

RESEARCH ARTICLE

ASSESSING GENETIC DIVERSITY IN *CHRYSANTHEMUM MORIFOLIUM* GENOTYPES BASED ON DUS DESCRIPTORS: PLANT, LEAVES AND FLORAL TRAITS

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ABSTRACT

Genetic diversity is essential to explore novelties in flower and plant architecture to develop new cultivars. 50 genotypes of *Chrysanthemum morifolium* were systematically characterized for accurate genotype identification based on DUS descriptors in UPOV guidelines. The experiment was conducted for two consecutive years. Significant variation was observed for fourteen quantitative traits. Shannon equitability index was > 0.75 for 6 out of 11 qualitative traits indicating good diversity. The highest GCV (67.52%) and PVC (68.05%) were recorded for number of flowers per plant. High estimates of heritability and genetic advance as percentage of mean were shown by number of flowers per plant, stipule size and ray floret width suggesting effective selection. Principal component analysis (PCA) for both quantitative and qualitative traits performed independently, resulted in 4 principal components accounting for 74.7% and 63.27% of total variability respectively. Factor loading for quantitative traits revealed that leaf characters have major contribution to germplasm variability. Among **qualitative characters**, **ray florets** traits have major contribution to **germplasm variability**. The genotypes were classified into 4 groups at Euclidean distance of 88.94. Cluster 1 has only genotype Gauri which is an outlier for plant height and number of flowers per plants. Pusa Shwet is alone in cluster 2 having unique combination of large sized and semi double type flower head. Cluster 3 and cluster 4 have 28 and 20 genotypes respectively. Grouping based on Jaccard's similarity coefficient for binary data gave nine clusters at similarity coefficient of 0.33 with genotype Gauri alone constituting cluster 3.

KEYWORDS

Chrysanthemum morifolium, Genetic diversity, PCA, UPGMA, Euclidean distance, Jaccard's similarity index

1. INTRODUCTION

Globally chrysanthemum ranks second in trade after rose (Jaime et al., 2013). The species is cultivated for cut flower, loose flower, pot plant, landscaping, culinary, medicinal and extraction of pyrethrum. It is a complex allohexaploid that often exhibits aneuploidy with chromosome numbers varying from 47 to 67 (Roxas et al., 1995). The polyploidy affects the response of species under variable environments. In India, it is grown in an area of 23.93 kha with a total production of 470.16 kt during year 2021-22 (NHB, 2022). The market demands for novelties in plant architecture, type of flower and color, and yield. Such demands persuade breeders to alter these traits to suit the purpose.

Germplasm serves as reservoir of alleles and studying this diversity is extremely important for crop improvement and evolutionary studies. Chrysanthemum has enormous phenotypic diversity worldwide for flower shapes (single, double, anemone, incurve, and pompon) as a result of different combinations of floret number, petal size and floral organ fusion (Dai et al., 2019). For genetic diversity, multivariate data analysis is used of which Principal component analysis (PCA) and cluster analysis are being commonly employed (Mohammadi and Prasanna 2003). PCA helps to identify patterns, relationships between traits, and the main sources of variation among different accessions (Hair et al., 1995). Clustering techniques, enable researchers to manage germplasm collections, enhance breeding, sustainable utilization and conservation of genetic resources

(Azad et al., 2012). The study aimed morphological characterization of 50 genotypes of *Chrysanthemum morifolium* focusing on the estimation of variability and heritability in relation to both quantitative and qualitative DUS characteristics along with principal component analysis and cluster analysis to classify genotypes into groups.

2. MATERIALS AND METHODS

The experiment comprising 50 diverse chrysanthemum genotypes was laid at the experimental farm, Division of Floriculture and Landscaping, IARI, New Delhi in randomized block design with two replications during two consecutive years (2021-2022 & 2022-23). The experimental plot (4.5 m × 1.8 m) comprised of three rows distanced 60 cm apart with plants spaced at 45 cm apart. Standard agronomic practices were followed during crop growth. Data were recorded on 10 randomly selected plants for 25 descriptors including 14 quantitative characters viz., Plant based traits: plant height (PH) & number of primary branches (NPB), Leaf based traits: stipule size (SZ), leaf lamina length (LL), leaf width (LW), ratio of leaf length/width (L/W), petiole length (PL), Terminal lobe length (TLL), length of lower lobe of leaf (LLL), flower based traits: number of flowers per plant (NFP), flower head diameter (FD), peduncle length (Pd. L), ray florets length (RL), ray florets width (RW) and 11 qualitative characters viz., Plant based trait: plant type (PT), leaf based trait: petiole attitude, leaf predominant shape of base, leaf color, flower based traits: flower head type, number of types of ray floret, predominant type of ray floret, cross

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section of ray floret, rolling of margins of ray floret, longitudinal axis of majority of ray florets and shape of tip of ray floret.

The picture of flower type of chrysanthemum genotypes is given in Figure 1. Plant characters, stem, stipule, leaf and petiole characters were observed when the terminal buds show color, just before they open. Plant height was measured from the tallest point of the canopy to the base of the plant. Plant type was visually observed as bushy (having branches and no main single stem) or non bushy (producing single stem). Leaf length was measured from the lamina tip to the intersection of lamina and petiole along the lamina midrib. Leaf width was measured from the widest lamina lobes. Petiole length was measured from the stem margin to the end of leaf lamina. The number of flowers was counted during the flowering time of each cultivar. Flower head diameter was measured from the widest point of the flower. Ray floret characteristics were observed on the outermost row of the floret. Both quantitative and qualitative traits were observed according to UPOV (International Union for the Protection of New Varieties of Plants) descriptors. The year-wise quantitative data were pooled and used for statistical Analysis. The analysis of variance for quantitative traits was computed as described (Panse and Sukhatme, 1967).

Phenotypic (σ^2p) and genotypic (σ^2g) variances were calculated using the method suggested by a researcher as $\sigma^2p = \sigma^2g + \sigma^2e$, $\sigma^2g = \text{MSG} - \text{MSe}/r$ and $\sigma^2e = \text{MSe}$, where MSG, MSe and r denote mean squares of genotypes, mean squares of error and number of replications respectively (Baye, 2002). The PCV and GCV were obtained by using the formulae, $\text{PCV}(\%) = \sqrt{\sigma^2p} / \bar{x} \times 100$, $\text{GCV}(\%) = \sqrt{\sigma^2g} / \bar{x} \times 100$, where $\bar{x}$ is the sample mean (Baye, 2002). GCV and PCV values were categorized as low (< 10%), moderate (10–20%) and high (> 20%) (Deshmukh et al., 1986). Estimates of broad sense heritability (h^2_b) were calculated according to the formulae: $h^2_b = \sigma^2g / \sigma^2p$ (Allard, 1999). The expected genetic advance (GA) under selection, assuming the selection intensity of 5% was calculated as proposed by a group researchers i.e. $GA = k (\sqrt{\sigma^2p})$. σ^2g / σ^2p , K is the standardized selection differential (2.5) (Johanson et al., 1955). Genetic advance as percent of mean, $GAM = (GA / \bar{x}) \times 100$. The Shannon equitability index (evenness) is simply the Shannon diversity index (H) divided by the maximum diversity ($\log k$). Shannon diversity index (H) is calculated as $H = \sum_{i=1}^k P_i \times \ln P_i$, where k denotes the number of group and P_i denoting the

proportion in group k.

This normalizes the Shannon diversity index to a value between 0 and 1. Evenness closer to 1 indicates more diversity (Shanon and Weaver, 1949). The mean values of each quantitative trait and transformed data (scores for the each descriptor state used for transforming qualitative data is given in supplementary table S3) of qualitative traits were used to perform PCA and cluster analyses using unweighted pair group method with arithmetic averages (UPGMA) clustering algorithm with dissimilarity between genotypes expressed as Euclidean distance (Kim et al., 2014; Kachare et al., 2016; Khatun et al., 2022). Euclidean distance between two genotypes i and j, having observations on morphological characters (p) was denoted by $x_1, x_2, \dots, x_p$ and $y_1, y_2, \dots, y_p$ for i and j, respectively, and was calculated with the following formula: $d(i, j) = [(x_1 - y_1)^2 + (x_2 - y_2)^2 + \dots + (x_p - y_p)^2]^{1/2}$. The computation of PCA, Euclidean distance matrices, construction of dendrogram and calculation of cophenetic correlation coefficient was done with the help of XL STAT software (Khatun et al., 2022). Additionally Jaccard's similarity coefficient was used to perform cluster analysis on binary data based on presence (1) or absence (0) of specific descriptor state (UPOV guidelines) for both qualitative and quantitative trait (Chung et al., 2019). A cluster analysis based on Jaccard's similarity index was achieved by using the UPGMA in NTSYS-pc Version 2.1q (Chen et al., 2013).

3. RESULTS

3.1 Morphological Characterization

A total of 25 morphological characteristics (14 quantitative and 11 qualitative traits) in 50 genotypes were evaluated. The Mean, range, treatment mean sum of square, standard error of difference (SED) and coefficient of variation (CV) for quantitative characters are given in Table 1. Significant variation was reported among the genotypes for all the quantitative traits. Plant height ranged from 24.64 cm (Basanti) to 92.95 cm (Gauri) which was divided into 19 short (20–40 cm), 27 medium (40–60 cm) and 6 tall (> 60 cm) genotypes. Observation on plant type showed that 44 genotypes were bushy type and 6 were non-bushy type. Number of primary branches varied from 3.5 (Thaichen queen) to 23 (Jaya) and the genotypes were grouped into 4 sparse (<6), 15 medium (6–12) and 31 dense (>12) branching.

