



## Differential Responses of Maize (*Zea mays* L.) Genotypes to Elevated Plant Density Combined with Deficit Irrigation

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### Authors' contributions

This work was carried out in collaboration between all authors. Author AMMAN designed the study, wrote the protocol and wrote the first draft of the manuscript. Authors MMMA and MAA managed the literature searches. Author ASMY managed the experimental process and performed data analysis. All authors read and approved the final manuscript.

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### ABSTRACT

High plant density and full irrigation along with the use of high density-tolerant genotype would lead to maximizing maize (*Zea mays* L.) grain productivity per unit land area. The objective of this investigation was to match the functions of optimum plant density and adequate irrigation with the greatest maize genotype efficiency to produce the highest possible yields per unit area. Six maize inbred lines differing in tolerance to water stress and high density (D) [three tolerant (T); L-20, L-53, Sk-5, and three sensitive (S); L-18, L-28, Sd-7] were chosen for diallel crosses. Parents and crosses were evaluated in the 2013 and 2014 seasons under three plant densities: low (47,600), medium (71,400), and high (95,200) plants ha<sup>-1</sup> and two irrigation regimes: water stress (at flowering stage) and non-stress (well watering). The T × T crosses were superior to the S × S and T × S crosses under the water stress–high D environment in most studied traits across seasons. The relationships between the six environments and grain yield per hectare (GYPH) showed near-linear regression functions for the tolerant high yielding group of hybrids with the optimum environment combination was well watering combined with high plant density (95,200 plants ha<sup>-1</sup>)

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and a curvilinear relationship for the sensitive low yielding group with the highest GYPH at a density of 71,400 plants ha<sup>-1</sup> combined with well irrigation. Cross L20 × L53 gave the highest grain yield in this study under both well watering– high-D (17.05 t ha<sup>-1</sup>) and well watering–medium-D environment (16.45 t ha<sup>-1</sup>).

*Keywords: Quadratic regression; high density tolerance; drought tolerance; flowering stage.*

## 1. INTRODUCTION

One of the potential methods to maximize total production of maize (*Zea mays* L.) in Egypt is through raising productivity per land unit area and thus upgrading our global rank in average productivity, especially with the irrigation system used in Egypt and good weather and soil conditions that suit maize crop as compared to other regions in the world. Grain yield per land unit area is the product of grain yield per plant and number of plants per unit area [1]. Maximum yield per unit area may be obtained by growing maize hybrids that can withstand elevated plant density up to 100,000 plants ha<sup>-1</sup> [2]. Average maize grain yield per unit area in the USA increased dramatically during the second half of the 20<sup>th</sup> century, due to improvement in crop management practices and greater tolerance of modern hybrids to high plant densities [3-5]. Growing commercial varieties of maize, released locally by the National Maize Breeding Program, at high plant density (HPD) and deficit irrigation causes a drastic reduction in grain yield per plant and grain yield per land unit area. The reason is probably due to the fact that these varieties are bred under low plant density and sufficient flood irrigation from River Nile, so they are not tolerant to both HPD and deficit irrigation; because of their tallness, one-eared, decumbent leaf and large-size type plants. On the contrary, modern maize hybrids in developed countries are characterized with high yielding ability from unit area under HPD, due to their high-density adaptive traits, such as early silking, short anthesis silking interval (ASI), less barren stalks and prolificacy [6]. Radenovic et al. [7] pointed out that maize genotypes with erect leaves are very desirable for increasing the population density due to better light interception.

The expected future shortage in irrigation water in Egypt necessitates that maize breeders should pay great attention to develop drought tolerant maize cultivars that could give high grain yield under both water-stress and non-stress conditions. Maize is particularly susceptible to

drought at the flowering stage [8]. Loss in grain yield is particularly severe when drought stress occurs at this stage [9-11]. Water stress at flowering, when silk growth, pollination, and kernel set occur, slows ear growth, and consequently silk emergence, more than tassel growth or anthesis, resulting in a widening interval between anthesis and silking (ASI) [12]. Drought tolerant genotypes of maize were characterized by having shorter anthesis-silking interval (ASI) [13] and more ears/plant [14].

The presence of genotypic differences in HPD and drought tolerance would help plant breeders in initiating successful breeding programs to improve such complicated characters. Differential responses of maize genotypes to high plant density combined with other abiotic stresses was reported by some investigators (e.g. [15-20]). The objectives of the present investigation were: (i) to evaluate the effects of stresses resulting from high plant density combined with deficit irrigation on traits of six inbreds and their diallel crosses, and (ii) to match the functions of appropriate plant density and adequate irrigation regime with greatest maize inbred or hybrid efficiency to produce the highest possible yields per unit area.

## 2. MATERIALS AND METHODS

This study was carried out 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), in 2012, 2013 and 2014 seasons.

### 2.1 Plant Materials

Based on the results of previous experiments [21], six maize (*Zea mays* L.) inbred lines in the 8<sup>th</sup> selfed generation (S<sub>8</sub>), showing clear differences in performance and general combining ability for grain yield/feddan(fed) under high plant density, were chosen in this study to be used as parents of diallel crosses (Table 1).

## 2.2 Making F<sub>1</sub> Diallel Crosses

In 2012 season, all possible diallel crosses (except reciprocals) were made among the six parents, so seeds of 15 direct F<sub>1</sub> crosses were obtained. Seeds of the 6 parents were also increased by selfing in the same season (2012) to obtain enough seeds of the inbreds in the 9<sup>th</sup> selfed generation (S<sub>9</sub> seed).

## 2.3 Evaluation of Parents and F<sub>1</sub>'s

Field evaluation experiments were carried out at the Agricultural Experiment and Research Station of Faculty of Agriculture, Cairo University, Giza, Egypt in 2013 and 2014 seasons. Each experiment included 15 F<sub>1</sub> crosses, their 6 parents and 2 check cultivars, *i.e.* SC 130(white), obtained from the Agricultural Research Center (ARC) and SC 2055(yellow) obtained from Hi-Tech Company-Egypt. Evaluation in each season was carried out under two water regimes (well watering; WW and water stress; WS at flowering stage by skipping the 4<sup>th</sup> and 5<sup>th</sup> irrigations) and three plant densities, (47,600, 71,400 and 95,200 plants/ha, representing low-, medium- and high-plant density, respectively).

A split-split plot design in randomized complete blocks (RCB) arrangement with three replications was used. Main plots were devoted to water stress (well watering and water stress). Sub-plots were assigned to plant density (D) (low-D, medium-D and high-D). Sub sub-plots were devoted to 23 maize genotypes (6 parents, 15 F<sub>1</sub>'s and 2 checks). Each sub sub-plot consisted of one ridge of 4 m long and 0.7 m width, *i.e.* the experimental plot area was 2.8 m<sup>2</sup>. Seeds were

sown in hills at 15, 20 and 30 cm apart, thereafter (before the 1<sup>st</sup> irrigation) were thinned to one plant/hill to achieve the 3 plant densities, *i.e.* 95,200, 71,400 and 47,600 plants/ha, respectively. Each main plot was surrounded with a wide alley (4 m width) to avoid interference of the two water treatments with irrigation water. Sowing date each season was on May 5 and May 8 in 2013 and 2014 seasons, respectively. The soil analysis of the experimental soil at the experimental site, as an average of the two growing seasons 2013 and 2014, indicated that the soil is clay loam (4.00% coarse sand, 30.90% fine sand, 31.20% silt, and 33.90% clay), the pH (paste extract) is 7.73, the EC is 1.91 dSm<sup>-1</sup>, soil bulk density is 1.2 g cm<sup>-3</sup>, calcium carbonate is 3.47%, organic matter is 2.09%, the available nutrient in mg kg<sup>-1</sup> are Nitrogen (34.20), Phosphorous (8.86), Potassium (242), hot water extractable B (0.49), DTPA - extractable Zn (0.52), DTPA - extractable Mn (0.75) and DTPA - extractable Fe (3.17). Meteorological variables in the 2013 and 2014 growing seasons of maize were obtained from Agro-meteorological Station at Giza, Egypt. For May, June, July and August, mean temperature was 27.87, 29.49, 28.47 and 30.33°C, maximum temperature was 35.7, 35.97, 34.93 and 37.07°C and relative humidity was 47.0, 53.0, 60.33 and 60.67% respectively, in 2013 season. In 2014 season, mean temperature was 26.1, 28.5, 29.1 and 29.9°C, maximum temperature was 38.8, 35.2, 35.6 and 36.4°C and relative humidity was 32.8, 35.2, 35.6 and 36.4%, respectively. Precipitation was nil in all months of maize growing season for both seasons. All other agricultural practices were followed according to the recommendations of ARC, Egypt.

