

# Estimates of heterosis and association of genetic distance with heterosis in durum wheat under different moisture regimes

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## SUMMARY

The objectives of the present study were to evaluate heterosis for grain yield and yield components in durum wheat, and to assess the prediction potential of amplified fragment length polymorphism (AFLP) based and agronomic trait based genetic distances (GD and MD, respectively) to F<sub>1</sub> performance, mid parent heterosis (MPH), and specific combining ability effects (SCA) under well-watered and moisture stress conditions. Six parental genotypes with different responses to moisture stress and their 15 F<sub>1</sub> crosses were evaluated for their responses to moisture stress conditions in a glasshouse. Some cross combinations showed significant MPH for grain yield and yield components. The expression of heterosis for grain yield was greater under moisture stress conditions than under well-watered conditions. Cluster analysis of the parental lines based on agronomic performance under stress conditions was similar to cluster analysis result based on AFLP marker profiles. F<sub>1</sub> performance was strongly correlated to both SCA effects and MPH under both stress and well water conditions. The correlation between SCA and MPH was very high under both treatment conditions for all traits. Correlation between GD and MD was significant only under stress conditions. Positive correlation was found only for the association between GD v. F<sub>1</sub> performance and GD v. SCA effects for harvest index (HI) under well-watered conditions. None of the correlations between MD and SCA effects were significant. The absence of association between GD and heterosis for yield and most agronomic traits implied that heterozygosity per se diversity is not a good predictor of heterosis or F<sub>1</sub> performance under both well-watered and stressed conditions.

## INTRODUCTION

One strategy to improve drought tolerance in many crops has been to exploit heterosis based on the premise that heterosis for yield and yield components increases under stress conditions. In maize (Betrán *et al.* 2003) and pearl millet (Yadaw *et al.* 2000), high estimates of heterosis for yield have been obtained under stress conditions. In barley, it has been shown that heterozygosity rather than heterogeneity is more important for grain yield under drought conditions (Einfeldt *et al.* 2005). At present, however, the information on the amount of heterosis in durum wheat, particularly under contrasting moisture regimes, is limited.

In cross-pollinated crops such as maize and sunflower, molecular markers have been successfully used to place genotypes into heterotic groups (Cheres *et al.* 2000). However, in many crops such as chickpea, cotton, pearl millet, rice and wheat, correlations of marker heterozygosity or genetic distance with heterosis have been low (Zhang *et al.* 1994; Ajmone-Marsan *et al.* 1998; Chowdari *et al.* 1998; Meredith & Brown 1998; Sant *et al.* 1999). An exception to this was sunflower, which had a high correlation value between genetic distance and heterosis (Cheres *et al.* 2000). In wheat, Martin *et al.* (1995) found significant associations between pedigree-based genetic distance and heterosis only for kernel weight and protein concentration.

Environment can differentially affect the performance of pure lines and hybrids, altering the relationship between genetic distance and heterosis. The

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influence of moisture stress on the use of amplified fragment length polymorphism (AFLP) based genetic distance (GD) as predictor of hybrid performance is poorly understood.

The objectives of the present study were to evaluate and quantify heterosis for grain yield and yield components under well-watered and stressed conditions, and to evaluate the prediction potential of AFLP based (GD) and agronomic trait based genetic distances (MD) to  $F_1$  performance, mid parent heterosis (MPH) and specific combining ability effects (SCA) under well-watered and moisture stress conditions.

## MATERIALS AND METHODS

Two drought-tolerant varieties, Boohai 'S' and DZ-2023, two moderately drought-tolerant varieties, DZ-1691 and DZ-320, and two susceptible varieties, Klinto and LD-357 (Solomon *et al.* 2003) were crossed in a half diallel design to generate 15  $F_1$  hybrids. Parental lines and their  $F_1$  progenies were evaluated under well-watered and simulated moisture stress conditions in the glasshouse during 2001/02 at the University of the Free State, South Africa (29°06'S, 26°18'E, 1351 m asl). The temperature in the glasshouse was maintained at an average of 25 and 15 °C day and night, respectively. Treatments were arranged in a randomized complete block design with three replications. Entries were planted in 10-litre plastic pots filled with 2 kg of gravel (to prevent soil leakage), and 7 kg of steam sterilized and air-dried sandy loam soil with proportions (by volume) of sand, silt and clay, of 0.8, 0.02 and 0.18 respectively. The air-dry moisture content of the soil was determined gravimetrically as 133 (field capacity) and 40 g moisture/kg (permanent wilting point). Pots were equally spaced (50 mm within a block, 1 m between blocks). Each genotype was sown into six pots; 12 seeds planted at equal spacing and equal depth. Seedlings were thinned to nine per pot at the two-leaf growth stage. Pots were maintained at 700–1000 g available soil moisture/kg for 1 month (up to about the four-leaf growth stage). Half of the experimental unit was then left without water until severe wilting was observed. The amount of water evaporated was monitored daily by weighing unplanted pots placed between planted pots in both the stress and the non-stress treatments in each block (six in each block). The amount of water transpired was determined by subtracting the weights of unplanted pots from the weights of the planted pots. Pots were replenished with the amount of water equivalent to the loss in weight to bring them to the predetermined level of moisture whenever the weight of pots fell to the lower limit established for the treatments. The moisture levels were 250–350 and 700–1000 g available soil moisture/kg for the stress and control treatments, respectively, until maturity.