Table 1: Showing treatment sum of square, range, mean, SED and CV of 14 quantitative characters

Characters	Treatment Mean sum of square		Min	Max	Mean	SED	CV
	Treatment, df - 49	Error, df - 49					
PH (cm)	355.23**	11.63	24.64	92.95	42.95	3.41	7.94
NPB	35.55**	2.65	3.5	23	13.26	1.63	12.28
LL (cm)	5.68**	0.11	3.2	10	6.25	0.33	5.23
LW (cm)	3.09**	0.12	2.36	7.9	4.69	0.35	7.45
PL (cm)	2.92**	0.13	0.83	6.05	2.58	0.37	14.38
L/W	0.23**	0.02	1.22	2.87	1.88	0.15	7.87
TLL (cm)	1.72**	0.10	1.3	5.02	2.84	0.32	11.29
LLL (cm)	2.31**	0.07	0.69	4.89	2.63	0.27	10.48
SZ (cm)	0.88**	0.007	0.45	2.9	1.21	0.09	7.11
NFP	8862.2**	69.74	34	400	98.19	8.35	8.50
FD (cm)	8.23**	0.10	2.7	11.47	6.16	0.32	5.18
Pd. L (cm)	7.69**	0.52	3.4	11.7	7.37	0.72	9.82
RFL(cm)	2.42**	0.01	1.42	6.1	3.04	0.12	3.93
RFW(cm)	0.18**	0.002	0.27	1.7	0.66	0.04	6.72

** means significant at 0.1 level of significance

Figure 2 shows the pictures of leaves. The shape of base of leaves was classified into 16 acute, 10 obtuse, 6 rounded, 5 truncate, 5 cordate and 8 asymmetric shape. Leaf color was categorized into three types light, medium and dark present in 11, 21 and 18 genotypes respectively. For petiole attitude, 18 genotypes showed horizontal petiole attitude, 15 very strongly upward attitude, 13 moderately upward attitude, 3 moderately

downward and 1 showed drooping petiole attitude. The leaf lengths ranged from 3.2 cm (Kaul) to 10 cm (Pusa Chitraksha), wherein 12 genotypes have short (< 5cm), 22 genotypes medium (5–7 cm) and 16 genotypes have long (>7cm) leaves. Leaf width varied from 2.36 cm (Kaul) to 7.9 cm (Star).

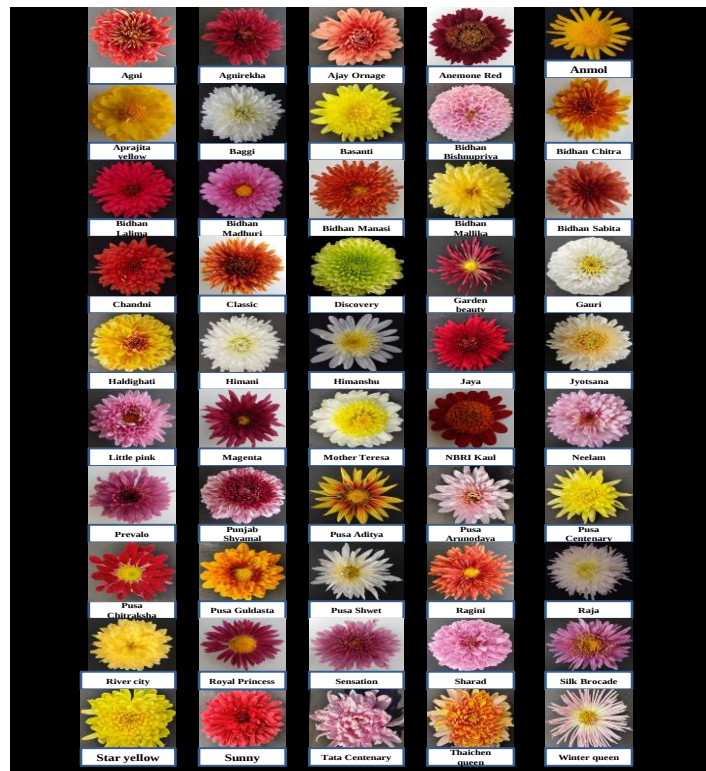


Figure 1: 50 Chrysanthemum genotypes with respective flower type

Table 2: Mean values of 25 quantitative and qualitative traits in cultivars *Chrysanthemum morifolium* (PT – Plant Type, BU – Bushy, NB- Non Bushy, PA – Petiole Attitude, VSU – Very Strongly Upward, MU- Moderately Upward, H- Horizontal, MD – Moderately Downward, D- Drooping)

Genotypes	Plant			Leaf										Flower		
	PH (cm)	NPB	PT	Leaf Shape	Leaf color	PA	LL (cm)	LW (cm)	L/W	PL	TLL (cm)	LLL (cm)	SS (cm)	NFP	FD (cm)	Pd.L (cm)
Agni	41.75	13.5	BU	Obtuse	dark	MD	4.5	3.45	1.94	2.2	1.3	1.55	0.75	129.5	6.95	7.7
Agnirekha	45.15	14.2	BU	Acute	dark	H	6.38	4.85	1.62	1.5	2.65	1.53	0.9	127.5	6.30	8.1
Ajay Orange	27.35	9.5	BU	Obtuse	medium	MU	4.72	4.75	1.22	1.1	2.05	2.5	1.4	48	4.93	6.47
Anemone red	35.15	12.65	BU	Asymmetrical	dark	D	9.25	6.35	1.83	2.4	3.2	1.25	2.7	52	6.99	7.38
Anmol	40.65	11.5	BU	Asymmetrical	light	VSU	3.8	3.6	1.75	2.47	3.1	1.85	0.63	149.5	4.14	3.4
Aprajita yellow	35.65	14.2	BU	Acute	medium	MU	5.67	3.2	2.38	1.95	1.9	3.25	0.86	102.5	4.9	6.35
Baggi	28.05	12	BU	Rounded	light	H	5.48	4.05	1.81	1.85	1.95	1.6	0.52	50.5	5.73	7.25
Basanti	24.64	18	BU	Acute	dark	VSU	5.88	5.45	1.45	2.05	2.12	3.1	0.9	76	4.35	8.49
Bidhan Bishnupriya	75.35	18	NB	Cordate	light	H	7.09	5.15	1.65	1.45	2.35	3.6	2.07	53.6	4.65	9.84
Bidhan Chitra	63.43	12	BU	Truncate	medium	H	6.05	4.4	1.72	1.55	2.61	3.22	1.65	66.4	5.5	4.35
Bidhan Lalima	46.5	19.9	BU	Obtuse	dark	H	5.3	4.9	1.56	2.35	2.83	2.35	0.55	171.5	6.35	7.35
Bidhan Madhuri	48	21	BU	Obtuse	dark	H	5.49	4.25	1.79	2.15	1.95	2.7	0.73	151.5	5.52	6.8
Bidhan Mallika	47.55	20	BU	Acute	medium	H	5.47	4.15	2.10	3.32	2.8	2.65	0.69	168	5	6.85
Bidhan Manasi	37.88	8.5	BU	Rounded	medium	H	3.86	3.35	1.41	1.4	2.23	2.67	1.7	57.5	4.35	6.30
Bidhan Sabita	46.35	19	BU	Rounded	medium	H	4.86	4.15	1.56	1.65	1.9	2.4	0.72	147	5.19	6.6

Table 2 (cont): Mean values of 25 quantitative and qualitative traits in cultivars *Chrysanthemum morifolium* (PT – Plant Type, BU – Bushy, NB- Non Bushy, PA – Petiole Attitude, VSU – Very Strongly Upward, MU- Moderately Upward, H- Horizontal, MD – Moderately Downward, D- Drooping)