**Table 1. Designation, origin and most important traits of 6 inbred lines (L) used for making diallel crosses of this study**

Entry designation	Origin	Institution (country)	Prolificacy	Productivity under high density	Leaf angle
L20-Y	SC 30N11	Pion. Int.Co.	Prolific	High	Erect
L53-W	SC 30K8	Pion. Int.Co.	Prolific	High	Erect
Sk5-W	Tepalcinco # 5	ARC-Egypt	Prolific	High	Erect
L18-Y	SC 30N11	Pion. Int.Co.	Prolific	Low	Wide
L28-Y	Pop 59	ARC-Thailand	Non-Prolific	Low	Wide
Sd7-W	A.E.D.	ARC-Egypt	Non-Prolific	Low	Erect

ARC = Agricultural Research Center, Pion. Int. Co. = Pioneer International Company in Egypt, SC = Single cross, A.E.D.= American Early Dent (Old local OPV), W = White grains and Y = Yellow grains.

## 2.4 Data Recorded

1. Days to 50% anthesis (DTA) (as number of days from planting to anthesis of 50% of plants per plot). 2. Days to 50% silking (DTS) (as number of days from planting to silking of 50% of plants/plot). 3. Anthesis-silking interval (ASI) (as number of days between 50% silking and 50% anthesis of plants per plot). 4. Plant height (PH) (cm) (measured from ground surface to the point of flag leaf insertion for five plants per plots). 5. Ear height (EH) (cm) measured from ground surface to the base of the top most ear relative to the plant height for five plants per plots. 6. Barren stalks (BS) (%) measured as percentage of plants bearing no ears relative to the total number of plants in the plot (an ear was considered fertile if it had one or more grains on the rachis). 7. Leaf angle (LANG) (o) measured as the angle between stem and blade of the leaf just above ear leaf according to Zadoks et al. [22]. The following grain yield traits were measured at harvest. 8. Number of ears per plant (EPP) calculated by dividing number of ears per plot on number of plants per plot. 9. Number of rows per ear (RPE) using 10 random ears/plot at harvest. 10. Number of kernels per row (KPR) using the same 10 random ears/plot. 11. Number of kernels per plant (KPP) calculated as: number of ears per plant  $\times$  number of rows per ear  $\times$  number of kernels per row. 12. 100-kernel weight (100-KW) (g) adjusted at 15.5% grain moisture, using shelled grains of each plot. 13. Grain yield per plant (GYPP) (g) estimated by dividing the grain yield per plot (adjusted at 15.5% grain moisture) on number of plants/plot at harvest. 14. Grain yield per hectare (GYPH) (ton), by adjusting grain yield/plot to grain yield per hectare. Stress tolerance index (STI): Stress tolerance index (STI) modified from equation suggested by Fageria [23] was used to classify genotypes for tolerance to stress (water stress and/or high density stress). The formula used is as follows:  $STI = (Y1/AY1) \times (Y2/AY2)$  Where,  $Y1$  = grain yield mean of a genotype at non-stress.  $AY1$  = average yield of all genotypes at non-stress  $Y2$  = grain yield mean of a genotype at stress.  $AY2$  = average yield of all genotypes at stress. When STI is  $\geq 1.0$ , it indicates that genotype is tolerant (T), If STI is  $< 1$ , it indicates that genotype is sensitive (S).

## 2.5 Biometrical Analyses

Combined analysis of variance of the split-split plot design in RCB arrangement on the basis of individual plot observation and combined

analysis of variance of RCBD for each of the six environments (WW-LD, WW-MD, WW-HD, WS-LD, WS-MD and WS-HD) across the two seasons were performed if the homogeneity test was non-significant using the MIXED procedure of SAS [24]. Least significant differences (LSD) were calculated according to Steel et al. [25].

## 3. RESULTS AND DISCUSSION

### 3.1 Analysis of Variance

Combined analysis of variance across years (Y) of the split-split plot design for the studied 23 genotypes (G) of maize (6 inbreds +15  $F_1$ 's + 2 check cultivars) under three plant densities (D) and two irrigation (I) regimes is presented in Table 2. Mean squares due to years were significant or highly significant for all studied 14 traits, except for anthesis-silking interval (ASI), barren stalks (BS), kernels/plant (KPP) and grain yield/ha (GYPH), indicating significant effect of climatic conditions on most studied traits. Mean squares due to irrigation regimes, plant densities and genotypes were significant or highly significant for all studied traits, except ASI, leaf angle (LANG) and rows/ear (RPE) for irrigation regimes, and ASI, RPE, indicating that plant density or irrigation regime had a significant effect on most studied traits and that genotype has an obvious and significant effect on all studied traits.

Mean squares due to the 1<sup>st</sup> order interaction, i.e.  $I \times Y$ ,  $D \times Y$ ,  $D \times I$ ,  $G \times Y$ ,  $G \times I$  and  $G \times D$  were significant ( $P \leq 0.05$  or  $0.01$ ) for all studied traits, except for 9 traits for  $I \times Y$ , 8 traits for  $D \times Y$ , 6 traits for  $D \times I$  and one trait (RPE) for  $G \times I$  and  $G \times D$  (Table 2). Mean squares due to the 2<sup>nd</sup> order interaction, i.e.  $G \times I \times Y$ ,  $G \times D \times Y$  and  $G \times D \times I$  were significant or highly significant for all studied traits, except RPE. However, mean squares due to  $D \times I \times Y$  and were insignificant for 4 traits, i.e. ASI, EPP, RPE and GYPH.

Mean squares due to the 3<sup>rd</sup> order interaction  $G \times I \times D \times Y$  were significant ( $P \leq 0.01$  or  $0.05$ ) for all studied traits, except for RPE trait only, indicating that the rank of maize genotypes differ from irrigation regime to another, from one density to another and from one year to another and the possibility of selection for improved performance under a specific combination between plant density and irrigation regime as proposed by several investigators [26,27] under a specific irrigation regime and [16-19,28-33] under a specific plant density.

**Table 2. Combined analysis of variance (% sum of squares) of split-split plot design for studied 23 maize genotypes under two irrigation regimes (I) and three plant densities (D) across 2013 and 2014 years**

SOV	df	% Sum of squares (SS)						
		DTA	DTS	ASI	PH	EH	BS%	LANG
Years (Y)	1	14.94**	10.76**	0.06	0.15**	0.80**	0.49	0.58**
Irrigation (I)	1	11.38**	13.05**	6.27**	0.77**	0.42**	4.63**	7.55**
IxY	1	1.61**	0.83**	0.47	0.04	0.001	0.268	0.64**
Error	8	0.09	0.14	0.98	0.07	0.08	2.06	0.35
Densities (D)	2	14.47**	27.85**	51.79**	4.65**	8.17**	4.01**	0.85**
DxY	2	0.35**	0.25**	0.04	0.01	0.001	0.13	0.16**
DxI	2	0.43**	0.38**	0.03	0.43**	0.08	0.03	0.001
DxIxY	2	0.25**	0.12**	0.08	0.001	0.13**	0.15	0.20**
Error	16	0.13	0.13	1.14	0.07	0.19	1.78	0.23
Genotypes (G)	22	26.09**	21.48**	2.34**	81.54**	77.35**	2.89	54.23**
GxY	22	8.04**	5.61**	2.03**	0.50**	1.70**	4.34**	7.58**
GxI	22	4.45**	3.78**	3.02**	2.27**	0.96**	3.72**	1.31**
GxIxY	44	2.49**	2.21**	3.13**	1.91**	2.24**	7.88**	5.01**
GxD	22	4.61**	3.57**	1.02	0.75**	0.74**	2.5	2.04**
GxDxY	44	2.94**	2.36**	3.07**	0.33	0.55**	5.68*	3.90**
GxDxI	44	2.22**	1.75**	2.1	1.72**	2.26**	5.24	4.09**
GxIxDxY	44	2.22**	1.92**	1.94	0.32	0.90**	6.00**	3.21**
Error	528	3.3	3.79	20.49	4.49	3.43	48.2	8.08
Total SS	827	9536	12716	970.3	691434	275820	27121	17829
CV%		1.23	1.44	17.36	3.29	4.14	39.48	5.81

DTA= Days to 50% anthesis, DTS = days to 50% silking, ASI = anthesis-silking interval, PH = plant height, EH = ear height, BS = barren stalks. LANG = leaf angle and \* and \*\* indicate significance at 0.05 and 0.01 probability levels, respectively

