

# Putative mechanisms of drought tolerance in maize (*Zea mays* L.) via root system architecture traits

## ABSTRACT

Identifying maize genotypes with favorable root architecture traits for drought tolerance is prerequisite for initiating a successful breeding program for developing high yielding and drought tolerant varieties of maize. The objectives of the present investigation were: (i) to identify drought tolerant genotypes of maize at flowering and grain filling, (ii) to elucidate the relationships between the drought tolerance and root architecture traits and (iii) to identify the putative mechanisms of drought tolerance *via* root system traits. A two-year experiment was carried out using a split plot experiment with three replications. The main plots were devoted to 3 irrigation regimes, *i.e.* well watering (WW), water stress at flowering (WSF) and at grain filling (WSG), and sub plots to 22 maize cultivars and populations. Drought tolerance index (DTI) had strong and positive associations with crown root length (CRL), root circumference (RC) and root dry weight (DRW) under both WSF and WSG, a negative correlation with brace root whorls (BW), and positive correlations with crown root number (CN) under WSF and brace root branching (BB) and crown root branching (CB) under WSG. These root traits could be considered as putative mechanisms of drought tolerance. The cultivars Pioneer-3444, SC-128, Egaseed-77, SC-10 and TWC-324 showed the most drought tolerant and the highest yielding in a descending order; each had a number of such drought tolerance mechanisms. Further investigation should be conducted to determine the underlying root mechanisms contributing to the selection of water-efficient hybrids of maize.

**Key words:** Corn, Crown and Brace roots, Correlations, Drought tolerance index.

## INTRODUCTION

Maize (*Zea mays* L.) in Egypt is mainly used for poultry industry and animal feed. For acreage and production, it ranks second to wheat among cereal crops in Egypt. It is grown as a summer season crop and well irrigated by water coming from Nile River and its branches and canals. Current maize hybrids cultivated in Egypt are selected under well irrigation and therefore are subject to yield losses when grown under water deficit. The amount of water available for irrigation is reducing, especially at the ends of canals and due to expanding maize cultivation into the deserts, where sandy soils are of low water holding capacity. In order to stabilize maize production in Egypt, there is a need to develop drought tolerant maize hybrids.

34 Maize is very sensitive to water stress during the flowering and grain-filling periods  
35 (Bai *et al.* 2006) [1]. However, Witt *et al.* (2012) [2] reported that the most critical period for  
36 yield production goes approximately from 2 weeks before flowering time until 2 weeks after  
37 flowering time. Developing maize varieties that are tolerant to drought is, therefore considered  
38 critical for increasing the maize production. Several investigations have been undertaken across  
39 the years to improve drought tolerance in breeding programs. Edmeades *et al.* (1993) [3]  
40 reported that germplasm developed from drought tolerant source populations performed  
41 significantly better under drought stress compared to conventional populations.

42 Root system architecture traits are important for plant productivity under drought stress  
43 (Lynch\_1995) [4]. Plants avoid dehydration by increasing water uptake in the soil profile and  
44 adapt to the chemical and physical soil constraints, particularly under drought conditions, *via* the  
45 morphological plasticity of their root system (Lynch 2007) [5]. The importance of a deep and  
46 vigorous root system for maintaining yield under drought stress has been reported in maize by  
47 Hund *et al.* (2011) [6]. Rauf and Sadaqat (2008) [7] stated that "drought tolerant genotypes  
48 generally increase the photosynthates allocation for root elongation under drought stress". Rauf *et al.*  
49 (2009) [8] reported that genetic variation for root elongation has been shown in maize. The  
50 effects of root architecture and size on maize yield also depend on the distribution of soil  
51 moisture and the competition for water resources within the plant community (King *et al.* 2009)  
52 [9].

53 Trait interrelationships in particular determine the degree of association among traits  
54 and how they may increase selection efficiency. It is useful if indirect selection for root traits  
55 gives greater response to selection for grain yield trait than direct selection for the same trait. The  
56 main criterion for drought tolerance selection is the association of each root trait with grain yield  
57 under stress conditions [9, 10] (King *et al.* 2009 and Trachsel *et al.*, 2011).

58 To start a successful breeding program for improving drought tolerance, available  
59 maize germplasm should be screened for related traits to drought tolerance; e.g. root architecture  
60 traits under deficit irrigation to identify the best ones for further use in extracting the best  
61 parental inbred lines for developing drought tolerant hybrids. The objectives of the present  
62 investigation were to: (i) characterize 22 maize genotypes for root architecture traits and  
63 tolerance to deficit irrigation at flowering and grain filling stages in order to identify drought

64 tolerant ones, (ii) elucidate the relationships between the drought tolerance and root traits and  
 65 (iii) identify the putative mechanisms of drought tolerance *via* root system architecture.

## 66 MATERIALS AND METHODS

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

### 71 Plant materials

72 Twenty two maize (*Zea mays* L.) genotypes were used, namely 15 Egyptian cultivars (10 single  
 73 crosses and 5 three-way crosses) and 7 open-pollinated populations (Table 1). These materials  
 74 were kindly provided by Hi-Tec Company (Hi-Tec-2031, Hi Tec-2066, Hi Tec 1100), DuPont  
 75 Pioneer Company (P-30K09, P-3444, P-32D99), Fine Seeds Company (Fine-1005), Egaseed  
 76 Company (Egaseed-77), Wataniya Company (Watania 11) and Agricultural Research Center-  
 77 Egypt (the rest of genotypes). These genotypes were chosen to represent the available germplasm  
 78 in Egypt and some of them could be considered sources for extracting drought tolerant inbred  
 79 lines.

80 **Table 1. Designation, origin and grain color of studied maize genotypes.**

Genotype No.	Designation	Origin	Genetic nature	Grain colour
1	Hi-Tec-2031	Hi-Tec, Egypt	Single cross	White
2	P-30K09	DuPont Pioneer	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	Yellow
8	P-3444	DuPont Pioneer	Single cross	Yellow
9	SC-166	ARC, Egypt	Single cross	Yellow
10	P-32D99	DuPont Pioneer	Single cross	Yellow
11	Hi-Tec 1100	Hi-Tec, Egypt	3-way cross	White
12	Watania 11	Watania Co., Egypt	3-way cross	White
13	TWC-324	ARC, Egypt	3-way cross	White
14	TWC-360	ARC, Egypt	3-way cross	Yellow
15	TWC-352	ARC, Egypt	3-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	USA	Population	Yellow

ARC = Agricultural Research Center, SC = Single cross, TWC = Three-way cross

## Experimental procedures

Sowing date was April 24<sup>th</sup> in the 1<sup>st</sup> season (2016) and April 30<sup>th</sup> in the 2<sup>nd</sup> 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 first 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 m<sup>2</sup>).

## 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 (4m width), to avoid water leaching between plots. Sub plots were devoted to twenty-two maize genotypes.

