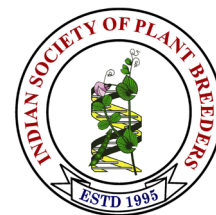


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## Research Note

### Discerning genetic diversity among sunflower germplasm accessions through multivariate analysis

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#### Abstract

The objective of the present investigation was to evaluate 135 sunflower germplasm accessions through Principal Component Analysis and Hierarchical cluster analysis to assess the extent of genetic diversity. The principal components with eigen values more than one *i.e.*, PC1 and PC2 describes 38.3% and 21.24% respectively with a cumulative effect of 59.62% of the total variation. The biplot analysis revealed positive association of seed yield per plant for 100 seed weight, head diameter, days to 50% flowering and it was negatively associated with oil content. By using Wards method of clustering, 135 sunflower accessions were aggregated in 12 clusters based on different traits in which cluster VIII includes 30 accessions and is the largest one. The accessions in cluster IV, I and VI had maximum values for 100 seed weight, seed yield per plant and oil content, respectively. The germplasm accessions 213R, 121R, RHA-GPR-110, CMS-107B, IB-11-3, SCG-107, 132R, NGM-15 and OPH-123 were identified to be potential accessions and these can be used for improvement of yield and its attributing traits in sunflower.

**Keywords:** Sunflower, Genetic diversity, Principal component analysis, Cluster analysis

Sunflower (*Helianthus annuus* L) is an important oil seed crop along with groundnut, mustard and soybean in the world. It is an annual crop belonging to Asteraceae family whose first distribution is reported in Central America (Ahmadpour *et al.*, 2022). It is a highly cross-pollinated crop with chromosome number ( $2n=34$ ) with high yield potential and is adapted to a variety of environmental conditions. The crop is majorly cultivated for edible oil, animal feed, ornamental, biodiesel production and medicinal value. Seeds are rich sources of proteins, vitamin E, minerals (31% selenium, 25% copper) and phytochemicals (Dutta and Sudhir, 2003). In world, sunflower crop contributes about 14% to 16 % of vegetable oils, 7% of the oilcake and meal produced from oil seeds. In India, the sunflower crop is occupied in an area of 226 lakh ha with a production of 2.28 lakh tonnes

and productivity of 1011 Kg/ha. (Directorate of Economics and Statistics, Govt. of India, 2022-23).

The sunflower cultivated area is reduced drastically in recent years. One of the reasons for such decline is the lack of good quality hybrids in sunflower. There is a lot of scope for exploring genetic diversity in sunflower to identify promising accessions for utilization in hybrid development in sunflower. Principal component analysis (PCA) is one of the multivariate techniques that analyses data and converts it into a series of new orthogonal variables called principal components by linearly combining the variables that account for majority of variance in the original variables (Abdi and Williams, 2010). The cluster analysis is pertinent method for concluding relationship among accessions and amount of genetic distance from

each other (Mellingers, 1972). Divergence analysis using PCA and hierarchical cluster analysis has been shown to be effective in determining potential accessions useful for hybridization (Chaudhary *et al.*, 2015). The prime objective of this investigation was to trace genetic diversity and identify the best divergent accessions for their inclusion in sunflower crop improvement programme.

The current investigation was conducted with 135 sunflower accessions at Regional Agricultural Research Station (RARS), Nandyal, Andhra Pradesh, India during *Rabi*, 2022-23. All genotypes were evaluated in Augmented block design with plot size of 5.0 × 1.8 m<sup>2</sup> per genotype with spacing of 60 × 30 cm<sup>2</sup>. All the agronomic practices recommended by Acharya N.G. Ranga Agricultural University were carried to raise a healthy crop. The data was collected from five randomly selected plants for yield and yield associated traits *viz.*, Plant height (PH), Head Diameter (HD), 100 Seed weight (HSW), Seed yield plant<sup>-1</sup> (SYP) and Oil Content (OC) while for days to 50% flowering data was recorded on plot basis. To assess genetic diversity among accessions, PCA (Hotelling, 1936) and cluster analysis were used to identify the most contributing traits for variation and diversity among accessions, respectively (Peeters and Martinelli, 1989). The data collected from experimental material was subjected statistical analysis to determine genetic diversity through PCA and hierarchical cluster analysis through JMP 17.0 statistical software (SAS Institute Inc., Cary, NC, USA). PCA for yield and yield associated traits resulted in development of biplot based on principal components. In hierarchical cluster analysis, constellation plot was developed using Ward's method.

