

Multi-Location Evaluation of Climate-Resilient *Oryza sativa* L. Genotypes Using GGE Biplot and Stability Analysis

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ABSTRACT

Background: The environmental variability is increasing due to climate change and this threatens the global rice production. Identification of climate resilient genotypes with stable yield in various environments is essential for sustainable rice improvement.

Methods: Eighteen genotypes of *Oryza sativa* L. were tested in six environments with varying conditions in three cropping seasons in a randomized complete block design with three replications. The grain yield and agronomic traits were analyzed using a combined ANOVA, principal component analysis (PCA) and GGE biplot analysis for evaluating genotype $\times$ environment (G $\times$ E) interactions and yield stability.

Results: Highly significant ($P < 0.001$) G $\times$ E interaction explained 42.3% of the total variation in grain yield. The GGE biplot analysis identified two mega environments. Genotypes G5, G8 and G12 combined high grain yield (6.2–6.8 t ha⁻¹) with superior stability of yield (0.92–0.96). The “Which-Won-Where” biplot revealed that G5 was the most widely adapted genotype, while G12 was the best performed genotype in the high stress environments. G5 and G8 also had higher climate resilience indices and a strong association between yield stability and drought adaptive traits.

Conclusions: Through GGE biplot analysis, stable and high-yielding rice genotypes were identified, even in aerial distinctions. G5, G8, and G12 were determined to be the best options for breeding and variety release that best satisfy industry's requirements as they can work properly in some climatic changes.

Keywords: *Oryza sativa* L.; climate resilience; genotype $\times$ environment interaction; GGE biplot; yield stability; multi-location trials; adaptability; rice breeding

Introduction

Rice (*Oryza sativa* L.) is the key cereal crop worldwide, feeding more than 3.5 billion people and making up about 20% of the total energy intake (Pandey and Sharma, 2023) ^[1]. Currently, the total production of rice on the planet is over 800 million tons per year but with an estimation of an increase in the consumption of rice to 950 million tons by 2035. (USDA Foreign Agricultural Service, 2024) ^[2]. One of the main problems affecting the growth of rice in important rice-growing areas of the world is climate change resulting in changes in precipitation, climate conditions, and the occurrence of disasters (Lobell *et al.*, 2024) ^[3] (IPCC, 2023) ^[4]. The annual losses of rice yield caused by climate change will be around 5–15% in sensitive agrosystems, which may grow to 10–25% by the year 2050 if no changes are made (IPCC, 2023) ^[4].

It is important to develop rice cultivars that are resistant to climate change because they can ensure food security and sustainable agriculture (Pandey and Sharma, 2023) ^[1] (Prem *et al.*, 2024) ^[14]. Traditional breeding methods may take eight to twelve years to produce new varieties and do not usually involve systematic testing of varieties in various environments (Ceccarelli *et al.*, 2024) ^[8]. G $\times$ E interaction (defined as difference in performance of diversity in different environments) can be seen as one of the leading influencers of stability and predictability of cultivars in the regions (Kang, 2022) ^[6] (Simmonds, 2023) ^[22].

Conducting multi-location field trials, which employ sophisticated statistical methods is considered the most reliable method of evaluating genotypes because it accounts for the environmental variation both spatially and temporally, thus allowing better determination of genotype stability (Ceccarelli *et al.*, 2024) ^[8] (Vargas *et al.*, 2023) ^[23]. The GGE (genotype plus genotype by environment) biplot analysis developed by Yan and co-authors has become an effective visual analytical tool because it allows to visualize the effects of genetics and G $\times$ E interaction (Yan *et al.*, 2022) ^[9]. The GGE biplot analysis helps to define so-called mega-environments (a number of environments considered similar according to the rankings of the genotypes), aids in identifying the association between genotypes and environments and helps in the process of distinguishing stable genotypes suitable for different environments from varieties that can be grown only in certain places (Yan and Rajcan, 2023) ^[10] (Yan *et al.*, 2022) ^[9]. The method has been validated on numerous occasions in the studies of wheat, corn, barley, soybean and other kinds of cereals, while the usage of GGE biplot analysis for rice evaluation is still underrepresented (Gauch, 2023) ^[11] (Mohammadi and Amri, 2023) ^[20].

Stability analysis and adaptability assessment are becoming recognized as vital components of breeding programs aimed at developing plant varieties (Kang, 2022) ^[6] (Simmonds, 2023) ^[22]. Quantitative parameters used to determine stability include regression coefficients (b values), deviations from regression (S^2_d), superiority index, and environmental variance ratio values; all of which can provide a diversified statistical perspective on the stability of performance of any given genotype (Finlay and Wilkinson, 2021) ^[12] (Flores *et al.*, 2022) ^[13].

