J. Hortl. Sci.
Vol. 10(2):177-182, 2015

Morpho-agronomic diversity in pole-type common bean (*Phaseolus vulgaris* L.) landraces from *Lushai* hills of North-East India

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ABSTRACT

The present study was based on morphological and agronomical characterization of 23 pole-type common bean (*Phaseolus vulgaris* L.) landraces collected from *Lushai* hills of North-East India. Extensive variation in plant and seed traits was found in 16 morphological and agronomical characters. Cluster analysis based on Euclidean distance grouped the genotypes into five main branches, reflecting their growth type and reproductive traits. Significant positive or negative correlation was observed among important traits. Principal component analysis was used for assessing patterns of variation by accounting for all the 10 quantitative and six qualitative variables together. Ordination among accessions showed that the first five principal components had Eigen values greater than one, and cumulatively accounted for 72% of the variation. Characterization based on quantitative and qualitative traits enabled separation of accessions into various groups representing landraces with distinct characters.

Key words: Common bean, pole-type, North-East India, landraces, principal components, morphological characterization

INTRODUCTION

Common bean (*Phaseolus vulgaris* L.), native to Central and South America, is widely cultivated in the temperate and subtropical regions of the globe. It is the most important food legumes in the world; its dry seed contains 21.1% protein, 69.9% carbohydrates, 1.7% fat, 381mg calcium, 425mg phosphorus and 12.4mg iron per 100g edible part (Ali and Kushwaha, 1987). Moreover, it serves as a functional food, as, it contains several bioactive compounds like enzyme inhibitors, lectins, phytates, oligosaccharides and various phenolic substances (Diaz-Batalla *et al*, 2006). Medicinally, it is used for controlling diabetics, certain cardiac conditions, it is a good natural cure for bladder burn and has both carminative and reparative properties against constipation and diarrhoea, respectively (Duke, 1981). Common bean is considered third in importance after soybean and peanut, but tops in direct consumption by humans (Broughton *et al*, 2003). Nutritionally, beans are often called the “poor man’s meat” as these are rich in proteins and minerals (especially iron and zinc), and vitamins (Beebe *et al*, 2000). In humans, iron is essential for preventing anemia, while, zinc is essential for adequate growth, sexual maturation, resistance to gastro-enteric and respiratory infections, especially in children (Bouis, 2003).

In India, Common bean is known by names like ‘Rajma’ and ‘French bean’ (green bean), and is grown mainly in North-Western Himalayan region comprising parts of Himachal Pradesh and Jammu & Kashmir, bearing a great diversity of agro-morphological traits like, seed size and colour (Kumar *et al*, 2008). A huge diversity in pole-type Common bean landraces exists in hilly states of North-East India, particularly in Mizoram, cultivated from a long time. In Mizoram, beans are primarily grown in the backyard and kitchen gardens for their green pods used as fresh vegetable, and, dried seeds as pulse or for seed purpose. The foliage is used as fodder and to restore soil fertility. In this state, a large pool of common bean landraces was found in farmers’ fields and forest areas. Nevertheless, these landraces were being lost to replacement by: high-value crops, drought, *jhum* fire, and human interference such as deforestation, etc. Therefore, a need is felt to characterize, conserve and maintain these landraces to serve as a source of desired traits for future breeding programme, and, to check further genetic erosion. Thus, a detailed description of accessions based on morpho-agronomical characters has tremendous bearing on conservation and genetic improvement in this crop. Until now, there have been no adequate studies on genetic diversity of pole-type Common bean landraces in Mizoram, which provides ample scope for studying variation in morphological and economic traits to be used for improvement of this crop. Therefore, the present investigation was aimed at studying vegetative and reproductive variation among pole-type Common bean landraces from the North-Eastern state of Mizoram, India.