**Table 2. (Continued)**

SOV	df	% Sum of squares (SS)						
		EPP	RPE	KPR	KPP	100-KW	GYPP	GYPH
Years (Y)	1	4.46**	0.60**	0.71**	0.24*	14.50**	0.18**	0.14**
Irrigation (I)	1	5.42**	5.64**	4.63**	9.32**	8.90**	8.94**	8.74**
IxY	1	0.47	0.066	0.001	0.05	4.453**	0.0076	0.0080*
Error	8	1.06	0.37	0.08	0.3	0.37	0.02	0.01
Densities (D)	2	30.24**	9.52**	5.70**	25.36**	13.46**	9.54**	7.04**
DxY	2	0.39**	0.01	0.02*	0.01	0.28**	0.001	0.01
DxI	2	1.01**	0.06	0.14**	0.16**	0.001	0.21**	0.27**
DxIxY	2	0.23**	0.15**	0.01	0.05*	0.029	0.04*	0.02
Error	16	0.3	0.15	0.04	0.12	0.09	0.08	0.06
Genotypes (G)	22	6.91**	53.32**	80.84**	48.44**	40.40**	73.56**	75.45**
GxY	22	4.59**	2.62**	1.65**	2.16**	3.70**	0.18**	0.19**
GxI	22	5.25**	3.01**	0.94**	0.97**	2.73**	2.03**	2.15**
GxIxY	44	3.91**	0.97	0.67**	1.15**	1.01**	2.76**	3.21**
GxD	22	4.47**	2.22**	0.31**	1.32**	2.72**	0.17**	0.14**
GxDxY	44	2.1	1.11	0.29	0.64	0.68**	0.26**	0.24**
GxDxI	44	3.84**	0.73	0.37	1.11**	1.05**	0.84**	1.06**
GxIxDxY	44	2.74*	0.64	0.38*	0.72	0.57	0.20**	0.17**
Error	528	22.62	18.81	3.22	7.89	5.07	0.98	1.09
Total SS	827	18.078	1460.019	50494.38	2533264	20245.32	3646883	142949.9
CV%		7.96	5.25	4.48	10.20	4.55	6.04	6.33

EPP = number of ears per plant, RPE = Number of rows per ear, KPR = Number of kernel per row, KPP = number of kernels per plant, 100-KW = 100-kernel weight, GYPP = grain yield per plant, GYPH = grain yield/ha, \* and \*\* indicate significance at 0.05 and 0.01 probability levels, respectively

It is observed from Table 2 that variance due to genotypes was the largest contributor to the total variance in this experiment for 11 out of 14 studied traits, as measured by percentage of sum of squares to total sum of squares. For the three traits ASI, BS and EPP, error variance was the largest contributor to the total variance; the reason might be due to the large value of C.V. for these characters (20.67, 23.19 and 20.13%, respectively). Comparing irrigation with density effect, it is clear from Table 2 that irrigation variance showed larger contribution to total variance than density variance for 7 traits (DTA, DTS, ASI, BS, RPE, 100KW and GYPH), indicating that water stress had more effect than elevated plant density on such traits, while density showed larger contribution to total variance than irrigation variance for the rest of studied traits (PH, EH, LANG, EPP, KPR, KPP and GYPP), indicating that high plant density had more effect than water stress on latter traits.

Combined analysis of variance of a randomized complete blocks design for studied traits and maize genotypes was performed across two seasons under each of the six environments (from E1 to E6); representing combinations of 3 plant densities  $\times$  2 irrigation regimes, i.e. E1 = well watering and low density, E2 = well watering and medium plant density, E3 = well watering and high plant density, E4 = water stress and low plant density, E5 = water stress and medium plant density and E6 = water stress and high plant density. Mean squares due to genotypes, parents and crosses under all environments were highly significant for all studied traits, except ASI under E1, E3 and E5 and EPP under E6, indicating the significance of differences among studied parents and among  $F_1$  diallel crosses in the majority of cases. Mean squares due to parents vs.  $F_1$  crosses were highly significant for all studied traits under all six environments, except for ASI under E1, E3 through E6, EPP under E3 and BS under E1, suggesting the presence of significant heterosis for most studied cases. Mean squares due to the interactions parents  $\times$  years ( $P \times Y$ ) and crosses  $\times$  years ( $F_1 \times Y$ ) were significant or highly significant for all studied traits under all environments, except DTS under E1 and E2 for  $F_1 \times Y$ , DTS under E5 for  $P \times Y$  and E1 for  $F_1 \times Y$ , ASI under E1, E3 and E5 for  $P \times Y$  and E3 for  $F_1 \times Y$ , BH under E1, E3, E4 and E5 for  $P \times Y$  and E2 through E6 for  $F_1 \times Y$ , EH under E1 through E4 for  $P \times Y$ , BS under E2, E5 and E6 for  $P \times Y$  and E3 and E6 for  $F_1 \times Y$ , EPP under E1, E2, E5 and E6 for  $P \times Y$  and E3 for  $F_1 \times Y$ , RPE under E2, E3, E5 and E6 for  $P \times Y$

and E2, E4 through E6 for  $F_1 \times Y$ , KPP under E1, E2, E3, E5 and E6 for  $P \times Y$  and E3 and E6 for  $F_1 \times Y$ , KPR under E3 and E6 for  $P \times Y$  and E6 for  $F_1 \times Y$ , 100KW under E3 and E4 for  $P \times Y$ , GYPP under E6 for  $P \times Y$ , GYPH under E1 for  $P \times Y$  and  $F_1 \times Y$ . Mean squares due to parents vs. crosses  $\times$  years were significant or highly significant in most studied cases. Such interaction was expressed in most environments for DTS, BS, LANG, EPP, KPR, KPP, 100KW and GYPH traits. This indicates that heterosis differ from season to season in these cases. The environment E6 (the most stressed environment) showed such interaction for all studied traits, except ASI, RPE and GYPP. Among genotypes components under all six environments, the largest contributor to total variance was parents vs.  $F_1$ 's (heterosis) variance, but the lowest contributor was parents.

### 3.2 Effects of Combinations of Irrigation Regimes and Plant Densities

The effects (relative change to non-stressed E1 environment) of stressed environments from E2 to E6 on the means of studied traits across all genotypes and across two years are presented in Table 3. The E1 represents the unstressed environment (well watered and low plant density) and will be used hereafter as control environment, E2 represents medium density stress only, E3 represents high density stress only, E4 represents water stress only, E5 represents water stress combined with medium density stress and E6 represents water stress combined with high density stress.

Both stresses combined together (water stress and plant density stress) were exhibited by E5 and E6 environments, with a maximum severity by E6 (water stress combined with high plant density), while E5 environment combined medium plant density with water stress. Comparing these two environments (E5 and E2) with the control non-stressed environment (E1) expressed in means and changes should give a picture of the effects of the two stresses combined together on different studied traits (Table 3). It can be observed that the rigidity of the stress combinations on GYPP was at maximum (49.71% reduction) under the environment E6 (WS-HD), where both severe stresses (highest plant density and deficit irrigation) existed. The reduction in GYPP due to the effect of water stress (WS) combined with medium plant density (MD) stress (E5) was 37.90%. Significant reductions in GYPP of maize

genotypes observed in environments E5 and E6 relative to E1 were due to both drought and high density stresses. On the contrary, GYPH under the environment E6 showed a tendency of non-significant increase (1.03%) over that under E1.

LANG. Maximum increases appeared under E6 and by ASI and BS traits. Increases in such traits are unfavorable. The reason for EH and PH increase under E5 and E6 may be attributed to elevated levels of plant density.

Reductions in grain yield resulted from both stresses (elevated plant densities and water stress) were associated with reductions in all yield components (EPP, RPE, KPR, KPP and 100-KW). Such reductions were more pronounced in E6 environment (maximum stresses) followed by E5 and were exhibited by kernels/plant (29.22 and 42.60%) under E5 and E6, respectively. On the other hand, the two stresses together (shown by the two environments E5 and E6) caused increases in DTA, DTS, ASI, PH, EH, BS and

Elevated plant density results in interplant competition that affects vegetative and reproductive growth [34]. An increase in the number of maize plants per unit area will enhance the competition among plants for resources within the maize canopy [5]. High plant density increases barrenness and results in smaller ears and reduced harvest index [3, 35-38]. High plant density also causes increased plant and ear heights, fewer EPP and later anthesis, with silk emergence delayed more than pollen shed [35].

