Detection of Genetic Components Through Triple Test Cross and Line X Tester Analysis in Bread Wheat

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Abstract: Studies were conducted in 2002/03,2003/04 and 2004/05 to detect epistasis and to estimate genetic components for five quantitative traits (days to heading, plant height (cm), number of spikes per plant, 100-grain weight (g) and grain yield per plant(g) using triple test cross analysis and to determine the superior parents and hybrid combinations in respect to grain yield and its components through line x tester analysis. Ten bread wheat varieties were crossed with three testers. Results revealed that significant epistasis is present for number of spikes per plant, 100-grain weight, plant height and days to heading. Additive × additive epistatic type was found to be much larger in magnitude than additive × dominance and dominance × dominance (J + L) epistatic types for grain yield per plant, number of spikes per plant and 100-grain weight. Both additive (D) and dominance (H) genetic components play an important role in the inheritance of number of spikes per plant, plant height and days to heading. The average degree of dominance $(H / D)^{1/2}$ was in the range of partial dominance for all traits studied. Line × tester analysis revealed that the nature of gene effects were predominantly non-additive for all traits studied. Three wheat varieties Jup / Biy, Giza – 164 and Sids 4, exhibited significant GCA effects for number of spikes per plant. The tester cultivar Giza 168 was at the top of GCA effects for yield and its components. The three -way superior crosses (F1 with each of Sids 4, Sids 9, Kauz/Ban, Cham4/Teve2, Prinia and Giza 164) showed the highest performance of their progenies and high values of selection index for grain yield and the most contributing characters.

Key words: Additive · combining ability · dominance · epistasis · line x tester · selection index · triple test cross *Triticum aestivum* L.

INTRODUCTION

Information of the type of gene action involved in the inheritance of a character is helpful in deciding the breeding procedures to be followed for plant improvement and is necessary for efficient utilization of available germplasm in a plant breeding programe.

In the genetic analysis, presence of epistasis is known to cause bias in the genetic components of variance [1]. Most of the genetic studies carried out by wheat breeders [2-7] demonstrated that three epistatic types (i.e. additive x additive, additive x dominance and dominance x dominance) of gene action were important in the inheritance of quantitative traits in bread wheat. Although, the designs used in the estimation of these genetic components assume the absence of epistasis.

Therefore, the presence of epistasis should be studied precisely before deciding any breeding programe. Among all the designs available for estimation of gene action, triple test cross is considered the most efficient model as it provides not only a precise test for epistasis, but also unbiased estimates of additive and dominance components if epistasis is absent [8].

Simmonds [9] emphasized the need to screen parents and crosses before their use in breeding program and suggested that combining ability analysis based on progeny test data is a useful method for evaluating parents and crosses for a wide range of quantitative characters.

The objectives of this study are to establish, 1- The existence of epistasis and to determine the additive (D) and dominance (H) variances conditioning quantitative traits using the triple test cross analysis. 2- Estimates of general and specific combining ability using line x tester analysis and selection index to select excellent parents and superior crosses before using in future breeding programe. 3- Broaden the genetic germplasm base in bread wheat by producing large hybrid progenies (single and three way crosses) and select the superior combinations for developing new breeding programes.

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MATERIALS AND METHODS

Three field experiments were conducted at the Experimental Research Station of the National Research Center at Shalakan El-Kalyoubia Governorate during the three successive seasons 2002/03, 2003/04 and 2004/05.

In the first season, two high yielding wheat cultivars, which showed wide adaptation in winter season and resistance to leaf rusts in Egypt [10] and difference in most agronomic traits, Gemmiza 9(P1) and Giza 168 (P2), were crossed to obtain their F1 (Gemmiza 9 x Giza 168) and hereafter used as three testers.

During 2003/04 season, each of the three testers P1, P2 and their F1 were crossed with ten divergent origin bread wheat lines, four from CYMMIT, Kauz/Fct, Kauz/Attila, Jup/Bjy and Tsh/Dove /Kauz ; three from ICARDA, Prinia, Cham4/Teraz and Bow/Mn 78131 and three Egyptian varieties Giza 164, Sids 4 and Sids 9, to produce 30 crosses i.e. 10L1, 10L2 and 10L3 progeny families of triple test cross design. The cross (P1 X P2) was also repeated to get fresh F1 grains.

