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MORPHOLOGICAL CHARACTERIZATION AND MULTIVARIATE ANALYSIS OF DIFFERENT GENOTYPES OF MUNGBEAN (VIGNA RADIATA (L.) R. WILCZEK)

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ABSTRACT

Mungbean (*Vigna radiata* (L.) R. Wilczek) is versatile crop mainly grown in subtropical regions with wideranging agricultural and nutritional benefits. Despite being the largest producer in the world, the productivity of mungbean is well below in India due to limited morphological variability observed in working collection of mungbean. In the present investigation, we have conducted morphological characterization and multivariate analysis to explore the genetic diversity among the 26 genotypes of mungbean (*Vigna radiata* (L.) R. Wilczek). These 26 accessions of mungbean procured from the Pulses Research Station, SDAU, Gujarat, and local markets were grown for evaluation in Randomized Block Design (RBD) with three replications during June to August 2021. Morphological observations recorded at different stages of life for the grown genotypes. Significant variations observed in morphological traits of different genotypes. Principal component analysis and cluster analysis discriminated following genotypes such as VM, GM-6, SKNM-1608, SKNM-1701, SKNM-1704, SKNM-1801, SKNM-1802, SKNM-1806, and SKNM-1808 based on the morphological observations noted above. These genotypes can be recommended to use as parent for further plant breeding programmes to develop new variety and conduct field trials for different locations and climates.

Keywords: Vigna radiata (L.) R. Wilczek, Morphological traits, Principal component analysis, Cluster analysis

INTRODUCTION

Mungbean (*Vigna radiata* (L.) R. Wilczek) is versatile and valuable crop with wide-ranging agricultural and nutritional benefits. This crop is mainly grown in pan-tropical regions of South Asia, Australia, North and South America, Central Africa, and Parts of China (Tahir *et al.*, 2020; Krishna *et al.*, 2023). Several key attributes including their adaptability to various environmental factors, contribution to soil fertility through nitrogen fixation, use in crop rotation practices, and highly nutritious properties make mungbeans an essential component of agricultural systems worldwide. In addition, their easy digestibility, and minimal flatulence contribute to their popularity among consumers, adding their overall importance in the field of leguminous crops (Lavanya *et al.*, 2008). It is a rich source of essential nutrients, minerals, and fibres, and play a crucial role in the humane diet. Their exceptional nutritional content positions them as a top choice among pulses, and underscores their potential to address protein deficiency in the humane diet (Divyaramakrishnan and Savithramma, 2014; Vir and Singh, 2016). Mungbean related research initiatives are actively being undertaken in various countries, with coordination made possible through an international network. This coordinated endeavour results from the understanding of the significant economic and nutritional value of mungbean. It is essential to increase yield per unit area to meet the growing demand for mungbeans. This can be achieved through the development of novel cultivars and improving cultural practices (Tahir *et al.*, 2020).

Mungbean occupies leading position as imported pulse crop in India. Despite being the largest producer in the world, the productivity of mungbean is well below in India than that of other countries. A critical obstacle to its improvement lies in the limited morphological variability observed in working collection of mungbean (Chattopadhyay *et al.*, 2011). This lack of polymorphism can be attributed to several factors, among which the major one is heavy reliance on limited number of genotypes with high degree of relatedness as parent for cultivar development programmes. In addition, the rich genetic diversity found in the gene pool of the Indian subcontinent, which serve as the epicentre of mungbean diversity, is underutilized (NB et al., 2015).