MATERIAL AND METHODS

Twenty three pole-type Common bean landraces (Table 1) were collected from different districts of Mizoram during August-September, 2013. Seeds were multiplied and plants were evaluated for qualitative and quantitative traits during September – December 2013. The study was carried out in the experimental field of ICAR RC NEH Region, Mizoram Centre, Kolasib district (24.2304°N, 92.6761°E) of Mizoram, India, in Completely Randomized Block Design, with ten replications. Twenty seeds of each genotype were sown at a depth of 3-4cm after field preparation, at a spacing of 40cm (between plants) and 90cm (between rows). Irrigation was applied at two-day intervals. Mineral fertilization included 50kg N, 75kg P_2O_5 and 75kg K_2O per hectare. IPGRI (1982) descriptor for *Phaseolus vulgaris* was used for morphological description, and the traits assessed are listed in Table 2. The quantitative variables evaluated were: number of flowers per plant, leaf length, leaf breadth, pod length, number of pods per plant, pod yield, number of seeds per pod, days to flowering, no. of nodes on main stem from base to first inflorescence, and number of flower buds per inflorescence. Qualitative variables studied were: colour of freshly-opened flower, pod colour, pod cross-section, pod curvature, seed-coat colour, and seed shape. Statistical analysis was based on descriptive analysis (SAS 9.3), Hierarchical cluster analysis (using the method of mean linkage in UPGMA with which dendogram was constructed). Correlation analysis and principal component analysis (PCA) was done using PRINCOMP procedure of SAS 9.3.

RESULTS AND DISCUSSION

Twenty three landraces of Common bean were collected from across the state of Mizoram including backyards of farmers and roadside markets. Of all the landraces, MZFB-43 and MZFB-47 were rare in occurrence and distinct, with purple coloured pods (Fig. 1). Pod yield ranged between 118.43 g/plant (MZFB-29) and 49.47 g/plant (MZFB-28). Descriptive statistics of 10 quantitative

Table 1. French bean landraces from Mizoram, their source, frequency of occurrence and yield

S. No.	Accession	Source	Frequency	Yield (g/plant)
1.	MZFB-27	Roadside market	Frequent	91.37
2.	MZFB-28	Roadside market	Frequent	49.47
3.	MZFB-29	Roadside market	Frequent	118.43
4.	MZFB-32	Backyard of farmer	Frequent	87.50
5.	MZFB-36	Roadside market	Frequent	67.37
6.	MZFB-38	Roadside market	Frequent	66.93
7.	MZFB-40	Backyard of farmer	Frequent	83.23
8.	MZFB-41	Backyard of farmer	Frequent	91.90
9.	MZFB-42	Backyard of farmer	Frequent	106.20
10.	MZFB-43	Sel. from IC-595238	Rare (Purple colour pod)	108.70
11.	MZFB-44	Backyard of farmer	Frequent	53.40
12.	MZFB-45	Roadside market	Frequent	89.67
13.	MZFB-47	Sel. from IC-595238	Rare (Purple colour pod)	98.77
14.	MZFB-48	Backyard of farmer	Frequent	84.97
15.	MZFB-49	Roadside market	Frequent	96.03
16.	MZFB-50	Backyard of farmer	Frequent	60.20
17.	MZFB-51	Backyard of farmer	Frequent	65.10
18.	MZFB-52	Backyard of farmer	Frequent	53.37
19.	MZFB-53	Roadside market	Frequent	65.43
20.	MZBF-70	Backyard of farmer	Frequent	93.97
21.	MZFB-74	Roadside market	Frequent	60.37
22.	MZFB-76	Backyard of farmer	Frequent	86.70

Table 2. List of morphological traits assessed during vegetative growth (assessment done in accordance with IPGRI recommendations, with some modification)

Sl. No.	Trait	Score code-descriptor state
1.	NF: Number of flowers per plant	20-40=1; 40-60=2; >60=3
2.	LL: Leaf length	10-15=1; 15-20=2
3.	LB: Leaf breadth	6-8cm=1; >8cm=2
4.	PL: Pod length	5-8cm=1; 8-11cm=2; >11cm=3
5.	NP: Number of pods per plant	5-15=1; 15-25=2
6.	PY: Pod yield	40-60g/plant=1; 60-80g/plant=2; >80g/plant=3
7.	NS: Number of seeds per pod	4-6=1; 6-8=2; 8-10=3; >10=4
8.	DF: Days to flowering	<45 days=1; 46-60 days=2
9.	NB: No. of nodes on the main stem from base to 1st inflorescence	3-4=1; >5=2
10.	FBI: No. of flower buds/inflorescence	1-2=1; 3-4=2; >4=3
11.	CF: Colour of freshly opened flower	White=1; Lilac=2; White with red stripes=3; Purple=4
12.	PC: Pod color	Dark purple=1; Purple stripe on green=2; Normal green=3
13.	PCS: Pod cross-section	Pear shaped=1; Round/elliptic=2
14.	PCu: Pod curvature	Slightly curved=1; Curved=2
15.	SC: Seed-coat color	Black=1; Brown (pale to dark)=2; Pale creamy=3
16.	SS: Seed shape	Cuboid=1; Kidney shaped=2

