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Genetic Analysis and RAPD Polymorphism in Some Durum Wheat Genotypes

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Abstract: Three Mexican and two Egyptian durum wheat genotypes were crossed using scaling test analysis during 2005/2006, 2006/2007 and were evaluated in 2007/2008 season at Assiut. Results showed that A, B and C scaling test were highly significant for most studied traits. Meanwhile, the additive-dominance model was adequate to demonstrate the genetic variation and it important in the inheritance of no. of spikes/plant and grain yield/plant. Both additive and dominance parameters were important in the inheritance of most studied traits. But the dominant component was the largest in magnitude in most crosses for most traits studied. The epistatic effects additive x additive [i] and dominance x dominance [l] were highly significant in most cases. The additive component (D) was greater than the dominance (H) for no. of spikes/plant in cross no. 1(Bani-Sweef-1 X Line-2) and for grain yield/plant in crosses 1(Bani-Sweef-1 X Line-2) and 3 (Sohag-3 X Line-4). Heritability values for no. of spikes/plant were low in crosses no. 1(Bani-Sweef-1 X Line-2) and 2(Sohag-3 X Line-3) and were moderately high for grain yield/plant in crosses no. 1(Bani-Sweef-1 X Line-2) and 3 (Sohag-3 X Line-4). Heterosis values over both mid-parent and the better parent were found for all studied traits. The inbreeding values were significant and coupled with a reduction in the mean of the F_2 generation for all studied traits. The genotypic (GCV) coefficient of variability was relatively low for all studied traits. DNA of eleven wheat genotypes (five parental genotypes, three F_1 crosses and three F_2 populations) was amplified with eight different random primers. Eight RAPD primers detected 129 fragments and 91 of them (70.125%) were polymorphic. Eleven wheat genotypes were grouped into two clusters using dendrogram analysis. The results of similarity indices were compared with those of genetic variation for the parents combined into three crosses. The parental with the lowest similarity indices had the highest score for genetic variation in most of the morphological and agronomical traits.

Key words: Durum wheat · Gene action · Genetic diversity · RAPD markers

INTRODUCTION

Wheat is the most important cereal crops; it is a stable diet for more than one third of the world population and contributes more calories and protein to the world diet than any other cereal crop. Nowadays, in Egypt there is an urgent need to increase the productivity level of wheat to reduce the food gap resulting from population increase. The breeders have to develop a new set of varieties with higher production. The true knowledge of the gene action for various durum wheat traits is useful in making decisions with regard to appropriate breeding system .

Singh *et al*. [1] showed that the additive gene effect was significant for heading date and plant height. Meanwhile, Khalifa *et al*. [2] found that both additive and dominance effects were important in the inheritance of flag leaf area, days to heading, days to maturity, plant height and grain yield/plant. Khalifa *et al*. [3] found that dominance gene effects were played the major role in controlling the genetic variation in the biological yield/plant. The study conducted by Amein [4] on six population of durum wheat found that the additive gene effects were played the major role in controlling the genetic variation in the plant height, no. of spikelets/spike and no. of spikes/plant. Meanwhile, the dominance gene

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effects were played the major role in controlling the genetic variation in the days to flowering, 1000-grain weight and grain yield /plant. Sallam [5] reported that the importance of both additive and dominance gene effects in the inheritance of 1000-grain weight, however the dominance gene effects were more than in magnitude in favorable condition.

Conventional breeding has accomplished a remarkable success in development of high yielding varieties. However, use of other non-conventional approaches may further accelerate the progress of such breeding programs. RAPD marker analysis provides virtually unlimited number of markers to compare individual genotypes and considering easy handling and cheaper cost per assay, it is possible to carry out large scale screening of breeding populations and genetic resources [6]. The present study aims to determine the types of gene action effects controlling morphological traits, yield and its components, as well as estimating heterosis, heritability, inbreeding depression and genotypic variability coefficient (GCV) of the studied traits. Moreover, the study aims to detect the genetic variation of the wheat genotypes under study using RAPD-PCR marker technique.

MATERIALS AND METHODS

The present study was carried out during the period of 2005/2006,2006/2007 and 2007/2008 growing seasons, at the Experimental Farm of the Faculty of Agriculture, Al-Azhar University, Assiut, Egypt. Five durum wheat (*Triticum turgidum*, sub species durum) genotypes were chosen for this study on the basis of their origin diversity. The origin and pedigree of these genotypes are presented in Table 1. In 2005/2006, the genotypes were sown and three crosses were made to produce F1 hybrid seeds and the crosses between the parental genotypes are presented in Table 2. In 2006/2007, crossing was made between the F_1 hybrids of each cross and its two respective parents to produce the first (F_1XP_1) and second (F_1XP_2) backcrosses (BC_1 and BC_2). At the same time, crossing was made among the parents to produce F_1 seeds. Some F_1 hybrids were selfed to produce the F_2 generation. In 2007/2008, the six basic generations (P_1, P_2, F_1, F_2, BC_1 and BC_2) of each of the three crosses were sown in a randomized complete block design with three replications. Each replicate consisted of one row of each of the parents and F_1's, two rows of each back-cross and three rows for the F_2 populations. Rows were 3 m long and 30 cm apart and 5 cm between plants. The recommended field practices for wheat production were adopted all over the growing seasons. Data were recorded on individual plant basis as follows, flag leaf area (cm), days to 50% heading, days to 50% maturity, plant height (cm), no. of spikes/plant, no. of spikelets/spike, biological yield (gm) per plant, which is the total biomass produced by the plant during the season (excluding the roots), 1000-grain weight (gm) and grain yield/plant (gm).

