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ESTIMATES OF HETEROSIS FOR SEED YIELD AND OIL CONTENTS IN SUNFLOWER (*Helianthus annuus* L.)

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ABSRACT

Eight sunflower parents and their sixteen F_1 hybrids were evaluated at Agricultural Research Institute, Tarnab, Peshawar during autumn 2003-04 to estimate mid and high parent heterosis for seed yield and oil content. A randomized complete block design with three replicates was used. Highly significant genetic differences (P<0.01) were observed among parents and F_1 hybrids for yield hectare^{-1}, harvest index, moisture factor and oil content. Mid parent and high parent heterosis estimates of F_1 hybrids ranged from 5.60 to 185.02% and –9.06 to 181.73% for yield hectare^{-1}, 23.33 to 171.66% and –43.91 to 127.36% for harvest index, 11.19 to –30.35 and 19.13 to –20.71% for moisture factor and –4.78 to 52.85% and –18.39 to 42.50% for oil content, respectively. Hybrid TS-18 x 291RGI showed highest positive mid and high parent heterotic effects for yield hectare^{-1}, TS-335 x 291RGI expressed highest positive mid and high parent heterotic magnitude for harvest index, TS-228 x 291RGI had highest negative mid and high parent heterosis for moisture factor and TS-335 x 291RGI expressed highest positive mid and high parent heterotic estimates for oil content. Based on mean performance and mid and high parent heterotic effects for seed yield and oil traits, parents of these three hybrids are suggested for use in sunflower breeding program.

INTRODUCTION

The cultivated sunflower (*Helianthus annuus* var. marcocarpus) ranks with soybean (*Glycine max* L.), rapeseed (*Brassica Campestris* L. and *B. niapus* L.) and peanut (groundnut) (*Arachis hypogae*) as one of the four most important annual crops in the world grown for edible oil (Heiser *et al;* 1969). Sunflower is the third major supplier of edible oil in the world after soybean and groundnut (Meric, 2003). It occupies an important place among oilseed crops in the world market and its production has multiplied by approximately 1.8 during last 20 years (Pouzet and Delplancke, 2000). The first stable source of cytoplasmic male sterility in sunflower was discovered by Leclercq from an interspecific cross involving *Helianthus petiolaris* Nutt. and *Helianthus annuus* L. (Leclercq 1969). Subsequent identification of genes for fertility restoration was made by Kinman (1970), which helped in efficient and economical production of hybrid seed. Cytoplasmic male sterility system is used for hybrid seeds production in *Helianthus annuus* L. since 1972 (Fick and Miller, 1997). The main objectives in sunflower breeding are; increase in seed yield, harvest index, assimilate acceptor, resistance to major diseases and pets, as well as early maturity, short stalk and uniform height. High oil content in seed and oil quality are important objectives when breeding high oil sunflower varieties and hybrids. Seed size, uniformity, appearance and colour and a high kernel-to-hull ratio are important when breeding non-oilseeds (Fick *et al*; 1978).

MATERIALS AND METHODS

Eight Sunflower parents i.e. four Cytoplasmic male sterile (CMS) lines TS-17, TS-18, TS-228 and TS-335 and four Restorer lines 291RGI, R-25, TR-9 and TR-6023 were manually crossed during autumn 2003 to produce 16 F_1 hybrids. These sixteen F_1 hybrids were TS-17 × 291RGI, TS-17 × R-25, TS-17 × TR-9, TS-17 × TR-6023, TS-18 × 291RGI, TS-18 × R-25, TS-18 × TR-9, TS-18 × TR-6023, TS-228 × 291RGI, TS-228 × R-25, TS-228 × TR-9, TS-228 × TR-6023, TS-335 × 291RGI, TS-335 × R-25, TS-335 × TR-9, TS-335 × TR-6023. The F_1 hybrids along with their eight parents were evaluated at Agricultural Research Institute, Tarnab, Peshawar during autumn 2004 using a Randomized Complete Block Design (RCBD) with three replicates. Each hybrid and parental line was planted in five-meter long rows with plant-to-plant distance of 0.30 m and row-to-row distance of 0.75 m. A basal fertilizer doze of 120 kg hectare^{-1} Nitrogen (Urea) and 60 kg hectare^{-1} of Phosphorus (Diammonium Phosphate) were applied. Full doze of DAP and half doze of Nitrogen was applied at the time of sowing, while the remaining half doze of nitrogen was applied just before head initiation.