Number of spikes per plant (spikes completely emerged from flag leaf ligules) was counted for all plants. At maturity, three plants per 10-litre pot were harvested and left to air dry for 10 days. Biomass was determined from plants harvested at soil level. Grain and biomass yield were determined as an average of the three randomly selected plants. Harvest index was calculated as the ratio of grain yield to biomass yield. Number of kernels per spike was calculated as total number of kernels to total number of spikes per plant. Individual kernel weight was determined from the weight of all seeds harvested divided by the total number of kernels.

Twenty  $F_1$  plants per cross and the six parental lines were grown in a glasshouse. DNA was extracted separately from 10 healthy plants of 15  $F_1$ s and six parental lines according to the modified monocot method (Edwards *et al.* 1991). Equal concentration of DNA from the 10 plants was bulked to form a sample for each  $F_1$  hybrids and parents. AFLP analysis (Vos *et al.* 1995) was carried out with fluorescent (FAM and NED) labelled +3/+3 *EcoRI* primers. A 25  $\mu$ l restriction digestion reaction was set up with 5  $\mu$ l of genomic DNA (250 ng), 5 $\times$ reaction buffer (50 mM Tris-HCL (pH 7.5), 50 mM Mg-acetate and 250 mM K-acetate), 2.5  $\mu$ l of DNA (100 ng/ $\mu$ l), 2  $\mu$ l of *EcoRI*/*MseI* (1.25 units/ $\mu$ l) restriction enzymes. The digested fragments were then ligated with 50 pMol *MseI* adapter (5'-GAC GAT GAG TCC TGA G-3', 3'-TA CTC AGG ACT CAT-5') and 5 pMol *EcoRI* adapter (5'-CTC GTA GAC TGC GTA CC-3', 3'-CAT CTG ACG CAT GGT TAA-5') and T4 DNA ligase (1 unit/ $\mu$ l). The ligation product was diluted 1:10 in TE (10 mM Tr-s-HCL (pH 8.0), 0.1mM EDTA) buffer.

Pre-selective reactions were performed with *EcoRI* + 1 and *MseI* + 1 selective primers in 51  $\mu$ l containing 5  $\mu$ l of diluted ligation product (1:10 in TE), 40  $\mu$ l of pre-amp primer mix (T4 polynucleotide kinase (10 units/ $\mu$ l in 50 mM Tris-HCl (pH = 7.6), 5 $\times$  kinase buffer (350 mM Tris-HCl (pH 7.6), 50 mM MgCL<sub>2</sub>, 500 mM KCl, 5 mM 2-mercaptoethanol)), 5  $\mu$ l of 10 $\times$ PCR buffer (100 mM Tris-HCl (pH 8.3), 15 mM MgCL<sub>2</sub>, 500 mM KCl) and 1 U of Ampli-Taq DNA polymerase (GibcoBRL). PCR amplification consisted of 20 cycles at 94 °C for 30 s, 56 °C for 60 s and 72 °C for 60 s. Pre-selective amplification products were diluted (1:50) in TE buffer (10 mM Tris-HCL (pH = 8.0) and 0.1 mM EDTA).

Selective PCR was performed in 20  $\mu$ l PCR reaction containing 5  $\mu$ l of the diluted pre-selective reaction product (1:50), 4.5  $\mu$ l of the *MseI* primer (6.7 ng/ $\mu$ l) (5'-GAT GAG TCC TGA GTA A-3') + 3 (CAG, CTA and CTT) and 1  $\mu$ l of *EcoRI* primer (27.8 ng/ $\mu$ l) (5'-GAC TGC GTA CCA ATT C-3') + 3 (ACA labelled with FAM and AAC NED) 2  $\mu$ l of 10 $\times$ PCR buffer (100 mM Tris-HCl (pH = 8.3), 15 mM MgCL<sub>2</sub> and 500 mM KCl) and 5U of Ampli Taq DNA polymerase. Reactions were performed on an ABI

2700 Perkin-Elmer thermal cycler at the following conditions: 12 cycles at 94 °C for 30 s, 65 °C for 30 s and 72 °C for 60 s, during which the annealing temperature was lowered by 0.7 °C per cycle followed by 23 cycles at 94 °C for 30 s, 56 °C for 30 s and 72 °C for 60 s. A total of six selective primer combinations were used. After selective PCR, 5 µl of the selective amplification product was added to 24 µl formamide and 1 µl of Rox standard size marker, denatured for 10 min at 94 °C and quickly cooled on ice. The AFLP fragments were resolved on an automatic ABI 310 capillary sequencer (PE Biosystems).

