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Genetic diversity and principal component analysis based on vegetative, floral and bulbous traits in narcissus (*Narcissus pseudonarcissus* L.)

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Abstract

The present investigation was carried out with the objective to assess the nature and magnitude of variability and extent of genetic divergence among different genotypes of narcissus on the basis of vegetative, floral and bulbous traits. Experimental results revealed significant differences among nine genotypes for different traits under study. Earliest scape emergence (64.43 days) and flowering (80.57 days) and highest bulb size (4.22 cm), weight of bulb (67.86 g) and bulblets (14.27 g) was recorded in the genotype Cumlaude, while largest flower size (8.37 cm) was observed in the cultivar Carlton. Further, significant positive correlations of scape length (0.825), weight of bulb (0.914) and root length (0.876) was recorded with bulb size. The principal component analysis (PCA) revealed that 90.97 % of total variations were explained by first four components. Further, loading of different traits based on first two principal components indicated that days to gose neck stage, bulb size, weight of bulb and root length are the main components of divergence among nine genotypes, whereas other traits had comparatively lesser contributions in the divergence. Based on the hierarchical cluster analysis, nine genotypes were grouped into two major clusters. Hence, crossing between the genotypes of these groups is expected to yield heterotic hybrids in future breeding programs.

Keywords: cluster analysis, correlation, dendrogram, genetic diversity, narcissus and PCA

Introduction

Narcissus (*Narcissus* sps.) is a bulbous perennial belonging to the monocotyledonous family Amaryllidaceae and considered as major ornamental crops of the temperate regions. The daffodil (*Narcissus pseudonarcissus* L.), is native to Western Europe, and is found in the area bounded by Portugal in the west, Germany in the east, and England and Wales to the north (Gul and Tahir, 2013) [9] and the highest diversity is found in the Iberian Peninsula and North Western Africa (Simon *et al.*, 2010) [14]. Based on consumer purchase figures, narcissus (daffodils and narcissi) were rated fourth in popularity and fifth in value of sales (www.flower.org.uk; Flower and Plants Association, Personal Communication, 2010) [16, 7]. The genus *Narcissus* consisting of about 63 species and many sub specific taxa and natural hybrids (Haddi-e-vincheh *et al.*, 2013) [10]. Various common names including daffodil, narcissus and Jonquil are used to describe all or some members of genus narcissus but the daffodil is now commonly used name for all the varieties of spring flowering bulbs in the genus *Narcissus* (Brickell, 1996; Spaulding and Barger, 2014) [4, 15]. Flowers are either solitary or in clusters of 2 or more, borne in spring sometimes autumn or winter. Leafless stems bear flowers each with 6 spreading perianth segments (petals), surrounding a corona which is also called as floral cup, tube or crown. The flowers are bright yellow, orange, red, pink, and white occasionally green (Spaulding and Barger, 2014) [15].

The different species and hybrids of *Narcissus* are popular as cut flower, garden flower, pot plant, rockeries and can be grown with little care in flower beds, around deciduous trees in temperate regions. Narcissus is also an important source of perfume (e.g. *N. poeticus*), lectins (important compounds in the study of plant-pathogen interactions) and a source of genes e.g. coding for phytoene synthase. The genus narcissus is well known for its morphological diversity due to vast amount of floral variations affecting both perianth and sex organs within and among species. Information on the magnitude of variability and extent of genetic divergence in a crop is an important consideration for planning any breeding programme and

ascertaining the scope of its improvement. Further the knowledge of interrelationship among different characters in the form of correlation is an important aspect for effective selection (Desh Raj *et al.*, 1997) ^[6]. The information on these aspects of narcissus in India particularly under agro-climatic conditions of Kullu-Valley of H.P. is completely lacking. Therefore, present investigation was carried out to assess the magnitude of variability and extent of genetic divergence among different genotypes of narcissus and to identify suitable genotypes for further use in future breeding programmes.

