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EXPLORING MORPHOLOGICAL TRAITS FOR ENHANCED YIELD POTENTIAL IN PAKISTANI RICE

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ABSTRACT

This study evaluated phenotypic variability among 14 selected rice (*Oryza sativa* L.) varieties using 16 quantitative morphological and physiological traits. Analysis of Variance (ANOVA) revealed significant differences ($p < 0.05$) across all traits, with coefficients of variation ranging from moderate to high, indicating substantial diversity. Physiological parameters measured at the reproductive stage, including photosynthetic and gas exchange traits, exhibited significant genotypic effects, while morphological descriptors reflected clear varietal differentiation despite potential environmental influences. Correlation analysis identified key trait associations relevant for yield improvement, highlighting their utility in breeding selection. Principal Component Analysis (PCA) revealed that the first five components (eigenvalues greater than 1) accounted for 81.8% of the total variance, with significant contributions from yield and phenological traits. Cluster analysis based on standardized data grouped the varieties into two major clusters, separating highly similar subgroups and identifying the most divergent pair—Basmati 2000 and Kisan Basmati. These findings demonstrate substantial phenotypic and genotypic divergence within the germplasm, providing valuable parental combinations for breeding programs that target yield potential and environmental adaptation.

Keywords: Rice (*Oryza sativa* L.), Agro-morphological traits, STRUCTURE analysis, Dendrogram clustering, Population structure, Yield potential.

INTRODUCTION

Over the next thirty years, the production of staple cereal grains, such as *Triticum aestivum*, *Zea mays*, and *Oryza sativa*, must be increased to keep pace with the growing global population and rising income levels. Overall, agriculture is jeopardized due to climate change caused by human activities, which forces farmers and plant breeders to compete with increased abiotic and biotic stresses. Breeding rice varieties native to the Asian tropics is much more susceptible to insect pests, viral, bacterial, and fungal agents than any other major cereal. The production of non-irrigated rice is more negatively affected by abiotic stress, such as drought and floods. Rice yield per unit area in Pakistan is very low compared to other developed countries of the world. The reduced production of rice in Pakistan is attributed to several environmental factors, including drought stress and various diseases caused by numerous microorganisms, which have led to a decline in rice yields every year (Khush and Virk, 2005; Asghar *et al*., 2007).

Smart plant breeders are giving more attention to simple and quantitative traits, which are used to create new rice lines guaranteeing better yield and quality enhancement over existing varieties. In Asia, the majority of public sector rice breeding programs employ conventional breeding methods, which is an extremely time-consuming process; on average, up to 10 years are required to produce and identify elite genotypes (Collard and Mackill, 2008).

With an increase in population, there is an alarming gap between the growing demand for rice and current production levels, posing a significant challenge for breeders. In order to meet the following challenge, we need to increase either the area under production or the yield, or both. It appears difficult to increase the area under production due to restraints such as waterlogging, drought, and salinity. Whereas increasing the yield seems to be a potential option, it can be achieved by using better management practices and enhanced varieties with higher yield potential. Management practices are efficient only if a variety has a genetic potential for higher yield. Enhancement in genetic potential depends on the genetic diversity & genetic maps of the variety that carries information of genes responsible for different agronomic characteristics (Reynolds and Borlaug, 2006).

To examine the germplasm reserves for crop consumption and its improvement programs, it is necessary to define and assess the phenotypic traits of the germplasm so that each cultivar can be classified and distinguished efficiently. The traits of panicle are instinctive and an illustrative part of the rice plant, which are used to determine its uniqueness, genetic diversity, quality, and yield (Kordrostami and Rahimi, 2015; Lei *et al.*, 2018). The selection of morphological characters for phenotyping the germplasm depends on the use of that germplasm. The traits related to yield components (grain yield) are chosen for the assessment of yield improvement.

The most essential step in plant breeding is the selection of plants with desired characteristics. For this purpose, the traditional method used by plant breeders depends on morphological traits, which are the visible indicators of genes, giving some idea about their genotype (Jehan and Lakhanpual, 2006).

MATERIALS AND METHODS

Seed Collection and Surface Sterilization

A total of fourteen rice (*Oryza sativa* L.) cultivars were used in this study. To minimize microbial contamination, seeds were surface-sterilized by immersion in a 2% sodium hypochlorite (NaOCl) solution for 20 minutes in sterile petri dishes. The seeds were then rinsed three times with distilled water for 30 seconds per rinse to remove residual NaOCl.

Field Layout and Experimental Design

The field experiment was laid out using a Randomized Complete Block Design (RCBD) with three replications. Each rice variety was transplanted in rows at a spacing of 25 cm × 25 cm. Each block consisted of five rows, with ten plants per row, for each variety. Standard agronomic practices were followed throughout the growing season, including the application of herbicides and insecticides to control weeds and pest infestations.

Phenotyping

Yield and Yield-Related Traits

Phenotypic data were recorded from ten randomly selected and tagged plants per plot. Sixteen agronomic traits were assessed, based on previously established protocols (Huang *et al*., 2010; Abou-Khalifa and El-Rewainy, 2012; Nguyen *et al*., 2018; Qaseem *et al*., 2019):

- **Days to 50% Flowering (DF):** The number of days from sowing to the date when 50% of plants in a plot had flowered.
- **Days to Maturity (DM):** Number of days from emergence to physiological maturity.
- **Plant Height (cm):** Measured at maturity from the soil surface to the tip of the tallest panicle (excluding awns) using a meter rod.
- **Number of Effective Tillers per Plant:** Counted as tillers bearing panicles.
- **Total Number of Tillers per Plant:** All tillers, productive and non-productive, were counted.
- **Panicle Length (cm):** Measured from the neck to the tip (excluding awns).
- **Panicle Weight (g):** Determined using a digital weighing balance.
- **Flag Leaf Area (cm^2):** Estimated using the formula:

Leaf area = K × length × width

Where K = 0.75 during vegetative and reproductive stages, and 0.67 at seedling and harvest stages (Bhan and Pande, 1996).

