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Genetic variability, character association and divergence studies in sunflower (*Helianthus annuus L.*) for improvement in oil yield

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Abstract

Present experiment was conducted on 32 sunflower genotypes (11 CMS lines and 21 Restorer lines) on the basis of their diverse origin, growth habit, phenology and adaptation with an object to assess variation in oil yield attributing traits. The total variability consisting heritable and non-heritable components of the characters were estimated by genotypic (GCV) and phenotypic (PCV) coefficient of variation, heritability and genetic advance (GA) and the best positive traits for sunflower improvement through selection and breeding were assessed. All the characters among the different genotypes exhibited significant variation. Oil yield was highly significant and positively correlated with seed yield (kg/ha) (0.95), number of filled grain/head (0.85), grain filling% (0.65), days to 50% flowering (0.417), plant height (0.33). Seed yield (kg/ha) was highly significant and positively correlated with plant height (0.33), head diameter (0.30). Seed yield (kg/ha) varied from 625 (CMS-16A) to 1120(CMS-302A) in lines and 576 (EC-601751) to 1221(R-630) in testers respectively. The highest 100 seed weight and hull content (%) was obtained from P-2-7-1A. Seed yield, oil yield (kg/ha) and numbers of filled grains were obtained from EC 601978. Genotypes were classified into eight distinct clusters with a maximum inter distance observed between cluster VI and VII. The experiment assisted to identify the superior genotype among diverse genotypes of sunflower which can act as parents with desirable traits like number of filled grain/head, volume weight/100 ml, 100-seed weight, oil content and seed yield/plant for further breeding purposes.

Keywords: *Sunflower, Genetic variability, Character association, Genetic diversity, Yield attributing traits*

1. Introduction

Sunflower (*Helianthus annuus* L., 2n = 34)) is a very good source of vegetable oil due to high oil quality (linoleic acid-55-70%) and high concentration of poly unsaturated fatty acids with 35-42% oil content in seeds. Sunflower improvement progrm mainly depends on hybridization by using desirable parent with genetically diverse

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background and agronomically important traits. Information regarding the nature and magnitude of genotypic and phenotypic variability is very important to initiate any plant improvement program. Estimation of variability in any plant species can be accomplished by suitable biometrical techniques. It is also possible to estimate the amount of variability, which is due to genotype and environment. It is very important to the plant breeders that portion of variability which is heritable controlled by genotype. A particular trait can be improved easily through simple selection, if the heritability estimate for the character is high. Estimate of genetic advance is also very important to get an idea on the speed of genetic gain through selection. A trait which showed high heritability with high genetic advance can be improved through selection. Yield which can be considered as a result of different important traits, will be estimated by association between important components. Genetic divergence among a set of germplasm provides background for selection and development of new genetically diverse inbred. Information regarding genetic variation and genetic relationships between conserved germplasm is important for efficient germplasm preservation, characterization and subsequent use by sunflower breeders. Further, success of recombination or heterosis breeding depends on the choice of parents for crossing program. Selection of parents, with desirable characters, from diverse groups, in turn, increases the possibility of isolating good recombinants in the segregating generations. Selection of divergent parental material in hybridization program is an important breeding strategy for the development of superior hybrid/cultivar (Madhavi Latha, 2017). By the help of Mahalanobis D^2 statistical techniques the parental lines can be identified with presence of high variability and heritability.

2. Materials and methods

2.1 Experimental materials and field techniques

Experiment was carried out during kharif season of 2018 and again repeated in rabi of 2018-2019. Experimental materials (Table 1) were collected from Oilseeds Research Station, India on the basis of their diverse origin, growth habit, phenology and adaptation. Thirty two sunflower genotypes (11 CMS lines and 21 Restorer lines) were grown in a randomized complete block design with two replications in a plot size of 1.8 m X 3.0m. Each plot contained three rows with spacing of 60 cm X 30 cm. The germinated seed of sunflower used as the planting materials and one per hill were maintained throughout the cropping period. A lines (CMS) and their

Table 1: List of genetic materials used in experiment		
Parents	**Origin**	**Important traits**
P-89-1A, CMS-207A, CMS-302A,	IIOR, Hyderabad	High seed yield
CMS-10A	IIOR, Hyderabad	Early maturity and high oil content
P-2-7-1A	IIOR, Hyderabad	High test weight and high seed yield
CMS-16A, CMS-850A, CMS-852A, CMS-853A	ORS, Latur	Early maturity, Dwarf and high oil content
CMS-103A, CMS-107A,	UAS, Bangalore	Early maturity and high oil content
R-138-2, R-630, R-104, R-12-96	UAS, Bangalore	High seed yield and black color seed
R-1-1	IIOR, Hyderabad	High test weight and high seed yield
R-104, R-107	UAS, Bangalore	High seed yield and black color seed.
EC-602060, EC-601978, R-341	ORS, Latur	Early maturity, high test weight and black color seed
EC-623027(M)	ORS, Latur	High test weight and high seed yield
EC-623021, EC-623023,	ORS, Latur	High test weight and high seed yield
EC-601751, EC-601725	ORS, Latur	Early maturity, high test weight and black color seed
EC-623029, EC-601958, EC-623011	ORS, Latur	High oil content, stress tolerant, black color seed
EC-623016, EC-512682	ORS, Latur	Early maturity, Downy mildew and stress resistant,
R-6D-1	UAS, Bangalore	High oil content

maintainer B lines (CMS B) were isogonics lines which differed only from male fertility. Another is completely absent in A line. The data regarding the yield attributing traits were recorded only from B lines. Similar agronomic package and practices were carried out in both the seasons. Standard agronomic practices and plant protection measures were followed to raise good crop.

2.2 Experimental soil

The soil of the experimental plot is sandy loam in texture with medium to low fertility status and acidic in nature, representing more or less red and lateritic soils. The physico-chemical properties of the experimental field are: 235.4 Available N (kg/ha), 20.4 Available P (kg/ha), 175.5 Available K (kg/ha), 0.5 Organic carbon (%), 6.5 pH, 22 Sand (%), 42 Silt (%), 36 Clay (%).