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Traditional breeding techniques have slowed down the development of novel mungbean cultivars with traits that considerably improve the yield quality. Accessing the detailed data on genetic diversity available in the germplasm repositories is therefore crucial. This is necessary to ensure the efficient use of germplasm and, ultimately successfully completion of mungbean breeding programmes (Mwangi *et al.*, 2021). A through characterization of existing germplasm holdings and collections is essential to exploit the potential of mungbean gene pool for the development of superior and promising cultivars. The goal of this characterization is to determine variations among genotypes that can be used in enhancement programmes. The best responsive results for genetic augmentation are frequently obtained via cross breeding techniques involving parents with higher genetic divergence. In this regards, agro-morphological characterization of genetic resources is playing a key role in developing novel and desirable plant varieties. These new plant varieties are essential for raising agricultural productivity and improving quality of obtained produces (Evgenidis *et al.*, 2011; Joshi *et al.*, 2022). In the present investigation, we have conducted multivariate analysis and explored the genetic diversity among the 26 genotypes of mungbean (*Vigna radiata* (L.) R. Wilczek) based on morphological characterization.

MATERIALS AND METHOD

Plant Material

The plant material contains 26 accessions of Mungbean (*Vigna radiata* (L.) R. Wilczek) from which 20 accessions procured from the Pulses Research Station, Sardarkrushinagar Dantiwada Agricultural University, Gujarat, and 06 accessions collected from the market and grown for evaluation in Randomized Block Design (RBD) with three replications during June to August 2021. Morphological observations recorded at different stages of life for the grown genotypes. The leaves, stems, pods, and seeds were also collected at different stages for qualitative and quantitative morphological measures. The names of genotypes are listed in Table 1.

Table 1 List of genotypes with their sample code

Sr. No.	Genotype	Sr. No.	Genotype
1	Western Mahavir (VM)	14	SKNM-1608
2	Vaishakhi Research Mungbean (VRM)	15	SKNM-1701
3	Neel++ Research Mungbean (NRM)	16	SKNM-1703
4	Avani-65 Research Moong (ARM)	17	SKNM-1704
5	Nidhi Seeds (NS)	18	SKNM-1705
6	Guj-4	19	SKNM-1801
7	GM-3	20	SKNM-1802
8	GM-4	21	SKNM-1803
9	K-851	22	SKNM-1806
10	GMM-5	23	SKNM-1808
11	GM-6	24	SKNM-1809
12	SKNM-1516	25	SKNM-1810
13	SKNM-1605	26	SKNM-1812

Record of Morphological Observations

The qualitative and quantitative morphological observations of the cultivated genotypes of mungbean were recorded at different stages in the field and in the laboratory. Qualitative characteristics such as growth habit, colour, shape, and lustre were recorded in natural daylight based on visual observations. The morphometric characteristics like height, length, and size were recorded in both field and laboratory. The following morphological observations recorded for the present study:

1) Time of Flowering:

The genotypes were categorized as early (<40 days), medium (40-50 days) and late (>50 days) based on observations recorded as visual assessment for the number of days required from sowing to flower emergence in 50% of plant population in each genotype.

2) Plant Growth Habit:

The genotypes were categorized as erect, semi-erect and spreading based on observations recorded at 50% flowering.

3) Plant Height (cm):

The genotypes were categorized as short (<50 cm), medium (50-70 cm), and long (>70 cm) based on height measured at maturity from ground level to the tip of the main stem of plant.

4) Stem Colour:

The genotypes were categorized as green, green with purple splashes, and purple based on visual observations recorded at maturity.

5) Stem Pubescence:



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The genotypes were categorized based on presence and absence of pubescence on mature stem of plants.

6) Leaflet Lobes:

The genotypes were categorized based on presence and absence of lobes in mature leaves.

7) Leaf Colour:

The genotypes were categorized based on visual assessment of green and dark green colour of leaves.

8) Leaf Vein Colour:

The genotypes were categorized as green, greenish purple, and purple based on visual assessment of vein colour in mature leaves of each genotype.

9) Petiole Colour:

The genotypes were categorized as green, green with purple splashes, and purple based on visual observations recorded at maturity.

10) Flower Petal Colour:

The genotypes were categorized based on visual assessment of yellow and light-yellow colour of flower petal at 50% maturity stage under natural day light.

11) **Pod Colour:**

The genotypes were categorized based on visual assessment of brown and black colour of mature pod observed under natural illumination.