## 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<sup>th</sup> and 5<sup>th</sup> 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 6<sup>th</sup> and 7<sup>th</sup> irrigations were withheld, resulting in 24 days water stress during grain filling stage.

## Agricultural practices

All other agricultural practices were followed according to the recommendations of ARC, Egypt. Nitrogen fertilization at the rate of 120 kg N/fed was added in two equal doses of Urea 46 % before the first and second irrigation. Triple Superphosphate Fertilizer (46% P<sub>2</sub>O<sub>5</sub>) at the rate of 30 kg P<sub>2</sub>O<sub>5</sub>/fed, 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

108 before the planting irrigation and manually by hoeing twice, the first before the first irrigation  
109 (after 21 days from sowing) and the second before the second irrigation (after 33 days from  
110 sowing). Pest control was performed when required by spraying plants with Lannate (Methomyl)  
111 90% (manufactured by DuPont, USA) against corn borers.

#### 112 **Soil analysis**

113 Physical and chemical soil analyses of the field experiments were performed at laboratories of  
114 Soil and Water Research Institute of ARC, Egypt. Across the two seasons, soil type was clay  
115 loam: Silt (36.4%), clay (35.3%), fine sand (22.8%) and coarse sand (5.5%), pH (7.92), EC (1.66  
116 dSm<sup>-1</sup>), SP (62.5), CaCO<sub>3</sub>(7.7 %), Soil bulk density (1.2 g cm<sup>-3</sup>), HCO<sub>3</sub> (0.71 mEqu/l), Cl (13.37  
117 mEqu/l), SO<sub>4</sub> (0.92mEqu/l), Ca<sup>++</sup> (4.7mEqu/l), Mg<sup>++</sup> (2.2mEqu/l), Na<sup>+</sup> (8.0mEqu/l), K<sup>+</sup>  
118 (0.1mEqu/l), N, P, K, Zn, Mn and Fe (371, 0.4, 398, 4.34, 9.08 and 10.14 mg/kg, respectively).

#### 119 **Data recorded:**

120 **1. Grain yield plant<sup>-1</sup> (GYPP) (g):** It was estimated by dividing the grain yield plot<sup>-1</sup> (adjusted at 15.5%  
121 grain moisture) on number of plants plot<sup>-1</sup> at harvest.

122 **2. Grain yield ha<sup>-1</sup> (GYPH) (ton):** It was estimated by adjusting grain yield plot<sup>-1</sup> at 15.5%  
123 grain moisture to grain yield ha<sup>-1</sup> (ton).

#### 124 **Root traits:**

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

134 **3. Number of above-ground whorls occupied with brace roots (BW).**

135 **4. Number of brace roots (BN).**

136 **5. Angle of 1<sup>st</sup> arm of the brace roots originating from whorl 1 (BA) (score).**

137 **6. Branching density of brace roots (BB) (score).**

138 **7. Number of crown roots (CN) (score).**

139 **8. Crown roots angle (CA) (score).**

140 **9. Branching density of crown roots (CB) (score).**

141 Traits from No. 5 to No. 9 were assigned values from one to nine according to  
142 Trachsel *et al.* (2011) [10], where one indicates shallow root angles (10°), low root  
143 numbers and a low branching density and nine indicates steep root angles (90°), high  
144 numbers and a high branching density (Fig.1).

145 **10. Crown root length (CRL) (cm).** The root length, measured as the distance between the last  
146 node to the end tip of the root.

147 **11. Root circumference (RC) (cm).** RC was measured from maximum root system width.

148 **12. Root (crown and brace) dry weight (RDW) (g).** The measured root was first spread out in  
149 the sun for partial drying and then put in an oven for total drying at 40°C for 24 hours. After  
150 drying the roots were weighed using an electronic scale.

151 **Drought tolerance index (DTI):**

152 Drought tolerance index is the factor used to differentiate between the genotypes from tolerance  
153 point of view and it is calculated by the equation of Fageria (1992) [11] as follows:

154 
$$DTI = (Y1/AY1) \times (Y2/AY2)$$

155 Where, Y1 = trait mean of a genotype at well watering. AY1 = average trait of all genotypes  
156 at well watering. Y2 = trait mean of a genotype at water stress. AY2 = average trait of all  
157 genotypes at water stress. When DTI is  $\geq 1$ , it indicates that genotype is tolerant (T) to  
158 drought. If DTI is  $<1$ , it indicates that genotype is sensitive (S) to drought.

159 **Biometrical analyses**

160 Analysis of variance of the split-split plot design in RCB arrangement was performed on the  
161 basis of individual plot observation using the MIXED procedure of MSTAT ®. Combined  
162 analysis of variance across the two growing seasons was also performed if the homogeneity test  
163 was non-significant. Moreover, combined analysis for each environment separately across  
164 seasons was performed as randomized complete block design. Least significant difference (LSD)  
165 values were calculated to test the significance of differences between means according to Steel *et*  
166 *al.* (1997) [12].



**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.**

Simple correlation coefficients were calculated between pairs of studied traits under well watering (WW), water stress (WS), severe water stress (SWS) and combined across all irrigation treatments according to Singh and Narayanan (2000) [13]. Spearman's rank correlation coefficients calculated among studied root traits and other studied traits under studied environments. It was

176 computed by using SPSS 17 computer software and the significance of the rank correlation  
177 coefficient was tested according to Steel *et al.* (1997) [12].

## 178 RESULTS AND DISCUSSION

### 179 3.1. Analysis of variance

180 Combined analysis of variance across seasons (S) of the split-split plot design (Table 2)  
181 indicated that mean squares due to seasons were significant ( $P \leq 0.05$  or  $P \leq 0.01$ ) for six out of  
182 studied 12 traits, namely brace root whorls (BW), brace root angle (BA), crown root angle (CA),  
183 crown root branching (CB), grain yield/plant and grain yield/ha. Mean squares due to irrigation  
184 regime were significant ( $P \leq 0.05$  or  $P \leq 0.01$ ) for six out of studied 12 traits, namely crown root  
185 number (CN), CB, root circumference (RC) and root dry weight (RDW), GYPP and GYPH.  
186 Mean squares due to genotype were significant ( $P \leq 0.01$ ) for all studied root and grain yield  
187 traits.

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

Variance Source	Mean Squares					
	BW	BN	BA	BB	CN	CA
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
I x S	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**
G x S	0.218	85.9	2.2	10.8**	4*	1.7
G x I	0.449	146.8	1.5	3.7	2.5	1.6
G x S x I	0.362	122.6	1.2	5.2*	2.3	1.1
	CB	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**
I x S	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**
G x S	4.7**	13.6	26.9	234.1**	3439.6**	46.4**
G x I	2.5	17.2	26.7	132.9	1335.8**	34.8**
G x S x I	1.8	23.1	32.2	142.4	1383.5**	19.6**

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



194 Mean squares due to the 1<sup>st</sup> order interaction were significant ( $P \leq 0.05$  or  $0.01$ ) for four  
195 traits (BN, RC, RDW and GYPH) due to  $I \times S$ , for six traits (BB, CN, CB, RDW, GYPP and  
196 GYPH) due to  $G \times S$  and two traits (GYPP and GYPH) due to  $G \times I$ . Mean squares due to the 2<sup>nd</sup>  
197 order interaction, *i.e.*  $G \times S \times I$ , were significant ( $P \leq 0.01$ ) for three traits, namely BB, GYPP and  
198 GYPH (Table 2).

