***Original Research Article***

**Phenotypic Variation and Genetic Divergence Among Tomato Genotypes: Implications for Parent Selection and Yield Enhancement**

# Abstract

This study investigated morphological diversity and genetic divergence among 25 tomato (*Solanum lycopersicum* L.) genotypes using 17 DUS-based descriptors and Mahalanobis D² statistics. The Shannon–Weaver diversity index (H′) revealed substantial phenotypic variability, especially in fruit traits. Vegetative traits, including growth habit and stem pubescence, were uniform (H′ = 0.00), reflecting strong selection for determinate, sturdy types suited to commercial production. In contrast, traits such as fruit shape (H′ = 1.61), locule number (H′ = 1.03), and total soluble solids (H′ = 1.00) showed considerable variation, while parameters like flowering time and leaf green intensity displayed moderate diversity.

Cluster analysis grouped the genotypes into five clusters with notable inter- and intra-cluster genetic divergence. Cluster IV exhibited the greatest intra-cluster distance (3.167), indicating higher internal genetic variability, while the largest inter-cluster distance was between Cluster II and Cluster V (5.519), suggesting these diverse groups could serve as promising parental combinations for heterosis. Cluster mean performance revealed substantial differences in yield-contributing traits. Cluster V was superior for most yield and quality attributes, including number of fruits per plant, fruit diameter, ascorbic acid content, and overall productivity. Cluster III excelled in average fruit weight and lycopene content, making it ideal for nutritional enhancement, while Cluster I had the shortest crop cycle.

These results highlight substantial genetic divergence and phenotypic variation, which can facilitate the selection of diverse parental lines for targeted hybridization and future tomato improvement programs.

***Keywords:*** Genetic diversity, Mahalanobis D² statistic, Morphological traits, Cluster analysis, Tomato breeding.

### INTRODUCTION

Tomato (*Solanum lycopersicum* L.), a member of the Solanaceae family, is widely grown and consumed around the world, earning its reputation as a vital component of the human diet and often labeled as a "protective food." It is one of the most significant vegetables used in processing industries. India currently holds the fourth position globally in terms of tomato cultivation area (Kumar *et al.,* 2023). South Mexico is the center of origin of tomato (Campos *et al.,* 2021).

Tomato landraces are known for their extensive morphological diversity, often displaying distinct characteristics even among closely related morphotypes. This variation plays a vital role in both taxonomic differentiation and the enhancement of their agricultural and commercial value. According to (Pereira-Dias *et al.,* 2020), the descriptors used for tomato exhibit a broad range of morphological traits, aiding in the classification of varietal groups and the assessment of phenotypic variability. Morphological characterization emphasizes traits that are highly heritable, visually discernible, and stable across diverse environments, making them reliable for evaluation (Grozeva *et al.,* 2020). As highlighted by (Corrado *et al.,* 2014), the detailed assessment of parental lines and hybrids can facilitate the identification of promising genotypes, which are essential for the genetic improvement and development of superior tomato cultivars.

A systematic and standardized approach to morphological characterization is essential for assessing variability among plant varieties. In India, the Distinctness, Uniformity, and Stability (DUS) testing system, implemented by the Protection of Plant Varieties and Farmers' Rights Authority (PPV&FRA), serves as a vital tool for varietal identification, registration, and protection (PPV&FRA, 2009). DUS descriptors facilitate the assessment of key morphological traits that are stable and heritable, thereby supporting varietal purity assessment and plant breeding efforts.

Although several studies have employed DUS-based morphological characterization for varietal evaluation, most have focused on a limited set of genotypes or traits, leaving a gap in comprehensive assessment across diverse germplasm collections. Moreover, the integration of such characterization with agronomic trait evaluation remains underexplored in many regional breeding programs. Thus, there is a pressing need to document morphological diversity in a structured manner to identify genotypes with high yield potential and desirable fruit quality traits for commercial cultivation.

Assessing genetic divergence among germplasm collections is also essential for identifying promising parental lines for future breeding initiatives. Information on the genetic diversity of important traits, particularly those that influence yield and quality can greatly enhance the efficiency of crop improvement programs. Mahalanobis D² statistic (1936) is a well-established multivariate tool for quantifying the genetic distance between genotypes. Grouping genotypes based on D² values enables breeders to select appropriate parents for heterosis breeding and targeted hybridization. This strategy also supports the recovery of superior transgressive segregants, which can ultimately contribute to the development of high-yielding, open-pollinated cultivars for commercial use. In view of these considerations, the present study was undertaken to evaluate the genetic diversity of the available germplasm based on fifteen qualitative and quantitative traits.

