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COLLECTION, CONSERVATION AND CHARACTERIZATION OF *CYMBIDIUM TRACYANUM* (L.) CASTLE FROM DARJEELING (WEST BENGAL)

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Abstract

Cymbidium tracyanum (L.) Castle is an early flowering, large flowered and scented orchid. Its 20 accessions from different parts of Darjeeling were studied for morphological characters. The results revealed wide variation in plant height, plant spread, pseudobulb size, inflorescence length, and flower count. The distance among various genotypes was determined by Hierarchical cluster analysis using Euclidian distance. The highest value (76.49) was observed for OCD - 1110 and the lowest (5.85) for OCD - 1135. The dendrogram revealed 4 clusters. The cluster 2 had largest membership followed by the clusters 1 and 3 and 4. The genotype OCD - 1110 was not part of any cluster.

Introduction

DARJEELING DISTRICT of West Bengal lies between 26° 31' and 27° 13' north latitude and between 87° 59' and 88° 53' east longitude. It comprises deep valleys and high hills with a wide spectrum of altitutional variation (300-3600 m). The district supports a unique diversity of climatic conditions ranging from subtropical to alpine. High humidity and sufficient sunlight are congenial factors for growth and development of cymbidiums.

Cymbidium is a genus of nearly 60 species, 6 of which (*Cymbidium eburneum, C. erythrostylum, C. hookerianum, C. insigne, C. lowianum,* and *C. tracyanum*) are responsible for the vast majority of our modern day hybrids. *C. tracyanum* is native to Myanmar, China and Malaysia. It is a large flowered, early flowering and fragrant species, and has been used in developing several cultivars like *Cymbidium wiganianum* (*C. eburneum* x *C. tracyanum*), *Cymbidium woodlandsense* (*C. mastersii* x *C. tracyanum*), *Cymbidium hanburyanum* (*C. erythrostylum* x *C, tracyanum*). It is mainly used as pollen rather than pod parent *(http://www. Geocities/pennypoint9/tracy.html)*. The species is under cultivation in some private nurseries and personal collections in Darjeeling. The advent of new exotic hybrids having greater appeal are exerting pressure on species and old varieties. Their sustainable development is desirable. The information on characterization and conservation of this species either on the basis of quantitative morphological characters or molecular basis is lacking. Hence, an attempt was made to collect, conserve and characterize it on the basis of morphological characters.

Material and Methods

The explorations for collection of *C. tracyanum* genotypes were conducted in five community development blocks namely Sukhia-Pokhari, Takdah, Kurseong, Mirik and Rungli Rungliot of Darjeeling district of West Bengal during the flowering season of 2001 - 2003. Twenty accessions were collected from the private nurseries and personal collections. The block wise collections were 5 from Kurseong, 6 from Sukhia-pokhari, 6 from Mirik, and 3 from Rungli and Rungliot (Takdah) (Fig. 1). The full grown plant or plant part having at least one backbulb, two leads and 1-2 vegetative shoots were collected. The duplication of collections was avoided by visual observation. The accessions were brought to National Research Centre for Orchids, Darjeeling Campus, Darjeeling (2150 m) and cleaned from dried or decayed leaves. If necessary, divided and then soaked in aqueous solution of Bavistin (1.5 g l-1) for 30 min and air dried. The plants were grown in plastic pot (12 inch dia) in potting mixture containing FYM: perlite: leaf mould (4: 1: 4) v/v. The watering was done at weekly/fortnightly intervals to keep potting mixture moist. The plants were fertilized with NPK (30:10:10) during active period of growth (March - September) and NPK (10:30:10) during flowering/ dormant season (October - February) with irrigation at 15 days interval. The accessions were grown in naturally ventilated polyhouse to protect them from rain. The observations on vegetative characters *i.e* plant height, plant spread, pseudobulb length, pseudobulb circumference, pseudobulb diameter, leaf length, leaf width, and reproductive characters namely inflorescence per pseudobulb, inflorescence length, raceme length, and flower number, flower height (dorsal sepal to lip), flower width (sepal to sepal distance) flower size across, dorsal sepal length, pedicel diameter and pedicel length were recorded for two consecutive years 2009 and 2010. The flower parts of the *Cymbidium tracyanum* and important observed parameters are shown in (Fig 2).The collected data were analyzed for range, mean, standard deviation and coefficient of variation. The various