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

204 Root system architecture is important for plant productivity under drought stress  
205 conditions [4] (Lynch, 1995). In order to improve plant performance, breeders need to select  
206 genotypes with a root architecture adapted to the conditions of the target environment. Results of  
207 the present study indicated that climatic conditions had a significant effect on BW, BA, CA, CB,  
208 GYPP and GYPH and that irrigation regime had a significant effect on CN, CB, RC, RDW,  
209 GYPP and GYPH. Moreover, genotype had an obvious effect on all studied traits. The role of  
210 maize genotype is in accordance with the finding of Trachsel et al. [10] (2011) for maize root  
211 traits and Al-Naggar et al. (2016a) [14, 15] for grain yield. Mean squares due to the 1<sup>st</sup> and  
212 2<sup>nd</sup> order interaction were significant for some root and yield traits, indicating that for such traits,  
213 the rank of maize genotypes differ from irrigation regime to another, and from one year to  
214 another and the possibility of selection for improved root and grain yield under a specific water  
215 stressed environment as proposed by Al-Naggar et al. (2009, 2011, 2016 b, 2017 a,b) [16-20].  
216 Combined analysis of variance of RCBD under each of the four environments indicated the  
217 significance of differences among studied genotypes for the majority of studied root traits and  
218 grain yield under each irrigation regime.

### 219 3.3. The effect of genotype

220 Average, minimum and maximum values of all studied traits of 22 genotypes across all  
221 irrigation treatments combined across two seasons are presented in Table (3).

222 **Table 3: Average, minimum (Min) and maximum (Max) values of all studied traits of**  
223 **each genotype combined across all irrigation regimes and across 2016 and 2017 seasons.**

Parameter	Traits					
	BW (No.)	BN (No.)	BA (score)	BB (score)	CN (score)	CA (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 <sub>05</sub>	0.36	6.8	0.74	1.09	0.86	0.76
	CB (score)	CRL (cm)	RC (cm)	RDW (g)	GYPP (g)	GYPH (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 <sub>05</sub>	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.

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 above-ground 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 1<sup>st</sup> 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

**Comment [w1]:** you can directly give the name of hybrid

245 by ARC-Egypt was the highest in GYPP and number of crown roots and second highest in crown  
246 root branching. The genotype No. 4 (Egaseed 77) developed by Fine Seed Co. showed the third  
247 highest in grain yield and the highest in brace root angle (BA). The genotype No. 5 (SC-10)  
248 developed by ARC-Egypt showed the highest means for one trait (crown root length; CRL); it  
249 gave the fourth highest grain yield per plant and per hectare.

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

254 Means of the 22 maize genotypes showed wide ranges of performance (difference  
255 between minimum and maximum values) for all studied root and yield traits across all irrigation  
256 treatments. Three commercial varieties showing the highest grain yield showed also the highest  
257 means for a number of root traits. The superiority of these three commercial varieties in six root  
258 traits (RC, CB, RDW, BB, CN and CRL) for Pioneer-3444, two traits (CN and CB) for SC-128,  
259 one trait (BA) for Egaseed 77 and one trait (CRL) for SC-10 might be the reason of their  
260 superiority in grain yield, because good roots may help the plants to uptake more water and  
261 nutrients from the soil for their biological activities, especially under drought conditions [4, 21,  
262 22] (Wright and Nageswara, 1994; Lynch, 1995; Henry et al., 2011).

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

#### 270 **Genotype × water stress interaction**

271 For root traits (Table 4), data were measured under WWF, WWG, WSF and WSG.  
272 Under WWF, WWG, WSF and WSG, for BW the lowest mean was exhibited by genotypes No.  
273 2, 13, 17 and 21 and the highest mean was shown by genotypes No. 17, 19, 4 and 10, for BN the  
274 lowest mean by genotypes No. 21, 12, 4 and 21 and the highest mean by genotypes No. 11, 11,  
275 10 and 10, for BA the lowest by genotypes No. 1, 9, 14 and 1 and the highest mean was shown

Comment [w2]: pls mention the hybrid name  
instead of genotype 1,2,3

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.

**Table 4. Average, minimum (Min) and maximum (Max) values under each irrigation treatment for all studied root traits and grain yield across two seasons.**

Parameter	WWF	WWG	WSF	WSG	WWF	WWG	WSF	WSG
<b>Brace Root Whorls No.</b>					<b>Brace Root No.</b>			
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 <sub>05</sub>	0.7	0.81	0.57	0.81	16.58	14.5	7.3	14.76
<b>Brace Root Angle (Score)</b>					<b>Brace Root Branching (Score)</b>			
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 <sub>05</sub>	1.62	1.88	1.02	1.25	2.38	2.66	1.66	2.02
<b>Crown Root Number (Score)</b>					<b>Crown Root Angle (Score)</b>			
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 <sub>05</sub>	2.2	1.8	1.3	1.47	1.6	1.92	1.2	1.25
<b>Crown Root Branching (Score)</b>					<b>Crown Root Length (cm)</b>			
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 <sub>05</sub>	1.95	2.35	1.49	1.54	6.67	5.1	4.1	4.4
<b>Root Circumference (cm)</b>					<b>Root Dry Weight (g)</b>			
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)

<b>LSD<sub>05</sub></b>	6.48	6.5	4.97	4.95	14.36	12.96	9.53	11.53
	<b>Grain Yield/Plant (g)</b>				<b>Grain Yield/ha(ton)</b>			
	<b>WW</b>	<b>WSF</b>	<b>WSG</b>		<b>WW</b>	<b>WSF</b>	<b>WSG</b>	
<b>Aver.</b>	128.2	91.4	102.2		9.03	5.8	6.72	
<b>Min.</b>	82.9 (19)	31.8 (22)	58.9 (15)		3.91 (22)	1.39 (22)	2.77 (22)	
<b>Max.</b>	168.1(1,5)	156.4(6,4)	179.7(8,6,4)		15.25(8,5,6)	10.55(4,8,6)	13.45(8,6)	
<b>LSD<sub>05</sub></b>	23	13.3	12.7		0.75	0.63	0.71	

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

For grain yield (Tables 5 and 6), data were measured under WW, WSF and WSG. The lowest mean GYPF 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.