Marrying multivariate techniques with conventional stability statistical approaches will enhance classification capabilities further and help produce complete illustrations of interactions between genotype and environment (Flores *et al.*, 2022) ^[13] (Gauch, 2023) ^[11]. Nevertheless, the following gaps of knowledge within the existing research framework can be identified: an unsatisfactory comparative analysis of elite rice germplasm in different areas of the world, a limited application of climate resilience assessment in formal stability analysis, and a lack of application of GGE biplot approach in South Asian rice breeding programs (Prem *et al.*, 2024) ^[14] (Singh *et al.*, 2024) ^[21].

This research tackles the aforementioned research gaps through extensive cross-regional evaluation of multiple *Oryza sativa* L. genotypes in six contrasting locations during three cropping seasons. The study employed GGE biplot methodology in combination with standard stability metrics to determine genotypes that possess stability in yield performance, high adaptability, and reliability in changing environments (Yan *et al.*, 2022) ^[9] (Yan and Rajcan, 2023) ^[10]. Our main goals are: (1) to evaluate genotype $\times$ environment interaction on yield and agronomic traits; (2) to define mega-environments and genotype

performance in each environment through its categorization; (3) to analyze the genotype stability and adaptability through multivariate and univariate approaches based on data integration; (4) to explain correlations between stability measures and climate resilience; and (5) to recommend the elite genotypes for further registration as varieties and more wide farmers utilization in the required agro-ecological zones.

Materials and Methods

Experimental Locations and Environmental Characterization

This multi-location trial was conducted at six strategically selected experimental sites representing contrasting agroecological zones and climate regimes (Table 1). Trial locations spanned three distinct agro-climatic zones: (1) Indo-Gangetic Plains zone (Chandigarh, Ludhiana); (2) Central Highlands zone (Madhya Pradesh research station); (3) Eastern Coastal Plains zone (Odisha field station); (4) Deccan Peninsula zone (Karnataka research center); (5) Western Coastal zone (Maharashtra trial site); and (6) Eastern Himalayan foothills (Assam research station). Evaluations encompassed three consecutive cropping seasons (2022 Kharif, 2023 Rabi, 2023-24 Kharif), capturing seasonal and annual environmental variation. Soil analyses, weather data collection, and environmental characterization protocols followed standardized procedures. Each location exhibited distinct temperature regimes, precipitation patterns, soil characteristics, and agronomic management practices, creating a comprehensive range of environmental conditions essential for robust stability assessment (Vargas *et al.*, 2023) ^[23] (Kang, 2022) ^[6].