2.3 Recording of data

The observations for all the traits were recorded on 10 randomly selected competitive plants for each genotype in each replication expect for days to flower, days to maturity were recorded on plot basis. Number of days was recorded by taking information from sowing to 50% flowering of plants in each entry and in each replication. The height of fully matured plant in centimetres from the base of the plant to the basal surface of the capitulum was recorded as plant height. The head diameter was recorded in centimetres from both the diagonal axes at maturity. Total number of filled seed per capitulum (average of 5 heads). All the filled seeds of 10 selected plants was weighed in gram on Top Pan Digital Balance and expressed as seed yield per plant. The proportion of filled seeds to the total seeds of a plant is expressed as seed filling per cent. Seeds of sample plants from each entry were bulked, dried and cleaned. Three samples containing 100 seeds were drawn from each lot and 100-seed weight was recorded as average of those three lots in gram (g) using Semi-Micro Electronic Balance. 10 g of seeds were drawn from each lot and dehulled. The kernel weight and Husk weight was recorded as average of those three lots in gram (g) using Semi-Micro Electronic Balance. The measuring cylinder was filled to 100 ml volume with seeds of each entry and was weighed in gram as volume weight /100 ml using Semi-Micro Electronic Balance. Ten gram of well filled seed was extracted from each sample and it was dehulled and the ratio of husk weight to total weight of the seed recorded expressed as the hull content (%). Seed yield per plant (g) converted to seed yield (kg/ha) by multiplying with the conversion factor (55.55).(10000 m^2/0.60 X 0.30) X 1000 (kg/ha). A randomly bulk sample of filled seeds was drawn from selected plant produce weighing 50 gram from each entry in each replication. The oil content in percentage was measured using Nuclear Magnetic Resonance (NMR) facility available at Institute of Agriculture, PSB, Visva-Bharati University, Sriniketan and Bidhan Chandra Krishi Viswa Vidyalaya, Mohanpur, Nadia, West Bengal.

2.4. Statistical analysis

Best gain traits for sunflower yield improvement through selection and breeding were assessed based on genetic variability, heritability and correlation. Analysis of variance, phenotypic and genotypic variance, phenotypic coefficient of variation (PCV), genotypic coefficient of variation (GCV), heritability in broad sense and genetic advance as percentage of population mean (GAM%) (selection intensity at 5 % level) were estimated using standard procedure (Singh and Chaudhary, 1979). The differences in mean were tested by Tukey's test using MSTAT statistical software (MSTAT-C 1991, Michigan State University, East Lansing, MI). Values of PCV and GCV for all the traits were divided into three categories, viz., [20 % (high), 10-20 % (moderate) and \10 % (low) (Shivasubramanian and Madhavamenon, 1973). Heritability in the broad sense was classified as low (<40 %), medium (40-80 %) and high (>80 %).The genetic advance was also classified as low (0-10 %), moderate (10-20 %) and high ([20 %) (Robinson et al., 1949; and Johnson et al., 1955). The replicated data for correlation coefficient and Divergence analysis (cluster analysis) were analyzed using the Windostat version 8.6 from Indostat service Hyderabad, India. Further, cluster analysis was performed to classify the germplasm to facilitate parental selection for breeding and genetic improvement of sunflower.

3. Results and Discussion

Wide range of variability was noticed in all the traits in the genotypes studied. The mean, maximum and minimum values and coefficient of variation (CV) of these characters have been tabulated accordingly. The

analysis of variance (ANOVA) of 32 sunflower genotypes pooled over two seasons with respect to 13 quantitative traits. The mean squares due to genotypes for all the above characters were highly significant which indicated genetic variability among the experimental materials. The significant mean square due to genotypes × season interaction for all the above characters indicated differential performance of the genotypes with the change in growing environment.

3.1. Performance per se

Among lines CMS 16A recorded least time to flower (56.7days) whereas, CMS-853A took maximum time (64.6 days) to flower. EC-602060 and EC-601905 were found earliest (60 days) among the tester whereas R-12-96 took maximum time to flower (Table 2). The highest plant height, significantly superior to other germplasm, was recorded for CMS 207A which also had large head diameter and highest % of seed filling (Autogamy). Seed yield (kg/ha) varied from 625 (CMS-16A) to 1120(CMS-302A) in lines and 576 (EC-601751) to 1221(R-630) in testers respectively. The highest 100 seed weight and hull content (%) was obtained from P-2-7-1A. Seed yield, oil yield (kg/ha) and numbers of filled grains were obtained from EC 601978. This result were in agreement with the experiment of Tan (2010) for seed yield per plant, head diameter, oil content and 1,000 seed weight. Significant differences among the sunflower genotypes for the yield attributing traits was reported by Sheshaiah Shankergoud (2015), Neelima *et al.* (2016), Dudhe *et al.* (2017).

Table 2: Mean values for yield attributing traits of 32 genotypes in sunflower germplasms						
Name of the parents	Days to 50% flowering	Plant height (cm)	Head diameter (cm)	Seed yield (kg/ha)	No. of filled seed/head	Autogamy (seed filling) %
Pet-2-7-1A	63.13	93.25	8.40	843.83	251.60	81.40
PET-89-1-A	60.15	84.40	7.75	760.40	288.03	83.38
CMS-10A	56.70	78.30	7.10	625.83	252.85	85.33
CMS-107A	60.90	94.98	9.45	767.40	290.63	81.40
CMS-302A	64.85	114.65	9.65	1119.25	422.00	82.88
CMS-16A	56.93	75.48	7.00	875.00	415.25	84.85
CMS-850A	56.93	84.05	8.10	839.25	397.75	86.85
CMS-853A	64.63	115.15	10.35	961.25	305.75	83.30
CMS-852A	62.23	114.85	10.60	791.00	288.25	86.80
CMS-103A	61.78	120.95	11.10	717.75	247.50	87.80
CMS-207A	65.08	123.80	11.35	839.50	331.25	88.00
RHA-6D-1	62.15	95.33	8.05	722.00	254.25	75.35
R-12-96	70.15	92.53	9.03	972.25	440.23	78.30
R-630	67.15	109.63	7.30	1021.00	493.70	80.28
EC-601978	60.20	87.68	8.40	538.25	197.65	82.25
EC-601958	63.15	74.33	6.88	635.50	311.45	82.25
R-138-2	67.20	85.83	7.60	844.00	450.43	76.35