**Table 3. Means of studied traits in six environments combined across all studied genotypes and across 2013 and 2014 seasons**

Trait	Parameter	E1	E2	E3	E4	E5	E6
		WW-LD	WW-MD	WW-HD	WS-LD	WS-MD	WS-HD
DTA (day)	Mean	60.41	61.75	63.03	62.11	64.15	65.81
	Change %	—	-2.22**	-4.34**	-2.81**	-6.19**	-8.93**
DTS (day)	Mean	62.77	64.97	67.25	64.97	67.91	70.62
	Change %	—	-3.50**	-7.14**	-3.50**	-8.18**	-12.50**
ASI (day)	Mean	2.36	3.21	4.22	2.86	3.75	4.81
	Change %	—	-36.25**	-78.96**	-21.17**	-59.14**	-103.84**
PH (cm)	Mean	230.98	234.61	241.64	220.89	230.46	240.68
	Change %	—	-1.57**	-4.61**	4.37**	0.22	-4.20**
EH (cm)	Mean	97.63	103.23	109.13	94.02	100.83	108.07
	Change %	—	-5.75**	-11.79**	3.69**	-3.28**	-10.70**
BS (%)	Mean	10.00	11.31	12.81	12.62	13.51	15.38
	Change %	—	-13.07	-28.11**	-26.18**	-35.08**	-53.79**
LANG (o)	Mean	27.73	26.83	26.92	30.34	29.34	29.45
	Change %	—	3.27**	2.93**	-9.41**	-5.80**	-6.19**
EPP	Mean	1.23	1.12	1.07	1.20	1.06	0.96
	Change %	—	8.74**	13.06**	2.76**	14.12**	21.71**
RPE	Mean	14.53	14.03	13.58	13.92	13.47	12.86
	Change %	—	3.42**	6.55**	4.23**	7.28**	11.48**
KPR	Mean	42.85	40.65	38.92	39.93	37.68	34.73
	Change %	—	5.13**	9.18**	6.82**	12.07**	18.96**
KPP	Mean	765.14	639.19	565.73	668.97	541.58	439.17
	Change %	—	16.46**	26.06**	12.57**	29.22**	42.60**
100-KW (g)	Mean	34.31	32.06	29.92	31.42	29.08	26.93
	Change %	—	6.57**	12.79**	8.41**	15.24**	21.51**
GYPP (g)	Mean	186.26	150.46	130.42	138.70	115.67	93.67
	Change %	—	19.22**	29.98**	25.53**	37.90**	49.71**
GYPH(ton)	Mean	8.62	10.40	11.94	6.39	8.10	8.71
	Change %	—	-20.59**	-38.48**	25.91**	6.13**	-1.03

WW = well watering, WS = water stress, LD = low density, MD = medium density, HD = high density, E = environment, DTA = days to 50% anthesis, DTS = days to 50% silking, ASI = anthesis-silking interval, PH = plant height, EH = ear height, BS = barren stalks, LANG = leaf angle, EPP = ears per plant, RPE = rows per ear, KPR = kernel per row, KPP = kernels per plant, 100-KW = 100-kernel weight, GYPP = grain yield per plant, GYPH = grain yield per hectare\* and \*\* indicate significance at 0.05 and 0.01 probability levels, respectively

### 3.3 Genotype × Irrigation × Plant Density Interaction

Mean grain yield/ha across years under six combinations of 2 irrigation regimes and 3 plant densities (E1 through E6) for all inbreds, hybrids and the check cultivars (SC130 and SC 2055) is presented in Table 4.

The rank of inbred parents for GYPH was approximately similar in all six environments, indicating less effect of interaction between inbreds, irrigation and plant density on GYPH. The percent reduction in GYPH due to both stresses relative to E1 (WW-LD) was smaller for the inbred lines L20, L28 and L53 than the inbreds L18, Sk5 and Sd7 in low-performing ones, which could be attributed to the higher potential yield of the first group of lines than the second one, under good environmental conditions. The first group of lines was therefore considered tolerant to both stresses expressed in GYPH, while the second one was considered sensitive. The best GYPH was obtained from E3 (WW-HD) for the inbreds L20, Sk5 and L53 followed by E2 (WW-MD) and E1 (WW-LD).

Regarding GYPH of the  $F_1$  crosses, the rank varied from one environment (a combination of irrigation regime with plant density level) to another, especially when comparing environments that combine two stresses with those having only one stress or no stress, indicating that the GYPH of a cross differs from one combination to another. Comparing to the non-stressed environment (E1), all 15  $F_1$  crosses showed an increase in their GYPH ranging from -5.75 to -35.99% under E2 and from -32.42 to -58.0% under E3 and 7 and 9 crosses showed an increase in GYPH under E5 and E6, respectively over that under E1. The increase in GYPH of these crosses under E2, E3, E5 and E6 over that under E1 could be attributed to the elevation of plant density even under water stress conditions existed in E5 and E6 environments. This indicates that the increase of GYPH due to the increase in plant density could compensate the reduction in GYPH due to water stress at flowering stage and even this could happen in some crosses if they have more tolerance to water stress and high density stress.

The highest GYPH in this experiment was obtained under E3 (well irrigation- high density) and the best crosses in this environment were L20 × L53 (17.05 t/ha), L53 × Sk5 (16.47 t/ha), L53 × Sd7 (16.30 t/ha), L20 × L18 (16.04 t/ha)

and Sk5 × L28 (15.45 t/ha), with a significant superiority over SC 2055 (the best check under this environment) by 22.9, 18.27, 17.52, 15.62 and 11.36%, respectively. Some hybrids in this experiment showed significant superiority over the best check in the respective environment (four crosses under E6, 11 crosses under E5, 10 under E4, 11 under E3, 7 under E2 and 4 under E1). These superiorities reached 51.92% over SC130 under E5 for the cross L20 × L53 (the best cross in the whole experiment under all six environments). The latter cross (L20 × L53) out-yielded the best check under the most stressed environment in this experiment (E6) by 30.67% (3.51 t/ha superiority). The cross L53 × Sk5 occupied the second rank after L20 × L53 under all environments and showed significant superiority in GYPH over the best check (SC130) by 11.13% (1.30 t/ha). It is worthy to note that the three crosses (L20 × L53) (L53 × Sk5) and (L53 × Sd7) were considered the highest responsive and the most tolerant ones to both stresses (water stress combined with high density).

Differential responses of maize genotypes to high plant density combined with other abiotic stresses were reported by some investigators [15-19]. Although high plant density results in interplant competition (especially for light, water, and nutrients), which affects vegetative and reproductive growth of maize [5], the use of hybrids tolerant of high density and using well irrigation during the whole plant life would overcome the negative impacts of such competition and lead to maximizing maize productivity per unit area [39]. As an alternative breeding strategy, tolerance to high plant population density has been suggested to improve performance under diverse abiotic stresses including drought [16-19,40].

### 3.4 Stress Tolerance of Inbreds and Hybrids

Stress tolerance index (STI) values of studied genotypes estimated using the equation suggested by Fageria [23] under the stressed environments E2 through E6 are presented in Table 5. According to our scale, when STI is  $\geq 1.0$ , it indicates that genotype is tolerant (T), if STI is  $< 1$ , it indicates that genotype is sensitive (S). The highest STI under all five stressed environments was exhibited by the inbred line L53, followed by inbred L20 and then Sk5. These three inbreds had STI value greater than unity under all five stressed environments and therefore could be considered tolerant to water



stress, medium and high plant density stress and water stress combined with medium and high density stresses.

On the contrary, the three inbred lines Sd7, L18 and L28 exhibited STI values less than unity under all five stressed environments and therefore could be considered sensitive to water stress, medium and high plant density stress and water stress combined with medium and high density stresses; with the most sensitive one was the inbred Sd7 under E4, E5 and E6 and inbred L18 under E2 and E3 environments. For F<sub>1</sub> crosses, the highest STI value was recorded by the cross L20 x L53 (TxT) under all stressed environments, followed by the cross L53 x Sk5 (TxT) and L53 x Sd7 (TxS) under all stresses. On the other hand, the most sensitive crosses under all stressed environments are L18 x L28 (S x S), L53 x L18 (T x S) and Sk5 x Sd7 (T x S). It is observed that all three T x T crosses (L20 x L53, L20 x Sk5 and L53 x Sk5) were tolerant under each (sole) stress and both stresses combined together, indicating hybrid accumulation of effects of stress tolerance genes from its two parents. Among the three S x S crosses, two (L18 x L28 and L18 x Sd7) were sensitive and one (L28 x Sd7) was tolerant to sole and combined stresses. The stress tolerance exhibited in the latter S x S hybrid could be attributed to epistasis effects. Among the nine T x S crosses, five (L20 x L28, L20 x Sd7, L53 x L28, L53 x Sd7 and Sk5 x L18) were tolerant in sole and combined stresses, while four (L20 x L18, L53 x L18, Sk5 x L28 and Sk5 x Sd7) were sensitive under each stress and combined stresses. The tolerance of the first five T x S crosses indicated accumulating of more genes of dominance effects of tolerance over sensitivity, while the tolerance of the latter four T x S crosses suggested accumulating less number of dominant tolerance genes.

A very strong association between tolerance to water stress and each of tolerance to density stress and to both stresses combined together was exhibited by inbred lines ( $r = > 0.96$ ) and hybrids ( $r = > 0.98$ ) (Table 6). The tolerant inbred or hybrid to water stress is also tolerant to elevated density and to water stress combined elevated density stresses and the *vice versa*.

The strong association between tolerance of hybrids and inbreds of maize to both water stress (drought stress) and high plant density stress

(water, nutrient and light stresses) in the present study was reported previously by many investigators [16-19,40].

