

## Diversity analysis of mungbean [*Vigna radiata* (L.) Wilczek] genotypes for bruchid resistance

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### ABSTRACT

Mungbean [*Vigna radiata* (L.) Wilczek] is one of the most important pulse crops in the tropical and sub-tropical areas. Although the total production of mungbean is not enough to supplement the nutritional requirement, an additional issue further augments the concern of its use: the post-harvest damage in the storage condition. Bruchids (*Callosobruchus chinensis*) are major polyphagous storage pest that causes substantial losses, both quantitatively and qualitatively. The objective of the current study was to classify the fifty-two mungbean genotypes into groups based on their bruchid resistant and susceptibility by using multivariate statistical analysis. Since the present study emphasizes the progressive breeding program, the multivariate analysis is expected to effectively demonstrate the diversity of bruchid attack and therefore identify and ensures the correct representation of the resistant genotypes. The Principal Component Analysis (PCA) displays the correlation of bruchid morpho-physiological traits and the diversity analysis groups the considered genotypes into four clusters (I-IV). The cluster III contains seven genotypes which are observed most effective against bruchid attack. The PC1 shows 70.83% of variability where PC2 shows 29.17% of variability. PCA picturize the most positive correlation between the number of egg laid (NOEL-0.922) and percentage of seed damage (POSD-0.975) among all the morpho-physiological traits. The cluster III thus helps selecting resistant parents for hybridization in future crop improvement program.

**Key words:** Mungbean, Bruchid, Biversity, Seed loss, Susceptible, Resistance.

**Abbreviations:** PCA: Principal component analysis, NOEL: Number of egg laid, POEH: Percentage of egg hatched, POSD: Percentage of seed damage, POSWL: Percentage of seed weight loss, MDD: Mean development days, POPT: Percentage of pest tolerance, SI: Susceptibility Index, R: Resistant, MR: Moderately Resistant, MS: Moderately Susceptible, S: Susceptible.

### INTRODUCTION

Mungbean [*Vigna radiata* (L.) Wilczek] is one of the most important short duration pulse crops due for it is the major source of inexpensive but unique quality of vegetable proteins, vitamins, and minerals. This is also the staple ingredients in the diet regimen of the growing population in the Asian subcontinent (Khajudpar and Tantasawat, 2011 and Siddique *et al.* 2006). Several host biotic and abiotic stress elements cause serious yield loss in the risk prone environment and among them the major yield loss (annually 35-40 %) is found during the postharvest storage condition due to bruchid infestation (Bahl, 2015). Bruchid (*Callosobruchus chinensis*) (Coleoptera: Bruchidae) are the major damaging polyphagous storage pest. It causes substantial post-harvest loss quantitatively (approximately around 3 million tons) and also deteriorates the quality of seeds. Bruchid infestation primarily starts in the field condition and the damage is carried into the storage condition where the seeds are completely damaged by perforation within 2-3 months. Damaged seeds lose their seed weight as

well as the nutritional quality. This affects the seed viability and consequently incurs direct losses from the farmers to consumers. So, the post-harvest losses are the major obstacles to reach the food security in the developing nations (Ponnusamy *et al.* 2014 and Somta *et al.* 2007). Multivariate analysis is a proven technique that can be applied for assembling genotypes as groups based on their trait similarity from a large number of genetic resources. This is also useful for genotype collection and utilization. Principle component analysis emphasizes on phenotypic traits association and provides plausible explanation to the distribution of correlated and uncorrelated traits. So, the genetic diversity is an evolutionary platform where the created groups transfer the desirable genes through the hybridization process from the parent to advanced progeny (Sharifi *et al.* 2018 and Perera *et al.* 2017). The current study aims to identify bruchid resistant and susceptible mungbean genotypes under the laboratory conditions through multivariate analysis and thus is expected to select bruchid resistant parents for future hybridization program.

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## MATERIALS AND METHODS

**Plant material:** Fifty-two mungbean genotypes were collected from NBPGR (New Delhi); Pulse and Oil Seed Research Station (Berhampore); and some local accessions of different districts of West Bengal.

**Experiment design and seasons:** The present experiment was carried out at the laboratory of the Department of Genetics and Plant Breeding at the Institute of Agricultural Science, University of Calcutta. Seeds of fifty-two mungbean genotypes were evaluated during the season of June to August from 2015 to 2017. The experiment was laid out in a Completely Randomized Design (CRD) in B.O.D condition for maintaining the temperature ( $28 \pm 2^\circ\text{C}$ ) and humidity ( $65 \pm 5\%$ ). The bruchid screening was performed by following the method of Dobie (1974) and Mensah *et al.* (1986).