Chandni	48.6 5	15.7	BU	Asymmetri c	dark	H	4.15	4.05	1.43	1.6 9	1.7	2.6	0.8	124	6.01	7.15
Classic	26	15.3 5	BU	Acute	mediu m	VS U	6.01	4.05	1.93	1.8 5	3.35	1.18	0.90	49	6.54	9.05
Dvery	40.4	9	BU	Acute	mediu m	MU	7.35	5.5	1.90	3.1 5	1.6	3.25	0.63	44	4.5	6.75
Garden Beauty	27.2 5	8.15	BU	Acute	dark	VS U	7.9	4.85	2.87	6.0 5	4.2	4.15	2.05	34	8.12	7
Gauri	92.9 5	14.5	BU	Acute	mediu m	VS U	5.25	3.9	2.68	5.1 5	3.15	2.35	1.26	400	2.7	5.55
Haldighati	44.2	13.3	BU	Asymmetri c	mediu m	H	7.25	5.35	1.95	3.0 5	3.15	3.05	0.95	106	4.42	5.25
Himani	45.4 0	17.5	BU	Cordate	mediu m	MU	6.32	4.67	1.70	1.6 6	3.01	2.97	1.75	104	8.5	11.1
Himanshu	25.8	14.1 5	BU	Acute	mediu m	MU	7.95	4.9	2.36	3.6	4	3.85	0.95	147. 5	8.65	11.6
Jaya	47.0 3	23	BU	Obtuse	mediu m	VS U	8.54	6.57	1.61	2.1	3	4.9	2.9	180. 5	5.57	10.9
Jyotsana	34.8	17.5	BU	Acute	dark	H	8.07	5.45	2.23	3.9 3	4.16	0.9	1.3	61	4.45	9.25
Little pink	50.3 5	13	BU	Cordate	light	MU	6.7	4.9	2	3.1	3.4	3.6	1.05	72	5.9	5.4
Magenta	36.5	13	BU	Asymmetri c	mediu m	VS U	4.8	3.6	1.83	1.8	3.1	1.15	0.69	50	4.38	7.5
Mother Teresa	40.1 7	14.4 5	BU	Asymmetri c	light	VS U	3.94	2.41	2.00	1.2 4	1.99	1.65	0.6	77.5	3.38	7.00
NBRI- Kaul	32.5	11.5	BU	Obtuse	light	MD	3.21	2.36	1.76	0.8 3	1.3	0.69	0.6	46	3.43	5.6
Neelam	35	14.9	BU	Asymmetri c	dark	MU	6.32	4.75	1.78	2.1 5	2.8	2.55	1.25	56	5.40	7.35
Prevalo	29	14.6	BU	Obtuse	dark	H	5.15	4.15	1.76	2.1 5	2.15	1	0.53	57.5	8.26	9.5
Punjab Shyamal	46.2 5	9.5	BU	Rounded	dark	H	8.1	4	2.53	2	2.16	2.1	0.47	67.5	4.56	11.4
Pusa Aditya	42.2 5	16	BU	Obtuse	dark	H	5.47	3.97	1.87	1.9 4	1.61	0.9	0.78	144	5.4	9.4
Pusa Arunodaya	30.1	7.5	N B	Cordate	light	H	5.85	4.95	1.85	3.3 5	3.9	2.45	2.05	40	9.27	6.7
Pusa Centenary	43.4	13.5	N B	Truncate	mediu m	MU	7.6	5.75	1.70	2.2 2	3.85	2.55	2.45	38.5	10.1	10.2
Pusa Chitraksha	49	14	BU	Rounded	light	MU	10	7.2	2.01	4.5	4.5	4.15	1.65	170	6.75	9.05
Pusa Guldasta	45.6	18	BU	Acute	mediu m	MD	6.75	4.85	2.47	5.2 5	3.3	3.2	1.82	137	5.35	8.75
Pusa Shwet	49.0 5	15.0 5	BU	Truncate	dark	MU	8.15	6.27	1.82	3.3	2.99	2.6	0.80	231	7.75	5.08
Ragini	571	13	BU	Acute	mediu m	MU	6.17	4.03	2.24	2.6 5	2.99	1.55	0.72	150. 5	6.42	10.0
Raja	25	12	BU	Rounded	light	MU	6.9	5.7	1.73	3	3.7	4.35	1.72	34	8.65	6.05
River city	49.1	10	BU	Asymmetri c	mediu m	VS U	3.62	3.2	1.48	1.1 5	1.55	2.65	0.65	127	4.97	5.91
Royal Princess	28.5	12	BU	Acute	mediu m	VS U	4.86	3.15	2.13	1.6	2.7	3.15	0.65	36	2.77	4.97
Sensation	56	12.5	BU	Obtuse	dark	H	4.33	3.45	1.79	1.8 7	2.09	1.55	1.05	164	6.3	9.35
Sharad	35.5	11.0 5	BU	Acute	dark	H	5.2	3.05	1.81	1.9	1.8	2	0.66	71.5	5.25	6
Silk Brocade	44.9 5	12.8	BU	Acute	dark	MU	5.80	5.3	1.61	2.3 5	2.85	2.55	0.45	93	7.75	7
Star Yellow	56.7 5	5	N B	Cordate	dark	VS U	9.65	7.9	1.67	3.6 5	5.02	4.1	2.1	61.5	10.2	7
Sunny	52.5	14	BU	Acute	light	VS U	5.35	4.8	2.33	5.8 5	4.55	2.5	1.15	38	5.45	6.1

Table 2 (cont): Mean values of 25 quantitative and qualitative traits in cultivars *Chrysanthemum morifolium* (PT – Plant Type, BU – Bushy, NB- Non Bushy, PA – Petiole Attitude, VSU – Very Strongly Upward, MU- Moderately Upward, H- Horizontal, MD – Moderately Downward, D- Drooping)

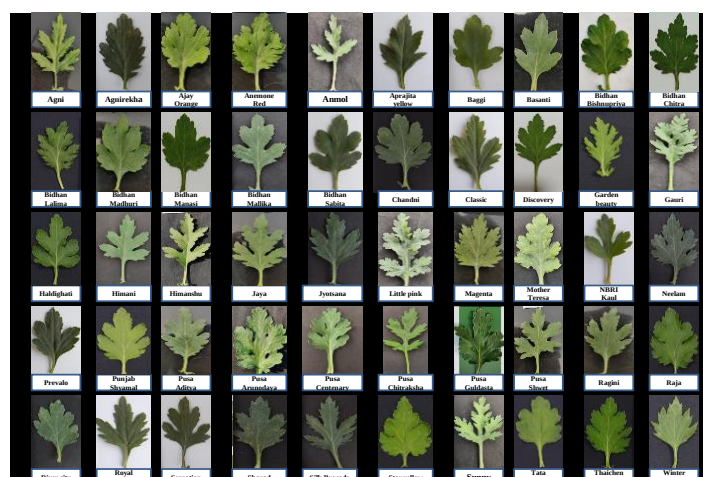
Tata Centenary	65.9	4.5	N B	Truncate	mediu m	VS U	9.05	7.45	1.53	2.4	4.15	4.6	2.05	45	10.4	6.55
Thaichen queen	42.95	3.5	N B	Truncate	light	VS U	8.65	6.95	1.66	2.95	3.85	4.55	2.1	63	11.4	3.6
Winter queen	28.2	5.5	BU	Obtuse	mediu m	VS U	8.05	5.3	2.26	3.95	3.5	3.9	2.45	38	7.97	6.05

Table 2 (cont): Mean values of 25 quantitative and qualitative traits in cultivars *Chrysanthemum morifolium* (PT – Plant Type, BU – Bushy, NB- Non Bushy, PA – Petiole Attitude, VSU – Very Strongly Upward, MU- Moderately Upward, H- Horizontal, MD – Moderately

	RFL (cm)	RFW (cm)	Flower head	No. of types of ray floret	Predominant ray floret	Ray floret cross section	Ray floret-rolling of margins	Ray floret longitudinal axis	Ray floret: shape of tip
Agni	3.25	0.68	Double	1	Incurved	Moderately concave	Moderately involute	Incurving	Emarginated
Agnirekha	3.05	0.56	Daisy-eyed double	2	Spatulate	Weakly conex	Flat	Straight	Pointed
Ajay Orange	3.2	0.5	Double	3	Ligulate	Weakly concave	Weakly involute	Straight	mamillate
Anemone red	3.14	0.53	Double	1	Spatulate	Weakly concave	Flat	Straight	dentate
Anmol	2	0.51	Double	1	Ligulate	Weakly concave	Flat	Straight	mamillate
Aprajita yellow	2.15	0.66	Semi double	1	Ligulate	Weakly conex	Flat	Straight	Emarginated
Baggi	2.59	0.62	Double	1	Ligulate	Moderately concave	Weakly revolute	Straight	Rounded
Basanti	2.38	0.72	Double	1	Ligulate	Flat	Flat	Straight	Pointed
Bidhan Bishnupriya	1.65	0.35	Double	2	Funnel shaped	Strongly concave	Moderately involute	Straight	Fringed
Bidhan Chitra	2.33	0.55	Daisy-eyed double	1	Ligulate	Flat	Flat	Straight	Emarginated
Bidhan Lalima	3.15	0.55	Double	1	Spatulate	Weakly concave	Flat	Straight	Pointed
Bidhan Madhuri	3	0.5	Semi double	1	Spatulate	Weakly concave	Flat	Straight	Emarginated
Bidhan Mallika	2.47	0.62	Daisy-eyed double	1	Ligulate	Flat	Flat	Straight	Pointed
Bidhan Manasi	1.78	0.5	Daisy-eyed double	2	Spatulate	Weakly concave	Flat	Straight	Emarginated
Bidhan Sabita	3.17	0.59	Double	1	Ligulate	Weakly conex	Flat	Straight	dentate
Chandni	3.35	0.65	Double	1	Spatulate	Weakly conex	Weakly revolute	Straight	dentate
Classic	3.25	0.42	Double	1	Ligulate	Weakly conex	Flat	Straight	Pointed
Discovery	1.89	0.61	Double	1	Incurved	Strongly concave	Moderately involute	Straight	Emarginated
Garden Beauty	4.75	0.3	Single	1	Spatulate	Flat	Flat	Straight	Pointed
Gauri	1.42	0.5	Double	1	Ligulate	Weakly conex	Weakly revolute	Reflexing	dentate
Haldighati	2.25	0.62	Double	3	Incurved	Moderately concave	Flat	Straight	Rounded
Himani	4.32	0.7	Double	3	Ligulate	Flat	Flat	Straight	Rounded
Himanshu	4.2	0.85	Double	2	Ligulate	Weakly conex	Weakly revolute	Straight	dentate
Jaya	2.85	0.42	Daisy-eyed double	1	Spatulate	Weakly conex	Flat	Straight	dentate
Jyotsana	3.05	0.52	Double	1	Ligulate	Weakly conex	Weakly revolute	Straight	dentate
Little pink	2.6	0.55	Semi double	2	Spatulate	Moderately concave	Weakly revolute	Straight	Rounded

Table 2 (cont): Mean values of 25 quantitative and qualitative traits in cultivars *Chrysanthemum morifolium* (PT – Plant Type, BU – Bushy, NB- Non Bushy, PA – Petiole Attitude, VSU – Very Strongly Upward, MU- Moderately Upward, H- Horizontal, MD – Moderately