**Table 5. Means of grain yield/plant and grain yield/ha for each genotype under each irrigation regime (well watering; WW, water stress at flowering; WSF and water stress at grain filling; WSG) across 2016 and 2017 seasons.**

Genotype	WW	WSF	Ch%	WSG	Ch%	WW	WSF	Ch%	WSG	Ch%
	<b>Grain yield/plant</b>					<b>Grain yield/ha</b>				
<b>1</b>	168.1	78.0	53.6	102.7	38.9	9.95	4.40	55.8	6.30	36.7
<b>2</b>	131.7	73.3	44.3	92.0	30.1	8.51	3.79	55.5	5.51	35.2
<b>3</b>	124.0	75.6	39.1	109.0	12.2	7.98	4.29	46.3	6.29	21.2
<b>4</b>	151.6	147.9	2.5	132.5	12.6	9.56	8.35	12.7	6.36	33.5
<b>5</b>	166.3	123.2	25.9	126.0	24.2	10.22	5.96	41.7	6.65	34.9
<b>6</b>	150.4	156.4	-4.0	168.7	-12.2	10.05	8.14	19.1	8.38	16.6
<b>7</b>	128.5	131.2	-2.1	106.8	16.9	7.34	6.41	12.6	4.76	35.2
<b>8</b>	150.4	137.6	8.5	179.7	-19.5	12.11	8.21	32.2	10.67	11.9
<b>9</b>	134.4	105.6	21.4	121.0	9.9	8.12	5.64	30.6	6.69	17.7
<b>10</b>	134.3	98.9	26.4	117.7	12.3	8.32	5.31	36.2	6.43	22.8
<b>11</b>	125.5	78.5	37.4	84.7	32.5	7.61	4.02	47.2	4.50	40.9
<b>12</b>	119.4	91.0	23.8	111.5	6.6	7.79	5.12	34.2	6.09	21.8
<b>13</b>	149.4	111.1	25.6	120.7	19.2	9.28	5.96	35.8	7.16	22.8
<b>14</b>	133.6	89.7	32.9	81.9	38.7	5.65	4.15	26.5	3.86	31.7
<b>15</b>	125.4	84.7	32.5	58.9	53.1	4.96	3.79	23.6	3.05	38.5
<b>16</b>	118.6	56.2	52.6	81.9	30.9	4.30	2.84	33.9	4.12	4.1
<b>17</b>	110.9	65.0	41.4	70.8	36.2	4.86	2.80	42.4	3.62	25.6
<b>18</b>	110.5	74.2	32.9	85.8	22.4	5.37	3.22	40.1	4.54	15.4
<b>19</b>	82.9	59.4	28.4	75.8	8.5	3.83	2.33	39.1	3.38	11.9
<b>20</b>	106.6	79.7	25.2	91.4	14.3	4.64	3.00	35.4	3.63	21.9
<b>21</b>	100.8	61.8	38.7	70.4	30.2	3.79	2.60	31.5	3.04	19.8

<b>22</b>	96.9	31.8	67.2	58.9	39.3	3.10	1.11	64.2	2.19	29.4
<b>Average</b>	128.2	91.4	28.7	102.2	20.3	7.15	4.61	35.5	5.33	25.5
<b>Min.</b>	82.9	31.8		58.9		3.10	1.11		2.19	
<b>Max.</b>	168.1	156.4		179.7		12.11	8.35		10.6	
<b>LSD<sub>.05</sub></b>	23	13.3		12.7		0.6	0.5		0.6	
<b>LSD<sub>.01</sub></b>	30.5	17.6		16.8		0.8	0.7		0.8	

Ch% = 100(WW-WSF or WSG)/WW

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.

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.

**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.**

WSF	WSG	WSF	WSG	WSF	WSG
<b>Brace root whorls No.</b>		<b>Brace root No.</b>		<b>Brace root angle (score)</b>	
		<b>Highest</b>			
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
		<b>Lowest</b>			
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
<b>Brace root branching (score)</b>		<b>Crown root number (score)</b>		<b>Crown root angle (score)</b>	
		<b>Highest</b>			
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 branching (score)		Crown root length (cm)		Root circumference (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/plant (g)		Grain yield/ha	
Highest					
SC-10	P-3444	SC-128	P-3444	Eg-77	P-3444
Fine 1005	HT-1100	Eg-77	SC-128	P-3444	SC-128
SC-128	SC-128	P-3444	Eg-77	SC-128	TWC-324
TWC-352	HT-2031	HT-2066	SC-10	HT-2066	SC-166
Lowest					
Midland	Nebraska	Golden	Pop-45	Pop-45	Nebraska
TWC-324	Midland	Nebraska	Golden	Golden	TWC-352
Golden	Nubaria	Giza	TWC-352	Nebraska	Golden
Nebraska	Golden	Sweep	Sweep	Sweep	Sweep

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.

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.

332 The best genotypes in grain yield under drought at either flowering or grain filling were  
 333 characterized by one or more desirable root architecture traits. Accumulating genes of more  
 334 desirable root characteristics in one genotype might help plants to search water and nutrients in  
 335 the soil and consequently help plant to accomplish its biological activities and achieve almost its  
 336 potential grain yield under drought stress at flowering or grain filling stages [4, 10, 21-24]  
 337 (Wright and Nageswara, 1994; Lynch,1995; Hund et al.,2009 b;Hund,2010; Henry et al.,  
 338 2011;Trachsel et al. (2011). The studied single-cross hybrids P-3444, Egaseed-77 and SC-128  
 339 were considered drought tolerant genotypes under drought stress at flowering and/or grain filling  
 340 stages and would be offered to future breeding programs to utilize their genes of desirable root  
 341 architecture and grain yield traits in improving maize drought tolerance under Egyptian  
 342 conditions. It should be mentioned that the hybrid P-3444 was characterized in this experiment  
 343 by its ability to stay green even under water stress, which might help it to tolerate water stress at  
 344 grain filling stage in a way much better than other tested hybrids and populations.

### 345 3.2. Drought tolerance index

346 Drought tolerance index (DTI) values of studied genotypes under the stressed  
 347 environments WSF and WSG are presented in Table (7). According to our scale, when DTI is  
 348  $\geq 1.0$ , it indicates that genotype is tolerant (T), if DTI is 1.0, it indicates that genotype is  
 349 moderately tolerant (MT) and if DTI is  $< 1.0$ , it indicates that genotype is sensitive (S).

350 Based on DTI values, the 22 studied maize genotypes were grouped into three categories  
 351 under water stress at flowering, namely tolerant (10 genotypes), moderately tolerant (two  
 352 genotypes) and sensitive (10 genotypes) (Table 7). Under water stress conditions at grain filling,  
 353 number of tolerant (T), and sensitive (S) genotypes were 11, and 11, respectively.