Each measured trait was subjected to ANOVA using the GLM procedure (SAS 1999). Least squares means were calculated for each entry. Mid-parent heterosis (MPH) values were calculated as a deviation percentage.

Mid-parent (MP) heterosis (%) was computed as: Mid-parent heterosis =  $[\text{Progeny}_{ij} - (p_i + p_j)/2] / [(p_i + p_j)/2] * 100$ , where  $p_i$  = performance of parental line  $i$ ,  $p_j$  = performance of parental line  $j$  and  $\text{progeny}_{ij}$  = progeny performance of a cross of parents  $i$  and  $j$ .

General (GCA) and specific combining ability (SCA) were calculated using SAS (Zhang & Kang 1997) based on the Griffing (1956) fixed effects model using both parents and  $F_1$ s without the reciprocals.

AFLP fragments were coded as 1 for presence and 0 for absence. Distance matrices were compiled for all pairs of genotypes from binary data based on both unique and shared bands (AFLP) using the Euclidean distance method (Kaufman & Rousseeuw 1990). The distance matrix generated using AFLP data is referred to as genetic distance (GD). In order to compute genetic distance matrices based on agronomic performance, mean traits performances were first (normalized),  $Z$  transformed to standardized units. The agronomic dissimilarity matrices based on all measured traits were calculated using the Euclidean distance method. The distance obtained using agronomic traits is termed morphological distance (MD). Heterosis and SCA distance matrices were created from SCA and mid-parent mean heterosis  $Z$  transformed values.

Cluster analysis was performed on the genetic distance matrices generated by the Euclidean distance method to reveal the pattern of genetic relationships among genotypes using the unweighted pair group method with arithmetic averages (UPGMA) in the NCSS computer package (Hintze 2000).

The correlation between the distance matrices was computed and its significance was determined based on 10 000 permutations according to Mantel (1967).

## RESULTS

### *Estimates of heterosis*

Mean performance of parental lines and their crosses is given in Table 1. Drought stress caused substantial

yield reduction. The extent of yield reduction due to moisture stress, however, was different among the parental lines and the various cross combinations.

Mid parent heterosis (MPH) represents the average deviation of an  $F_1$  compared with the mean performance of its two parents. The average MPH for grain yield was 24.5 and 39.8% under well-watered (WW) and moisture stress (S) conditions, respectively. Significant MPH for grain yield under well-watered conditions ranged from -34% (DZ-1691 × LD-357) to 72% (DZ-2023 × DZ-1691). Under stress conditions, significant MPH ranged from -42% (Boohai'S' × LD-357) to 127% (DZ-2023 × DZ-320) (Table 2). Estimates of MPH were generally higher under moisture stress conditions compared with the well-watered conditions. The most important yield components, kernel number and kernel weight, also exhibited different MPH values among crosses within, and sometimes between, well-watered and moisture stress conditions (Table 2). Heterosis for grain yield appears to be associated with kernel number, kernel weight, or both, depending on the cross combinations and environmental conditions. Mid-parent heterosis for grain yield under well-watered conditions seems to be associated mainly with kernel number. However, under moisture stress, the high MPH for grain yield was mainly due to the high MPH for kernel weight ( $r = 0.60$ ;  $P < 0.05$ ).

Mean squares due to GCA and SCA effects as well as the interactions of combining abilities effects with moisture treatments were highly significant for all traits ( $P < 0.001$ ). The relative importance of non-additive and additive gene actions or SCA/GCA ratios was different for the different traits and treatment conditions. The SCA/GCA ratio for grain yield was higher (0.43) under well-watered conditions compared with moisture stress conditions (0.31). To visualize SCA advantages for observed grain yield performance of  $F_1$  hybrids, the expected yield of each cross was plotted against its observed yield (Fig. 1). Under well-watered conditions, there was high variability in SCA values among the different crosses. DZ-2023 × DZ-1691, Boohai'S' × DZ-1691 and Boohai'S' × DZ-320 showed positive and significant deviations from the line of expectancy (Fig. 1a). These crosses also showed the highest MPH for grain yield under well-watered conditions (Table 2). Boohai'S' × Klinto and DZ-1691 × LD-357 deviated significantly below the expected line (Fig. 1a); and these crosses had the most negative MPH under well-watered conditions (Table 2). Only a few crosses; DZ-2023 × Boohai'S', DZ-320 × LD-357 and DZ-1691 × LD-567 showed clear SCA advantages for grain yield under moisture stress conditions. Boohai'S' × LD-357 had the least SCA advantage for grain yield under stress conditions, which resulted in the lowest MPH for grain yield. All the other crosses showed limited variability regarding SCA effects, as the observed values fell more or less

