Egyptian Journal of Agronomy

Enhancing Wheat Genetic Resources through Diallel Crosses for Production of New Lines

Ramadan M. Esmail, F.B. Fathallah, Adel A. El-Shafei, R.M. Shoaib, Sherin A. Mahfouze, I.S. EL-Demardash#

Genetics and Cytology Department, Biotechnology Research Institute, National Research Centre, 12622 Dokki, Giza, Egypt.

SEVEN bread wheat (*Triticum aestivum* L.) genotypes were evaluated by Griffin's Diallel analysis for combining ability of grain yield, and its components to estimate their potential as sources of new germplasm for wheat breeding programs. Analysis of variance due to GCA and SCA was significant for all characters revealed difference among of parents in GCA and difference between crosses for SCA. The variance of yield analysis revealed highly significant differences among the progenies. The majority of the differences between the crosses were decided by general combining ability (GCA). SCA (specific combing ability) was also significant, but less so. GCA effect estimates revealed that one line was the best general combiner for grain yield. Obtained results showed the two parental lines no 1 and 4 proved to be good combiners for grain yield and some yield-related traits. Six crosses (no. 1, 2, 3, 5, 11 and 17) revealed high performance and good specific combiners for grain yield and some of its components. Consequently, these new bread wheat genotypes can be used to enhanced further wheat breeding program and exploited it for release a new wheat cultivars. This research used 29 simple sequence repeat (SSR) and EST-SSR markers to identify the 7 wheat genotypes according to their associations and choose those with more genetic diversity. The polymorphism detected by EST-SSR primers was greater than that detected by ESTSSR markers, according to the current study. Assorted genotypes could be distinguished using these. The dendrogram and PCoA both revealed that there was some variations among the wheat genotypes.

Keywords : EST-SSR, GCA, Genetic diversity, SCA, Wheat.

Introduction

Wheat is one of the main staple crops that are critical to global food security. The majority of the world's population relies on wheat as food, which occupies a prominent place in international food grain trade.

The challenges facing scientists interested in increasing wheat production locally and globally are the lack of adequate agricultural land availability, climate change and biotic &abiotic stresses. There is no doubt that the Russo-Ukrainian war has caused great suffering to humanity on the planet, as well as harming global trade, especially wheat, which often has a significant impact on developing countries. This gives every country caution and the right to manage and provide its population's need for food, especially if it imports large quantities of grain like Egypt. Therefore, continuous research on the development of new genetic resources from wheat germplasm must continue, through breeding programs to develop high yielding and disease-resistant varieties to cope with climate change.

Diallel mating designs is a useful biometrical tool are frequently used in plant breeding to study the patterns of inheritance of different traits in

many crops (Moterle et al., 2012; Townsend et al., 2013). Also, supply useful genetic information for breeders, such as general combining ability GCA and specific combining ability SCA, to help them develop appropriate breeding and selection strategies to identify the superior parents to be used in breeding programs and promising cross combinations for development new germplasm (Esmail, 2002; Zhang et al., 2005; Acquaah, 2012, Fathallah et al., 2021; Kajla et al., 2022).

Phenotypic and molecular approaches, either alone or in combination, have been used to identify of the   genetic diversity and deviation among wheat genetic resources (Salem et al., 2008; Ayed et al., 2010; Barakat et al., 2010; Dodig et al., 2010; Barakat et al., 2013; Laido et al., 2013; Arora et al., 2014). Several types of molecular markers, including random amplified polymorphic DNA (RAPD), ISSR, (Al-Doss et al., 2009; Barakat et al., 2010), SRAP (Sequence-Related Amplified Polymorphism), TRAP (Target Region Amplified Polymorphism), (Al-Doss et al., 2010) amplified fragment length polymorphism (AFLP) (Bertan et al., 2009), single nucleotide polymorphism (SNP) (Nie et al., 2019), EST-SSR and simple sequence repeat (SSR) (Elshafei et al., 2019a, b) have been used in the genetic analysis of bread wheat.SSR markers are multi-allele, co-dominant, highly informative, reasonably numerous, extensively dispersed across the genome, and reproducible (Powell et al., 1996a; Chen et al., 2012). SSRs are useful for a variety of purposes, including genetic research, aided selection for crop improvement, and genetic diversity estimate of wheat cultivars and lines (Eivazi et al., 2008; Al-Ashkar et al., 2020). Under non-stressed and abiotic-stressed environments, Semahegn et al. (2020) found basic genotypic variations among groups of bread wheat cultivars for important agronomic parameters.

The following goals were established:

1- To assess performance of seven spring wheat genotypes and their F1s to recognize the best performing genotypes,

2- To study the GCA and SCA to discover the best general combiners and hybrid combinations for yield and its related components,

3- To estimate the genetic distance among the 7 spring wheat parental lines using SSR and EST-SSR molecular markers.

4- To get a base population for development a new improved bread wheat cultivars.

Materials and Methods

Plant materials

The experimental material included seven different lines/varieties of spring wheat, Symbol, Name and Pedigree are presented in Table 1.

Experimental design

The statistical analysis of data obtained is conducted according to Gomez & Gomez (1984) using excel statistical program. A two-step analysis was performed for each evaluated trait. The first step consisted of analysis of variance and test of significance. Whereas the second step including estimates of general and specific combining ability in the parents and their F1crosses using Griffing (1956) diallel analysis, model I method II .

Data collection

Data were measured on an individual plants basis for the following traits: days to heading, plant height (cm), number of spikes/plant, 100-kernel weight (g), spike yield (g) and grain yield/plant (g).

TABLE 1. Symbol, name and pedigree of seven genotypes used in this study

Name	Pedigree
Line-20 (P1)	Advanced breeding line –dwarf plant type
Line-R1(P2)	Advanced breeding line –rust resistance
Misr -1 (P3)	Oasis/Skauz//4*BCN/3/2*PASTOR.CMSSOYO1881T- 050M-030Y–O3OM-030WGY-33M-0Y–0S
Line white M (P4)	Advanced breeding line
Sids -14 (P5)	SW8488*2/KUKUNA-CGSS01Y00081T-099M-099Y- 099M-099B–9Y–0B-0SD
Giza -171 (P6)	SAKHA93 /GEMMEIZA 9 S.6-1GZ- 4GZ- 1GZ- 2GZ-0S
Line- 37 (P7)	Advanced breeding line –long spike

DNA sampling, SSR and EST-SSR markers amplification

Molecular analysis

The Biotechnology Lab, Department of Genetics and Cytology, National Research Center (NRC), Dokki, Giza, Egypt, did the molecular analysis. The Wizard Genomic DNA Purification Kit was used to extract DNA from wheat genotypes (Promega Corporation Biotechnology, Madison, WI, USA). The isolated DNA was then treated with RNase and kept at 20°C in the refrigerator. The DNA was diluted to 25ng/μL before the SSR and EST-SSR analyses. The primers utilized were nine SSR primers (Somers et al., 2004) and twenty EST-SSR primers (Peng & Lapitan, 2005) (Tables 6, 7). The PCR mixture comprised 50ng of genomic DNA, 1X PCR buffer, 1.5mM $MgCl_2$, 0.1mM each dNTP, 0.5μM each of forward and reverse primers, and 1U Taq polymerase in a volume of $0.010cm^3$. The PCR program for the SSR and EST-SSR analyses involved a primary denaturation at 94°C for 3min, followed by 35 cycles of denaturation at 94°C for 1min, annealing at 50, and 55°C (dependent on SSR and EST-SSR primers) for 1min, and extension at 72°C for 2min, followed by a final extension at 72°C for 10min. The amplified PCR products were applied to 3% (m/v) agarose gel containing $0.1μgcm^{-3}$ ethidium bromide in TBE buffer. After electrophoresis, a photograph of the gel was captured using a UV trans-illuminator. The EST-SSR and SSR data were scored on the basis of presence (1) or absence (0) of a given marker, after excluding un-reproducible bands.

A similarity matrix was estimated according to Nei & Li (1979) using molecular marker data as follows:

$$SM = 2N_{ij}/(N_i + N_j)$$

where, N_{ij} is the number of alleles present in both the ith and jth genotypes, N_i is the number of bands present in the ith genotype, and N_j is the number of alleles present in the jth genotype. The similarity matrix was then subjected to the rate unweighted pair group method with arithmetic average (UPGMA) grouping algorithm. Principal coordinate analysis (PCoA) was used as an alternative to hierarchical clustering in that the similarity matrix was used to obtain the coordinates. These coordinates were then used to create scatter plots that represent the relationships among genotypes. Both UPGMA and PCoA were conducted using PAST version 1.62 (Hammer et al., 2001). Furthermore, to ensure

the reliability of the generated dendrogram, 1000 simulations were performed using PAUP* version 4.0.b5 (Swofford, 2001).