### 3.5 Superiority of Tolerant (T) Over Sensitive (S) Genotypes

To describe the differences between tolerant (T) and sensitive (S) inbreds and hybrids, data of the selected characters were averaged for the two groups of inbreds and hybrids differing in their high density tolerance, namely in grain yield/plant under high density stress (E3), water stress (E4), and combined between water stress and high density stress (E6) (Table 7). Based on STI index, the high-density tolerant (T) inbred lines were L20, L53 and Sk5 and the high-density sensitive (S) inbred lines were Sd7, L18 and L28. Moreover, the 3 F<sub>1</sub> crosses L20 x L53, L53 x Sk5 and L53 x Sd7 were considered the most tolerant to high density, while the crosses L18 x L28, L53 x L18 and Sk5 x Sd7 were considered as the most high-density sensitive crosses (Table 8). Based on stress tolerance index, the tolerant inbreds and hybrids for both stresses were the same tolerant ones to either drought or high density stress alone and the sensitive ones for both stresses were the same sensitive to either ones.

Data averaged for each of the two groups (T and S) of inbreds and crosses differing in tolerance to combined stress of high density and water stress (E6) indicate that grain yield/ha of combined stress tolerant (T) was greater than that of the combined stress sensitive (S) inbreds and crosses by 206.90 and 60.25%, respectively under combined stress (water stress and 95,200 plants/ha) conditions. Superiority of combined stress tolerant (T) over sensitive (S) inbreds in GYPH under combined stress (E6) was due to their superiority in GYPP (233.72%), EPP (22.50%), RPE (25.53%), KPR (36.67%), KPP (62.81%), 100-KW (27.15%), *i.e.* in all studied yield component traits. Likewise, under combined stress, the tolerant inbreds showed 3.03% shorter ASI, 23.66% smaller leaf angle than the sensitive inbreds (Table 7). Superiority of T over S hybrids in GYPH under combined stress was due to their superiority in GYPP (60.25%), EPP (15.18%), RPE (17.92%), KPR (26.08), KPP (32.30%), 100-KW (27.95%), BS (-50.56%) and ASI (-9.39%), DTA (-9.00%), DTS (-9.02%), PH (-9.67%), EH (-20.77%) and LANG (-26.33%) than sensitive F<sub>1</sub> crosses (Table 7).

**Table 4. Mean grain yield/ha (ton) across two seasons and percentage change (Ch%) from non-stressed (E1) to stressed environments (E2 through E6)**

Genotypes	E1		E2		E3		E4		E5		E6	
	(WW-LD)		(WW-MD)		(WW-HD)		(WS-LD)		(WS-MD)		(WS-HD)	
	Mean	Ch	Mean	Ch	Mean	Ch	Mean	Ch	Mean	Ch	Mean	Ch
<b>Inbreds</b>												
L20	4.95		6.41	-29.5**	6.64	-34.1**	2.39	51.6**	2.76	44.2**	3.86	22.0**
L53	6.13		6.47	-5.5**	6.66	-8.6**	3.52	42.5**	3.87	36.9**	4.73	22.9**
Sk5	3.6		4.48	-24.5**	4.92	-36.6**	2.17	39.8**	2.47	31.5**	2.64	26.6**
L18	2.16		1.85	14.5**	1.86	13.9**	1.49	31.3**	1.47	32.3**	0.98	54.6**
L28	2.06		2.44	-18.5**	2.83	-37.3**	0.87	57.7**	1.28	37.9**	1.64	20.4**
Sd7	2.01		2.5	-24.1**	3.05	-51.7**	0.63	68.5**	0.72	64.4**	1.04	48.4**
<b>F<sub>1</sub> crosses</b>												
L20 X L53	12.88		16.45	-27.71**	17.05	-32.42**	11.23	12.81**	14.24	-10.57**	14.95	-16.13**
L20 XSK5	10.22		12.59	-23.19**	14.21	-39.11**	7.75	24.19**	10.22	-0.03	10.76	-5.29**
L20 X L18	10.15		13.38	-31.77**	16.04	-58.00**	8.33	17.99**	10.76	-5.94**	12.04	-18.59**
L20 X L28	10.81		12.88	-19.17**	14.51	-34.26**	7.97	26.23**	10.77	0.3	10.57	2.23*
L20 X Sd7	10.53		12.6	-19.67**	14.85	-41.05**	8.31	21.05**	10.07	4.29**	11.28	-7.21**
L 53 X Sk5	11.4		15.5	-35.99**	16.47	-44.48**	9.31	18.34**	12.16	-6.71**	12.72	-11.56**
L53 X L18	8.99		10.2	-13.38**	12.85	-42.82**	6.45	28.30**	8.23	8.48**	8.85	1.62
L53 X L28	11.03		11.66	-5.75**	14.99	-35.90**	7.95	27.88**	10.9	1.19	9.93	9.98**
L53 X Sd7	11.19		15.13	-35.24**	16.3	-45.74**	8.96	19.92**	11.82	-5.69**	12.3	-9.94**
Sk5 X L18	10.9		13.6	-24.77**	15.18	-39.20**	8.43	22.68**	10.9	0.03	11.44	-4.95**
Sk5 X L28	10.34		13.9	-34.41**	15.45	-49.40**	8.17	20.99**	10.59	-2.44*	11.52	-11.36**
Sk5 X Sd7	9.58		10.88	-13.59**	13.48	-40.77**	6.86	28.40**	8.92	6.82**	9.26	3.32**
L18 X L28	7.91		8.59	-8.60**	11.42	-44.37**	5.76	27.22**	6.3	20.37**	6.84	13.56**
L18 X Sd7	9.88		11.17	-13.08**	13.8	-39.66**	7.16	27.53**	9.41	4.71**	9.44	4.44**
L28 X Sd7	10.49		12.67	-20.75**	14.67	-39.84**	7.97	24.08**	10.6	-1.02	10.95	-4.40**
<b>Checks</b>												
SC 130	10.67		11.76	-10.2**	13.59	-27.4**	7.62	28.5**	9.37	12.1**	11.44	-7.3**
SC 2055	10		12.58	-25.8**	13.87	-38.7**	6.89	31.1**	8.92	10.8**	11.44	-14.4**

WW = well watering, WS = water stress, LD = low density, MD = medium density, HD = high density, E = environment, Ch% =  $100 \times (E1 - RE) / E1$ , RE = Respective environment, \* and \*\* significant at 0.05 and 0.01 probability levels

**Table 5. Stress tolerance index (STI) of maize inbreds and hybrids under sole (E2, E3 and E4) and combined (E5 and E6) stress conditions**

Genotype	E2	E3	E4	E5	E6
	WW-MD	WW-HD	WS-LD	WS-MD	WS-HD
<b>Inbreds</b>					
L20	2.25	2.12	1.85	1.72	2.24
L53	2.81	2.64	3.39	2.95	3.40
Sk5	1.14	1.14	1.09	1.27	1.02
L18	0.29	0.26	0.49	0.42	0.25
L28	0.36	0.38	0.28	0.37	0.38
Sd7	0.36	0.5	0.22	0.31	0.22
<b>F<sub>1</sub> crosses</b>					
L20 × L53	1.59	1.46	1.71	1.64	1.70
L20 × SK5	1.00	1.00	1.00	1.00	1.01
L20 × L18	0.89	0.92	0.92	0.93	0.89
L20 × L28	1.08	1.07	1.07	1.10	1.10
L20 × Sd7	0.99	1.00	1.02	1.01	1.02
L 53 × Sk5	1.33	1.25	1.27	1.27	1.28
L53 × L18	0.70	0.75	0.70	0.70	0.72
L53 × L28	1.22	1.17	1.14	1.17	1.17
L53 × Sd7	1.27	1.21	1.21	1.23	1.21
Sk5 × L18	1.13	1.14	1.10	1.13	1.13
Sk5 × L28	0.94	0.96	0.98	0.97	0.97
Sk5 × Sd7	0.79	0.83	0.78	0.80	0.79
L18 × L28	0.51	0.58	0.54	0.46	0.48
L18 × Sd7	0.83	0.87	0.84	0.86	0.82
L28 × Sd7	1.01	1.04	1.03	1.01	1.02
<b>Checks</b>					
SC 130	0.99	1.01	1.07	1.05	1.02
SC 2055	1.01	0.99	0.93	0.95	0.98

WW = well watering, WS = water stress, LD = low density, MD = medium density, HD = high density, E = environment

**Table 6. Rank correlation coefficients among six environments for STI of parental inbreds (above diagonal) and F<sub>1</sub> crosses (below diagonal) across two seasons**

Environment	E2	E3	E4	E5	E6
	WW-MD	WW-HD	WS-LD	WS-MD	WS-HD
E2		0.998**	0.97**	0.97**	0.99**
E3	0.99**		0.96**	0.97**	0.986**
E4	0.98**	0.98**		0.99**	0.99**
E5	0.99**	0.99**	0.99**		0.985**
E6	0.98**	0.98**	0.99**	0.99**	

WW = well watering, WS = water stress, LD = low density, MD = medium density, HD=high density, E = environment