- **Number of Seeds per Panicle:** Counted manually.
- **Seed Weight per Panicle (g):** Weighed using a digital balance.
- **1000-Grain Weight (g):** Weight of 100 grains (in triplicate) averaged and multiplied by 10.
- **Seed Length and Width (mm):** Measured using a digital Vernier caliper.
- **Seed Length/Width Ratio:** Calculated as the ratio of seed length to width.
- **Yield per Plant (g):** Average grain yield from ten tagged plants.
- **Yield per Square Meter ($g\ m^{-2}$):** Calculated using the formula:

$$\text{Yield} = \frac{\text{No of head per sq meter x Av.no.of grains per head}}{100} \times \text{Weight of 100 grains}$$

Physiological Traits

Photosynthetic and gas exchange parameters were recorded from fully expanded leaves using an Infrared Gas Analyzer (IRGA), following the methodology of Moon *et al*. (1987) and Pandey *et al*. (2017). The traits measured included:

- **Change in CO_2 Concentration (ΔCO_2, µmol mol⁻¹):** Calculated as the difference between reference and sample CO_2 concentration.
- **Photosynthetic Rate (A, µmol m^{-2} s^{-1})**
- **Transpiration Rate (E, mmol m^{-2} s^{-1})**
- **Stomatal Conductance (Gs, mmol m^{-2} s^{-1})**
- **Intercellular CO_2 Concentration (Ci, µmol mol^{-1})**
- **Conductance to Water Vapor (Gj, mol m^{-2} s^{-1})**
- **Stomatal Respiration Rate (R, µmol m^{-2} s^{-1})**

Measurements were taken under controlled conditions: ambient CO_2 concentration (C_ref) of 328.39 µmol mol^{-1}, leaf surface area of 6.25 cm^2, leaf chamber temperature between 37.9°C and 48.3°C, chamber gas flow rate of 389 mL min^{-1}, molar gas flow rate (U) of 239 µmol s^{-1}, and a light intensity (PAR) of up to 921 µmol m^{-2} s^{-1}. Ambient pressure was maintained at 99.23 kPa.

Chlorophyll Content

Chlorophyll content (µmol m^{-2}) was estimated using the acetone extraction method (Rajalakshmi and Banu, 2015). Fresh leaf samples were homogenized in 25 mL of chilled 80% acetone, filtered, and the absorbance was measured at 645 nm and 663 nm using a UV-Vis spectrophotometer. Chlorophyll concentrations were calculated using the formula:

- Chlorophyll a = $12.7(A_{663}) - 2.69(A_{645})$
- Chlorophyll b = $22.9(A_{645}) - 4.68(A_{663})$
- Total Chlorophyll = $20.2(A_{645}) + 8.02(A_{663})$

Relative Water Content (RWC)

Leaf water status was determined as Relative Water Content (RWC) following Barr and Weatherley (1962). Fully expanded leaves (1 × 7 cm segments) were collected and immediately weighed (W), then hydrated for 3–4 hours to full turgidity (TW), and finally oven-dried at 80°C for 24 hours to obtain dry weight (DW). RWC was calculated using the formula:

$$RWC (\%) = [(W-DW) / (TW-DW)] \times 100$$

Where W stands for sample fresh weight, TW stands for sample turgid weight, and DW is used for sample dry weight.

Statistical Analysis

For testing differences among rice varieties, two-way Analysis of Variance (ANOVA) was performed for each trait. Where significant differences were detected, post-hoc pairwise comparisons were carried out using Tukey’s Honestly Significant Difference (HSD) test, which controls for family-wise error rate while providing robust detection of significant differences between means. Pearson’s correlation coefficients (r) were calculated using SPSS Statistics (Version 21, IBM Corp.) to evaluate interrelationships among traits. The same software was also employed to construct a dendrogram through hierarchical cluster analysis, using Ward’s linkage and squared Euclidean distance as the similarity measure. This allowed visualization of genetic relatedness and grouping patterns among rice varieties.

To investigate overall genetic variability and to reduce dimensionality, a Principal Component Analysis (PCA) and biplot were carried out in R Studio (Version 2023.09.1+494, R Foundation for Statistical Computing). The PCA was based on the correlation matrix of standardized quantitative traits, and principal components with eigenvalues > 1 were retained according to the Kaiser criterion. Biplot were generated to display both variety distributions and trait loadings.

RESULTS

Morphological traits

Comparative performance of morphological traits

Two-way ANOVA (Analysis of Variance) indicated significant ($P<0.01$ and 0.05) effects of varieties in almost all the traits studied (Table 1). All the varieties showed highly significant variations for all the traits studied. The extent of variability for any character is very important for the improvement of crops through breeding. Significant variation in all the traits studied indicated the presence of high genetic diversity among the varieties under study. The sum of Squares (SS) determined the total variation in data for each trait; larger values indicated high variability, and smaller values showed that there was less variability. Mean Square (MS) provided a means to measure the average variation among each group; the greater the value, the greater the difference among groups. The F-value specified the ratio of the variance explained by the model to the variance within the samples, offering an insight into the statistical significance of the results.

Table 1. Analysis of Variance in Selected Rice Varieties for sixteen Morphological Traits.

Trait	Sum of Squares (SS)	Mean Square (MS)	df	F-value	Significance
Days to 50% flowering	169	13.00	13	6.53	0.033
Days to maturity	177	13.62	13	6.85	0.04
Plant Height	228	17.54	13	8.82	0.007**
No. of Effective Tillers	46	3.54	13	1.78	0.057.
Panicle Length	192	14.77	13	7.42	0.002**
Panicle Weight	159	12.23	13	6.15	0.074.
Flag Leaf Area	228	17.54	13	8.82	0.024*
No. of Seed per Panicle	228	17.54	13	8.82	0.466
Weight of Seed per Panicle	199	15.31	13	7.70	0.026*
Thousand Grain Weight	196	15.08	13	7.58	0.01**
Seed Length	108	8.31	13	4.18	0.04
Seed Width	129	9.92	13	4.99	0.02*
Seed Ratio	213	16.38	13	8.23	0.02*
Yield per Plant	228	17.54	13	8.82	0.04
No. of Tiller per Plant	127	9.77	13	4.91	0.03
Yield per Square Meter	228	17.54	13	8.82	0.03

p ≤ 0.01 (**) = Highly significant, **p ≤ 0.05 (*)** = Significant, **p ≤ 0.1 (.)** = Marginally significant, **p > 0.1 (blank)** = Not significant

Comprehensive Trait Analysis of Rice Varieties

Fourteen rice varieties under study were evaluated for sixteen different traits as shown in Fig. **1-16**. Among the varieties evaluated, Basmati 2000 exhibited the highest yield per square meter (1636.80 g/m²), followed closely by Super Basmati (1775.67 g/m²) and BAS 198 (1621 g/m²). The high yield of Basmati 2000 was attributed to its superior panicle length (31.30 cm), panicle weight (5.90 g), and the highest number of seeds per panicle (176). Additionally, it had a relatively higher thousand-grain weight (31 g) and a moderate plant height (159 cm), which contributed to its high grain production. Similarly, Super Basmati, which ranked second in yield, had relatively longer maturity duration (140 days), a moderate number of tillers per plant (32), and a high number of seeds per panicle (179), making it a high-yielding variety.