Marker efficiency analysis

The performance of the primers was assessed using the iMEC program, which calculated various metrics such as polymorphic Information Content (PIC), Discriminating Power (DP), and expected heterozygosity (H) for each primer.

Amiryousei et al. (2018) his program calculates PIC using Botstein et al. (1980) formula:

$$PIC = 1 - \Sigma\ p_i^2 - \Sigma\Sigma\ p_i^2\ p_j^2$$

where, p_i and p are the population frequency of the ith and jth allele. Heirst summation is over the total number of alleles, whereas the two subsequent summations denote all the i and j where, i = j.

E (EMR) was calculated using Powell et al. (1996b) formula:

$$EMR = n\ \beta$$

where, n is the average number of fragments amplified by an individual to a specific system marker (multiplex ratio) and β is estimated from the number of polymorphicloci (np) and the number of non-polymorphic loci (nnp); β = np/ (np+ nnp).

Marker index was calculated using Powell et al. (1996b) formula: MI = E Havp. The product of the effective multiplex ratio and the average expected heterozygosity for polymorphic markers, where H denotes the average expected heterozygosity for the polymorphic markers. It is equal to Σ Hp/np, where the summation is over all polymorphic sites with Hp and np defined as above.

Discriminating power was calculated using Tessier et al. (1999) formula DP = 1 – C, The probability that two randomly chosen individuals exhibit different banding patterns and are thus distinguishable from one another. C is defined as the confusion probability. For the i-th pattern of the given j-th primer, present at frequency pi in a set of varieties, the confusion probability is C = Σ ci= Σ pi(Npi−1)/(N−1), where, for N individuals, C is equal to the sum of all c for all of the patterns generated by the primer.

Expected heterozygosity was calculated using

Liu (1998) formula: H = 1 − Σ p2. The probability that an individual is heterozygous for the locus in the population. p is the allele frequency for the i-th allele, and the summation is over all available alleles.

Results and Discussion

The analysis of variance for all traits studied is presented in Table 2. Highly significant differences were recorded for genotypes and its partitioning parents, crosses and parent vs. crosses for all characters except 100 grain weight for parents and number of spikes and grain yield /plant for parents vs. crosses indicating the presence of considerable variability between the tested genotypes.

Analysis of variance due to GCA and SCA was significant for all characters except 100 grain weight for general combining ability revealed difference between parents for GCA and difference between crosses for SCA. Also, results of combining abilities revealed the importance of both additive and non-additive gene actions in the inheritance of all traits. The GCA variances are higher than SCA variances (GCA/SCA) for the most traits studied, revealed the predominance of additive gene action and progeny selection will be effective for improvement of these traits. This ratio was <1.0 for 100 grain weight implying a significant function of non-additive(dominance and epistasis) genetic effect on this trait (Table 2).

These results agree with the findings of Esmail (2002), Zhang et al. (2005), Acquaah (2012), Tayade et al. (2020) and Fathallah et al. (2021). They found that the ratio of GCA/SCA was higher than unity. However, the GCA/SCA variance ratio was less than unity emphasizing the role of non-additive gene action for all the traits (Kajla et al., 2022)

The parental mean performance (Table 3) varied greatly across all traits studied. Furthermore, the presence of non-additive gene effects and/or transgressive segregation in F1 crosses located outside the parental range indicates the presence of non-additive gene effects and/or transgressive segregation. These findings concur with those reported by Rahul & Kandalkar (2018).

Mean performance for days to heading was differed among the tested 7 genotypes ranged between 71.0-91.0 and 67.31 the correct range

is 67.33-87.03 between parental lines and their hybrid combinations, respectively. P1XP4 displayed the earliest heading, whereas p2xp3 and p3xp5 exhibited the latest heading (Table 3). Plant height mean performance among parental lines used revealed that Line 20 had the shortest plant height (71cm) while , the taller taken from Line 37 (119.67cm). Cultivars with short stature are required to restrain lodging in wheat. A suitable plant height is associated with a narrower range of lodging, a greater number of grains per spike, a higher harvest index, and higher grain yield and quality (Hedden, 2003; Griffiths et al., 2012). However, Jaradat et al. (1996) found that plant height reduced grain yield due to a negative correlation with grain yield. Semi-dwarf stature is a desirable trait in wheat because it provides not only resistance to lodging but also a mechanism for efficiently utilizing nitrogen fertilizer. Medium statured genotypes had higher grain yield than tall statured genotypes, according to Siyal et al. (2020) and Zhao et al. (2018). Number of spike per plant was significantly varied among genotypes, and varied between 6.0-12.0 spikes. The parental lines P2 and P5 had the highest spike numbers, whereas P1XP4, and P1 XP6 displayed also the highest spikes. P7 among the seven parents studied was high in magnitude in spike yield, while four F1 crosses contain the P7 i.e., (P1XP7, P2XP7, P3XP7, and P4XP7) were is the heaviest in spike yield. For100-grain weight, the average of parents ranged from 5.12 to 6.07g whereas from hybrids ranged from 5.14 to 7.33g. Concerning, grain yield /plant seven F1 hybrids exceeded the overall mean crosses (42.55), the best hybrids were P1XP4 (60.96g) followed by P1XP6 (55.18g) and P1XP2 (54.08). The mean performance of parents ranged from (38.22g) for Giza 171 to (47.69g) for Sids 14.

Results of general combining ability effects (GCA) for seven parental lines are presented in (Table 4 and Figs., 1-6). Studying the combining ability allows the breeder to characterize and classify the parental genotypes used in terms of their ability to produce crosses of different quality. Negative values are preferred for days to heading and plant height which are displayed on the downward trend of the diagram. Generally, in our research, it was observed that no parent was found as a good combiner for all the traits under study because the combined ability of the two parents was not consistent with all the yield component traits except days to heading, plant height and grain yield/ plant for parent (p1) line -20

TABLE 2. Analysis of variance of half diallel cross of seven bread wheat genotypes for all traits studied

S.O.V	DF	Days to heading	Plant height (cm)	Spikes no. / plant	Spike yield (g)	100 grain weight (g)	Grain yield /plant (g)
Reps	2	1.65	4.15	1.58	0.142	0.105	1.22
Genotypes	27	120.30**	439.49**	7.23**	1.62**	0.924**	161.62**
Parents	6	203.38**	821.41**	4.43*	0.657**	0.342	29.65**
Crosses	20	94.23**	319.73**	8.37**	1.507**	1.029**	208.57**
P.vs.Cr.	1	143.25**	543.25**	1.28	9.758**	2.32*	14.78
GCA	6	426.89**	801.65**	16.24**	3.02**	0.611	224.74**
SCA	20	34.34**	352.82**	4.89**	1.29**	1.064**	150.76**
Error	54	1.73	3.50	2.06	0.064	0.350	3.58
GCA/SCA	-------	12.43	2.27	3.32	2.34	0.57	1.49

*and ** significant at 0.05 and 0.01 levels of probability, respectively.

TABLE 3. Mean performance of half diallel cross analysis in bread wheat for all traits studied.