### MATERIALS AND METHODS

The experiment was conducted during the *Rabi* season of 2023–24 at the Main Experimental Research Station, College of Horticulture, Acharya Narendra Deva University of Agriculture and Technology, Kumarganj, Ayodhya, U.P. (24.56°N latitude, 81.84°E longitude; 113 m AMSL). The site experiences a humid subtropical climate with an average annual rainfall of approximately 1200 mm, and temperature fluctuations ranging from 5.3°C to 40.9°C during the cropping period.

A total of 25 tomato genotypes, including released varieties, advanced breeding lines, local landraces and 1 check variety (Kashi Aman) were evaluated in a randomized block design (RBD) with three replications. Each plot consisted of two rows with a spacing of 60 cm between rows and 50 cm between plants, and standard cultural practices were uniformly followed.

Observations were recorded on 17 morphological traits related to leaves, flowers, and fruits, following the DUS guidelines of (PPV&FRA, 2009), using visual (VG, VS) and measurement-based (MG, MS) assessment methods on five randomly selected plants per replication. Color attributes were assessed using the RHS Color Chart (2001). All qualitative traits were numerically coded for statistical analysis.

**Statistical Analysis**  
Morphological diversity was estimated using the Shannon–Weaver diversity index (H′) in RStudio. Genetic divergence was determined by Mahalanobis D² statistics, and UPGMA cluster analysis was also conducted in RStudio to group the genotypes into distinct clusters.

**RESULTS AND DISCUSSION**

**Morphological characterization**

A comprehensive evaluation of 25 tomato genotypes based on 17 morphological descriptors, following the DUS guidelines, revealed significant phenotypic diversity (Table 1). The observed variability was supported by the number of trait variants, Shannon–Weaver diversity index (H′), and frequency distribution, collectively reflecting broad genetic richness across the genotypes. These findings were further substantiated through visual representation (Figure 1). The Shannon–Weaver diversity index (H′) is a well-established metric for assessing phenotypic variation, based on the assumption of random sampling from independent populations (Shannon & Weaver: 1949; Clarke & Warwick: 2001).

Vegetative traits such as growth habit and stem pubescence were uniform across all genotypes (H′ = 0.00), indicating directional selection for determinate types with sturdy stems. This growth habit is preferred in commercial production for its uniform flowering and fruiting, allowing synchronized harvests and efficient crop management. Likewise, uniform stem pubescence may help protect the plant surface from insect damage, harsh weather, and excessive transpiration, especially in open-field conditions (Peralta & Spooner, 2000). Moderate diversity was observed in leaf green intensity (H′ = 0.70), with a majority of genotypes exhibiting medium green coloration. Leaf structure (H′ = 0.48) and leaflet serration (H′ = 0.44) also showed limited diversity. Flowering time demonstrated moderate variation (H′ = 0.50), with most genotypes categorized as medium and a minority as early flowering. Early flowering is responsible for early fruiting and may be beneficial for breeders and farmers (Rashid *et al.,* 2016). The peduncle abscission layer was present in 92% of genotypes (H′ = 0.24), a trait favored for ease of harvest.

Fruit traits exhibited greater morphological diversity. Fruit shape in longitudinal section showed the highest variation (H′ = 1.61), with genotypes displaying circular (28%), slightly flattened (24%), obovoid (16%), and other forms, making it a key differentiator. High diversity was also evident in locule number (H′ = 1.03) and TSS content (H′ = 1.00), both critical for market and processing traits. Fruit size, width, and length presented moderate diversity (H′ = 0.62, 0.62 and 0.42 respectively), with the majority in medium to large categories. Peduncle-end depression (H′ = 0.88), shape at blossom end (H′ = 0.77), and fruit color at maturity (H′ = 0.62) added further phenotypic distinction. The dominance of red (68%) and orange (32%) fruit color indicates the presence of high level of lycopene and beta-carotene (Nasir *et al.,* 2015). Notably, 48% of genotypes recorded high TSS values, further reinforcing their potential in processing and fresh market applications.