**Data collection:** The data were recorded from each replication on different morpho-physiological traits such as number of egg laid (NOEL), number of adult emerge (NOAE), percentage of egg hatched (POEH), number of seed damage (NOSD), percentage of seed damage (POSD), initial seed weight (ISW), percentage of seed weight loss (POSWL), mean development days (MDD), percentage of pest tolerance (POPT) and the susceptibility index (S.I.). These parameters were calculated using the formula of Dobie (1974) and the mean values were computed from the observations of all three seasons. According to Dobie (1974) and Mensah (1986), the susceptibility index is classified into five groups

with slight modification such as 0 to 3 (Resistant), 3.1 to 6 (Moderately Resistant), 6.1 to 8 (Moderately Susceptible), 8.1 to 10 (Susceptible) and  $> 10$  (Highly Resistant).

**Statistical analysis:** The descriptive statistics including mean, standard error (SE) and range in standard unit were calculated using SPAR 2.0 software package. The Principal component analysis (PCA) and k-means clustering (combined data over three seasons used for each trait) were done using IBM SPSS 20.0. The tree diagram (dendrogram) is based on Unweighted Pair Group Method with Arithmetic Mean (UPGMA) method with the Euclidean distance matrix (Sneath and Sokal, 1973) constructed by Darwin version 6. The first two principal components were plotted against each other to find the patterns of bruchid morpho-physiological traits variability among the mungbean genotypes for bruchid resistant using IBM SPSS 20.0.

## RESULTS AND DISCUSSION

The standardized data of fifty-two mungbean genotypes were evaluated and collated in Table 1. The results display the variation of observables of ten morpho-physiological traits of bruchid. The maximum variations are found into percentage of seed damage (POSD), number of adult bruchid emerge (NOAE) and percentage of pest tolerance (POPT). Other morpho-physiological traits are shown to have medium to low variation. Based on the mean value of bruchid morpho-physiological traits through the UPGMA cluster method, the fifty-two mungbean genotypes

**Table 1:** Basic statistics of seven morpho-physiological traits in bruchid resistance screening.

Traits	Pooled Mean $\pm$ Standard Error	Minimum	Maximum
Number of Egg Laid (NOEL)	49.22 $\pm$ 1.14	20.85	76.51
Number of Adult Emerge (NOAE)	30.88 $\pm$ 2.58	8.40	62.63
Percentage of Egg Hatched (POEH)	60.78 $\pm$ 0.99	34.55	85.28
Number of seed Damage (NOSD)	24.46 $\pm$ 1.82	5.71	45.10
Percentage of Seed Damage (POSD)	49.12 $\pm$ 2.66	11.41	90.21
Initial Seed Weight (ISW)	1.80 $\pm$ 0.56	1.00	3.16
Percentage of Seed Weight Loss (POSWL)	43.53 $\pm$ 1.59	5.68	82.96
Mean Development Days (MDD)	24.74 $\pm$ 0.49	17.84	43.09
Percentage of Pest Tolerance (POST)	50.88 $\pm$ 2.15	9.79	88.59
Susceptibility Index (SI)	6.26 $\pm$ 1.69	2.14	9.68

**Table 2:** Cluster analysis and classification with regards to screening of bruchid resistant in mungbean.

Genotypes			
Cluster No.	No. of Genotype	Percentage of Contribution	Name of Genotypes
I	2	3.85	CUM4, PM-11-51.
II	22	42.31	Sonali(B1), PS-16, APDM-116, MC-39, K-851, PM-2, TM-98-20, Howrah Local, Purulia Local, Pusa Vishal, IPM-99-125, IPM-5-7, Pusa-1431, SML-115, NDML-13-11, Pusa-1432, Samrat, MH-909, Sukumar, CUM5, CUS1, CUS3.
III	7	13.46	APDM-84, MH-98-1, SML-302, HUM-8, Panna, KM-139, CUM2.
IV	21	40.38	PTM-11, ML-5, UPM-993, Pusa Baishakhi, Pusa-9632, A-82, Sonamung-1, Baraipur Local, Bankura Local, PantMung-5, VC-639, IPM-205-07, HUM-16, WBM-045, PDM-54, Sonamung-2, CUM3, CUM6, CUS2, CUS4.