Table 1: The entry name, pedigree and source of the five durum wheat genotypes

No.	Genotype	Pedigree	Origin
1	Bani-Sweef-1	Jo "S"/ AA "S"// FG- BITTERN "S" CD 21831- 2Sh-1Sh-oSh	Egypt
2	Sohag-3	MEXI "S"/ MGHA / 51792 // Durum 6 CD 9799	Egypt
3	Line-2	JABUL / 5GDVZ 512 / CIT // RUFF / FG / 4 / BY* 2 / TOB // AA / 3 / TEL CD 3087 – 1sd -1sd – 1sd – osd.	Mexico
4	Line-3	CRONOS CD 3094 – 9sd – 1sd -1sd –osd.	Mexico
5	Line-4	BOOMER – 241AJ AIA -9 // ACO89 CD 3103 -1sd – 1sd -1sd – osd.	Mexico

Table 2: Crosses established between the five durum wheat genotypes

Cross No	Cross	Cross name
1	P_1 X P_3	Bani-Sweef-1 X Line-2
2	P_2 X P_4	Sohag-3 X Line-3
3	P_2 X P_5	Sohag-3 X Line-4

DNA Extraction and RAPD Amplification Conditions: Leaves were obtained from 14 days old plants and ground to a fine powder in liquid nitrogen. The genomic DNA was extracted using CTAB method [7]. RAPD analysis was performed using eight 10-mer random primers (Table 1) procured from Operon Technologies Inc. (Alabameda, CA).

PCR reaction was used in a final volume of 25 µl containing 12.5 µl of Master Mix (Fermentas),2.5 µl of 5 µM of each primer, 50 ng of template DNA. Reactions were performed in a thermocycler (B iometra T1, G mbH). RAPD-PCR was performed according to Williams *et al.* [8] as one cycle of 95°C for 5 min (denaturation), 36 cycles of {94°C for 1 min, 36°C for 1 min and 72°C for 1 min (annealing)} and a final extension of 2 min at 72°C. PCR products were analyzed using 1% agarose gel electrophoresis and visualized with ethidium bromide staining. The sizes of the fragments were estimated based on a DNA ladder of 100 bp (MBI, Fermentas).

Statistical and Genetic Analysis: The analysis of variance of the six basic generations (P_1, P_2, F_1,F_2, BC_1 and BC_2) were statistically analyzed using (RCBD) analysis of variance. The scaling tests (A, B and C) were calculated for each trait to detect the adequacy of the additive-dominance model or the presence of non-allelic gene interaction according to Mather and Jinks [9]. The six parameters genetic model (m, d, h,i, j and l) were computed according to Jinks and Jones [10] as follows, [m]=mean, [d]=additive effect $= \overline{BC_1} - \overline{BC_2}$, [h] = dominance effect $= \overline{F_1} - 4\overline{F_2} - ½\ \overline{P_1} - ½\ \overline{P_2} + 2\overline{BC_1} + 2\overline{BC_2}$, [i]=additive x additive type of gene interaction $= 2\overline{BC_1} + 2\overline{BC_2} - 4\overline{F_2}$, [j]=additive x dominance type of gene interaction $= \overline{BC_1} - ½\ \overline{P_1} - \overline{BC_1} + ½\ \overline{P_2}$ and [l] = dominance x dominance type of gene interaction $= \overline{P_1} + \overline{P_2} + 2\overline{F_1} + 4\overline{F_2} - 4\overline{BC_1} - 4\overline{BC_2}$. Whenever the additive-dominance model proved to be adequate, the phenotypic variance for each character was partitioned into additive (D), dominance (H) and environmental (E) using Mather and Jinks [9] as follows, $E = 1/3\ (V_{\overline{F1}} + V_{\overline{F2}} + V_{\overline{F1}})$, $D = 4\ V_{\overline{F2}} - 2(V_{\overline{BC_1}} + V_{\overline{BC_2}})$, $H = 4(V_{\overline{F2}} - ½\ V_D - V_E)$ and $h^2_{(NS)} = \frac{½\ D}{½\ D + ¼\ H + E} \times 100$. The T test was performed as follows, $\pm T = \frac{\text{effect}}{\sqrt{\text{variance of effect}}}$

Estimates of heterosis (%) were calculated as the percent deviation of F_1 mean performance over that of either better or mid parent as follows,

Heterosis from the better-parent,

$$H(\overline{B.P})\% = \frac{\overline{F_1} - \overline{B.P}}{\overline{B.P}} x100$$

Heterosis from the mid-parents,

$$H(\overline{M.P})\% = \frac{\overline{F_1} - \overline{M.P}}{\overline{M.P}} x100$$

Inbreeding Depression: Its values were measured from the following equations,

Inbreeding depression of $F_1 = \frac{\overline{F_1} - \overline{F_2}}{\overline{F_1}} x100$

$$t.I.D = \frac{\overline{F_1} - \overline{F_2}}{\sqrt{V.I.D}}$$

Estimation of Genotypic Coefficient of Variability: The genotypic Variability Coefficient (GCV) calculated according to Singh and Chaudhary [11] as follows, $GCV = \frac{\sqrt{V\overline{F_2} - V\overline{E}}}{\overline{F_2}}$.