Materials and Methods

The present investigation was carried at ICAR-Indian Agricultural Research Institute (IARI) Regional Station, Katrain, Kullu-Valley, Himachal Pradesh, India during the year 2016-17 and 2017-18. The study was carried out on nine diverse genotypes of daffodils collected from different sources exhibiting significant differences in the characters considered for the study. The bulbs of nine daffodil cultivars (Fig. 1) viz. Carlton, Dick Wilden, Golden Ducot, Yellow Cheerfulness, Lemon Beauty, Prof. Einstein, Dbdam, Publeo and Cumlaude were planted each year during the month of November at the Sarsai Research Farm of the station under natural growing conditions in Randomized Block Design with three replications. Ten plants were selected for recording the observations and data were collected on 14 different parameters pertaining to growth, flowering and bulb production of different genotypes. Uniform package of practices were followed to raise the successful crop stand. The data recorded was subjected to statistical analyses using OP Stat and SPSS 16.0 software. The variability estimates were worked out through Analysis of Variance (ANOVA) in a Randomized Block Design, while correlation coefficients were determined by co variance and variance between the traits.

Results and Discussion

Mean performance

Evaluation of narcissus genotypes revealed that there was significant variation among the genotypes with respect to growth and bulb attributes. These variations are owing to genetic background of the genotypes. Data presented in table 1 divulged that different vegetative traits varied significantly among different genotypes of narcissus. Earliest scape emergence (64.43 days) was recorded in cv. Cumlaude while delayed scape emergence was noticed in cv. Publeo (102.47 days). Scape length varied significantly among the genotypes and recorded maximum in the cv. Golden Ducot (32.53 cm) followed by Dbdam (29.37 cm) while minimum scape length (16.37 cm) was observed in cv. Publeo. Siddique *et al.* (2012) ^[13] and Priyanka *et al.*, (2012) ^[11] had reported similar variation in scape length of narcissus genotypes. Genotype Yellow Cheerfulness was found superior for number of leaves per plant (5.98) which was at par with Lemon Beauty (5.18) and Cumlaude (5.78) while least number of leaves per plant were recorded in Carlton (3.47) followed by Golden Ducot (4.35) and Publeo (4.38). Longest leaves were observed in Golden Ducot (23.83 cm) which was at par with Cumlaude (23.50 cm) and Dbdam (22.17 cm). Similar variation in leaf length was also reported earlier by Addai (2011) ^[1] in Hyacinth. Leaf width was noticed maximum in Golden Ducot (1.57 cm) and it was found was at par with Dick Wilden (1.48 cm) and Yellow Cheerfulness (1.45 cm).

The evaluated genotypes exhibited wide variations for floral attributes (Table 1). Narcissus flower earliest among the temperate bulbous flowers, which is matchless characteristic and most liked. It was noticed that minimum number of days were taken to reach goose neck stage by Cumlaude (70.38 days) whereas maximum number of days to reach goose neck stage were taken by Dick Wilden (109.33 days). It may be due to different genetic composition and chilling requirement of the genotypes. These can be utilized for extending period blooming period of narcissus, which is a major constraint in its exploitation. Similar variations in days taken to reach the goose neck stage reported by Priyanka *et al.* (2012) ^[11] in narcissus cultivars under mid hill conditions of Himachal Pradesh. Earliest flowering was also noticed in cv. Cumlaude (80.57 days) whereas cv. Yellow Cheerfulness was the last to flower (122.87 days). Variation in days to flowering in narcissus was also observed by Fry (1975) ^[8] and Priyanka *et al.*, (2012) ^[11]. Earliness in narcissus genotypes was also reported by Rees *et al.* (1972) ^[12]. Flower size is an important quality criterion of narcissus cut flowers. The largest flower size (8.37 cm) was noticed in the cv. Carlton. Cohen *et al.* (2009) ^[5] also found variation in flower size, flowering time and other morphological traits in narcissus germplasm.

Wide variation was observed in bulb characteristics like size of bulb, size of bulblets, weight of bulb, weight of bulblets and root length (Table 2) among the genotypes. In narcissus bulb multiplication rate is slow, which hinders its commercial exploitation. Size of bulb was found significantly high in cv. Cumlaude (4.22 cm) and it was found at par with cv. Prof. Einstein (4.04 cm). Highest value for bulb weight 67.86 g, weight of bulblets (14.27 g) and root length (9.58 cm) was observed in cv. Cumlaude while for bulblets size it was noticed maximum (2.69 cm) in cv. Yellow Cheerfulness. Variability in bulb size and weight, size and weight of bulblets was also reported earlier by Priyanka *et al.* (2012) ^[11] in narcissus genotypes.