12) Pod Length (cm):

The genotypes were categorized as short (<8 cm), medium (8-10 cm), and long (>10 cm) based on length of mature pod measured at maturity.

13) **Pod Curvature:**

The genotypes were categorized as straight and curved based on curvature observed in mature pods.

14) Seed Colour:

The genotypes were categorized as yellow, green, mottled and black based of visual observation of seed colour recorded under natural illuminations.

15) Seed Lusture:

The genotypes were categorized based on shiny and dull lusture of seeds.

16) Seed Shape:

The genotypes were categorized based on oval and drum shaped seeds of each genotype observed under magnifying lens.

17) Seed Size (g):

The genotypes were categorized as small (<3 g), medium (3-5 g), and large (>5 g) based on weight of 100 seeds of each genotype.

Statistical Analysis

Two techniques of multivariate analysis, Principal Component Analysis (PCA) and Cluster Analysis were performed by PAST version 4.03 software.

RESULTS AND DISCUSSION

Morphological Observations

Mungbean (*Vigna radiata* (L.) R. Wilczek) genetic improvement initiatives began in Indian in third decade of 20th century, but they really took off after 1967 with the establishment of All India Coordinated Pulse Improvement Programme. In the past, varietal development in mungbean relied heavily on the practices of hybridization and selection, which was centred on achieving objectives of high grain yield, disease resistance, dwarf plant types, spreading branches and other conventional traits. However, significant transformations in breeding strategies occurred past-1990s, a trend shifted towards reduced plant height, maturity duration, non-twining and erect growth habit, non-droopy pods, synchronous maturity, and determinacy, in addition to conventional yield contributing traits. Such breeding practices led to notable increase in the utilization of intraspecific and interspecific hybridization, and conventional approach direct selection from landraces was gradually pushed to the backseat. As result, a large number of varieties were developed in India by different organizations (Mehandi *et al.*, 2019; Pratap *et al.*, 2021). However modern mungbean varieties, which makeup the majority of overall mungbean production, were developed only using narrow spectrum of mungbean variability. This means that there is still a lot of untapped potential in mungbean variability that could be used to develop new varieties with improved traits.

Qualitative morphological traits were recognized for their important role in the evolution of diversity, influenced by both natural selections and deliberate human intervention. Significant variations observed among studied landraces, primarily based on their morphological characters. These characters have been carefully documented through visual assessment and quantitative approaches at different stages of plant growth. Table 2 below presents the recorded phenotypic results derived from observations made in the field and laboratory.



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Consumer preferences are greatly influenced by the qualitative characteristics of mungbean seeds such as, seed shape, colour, and lustre. For example, in the eastern states of India, small mungbean seeds are in high demand and commands a price premium compared to bold seeds. Likewise, the green hypocotyl kind of mungbeans were preferred above purple ones in bean sprouting industries. The bright green seed lustre is generally picked over dull lustre, and mature pods with black colour are coveted for their capacity to lessen seed discoloration. A number of features, including early flowering, early maturation, pod length, seed weight etc., are directly relevant to crop improvement (Tripathy et al., 2015; Gayacharan et al., 2020). Several other variables, including growth habit, leaflet shape and size, leaf and pod pubescence, are allied to plant architecture. These traits are prime indicator of variation in development of improved cultivars through distant hybridization (Pratap et al., 2021). Majority of the genotypes in the current study rendered aforementioned traits, with many of them exhibiting bright shiny seed lustre (VM, NRM, GM-4, SKNM-1516, SKNM-1608, SKNM-1701, SKNM-1704, SKNM-1705, SKNM-1801, SKNM-1802, SKNM-1803, SKNM-1810, SKNM-1812), although only single genotype was found to have small seed shape viz SKNM-1704 (Fig 1). The dark black colour of pods was observed in VRM, ARM, Guj-4, GMM-5, GM-6, SKNM-1516, SKNM-1605, SKNM-1608, SKNM-1806, SKNM-1808, SKNM-1809 (Fig 2). High demand early flowering trait was also observed in VM, VRM, NRM, ARM, GM-4, K-851, SKNM-1516, SKNM-1605, SKNM-1705, SKNM-1801, SKNM-1803, SKNM-1806, SKNM-1812. Promising accessions of mungbean have been identified that exhibit high grain weight (VM, VRM, NRM, GM-3, GM-4, GMM-5, GM-6, SKNM-1516, SKNM-1801, SKNM-1806, SKNM-1808, SKNM-1812), and longer pods (Guj-4, GM-6, SKNM-1704, SKNM-1801, SKNM-1806, SKNM-1808). These accessions can be subjected to multilocation varietal yield trials, which could potentially lead to their direct release as varieties.