RAPD Data Analysis: RAPD data were scored for presence (1), absence (0). Cluster and genetic similarity analyses were performed using SIMQUAL; SAHN and TREE for NTSYS-pc ver 2.10 (Applied Biostatics, Setauket, New York, USA). UPGMA; WPGMA; Complete-link and Singel-link were applied in all possible combinations with the similarity coefficients, Jaccard and simple matching [12].

RESULTS AND DISCUSSION

Genetic Analysis of Quantitative Characters: The mean characters of the six generations for each of the three crosses are presented in Table 3. The results indicated that means of the F_1's were higher than either the highest parent or mid-parent value, indicating over-dominance or partial dominance, respectively towards the respective parents for all studied traits. Similar results were obtained by Khalifa *et al.*[2], Khalifa *et al.* [3] and Menon and Sharma [13].

The results of the A, B and C scaling tests for assessing the validity of additive-dominance models are given in Table 4. The non-allelic interaction was found to be operating in the control of genetic variation among the six generations for most studied traits. Meanwhile, crosses no. 1 and 2 for no. of spikes/plant and crosses no. 1 and 3 for grain yield/plant, the values of the A,B and C scaling tests were not significant, indicating the absence of non-allelic interaction and the additive-dominance model was adequate to demonstrate the genetic variation and it is important in the inheritance of the two studied traits in such crosses. These results are in agreement with those obtained by Khalifa *et al.* [3], Raghavaiah and Joshi [14] and Bakheit *et al.*[15].

The six parameters of gene effect conducted by using the population means are presented in Table 4. The mean effect [m] was highly significant for all studied traits, except cross no. 1 for grain yield /plant, indicated that all studied traits were quantitatively inherited. Both additive [d] and dominance [h] parameters were significant or highly significant in most crosses for most studied traits, indicating that both additive and non-additive effects were important in the inheritance of all studied traits. Similar results were found by Amein [4] and Parakash and Joshi [16]. The dominance parameter [h] showed the largest in magnitude in most crosses for most studied traits, indicating that dominance gene effects play the major role in controlling the genetic variation of the most studied traits. With regard to the negative value of [h] observed for some studied traits indicated that the alleles responsible for less value of traits were dominant over the alleles controlling high value. Meanwhile, the absence

Table 3: Mean performance of parents, F_1, F_2 and backcross generations in three durum wheat crosses for all studied traits

