Manish Sharma*, Shadakshari Yediyur Gavisiddaiah, Annabathula Mohan Rao and Sampangi Ramesh

Utilization of wild species for diversifying the cytoplasmic male sterility source of sunflower (*Helianthus annuus* L.) hybrids

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Abstract: The present investigation is directed at improving sunflower using hybrids having diverse cytoplasmic male sterile (CMS) sources from *Helianthus annuus* and *H. argophyllus*. The aim is to develop and identify promising lines and hybrids with a high combining ability and to obtain highly productive oilseed sunflower hybrids having diverse CMS sources. Five diversified CMS lines and 10 testers were crossed in a line × tester mating design to develop 50 F_1 hybrids for genetic analysis. The pooled analysis of variance revealed significant differences among hybrids for the traits studied. The mean squares of lines and testers from crosses and GCA variance components revealed the prevalence of additive variances and additive gene action. The mean squares of lines × tester interactions were also significant for all the traits considered. The significance of lines × tester interactions and SCA variance components indicated that SCA is also important in the expression of traits and demonstrated the role of dominant and epistatic genes in controlling the various traits. Among the diverse CMS lines, ARG-6-3-1-4was identified as the best general combiner for stem diameter, volume weight, seed yield, hull content and oil content. While, the line ARG-2-1-2 was the best general combiner for days to 50 per cent flowering, head diameter, 100 seed weight and seed filling per cent. The tester M17-R was observed to be the best general combiner for earliness and volume weight, while RHA 93 was the best general combiner for plant height, seed yield and oil content. The crossMUT-2-8-3-2 × GKVK 3 was found to be a good specific combiner for stem diameter, 100 seed weight, seed yield, seed filling percentage and oil content while, ARG-6-3-1-4 × GKVK 3 was a good specific

***Corresponding author: Manish Sharma**, Department of Genetics and Plant Breeding, UAS, Bangalore, India, E-mail: manisharmagpb@sdau.edu.in. https://orcid.org/0000-0002-2706-8641
Shadakshari Yediyur Gavisiddaiah, UAS, Bangalore, India
Annabathula Mohan Rao and Sampangi Ramesh, Department of Genetics and Plant Breeding, UAS, Bangalore, India

combiner for days to 50% flowering, plant height and 100 seed weight. The highest standard heterosis for seed yield was observed in the hybrid MUT-2-8-3-2 × GKVK 3 followed by ARG-2-1-2 × LTRR 822 while for oil content the highest was seen in the cross ARG-6-3-1-4 × RHA95-C-1 and thus these hybrids can be exploited for sunflower improvement and diversifying the cytoplasmic male sterile sources of this valuable oilseed crop.

Keywords: combining ability; cytoplasmic male sterility; hybrids; sunflower.

Introduction

Sunflower (*Helianthus annuus* L.) is an oilseed crop used for an edible purposes and other industrial use. The increase in sunflower production and seed quality has been largely connected to the inclusion of wild *Helianthus* species into the improvement programs. Using this approach, the diversity in sunflower was enriched and the possibility of heterosis breeding was created through the identification of CMS – restorers of the fertility genetic system (Christov 2008). In India, the sunflower was introduced in 1969 from Russia because of its distinct advantages, *viz*., photo insensitivity, short duration, high seed multiplication ratio, high seed yield and better quality of the oil. However, commercial cultivation of sunflower in India started in 1972 with the introduction of Russian varieties. A major event in sunflower history was the discovery of cytoplasmic male sterility (CMS) in a wild prairie sunflower, *Helianthus petiolaris* Nutt. (Leclercq 1969), and subsequent identification of genes for fertility restoration by Kinman (1970); Enns et al. (1970), Leclercq (1971) and Vranceanu and Stoenescu (1971) that led to the production of commercial hybrids. Sunflower hybrids are preferred over open-pollinated varieties since hybrids offer several benefits in terms of growth, development, synchronous flowering, early maturity, higher seed set, increased productivity, and resistance to major foliar diseases and response to higher levels of chemical fertilizer application. In India, the first-ever CMS based sunflower hybrid BSH-1 was released from the University of Agricultural Sciences, Bangalore (Seetharam et al. 1980) which provided the required impetus to expand sunflower cultivation in the country. Since then, many hybrids have been released for commercial cultivation by utilizing the cytoplasmic genetic male sterility system. The area under sunflower cultivation in India was 2.5 lakh hectares with a production of 2.2 lakh tonnes and productivity of 0.9 t/ha (Anonymous 2019). Over 70% of the sunflower crop is being grown across the states of Karnataka, Maharashtra and Andhra Pradesh. In Karnataka, it occupies an area of about 1.60 lakh ha. with a

production of 1.20 lakh tonnes and a productivity of 0.75 t/ha. Karnataka, popularly known as the "Sunflower State" is the leading producer of oilseed sunflower, accounting for 63% of the total acreage and 53% of the national production.

Much of the current germplasm used in sunflower breeding programs originated from limited genetic resources, resulting in a crop with an extremely narrow genetic base. At present, only one CMS source (i.e., PET 1) is being widely used for the sunflower hybrid breeding program (Seiler et al. 2017) which poses a potential risk to the narrow genetic base for hybrid sunflower production. Prevalence of genetic uniformity of this kind over a large area could result in the genetic vulnerability of hybrids to a new strain of disease or pest similar to that happened in maize when 'Texas' cytoplasm become susceptible to *Helminthosporium maydis* in the USA (Tatum 1971). Among several strategies available to overcome this problem, diversification of CMS sources is possibly the most economic and effective method. The utilization of different cytoplasmic backgrounds in hybrid development will improve the general variability of sunflower hybrids and enhances their tolerance to diseases and pests.

Seventy-two CMS sources, 38 from wild *H. annuus*, 24 from other wild annual species, and only 10 from perennial species, have been identified in progenies of crosses between wild *Helianthus* populations and cultivated lines, or from induced mutation (Serieys 2002; Serieys and Christov 2005). Utilization of these CMS sources requires the development of lines having different cytoplasms in a common nuclear genetic background (isonuclear alloplasmic lines) to understand the impact of alien cytoplasm on seed yield and its related traits. Even in hybrids differing in their CMS source a thorough understanding of the interaction between the alien cytoplasm and nuclear genes from commercially cultivated sources and the impact of this interaction on heterosis for yield-related traits is required for utilizing these alloplasmic lines in hybrid development. Jan et al. (2014), developed and compared 20 diverse alloplasmic cytoplasmic substitution lines from annual and perennial wild species for agronomic traits with the inbred line HA89 over four environments. Lines having annual species cytoplasm did not affect agronomic traits compared with the currently used PET1 cytoplasm which meant that most cytoplasm of wild annual *Helianthus* species can accommodate cultivated nuclear genes without significant adverse interactions and are potential sources of cytoplasmic diversity for sunflower breeding. Considering this, the current investigation aims to demonstrate that by transferring a CMS source from wild *Helianthus* species into cultivated sunflower, new sunflower lines and hybrids can be developed, which have the good combining ability, high production potential and high oil content that are suitable for growing in Karnataka and other sunflower producing regions worldwide.