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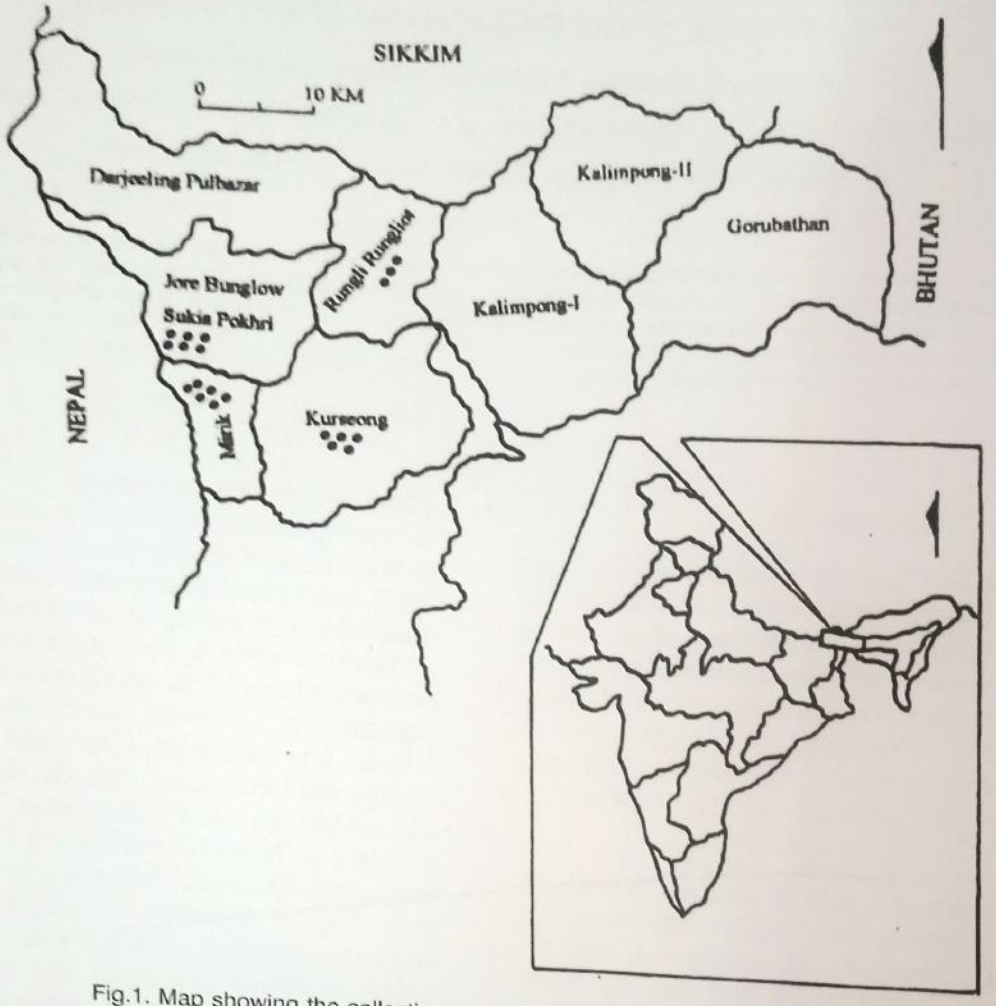

Fig.1. Map showing the collection sites of *Cymbidium tracyanum* genotypes.

morphological parameters were subjected to Hierarchical cluster analysis using Euclidian distance (SPSS 12.0 Version).

Results and Discussion

The accessions showed wide variability with respect to different morphological traits (Table 1). The plant height varied from 26.2 - 67.8 cm, plant spread from 24.6 - 57.4 cm, leaf length from 43.2 - 76.3 cm and sheath length from 5.8 - 10.2 cm. In sympodial orchids, like *Cymbidium tracyanum* the inflorescence or vegetative shoots arise from the base of pseudobulb. The length, width and circumference are a good indicator for this purpose. The length of pseudobulbs was found between 6.0 cm and 12.5 cm, diameter between 2.62 cm and 4.96 cm, and circumference between 12.3 cm and 18.3 cm. The plant height was found to be most variable trait (cv 25.35 %) and it was followed by plant spread (cv 22.23 %). The least variation was seen in functional leaves (cv 9.88 %). In floricultural crops especially for cut flower production, the prime concern relates to inflorescence and its attributes. The market requires optimum flower count (not less than 12), flower balance (60 flowering zone: 40 non flowering zone) sturdy and

a, Flower height; b, Flower width; c, Lip lobe distance

Fig. 2. Showing floral parts of *Cymbidium tracyanum* and important observed parameters

Table 1. Plant parameters in *Cymbidium tracyanum* grown *ex situ*.