parameters among the 23 Common bean landraces revealed that all the parameters had a highly significant (P< .0001%) variation among them (Table 3). Number of flowers averaged 43.5 and ranged from 20.33 (MZFB-52) to 61.33 (MZFB-45). Leaf length averaged 15.4cm, with a range of 12cm (MZFB-74) to 18cm (MZFB-29). Average leaf breadth was 10.41cm, and ranged from 8cm (MZFB-74) to 12.77cm (MZFB-29). Pod length averaged 11.24cm, with a range of 8.13cm (MZFB-43) to 14.63cm (MZFB-29). Average number of pods per plant was 13.02, and ranged from 7.67 (MZFB-51 and 52) to 23.67 (MZFB-38). Pod yield averaged 80.59g/plant, and ranged from 49.47g/plant (MZFB-28) to 118.43g/plant (MZFB-29). Number of seeds per pod averaged 5.69, and varied from 4.17 (MZFB-45) to 10.77 (MZFB-29). Average days to flowering were 49.43, and ranged from 43 (MZFB-45) to 54 days (MZFB-51 and 52). Number of nodes on the main stem from the base to the first inflorescence averaged 5.25, and varied from 3.8 (MZFB-28) to 6.7 (MZFB-40). Number of flower buds/ inflorescence averaged 3.42, and ranged from 2.7 (MZFB-43) to 4.2 (MZFB-30, 44 and 74).

In Fig. 1 are depicted the distinct pod, flower and seed, respectively, of MZFB-43 (a, b and c) and MZFB-47 (d, e and f). Our results prove that landraces of Common bean, grown by farmers in the *Lushai* hill region of North-East India, are valuable sources of genetic variation for breeding in this crop. This variation is due to inherent genetic differences among landraces grown throughout this area. Morphologically, Common bean differs in its growth habit (Singh, 1982), vegetative characters, and flower, pod and seed traits (Singh *et al*, 1991a, b) useful in selection in breeding programmes. Besides, this result was expected, as, landraces are a result of decades of natural and artificial selection by the farmer for a better adaptation to the local growing conditions, with different types being preferred by farmers in various regions (Harlan, 1976). Similar findings were reported by Stoilova *et al* (2013), where landraces were evaluated for 16 morphological characters, and considerable variation was found among genotypes. A majority of landraces had white seed-colour, but some also had cream, purple, and white with red colour around the hilum. The predominant seed-shape was long, but three accessions had round seeds.

Based on Euclidean distance, five clusters were formed which, in turn, fell into sub-groups, as depicted in Fig. 2. Number of genotypes in the clusters varied from one (Cluster II) to eight (Cluster IV). Cluster I was predominant in landraces with similar number of days to flowering, number of nodes on the main stem from the base to the first inflorescence, pod curvature, seed-coat color, etc. Cluster II consisted of a single landrace, MZFB-29, having the highest recorded leaf length (18cm), leaf breadth

Table 3. Descriptive statistics of 10 quantitative parameters in 23 Common bean landraces

Variable	Mean	Variance	Minimum	Maximum	t Value	Pr > |t|
NF	43.50	138.79	20.33	61.33	17.70	<.0001
LL	15.40	2.94	12.00	18.00	43.10	<.0001
LB	10.41	0.99	8.00	12.77	49.97	<.0001
PL	11.24	3.27	8.13	14.63	29.82	<.0001
NP	13.02	17.25	7.67	23.67	15.04	<.0001
PY	80.59	372.96	49.47	118.43	20.01	<.0001
NS	5.69	1.91	4.17	10.77	19.75	<.0001
DF	49.43	7.34	43.00	54.00	87.46	<.0001
NB	5.25	0.56	3.80	6.70	33.42	<.0001
FBI	3.42	0.25	2.70	4.20	32.47	<.0001

