

Genetic Diversity in Mungbean [Vigna radiata (L.) Wilczek] Genotypes based on K-Mean Determination

Chandra Mohan Singh*1, S.B. Mishra² and Anil Pandey²

¹Department of Plant Breeding and Genetics, Rajendra Agricultural University, Bihar, Pusa (Samastipur)- 848 125, India. ²Department of Plant Breeding and Genetics, Tirhut College of Agriculture, Dholi – 843 121, Muzaffarpur, Bihar, India.

The present study was conducted with 36 genotypes of mungbean to assess the extent and pattern of genetic divergence. Cluster analysis exhibited that genotypes fall into seven clusters, in which cluster I is mono genotypic, indicated the distinctness of this genotype from others. Principle component analysis (PCA) showed that the first four principle components (PC) had eigen value >2.00 and accounted 82.29% of total variation. Based on 2-D/ 3-D diagram and their cluster means, the suitable genotypes may be chosen and involved in hybridization programme for mungbean improvement. Analysis based on harvest index indicated that maximum selection score (SCs) was recorded for 40.1 – 45.0% class interval, can be used as a selection criteria for mungbean improvement.

Key words: Mungbean, Genetic diversity, K-Mean clustering, Selection Score, Trait optimization.

Mungbean [Vigna radiata (L.) Wilczek] is an important short duration grain legume grown in Asia as a sole crop as well as in multiple cropping systems. It remains as an income generator for majority of marginal farmers. It is a substitute for meat in most of the Asian diet, and a significant component of various cropping systems (Srinives et al., 2000; Rudy et al., 2006). The grains and plant of mungbean are also used as a feed and fodder for cattle. Further, crop residues of mungbean after picking pods are used as green manure besides, fixing the atmospheric nitrogen into the soil. It is cultivated in kharif as well as zaid (spring/ summer) season. But now, farmers prefer mungbean in zaid season, because of their dwarf behavior to utilize the gap between the period of harvest of wheat and sowing / planting of rice. In turn, this method increases the cropping intensity and improves the soil health.

Knowledge of genetic variability is a pre requisite of any crop improvement programme. Therefore, grouping of genotypes based on major yield attributing traits may be useful. Genetic diversity study includes understanding the crop performance leading to crop improvement (Aremu, 2005). Knowledge of crop performance in genetic diverse population reveals the differences in the nature of genetic materials used. Genetic diversity study, is a step wise process through which existing variations in the nature of individual or group of individual crop genotypes are identified using specific statistical method or combination of methods (Warburton and Crossa, 2000; Aremu, 2005). It is expected that the identified variations would form a pattern of genetic relationship usable in grouping genotypes. The first step of any crop improvement programme is to identify crop plants that exhibit exploitable variation for the trait(s) of interest.

¹*Corresponding author email: cmsingh.gpb@gmail.com

Genetic diversity is useful for developing germplasm conservation strategies, sorting out trait specific groups and their utilization. Keeping the above facts under consideration the present investigation was undertaken to study genetic diversity and isolate the suitable parents for mungbean crop improvement.

Materials and Methods

The present investigation was conducted with 36 genotypes of mungbean viz., HUM 12, IPM 02-14, NDM 9-18, ML 1666, DM 05-12-1-42-3, DMS 01-34-2, DMS 03-17-2, DM 99-11-5, SML 668, Samrat, DMC 17, Meha, Sona, IPM 2K-14-9, DM 05-74-11, IPM 99-01-10, PM 2, P 1131, DMS 02-11-4, IPM 99-1-6, P 1232, P. Vishal, P 1131, IPM 2K-15-4, P 9531, PM 08-2, NDM 12-308, DMS 02-11-13, IPM 99-394, SML 1186, PM 5, SML 1151, P Baishakhi, AKM 8803, HUM 16 and TMB 37. The genotypes were received from Pulse Breeding Section, Department of Plant Breeding and Genetics, Tirbut College of Agriculture, Dholi, Muzaffarpur, Bihar, India and numbered serially 1 to 36. The experiment was conducted at Crop Research Farm of TCA, Dholi (RAU, Pusa), which is situated (25.50N, 35.40E, 52.12 m MSL) in Muzaffarpur district of North Bihar, India.