The descriptive statistics of six measured traits among 135 studied accessions is summarized in **Table 1**. The accessions EC-601624 (50 days), P-147R and IC-502013 (51 days) were observed to be early flowering, while OPH-

86 was late flowering (65 days). The plant height ranged from 53 cm (RHA-GPR-118) to 150 cm (213 R) with an average of 94.27 cm. The mean value of head diameter was found to be 9.13 cm with range from 3 cm (CMS-107 B) to 16 cm (213 R). The 100 seed weight ranged from 3.8 g (121 R) to 9.2 g (OPH 137) with mean value of 7.55 g. The mean seed yield per plant was recorded as 17.34 g with range from 5.24 g (CMS 107B) to 30.35 g (RHA GPR-3 and 213 R) among the studies accessions. The oil content ranged from 29.6% (GP-6-116) to 38.49% (OPH-74) with average value of 35.2%. The study showed that the genotype, 213 R recorded highest means for plant height, head diameter and seed yield per plant. The presence of significant variability among the accessions is the key for success in crop improvement programme. Further, the presence of variability for head diameter, seed yield and oil content provides the better scope for improvement through simple selection of inbreds in sunflower (Varalakshmi *et al.*, 2020). The availability of diverse inbreds enables the development of potential heterotic hybrid combinations in sunflower (Reavanth *et al.*, 2021).

The PCA is regarded as an authentic tool for efficacious selection of accessions for utilization in crop improvement programme. The results of PCA revealed the significance of first two principal components in discriminating sunflower germplasm accessions. The first two PCs, PC1 and PC2 exhibited eigen value greater than one explaining 59.62% of total variation. The eigen values and total cumulative percentage of variances explained by PCs is furnished in **Table 2**. The first PC displayed high positive weight to plant height (0.524), head diameter (0.486) and also to seed yield per plant (0.445). Similarly, second, third and fourth PCs recorded highest positive loading to oil content (%). Likewise, fifth and sixth PCs gave positive loading to 100 seed weight (0.648) and head diameter (0.562), respectively (**Table 3**). Out of five PCs, the first PC is

**Table 1. Descriptive statistics of the measured characters in sunflower accessions**

S. No.	Characters	Mean	Maximum	Minimum	SD	SE
1	Days to 50% flowering	57.26	65	50	3.39	0.29
2	Plant height (cm)	94.27	150	53	17.92	1.55
3	Head diameter (cm)	9.13	16	3	2.34	0.20
4	100 seed weight (g)	7.55	9.2	3.8	1.10	0.10
5	Seed yield / plant (g)	17.34	30.35	5.24	5.80	0.50
6	Oil content (%)	35.20	38.49	29.60	2.30	0.20

**Table 2. Total variances explained by different principal components in sunflower accessions**

Components	PC1	PC2	PC3	PC4	PC5	PC6
Eigen values	2.303	1.274	0.842	0.798	0.501	0.281
Proportion variance %	38.38	21.24	14.04	13.30	8.36	4.68
Cumulative variance %	38.38	59.62	73.66	86.96	95.32	100.00