BAS 198, which also exhibited a high yield (1621 g/m²), had a considerable number of seeds per panicle (157) and a high thousand-grain weight (35 g). Despite having a lower flag leaf area (56.70 cm²) compared to others, its superior grain weight and seed characteristics contributed to its higher yield. On the other hand, Basmati 515 and BAS Pak also performed well in terms of yield, producing 1117.20 g/m² and 1122.09 g/m², respectively. Basmati 515 had a relatively taller plant height (165 cm) and a high number of tillers per plant (28), while BAS Pak exhibited a significantly larger flag leaf area (71.35 cm²), which likely supported better photosynthetic efficiency and grain filling.

Contrastingly, PK 1121 Aromatic and PK 386 exhibited lower yield per square meter, with values of 744 g/m² and 802.72 g/m², respectively. They had relatively fewer seeds per panicle (93 and 173, respectively) and moderate to lower panicle weight. Although Super Gold had the highest plant height (178 cm) and a large flag leaf area (198.80 cm²), its yield (1009 g/m²) was only moderate, suggesting that plant height alone does not directly translate to yield potential. Similarly, Chenab Basmati had a lower yield (893.50 g/m²), despite having the highest number of seeds per panicle (201), which might indicate poor grain filling or other physiological limitations.

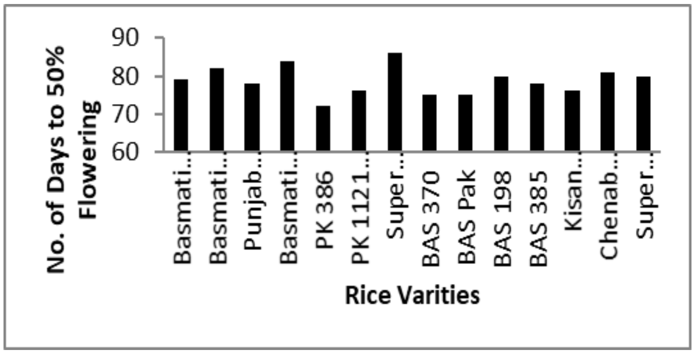

Fig 1. Comparison of days to 50% flowering among different rice varieties.

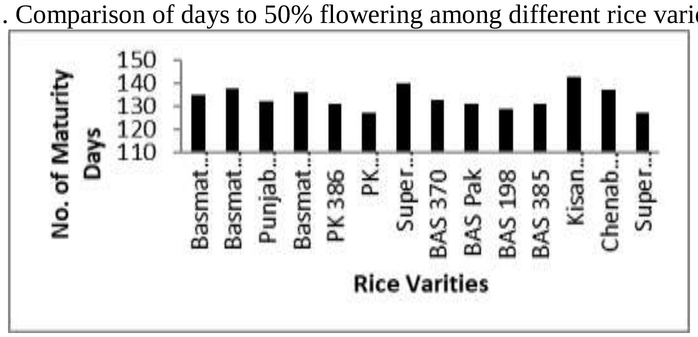

Fig 2. Comparison of days to maturity among different rice varieties.

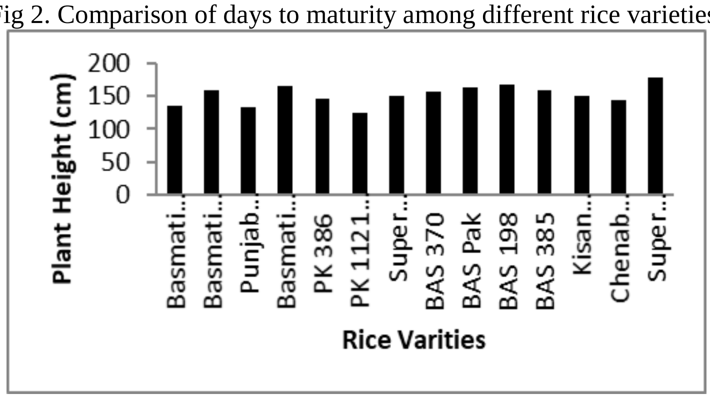

Fig 3. Comparison of plant height among different rice varieties.

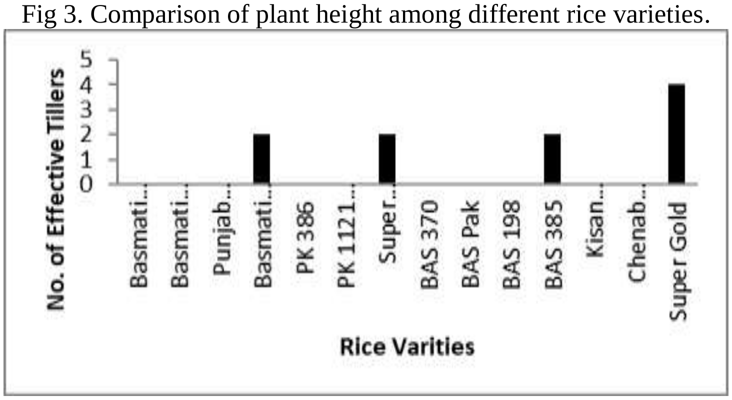

Fig 4. Comparison of no. of effective tiller among different rice varieties.

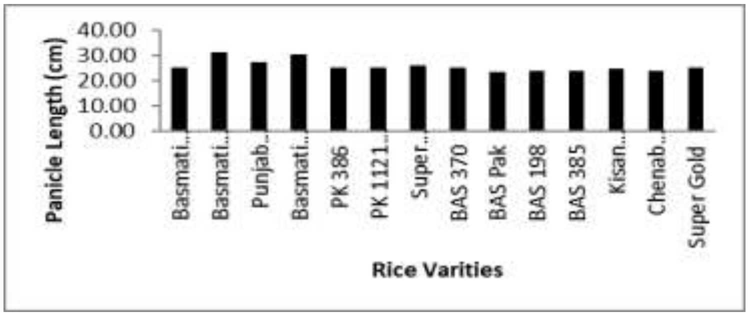

Fig 5. Comparison of panicle length among different rice varieties.

