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Multivariate Clustering and Genetic Divergence Assessment of Momordica Charantia L. Germplasm in Northern Kashmir

Dr. Siti Nur Aisyah
Indonesian Institute of Horticultural Sciences.Bogor, Indonesia

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Abstract

Genetic diversity is a fundamental consideration in bitter gourd improvement because the identification of genetically divergent germplasm can support parent selection, trait-based breeding, and the development of improved cultivars. Northern Kashmir represents a distinctive agroecological setting in which the evaluation of bitter gourd genotypes may help identify promising materials for local cultivation and future breeding programmes. This paper examines a multivariate framework for assessing genetic divergence and clustering among Momordica charantia L. genotypes, with particular emphasis on the application of Mahalanobis' generalized distance, D^2 statistics, and cluster-based interpretation. The methodological foundation draws on classical multivariate statistical analysis and established agricultural approaches to quantitative variation (Mahalanobis, 1936; Panse & Shukhatme, 1967; Rao, 1952). The supplied literature on bitter gourd provides context for evaluating yield performance, genetic variability, heritability, and divergence among genotypes (Rani, 2014; Resmi & Sreelathakumary, 2012; Talukder et al., 2018).

Because no genotype-level observations, experimental design, cluster assignments, or calculated D^2 values were supplied, this paper develops a research-oriented analytical framework rather than presenting fabricated empirical results. The findings section therefore distinguishes established methodological implications from results that require field data. The proposed approach emphasizes standardized quantitative characterization, multivariate distance estimation, cluster formation, within- and between-cluster comparisons, and the identification of traits contributing to genetic divergence. The framework can support more systematic selection of parental material while recognizing that statistical divergence alone does not guarantee superior hybrid performance or agronomic adaptation.

Keywords: Bitter gourd; Momordica charantia L.; genetic diversity; multivariate clustering; Mahalanobis D^2 statistics; germplasm characterization; genetic divergence; quantitative traits; plant breeding.

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1. Introduction

1.1 Background and research problem

Bitter gourd (Momordica charantia L.) is an important vegetable crop whose improvement depends on the availability of diverse genetic material and the effective identification of desirable agronomic characteristics. Differences among genotypes may occur in growth, flowering, fruit morphology, maturity, yield, and related quantitative traits. Systematic characterization of these differences is essential because visual assessment or comparison of individual traits may not adequately represent the overall genetic divergence among candidate breeding materials.

Genetic diversity analysis provides a structured basis for comparing genotypes across several characteristics simultaneously. In agricultural research, multivariate methods are particularly useful when traits are correlated or when individual measurements provide incomplete evidence of overall differentiation. Mahalanobis' generalized distance established a statistical foundation for measuring multivariate separation while accounting for relationships among measured variables (Mahalanobis, 1936). Its application to crop germplasm assessment enables researchers to compare genotypes through a combined representation of their quantitative characteristics.

The central research problem addressed here is how a multivariate clustering framework can be used to characterize the divergence of bitter gourd germplasm under the agroecological conditions of Northern Kashmir. The question has practical importance because locally relevant germplasm may contain useful combinations of yield-related and adaptive traits that are not readily identified through single-trait selection.

1.2 Research relevance and rationale

Previous studies supplied for this paper address bitter gourd performance, genetic divergence, and quantitative variability. Rani (2014) and Saho (2015) focus on genotype performance for yield and earliness, while Sagar et al. (2022) examine mean performance across growth and yield characteristics. Resmi and Sreelathakumary (2012), Singh and Kandasamy (2020), and Tyagi et al. (2018) address genetic divergence or diversity in bitter gourd germplasm. Research on genetic variability, heritability, and genetic advance provides an additional perspective on the usefulness of quantitative traits for selection (Talukder et al., 2018; Triveni et al., 2021).

These research directions collectively support the relevance of combining performance evaluation with multivariate diversity analysis. However, differences in geographical setting, genotype composition, environmental conditions, and trait measurement can limit direct comparisons between studies. A dedicated analysis of Northern Kashmir should therefore be based on locally collected and consistently measured observations rather than assuming that diversity patterns reported elsewhere will apply unchanged.

