Electronic Journal of Plant Breeding, 8(1): 324-330 (March 2017)
ISSN 0975-928X
DOI: 10.5958/0975-928X.2017.00048.5

Research Note

Classification and characterization of chilli (*Capsicum annuum* L.) found in Manipur using multivariate analysis

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(Received: 11 July 2016; Revised: 10 Feb 2017; Accepted: 22 Feb 2017)

Abstract
The present study was undertaken to understand and find out the morphological characters contributing to the diversity of chilli germplasm of Manipur. A total of 20 chilli cultivars were collected from different districts of Manipur and characterized using 41 morphological characters (both qualitative and quantitative) based on IPGRI Descriptor for chilli. Cluster analysis using NTSYS revealed grouping of 20 chilli cultivars into 3 major groups at a distance co-efficient of 0.04 - 0.05. The first major group consist of 5 cultivars which are all bigger chilli cultivars and the second major cluster consist of 4 cultivars which are mainly consume in daily cuisine and the last major cluster consist of 11 cultivars which are small to medium in size. The first 2 principal components explained 97% of total variance. Based on eigen values greater than (±0.6), all the 41 characters used were informative and contributes highly to chilli diversity. So, the present study revealed that all the 41 morphological characters proposed by IPGRI descriptors can be successfully used in classifying bigger chilli cultivars from medium and small size chilli and daily used chilli cultivars.

Key words
Characterization, chilli, cluster analysis, descriptor, germplasm, Manipur

Chilli (*Capsicum annuum* L.), a native crop of tropical America and West Indies, which is also known as Capsicum, Papkrika, Cayenne, etc., belongs to family Solanaceae. It was first introduced in India by Portuguese towards the end of 15th century (Basu and Krishna, 2003). India is considered to be secondary centre of diversity for chilli (IBPGR, 1983) and it contributes one fourth of total world production of chilli (FAOSTAT, 2012). North-eastern states are home to much genetic variability where several interspecific hybrids/derivatives of chilli originated (Daliwal, 2014). It is believed that wide spectrum of chilli cultivars available in Manipur may be due to the fact that Manipur is at the confluence point of Indian and South Asian civilizations. Taxonomic identification of chilli using morphological characters is difficult as it displays wide variations and also influence by environmental factors. However, characterizations using morphological traits/characters are the basic and most cost effective method used in classification and evaluation of the germplasm (Smith *et al.,* 1991). Multivariate analysis using clustering and principal component analysis is the most widely used method (Mohammadi and Prasanna, 2007). To this effect, genetic diversity analysis of chilli using morphological traits has proven useful and has been reported by many researchers.(Chandal et al., 2016; Jyothi et al., 2011; Dudonte et al., 2008; Peeraullee and Sanmukhiya, 2013; Deka et al., 2016).

Since, less work has been reported on chilli diversity from Manipur, the present work was undertaken with the objectives for proper collection, characterization and to estimate the level of chilli diversity using morphological characters.

The experiment consists 20 chilli cultivars collected from nine (9) districts (Imphal West, Imphal East, Thoubal, Tamenglong, Bishnupur, Senapati, Chandel, Ukhrul and Churachandpur) of Manipur (Table 1). The chilli cultivars were cultivated and maintained during July to October (2015) in a germplasm with 2×2 ft spacing in rows under the same environmental condition for continuous evaluation and further analysis.

Morphological characters: All the 41 morphological descriptors proposed by IPGRI (1983) were used to score the chilli cultivars which include both qualitative and quantitative characters. The characters were scored according to capsicum descriptors and actual data were used for morphological analysis. The characters used were Hypocotyl, Cotyledonous leaf colour, Cotyledonous leaf shape, Stem colour, Nodal anthocyanin (whole plant), Stem shape, Stem pubescence, Plant height, Plant growth habit, Branching habit, Leaf density, Leaf colour, Leaf shape, Lamina margin, Leaf pubescence, Number of flowers per axil Flower position, Corolla colour, Corolla spot colour, Male sterility, Calyx pigmentation, Calyx margin, Calyx annular constriction, Anthocyanin spots or stripes, Fruit colour at intermediate stage, Fruit set, Fruit colour at mature stage, Fruit shape, Fruit width, Fruit shape at pedicel attachment, Neck at base of fruit, Fruit blossom end appendage, Fruit cross-sectional, corrugation, Fruit surface, Fruit shape at blossom end, Pedicel with stem, Seed colour, Seed

surface, Seed size, Pedicel with fruit and number of seeds per fruit.

