



Plant Archives

Journal homepage: <http://www.plantarchives.org>

DOI Url : <https://doi.org/10.51470/PLANTARCHIVES.2026.v26.supplement-2.240>

GENETIC DIVERGENCE ANALYSIS AND EVALUATION OF RESISTANCE TO MUNGBEAN YELLOW MOSAIC VIRUS (MYMV) IN GREEN GRAM (*Vigna radiata* L. Wilczek)

Katariya Maheshkumar Atubhai¹, Manju Singh², Chanchal Shakyawal¹, Rishabh Shukla^{1*} and Simran Singh¹

¹Department of Genetics and Plant Breeding, Navsari Agricultural University, Navsari, Gujarat, India, 396450

²ASPEE Shakilam Biotechnology Institute, Navsari Agricultural University, Surat, Gujarat, India, 395007

*Corresponding author E-mail: shuklarishabh1997@gmail.com

(Date of Receiving : 05-04-2026; Date of Revision : 25-05-2026; Date of Acceptance : 10-06-2026)

ABSTRACT

Mungbean (*Vigna radiata*) is a nutritionally important pulse crop widely cultivated in tropical and subtropical regions is constrained by low productivity arising from narrow genetic variability and susceptibility to major biotic stresses particularly Mungbean Yellow Mosaic Virus (MYMV). In this study genetic diversity among 96 mungbean genotypes was investigated using multivariate cluster analysis and the usefulness of genetic divergence for identifying MYMV-responsive materials was assessed. Genotypes were evaluated under field conditions and analyzed through Mahalanobis' D² distance and K-means clustering based on morphological and agronomic traits. The clustering partitioned the genotypes into four distinct, non-overlapping clusters, demonstrating substantial genetic divergence within the germplasm. Cluster II comprised the highest number of genotypes (38) indicating relatively closer relatedness among its members whereas Cluster I contained the fewest genotypes (13) suggesting comparatively higher divergence. Intra- and inter-cluster distance estimates revealed that inter-cluster distances exceeded intra-cluster distances with the largest genetic separation observed between Cluster I and Cluster IV followed by Cluster I and Cluster III. Such divergence patterns imply that crosses between genotypes belonging to widely separated clusters may enhance heterosis and increase the probability of obtaining superior recombinants. Screening against MYMV further showed a wide range of disease incidence and genotypes were categorized into different resistance classes using a standard rating scale identifying several highly resistant entries along with highly susceptible materials. Overall, the combined evidence of genetic divergence and MYMV reaction provides a practical basis for selecting diverse parents and resistant donors to accelerate mungbean breeding.

Keywords: Genetic diversity, Cluster analysis, Mahalanobis' D², K-means clustering, Mungbean Yellow Mosaic Virus (MYMV), Disease resistance.

Introduction

Green gram [*Vigna radiata* (L.) Wilczek] $2n = 2x = 22$ is an important edible legume crop cultivated almost all over India after chickpea and pigeon pea (Singh *et al.* 2026). Mungbean is an important pulse crop widely cultivated for its high nutritional value and adaptability to diverse agro-climatic conditions. Every crop needs genetic diversity because it creates opportunities for growth, selection and hybridization (Shakyawal *et al.*, 2024). Cluster analysis, a multivariate statistical technique, is widely used to

classify genotypes into distinct groups based on their genetic divergence (Rao, 1952). The application of Mahalanobis' D² statistics provides a reliable measure of genetic distance and helps in grouping genotypes into clusters with similar characteristics (Mahalanobis, 1936). Such classification facilitates the identification of genetically diverse parents, which are crucial for achieving higher heterosis and generating superior recombinants. In mungbean, cluster analysis has been effectively utilized to assess genetic divergence and to group genotypes into distinct clusters, reflecting their

genetic relationships and variability. Genotypes belonging to widely separated clusters are considered more diverse and are likely to produce better segregants in breeding programmes. Therefore, combining genetic divergence analysis with disease resistance screening provides a strategic approach for selecting promising genotypes. Despite its significance, productivity remains low due to limited genetic variability and vulnerability to several biotic stresses. Among these, *Mungbean Yellow Mosaic Virus* (MYMV) is one of the most devastating diseases, causing substantial yield losses in mungbean-growing regions. The disease is transmitted by the whitefly (*Bemisia tabaci*) and is characterized by yellow mosaic symptoms on leaves, leading to reduced photosynthetic efficiency and poor seed development. Therefore, systematic screening of germplasm against MYMV is essential for identifying resistant genotypes and developing durable resistant cultivars.