**Table 3:** K-Mean presented to morpho physiological traits of four different clusters in mungbean genotypes for screening of bruchid resistance.

Cluster	NOEL	NOAE	POEH	NOSD	POSD	ISW	POSWL	MDD	POPT	SI	Class of SI
I	74.99±2.46	57.91±2.48	77.39±3.22	42.64±2.82	85.27±2.31	1.61±0.52	77.26±5.42	19.26±2.32	14.73±1.45	9.26±1.29	S
II	60.14±1.68	41.67±1.79	69.43±3.01	33.42±2.34	66.85±4.53	1.77±0.82	57.71±4.26	21.30±2.04	33.15±1.75	7.65±2.45	MS
III	27.50±1.18	15.28±1.21	43.75±1.51	11.58±2.17	17.96±2.64	1.86±0.47	11.14±1.28	37.67±3.41	82.04±3.66	2.90±1.05	R
IV	42.58±2.20	22.77±1.56	55.82±2.74	18.07±1.55	37.49±2.02	1.85±0.85	36.25±2.62	24.67±1.52	62.51±2.51	5.64±2.18	MR

**Note:** NOEL: Number of egg laid, NOAE: Number of adult emerge, POEH: Percentage of egg hatched, NOSD: Number of seed damage, POSD: Percentage of seed damage, ISW- Initial Seed Weight, POSWL: Percentage of seed weight loss, MDD: Mean development days, POPT: Percentage of pest tolerance, SI: Susceptible Index, R: Resistant, MR: Moderately Resistant, MS: Moderately Susceptible, S: Susceptible.

have been hierarchically classified into 4 clusters as shown in Fig 1. It displays two major clusters, one medium and one smaller cluster. The results of cluster analysis are shown in Table 2 and the analysis reflects the variation pattern among all the mungbean genotypes for bruchid resistant. The cluster I consists of two genotypes (3.85%) and cluster II consists of twenty two, the maximum number of mungbean genotypes, contributing 42.31%. The cluster III and cluster IV represent seven (13.46%) and twenty one (40.38%) genotypes, respectively. The k-mean cluster shows ten morpho-physiological traits. The mean value of four different clusters are shown in Table 3 and Fig 2. The k-mean cluster identifies cluster III as the most interesting group where seven genotypes exhibited with the low susceptibility index (2.90), very high pest tolerance percentage (82.04), long mean development period in days (37.67), less number of adult bruchid emerge (15.28) and low seed damage percentage (17.96) against bruchid attack. The highest bruchid susceptible group belongs to cluster I (9.68) with maximum seed damage percentage (90.21), seed weight loss percentage (82.96), short mean development period (17.84) and very low pest tolerance percentage (9.79). Cluster II (7.70) and cluster IV (5.64) exhibits respectively, moderately bruchid susceptible and moderately bruchid resistance. The maximum (114.07) inter cluster distance is shown between cluster I and cluster III while minimum (26.83) inter cluster distance is found between cluster III and cluster IV as shown in Table 4. The Eigen values of ten components are presented in the scree plot (Fig 3). Principle component analysis (PCA) shows the relationship of ten bruchid morpho-physiological

**Table 4:** Inter cluster distance between four different clusters basis on the mean values of seven morpho physiological traits in Mungbean genotypes for bruchid resistance.

Cluster	II	III	IV
I	44.08	114.07	87.97
II		71.09	45.42
III			26.83

**Table 5:** Two principle components and their seven morpho-physiological traits in mungbean genotypes.

Traits	PC1	PC2
Number of Egg Laid (NOEL)	0.999	0.046
Number of Adult Emerge (NOAE)	0.888	0.459
Percentage of Egg Hatched (POEH)	0.697	0.717
Number of Seed Damage (NOSD)	0.292	0.956
Percentage of Seed Damage (POSD)	0.977	0.215
Initial Seed Weight (ISW)	0.050	0.999
Percentage of Seed Weight Loss (POSWL)	0.999	-0.053
Mean Development Days (MDD)	-0.039	-0.999
Percentage of Pest Tolerance (POST)	-0.977	-0.215
Susceptibility Index (SI)	0.180	0.984
Eigen Values	6.89	3.11
% of Variance	68.91	31.09
Cumulative %	68.91	100.00