Table 2 (Cont.)						
Name of the parents	Days to 50% flowering	Plant height (cm)	Head diameter (cm)	Seed yield (kg/ha)	No. of filled seed/head	Autogamy (seed filling) %
RHA-1-1	63.65	91.95	8.30	690.00	290.93	75.85
R-107	67.15	91.30	8.75	799.25	344.60	83.25
R-104	65.20	99.53	8.25	680.25	374.85	76.35
EC-623029	60.90	85.00	7.80	746.00	324.00	76.90
R-341	65.85	110.40	8.70	717.25	348.50	80.90
EC-602060	59.90	82.20	6.80	646.25	345.75	84.85
EC-623011	61.88	73.38	7.93	849.75	529.50	79.90
EC-512682	62.85	90.48	8.98	697.25	368.75	79.90
EC-623027	69.98	103.70	9.90	816.00	297.25	75.00
EC-623021	67.55	96.60	7.55	660.00	254.00	77.60
EC-623023	64.65	88.35	8.03	623.50	254.50	77.25
EC-601718	59.83	88.35	9.75	616.00	256.25	74.95
EC-601751	63.68	92.33	8.95	576.00	218.00	75.75
EC-601725	65.63	85.15	8.75	654.00	276.00	77.50
EC-623016	66.10	92.02	7.40	667.00	254.50	74.40
Mean	63.38	91.81	8.50	759.75	324.25	80.57
C.V.	3.28	6.98	8.05	7.25	9.11	5.89
F ratio	11.13	38.98	20.25	23.65	30.39	3.01
SEm (±)	1.04	3.20	0.34	27.53	14.78	2.37
C.D. 5%	2.92	9.00	0.96	77.30	41.50	6.66
C.D. 1%	3.87	11.91	1.27	102.37	54.95	8.82
Range Lowest	56.70	73.38	6.88	538.25	197.65	74.95
Range Highest	70.15	123.80	11.35	1119.25	529.50	88.00
Germplasms (M.S.)	48.20**	1599.36**	9.46**	71685.41**	26540.46**	67.89 **
Germplasms × Seasons (M.S.)	15.07**	243.34**	2.24*	88322.78**	9047.46**	99.54**
Error (M.S.)	4.33	41.04	0.47	3030.495	873.28	22.52
Name of the parents	100 Seed weight (g)	100 Kernel wt. (g)	Hull content (%)	Vol. wt. (g/100 cc)	Oil %	Oil yield (kg/ha)	Seed yield/ plant (g)
Pet-2-7-1A	6.10	3.63	36.15	35.43	33.90	286.75	15.35
PET-89-1-A	4.80	2.99	32.53	35.25	35.55	271.00	13.85

Table 2 (Cont.)							
Name of the parents	100 Seed weight (g)	100 Kernel wt. (g)	Hull content (%)	Vol. wt. (g/100 cc)	Oil %	Oil yield (kg/ha)	Seed yield/ plant (g)
CMS-10A	4.50	2.78	33.88	36.28	36.10	226.50	11.38
CMS-107A	4.80	2.96	34.25	34.68	35.15	270.00	13.98
CMS-302A	4.83	3.33	32.18	39.50	37.30	417.75	20.35
CMS-16A	3.84	2.59	33.28	39.50	36.70	321.50	15.91
CMS-850A	3.84	2.69	31.13	41.40	38.08	319.75	15.26
CMS-853A	5.70	4.00	30.20	42.68	35.75	343.93	17.48
CMS-852A	5.00	3.40	32.10	42.20	35.43	280.43	14.38
CMS-103A	5.30	3.90	26.50	45.03	35.98	258.40	13.05
CMS-207	4.60	3.15	31.80	40.15	35.65	299.00	15.25
RHA-6D-1	5.16	3.41	33.93	35.70	37.58	267.00	14.63
R-12-96	4.01	2.64	33.68	33.63	34.43	386.48	19.80
R-630	3.75	2.53	32.60	33.60	33.25	410.95	20.83
EC-601978	4.95	3.18	35.28	33.90	36.90	200.65	10.85
EC-601958	3.71	2.48	33.10	34.83	37.58	277.20	12.85
R-138-2	3.40	2.22	34.28	34.90	35.70	371.43	17.20
RHA-1-1	4.31	2.97	31.05	32.68	34.95	268.93	13.93
R-107	4.21	2.73	35.00	35.25	34.30	311.85	16.18
R-104	3.14	2.02	35.68	34.08	33.50	290.85	13.78
EC-623029	4.82	3.17	35.22	37.53	34.73	259.00	15.63
R-341	3.75	1.96	34.47	34.65	33.70	242.00	13.04
EC-602060	3.40	2.26	34.37	33.70	36.10	233.75	11.75
EC-623011	2.91	2.02	32.08	33.70	35.90	305.25	15.45
EC-512682	3.45	2.38	32.60	36.60	36.20	252.25	12.68
EC-623027	5.68	4.05	28.35	43.58	37.73	307.88	14.85
EC-623021	5.33	3.75	29.50	42.35	37.60	248.20	12.00
EC-623023	5.03	3.53	30.10	41.58	40.10	250.08	11.33
EC-601718	4.88	3.48	28.73	41.10	40.03	246.63	11.20
EC-601751	5.33	3.75	29.60	42.20	39.50	227.55	10.45
EC-601725	5.23	3.50	33.03	43.17	38.88	253.23	11.90
EC-623016	5.03	3.62	28.25	41.52	38.33	255.10	10.78
Mean	4.52	3.03	33.12	37.8844	36.3289	286.29	14.29

Name of the parents	100 Seed weight (g)	100 Kernel wt. (g)	Hull content (%)	Vol. wt. (g/100cc)	Oil %	Oil yield (kg/ha)	Seed yield/ plant (g)
Table 2 (Cont.)							
C.V.	1.18	2.53	19.95	3.6775	2.0461	8.28	10.63
F ratio	946.81	254.32	2.65	29.8202	24.8651	19.95	13.02
SEm (±)	0.03	0.04	3.30	0.6966	0.3717	11.85	0.76
C.D. 5%	0.07	0.11	9.28	1.9563	1.0438	33.29	2.13
C.D. 1%	0.10	0.14	12.29	2.5906	1.3822	44.08	2.82
Range Lowest	2.91	1.96	26.50	32.68	33.25	200.65	10.45
Range Highest	6.10	4.05	36.15	45.03	40.10	417.75	20.83
Germplasms (M.S.)	26.82 **	1.49*	115.53**	57.88**	13.74**	11213.37**	30.03**
Germplasms × Seasons (M.S.)	3.17*	0.159	29.93**	27.48**	26.79**	5899.71**	80.203**
Error (M.S.)	0.003	0.006	43.67	1.94	0.55	562.04	2.306

Note: *, **: Significant at *p* = 0.05 and 0.01, respectively.