All plant materials, the thirty families (crosses), ten parents and the three testers, were grown in a Randomized Complete Block Design with three replicates in 2004/2005. Each progeny family were grown in a 3 m long row. The spacing between (row to row distance) and within (plant to plant distance) rows were maintained at 30 and 10 cm, respectively. All the normal agronomic practices were followed as usual in the ordinary wheat field in the area of study. Data were scored on fifteen competitive plants from each row in each replications for the five characters, days to heading, plant height (cm), number of spikes per plant, 100-grain weight (g) and grain yield per plant(g).

Statistical analysis: The procedure followed for detecting epistasis was done according to the method outlined by Kearsey and Jinks [11]. The analysis is based on the following model,

$$Lijk = M + GIJ + rk + eijk$$
where;

- Lijk : Denotes the phenotypic value of the cross between tester Li and line J in replication k.
- M : Denotes the mean of all single and three way crosses
- Gij : Denotes the genotypic value of the cross between tester Li and line j
- Rk : Denotes the effect of replication K and

- Eijk : Denotes the error associated with the cross ij in replication k.

Data of the triple test cross families (L1i, L2i and L3i) were firstly subjected to the conventional analysis of variance to test the significance between families. The mean squares of deviations L1i + L2i - 2L3i (overall epistasis) were tested against pooled error to determine the presence of epistasis.

The overall epistasis was partitioned into (I) type of epistasis (additive x additive) and (J and L) types of epistasis i.e. additive x dominance and dominance x dominance; respectively. The estimation of additive (D) and dominance (H) genetic components and the correlation coefficient (r) between sums L1i + L2i and differences L1i - L2i were obtained to detect the direction of dominance, according to Jinks and Perkins [12]. Average degree of dominance was calculated as (H/D)1/2. The data were also subjected to line x tester analysis and selection index using methodology which is fully described by Singh and Chaudhary [13].

RESULTS AND DISCUSSION

The analysis of variance for all traits studied are presented in Table 1. Genotypes, hybrids and parents mean square estimates were found to be highly significant for all traits studied, indicating the presence of variability among hybrids and their parents. Hybrids Vs parents mean square estimates, as an average heterosis overall crosses, were found to be highly significant for all traits except days to heading. Also, data given in Table 1 indicated highly significant mean squares for lines for all traits studied. Testers mean squares were found to be highly significant for all traits except grain yield per plant.

Lines Vs testers mean squares were highly significant for all traits except days to heading. The mean performance of the two parents P1(Gemmiza 9)Vs P2 (Giza 168) were significantly different from each other in all traits except grain yield per plant. The unbiased estimates of additive and dominance gene action and the unambiguous test of epistasis would only be achieved when the testers are different from each other. However, when this condition of difference between two parents is not met, the estimates are biased to an unknown extent [11, 14].

The estimates of general (GCA) and specific (SCA) combining ability variances Table 1 revealed that the nature of the gene effects was predominantly non-additive for all traits under investigation.

Table 1: Mean squares of the analysis of variance of (L1i, L2i and L3i) triple test cross hybrids and line x tester analysis for all traits studied

Source of variance	D.F.	Grain yield per plant (g)	No. of spikes per plant	100-grain weight (g)	Plant height (cm)	Days to heading
Replications	2	4.70	0.209	0.008	1.02	8.25
Genotypes	42	53.11**	5.83**	0.839**	216.05**	67.90**
Hybrids	29	39.85**	3.53**	0.313**	133.55**	26.42**
Parents	12	69.15**	9.77**	0.969**	399.60**	173.44**
Hybrid Vs.Par.	1	245.13**	25.05**	14.51**	405.91**	4.35
Lines	9	80.18**	11.53**	0.899**	461.20**	212.95**
Testers	2	31.05	4.40**	1.63**	307.11**	82.11**
LineVs Testers	1	46.04**	4.71**	0.267**	30.21**	0.578
P1 Vs P2	1	6.51	4.27**	1.62**	300.0**	75.0**
Error	84	12.48	0.669	0.029	4.515	1.703
S^2 GCA		0.373	0.027	0.003	0.838	0.335
S^2 SCA		2.48	0.479	0.049	28.08	2.26
GCA/ SCA		0.150	0.056	0.061	0.029	0.148

*, ** Significant at 0.05 and 0.01 probability levels, respectively

Table 2: Analysis of variance for testing the presence of epistasis in a triple test cross for all traits studied

