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Influence of Testers and Watering Regimes on Combining ability and Heterosis of Maize Top Crosses

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Abstract

Thirty-six top crosses of white maize were produced as the combinations of four male testers and nine newly developed inbreds as female parents. The four testers included an inbred, a single cross, a three-way cross, and a synthetic variety. The parents and crosses were evaluated in two separate field irrigation trials to explore the extent of general combining ability and heterosis of crosses using narrow, medium, and broad base testers under normal and stressed watering regimes. Watering regimes and genotypes as sources of variation of combined analysis were highly significant for all traits. The (Parents vs. Crosses) with watering regimes interaction possessed highly significant mean squares for dates of tasseling and silking. Significant negative GCA effects were recorded for tasseling date and silking date under both conditions. The grain yield of crosses varied differently between regimes. Two testers (I.272 and G.2) and two lines (I.276 and I.278) recorded significant negative GCA effects (favorable) for dates to tasseling (TD) and silking (SD) under both watering regimes. The SC.10 (as tester) and three female lines (I.274, I.277, and I.281) exhibited significant positive (unfavorable) GCA effects on flowering dates under both conditions. Variable GCA effects for ASI were observed among the tested maize genotypes, particularly under normal conditions. However, under-stressed one, all tested genotypes showed insignificant GCA effects for ASI except I.272, which may be shorted the ASI. Out of thirty-six top crosses, thirty and twenty-eight crosses recorded significantly unfavorable (favorable) heterosis under normal irrigated trial comparing to twenty-two, and twenty-one crosses in stressed watering regime for TD and SD, respectively. However, for ASI, eleven and nine crosses were significantly showed favorable flowering intervals than corresponding mid-parents under normal and stressed conditions respectively. For grain yield per plot about 25% significant superiority in performance of all top crosses over corresponding mid-parents was recorded under either normal or stress conditions. The investigated ten inbred lines may be of great benefit for hybrid water-saving maize breeding program. Narrow genetic base tester (I.272) resulted in favorable heterosis of crosses and could be recommended in advanced generations of homozygosity, whereas mostly based ones may be valid for screening during the first generations of selfing.

Introduction

Maize (*Zea mays L.*) is the third most important cereal crops in the world after wheat and rice. Owing to limited water resources, the development of drought-tolerant varieties is an essential goal of plant breeding to alleviate the effects of water deficit.

Water-deficit stress negatively affects the growth and development of maize plants Moreno et al. (2005). Water stress reflected considerably in delaying silking and increase the anthesis-silking interval (ASI), with yield failure according to Byrne et al. (1995), Kahiu et al. (2013) and Darwish et al. (2015).

Recent maize hybrids may be bred using drought-tolerant parental inbreds either under regular irrigation or water deficit conditions. Drought tolerant germplasm might be specially adapted to low yield environments as reported by Moreno et al. (2005) and Kiani (2013).

Combining ability studies enable the breeders to select suitable cross combinations. General combining ability refers to the average performance of the genotype in a series of hybrid combinations and is a measure of additive gene action Sharief et al. (2009).

The active breeding program for maize drought-tolerant hybrids necessitates determining the performance and variation among newly developed maize inbred lines and their test crosses under normal and water-stressed conditions. Thus, objectives of the present studies were to explore general combining ability of some newly developed maize inbred lines and the extent of heterosis of their top crosses using narrow, medium and broad base testers under two watering regimes

Materials and Methods

The field experiments were conducted at the Agricultural Experiments and Research Farm of the Faculty of Agriculture, Minia University, El-Minia, Egypt during 2013 and 2014 summer seasons as illustrated by Darwish et al. (2015).

Top crosses production

During the summer season of 2013, thirty-six top crosses of white maize were produced as the combinations of four male testers and nine inbreds as female parents. The male testers included I.272, SC.10, TWC.310, and G.2 variety. Another nine inbred lines (I.273, I.274, I.275, I.276, I.277, I.278, I.279, I.280, and I.281) were used as female parents of the top crosses. The inbred lines were developed by Maize Research Section, ARC under drought conditions. The original seed of these parents was kindly provided by Maize Research Section, Field Crops Research Institute, ARC.