Sr. No.	Characteristics	States	Example Varieties
1	Time of flowering	Early (<40 days)	VM, VRM, NRM, ARM, GM-4, K-851, SKNM-1516, SKNM- 1605, SKNM-1705, SKNM-1801, SKNM-1803, SKNM-1806, SKNM-1812
		Medium (40- 50 days)	NS, Guj-4, GMM-5, SKNM-1608, SKNM-1701, SKNM-1703, SKNM-1704, SKNM-1808, SKNM-1810
		Late (>50 days)	GM-3, GM-6, SKNM-1802, SKNM-1809
2	Plant: Growth habit	Erect	VM, GM-3, GMM-5, SKNM-1605, SKNM-1701, SKNM-1703, SKNM-1704, SKNM-1808
		Semi-erect	VRM, NRM, ARM, NS, GM-4, GM-6, SKNM-1608, SKNM- 1801, SKNM-1803, SKNM-1806, SKNM-1810
		Spreading	Guj-4, K-851, SKNM-1516, SKNM-1705, SKNM-1802, SKNM- 1809, SKNM-1812
	Plant: Height	Short (<50 cm)	GM-6, SKNM-1810, SKNM-1812
3		Medium (50- 70 cm)	VRM, Guj-4, K-851, SKNM-1516, SKNM-1701, SKNM-1704, SKNM-1806, SKNM-1808
		Long (>70 cm)	VM, NRM, ARM, NS, Guj-4, GM-3, GM-4, GMM-5, SKNM- 1605, SKNM-1608, SKNM-1703, SKNM-1705, SKNM-1801, SKNM-1802, SKNM-1803, SKNM-1809
4	Stem: Colour	Green	VM, NRM, ARM, NS, GM-4, GM-6, SKNM-1516, SKNM-1801, SKNM-1803, SKNM-1808, SKNM-1809, SKNM-1810, SKNM- 1812
		Green with purple splashes	VRM, GM-3, K-851, GMM-5, SKNM-1605, SKNM-1608, SKNM-1701, SKNM-1703, SKNM-1704, SKNM-1705, SKNM- 1802, SKNM-1806
		Purple	
5	Stem: Pubescence	Absent Present	K-851 VM, VRM, NRM, ARM, NS, Guj-4, GM-3, GM-4, GMM-5, GM- 6, SKNM-1516, SKNM-1605, SKNM-1608, SKNM-1701, SKNM-1703, SKNM-1704, SKNM-1705, SKNM-1801, SKNM- 1802, SKNM-1803, SKNM-1806, SKNM-1808, SKNM-1809, SKNM-1810, SKNM-1812