Ten plants were randomly selected from each plot to record data on yield hectare^{-1} (kg), harvest index and moisture factor. Oil content (%) was calculated on three samples from the bulked seeds of each genotype and analyzed at Pakistan Council of

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Scientific and Industrial Research (PCSIR) Laboratories Complex Peshawar, where oil content was determined on a 10 g oven dried achene sample by Nuclear Magnetic Resonance (NMR) procedure (Granlund and Zimmerman, 1975).

The data were analyzed using MSTAT-C statistical software and least significance difference (LSD) test was used for mean separation. Mid parent and High parent heterosis (heterobeltiosis) were computed for each trait using the following formula (Sharma and Singh 1978).

a. Mid-Parent Heterosis (%) = (F_1 – MP/MP) x 100
b. High-Parent Heterosis or heterobeltiosis (%) = (F_1 – HP/HP) x 100

The significance of F_1 hybrids vs Mid-Parents and High-Parents means was determined via t-test of Wynne *et al*., (1970) as follow: -

a. t-test for mid parent = $(F_1 – MP)/\sqrt{3/8 \times \delta^2_E}$

b. t-test for high parent = $(F_1 – HP)/\sqrt{3/8 \times \delta^2_E}$

Wherein,

F_1 = mean of F_1 hybrid

MP = mean of two parents for a trait in a cross (parent 1 + parent 2)/2

HP = mean of high parent for a trait in a cross.

δ^2_E = mean squares of the pooled error (MSE)

RESULTS AND DISCUSSION

Yield Hectare^{-1}

Highly significant differences (P<0.01) were observed among parents and their F_1 hybrids for yield hectare^{-1} (Table-I). Lowest yield hectare^{-1} was observed for 291RGI (1937.33 kg), which was significantly lower than all parents except TS-18 (1983.17 kg) and TS-228 (2068.83 kg), followed by R-25 (2191.67 kg) and TS-335 (2050.00 kg). Highest yield hectare^{-1} was observed for TS-17 (3628.00 kg), which was significantly higher than all parents followed by TR-6023 (2621.17 kg) and TR-9 (2572.83 kg). Mean values of yield hectare^{-1} for F_1 hybrids ranged from 2651.00 to 5587.17 kg representing a net difference of 2936.17 kg. The lowest yield hectare^{-1} was observed for TS-228 x R-25 (2651.00 kg), which was significantly lower than all F_1 hybrids except TS-228 x TR-6023 (2703.17 kg) followed by TS-335 x TR-6023 (2866.67 kg) and TS-335 x R-25 (2937.00 kg) while the highest yield hectare^{-1} was recorded for TS-18 x 291RGI (5587.17 kg), which was significantly higher than all F_1 hybrids except TS-18 x TR-6023 (5509.00 kg) followed by TS-18 x R-25 (5241.50) and TS-18 x TR-9 (5111.67 kg) (Table-I).

For yield hectare^{-1} positive values of heterosis and heterobeltiosis are desirable. Analysis of variance revealed highly significant differences among the genotypes for yield hectare^{-1}. All the F_1 hybrids showed highly significant heterotic effect whereas heterobeltiotic effect was also highly significant except for TS-228 x TR-6023. All the F_1 hybrids expressed positive heterotic effect and ranged from 5.60 to 185.02%. Maximum positive heterosis was observed for TS-18 x 291RGI (185.02%) followed by TS-18 x R-25 (151.10%). Heterobeltiotic effect was positive for all F_1 hybrids except TS-17 x TR-6023 (-9.06%) and ranged from 3.13 to 181.73%. Maximum positive heterobeltiotic effect which observed for TS-18 x 291RGI (181.73%) followed by TS-18 x R-25 (139.16%). Among the F_1 hybrids, TS-18 x 291RGI manifested highest positive heterosis percentage from mid and high parent indicating that among the hybrids, it outperformed other hybrids in yield hectare^{-1}. The present study is supported by the work of Singh *et al.* (2002) and Goksoy (1999), who have reported 278.0% and 15.9 to 178.1% heterosis, respectively for this character. Kumar *et al.* (1999), Yilmaz and Emiroglu (1995) and Yenice and Arslan (1997) have reported heterosis of –24.75 to 40.36% and 65.7 to 77.90% respectively. The work of Cheres *et al.* (2000), Kandhola *et al.* (1995), Sugoor *et al.* (1994), Gangappa *et al.* (1997), Madrap *et al* (1994) also support the findings of the current study.