Superiority of T to S inbreds and crosses may be attributed to the high water use efficiency traits of the hybrids, due to heterosis, relative to their inbred parents. These results are in agreement with those reported by several investigators [41,42]. The superiority of modern maize hybrids tolerant of high plant density has also been attributed to decrease barrenness [40], more leaf erectness [7], synchronization of 50% anthesis with 50% silking [41] and increased prolificacy (more ears per plant) [42]. A shortened ASI is considered an indication of higher flow of assimilates to the developing ears during the

early reproductive stage under conditions of high density stress [43,44]. High plant density-tolerant genotypes display shorter ASI than intolerant ones [45–47]. Al-Naggar et al. [33] also reported that under high plant density, tolerant testcrosses showed 314.4% more GYPP, 115.0% more KPP, 48.4% heavier 100-KW, 42.9% more EPP, 98.2% less BS and 63.3% shorter ASI than sensitive testcrosses. Mansfield and Mumm [48] reported that in U. S. maize germplasm evaluated for plant density tolerance, a subset of traits including leaf angle, upper stem diameter, leaf area required to produce a gram of

grain, kernel rows per ear, days to canopy closure, barrenness, kernels plant<sup>-1</sup>, kernel length, leaf number, upper leaf area, stay green, zipper effect, kernels per row, and anthesis-to-silking interval were associated with grain yield across plant densities ranging from 47,000 to 133,000 plants ha<sup>-1</sup>. Barrenness, anthesis-silking interval (ASI), leaf senescence and leaf rolling were proposed by Bolanos and Edmeades [13] and Edmeades et al. [46] as secondary traits to improve yield in drought-prone environments. These results are consistent with those reported by Al-Naggar et al. [21]. Reduction in barren stalks and shortening in ASI of tolerant as compared to sensitive inbreds and hybrids in the present study are desirable and may be considered as important contributors to water deficit as well as to high-density tolerance. Similar conclusions have been reported by several investigators [33,43–51].

### 3.6 Differential Response of TxT, TxS and SxS Crosses

Mean performance of traits were averaged across three groups of F<sub>1</sub> crosses, *i.e.* TxT, TxS and SxS groups based on parental tolerance to each stress (either high density or water stress) or both stresses together and presented in Table (8). Number of crosses was 3, 9 and 3 for the TxT, TxS and SxS groups, respectively. In general, TxT crosses had favorable (higher) values for grain yield and its attributes and lower (favorable) values for DTA, DTS, ASI, PH, EH,

BS and LANG than SxS and TxS crosses under stressed environments (E5 and E6) and non-stressed environment (E1).

In general, under the most severe environment (E6) where both severe stresses (water stress and density of 95,200 plants/ha) existed, water stress and high density TxT crosses were the most superior for all studied traits as compared to T x S and S x S crosses (Table 9). The TxS crosses for both stresses came in the second rank for superiority in all traits and the SxS crosses were in the last rank.

Under water deficit and high density stresses together (E6), grain yield/ha of water stress and high-D TxT crosses (11.81 t/ha) was greater than that of SxS (9.08 t/ha) and TxS (10.80 t/ha) by 41.13 and 18.61%, respectively. This indicates that to obtain a tolerant cross to both stresses in the same time, it is preferable that its two parental inbred lines should be tolerant to both stresses. This assures that water stress combined with density stress tolerance traits are quantitative in nature, so the tolerant cross accumulates additive genes of both water stress and high density tolerance from both parents.

Superiority of water stress and high-D TxT and TxS over SxS crosses in GYPH under low-N and high-D stresses (41.13 and 18.99%, respectively) was due to their superiority in GYPP by 41.13 and 18.97%, KPP by 25.39 and 12.01%, 100-KW by 17.26 and 5.65%, EPP by 12.63 and

**Table 7. Superiority (%) of the three most tolerant (T) over the three most sensitive (S) inbreds and crosses for studied characters under the stressed environment E6 combined across 2013 and 2014 seasons**

Trait	Inbreds			Crosses		
	T	S	% Superiority	T	S	% Superiority
<b>E6 (WS-HD)</b>						
DTA (day)	64.97	65.5	-0.81**	62.94	69.17	-9.00**
DTS (day)	68.83	68.89	-0.08	67.5	74.19	-9.02**
ASI (day)	4.44	4.58	-3.03	4.56	5.03	-9.39**
PH (cm)	206.17	197.06	4.62**	242.33	268.28	-9.67**
EH (cm)	93.14	81.83	13.83**	100.87	127.31	-20.77**
BS (%)	12.33	16.81	-26.63**	10.09	20.4	-50.56**
LANG (°)	24.56	32.17	-23.66**	25.33	34.39	-26.33**
EPP	0.97	0.79	22.50**	1.07	0.93	15.18**
RPE	13.36	10.64	25.53**	14.21	12.05	17.92**
KPR	26.47	19.37	36.67**	43.68	34.64	26.08**
KPP	309.68	190.22	62.81**	575.34	434.86	32.30**
100-KW (g)	26.95	21.2	27.15**	31.48	24.6	27.95**
GYPP (g)	39.55	11.85	233.72**	143.49	89.54	60.25**
GYPH (ton)	3.74	1.22	206.90**	13.32	8.31	60.25**

$$\% \text{ Superiority} = 100 \times [(T - S)/S]$$

4.21%, RPE by 12.20 and 4.25% and KPR by 18.29 and 6.35%, respectively (Table 9). Moreover, under the most severe stresses in this experiment existed in E6 environment, water stress and high-D TxT and T x S crosses were earlier in DTA by 8.56 and 6.43%, DTS by 7.99 and 5.37%, shorter in PH by 6.93 and 4.39%, lower in EH by 18.16 and 6.45%, lower in BS by 41.53 and 23.70% and narrower in LANG by 21.02 and 8.59% than SxS crosses, respectively.

In general, crosses classified as water stress and high-density tolerant x water stress and high-

density tolerant crosses in terms of grain yield under water stress and high density stresses had a better drought adaptive traits and high density adaptive traits such as higher values of all grain yield components and lower values of DTA, DTS, ASI, PH, EH, BS and LANG as compared with water stress and high density sensitive x water stress and high density sensitive crosses. Maize adaptive traits to high density stress seem to be generally similar to those adaptive traits to water stress as cleared from the results of the present study. Some investigators [7,21,44] reached to a similar conclusion.

**Table 8. Trait differences averaged across two seasons for TxT, TxS and SxS groups of F<sub>1</sub> crosses for combinations of water stress and plant density stress**

Trait	WW-LD (E1)			WS-MD (E5)			WS-HD (E6)		
	T xT	T xS	S xS	T xT	T xS	S xS	T xT	T xS	S xS
DTA (day)	58.67	59.49	60.44	61.67	63.18	67.22	63.17	64.64	69.08
DTS (day)	60.78	61.69	62.72	65.39	66.99	71.19	67.83	69.76	73.72
ASI (day)	2.11	2.2	2.28	3.72	3.81	3.97	4.67	5.12	4.64
PH (cm)	227.78	245.28	257.17	237.72	245.33	252.61	246.17	252.89	264.5
EH (cm)	92.37	106.86	114.75	94.84	109.68	116.4	101.35	115.85	123.84
BS (%)	8.36	9.84	11.99	9.97	12.87	16.05	11.05	14.42	18.9
LANG (°)	24.39	28.52	31.33	25.06	30.07	33.17	26.11	30.22	33.06
EPP	1.36	1.22	1.16	1.2	1.06	1.02	1.07	0.99	0.95
RPE	15.74	14.59	13.58	14.58	13.53	12.87	13.98	12.99	12.46
KPR	49.68	45.3	43.46	45.07	41.06	39.01	42.82	38.5	36.2
KPP	918.58	818.1	752.37	689.37	598.56	547.42	563.89	503.71	449.71
100KW (g)	38.14	35.92	34.3	32.47	29.93	28.08	30.5	27.48	26.01
GYPP (g)	248.19	224.43	204.01	171.56	147.86	124.17	137.95	116.29	97.75
GYPH (ton)	11.50	10.39	9.43	12.21	10.33	8.77	12.81	10.80	9.08

T = tolerant, S = sensitive, LD = low density (47,600 plants/ha), MD = medium density (71,400 plants/ha) and HD = high density (95,200plants/ha)

**Table 9. Superiority (%) of T x T and T x S over S x S crosses for selected traits under combination of plant densities and irrigation regimes across two seasons**