Correlation studies

The nature and magnitude of association between growth, flowering and bulbous parameters was worked out using Pearson's correlation coefficient (Table 3). Scape length revealed a highly significant and positive correlation with weight of bulb (0.825). Earlier, Bhatia *et al.*, (2013) ^[3] also observed highly significant and positive correlation of scape length with bulb size in tulip. Scape length was found to be negatively associated with bulb size, weight of bulblets and root length. Days to scape emergence exhibited significant negative association with days to goose neck stage, days to flowering, and bulblets size. Days to goose neck stage showed highly significant and positive correlation with days to flowering (0.940). Flower size revealed negative association with number of bulblets per plant. Narcissus is commercially propagated via bulbs. However, multiplication rate in narcissus is quite poor. So there is a need to improve the multiplication potential either by selecting the genotypes with high bulb production coefficient or via indirect improvement in the traits contributing for number of bulbs per plant. Number of bulblets showed highly significant and positive correlation with size of bulblets (0.749). Further trait like bulb size revealed highly significant and positive correlation with weight of bulb (0.914) and root length (0.876). Size of bulblets exhibited a significant and positive correlation with weight of bulblets (0.840). While, weight of bulbs revealed a highly significant and positive correlation with weight of bulblets (0.749) and root length (0.893). It indicated that improvement

in these traits could lead to an increase in number of bulbs per plant. Similar trends of associations of bulbous traits have been earlier reported in tulip by Bhatia *et al.* (2013) [3].

Principal component and cluster analyses

In the present study, first four principal components with eigen values more than one contributed to 90.97 per cent of cumulative variability among the nine genotypes evaluated for 14 morphological and bulbous traits (Table 4). The first principal component accounted for 45.65 per cent of variance, while the second accounted for 28.75 per cent of total variability. The per cent of variability from third and fourth principal components accounted for 8.80 and 7.77 in decreasing order, respectively. It was therefore inferred that essential features of data set had been represented in the first four PCs. Number of bulblets (0.56), size of bulb (0.85), size of bulblets (0.72), weight of bulb (0.92), weight of bulblets (0.89) and root length (0.86) explained the maximum variance in PC1. The PC2 which accounted for 28.75 per cent of total variance showed higher variance for days to scape emergence (0.80), days to goose neck stage (0.92), days to flowering (0.91) and flower size (0.41) signifying their importance for

quality improvement in narcissus. The PC3 reflected significant high loading for traits like number of leaves (0.87) and leaf length (0.13). The PC4 showed significant variance for scape length (-0.11) and leaf width (0.41). Further, loading of different traits based on first two principal components indicated that days to goose neck stage, bulb size, weight of bulb and root length are the main components of divergence among nine genotypes, whereas other traits had comparatively lesser contributions in the divergence (Fig 2). Similar results related to principal component analysis in tulip were also reported by Bhatia *et al.* (2017) [2].

Based on the hierarchical cluster analysis, nine genotypes were divided into two major clusters at a distance of 25 (Fig 3). The first cluster had one genotypes i.e. Cumlaude. The second cluster had two sub-clusters with one genotype in first sub-cluster (Pubelo) and rest of the seven genotypes (Dick Wilden, Carlton, Golden Ducot, Dbdam, Prof. Einstein, Lemon Beauty, and Yellow Cheerfulness) were accumulated in second sub-cluster. Clustering pattern of the dendrogram would facilitate the selection of suitable cultivars for future breeding programs. Similar results were also reported by Bhatia *et al.* (2017) [2] in tulip.

Table 1: Mean performance of daffodil genotypes for different vegetative and floral traits