Field trials, watering regimes, and analyzed variables

In the summer season of 2014, the 36 top crosses along with parental genotypes were evaluated under two separate irrigation trials, *i.e.*, normal (N) and stressed (S). The irrigation treatments were adopted after the 1^{st} irrigation, 31 days after sowing. The irrigation of ordinary (N) and stress (S) experiments were conducted at 14 and 21days intervals, respectively. Soil type was clay loam, and the average depletion of soil moisture reached to 65-72 and 92-95.0% in N and S conditions, respectively. Each trial was conducted as Randomized Complete Blocks Design (RCBD) with three replications. The plot size comprised three ridges, each three meters long and 70 cm apart ($6.3m^2$). The seeds were dry planted on 27^{th} May on one side of the ridge in hills distanced 25 cm. Seedlings were thinned to one plant/hill three weeks after sowing.

The dates of flowering were recorded as the numbers of days to anthesis (pollen shed) and silking of 50% plants per plot. The difference between these dates was considered as an anthesis-silking interval (ASI). Grain yield per plot was recorded as the grain weight of all plants per plot adjusted to 15.5% grain moisture.

Statistical analysis

The line × tester analysis was performed for obtaining data of each trial, then the combined across trials was followed according to Kempthorne (1957) and following by Singh and Chaudhary (1977).

However, to determine the influences due to irrigation treatments (regimes) and its interactions with other investigated factors, the combined analysis over environments was performed. Homogeneity test was adopted of the error terms of both trials prior analysis of variance, which indicates the allowance of combined analysis.

Heterosis was calculated for each cross as the percent deviation of the mid-parent average from F_1 mean performance for each experiment and combined over both conditions.

Appropriate L.S.D values were computed to test the significance of the difference between the average of both parents and corresponding F_1.

Results and discussion

Significance of Mean Squares

Mean squares due to line × tester analyses combined over watering regimes are presented in Table 1. Genotypes (G.) included parents, either lines or testers and resulted in F_1 crosses varied highly significantly for all studied traits. This indicates that the tested maize genotypes varied considerably over both watering conditions. Watering regimes (W.) as a source of variation of combined analysis, recorded highly significant mean squares for all traits. The magnitudes of mean squares due to water regimes are higher than

Table 1 - Significance of mean squares due to different sources of line × tester combined over normal and stressed watering regimes (W.) trials for tasseling dates (T.D), silking dates (S.D), anthesis-silking interval (ASI) and yield per plot (GY plot) during 2014 season.

S.O. V	d.f	T. D	S. D	ASI	GY plot kg
Watering (W.)	1	594.3**	1014.0**	55.73**	33.67**
Geno. (G.)	48	41.5**	46.5**	0.81**	3.10**
Parents (P.)	12	52.8**	57.3**	0.96**	5.77**
P.vs. C.	1	669.2**	729.7**	1.31ns	42.57**
Crosses (C.)	35	19.7**	23.3**	0.75**	1.05**
Lines (L.)	8	18.8**	22.9**	0.60 ns	0.63**
Testers (T.)	3	64.1**	76.5**	0.68 ns	5.31**
L.xT.	24	14.4**	16.7**	0.80 ns	0.66**
G.xW.	48	1.7**	2.3**	0.53*	0.42**
P.xW.	12	1.1ns	3.2**	1.12**	1.00**
(P.vs. C.) xW.	1	45.2**	35.1**	0.64 ns	0.34 ns
C.xW.	35	0.7ns	1.0 ns	0.33ns	0.22*

ns, * and ** indicate insignificant, significant at 5% and 1% levels of probability respectively.

Table 2 -Estimates of general combining ability effects of parental maize genotypes for studied traits under normal (N) and stressed (S) conditions during 2014 summer season.