Trait	WW-LD (E1)		WS-MD (E5)		WS-HD (E6)	
	T xT	T xS	T xT	T xS	T xT	T xS
DTA	-2.93**	-1.57*	-8.26**	-6.01**	-8.56**	-6.43**
DTS	-3.09**	-1.64	-8.15**	-5.90**	-7.99**	-5.37**
ASI	-7.46	-3.51	-6.30	-4.03	0.65	10.34
PH	-11.43**	-4.62**	-5.89**	-2.88	-6.93**	-4.39
EH	-19.50**	-6.88**	-18.52**	-5.77**	-18.16**	-6.45*
BS	-30.28*	-17.93	-37.88	-19.81	-41.53*	-23.70
LANG	-22.15**	-8.97**	-24.45**	-9.35**	-21.02**	-8.59**
EPP	17.24**	5.17	17.65**	3.92	12.63	4.21
RPE	15.91**	7.44*	13.29**	5.13	12.20**	4.25
KPR	14.31**	4.23*	15.53**	5.26*	18.29**	6.35
KPP	22.09**	8.74	25.93**	9.34	25.39**	12.01
100-KW	11.20**	4.72	15.63**	6.59*	17.26**	5.65
GYPP	21.66**	10.01**	38.17**	19.08**	41.13**	18.97**
GYPH	21.96**	10.22**	39.19**	17.79**	41.13**	18.99**

% Superiority = 100 x [(TxT) or (TxS) - (SxS)/(SxS)], T = tolerant, S = sensitive, WW = well watering, WS = water stress, LD = low density (47,600 plants/ha), MD = medium density (71,400 plants/ha) and HD = high density (95,200 plants/ha)

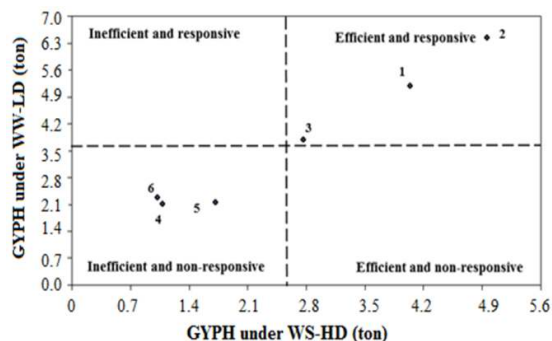
### 3.7 Grouping Genotypes Based on Efficiency and Responsiveness

According to efficiency under high density combined with water stress and responsiveness to low density with well watering, studied inbreds and crosses were classified into four groups, *i.e.* high density with water stress efficient and responsive to low density with well water (E-R), high density with water stress efficient and non-responsive (E-NR), high density with water stress inefficient and responsive (IE-R) and high density with water stress inefficient and non-responsive (IN-NR) based on GYPH trait. The inbreds No.2 (L53), No.1 (L20) and No.3 (Sk5) were classified as high density with water stress efficient and responsive, while inbreds No.4 (L18), No.5 (L28) and No.6 (Sd7) were classified as high density with water stress inefficient and non-responsive (Fig. 1). The F<sub>1</sub> crosses No. 1 (L20 × L53), No. 6 (L 53 × Sk5), No. 9 (L53 × Sd7), No. 10(Sk5 × L18) and No.5 (L20 ×Sd7) had the highest GYPH under high-D-water stress and low-D-well water, *i.e.* they could be considered as the most high-D with water stress efficient and the most responsive genotypes to the good environment in this study (Fig. 2). On the contrary, the F<sub>1</sub> crosses No.13 (L18 × L28), No.7 (L53 × L18), No.12 (Sk5 × Sd7), No.14 (L18 × Sd7) and No.2 (L20 ×Sk5) had the lowest GYPH under both low D-WW and high D-WS and therefore could be considered inefficient and non-responsive. The crosses No.3 (L20 × L18) and No.11 (Sk5 × L28) occupied the group of density with water efficient and non-responsive (high GYPH under HD-WS but low GYPH under LD-WW). The crosses No.4 (L20 × L28), No.8 (L53 × L28) and No.15 (L28 × Sd7) had low GYPH under LD-WW and under HD-WS, *i.e.* high density and water stress inefficient and responsive to LD-WW environment.

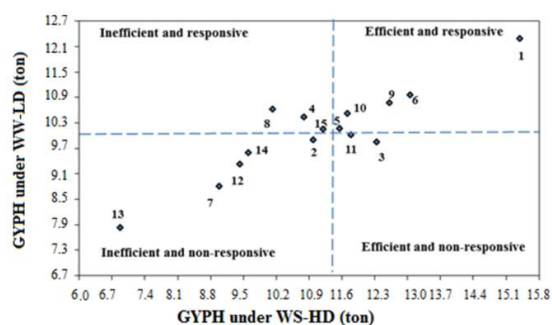
Summarizing the above-mentioned classifications, it is apparent that the three parents L20, L53 and Sk5 and the F<sub>1</sub> crosses No.1(L20 × L53), No.6(L 53 × Sk5), No.9(L53 × Sd7), No.10(Sk5 × L18) and No.5(L20 × Sd7) occupied the first group in all classifications; they are the most efficient, most tolerant to high density, water stress and combination between water and density stresses, responsive to the good environment and high yielders under the stressed environment.

According to Fageria and Baligar [52-54] genotypes (progenies) belonging to the group "efficient and responsive" appear to be the most

desirable materials for breeding programs that deal with adaptation to high density stress, water stress or both stresses together. On the contrary, the three parents L18, L28 and Sd7 and the crosses No.13(L18 × L28), No.7(L53 × L18), No.12(Sk5 × Sd7), No.14(L18 × Sd7) and No.2(L20 ×Sk5) occupied the fourth group in all classification; they are the most inefficient, most sensitive to high density, water stress and combination between water and density stresses, non-responsive to the good environment and low yielders under the stressed environment.



**Fig. 1. Relationships between grain yield/ha (GYPH) of 6 parental inbreds under non stress (WW-LD) and stress for both water and high density (WS-HD) combined across 2013 and 2014 seasons. Broken lines represent mean of GYPH. Numbers from 1 to 6 refer to parental inbreds names**



**Fig. 2. Relationships between grain yield/ha (GYPH) of 15 F<sub>1</sub> maize hybrids under both stress (WW-LD and WS-HD) combined across 2013 and 2014 seasons. Broken lines represent mean of GYPH. Numbers from 1 to 15 refer to F<sub>1</sub> hybrids names**

### 3.8 Regression of Grain Yield on Elevated Levels of Stress

Data were reanalyzed to evaluate GYPH responses of inbreds and hybrids across varying

levels of stress *via* regression technique. For each genotype or group of genotypes, quadratic regression function was performed for irrigation regime × plant density interaction. The regression functions were used to identify which treatment(s) provide optimum GYPH for each genotype (or group of genotypes). The relationship between the six environments (combinations of two irrigation regimes and 3 plant densities) and grain yield/ha of inbreds across years are illustrated in Fig. 3. The 6 environments were arranged in Fig. 3 based on the severity of both water and plant density stresses together, where the poorest environment (WS-HD) represents maximum stress (water stress and highest plant density), while the best environment (WW-LD) represents the most favorable one (well watering and lowest plant density). The three inbred parents (L20, L53 and Sk5) showed a quadratic regression function, with an optimum combination of well watering and close to 95,200 plants/ha plant density. While, the inbreds L18, L28 and Sd7 showed a weak quadratic regression very close to linear response, with an optimum environment of combination between well watering and density between low D (L18) and high D (L28 and Sd7) (Fig. 3).

The relationships between the six environments (combinations of 2 irrigation regimes and 3 plant densities) and grain yield/ha of F<sub>1</sub> crosses across years are illustrated in Fig. 4. The grain yield/ha

across years of the 4 groups of F<sub>1</sub> crosses showed a quadratic regression function under the six combinations of water regimes and plant densities. The optimum density and watering combination was about 88,000 plants/ha when giving well watering across the four groups of F<sub>1</sub> crosses. The most responsive group of hybrids to the improvement of environmental conditions was E-R followed by E-NR and IE-R groups, while the lowest responsive group was IE-NR.

According to tolerance to high density combined with water stress and high yielding under both stresses together, studied crosses were classified into three groups, *i.e.* tolerant to high density with water stress and high yielding (T-HY), sensitive to high density with water stress and high yielding (S-HY) and sensitive to high density with water stress and of low GYPH (S-LY). The grain yield/ha across years of the 3 groups of F<sub>1</sub> crosses showed a clear quadratic curvilinear regression function under the six studied environments (Fig. 5). The optimum environment combination was well watering combined with high plant density (95,200 plants/ha) for the T-HY and S-HY groups and well watering combined with medium plant density (71,400 plants/ha) for the group S-LY. The most responsive group of hybrids to the elevated plant density combined with irrigation regime was T-HY followed by S-HY group, while the lowest responsive group was S-LY.