Materials and methods

The present investigations were undertaken at the Zonal Agricultural Research Station, University of Agricultural Sciences, Gandhi Krishi Vigyana Kendra, Bangalore, India. The research station is geographically situated at 12°58′ N latitude and 77°35′ N longitude at an altitude of 930 m above mean sea level. Materials for the study consisted of five male sterile lines, ARG-2-1-2, MUT-2-8-3-2, E002, ARG 3 and ARG-6-3-1-4 (Supplementary Figures 1–5) derived from *H. argophyllus* and *H. annuus* developed from diverse cytoplasmic male sterile sources in the different nuclear genetic background (Table 1a). Diverse CMS lines were obtained from the Directorate of Oilseeds Research (DOR), Hyderabad and the Department of Oilseeds, TNAU, Coimbatore. These CMS lines were developed by crossing with the maintainer line (*H. annuus* genetic background), followed by repeated backcrossing to the maintainer parent (Figure 1). Phenotypic uniformity concerning morphological characters within these CMS lines was obtained in BC_6 progenies which were then selfed for one generation to make the lines stable. Characterization of these lines is presented in Table 1b. Ten restorers/testers lines, GKVK-3, RHA 6D-1, RHA 95-C-1, LTRR 822, M-17R, MR-1, RHA-272-II, X-15-NB-10, GKVK-2 and RHA-93 were used to study combining ability. These testers were selected based on their ability to restore fertility in these diversified CMS sources (Nandini et al. 2017). To estimate economic heterosis three standard checks *viz.*, KBSH-44 (National check), KBSH-53 and KBSH-78 (Local checks) were used in the experiment.

During *Rabi*, 2015–16, all the diverse CMS lines and 10 restorer lines were sown in the field to cross in a Line × Tester mating design (Kempthorne 1957). Staggered sowing of all inbred lines was carried out three times at an interval of two days to ensure flowering synchronization with diverse CMS lines. To prevent undesirable pollination, heads of CMS lines were covered with cloth bags a day before the opening of the first ray florets. Similarly, the heads of inbred lines were also covered with cloth bags to collect pollen. Pollen from the inbred lines was collected in Petri plates and applied to the flowers of female lines using camel hairbrushes during morning hours. The pollination was repeated for five to six days in each combination to ensure sufficient seed set and simultaneously, all inbreds were sib pollinated. At maturity, the crossed seeds of 50 combinations were collected for future evaluation. Hybrids and checks were evaluated in *Kharif* 2016 and *Rabi/Summer* 2016–17 with two replications each and a randomized block design. Each genotype was sown in a single row of 3-m length with a spacing of 60 and 30 cm between plants and within row respectively. All the recommended agronomic practices were followed for raising a successful crop under irrigated conditions. The data during germination till maturity were obtained from F_1 plants and their parents for days to 50% flowering, plant height (cm), head diameter (cm), stem diameter

Table 1a: Diversified CMS lines.

Sl. no	CMS designation	Cytosterility source	Nuclear Genetic background
1	ARG-2-1-2	*H. argophyllus*	234 B
2	MUT-2-8-3-2	*H. annuus*	234 B
3	E002	*H. annuus*	DS2 B
4	ARG 3	*H. argophyllus*	DS2 B
5	ARG-6-3-1-4	*H. argophyllus*	REC 428 B

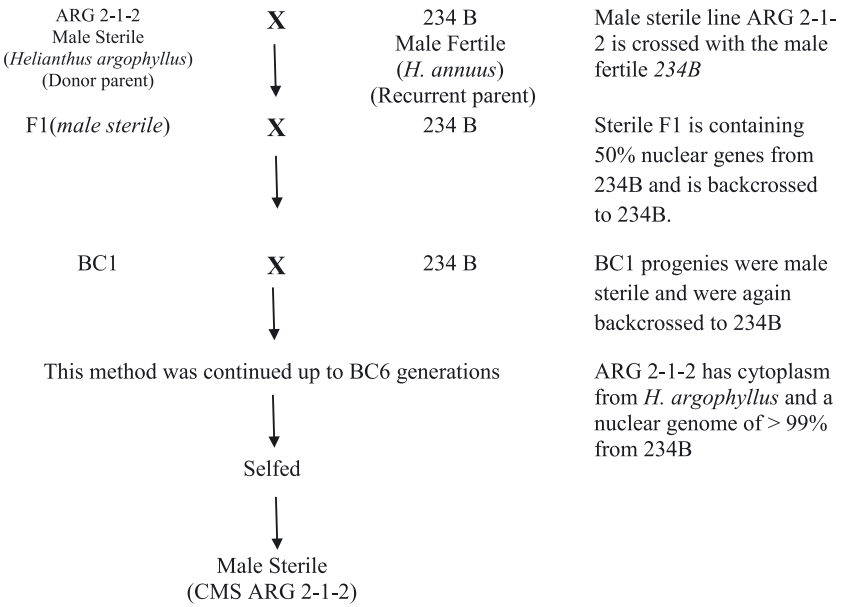

Figure 1: Transfer of male-sterile cytoplasm into the nuclear genome of cultivated 234B. This procedure was followed for the development of all the five CMS lines separately.

Table 1b: Characterization of diversified CMS lines.

S. no.	Characteristics	ARG-2-1-2	MUT2-8-3-2	E002	ARG-3	ARG-6-3-1-4
1	Leaf: Size	15.5 cm (medium)	19.5 cm (medium)	20.3 cm (medium)	21.3 cm (medium)	19.8 cm (medium)
2	Leaf: Shape	Cordate	Cordate	Cordate	Cordate	Cordate
3	Leaf: Colour	Med. Green	Dark green	Med. Green	Med. Green	Dark green
4	Leaf: Blistering	Medium	Medium	Medium	Medium	Strong
5	Leaf: Serration	Medium	Medium to coarse	Medium	Medium	Medium
6	Leaf: Angle of lateral veins	Acute	Obtuse	Acute	Acute	Acute
7	Lea: Orientation of blade	Erect	Erect	Erect	Erect	Erect
8	Leaf: Petiole anthocyanin pigmentation	Absent	Absent	Absent	Absent	Absent
9	Stem: Pigmentation	Absent	Absent	Absent	Absent	Absent
10	Ray floret: Number	34 (medium)	44 (many)	34 (medium)	41 (many)	37 (medium)

Table 1b: (continued)

S. no.	Characteristics	ARG-2-1-2	MUT2-8-3-2	E002	ARG-3	ARG-6-3-1-4
11	Ray floret: Shape	Elongated	Elongated	Elongated	Elongated	Elongated
12	Ray floret: Colour	Yellow	Yellow	Yellow	Yellow	Yellow
13	Disk floret: Colour	Yellow	Yellow	Yellow	Yellow	Yellow
14	Disk floret: Anthocyanin pigmentation of stigma	Absent	Absent	Absent	Absent	Absent
15	Bract: Shape	Elongated	Elongated	Elongated	Elongated	Elongated
16	Bract: Anthocyanin pigmentation	Absent	Absent	Absent	Absent	Absent
17	Head: Attitude	Vertical	Vertical	Vertical	Vertical	Vertical
18	Head: Diameter	11.5	13.2	14.2	14.1	14.5
19	Head: Shape of the grain side	Convex	Convex	Convex	Convex	Convex
20	Plant: Branching	Non-branch	Non-branch	Non-branch	Non-branch	Non-branch
21	Plant: Height	115.5 (short)	79.8 (very short)	120.5 (medium)	108.3 (short)	154.2 (tall)
22	Seed weight (100 seed weight in grams)	5.07 (medium)	4.56 (medium)	5.15 (medium)	4.98 (medium)	4.21 (medium)
23	Seed colour	Black	Black	Black	Black	Black

(cm), days to maturity, 100-seed weight (g), volume weight (g 100 mL^{-1}), seed yield (kg ha^{-1}), hull content (%), seed filling percentage and oil content (%). The seed oil content was determined by nuclear magnetic resonance spectrometry (NMR). The data were analyzed to determine the differences among genotypes, parents cross, parents versus crosses according to Gomez and Gomez (1984), while mean squares for GCA were determined from lines and testers and specific combing ability from lines × tester interactions according to statistical procedures developed by Kempthorne (1957) and adopted by Singh and Choudhary (1984). Heterosis over standard checks (SC) was computed by the method suggested by Turner (1953) and Hayes et al. (1955).