Parent	T. D		S. D		ASI		GY plot	
	N	S	N	S	N	S	N	S
I.272	-0.61**	-0.42**	-0.79**	-0.67**	-0.18**	-0.25*	0.03	0.07
I.273	0.22	0.90**	0.28	0.99**	0.06	0.08	0.19	0.41**
I.274	1.47**	1.15**	1.45 **	1.40**	-0.01	0.25	-0.17	-0.17
I.275	-0.19	-0.42	-0.29	-0.42	-0.10	0.00	-0.24*	-0.10
I.276	-0.86**	-0.84**	-0.62*	-0.75*	0.23**	0.08	0.06	-0.04
I.277	0.97**	0.90**	1.20 **	1.15**	0.23**	0.25	0.24*	-0.18
I.278	-1.4**	-1.50**	-1.37**	-1.84**	0.06	-0.33	-0.16	-0.06
I.279	0.30	-0.00	0.03	-0.09	-0.26**	-0.08	0.18	0.15
I.280	0.13	0.24	0.12	0.24	-0.01	0.00	-0.13	-0.06
I.281	0.97**	0.58*	0.93**	0.73*	-0.03	0.14	-0.54**	-0.35**
SC.10	0.86**	0.80**	1.08**	0.84**	0.22**	0.03	0.01	0.03
TWC.310	-0.25	0.02	-0.32	-0.04	-0.07**	-0.07	0.32**	0.17*
G.2	-1.58**	-1.41**	-1.69**	-1.52**	-0.11**	-0.11	0.20**	0.14

* and ** indicate significant at 5% and 1% levels of probability respectively.

those of genotypes (G.). Watering regimes (W.) seemed affected the performance of maize traits greater than genotypic differences. It recorded 14,22,69 and 11 folds higher as much as those of genotypes for tasseling date (T.D), silking date (S.D), anthesis-silking interval (ASI) and yield per plot (GY plot), respectively.

Parental genotypes either lines or testers recorded highly significant mean squares for all studied traits except for ASI. The variances due to constructed crosses are highly significant for all traits. It is worth to mention that variances due to crosses are lower in magnitude than those of their parents.

The ratio of parents to crosses mean squares are 2.7, 2.5, 1.3, and 5.5 for T.D, S.D, ASI, and GY plot, respectively. Similar higher ratios could be observed of testers to lines (3.4, 3.1, 1.1 and 8.4 for studied traits in the same order). Thus, testers seemed to be contributed more than lines in the performance of top crosses, which may be due to that the genetic variation of testers is more extensive (from inbred to synthetic) than lines (all inbreds).

The line × tester interaction combined over regimes, recorded highly significant mean squares for all studied traits except ASI. This indicates that the performance of top cross combinations varied due to each mated line and used the tester. In other words, the performance of crosses varied according to the combination of both parental genotypes.

The single degree of freedom comparison P.vs. C. showed highly significant mean squares for the tabulated traits except for ASI. The G. × W. interactions recorded highly significant mean squares for all studied traits, which indicated that the studied genotypes performed differently from water regime to another. The interaction between P. × W. recorded highly significant mean squares for all studied traits except T.D which proved that parental genotypes (either lines or testers) performed differently from watering regime to another except for T.D. However, the interaction between (P. *vs.* C.) × W. recorded highly significant mean squares for dates of tasseling (T.D) and silking (S.D). Thus, the extent of heterosis for both traits due to crossing pronounced irrespective of water treatments. The C. × W. interaction recorded highly significant mean squares only for grain yield. This indicates that the productivity of the investigated crosses varied differently between watering regimes.

General Combining Ability Effects:

Four maize genotypes recorded significant negative GCA effects (favorable) for dates to tasseling (T.D) and silking (S.D) under both watering regimes (Table 2). These genotypes included two testers (I.272 and G.2) and two lines (I.276 and I.278). Thus, these four parents may able to produce earlier hybrids when crossed with other genotypes or lines. Moreover, such a favorable effect of these parents varied in magnitude from less than one day of I.272 and I.276 to about 1.5 days of I.278 and G.2.