**Table 3. Factor loading of different characters with respect to different principal factor in sunflower accessions**

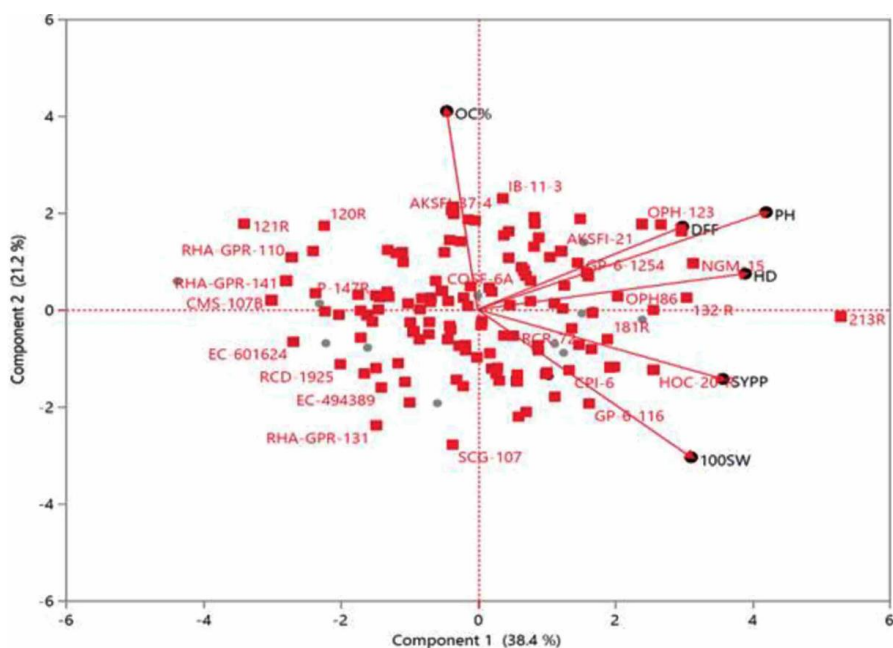
Principal components	PC1	PC2	PC3	PC4	PC5	PC6
DFF	0.372	0.287	-0.716	0.325	-0.266	0.302
PH (cm)	0.524	0.337	-0.094	-0.228	0.302	-0.679
HD (cm)	0.486	0.124	0.298	-0.583	-0.066	0.562
100 SW (g)	0.388	-0.510	0.006	0.352	0.648	0.215
SY/P (g)	0.445	-0.240	0.434	0.3872	-0.594	-0.230
OC %	-0.056	0.686	0.448	0.479	0.249	0.185

strongly correlated with Plant height, head diameter and seed yield per plant. Harshavardhan and Shobha (2022) assessed the variability of nine plant traits for growth of sunflower and reduced the dimensionality to four principal components, which extracted about 83% of variance in the original data. The obtained results in the study were in confirmation with Sasikala *et al.* (2022) suggesting that selection of principal components with high scores would be rewarding for sunflower crop improvement programme.

The biplot depicted the relationship of 135 sunflower accessions for six traits (Fig. 1). From the biplots, the six yield and yield attributing traits were differentiated into three groups. Seed yield per plant and 100 seed weight were grouped in same cluster. Days to 50% flowering, plant height and head diameter were grouped in single cluster. Whereas, oil content alone grouped as one cluster. The selection of accessions with highest score

in PC1 will be desirable for developing high seed yield types in sunflower. The study showed that the accessions viz., 213R, 121R, RHA-GPR-110, CMS-107B, IB-11-3, SCG-107, 132R, NGM-15 and OPH-123 were located at extreme ends of distinct quadrants of the plot. Hence, these genotypes were recognized as highly divergent and found to be potential for exploitation in hybridization programme to enhance heterotic potential in sunflower crop. These results are in congruence with findings of Ahmad *et al.* (2021) in sunflower. This showed that PCA is a reliable method in identifying few key traits contributing to the largest variation and could be a reliable method in predicting the important traits influencing clustering of different cultivars.