A field experiment was conducted in Randomized Block Design with three replications during summer, months of 2012. Each genotype was sown in six rows in 4 m length plot with 30 x 10 cm spacing. The observations were recorded on 26 agromorphological traits *viz.*, days to first flower opening (DFFO), plant height (PH), number of primary branches per plant (NPBP), number of secondary branches per plant (NSBP), petiole length (PetL), leaf area (LA), number of nodes on main stem (NNMS), average internodal length (AIL), primary branch angle with main stem (PBAMS), node for first productive

peduncle (NFPP), peduncle length (PedL), pod angle with peduncle (PAP), leaf pubescence density (LPD), stem pubescence density (SPD), petiole pubescence density (PetPD), pod pubescence density (PPD), number of clusters per plant (NCP), number of pods per cluster (NPC), pod length (PL), beak length (BL), number of seeds per pod (NSP), pod filling index (PFI), seed index or 100-seed weight (SI), biological yield per plant (BYP), harvest index (HI) and seed yield per plant (SYP) for quantitative analysis. Five random plants were tagged from each plot to record the data (quantitative traits) for all the yield and agro-morphological traits except DFFO. DFFO was recorded on plot basis, when majority of the plants (about 80%) attained the stage with minimum one open flower. For scoring of the data leaf sample was taken from 5th node from the base. Leaf area (LA) was calculated by the formula suggested by Yoshida et al. (1972). The data on yield and its other related traits were subjected to genetic divergence analysis based on K-mean determination by using statistical package WINDOSTAT 8.6 version. To calculate the selection scores (SCs) ranking for some important agro-morphological traits within HI range, top three values for individual traits were picked and multiplied in reverse order, *i.e.*, the 1st rank trait(s) multiplied with 3, 2nd rank trait(s) with 2 and 3rd rank trait(s) with 1. To minimize error, the products were multiplied with the communality values observed in PCA and the SCs were calculated.

Results and Discussion

A wide range of variability was exhibited by most of the traits under study (Table 1). The coefficient of variation (CV) was moderate (20.80 - 24.19%) for NPBP, NSBP and LA, whereas, the other traits showed low CV. DFFO ranged from 34 to 61 days with a mean of 47 days. Plant height ranged from 26.27 to 47 with a mean of 32.25 cm. NPBP, NSBP and PBAMS varied from 1-6, 1-10 branches per plant with a range of 290 to 980. Assesing NNMS and AIL is an important parameter for lodging tolerance, which varied from 6.30 to 14.33 with a mean of 8 nodes per plant; and 2.26 to 5.53 cm with a mean of 4.13 cm, respectively. LA and PetL ranged from 11.23 to 52.93 with a mean of 28.55 cm² and 5.20 to 11.33 with a mean of 8.40 cm. NFPP, and PedL ranged from 3-5 with a mean of 4^{th} node, and 3.83 to 12.47 cm with a mean of 8.73 cm, respectively. NCP and NPC 3-11 with a mean of 7 clusters per plant, and 3-6 with a mean of 4 pods per cluster, respectively. PL varied from 5.25 to 8.45 cm with a mean of 6.68 cm, and NSP varied from 3 to 12 with a mean of 9 seeds per pod. SI ranged from 2.67 to 5.53 g with a mean of 4.38 g. BYP and HI are very important traits for improvement of SYP and ranged from 11.39 to 28.48 g, with a mean of 21.05 g and 29.39 to 57.47%, with a mean of 41.34%, respectively. Seed yield is the ultimate objective of any crop improvement programme and it varied from 5.61 to 13.99 g, with a mean of 8.57 g per plant.