Magenta	1.90	0.38	Semi double	1	Ligulate	Flat	Flat	Reflexing	Emarginated
Mother Teresa	1.56	0.49	Double	1	Ligulate	Flat	Flat	Straight	Rounded
NBRI- Kaul	1.55	0.54	Double	1	Ligulate	Flat	Flat	Reflexing	mamillate
Neelam	3.75	0.68	Double	1	Spatulate	Weakly conex	Flat	Straight	Pointed
Prevalo	3.19	0.59	Double	1	Spatulate	Weakly conex	Flat	Straight	Emarginated
Punjab Shyamal	1.43	0.57	Double	1	Ligulate	Flat	Flat	Reflexing	Rounded
Pusa Aditya	2.95	0.68	Single	1	Spatulate	Moderately concave	Weakly revolute	Straight	mamillate
Pusa Arunodaya	4.1	1.1	Semi double	1	Ligulate	Flat	Flat	straight	dentate
Pusa Centenary	5.51	1.4	Double	1	Incurved	Strongly concave	Moderately involute	Incurving	Rounded
Pusa Chitraksha	3.15	0.72	Single	3	Spatulate	Flat	Flat	Straight	Pointed
Pusa Guldasta	2.35	0.92	Semi double	1	Ligulate	Weakly conex	Flat	Straight	mamillate
Pusa Shwet	4	0.75	Semi double	1	Ligulate	Weakly concave	Flat	Straight	Pointed
Ragini	2.95	0.57	Semi double	1	Ligulate	Flat	Flat	Twisted	Rounded
Raja	3	0.57	Double	1	Ligulate	Flat	Flat	Straight	Rounded
River city	2.85	0.5	Daisy-eyed double	1	Ligulate	Weakly conex	Flat	Twisted	Pointed
Royal Princess	2.05	0.48	Single	1	Ligulate	Flat	Flat	Straight	Pointed
Sensation	2.75	0.63	Semi double	1	Spatulate	Weakly concave	Flat	Straight	Emarginated
Sharad	3.10	0.57	Double	1	Spatulate	Moderately concave	Flat	Reflexing	Emarginated
Silk Brocade	2.27	0.55	Double	2	Spatulate	Weakly concave	Flat	Straight	Pointed
Star Yellow	4.45	1.7	Double	1	Incurved	Strongly concave	Moderately involute	Incurving	Pointed
Sunny	3.1	0.57	Double	1	Ligulate	Flat	Flat	Straight	Rounded
Tata Centenary	6.1	1.55	Double	1	Ligulate	Moderately concave	Moderately involute	Incurving	Rounded
Thaichen queen	5.97	1.57	Double	1	Ligulate	Moderately concave	Moderately involute	Incurving	dentate
Winter queen	4.55	0.27	Single	1	Quilled	NA	NA	Straight	Rounded

**Figure 2:** 50Chrysanthemum genotypes with respective Leaf type

Yellow where 13 genotypes bear narrow leaf (<4cm), 21 genotypes medium leaf (4-5 cm) and 16 genotypes have broad leaf (> 5 cm). Leaf L/W ratio ranged from 1.2 (Ajay orange) to 2.87 (Garden Beauty) and genotypes were described as 5 low (<1.5), 41 medium (1.5 – 2.5) and 4 high (>2.5) L/W. For petiole length the genotypes were grouped as 33 short (< 2.5 cm), 13 medium (2.5 cm – 4.5) and 4 long (>4.5 cm). Shortest petiole length was observed in Kaul (0.83 cm) and longest in Garden beauty (6.05cm) which was at par with Sunny (5.85 cm). For terminal lobe length genotypes were evaluated as 12 short (< 2 cm), 17 medium (2 cm – 3 cm) and 21 long (> 3cm). Agni and Kaul had shortest terminal lobe of 1.3 cm and Star Yellow had longest one i.e. 5.02cm and was at par with Sunny (4.55cm) and Chitraksha (4.5 cm). The lower lobe length ranged from 0.69 cm in Kaul to 4.6 cm in Tata Centenary and genotypes were categorized as 14 short (< 2 cm), 17 medium (2 cm – 3 cm) and 19 long (> 3cm). The stipule size varied from 0.45 cm in Silk Brocade to 2.9 cm in Jaya and genotypes were grouped as 27 small (< 1 cm), 13 medium (1 cm – 2 cm) and 10 large (> 2 cm).

Evaluation of number of flowers per plant showed that 12 genotypes bear few flowers (< 50), 17 medium (50 – 100) and 21 genotypes had many flowers (>100). Gauri was an outlier with highest number of flowers (400) whereas Raja and Garden Beauty had the least (34). Flower diameter was observed as small (< 4 cm), medium (4 cm – 8 cm) and large (>8 cm) in 4, 36 and 10 genotypes respectively. Gauri had the smallest flower head diameter of 2.7 cm and Thaichen queen had the widest diameter of 11.4 cm followed by Tata Centenary and Star Yellow. Based on peduncle length genotypes were grouped as 4 short (< 5 cm), 33 medium (5 cm – 9 cm) and 13 long (> 9 cm). Shortest peduncle length was observed in Anmol (3.4 cm) and longest in Himanshu (11.7 cm). Flower head was classified into 4 categories viz. single type 5 genotypes, semi double 9, daisy eyed double 6 and 30 genotypes were double type. Different genotypes had wide range of flower color (supplementary table S1). Ray floret length was observed as short (< 2 cm), medium (2 cm – 3 cm) and long (> 3 cm) present in 8, 18 and 24 genotypes respectively. Longest ray floret was in Tata Centenary (6.1 cm) and the smallest in Gauri (1.42 cm).

The ray floret width ranged from 0.27 cm in Winter queen to 1.7 cm in Star Yellow. Based on ray floret width genotypes were classified as 11 narrow (<0.5 cm), 34 medium (0.5 cm – 1 cm) and 5 broad (> 1 cm). Out of 50 cultivars, 6 cultivar consist of two types of ray floret, among these Agnireka and Silk Brocade had spatulate and ligulate ray floret, Bidhan Manasi and Little pink had spatulate and quilled ray floret and Himanshu and Bidhan Bishnupriya had Ligulate and funnel shaped ray floret. Four genotypes comprised of three types of ray floret, out of which 3 genotypes namely Pusa Chitraksha, Ajay orange and Himani contain spatulate, ligulate and quilled ray floret, whereas Haldighati consist of incurved, spatulate and quilled type of ray floret. The remaining genotypes constitute of only one type of ray floret. Ligulate, spatulate, incurved, funnel shaped and quilled were predominant ray floret in 27, 16, 5, 1 and 1 genotype respectively. For the ray floret-cross section, 4 genotypes were with strongly concave ray floret, 8 had moderately concave, 9 showed weakly concave, 15 had flat and 13 exhibited weakly convex ray floret-cross section, the variety Winter Queen was quilled type, therefore this character was not applicable in this.

Besides flat margins of ray floret in 34 genotypes, 3 more types: moderately involute in 7, weakly revolute in 7 and weakly involute in 1 genotype, this character was also not applicable in cultivar Winter Queen. The longitudinal axis of ray floret was classified into four types, out of 50 genotypes studied, 1 genotype had twisted longitudinal axis, 6 genotypes showed incurving longitudinal axis, 5 cultivars showed reflexing longitudinal axis and remaining 38 genotypes showed straight longitudinal axis. Depending on the shape of tip of ray floret, 50 genotypes were categorized into five types viz. emarginated (10), pointed (13), mammillate (5), dentate (9), rounded (12) and fringed (1). The above classification of quantitative and qualitative traits is based on descriptor state for different characters given in UPOV DUS guidelines. The mean performance of various genotypes for 14 quantitative characters and observed trait for 11 qualitative trait is given in table 2. Figure 3 gives box plot depicting data dispersion for quantitative traits.

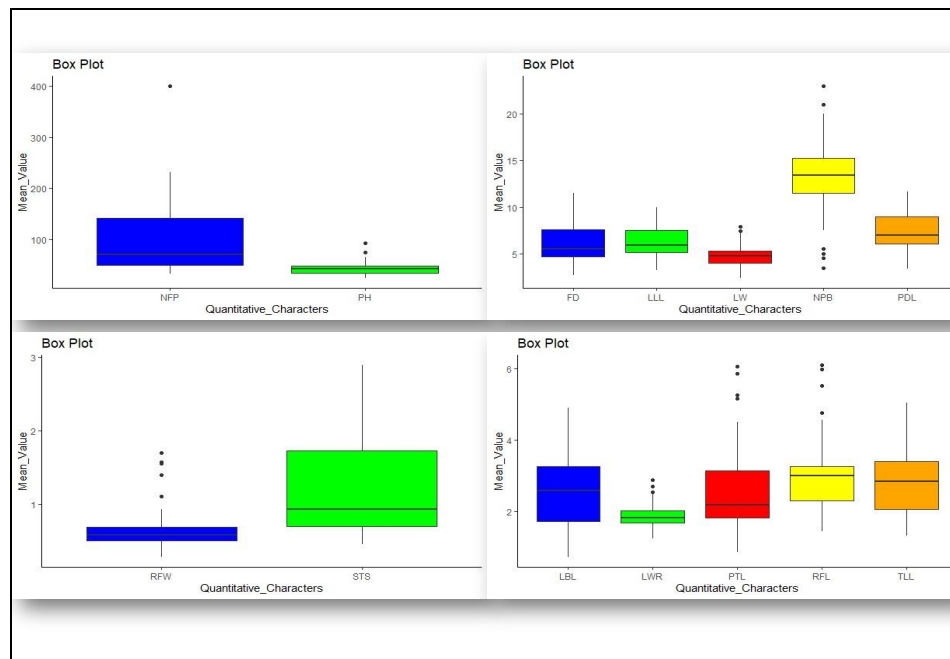


Figure 3: Box plot depicting data dispersion for quantitative traits. NFP - Number of flowers per plant, PH - Plant height, FD - Flower diameter, LLL - Leaf lamina length, LW - Leaf width, NPB - Number of primary branches, PDL - Peduncle length, RFW - Ray floret width, STS - Stipule size, LBL - Lower lobe length, LWR - Leaf width ratio, PTL - Petiole length, RFL - Ray floret length, TTL - Terminal lobe length

3.2 Shannon Equitability Index for qualitative traits

The Shannon Equitability (E_H) was estimated for 11 qualitative characters to measures the evenness of different types in the population. It varied from 0.52 to 0.96. Six characters showed E_H greater than 0.75 viz. leaf shape of base (0.94), leaf green color of upper surface (0.96), petiole attitude (0.82), flower head type (0.79), ray floret profile in cross section (0.94) and ray floret shape of tip (0.91). Diversity of phenotypic classes for qualitative trait (supplementary table S2).