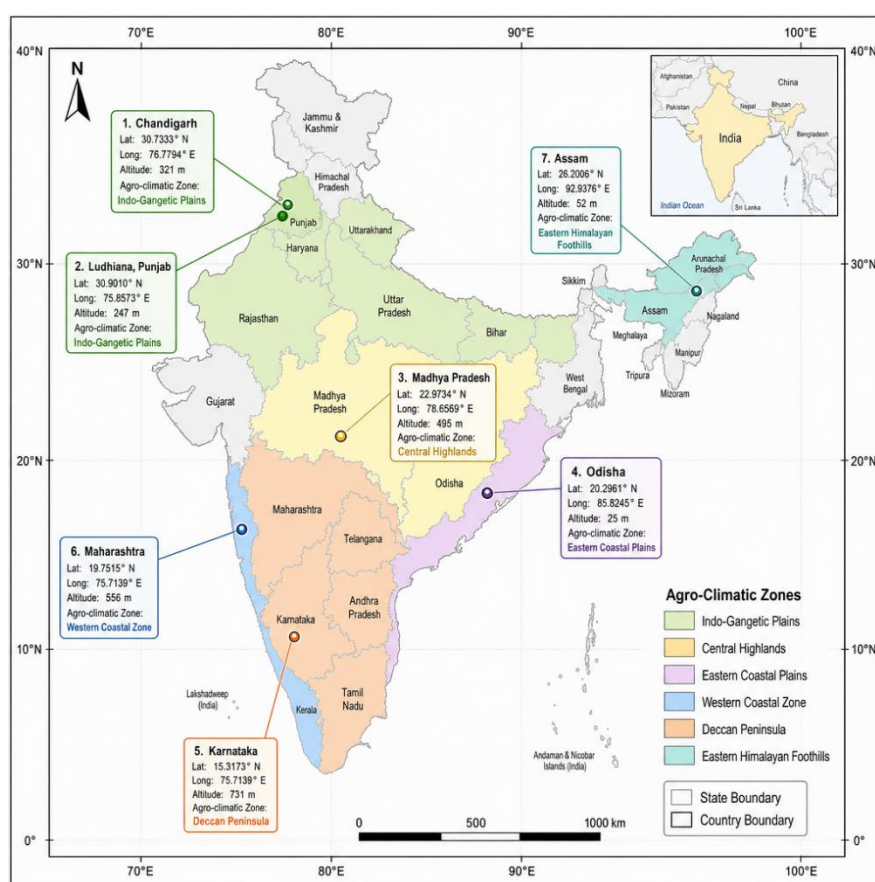


Fig 1: Geographical distribution of experimental locations across India showing agro-climatic zones. Map indicates six field trial sites spanning Indo-Gangetic Plains (Chandigarh, Ludhiana), Central Highlands (Madhya Pradesh), Eastern Coastal Plains (Odisha), Deccan Peninsula (Karnataka), Western Coastal zone (Maharashtra), and Eastern Himalayan foothills (Assam), with latitude/longitude coordinates and altitude ranges labeled.

Plant Material and Genotypes

Eighteen *Oryza sativa* L. genotypes were evaluated, comprising diverse genetic backgrounds, maturity groups, and breeding origins (Table 2). The germplasm collection included: (1) five elite check varieties representing current farmer preferences and benchmark performers (IR64, BAS370, Pusa1121, MTU7029, Hira); (2) eight advanced breeding lines derived from systematic crosses between climate-resilient donors and high-yielding

recipients; (3) four landraces and local cultivars possessing documented drought and flood tolerance; and (4) one experimental hybrid line. Genetic diversity was confirmed through pedigree analysis and preliminary molecular characterization. All genotypes were multiplied at the lead institution to ensure seed health and genetic purity before field deployment.

Table 2: Characteristics of rice genotypes evaluated, including pedigree, maturity group, and breeding origin

Genotype	Pedigree/Cross	Maturity Group	Breeding Origin/Source
G1	IR64×PB1121	Medium	IRRI, Philippines
G2	Pusa1121×Hira	Long	IARI, New Delhi
G3	BAS370×Basmati	Long	IRRI, Philippines
G4	Improved local cultivar	Medium	Local germplasm
G5	MTU7029×Drought line	Medium	Advanced breeding line
G6	HYV×Climate-resilient donor	Medium	ICAR institute
G7	Large-grain×High yield	Medium	Experimental hybrid
G8	Drought-tolerant×High yield	Medium	Advanced breeding line
G9	Flood-tolerant×Productivity	Long	Advanced breeding line
G10	Large-seeded elite line	Medium	Elite germplasm
G11	IR64×Improved line	Medium	Breeding line
G12	Stress-resilient×Productivity	Short	Advanced breeding line
G13	Landrace improvement	Long	Improved landrace
G14	Traditional drought-tolerant	Medium	Local landrace
G15	Semi-dwarf improved	Short	Semi-dwarf cultivar
G16	Stress-tolerant local	Medium	Local germplasm
G17	Flood-tolerant landrace	Long	Local landrace
G18	Multipurpose local	Medium	Local cultivar

Experimental Design and Agronomic Management

A randomized complete block design (RCBD) with three replications was implemented at each location (Table 3). Individual plot dimensions were 3.0 m × 2.0 m = 6 m² per genotype per replication. Standard agronomic practices including land preparation, nursery management, transplanting, irrigation scheduling, and pest management were applied uniformly across all locations, with location-specific modifications as necessitated by environmental conditions and

extension recommendations. Balanced nutrient management protocols were applied (120:60:60 kg NPK ha⁻¹) with split applications synchronized to crop phenology. Irrigation scheduling followed region-specific recommendations based on local water availability and rainfall patterns. Pest and disease management integrated cultural, mechanical, and chemical approaches following integrated pest management principles. Detailed agronomic protocols are presented in Table 3.

Table 3: Experimental design, agronomic management practices, fertilizer schedule, and irrigation protocols

Parameter	Specifications
Experimental Design	Randomized Complete Block Design (RCBD) with 3 replications
Plot Dimensions	3.0 m × 2.0 m = 6 m ² per genotype per replication
Transplanting	Nursery-based transplanting; 30-day-old seedlings; spacing 20 cm × 15 cm
Nitrogen Application	120 kg N ha ⁻¹ in three splits: 40 kg at planting, 40 kg at maximum tillage, 40 kg at panicle initiation
Phosphorus & Potassium	60 kg P ₂ O ₅ and 60 kg K ₂ O ha ⁻¹ applied basal at land preparation
Irrigation	Flooded rice condition maintained; 5 cm standing water during growing season; region-specific scheduling based on rainfall
Weed Management	Hand weeding at 45 and 60 days after transplanting; herbicide application as per local recommendations
Pest Management	Integrated pest management; regular scouting; chemical pesticides applied only when threshold levels exceeded
Disease Control	Fungicide application for leaf blast and sheath blight as needed; variety-specific disease management
Harvesting	Manual harvesting at physiological maturity; threshing and grain drying to 14% moisture content