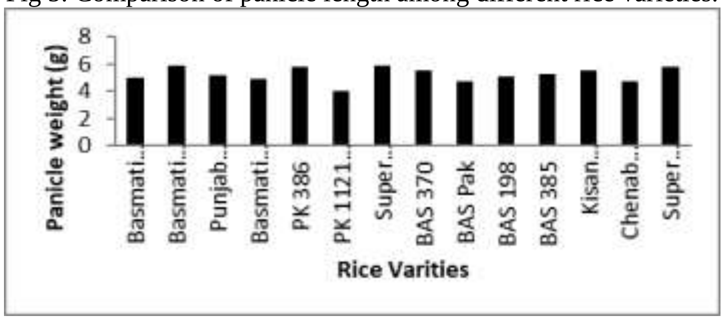

Fig 6. Comparison of panicle weight among different rice varieties.

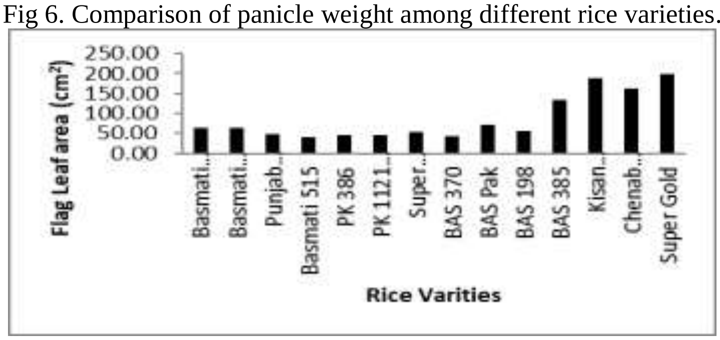

Fig 7. Comparison of flag leaf area among different rice varieties.

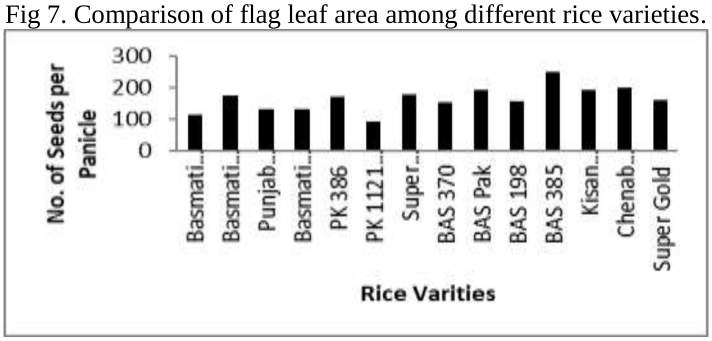

Fig 8. Comparison of the number of seeds per panicle among different rice varieties.

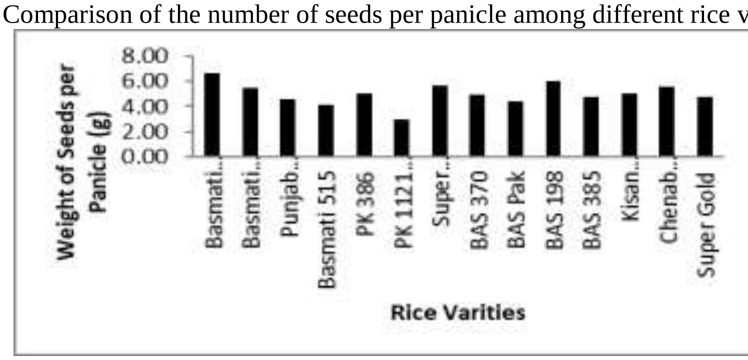

Fig 9. Comparison of the weight of seeds per panicle among different rice varieties.

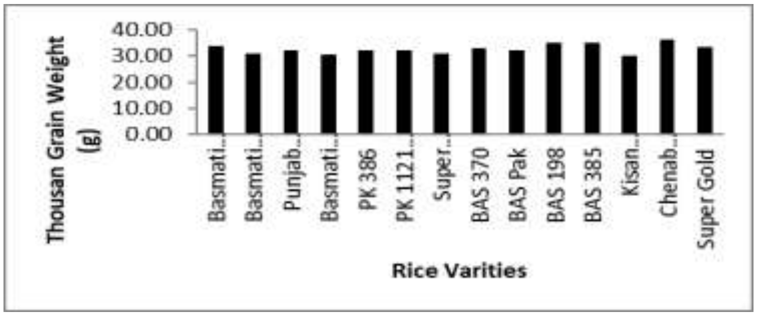

Fig 10. Comparison of thousand-grain weight among different rice varieties.

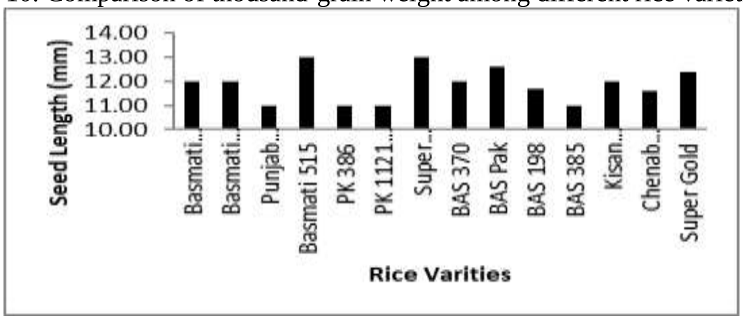

Fig 11. Comparison of seed length among different rice varieties.

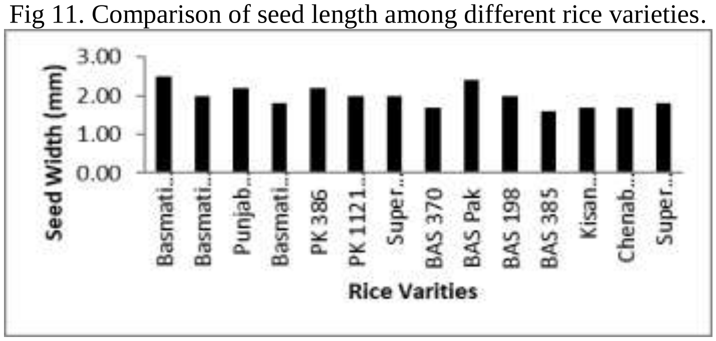

Fig 12. Comparison of seed width among different rice varieties.