NF= No. of flowers per plant, LL= Leaf length, LB= Leaf breadth, PL= Pod length, NP= No. of pods per plant, PY= Pod yield, NS= No. of seeds per pod, DF= Days to flowering, NB= No. of nodes on the main stem from the base to the first inflorescence, FBI= No. of flower buds/ inflorescence

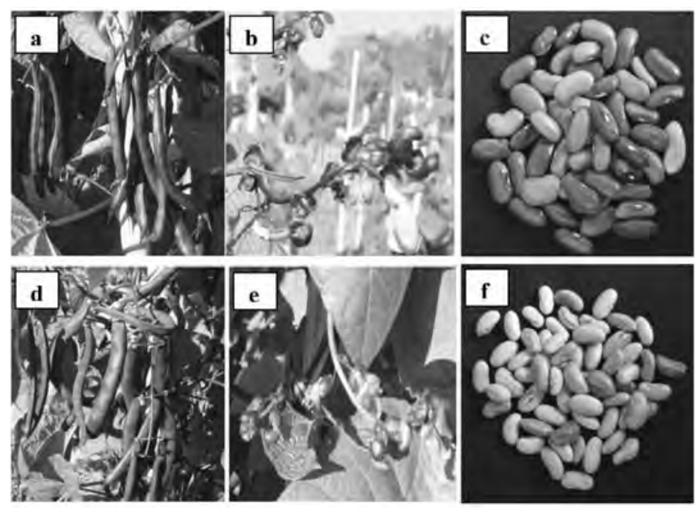

Fig. 1. Distinct pod, flower and seed, respectively, of MZFB-43 (a, b and c) and MZFB-47 (d, e and f)

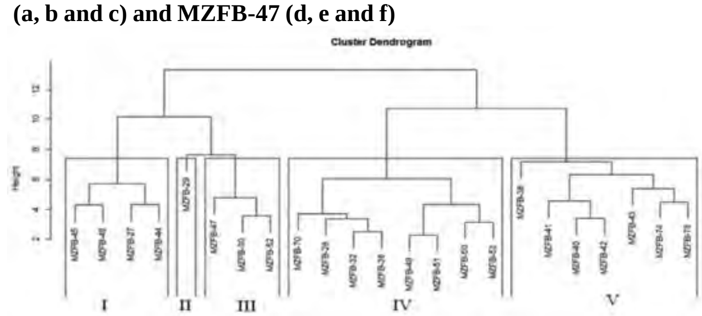

Fig. 2. Grouping of Common bean landraces on the basis of morphological traits: Prior to clustering data, variable rescaling was done for comparability; Hierarchical Cluster Analysis: Distance Calculation method - "Euclidean", Hierarchical cluster analysis method - "Ward"; Based on Euclidean distance (7.856), 5 clusters were formed

Table 4. Pearson Correlation Coefficients of quantitative variables in 23 Common bean landraces

	NF	LL	LB	PL	NP	PY	NS	DF	NB	FBI
NF	1	0.11	0.07	0.27	0.37	0.52*	0.25	(-0.48)*	0.17	0.30
LL		1	0.83**	0.62**	(-0.43)*	0.13	0.54**	0.25	-0.20	-0.02
LB			1	0.54**	-0.33	0.09	0.60**	0.14	-0.36	-0.11
PL				1	-0.35	0.41	0.55**	0.18	-0.03	0.11
NP					1	0.53**	0.02	(-0.42)*	0.35	0.25
PY						1	0.59**	-0.16	0.25	0.42*
NS							1	0.24	-0.07	0.19
DF								1	-0.15	-0.03
NB									1	0.05
FBI										1

NF= No. of flowers per plant, LL= Leaf length, LB= Leaf breadth, PL= Pod length, NP= No. of pods per plant, PY= Pod yield, NS= No. of seeds per pod, DF= Days to flowering, NB= No. of nodes on the main stem from the base to the first inflorescence, FBI= No. of flower buds/ inflorescence