1.3 Objectives and scope

The principal objective is to develop a statistically grounded approach for assessing genetic divergence and clustering among bitter gourd genotypes relevant to Northern Kashmir. Specifically, the paper aims to explain the role of multivariate characterization in germplasm evaluation, establish a reproducible analytical procedure based on D^2 statistics, describe how clusters and inter-cluster distances should be interpreted, and identify the implications for parental selection and crop improvement.

The scope is methodological and interpretive. The supplied references establish the conceptual basis, but no original field dataset was provided. Accordingly, the paper does not claim to have measured actual genetic distances, identified particular superior genotypes, or established the number and composition of clusters in Northern Kashmir.

2. Literature Review

2.1 Statistical foundations of genetic divergence

Multivariate analysis provides a means of examining the collective differences among biological units characterized by several quantitative variables. In crop improvement, this is important because genotypes rarely differ in only one characteristic. A genotype with relatively high yield may also differ in maturity, fruit dimensions, growth habit, or other traits relevant to production. Evaluating each trait separately can therefore produce an incomplete assessment of overall differentiation.

Mahalanobis (1936) introduced a generalized statistical distance for multivariate observations. Unlike ordinary Euclidean distance, this approach accounts for the covariance structure of the measured variables. When two traits are strongly correlated, their shared information should not be counted as though it represented two entirely independent sources of differentiation. Mahalanobis distance addresses this issue by incorporating the variability and interrelationships of the observations.

Rao (1952) provides a broader foundation for advanced statistical methods in biometrical research, while Panse and Shukhatme (1967) establish the agricultural statistical context in which experimental observations, variability, and comparisons among crop materials can be evaluated. These works support a rigorous approach to germplasm assessment, although the reliability of any particular analysis depends on sampling, experimental design, measurement quality, and the suitability of the

statistical assumptions.

2.2 Research on bitter gourd performance and diversity

The supplied literature covers several complementary dimensions of bitter gourd improvement. Rani (2014) and Saho (2015) examine genotype performance in relation to yield and earliness. These characteristics are relevant to selection because production potential depends not only on final yield but also on the time required to reach harvestable maturity. Nevertheless, performance comparisons alone do not establish the genetic distance between genotypes. Two high-performing genotypes may be relatively similar across most measured traits, while a lower-performing genotype may possess useful characteristics absent from the leading materials.

Sagar et al. (2022) extend this performance-oriented perspective by considering mean values for growth and yield characters. Such evaluations can identify traits and genotypes deserving further investigation. Their usefulness for breeding decisions increases when mean performance is interpreted alongside multivariate divergence, since breeding programmes must consider both the desirability of traits and the distinctiveness of potential parents.

Resmi and Sreelathakumary (2012) specifically investigate genetic divergence in bitter gourd. Singh and Kandasamy (2020) address genetic diversity under coastal ecosystems, while Tyagi et al. (2018) also examine genetic divergence in bitter gourd. Collectively, these studies establish genetic differentiation as a relevant topic in bitter gourd research. However, findings from other ecological settings should not be treated as direct evidence of the clustering pattern expected in Northern Kashmir.

Talukder et al. (2018) examine genetic variability, heritability, and genetic advance in relation to yield and yield-contributing traits. Triveni et al. (2021) address similar quantitative-genetic considerations. These perspectives are complementary to multivariate clustering: variability describes the extent of observed differences, heritability concerns the proportion of variation attributable to genetic differences under a particular analytical framework, and genetic advance concerns the expected response to selection under stated assumptions. None of these measures is interchangeable with multivariate distance.

2.3 Research gap and theoretical positioning

The principal gap relevant to the present title is the absence, in the supplied material, of a documented genotype-level dataset specifically representing Northern Kashmir. The references provide statistical foundations and examples of bitter gourd performance and divergence research, but they do not supply the observations required to calculate local cluster membership or D^2 distances.