	Cross 1 Mean±S.E	Cross 2 Mean±S.E	Cross 3 Mean±S.E	Cross 1 Mean±S.E	Cross 2 Mean±S.E	Cross 3 Mean±S.E	Cross 1 Mean±S.E	Cross 2 Mean±S.E	Cross 3 Mean±S.E
Generation	Flag leaf area			Days to 50% heading			Days to 50% maturity		
P_1	24.99±1.47	24.10±1.07	24.10±1.07	88.86±0.71	88.50±0.70	96.14±1.15	132.42±0.25	133.86±0.30	136.50±0.90
P_2	18.38±0.63	21.05±1.13	23.60±1.55	91.76±0.46	96.14±1.15	99.61±0.57	141.25±0.96	138.47±1.30	138.47±1.30
F_1	34.01±1.27	28.81±1.58	37.71±1.70	89.50±0.13	92.15±0.42	97.55±0.28	134.89±0.33	132.17±0.41	134.00±0.54
F_2	31.70±0.89	28.11±0.68	35.77±1.24	93.64±0.31	93.63±0.40	98.43±0.18	139.40±0.33	140.20±0.23	140.44±0.55
BC_1	37.81±0.26	42.21±0.59	29.90±0.67	98.32±0.49	92.32±0.49	96.28±0.44	148.47±0.46	137.00±0.24	139.50±0.18
BC_2	35.98±0.43	33.99±0.46	27.93±0.64	99.09±0.25	95.78±0.53	96.28±0.75	149.67±0.32	147.55±0.22	151.20±0.25
LSD5%	1.85	1.93	3.10	0.77	2.24	0.89	1.07	1.02	1.24
	Plant height			No.of spikes/plant			No.of spikelets/spike		
P_1	93.64±1.02	93.17±1.33	90.27±1.12	8.00±0.34	7.33±0.28	7.62±0.39	22.30±0.34	20.65±0.34	20.95±0.36
P_2	93.63±0.65	88.97±1.26	88.97±1.26	6.24±0.30	7.10±0.34	7.10±0.34	20.91±0.31	19.56±0.36	20.65±0.43
F_1	94.88±0.77	104.13±1.35	93.45±1.30	8.67±0.23	9.22±0.25	9.08±0.30	21.96±0.23	22.81±0.50	21.52±0.31
F_2	94.62±0.82	94.02±1.45	89.48±1.11	8.19±0.24	8.27±0.25	7.100.26±	19.85±0.42	22.61±0.51	21.23±0.56
BC_1	99.47±0.73	91.18±0.58	97.36±1.03	8.49±0.24	7.93±0.21	7.26±0.27	19.88±0.31	23.36±0.45	23.81±0.45
BC_2	98.94±0.69	85.00±1.05	88.77±1.03	7.40±0.20	7.59±0.27	6.93±0.20	18.76±0.24	22.23±0.55	21.74±0.46
LSD5%	0.80	4.38	2.12	0.25	0.19	0.32	0.92	1.45	1.22
	Biological yield			1000-grain weight			Grain yield/plant		
P_1	33.26±1.10	44.29±1.48	44.29±1.48	57.18±0.96	59.18±2.46	61.86±2.20	9.50±0.43	15.80±0.67	13.56±0.66
P_2	27.80±1.19	38.16±1.51	33.30±1.78	55.33±0.92	54.15±1.13	54.15±1.13	9.14±0.52	13.56±0.66	13.36±0.86
F_1	50.13±1.39	51.18±2.45	50.43±1.95	64.10±1.34	64.30±0.84	61.21±1.10	13.72±0.99	15.47±0.80	15.59±1.19
F_2	43.57±1.12	44.04±1.13	41.76±1.25	54.89±0.43	55.24±0.64	55.53±0.79	10.51±0.78	14.50±1.06	14.85±0.91
BC_1	41.92±1.13	42.28±1.14	42.18±1.13	61.00±1.00	58.21±0.72	57.00±0.52	11.99±0.78	15.05±0.96	14.25±0.94
BC_2	41.82±0.95	36.65±0.97	41.91±1.21	59.64±1.20	55.81±0.56	55.00±0.57	11.16±0.65	12.49±0.83	13.59±0.87
LSD5%	1.50	2.65	3.47	1.59	3.00	3.47	1.53	1.41	2.01

Table 4: The scaling test and estimates of the additive,dominance and interaction parameters in three durum wheat crosses for all studied traits