 Table 1. Range, mean and coefficient of variation

 of morphological traits in mungbean

Troite	Rar	nge	Maan	CV (0()
Traits -	Min	Max	Mean	CV (%)
Days to first flower opening	34.67	61.67	47.03	5.11
Plant height (cm)	26.27	47.00	32.45	9.34
Number of primary branches/ plant	0.40	6.33	2.65	20.85
Number of secondary branches/ plant	0.13	10.00	3.11	20.80
Petiole length (cm)	5.20	11.33	8.40	9.83
Leaf area (cm ²)	11.23	52.93	28.55	24.19
Number of nodes on main stem	3.60	14.33	8.01	7.76
Average intermodal length (cm)	2.26	5.53	4.13	11.56
Primary branch angle with stem (Degree)	29.00	98.00	70.85	7.78
Node for first productive peduncle	3.13	5.57	4.24	12.57
Peduncle length (cm)	3.83	12.47	8.73	11.52
Pod angle with peduncle (Degree)	56.67	110.67	81.34	7.64
Leaf pubescence density (cm ²)	29.13	153.13	84.21	5.67
Stem pubescence density (cm ²)	25.33	235.00	133.01	6.04
Pod pubescence density (cm ²)	101.33	175.00	145.89	3.41
Petiole pubescence density (cm ²)	24.33	94.80	61.08	8.74
Number of clusters/ plant	3.93	11.92	7.95	19.28
Number of pods/ cluster	3.80	6.23	4.86	13.02
Pod length (cm)	5.25	8.45	6.68	7.02
Beak length (mm)	1.75	7.50	4.15	12.74
Number of seeds/ pod	3.07	11.60	9.60	9.34
Pod filling index (%)	54.30	89.87	69.09	7.35
100-SW (g)	2.67	5.53	4.38	10.00
Biological yield/ plant (g)	11.39	28.48	21.05	11.80
Harvest index (%)	29.39	57.47	41.34	13.48
Seed yield/ plant (g)	5.61	13.99	8.57	13.69

Cluster Analysis

Cluster analysis helps to select the suitable genotypes for hybridization to manipulate the important traits. Selection of proper parents plays a vital role for a successful plant breeding programme. Parents with more genetic distance can create higher varibility, which increases the genetic gain in selection. The grouping of the genotypes was done by K-mean clustering pattern. The distribution of 36 mungbean genotypes in different clusters and their cluster means are presented in Table 2 and 3, respectively. The total genotypes of mungbean fall into seven clusters. Cluster I comprised only one genotype (i.e. Sona) forming a mono genotypic cluster, whereas cluster II and cluster IV comprised eight genotypes in each. Cluster VI and cluster VII consist of three genotypes each. Genotype Pusa Vishal also exhibited good diversity with Sona (Fig 1 & 2) and found suitable for important in some other agro-morphological traits. This genotype falls in cluster VI and this is noted with less NPBP, NCP but high PL, SI and moderate SYP as compared to Sona. Based on cluster mean, it was noted that the cluster I and VII showed average diversity for SYP which indicates that obtaining transgressive segregants in early segregating and subsequent generations is possible. These clusters were also found suitable for Table 2. Distribution of members in various clusters

K-Group	No. of Entries	Cluster Members					
I	1	15					
11	8	4, 8, 9, 10, 11, 12, 35, 36					
111	6	16, 17, 25, 26, 28, 31					
IV	8	5, 14, 15, 18, 19, 20, 23, 27					
V	7	1, 3, 21, 29, 30, 32, 34					
VI	3	2 , 22, 24					
VII	3	6, 7, 33					