Cluster and Principal component analysis: Hierarchical cluster analysis was done using unweighted pair group method (UPGMA) (Sneath and Sokal, 1973). From the morphological data a simple matching (SM) correlation coefficient using 'Simqual' module was used to generate the pairwise similarity matrix. All the statistical analysis was carried out using Numerical Taxonomy and Multivariate Analysis System (NTSYS-PC) software (Rohlf, 2000). Principal components (PCs) containing information from all the characters in varying proportion was extracted using principal component extraction methods (Sneath and Sokal,1973) and the principal components showing the highest variation was used for representing the distribution of samples in a 2-D and 3D scatter plot. The statistical analysis was performed with using IBM SPSS (Statistical Package for Social Sciences) version 20.0.

Cluster analysis: The dendrogram generated by Unweighted Pair Group Method using Average (UPGMA) based on SM (Simple Matching) correlation showed clustering of 20 chilli cultivars into three major groups at a distance co-efficient ranging from 0.06 to 0.07 (Figure 1). The first major group consist of 5 cultivars -U morok, Chakhou u-morok, U morok achouba, U morok macha and Bell shape which are all bigger chilli cultivars and the second major cluster consist of 4 cultivars namely - Meitei morok, Morok asangbi, Yensang morok, Churachandpur morok (which are mainly consume in daily cuisine) and the last major cluster consist of 11 cultivars which are small to medium in size namely- U-chimorok, small round chilli, Ukhrul morok asangbi, black bird eye, Morok macha,Crispy long, Fireball, Meitei morok macha,Colour xanadu, Meitei morok asaba, Meitei morok 1. From this result, it can be interpret that using 41 morphological characters, bigger chilli cultivars can be successfully differentiate from small and medium size chillies. Similar work has been reported by Wang and Bosland, 2006 that fruit size played an important role in variability of chillies.

The positions of 20 samples in the scatter plot with respect to 1st/ 2nd principal component in 2D and 3D from the original similarity correlation showed the samples fell into three (3) groups similar to groups formed by the cluster analysis (Fig 2 and Fig 3).

Principal component analysis: Based on the eigen value greater than one as a measure of significance of the principal component analysis (PCA), two (2) principal components were extracted which accounted for 97.9% of total variance in the present study (Table 3). The first principal component explained 73% of total variance and the second principal component explained 32.8% of total variance. Eigen vectors of each of the principal components revealed that all the 41 morphological characters contribute significantly. The 41 characters whose eigen vector were greater than ±0.6 were found to be useful in differentiating the chilli cultivars (Table 4). Similar works were also reported by Rego et al., 2003; Lefebvre et al., 1993 that principal component values/scores used for genetic divergence studies in chilli to avoid replicates and narrow down the variables.

The present study describes and estimates the extent of variation among chilli cultivars found in Manipur using 41 characters of IPGRI descriptors. It confirmed high, heritability and variation in 20 samples suggesting that classification based cluster analysis and principal component analysis is a good way to classify chilli cultivars. The information gathered in this study will be useful in future cultivar identification, breeding programme and sustainable utilization.

Acknowledgement
The authors are thankful to DBT for the DBT-RA fellowship and also express gratitude to Department of Plant Breeding and Genetics, CAU, Imphal for lab facility.

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Electronic Journal of Plant Breeding, 8(1): 324-330 (March 2017)
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Table 1. Distribution of chilli in different districts of Manipur

S. No	Cultivars	Voucher no.	IW	IE	TH	BIS	TAM	UKH	SEN	CHA	CHU
1.	U-Morok	CAU1	+	+	+	+	+	+	+	+	+
2.	Morok asangbi	CAU2	+	+	+	+	+	+	+	+	+
3.	Churachandpur morok	CAU3	+	+	+	-	-	-	-	-	+
4.	Bell shape	CAU4	+	+	-	-	-	-	-	-	-
5.	Yensang morok	CAU5	+	+	-	-	-	-	-	+	-
6.	Meitei morok	CAU6	+	+	+	+	+	+	+	+	+
7.	Chakhou U-morok	CAU7	+	+	-	-	-	-	-	-	+
8.	U-morok macha	CAU8	+	+	+	+	+	+	-	-	-
9.	Morok macha	CAU9	+	-	-	-	-	-	-	+	+
10.	Meitei morok 1	CAU10	+	+	+	+	+	+	+	+	+
11.	U-chi morok	CAU11	+	+	+	+	+	+	+	+	+
12.	Small round chilli	CAU12	+	-	-	-	+	+	-	-	+
13.	U morok achouba	CAU13	+	+	+	+	+	+	+	+	+
14.	Meitei morok macha	CAU14	+	+	+	+	+	+	+	+	+
15.	Ukhrul morok asangbi	CAU15	+	-	-	-	-	+	-	-	-
16.	Colour Xanadu	CAU16	+	-	-	-	-	-	-	-	-
17.	Crispy long	CAU17	+	-	-	-	-	-	-	-	-
18.	Fireball	CAU18	+	-	-	-	-	-	-	-	-
19.	Colour xanadu	CAU19	+	+	+	+	+	+	+	-	-
20	Black bird eye	CAU20	+	+	+	-	-	-	-	-	+