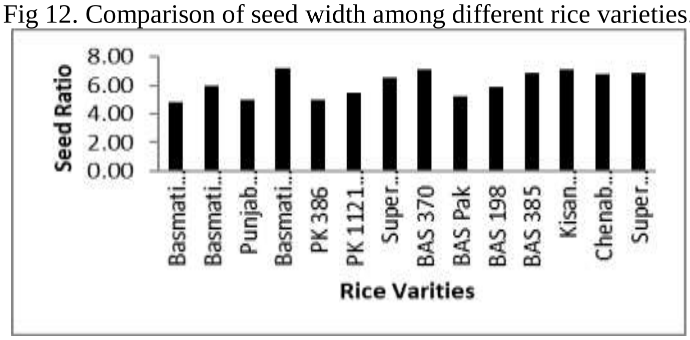

Fig 13. Comparison of seed ratio among different rice varieties.

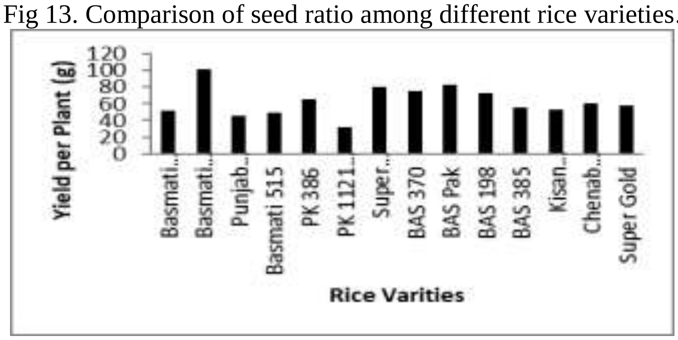

Fig 14. Comparison of the yield per plant among different rice varieties.

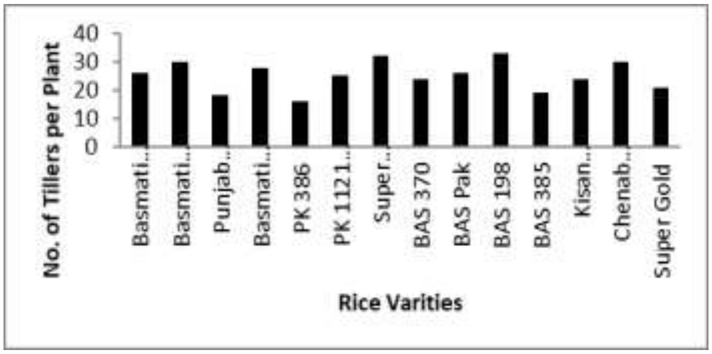

Fig 15. Comparison of the number of tillers per plant among different rice varieties.

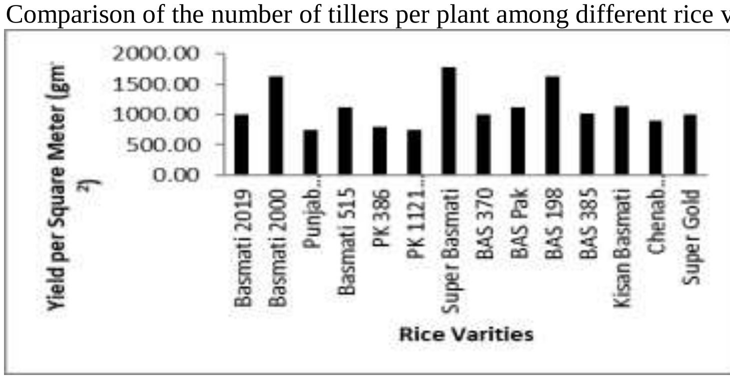

Fig 16. Comparison of yield per square meter among different rice varieties.

Correlation Coefficient Analysis

In order to find an essential relationship between two variables, a correlation study offers the best insight. It provides a better understanding of the association between different yield and yield-related components; in addition, it is also useful for plant breeders to select a variety with desired characteristics. Correlation coefficient analysis between all traits that were studied in this work is presented in Fig. 17.

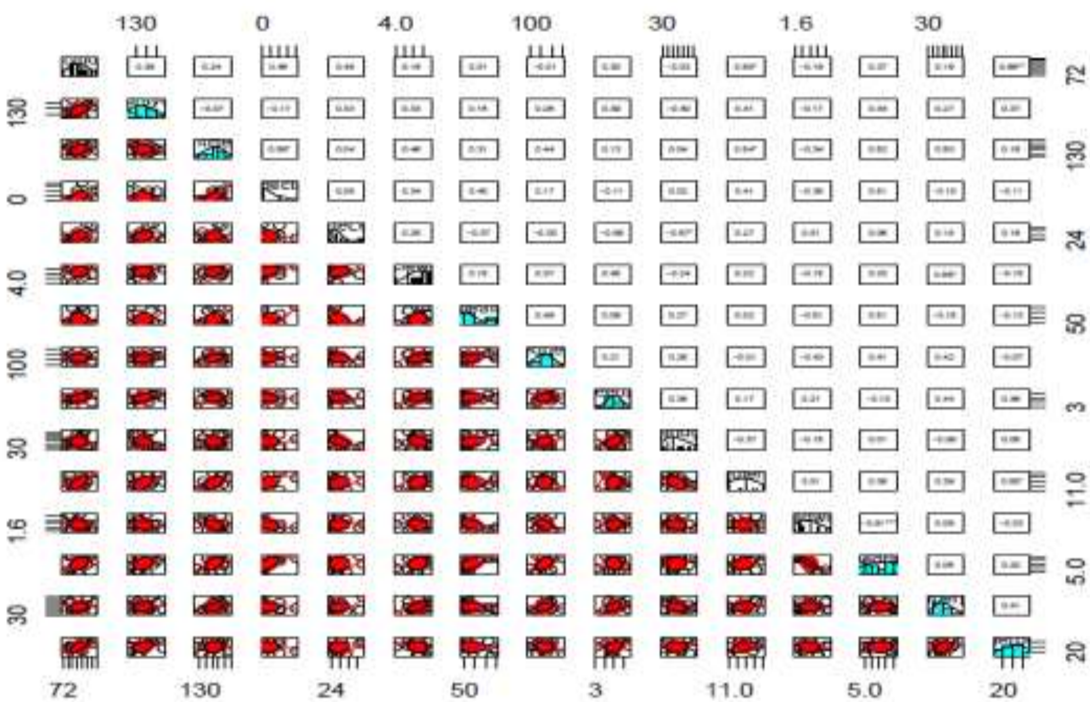

Fig 17. Pearson correlation among different physio-morphological traits of rice (*Oryza sativa* L.) under field conditions.