*Significant at 5%, **Significant at 1%

(12.77), pod yield (118.43 g/plant) and number of seeds per pod (10.77). Cluster III grouped three accessions with similar traits like normal, green pod-color, pear-shaped pod cross-section, and brown (pale to dark) seed-coat color. Cluster IV was dominated by kidney-shaped seeds and white-coloured freshly-opened flowers. Slightly curved pod, pod length 8-11cm and 4-6 seeds per pod were the dominant traits in Cluster V. Traits that showed highest difference among clusters were those related to reproduction, like pod yield, number of flowers per plant, number of pods per plant, etc. Characterization based on qualitative traits enable separation of the accessions into various groups representing accessions from different geographical areas of the country (Beyene, 2013). Consequently, our results can give an index for identifying characters helpful for characterizing Common bean.

Pearson Correlation Coefficients of quantitative variables in 23 Common bean landraces are presented in Table 4. A significant (P <0.05%) positive correlation was found between number of flowers per plant and pod yield, while, a significant (P <0.05%) negative correlation was found between number of flowers per plant and days taken to flowering. Leaf length exhibited a highly significant (P <0.01%) positive correlation with leaf breadth, pod length and number of seeds per pod, while, number of pods per plant showed a significant (P <0.05%) negative correlation for this trait. Leaf breadth showed a highly significant (P <0.01%) positive correlation with pod length and number of seeds per pod. Pod length and number of seeds per pod, number of pods per plant, pod yield, pod yield and number of seed per pod exhibited a highly significant (P <0.01%) positive correlation. A strong correlation between these traits allows selection of the related trait/s interchangeably in a breeding programme. If two strongly correlated traits are desired, both can be selected simultaneously, based on one of the traits (Okii *et al*, 2014). Miko (2008) reported strongly correlated traits to be possibly under the influence of the same genes or showed pleiotropic effects.

Principal component analysis was used for assessing patterns of variation, by accounting for all the 10 quantitative and 6 qualitative variables together. Also, Eigen vectors, Eigen values, differences, proportions and cumulative percentages of variation were explained by the principal components. Ordination among accessions showed the first five principal components as having Eigen values greater than one, and these cumulatively accounted for 72% of the variation (Table 5). The first component alone explained 21% of the total variation and was mainly associated with characters such as pod length, number of seeds per pod, leaf length, leaf breadth and pod yield. The second principal component explained 19% of the variation and was associated with number of pods per plant and number of flowers per plant. The third principal component explained 12% of the variation, and was associated with pod curvature, pod color, number of nodes on the main stem from the base to the first inflorescence, and seed-coat color. The fourth and fifth principal components together explained 20% of the variation, and were associated with seed shape, pod curvature, number of nodes on the main stem from the base to the first inflorescence, and seed-coat color. Our study revealed that the first five principal components had Eigen values greater than one, and cumulatively accounted for 72% of the variation. The first component explained 21%, second 19%, third 12%, and, fourth and fifth combined 20% of the total variation. Principal component analysis has been applied for studying germplasm collections in several species,

Table 5. Component scores, Eigen values, differences, proportions and cumulative percentage of variation explained by the first five principal components (PC) in Common bean landraces

Trait	PC 1	PC 2	PC 3	PC 4	PC 5
LB	0.371	-0.296	-0.044	-0.066	0.190
PL	0.432	-0.141	0.055	0.113	0.019
NP	-0.037	0.452	0.193	-0.186	0.044
NF	0.281	0.350	0.025	-0.117	0.261
LL	0.383	-0.301	0.067	0.021	0.165
PY	0.337	0.284	0.198	-0.154	-0.233
NS	0.396	-0.093	0.269	-0.225	-0.125
DF	0.002	-0.343	0.192	-0.115	-0.266
NB	-0.027	0.257	0.365	0.394	-0.119
FBI	0.160	0.215	0.122	0.008	-0.337
CF	0.217	0.254	-0.437	0.159	-0.166
PC	-0.300	-0.221	0.380	-0.148	0.087
PCS	-0.092	0.100	0.210	-0.499	0.152
PCu	-0.007	-0.090	0.417	0.433	-0.286
SC	0.043	0.142	0.313	0.104	0.527
SS	0.040	0.021	0.032	0.441	0.420
Eigen value	3.47	3.15	1.95	1.83	1.25
Difference	0.31	1.19	0.12	0.57	
Proportion	0.21	0.19	0.12	0.11	0.09
Cumulative	0.21	0.41	0.53	0.65	0.72