**Table 1. Scoring system for morphological characterization of tomato genotypes**

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **S.**  **No.** | **Characteristics** | **Code** | **Type of assessment** | **Shannon Index (H')** | **States** | **Notes** | **No. of variants** | **Frequency %** |
| **1.** | Leaf: Intensity of green color | LIGC | VG | 0.70 | Light  Medium  Dark | 3  5  7 | 1  14  10 | 4%  56%  40% |
| **2.** | Plant: Growth type | GH | VG | 0.00 | Determinate  Semi determinate | 1  2 | 25  - | 100%  - |
| **3.** | Stem: Pubescence | SP | VS | 0.00 | Absent  Present | 1  9 | 25  - | 100%  - |
| **4.** | Leaflet: Serration | LLS | VS | 0.44 | Absent (potato type)  Less serrated  Highly serrated | 1  3  7 | 1  21  3 | 4%  84%  12% |
| **5.** | Leaf: Structure | LS | VG | 0.48 | Open  Intermediate  Closed | 3  5  7 | 6  19  - | 24%  76%  - |
| **6.** | Peduncle: Abscission layer | PA | VS | 0.24 | Absent (joint less)  Present (jointed) | 1  9 | 2  23 | 8%  92% |
| **7.** | Time of flowering (50% of the plants with at least one open flower from seed sowing) (days) | TOF | VG | 0.50 | Early (<65)  Medium (65-80)  Late (>80) | 3  5  7 | 5  20  - | 20%  80%  - |
| **8.** | Fruit: Size (average weight of 10 fruits) (g) | FS | MG | 0.62 | Very small (<100)  Small (100-200)  Medium (201-700)  Large (701-1000)  Very large (>1000) | 1  3  5  7  9 | -  -  17  8  - | -  -  68%  32%  - |
| **9.** | Fruit: Length (cm) | FL | MS | 0.42 | Very short (< 3.0)  Small (3.0-5.0)  Medium (5.1-7.0)  Large (7.1-9.0)  Very large (> 9.0) | 1  3  5  7  9 | -  20  5  -  - | -  80%  20%  -  - |
| **10.** | Fruit: Width (cm) | FW | MS | 0.62 | Very short (< 3.0)  Small (3.0-5.0)  Medium (5.1-7.0)  Large (7.1-9.0)  Very large (> 9.0) | 1  3  5  7  9 | -  17  8  -  - | -  68%  32%  -  - |
| **11.** | Fruit: Shape in longitudinal section | FSLS | VS | 1.61 | Flattened  Slightly flattened  Circular  Rectangular  Cylindrical  Heart shaped  Obovoid  Ovoid  Pear shaped | 1  2  3  4  5  6  7  8  9 | 4  6  7  -  -  1  4  2  1 | 16%  24%  28%  -  -  4%  16%  8%  4% |
| **12.** | Fruit: Depression at peduncle end | FDPE | VS | 0.88 | Absent  Shallow  Medium  Deep | 1  3  5  7 | 3  15  7  - | 12%  60%  28%  - |
| **13.** | Fruit: Shape at blossom end | FSBE | VS | 0.77 | Indented  Indented to flat  Flat  Flat to pointed  Pointed  Circular | 1  2  3  4  5  6 | -  2  21  1  1  - | -  8%  84%  4%  4%  - |
| **14.** | Fruit: Thickness of the pericarp (cm) | FTP | MG | 0.42 | Thin (<0.3)  Medium (0.3 to 0.6)  Thick (>0.6) | 3  5  7 | 5  20  - | 20%  80%  - |
| **15.** | Fruit: Number of locules | FNOL | VS | 1.03 | 2  3-4  >4 | 1  2  3 | 6  7  12 | 24%  28%  48% |
| **16.** | Fruit: Color at maturity | FCM | VG | 0.62 | Yellow  Orange  Pink  Red | 1  2  3  4 | -  8  -  17 | -  32%  -  68% |
| **17.** | Fruit: Total soluble Solids (O Brix) | FTSS | MG | 1.00 | Low (<3)  Medium (3.1 -4)  High (4.1-5.0)  Very high (>5) | 3  5  7  9 | -  4  9  12 | -  16%  36%  48% |

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| **Figure 1.** Morphological Profiling of Tomato genotypes: (a) Variation in number of locules, (b). Differences in leaf margin serration,(c). Measurement of fruit length, width, and pericarp thickness, (d). Presence of stem pubescence, (e). Diversity in fruit shape, (f). Diversity in fruit shape at the blossom end. | |