	DFFO	PH	NPBP	NSBP	PeL	LA	NMS	AIL	PBAMS	NFP	PedL	PAP	LPD
Cluster I	61.67	31.73	6.33	10.00	5.20	11.23	14.33	2.26	85.67	5.17	3.83	82.30	84.93
Cluster II	41.88	33.60	1.66	1.08	9.32	37.77	7.90	4.26	77.42	4.77	8.46	75.95	80.17
Cluster III	48.28	32.82	2.93	3.88	7.66	22.87	7.88	4.23	71.00	4.02	9.78	90.67	75.86
Cluster IV	50.75	29.77	3.19	4.02	9.22	31.53	7.35	4.06	71.35	4.05	9.25	77.77	93.53
Cluster V	47.91	31.25	2.89	3.71	7.72	24.02	8.12	3.88	70.89	4.08	8.53	82.85	100.85
Cluster VI	50.56	30.08	3.00	3.16	6.95	19.48	7.24	4.23	69.61	3.86	7.27	86.93	63.76
Cluster VII	37.89	41.22	1.19	0.84	9.31	32.78	8.71	4.83	47.88	4.20	9.51	77.20	68.26
	SPD	PPD	PePD	NCP	NPP	PL	BL	NSP	PFI	SI	BYP	НІ	evr.
	SPD	PPD	PePD	NCP	NPP	PL	BI	NSP	PFI	SI	BYP	ы	SYP
Cluster I	104.07							-					-
	104.07	126.67	74.77	9.50	4.17	5.53	3.57	3.67	72.82	2.67	15.33	36.71	5.63
	148.46	126.67 147.97	74.77 71.26	9.50 7.03	4.17 4.67	5.53 6.16		-	72.82 67.63	2.67 4.18			-
Cluster II Cluster III							3.57	3.67			15.33	36.71	5.63
Cluster II	148.46	147.97	71.26	7.03	4.67	6.16	3.57 3.94	3.67 9.18	67.63	4.18	15.33 18.86	36.71 40.56	5.63 7.46
Cluster II Cluster III	148.46 174.31	147.97 141.86	71.26 49.98	7.03 8.83	4.67 4.86	6.16 6.94	3.57 3.94 4.18	3.67 9.18 9.92	67.63 69.95	4.18 4.60	15.33 18.86 23.20	36.71 40.56 39.68	5.63 7.46 9.15
Cluster II Cluster III Cluster IV	148.46 174.31 162.51	147.97 141.86 156.63	71.26 49.98 78.79	7.03 8.83 9.34	4.67 4.86 5.03	6.16 6.94 6.53	3.57 3.94 4.18 4.50	3.67 9.18 9.92 9.91	67.63 69.95 66.17	4.18 4.60 4.39	15.33 18.86 23.20 24.98	36.71 40.56 39.68 38.50	5.63 7.46 9.15 9.64

Table 3. Mean characteristics (K-Mean) on various agro-morphological traits for each clusters in mungbean

improvement of DFFO and HI, ultimately it is possible to improve mungbean with early maturing varieties/ lines with high harvest index. Several researchers reported the significance of high harvest index to improve the seed yield in mungbean (Ghafoor *et al.*, 1993; and Singh *et al.*, 2009). The green revolution

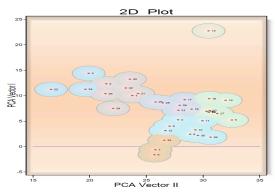


Fig. 1. Distribution of 36 mungbean genotypes between first and second principle components (2-D Plot Diagram).

in cereals is largely supported by a tremendous increase in HI that enhanced cereal productivity. Similar emphasis is being given in legumes to select genotypes with appropriate harvest index (Zubair *et al.*, 2007). Representative genotypes may be chosen from particular diverse groups based on their cluster mean in hybridization programme for mungbean improvement.

Principle Component Analysis (PCA)

In order to maintain, evaluate and utilize germplasm

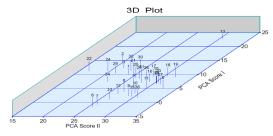


Fig. 2. Distribution of 36 mungbean genotypes between first second and third principle components (3-D Plot Diagram).

effectively, it is important to investigate the available genetic diversity (Mohammadi, 2003). PCA was used to identify the most significant variables in the traits studied. It was also used for establishing the genetic relationship among the genotypes. Association between traits emphasized by this method may correspond to genetic linkage between loci controlling the traits or a plieotropic effect (lezzoni and Pritts, 1991). The genetic variation present in breeding material was divided into eight principle components which explained 86.52% of total variation (Table 4). The first four principle components had eigen value >2.00 and accounted 82.29% of total variation. The first principle component (PC 1) explained 28.75% of the total variation and was mainly associated with NPBP, NSBP, SYP, DFFO and BYP. The second PC (PC 2) was responsible for 14.90% variation and was related to NFPP, SPD, PetPD, HI and LA. The third PC (PC 3) contributed about 11.81% variation due to effect of HI, BYP and NPC. The fourth PC (PC 4) explained 8.55% variation and associated with PPD and NFPP. This showed that higher loading values are attributed to yield parameters i.e. NPBP, NSBP, SYP, DFFO, NFPP, HI, LA and pubescence traits i.e. SPD, PetPD, which indicated the importance of these traits. The dispersion of the genotypes showed considerable amount of variability present in breeding material (Fig 1). Two genotypes i.e. DMS 01-34-2 and DMS 03-17-2 had negative values for PC 1. These genotypes were found to be early maturing.