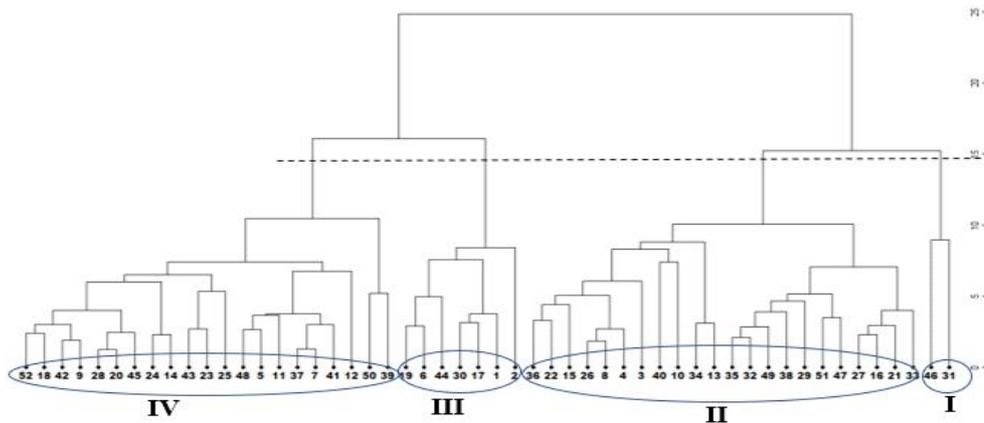


Fig 1: Dendrogram showing four clusters of fifty-two mungbean genotypes for screening of bruchid resistance.

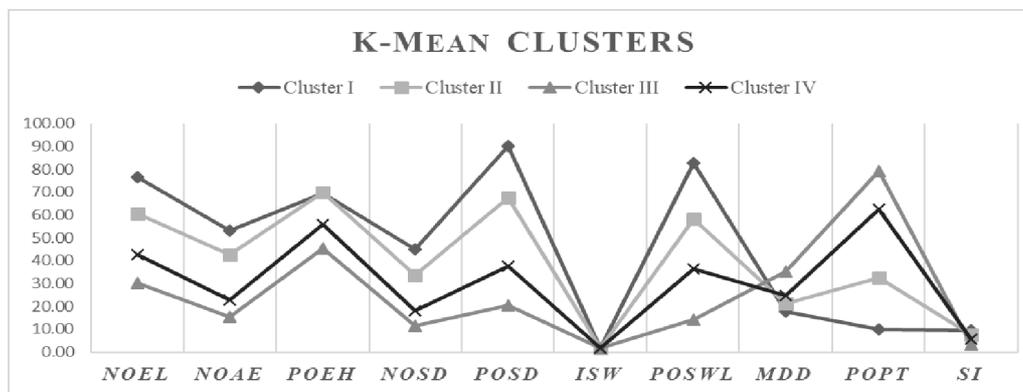


Fig 2: Mean values of ten morpho physiological traits of mungbean genotypes classified into four cluster for bruchid resistant.

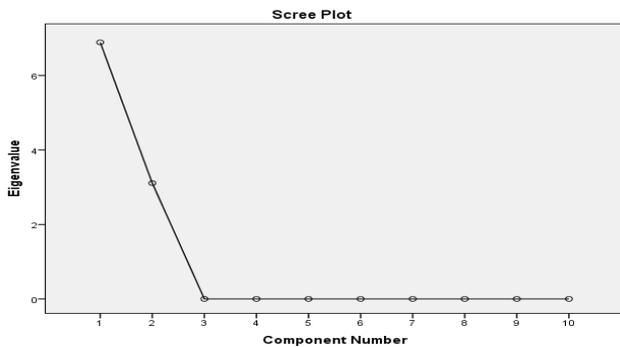
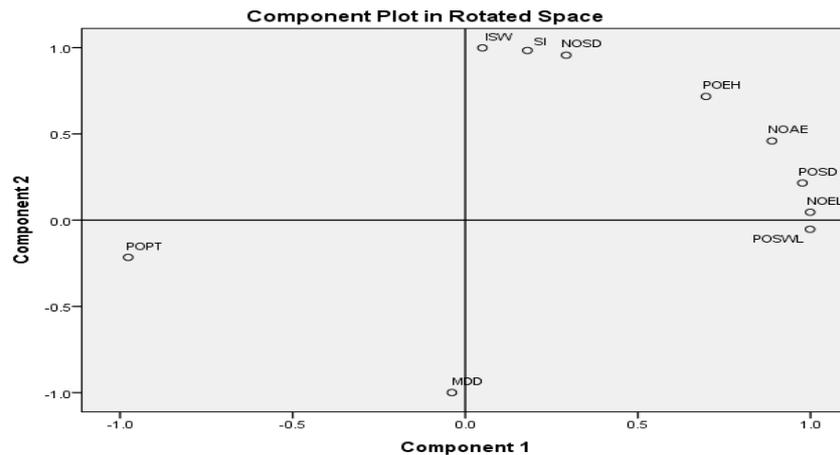