Scaling test and parameters	Cross 1 Mean±S.E	Cross 2 Mean±S.E	Cross 3 Mean±S.E	Cross 1 Mean±S.E	Cross 2 Mean±S.E	Cross 3 Mean±S.E	Cross 1 Mean±S.E	Cross 2 Mean±S.E	Cross 3 Mean±S.E
	Flag leaf area			Days to 50% heading			Days to 50% maturity		
A	16.62±2.01**	31.51±2.24**	-2.01±2.41	18.28±1.22**	3.99±1.28**	-1.13±1.47	29.63±1.01**	7.97±0.70**	8.50±1.11**
B	19.57±1.65**	18.12±2.14**	-5.45±2.63*	16.92±0.69**	3.27±1.62*	-4.60±1.62**	23.20±1.20**	24.46±1.43**	29.93±1.50**
C	15.41±4.66**	9.67±4.44**	19.96±6.29**	14.94±1.53**	5.58±2.25*	2.87±1.57	14.15±1.76**	24.13±1.82**	18.79±2.91**
[m]	31.70±0.89**	28.11±0.68**	35.77±1.24**	93.64±0.31**	93.63±0.40**	98.43±0.18**	139.40±0.33**	140.33±0.20**	140.44±0.55**
[d]	1.83±0.50**	8.22±0.75**	1.97±0.93*	-0.77±0.55	-3.46±0.72**	0.00±0.86	-1.20±0.56*	-6.87±0.61**	-4.28±1.10**
[h]	33.11±3.99**	46.20±3.56**	-13.56±5.64*	19.45±1.72**	1.51±2.30	-8.93±1.99**	36.73±1.81**	16.10±1.65**	30.99±3.18**
[i]	20.78±3.70**	39.96±3.09**	-27.42±5.29**	20.26±1.66**	1.68±2.16	-8.60±1.87**	38.68±1.71**	20.10±1.45**	34.48±3.04**
[j]	-1.47±0.94	6.70±1.08**	1.72±1.32	0.68±0.70	0.36±0.99	1.74±1.08	3.22±0.75**	-4.57±0.90**	-3.30±1.32*
[l]	-56.97±5.10**	-89.59±5.35**	34.88±7.30**	-55.46±2.69**	-8.94±3.67*	14.33±3.80**	-91.51±2.84**	-64.85±3.00**	-87.75±5.11**
	Plant height			No.of spikes/plant			No.of spikelets/spike		
A	10.42±1.93**	-14.94±2.22**	11.00±2.67**	0.31±0.63	-0.69±0.56	-2.18±0.74**	-4.50±0.74**	3.26±1.12**	5.15±1.01**
B	9.37±1.71**	-23.10±2.79**	-4.88±2.75	-0.11±0.55	-1.14±0.68	-2.32±0.61**	-5.35±0.62**	2.09±1.25	1.31±1.10
C	1.45±3.82	-14.32±6.65*	-8.22±5.42	1.18±1.16	0.20±1.19	-4.48±1.32**	-7.75±1.80**	4.61±2.33*	0.28±2.4
[m]	94.62±0.82**	94.02±1.45**	89.48±1.11**	8.10±1.17**	9.62±1.21**	7.78±0.23**	20.43±0.31**	19.10±0.23**	19.46±0.24**
[d]	0.53±1.0	6.18±1.19**	8.59±1.46**	0.88±0.22**	0.12±0.22	0.33±0.34	1.31±0.68	1.13±0.71	2.07±0.64**
[h]	19.59±3.97**	-10.66±6.48	18.17±5.54**	-0.21±2.79	-4.62±2.91	-1.02±1.21	8.34±1.88	17.49±1.78**	13.98±1.65**
[i]	18.34±3.85**	-23.72±6.27**	14.34±5.32**	----	----	-2.74±1.14*	7.98±1.85**	14.78±1.69**	13.26±1.62**
[j]	0.52±1.17	4.08±1.51**	7.94±1.68**	----	----	0.07±0.43	0.61±0.72	0.58±0.76	1.92±0.70**
[l]	-38.13±5.54**	61.76±8.19**	-20.46±7.96*	----	----	7.24±1.82**	-10.55±3.10**	-20.13±3.19**	-19.72±2.87**
	Biological yield			1000-grain weight			Grain yield/plant		
A	0.45±2.87	-10.91±3.65**	-10.36±3.33**	5.28±1.81**	-7.06±2.98*	-9.07±2.67**	0.76±1.87	-1.17±2.18	-0.65±2.33
B	5.71±2.64*	-16.04±3.46**	0.09±3.59	6.35±1.92**	-6.83±1.92**	-5.36±2.10*	-0.54±1.71	-4.05±1.95*	-1.77±2.28
C	12.96±5.52*	-8.65±6.99	-11.41±6.74	-8.87±3.62*	-20.97±4.13**	-16.31±4.62**	-4.04±3.74	-2.30±4.63	1.30±4.47
[m]	43.57±1.12**	44.04±1.13**	41.76±1.25**	54.89±0.43**	55.24±0.64**	55.53±0.79**	4.58±2.87	14.50±0.95**	16.70±3.39**
[d]	0.10±1.48	5.63±1.49**	0.27±1.66	1.36±1.56	2.40±0.91**	2.00±0.77*	0.18±0.34	2.56±1.27*	0.10±0.55
[h]	12.80±5.60*	-8.34±6.03	12.78±6.40**	29.57±3.85**	14.72±3.53**	5.09±3.91	14.10±7.39	-2.13±4.65	-6.77±9.04
[i]	-6.80±5.37	-18.30±5.41**	1.14±5.98	21.72±3.55**	7.08±3.13*	1.88 ±3.53	----	-2.92±4.56	----
[j]	-2.63±1.68	2.57±1.83	-5.23±2.02*	0.43±1.70	-0.12±1.67	-1.86±1.50	----	1.44±1.35	----
[l]	0.64±8.10	45.25±9.18**	9.13±9.46	-22.29±7.12**	6.81±5.52	12.55±5.56*	----	8.14±6.59	----

Table 5: Components of variance for no. of spikes /plant and grain yield /plant for the durum wheat crosses showing no interaction

Components of variance	No.of spikes/plant		Grain yield/plant	
	Cross 1 (Bani-Sweef-1 / Line-2)	Cross 2 (Sohag-3 / Line-3)	Cross 1 (Bani-Sweef-1 / Line-2)	Cross 3 (Sohag-3 / Line-4)
D	2.12	0.94	36.28	32.52
H	-0.44	2.88	-14.11	-3.21
E	2.45	2.51	9.49	17.35
h^2	0.31	0.13	0.76	0.50

Table 6: Heterosis, inbreeding depression (%) and genotypic coefficient of variability (GCV) in three durum wheat crosses for all studied traits