Table 1. Mean values of grain yield, kernel number per spike and kernel weight for parents and F<sub>1</sub> hybrids of durum wheat grown at well-watered (WW) and stress (S) moisture conditions

Parent/cross	Grain yield (g/plant)		Kernels/spike		Kernel weight (mg)	
	WW	S	WW	S	WW	S
DZ-2023	2.7	0.6	22	13	37	26
Boohai'S'	2.6	0.8	16	11	51	55
Klinto	3.3	0.3	16	10	33	34
DZ1691	2.5	0.5	15	6	36	36
LD-357	1.0	0.2	12	6	27	32
DZ-320	1.6	0.6	13	10	40	31
1 × 2	2.8	1.3	24	11	46	44
1 × 3	3.0	0.7	32	18	36	45
1 × 4	4.5	0.9	41	9	47	30
1 × 5	2.8	0.6	12	6	55	41
1 × 6	3.4	1.3	19	22	47	46
2 × 3	2.7	0.6	22	10	45	34
2 × 4	3.9	0.5	32	10	42	38
2 × 5	2.1	0.3	10	6	53	18
2 × 6	3.4	0.9	21	11	52	63
3 × 4	3.8	0.4	19	11	68	23
3 × 5	2.7	0.3	14	5	50	53
3 × 6	2.6	0.8	15	13	44	35
4 × 5	1.1	0.4	7	5	55	45
4 × 6	2.5	0.6	23	10	41	26
5 × 6	1.5	0.8	10	7	47	54
Mean	2.69	0.64	19	10	45	39
s.d.	0.89	0.30	8.3	4.2	9.1	11.7

Table 2. Estimate of mid parent heterosis (MPH) of grain yield, kernel number and kernel weight for 15 F<sub>1</sub> hybrids of durum wheat grown in well-watered (WW) and stress (S) moisture conditions. Probability values are indicated in parentheses

Cross	Grain yield (g/plant)		Kernels/spike		Kernel weight (mg)	
	WW	S	WW	S	WW	S
1 × 2	6.6	94.1 (0.001)	30.9 (0.05)	-10.0	5.6	7.7
1 × 3	0.8	69.8 (0.001)	70.2 (0.001)	58.1 (0.001)	2.9	49.9 (0.001)
1 × 4	71.9 (0.001)	66.3 (0.001)	122.0 (0.001)	-4.5	30.5 (0.01)	-2.1
1 × 5	52.8 (0.001)	55.0 (0.001)	-27.5 (0.05)	-39.9 (0.001)	71.0 (0.001)	43.4 (0.01)
1 × 6	55.7 (0.001)	126.7 (0.001)	9.4	89.3 (0.001)	23.2 (0.05)	61.6 (0.001)
2 × 3	-10.1 (0.05)	2.8	37.8 (0.01)	-1.2	7.5	-25.1 (0.05)
2 × 4	54.1 (0.001)	-22.5	107.3 (0.001)	13.1	-2.6	-17.2
2 × 5	15.6 (0.01)	-41.6 (0.01)	-26.3 (0.05)	-27.3 (0.05)	36.3 (0.01)	-59.2 (0.001)
2 × 6	62.0 (0.001)	35.9 (0.05)	42.3 (0.01)	5.6	14.1	46.0 (0.001)
3 × 4	31.7 (0.001)	9.1	22.0	35.7 (0.01)	97.0 (0.001)	-36.0 (0.01)
3 × 5	24.3 (0.001)	-3.7	2.3	-34.7 (0.01)	66.1 (0.001)	59.0 (0.001)
3 × 6	5.0	85.7 (0.001)	6.0	28.9 (0.01)	19.6	6.1
4 × 5	-34.5 (0.001)	11.0	-51.3 (0.001)	-21.0 (0.05)	74.2 (0.001)	33.4 (0.01)
4 × 6	20.1 (0.001)	15.1	61.5 (0.001)	38.4 (0.001)	7.3	-23.7
5 × 6	11.4 (0.05)	93.5 (0.001)	-23.0	-16.3	39.5 (0.001)	70.3 (0.001)
Mean	24.50	39.82	25.57	7.61	32.81	14.27
s.e.	4.641	14.279	12.558	10.143	9.889	11.963

Parents are: 1 = DZ-2023, 2 = Boohai'S', 3 = Klinto, 4 = DZ-1691, 5 = LD-357 and 6 = DZ-320.