Hierarchical cluster analysis was conducted with 135 sunflower germplasm accessions using Wards method which provides the best result to get the finest

**Fig. 1. Biplot comprising of 135 sunflower accessions studied for six traits**

DFF-Days to 50% flowering, PH-Plant height (cm), HD- Head Diameter (cm), 100 SW-100 seed weight (g), SY/P- Seed yield per plant (g), OC%- Oil content %

Table 4. Grouping of different accessions into different clusters

Cluster	Number of accessions	Accessions
I	19	RCR-72, EC-198072, RHA-272-1, OPH-150, GMU-324, NDI-7, CPI-6, OPH-137, GMU-1032, RCB-19-40, RCR-19-25, RHA-GPR-3, RCB-19-25, 160R, SCG-107, EC-276583, IC-502013, SS-2038(C), GMU-450.
II	11	GMU-258, IC-502017, HOC-20-R, RHA-GPR-118, EC-198078, GMU-734, EC-93606, GPR-78, AKSFI-21, EC-178168, 107R.
III	9	GP-6-116, OPH-67, GMU-428, EC-444424, RCR-73, GMU-461, NDSI-8, RHA-378, GMU-488.
IV	10	CSFI-99, NGM-15, 151R, OPH-123, GMU-780, GMU-689, 132R, OPH-86, 181R, 213R.
V	8	COSF-6B, GMU-1031, GMU729, ARM-243A, IB-11-3, AKSFI-22, GMU-575, AKSFI-23.
VI	7	OPH-74, AKSFI-37-4, EC-512687, RHA-138-2R, GMU-713, 120R, CMS-911B.
VII	9	ARM-2508, GMU-502, GP-6-1254, COSF-7A, NDI-6, AKSFI-51-6-3, EC-6023072, AKSF 14-1R, AKSFI-174.
VIII	30	RCR-19-12, GMU-266, JD-32, GMU-1108, GMU-118, GKVK-3, RHA-GPR-37, CPI-2, EC-399272, MRHA-2, COSF-7B, EC-494377, IC-75646, CSFI-5315, RCR-72, COSF-1A, COSF-6A, CMS-234A, TSG-298R, CMS-607B, EC-399969, RHA-GPR-54, NDSI-15, GMU-520, NDCMS-2B, NO-27, GMU-1000, GMU-510, CMS-338-1B, P-122-R.
IX	9	OPH-67, RHA-GPR-131, COSF-12A, SCG-107, EC-279902, RHA-1232, CMS-898, EC-494389, P-70R, RCD-1925, GMU-1032.
X	6	CMS-265A, RHA-6D-1, GMU-896, CSFI-99, REC-428, EC-601624.
XI	10	121R, RHA-GPR-110, EC-601616, RHA-GPR-118, R83BR, NGM-21, GMU-500, GP-53-18, RHA-GPR-141, CMS107B.
XII	7	CSFI-99, P-148R, P-147R, RHA-850, GP6-374, RHA-GPR-72, GMU-258.