Results and discussion

Pooled analysis of variance

The presence of genetic variability is the basic requirement for developing high yielding hybrids in sunflower breeding programs. Pooled analysis of variance for the experimental design shown in Table 2 indicated significant differences among genotypes, parents and crosses for all the studied traits confirming that the data is

valid for genetic analysis. The pooled analysis of variance for combining ability due to different sources for all the characters (Table 3) revealed that the mean squares of lines and testers from crosses to determine the GCA were significant for most of the traits and mean squares of lines × tester interactions were significant for all the traits. The significance of lines × tester interactions indicated that SCA is also important in the expression of traits and demonstrated the value of non-additive variances and dominant genes controlling the traits. Memon et al. (2015), Lakshman et al. (2019) and Hilli et al. (2020) also observed that both additive and non-additive genetic variations were equally important for yield and its contributing traits in sunflower.

General combining ability effects

Combining the ability of a line/strain to produce superior progenies upon hybridization with other lines/strains is an important criterion to select parents for developing superior new hybrids (Sprague and Tatum 1942). To reduce the crop growth period, fewer days to flowering and maturity are preferred. The sunflower growers prefer short duration hybrids because they reduce the incidence of insect-pest, disease attacks and adverse environmental effects (Memon et al. 2015). For days to 50% flowering and days to maturity only line E002 exhibited a significant negative GCA effect (Table 4) for both these traits, while among testers the highest significant negative GCA effect was recorded for M17-R (Table 5) for both the traits followed by GKVK 2 and RHA-272-II. Thus, these lines E002 and testers M17-R, GKVK 2 and RHA-272-II were found to be good general combiners for earliness. Therefore, these lines could be used in the development of early maturing hybrids. Meena et al. (2013) and Azad et al. (2016) have also identified good general combiners for early flowering.

Since reduced plant height promotes resistance to lodging, there is also considerable interest in the development of semi-dwarf hybrids. The most prominent negative effect of GCA on plant height was found in the CMS lines ARG 3 (–11.390) and testers in RHA-93 (–15.748), hence, the lines and testers with negative GCA effects can be used in hybridization programs to develop medium stature plants. For head diameter, line ARG-2-1-2 (0.270) exhibited significant positive GCA effects while line ARG 3 (–0.272) exhibited significant negative GCA effects while among testers, only RHA-95-C-1 (0.545) and RHA 272-II (0.235) exhibited significant positive GCA effect. Riaz et al. (2017) have also reported similar results and inferred that these identified lines and testers with a positive GCA effect could be used in a further breeding programme to synthesise hybrids with large head size, in turn contributing to increased yield. For stem diameter

Table 2: A pooled analysis of variance for seed yield and component traits in sunflower.

Source of Variations	d.f	Days to 50% flowering	Plant height (cm)	Head diameter (cm)	Stem diameter (cm)	Days to Maturity	100 seed weight (g)	Volume weight (g/100 mL)	Seed yield (kg/ha)	Hull content (%)	Seed filling per cent	Oil content (%)
Replicates	1	0.65	87.08	2.94	0.05	3.02	0.05	2.66	171,553.20	20.28	9.89	15.15**
Season	1	973.11**	6509.30**	18.72**	0.07**	1024.06**	16.18**	17.86*	100,0458.85**	138.82**	0.0002	46.07**
Genotypes	64	29.22**	1411.20**	7.97**	0.15**	40.95**	1.64**	27.02**	1,321,399.76**	34.35**	37.25**	42.01**
Crosses	49	22.74**	558.12**	1.05**	0.05**	34.67**	0.69**	29.43**	147,303.15**	30.69**	37.99**	42.65**
Parents vs. crosses	1	21.34**	25165.55**	204.28**	3.74**	4.02	25.37**	35.51**	54,769,267.49**	316.93**	190.94**	469.24
Error	128	1.08	47.47	0.21	0.01	1.70	0.08	3.29	36,192.46	0.92	1.77	0.22

**significant at $p \leq 0.01$. *significant at $p \leq 0.05$.

Table 3: Pooled Analysis of variance for combining ability for seed yield and component traits in sunflower.

Source of Variations	d.f	Days to 50% flowering	Plant height (cm)	Head diameter (cm)	Stem diameter (cm)	Days to maturity	100 seed weight (g)	Volume weight (g/100 mL))	Seed yield (kg/ha)	Hull content (%)	Seed filling per cent	Oil content (%)
Replicates (R)	1	1.280	0.551	1.84	0.03	1.45	0.048	10.37	289,205.52	29.28**	28.74	17.81
Season (S)	1	1003.52**	12987.88**	1.51**	0.002	804.01**	12.370**	4.14	9,889,505.58**	140.47**	0.05	51.69**
Crosses (C)	49	22.74**	558.12**	1.05**	0.05**	34.67**	0.686**	29.43**	147,303.15**	30.69**	37.99**	42.65**
Lines (L)	4	80.04**	2723.99**	1.67	0.03	111.79**	2.301**	101.65**	341,161.03*	103.75**	131.40**	430.24**
Tester (T)	9	69.62**	1088.27**	1.61	0.10*	89.41**	0.790	88.19**	252,230.83*	32.48	61.86*	10.73
Lines × tester	36	4.66**	184.93**	0.84**	0.04**	12.42**	0.481**	6.71**	99,531.47**	22.14**	21.65**	7.57**
S × C	49	2.64*	132.63**	0.42**	0.003	5.64**	0.043	1.42	24,376.23	1.14	1.31	0.18
S × L	4	3.66	124.19	0.29	0.002	1.22	0.02	11.84**	40,277.69	0.23	1.45	0.07
S × T	9	4.63*	168.23	0.68	0.001	9.03	0.07	0.51	42,703.38*	1.01	0.85	0.10
S × LxT	36	2.03*	124.67**	0.37*	0.001	5.28**	0.03	0.48	18,027.61	1.27	1.40	0.21
Error	98	1.15	41.21	0.20	0.007	2.04	0.083	3.21	44,015.29	0.88	1.32	0.17

**significant at $p \leq 0.01$. *significant at $p \leq 0.05$.

Table 4: Estimates of general combining ability effects of lines for seed yield and component traits in sunflower.

Lines	Days to 50% flowering	Plant height (cm)	Head diameter (cm)	Stem diameter (cm)	Days to maturity	100 seed weight (g)	Volume weight (g/100 mL)	Seed yield (kg/ha)	Hull content (%)	Seed filling per cent	Oil content (%)
ARG-2-1-2	−1.670**	4.088**	0.270**	0.0001	2.580**	0.363**	0.473	93.574**	−0.969**	1.686**	2.571**
MUT-2-8-3-2	0.155	4.430**	−0.075	−0.005	0.670**	0.046	1.019**	−60.981	−0.190	0.523**	−3.967**
E002	−0.995**	−5.713**	−0.035	−0.008	−0.630**	−0.211**	−1.869**	−50.603	1.465**	0.377*	0.444**
ARG 3	0.530**	−11.390**	−0.272**	−0.029*	0.795**	−0.225**	−1.446**	−88.253**	1.745**	−3.101**	−2.688**
ARG-6-3-1-4	1.980**	8.585**	0.113	0.041**	1.745**	0.027	1.822**	106.263**	−2.051**	0.515**	3.640**
SE ±	0.170	1.015	0.071	0.013	0.226	0.046	0.283	33.172	0.148	0.182	0.065

**significant at $p \leq 0.01$. *significant at $p \leq 0.05$.

Table 5: Estimates of general combining ability effects of testers for seed yield and component traits in sunflower.