characters	Crosses	Heterosis M.P	Heterosis B.P	Inbreeding depression (%)	Genotypic coefficient variability (GCV)
Flag leaf area	Cross1	56.84**	36.09**	6.79**	4.91
	Cross2	27.62**	19.54**	2.43*	3.12
	Cross3	58.11**	56.47**	5.14**	6.64
Days to 50% heading	Cross1	-0.90**	0.72**	-1.33**	3.35
	Cross2	-0.18	4.12**	-1.49	5.59
	Cross3	-0.33**	1.47**	-0.14	5.49
Days to 50% maturity	Cross1	-1.42**	1.87**	-3.34**	2.26
	Cross2	-2.93**	-1.26**	-6.08**	1.57
	Cross3	-2.53**	-1.83**	-4.81**	3.78
Plant height	Cross1	-18.28**	-42.27**	-201.79**	5.10
	Cross2	8.80**	3.07	-204.87**	8.88
	Cross3	17.53**	32.81**	-96.51**	6.78
No.of spikes/plant	Cross1	21.77**	8.38**	5.54**	1.56
	Cross2	27.79**	25.78**	10.30**	1.62
	Cross3	23.37**	19.16**	21.81**	1.52
No.of spikelets/spike	Cross1	1.64**	-1.52**	9.61**	2.51
	Cross2	13.45**	10.46**	0.88	2.95
	Cross3	3.46**	2.72**	1.35*	3.37
Biological yield	Cross1	64.20**	50.72**	13.09**	9.59
	Cross2	24.15**	15.56**	13.95**	8.34
	Cross3	30.00**	13.86**	17.19**	9.62
1000-grain weight	Cross1	13.94**	12.10**	9.58**	2.53
	Cross2	13.47**	8.65**	14.10**	3.10
	Cross3	5.53**	-1.10	9.28**	3.94
Grain yield/plant	Cross1	47.21**	44.42**	23.40**	4.01
	Cross2	5.38**	-2.10**	6.27**	6.0
	Cross3	15.82**	14.97**	4.75**	4.56

of a significant [h] component would imply no dominance genetic differences or presence of ambidirctional dominance between the two parents and the dominant effects seemed to be not important in the genetic control of these crosses. The epistatic effects additive x additive [i] and dominance x dominance [l] were highly significant in most cases, it could be concluded from the above – mentioned results that dominance as well as epistatic effects additive x additive [i] and dominance x dominance [l] were very important in the inheritance of these studied traits. These results were opposite with Menon and Sharma [13], Hassan [17], Khalifa *et al.* [18], Saad [19] and Motawea [20]. The [j] parameter additive x dominance was significant positive or negative, indicating that dominance was towards direction of increasing and decreasing, for these studied traits, respectively.

The estimates of the components of variance for the interaction for no. of spikes/plant (crosses no. 1and 2) and for grain yield/plant (crosses no. 1and 3) are listed in Table 5. The additive component (D) was greater than the dominance (H) for no. of spikes/plant in cross no. 1 and for grain yield/plant in crosses No. 1 and 3, indicating the important role of additive variance for controlling of these traits. Accordingly, the narrow-sense heritability value for no. of spikes/plant were low in cross 1 and 2 (0.31 and 0.13 respectively) while, for

Table 7: The nucleotide sequence used for RAPD

Primer name	Sequence(5'-3')
OPA10	GTGATCGCAG
OPA19	CAAACGTCGG
OPC16	CACACTCCAG
OPC19	GTTGCCAGCC
OPD4	TCTGGTGAGG
OPD11	AGCGCCATTG
OPR2	CACAGCTGCC
OPR3	ACACAGAGGG

grain yield/plant the narrow-sense heritability value was moderately high in crosses No. 1 and 3 (0.76 and 0.50 respectively). Similar results were found by Shoran *et al.* [21].

Heterosis (%), inbreeding depression % and genotypic (GCV) coefficient of variability are presented in Table 6. Heterosis relative to mid-parents was significantly negative for days to 50% heading in both crosses no. 1 and 3, days to 50% maturity in all three crosses and plant height in cross no. 1.These results indicated that dominance direction was toward the low respective parent. Heterosis above the better parent was significantly negative for days to 50% maturity in both crosses no. 2 and 3, plant height in cross no. 1 indicating that dominance direction was toward the best parent. While, the heterosis above the better parent for no. of spikelets/spike in cross no. 1 and grain yield/plant in cross no. 2 was also significantly negative indicating that dominance direction was toward the worst parent. Meanwhile, it was significantly positive for flag leaf area, no. of spikes/plant, biological yield and 1000-grain weight indicating that the importance of hybrid vigor for these traits. These results go in line with those reported by Khalifa *et al.* [2], Khalifa *et al.* [3] and Sharma *et al.*[22].

Inbreeding depression values were positive and highly significant for all the studied traits in most crosses, except for days to 50% heading in cross no. 1, days to 50% maturity and plant height in all three crosses. These results were coupled with a reduction in the mean in the F_2 generation for all studied traits in all crosses. This is expected as the expression of heterosis in F_1 will be followed by a respectively reduction in F_2 due to the direct effect of homozygosity. These results are in line with those reported by Khalifa *et al.* [2], Khalifa *et al.* [3] and Sharma *et al.*[22].

The genotypic coefficients (GCV) of variability values were low for most studied traits in most crosses, indicating the decrease of genetic diversity.

RAPD-PCR Analysis: A total of 129 fragments were generated, with an average of 16.125 fragments/primer (Table 8 and Fig. 1). Among the eleven wheat genotypes, OP-R2 primer produced the largest number of DNA-amplified fragments (18), while the smallest number (14) was produced by OP-A10, OP-A19 and OP-C16. Ten marker specific-genotypes were obtained in some lines with 7.75%. Whereas, one in Sohag-3 (2013 bp), Line-2 (296 bp), line-3 (204 bp) and F_2 of Bani Sweef-1 X Line-2 (885 bp) while Line-4 and F_1 of Sohag-3 X Line-3 gave three bands (688, 870, 150 bp and 768, 555 and 500 bp,respectively).