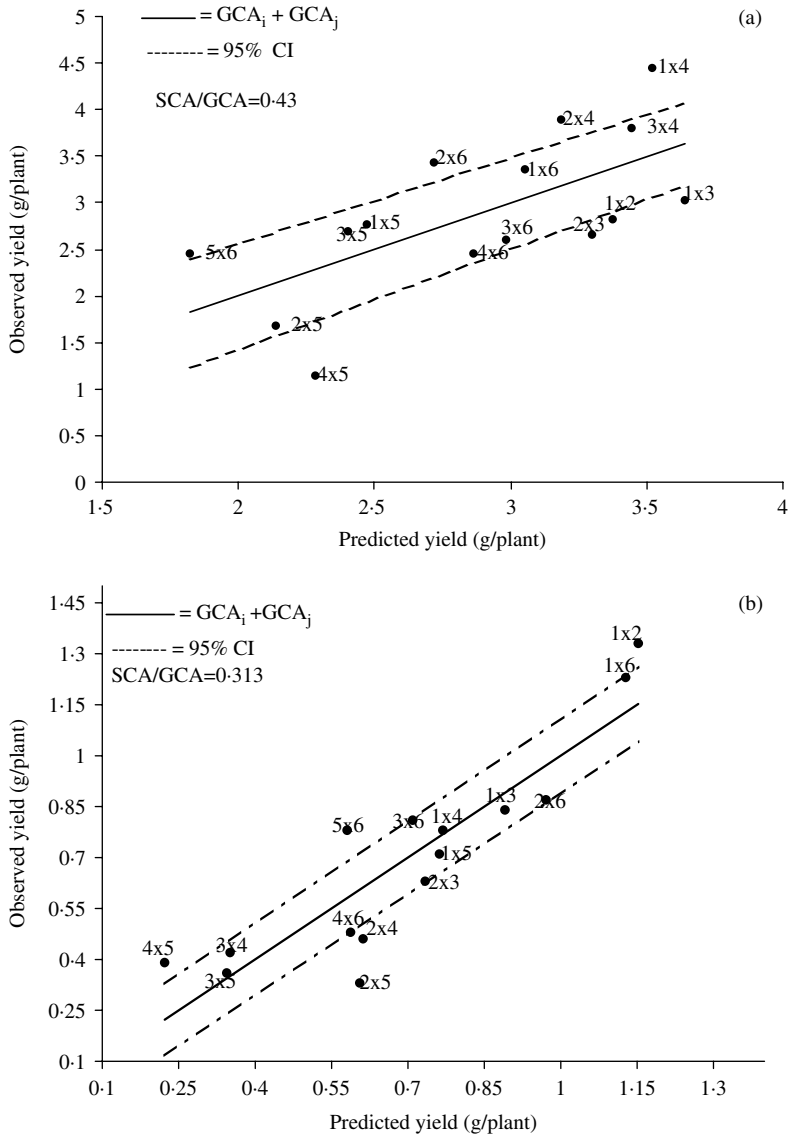


Fig. 1. Observed versus predicted relationships for (a) grain yield under well-watered conditions and (b) grain yield under stress conditions. Parents are: 1 = DZ-2023, 2 = Boohai'S', 3 = Klinto, 4 = DZ-1691, 5 = LD-357 and 6 = DZ-320.

within 95% confidence interval of the predicted values (Fig. 1b). The observed high estimate of MPH for grain yield for many of these cross combinations under stress conditions (Table 2) was mainly due to high GCA effects of the corresponding parental lines.

*Genetic analyses*

Out of 465 fragments obtained from the six primer combinations, on average, 0.3 of the fragments were polymorphic across the six parental lines and 15

hybrids. The dendrogram generated from combined polymorphic genetic distance matrix resulted in two clusters (Fig. 2). DZ-2023, Boohai'S' and DZ-320 were placed in the first cluster, while the other moderately tolerant, DZ-1691 and the two susceptible parental lines were grouped together: The cophenetic correlation for the cluster analysis was very strong ( $r=0.94$ ).

The dendrogram generated from agronomic traits distance matrices failed to clearly group the parental genotypes under well-watered conditions (Fig. 3a).