3.3 Genetic Variability, Heritability and Genetic Advance

Estimates of genotypic coefficient of variance (GCV) and phenotypic coefficient of variance (PCV) of different traits are given in Table 3. The highest GCV and PVC values were found particularly for number of flowers

per plant (67.52% and 68.05%), stipule size (54.33% and 54.76%), petiole length (45.8% and 48.00%) and Ray floret width (45.71% and 46.21%) respectively. Whereas moderate GCV and PCV were recorded for leaf lower lobe length (40.17% and 41.51%) and ray floret length (36.08% and 36.29%) respectively. Low GCV and PCV viz. 17.28% and 18.98% respectively was recorded for ratio of leaf length to leaf width indicating existence of less variability. Most of the traits in this study showed broad sense heritability > 90%. Ray floret length (98.84%) exhibited highest heritability followed by number of flowers per plant (98.43), stipule size (98.42) and ray floret width (97.84%). Leaf length to width ratio showed relatively low heritability. Genetic advance as percent of mean was highest for number of flowers per plant (138.01) followed by stipule size (111.03) and ray floret width (93.13).

Table 3: Estimates of genetic variability, heritability and genetic advance

Characters	var (g)	var (p)	Heritability (%)	GA	GA% mean	GCV (%)	PCV (%)
Plant: Height(cm)	171.8	183.43	93.65	26.12	60.83	30.51	31.53
No. of primary Branches	16.45	19.10	86.12	7.75	58.51	30.59	32.96
Leaf Length (cm)	2.78	2.89	96.30	3.37	54.04	26.73	27.24
Leaf Width (cm)	1.48	1.60	92.33	2.41	51.37	25.95	27.01
Petiole length (cm)	1.39	1.23	91.03	2.31	90.02	45.80	48.0
Leaf: ratio length/width	0.106	0.128	82.81	0.61	32.39	17.28	18.98
Terminal Lobe length (cm)	0.806	0.909	88.33	1.73	61.09	31.62	33.58
Lower lobe length (cm)	1.116	1.192	93.62	2.10	80.07	40.17	41.51
Stipule size (cm)	0.437	0.444	98.42	1.35	111.03	54.33	54.76
No. of flowers per plant	4396.2	4465.9	98.43	135.51	138.01	67.52	68.05
Flower head: diameter (cm)	4.063	4.165	97.55	4.10	66.58	32.72	33.13
Peduncle length (cm)	3.583	4.107	87.241	3.64	49.41	25.90	27.49
Ray floret: length (cm)	1.2015	1.2155	98.84	2.24	73.90	36.08	36.29
Ray floret: width (cm)	0.091	0.093	97.849	0.614	93.13	45.70	46.20

3.4 PCA Results

PCA identifies variables that are most significant in describing the overall variability in the dataset. PCA was performed separately on 14 quantitative characters and 11 qualitative traits is given in Table 4 & 5 respectively showing eigen values, factor loading, proportion of variability and cumulative variability.

3.4.1 Quantitative traits

A total of 14 quantitative characteristics of Chrysanthemum were evaluated for classification in multivariate analysis. Eigen values varied from 0.01 to 5.33 (Figure 5 A). 4 principal components were having Eigen values more than 1.00 viz. PC 1 (5.33), PC 2 (2.24), PC 3 (1.52) and PC 4 (1.37) which together explaining 74.7% of the phenotypic variation

present in the data. The PC 1 accounted for 38.09% of the total phenotypic variability with major contribution from 6 characters namely leaf lamina length, leaf width, Terminal lobe length, stipule size, flower diameter and ray floret length. The PC 2 covered 16 % of the total variation and was closely related with 4 characters viz. petiole length, leaf length to width ratio and number of flowers per plant. The PC 3 constituting 10.83% of the total phenotypic and is mainly contributed by leaf length to width ratio. PC 4 explains 9.83% of variability with major contribution from peduncle length. The biplot (Figure 4) provides an insight into the direction of correlation between variables. Flower diameter showed highly positive correlation with ray floret length and ray floret width. Number of flowers per plant is positively correlated with number of primary branches. Leaf lamina length strongly and directly correlated with terminal lobe length.

Table 4: Factor loadings, Eigen values and cumulated total variation of the first four principal components for quantitative traits

Characters	F1	F2	F3	F4
Plant Height	0.072	0.506	-0.495	-0.524
Number of primary branches	-0.408	0.436	-0.484	0.407
Leaf lamina length	0.836	0.218	-0.047	0.220
Leaf width	0.875	0.053	-0.247	0.021
Petiole length	0.517	0.652	0.424	-0.048
L/W	0.073	0.697	0.586	0.145
Terminal lobe length	0.810	0.270	0.216	0.042
Lower lobe length	0.665	0.107	-0.052	-0.217
Stipule size	0.735	0.016	-0.093	0.082
Number of flowers per plant	-0.184	0.677	-0.442	-0.289
Flower diameter	0.813	-0.305	-0.106	0.090
peduncle length	0.059	0.195	-0.356	0.814
Ray floret length	0.790	-0.294	-0.073	0.057
Ray floret width	0.680	-0.270	-0.231	-0.225
Eigen values	5.333	2.241	1.517	1.376
Cumulated variation (%)	38.094	54.100	64.934	74.760

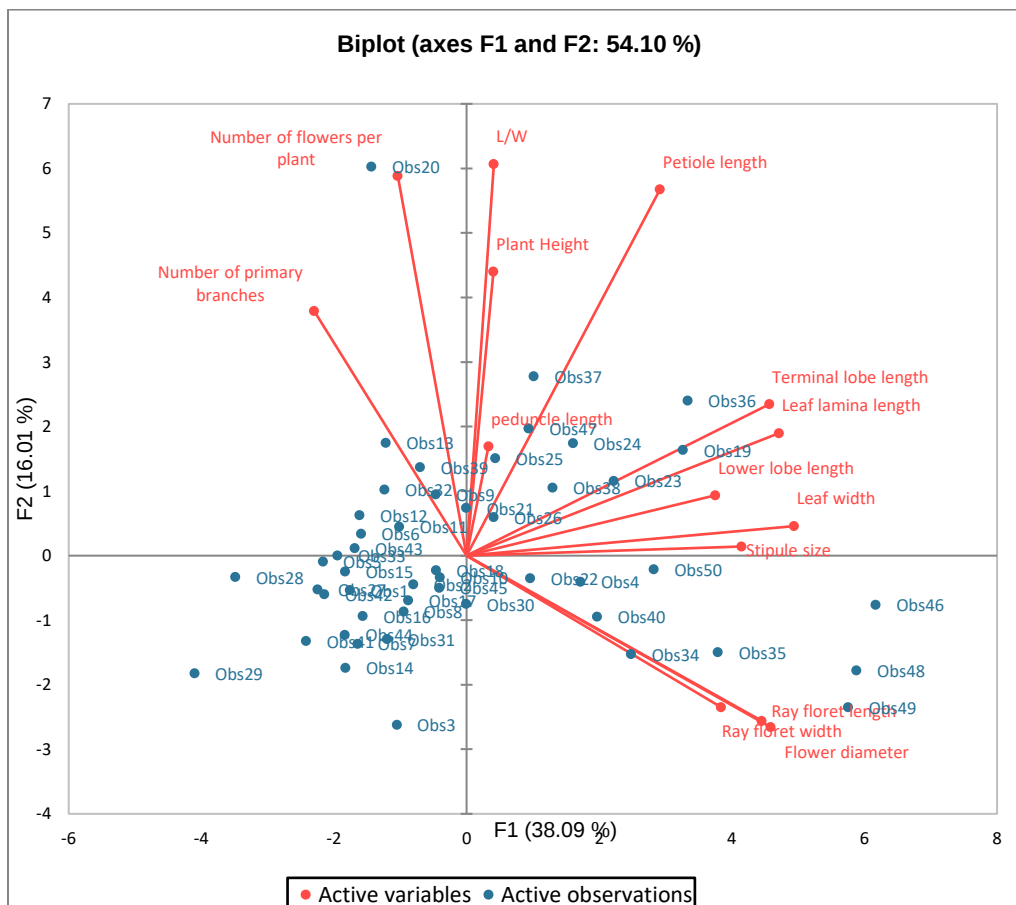


Figure 4: The biplot of 50 Chrysanthemum genotypes for principal components one (F1) and two (F2) for the quantitative traits. The lines show the contribution (magnitude and direction) of the 14 quantitative traits