Genotypes	Days to scape emergence(days)	Scape length (cm)	Number of leaves	Leaf length (cm)	Leaf width (cm)	Days to goose neck stage (days)	Days to flowering (days)	Flower size (cm)
Carlton	68.50	27.73	3.47	17.00	1.37	94.93	97.45	8.37
Dick Wilden	73.42	18.67	4.35	19.30	1.48	109.33	117.50	6.36
Golden Ducot	93.17	32.53	3.67	23.83	1.57	102.67	110.33	6.68
Yellow cheerfulness	98.72	22.90	5.98	16.13	1.45	107.37	122.87	6.93
Lemon Beauty	98.10	24.47	5.18	17.60	1.43	107.83	118.30	7.08
Prof. Einstein	93.47	26.55	5.37	17.87	1.37	98.43	107.52	7.92
Dbdam	101.57	29.37	4.90	22.17	1.43	106.56	110.13	7.93
Pubelo	102.47	16.37	4.38	13.47	0.58	108.07	110.17	3.72
Cumlaude	64.43	24.88	5.78	23.50	1.43	70.38	80.57	4.13
CD at (P=0.05)	2.90	2.81	0.72	2.07	0.13	1.16	1.21	0.72

Table 2: Mean performance of daffodil genotypes for different bulbous traits

Genotypes	Number of bulblets	Size of bulb (cm)	Size of bulblets (cm)	Weight of bulbs(g)	Weight of bulblets (g)	Root length (cm)
Carlton	0.10	3.90	0.89	47.05	3.33	6.87
Dick Wilden	0.10	3.40	1.49	33.07	4.17	4.62
Golden Ducot	0.00	3.45	0.00	36.67	0.00	3.19
Yellow cheerfulness	0.80	3.63	2.69	37.00	9.33	3.32
Lemon Beauty	0.28	3.87	2.01	42.67	5.00	6.08
Prof. Einstein	0.73	4.04	2.07	43.50	4.55	6.69
Dbdam	0.33	3.65	0.53	36.47	1.38	6.75
Pubelo	0.40	2.58	0.57	15.23	0.50	1.45
Cumlaude	0.65	4.22	2.35	67.86	14.27	9.58
CD at (P=0.05)	NS	0.54	1.67	9.58	6.25	1.74

Table 3: Pearson's correlation (r) among different morphological and bulbous traits in daffodil cultivars

Characters	Days to scape emergence (days)	Scape length (cm)	Number of leaves	Leaf length (cm)	Leaf width (cm)	Days to goose neck stage (days)	Days to flowering (days)	Flower size (cm)	Number of bulblets	Size of Bulb (cm)	Size of Bulblets (cm)	Weight of bulbs (g)	Weight of bulblets (g)	Root length (cm)
Days to scape emergence (days)		-0.006	0.173	-0.316	-0.314	-0.691*	-0.682*	0.100	-0.541	-0.144	-0.668*	-0.115	-0.204	0.175
Scape length (cm)			-0.200	0.665	0.644	-0.223	-0.222	0.588	-0.554	0.839**	-0.554	0.825**	-0.687*	-0.862**
Number of leaves				-0.018	0.113	-0.245	0.009	-0.181	-0.054	0.122	-0.093	0.025	-0.009	0.038
Leaf length (cm)					0.665	-0.463	-0.417	0.058	0.054	-0.698*	-0.219	-0.750*	-0.434	-0.924**
Leaf width (cm)						-0.173	0.005	0.585	-0.277	-0.554	-0.431	-0.824**	-0.820**	-0.691*
Days to goose neck stage (days)							0.940**	0.324	-0.381	0.126	-0.531	0.208	-0.310	0.416

Days to flowering (days)								0.317	-0.362	0.061	-0.591	0.076	-0.418	0.369
Flower size (cm)									-0.809**	-0.297	-0.597	-0.453	-0.796*	-0.282
Number of bulblets										0.280	0.749*	0.250	0.642	0.196
Size of Bulb (cm)											0.536	0.914**	0.607	0.876**
Size of Bulblets (cm)												0.486	0.840**	0.373
Weight of bulbs (g)													0.749*	0.893*
Weight of bulblets (g)														0.568
Root length (cm)														

*Significant at $p < 0.05$, **Significant at $p < 0.01$

Table 4: Component loading of 14 traits, eigen values, proportion of total variability represented by first four principle components (PC), cumulative variability in nine daffodil genotypes.