**Composition of Clusters**

The grouping of the twenty-five tomato genotypes based on their performance for multiple traits is presented in Table 2and also validated through UPGMA cluster dendogram in Figure 2. Five distinct clusters (I–V) were formed. Cluster IV contained the greatest number of genotypes (7), followed by Cluster II (6), Cluster V (5), Cluster III (4), and Cluster I (3). The genotypes assembled within each cluster shared considerable genetic similarity, resulting in their close association. Comparable findings regarding the clustering of tomato genotypes based on genetic divergence have been documented previously by (Reddy *et al.*, 2013).

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| **Figure 2.** Cluster dendrogram of 25 tomato genotypes based on Mahalanobis D² distances using UPGMA method |

**Table 2.** Cluster-wise distribution of 25 tomato genotypes

|  |  |  |
| --- | --- | --- |
| **Clusters** | **No of genotypes** | **Genotypes** |
| I | 3 | NDT-22-3, NDT-22-14 and NDT-22-25 |
| II | 6 | NDT-22-16, NDT-22-17, NDT-22-21, NDT-22-22, NDT-22-223 and NDT-22-24 |
| III | 4 | NDT-22-7, NDT-22-26, NDT-22-27 and NDT-22-29 |
| IV | 7 | NDT-22-1, NDT-22-2, NDT-22-4, NDT-22-15, NDT-22-20, NDT-22-28 and Kashi Aman |
| V | 5 | NDT-22-5, NDT-22-9, NDT-22-10, NDT-22-12 and NDT-22-13 |

**Intra and Inter-Cluster genetic divergence (D²)**

Genetic divergence among 25 tomato genotypes was assessed using Mahalanobis D² statistic, and five distinct clusters were formed presented in Table 3. The intra-cluster distance was highest in Cluster IV (3.167), followed by Cluster V (2.927), Cluster III (2.706), Cluster II (2.686), and lowest in Cluster I (2.281), indicating greater genetic diversity within Cluster IV and higher genetic similarity within Cluster I.

Among the inter-cluster distances, the divergence between Cluster II and Cluster V was the greatest (5.519), followed by the divergence between Cluster II and Cluster III (5.113), and between Cluster I and Cluster III (4.626), while the least divergence was observed between Cluster III and Cluster IV (3.562). These high inter-cluster distances suggest that crosses between these genetically distant clusters could produce superior heterotic combinations. Similar findings were also reported by (Yadav *et al.,* 2020), and (Verma *et al.,* 2023).

**Table 3.** Intra and inter-cluster distances (D² values) among five clusters

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| **Clusters** | **I** | **II** | **III** | **IV** | **V** |
| I | 2.281 |  |  |  |  |
| II | 3.685 | 2.686 |  |  |  |
| III | 4.626 | 5.113 | 2.706 |  |  |
| IV | 4.344 | 4.179 | 3.562 | 3.167 |  |
| V | 3.620 | 5.519 | 4.200 | 3.752 | 2.927 |

**Cluster mean analysis**

The mean performance of each cluster for different traits is presented in Table 4. **Cluster V** had the highest mean for primary branches (5.59), polar fruit diameter (5.73 cm), number of fruits per plant (24.22), ascorbic acid (18.50 mg/100 g), and fruit yield per plant (1.68 kg), yield per plot (9.87 kg), and yield per hectare (282.87 q/ha), but the lowest for equatorial fruit diameter (4.16 cm), pericarp thickness (3.18 mm), number of locules (2.93), and lycopene content (3.83 mg/100 g). **Cluster IV** showed the highest mean for number of locules (4.71), TSS (5.29 ºBrix), days to 50% flowering (44.05), and days to first fruit harvest (81.95), while ascorbic acid was lowest (16.94 mg/100 g). **Cluster III** excelled for equatorial fruit diameter (5.15 cm), pericarp thickness (4.59 mm), average fruit weight (77.82 g), and lycopene (5.69 mg/100 g), but had the lowest mean value for plant height (61.99 cm) and primary branches (3.87). **Cluster II** showed the lowest mean for polar fruit diameter (3.91 cm), average fruit weight (42.68 g), fruits per plant (18.45), and fruit yield per plant (0.81 kg), yield per plot (5.31 kg), and yield per hectare (148.66 q/ha). **Cluster I** had the shortest crop cycle with the least days to 50% flowering (36.00) and days to first fruit harvest (72.67), lowest TSS (3.90 ºBrix), and the tallest plants (79.52 cm). These patterns indicate substantial genetic divergence and underline the potential for selecting diverse parents for hybridization. Similar findings have been reported by (Pushpam *et al.,* 2017), (Narayan *et al.*, 2018), and (Naveen *et al.,* 2018).