The proposed study is therefore positioned at the intersection of quantitative characterization, multivariate statistical analysis, and applied crop improvement. It treats clustering as a tool for organizing observed phenotypic differentiation rather than as automatic proof of molecular or evolutionary relationships. This distinction is important because phenotypic differences can reflect genetic effects, environmental variation, genotype-by-environment interaction, or measurement error.

The contribution of a Northern Kashmir assessment would be strongest if it integrated standardized trait measurement, appropriate multivariate distance estimation, and agronomic interpretation. Such integration would connect the statistical identification of divergent groups with the practical question of whether their members possess complementary traits that justify their inclusion in a breeding programme.

3. Methodology

3.1 Research design and study material

A suitable investigation would employ a comparative field evaluation of bitter gourd genotypes grown under documented Northern Kashmir conditions. The germplasm collection should include clearly identified accessions or cultivars representing the available diversity. Each genotype should have a unique identifier, and seed source, collection history, and relevant passport information should be recorded wherever available.

A randomized experimental design with adequate replication would help separate genotype-associated differences from within-field variability. The specific design, number of genotypes, replication level, plot dimensions, and management practices must be determined according to the available germplasm, field resources, and experimental objectives. These details cannot be specified as completed procedures without the original study records.

Where the research aims to identify genotypes adapted to

the Northern Kashmir environment, the location, elevation, planting date, soil characteristics, weather conditions, irrigation, and crop management should be documented. If the aim extends to adaptation across environments, observations from multiple locations or seasons would be needed. A single-location evaluation can describe performance under its test conditions but cannot independently establish broad environmental stability.

3.2 Selection and measurement of quantitative traits

Trait selection should be guided by breeding objectives and measurement reliability. Candidate variables may include vine growth, branching, days to first flowering, days to first harvest, fruit length, fruit diameter, fruit weight, fruit number per plant, and total yield per plant or unit area. These are proposed measurement categories, not traits confirmed to have been collected in the present case.

Each trait requires an explicit operational definition. For example, days to first flowering should be calculated from a defined planting or emergence reference point, and fruit weight should be measured using a consistent weighing procedure. Yield measurements must be expressed on a comparable basis across plots. Ambiguous definitions can introduce artificial differences and weaken the validity of subsequent clustering.

Measurements should be collected from comparable developmental stages and according to a consistent sampling protocol. Missing observations, extreme values, transcription errors, and inconsistent units should be examined before analysis. A genuine extreme genotype should not be removed merely because its measurements are unusual; exclusion requires a defensible reason, such as a documented measurement error.

3.3 Data preparation and exploratory analysis

The initial dataset should be organized so that each row represents a genotype or experimental unit, as appropriate, and each column represents a measured trait. Replicated plot-level observations should be retained for statistical assessment before genotype means are prepared. If environmental or block effects are present, they should be considered when estimating genotype-level values.

Descriptive statistics should summarize trait means, standard deviations, ranges, and distributions. Correlation analysis can reveal strong relationships among variables and identify possible redundancy. For instance, fruit length and fruit weight may be associated, while days to flowering and days to first harvest may represent overlapping aspects of crop development. Such relationships should be investigated rather than assumed.

Variables measured in different units require careful treatment. Mahalanobis distance accounts for the covariance structure, but it does not remove the need for sound data preparation. Poorly estimated covariance matrices, excessive redundant variables, missing observations, or a sample size that is inadequate relative to the number of traits can make the distance estimates unstable.

3.4 Mahalanobis D^2 distance estimation

The theoretical foundation of the proposed analysis is Mahalanobis' generalized distance (Mahalanobis, 1936). For a pair of genotype profiles, the distance reflects their multivariate separation while accounting for the covariance among the selected traits. In practice, researchers estimate the relevant covariance structure from the experimental observations and use it to calculate pairwise distances.

The distinction between univariate and multivariate comparison is central. A simple comparison may show that two genotypes differ in fruit length, but it cannot establish whether they are distinct when all selected traits are considered together. D^2 analysis provides a joint assessment of their separation across the measured variables.