Genotypes	Days to heading	Plant height (cm)	No. of spikes/ plant	Spike yield (g)	100 grain weight (g)	Grain yield/plant (g)
P1- Line20	74.33	71.00	9.00	4.53	5.44	40.77
P2- Line 1	90.33	114.3	10.69	3.78	5.67	40.41
P3- Misr1	88.00	107.7	8.93	4.38	5.90	39.11
P4-Whitem	71.00	107.31	8.71	4.64	5.99	40.41
P5- Sids14	91.00	113.67	10.30	4.63	5.53	47.69
P6-Giza171	76.30	115.01	7.69	4.97	6.07	38.22
P7- Line 37	78.70	119.67	8.27	5.27	5.12	43.58
Par, mean	81.38	106.94	9.08	4.60	5.67	41.45
p1xp2	82.02	94.33	10.02	5.4	5.87	54.08
p1xp3	75.31	97.0	11.0	4.84	5.67	53.23
p1xp4	67.33	91.02	12.00	5.08	5.63	60.96
p1xp5	71.66	101.00	8.00	5.41	7.33	43.27
p1xp6	74.3	94.02	10.03	5.50	5.27	55.18
p1xp7	71.00	92.02	6.33	6.24	6.58	39.49
p2xp3	87.03	111.6	7.67	4.41	6.09	33.82
p2xp4	78.3	125.3	8.67	5.12	6.78	44.37
p2xp5	84.67	114.67	8.33	3.83	6.41	31.90
p2xp6	81.33	104.66	6.33	5.99	5.95	37.89
p2xp7	83.3	123.67	7.02	6.77	5.67	47.52
p3xp4	81.00	103.68	9.00	4.55	6.30	40.95
p3xp5	87.00	102.66	7.00	4.46	6.67	31.22
p3xp6	75.3	105	7.00	5.97	6.09	41.79
p3xp7	83.67	121.00	6.00	6.18	5.37	37.09
p4xp5	75.33	108.00	7.66	5.23	6.0	40.06
p4xp6	73.30	110.66	10.0	5.23	5.14	52.29
p4xp7	71.71	116.23	6.33	6.02	5.67	38.09
p5xp6	80.3	120.31	7.66	5.54	5.76	42.42
p5xp7	79.00	118.66	6.33	5.56	7.10	35.19
p6xp7	82.66	115.00	6.33	5.77	5.85	36.52
Crosses Mean	78.35	108.12	8.03	5.39	6.05	42.73
Grand mean	79.85	107.53	8.55	4.99	5.86	42.55
LSD 0.05	2.14	3.09	2.36	0.42	0.97	4.28
LSD 0.01	2.86	4.13	3.16	0.56	1.30	5.72

TABLE 4. Estimates of general combining ability (GCA) effects of seven parental lines used in diallel cross for all traits studied

Parents	Days to heading	Plant height (cm)	Spikes no./ plant	Spike yield (g)	100 grain weight (g)	Grain yield/ plant (g)
P1	-1.57**	-1.59**	-0.005	0.001	-0.016	1.80**
P2	1.64	-3.13**	0.414	-0.090	0.016	-0.30
P3	1.19	1.25	0.007	-0.087	0.011	-0.86
P4	-1.63**	-0.11	0.106	-0.037	-0.006	0.67
P5	1.01	0.46	0.007	-0.082	0.095	-0.76
P6	-0.48*	0.96	-0.067	0.089	-0.056	0.10
P7	-0.16	2.16	-0.462	0.206**	-0.044	-0.65
LSD.05(gi)	0.47	0.67	0.51	0.09	0.213	0.68
LSD.01(gi)	0.63	0.90	0.69	0.12	0.284	0.91
LSD.05(gi-gj)	0.82	1.16	0.89	0.16	0.36	1.18
LSD.01(gi-gj)	1.09	1.56	1.19	0.21	0.49	1.57
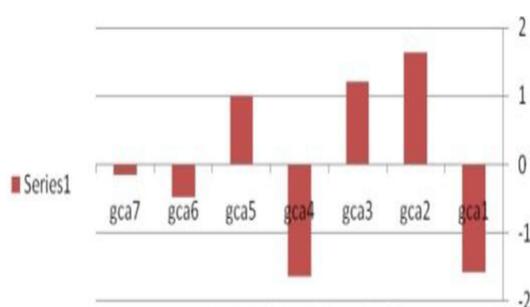
Fig. 1. GCA effects of days of heading
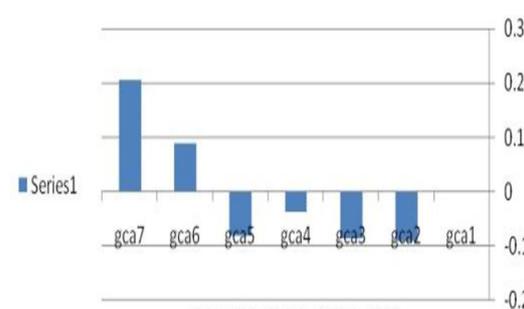
Fig. 4. GCA effects of days of spike yield
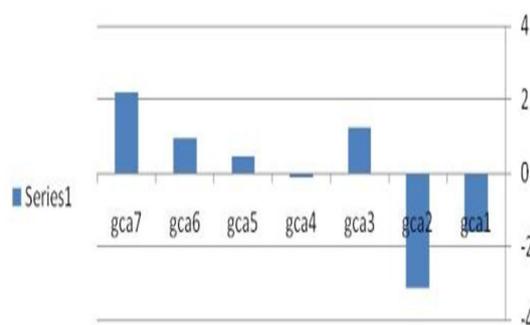
Fig. 2. GCA effects of days of plant height (cm)
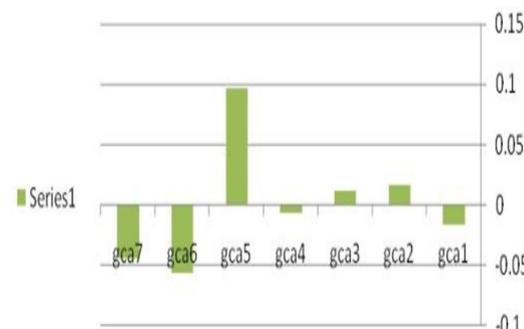
Fig. 5. GCA effects of 100 grain weight (g)
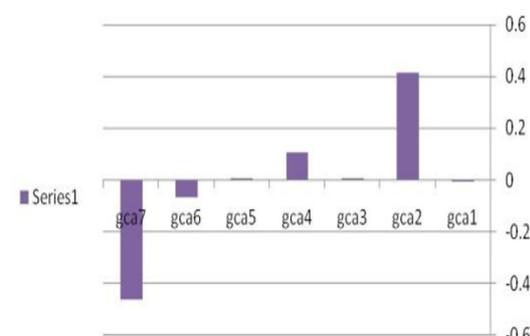
Fig. 3. GCA effects of days of spikes/plant
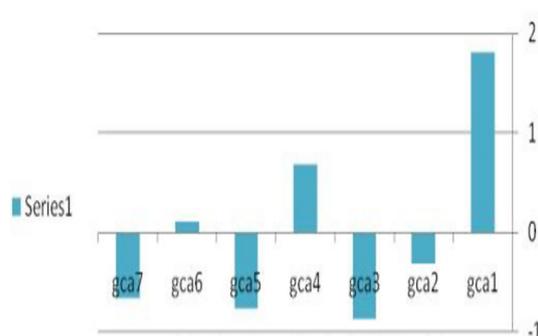
Fig. 6. GCA effects of grain yield/plant (g)

The largest and negative values for general combining ability effects was in the P1 and P4 for days to heading,P1 and P2 for plant height (Figs. 1, 2). Days to heading and plant height having negative values of GCA effects are indicates of better contribution to earliness and tolerance to loading. The best general combining ability effects with positive values were P2 for no of spikes per plant , P6 and P7 for spike yield (Fig. 4), P5 for 100 grain weight and P1 & P4 for grain yield/ plant as revealed in (Fig. 6) where, bars in upward direction.

GCA is an effective tool for selecting parents based on the performance of their offspring, typically F1s, but it has also been used in F2 and later generations (Fn). A low GCA value, whether positive or negative, indicates that the mean of a parent's crossing with the other does not deviate significantly from the overall mean of the crosses. A high GCA value, on the other hand, indicates that the parental mean is superior or inferior to the general mean. This represents information about the concentration of predominantly additive genes and indicates strong evidence of desirable gene flow from parents to offspring (Franco et al., 2001). A good performance parent per si may not necessarily produce better hybrids when used in hybridization (Esmail, 2002; Tyagi & Lal, 2005; Shukla & Pandey, 2008; Kumar et al., 2017; Yadav et al., 2017; Kajla et al., 2022) provided similar information about GCA in wheat. The information resulting from the estimation of the general ability effects of parents is great importance for the breeder to enable him to predict the new recombination's in the next generations. The value and direction of combining ability effects are also, guiding principles for the breeder to use the best parents in any breeding program.

Bars in upward direction show positive GCA while bars in downward direction show negative GCA values.

Estimates of the specific combining ability (SCA) effects are useful in identifying the best hybrid combinations with desirable traits. Results of the estimates of specific combining ability effects of the 21 F1 crosses for all traits studied are presented in (Table 5 and Figs. 7-12). Two crosses had the best specific combiners for days to heading i.e, cross-4 (P1, Line 20 xP4, White M.) and cross-14(P3,Misr 1 x P6, Giza 171). These had the lowest days to heading 67.33 and 75.3

,as revealed in Fig. 7 where, bars in downward direction shows negative values of SCA effects of these crosses.

As regard to plant height, SCA effects negative values is preferred results in Fig.8 showed that five crosses no.8, 10,11,14 and 16 were lie on downward direction and had 17 high negative values of SCA effects also , considered as the best hybrid combinations useful in breeding and developing a new semi dwarf cultivars.