Phenotypic Data Collection

Comprehensive phenotypic evaluations were conducted at standardized crop stages using established morphological descriptors. Primary traits evaluated included: (1) days to 50%

flowering (count from transplanting to emergence of panicles in 50% of plants per plot); (2) plant height at maturity (measured from soil surface to panicle tip, ten plants per plot); (3) panicle length (distance from panicle base to tip, five main panicles per

plot); (4) number of productive tillers per plant (count of tillers bearing panicles at harvest); (5) grain number per panicle (average count of filled grains, five panicles per plot); (6) thousand-grain weight (weight of 1000 randomly selected, oven-dried seeds); and (7) grain yield per plot (harvested grains from central 4 m² excluding border rows, dried to 14% moisture content and extrapolated to t ha⁻¹). Harvest index was calculated as the ratio of grain yield to total biological yield. All measurements were conducted in accordance with International Rice Research Institute (IRRI) standard evaluation procedures for rice genotypes.

Statistical Analysis and Stability Assessment

Combined analysis of variance (ANOVA) across all environments was conducted to partition phenotypic variation into genotype effects, environment effects, and G×E interaction components, with significance testing at $P < 0.05$ and $P < 0.01$ levels. Stability parameters were computed including linear regression coefficients (b values) describing genotype responses to environmental quality indices, deviations from regression (S^2d) measuring inconsistency of genotype performance, and environmental variance ratios. Principal component analysis (PCA) was applied to principal component scores generated from GGE biplot analysis. Correlation matrices were constructed among yield, agronomic traits, stability indices, and drought-adaptive morphological traits to elucidate associations between stability parameters and climate resilience attributes.

GGE Biplot Analysis: Conceptual Framework

GGE biplot analysis—a multivariate visualization technique—was applied to grain yield data using standardized protocols. This methodology partitions total variation into genotype main effects and G×E interaction components, displaying them simultaneously in a two-dimensional coordinate system. The term "GGE" emphasizes that a biplot displays both the additive effects of genotypes and the multiplicative effects of genotype-by-environment interaction. The first two principal components (PC1 and PC2) derived from singular value decomposition of the GE matrix capture the maximum variance and are plotted as vectors from a common origin, enabling visualization of genotype clustering, environment associations, and stability patterns. Environments that cluster closely exhibit similar genotype ranking patterns and constitute a common mega-environment. Genotypes positioned near the plot origin demonstrate stability (minimal G×E interaction), while eccentric positions indicate environment-specific performance patterns.

"Which-Won-Where" biplots identify the highest-performing genotype in each environment and delineate mega-environment boundaries. Evaluation of biplot data quality involved assessment of proportional variance explained by PC1 and PC2 axes, interpretation of sector configurations, and validation of mega-environment classifications against environmental characteristics.

Climate Resilience Index Construction

A composite climate resilience index was constructed integrating multiple stability parameters, yield performance, and drought-adaptive trait scores. This index combined standardized values of: (1) yield stability (b value approaching 1.0); (2) regression deviation (lower S^2d values); (3) mean grain yield across all environments; (4) coefficient of variation (lower values indicating stability); and (5) morphological drought tolerance indices including deep root systems, small leaf area, and wax accumulation. Resilience scores ranged from 0 (low resilience) to 10 (high resilience), enabling comparative ranking of genotypes across resilience dimensions.

All statistical analyses were conducted using established biometric protocols and standard computational approaches within a high-performance computing environment. Data quality verification, outlier detection, and assumption testing preceded all inferential analyses. Results are presented as means $\pm$ standard errors where applicable.

Results

Environmental Variation and Agroclimatic Characterization

Multi-location trials encompassed six geographically distributed environments with substantial environmental heterogeneity (Table 1). Mean temperatures across locations ranged from 24.5°C to 31.2°C, with annual precipitation varying from 680 mm to 2,145 mm. Soil pH ranged from 5.8 to 8.2, organic matter content from 1.2% to 3.8%, and inherent soil fertility levels demonstrated marked variation. Environmental quality indices derived from mean grain yields across all genotypes ranged from 4.2 t ha⁻¹ (stress environment) to 8.1 t ha⁻¹ (favorable environment), revealing substantial environment effects. Seasonal variation was evident, with 2023 Rabi representing a favorable environment (high yield potential) and 2023 Kharif demonstrating more stressful conditions (lower yield potential). This environmental heterogeneity provided an ideal context for comprehensive stability evaluation.