Similarity indices were developed on the basis of the obtained amplified fragments of the eleven wheat genotypes using the eight RAPD primers as shown in Table 9. The genetic similarity values ranged from 0.62 to 0.81, with the mean of 0.707. The highest value was found between F_2 of Bani Sweef-1 X line-2 and F_2 of Sohag-3 X line-3, while the lowest genetic similarity was observed between Bani Sweef-1 and F_2 of Sohag-3 X line-3. The dendrogram (Fig. 2) constructed with UPGMA method revealed that 11 genotypes fell into two distinct groups. The main cluster I included only Bani Sweef-1. Cluster II, in its order, was divided into 2 sub-cluster, the first sub-cluster included Sohag-3, F_1 of Bani Sweef-1 X Line-2 and F_1 of Sohag-3 X Line-4 and the second sub-cluster included, Line-2, F_2 of Bani Sweef-1 X Line-2, F_2 of Sohag-3 X Line-3, F_2 Sohag-3 X Line-4, line-3, Line-4 and F_1 of Sohag-3 X Line-3. The random amplified polymorphic DNA (RAPD) technique has been successfully used for the assessment of genetic diversity in diploid, tetraploid and hexaploid wheat [23-26].

On the other hand, the genetic diversity in cultivated crops is essential for successful breeding and creation of new cultivars. Knowing the genetic diversity of wheat germplasm is necessary for identifying diverse parental combinations and creating segregating progeny with high genetic variability for selection. The results obtained by RAPD analysis were used to compare the genetic similarity between the five parents. These values ranged from 0.67 (between sohag-3 and line-4) to 0.77 (between line-2 and line-3). The values obtained between Mexican parents 0.77, 0.75, 0.71 for line-2 and line-3, line-2 and line-4 and line-3 and line-4 respectively were relatively higher from those obtained between Egyptian parents 0.68 (between Bani Sweef-1 and Sohag-3). The three crosses of the present study were made between Egyptian and Mexican parents. The similarity values between the combined parents for each cross were 0.70, 0.71 and 0.67 for cross no. 1, 2 and 3,respectively. Similarity values

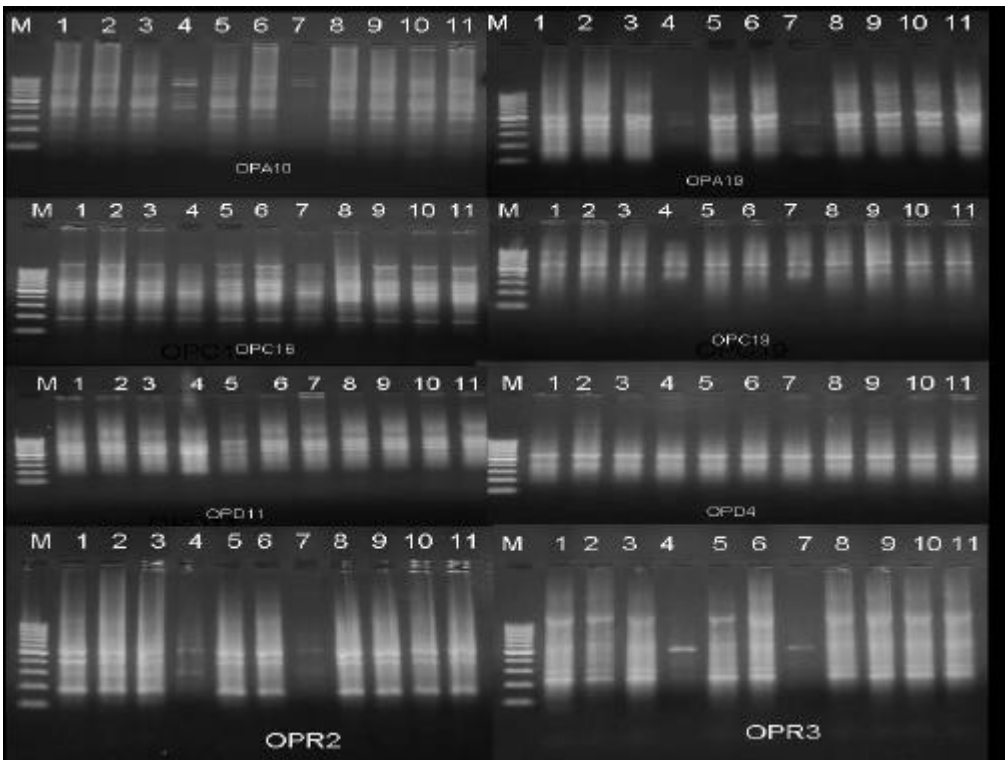

Fig. 1: RAPD amplified products of the eleven wheat lines using eight random primers 1,Bani Sweef-1, 2, Sohag-3, 3, Line-2, 4, line-3, 5, Line-4, 6, F1 of Bani Sweef-1 X Line-2, 7, F1 of Sohag-3 X Line-3, 8, F1 Sohag-3 X Line-4, 9, F2 of Bani Sweef-1 X Line-2, 10, F2 of Sohag-3 X Line-3 and 11, F2 Sohag-3 X Line-4