Plant Characters	Range	Mean*(± Std. Deviation)	*CV
Plant height (cm)	26.20 - 67.80	31.19 ± 9.68	25.35
Plant spread (cm)	24.60 - 57.40	41.03 ±9.122	22.23
Pseudobulb length (cm)	6.00 - 12.50	9.05 ±1.730	19.12
Pseudobulb diameter (cm)	2.62 - 4.96	3.74 ±0.515	13.81
Pseudobulb circumference (cm)	12.3 - 18.20	15.27 ±1.669	10.93
Functional leaf number	8.00 - 11.00	9.33 ±0.922	9.88
Leaf length (cm)	43.20 - 76.30	60.01 ±8.797	14.66
Leaf width (cm)	1.80 - 3.60	2.72 ±0.475	17.42
Sheath length (cm)	5.80 - 10.20	8.12 ±1.109	13.66

Average of (n=20) ± standard deviation of the mean
*Coefficient of variation

straight stem (10 mm diameter), and colour (depending on the market) with fragrance as an added advantage. Hence, a breeder will look for the traits which could fulfill these objectives. The observation recorded with respect to flower spike parameters are presented in Table 2. The variation in length of inflorescence found between (48.6 - 99.1 cm), raceme or flowering zone (20.9 - 64.3 cm), non flowering zone or stalk length (21.0 - 51.0 cm), inflorescence diameter at base (0.72 - 1.48 cm) and flower count (6.6 - 13.5). The inflorescence in these accessions varied from arching to straight. The flowers were variously spaced (3.5 - 5.0 cm) on the inflorescence. Among the inflorescence characters the raceme length was most variable (cv 25.79 %) followed by bract number (cv 21.5 %) and stalk length (cv 20.78 %). The preferred raceme and stalk ratio for cut flower cultivar is (1.5). The genotype OCD - 1110 had a ratio of 1.85 and it was followed 1.45 in the genotype OCD - 1187. A proper selection of parents in hybridization may yield better cultivars as per demand of the market. The flowers of *Cymbidium tracyanum* were spreading and greenish to greenish yellow in colour. The sepals and petals were greenish yellow dotted or dotted streaked with dark maroon color. The lip was hairy white to ochre yellow in color. The data on flower parameters are mentioned in Table 3. The flower size across varied from 9.40 to 12.40 cm. Veitch had found *Cymbidium lowianum*, *C. giganteum* (= *iridoides*) and *C. tracyanum* to be pod-sterile, and only useful as pollen-parents. However, of the eleven hybrids registered from

Table 2. Inflorescence characters of *Cymbidium tracyanum* grown *ex situ*.

Plant Characters	Range	Mean* (± S.D.)	CV
Inflorescence length (cm)	48.60 - 99.10	72.37 ±14.391	19.89
Raceme length (cm)	20.90 - 64.30	37.95 ±9.787	25.79
Stalk length (cm)	21.00 - 51.00	34.42 ±7.153	20.78
Inflorescence diameter at base (cm)	0.72 - 1.48	0.96 ±0.183	19.21
Bract number	4.00 - 9.00	5.85 ±1.258	21.5
Bract length (cm)	8.50 - 17.40	13.31 ±2.737	20.56
Flower number	6.60 - 13.50	9.68 ±1.928	19.93
Flower node distance	3.50 - 5.00	4.16 ±0.405	19.89

Average of (n=20) ± standard deviation of the mean

Table 3. Flower parameters of *Cymbidium tracyanum* grown under *ex situ* conditions.