Harvest Index

Genetic differences for harvest index were highly significant (P<0.01) among parents and hybrids (Table-I). The minimum percentage of harvest index was observed for 291RGI (7.14%), which was significantly lower than all parents except TS-228 (9.77%), while maximum percentage of harvest index was observed for TS-18 (37.76%). Among the F_1 hybrids, the harvest index ranged from 14.84 to 44.11%, representing a net difference of 29.27%. Minimum heterosis was observed for TS-17 x 291RGI (14.84%), which was significantly lower than all F_1 hybrids except TS-17 x TR-6023 (15.22%) and TS-335 x TR-6023 (18.18%). The maximum harvest index was observed for TS-18 x R-25 (44.11%), which was significantly higher than all F_1 hybrids (Table-I). All the F_1 hybrids expressed highly significant heterosis for harvest index except TS-17 x 291RGI, TS-17 x TR-9 and TS-18 x 291RGI whereas heterobeltiotic effect was highly significant for harvest index except TS-17 x R-25, TS-17 x TR-9, TS-228 x TR-6023 and TS-335 x TR-6023. The F_1 hybrids had an increase of 5.42% of harvest index from the mean values of parents.

Plants with maximum biological yield may not necessarily bear maximum economic yield, therefore, breeders are interested in maximum harvest index for sunflower hybrids in relation to lower biological yield. For harvest index positive values of heterosis and heterobeltiosis are desirable. The difference among the mean values for harvest index among the parents and F_1 hybrids were highly significant. Thirteen F_1 hybrids expressed highly significant heterosis for harvest index whereas twelve F_1 hybrids showed heterobeltiotic effect as highly significant for harvest index. Twelve F_1 hybrids produced positive heterotic effect and ranged from –1.72 to 171.66%. Maximum positive heterosis was produced by TS-335 x 291RGI (171.66%). Heterobeltiotic effect was positive for ten F_1 hybrids and ranged from –0.48 to 127.36%. Maximum positive heterobeltiotic magnitude was observed for TS-335 x 291RGI (127.36%). Among the F_1 hybrids TS-335 x 291RGI outperformed other hybrids by expressing maximum positive value from the mid and high parents indicating that harvest index was increased considerably by this F_1 hybrid. The finding in the present study is supported by the work of Khan *et al.* (2004) who found that TS-4 x TR-11 manifested maximum positive heterosis for harvest index (128.6%). Andrei (2003) and Madrap *et al.* (1994) also found the similar results of heterosis for harvest index.

Moisture Factor

Differences among parents and hybrids for moisture factor were highly significant (P<0.01) (Table-II). Minimum moisture factor was observed for TR-9 and TR-6023 (0.64) which was significantly lower than all parents except TS-17 and TS-18 (0.67), followed by TS-228 and TS-335 (0.73), while maximum moisture factor was observed for 291RGI (0.93), which was significantly higher than all parents followed by R-25 (0.75) and TS-228 and TS-335 (0.73). Among the F_1 hybrids, moisture factor ranged from 0.50 to 0.78 showing a net difference of 0.28. Minimum moisture factor among F_1 hybrids was observed for TS-17 x TR-6023 (0.50), which was significantly lower than all F_1 hybrids except TS-335 x TR-9 (0.51) and TS-228 x 291RGI and TS-18 x 291RGI (0.58) followed by TS-18 x TR-6023 (0.60) and TS-18 x TR-9 (0.62). Maximum moisture factor was observed for TS-228 x R-25 (0.78), which was significantly higher than all F_1 hybrids except TS-228 x TR-6023 (0.76) and TS-228 x TR-9 (0.73) followed by TS-335 x TR-6023 (0.70) as shown in Table-II.

For moisture factor negative effects of heterosis and heterobeltiosis is desirable. Sufficient genetic variability existed among the sunflower genotypes for moisture factor. Heterotic effect of moisture factor was significant for eleven crosses whereas heterobeltiotic effect was also significant for ten F_1 hybrids. Heterosis was negative for eleven F_1 hybrids and ranged from –30.35 to 11.19%. Maximum negative heterosis was observed for TS-228 x 291RGI (-30.35%), followed by TS-18 x 291RGI (-27.83%). Heterobeltiosis ranged from –20.30 to 19.13%. Maximum negative heterobeltiosis was observed for TS-228 x 291RGI (-20.30%). Among the F_1 hybrids, TS-228 x 291RGI decreased moisture factor from the mid and high parent in comparison to other F_1 hybrids.