Source of variance	D.F.	Grain yield per plant (g)	No. of spikes per plant	100-grain weight (g)	Plant height (cm)	Days to heading
Total of pistasis	10	241.46	24.872*	1.904**	870.26**	60.80**
I types epistasis	1	847.48**	155.36**	6.57**	448.53**	4.80
J + L epistasis	9	174.13	10.37	1.38**	917.13**	67.02**
I- type epistasis x block	2	220.78	3.01	0.008	31.03	1.60
J + L epistasis x block	18	144.05	9.99	0.2395	14.52	8.82
Total epistasis x block	20	151.73	6.59	0.216	16.16	8.10

*,** Significant at 0.05 and 0.01 probability levels, respectively   (I) = additive x additive,   (J) = additive x dominance,   (L) = dominance x dominance

The analysis of variance for testing the presence of epistasis in the inheritance of all traits studied is presented in Table 2. The mean square for the deviations L1i + L2i - 2L3i revealed presence of significant epistasis for days to heading, plant height, number of spikes per plant and 100- grain weight. Partitioning of the total epistatic effect revealed the presence of highly significant additive x additive (I) type of epistasis for all characters except days to heading. Also, estimates of additive x dominance and dominance x dominance, J+L type of epistasis mean square were highly significant for 100- grain weight, plant height and days to heading.

The additive x additive epistatic type (I) was found to be much larger in magnitude than additive x dominance and dominance x dominance (J+L) epistatic types for grain yield per plant, number of spikes per plant and 100-grain weight, indicating that fixable components of epistasis were more important than non-fixable one in the inheritance of these traits. These results would ascertain the results previously obtained from the line x tester analysis, Table 1.

In a self pollinated crop such as wheat, where the production of commercial hybrids is still far to be relized, the fixable component of epistasis (I) type could be easily exploited in evolving homozygous cultivars by following hybridization and selection procedures. Most of the new Egyptian wheat cultivars developed following the principles of pedigree method, Mossad et al. [10]. Goldringer et al. [15] reported that, epistasis could be expected to be quite important in self pollinated crops because of selection history on a fixed genome with only few recombination events, while the second type of epistasis (J+L) is useful in the development of hybrids.

The results obtained here revealed the importance of epistasis in the inheritance of most characters in wheat. Moreover Eissa [4], Comber [5], El - Nahas [7], Singh et al. [16], Singh and Nanda [17] and Singh et al. [18] found similar results.

Detection of genetic variance components: The mean squares due to sums (L1i + L2i) were found to be highly significant for all traits except 100- grain weight Table 3,

Table 3: Mean square from analysis of variance for sums and difference and estimates of additive (D),dominance (H) and degree of dominance in triple test cross
for all traits studied

Source of variance	D.F.	Grain yield per plant (g)	No. of spikes per plant	100-grain weight (g)	Plant height (cm)	Days to heading
Sums ($L_{1i}+L_{2i}$)	9	101.49**	6.56**	0.798	446.31**	96.79**
Error	18	21.68	1.77	0.818	13.83	3.04
Difference ($L_{1i}-L_{2i}$)	9	21.69	4.96**	0.243**	49.33**	11.57**
Error	18	19.37	0.858	0.061	5.81	2.84
D	-	106.41**	6.38*	0.954	576.63**	124.99**
H	-	3.09	5.48**	0.243**	58.03**	11.64**
(H/D) ½	-	0.17	0.93	0.51	0.32	0.31
R	-	0.205	0.372	0.534*	- 0.449	- 0.656**

*,** Significant at 0.05 and 0.01 probability levels, respectively,   (r) = Correlation coefficients between sums ($L_1 i + L_2 i$) and differences (L_{1i}- L_{2i})

Table 4: General combining ability effects and selection index (SI)of the parental lines and testers for all traits studied.