Hence, the present study emphasizes the simultaneous evaluation of mungbean genotypes through cluster analysis and screening against MYMV to identify genetically diverse and disease-resistant genotypes for future breeding programmes.

Material and Methods

The experimental material for the present study includes 96 greengram genotypes. The experiment was carried out during the Summer 2021 season at the College Farm, N.M. College of Agriculture, Navsari Agricultural University, located in Navsari. The prevailing environmental conditions during the crop season were favourable for normal growth and development of the crop.

The experimental material comprised 96 derived genotypes along with four standard checks including GM-4, GM-6, GM-7 and 40 C were planted using Augmented Block Design, where each and every check was replicated in four blocks. Each genotype was sown in a single row consisting of 20 plants, maintaining a spacing of 60 cm between rows and 15 cm between plants. All recommended agronomic practices were followed to ensure optimal crop growth. Observations were recorded on five randomly selected plants per genotype for various quantitative traits including Days to 50 % flowering, Days to maturity, Plant height (cm), Primary branches per plant, Pods per cluster, Pods per plant, Pod length (cm), Seeds per pod, 100 seed weight (g), Seed yield per plant (SYPP) (g), and Mungbean Yellow Mosaic Virus (MYMV) disease incidence (%). The data recorded for the different characters excluding MYMV incidence were subjected to analysis of variance. Different parameters of the genetic

variability were computed by standard statistical procedures. MYMV disease incidence recorded at 60 days after sowing and at the time of peak reproductive stage.

Statistical Analysis

Cluster analysis was employed to classify genotypes into distinct groups based on the degree of genetic divergence (Rao, 1952). The Mahalanobis D² statistics were computed for all possible pairwise combinations (Mahalanobis, 1936). In the present study, non-hierarchical clustering was performed using the K-means clustering algorithm, as described by Forgy (1965) and MacQueen (1967). This method partitions the entire dataset into a predefined number of clusters (K) by minimizing the within-cluster sum of squares (WCSS) and maximizing the variation between clusters. Each cluster is represented by a centroid, and genotypes are assigned to clusters based on their Euclidean distance from the respective centroids (MacQueen, 1967).

Result and Discussion

Divergence Analysis

The genetic divergence among 100 mungbean genotypes of *Vigna radiata* was assessed using K-means clustering based on 19 morphological, physiological, and yield-related traits. The analysis grouped all genotypes into four distinct and non-overlapping clusters indicating the presence of substantial genetic variability among the experimental material (Table 1). The distribution of genotypes across clusters was uneven. Cluster II was the largest, comprising 38 genotypes followed by Cluster IV with 31 genotypes, Cluster III with 18 genotypes and Cluster I with 13 genotypes. The predominance of genotypes in Cluster II suggests a higher degree of genetic similarity among its members indicating a comparatively narrow genetic base. In contrast Cluster I, with the least number of genotypes represents relatively more diverse and unique genotypes. Such uneven distribution of genotypes among clusters reflects the existence of substantial variability which is essential for effective selection and hybridization programmes. Similar clustering patterns in mungbean have been reported by Abna *et al.* (2011) and Verma *et al.* (2016) who also observed unequal distribution of genotypes across clusters indicating differential genetic diversity. The intra- and inter-cluster distances provided a clear understanding of the magnitude of genetic divergence among and within clusters (Table 2). The intra-cluster distance was highest in Cluster II (18.16), followed by Cluster IV (17.86), Cluster III (17.32), and Cluster I (16.94), suggesting relatively