For the spike yield the crosses 10-(2x5),11-(2x7) and 14-(3x6) had positive and significant SCA effects (Favorable) while, crosses 2-(1x3),3-(1x4),5-(1x6),11-(2x7) and 17-(4x6) had positive and significant values for grain yield/plant indicate that parent line 20 (p1) had a highest SCA effects for this trait and the same time ranks the best for it is performance in thesame related trait, such line would be considered as a good breeding material to improve this specific trait.

SSR analysis

Nine pairs of primers were employed to amplify all of the genotypes in this study, with 21 providing repeatable and well-resolved bands (Table 6). Wmc44 (4 alleles) had the largest number of SSR markers, followed by barc130, barc173, and wmc169 (3 alleles), with the other markers scoring low (2 and 1 alleles). Each marker had an average of 2.33 enhanced markers. These markers yielded 21 bands, 10 of which were polymorphic alleles had an average of 1.11, accounting for 47.62% of the total alleles (Table 6).

The discriminating power (DP) of twenty EST-SSR markers was determined by calculating their PIC. Using 20 EST-SSR markers, a total of 36 alleles were amplified among the seven wheat genotypes. The number of amplified bands (alleles) per marker ranged from one for six EST-SSR markers like'xcwem11 ' to three for two markers,'xcwem14 and xcwem16,' with an average value of 1.8 alleles/primer. These markers revealed 36 bands, 23 of which were polymorphic alleles with an average of 1.15 and accounted for 63.90 percent of alleles. Amplified alleles ranged in size from 80 to 320 bp (Table 7).

Marker efficiency analysis (MEA)

iMEC is an easy route for determining the effectiveness of specific markers' polymorphisms. The polymorphism indices of selected SSR

markers are shown in Table 6. For each marker, PIC is a measure of the diversity and occurrence of alleles produced among genotypes. PIC was 0.315 on average, with the highest value being 0.520 for wmc169, followed by 0.444 for wmc44, and the lowest for 0.152 for barc147. The average heterozygosity (H) per allele ranged from 0.165 (barc147) to 0.592 (wmc169). The marker index (MI) was determined to measure the effectiveness of the SSR marker system on wheat genotypes and found to be highest for wmc169 (MI =0.592), followed by wmc44 (MI = 0.512), and the lowest for barc200 (MI = 0.162), with a mean of 0.355 per marker. We estimated discriminative power

(DP) using a mean index of DP = 0.0404 and a range of 0.0 to 0.078 to determine the judicious profundity of the ssr marker. The ratio of effective multiplexes (E). It was 1 for all SSR or EST-SSR markers, also abbreviated as (EMR). It is the result of an individual screening's polymorphic loci component. Except in the case of SSRs (or other co-dominant markers), when E is 1 because each assay exposes a single locus. A significant positive correlation was found between PIC and MI (r = 0.998, p 0.05); D and PIC values (r = 0.435, p 0.05); PIC and H (r = 0.91, p 0.05); and MI and D (r = 0.413, p 0.05).

TABLE 5. Estimates of specific combining ability (SCA) effects of 21 F1s cross combination for all traits studied

21 F1 crosses	Days to heading	Plant height (cm)	No. of spikes / plant	Spike yield (g)	100 grain weight (g)	Grain yield/ plant (g)
1- S1x2	0.97	6.90	-0.512	0.159	-0.030	2.64**
2- S13	-0.79	0.29	-0.216	-0.030	-0.091	2.74**
3- S14	-1.31	1.54	-0.093	0.001	-0.086	3.34**
4- S15	-1.82*	3.08	0.339	0.154	0.377	-0.69
5- S16	0.55	3.02	-0.253	0.014	-0.157	2.31*
6- S17	-0.88	3.36	0.031	0.144	0.271	-2.21
7- S23	-0.13	-1.72	0.586	-0.083	0.015	-1.83
8- S24	-0.86	-2.36	0.820	0.104	0.265	0.53
9-S25	-0.71	0.40	-0.413	-0.279	0.038	-2.77
10-S26	-0.34	-2.44	0.327	0.267*	0.038	-1.39
11-S27	0.003	-4.31	-0.501	0.411**	-0.068	2.78**
12-S34	0.47	4.70	0.117	-0.089	0.106	-0.40
13-S35	0.51	0.57	0.105	-0.075	0.131	-2.03
14-S36	-1.89**	-3.27	-0.487	0.260*	0.088	0.28
15-S37	0.56	1.86	0.129	0.212	-0.164	-0.16
16-S45	-1.22	-2.07	-0.438	0.134	-0.076	-0.58
17-S46	-0.40	-1.80	-0.364	-0.039	-0.208	2.63*
18-S47	-1.28	2.33	-0.302	0.106	-0.047	-1.67
19-S56	-0.03	-0.48	0.734	0.110	-0.125	0.45
20-S57	-0.79	0.20	-0.092	-0.001	0.327	-1.01
21-S67	1.90**	0.48	-0.018	-0.101	0.065	-1.29
LSD (Sij)0.05	1.37	1.96	1.50	0.26	0.61	1.98
LSD (sij)0.01	1.84	2.62	2.01	0.35	0.83	2.64
LSD 0.05 (sij-sik)	2.04	2.91	2.23	0.39	0.92	2.94
LSD0.01(sij-sik)	2.73	3.88	2.98	0.53	1.23	3.93
LSD0.05 (sij-skl)	1.91	2.72	2.09	0.37	0.86	2.75
LSD0.01(sij-skl)	2.55	3.64	2.79	0.49	1.15	3.68
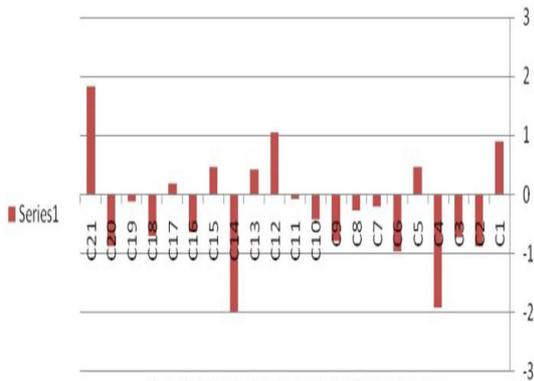
Fig. 7. SCA effects of 1 F1 crosses for days to heading

Fig. 10. SCA effects of 21 F1 crosses for spike yiled

Fig. 8. SCA effects of 21 F1 crosses for plant height (cm)
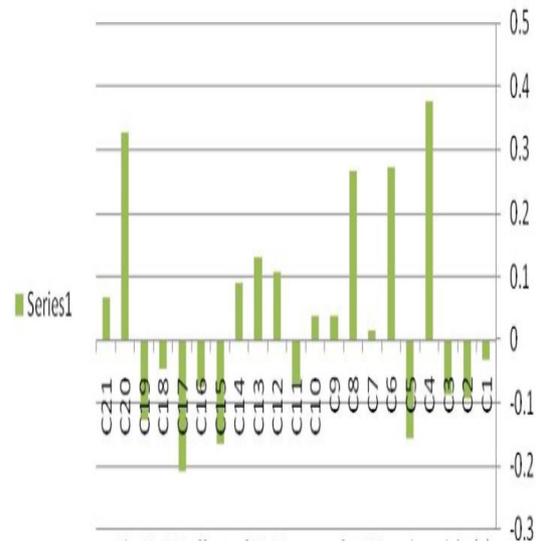
Fig. 11. SCA effects of 21 F1 crosses for grain weight (g)

Fig. 9. SCA effects of 21 F1 crosses for no. of spikes/ plant
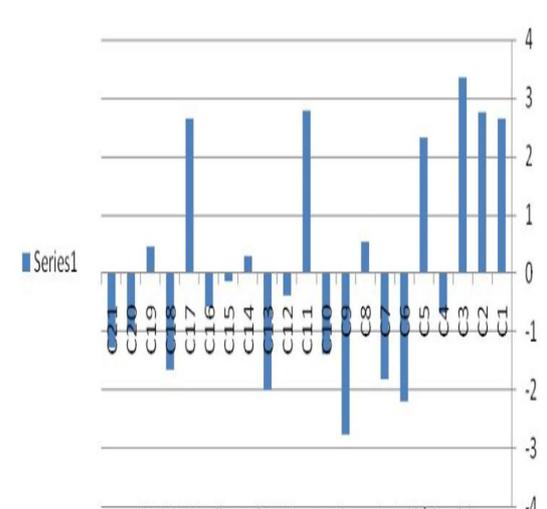
Fig. 12. SCA effects of 21 F1 crosses for yield/ plant (g)