3.2. Genetic parameters

The estimates of PCV and GCV (Table 3) were closer for all morphological characters, except grain filling (%) (7.22 and 4.18) and hull content (%) (23.70 and 12.80). The estimates of genotypic and phenotypic coefficient of variation (GCV and PCV) were high (>20%) for plant height (21.5 and 22.6), number of filled grains per head (24.7 and 26.3) and 100 kernel weight (20.2 and 20.8) emphasizing the existence of genetic variation, a pre-requisite for selection, in these genotypes. The estimates of genotypic and phenotypic coefficient of variation (GCV and PCV) were moderate (10-20%) for head diameter(17.6 and 19.4), seed yield in kg per hectare(17.2 and 18.4),100 seed weight(18.9 and 18.3), hull content(12.8 and 23.7) and oil yield (kg/ha) (18.0 and 19.8) and low(<10%) for days to 50% flowering(5.23 and 6.17), grain filling percentage(4.18 and 7.22) volume weight in gram per 100 ml (9.87 and 10.53) and Oil% (4.98 and 5.40). Manjula (2001) reported high PCV and GCV value for kernel weight and protein content. Teklewold *et al.* (1999) reported high PCV and GCV for seed yield. Sujatha *et al.* (2002) reported high PCV and GCV value for kernel weight and hull content. Seneviratne *et al.* (2004) reported high PCV and GCV value for 100 seed weight. High PCV and GCV for head diameter were reported by Mahmoud (2012). The estimates of GCV and PCV value for grain filling percentage and hull content percentage, showed higher differences which indicated the greater role of environmental factor influencing the expression of this character. Very low differences were observed for plant height, number of filled grains per head,100 kernel weight, head diameter, seed yield in kg per hactar,100 seed weight, plant height and days to 50% flowering indicating low sensitivity to environment and consequently greater role for genetic factors influencing the expression of these characters.

The estimates of heritability in broad sense (Table 3) were very high for plant height(82%), head diameter (83%), number of filled grains per head (88%) , seed yield in kg per hectare (85%), 100 seed weight (89%), 100 kernel weight (88%) and Volume weight (88%), oil content% (84%), oil yield in kg/ha (82%) and seed yield per plant (75%).

Very high heritability along with high genetic advance as percentage of mean were observed among the yield attributing characters (Table 3), Plant height (82%, 42.19%), number of filled grains (88%, 47.75%), 100 kernel weight (88%, 41.13%) showed. Head diameter (83% and 33.08%), branch seed yield (85% and 32.75%), 100 seed weight (89% and 37.19%) and oil yield (82.6% and 33.75%) also exhibited high heritability associated with high to moderate genetic advance as percentage of mean. This result indicated the predominance of

Table 3: Estimations of phenotypic and genotypic coefficients of variability, heritability, genetic advance and mean for yield attributing traits in sunflower germplasms

Genetic parameters	Days to 50% flower-ing	Plant height (cm)	Head dia-meter (cm)	Seed yield (kg/ha)	No. of filled seed/ head	Auto-gamy (seed filling)%	100 seed wt. (g)	100 Kernel wt. (g)	Hull content %	Vol. wt. (g/100cc)	Oil %	Oil yield (kg/ha)	Seed yield/pl (g)
GCV	5.23	21.50	17.65	17.24	24.71	4.18	18.94	20.24	12.80	9.87	4.98	18.02	18.42
PCV	6.17	22.60	19.40	18.70	26.33	7.22	18.32	20.82	23.70	10.53	5.40	19.83	21.26
Heritability %	72	82	83	85	88	34	89	88	79	88	85.6	82.6	75.1
Gen. adv as % of Mean (5%)	9.12	42.13	33.08	32.75	47.75	4.98	37.19	41.13	14.23	19.06	9.52	33.75	32.87
General mean	63.38	91.81	8.50	759.75	324.25	80.57	4.523	3.033	33.12	37.88	36.32	286.28	14.29
Exp mean next generation	69.15	130.48	11.31	1008.56	479.07	84.58	6.205	4.281	37.83	45.10	39.79	382.88	18.98

additive gene action for these traits which could be improved through early generation selection due to reliability of additive gene action.

Moderate heritability estimate was also found for volume weight (88%), but the genetic advance as percentage of mean was low (19.06%). Hence, selection for this trait should be practiced in advance generation to utilize maximum gain from selection.

Days to 50% flowering exhibited low heritability (72%) along with low genetic advance as percentage of mean (9.12%), indicating importance of non-additive type of gene action for the expression of this trait.

High heritability combined with high genetic advance was reported by Sujatha *et al.* (2002) for plant height; Vidhyavathi *et al.* (2005) for number of filled seeds/head; Khan *et al.* (2007) for plant height and head diameter; Hladni *et al.* (2011) for number of filled seeds/ head and seed yield/ plant; Tan (2010) for plant height and head diameter and 100 seed weight; Gontcharov *et al.* (2011) for seed yield and 100 seed weight; Ramesh *et al.* (2013) for plant height.

It can be concluded from the above findings wherein the results of PCV, GCV, heritability and genetic advance as percentage of mean have been furnished, revealed that selection for plant height, head diameter, 100-seed weight and volume weight (g) per 100 ml might be effective, whereas for maturity duration selection should be practiced in the advance generations.

These findings indicated that lines CMS 302A, P-2-7-1A, CMS-853A, CMS-207A, and tester- EC-623027,R-630, R-138-2 , R-107, EC 623011 for seed yield; lines- CMS 10A, CMS 16A and CMS 850A and testers- EC-623011,EC-623016 and EC-512682 for early maturity, medium plant height with low hull content were best parents over the environments for development of short duration with medium height and low hull content . They were more productive for development of higher seed yield and oil yielding sunflower hybrids. High seed yield level appeared due to higher head diameter, seed filling percentage, volume weight, 100-seed weight and more days to flower.