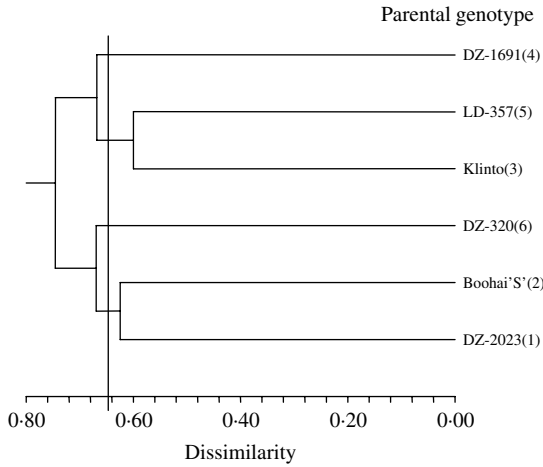


Fig. 2. Dendrogram depicting genetic relationships of six durum wheat genotypes differing in drought tolerance based on AFLP analysis. Numbers in parentheses indicate parent number in the cross. The vertical line indicates the limit of significance.

Under stressed conditions, however, the two tolerant parents, DZ-2023 and Boohai'S' were grouped together while the susceptible parent; LD-357 and the two moderately tolerant parents were clustered together. The other susceptible parent, Klinto, was found to be unique (Fig. 3b).

*Correlation of distance matrices*

F<sub>1</sub> performance was strongly correlated with both MPH and SCA effects distance matrices for all analysed traits and treatment conditions (Table 3). Moreover, the associations between MPH and SCA matrices also were positive and significant (Table 3). The correlation between GD and F<sub>1</sub> performance was positive and significant only for HI under well-watered conditions. The association between MD and F<sub>1</sub> was significant but negative only for grain yield and kernel number under well-watered conditions (Table 3). Neither GD nor MD was correlated with MPH under any of the conditions, except for a negative association between MD and MPH for kernel number under well-watered conditions. None of the associations between SCA and GD or between SCA and MD matrices was significant, except for the association between SCA and GD for HI under well-watered conditions (Table 3). The correlation between MD and GD was significant ( $r=0.54$ ;  $P<0.05$ ) only at moisture stress conditions.

**DISCUSSION**

*Estimates of heterosis*

Hybrid wheat has the potential for improved performance and stability across different environments.

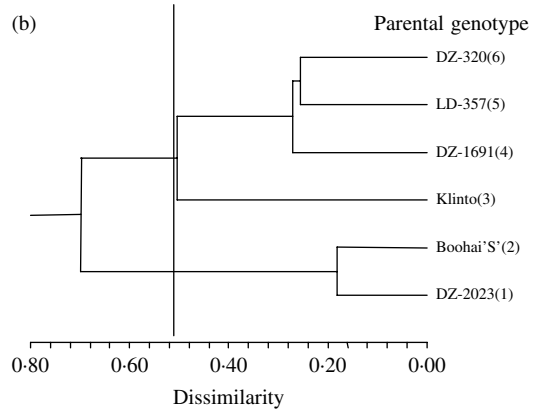
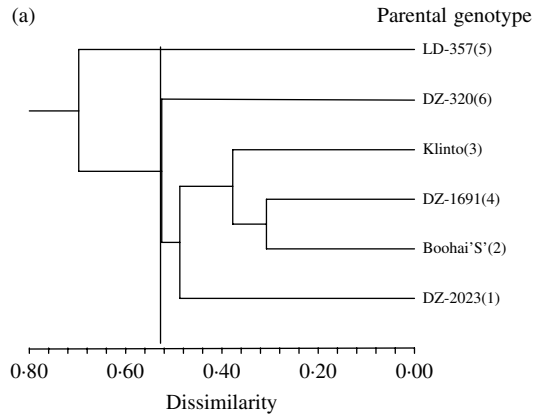


Fig. 3. Dendrograms computed from agronomic traits for six durum wheat genotypes grown (a) under well-watered and (b) stress conditions. Numbers in parentheses indicate parent number in the cross. The vertical line indicates the limit of significance.

However, according to Dreisigacker *et al.* (2005), three prerequisites are considered crucial for its successful implementation: (i) a cost effective system of seed production, (ii) an adequate amount of heterosis and (iii) the development of heterotic groups and patterns to ensure future progress through hybrid breeding. These conditions are met in cross-pollinated crops such as maize. However, in self-pollinated crops although progress has been made through the use of chemical hybridizing agents, the level of heterosis and absence of heterotic groups remain the limiting factors in hybrid wheat breeding programmes. Some (Austin 1999; Maluszynski *et al.* 2001; Fasoula & Fasoula 2002) also believe that a large part of heterosis should be fixed in pure lines through conventional breeding.