Testers	Days to 50% flowering	Plant height (cm)	Head diameter (cm)	Stem diameter (cm)	Days to maturity	100 seed weight (g)	Volume weight (g/100 mL)	Seed yield (kg/ha)	Hull content (%)	Seed filling per cent	Oil content (%)
GKVK-3	3.480**	−1.608	−0.100	0.042*	3.695**	0.232**	2.071**	88.047	−0.088	1.654**	−0.605**
RHA-6D-1	0.880**	1.233	0.135	−0.003	1.845**	−0.080	0.245	−89.587	−1.053**	1.047**	0.870**
RHA-95-C-1	1.730**	13.943**	0.545**	0.106**	2.095**	0.184**	0.703	72.991	−0.261	1.277**	0.942**
LTRR-822	1.730**	5.662**	0.055	0.054**	1.145**	0.021	−3.152**	13.824	−2.018**	−0.755**	0.088
M17-R	−2.120**	−3.093**	−0.410**	−0.074**	−2.805**	−0.440**	3.166**	−133.831**	−0.646**	−0.732**	0.210*
MR-1	−1.270**	0.663	0.125	0.052**	−0.855**	−0.117	0.145	105.547*	0.367	1.182**	0.279**
RHA-272-II	−1.470**	−1.077	0.235*	−0.002	−1.955**	0.198**	−0.505	−159.909**	2.121**	−3.848**	−1.152**
X-15-NB-10	−0.070	0.093	−0.070	0.028	−0.105	−0.092	−3.161**	−75.009	1.113**	1.187**	0.201*
GKVK-2	−1.520**	−0.068	−0.355**	−0.116**	−1.955**	0.023	1.745**	−0.842	1.451**	−1.760**	0.281**
RHA-93	−1.370**	−15.748**	−0.160	−0.086**	−1.105**	0.073	−1.256**	178.769**	−0.984**	0.748**	1.115**
SE±	0.240	1.435	0.101	0.019	0.319	0.064	0.401	46.912	0.210	0.257	0.092

**significant at $p \leq 0.01$. *significant at $p \leq 0.05$.

lines ARG-6-3-1-4 (0.041) exhibited the highest significant positive effect and ARG 3 (–0.029) exhibited a significant negative GCA effect. Testers RHA-95-C-1 (0.106) and LTRR 822 (0.054) followed by MR-1 (0.052) exhibited the highest significant positive GCA effect as also observed in the studies of Lakshman et al. (2019) indicating the preponderance of additive effects in the inheritance of this character.

The seed weight of a genotype serves as an indicator of the expression of an end product i.e., seed yield since it is an important character contributing to seed yield. Lines ARG-2-1-2 (0.363) exhibited the highest significant positive GCA effect while testers, GKVK 3 (0.232), RHA 272-II (0.198) and RHA-95-C-1 (0.184) had significant positive *GCA* effect indicating their high utility in the breeding programme. For volume weight lines ARG-6-3-1-4 (1.822) and MUT-2-8-3-2 (1.019) exhibited a significant positive *GCA* effect. Three testers showed a significant positive *GCA* effect, the highest in M17-R (3.166) followed by GKVK 3 (2.071) and GKVK-2 (1.745). Hence, these lines and testers showing positive GCA effects could be used in a hybridization programme to develop hybrids with high seed weight and volume weight. Patil et al. (2012) have also reported good general combiners for these yield contributing traits.

The *GCA* effects for seed yield varied both in magnitude and direction among both lines and testers. Lines ARG-6-3-1-4 (106.263) and ARG-2-1-2 (93.574) expressed significant positive *GCA* effects while ARG 3 (–88.253) exhibited a significant negative GCA effect. Among testers, GCA effects ranged from 178.769 (RHA 93) to –159.909 (RHA 272-II). Testers RHA 93 and MR-1 recorded positive *GCA* effects while two testers showed negative *GCA* effects. It is interesting to note that the line ARG-6-3-1-4 is a good general combiner for most of the yield contributing characters, showing that a positive association exists between seed yield and its attributes such as plant height, stem diameter, head diameter and volume weight. Hence, ARG-6-3-1-4 could also be used in breeding for the development of hybrids with higher seed yields. In earlier reports, Salem and Ali (2012), Memon et al. (2015) and Chahal et al. (2019) also reported good general combiners for seed yield. Patil et al. (2012) in their study observed significant negative GCA effects for hull content. In our results also lines ARG-6-3-1-4 (–2.051) and ARG-2-1-2 (–0.969) exhibited a significant negative *GCA* effect which is desirable. Four of the 10 testers had a significant negative GCA effect, the highest being observed for LTRR 822 (–2.018) followed by RHA 6D-1 (–1.053), RHA 93 (–0.984) and M17-R (–0.646). Parents showing negative GCA for this trait can be considered to develop hybrids having low hull content. For seed filling percentage lines ARG-2-1-2 (1.686), MUT-2-8-3-2 (0.523), ARG-6-3-1-4 (0.515) and E002 (0.377) had significant positive *GCA*. whereas, line ARG 3 (–3.101) exhibited a significant negative GCA effect. All the testers, recorded a significant *GCA* effect, with six being positive and four being

negative. The highest positives were GKVK 3 (1.654) followed by RHA-95-C-1 (1.277). Lakshman et al. (2019) reported similar results for seed filling percentage inferring that the lines and tester having positive significant GCA effects appeared to transfer the alleles with additive effects.

Since sunflower is an oilseed crop, oil is the ultimate end product, hence, increased oil content is of prime importance. All the lines tested expressed significant *GCA* effects, three positive and two negative. The line ARG-6-3-1-4 (3.640) manifested the highest positive significant *GCA* effect followed by ARG-2-1-2 (2.571) and E002 (0.444). Seven testers had significant positive *GCA* effects. The testers RHA-93 (1.115) followed by RHA 95-C-1 (0.942) and RHA 6D-1 (0.870) were the best general combiners for oil content and thus would be desirable parents to be used for developing sunflower hybrids with high oil content. Similar findings for oil content were reported by Azad et al. (2016) and Attia et al. (2020).

Specific combining ability effects

The relative performance of any cross combination is expressed as a specific combining ability and is denoted in terms of *SCA* effects and SCA variance. The SCA variance denotes the non-additive or dominance portion of variance and is generally non-fixable on selfing but can be exploited in a hybrid combination. Out of 50 crosses, only five hybrids recorded the desirable significant negative *SCA* effects and three hybrids exhibited significant positive *SCA* effects for days to 50% flowering (Table 6). The crosses which exhibited the highest significant negative *SCA* effects for earliness are MUT-2-8-3-2 × M-17-R (–1.805) followed by ARG-6-3-1-4 × GKVK-3 (–1.730) and ARG-2-1-2 × GKVK-2 (–1.580). The parents of the best specific combinations, MUT-2-8-3-2 × M-17-R (Table 7) were of low × low general combiners indicating the involvement of non-additive gene action and over dominance in the expression of this trait. Concerning days to maturity, 17 out of 50 crosses manifested significant *SCA* effects, of which the highest negative *SCA* effect was manifested by ARG 3 × GKVK-2 (–2.545), followed by ARG 3 × LTRR-822 (–2.395) and ARG-2-1-2 × RHA 6D-1 (–2.220). Ghaffari et al. (2020) also obtained similar results and concluded that crosses showing significant negative SCA effects and variances possess dominant or over dominant types of genes with decreasing effects, hence, may be exploited for earliness in sunflower.