Characters	Principal component*			
	PC ₁ #	PC ₂	PC ₃	PC ₄
Days to scape emergence (days)	-0.08	0.80	0.41	-0.38
Scape length (cm)	-0.91	-0.17	-0.12	-0.11
Number of leaves	0.05	0.00	0.87	0.48
Leaf length (cm)	-0.69	-0.58	0.13	-0.17
Leaf width (cm)	-0.82	-0.18	-0.04	0.41
Days to goose neck stage (days)	0.02	0.92	-0.17	-0.19
Days to flowering (days)	-0.06	0.91	0.04	-0.04
Flower size (cm)	-0.67	0.41	-0.41	0.41
Number of bulblets	0.56	-0.60	0.03	-0.20
Size of Bulb (cm)	0.85	0.16	-0.12	0.42
Size of Bulblets (cm)	0.72	-0.62	-0.21	0.09
Weight of bulbs (g)	0.92	0.25	-0.05	0.07
Weight of bulblets (g)	0.89	-0.33	0.04	-0.20
Root length (cm)	0.86	0.45	-0.10	0.18
Eigen Value	6.39	4.02	1.23	1.08
Percentage of variance	45.65	28.75	8.80	7.77
Cumulative % of variance	45.65	74.40	83.20	90.97

#PC: Principal component

*Extracted through principle component analysis

**Bold value indicates the highest Eigen vector for the corresponding charcters amongst the four principal components

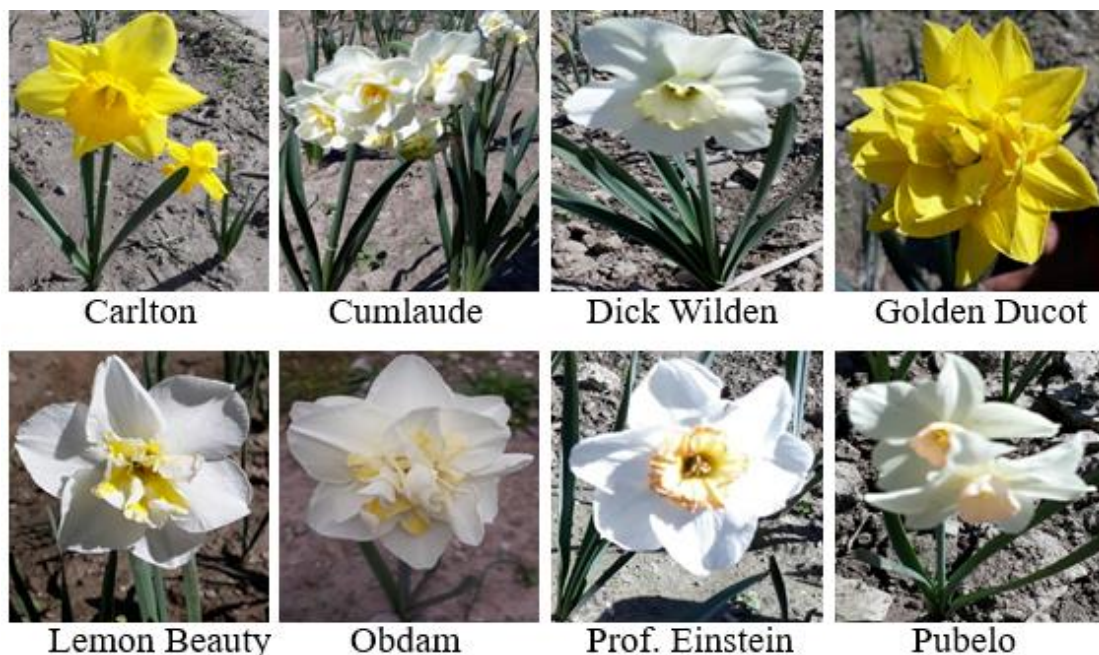