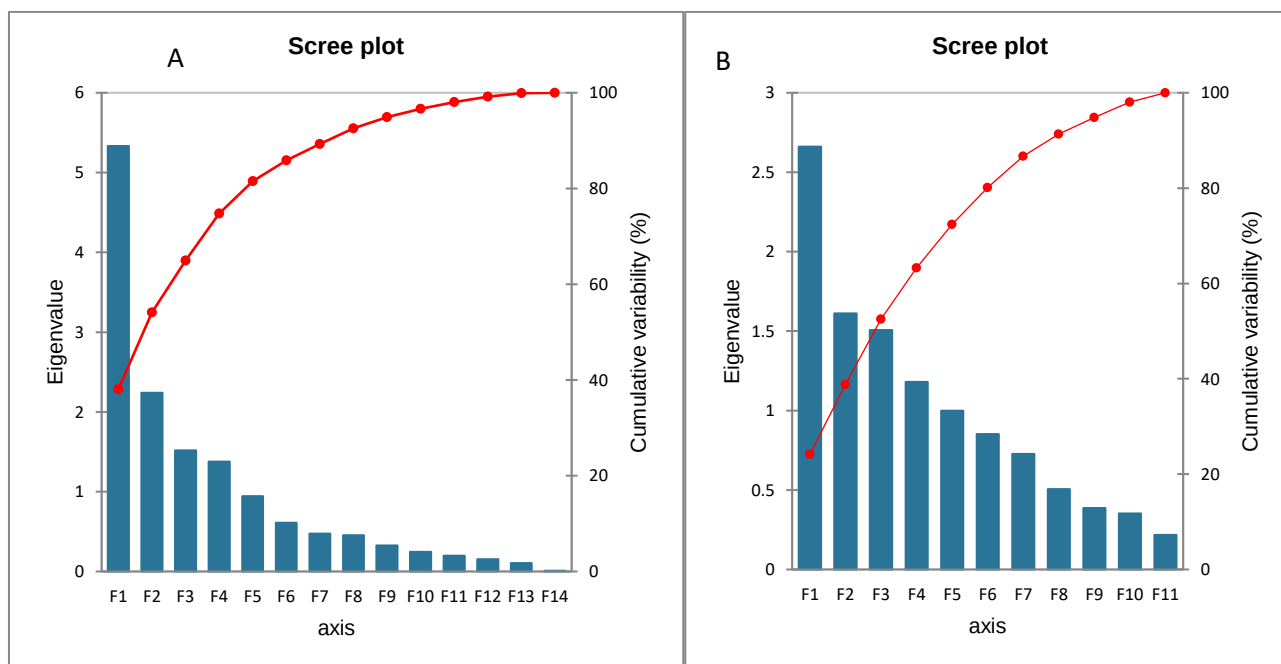


Figure 5: The screen plot shows the eigen values of different principal components(bars in the figure) and the percent cumulative variability (line in the figure) as shown by PCA A: based on quantitative character B: based on qualitative morphological traits.

3.4.2 Qualitative characters

PCA for qualitative traits was conducted using harmonized values (transformed data). Eigen value ranged from 0.22 to 2.66 (Figure 5 B). 4 principal components viz. PC 1 (2.66), PC 2 (1.61), PC 3 (1.51) and PC 4 (1.2) were having Eigen values more than 1.00 which cumulatively explained 63.27% of the total phenotypic variation present in the data. PC 1 explains 24.18% of the total variation and was correlated with ray floret

rolling of margins, ray floret cross section, plant type and ray floret longitudinal axis. PC 2 accounted for 14.6518% of total phenotypic variability and mainly contributed by petiole attitude, leaf color and predominant type of ray floret. PC 3 contributes 13.71% of total variability and is associated with flower head and ray floret: shape of tip. PC 4 constitutes 10.73% with major contribution from number of types of ray floret. The biplot for qualitative trait is given in Figure 6.

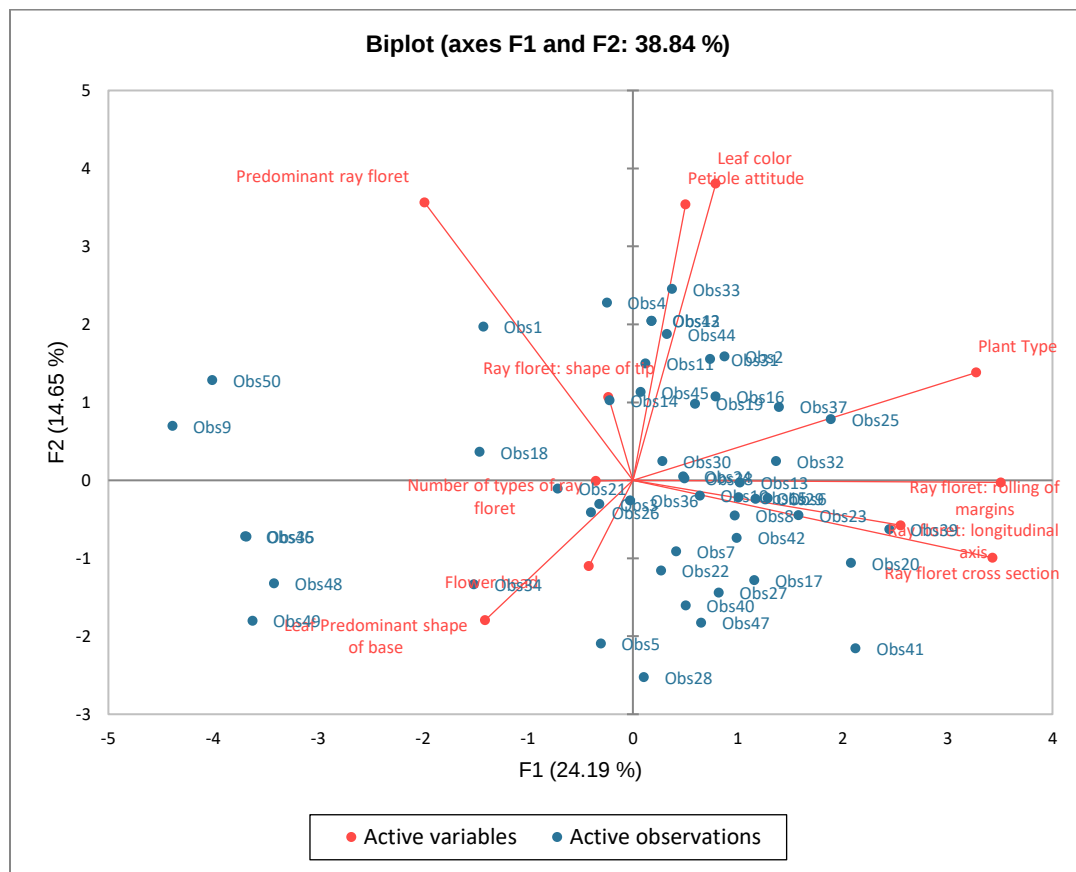


Figure 6: The biplot of 50 Chrysanthemum genotypes for principal components one (F1) and two (F2) for the quantitative traits. The lines show the contribution (magnitude and direction) of the 11 qualitative traits

Table 5: Factor loadings, Eigen values and cumulated total variation of the first four principal components for qualitative traits

Characters	F1	F2	F3	F4
Plant Type	0.767	0.252	-0.147	0.218
Petiole attitude	0.118	0.645	0.579	0.011
Leaf Predominant shape of base	-0.330	-0.327	0.355	0.310
Leaf color	0.185	0.694	-0.239	-0.274
Flower head	-0.098	-0.200	0.531	-0.290
Number of types of ray floret	-0.082	-0.001	0.163	0.839
Predominant ray floret	-0.465	0.650	-0.105	0.308
Ray floret cross section	0.804	-0.181	0.201	-0.091
Ray floret: rolling of margins	0.822	-0.005	0.252	0.156
Ray floret: longitudinal axis	0.598	-0.105	-0.179	0.178
Ray floret: shape of tip	-0.054	0.195	0.715	-0.125
Eigen values	2.661	1.611	1.508	1.181
Cumulated variation (%)	24.187	14.649	13.706	10.733

3.5 Cluster analysis based on Euclidean distance

Unweighted pair group method with arithmetic average (UPGMA) was used for cluster analysis and Euclidean distance matrices were also constructed. The harmonized values for qualitative trait and measured value of quantitative traits were used for analysis. The Euclidean distance ranged from 5.37-372.36. The maximum distance of 372.36 was observed between Gauri and Raja belonging to cluster 1 and cluster 3 respectively. This was followed by a distance of 372.02 between Gauri (cluster 1) and Garden beauty (cluster 3) and a distance of 369.72 between Gauri and Royal Princess (cluster 3). The closest related cultivars were *Bidhan Lalima* and *Bidhan Mallika* with a distance of 5.37 followed by *Bidhan Madhuri* and *Bidhan Sabita* with a distance of 6.7 and all four of them were grouped together in cluster 4. The UPGMA dendrogram aligned with the distance matrix as indicated by cophenetic correlation coefficient value of 0.92.