For plant height, the best crosses which exhibited a high negative *SCA* effect were ARG-6-3-1-4 × GKVK-3 (–15.125) followed by ARG-6-3-1-4 × RHA 6D-1 (–12.465) and ARG 3 × GKVK-2 (–10.265). Bhoite et al. (2018) also reported good specific combiners for plant height. Head diameter in the case of sunflower is an important yield attributing character since there is a positive correlation of head size with the

Table 6: Estimates of specific combing ability effects of crosses for seed yield and component traits in sunflower.

Hybrids	Days to 50% flowering	Plant height (cm)	Head diameter (cm)	Stem diameter (cm)	Days to maturity	100 seed weight (g)	Volume weight (g/100 mL)	Seed yield (kg/ha)	Hull content (%)	Seed filling per cent	Oil content (%)
ARG-2-1-2 × GKVK-3	−0.330	0.048	−0.335	−0.019	−0.570	−0.119	−0.020	−244.963*	0.491	−2.268**	−1.104**
ARG-2-1-2 × RHA 6D-1	−0.230	−0.793	0.530*	0.109*	−2.220**	0.535**	0.589	6.281	0.928	−0.087	0.681**
ARG-2-1-2 × 95-C-1	−0.330	−0.002	−0.030	−0.040	−1.970**	0.413**	−0.377	62.870	−0.784	1.233*	−1.073**
ARG-2-1-2 × LTRR-822	−0.080	−3.123	0.060	0.011	0.980	0.217	1.416	295.926**	−2.097**	3.556**	−0.457*
ARG-2-1-2 × M-17-R	1.770**	−10.118**	−0.475*	0.010	0.930	−0.220	−0.305	−180.030	1.424**	1.058	0.771**
ARG-2-1-2 × MR-1	0.420	3.178	−0.060	0.051	0.230	−0.350*	0.741	−145.797	0.924	−2.747**	1.047**
ARG-2-1-2 × RHA 272-II	0.620	−0.283	0.080	−0.015	2.580**	0.074	−0.504	136.325	1.335**	0.863	−1.429**
ARG-2-1-2 × X-15-NB-10	0.470	7.347*	−0.165	−0.012	1.480*	−0.633**	−1.746	−98.575	1.393*	−2.081**	0.155
ARG-2-1-2 × GKVK-2	−1.580**	5.183	0.220	−0.073	−0.670	−0.038	0.771	99.481	1.030*	−1.739**	−0.550**
ARG-2-1-2 × RHA-93	−0.730	−1.438	0.175	−0.018	−0.770	0.122	−0.563	68.481	−4.646**	2.213**	1.961**
MUT-2-8-3-2 × GKVK-3	−0.405	1.830	1.160**	0.141**	−0.820	0.395**	0.626	404.036**	−2.191**	1.932**	2.070**

Table 6: (continued)

Hybrids	Days to 50% flowering	Plant height (cm)	Head diameter (cm)	Stem diameter (cm)	Days to maturity	100 seed weight (g)	Volume weight (g/100 mL)	Seed yield (kg/ha)	Hull content (%)	Seed filling per cent	Oil content (%)
MUT-2-8-3-2 × RHA 6D-1	0.945	3.665	−0.025	−0.056	0.530	−0.453**	0.427	−205.830	1.754**	−1.804**	−1.771**
MUT-2-8-3-2 × 95-C-1	−0.905	−1.970	−0.585*	0.015	−1.220	0.268	−0.376	−0.352	0.0001	−2.994**	0.285
MUT-2-8-3-2 × LTRR-822	−0.905	−0.765	−0.045	−0.031	−2.020**	−0.041	2.049*	−130.075	−1.973**	0.704	−1.271**
MUT-2-8-3-2 × M-17-R	−1.805**	−7.210*	−0.405	−0.070	−0.570	−0.565**	−1.094	−258.808*	−2.120**	−4.747**	−0.128
MUT-2-8-3-2 × MR-1	−0.155	−8.965**	−0.390	−0.032	0.230	−0.076	2.157*	65.703	−0.023	0.746	−0.562**
MUT-2-8-3-2 × RHA 272-II	0.545	5.175	−0.200	0.017	0.580	0.241	0.637	65.881	3.398**	5.329**	0.312
MUT-2-8-3-2 × X-15-NB-10	−0.355	−4.945	0.380	−0.082	−0.520	0.456**	−1.847*	99.037	3.731**	−1.058	−0.732**
MUT-2-8-3-2 × GKVK-2	3.095**	7.265*	−0.135	0.027	2.830**	−0.463**	−1.723	−225.130*	−3.062**	0.714	0.579**
MUT-2-8-3-2 × RHA-93	−0.055	5.920	0.245	0.072	0.980	0.236	−0.857	185.537	0.485	1.176*	1.217**
E002 × GKVK-3	0.745	10.748**	0.070	0.031	0.230	−0.212	−0.485	−135.508	−1.006*	−1.744**	0.631**

Table 6: (continued)

Hybrids	Days to 50% flowering	Plant height (cm)	Head diameter (cm)	Stem diameter (cm)	Days to maturity	100 seed weight (g)	Volume weight (g/100 mL)	Seed yield (kg/ha)	Hull content (%)	Seed filling per cent	Oil content (%)
E002 × RHA 6D-1	−0.905	5.708	−0.240	0.037	−1.670*	−0.065	−1.437	3.403	−0.401	1.428*	0.326
E002 × 95-C-1	−0.255	1.447	0.200	0.048	−0.170	−0.020	0.328	14.270	1.410**	0.575	−2.151**
E002 × LTRR-822	0.495	−0.372	0.065	0.019	2.280**	−0.089	−2.098*	−72.397	0.062	−2.250**	1.218**
E002 × M-17-R	−0.405	1.658	0.305	−0.125**	−1.520*	0.300*	1.825*	254.425*	1.818**	1.370*	−1.579**
E002 × MR-1	0.495	−4.298	−0.030	−0.039	0.530	0.312*	−1.912*	−9.952	0.327	−0.035	−0.465*
E002 × RHA 272-II	−1.055	−5.383	−0.190	−0.135**	−0.870	−0.019	0.278	40.225	−1.262**	−4.392**	1.276**
E002 × X-15-NB-10	−0.205	−1.753	0.015	0.076	−0.220	−0.101	2.816**	−26.619	−2.401**	2.718**	2.520**
E002 × GKVK-2	0.745	−5.293	−0.075	0.055	1.130	0.264	−0.072	76.992	0.173	2.438**	−0.072
E002 × RHA-93	0.345	−2.463	−0.120	0.035	0.280	−0.371*	0.759	−144.841	1.280**	−0.110	−1.704**
ARG 3 × GKVK-3	1.720**	2.500	0.358	0.035	2.555**	−0.589**	−1.159	104.920	−0.943*	1.901**	0.080
ARG 3 × RHA 6D-1	0.320	3.885	0.348	0.138**	4.405***	0.098	−0.583	126.998	1.142*	1.403*	−0.433*
ARG 3 × 95-C-1	0.470	−6.375*	0.262	−0.021	1.155	−0.231	0.289	−74.469	1.998**	1.120	1.176**