 Table 2 Morphological Observations



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6	Leaflet: Lobes	Absent	VM, NRM, ARM, NS, Guj-4, GM-3, GM-4, GMM-5, GM-6, SKNM-1516, SKNM-1605, SKNM-1608, SKNM-1701, SKNM- 1703, SKNM-1704, SKNM-1705, SKNM1801, SKNM-1802, SKNM-1803, SKNM-1806, SKNM-1808, SKNM-1810, SKNM 1812		
		Present	VRM, NS, K-851, SKNM-1809		
7		Green	ARM, GMM-5, SKNM-1605, SKNM-1704, SKNM-1801, SKNM-1802, SKNM-1809		
	Leaf: Colour	Dark green	VM, VRM, NRM, NS, Guj-4, GM-3, GM-4, K-851, GM-6, SKNM-1516, SKNM-1608, SKNM-1701, SKNM-1703, SKNM- 1705, SKNM-1803, SKNM-1806, SKNM-1808, SKNM-1810, SKNM1812		
8		Green	Guj-4, GM-4, GM-6, SKNM-1516, SKNM-1701, SKNM-1801, SKNM-1803, SKNM-1810, SKNM-1812		
	Leaf: Vein colour	Greenish purple	K-851, GMM-5, SKNM-1605, SKNM-1608, SKNM-1704, SKNM-1802, SKNM-1806, SKNM-1808, SKNM-1809		
		Purple	VM, VRM, NRM, ARM, NS, GM-3, SKNM-1703, SKNM-1705		
		Green	NRM, Guj-4, GM-4, GM-6, SKNM-1516, SKNM-1801, SKNM- 1803, SKNM-1806, SKNM-1810, SKNM-1812		
9	Petiole: Colour	Green with purple splashes	VM, VRM, ARM, NS, GM-3, K-851, GMM-5, SKNM-1605, SKNM-1608, SKNM-1701, SKNM-1704, SKNM-1802, SKNM- 1808, SKNM-1809		
		Purple	SKNM-1703, SKNM-1705		
10	Flower: Petal colour	Yellow	VM, NRM, Guj-4, GM-3, GM-4, K-851, GMM-5, GM-6, SKNM- 1516, SKNM-1608, SKNM-1701, SKNM-1705, SKNM-1803, SKNM-1809, SKNM-1810		
		Light yellow	VRM, ARM, NS, SKNM-1605, SKNM-1703, SKNM-1704, SKNM-1801, SKNM-1802, SKNM-1806, SKNM-1808, SKNM- 1812		
11	Pod: Colour	Brown	VM, NRM, NS, GM-3, GM-4, K-851, SKNM-1608, SKNM- 1701, SKNM-1703, SKNM-1704, SKNM-1705, SKNM-1801, SKNM-1802, SKNM-1803, SKNM-1810, SKNM-1812		
		Black	VRM, ARM, Guj-4, GMM-5, GM-6, SKNM-1516, SKNM-1605, SKNM-1708, SKNM-1806, SKNM-1808, SKNM-1809		
12	Pod: Length	Short (<8 cm)	GM-3, SKNM-1701, SKNM-1802		
		Medium (8- 10 cm)	VM, VRM, NRM, ARM, NS, GM-4, K-851, GMM-5, SKNM- 1516, SKNM-1605, SKNM-1608, SKNM-1703, SKNM-1704, SKNM-1803, SKNM-1809, SKNM-1810, SKNM-1812		
		Long (>10 cm)	Guj-4, GM-6, SKNM-1705, SKNM-1801, SKNM-1806, SKNM- 1808		
13	Pod: Curvature	Straight	VM, VRM, NRM, ARM, NS, GM-4, SKNM-1516, SKNM-1605, SKNM-1608, SKNM-1703, SKNM-1802, SKNM-1803, SKNM- 1809, SKNM-1810, SKNM-1812		
		Curved	Guj-4, GM-3, SKNM-1701, SKNM-1705, K-851, GMM-5, GM- 6, SKNM-1704, SKNM-1801, SKNM-1806, SKNM-1808		
14	Seed: Colour	Green	VM, VRM, NRM, ARM, NS, Guj-4, GM-3, GM-4, K-851, GMM-5, GM-6, SKNM-1516, SKNM-1605, SKNM-1608, SKNM-1701, SKNM-1703, SKNM-1704, SKNM-1705, SKNM- 1801, SKNM-1802, SKNM-1809, SKNM-1810, SKNM-1812		
		Mottled	SKNM-1803, SKNM-1806, SKNM-1808		
15	Seed: Lusture	Shiny	VM, NRM, GM-4, SKNM-1516, SKNM-1608, SKNM-1701, SKNM-1704, SKNM-1705, SKNM-1801, SKNM-1802, SKNM- 1803, SKNM-1810, SKNM-1812		
		Dull	VRM, ARM, NS, Guj-4, GM-3, k-851, GMM-5, GM-6, SKNM- 1605, SKNM-1703, SKNM-1806, SKNM-1808, SKNM-1809		
16	Seed: Shape	Oval Drum shaped	VM, NRM, GMM-5, SKNM-1701, SKNM-1704 VRM, ARM, NS, Guj-4, GM-3, GM-4, K-851, GM-6, SKNM-		