At a Euclidean distance of 88.94 the genotypes were grouped into four clusters. The distribution pattern revealed maximum number of genotypes *i.e.*, 28 in cluster 3 followed by cluster 4 having 20 genotypes and cluster 1 and cluster 2 have Gauri and Pusa Shweta respectively. Cluster 3 has 28 genotypes viz. Ajay orange, Anemone red, Baggi, Basanti, Bidhan Bishnupriya, Bidhan Chitra, Bidhan manasi, Classic, Discovery, Garden beauty, Jyotsana, Little pink, Magenta, Mother Teresa, Kaul, Neelam, Prevalo, Punjab Shyamal, Pusa Arunodaya, Pusa Centenary, Raja, Royal princess, Sharad, Star yellow, Sunny, Tata centenary, Thaichen queen and Winter queen. Cluster 4 comprise of 20 genotypes namely Agni, Agnirekha, Aprajita yellow, Bidhan Lalima, Bidhan Madhuri, Bidhan Mallika, Bidhan Sabita, Chandni, Haldighati, Himani, Himanshu, Jaya, Pusa Aditya, Pusa Chitraksha, Pusa Guldasta, Ragini, River city, Sensation and Silk Brocade. Figure 7 represents clustering based on Euclidean distance.

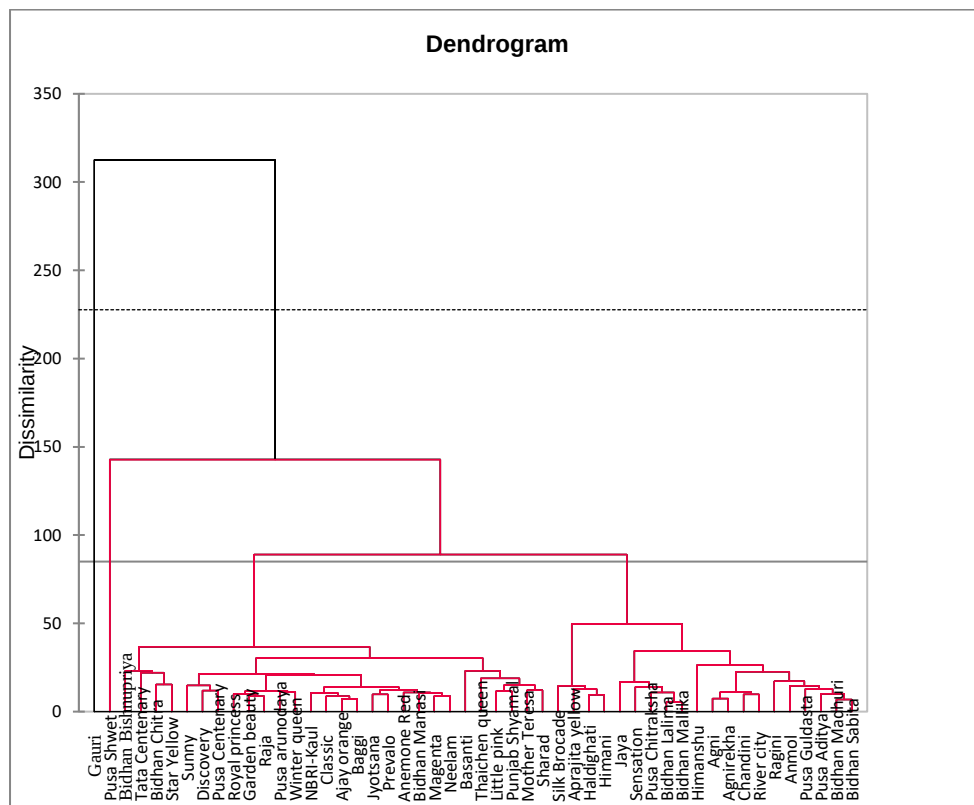


Figure 7: Dendrogram of 50 chrysanthemum cultivars based on Euclidean distance (UPGMA)

3.6 Cluster analysis based on jaccard's similarity index

UPGMA cluster analysis was also performed based on Binary data to resolve the genetic relationships among the 50 genotypes. The Jaccard's similarity coefficient varied from 0.04 to 0.69. The dendrogram divided the genotypes into 9 main clusters at the similarity coefficient of 0.33. The minimum Jaccard's similarity coefficient was 0.04 between genotype Bidhan Manasi and Thaichen Queen which belong to cluster 5 & 1 respectively, whereas maximum Jaccard's similarity coefficient of 0.69 was same between Agnirekha and Bidhan Lalima, Bidhan Lalima and Bidhan Madhuri and Bidhan Sabita and Chandini. Cluster 1 contained 5 genotypes namely Thaichen Queen, Star Yellow, Tata Centenary, Pusa Centenary and Pusa Arunodaya. Cluster 2, 4 and 5 have two genotypes each. Cluster 2 consisted of Garden Beauty and winter Queen. Bidhan Bishnupriya and Jaya made cluster 4. Ajay orange and Bidhan Manasi

constituted Cluster 5. Cluster 3 and 6 had only one genotype each viz. Gauri and Punjab Shyamal respectively. Cluster 7 had 6 genotypes namely Anmol, Magenta, Mother Teresa, Kaul, Royal Princess and Rivercity. Cluster 8 comprised of 7 genotypes namely Discovery, Haldighati, Himani, Sunny, Raja, Pusa Chitaksha, Little Pink. There were 24 genotypes in cluster 9 namely Agni, Pusa Aditya, Sensation, Agnirekha, Bidhan Lalima, Bidhan Madhuri, Neelam, Prevalo, Bidhan Sabita, Chandini, Basanti, Silk Brocade, Bidhan Mallika, Ragini, Pusa Shwet, Baggi, Sharad, Aprajita yellow, Pusa Guldasta, Bidhan Chitra, Anemone Red, Jyotsana, Classic and Himanshu. Cluster 9 showed sub-clustering at Jaccard's similarity index of 0.35 with two sub clusters i.e. cluster 9a and cluster 9b. Cluster 9a included 4 genotypes namely Anemone Red, Jyotsana, Classic and Himanshu. Cluster 9b contain rest of the 20 genotypes. Fig 8 represent clustering of 50 chrysanthemum genotypes based on Jaccard's similarity index.

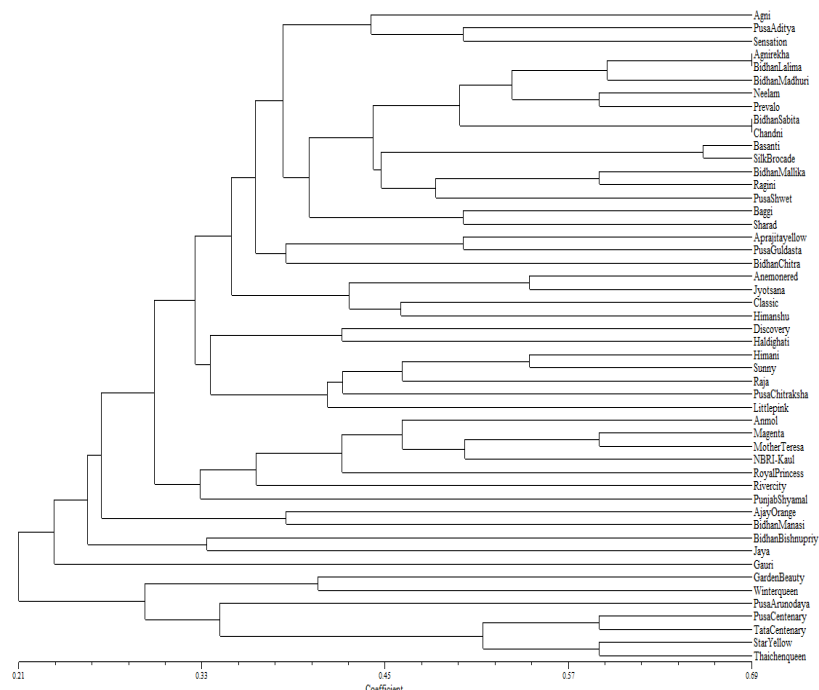


Figure 8: Dendrogram of 50 chrysanthemum cultivars based on Jaccard's similarity index (UPGMA)

4. DISCUSSION

Assessing the diversity within a crop species is crucial for identifying unique traits or alleles that can be used to enhance desirable characteristics. Crossing genotypes from different clusters with greater genetic distance can enhance genetic diversity and potentially result in improved crop varieties with desirable traits.

4.1 Morphological characterization

The 50 accessions showed significant variation with respect to 14 quantitative characters. Shannon equitability index showed that 6 out of 11 qualitative traits had equitability index of more than 0.75 indicating high level of diversity. Reasonable diversity was exhibited by predominant type of ray floret (0.67) and ray floret rolling of margins (0.63). Plant attributes like plant height, plant type and number of primary branches along with floral characteristics determines the end use of chrysanthemum as pot plant, loose flower, garden display and cut flower. Basanti, Himanshu, Prevalo, Sharad and Jyotsana could be used for pot purpose based on plant height, flower color and good number of flowering. Based on number of flowers, Gauri, Pusa Shwet, Himani, Jaya, Bidhan Lalima, Bidham Madhuri, Bidhan Mallika, Bidhan Sabita, and Chandni are suitable for loose flower purpose. Similar evaluation of genotype was done (Suvija et al., 2016). Leaf characteristics enable early identification of varieties which helps in early selection in breeding (Gao et al., 2020). It was found that leaf length and petiole length were important parameters in the evaluation of hybrid varieties in breeding studies. Differences were observed in terms of both leaf size and leaf shape. Morphological variation for leaf characteristics in Chrysanthemum have also been reported (Zhen et al., 2013). Variation in flower characteristics among Chrysanthemum genotypes were reported earlier and might vary depending on climatic conditions (Guo et al., 2008; MacDonald et al., 2017; Wang et al., 2021). Ray floret characteristics determine aesthetic value of cultivar and maybe useful in crop improvement program. Color variations in ray florets were noted among cultivars. Pusa Aditya, Pusa Guldasta, Punjab shyamali and River city showed presence of secondary color in the inner side of the ray floret. Large genetic variation for ray floret traits was previously observed (Lim et al., 2014).