Parental lines	Grain yield per plant (g)	No. of spikes per plant	100-grain weight (g)	Plant height (cm)	Days to heading	SI
Kauz/ Fct (L1)	-2.75	-0.78	-0.26	-7.08**	2.26	32.68
PRINTIA (L2)	-0.97	-0.27	0.14*	-6.37**	2.15	27.79
Giza. 164 (L3)	0.74	0.43	-0.13	4.30	0.71	35.27
Sids -9(L4)	3.59**	-0.52	0.43**	-2.81**	-3.18**	19.41
Cham4/ Teve2(L5)	2.27	0.14	-0.22	-0.92	-0.18	30.40
Tsh/ Dove (L6)	-5.41	-0.75	-0.08	9.30	0.71	33.58
Jup/ Bjy (L7)	-0.92	0.90**	-0.05	4.52	0.71	31.23
Bow/ Mn 7 2131 (L8)	-0.26	-0.10	0.11**	-1.03	1.49	32.13
Kauz/ Ban (L9)	0.89	0.27	0.19**	0.63	-0.18	27.35
Sids -4 (L0)	2.81*	0.68*	-0.14	-0.58	-4.51**	21.75
C. Testers Gimmeza 9 (p1)	-1.89	-0.81	0.05	1.02	1.96	29.23
Giza. 168 (p2)	1.77	0.76**	0.16**	1.29	-0.13	31.23
Gim9 x G168 (F1)	0.12	0.05	-0.21	-2.31**	-1.83**	32.78
L.S.D (lines) 0.05	2.34	0.54	0.11	1.41	1.71	
0.01	3.11	0.72	0.15	1.87	2.27	
L.S.D (testers) 0.05	1.28	0.29	0.06	0.77	0.94	
0.01	1.70	0.39	0.08	1.02	1.25	

*, ** Significant at 0.05 and 0.01 levels of probability, respectively

indicating the presence of additive genetic variance for grain yield per plant, number of spikes per plant, plant height and days to heading. The mean squares due to difference (L1i-L2i) were highly significant for all traits except grain yield per plant, indicating the importance of dominance genetic variance for these traits.

The estimates of additive (D) and dominance (H) components Table 3, indicated that both additive and dominance (H) components play an important role in the inheritance of number of spikes per plant, plant height and days to heading. The (D) component was significant for grain yield per plant, while the (H) component was significant for 100-grain weight. However, the additive components were larger than dominance for all traits studied and that would ascertain the previous results obtained by line x tester analysis.

Consequently, it could be concluded that selection procedures based on accumulation of additive effects would be successful in improving all traits studied. However, to maximize selection advance, procedures which are known to be effective in shifting gene frequency when both additive and non-additive genetic variances are involved would be preferred. The same results were also obtained by Eissa [4], Comber [5], Esmail [6] , El -Nahas [7], Singh *et al.* [16] and Nanda *et al.* [19]. The degree of dominance (H/D)1/2 was less than unity for all traits studied suggesting the role of partial dominance in the inheritance of these traits and ascertain the fact that in self pollinated crops,most genes are homozygous and the over-dominance is rare. Genetic advance in genetic systems with over-dominance and epistasis are slower than when gene effects are purely additive or partially dominant [20].

Table 5: Specific combining ability effects and selection index (SI) of different crosses for all traits studied.

Crosses	Grain yield per plant (g)	No. of spikes per plant	100-grain weight (g)	Plant height (cm)	Days to heading	SI
Gim.9(p1) X L1	-0.312	0.061	-0.011	5.860	0.033	29.010
X L2	-0.099	-0.396	-0.035	-1.130	0.144	30.650
X L3	4.680*	0.941*	0.022	0.867	-0.740	36.010
X L4	-1.110	0.167	0.203*	3.640	-1.190	33.590
X L5	-0.440	0.366	-0.361	-0.578	0.478	32.230
X L6	-1.520	-1.084	-0.037	-0.800	-0.078	26.070
X L7	-0.820	1.268**	0.009	2.640	0.922	34.960
X L8	0.951	0.131	0.222*	0.533	-0.189	32.970
X L9	-0.920	-0.922	0.119	-6.130**	-0.189	31.370
X L10	-0.404	-0.461	-0.131	-4.910**	0.811	32.600
F1(Gim.9xG.168) XL1	0.068	-0.172	0.055	-8.070**	-2.870	33.110
X L2	-0.003	-0.129	0.088	1.270	-1.420	35.650
X L3	-4.220	0.341	-0.255	-0.400	-0.980	35.410
X L4	-0.879	0.554	0.060	1.380	1.580	38.250
X L5	1.039	-0.427	0.352**	1.490	-0.756	37.150
X L6	1.577	0.933	-0.044	-4.400**	0.356	34.870
X L7	-1.312	-0.845	-0.395	-7.960**	-0.311	34.510
X L8	-2.733	-0.962	-0.052	-0.400	0.578	33.400
X L9	1.526	0.201	-0.105	10.270	1.578	38.040
X L10	4.940*	0.506	0.295**	6.820	2.240	41.310
G.168 X L1	0.243	0.111	-0.044	2.200	2.830	30.850
X L2	0.102	0.524	-0.054	-0.133	1.280	33.880
X L3	-0.459	-1.283	0.233*	-0.467	1.720	32.420
X L4	1.989	-0.720	-0.263	-5.020**	-0.390	33.810
X L5	-0.596	0.062	0.009	-0.911	0.278	34.070
X L6	-0.061	0.152	0.081	5.200	-0.278	30.590
X L7	2.133	-0.423	0.336**	5.310	-0.611	35.330
X L8	1.782	0.830	-0.171	-0.133	-0.389	35.250
X L9	-0.602	0.791	-0.014	-4.130**	-1.389	35.880
X L10	-4.530	-0.045	-0.164	-1.910	-3.060*	33.380
L.S.D. 0.05	4.060	0.939	0.197	2.440	2.970	---
L.S.D. 0.01	5.380	1.250	0.261	3.240	3.940	---