Plant Characters	Range	Mean* (± Std. Dev)	CV
Flower height (cm)	3.20 - 4.70	3.79 ±0.425	11.22
Flower width (cm)	9.50 - 11.50	10.35 ±0.533	5.15
Flower size across (cm)	9.40 - 12.40	10.90 ±0.724	6.64
Lip lobe distance (cm)	1.60 - 2.80	2.47 ±.0297	12.02
Dorsal sepal length (cm)	6.00 - 7.70	6.93 ±0.461	6.67
Pedicellate ovary length (cm)	4.50 - 6.80	6.16 ±0.665	10.81
Pedicellate ovary diameter (cm)	0.24 - 0.44	0.38 ±0.053	14.00

Average of (n=20) ± standard deviation of the mean

1946-1960, only three appear to have had the *C. tracyanum* as the pod-parent, and of the five hybrids registered from 1961-1970, none had *Cymbidium tracyanum* as pod-parent. It would therefore seem that *C. tracyanum* can be used as a pod parent, although it clearly works better as a pollen-parent. Perhaps there is variability in fertility between different clones .Vij and Shekhar (1987) reported a somatic chromosome number of 2n=40 in *C. tracyanum*. The other Indian species having similar chromosome number are *C. aloifolium*, *C. bicolor*, *C. cochleare*, *C. cyperifolium*, *C. dayanum*, *C. devonianum*, *C. eburneum*, *C. elegans*, *C. ensifolium*, *C. gammieanum*, *C. iridoides* (=*giganteum*), *C. hookerianum* (=*grandiflorum*), *C. lowianum*, *C. mastersii*, *C. pendulum*, and *C. whiteae*.

The morphological data collected from various genotypes were subjected to Hierarchical cluster analysis using Euclidian distance to determine the distance among various genotypes (Table 4). A range of 5.85 to 76.49 Euclidian distance values were observed. The OCD - 1110 had the highest (76.49) and OCD - 1135 had lowest (5.85) Euclidian distance value. The genotype OCD - 1186 and OCD - 1135 (5.85), OCD - 1182 and OCD - 2134 (6.84) and OCD - 1161 and OCD - 2134 (8.04) were found to closely related. The genotype OCD - 1110 was found to have highest dissimilarity value and appears to be distantly related with other genotypes. The dendrogram revealed 4 clusters (Fig. 3). The cluster membership and desirable traits of each group are summarized in Table 5. The genotype OCD - 1110 did not cluster with any of the group. The grouping of germplasm in to different

Table .5. Cluster membership and desirable traits of each cluster.

Cluster Number	Genotypes	Desirable traits of the group
I	OCD-1158; OCD-1179; OCD-2149; OCD-1171; OCD-1151	Small size plants with small pseudobulbs comparatively less fleshy having short inflorescence and raceme and stalk length, low flower count.
II	OCD-1186; OCD-1191; OCD-1145; OCD-1135; OCD-1187; OCD-1185	Tall plants with medium size pseudobulb having long flower spike with long raceme with long stalk, higher flower bud count.
III	OCD-1193; OCD-1133; OCD-1161; OCD-1182; OCD-2134	Medium height plants having long pseudobulbs of large girth, medium size flower spike having medium size raceme and high flower bud count.
IV	OCD-1141; OCD-2138; OCD-2117	Short height plants with medium size pseudobulbs having medium size flower spike with medium size raceme, moderate flower bud count.
V	OCD-1110	Tall plant with medium size pseudobulb; very long flower spike, very long raceme and medium size flower stalk and high flower bud count.

Table 4. Dissimilarity matrix showing Euclidean distance among various genotypes of *Cymbidium tracyanum*.