354 **Table 7. Drought tolerance index (DTI) of each genotype under WSF and WSG environments.**

Genotype No.	Designation	WSF	WSG	Genotype No.	Designation	WSF	WSG
1	Hi-Tec-2031	1.3	1.6	12	Watania -11	1.2	1.2
2	P-30K09	1.0	1.2	13	TWC-324	1.7	1.7
3	Fine 1005	1.0	1.3	14	TWC-360	0.7	0.6
4	Egaseed-77	2.4	1.6	15	TWC-352	0.6	0.4
5	SC-10	1.8	1.8	16	Giza Baladi	0.4	0.5
6	SC-128	2.5	2.2	17	Population-45	0.4	0.5
7	Hi-Tec-2066	1.4	0.9	18	Nubaria	0.5	0.6
8	P-3444	3.0	3.4	19	Nebraska Midland	0.3	0.3



9	SC-166	1.4	1.4	20	Midland Cunningham	0.4	0.4
10	P-32D99	1.3	1.4	21	Golden Republic	0.3	0.3
11	Hi-Tec-1100	0.9	0.9	22	Sweepstakes 5303	0.1	0.2

The highest DTI under both the two stressed environments (WSF and WSG) was exhibited by the genotype No. 8 (P-3444). The 2<sup>nd</sup> and 3<sup>rd</sup> highest genotypes in DTI were SC-128 and Egaseed-77 under WSF and SC-128 and SC-10 under WSG. For productivity (grain yield/plant) under WSF, the genotype Egaseed-77 ranked 1<sup>st</sup>, but P-3444 and SC-128 ranked 3<sup>rd</sup>. Under WSG, P-3444, SC-128 and SC-10 ranked 1<sup>st</sup>, 2<sup>nd</sup> and 3<sup>rd</sup>, for productivity as well as drought tolerance index.

On the contrary, the most drought sensitive genotypes were the open-pollinated populations Sweepstakes 5303, Golden Republic and Nebraska Midland under both water stress environments (WSF and WSG); their grain yield were the lowest.

### 3.3. Superiority of drought tolerant (T) to sensitive (S) genotypes

Based on grain yield/plant and drought tolerance index (DTI) the best three genotypes were the single cross hybrids P-3444, SC-128 and Egaseed-77 under WSF and P-3444, SC-128 and SC-10 under WSG, while the most drought sensitive and lowest yielding genotypes were the populations Sweepstakes, Golden Republic and Nebraska Midland under both water stress environments (WSF and WSG). Data averaged for each of the two groups (T and S) under WSF and under WSG indicated that GYPP of drought tolerant (T) was greater than that of the sensitive (S) genotypes by 189.0 and 131.3 % under drought at flowering (WSF) and grain filling (WSG), respectively (Table 8).

**Table 8. Superiority (Sup.%) of the three most tolerant (T) to the three most sensitive (S) genotypes for selected traits under the stressed environments WSF and WSG, combined across 2016 and 2017 seasons.**

Trait	WSF			WSG		
	T	S	Sup. %	T	S	Sup. %
Grain yield/plant	147.3	51.0	189.0**	158.1	68.3	131.3**
Crown root number	4.2	2.4	76.7**	3.4	2.3	45.2*
Crown root branching	5.4	3.8	42.6*	4.6	2.5	84.4**
Crown root length	25.6	22.9	11.3*	23.3	18.6	25.4*
Root circumference	35.6	28.4	25.4**	32.6	26.4	23.6*
Root dry weight	20.1	10.7	86.7*	33.1	14.6	126.3**

\* and \*\* indicate significance at 0.05 and 0.01 probability levels, respectively.

Significant superiority of drought tolerant (T) over sensitive (S) genotypes in GYPP under drought at flowering and grain filling was associated with significant superiority in higher CN (76.7 and 45.2%), CB (42.6 and 84.4%), higher CRL (11.3 and 25.4 %), higher RC (25.4 and 23.6%) and higher RDW (86.7 and 126.3%), respectively.

### 3.4. Correlations between drought tolerance and root traits

Drought tolerance index had a strong significant ( $p \leq 0.01$ ) and positive correlation with grain yield/plant ( $r = 0.912^{**}$  and  $0.941^{**}$ ) under WSF and WSG conditions, respectively (Table 9). Drought tolerance had a significant and positive correlation coefficient, with crown root length ( $r = 0.693^{**}$  and  $0.561^{**}$ ), root circumference ( $0.440^*$  and  $0.499^*$ ) crown root dry weight ( $r = 0.410^*$  and  $0.592^{**}$ ) under WSF and WSG conditions, respectively.

Moreover, drought tolerance index had a significant and negative correlation coefficient with brace root whorls; BW ( $-0.598^{**}$ ) and a significant and positive correlation coefficient with brace root branching; BB ( $0.506^*$ ) and crown root branching ( $0.489^*$ ) under WSG.

**Table 9. Correlation coefficients between drought tolerance index (DTI) and means of studied traits of all genotypes under water stress at flowering (WSF) and at grain filling (WSG) across seasons.**

Trait	WSF	WSG	Trait	WSF	WSG
Grain yield/plant	.912**	.941**	Crown root angle	-.319	-.203
Brace root whorls number	-.598**	-.288	Crown root branching	.381	.489*
Brace root Number	-.250	-.231	Crown root length	.693**	.561**
Brace root angle	-.183	-.193	Root circumference	.440*	.499*
Brace root Branching	.169	.506*	Root dry weight	.410*	.592**
Crown root number	.469*	.320			

\* and \*\* indicate significance at 0.05 and 0.01 probability levels, respectively.

### 3.5. Correlations between grain yield and root traits

Estimates of rank correlation coefficients among grain yield/plant and all studied root traits across the two seasons under well watering, water stress at flowering (WSF) and grain filling (WSG) were calculated across all genotypes and presented in Table (10). Under well watering, grain yield/plant had a significant ( $p \leq 0.01$ ) and positive association with the root dry weight (RDW) (0.42), root circumference (RC) (0.43), crown root length (0.26), crown root branching (CB) (0.27), number of crown roots (CN) (0.23) and brace root branching (BB) (0.34).

Data in Table (10) showed that under WSF, grain yield/plant was significantly ( $P \leq 0.01$ ) and positively correlated with each of RC ( $r=0.33$ ) and CN ( $r=0.27$ ). Under water stress at

grain filling (WSG), grain yield/plant had a significant and positive correlation ( $p \leq 0.01$  or  $p \leq 0.05$ ) with CRL ( $r=0.33$ ), CB ( $r=0.25$ ), RDW ( $r=0.23$ ), BB ( $r=0.18$ ) and RC ( $r=0.17$ ).