Table 6: (continued)

Hybrids	Days to 50% flowering	Plant height (cm)	Head diameter (cm)	Stem diameter (cm)	Days to maturity	100 seed weight (g)	Volume weight (g/100 mL)	Seed yield (kg/ha)	Hull content (%)	Seed filling per cent	Oil content (%)
ARG 3 × LTRR-822	0.470	−0.820	−0.147	−0.050	−2.395**	−0.103	−1.127	−28.635	2.177**	0.400	−0.600**
ARG 3 × M-17-R	−0.180	4.985	0.442	0.136*	−0.445	0.228	0.246	104.909	2.466**	1.710**	1.052**
ARG 3 × MR-1	−1.280*	−0.420	−0.167	−0.038	−1.895**	−0.095	−0.604	−9.802	−1.647**	−0.480	−0.056
ARG 3 × RHA 272-II	−0.830	3.345	−0.027	0.061	−1.545*	−0.095	0.041	−87.402	−2.341**	−2.137**	1.047**
ARG 3 × X-15-NB-10	0.020	1.300	−0.223	0.002	0.355	0.220	1.932*	−13.969	−3.501**	−0.019	−0.361
ARG 3 × GKVK-2	−1.530**	−10.265**	−0.738**	−0.254**	−2.545**	0.210	0.391	−4.802	0.394	−1.052	0.622**
ARG 3 × RHA-93	0.820	1.865	−0.107	−0.009	0.355	0.355*	0.572	−117.747	0.256	−2.848**	−2.528**
ARG-6-3-1-4 × GKVK-3	−1.730**	−15.125**	−1.253**	−0.187*	−1.395	0.525**	1.038	−128.486	3.648**	0.178	−1.678**
ARG-6-3-1-4 × RHA 6D-1	−0.130	−12.465**	−0.613**	−0.227**	−1.045	−0.116	1.004	69.148	−3.424**	−0.941	1.197**
ARG-6-3-1-4 × 95-C-1	1.020	6.900*	0.153	0.001	2.205**	−0.430**	0.136	−2.319	−2.624**	0.064	1.763**
ARG-6-3-1-4 × LTRR-822	0.020	5.080	0.068	0.051	1.155	0.016	−0.240	−64.819	1.831**	−2.411**	1.109**

Table 6: (continued)

Hybrids	Days to 50% flowering	Plant height (cm)	Head diameter (cm)	Stem diameter (cm)	Days to maturity	100 seed weight (g)	Volume weight (g/100 mL)	Seed yield (kg/ha)	Hull content (%)	Seed filling per cent	Oil content (%)
ARG-6-3-1-4 × M-17-R	0.620	10.685**	0.132	0.049	1.605*	0.257	−0.672	79.504	−3.588**	0.609	−0.116
ARG-6-3-1-4 × MR-1	0.520	10.505**	0.648**	0.058	0.905	0.209	−0.382	99.847	0.419	2.514**	0.036
ARG-6-3-1-4 × RHA 272-II	0.720	−2.855	0.338	0.072	−0.745	−0.202	−0.452	−155.030	−1.130*	0.337	−1.206**
ARG-6-3-1-4 × X-15-NB-10	0.070	−1.950	−0.008	0.017	−1.095	0.058	−1.156	40.126	0.778	0.440	−1.582**
ARG-6-3-1-4 × GKVK-2	−0.730	3.110	0.728**	0.246**	−0.745	0.026	0.633	53.459	1.465**	−0.361	−0.579**
ARG-6-3-1-4 × RHA-93	−0.380	−3.885	−0.193	−0.079	−0.845	−0.342*	0.089	8.570	2.624**	−0.431	1.054**
SE±	0.536	3.210	0.226	0.042	0.714	0.144	0.896	104.899	0.470	0.575	0.205

**significant at $p \leq 0.01$. *significant at $p \leq 0.05$.

Table 7: Variance due to general and specific combining ability for seed yield and component traits in sunflower.

Characters	Variance due to GCA	Variance due to SCA	GCA/SCA
Days to 50% flowering	2.456	0.877	2.800
Plant height (cm)	62.160	35.930	1.730
Head diameter (cm)	0.048	0.158	0.304
Stem diameter (cm)	0.002	0.009	0.222
Days to maturity	3.285	2.594	1.266
100 seed weight (g)	0.049	0.100	0.490
Volume weight (g/100 mL)	3.057	0.875	3.494
Seed yield (kg/ha)	8422.689	13,879.046	0.607
Hull content (%)	2.241	5.314	0.422
Seed filling (%)	3.177	5.083	0.625
Oil content (%)	1.850	7.344	0.252

number of seeds per head and in turn with seed yield. The hybrid MUT-2-8-3-2 × GKVK-3 (1.160) topped the list of crosses that showed the highest significant positive *SCA* effects followed by ARG-6-3-1-4 × GKVK-2 (0.728) and ARG-6-3-1-4 × MR-1 (0.648). The prevalence of non-additive gene action for this trait was also observed by Parameshwarappa et al. (2008) and Machikowa et al. (2011). Ten out of 50 hybrids showed a significant *SCA* effect for stem diameter, of which five were in the positive and five were in the negative direction. The hybrid ARG-6-3-1-4 × GKVK-2 (0.246) expressed a significant positive *SCA* effect followed by MUT-2-8-3-2 × GKVK-3 (0.141) and ARG 3 × RHA 6D-1 (0.138). Contrary to this, cross ARG 3 × GKVK-2 (–0.254) exhibited the highest significant negative *SCA* effects followed by ARG-6-3-1-4 × RHA-6D-1 (–0.227). However, the magnitude of *SCA* effects among the hybrids was very low for this trait. These results confirm those observed in the studies of Shankar et al. (2007).

Eight cross combinations showed significant positive *SCA* effects for 100 seed weight. Of these, ARG-2-1-2 × RHA 6D-1 (0.535), ARG-6-3-1-4 × GKVK-3 (0.525) and MUT-2-8-3-2 × X-15-NB-10 (0.456) were the best specific combiners. Of the top three ranked hybrids for the trait two crosses involved at least one parent with a low *GCA* effect i.e., these crosses were of high × low or low × high type of specific combinations suggesting the involvement of non-additive gene action in the inheritance of this trait. Patil et al. (2017) reported good specific combiners for 100 seed weight and also reported the existence of non-additive gene action in the inheritance of this trait. The cross combination E002 × X-15-NB-10 (2.816) was the best specific combiner for volume weight followed by MUT-2-8-3-2 × MR-1 (2.157) and MUT-2-8-3-2 × LTRR-822 (2.049). All three of the best crosses involved at least one parent

with low *GCA* effects, clearly suggesting the involvement of non-additive gene action in the inheritance of the trait. Similar results were obtained by Chandra et al. (2011) and Lakshman et al. (2019).