TABLE 6. Efficacy of nine SSR markers polymorphism calculated with IMEC of wheat genotypes, SB (scored bands), PB (polymorphic bands), H (expected heterozygosity), E or EMR (Effective multiplex ratio), PIC polymorphism information content, MI (marker index) and DP(discriminating power)

SSR markers	SB	PB	H	E or EMR	PIC	MI	DP
Barc130	3	1	0.247	1.000	0.380	0.427	0.026
Barc147	1	0	0.165	1.000	0.152	0.165	0.000
Barc167	2	0	0.297	1.000	0.253	0.297	0.000
Barc173	3	2	0.420	1.000	0.365	0.420	0.078
Barc180	2	1	0.310	1.000	0.280	0.310	0.052
Barc200	1	1	0.170	1.000	0.162	0.162	0.052
Wmc44	4	2	0.512	1.000	0.444	0.512	0.052
Wmc169	3	2	0.592	1.000	0.520	0.592	0.052
Wmc175	2	1	0.310	1.000	0.280	0.310	0.052
Total	21	10	3.023	9.000	2.836	3.195	0.364
Average	2.333	1.111	0.336	1.000	0.315	0.355	0.040

TABLE 7. Efficacy of twenty EST-SSR markers polymorphism calculated with iMEC of wheat genotypes, SB (scored bands), PB (polymorphic bands), H (expected heterozygosity), E or EMR (Effective multiplex ratio), PIC polymorphism information content, MI (marker index) and DP(Discriminating power)

EST-SSR markers	SB	PB	H	E or EMR	PIC	MI	DP
Xcwem1	1	0	0.165	1.000	0.152	0.165	0.000
Xcwem2	1	0	0.165	1.000	.0152	0.165	0.000
Xcwem3	2	2	0.314	1.000	0.292	0.314	0.0866
Xcwem4	2	2	0.314	1.000	0.292	0.314	0.0520
Xcwem5	2	2	0.306	1.000	0.272	0.306	0.0520
Xcwem6	2	2	0.313	1.000	0.290	0.313	0.0952
Xcwem7	2	1	0.311	1.000	0.285	0.311	0.0779
Xcwem8	2	1	0.302	1.000	0.264	0.302	0.026
Xcwem9	2	2	0.313	1.000	0.289	0.313	0.0952
Xcwem10	1	0	0.165	1.000	0.152	0.165	0.000
Xcwem11	2	1	0.309	1.000	0.279	0.310	0.052
Xcwem12	2	1	0.313	1.000	.0289	0.313	0.0433
Xcwem13	1	1	0.170	1.000	0.161	0.170	0.0433
Xcwem14	3	2	0.410	1.000	0.344	0.410	0.0520
Xcwem15	1	0	0.165	1.000	0.152	0.165	0.000
Xcwem16	3	2	0.432	1.000	0.390	0.432	0.0952
Xcwem17	2	1	0.313	1.000	0.290	0.313	0.0431
Xcwem18	2	1	0.314	1.000	0.291	0.314	0.0259
Xcwem19	1	1	0.169	1.000	0.161	0.169	0.0433
Xcwem20	2	1	0.302	1.000	0.263	0.302	0.0230
Total	36	23	5.565	20.00	4.6631	5.566	0.906
Average	1.80	1.15	0.278	1.000	0.233	0.278	0.045

IMEC is a basic approach for evaluating the polymorphism efficiency of individual markers. In this table, specific polymorphism indices of chosen EST-SSR markers are shown (Table 7). With an average of 0.278 per allele, the heterozygosity (H) ranged from 0.165 (xcwem1, 2, 10, and xcwem15) to 0.432 (xcwem16). PIC is a measure of the diversity and recurrence of produced alleles among genotypes for each marker. The highest PIC value was recorded for xcwem16 (PIC = 0.390), followed

by xcwem14 (0.344), while the lowest PIC value was reported for the four markers xcwem1, 2, 10, and xcwem15 (PIC = 0.152). The marker index (MI) was investigated to determine the utility of the EST-SSR marker system on wheat genotypes, and it was found to be highest for xcwem16 (MI= 0.432), followed by xcwem14 (MI = 0.410), and lowest for four markers (xcwem1, 2, 10, and xcwem15) (MI = 0.165), with an average of 0.278 per marker. We calculated discriminative power (DP) with a mean index of 0.045 and a range of 0.0 to 0.0952 to determine the appropriate profundity of the EST-SSR marker. PIC vs MI (r = 0.763, p 0.05); D vs PIC values (r = 0.634, p 0.05); PIC vs H (r = 0.763, p 0.05); and MI vs D (r = 0.679, p 0.05) all had a positive significant connection.

Cluster analysis

SSR and EST-SSR data sets were combined to determine the genetic relationship between the different wheat genotypes. The correlation between the SSR and EST-SSR similarity coefficient matrices was 0.65. The similarity coefficients, which were calculated based on the combined SSR and EST-SSR data ranged from 0.41to 0.87with an average of 0.80 among all of the seven wheat genotypes. The nearest genetic distance (0.87) occurred between L-37 and Sdis-14 genotypes. The largest genetic distance (0.41) occurred between L-1 and White M (Table 8).

All of the above revealed a minor biological variation between genotypes. The dendrogram generated by UPGMA shows the genetic relatedness between the genotypes. The correlation coefficient between the dendrogram and its similarity matrix was 0.9507, indicating that the dendrogram may represent the genetic relationship well. At a similarity coefficient of 0.80, the dendrogram revealed that all genotypes were divided into two distinct clusters (Fig. 13). Cluster I had only one genotype (White M), which had the shortest time to head and was originally from Saudi Arabia.

Cluster II consisted of six genotypes divided into five subgroups. The genotype was found in the first subgroup, which had a bootstrap value of 24%. (Misr-1). The second subgroup, which had a bootstrap value of 58 percent, featured the two genotypes with the highest grain yield/p (Sids-14 and L-37). Only one genotype was included in the third subgroup, which had a bootstrap value of 28%. (L-20). The genotype with the highest 100 grain weight (6.07g), Giza-171, was included in the fourth subgroup, which had a bootstrap value of 46%. One genotype (L-1) in the fifth subgroup had a significant number of days to heading.

The PCoA result was consistent with the UPGMA cluster algorithm. The dendrogram grouping corresponded to the scatterplot grouping (Fig. 14). The PCoA also classified all genotypes into three different groups. The first three principal coordinates accounted for 50.33% of the total variation (30.85, 13.33, and 6.14% by the first, second, and third principal coordinates, respectively). The cluster analysis and PCoA both revealed that there was some variation among the wheat genotypes.

Perenzin et al. (1998) used molecular markers to determine parental variety and F1 hybrid performance. RAPD was tested on the parents using 87 primers, yielding 304 polymorphic bands. The genetic similarity between parents was evaluated using the similarity coefficient on the basis of shared bands and ranged from 0.25 to 0.57. A total of 75 bands were found among the analyzed cultivars for RAPD analysis, with 53 bands showing polymorphism, while a total of 11 alleles were detected among the studied genotypes for SSR markers, with 8 alleles showing polymorphism (Harby, 2020). The 40 SSR markers tested on the eight genotypes found a total of 112 alleles. This equated to an average of 2.8 alleles per site, with a single SSR locus detecting anywhere from two to five alleles (Ramu et al., 2013).