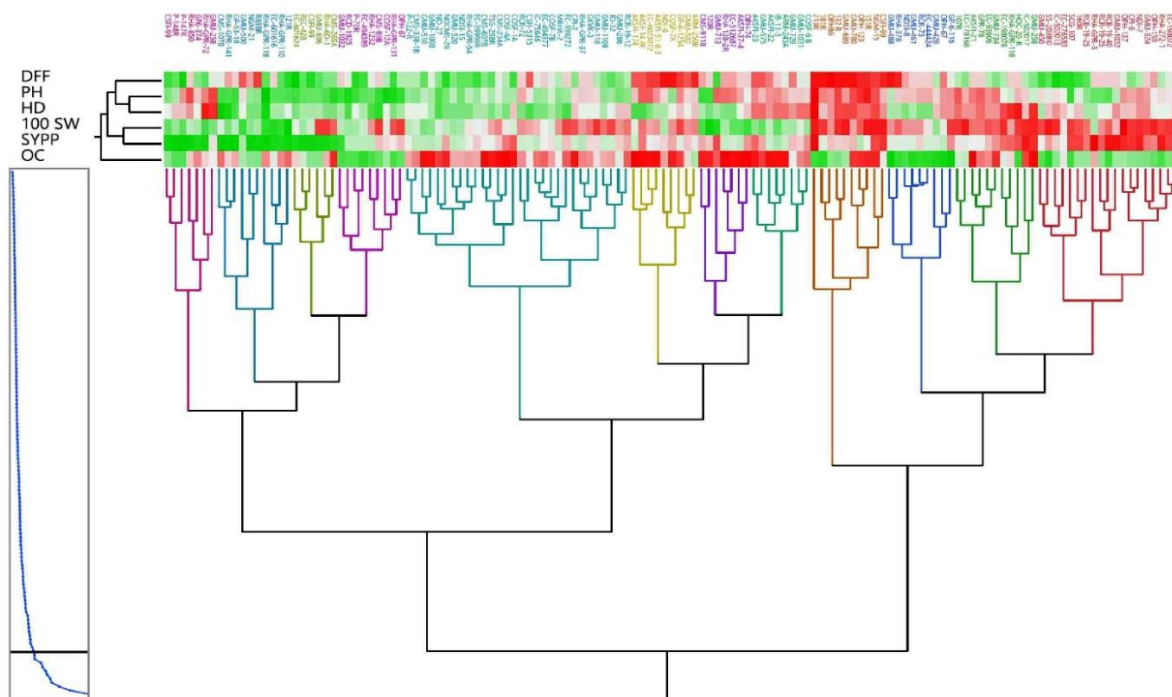


Fig. 2. Dendrogram showing clustering by Wards method

DFF-Days to 50% flowering, PH-Plant height (cm), HD- Head Diameter (cm), 100 SW-100 seed weight (g), SY/P- Seed yield per plant (g), OC%- Oil content %

possible classification. The cluster analysis revealed the aggregation of accessions into 12 clusters (Table 4 and Fig. 2). The cluster means computed for six major yield attributing characters revealed the existence of ample amount of variation among the clusters (Table 5). The highest and lowest cluster means were recorded for the traits plant height (123.10 cm) and 100 seed weight (5.52g), respectively. Maximum cluster mean was noticed in cluster IV (43.85) followed by cluster II (40.00). The

least cluster mean was displayed by cluster X (30.75). This clearly infers the existence of sufficient genetic divergence in the accessions of these clusters. Further, among the 12 divergent clusters, the highest numbers of accessions were grouped in cluster VIII with 30 accessions followed by 19 in cluster I. The accessions in cluster IV showed highest mean values for days to 50% flowering, plant height, head diameter and 100 seed weight. The accessions of cluster I and cluster VI showed

Table 5. Cluster means of various characters of sunflower hybrids under study

Cluster No.	DFF	PH	HD	100 SW	SY/P	OC %	Mean
Cluster I	55.58	92.58	9.58	8.38	<b>24.09</b>	33.29	37.25
Cluster II	56.45	105.73	12.18	8.48	21.24	35.92	40.00
Cluster III	59.22	102.11	8.56	7.84	17.38	<b>31.00</b>	37.69
Cluster IV	<b>63.60</b>	<b>123.10</b>	<b>12.40</b>	<b>8.60</b>	20.53	34.92	<b>43.85</b>
Cluster V	58.88	105.00	11.75	6.63	18.66	37.19	39.68
Cluster VI	57.71	98.57	8.00	<b>5.52</b>	18.91	<b>37.96</b>	37.77
Cluster VII	62.22	104.78	8.11	7.88	18.36	37.04	39.73
Cluster VIII	55.77	87.70	7.96	7.66	17.50	36.64	35.53
Cluster IX	55.56	<b>68.00</b>	7.33	7.63	16.28	32.52	31.22
Cluster X	<b>53.33</b>	73.33	7.50	7.82	7.46	35.11	<b>30.75</b>
Cluster XI	57.40	75.50	<b>6.50</b>	5.84	<b>7.35</b>	35.30	31.31
Cluster XII	54.57	106.29	11.14	6.30	8.14	34.44	36.81
Mean values	57.52	95.22	9.25	7.38	16.32	35.11	