Fig 3: Scree plot constructed for ten principle components.

parameters within PC1 and PC2 among the mungbean genotypes for resistance of bruchid. The eigen values of PC1 and PC2 are found greater than 1 and shown in Table 5. In PC1, five components were more positively contributed with 68.91% of variability while rest of the other five components in PC2 were contributed 31.09% of the variability. The morpho-physiological traits of bruchid such as number of eggs laid (NOEL), percentage of seed damage (POSD), percentage of egg hatched (POEH) are more positively correlated to PC1 whereas the bruchid susceptibility index is correlated more strongly with PC2 as shown in Fig 4. This

result reflects the fact that the most positive correlations are observed between the NOEL and the POSD.

**Sarkar and Bhattacharyya, (2015):** Reported that multivariate analysis aligned the characters as well genotypes in an appropriate manner which is visible proof of conceivable utility of a group. The present study shows medium to low variation in bruchid morphophysiological traits and the dendrogram displays four clusters. Among the four clusters, cluster II exhibits the largest population (42.31%) based on their similarity of morpho-physiological traits whereas cluster III consists of a robust genotypes group. K-mean cluster shows the mean value of ten morpho-physiological traits. Among the four cluster, cluster III shows the low level of seed damage, long mean development period (the period count down starts from the egg laid period to adult bruchid emerge) with low level of seed damage percentage, and high level of pest tolerance. So, these parameters of cluster III reflect low level of bruchid susceptibility index i.e. bruchid resistant (R). However, the opposite trend is observed for cluster I. Fujii and Miyazaki, (1987) reported that TC 1966 was the first bruchid resistant wild cultivar. Ponnusamy *et al*, (2014) reported KPS1 is bruchid susceptible where V1128 and V2817 are the newly reported and V2709 and V2802 were previously reported



**Fig 4:** Scattered diagram of two principle components indicating relationship between ten morpho physiological traits of bruchid resistant. (Note: NOEL: Number of egg laid, POEH: Percentage of egg hatched, POSD: Percentage of seed damage, POSWL: Percentage of seed weight loss, MDD: Mean development days, POPT: Percentage of pest tolerance, SI: Susceptible Index.)

bruchid resistant mungbean lines. The present experiment shows that the seven genotypes in cluster III are also resistant to bruchid. The inter cluster distance are shown to depend on the phenotypic traits variation. When the maximum morphophysiological traits variation found in clusters I and III, the maximum inter cluster distance is found between these clusters (114.07). Principle component analysis reflects the distribution system on the basis of seven morpho-physiological traits of bruchid in mungbean (Mohsen *et al*, 2014). PC1 and PC2 show the relationship among ten morpho-physiological traits where maximum variation (68.91%) is found in PC1. In PC1, the percentage of seed damage (POSD, 0.977) positively correlates with number of eggs laid (NOEL) and number of adults emerge (NOAE) (0.888). This correlation provides the support that POSD depends on both NOEL and NOAE. On the other hand, in PC2 the initial seed weight (ISW, 0.999) and the number of seed damage (NOSD, 0.956) positively correlate with bruchid susceptibility index but the mean development period (MDD, -0.999) shows a negative correlation with bruchid susceptibility index. Therefore, the principle component analysis (PCA) is found to be an important tool

for the identification, utilization, and conservation of bruchid resistant mungbean genotypes (Badii *et al*, 2013 and Mohsen *et al*, 2014). Tomooka *et al*. (2000) exhibited the diversity of evaluated *Vigna sp.* for bruchid resistance and their findings also represented the similar trends of bruchid resistant like of our present study.

## CONCLUSION

The present study was formulated to support the progressive breeding program where multivariate analysis clearly shows a genotypical diversity among a large collection of mungbeans. This in turn identifies the resistant mungbean genotypes in cluster III against the cosmopolitan virulent storage pest bruchid. Thus, mungbean genotypes in cluster III could be a natural choice as potential parent for future hybridization program that provide an opportunity to develop new bruchid resistant lines for mungbean improvement program.

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