Oil Content

Variation for oil content was highly significant (P<0.01) among parents and their F_1 hybrids (Table-II). Among the parents 291RGI showed minimum oil content (20.75%) which was significantly lower than all parents followed by TR-9 (22.99%) and TS-228 (23.35%). Maximum oil content was observed for TS-17 (33.47%), which was significantly higher than all parents followed by TS-18 (32.19%), R-25 (26.82%), TR-6023 (26%) and TS-335 (24%). Among the F_1 hybrids, oil content ranged from 27.75 to 34.20% representing a net difference of 6.45%. Hybrid TS-17 x 291RGI (27.75%) expressed minimum oil content while maximum oil content was observed for TS-335 x 291RGI and TS-335 x TR-6023 (34.20%), which were significantly higher than all F_1 hybrids (Table-II).

Oil percentage of whole sunflower seeds depends on both the percentage of hull and the percentage of oil in the kernel. Hull percentage among genotypes may vary from 10 to 60%, while oil percentage in kernel from 26 to 72%. Hull percentage of seeds and oil percentage in the kernels of present high oil cultivars or hybrids is in the range of 20 to 25% and 57 to 67%, respectively. Oil percentage generally is considered to be quantitatively inherited. Sunflower hybrids with high oil content are the main target of sunflower breeders (Gundaev, 1971). For oil content positive values of heterosis and heterobeltiosis are desirable. The differences among the mean values for oil content were highly significant among the sunflower genotypes. Heterotic effect for all F_1 hybrids was highly significant except TS-17 x TR-6023 whereas heterobeltiotic effect was also highly significant except TS-17 x R-25. Heterosis was positive for all F_1 hybrids except TS-18 x R-25 and TS-18 x TR-9 and ranged from –4.78 to 52.85%. Maximum positive heterotic effect was expressed by TS-335 x 291RGI (52.85%). Heterobeltiotic magnitude for ten out of sixteen F_1 hybrids was observed as positive. Heterobeltiosis ranged from –18.39 to 42.50%. Maximum positive heterobeltiotic effect was observed for TS-335 x 291RGI (42.50%). The F_1 hybrids TS-335 x 291RGI expressed maximum positive heterosis from the mid and better parent indicating that this cross has increased oil content than other F_1 hybrids. The present study is supported by the previous research findings of Sakthivel (2003) who concluded –16.70 to 26.82% heterobeltiosis for oil content and recommended CMS6A x GP270 as the best cross for oil content. Singh *et al.* (2002) found maximum heterobeltiosis 30.08% for oil content. Nehru *et al.* (2000) found positive heterobeltiosis for oil content and the hybrid BLC-5R-2-7-3 x HA234B was found to be good specific combiner while HA234B was good general combiner. Similarly Kumar *et al.* (1999) found –5.56 to 29.4% heterobeltiosis for oil content.

The present study was inspired keeping in view the edible oil needs along with the shortage of quality hybrid seed to cater the increasing gap between consumption and production in Pakistan. Consumption of vegetable edible oil is getting popularity with the passage of time while production is lingering behind. To bridge this gap, therefore, the availability of quality hybrid sunflower seed needs to be ensured. In this backdrop the present study was conducted, so that the available sunflower genotypes may properly be improved and subsequently exploited locally in heterosis breeding program.

Table-I: Mean values, mid-parent heterosis (MPH%) and high-parent heterosis (HPH%) for yield hectare^{-1} (YPH) and harvest index (HI) in sunflower genotypes during 2003-04.

GENOTYPE	YPH(Kg)	MPH(%)	HPH(%)	HI(%)	MPH(%)	HPH(%)
PARENTS						
TS-17	3628.83 F	-	-	19.22 GHIJ	-	-
TS-18	1983.17 M	-	-	37.76 B	-	-
TS-228	2068.83 LM	-	-	9.77 OP	-	-
TS-335	2250.00 K	-	-	10.59 NO	-	-
291RGI	1937.33 M	-	-	7.14 P	-	-
R-25	2191.67 KL	-	-	13.26 MN	-	-
TR-9	2572.83 J	-	-	18.63 HIJ	-	-
TR-6023	2621.17 J	-	-	17.49 JKL	-	-
Mean	**2406.73**	-	-	**16.73**	-	-
HYBRIDS						
TS-17×291RGI	4567.17 D	64.1**	25.86**	14.84 LM	12.6	-22.78**
TS-17×R-25	4213.33 E	44.78**	16.11**	19.12 GHIJ	17.79*	-0.48
TS-17×TR-9	4637.67 CD	49.56**	27.80**	19.73 FGHIJ	4.26	2.67
TS-17×TR-6023	3300.00 G	5.60**	-9.06**	15.22 KLM	-17.04*	-20.78**
TS-18×291RGI	5587.17 A	185.02**	181.73**	22.07 DEFG	-1.72	-41.57**
TS-18×R-25	5241.50 B	151.10**	139.16**	44.11 A	72.92**	16.81**
TS-18×TR-9	5111.67 B	124.39**	98.68**	24.17 D	-14.30**	-36.01**
TS-18×TR-6023	5509.00 A	139.30**	110.17**	21.18 DEFGHI	-23.33**	-43.91**
TS-228×291RGI	4765.50 C	137.91**	130.35**	20.69 EFGHIJ	144.79**	111.83**
TS-228×R-25	2651.00 J	24.45**	20.96**	28.67 C	149.06**	116.30**
TS-228×TR-9	3658.33 F	57.63**	42.19**	21.82 DEFGH	53.70**	17.14*
TS-228×TR-6023	2703.17 IJ	15.27**	3.13	19.00 GHIJ	39.44**	8.67
TS-335×291RGI	4533.33 D	116.53**	101.48**	24.08 DE	171.66**	127.36**
TS-335×R-25	2937.00 H	32.25**	30.53**	18.49 HIJK	55.03**	39.45**
TS-335×TR-9	4700.00 CD	94.91**	82.68**	23.10 DEF	58.08**	23.97**
TS-335×TR-6023	2866.67 HI	17.70**	9.37**	18.18 IJKL	29.52**	3.99
Mean	**4186.41**			**22.15**		
LSD%	**170.568**			**3.393**		