TABLE 8. Simple matching coefficients of similarity matrix created from study utilizing EST-SSR and SSR combinations markers

	L-1	Misr-1	L-20	Sids-14	L-37	White M	Giza171
L-1	1.00						
Misr-1	0.73	1.00					
L-20	0.67	0.75	1.00				
Sids-14	0.75	0.78	0.77	1.00			
L-37	0.70	0.82	0.80	0.87	1.00		
White M	0.41	0.48	0.56	0.54	0.53	1.00	
Giza 171	0.66	0.70	0.68	0.83	0.74	0.58	1.00
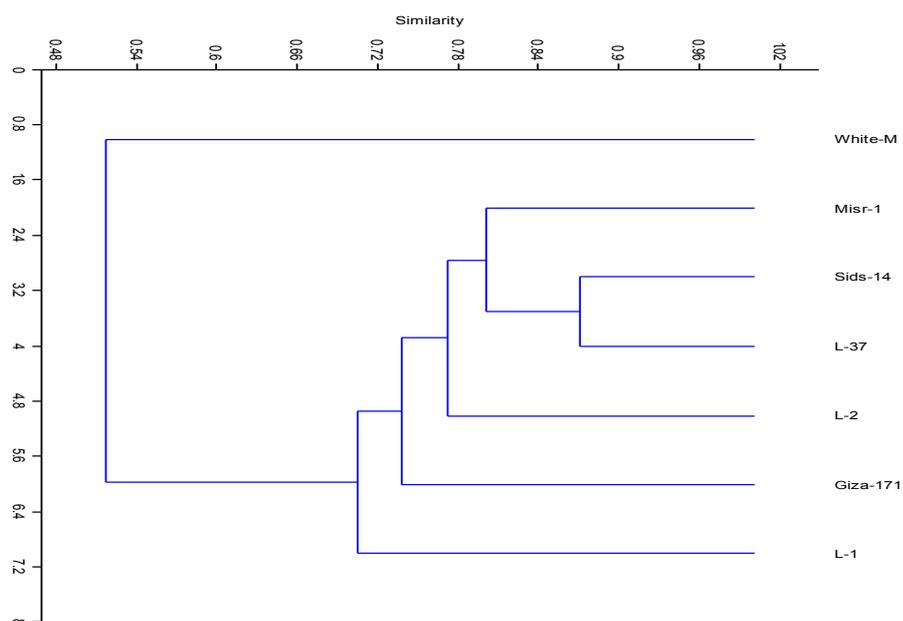
Fig. 13. Dendrogram based on the similarity coefficient of save genotypes based on the allelic data of 29 SSR and EST-SSR combinations
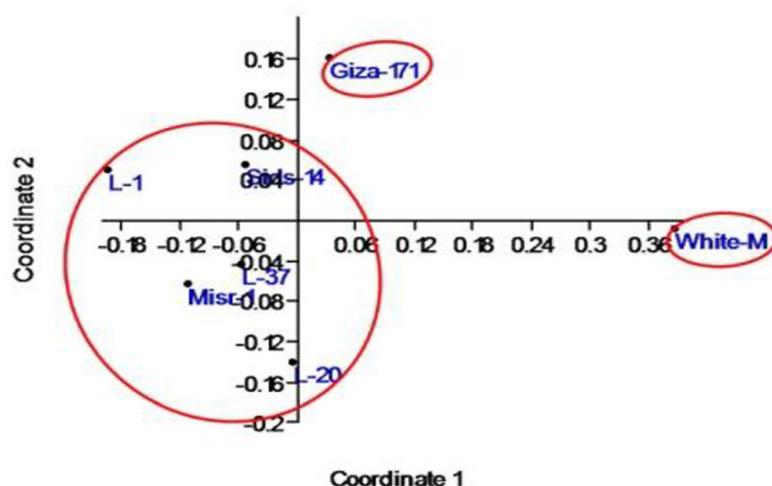
Fig. 14. Principal coordinate analysis (PCoA) of the 7 wheat genotypes with 29 SSR and EST-SSR combinations

The genetic distance between the eight genotypes, as determined by SSR markers, ranged from 0.20 to 0.48. For grain yield and heterosis , the correlation between Nei's genetic distance and SCA ranged from 0.4 to 0.5. These findings suggest that the level of SCA and heterosis is determined by the genetic diversity of the wheat genotypes studied by El-Maghra (2005). Among the ten sorghum genotypes, the ISSR profile analysis revealed 38 monomorphic and 113 polymorphic with 74.83 percent variation (Tawfik & El-Mouhamady, 2019). For PCR amplification of the genome of these 12 selections and their parents, fifteen SSR markers were used. With an average polymorphism of 86.67 percent, the SSR analysis revealed that the 12 families are genetically distinct from their 7 parents. The percentages of genetic similarity (Gs) ranged from 30% to 88 percent. Both the SF3 and SF4 mutants showed a lot of potential (Al-Naggar et al., 2015).

The genetic difference between the 16 WHEAT genotypes ranged from 0.235 to 0.911.

Cluster analysis employing SSRs to measure genetic distance divided the genotypes into three major groups and six sub-groups, which was almost identical to the results of principal coordinate analysis (Al-Ashkar et al., 2020). SSR markers are used. Five SSR markers were used to amplify a total of 48 alleles. Thirty-four of the 48 fragments (70.83%) were monomorphic bands, leaving 14 loci polymorphic (29. 17 percent polymorphasim). The five SSR primers had PIC values ranging from 0.863 to 0.881, with an average of 0. 870. The genetic identity index of the Nei family ranged from 0.81 to 0.95. The genetic closeness of nine wheat parents was divided into three groups (Fathallah et al., 2021). The average genetic similarity among the twelve wheat genotypes was 0.50, with values ranging from 0.34 to 0.68 in a study of genetic diversity among 12 Saudi wheat cultivars using RAPD and ISSR primers (Barakat et al., 2010). To locate the genetic diversity of 6 durum wheat genotypes, 19 SRAP and 9 TRAP markers were evaluated. As a result, the coefficient of similarity ranged from 0.71 to 0.93 for SRAP and 0.46 to 0.84 for TRAP markers, and the cluster based on SRAP markers differed from the cluster based on TRAP markers (Al-Doss et al., 2010).

Cluster analysis was performed using 242 SSR markers on 43 wheat genotypes, which were then divided into three groups: two groups for winter wheat (18 and 10 genotypes) and one group for spring wheat (15 genotypes) (Chao et al., 2007). The use of forty EST-SSR and SSR markers to assess genetic diversity in sorghum. Cluster analysis revealed that the majority of genotypes within geographic origins were mostly based on race. The EST-SSR markers utilized in this study were shown to have greater discriminating power than the genomic SSRs (Ramu et al., 2013).

PIC values from eight SRAP combination primers ranged from 0.445 to 0.896, with an average of 0.764 per primer. The PIC values were correlated (r = 0.896) in a positive way (Elshafei et al., 2019a). All of the SSR loci studied had a wide range of PIC values. Sixteen SSR markers detected only one allele and had zero PIC values. The remaining 7 markers had PIC values ranging from 0.18 to 0.576. The number of amplified alleles per primer was strongly linked (r = 0.95) with the PIC values (Elshafei et al., 2019b). PIC was 0.82, with a range of 0.70 to 0.89. (Salehi et al., 2018). 230 Nebraska winter wheat genotypes were studied for genetic diversity using single nucleotide polymorphisms (SNPs). The average PIC across chromosomes was 0.23, with a range of 0.09 to 0.37. (Eltaher et al., 2018).

The genotypes were not separated according to their designated mega environments (MEs) using PCoA based on modified Rogers' distances (Dreisigacke, 2004). With 2010 SSR alleles, the principal coordinates analysis for 20 early and current wheat cultivars. Not only did the main coordinate plot disclose two completely separate groups of 20 early and recent cultivars, but it also revealed two completely separate groups of 20 early and recent cultivars (Fu & Somers, 2009). Synthetic hexaploid wheat (SHWs) were divided into two subgroups (Spring SHW and Winter SHW) using principal coordinate analysis (PCoA) and cluster analysis, similar to Bayesian clustering. Data from PCoA of 18 faba bean genotypes using eight SRAP primers. In the PCoA scatter plot, the first three PCoA account for 63.8 percent of the total variation in accessions (Elshafei et al., 2019a).

Each marker's measured DP ranged from 0.033 to 0.067, with an average of 0.042. (ELshafei et al., 2019b). Ammar et al. (2015) that the DP of forty faba beans ranged from 0.29 to 0.05 using six SRAP markers and 0.13 to 0.42 using four AFLP markers. Using six (AFLP) combination primers, (Khierallah et al., 2011) found a DP range of 0.31 to 0.06 among eleven date palms. The DP of the primer ranged from 0.0435 to 0.195, with 0.125 being the average (Elshafei et al., 2019a).

Conclusion

Results obtained here indicated that the two parental lines no 1 and 4 proved to be good combiners for grain yield and some yield-related traits. Six crosses (no.1,2,3,5,11and 17) revealed high performance and good specific combiners for grain yield and some of its components. Consequently, these new bread wheat genotypes can be used to enhanced further wheat breeding program and exploited it for release a new wheat cultivars. At molecular level results revealed that , the polymorphism found by EST-SSR primers was larger than that detected by gSSR markers. These could be used to distinguish various genotypes. There was some variability among the wheat genotypes, according to both cluster analysis and PCoA. Pre-breeding or genetic enhancement of

any crop species requires the incorporation of a gene or genes from unmodified genetic resources into the improved commercial released varieties.