Four other genotypes exhibited significantly positive(unfavorable) GCA effects on flowering dates under both conditions. These genotypes included SC.10 (as tester) and three female lines (I.274, I.277, and I.281). Such parental genotypes may prolong the dates of tasseling and silking when including in cross combinations either cultivated under normal or stressed irrigation conditions. Other third four parents, included TWC 310 (used as tester) and three female lines (I.275, I.279, and I.280) seemed did not affect the flowering dates of their crosses due to insignificant GCA under both investigated regimes. The rest female parent, i.e. I.273 possessed significant positive (unfavorable) GCA effects for flowering dates only under stress conditions

Table 3 -Means of relative mid-parent heterosis (MHet) and the numbers of crosses exhibited negative (NG) and positive (PS) significant as well as insignificant (NS) heterosis of each group of test crosses under normal (N) and stressed (S) conditions during 2014 summer season

Trait	conditions	SC's				TWC's				DC's				Synthetics				Overall			
		MHet	NG	PS	NS	MHet	NG	PS	NS	MHet	NG	PS	NS	MHet	NG	PS	NS	MHet	NG	PS	NS
TD	N	-7.73*	9	0	0	-2.68*	6	0	3	-5.50*	6	0	3	-6.97*	9	0	0	-6.15*	30	0	6
	S	-6.95*	9	0	0	0.47	2	3	4	-2.46*	5	2	2	-3.63*	6	0	3	-3.54*	22	5	9
SD	N	-7.71*	9	0	0	-2.21*	4	0	5	-5.57*	6	0	3	-7.24*	9	0	0	-6.07*	28	0	8
	S	-7.66*	8	0	1	0.67*	2	4	3	-2.16*	5	2	2	-4.44*	6	0	3	-3.74*	21	6	9
ASI	N	-6.40*	1	3	5	19.48*	2	6	1	-7.56*	3	2	4	-17.52*	5	1	3	-2.37*	11	12	13
	S	-21.68*	3	0	6	4.56	2	2	5	6.45	1	2	6	-21.45*	3	0	6	-8.76*	9	4	23
GY/plot	N	45.24*	0	7	2	0.72	0	1	8	8.42*	0	2	7	9.09*	0	3	6	25.59*	0	13	23
	S	29.65*	0	5	4	13.14*	0	3	6	10.91*	0	4	5	25.04*	0	7	2	25.27*	0	19	17

* indicates significant of F1- mid-parent difference at 5% level of probability.

by about one day.

For ASI, variable GCA effects could be observed among the tested maize genotypes, particularly under normal conditions. Under regular irrigation, four genotypes (L.272, I.279, TWC.310, and G.2) recorded significant negative (favorable) GCA effects for ASI. Another three genotypes (I.276, I.277, and SC.10) recorded significantly positive (unfavorable) GCA effects on ASI under normal condition. However, under-stressed one, all tested genotypes showed insignificant GCA effects for ASI except I.272 which may be shorted the ASI interval by about 0.25 day.

For grain yield per plot, TWC.310 (as a tester) and I.281 recorded significant positive and negative GCA effects under both (normal and stressed) conditions, respectively. On the other hand, I.277 and G.2 recorded significant positive GCA effects only under normal conditions. However, I.273 possessed significant positive GCA effects only under a stressed condition. On the contrary, the I.275 inbred line recorded significant negative GCA effects only under normal irrigation condition.

Mid-Parent Heterosis:

The magnitudes and significance of mid-parent heterosis overall cross and for each group of crosses (based on testers) under normal (N), and stress (S) conditions are presented in Table 3. The numbers of crosses that recorded significantly negative (NG) or positive (PS) and insignificant (NS) mid-heterosis in each category of using I.272 (SC'), SC.10 (TWC's). TWC.310 (DC's) and G.2 (Synthetics) testers are also presented in this table.