During the present study, days to 50% flowering were positively and significantly correlated with seed length, number of tillers per plant, and yield per square meter (r= 0.580*, r= 0.664**, and r= 0.626*). Plant height was positively and significantly correlated with the number of effective tillers and seed length (r= 0.576*, r= 0.537*). Panicle length was negatively and significantly correlated with 1000 grain weight (r= -0.567*). Panicle weight was positively correlated with yield per plant (r= 0.551*). Seed length was positively and significantly correlated with the number of tillers per plant and yield per square meter (r= 0.554*, r= 0.557*). Seed weight was negatively and significantly correlated with seed length-width ratio (r=-0.913**). On the other hand, yield per square meter was positively and significantly correlated with yield per plant and tillers per plant (r = 0.709**, r = 0.730**). Some physiological traits were also studied at the reproductive stage of each variety. Correlation studies data were recorded in Table 2. Results showed that the intercellular carbon dioxide concentration exhibited a positive and significant correlation with the change in carbon dioxide concentration and a negative and significant correlation with the transpiration rate (r = 0.714**, r = -0.547*). Whereas, a change in carbon dioxide concentration showed a significantly negative correlation with the transpiration rate (r = -0.647*).

Table 2. Simple correlation studies at the reproductive stage of experimental rice varieties.

	Ci	ΔCO_2	E	A	CC	Gs	ΔH_2O	R	Gj
Ci	1								
ΔCO_2	0.714**	1							
E	-0.547*	-0.647*	1						
A	-.0366^{ns}	-0.315^{ns}	-0.267^{ns}	1					
CC	0.286^{ns}	-0.121^{ns}	0.221^{ns}	-0.021^{ns}	1				
Gs	-0.406^{ns}	-0.387^{ns}	0.278^{ns}	0.131^{ns}	-0.013^{ns}	1			
ΔH_2O	-0.343^{ns}	-0.181^{ns}	0.242^{ns}	0.040^{ns}	-0.157^{ns}	-0.125^{ns}	1		
R	-0.186^{ns}	0.196^{ns}	-0.153^{ns}	0.196^{ns}	-0.227^{ns}	-0.413^{ns}	-0.311^{ns}	1	
Gj	0.03^{ns}	0.435^{ns}	-0.47^{ns}	-0.342^{ns}	-0.204^{ns}	-0.363^{ns}	0.045^{ns}	0.425^{ns}	1

** Correlation is significant at the 0.01 level (2-tailed); * Correlation is significant at the 0.05 level (2-tailed).

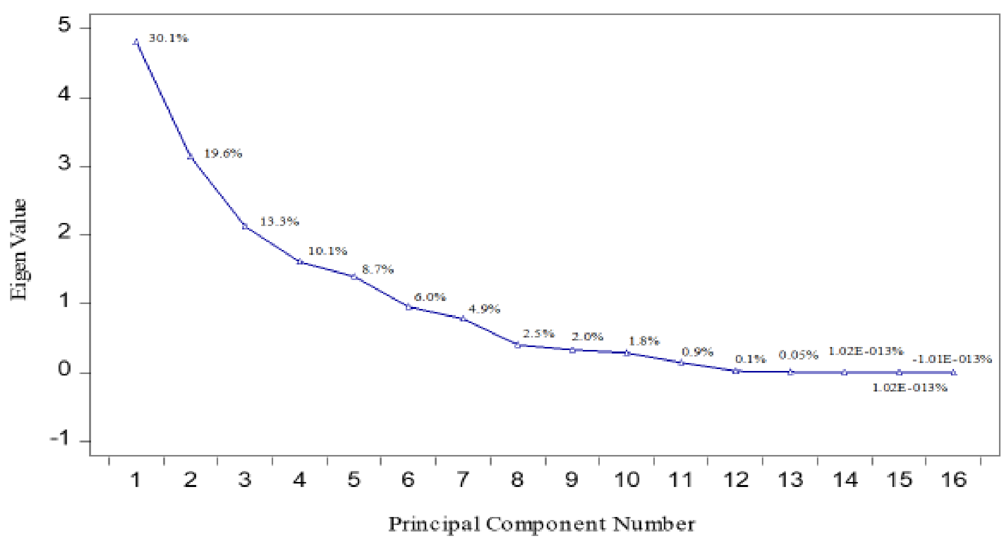

Fig 18. Scree plot of principal component analysis of experimental rice varieties between their eigenvalues and the number of principal components.

Principal Component Analysis

Principal Component Analysis (PCA) is used to evaluate the total variance of a specific trait, and each vector's coefficient indicates the effect of each variable on all related principal components. Coefficients are not dependent on direction (either positive or negative); a higher coefficient shows much more efficiency in order to distinguish different varieties.

For all morphological indices of different rice varieties, Principal Component Analysis (PCA) was executed as shown in Tables 1 and 2. Out of sixteen, five Principal Components (PCs) showed more than 1 eigenvalue and exhibited about 81.8% variability among the study traits for each rice variety. So, for further elucidation, these five PCs were granted due importance. The PC1 showed 30.1%, PC2 exhibited 19.6%, PC3 had 13.3%, PC4 showed 10.1% and PC5 displayed 8.7% variability among the study rice varieties. Eigenvalue and variance related to each principal, reduced gradually and stopped at -1.001E-013 and -1.01E-013, respectively (Fig. 18).

Bi-plot

Traits that are closer together on the plot were more positively correlated with each other. Traits like Yield per plant and Yield per square meter, Days to 50% flowering, and Seed length were closely aligned, suggesting a positive correlation. The length of the vectors (lines) from the origin indicated the contribution of each trait to the two dimensions. Longer vectors contributed more to the variation. Dim1 and Dim2 represented underlying factors or principal components that summarized the data. Dim1 was more related to traits affecting overall plant structure, while Dim2 captured specific yield-related traits. Dim 1 accounted for 30.1% of the variation in the dataset, whereas Dim 2 accounted for 19.6% of the variation in the dataset. Together, those two dimensions captured roughly 49.7% of the total variation, which suggested that they provided a good summary of the data (Fig. 19).