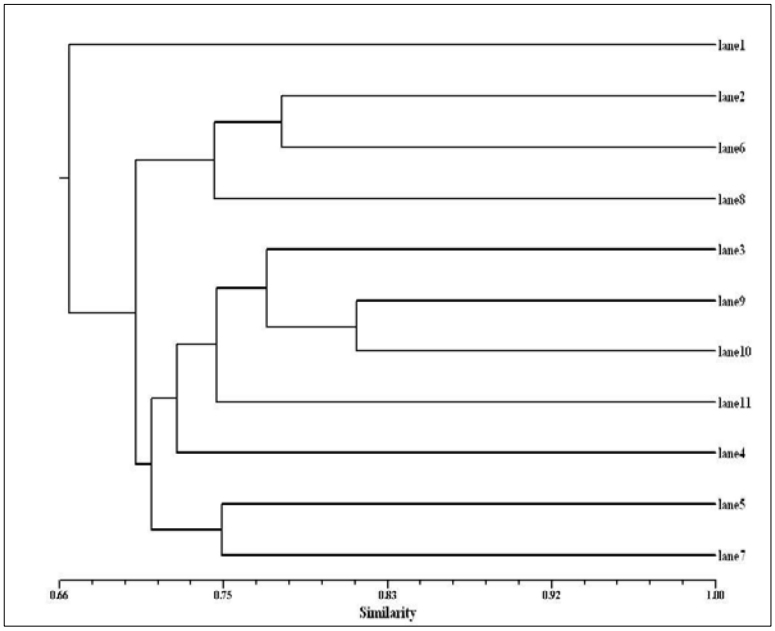

Fig. 2: Dendrogram showing the genetic relationships among 11 lines of wheat based on RAPD analysis Lane 1,Bani Sweef-1, Lane 2, Sohag-3, Lane 3, Line-2, Lane 4, line-3, Lane 5, Line-4, Lane 6, F1 of Bani Sweef-1 X Line-2, Lane 7, F1 of Sohag-3 X Line-3, Lane 8, F1 Sohag-3 X Line-4, Lane 9, F2 of Bani Sweef-1 X Line-2, Lane 10, F2 of Sohag-3 X Line-3 and Lane 11, F2 Sohag-3 X Line-4

Table 8: Number of bands generated and polymorphism percentage as revealed by RAPD among the eleven wheat lines

No.	Primers	Total bands	Monomorphic bands	Polymorphic bands	Polymorphism%
1	OPA10	14	4	10	71.4
2	OPA19	14	3	11	78.5
3	OPC16	14	7	7	50
4	OPC19	17	4	13	76.4
5	OPD4	16	6	10	62.5
6	OPD11	17	7	10	58.8
7	OPR2	18	2	16	88.9
8	OPR3	19	5	14	73.6
Total		129	38	91	70.5

Table 9: RAPD-based genetic similarity within groups

	1	2	3	4	5	6	7	8	9	10
2	0.68									
3	0.70	0.71								
4	0.68	0.71	0.77							
5	0.67	0.67	0.75	0.71						
6	0.66	0.78	0.71	0.70	0.68					
7	0.66	0.68	0.70	0.68	0.74	0.75				
8	0.68	0.74	0.66	0.69	0.71	0.74	0.68			
9	0.64	0.67	0.77	0.71	0.72	0.71	0.74	0.75		
10	0.62	0.66	0.77	0.71	0.72	0.70	0.71	0.71	0.81	
11	0.67	0.67	0.74	0.71	0.69	0.76	0.65	0.71	0.75	0.73

1,Bani Sweef-1, 2, Sohag-3, 3, Line-2, 4, line-3, 5, Line-4, 6, F1 of Bani Sweef-1 X Line-2, 7, F1 of Sohag-3X Line-3, 8, F1 Sohag-3 X Line-4, 9, F2 of Bani Sweef-1 X Line-2, 10, F2 of Sohag-3 X Line-3 and 11, F2 Sohag-3 X Line-4

between the parents combined in the three crosses were compared with values of the genetic variation obtained in segregation generations for morphological traits, yield and its components (Table 6). It could expect that cross 3 has the highest values for genetic variations in most studied traits as the parents of cross 3 had the lowest similarity index value. The results of genetic variation indicated that cross 3 had the highest score of variations in 5 out of 9 of the studied traits. Then, cross 2 had the highest score of variation in the other four traits. Lru *et al.* [27] used RAPD markers to studying the differences among 20 wheat parents with different yield characteristics and predicting the yield performance of hybrids produced from these parents. The twenty wheat lines were divided into four groups. Hybrids from parents in different groups were generally superior to most hybrids from parents in the same group. On the other hand, SSR microsatellites and morphological traits were used to evaluate the genetic diversity of seven bread wheat genotypes [28], they found that the range of the genetic distance based on morphological traits was on average higher than SSR markers, which may reflect the influence of the environment on the performances of the materials. The present study shows that analyzing higher number of parental genotypes by RAPD markers could assist for rapid predicting the genetic diversity among their crosses.

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