Significant negative (favorable) overall performance of hybrids over corresponding mid-parents could be observed for flowering dates (T.D and S.D) and intervals (ASI). The magnitudes of overall heterosis were one and a half folds under normal (≈ -6.0%) irrigation as much as in stress (≈ -4.0%) condition for T.D and S.D. However, ASI under stress trial recorded four folds (-8.76 %) as much as under normal (-2.37%) condition. Out of thirty-six top crosses, thirty and twenty-eight crosses recorded significantly negative (favorable) heterosis under normal irrigated trial comparing to twenty-two, and twenty-one crosses in the stressed watering regime for TD and SD, respectively. Only five or six hybrids showed significant positive (unfavorable) heterosis under stress compared to null crosses in normal conditions for flowering dates either T.D or S.D. However, for ASI, eleven and nine crosses showed significantly favorable flowering intervals than corresponding mid-parents under normal and stressed conditions respectively. On the other hands, twelve and four hybrids recorded significantly wider ASI than their mid-parents in normal and stress conditions, respectively.

For grain yield per plot, about 25% significant superiority in performance of all top crosses over corresponding mid-parents was recorded under either normal or stress conditions (Table 3). Such grain yield heterosis of hybrids was reached to about 45 and 30% in single-cross combinations under normal and stressed conditions, respectively. However, the other three groups of crosses exhibited significant positive heterosis for grain yields much higher under stress conditions than under normal one. Surprisingly, grain yield per plot recorded overall heterosis% (25%) with pronounced % of SC's combinations (30-45%). This may be due to that heterosis was calculated relative to corresponding mid-parental performance, which performed low under both conditions. The numbers of crosses that exhibited significant positive heterosis were higher under normal than under stress conditions only in SC's combination of top crosses. However, TWC's, DC's and synthetic combinations showed higher numbers of crosses possessed significant positive heterosis under stressed than under normal one. It is worthy of mentioning that none of the investigated top crosses exhibited significantly negative heterosis for grain yield neither in normal irrigated regime nor stressed one.

Durães et al. (2002), Betran et al. (2003), Makumbi et al. (2011), Kuchanur et al. (2013), Dao et al. (2014) and Adebayo et al. (2014) found that GCA and heterosis in maize greatly influenced by the level of irrigation for most of the studied traits.

The utilization of diverse maize inbreds from as the related or unrelated genotypes which exhibited combining ability and heterotic response when crossed with genotypes for other distinct groups (Melchinger and Gumber, 1998; Dao et al. 2014 and others). They pointed out that the superiority of inter-group over intra-group crosses supported the superior identification hybrids.

The inclusion of drought-tolerant inbreds from variable gene pools will be of great benefit for the hybrid water-saving breeding program.

The investigated ten inbred lines descended to 3 different origins, G.2, Tep.5, and American new dent (A.E.D). Four of which (I.272, I.273, I279, and I.280) were developed from the synthetic variety Giza 2, and other four lines (I.274, I.275, I.276 and 281) were prolonged to Tep.5 population. The remainder two lines, i.e. I.277 and I.278 traced to A.E.D. Despite the relativeness between I.272 and G.2 as testers and 3 of female lines (I.273, I.279, and I.280), the obtained data proved that selection practiced along with selfing during the development of these inbreds reflected in genetic divergence. The obtained data under regular and stressed irrigation as well line × tester analyses proved that this collection possessed encouraging potentiality for enrolling in maize hybrid breeding program. GCA effects, as well as the extent of heterosis, supported these arguments. Judging by the estimates of GCA, G.2, and I.272 as testers exhibited favorable combining abilities for flowering dates and intervals in addition to grain yield than SC.10 as tester under both conditions. The TWC.310 showed significant GCA only for GY under both investigated trials. The tested female inbreds exhibited variable degrees of GCA for T.D, S.D, ASI, and GY under both investigated conditions.

It may be concluded that the utilization of narrow genetic base tester (I.272) resulted in favorable heterosis of crosses for earliness (TD & SD) by about 7% accompanied with shorter ASI and positive grain yield by more than 29.0% under normal and stress condition. Normal heterosis was obtained by using the broad genetic base G.2 as a tester compared that obtained by using medium genetic base (SC.10 or TWC.310). Narrow genetic base testers could be recommended in advanced generations of homozygosity, whereas mostly based ones may be valid for screening during the first generations of selfing.

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