LB= Leaf breadth, PL= Pod length, NP= No. of pods per plant, NF= No. of flowers per plant, LL= Leaf length, PY= Pod yield, NS= No. of seeds per pod, DF= Days to flowering, NB= No. of nodes on the main stem from the base to the first inflorescence, FBI= No. of flower buds/ inflorescence, CF= Colour of freshly-opened flower, PC= Pod color, PCS= Pod cross-section, PCu= Pod curvature, SC= Seed-coat color, SS= Seed shape

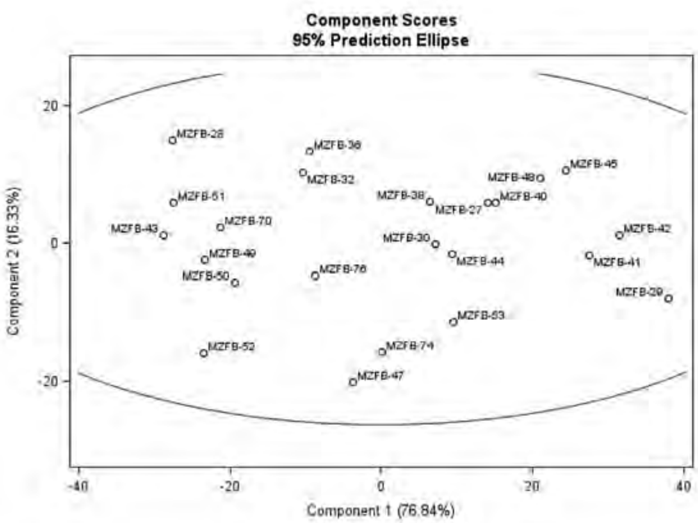

Fig. 3. Representation of Common bean landraces in a space defined by the first two principal components (principal components 1 vs. principal components 2)

including horticultural and arboreal species (Iezzoni and Pritts, 1991; Pe´rez-Gonza´lez, 1992; Badenes *et al*, 1998; Mars and Marrakchi, 1999; Leguizamo´n and Badenes, 2003). Principal component analysis allows better comprehension of relations, correlations and significance among the variables studied; applied to the germplasm collections, it shows the structure of a collection by identifying the most relevant variables, relationship among accessions, and, identification of possible mistakes. To assess the score of individual landraces, principal component 1 and principal component 2 are plotted in Fig. 3. Landraces MZFB-27, MZFB-30, MZFB-38, MZFB-40, MZFB-42, MZFB-45 and MZFB-48 occupied the right top corner of the plot, indicating higher scores for principal component 1. These clustered genotypes have positive scores for pod length, number of seeds per pod, leaf length, leaf breadth and pod yield. Also, landraces MZFB-28, MZFB-32, MZFB-36, MZFB-51, MZFB-43 and MZFB-70 occupied the left top corner of the plot, indicating higher scores for principal component 2 with similar number of pods per plant and number of flowers per plant.

Characterization based on quantitative and qualitative traits enables separation of accessions into different groups representing landraces with distinct characteristics. Therefore, the present finding can help identify characters necessary to characterize Common bean landraces. A broad genetic diversity of Common bean landraces exists in Mizoram state of India. The germplasm clustered into five major groups, with the most variations attributed to days to flowering, node number on the main stem from the base to first inflorescence, pod curvature, seed-coat colour, pod yield, number of seeds per pod, etc. These traits can be used for characterization; conservation and breeding in Common bean landraces. Similarity in clustering and correlation information on phenotypic traits can be used for parent selection in breeding programmes. These results show the use of morpho-agronomic information for characterization of Common bean landraces of Mizoram.

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(MS Received 27 November 2014, Revised 18 May 2015, Accepted 02 July 2015)