Concerning seed yield, *SCA* ranged from –258.81 to 404.04 with the best specific combiner being MUT-2-8-3-2 × GKVK-3 (404.036), followed by ARG-2-1-2 × LTRR-822 (295.926) and E002 × M-17-R (254.425). A large pool of variability was evident by how wide the range, as well as the magnitude and direction of *SCA* effects on the character, were. In the first of two top crosses, MUT-2-8-3-2 × GKVK-3 and ARG-2-1-2 × LTRR-822, at least one parent with high and another parent with low *GCA* effects were present. This could be attributed to the involvement of non-additive gene action. However, it was interesting to note that in the third cross, E002 × M-17-R both the parents with low *GCA* effects were involved, suggesting the prevalence of overdominance and epitasis. Dhillon and Tyagi (2016) also reported good specific combiners for seed yielding sunflowers.

Thirty-five out of 50 crosses showed significant *SCA* effects for hull content, of which ARG-2-1-2 × RHA-93 (–4.646) topped the list of hybrids expressing a significant negative *SCA* effect followed by ARG-6-3-1-4 × M-17-R (–3.588) and ARG-2-1-2 × X-15-NB-10 (–3.501). Contrarily, cross MUT-2-8-3-2 × X-15-NB-10 (3.731) and ARG-6-3-1-4 × GKVK 3 (3.648) exhibited the highest significant negative *SCA* effects. Bhoite et al. (2018) reported desired negative specific combiners for this trait. Concerning seed filling percentage, 14 and 13 crosses exhibited significant positive and negative *SCA* effects, respectively. The cross, MUT-2-8-3-2 × RHA 272-II (5.329), ARG-2-1-2 × LTRR-822 (3.556) and ARG-2-1-2 × X-15-NB-10 (2.718) were the best specific combiners for seed filling percentage. Meena et al. (2013) and Sharma and Shadakshari (2021) also reported good specific combiners for seed filling percentage.

Highly significant *SCA* effects for oil content were observed in 39 crosses, of which 19 and 20 crosses expressed positive and negative significant *SCA* effects, respectively. The hybrids E002 × X-15-NB-10 (2.520), MUT-2-8-3-2 × GKVK-3 (2.070) and ARG-2-1-2 × RHA-93 (1.961) topped the list of crosses expressing significant positive SCA effects. The best specific combination of E002 × X-15-NB-10 was from parents with a low combining ability which indicated the involvement of non-additive gene action and also the existence of overdominance and epitasis in the inheritance of this trait. Non-additive gene action for oil content was also reported by Azad et al. (2016) and Hilli et al. (2020). None of the hybrids was good specific combiners for all the characters studied. However, the cross combination MUT-2-8-3-2 × GKVK 3 was found to be a good specific combiner for stem diameter, 100 seed weight, seed yield, seed filling percentage, oil content and oil yield hence it is a candidate to be tested in large scale yield trials over locations and seasons to confirm its potential for commercial cultivation. Since these hybrids are based on

diverse cytosterile sources, even their on par performance with PET 1 cytoplasm based hybrids will be sufficient enough so that the variability for cytoplasmic male sterility of sunflower hybrids can be expanded.

Variance due to general and specific combining ability effects

The ratio of *GCA* to *SCA* is used to indicate the predominance of non-additive gene action in the inheritance of the traits. The results revealed that, among the 12 characters studied, characters, days to 50% flowering, plant height, days to maturity and volume weight there was a preponderance of additive gene action as indicated by greater than unity GCA to SCA ratio (Table 7) while remaining characters *viz.*, head diameter, stem diameter, 100 seed weight, seed yield, hull content, seed filling per cent, oil content and oil yield manifested a higher magnitude of SCA variance compared to GCA variance. Similar to the present findings, non-additive gene actions were documented for head diameter, stem diameter, 100 seed weight, seed yield, hull content, seed filling per cent and oil content, by Bhoite et al. (2018) and Hilli et al. (2020).

Proportional contribution of lines, testers and line × tester interaction for the performance of hybrids.

The data on the proportional contribution of lines, testers and line × tester interaction for studied traits revealed that the line × tester interaction contributed more to the performance of hybrids for most of the characters such as head diameter, stem diameter, 100 seed weight, seed yield, hull content, seed filling per cent, oil content and oil yield (Table 8). However, for days to 50% flowering, days to maturity and volume weight contribution of the testers was greater when compared to lines and line × tester interaction. Shankar et al. (2007) observed similar results and emphasized that due care needs to be excised when selecting the inbreds/lines to be used as parents for hybridization and to safely use these sources to broaden the genetic base of CMS source so that this valuable oilseed crop can be safeguarded from any eventuality of biotic and abiotic threats in the future.

Estimation of standard heterosis

Increased seed yield and oil content are the ultimate objectives in oilseed breeding hence high heterosis for these characters is always the goal. For seed yield, only two hybrids exhibited significant positive heterosis over the check hybrid KBSH-44. The hybrid MUT-2-8-3-2 × GKVK-3 (16.39%) exhibited the highest

Table 8: Proportional contribution of lines, testers and L × T interaction to the total variance among the hybrids.

Sl. No.	Characters	Lines (L)	Testers (T)	L × T interaction
1	Days to 50% flowering	28.730	56.228	15.042
2	Plant height (cm)	39.842	35.814	24.344
3	Head diameter (cm)	13.012	28.252	58.736
4	Stem diameter (cm)	3.950	35.319	60.732
5	Days to maturity	26.322	47.364	26.314
6	100 seed weight (g)	27.379	21.159	51.462
7	Volume weight (g/100 mL)	28.199	55.045	16.756
8	Seed yield (kg/ha)	18.907	31.451	49.643
9	Hull content (%)	27.589	19.434	52.977
10	Seed filling (%)	28.229	29.903	41.868
11	Oil content (%)	4.622	13.035	82.344

significant positive heterosis followed by ARG-2-1-2 × LTRR-822 (15.04%) (Table 9). Concerning check KBSH 53 and KBSH 78, the standard heterosis of the hybrid MUT-2-8-3-2 × GKVK-3 was 15.11 and 20.99% respectively. Hybrid ARG-2-1-2 × LTRR-822 ranked second for seed yield with standard heterosis of 15.04, 13.78 and 19.59% over the checks KBSH 44, KBSH 53 and KBSH 78 respectively. For KBSH-78, three hybrids expressed significant positive heterosis and four showed significant negative heterosis with the range of heterosis being –23.67 to 20.99%. Both the hybrids MUT-2-8-3-2 × GKVK-3 (LxH) and ARG-2-1-2 × LTRR-822 (HxL) had only one of the parents as a good general combiner suggesting the preponderance of non-additive gene action. Awaad et al. (2016), Rathi et al. (2016) and Ailwar et al. (2020) in their respective studies have reported high levels of standard heterosis for seed yield.

In oilseed crops, oil is the ultimate end product and hence, increasing oil content is of prime importance. Seven out of 50 hybrids exhibited significant positive heterosis better than KBSH-44 for oil content (Supplementary Table 3). The cross ARG-6-3-1-4 × RHA95-C-1 (8.42%) exhibited highest positive heterosis followed by ARG-6-3-1-4 × RHA 6D-1 (6.74%), ARG-6-3-1-4 × LTRR-822 (4.44%) and ARG-2-1-2 × RHA 6D-1 (2.55%). However, 37 hybrids had significant negative heterosis, and cross ARG-3 × RHA-93 (–25.07%) showed the highest significant negative heterosis over KBSH-44. Over KBSH-53 and KBSH 78, only two hybrids ARG-6-3-1-4 × RHA 95-C-1 and ARG-6-3-1-4 × RHA 6D-1 exhibited significant positive heterosis, with heterosis ranging from –27.25 to 5.28% in case of KBSH 53 and -28.22–3.87% concerning KBSH 78. Parameshwarappa et al. (2008); Rathi et al. (2016) and Ailwar et al. (2020) reported significant heterosis for oil content.