References

Acquaah, G. (2012) "*Principles of Plant Genetics and Breeding*". 2nd ed., Wiley-Blackwell, John Wiley & Sons, Chichester. https://doi.org/10.1002/9781118313718

Al-Ashkar, I., Alderfasi, A., Ben Romdhane, W., Seleiman, M.F., El-Said, R.A., Al-Doss, A. (2020) Morphological and genetic diversity within salt tolerance detection in eighteen wheat genotypes. *Plants*, **9**(3), 287.

Al-Doss, A.A., Moustafa, K.A., Ahmed, E.I., Elshafei, A.A., Barakat, M.N. (2009) Assessment of genetic diversity in Saudi wheat genotypes under heat stress using molecular markers and agronomic traits. *Global Science Books. International Journal of Plant Breeding,* **3**, 103-110.

Al-Doss, A.A., Saleh, M., Moustafa, K.A., Elshafei, A.A., Barakat, M.N. (2010) Grain yield stability and molecular characterization of durum wheat genotypes under heat stress conditions. *African Journal of Agricultural Research,* **5**(22), 3065-3074.

Al-Naggar, A.M.M., Al-Azab, K.F., Sobieh, S.E.S., Atta, M.M.M. (2015) Morphological and SSR assessment of putative drought tolerant M3 and F3 families of wheat (*Triticum aestivum* L.). *British Biotechnology Journal,* **6**(4), 174-190.

Ammar, M.H., Alghamdi, S.S., Migdadi, H.M., Khan, M.A., El-Harty, E.H., Al-Faifi, S.A. (2015) Assessment of genetic diversity among faba bean genotypes using agro-morphological and molecular markers. *Saudi Journal of Biological Sciences,* **22**(3), 340-350.

Amiryousei, A., Hyvnen, J., Poczai, P. (2018) iMEC: Online marker eiciency calculator. *Applications in Plant Sciences,* **6**(6), e01159.

Arora, A., Kundu, S., Dilbaghi, N., Sharma, I., Tiwari, R. (2014) Population structure and genetic diversity among Indian wheat varieties using microsatellite (SSR) markers. *Australian Journal of Crop Science,* **8**(9), 1281-1289.

Ayed, S., Karmous, C., Slim, A., Amara, A.S. (2010) Genetic variation of durum wheat landraces using morphological and protein markers. *African Journal of Biotechnology,* **9**, 8277-8282.

Barakat, M., Al-Doss, A.A., Moustafa, K., Ibrahim, E. (2010) Morphological and molecular characterization of Saudi wheat genotypes under drought stress. *Journal of Food, Agriculture and Environment,* **8**(1), 220-228.

Barakat, M.N., Al-Doss, A.A., Elshafei, A.A., Ghazy, A.I., Moustafa, K.A. (2013) Assessment of genetic diversity among wheat doubled haploid plants using TRAP markers and morpho-agronomictraits. *Australian Journal of Crop Science,* **7**(1), 104-111.

Bertan, I., Carvalho, F.I.F.D., Oliveira, A.C.D., Benin, G., Vieira, E.A., Valério, I.P. (2009) Morphological, pedigree, and molecular distances and their association with hybrid wheat per formance. *Pesquisa Agropecuária Brasileira*, **44**(2), 155-163.

Botstein, D., White, R.L., Skolnick, M., Davis, R.W. (1980) Construction of a genetic linkage map in man using restriction fragment length polymorphisms. *American Journal of Human Genetics,* **32**, 314–331.

Chao, S., Zhang, W., Dubcovsky, J., Sorrells, M. (2007) Evaluation of genetic diversity and genome-wide linkage disequilibrium among US wheat (*Triticum aestivum* L.) germ plasm representing different market classes. *Crop Science*, **47**(3), 1018-1030.

Chen, X., Min, D., Yasir, T.A., Hu, Y-G. (2012) Genetic diversity, population structure and linkage disequilibrium in elite Chinese winter wheat investigated with SSR markers. *PLOS One*, **7**(9), e44510.

Dodig, D., Zorić, M., Kobiljski, B., Šurlan-Momirović, G., Quarrie, S.A. (2010) Assessing drought tolerance and regional patterns of genetic diversity among spring and winter bread wheat using simple sequence repeats and phenotypic data. *Crop & Pasture Science,* **61**, 812–824.

Dreisigacker, S. (2004) SSR and pedigree analyses of genetic diversity among CIMMYT wheat lines targeted to different mega environments. *Crop Science,* **44**(2), 381-388.

Eivazi, A., Naghavi, M., Hajheidari, M., Pirseyedi,

S., Ghaffari, M., Mohammadi, S., et al. (2008) Assessing wheat (*Triticum aestivum* L.) genetic diversity using quality traits, amplified fragment length polymorphisms, simple sequence repeats and proteome analysis. *Annals of Applied Biology,* **152**(1), 81–91.

El-Maghraby, M.A., Moussa, M.E., Hana, N.S., Agrama, H.A. (2005) Combining ability under drought stress relative to SSR diversity in common wheat. *Euphytica*, **141**(3), 301-308.

Elshafei, A.A.M., Amer, M.A.E., Elenany, M.A.M. Helal, A.G.A.E. (2019a) Evaluation of the genetic variability of faba bean (*Vicia faba* L.) genotypes using agronomic traits and molecular markers. *Bulletin of the National Research Centre,* **43**(1), 1-10.

ELshafei, A.A., Afiah, S.A., Amer, M.A., Magda, A.M. (2019b) Validation of molecular markers linked with salinity tolerance in wheat (*Triticum aestivum* L.) grown on saline soil. *Bioscience Research,* **16**(2), 963-978.

Eltaher, S., Sallam, A., Belamkar, V., Emara, H.A., Nower, A.A., Salem, K.F., Poland, J., Baenziger, P.S. (2018) Genetic diversity and population structure of F3: 6 Nebraska winter wheat genotypes using genotyping-by-sequencing. *Frontiers in genetics,* **9**, 76.

Esmail, R.M. (2002) Estimation of genetic parameters in the F1 and F2 generations of diallel crosses in bread wheat (*Triticum aestivum* L.). *Bulletin of the National Research Centre,* **27**(1), 85-106 .

Fathallah, F.B., ElHosary, A.A., ELBadawy, M., ElHosary, A.A.A., Esmail, R.M., Mahfouze, Sherin A. (2021) Validation of simple sequence repeats (SSRs) linked to drought susceptibility index in diallel cross wheat. *Annals of Agricultural Science, Moshtohor,* **59**(2), 211-224.

Franco, M.C., Cassini, S.T., Oliveira, V.R., Vieira, C., Tsai, S.M., et al. (2001) Combining ability for nodulation in common bean (*Phaseolus vulgaris* L.) genotypes from Andean and Middle American gene pools. *Euphytica*, **118**(3), 265-270.

Fu, Y.B., Somers, D.J. (2009) Genome-wide reduction of genetic diversity in wheat breeding. *Crop Science,* **49**(1), 161-168.

Gomez, K.A., Gomez, A.A. (1984) "*Statistical Procedures for Agricultural Research*". John Wiley and Sons. New York. 680p.

Griffing, B. (1956) Concept of general and specific combining ability in relation to diallel crossing systems. *Australian. Journal of Biological Sciences,* **9**, 463-493.

Griffiths, S., Simmonds, J.R., Leverington, M., Wang, Y., Fish, L., Sayers, L., et al. (2012) Meta-QTL analysis of the genetic control of crop height in elite European winter wheat germplasm. *Molecular Breeding*, **29**, 159–171.

Hammer, D., Harper, A., Ryan, P. (2001) PAST: palaeontological statistics software package for education and data analysis. *Palaeontologia Electronica*, **4**(1), http://palaeo-electronica. org/2001_1/past/issue1_01.htm.

Harby, M.H. (2020). Molecular and cytogenetic studies on abiotic stress tolerance in wheat. *Alexandria Science Exchange Journal,* **41**, 479-487.

Hedden, P. (2003) The genes of the Green Revolution. *Trends in Genetics,* **19**, 5–9.

Jaradat, A.A., Ajlun, M.M., Karak, G.(1996) Genetic structure of Durum wheat landraces in a center of diversity. 5th *International Wheat Conference,* Abstracts: pp. 10-14.

Kajla, S.L., Sharma, A.K., Hoshiyar, S., Mukesh, B., Neelu, K., Moti, R.N. (2022) Combining ability analysis for grain yield and it's contributing traits in bread wheat (*Triticum aestivum*) over environments. *The Pharma Innovation Journal,* **11**(1), 1797-1800.

Khierallah, H., Bader, B.M., Hamwieh, A. (2011) Assessment of genetic diversity for some Iraqi date palms (*Phoenix dactylifera* L.) using amplified fragment length polymorphisms (AFLP) markers. *African Journal of Biotechnology,* **10**, 9570-9576.