Mid-parent heterosis for grain yield was found to be as high as 72 and 127% under well-watered and moisture stress conditions, respectively. The highest

Table 3. Significant correlation coefficient (*r*) among *F*<sub>1</sub> performance (*F*<sub>1</sub>), specific combining ability (SCA), mid parent heterosis (MPH), genetic distance (GD) and morphological distance (MD) estimated from 15 *F*<sub>1</sub> hybrids of durum wheat evaluated under well-watered (WW) and stressed (S) conditions. Probability values are indicated in parentheses

Trait	Moisture	<i>r</i> ( <i>F</i> <sub>1</sub> MPH)	<i>r</i> ( <i>F</i> <sub>1</sub> SCA)	<i>r</i> (MPH SCA)	<i>r</i> ( <i>F</i> <sub>1</sub> GD)	<i>r</i> ( <i>F</i> <sub>1</sub> MD)	<i>r</i> (GD MPH)	<i>r</i> (MD MPH)	<i>r</i> (GD SCA)	<i>r</i> (MD SCA)
Grain yield	WW	0.78 (0.01)	0.78 (0.01)	0.96 (0.001)	ns	-61 (0.05)	ns	ns	ns	ns
	S	0.87 (0.001)	0.89 (0.001)	0.91 (0.001)	ns	ns	ns	ns	ns	ns
Kernels/spike	WW	0.97 (0.001)	0.90 (0.001)	0.93 (0.001)	ns	-67 (0.05)	ns	-0.73 (0.01)	ns	ns
	S	0.92 (0.001)	0.85 (0.001)	0.90 (0.001)	ns	ns	ns	ns	ns	ns
Kernel weight	WW	0.85 (0.001)	0.96 (0.001)	0.92 (0.001)	ns	ns	ns	ns	ns	ns
	S	0.89 (0.001)	0.95 (0.001)	0.92 (0.001)	ns	ns	ns	ns	ns	ns
Harvest Index	WW	0.78 (0.01)	0.90 (0.001)	0.93 (0.001)	0.56 (0.05)	ns	ns	ns	0.62 (0.01)	ns
	S	0.83 (0.001)	0.81 (0.001)	0.95 (0.001)	ns	ns	ns	ns	ns	ns

estimate of heterosis obtained at well-watered conditions is comparable with 87% MPH for grain yield in similar studies in durum wheat based on field studies (Budak 2001). However, higher (>100%) MPH for grain yield in bread wheat has been reported (Akhter *et al.* 2003; Singh *et al.* 2004). The average MPH for grain yield was higher in moisture stress conditions compared with absence of moisture stress. The highest level (72%) MPH for grain yield of the cross, DZ-2023 × DZ-1691, was almost doubled (127%) due to moisture stress. Heterosis is the manifestation of heterozygosity. Greater MPH due to moisture stress is to be expected due to the comparatively poor performance of the homozygous parental lines. Einfeldt *et al.* (2005) also concluded that heterozygosity is more important for grain yield improvement under drought conditions. In the present experiment, it was observed that some crosses showed high heterosis for grain yield both under stress and well watered conditions. However, few crosses showed a high amount of MPH only under stressed or well-watered conditions (Table 2). This suggests that some cross combinations could be identified for wider adaptation. Nevertheless, evaluating the materials in their target environments could also help identify specifically adapted hybrids.

Mid-parent heterosis for grain yield could be due to high MPH for one or more of the yield components (Kindred & Gooding 2005; Singh *et al.* 2004). Mid-parent heterosis for kernel number and kernel weight was not consistent across environments and cross combinations. Depending on the cross combinations, MPH for grain yield under well-watered conditions seems to be due to high MPH for kernel number. Under stressed conditions, however, the correlation between MPH for grain and kernel weight was significant (*r* = 0.60; *P* < 0.05). This suggests that heterosis in grain yield under a stress situation could be mainly due to high heterosis for kernel weight. Singh *et al.* (2004) also reported the inconsistency of the role of various yield components in the expression of heterosis in grain yield between early, normal and late planting dates in bread wheat. Prasad *et al.* (1998) reported that heterosis for grain number and kernel weight was independently associated to heterosis for grain yield per plant.

Analysis of the relative importance of GCA v. SCA effects provides an indication of the type of gene action involved in the expression of traits and allows inferences in the optimum allocation of resources in the hybrid-breeding programme. In the present experiment, both GCA and SCA effects for grain yield were important, suggesting both additive and non-additive genetic effects are important for inheritance of grain yield. However, the relative importance of SCA effects was lower than that of GCA effects. This agrees with previous studies in maize and wheat (Betrán *et al.* 2003; El-Maghraby *et al.* 2005;

Dreisigacker *et al.* 2005). The relationship between observed *v.* predicted  $F_1$  yield performance showed that SCA advantage is high when parents from different drought-tolerance categories are crossed. Theoretical and experimental results in maize show that SCA effects are of primary importance in intra-group crosses, whereas GCA effects are predominant in inter-group crosses (Melchinger & Gumber 1998; Betrán *et al.* 2003).