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			1516, SKNM-1605, SKNM-1608, SKNM-1703, SKNM-1705, SKNM-1801, SKNM-1802, SKNM-1803, SKNM-1806, SKNM- 1808, SKNM-1809, SKNM-1810, SKNM-1812		
17	Seed: Size (weight of 100 seeds)	Small (<3 g) Medium (3-5 g)	SKNM-1704 ARM, NS, Guj-4, K-851, SKNM-1605, SKNM-1608, SKNM- 1701, SKNM-1703, SKNM-1705, SKNM-1802, SKNM-1803, SKNM-1809, SKNM-1810		
		Large (>5 g)	VM, VRM, NRM, GM-3, GM-4, GMM-5, GM-6, SKNM-1516, SKNM-1801, SKNM-1806, SKNM-1808, SKNM-1812		

Fig 1 Seed of Different Mungbean Genotypes



Fig 2 Pods of Different Mungbean Genotypes



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Principal component analysis

Principal component analysis (PCA) was performed to discriminate the genotypes based on morphological observations (Fig 3). The first components PC1, PC2, PC3 and PC4 accounted 26.09%, 13.34%, 12.20% and 11.55% of total variance, respectively with cumulative variance of about 63.18%. Significant variation was observed among different genotypes because the genotypes were found more scattered rather than clustered in the scatter plot. The genotype SKNM-1812, SKNM-1516, SKNM-1801, SKNM-1803, and GM-4 were found be more closely similar. However, significant similarity observed between ARM, VM and SKNM-1705, between SKNM-1605 and NS, and between SKNM-1809, SKNM-1802, and GMM-5. The PCA analysis discriminated genotype NRM, GM-3, GM-6, SKNM-1608, SKNM-1701, SKNM-1703, and SKNM-1704 as most diverse among given samples based on morphological characteristics.