Table 4. Eigen values and variability explained by each principle components (PCs)

				- 1	- /			
	PC1	PC2	PC3	PC4	PC5	PC6	PC7	PC8
Eigen Value (Root)	7.47	3.88	3.07	2.22	1.86	1.51	1.37	0.94
% Var. Exp.	28.75	14.90	11.81	8.55	7.17	5.83	5.27	3.63
Cum. Var. Exp.	28.75	43.65	55.47	64.02	71.19	77.02	82.29	85.92

The maximum positive values for PC 1 and PC 2 was exhibited by Sona mung. The study through canonical vector analysis revealed that, on the basis of two axis the cumulative percentage of variation observed by PC 1 and PC 2 was 43.65% and the maximum contribution of NPBP and NFPP was found towards total divergence, whereas based on 3-D plot analysis, the cumulative variation was 55.47% and maximum contribution of NPBP, NFPP and HI was recorded towards the total divergence. The 3-D

Table 5. Correlation coefficient of each agromorphological trait with respect to its principle components (PC)

Traits	Comm unality	PC1	PC2	PC3
Days to first flower	0.262	0.287	0.076	0.026
Plant height (cm)	0.318	-0.155	-0.032	-0.230
Number of primary branches/ plant	0.164	0.309	0.102	0.050
Number of secondary branches/ plant	0.141	0.309	0.102	0.116
Petiole length (cm)	0.146	-0.304	0.063	0.040
Leaf area (cm2)	0.431	0.068	0.211	0.075
Number of nodes on main stem	0.298	0.186	0.103	-0.289
Average intermodal length (cm)	0.147	0.324	0.135	0.026
Primary branch angle with stem (Degree)	0.624	0.135	0.149	0.194
Node for first productive peduncle	0.366	0.074	0.307	-0.055
Peduncle length (cm)	0.418	-0.190	0.021	0.034
Pod angle with peduncle (Degree)	0.281	0.233	-0.068	0.143
Leaf pubescence density (cm2)	0.447	0.031	0.041	-0.280
Stem pubescence density (cm2)	0.330	-0.133	0.305	0.200
Pod pubescence density (cm2)	0.361	0.121	0.222	-0.013
Petiole pubescence density (cm2)	0.241	-0.210	0.302	0.052
Number of clusters/ plant	0.311	-0.107	0.129	0.408
Number of pods/ cluster	0.236	-0.238	0.206	0.231
Pod length (cm)	0.337	-0.043	-0.289	0.315
Beak length (mm)	0.428	-0.084	0.247	-0.116
Number of seeds/ pod	0.397	-0.132	-0.104	0.198
Pod filling index (%)	0.209	-0.106	-0.394	0.150
100-SW (g)	0.394	0.085	-0.319	0.151
Biological yield/ plant (g)	0.233	0.268	0.025	0.263
Harvest index (%)	0.273	-0.070	0.208	0.382
Seed yield/ plant (g)	0.207	0.291	-0.151	0.154

diagram (Fig 2) showed that the maximum diversity was recorded between Sona and DMS 03-17-2, DMS 01-34-2 and Pusa Baishakhi. These genotypes may be used in hybridization programme to improve characters in different principle component depends upon the nature of breeding material. Abna *et al.* (2012) conducted the experiment for divergence analysis with 20 genotypes and observed that the first principal component (PC1) is related to number of fruiting branches per plant, number of pod per plant and number of pod cluster per plant that explained 39.4% of total variability. The characters with greatest positive weight on second principal component (PC2) were 100 seed weight, seed yield and number of pod per plant.