The statistical interpretation depends on the data and estimation procedure. When genotype means are used, researchers must state how experimental error and replication were incorporated. When the number of traits is high relative to the number of independent observations, the covariance estimate may be singular or poorly conditioned. In such circumstances, the trait set, sample size, or estimation strategy must be reconsidered before clustering proceeds.

3.5 Cluster formation and validation

After estimating the pairwise distances, the researcher can group genotypes according to a clearly specified clustering procedure. The method and decision rules used to form clusters must be reported so that the analysis can be reproduced. The number of clusters should not be

selected merely because a particular grouping produces an appealing narrative.

The resulting groups can be examined through cluster membership, within-cluster dispersion, and between-cluster separation. A useful clustering solution should provide interpretable differentiation without creating unnecessarily small or unstable groups. Alternative clustering specifications and sensitivity checks can help determine whether the broad pattern is robust to reasonable analytical choices.

Mahalanobis distance and clustering should be treated as related but distinct stages. The distance measure quantifies separation between multivariate profiles; the clustering procedure organizes those distances into groups. Different clustering algorithms or decision rules may produce different partitions from the same distance matrix.

3.6 Trait contribution and breeding interpretation

Cluster means should be calculated for the traits included in the study, and between-cluster comparisons should identify characteristics that distinguish the groups. The objective is not simply to rank clusters by distance but to determine which traits make the groups agronomically meaningful.

If a divergent group combines desirable yield with suitable maturity, it may provide useful parental material. Alternatively, a highly divergent group may have limited immediate breeding value if its distinguishing characteristics are undesirable. Trait desirability must therefore be assessed separately from statistical divergence.

The Mahalanobis framework supports the joint evaluation of traits and their relationships, but a distance value alone does not identify causal genes or establish the inheritance of a particular characteristic. Selection decisions should combine divergence estimates with replicated performance, trait heritability evidence where available, and the objectives of the breeding programme (Panse & Shukhatme, 1967; Rao, 1952).

4. Results

4.1 Status of the empirical evidence

The primary finding of the present work is the development of a structured analytical framework for evaluating multivariate genetic divergence in bitter gourd germplasm from Northern Kashmir. The supplied references establish the relevance of statistical distance, agricultural biometrics, genotype performance, and genetic variability. However, the available material does not contain raw observations, genotype identifiers, calculated distances, or cluster assignments. Consequently, numerical findings about the local germplasm cannot be reported responsibly.

This limitation is important because a research paper should distinguish methodological conclusions from experimentally observed results. Reporting a specific number of clusters, identifying the most divergent genotypes, or claiming that a particular trait contributed most strongly to divergence would require data that are not available here.

4.2 Expected analytical outputs

The proposed multivariate assessment would generate several complementary outputs. The first would be a standardized genotype-by-trait dataset, accompanied by descriptive statistics that characterize the observed range of agronomic variation. These statistics would establish whether the evaluated germplasm exhibits meaningful differences in the selected characteristics and whether any variables require further scrutiny.

The second output would be a pairwise D^2 distance matrix. This matrix would quantify the multivariate separation between genotype profiles, subject to the adequacy of the covariance estimates. A larger distance would indicate greater statistical separation across the selected traits, but it would not, by itself, establish the genetic mechanism responsible for the difference. The conceptual basis follows Mahalanobis (1936), whose generalized distance accounts for relationships among multiple measurements.

The third output would be a cluster classification. Each genotype would be assigned to a group according to the selected clustering procedure. Cluster summaries would then reveal whether the groups differ in overall trait profiles and whether some clusters contain relatively homogeneous or heterogeneous material. These findings would need to be evaluated alongside the stability of the clustering solution.

4.3 Interpretation of possible divergence patterns

Several possible outcomes have different implications. If the analysis identifies clearly separated clusters with contrasting yield and maturity profiles, the results could help breeders identify parents with complementary

characteristics. If high-yielding genotypes occur in several clusters, selection could potentially draw on multiple distinct backgrounds rather than relying on a single performance group. Conversely, if the clusters overlap substantially in their agronomic characteristics, the apparent multivariate separation may have limited practical value for selection.

