC-324 | Midland | Golden | Golden | HT-2066 | HT-2066 | |
| Crown root b | pranching (score) | Crown root I | ength (cm) | Root circum | ference (cm) | |
| Highest | | | | | | |
| SC-128 | P-3444 | P-3444 | Eg-77 | HT-2066 | P-3444 | |
| P-3444 | HT-1100 | SC-166 | P-3444 | TWC-352 | 30K09 | |
| TWC-352 | HT-2066 | SC-10 | HT-1100 | TWC-352 | TWC-352 | |
| SC-166 | SC-128 | Eg-77 | SC-10 | SC-128 | HT-2031 | |
| Lowest | | | | | | |
| Fine 1005 | Golden | Pop-45 | Nubaria | Nubaria | Nebraska | |
| Eg-77 | 32D99 | HT-2066 | Golden | Midland | Midland | |
| TWC-324 | TWC-324 | Midland | Giza | Golden | Nubaria | |
| Nebraska | Nebraska | Sweep | Sweep | Nebraska | Golden | |
| Root dry weight (g) | | Grain yield/p | lant (g) | Grain yield/h | а | |
| Hignest | D 0444 | 00.400 | D 0 4 4 4 | | D 0 4 4 4 | |
| SC-10 | P-3444 | SC-128 | P-3444 | Eg-// | P-3444 | |
| Fine 1005 | HI-1100 | Eg-// | SC-128 | P-3444 | SC-128 | |
| SC-128 | SC-128 | P-3444 | Eg-77 | SC-128 | 100-324 | |
| 100-352 | HT-2031 | HI-2066 | 50-10 | HI-2066 | 50-100 | |
| LOWEST | Nobraslis | Colder | | Dan 15 | Nobrestie | |
| | Nedraska | Golden | Pop-45 | Pop-45 | INEDFASKA | |
| 1 VV U-324 | Nubaria | Cizo | | Goluen | TVVU-352 | |
| Nebracka | Colden | Giza | 100-302 Sween | Swoon | Sween | |
| Fine 1005 SC-128 TWC-352 Lowest Midland TWC-324 Golden Nebraska | HT-1100 SC-128 HT-2031 Nebraska Midland Nubaria Golden | Eg-77 P-3444 HT-2066 Golden Nebraska Giza Sweep | SC-128 Eg-77 SC-10 Pop-45 Golden TWC-352 Sweep | P-3444 SC-128 HT-2066 Pop-45 Golden Nebraska Sweep | SC-128 TWC-324 SC-166 Nebraska TWC-352 Golden Sweep | |

Table 6. The four highest and the four lowest genotypes for studied traits under water stress at flowering (WSF) and grain filling (WSG) across seasons

4. DISCUSSION

Results of the present study indicated that climatic conditions had a significant effect on BW, BA, CA, CB, GYPP and GYPH and that irrigation regime had a significant effect on CN, CB, RC, RDW, GYPP and GYPH. Moreover,

genotype had an obvious effect on all studied traits. The role of maize genotype is in accordance with the finding of Trachsel et al. [9] for maize root traits and Al-Naggar et al. [3] for grain yield. Mean squares due to the the 1^{st} and 2^{nd} order interaction were significant for some root and yield traits, indicating that for such traits,

the rank of maize genotypes differ from irrigation regime to another, and from one year to another and the possibility of selection for improved root and grain yield under a specific water stressed environment as proposed by Al-Naggar et al. [3-5, 17]. Combined analysis of variance of RCBD under each environment indicated the significance of differences among studied genotypes for the majority of studied root traits and grain yield under each irrigation regime.

The significant reductions of 28.69 and 20.26% in grain yield/plant and 35.53 and 25.51% in grain yield/ha, due to water stress conditions imposed during flowering and grain filling stages, respectively indicate that drought stress at flowering had more severe effect on yield than drought at grain filling. This result is in accordance to Denmead and Shaw [18], who noted that water stress during the vegetative stage of corn production reduced grain yield by 25%, water stress during silking reduced grain vield by 50%, while water stress during grain fill reduced grain yield by 21%. El-Ganayni et al. [19] also reported 33% yield reduction due to water stress at flowering. On the contrary, Al-Naggar et al. [3] found that drought stress at grain filling stage had more severe effect on yield than drought at flowering. They attributed that to stopping irrigation after flowering stage until the end of the season in their experiment and not giving the water required by plants for grain filling. Differences in results of different investigators might be due to differences in soil properties and climate conditions prevailed during the seasons and locations of different studies. Reduction in grain yield due to water deficit at flowering and grain filling stages is in agreement with those of several investigators [19-24].