Bold figures indicate maximum and minimum values in each character. DFF-days to 50% flowering, PH-plant height, HD-head diameter, 100 SW-100 seed weight, SY/P-seed yield per plant, OC%-Oil content

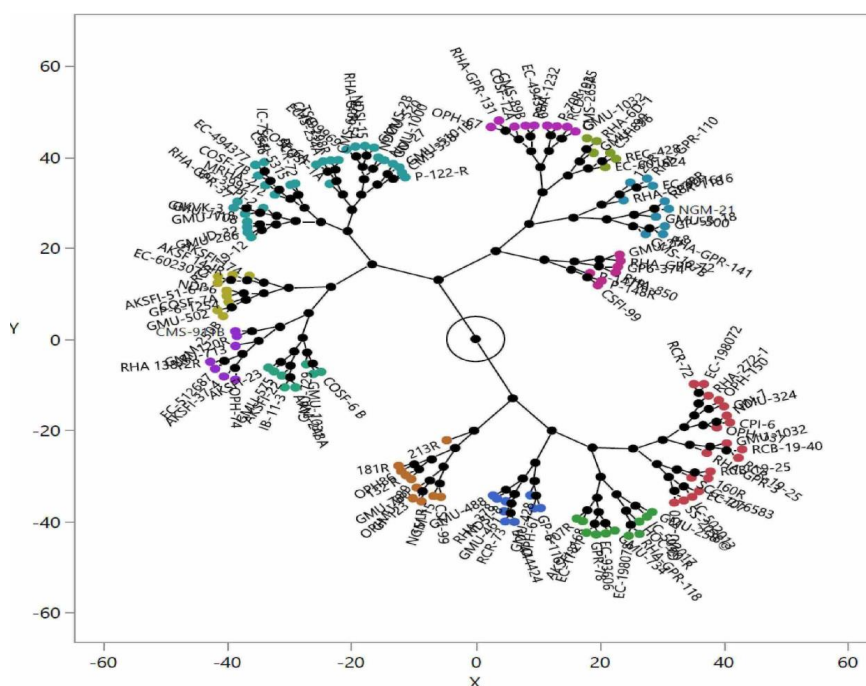


Fig.3. Constellation plot of 135 Sunflower accessions in to 12 clusters based on Euclidean distances



maximum value for seed yield per plant and oil content, respectively. The reported results are in agreement with the earlier findings of (Dudhe *et al.*, 2019) in sunflower. The hierarchical cluster analysis showed the presence of greater genetic distance among the sunflower accessions, which will uplift the hybridization programme in sunflower (Reddy *et al.*, 2012).

The relationship among the 135 sunflower accessions is depicted in constellation plot based on Wards method (Fig. 3). The accessions are grouped as end points and every cluster join as a new point with lines drawn will act as membership in constellation plot. The plot divided the total accessions into 12 clusters with membership of 19,11,9,10,8,7,9,30,9,6,10 and 7. The Clustering pattern divulged that majority of accessions congregated in cluster VIII (30), followed by Cluster I (19) and Cluster II (11). The accessions with longer line representing greater genetic distance between the clusters. Further, the accessions with maximum genetic distance are promoted in crossing programmes to develop sunflower hybrids with desirable combination of traits in them (Singh *et al.*, 2019).

PCA revealed that the first two principal components attributed 59.62% genetic variability for yield and yield attributing traits in sunflower accessions. The PC1 contributed the maximum towards diversity (38.38%) followed by PC2 (21.24%). The biplot analysis revealed the positive association of seed yield per plant with head diameter, 100 seed weight, plant height, days to 50% flowering. Whereas, negative association with oil content among 135 sunflower accessions. The cluster analysis of 135 sunflower accessions exhibited high genetic diversity, indicating a great chance for crop improvement by crossing accessions from other clusters. The accessions 213R, CMS-107B, 132R, NGM-15, RHA-GPR-3, OPH-74 and OPH-123 were identified as promising accessions and could be used for hybrid breeding programme in sunflower.

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