*, ** MPH and HPH effects significant at 5 and 1% probability levels, respectively.
Means in a column sharing same letter(s) are not significantly different at 5% probability level.

Table-II: Mean values, mid-parent heterosis (MPH%) and high-parent heterosis (HPH%) for moisture factor (MF) and oil content (OC) in sunflower genotypes during 2003-04.

GENOTYPE	MF	MPH(%)	HPH(%)	OC(%)	MPH(%)	HPH(%)
PARENTS						
TS-TS-17	0.67 DEFG	-	-	33.47 B	-	-
TS-TS-18	0.67 DEFG	-	-	32.19 E	-	-
TS-TS-228	0.73 BCDE	-	-	23.35 Q	-	-
TS-TS-335	0.73 BCDE	-	-	24.00 P	-	-
291RGI	0.93 A	-	-	20.75 S	-	-
R-R-25	0.75 BCD	-	-	26.82 M	-	-
TR-TR-9	0.64 FGH	-	-	22.99 R	-	-
TR-TR-6023	0.64 FGH	-	-	26.00 O	-	-
Mean	**0.72**	-	-	**26.20**	-	-
HYBRIDS						
TS-17×291RGI	0.64 FGH	-20.5**	-5.16	27.75 L	2.4**	-17.09**
TS-17×R-R-25	0.66 EFGH	-7.74*	-2.46	33.43 BC	10.90**	-0.12
TS-17×TR-TR-9	0.67 EFG	2.11	5.11	30.87 G	9.35**	-7.77**
TS-17×TR-6023	0.50 J	-23.71**	-21.73**	29.80 I	0.22	-10.97**
TS-18×291RGI	0.58 HIJ	-27.83**	-13.82**	32.36 D	22.25**	0.53**
TS-18×R-R-25	0.55 IJ	-22.36**	-17.88**	28.12 K	-4.69**	-12.64**
TS-18×TR-TR-9	0.62 FGHI	-5.90	-3.17	26.27 N	-4.78**	-18.39**
TS-18×TR-6023	0.60 GHI	-9.00*	-6.67	33.32 C	14.52**	3.51**
TS-228×291RGI	0.58 HIJ	-30.35**	-20.71**	30.93 G	40.27**	32.46**
TS-228×R-25	0.78 B	5.80	7.16	31.88 F	27.09**	18.87**
TS-228×TR-9	0.73 BCDE	6.63	14.64**	30.39 H	31.16**	30.15**
TS-228×TR-6023	0.76 BC	11.19**	19.13**	33.30 C	34.95**	28.08**
TS-335×291RGI	0.64 FGH	-23.50**	-12.92**	34.20 A	52.85**	42.50**
TS-335×R-25	0.65 EFGH	-11.96**	-10.83**	29.50 J	16.10**	9.99**
TS-335×TR-9	0.51 J	-25.87**	-20.30**	33.40 BC	42.16**	39.17**
TS-335×TR-6023	0.70 CDEF	1.51	8.75*	34.20 A	36.80**	31.54**
Mean	**0.63**			**31.23**		
LSD%	**0.081**			**0.131**		

*, ** MPH and HPH effects significant at 5 and 1% probability levels, respectively.
Means in a column sharing same letter(s) are not significantly different at 5% probability level.

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