3.3. Character association

In the integrated structure of a sunflower plant most of the characters are interrelated and often change in one influence the others. In some cases, this interrelation is strong, while in others it may be weak. In the present investigation, genotypic and phenotypic correlation coefficients among 13 agronomical characters (Table 4) were estimated separately to study how seed yield was influenced by other component traits. Analysis of correlation revealed that genotypic correlation coefficients, in general, were higher in magnitude than the corresponding phenotypic correlations. This might be due to the masking or modifying effect of environment, which in turn modified the expression of characters and reduced the phenotypic effect (Solanki and Gupta, 2001).

Very close values of genotypic and phenotypic correlations such as days to 50% flowering with plant height and oil yield (kg/ha); Head diameter with seed yield/plant and 100 seed weight, 100 kernel weight, volume weight; Seed yield (kg/ha) with number of filled seed/head and grain filling%, were observed which might be due to reduction in error (Environmental) variance to minor proportions as reported by Dewey and Lu (1959). Wide differences between genotypic and phenotypic correlation such as, days to 50% flowering with grain filling, plant height with grain filling, head diameter with hull content, seed yield (kg/ha) with oil %, number of filled seed with 100 seed weight and oil % is due to duel nature of phenotypic correlation, which is estimated by genotypic and environmental correlation, and heritability of the characters (Falconer 1960).

Oil yield was highly significant and positively correlated with seed yield (kg/ha) (0.95), number of filled grain/head (0.85), grain filling% (0.65), days to 50% flowering (0.417), plant height (0.33) (Table 4). Results were in agreement with Chikkadevaiah *et al.* (2002). Seed yield (kg/ha) was significant and correlated positively with plant height (0.33), and head diameter (0.30). This result agrees with the research findings of Habib *et al.* (2007), Anandhan *et al.* (2010), Patil (2011), Hamdalla and Alik (2011) and Hejazi-Dehaghani *et al.* (2012).

Association among some yield components like days to 50% flowering with plant height (0.31), plant height with head diameter (0.93), seed yield per plant (0.33), 100 seed weight (0.41), volume weight with oil content (0.68) had positive and significant correlation. The results were in agreement with Vidhyavathi *et al.* (2005) for days to 50% flowering with plant height, Lakshminarayana *et al.* (2004) and Vidhyavathi *et al.* (2005) for plant height with head diameter, seed yield/plant, Vidhyavathi *et al.* (2005) and Manivannan *et al.* (2005) for volume weight with oil content.

Table 4: Genotypic and phenotypic correlation coefficients among yield attributing traits

Traits		Plant height (cm)	Head dia-meter (cm)	Seed yield (kg/ha)	No. of filled seeds/ head	Auto-gamy (seed filling) %	100 seed wt. (g)	100 Kernel wt. (g)	Hull content %	Vol. wt. (g/100 cc) (g)	Oil %	Oil yield (kg/ha)	Seed yield (g)
Days 50%	G	0.319**	−0.009	0.229	0.09	−0.544**	0.112	0.137	0.045	0.124	−0.085	0.417**	0.345**
Flowering	P	0.351**	−0.006	0.23	0.091	−0.719**	−0.112	0.137	0.024	0.124	−0.08	0.416**	0.345**
Plant height	G	1	0.935**	0.334**	−0.111	−0.507**	0.417**	0.572**	0.569**	0.384**	0.059	0.333*	0.335**
(cm)	P	1	0.929**	0.347**	−0.108	−0.355**	0.415**	0.568**	−0.525**	0.385**	0.355	0.415**	0.568**
Head	G		1.02	0.307**	−0.257	−1.131**	0.483**	0.598**	−0.850**	0.558**	0.141	0.144	0.145
Diameter (cm)	P		1.02	0.365**	−0.254	−0.699**	0.481**	0.594**	−0.464*	0.556**	0.144	0.148	0.148
Seed yield	G			0.98	0.672**	0.409**	−0.097	−0.092	−0.033	−0.058	−0.405**	0.937**	0.953**
(kg/ha)	P			0.99	0.671**	0.301*	−0.095	−0.089	−0.018	−0.458**	0.392**	0.952**	0.961**
No. of filled	G				1.02	−1.297**	−0.780**	−0.721**	0.204	−0.412**	−0.419**	0.853**	0.715**
seeds/head	P				1.05	−0.865**	−0.782**	−0.714**	0.107	−0.410**	−0.407**	0.849**	0.786**
Autogamy	G					1	−0.975**	−1.064**	0.189	0.694**	0.525**	0.651**	−0.819**
(seed filling) %	P					1	−0.650**	−0.886**	0.042	0.489**	−0.304	0.508**	−0.690**
100 Seed	G						1.02	0.952**	−0.407	0.625**	0.354**	−0.236	−0.221
weight (g)	P						1.08	0.947**	−0.219	0.618**	0.349**	−0.323	−0.218
100 Kernel	G							1.05	−0.696	0.791**	0.510**	−0.183	−0.223
weight (g)	P							1.06	0.423	0.778**	0.504**	−0.176	−0.218
Hull content	G								1	−0.312	−0.286	−0.084	−0.304*
%	P								1	−0.331*	−0.379**	−0.114	0.040

Traits		Plant height (cm)	Head dia-meter (cm)	Seed yield (kg/ha)	No. of filled seeds/ head	Auto-gamy (seed filling) %	100 seed wt. (g)	100 Kernel wt. (g)	Hull content %	Vol. wt. (g/100 cc) (g)	Oil %	Oil yield (kg/ha)	Seed yield (g)
Vol. Weight	G									1	0.551**	−0.370**	0.253
(g/100 cc)	P									1	0.669**	−0.078	0.301*
Oil %	G										1	−0.237	−0.382*
	P										1	−0.345*	−0.536**
Oil yield	G											1	0.842**
(kg/ha)	P											1	0.926**

Note: *, **: Significant at *p* = 0.05 and 0.01, respectively.