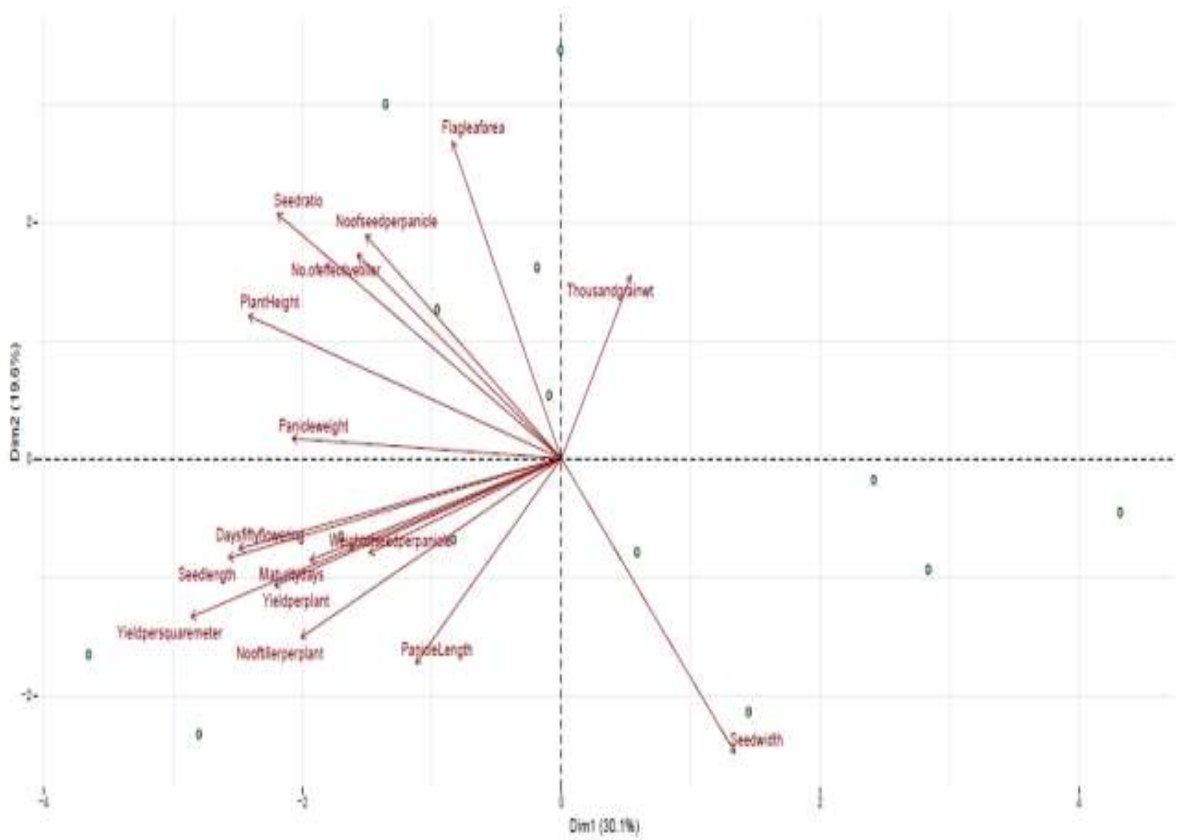

Fig 19. Bi-plot of 14 varieties of rice based on the first and second PC by the combined PCA for the sixteen physio-morphological traits.

Cluster Analysis

Cluster analysis based on quantitative morphological traits grouped the fourteen rice varieties into two main clusters, reflecting phenotypic variability within the collection (Fig. 20). In the dendrogram, the y-axis lists the rice varieties, while the x-axis represents the Euclidean distance (dissimilarity coefficient), ranging from 0 to 25. Smaller values indicate greater phenotypic similarity, whereas larger values denote greater dissimilarity. Horizontal lines represent the points at which clusters merge; longer lines indicate higher dissimilarity at the point of fusion.

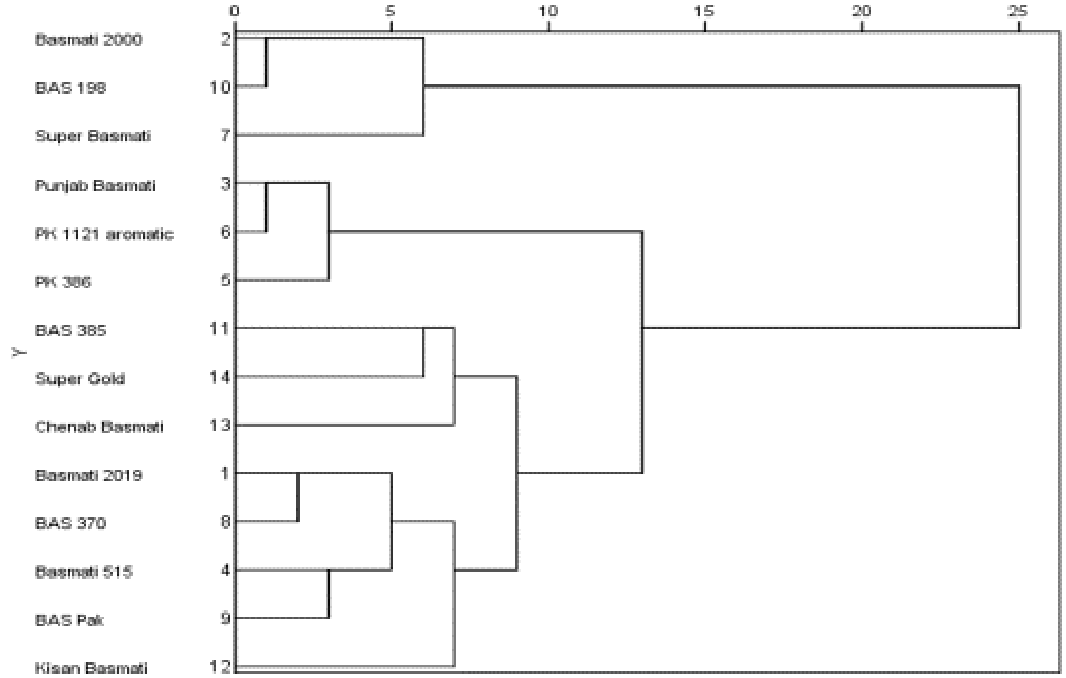

Fig. 20. Dendrogram of experimental rice varieties based on their morphological traits**.**

The dendrogram separated the varieties into two major clusters at a high dissimilarity level (~20). Cluster I (left branch) included: Basmati 2000, BAS 198, Super Basmati, Punjab Basmati, PK 1121 Aromatic, and PK 386. Within this cluster, Basmati 2000 and BAS 198 were the most closely related, joining at a low dissimilarity level (~2.5). Super Basmati merged with this subgroup at a slightly higher level, indicating high overall similarity among the three. Punjab Basmati and PK 1121 Aromatic formed another close subgroup, which subsequently merged with PK 386.