#### *Genetic analyses*

The absence of clear clustering based on the analysis of agronomic traits under well-watered conditions implies that the parental lines perform similarly under normal conditions. Clear separation among parental genotypes under stressed conditions, nevertheless, implies that those genotypes had different responses to imposed moisture stress (Solomon *et al.* 2003). The six primer combinations employed to analyse the diversity among the six parental lines and their 15  $F_1$ s demonstrated that the proportion of AFLP fragments that were polymorphic was 0.30. The amount of polymorphism obtained in the present study was comparable to previous diversity studies in durum wheat (Medini *et al.* 2005). The clustering of parents based on agronomic traits and AFLP markers profile was very similar. Moghaddam *et al.* (2005) also reported similarity between phenotypic and AFLP profile-based clustering for some drought tolerant Iranian wheat materials.

The significant correlation between AFLP based (GD) and agronomic trait based (MD) distances also points to the existence of genetic variation with respect to performance under moisture stress conditions among parental lines. A similar study in rice showed that difference in performance among genotypes differing in their response to moisture stress was also due to their genetic difference (Mathew *et al.* 2000). Nachit *et al.* (2000) demonstrated that differences in adaptation and productivity in dry land environments in durum wheat is strongly correlated with genetic variability based on AFLP data.

#### *Correlation of distance matrices*

$F_1$  performance was significantly and positively correlated to both SCA effects and MPH for both treatment conditions and for all the traits analysed. This agrees with similar experiments in maize (Betrán *et al.* 2003). Moreover, much of the variation in SCA effects for grain yield was within 95% confidence interval (CI) boundaries predicted by  $GCA_i + GCA_j$  (Fig. 1). In wheat, positive associations between  $F_1$  performance and mid parent values have been reported (Dreisigacker *et al.* 2005). This suggests that relatively better  $F_1$  progenies are likely to be obtained if parents from the different drought-tolerance

categories with good GCA effects are crossed. Thus, hybrid breeding should be relatively efficient based on selection for parental performance under the target environment and a relatively smaller number of test crosses involving outstanding parental lines under the target environment. The magnitude of correlation between SCA and MPH was very similar to the magnitude of correlation coefficients between  $F_1$  *v.* SCA and  $F_1$  *v.* MPH. This suggests that  $F_1$  performance can be predicted using either SCA or MPH estimates. However, prediction based on SCA effects could be more reliable as MPH estimates are dependent on the performance of parental lines (Betrán *et al.* 2003).

In the present experiment, it was found that AFLP based genetic distance (GD) was correlated significantly only to  $F_1$  performance and SCA effects for HI under well-watered conditions. The correlation between GD and MPH in the present study was not significant under any of the treatment conditions. In many crop species, such as chickpea, cotton, maize, pearl millet, rice and wheat, correlations of genetic distance based on markers' heterozygosity between individuals with heterosis have been of no or low significance (Zhang *et al.* 1994; Ajmone-Marsan *et al.* 1998; Chowdari *et al.* 1998; Meredith & Brown 1998; Sant *et al.* 1999; Betrán *et al.* 2003; Dreisigacker *et al.* 2005; El-Maghraby *et al.* 2005). Some studies in rice (Zhang *et al.* 1994) and alfalfa (Riday *et al.* 2003) have shown that distance based on random genetic markers is a poor predictor of heterosis, probably due to noise resulting from markers not linked directly to the trait being studied. The correlation between MD and  $F_1$  was also not significant for most of the traits. The significant correlation coefficients observed between MD and  $F_1$  for grain yield and kernel number at well-watered conditions were negative indicating that increases in parental lines divergent in terms of yield or kernel number would result in poor  $F_1$  hybrids. The correlations between MPH and MD for all traits under both stress and well-watered conditions were not significant, except for kernel number at well-watered conditions. Earlier studies in wheat (Fabrizius *et al.* 1998) also showed that there was no linear association between MD and heterosis. This implies that morphological trait-based trends of diversity cannot be used to predict heterosis.

In conclusion, the present study showed that considerably high-level heterosis for grain yield was found under the different treatment conditions for some crosses of durum wheat. This highlights the potential of some crosses in hybrid productions. However, further evaluation under diverse field conditions may prove essential before a decision is made to use them in hybrid breeding programmes. The absence of significant correlation between GD and MD with SCA,  $F_1$  performance or MPH for most of the traits under both stress and well-watered conditions indicates that simple diversity measures based on



general heterozygosity among parental lines cannot be a useful predictor of heterosis or hybrid performance under either moisture stress or normal growing conditions.

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