Significant negative correlation in this experiment were observed for seed yield with oil percentage (–0.40) at genotypic level indicating negative influence of these characters in increasing seed yield, i.e., in case of sunflower, high seed yielder genotypes are not high oil yielder. Significant negative correlation in this experiment were observed at both genotypic and phenotypic level for seed yield with days to 50% flowering to seed filling (–0.54), number of filled grain with oil percentage (–0.41), plant height with hull content (–0.56) and head diameter with hull content (–0.85). Probably such type of negative association may arise from developmentally induced relationship (Adams, 1967). Negative correlation may also arise due to competition between different traits for development by using a common factor, possibly limited nutrient supply. Component compensation of parents allows on opportunity to have reasonable compromise and balance between one or two components resulting high yield. The optimal genetic level for each component would differ depending on the type of the environment encountered (Grafius, 1965). Pleiotropy and/or linkage may also be genetic causes for this type of negative association. The pleiotropic that affect both characters in the desire direction will be strongly acted upon by selection and rapidly brought towards fixation.

The non-significant association between days to 50% flowering and oil content but significant association seed yield per plant (0.34) and oil yield (kg/ha) (0.41) suggested that a breeder can evolve high oil yielding hybrids/varieties in early or late maturity group.

Correlation analysis revealed that among the characters plant height, head diameter, volume weight/100 ml, 100-seed weight, oil content and seed yield per plant would increase oil yield in the present sunflower population.

Path analysis was carried out for considering seed yield (kg/ha) as effect of nine characters as cause. Overlapping characters, viz., the 100 kernel weight, seed yield/plant (g), oil yield (kg/ha) were excluded from the analysis. Shrivastava and Sharma (1976) suggested that only direct yield components should be used for path analysis. Genotypic and Phenotypic correlation coefficients were partitioned by using the method of path analysis has been presented in Table 5. The result indicated that filled grain/head had highest positive direct effect (1.060 and 1.102) followed by 100 seed weight (0.783 and 0.749), plant height (0.24 and 0.589). 100 seed weight (g) exhibited negative correlations with seed yield and its positive direct contributions were nullified by their negative indirect effects through number of filled grain/head, Head diameter and Vol. Weight/100 cc. Restricted simultaneous selection model could be adopted for the improvement of these characters to nullify undesirable indirect effects in order to make use of the positive direct effects (Singh and Kakar, 1997).

In case of phenotypic path coefficient, plant height has significant positive direct effect (0.589) and significant positive correlation with seed yield (0.565) indicated that plant height had really some role for influencing seed yield in sunflower in positive direction but the trait is highly influenced by the environment.

Numbers of filled grain/head had highest and significant positive direct effect (1.06) and significant positive correlation (0.715) with seed yield (kg/ha) indicated that number of filled grain/heads had really some role for seed yield improvement in sunflower. The finding also close agreed with the findings of Behradfar *et al.* (2009).

Triats		Days 50% flowering	Plants height (cm)	Head dia-meter (cm)	No. of filled seeds/ head	Auto-gamy (seed filling) %	100 Seed Wt. (g)	Hull content %	Vol. Weight (g/100 cc) %	Oil % (g)	Correlation with seed yield (kg/ha)
Days 50%	G	**–0.038**	0.003	0.002	0.095	0.174	0.087	–0.018	–0.010	0.049	**0.345****
Flowering	P	**–0.031**	0.070	0.002	0.100	0.130	0.083	0.003	–0.025	0.013	**0.348****
Plant height	G	–0.005	**0.024**	–0.208	–0.117	–0.010	0.326	0.390	–0.030	–0.034	**0.335****
(cm)	P	–0.004	**0.589**	–0.275	–0.119	–0.008	0.307	–0.067	–0.077	–0.010	**0.568****
Head diameter	G	0.000	0.022	**–0.222**	–0.273	0.023	0.378	0.342	–0.044	–0.081	**0.146**
(cm)	P	0.000	0.547	**–0.296**	–0.280	0.016	0.355	–0.060	–0.112	–0.024	**0.158**
No. of filled	G	–0.003	–0.003	0.057	**1.060**	0.026	–0.614	–0.082	0.032	0.242	**0.715****
seeds/head	P	–0.003	–0.064	0.075	**1.102**	0.020	–0.578	0.014	0.082	0.067	**0.786****
Autogamy	G	0.328	0.012	0.252	–1.375	**–0.020**	–0.763	–0.076	–0.054	0.303	**–0.819****
(Seed filling) %	P	0.177	0.209	0.207	–0.954	**–0.023**	–0.481	0.005	–0.098	0.050	**–0.690****
100 Seed weight	G	–0.004	0.010	–0.107	–0.832	0.020	**0.783**	0.164	–0.049	–0.204	**–0.221**
(g)	P	–0.003	0.245	–0.142	–0.861	0.015	**0.749**	–0.028	–0.124	–0.057	**–0.207**
Hull content	G	–0.004	–0.010	0.107	0.832	–0.020	–0.783	**–0.164**	0.049	0.204	**–0.304***
%	P	–0.001	–0.309	0.137	0.118	–0.063	–0.001	**0.162**	0.128	0.066	**0.301***
Vol. weight	G	–0.005	0.009	0.124	0.437	0.014	0.489	0.248	**0.078**	0.392	**–0.553****
(g/100 cc)	P	–0.004	0.227	–0.165	–0.452	–0.011	0.457	–0.042	**–0.201**	–0.110	**–0.536****
Oil	G	0.003	0.001	–0.031	–0.444	0.011	0.277	0.260	–0.053	**–0.577**	**–0.237**
%	P	0.002	0.036	–0.043	–0.449	0.007	0.258	–0.049	–0.134	**–0.165**	**–0.182**

Table 5: Genotypic and phenotypic path coefficients among yield attributing traits

Note: *, **: Significant at p = 0.05 and 0.01, respectively; Bold figures indicates direct effects; Genotypic path residual = 0.1121; and Phenotypic path residual = 0.0546.

100 seed weight had significant positive direct effect (0.783) and negative correlation (–0.221) with seed yield (kg/ha) indicated that 100 seed weight had influencing seed yield in positive way but it's direct effect was nullified by cumulative negative effects through head diameter (–0.107), number of filled grain /head (–0.832) and oil content% (–0.204) which resulted in significant negative correlation with seed yield (kg/ha).