higher heterogeneity within Cluster II. The inter-cluster distances were comparatively higher than intra-cluster distances indicating substantial genetic divergence among clusters. The maximum inter-cluster distance was observed between Cluster I and Cluster IV (28.81) followed by Cluster I and Cluster III (19.37) suggesting that genotypes belonging to these clusters are genetically most diverse. Conversely the minimum inter-cluster distance was recorded between Cluster II and Cluster IV (13.41) indicating closer genetic relationship between these clusters. The presence of wide inter-cluster distances suggests that hybridization between genotypes from genetically distant clusters would result in greater heterosis and increased chances of obtaining superior transgressive segregants. This observation is in agreement with the findings of Murty and Arunachalam (1966) who emphasized that selection of parents based on genetic divergence rather than geographical origin is more effective in crop improvement programmes. Similar conclusions were also reported by Dhivyabharathi *et al.* (2019), Datta *et al.* (2012), Gadakh *et al.* (2013), Bisti *et al.* (2022), Patel *et al.* (2021), Shulee *et al.* (2020) and Singh *et al.* (2015) who highlighted the importance of inter-cluster distances in selecting diverse parents. The cluster mean values for ten important characters revealed considerable variation among clusters in mungbean (*Vigna radiata*) indicating the presence of substantial genetic diversity among the genotypes (Table 3). Among the four clusters, Cluster I exhibited superior performance for most of the yield and yield-contributing traits recording the highest grain yield per plant (9.12 g), plant height (73.81 cm), pods per plant (19.56), pods per cluster (3.57), pod length (6.85 cm), seeds per pod (9.85), and seed index (4.13 g). These results suggest that genotypes grouped in Cluster I possess better yield potential and overall agronomic performance. In contrast Cluster III recorded the lowest grain yield per plant (5.78 g) along with comparatively higher values for days to first flowering (51.94 days) and days to maturity (76.33 days) indicating that genotypes in this cluster are late maturing and relatively poor yield. Cluster II showed moderate performance with grain yield per plant of 7.92 g and relatively lower days to first flowering (40.76 days), and maturity (69.65 days) indicating early to medium maturity along with acceptable yield levels. Similarly Cluster IV was characterized by the lowest plant height (46.40 cm), pods per plant (12.79), pods per cluster (2.66), and seeds per pod (8.51) resulting in comparatively lower grain yield (6.02 g) reflecting poor yield performance of genotypes in this cluster. With respect to earliness Cluster II (40.76 days) and Cluster IV (40.78 days) recorded minimum

days to first flowering indicating early flowering behaviour whereas Cluster III (51.94 days) was the highest. A similar result was observed for days to maturity where Cluster IV (69.26 days) was earliest and Cluster III (76.33 days) was highest. Earliness is an important trait for escaping terminal drought and fitting into multiple cropping systems. The variation observed among cluster means for yield and its component traits indicates that traits such as pods per plant, seeds per pod, and seed index played a major role in determining yield performance. The superior performance of Cluster I for these traits clearly explains its highest grain yield. The results are in agreement with earlier findings of Gadakh *et al.* (2013), Kanavi *et al.* (2020), Mohan *et al.* (2019), Shibli *et al.* (2021), and Prasanna *et al.* (2013) who also reported significant variation among cluster means and emphasized the importance of yield-contributing traits in determining genetic divergence in mungbean.

MYMV Analysis

The incidence of Mungbean Yellow Mosaic Virus (MYMV) was recorded in one hundred mungbean genotypes of *Vigna radiata* under natural field conditions. The disease transmitted by the whitefly (*Bemisia tabaci*) is one of the most serious constraints in mungbean production causing significant yield losses due to reduced photosynthetic area, stunted growth, and poor pod and seed development (Malathi *et al.*, 2017). A wide range of variation was observed among genotypes for MYMV incidence indicating the presence of substantial genetic variability for disease resistance. The percent disease incidence ranged from 0 to more than 70%, clearly demonstrating differential response of genotypes to MYMV infection (Table 4). Based on the standard disease rating scale (0–5), the genotypes were classified into six categories viz., highly resistant (HR), resistant (R), moderately resistant (MR), moderately susceptible (MS), susceptible (S), and highly susceptible (HS) (Sekar and Nalini, 2017). The categorization revealed that four genotypes (NMS-21-1, NMS-21-4, NMS-21-53 and NMS-21-94) were found highly resistant showing no visible symptoms of MYMV infection. A large number of genotypes were grouped under the resistant category (1–10% infection) indicating the presence of considerable resistance sources in the studied material. Several genotypes also exhibited moderately resistant reaction (11–20%), suggesting partial resistance which could be useful in breeding programmes. However, a considerable proportion of genotypes fell under moderately susceptible (21–30%) and susceptible (31–50%) categories indicating moderate vulnerability to MYMV. Furthermore a group of genotypes including