These are conditional interpretations, not observations from the present dataset. The distinction matters because the literature on bitter gourd performance and diversity establishes the relevance of examining such characteristics but does not determine the outcome for Northern Kashmir (Rani, 2014; Resmi & Sreelathakumary, 2012; Sagar et al., 2022).

A further expected output would be a comparison of cluster means for individual traits. Such comparisons could identify whether divergence is associated mainly with fruit characteristics, crop duration, yield components, or a broader combination of features. The actual contribution of individual traits must be established from the study data and the chosen statistical procedure.

4.4 Implications for selection

The framework supports a two-stage selection strategy. First, genotypes can be screened for desirable agronomic performance. Second, the multivariate distance analysis can identify candidates that are distinct from one another while remaining relevant to the breeding objective. This approach avoids treating statistical distance as a substitute for practical merit.

Research on genetic variability, heritability, and genetic advance provides a complementary basis for assessing the potential usefulness of observed trait differences (Talukder et al., 2018; Triveni et al., 2021). A genotype that appears distinctive may warrant further evaluation, but its value for improvement depends on trait desirability, the reliability of the observations, and the inheritance of the characteristics concerned.

4.5 Evidence boundaries

The findings that can be supported at this stage are methodological: D^2 statistics provide a principled basis for multivariate separation; clustering can organize the resulting distances into interpretable groups; and agronomic performance must be considered when translating those groups into breeding decisions. No empirical conclusion about the actual diversity level, cluster composition, superior genotype, or dominant trait in Northern Kashmir can be established without original observations.

A completed empirical version should report the number of evaluated genotypes, trait definitions, experimental design, estimated distances, cluster sizes, cluster means, between-cluster comparisons, and relevant uncertainty measures. It should also include tables showing the observed values rather than illustrative figures presented as genuine results.

5. Discussion

5.1 Integrating multivariate divergence with agronomic performance

The central methodological implication is that genotype selection should consider both performance and multivariate distinctiveness. Performance-oriented studies of bitter gourd address characteristics relevant to yield and earliness, while divergence studies address differences among germplasm materials (Rani, 2014; Saho, 2015; Resmi & Sreelathakumary, 2012). Combining these perspectives offers a more informative basis for selection than either approach alone.

A genotype with excellent yield but limited divergence from existing breeding material may still be valuable for direct cultivation or selection. Conversely, a highly divergent genotype may be useful as a parent if it contributes a desirable characteristic absent from the other parent. The breeding objective therefore determines how much importance should be assigned to distance relative to mean performance.

Mahalanobis' generalized distance is appropriate for this type of multivariate assessment because it incorporates covariance among traits (Mahalanobis, 1936). However, the statistical sophistication of the method does not eliminate the need for careful experimental design. Unrepresentative germplasm, inconsistent measurement, or inadequate replication can undermine the biological interpretation of otherwise correctly calculated distances.

5.2 Northern Kashmir as an agroecological context

The regional focus is relevant because genotype performance and phenotypic expression can vary with environmental conditions. A germplasm collection evaluated under Northern Kashmir conditions may display a different pattern of observed variation from material studied in coastal or other production environments. Singh and Kandasamy (2020), for

example, address bitter gourd diversity under coastal ecosystems, highlighting the importance of interpreting diversity research within its environmental setting.

Nevertheless, a regional field evaluation should not automatically interpret all observed variation as genetically determined. Differences may arise from environmental heterogeneity, interactions between genotype and environment, or measurement error. Replication and appropriate statistical treatment are therefore essential. If the objective is to recommend cultivars for the region, evaluation over multiple seasons or locations would provide stronger evidence of adaptation than a single trial.

The National Horticulture Board (2022) provides a horticultural information reference that may help establish the broader crop context. However, general crop information cannot substitute for local genotype-level measurements or demonstrate the diversity of a particular germplasm collection.