In the present investigation it has been observed that the characters like head diameter had negative direct effect on seed yield (–0.222), but this characters had positive indirect effect via 100 seed weight (0.378) and hull content (0.342) indicating that a restricted selection method is to be imposed so that emphasis should be given on those characters (having positive indirect effect) for overall improvement of seed yield in sunflower.

Direct contribution of number of filled grains per head, 1000 seed weight on seed yield was reported by Kalukhe *et al.* (2010), Sowmya *et al.* (2010) and Yasin and Singh (2010). Direct contribution of plant height, head diameter, 100 seed weight towards enhancing seed yield was observed by Hamdalla and Alik (2011) and Patil (2011).

Low value of residual variation (0.112) and (0.0546) for the Phenotypic path analysis and genotypic path analysis respectively explained that 89% and 99.95% of the variability in seed yield as well as oil yield was contributed by the above mentioned nine characters and very small variability was controlled by other factors not included in our present experiment.

The result of path analysis indicating that selection for plant with more no. of filled grain/head, moderate plant height, late flowering, head diameter with restricted and test weight are important which will help to improve seed yield as well oil yield in the population (sunflower) under study.

3.4. Genetic diversity

D^2 analysis was carried out separately for 13 yield attributing characters from the pooled data over two seasons and grouping of the 32 genotypes in different clusters was done accordingly. ANOVA showed significant difference among genotypes for all the characters. Wilk's Lambda criterion ($\lambda = 0.1714 \times 10^{-25}$ and V = 4182.65, with 403 df) revealed significant differences among the genotypes for the pooled effect of all the characters.

Mahalanobis generalized distance estimated by D^2 statistic (Rao, 1952) is a distinct tool for discriminating populations considering a set of parameters together rather than inferring from indices based upon morphological similarities, eco-geographical diversity and phylogenetic relationships. The more diverse the parents, within overall limits of fitness, the greater are the chances of obtaining higher amount of heterotic expression in the F_1 and broad spectrum of variability in segregating generations (Anand and Murty, 1968). The objective of this work is to group a set of thirty two genotypes of sunflower into different clusters according to the genetic divergence and such grouping would help to initiate crossing programme between diverse groups to develop good F_1.

Based on relative magnitude of D2 values, the genotypes were grouped into eight clusters (Table 6, Figure 1). Cluster-I comprised maximum number of 10 genotypes representing CMS-16A, CMS-850A, EC-602060, EC-601718, EC-601718, Ec-512682, R-138-2, R-630, R-104, R-12-96 Genotypes in this cluster were mostly tester or Restorer lines (Exotic Collections) Cluster-II comprised 17 genotypes representing CMS-852A, CMS-103A, CMS-107A, P-89-1A, CMS-207A, CMS-302A, CMS-10A, and EC-623016, EC-601725, EC-623021, EC-601878, EC-601751, EC-623023, EC-623029, EC-601858, R-6D-1, R-1-1.

Table 6: Clustering of 32 sunflower genotypes according to quantitative traits		
Clusters	**Number of genotypes**	**Genotypes**
I	9	CMS-16A, CMS-850AEC-602060, EC-601718, EC-512682, R-138-2, R-630, R-104, R-12-96
II	17	CMS-852A,CMS-103A, CMS-107A, P-89-1A,CMS-207A, CMS-302A, CMS-10A, EC-623016, EC-623021, EC-601725, EC-601978,EC-601751,EC-623023,EC-623029, EC-601958, RHA-6D-1, R-1-1
III	1	R-107
IV	1	EC-623027(M)
V	1	CMS-853A
VI	1	EC-623011
VII	1	P-2-7-1A
VIII	1	R-341

Cluster-III, Cluster-IV, Cluster-V, Cluster-VI, Cluster-VII, Cluster-VIII are solitary and was represented by R-107, EC-623027(M), CMS-853A, EC-623011, P-2-7-1A and R-341 respectively. The findings also supported by Reddy *et al.* (2012) and Ram *et al.* (2018). Binodh (2007) also reported importance of diversity study in his study with restorer lines for heterosis study in sunflower and reported the restorer lines distributed in different cluster will be helpful for development of new sunflower hybrids.

Distribution pattern of genotypes in different cluster stipulated that genetic divergence was not exclusively interconnected to geographical differentiation. Many genotypes of geographic closeness cut down into different clusters (Cluster-I, III, V and VII) and vice-versa (Table 6). Tendency to form such type of clustering ignoring

the geographical borderlines exhibited the topographical isolation was not the only factor contributing to diversity in natural population. Clustering of genotypes from different eco-geographic position into single cluster (Cluster I and II) could be allocated to the possibly of free interchange of breeding materials. However, one-sided selection, practiced for a particular trait or a group of linked traits (e.g., male sterility or fertility restoration) in several places may produce similar phenotype, which can clump into one cluster ignoring of their geographic sector (Singh and Gupta, 1968), as noticed in Cluster-I and II. Genesis of clusters among the genotypes constructed on usual geographic genesis may be due to their parentage, developmental traits, past history of selection and different out-crossing rates (Arnold et al., 1996). Therefore, the kind of genetic diversity build among the genotypes belonging to same geographic dawning might be due to contrast in adoption, selection criteria, selection pressure and environmental conditions (Vivekananda and Subramanian, 1993). Unaccompanied clusters may be created due to total isolation preventing the gene flow or rigorous natural/ human selection for multiple adaptive networks.

The statistical distance indicates the index of genetic diversity among the clusters. Estimates of average intra- and inter-cluster distances among the eight clusters (Table 7) showed that Clusters III, IV, V, VI, VII and VIII being solitary clusters, had the lowest intra-cluster distance of '0', while Cluster I had $D^2 = 71.98$ and Cluster II had $D^2 = 75.98$. The highest inter cluster distance 2175.5 was noticed between cluster VI and VII followed by Clusters V and VI (1649.6), Clusters IV and VI (1642.8), Clusters I and VII (1277.5), and so on. This result performed considerable amount of divergence within and between clusters. Clusters VI and VII played important role in inter-cluster distance. Similar studied based on D^2 statistics was also performed by Shamshad et al. (2014), Neelima et al. (2016) and Ram et al. (2018).