**Table 10. Correlation coefficients between grain yield/plant and each of studied root traits of maize under well watering (WW), water stress at flowering (WSF) and water stress at grain filling (WSG) across two years.**

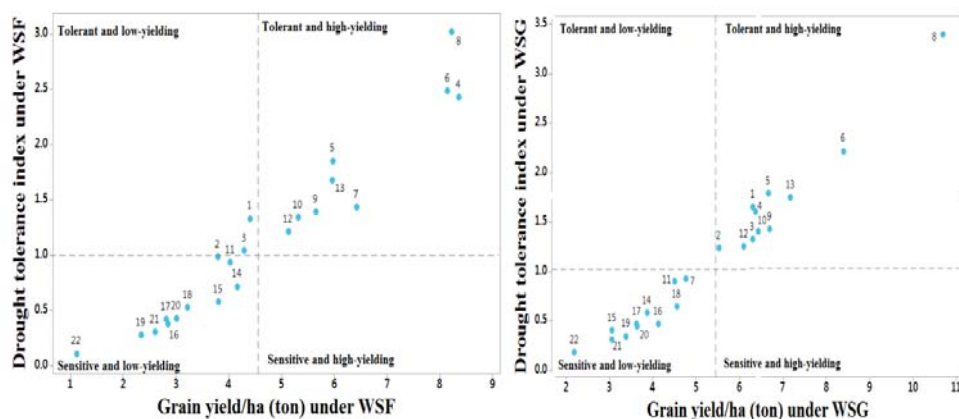
Environment	BW	BN	BA	BB	CN	CA	CB	CRL	RC	RDW
WW	-0.2	-0.07	-0.09	0.34**	0.23**	-0.14	0.27**	0.26**	0.43**	0.42**
WSF	-0.07	0.01	-0.2	0.13	0.27**	-0.03	0.08	-0.03	0.33**	0.13
WSG	-0.14	-0.12	-0.02	0.18*	0.21**	-0.08	0.25**	0.33**	0.17*	0.23**

\* and \*\* indicate significance at 0.05 and 0.01 probability levels, respectively. GYPP = grain yield per plant, BW= Number of above-ground whorls occupied with brace roots, BN= Number of brace roots, BA= Angle of 1st arm of the brace roots originating from whorl 1, 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.

## Grouping genotypes

### Based on drought tolerance and grain yield

Mean grain yield/fed of studied genotypes under water stress at flowering (WSF) and grain filling (WSG), was plotted against drought tolerance index of the same genotypes under WSF and WSG; respectively (Fig. 2), which made it possible to distinguish between four groups, namely tolerant and high- yielding, tolerant and low-yielding, sensitive and high-yielding and sensitive and low-yielding according to Sattelmacher *et al.*, 1994 [25], Worku *et al.* (2007) [26] and Al-Naggar *et al.* (2015) [27].



**Fig. 2. Relationships between drought tolerance index (DTI) and means of GYPH of genotypes (from No.1 to No.22) under water stress at flowering (WSF) and grain filling (WSG) combined across seasons. Broken lines represent mean grain yield/fed and DTI.**

Under water stress at flowering (WSF), the genotypes No 8 followed by No. 4, 6, 5, 7, 13, 9, 10 and 12 were classified as the drought tolerant and high yielding genotypes, *i.e.* they could be considered as the most water stress tolerant and the most responsive genotypes to water stress at flowering in this study (Fig. 2). There was no genotype belonging to the group of sensitive and high yielding genotypes under WSF. The genotypes No. 1 and 3 occupied the group of tolerant and low yielding under WSF. The genotypes No 22, 19, 21, 16, 17, 20, 18, 15, 14, 11 and 2 were classified as water stress sensitive and low yielding and therefore could be considered sensitive and low yielding.

Under water stress at grain filling (WSG), the genotypes No. 8 followed by 6, 13, 5, 1, 4, 9, 10, 3, 12 and 2 were classified as drought tolerant and high yielding, they could be considered as the most water stress tolerant and the most responsive genotypes to water stress at grain filling in this study (Fig. 3). On the contrary, genotypes No. 22, 21, 15, 19, 20, 17, 16, 14, 18, 11 and 7 were classified as water stress sensitive and low yielding (Fig. 2).

According to Fageria and Baligar (1994 and 1997a and b) [28-30] genotypes belonging to the 1<sup>st</sup> group "tolerant and high yielding" (above all) and 2<sup>nd</sup> group "tolerant and low yielding" (to a lesser extent) (we did not have) appear to be the most desirable materials for breeding programs that deal with adaptation to water stress. It was observed that the genotypes No. 8, 6, 4, 13, 5, 9, 10 and 12 occupied the first group (E-R) under both WSF and WSG conditions; they had genes of high water efficiency; *i.e.* drought tolerance to both WSF and WSG stages and genes for high yield under well watering conditions.

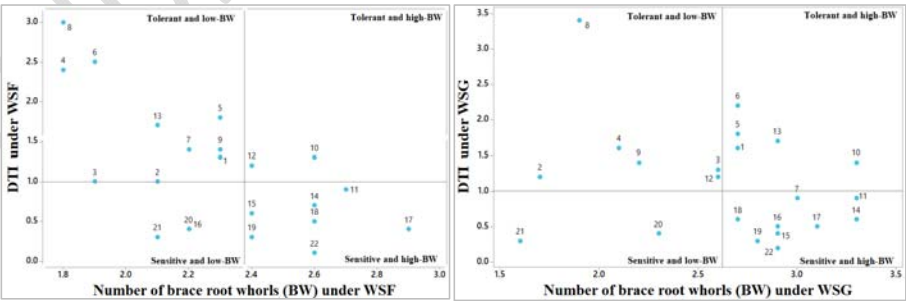
Summarizing the above-mentioned classifications, it is apparent that the genotypes No. 8 (P-3444) followed by 6 (SC-128), 4 (Egaseed-77), 5 (SC-10), 13 (TWC-324), 7 (Hi Tec-2066), 9 (SC-166), 10 (P-32D99) and 12 (Watania 11) were the best genotypes that occupied the first group (best one) in both classifications; they are the most efficient, most drought tolerant, the highest yielder under WSF as well as WW. The genotypes No. 8 (P-3444) followed by 6 (SC-128), 13 (TWC-324), 5 (SC-10), 1 (Hi Tec-2031), 4 (Egaseed-77), 9 (SC-166), 10 (P-32D99), 3 (Fine 1005), 12 (Watania 11) and 2 (P-30K09) were the best genotypes that occupied the first