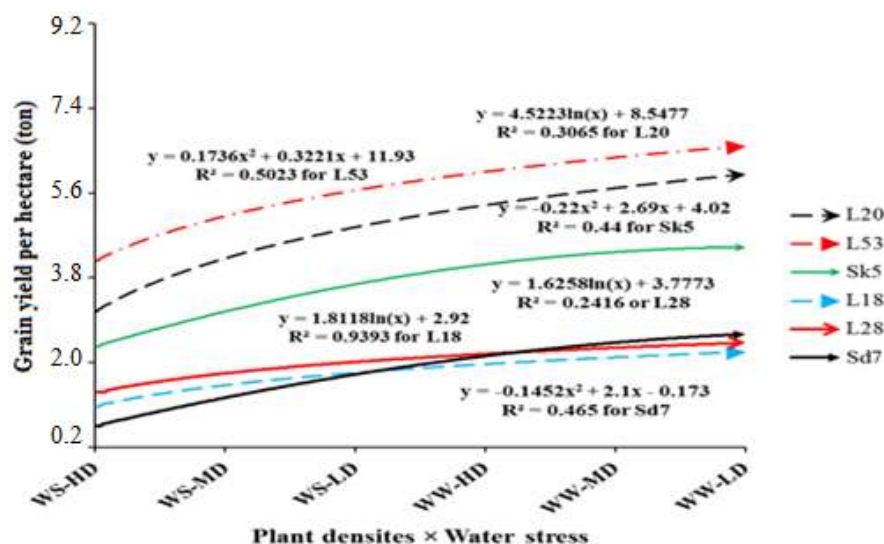


Fig. 3. Relationship between grain yield/ha of inbreds and six environment combinations between three plant densities and two water regimes across two seasons

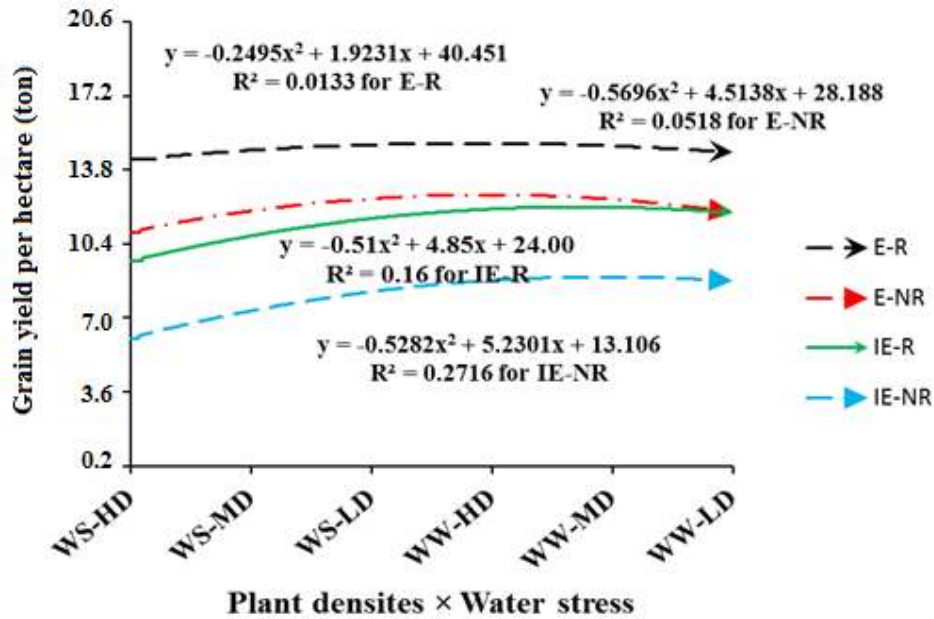


Fig. 4. Relationship between grain yield/ha of four groups of F<sub>1</sub> crosses, namely, efficient and responsive (E-R), efficient and non-responsive (E-NR), inefficient and responsive (IE-R), and inefficient and non-responsive (IE-NR) crosses and six combinations among three plant densities and two irrigation regimes across two seasons

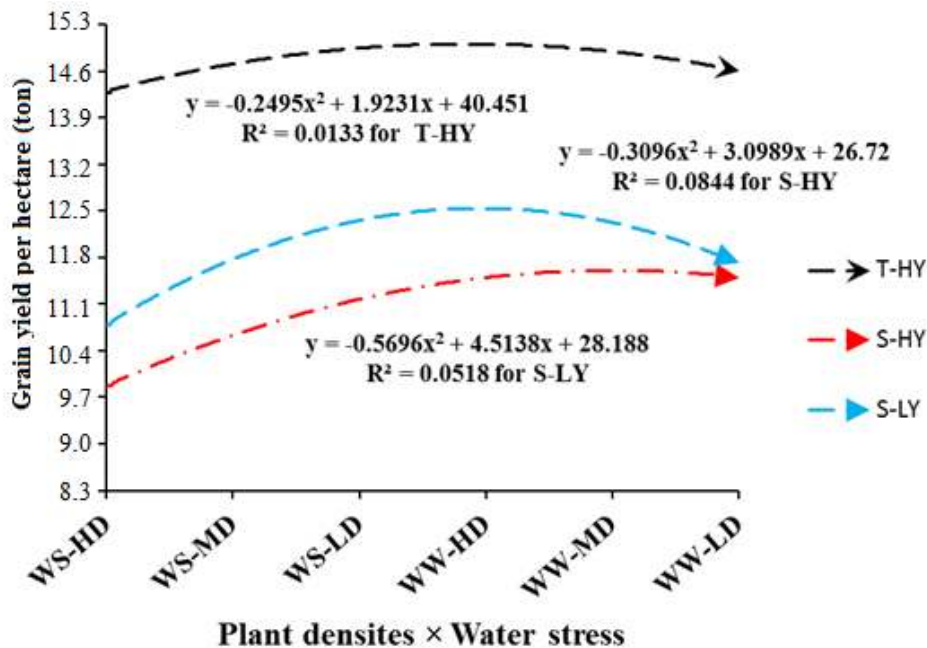


Fig. 5. Relationship between grain yield/ha of four groups of F<sub>1</sub> crosses, namely, tolerant and high yield (T-HY), sensitive and high yield (S-HY), tolerant and low yield (T-LY) and sensitive and low yield (S-LY) across environment combinations between two plant densities and two water regimes across two seasons



In this context, Shapiro and Wortmann [55] reported that the corn grain yield typically exhibits a quadratic response to plant density with a near-linear increase across a range of low densities, a gradually decreasing rate of yield increase relative to density increase and finally a yield plateau at some relatively high plant density. Boomsma et al. [56] showed that under large ranges of plant density (54,000-104,000 plants/ha) and N rate (0-330 kg N/ha), higher densities required more N. This seems logic, given the prevailing belief that high yields require more plants, and that more plants require more N.

Clark [39] mentioned that there was little yield response to N rates above 90 kg N/ha at the low and high densities, as there was a curvilinear increase until yield plateau at the low density (8.1 Mg/ha at 133 kg N/ha) and the high density (5.9 Mg/ha at 102 kg N/ha). He added that response to N was greatest at the middle density (83,980 plants/ ha), as there was a quadratic response with maximum yield at 188 kg N/ha (8.7 Mg/ha). He found that across the low-stress environments, the lowest density (44,460 plants/ha) responded little to N rates above 90 kg N/ha, while there was greater response to N rates at the middle density (13.5 Mg/ha at 162 kg N/ha) and the high density (13.4 Mg/ha at 174 kg N/ha). He concluded that no support was found for the idea that increasing corn yield requires increases in both plant density and N rate above rates typically used. Their and our results advance our understanding of irrigation regime-plant density interaction within contrasting environmental conditions, but understanding the complexities of hybrid interactions with irrigation regime and plant density will require additional work.

#### 4. CONCLUSION

Some maize single cross hybrids resulted from this study could maximize maize productivity, reaching 17.05 t ha<sup>-1</sup> in the cross L20 × L53 on the same land unit area, if they are grown at twice the plant population density of 95,200plants ha<sup>-1</sup> used in Egypt, but provided they are given the full irrigation at all growth stages of maize plant. The same cross also gave the highest superiority in grain yield (51.92%) over the best check in this experiment under water stress combined with high plant density (95,200 plants ha<sup>-1</sup>). This indicates that the increase of GYPH due to the increase in plant density could compensate the reduction in GYPH due to water

stress at flowering stage. A very strong association was exhibited between tolerance to water stress and each of density stress and both stresses combined together, indicating that the tolerant inbred or hybrid to water stress is also tolerant to elevated density and to water stress combined with elevated density stress and the *vice versa*. The results also indicates that to obtain a tolerant cross to both stresses in the same time, it is preferable that its two parental inbred lines should be tolerant to both stresses. Maize adaptive traits to high density stress seem to be generally similar to those adaptive traits to water stress as cleared from the results of the present study. The best combination of plant population density and irrigation regime for giving the highest grain yield per unit land area in this study was identified for the studied maize genotypes. The optimum environment combination was well watering combined with high plant density (95,200 plants ha<sup>-1</sup>) for the tolerant high yielding group of hybrids and well watering combined with medium plant density (71,400 plants ha<sup>-1</sup>) for the sensitive low yielding group.

#### COMPETING INTERESTS

Authors have declared that no competing interests exist.

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