*, ** Significant at 0.05 and 0.01 levels of probability, respectively

Further, the correlation coefficient between the sums (L1i + L2i) and differences (L1i -L2i) were found to be positive and significant for 100 grains weight indicating that dominance seemed to be acting in one direction. On the contrary, the correlation was negative and significant for days to heading suggesting increasing type of genes are dominant. However, the correlation coefficient for the remaining traits were insignificant indicating the genes with positive and negative effects were equally distributed among the genotypes including in this investigation.

Estimates of general combining ability (GCA) effects of the ten parental lines and three testers are presented in Table 4. High positive values of GCA effects would be of interest in all traits studied except plant height

and days to heading, high negative value would be preferred from the wheat breeder point of view. Results revealed that the two long spike varieties (Sids 4 and Sids 9) and Giza 168 were good general combiners for grain yield per plant. Three wheat varieties Jup/Bjy, Giza 164 and Sids 4 exhibited significant GCA effects for number of spikes per plant proving to be good combiners for this trait. Five parental lines showed good combiner for 100-grain weight. KAUZ/Fct, PRINIA and Sids 9 giving significant negative GCA effects for plant height proving to be good combiners for wheat breeding to develop dwarf or semi-dwarf cultivars. For days to heading the best general combiner, Sids 4, Sids 9 and Giza 168 exhibited highly significant negative GCA effects. The tester cultivar Giza 168 was at the top of GCA effects

for yield and its components and followed by Giza 164 and Sids 4.

Selection index, is a statistical tool, which enables the wheat breeder to discriminate and select the superior genotypes would be exploited in the future breeding program based on selection of yield characters simultaneously using phenotypes performance. Results of selection index of the parental lines and testers revealed that Giza 164 followed by Tsh/Dove and KAU Z/Fct, have the highest value of selection index. Among the testers Giza 168 and the F1(Giza 168 x Gemmiza 9) possessed high selection index.

Once identified the best parental combiners can be crossed to identify optimal hybrid combinations or hybridized with the intent of selecting promising genotypes within the segregating generations. In recurrent selection schemes, parents possessing high combining ability can be crossed with one another to accumulate desirable alleles within a base population [21].

Estimates of specific combining ability effects (SCA) of the thirty crosses for all traits studied are presented in Table 5. Significant positive SCA effects were observed in two crosses (Gemmiza 9 x Giza 164) and (Sids 4 x F1) for grain yield per plant, two single crosses for number of spikes per plant, six crosses for 100-grain weight. However, highly significant negative SCA effects were obtained in seven crosses for plant height and one cross for days to heading. Therefore, most of the previous crosses showed high positive SCA effects for yield and its components and high negative values for plant height and days to heading could be exploited in wheat breeding programs.

Among the hybrid combinations, six of ten three way crosses and four of the twenty single crosses were selected Table 5. These three-way superior crosses (F1 with each of Sids 4, Sids 9, Kauz/Ban, Cham4/Teve2, Prinia and Giza 164) showed the highest performance of their progenies and high values of selection index for grain yield and the most contributing characters, indicated that better homozygous progenies would be derived from the three -way crosses as compared to the single crosses. Moreover some of these crosses exhibited high SCA effects for yield characters. Therefore, it is more advantageous to attempt the three-way crosses in future wheat breeding program to improve yield and its components.

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