Kumar, J., Singh, S.K., Singh, L., Kumar, M., Srivastava, M., Singh, J. (2017) Combining ability analysis for yield and its components in bread wheat (*Triticum aestivum* L.). *International journal of Current Microbiology and Applied Science,* **6**(3), 24-39.

Laido, G., Mangini, G., Taranto, F., Gadaleta, A., Blanco, A., Cattivelli, L., et al. (2013) Genetic

diversity and population structure of tetraploid wheats (*Triticum turgidum* L.) estimated by SSR, DArT and Pedigree data. *PLoS One,* **8**(6), e67280.

Liu, B.H. (1998) "*Statistical Genomics: Linkage, Mapping and QTL Analysis*". CRC Press, Boca Raton, Florida, USA.

Moterle, L.M., Braccini, A.L., Scapim, C.A., et al. (2012) Combining ability of popcorn lines for seed quality and agronomic traits. *Euphytica*, **185**(3), 337–347.

Nei, M., Li, W.H. (1979) Mathematical model for studying genetic variation in terms of restriction endonucleases. *Proceedings of the National Academy of Sciences* ,**76**(10), 5269–5273.

Nie, Y., Ji, W., Ma, S. (2019) Assessment of heterosis based on genetic distance estimated using SNP in common wheat. *Agronomy,* **9**(2), 66.

Peng, J.H., Lapitan, N.L. (2005) Characterization of EST-derived microsatellites in the wheat genome and development of eSSR markers. *Functional & Integrative Genomics,* **5**(2), 80–96.

Perenzin, M., Corbellini, M., Accerbi, M., Vaccino, P., Borghi, B. (1998) Bread wheat: F1 hybrid performance and parental diversity estimates using molecular markers. *Euphytica*, **100**(1), 273-279.

Powell, W., Machray, G.C., Provan, J. (1996a) Polymorphism revealed by simple sequence repeats. *Trends in Plant Science*, **1**(7), 215–22.

Powell, W., Morgante, M., Andre, C., Hanafey, M., Vogel, J., Tingey, S., Rafalski, A. (1996b) The comparison of RFLP, RAPD, AFLP and SSR (microsatellite) markers for germplasm analysis. *Molecular Breeding*, **2**, 225–238.

Rahul, S.R., Kandalkar, V.S. (2018) Combining ability and heterosis for grain yield and its attributing traits in bread wheat (*Triticum aestivum* L.). *Journal of Pharmacognosy and Phytochemistry*, **7**(2), 113-119.

Ramu, P., Billot, C., Rami, J.F., Senthilvel, S., Upadhyaya, H.D., Reddy, L.A., Hash, C.T., (2013) Assessment of genetic diversity in the sorghum reference set using EST-SSR markers. *Theoretical and Applied Genetics,* **126**(8), 2051-2064.

Salehi, M., Arzani, A., Talebi, M., Rokhzadi, A. (2018)

Genetic diversity of wheat wild relatives using SSR markers. *Genetika*, **50**(1), 131-141.

Salem, K.F.M., El-Zanaty, A.M., Esmail, R.M. (2008)Assessing wheat (*Triticu maestivum* L.) genetic diversity using morphological characters and microsatallite markers. *World Journal of Agricultural Sciences,* **4**(5), 538-544.

Semahegn, Y., Shimelis, H., Laing, M., Mathew, I. (2020) Evaluation of bread wheat (*Triticum aestivum* L.) genotypes for yield and related traits under drought stress conditions. *Acta Agricultur? Scandinavica. Section B, Soil and Plant Science,* **70**(6), 474-484.

Shukla, S.K., Pandey, M.P. (2008) Combining ability and heterosis over environments for yield and yield components in two-line hybrids involving thermosensitive genic male sterile lines in rice (*Oryza sativa* L.). *Plant Breeding*, **127**(1), 28–32.

Siyal, A.L., Siyal, F.K., Jatt, T. (2020) Yield from genetic variability of bread wheat (*Triticum aestivum* L.) genotypes under water stress condition: A case study of Tandojam, Sindh. *Pure and Applied Biology,* **10**(3), 841

Somers, D.J., Isaac, P., Edwards, K. (2004) A high-density microsatellite consensus map for bread wheat (*Triticum aestivum* L.). *Theoretical and Applied Genetics,* **109**(6), 1105–1114.

Swofford, D.L. (2001) PAUP: phylogenetic analysis using parsimony (and other methods), Version 4.0b8, Sinauer Associates, Sunderland, MA.

Tawfik, R.S., El-Mouhamady, A.B.A. (2019) Molecular genetic studies on abiotic stress resistance in sorghum entries through using half diallel analysis and inter-simple sequence repeat (ISSR) markers. *Bulletin of the National Research Centre,* **43**(1), 1-17.

Tayade, S.D., Potdukhe, N.R., Bharad, S., Phuke, R.M., Tayade, N.R. (2020) Combining ability analysis for yield and yield components in bread wheat. *International Journal of Chemical Studies,* **8**(6), 949-953.

Tessier, C., David, J., This, P., Boursiquot, J.M., Charrier, A. (1999) Optimization of the choice of Molecular markers for varietal identification in *Vitis vinifera* L. *Theoretical and Applied Genetics,* **89**, 171 -177.

Townsend, T., Segura, V., Chigeza, G., et al. (2013) The use of combining ability analysis to identify elite parents for Artemisia annua F1 hybrid production. *PLoS One,* **8**(4), e61989.

Tyagi, A.P., Lal, P. (2005). Line × tester analysis in sugar cane (Saccharumofficinarum). *South Pacific Journal of Natural Science,* **23**(1), 30–36.

Yadav, J., Sharma, S.N., Jakhar, M.L., Shweta (2017) Combining ability analysis for yield and its components in bread wheat over environments. *International Journal of Plant Sciences,* **12**(2), 95-101.

Zhang, Q.P., Li, Q., Wang, X.E., Wang, H.Y., Lang, S.P., Wang, Y.L., Wang, S.L., Chen, P.D., Liu, D.J. (2005) Development and characterization of a Triticumaestivum-Haynaldiavillosa translocation line T4VS.4DL conferring resistance to wheat spindle streak mosaic virus. *Euphytica*, **145**, 317–320.

Zhao, C., Bao, Y., Wang, X., Yu, H., Ding, A., Guan, C., Cui, J., et al. (2018) QTL for flag leaf size and their influence on yield-related traits in wheat. *Euphytica*, **214**, 1–15.

تعزيزالأصول الوراثية للقمح من خلال الهجن التبادلية لإنتاج سلالات جديدة

رمضان محمد إسماعيل، فتح الله برهامي فتح الله، عادل أحمد الشافعي، رشاد محمد شعيب، شيرين امين محفوظ، ابتهال صلاح الدمرداش

قسم الوراثة والسيتولوجي- معهد بحوث التقنيات الحيوية- المركزالقومي للبحوث- الجيزة- مصر.

أجريت هذه الدراسة بهدف تقييم القدرة علي التآلف لصفة المحصول ومكوناته لعدد من الهجن التبادلية الناتجة بالتهجين بين سبعة اصناف مختلفة من قمح الخبز كخطوة اولية لانتاج سلالات جديدة من قمح الخبز عالية الانتاجية ومقاومة للظروف البيئية الغير ملاءمة ومتحملة لظروف نقص المياه. واظهر تحليل التباين لكل من القدرة العامة والخاصة علي التآلف وجود فروق معنوية لكل الصفات تحت الدراسة مما يؤكد وجود اختلافات بين الاباء في قدرتها العامة والهجن في قدرتها الخاصة. وكانت غالبية الفروق بين الهجن راجعة لعلو قيم للقدرة العامة للأباء عن القدرة الخاصة للهجن وكفاءتها علي التوريث للنسل. واظهر اثنين من الآباء (الاول والرابع) قدرة عامة عالية مقارنة بباقي الآباء السبعة. كما تم التوصل إلى ستة هجن جديدة من القمح (1،2،3،5،11،17) ذات قدرة خاصة عالية جاري استغلالها في انتاج سلالات جديدة من قمح الخبز. وعلي المستوي الجزيئي والمعلمات الجزيئية تم استخدام 29 تسلسل بسيط التكرار SSR and EST-SSR لمعرفة التباعد الوراثي بين الاباء بتحليل الحمض النووي DNA. واظهرت النتائج فاعلية ال EST-SSR في التفريق بين الاباء علي المستوي الجزيئي.