*IW=Imphal west; IE=Imphal east; TH=Thoubal; BIS=Bishnupur; TAM=Tamenglong; CHU=Churachandpur; CHA=Chandel; UKH=Ukhrul; SEN=Senapati;
+ = present; − = absent

Table 2. Details of clusters between 20 cultivars by UPGMA clustering of correlation coefficients

S. No	Major cluster no.	Cluster samples (allowed distance co-efficient = 0.04 to 0.05)	No. of cultivars
1.	1	U morok, Chakhou u-morok, U morok achouba, U morok macha and Bell shape	5
2.	2	Meitei morok, Morok asangbi, Yensang morok, Churachandpur morok	4
3.	3	U-chimorok, small round chilli, Ukhrul morok asangbi, black bird eye, Morok macha,Crispy long, Fireball, Meitei morok macha,Colour xanadu, Meitei morok asaba, Meitei morok 1	11

 DOI: 10.5958/0975-928X.2017.00048.5

Table 3. Principal components showing the total variance (SPSS softwares)

Total Variance Explained									
CP	Initial Eigen values			Extraction Sums of Squared Loadings			Rotation Sums of Squared Loadings		
	Total	% of Variance	Cumulative %	Total	% of Variance	Cumulative %	Total	% of Variance	Cumulative %
1	31.01	75.63	75.63	31.01	75.63	75.63	30.39	74.13	74.13
2	9.16	22.34	97.97	9.16	22.34	97.97	9.78	23.84	97.97
3	0.55	1.33	99.31						
4	0.18	0.45	99.75						
5	0.08	0.19	99.94						
6	0.01	0.03	99.97						
7	0.01	0.02	99.99						
8	0.002	0.004	99.99						
9	0.001	0.004	99.99						
10	0.001	0.002	99.99						
11	0.000	0.001	99.99						
12	0.000	0.001	99.99						
13	0.000	0.000	100.00						

Table 4. Details of morphological characters with eigen vectors and eigen values

S. No.	Characters	Principal component	
		PC 1	PC 2
1	Hypocotyl	0.965	0.239
2	Cotyledonous leaf colour	0.990	-0.130
3	Cotyledonous leaf shape	0.992	-0.122
4	Stem colour	0.907	0.415
5	Nodal anthocyanin (whole plant)	0.799	0.593
6	Stem shape	0.981	-0.182
7	Stem pubescence	0.981	-0.182
8	Plant height	0.992	-0.086
9	Plant growth habit	0.848	0.524
10	Branching habit	-0.542	0.820
11	Leaf density	-0.419	0.899
12	Leaf colour	0.784	0.572
13	Leaf shape	0.960	0.251
14	Lamina margin	0.980	0.179
15	Leaf pubescence	0.980	0.179
16	Number of flowers per axil	0.831	0.546
17	Flower position	-0.383	0.906

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Table 4. Contd.,

S. No.	Characters	Principal component	
		PC 1	PC 2
18	Corolla colour	0.927	0.370
19	Corolla spot colour	0.981	-0.185
20	Male sterility	0.981	-0.186
21	Calyx pigmentation	0.985	-0.167
22	Calyx margin	0.952	-0.288
23	Calyx annular constriction	0.961	-0.259
24	Anthocyanin spots or stripes	0.972	-0.216
25	Fruit colour at intermediate stage	0.976	-0.213
26	Fruit set	0.937	0.321
27	Fruit colour at mature stage	-0.485	0.869
28	Fruit shape	-0.609	0.714
29	Fruit width	0.920	0.180
30	Fruit shape at pedicel attachment	0.941	0.192
31	Neck at base of fruit	0.965	-0.251
32	Fruit blossom end appendage	0.987	-0.152
33	Fruit cross-sectional	0.983	-0.153
34	corrugation	0.787	0.603
35	Fruit surface	0.981	-0.189
36	Fruit shape at blossom end	0.419	0.877
37	Pedicel with stem	0.097	0.984
38	Seed colour	0.977	0.067
39	Seed surface	0.991	-0.123
40	Seed size	0.077	0.981
41	Number of seeds per fruit	0.991	-0.053