4.2 Genetic variation, Heritability and Genetic advance

High values of PCV and GCV value indicates high variability and vice versa. Presence of high variability indicates effective selection for the character. Moderate to low variability indicates the need for improvement of base population (Chauhan et al., 2020). The results indicated that PCV are slightly greater than the GCV for all the traits, this mean that the traits under study were less influenced by environment. Similar PCV & GCV values for growth and floral characters were observed (Sarkar et al., 2005). High estimates for heritability and genetic advance as percentage of mean was recorded for number of flowers per plant, stipule size and ray floret width. In chrysanthemum, the high heritability values and genetic advance as per cent of mean for number of flowers per plant was also reported (Henny et al., 2021).

4.3 PCA Analysis

PCA is a valuable tool in multivariate analysis to reduce the dataset's dimensionality without losing important information about the relationships between variables. PCA was done using a correlation matrix as it helps to ensure that the PCA results are robust, interpretable, and not biased by the original measurement units of the variables when dealing with variables measured on different scales. PCA for quantitative traits revealed that leaf characters have major contribution to germplasm variability. For qualitative characters, ray florets traits have major contribution to germplasm variability. A group researcher conducted PCA of 35 morphological characters in 15 taxa of Chrysanthemum species and

identified 12 principle components explaining 99.4% of variation (Kim et al., 2014).

4.4 Cluster analysis based on Euclidean distance

Cophenetic correlation coefficient value equal to or greater than 0.85 is considered good ensuring the consistency of the dendrogram with the distance matrices (Stuessy, 1990). Cluster 1 consist of only one genotype i.e. Gauri which bear white colour small sized double type flower, longest plant height of 92.95 cm, petiole length (5.15 cm), smallest flower diameter (2.7 cm) and highest number of flowers per plant (400). Pusa Shwet alone belongs to cluster 2 which has white color semi double flower (2-3 rows of ray floret), long leaf (8.15 cm), wide leaf (6.27 cm), higher number of flower (231) and large flower diameter (7.75 cm). Cluster 3 constitute of 28 genotypes and demonstrated comparatively less mean values for plant height (39.88 cm), number of branches per plant (11.62) and number of flowers per plant (54.76). Moderate mean value for flower diameter was (6.3 cm) and peduncle length (7.15 cm). With respect to qualitative characters, most genotypes in cluster 3 showed upward petiole attitude, flat cross section of ray floret and round shape of tip of ray floret.

Cluster 4 comprise of 20 genotypes having adequate number of primary branches per plant with mean value 15.66, good number of flowers per plant with average value 139.72 and long peduncle with mean value 7.93. All the plants in cluster 4 are bushy type, majority of plants have downward petiole attitude and weakly convex cross section of ray floret. Fifteen taxa of Chrysanthemum species were classified into three groups through PCA and cluster analysis based on 35 qualitative and quantitative traits (Kim et al., 2014). Mean value of quantitative traits for different cluster is given in Table 6.

4.5 Cluster analysis based on jaccard's similarity index

The range of Jaccard's similarity coefficients (0.04 – 0.69) indicated significant genetic diversity between chrysanthemum cultivars. In cluster 1 all 5 genotypes are of non bushy types, large diameter double type flower, large stipule size, long and wide ray floret and a less number of flowers. All except Pusa Arunodaya exhibited incurving type of ray floret, moderately involute rolling of margins of ray floret and concave cross section of ray floret. Cluster 2 had single flower type, short plants, few numbers of flowers and large stipule size. Genetic relationships within chrysanthemum were partly indicated by their ray floret type was also shown (Chen et al., 2013; Mia et al., 2007). Cluster 3 had only genotype Gauri which showed Jaccard's similarity index of 0.38 or less with all the genotypes showing exclusive higher value for plant height (92.95 cm) and number of flowers (400). Petiole length, flower diameter, ray floret length and ray floret longitudinal axis in Gauri varied from majority of the genotypes. Cluster 4 showed higher number of primary branches, long and wide leaf, medium terminal lobe length of leaf and long lower lobe length. Clustering based on various leaf characteristics was also performed (Kim et al., 2014). Genotypes in cluster 5 exhibited short height, medium number of primary branches, short leaf length, low leaf length to width ratio and medium stipule size. Punjab Shyamal, the only cultivar in cluster 6 showed reflexing longitudinal axis of ray floret, 2 types of color on the inner side of ray floret, short ray floret along with long and narrow length. All the genotypes in cluster 7 showed long and narrow leaves with short petiole length and medium length of ray floret. In cluster 8 all cultivars exhibited bushy plant type, straight longitudinal axis of ray floret, medium leaf length to width ratio, medium width of ray floret and also all genotypes had medium to large size flowers. Majority of cultivars in cluster 9 have medium sized flowers, many primary branches, many number of flowers and flat margins of ray floret. The mean value of quantitative traits for different cluster is given in Table 7.

Table 6 : Mean values of clusters (based on Euclidean distance) for different characters

Cluster	1	2	3	4
Plant: Height(cm)	92.95	49.05	39.88	45.33
No. of primary Branches	14.5	15.05	11.62	15.66
Leaf Length (cm)	5.25	8.15	6.42	5.89
Leaf Width (cm)	3.9	6.27	4.8	4.5
Petiole length (cm)	2.68	1.82	1.86	1.86
Leaf: ratio length/width	5.15	3.3	2.5	2.47
Terminal Lobe length (cm)	3.15	2.99	2.99	2.61
Lower lobe length (cm)	2.35	2.6	2.62	2.61
Stipule size (cm)	1.26	0.8	1.33	1.01

Table 6 (cont): Mean values of clusters (based on Euclidean distance) for different characters

No. of flowers per plant	400	231	54.76	139.72
Flower head: diameter (cm)	2.7	7.75	6.3	6.02
Peduncle length (cm)	5.55	5.08	7.15	7.93
Ray floret: length (cm)	1.42	4	3.1	2.92
Ray floret: width (cm)	0.5	0.75	0.7	0.62

Table 7: Mean values of clusters (based on Jaccard's similarity index) for different characters

Cluster	1	2	3	4	5	6	7	8	9
Plant: Height(cm)	47.82	27.72	92.95	61.19	32.62	46.25	37.90	43.84	41.33
No. of primary Branches	6.8	6.82	14.5	20.5	9	9.5	12.07	13.26	15.29
Leaf Length (cm)	8.16	7.97	5.25	7.82	4.29	8.1	4.04	7.12	5.99
Leaf Width (cm)	6.6	5.07	3.9	5.86	4.05	4	3.05	5.44	4.48
Petiole length (cm)	2.91	5	5.15	1.77	1.25	2	1.52	3.47	2.40
Leaf: ratio length/width	1.69	2.57	2.685	1.63	1.32	2.53	1.83	1.95	1.87
Terminal Lobe length (cm)	4.15	3.85	3.15	2.67	2.14	2.16	2.29	3.41	2.54
Lower lobe length (cm)	3.65	4.02	2.35	4.25	2.59	2.1	1.86	3.41	2.17
Stipule size (cm)	2.15	2.25	1.265	2.49	1.55	0.47	0.64	1.27	0.96
No. of flowers per plant	49.6	36	400	117.05	52.75	67.5	81	81.14	113.68
Flower head: diameter (cm)	10.29	8.05	2.7	5.11	4.64	4.56	3.85	6.31	6.09
Peduncle length (cm)	6.81	6.52	5.55	10.37	6.39	11.4	5.73	7.11	7.78
Ray floret: length (cm)	5.23	4.65	1.425	2.25	2.49	1.43	1.98	2.90	2.99
Ray floret: width (cm)	1.46	0.28	0.5	0.39	0.5	0.57	0.48	0.62	0.62

5. CONCLUSION

The 50 genotypes of *Chrysanthemum* varied significantly for 25 Morphological characters. The PCVs were slightly higher than GCVs indicating correspondence between genotype and phenotype. PCA showed that among quantitative traits, leaf characters and **ray florets** characters among qualitative trait have major contribution to germplasm variability. Clustering of genotypes based on Euclidean distance and Jaccard's similarity coefficient gave 4 and 9 clusters respectively. Gauri was an outlier for plant height and number of flowers per plants and thus was grouped alone in both type of clustering. The morphological identification of genotypes will enable efficient utilization of genetic resources, selection of parents for hybridization that will help in improvement of desired traits.

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COMPETING INTEREST

No potential conflict of interest was reported by the author(s).

AUTHORS CONTRIBUTION

Conceptualization: Gunjeet Kumar, Vartika Budhlakoti; methodology: Gunjeet Kumar, Vartika Budhlakoti, A.K. Tiwari, V.M. Hiremath; data analysis: Gunjeet Kumar, Vartika Budhlakoti, V.M. Hiremath, Saipriya Panigrahi, Shreekant; writing: original draft preparation, Gunjeet Kumar, Vartika Budhlakoti, A.K. Tiwari, V.M. Hiremath; Review and editing: Gunjeet Kumar, Markandey Singh. All authors read and made suggestions which were incorporated in the final manuscript.

DATA AVAILABILITY

All data pertaining to this research is available in the manuscript or list of supplementary table.

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