Table 9: Top ranking hybrids with desirable standard heterosis compared to KBSH-44, KBSH-53 and KBSH-78 for seed yield and component traits.

Characters	Crosses	% standard heterosis over KBSH-44	% standard heterosis over KBSH-53	% standard heterosis over KBSH-78	Type of combinations
Days to 50% flowering	ARG-2-1-2 × GKVK-2	−15.08**	−18.94**	−2.28	H × H
	MUT-2-8-3-2 × M-17-R	−13.49**	−17.42**	−0.46	L × L
	ARG-2-1-2 × RHA-93	−13.49**	−17.42**	−0.46	L × H
	E002 × M-17-R	−13.10**	−17.05**	0.00	L × L
	E002 × RHA 272-II	−13.10**	−17.05**	0.00	L × L
Plant height (cm)	ARG 3 × RHA-93	−27.33**	−30.69**	−8.59**	H × H
	E002 × RHA-93	−26.61**	−30.00**	−7.68*	L × H
	ARG 3 × GKVK-2	−25.44**	−28.88**	−6.21*	H × H
	ARG-2-1-2 × RHA-93	−20.84**	−24.49**	−0.42	L × H
	E002 × RHA 272-II	−20.34**	−24.02**	0.20	L × L
Head diameter (cm)	MUT-2-8-3-2 × GKVK-3	−1.81	0.67	3.91	L × H
	ARG-2-1-2 × RHA 6D-1	−2.13	0.34	3.56	H × L
	ARG-6-3-1-4 × MR-1	−2.46	0.00	3.21	H × H
	ARG-6-3-1-4 × RHA95-C-1	−2.96	−0.51	2.69	H × H
	ARG-2-1-2 × RHA95-C-1	−3.12	−0.67	2.52	H × H
Stem diameter (cm)	MUT-2-8-3-2 × GKVK-3	−0.31	−0.71	18.57**	L × H
	ARG-6-3-1-4 × GKVK-2	−0.61	−1.02	18.20**	H × H
	ARG-6-3-1-4 × MR-1	−1.43	−1.83	17.23**	H × H
	ARG-6-3-1-4 × 95-C-1	−1.63	−2.03	16.99**	H × H
	ARG-6-3-1-4 × LTRR-822	−1.63	−2.03	16.99**	H × L

Table 9: (continued)

Characters	Crosses	% standard heterosis over KBSH-44	% standard heterosis over KBSH-53	% standard heterosis over KBSH-78	Type of combinations
Days to maturity	ARG-2-1-2 × GKVK-2	−9.79**	−11.66**	0.00	H × H
	E002 × M-17-R	−9.52**	−11.40**	0.29	L × L
	ARG-2-1-2 × RHA-93	−8.99**	−10.88**	0.88	H × H
	ARG-2-1-2 × M-17-R	−8.99**	−10.88**	0.88	H × L
	ARG 3 × GKVK-2	−8.20**	−10.10**	1.76	H × H
100 seed weight (g)	ARG-2-1-2 × RHA95-C-1	−1.68	19.96**	7.94*	H × H
	ARG-2-1-2 × RHA 6D-1	−4.12	16.98**	5.26	H × L
	ARG-6-3-1-4 × GKVK-3	−4.71	16.26**	4.61	H × H
	MUT-2-8-3-2 × GKVK-3	−6.60	13.95**	2.54	L × H
	ARG-2-1-2 × RHA 272-II	−7.25	13.17**	1.83	H × L
Volume weight (g/100 mL)	ARG-6-3-1-4 × GKVK-3	4.83	16.35**	9.20**	H × H
	ARG-6-3-1-4 × M-17-R	3.51	14.88**	7.83**	H × L
	ARG-6-3-1-4 × GKVK-2	3.26	14.61**	7.57**	H × H
	MUT-2-8-3-2 × GKVK-3	2.22	13.45**	6.49*	L × H
	ARG-2-1-2 × M-17-R	1.40	12.54**	5.63*	H × L
Seed yield (kg/ha)	MUT-2-8-3-2 × GKVK-3	16.39*	15.11*	20.99**	L × H
	ARG-2-1-2 × LTRR-822	15.04*	13.78	19.59**	H × L
	ARG-2-1-2 × RHA-93	12.00	10.78	16.43*	H × H
	ARG-6-3-1-4 × MR-1	10.59	9.38	14.96	H × H
	MUT-2-8-3-2 × RHA-93	10.18	8.98	14.54	L × H

Table 9: (continued)

Characters	Crosses	% standard heterosis over KBSH-44	% standard heterosis over KBSH-53	% standard heterosis over KBSH-78	Type of combinations
Hull content (%)	ARG-2-1-2 × RHA-93	−22.50**	−23.84**	−18.34**	H × H
	ARG-6-3-1-4 × RHA 6D-1	−22.26**	−23.61**	−18.09**	H × L
	ARG-6-3-1-4 × M-17-R	−21.43**	−22.79**	−17.21**	H × L
	ARG-2-1-2 × LTRR-822	−17.31**	−18.75**	−12.88**	H × L
	ARG-6-3-1-4 × RHA95-C-1	−16.81**	−18.26**	−12.35**	H × L
Seed filling per cent	ARG-2-1-2 × RHA-93	−0.47	1.05	0.45	H × H
	ARG-2-1-2 × LTRR-822	−0.64	0.88	0.28	H × L
	E002 × X-15-NB-10	−0.85	0.66	0.06	L × L
	ARG-6-3-1-4 × MR-1	−0.92	0.59	−0.01	H × H
	ARG-2-1-2 × RHA95-C-1	−0.94	0.57	−0.03	H × H
Oil content (%)	ARG-6-3-1-4 × RHA95-C-1	8.42**	5.28**	3.87**	H × H
	ARG-6-3-1-4 × RHA 6D-1	6.74**	3.64**	2.26**	H × L
	ARG-6-3-1-4 × LTRR-822	4.44**	1.41	0.06	H × L
	ARG-2-1-2 × RHA 6D-1	2.55**	−0.42	−1.75*	H × L
	ARG-6-3-1-4 × MR-1	2.11**	−0.85**	−2.18**	H × H

Conclusions

The diversified CMS line ARG-6-3-1-4was identified as the best general combiner for stem diameter, volume weight, seed yield, hull content and oil content while tester RHA 93 was the best general combiner for plant height, seed yield and oil content so these can be utilized as desirable parents for developing commercial

sunflower hybrids. However none of the hybrids were good specific combiners for all the characters, the cross MUT-2-8-3-2 × GKVK 3 was found to be a good specific combiner for stem diameter, 100 seed weight, seed yield, seed filling percentage and oil content, while, ARG-6-3-1-4 × GKVK 3 was a good specific combiner for days to 50% flowering, plant height and 100 seed weight. The highest standard heterosis for seed yield was observed in the hybrid MUT-2-8-3-2 × GKVK 3 followed by ARG-2-1-2 × LTRR 822, while the highest oil content was seen in the cross ARG-6-3-1-4 × RHA95-C-1 so these hybrids can be exploited for sunflower improvement to enhancing the variability for cytoplasmic male sterility of oilseed sunflower. Further, as these lines, hybrids are based on wild/non-conventional cytoplasmic sources of sunflower they can be screened for pests and diseases as well as for abiotic stress because these wild species are the storehouse of valuable genes helping them survive in extreme climatic conditions.

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