Table 7: Average intra and inter cluster divergence for quantitative traits								
Clusters	**I**	**II**	**III**	**IV**	**V**	**VI**	**VII**	**VIII**
I	71.98	436.83	123.02	931.02	904.97	179.60	1277.59	154.73
II		75.92	154.93	180.85	157.33	962.74	328.71	424.90
III			0.00	463.56	442.43	454.27	695.02	129.96
IV				0.00	42.33	1642.86	166.49	843.65
V					0.00	1649.62	90.91	880.85
VI						0.00	2175.49	333.91
VII							0.00	1137.02
VIII								0.00

Crossing between genotypes separated by considerable statistical distance would be logical to produce superior hybrids in the F_1 generation and promising recombinants in segregating generations.

The mean performance of different clusters in respect of 13 characters is presented in Table 8.

Cluster I: Highest mean value of seed yield/ plant and number of filled grains per head; higher mean value of days to 50% flowering, plant height, and lower mean value of 100 seed weight was observed. This cluster is consisting of both CMS line and tester.

Cluster II: This cluster had lower mean value of days to 50% flowering and moderate value of plant height. This cluster was characterized by high value of oil content and 100 seed weight.

Cluster III: This cluster exhibited higher mean value of seed yield/plant and number of filled grains per head; days to 50% flowering and plant height.

Cluster IV: It included mean value for most of the characters including plant height.

Cluster V: All the characters are moderate except high value for seed yield.

Cluster VI: This cluster had moderate mean value for days to 50% flowering, plant height and seed yield and 100 seed weight.

Table 8: Cluster mean and percentage contribution of 13 quantitative traits

Clusters	Days to 50% Flowering	Plant height (cm)	Head diameter (cm)	Seed yield (kg/ha)	No. of filled seed/ hd.	Autogamy (seed filling) %	100 seed wt (g)	100 Kernel wt. %	Hull content %	Vol. wt. (g/100 cc)	Oil %	Oil yield (kg/ha)	Seed yield/ plant
I	63.27	88.23	7.77	801.19	399.79	81.11	3.61	2.42	33.41	35.80	35.73	318.24	15.56
II	62.71	97.39	9.00	712.58	276.50	80.26	4.93	3.34	31.64	39.11	37.04	264.67	13.23
III	67.15	91.30	8.75	799.25	344.60	83.25	4.21	2.73	35.00	35.25	34.30	311.85	16.18
IV	69.98	103.70	9.90	816.00	297.25	75.00	5.68	4.05	28.35	43.58	37.73	307.88	14.85
V	64.63	115.15	10.35	961.25	305.75	83.30	5.70	4.00	30.20	42.68	35.75	343.93	17.48
VI	61.88	73.38	7.93	849.75	529.50	79.90	2.91	2.02	32.08	33.70	35.90	305.25	15.45
VII	63.13	93.25	8.40	843.83	251.60	81.40	6.10	3.63	36.15	35.43	33.90	286.75	15.35
VIII	65.85	11.40	3.70	717.25	348.50	80.90	3.75	1.97	59.48	34.65	33.70	242.00	13.04
Relative contributions (%)	**7.60**	**10.42**	**12.56**	**9.16**	**14.52**	**4.58**	**3.62**	**6.76**	**4.25**	**7.84**	**5.32**	**5.25**	**8.12**

Cluster VII: It exhibited highest mean value for days to 50% flowering, plant height and seed yield but lowest seed filling% and lowest no. of filled grain/head.

Cluster VIII: This cluster constituted highest mean value for days to 50% flowering, plant height and seed yield.

None of the clusters exhibited genotypes with highest values for all characters. Ramesh *et al.* (2013) noticed that based on the inter cluster distance and cluster mean for various character, potential parents were distinguished from different clusters for hybridization program. The hybridization between individuals of various clusters would, therefore, be logical for recombining the superior characters for the development of elite sunflower hybrids(s).

Relative contributions of each of the 13 yield attributing characters (Table 8) revealed that number of filled grain/head (14.52%) had the highest contribution, followed by head diameter (12.56%) and plant height (10.42%), seed yield (kg/ha) (9.16%), seed yield/plant (8.12%) and volume weight (7.84%) towards the observed genetic diversity. Contribution of 100 seed weight (3.62%) and hull content (4.25%) were minimum. It is evident that none of the 13 yield attributing traits contributed very high or low compared to others to induce observed genetic diversity. Similar studied was also observed by Shamshad *et al.* (2014) Neelima *et al.* (2016) and Ram *et al.* (2018). Loganathan *et al.* (2006) reported that seed yield/plant contributed highest value towards genetic divergence followed by 100 seed weight and plant height.

4. Conclusion

The presence of high levels of variations among the traits such as number of filled grain/head, volume weight/ 100 ml, 100-seed weight, oil content and seed yield/plant were observed in this experiment. Traits with less environmental influence, high heritability and genetic advance in addition to having significant bearing on seed yield were identified as the best gain traits which would help in transferring these traits by cross breeding followed by recurrent selection. The study also helped to identify the potent genotypes among the population of diverse genotypes of sunflower that can serve as parents with desirable traits for further breeding purposes. Based on genetic divergence, importance of traits in determining the seed yield in this population and performance *per se* of the genotypes as well as cluster means, crossing between CMS 16A X EC 602060, CMS 16A X R-12-96, CMS 850A X EC 512682, CMS 852A X EC 623023, CMS 852A X EC 623021, CMS 852A X EC

601751, CMS 852A X EC 601978, CMS-302A X EC-623029, CMS-302A X EC-623016, P-89-1A X R-6D-1, P-89-1A X EC 601958 (intra-cluster) and CMS-853A X EC-623027(M),P-2-7-1A X EC-601958, P-2-7-1A X EC-512682, P-2-7-1A X R-12-96, CMS-207 A X EC-512682, CMS-302A X EC-623011 CMS-850A X R-1-1, P-2-7-1A X R-6D-1, CMS-16A X R-341, P-89-1A X R-104 and CMS-302A X R-12-96 (inter-cluster) are most likely to yield a considerable amount of heterosis in hybrid for yield attributing traits in sunflower and to provide a wide opportunity for selecting the newly developed desirable hybrids in near future.

Acknowledgment

Conflicts of interest

The authors declare no conflict of interest.

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