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20
OCD-1158	0.00																			
OCD -1185	53.34	0.00																		
OCD-1186	38.42	19.81	0.00																	
OCD-1193	45.67	27.21	24.30	0.00																
OCD-1133	53.64	30.11	29.66	18.19	0.00															
OCD-1141	24.24	34.24	21.08	37.59	42.28	0.00														
OCD-1110	76.49	43.14	42.85	53.23	56.11	59.89	0.00													
OCD-1191	57.33	24.11	21.89	31.23	34.31	40.76	24.36	0.00												
OCD-1179	14.32	49.73	35.25	39.77	51.46	27.52	71.27	53.01	0.00											
OCD-1145	50.20	22.76	15.25	34.28	34.23	30.87	33.77	17.07	48.02	0.00										
OCD-1161	45.28	21.00	16.54	12.92	16.25	32.53	46.49	23.16	41.83	24.30	0.00									
OCD-1135	37.75	21.19	5.85	21.84	30.44	23.00	42.61	21.61	33.03	18.68	16.32	0.00								
OCD-1187	39.15	26.35	10.58	30.34	33.17	22.53	41.05	22.45	37.62	12.66	21.56	13.19	0.00							
OCD-1182	48.12	20.68	18.06	18.67	14.80	33.09	47.86	24.68	45.97	22.91	9.21	19.59	22.85	0.00						
OCD-2149	21.54	48.60	38.10	35.21	47.59	32.29	75.84	55.17	16.21	52.66	39.63	35.44	42.92	43.90	0.00					
OCD-2134	51.03	17.73	18.89	16.66	14.96	36.00	45.01	22.29	47.65	23.04	8.04	19.69	23.92	6.85	45.57	0.00				
OCD-2138	38.84	26.52	17.08	24.61	22.81	24.48	55.15	33.52	37.94	24.44	18.43	19.83	20.17	15.50	38.54	19.00	0.00			
OCD-2117	32.89	28.51	16.91	21.79	23.57	21.48	56.23	33.06	33.21	27.53	16.18	17.94	20.16	16.21	31.76	20.40	12.91	0.00		
OCD-1171	11.41	53.66	38.18	46.25	55.36	26.13	75.11	57.51	10.98	49.83	46.71	37.26	39.42	49.55	23.25	52.01	39.08	35.90	0.00	
OCD-1151	13.98	45.26	31.90	35.80	45.53	22.73	71.68	51.32	11.64	45.50	37.28	30.46	35.43	40.06	13.41	42.52	31.29	26.52	14.45	0.00

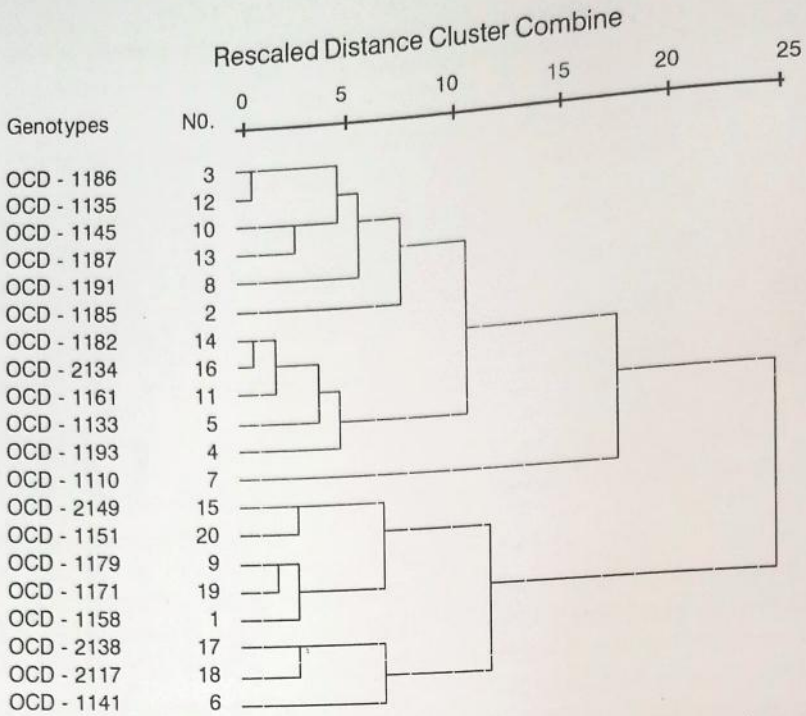

Fig. 3: Dendrogram illustrating dissimilarity among genotypes of *Cymbidium tracyanum* (L.) Castle.

clusters was not found to have relation with the place of collection. It suggests that plants have been frequently exchanged among the growers. Hence, a lot of variability exists with respect plant and inflorescence and flower characters and efforts should be made to utilize the variability available in the species for developing superior quality of hybrids. Parab and Krishnan (2008) assessed the genetic variation among populations of *Rhynchostylis retusa* using IISR and RAPD markers and concluded that higher genetic variability exists among the population of the species within small region. Similarly, variability in the population of *Bletia purpurea* was documented by Rene and Victoria (2002). Rao (2006) also observed that some of the species exhibit lot of variation in wild population which are very useful for breeding programmes to develop hybrids of commercial value in the international market.

Conclusion

Though the breeding of cymbidiums has not yet received momentum but the efforts to evolve new cultivars are continuing with several institutions in the country. *C. tracyanum* showed wide variability with respect to various morphological characters hence a careful selection of genotypes should be made using it as either parent for hybridization.

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