NMS-21-23, NMS-21-24, NMS-21-40, NMS-21-49, NMS-21-66, NMS-21-67, NMS-21-68, NMS-21-69, NMS-21-70, NMS-21-84, NMS-21-88, NMS-21-89, NMS-21-90 and check GM-4 were categorized as highly susceptible (>50% infection) reflecting their poor performance under disease pressure. The presence of both highly resistant and highly susceptible genotypes in the same population indicates a wide genetic base and segregation for MYMV resistance. Such variability is highly desirable for breeding programmes aimed at developing resistant varieties. The observed variation in disease response may be attributed to genetic differences among genotypes as well as environmental influence and vector population dynamics. Similar variability for MYMV resistance in mungbean has been reported by Singh *et al.* (2015) and Prasanna *et al.* (2013) who emphasized the importance of screening germplasm under natural epiphytotic conditions for identifying stable resistance sources. Overall, the cluster mean analysis clearly demonstrated the presence of wide variability among clusters and significant variability for MYMV resistance providing ample scope for selection and hybridization in mungbean improvement programmes.

Conclusion

The present investigation confirmed that the evaluated mungbean genotypes harbor substantial genetic diversity as reflected by K-means clustering into four distinct groups and by consistently higher inter-cluster than intra-cluster distances. The uneven cluster membership suggests varying levels of genetic relatedness among genotypes with Cluster I representing the most diverse set and Cluster II representing comparatively closer genetic relationships. The maximum divergence detected between Cluster I and Cluster IV (and also between Cluster I and Cluster III) indicates that hybridization using parents from these widely separated clusters is likely to generate greater heterosis and improved segregants. In addition, MYMV screening demonstrated substantial phenotypic variation in disease response enabling the identification of resistant sources alongside susceptible genotypes. Therefore, for future breeding it is recommended to prioritize crosses that combine genotypes from genetically distant clusters with those exhibiting low MYMV incidence. This integrative strategy linking genetic divergence with disease resistance will improve the efficiency of parent selection and enhance the development of durable MYMV-resistant mungbean cultivars.

Table 1: Clustering pattern of 100 mungbean genotypes on the basis of k-means cluster analysis for all traits

Cluster	No. of genotypes included	Members
I	13	NMS-21-9, NMS-21-20, NMS-21-21, NMS-21-22, NMS-21-23, NMS-21-24, NMS-21-32, NMS-21-47, NMS-21-48, NMS-21-71, NMS-21-76, NMS-21-81, NMS-21-82
II	38	NMS-21-1, NMS-21-2, NMS-21-3, NMS-21-14, NMS-21-15, NMS-21-16, NMS-21-17, NMS-21-18, NMS-21-19, NMS-21-25, NMS-21-27, NMS-21-29, NMS-21-30, NMS-21-31, NMS-21-33, NMS-21-34, NMS-21-40, NMS-21-42, NMS-21-43, NMS-21-51, NMS-21-52, NMS-21-53, NMS-21-54, NMS-21-56, NMS-21-57, NMS-21-58, NMS-21-59, NMS-21-61, NMS-21-69, NMS-21-70, NMS-21-72, NMS-21-80, NMS-21-83, NMS-21-93, NMS-21-94, NMS-21-95, NMS-21-96, 40C
III	18	NMS-21-8, NMS-21-28, NMS-21-36, NMS-21-41, NMS-21-44, NMS-21-45, NMS-21-46, NMS-21-50, NMS-21-62, NMS-21-63, NMS-21-64, NMS-21-65, NMS-21-66, NMS-21-67, NMS-21-68, NMS-21-74, NMS-21-75, NMS-21-78
IV	31	NMS-21-4, NMS-21-5, NMS-21-6, NMS-21-7, NMS-21-10, NMS-21-11, NMS-21-12, NMS-21-13, NMS-21-26, NMS-21-35, NMS-21-37, NMS-21-38, NMS-21-39, NMS-21-49, NMS-21-55, NMS-21-60, NMS-21-73, NMS-21-77, NMS-21-79, NMS-21-84, NMS-21-85, NMS-21-86, NMS-21-87, NMS-21-88, NMS-21-89, NMS-21-90, NMS-21-91, NMS-21-92, GM-4, GM-5, GM-6