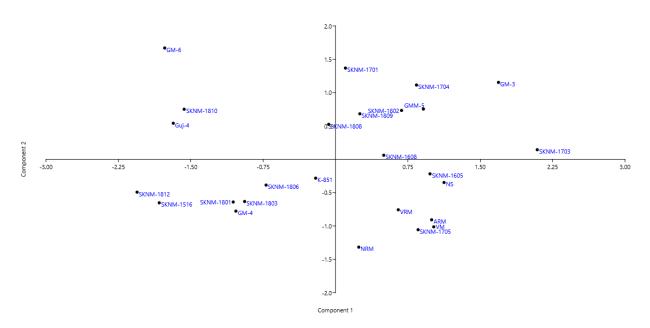


Fig 3 Scatter plot (Principal components analysis) Cluster analysis

A dendrogram was constructed with cophenetic correlation coefficient of 0.72 based on UPGMA (Unweighted Pair-Group Method with Arithmetic mean) using Jaccard's similarity coefficient by PAST (version 4.03) software as presented in Fig 4. The results obtained were in consistent with principal component analysis. The genotypes grouped into two major clusters A and B at similarity distance of 45%. The cluster A comprised of total 10 genotype, and was further divided into two clusters A1 and A2 at similarity index of 50%. The A2 cluster contains only two genotypes VM and NRM, while A1 divided into two more clusters A1.1 and A1.2 at genetic distance of 59%. The cluster A1.1 comprised of only two genotypes named Guj-4 and GM-6 at genetic distance of 91%. However, the cluster A1.2 also further divided into two more clusters, one cluster comprised of only single genotype named SKNM-1801, while the other cluster also further made of two groups, the first one contains two genotype named SKNM-1810 and SKNM-1812, while second group contains three genotypes named GM-4, SKNM-1516 and SKNM-1803. The Cluster B was largest cluster further divided into two more cluster B1, and B2 at genetic distance of 51%. B2 cluster was again divided into two cluster B2.1 and B2.2. B2.1 comprised of two genotypes GMM-5 and SKNM-1704, while cluster B2.2 was comprised of single genotype named SKNM-1701. The cluster B1 further divided into two major clusters B1.1 and B1.2 at genetic distance of 61%. The cluster B1.1 further divided into two clusters B1.1a and B1.1b at genetic distance 64%, among which cluster B1.1b contains two genotypes named SKNM-1806 and SKNM-1808 at genetic distance of 75%, while cluster B1.1a again divided into two more cluster. The one cluster contains two genotypes named ARM and SKNM-1605 at similarity distance index of 83%, while another cluster was made from three genotypes named SKNM-1809, VRM and NS. The cluster B1.2 also further divided into two more clusters B1.2a and B1.2b at genetic distance of 62%. The cluster B1.2a again grouped into two more clusters at genetic distance of 65%, among which one cluster contain two genotypes GM-3 and SKNM-1703, while another cluster contains three genotypes K-851, SKNM-1608 and SKNM-1705. However, only one genotype SKNM-1802 falls in cluster B1.2b



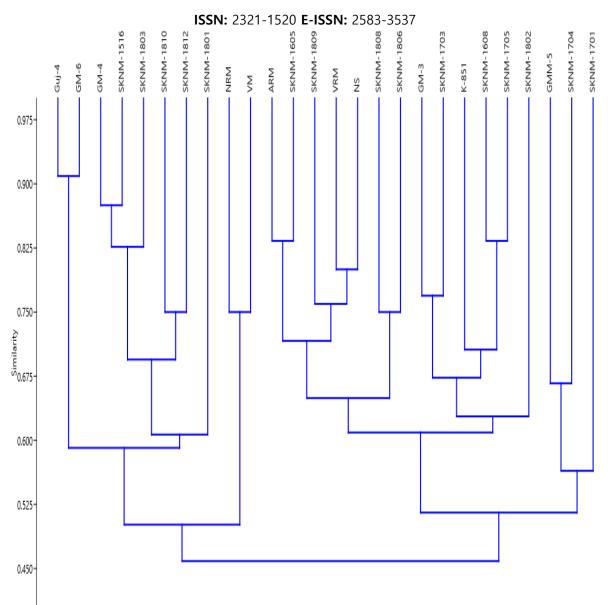


Fig 4 Dendrogram constructed using UPGMA analysis based on morphological observations

CONCLUSION

The present investigation focused on the diversity analysis based on morphological observations in 26 different genotypes of mungbean (*Vigna radiata* (L.) R. Wilczek). The significant variations were observed among different genotypes for each morphological trait. The most diverse genotypes revealed to be are VM, GM-6, SKNM-1608, SKNM-1701, SKNM-1704, SKNM-1801, SKNM-1802, SKNM-1806, and SKNM-1808 and all of them were found with promising morphological traits. These genotypes can be recommended to use as parent for further plant breeding programmes to develop new variety and conduct field trials for different locations and climates. These genotypes might be good choice for genetic stock conservations.

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AUTHOR CONTRIBUTION

Both authors equally contributed to the paper.

CONFLICT OF INTEREST

The authors declare no conflict of interest.



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