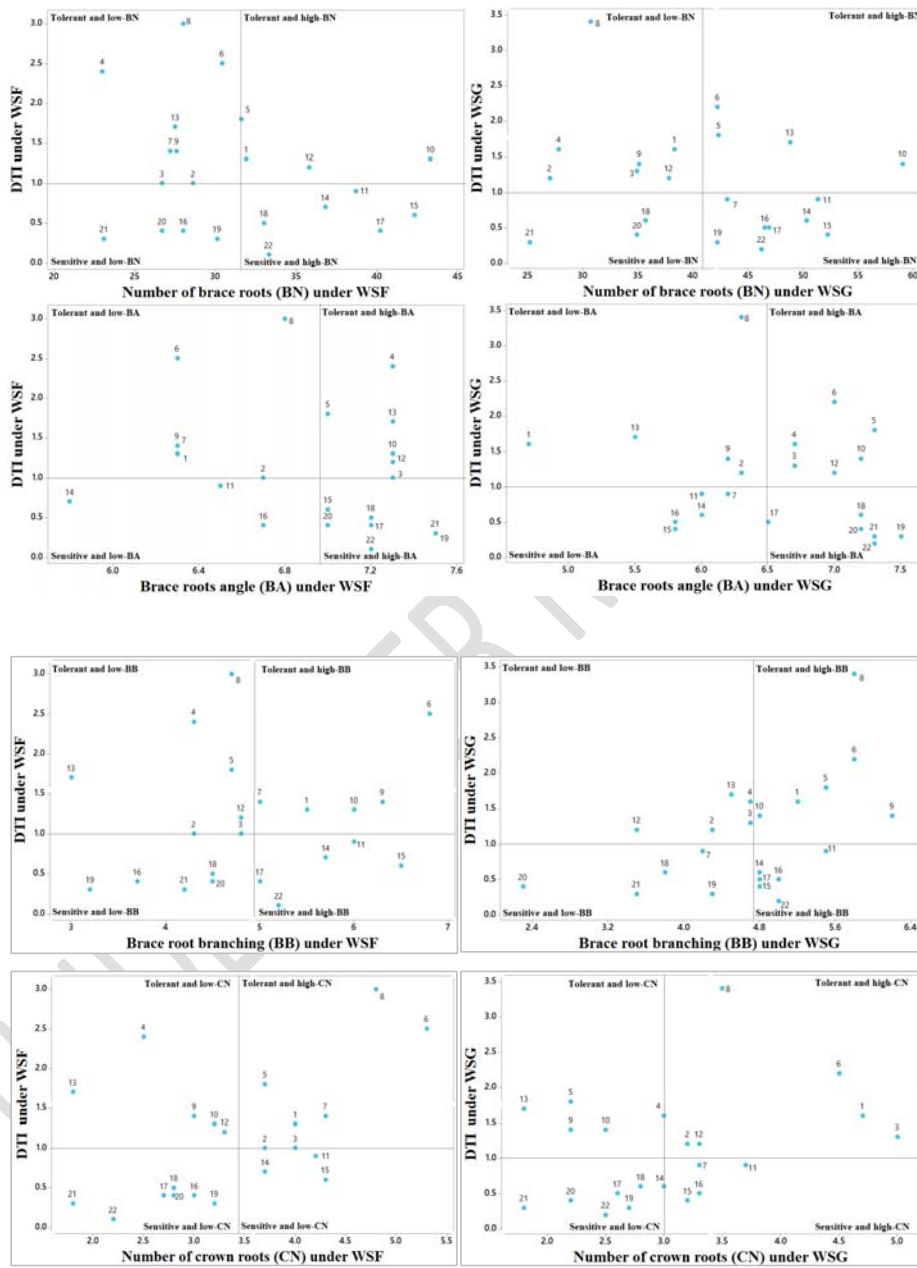
group (best one) in both classifications; they are the most efficient, most drought tolerant, the highest yielder under WSG as well as WW.

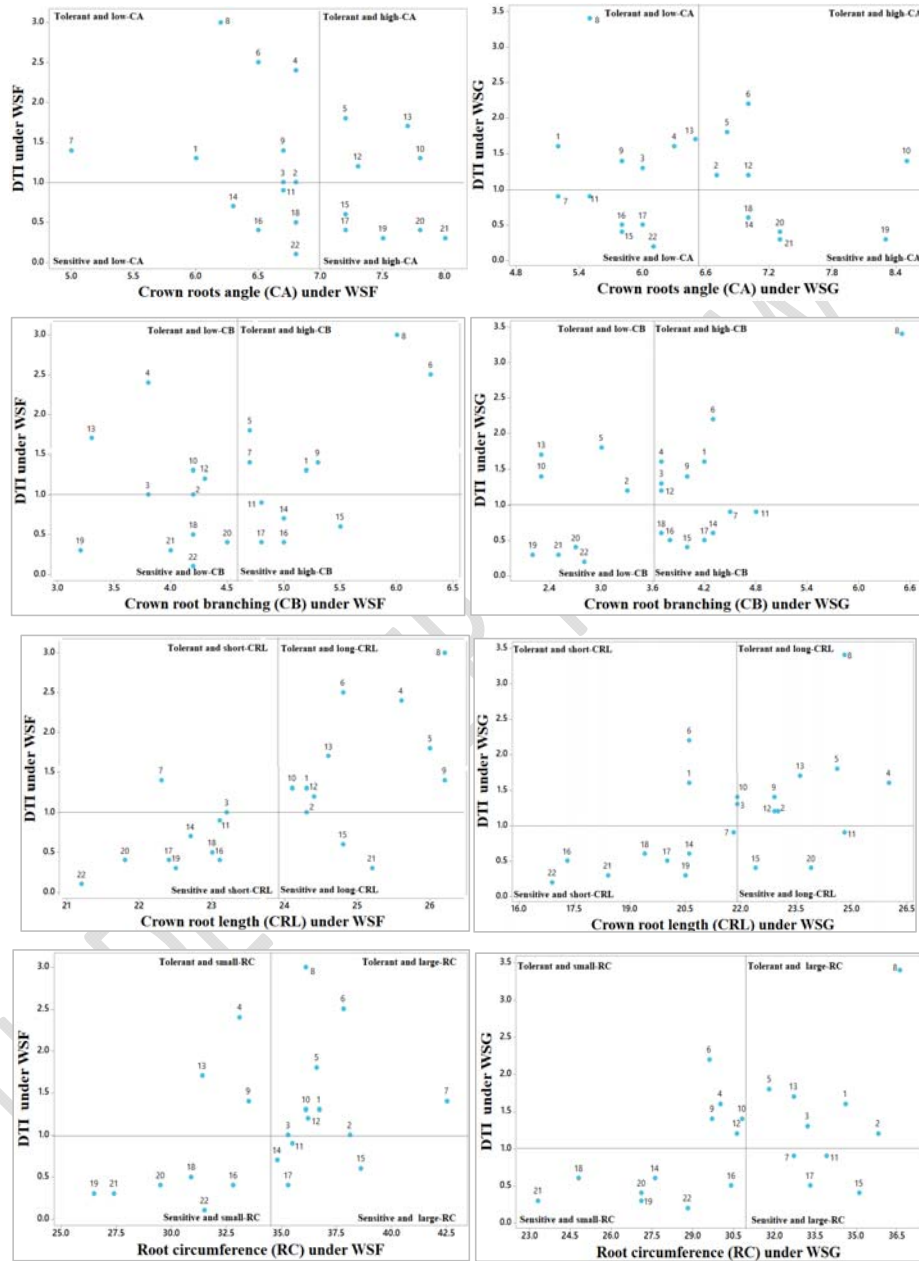
It was observed that the genotypes No 8 (P-3444) followed by 6 (SC-128), 4 (Egaseed-77), 5 (SC-10), 13 (TWC-324), 7 (Hi Tec-2066), 9 (SC-166), 10 (P-32D99) and 12 (Watania 11) were the best in the first group for both stresses WSF and WSG; they are the most efficient, most drought tolerant and the highest yielders under WSF and WSG as well as WW. In accordance to these results, a previous study by Al-Naggar *et al.* (2011) [17], proved that the single cross hybrid SC-128 (genotype No. 6 in the present study) was the most water efficient (drought tolerant) under WSF and the most responsive to WW based on grain yield, ears/plant, kernels/plant, ASI and leaf senescence.