**Table 4.** Mahalanobis D² Cluster Mean Analysis

|  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **Clusters** |  | **Days to 50% flowering** | **Days to first fruit harvest** | **Plant height (cm)** | **No. of primary branches/plant** | **Polar fruit diameter (cm)** | **Equatorial fruit diameter (cm)** | **Pericarp thickness (mm)** | **No. of locules/fruit** | **Average fruit weight (g)** | **No. of fruits/plant** | **TSS content (Brix)** | **Ascorbic acid (100mg/fruit)** | **Lycopene content (mg/100gm)** | **Fruit yield/plant (Kg)** | **Fruit yield/plot (kg)** | **Fruit yield (q/ha)** |
| **I** | **Mean** | 36.00 | 72.67 | 79.52 | 4.37 | 4.94 | 4.19 | 3.81 | 3.00 | 47.47 | 22.34 | 3.90 | 17.11 | 5.27 | 1.06 | 7.83 | 213.16 |
|  | **SE±** | 3.18 | 4.63 | 6.92 | 0.97 | 0.65 | 0.34 | 0.32 | 1.73 | 2.04 | 3.26 | 0.43 | 0.77 | 0.46 | 0.18 | 0.86 | 31.31 |
| **II** | **Mean** | 42.06 | 79.83 | 71.19 | 4.37 | 3.91 | 4.25 | 3.53 | 3.39 | 42.68 | 18.45 | 5.02 | 18.41 | 4.73 | 0.81 | 5.31 | 148.66 |
|  | **SE±** | 3.18 | 3.02 | 4.92 | 0.54 | 0.55 | 0.94 | 0.71 | 0.98 | 6.02 | 2.35 | 0.42 | 3.35 | 1.29 | 0.08 | 1.22 | 36.57 |
| **III** | **Mean** | 40.67 | 78.75 | 61.99 | 3.87 | 4.93 | 5.15 | 4.59 | 3.83 | 77.82 | 20.72 | 5.24 | 17.45 | 5.69 | 1.63 | 9.80 | 275.95 |
|  | **SE±** | 5.99 | 6.39 | 7.67 | 0.13 | 0.25 | 0.62 | 0.77 | 0.58 | 2.30 | 1.98 | 0.58 | 4.57 | 0.78 | 0.14 | 0.70 | 12.01 |
| **IV** | **Mean** | 44.05 | 81.95 | 77.67 | 6.30 | 4.41 | 5.07 | 3.74 | 4.71 | 65.95 | 21.61 | 5.29 | 16.94 | 4.83 | 1.42 | 8.60 | 248.90 |
|  | **SE±** | 2.01 | 1.75 | 8.29 | 0.78 | 0.54 | 1.04 | 0.91 | 1.01 | 8.92 | 3.02 | 0.74 | 3.00 | 1.10 | 0.31 | 1.78 | 50.68 |
| **V** | **Mean** | 37.53 | 74.13 | 77.65 | 5.59 | 5.73 | 4.16 | 3.18 | 2.93 | 69.50 | 24.22 | 4.71 | 18.50 | 3.83 | 1.68 | 9.87 | 282.87 |
|  | **SE±** | 5.38 | 5.07 | 3.45 | 1.07 | 1.03 | 0.59 | 0.56 | 1.28 | 8.16 | 1.97 | 0.87 | 3.32 | 0.76 | 0.19 | 0.62 | 5.43 |

### ****Conclusion****

This study underscores the significance of morphological characterization and genetic divergence analysis in identifying diverse and agronomically superior tomato genotypes. Morphological traits effectively distinguished genotypes based on key yield and quality attributes, while multivariate clustering revealed substantial genetic variability across five distinct groups. The integration of these approaches provides a valuable framework for parent selection and hybridization. Exploiting this diversity offers strong potential for developing high yielding, early maturing, and quality rich tomato cultivars in future breeding programs.

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