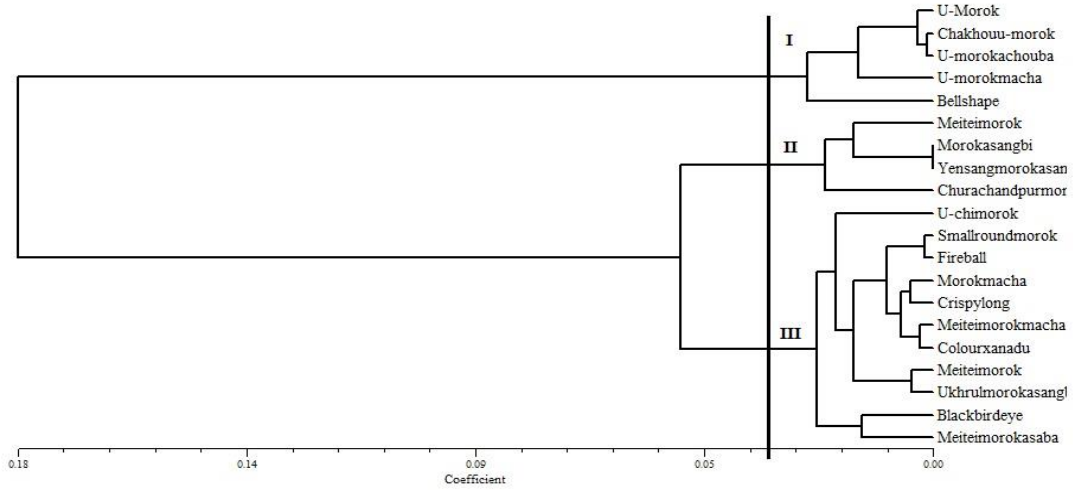

Fig 1. Dendrogram of 20 chilli cultivar based on 41 IPGRI morphological characters (NTSYS)

DOI: 10.5958/0975-928X.2017.00048.5

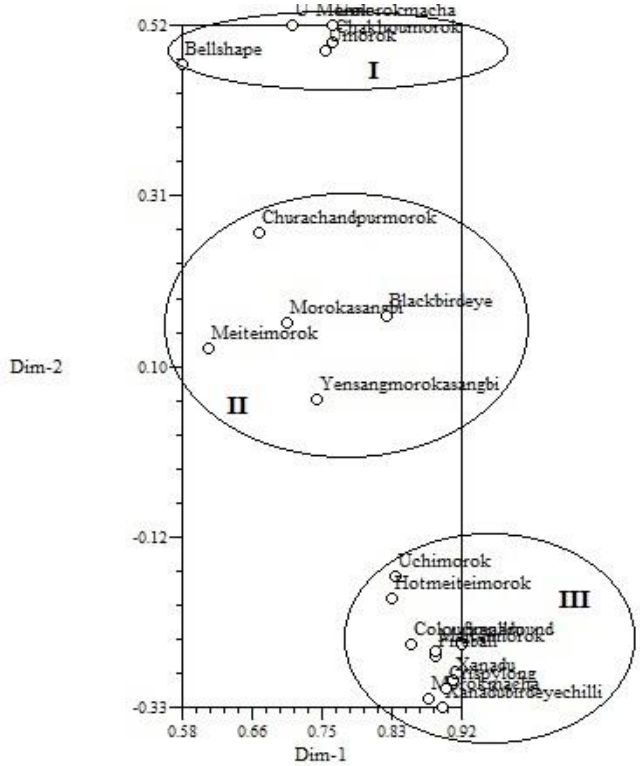

Fig 2. Two Dimentional distributions of chilli cultivars using NTSYS software (Simqual method)

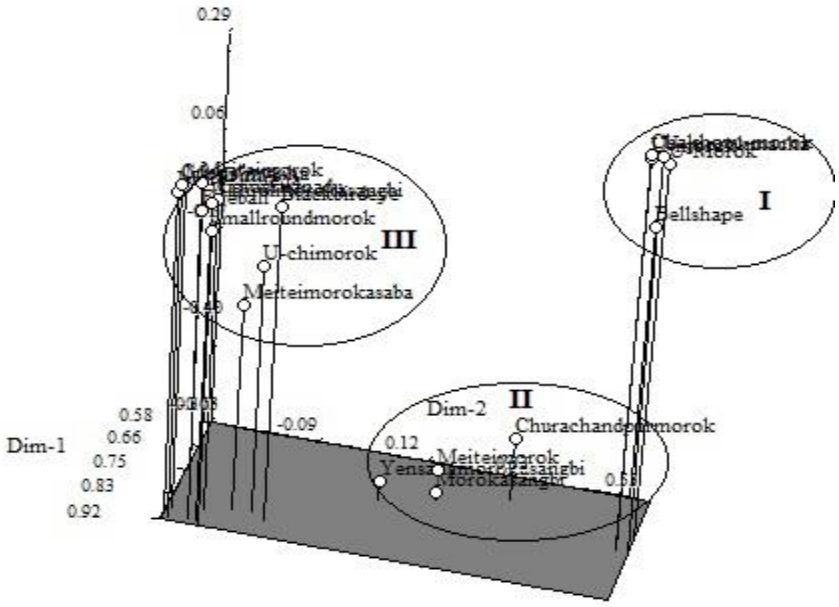

Fig 3. Three Dimentional representations of chilli cultivars