5.3 Relationship with quantitative-genetic research

Divergence, heritability, and genetic advance answer different questions. Multivariate divergence concerns the separation of observed trait profiles; heritability addresses the genetic contribution to observed variation under a specified population and environment; and genetic advance concerns expected selection response. Talukder et al. (2018) and Triveni et al. (2021) provide relevant context for the latter two dimensions.

Integrating these perspectives can improve breeding decisions. A divergent cluster characterized by desirable fruit traits may deserve further consideration, but the expected response to selection depends on the genetic basis of those traits and the reliability of their measurement. Similarly, a trait that strongly distinguishes clusters is not necessarily the trait that contributes most to yield improvement. Statistical differentiation and breeding value should therefore remain separate analytical concepts.

5.4 Limitations and methodological trade-offs

Several limitations require attention. First, distance estimates depend on the traits included in the analysis. Omitting important characteristics may produce an incomplete representation of divergence, while including many redundant or poorly measured traits may destabilize covariance estimation. Trait selection should consequently follow the research objective rather than a preference for maximizing the number of variables.

Second, cluster formation depends on the chosen algorithm and its decision rules. A cluster is an analytical grouping, not necessarily a naturally discrete biological population. Robustness checks should determine whether the major groups remain reasonably consistent under alternative defensible specifications.

Third, phenotypic divergence does not necessarily equal molecular genetic divergence. The proposed analysis can characterize quantitative phenotypic differentiation, but claims about genomic relationships would require suitable molecular evidence. This distinction is especially important when interpreting clusters as evidence of ancestry or genetic relatedness.

Fourth, geographic isolation should not be assumed to guarantee genetic distinctiveness. Genotypes from different locations may share similar trait profiles, while genotypes collected from a relatively restricted area may differ considerably. Actual measurements must determine the observed pattern.

5.5 Implications for practical breeding

The framework offers a decision-support approach for selecting germplasm that combines desirable performance with useful differentiation. Researchers should first identify genotypes that meet minimum agronomic requirements, then assess their multivariate relationships, and finally evaluate promising parental combinations through controlled crossing and progeny assessment where appropriate.

A greater distance between candidate parents may justify investigation, but it does not guarantee heterosis, favourable trait inheritance, or superior offspring. Those outcomes require experimental verification. Similarly, a cluster characterized by high mean yield may still contain individual genotypes with substantially different maturity or fruit characteristics. Selection should therefore operate at both the cluster and genotype levels.

The strongest practical contribution of this approach is the creation of a transparent link between field characterization and breeding decisions. Its usefulness will depend on the quality of the local dataset, appropriate statistical analysis, and independent confirmation of the agronomic value of selected materials.

6. Conclusion

Multivariate clustering and genetic divergence assessment provide a systematic basis for characterizing bitter gourd germplasm and supporting informed parental selection. The application of Mahalanobis D^2 statistics enables researchers to evaluate differences across several quantitative traits while accounting for their covariance structure (Mahalanobis, 1936). When combined with a transparent clustering procedure and agronomic evaluation, this approach can help identify potentially useful combinations of characteristics for crop improvement.

For Northern Kashmir, the proposed framework emphasizes standardized field observations, reliable trait measurement, appropriate distance estimation, cluster validation, and interpretation of cluster means in relation to breeding objectives. It also highlights the importance of distinguishing statistical divergence from genetic inheritance, adaptation, and actual breeding value. Research on bitter gourd performance, diversity, and quantitative-genetic variation provides a relevant foundation for this integrated approach (Resmi & Sreelathakumary, 2012; Sagar et al., 2022; Talukder et al., 2018).

The principal limitation is the absence of original genotype-level observations. Consequently, the present paper establishes an analytical framework rather than demonstrating empirical cluster patterns or identifying superior genotypes. Future research should apply the framework to replicated field data from Northern Kashmir, validate clustering stability, and evaluate promising materials across seasons or locations. Where feasible, molecular characterization and progeny testing could complement phenotypic divergence analysis.

Ultimately, the value of genetic divergence assessment lies not in maximizing statistical distance alone but in identifying distinct, agronomically useful germplasm that can contribute to measurable improvements in bitter gourd production.

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