Table 2: Estimates of average inter-cluster distance for 4 clusters in mungbean genotypes

Cluster	I	II	III	IV
I	-	15.92	19.37	28.81
II		-	13.48	13.41
III			-	17.41
IV				-

Table 3: Cluster means of ten characters in mungbean

Clusters	Centres									
	DFF	DM	PH	PBPP	PPC	PPP	PL	SPP	SI	SYPP
I	44.54	71.77	73.81	2.91	3.57	19.56	6.85	9.85	4.13	9.12
II	40.76	69.65	58.67	2.83	3.52	17.60	6.78	9.71	3.87	7.92
III	51.94	76.33	57.38	2.72	3.05	15.32	6.73	9.61	3.79	5.78
IV	40.78	69.26	46.40	2.57	2.66	12.79	6.51	8.51	3.86	6.02

Table 4: Categorization of mungbean genotypes based on MYMV incidence percentage

Disease scale	Percent infection	Infection category	Reaction group	Genotypes
0	All plant free of disease symptoms	Highly resistant	HR	NMS-21-1, NM-21-4, NMS-21-53, NMS-21-94
1	1-10	Resistant	R	NMS-21-2, NMS-21-5, NMS-21-6, NMS-21-7, NMS-21-8, NMS-21-10, NMS-21-11, NMS-21-12, NMS-21-14, NMS-21-15, NMS-21-20, NMS-21-22, NMS-21-28, NMS-21-32, NMS-21-33, NMS-21-34, NMS-21-35, NMS-21-36, NMS-21-38, NMS-21-42, NMS-21-44, NMS-21-46, NMS-21-48, NMS-21-50, NMS-21-52, NMS-21-54, NMS-21-55, NMS-21-59, NMS-21-61, NMS-21-63, NMS-21-64, NMS-21-71, NMS-21-72, NMS-21-73, NMS-21-74, NMS-21-76, NMS-21-78, NMS-21-80, NMS-21-85, NMS-21-86, NMS-21-87, NMS-21-92, NMS-21-95, NMS-21-96, 40C
2	11-20	Moderately resistant	MR	NMS-21-3, NMS-21-9, NMS-21-13, NMS-21-21, NMS-21-27, NMS-21-29, NMS-21-30, NMS-21-31, NMS-21-37, NMS-21-56, NMS-21-57, NMS-21-60, NMS-21-62, NMS-21-65, NMS-21-75, NMS-21-77, NMS-21-79, NMS-21-81, NMS-21-82, NMS-21-83, NMS-21-91, NMS-21-93, GM-6
3	21-30	Moderately susceptible	MS	NMS-21-17, NMS-21-18, NMS-21-19, NMS-21-25, NMS-21-26, NMS-21-39, NMS-21-45, NMS-21-47, NMS-21-51, NMS-21-58, GM-7
4	31-50	Susceptible	S	NMS-21-16, NMS-21-41, NMS-21-43
5	More than 50	Highly susceptible	HS	NMS-21-23, NMS-21-24, NMS-21-40, NMS-21-49, NMS-21-66, NMS-21-67, NMS-21-68, NMS-21-69, NMS-21-70, NMS-21-84, NMS-21-88, NMS-21-89, NMS-21-90, GM-4

Acknowledgement

The authors would like to thank the Department of Genetics and Plant breeding, N.M.C.A., NAU, Navsari for providing the necessary facilities required for this study.

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