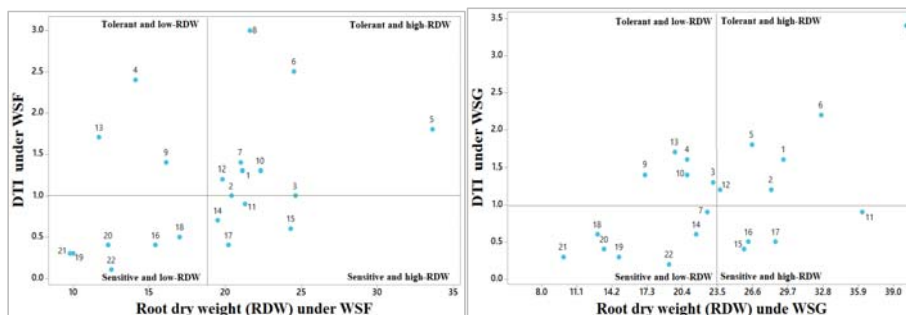
**Based on drought tolerance and root traits**

Means of root traits of studied genotypes under water stress at flowering (WSF) and grain filling (WSG), were plotted against drought tolerance index (DTI) of the same genotypes under WSF and WSG; respectively (Fig. 3), which made it possible to distinguish between four groups, namely tolerant and high value of root trait, tolerant and low value of root trait, sensitive and high value of root trait and sensitive and low value of root trait. According to Fageria and Baligar [29] (1997a), genotypes belonging to the 1<sup>st</sup> group "tolerant and high value of root trait" (above all) appear to be the most desirable materials for breeding programs.









**Fig. 3. Relationships between drought tolerance index (DTI) and means of number of whorls carrying brace roots, brace root branching, crown root number, crown root branching, root circumference, crown root length, and root dry weight, of genotypes (from No. 1 to No.22) under water stress at flowering (WSF) and grain filling (WSG) combined across seasons. Broken lines represent mean DTI and root trait.**

Figure (3) indicates that the 1<sup>st</sup> group "tolerant and high value of root trait" included the genotypes No. 10 and 12 under WSF, No. 10, 13, 6, 5 and 1 under WSG for number of whorls carrying brace roots, No. 10, 12, 1 and 5 under WSF, No. 10, 13, 1, 5 and 6 under WSG for number of brace roots, No. 4, 13, 10, 12, 3 and 5 under WSF, No. 5, 6, 10, 12, 4 and 3 under WSG for brace root angle, No. 6, 9, 10, 1 and 7 under WSF, No. 9, 6, 5, 1 and 10 under WSG for brace root branching, No. 6, 8, 7, 1, 5, 3 and 2 under WSF, No. 3, 1, 6, 8, 12 and 2 under WSG for number of crown roots, No. 10, 13, 12 and 5 under WSF, No. 10, 6, 12, 5 and 2 under WSG for crown root angle, No. 6, 8, 9, 1, 7 and 5 under WSF, No. 8, 6, 1, 9, 4, 3 and 12 under WSG for crown root branching, No. 8, 4, 5, 6, 9, 12, 1, 10 and 2 under WSF, No. 8, 4, 5, 13, 9, 2, 3, 10 and 12 under WSG for crown root length, No. 7, 6, 8, 5, 1, 10, 12, 2 and 3 under WSF, No. 8, 2, 1, 3, 13 and 5 under WSG for root circumference and No. 5, 6, 8, 10, 7, 1, 12, 3 and 2 under WSF, No. 8, 6, 1, 5, 2 and 12 under WSG for root dry weight.

#### **Mechanisms of drought tolerance of the most tolerant and high-yielding genotypes:**

The above-mentioned results (Figs. 2 and 3) helped us to identify the root traits that characterize the most drought tolerant and high-yielding genotypes, in descending order, as follows:

- 1. Genotype No. 8 (SC-P-3444):** Five traits (high CN, CB, large RC, long CRL and heavy RDW) under both WSF and WSG.
- 2. Genotype No. 6 (SC-128):** Four traits (high CN, CB, BB, large RC and heavy RDW) under both WSF and WSG.



- 510 **3. Genotype No. 4 (SC-Egaseed-77):** Two traits (steep brace root; i.e. large BA and long CRL)  
511 under both WSF and WSG.
- 512 **4. Genotype No. 5 (SC-10):** Six traits (high CN, CB, BA,RC, long CRL and heavy RDW)  
513 under WSF and five traits (high BA, CA, large RC, long CRL and heavy RDW) under WSG.
- 514 **5. Genotype No. 13 (TWC-324):** Two traits (steep brace root; i.e. large BA and long crown  
515 root (CRL) under WSF and two traits (large RC and long CRL) under WSG.
- 516 **6. Genotype No. 9 (SC-166):** Two traits (high CB and long crown root CRL) under both WSF  
517 and WSG.
- 518 **7. Genotype No. 10 (SC-P-32D99):** Four traits (steep crown root; CA steep brace root; BA,  
519 long crown root; CRL and heavy root dry weight; RDW) under both WSF and WSG and one  
520 trait (heavy RDW) under WSF.
- 521 **8. Genotype No. 12 (Watania TWC-11):** Seven traits (BW, BN, BA, CA, CRL, RC and  
522 RDW) under WSF and six traits (BA, CN, CA, CB, CRL and RDW) under WSG.

523 The present study suggested that further investigation should be conducted to determine the  
524 underlying root mechanisms contributing to the selection of water-efficient hybrids of maize.

525 In a recent study [31] (Shao et al., 2019), maize genotypes with less variation in root  
526 size, medium root size, medium broad root system and more inter-row root distribution help to  
527 reduce root-to-root competition and tend to have higher yield at high planting density.

## 528 CONCLUSIONS

529 Correlation analysis of the present study concluded that drought tolerance in maize had a  
530 strong and positive association with crown root length, root circumference and root dry weight  
531 under both WSF and WSG, a negative correlation with brace root whorls, and a positive  
532 correlation with crown root number under WSF and brace root branching and crown root  
533 branching under WSG. These root traits could be considered as putative mechanisms of drought  
534 tolerance. The present study suggested that further investigation should be conducted to  
535 determine the underlying plant mechanisms contributing to the selection of water-efficient  
536 hybrids of maize. The cultivars Pioneer-3444, SC-128, Egaseed-77, SC-10 and TWC-324  
537 showed the most drought tolerance and the highest yielding in a descending order; each had a  
538 number of such drought tolerance mechanisms. These cultivars should be retested for drought  
539 tolerance and grain productivity under drought stress and could be offered to plant breeding  
540 programs for improving tolerance to drought and high grain yield.

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