Water stress at flowering stage (WSF) caused a significant reduction in root traits, namely BW (9.31%), BN (18.27%), CN (11.50%) and RDW (28.31%), but caused a significant increase (elongation) in CRL (9.90%). On the contrary, water stress at grain filling (WSG) caused a significant increase in BN (10.10%), CN (14.71%) and RDW (11.60%), but caused a significant reduction in CB (10.05%). It is observed that significant changes in root traits caused due to water stress at flowering were, in opposite direction to those caused due to water stress at grain filling; they were in the direction of increase for WSG (in 4 out of 5 traits) and in the direction of decrease for WSF (in 3 out of 4 traits). The trend was that WSF caused a reduction, while WSG caused an increase in most studied root traits as compared to its corresponding controls. The reason of differences between the effects of water stress at flowering and at grain filling might be due to the age of plant when water stress occurred. Ageing of corn plant from 80 to 104 days age caused a significant reduction in five traits, namely BB (12.62%), CN (30.45%), CB (10.81%), RC (10.98%) and RDW (20.16%). Replacement of older roots with newly formed roots is referred to as root turnover [25]. Root turnover is important to the success of individual plants in obtaining water and nutrients. Root turnover is estimated from the difference between cumulative births and deaths, which represents either a net accumulation or disappearance of roots [26,27]. It seems in the present study that the difference is in the negative direction, i.e. root deaths are faster than accumulation, especially under water stress conditions.

Means of the 22 maize genotypes showed wide ranges of performance (difference between minimum and maximum values) for all studied root and yield traits across all irrigation treatments. Three commercial varieties showing the highest grain yield showed also the highest means for a number of root traits. The superiority of these three commercial varieties in six root traits (RC, CB, RDW, BB, CN and CRL) for Pioneer-3444, two traits (CN and CB) for SC-128, one trait (BA) for Egaseed 77 and one trait (CRL) for SC-10 might be the reason of their superiority in grain yield, because good roots may help the plants to uptake more water and nutrients from the soil for their biological activities, especially under drought conditions [6,7,28].

In general, the commercial varieties P-3444, SC-128, Egaseed-77 and SC-10 were the best genotypes in our experiment; they showed the highest grain yield and the best root architectural traits across all studied irrigation treatments; they could be recommended for farmers use under a range of different environments as well as for maize breeding programs. On the contrary, it is observed that most of root and yield traits with undesirable mean values were exhibited by populations and the vice versa for traits with desirable means, which were mostly shown by the single crosses.

Results from Tables 4 and 5 concluded that the best genotypes were No. 8 (P-3444) in 5 traits (GYPP, GYPH, CB, RC, RDW) under WSG, 4 traits (CN, CB, RC, RDW) under WWG, 3 traits

(CA, CRL, RDW) under WWF and one trait (GYPH) under WW, the genotype No. 6 (SC 128) in 4 traits (GYPP, BB, CA, CB) under WSF, the genotype No.5 (SC 10) in two traits (BB and CRL) under WWF and WWG, respectively, the genotype No. 7 (Hi-Tec 2066) in one trait (RC) under WSF and RC under WWF, the genotype No. 4 (Egaseed 77) in one trait (GYPH) under WSF, and the genotype No. 2 (30K09) in one trait (GYPH) under WSF.

The best genotypes in grain yield under drought at either flowering or grain filling were characterized by one or more desirable root architecture traits. Accumulating genes of more desirable root characteristics in one genotype might help plants to search water and nutrients in the soil and consequently help plant to accomplish its biological activities and achieve almost its potential grain yield under drought stress at flowering or grain filling stages [6-9,15,28]. The studied single-cross hybrids P-3444, Egaseed-77 and SC-128 were considered drought tolerant genotypes under drought stress at flowering and/or grain filling stages and would be offered to future breeding programs to utilize their genes of desirable root architecture and grain yield traits in improving maize drought tolerance under Egyptian conditions. It should be mentioned that the hybrid P-3444 was characterized in this experiment by its ability to stay green even under water stress, which might help it to tolerate water stress at grain filling stage in a way much better than other tested hybrids and populations.

5. CONCLUSION

This study concluded that WSF caused a significant reduction in four root traits, namely BW, BN, CN, RDW, but caused a significant increase (elongation) in CRL. On the contrary, WSG caused a significant increase in three root traits, namely BN, CN and RDW, but caused a significant reduction only in CB. The study concluded that the single cross cultivars P-3444, Egaseed-77 and SC-128 were considered tolerant to drought at flowering and/or grain filling and would be recommended to future breeding programs to utilize their desirable root system architecture and high grain yield traits in improving maize drought tolerance under Egyptian conditions.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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