Fig 1: Daffodil cultivars used for present study

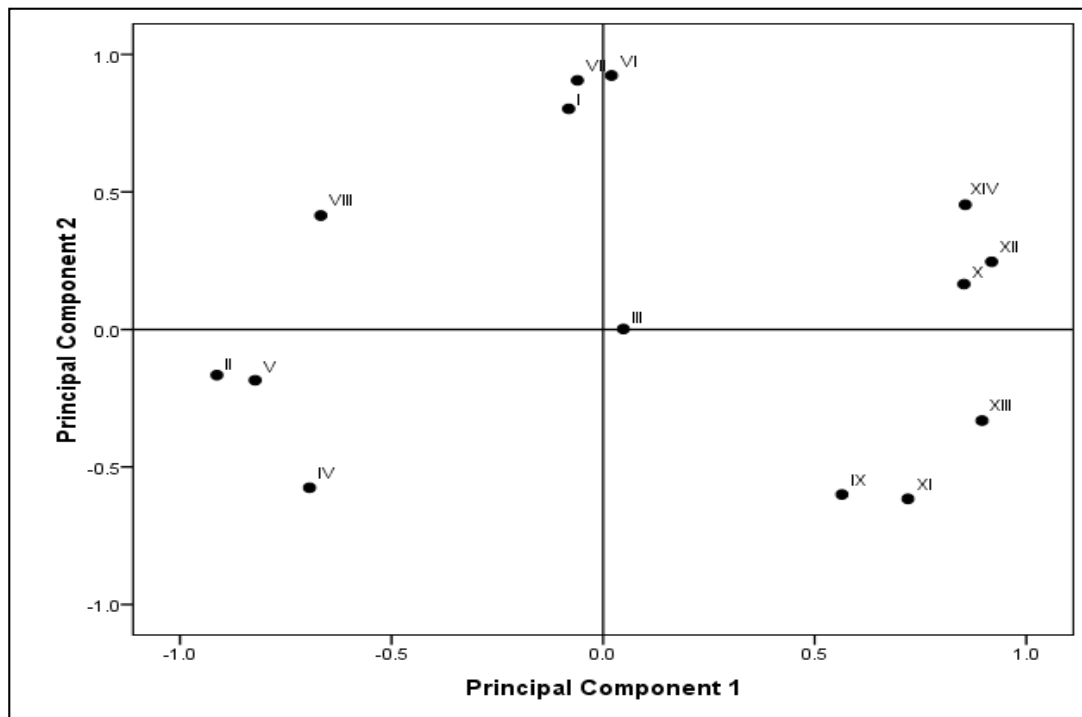


Fig 2: Loading of different traits based on first two principle components

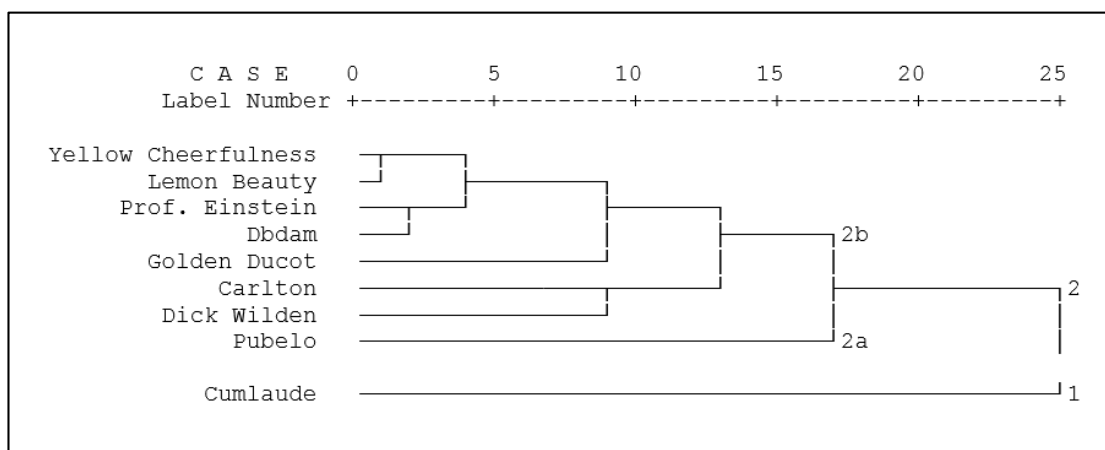


Fig 3: Dendrogram showing clustering pattern of 9 daffodil cultivars based on 14 morphological and bulbous traits constructed using complete linkage Euclidean distance method

Conclusion

The study revealed wide variations for majority of traits indicating sufficient genetic variability to be exploited in future breeding programs. The commercially important traits like scape length, days to goose neck stage, number and weight of bulb and bulblets exhibited a high degree of significant positive correlations. Hence, direct selection from germplasm lines may be effective for improving the cut flower quality. Besides this, hierarchical cluster analysis grouped nine genotypes into two major clusters. Hence, distant parental lines can be selected from these for F₁ hybrid breeding in narcissus.

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