Optimization of HI

HI is a very important trait to increase the grain yield potential in crops; but it is a very complex trait in legumes, unpredictable and sensitive to environmental fluctuations (Ghafoor et al., 2000, Ghafoor et al., 2001). Singh et al. (2006) also reported significant differences in seed yield of mungbean varieties mainly because of differences in HI. In order to find out the best HI along with other traits, the breeding materials were classified into various groups (Table 6). The maximum genotypes (nine in each group) fell between 35.1 - 40.0% and 40.1 – 45.0% HI range. Genotypes with <30.00% HI was characterized by tall plant structure, high NCP, NPC, PL, NSP, SI and BYP. The genotypes which fell in 40.1 - 45.0 % HI range gave higher values for PBAMS and were found to be early flowering,

Table 6. Analysis of mungbean genotypes based on harvest index (HI) class interval

	HI Class Interval											
Traits	< 30.00		30.10- 35.00		35.10 - 40.00		40.10 - 45.00		45.1 - 50.00		> 50.00	
	M	SE	М	SE	М	SE	М	SE	М	SE	М	SE
Days to first Flower Opening	51.22	1.70	47.25	1.01	48.56	1.30	45.59	1.55	45.62	1.36	45.92	1.49
Plant height (cm)	33.98	2.18	31.79	0.95	32.23	2.17	32.63	1.58	32.36	1.85	32.08	1.41
Leaf area (cm ²)	22.25	1.14	29.92	3.21	30.92	7.07	30.00	3.38	23.34	2.82	37.12	3.65
Primary branch angle with stem	68.33	2.94	70.09	6.15	63.27	3.86	75.72	3.43	66.88	0.28	61.53	2.88
Number of clusters/ plant	4.71	0.48	8.58	1.01	7.77	0.65	8.67	0.79	7.77	3.44	8.87	1.11
Number of pods/ cluster	5.38	0.39	4.83	0.28	4.61	0.38	5.2	0.48	4.68	1.17	4.48	0.20
Pod length (cm)	7.33	0.08	5.74	1.23	6.26	0.26	6.79	0.28	6.76	0.42	7.12	0.38
Number of seeds/ pod	10.51	0.15	9.63	0.42	9.02	0.54	9.63	0.61	9.46	0.42	10.40	0.79
100-Seed weight (g)	4.66	0.27	4.52	0.25	4.1	0.29	4.57	0.24	4.16	0.21	4.60	0.11
Biological yield/ plant (g)	22.3	0.91	17.17	1.99	21.37	1.01	22.04	1.31	18.88	2.11	20.37	0.67
Seed yield/ plant (g)	6.53	0.39	7.25	1.00	7.86	0.62	9.37	0.75	8.78	0.63	10.75	0.48
Number of Genotypes	0	3	0-	4	0	9	0	9	0	7	0-	4

M= Mean, SE= Standard error of mean

the NPBP, NFPP and HI. In addition, the genotype Pusa Vishal also exhibited good diversity with Sona mung. Lower placement of productive peduncle may help to improve the seed traits by good translocation of nutrient from source to sink. The association of

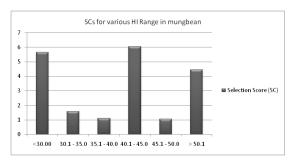


Fig. 3. Selection score based on harvest index (HI) in mungbean

indicating the good use of solar radiation / energy mainly due to its canopy and improved the seed yield. The genotypes with > 50.0% HI gave high LA along with early flowering, NCP and SYP. Thus, there is a need to find out the best range of HI by calculating the selection scores (Ghafoor *et al.*, 2001). SCs for each HI class interval have been presented in Fig. 3. The present investigation indicated that 40.1 - 45.0 % HI range had the maximum SCs (6.05) followed by <30.0 (5.67) and >50.0% (4.43). Ghafoor *et al.* (2001) reported best SC for 30.1 – 35.0 % HI class interval in *Vigna mungo* L. Hepper. High SCs gave clue for the future selection for mungbean crop improvement

Conclusion

Sona mung exhibited more diversity with other genotypes present in different clusters. PCA revealed that the first four principle components accounted 82.29% of total variation and indicated the possibility of improvement of mungbean by manipulation of the traits *viz.*, NPBP, NSBP, SYP, DFFO, NFPP, SPD, PetPD, HI, LA and NCP. Suitable genotypes chosen from particular diverse groups based on their cluster mean may be involved in hybridization programmes for mungbean crop improvement. Analysis on HI indicated that HI range with 40.1 – 45.0 % exhibited the maximum selection score because of its canopy development, which could be used as selection criteria for yield improvement in mungbean.

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