Cluster II (right branch) included: BAS 385, Super Gold, Chenab Basmati, Basmati 2019, BAS 370, Basmati 515, BAS Pak, and Kisan Basmati. Within this cluster, BAS 385 was somewhat distinct, joining the main group later in the hierarchy. Super Gold and Chenab Basmati formed a tight subcluster, indicating high similarity. A broader subcluster was formed by Basmati 2019, BAS 370, Basmati 515, BAS Pak, and Kisan Basmati, reflecting relatively close phenotypic resemblance among these varieties.

Varieties located at opposite ends of the dendrogram, such as Basmati 2000 (Cluster I) and Kisan Basmati (Cluster II), exhibited the highest dissimilarity (~25), suggesting substantial genetic or phenotypic divergence. These differences may be attributed to contrasting breeding origins, environmental adaptations, or targeted selection for specific agronomic traits.

DISCUSSION

In the present study, sixteen quantitative traits were evaluated to assess phenotypic variability among selected rice varieties. Analysis of Variance (ANOVA) revealed significant differences ($p < 0.05$) among varieties for all traits, with coefficients of variation (CV%) ranging from moderate to high. Comparable levels of variation in quantitative characteristics of fragrant rice cultivars were previously reported by Ashrafuzzaman *et al*., (2009). Both morphological and physiological traits contributed to the observed diversity, and several varieties exhibited particularly high values for yield and yield-related traits, reflecting their strong genetic potential. The presence of both phenotypic and genotypic divergence across all varieties is consistent with findings from Shahidullah *et al*., (2009), Huang *et al*., (2010), Hossain *et al*., (2020), Ara *et al*., (2022), and Fazal *et al*., (2023).

Morphological and physiological traits are widely utilized in rice germplasm characterization due to their ease of measurement and direct relevance to yield improvement (Peng *et al*., 2008; Yang and Hwa, 2008). In this study, physiological parameters—including change in CO_2 concentration, transpiration rate, photosynthetic rate, water conductance, intercellular CO_2 concentration, stomatal conductance, and stomatal respiration rate—were assessed at the reproductive stage and demonstrated significant genotypic effects. Similar responses for these traits have been documented by Basu *et al*., (2017) and Piveta *et al*., (2021).

Morphological traits also provide a direct means of detecting genetic differences among varieties and of evaluating performance under typical cultivation conditions (Anas and Yoshida, 2004; Li *et al*., 2009). However, their use in diversity studies can be limited by environmental influences (Naghavi *et al*., 2009) and pleiotropic gene effects. Despite these constraints, previous work has shown that morphological descriptors remain valuable in assessing genetic diversity and cultivar evolution (Fufa *et al*., 2005), and their utility persists even in the presence of advanced molecular markers (Benesi *et al*., 2004).

To further explore trait relationships, correlation coefficient analysis was performed for healthy and infected plants. The correlation coefficient (r) describes the degree and direction of association between two traits (Khatun *et al*., 2015). Given the complex and polygenic nature of grain yield, correlation analysis is particularly important for identifying yield-related traits that can be targeted in breeding programs (Hossain *et al*., 2015; Girma *et al*., 2018). This approach provides insights into both dependent and independent variables, enabling selection of genotypes with desirable trait combinations (Nihad *et al*., 2020; Ara *et al*., 2022).

Principal Component Analysis (PCA) was employed to partition total variance among traits. Of the 16 principal components extracted, five had eigenvalues greater than 1 (4.809, 3.133, 2.123, 1.611, and 1.396), together explaining 81.8% of the total variability. The magnitude of each variable's coefficient within a principal component reflects its contribution to differentiating varieties, regardless of the sign of the coefficient (Vishnu *et al*., 2014). Similar high contributions from early components were observed by Saleh *et al*., (2021), where the first four PCs accounted for 80.3% of total variability; by Behera *et al*., (2022), where they explained 81.11% of variance across 21 varieties; and by Yadav *et al*. (2024), who reported four PCs (eigenvalues 3.09, 2.64, 1.97, and 1.14) cumulatively explaining 80.58% of variance.

Cluster analysis, based on standardized quantitative trait data, grouped the varieties into two major clusters. Cluster I contained six varieties (Basmati 2000, BAS 198, Super Basmati, Punjab Basmati, PK 1121 Aromatic, and PK 386), with Basmati 2000, BAS 198, and Super Basmati showing the highest similarity. Cluster II included eight varieties (BAS 385, Super Gold, Chenab Basmati, Basmati 2019, BAS 370, Basmati 515, BAS Pak, and Kisan Basmati), where BAS 385 was distinct, clustering separately before merging with the group. Super Gold and Chenab Basmati formed a close subcluster, while Basmati 2019, BAS 370, Basmati 515, BAS Pak, and Kisan Basmati were highly similar. The most pronounced dissimilarity was observed between Basmati 2000 (Cluster I) and Kisan Basmati (Cluster II).

These clustering patterns are comparable to earlier reports. Tiwari *et al*., (2013) grouped four varieties into two major clusters, with subclusters based on similarity percentages. Shah *et al*., (2016) analyzed 105 accessions and identified five major clusters corresponding to indica, japonica, aromatic, aus, and wild rice. Yadav *et al*., (2024) classified 20 varieties into seven clusters, with Cluster III containing the largest number of varieties and Cluster VII being monospecific, representing the most distinct variety.

Overall, the combined use of ANOVA, correlation analysis, PCA, and cluster analysis provided a comprehensive view of the genetic diversity and trait associations among the studied rice varieties. The identification of highly divergent and high-performing genotypes provides valuable resources for targeted breeding programs aimed at improving yield and adapting to environmental challenges.

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