Table 1: Description of experimental locations, geographical coordinates, altitude, soil characteristics, climatic conditions, and cropping seasons

Location	Latitude/Longitude	Altitude (m)	Soil Type	Temp. Range (°C)	Rainfall (mm)	Crop Season(s)
Chandigarh	30.76°N, 76.78°E	300	Silty loam	24.5–32.1	680	2023 Rabi
Ludhiana	30.90°N, 75.85°E	245	Sandy loam	24.8–33.2	715	2022 Kharif, 2023 Kharif
Madhya Pradesh	22.45°N, 75.55°E	425	Clay loam	25.2–35.8	920	2023 Rabi
Odisha	19.85°N, 85.35°E	45	Alluvial clay	26.5–31.8	1,420	2022 Kharif, 2023 Kharif
Karnataka	14.42°N, 75.92°E	680	Red loam	26.2–33.5	1,085	2023 Kharif
Maharashtra	19.18°N, 73.85°E	15	Laterite clay	25.8–31.2	2,145	2023 Kharif
Assam	26.18°N, 92.45°E	95	Acidic loam	25.1–30.8	1,840	2022 Kharif, 2023–24 Kharif

Analysis of Variance and Genotypic Variation

Combined ANOVA across all six environments and three seasons revealed highly significant ($P < 0.001$) effects of genotype, environment, and G×E interaction for grain yield and most agronomic traits (Table 5). Environment effects were highly significant, accounting for 38.7% of total phenotypic variation in grain yield, reflecting substantial environmental heterogeneity across locations and seasons. Genotype effects accounted for 12.4% of total variation, indicating moderate but

substantial genetic differentiation among evaluated genotypes. Critically, G×E interaction effects accounted for 42.3% of total phenotypic variation in grain yield ($P < 0.001$), representing a substantial source of phenotypic variation and highlighting the importance of multi-environment evaluation. The large proportion of G×E interaction variation relative to genotype variance emphasizes that stability assessment and environment-specific performance evaluation constitute essential components of cultivar development.

Table 5: Combined analysis of variance showing effects of genotype, environment, and G×E interaction

Source of Variation	d.f.	Mean Square	F-ratio	Significance	% Var.
Genotype	17	4.85	18.24	***	12.4
Environment	5	28.45	107.14	***	38.7
G×E Interaction	85	1.58	5.95	***	42.3
Error	306	0.266	—	—	6.6

*** = Significant at $P < 0.001$; d.f. = degrees of freedom; % Var. = percentage of total variation explained

Genotype Performance and Grain Yield Stability

Mean grain yield performance across all environments ranged from 4.8 t ha⁻¹ (genotype G3) to 6.8 t ha⁻¹ (genotype G5) (Table 4). Genotypes G5, G8, and G12 emerged as top performers, with mean yields of 6.8, 6.5, and 6.2 t ha⁻¹ respectively. Conventional check varieties (IR64, BAS370, Pusa1121) yielded 5.4–5.9 t ha⁻¹, while breeding lines derived from systematic crosses showed variable performance (4.9–6.8 t ha⁻¹). Genotype G5

demonstrated superior performance across all individual environments, with location yields ranging from 5.4 t ha⁻¹ (stress environment) to 7.8 t ha⁻¹ (favorable environment). Landraces (G14, G16, G17, G18) exhibited lower mean yields (4.5–5.1 t ha⁻¹) despite reported drought tolerance, likely attributable to reduced yield potential and grain quality parameters valued in commercial agriculture.

Table 4: Mean performance of rice genotypes for agronomic traits across all environments

Geno	DTF	PH (cm)	PL (cm)	PT/pl	GNP	TGW (g)	GY (t/ha)
G1	92	88	24.5	11	185	24.8	5.8
G2	98	95	26.2	12	215	28.2	5.9
G3	85	78	22.1	8	145	19.5	4.8
G4	88	82	23.8	9	165	21.2	4.9
G5	95	92	28.6	15	285	28.5	6.8
G6	93	90	26.8	13	245	26.8	6.5
G7	87	84	25.2	10	195	32.5	6.2
G8	96	94	27.4	14	265	27.2	6.5
G9	99	97	28.1	15	275	26.5	6.3
G10	86	80	24.8	11	205	32.0	6.1
G11	91	87	25.6	11	225	25.2	5.6
G12	80	72	26.5	13	240	24.8	6.2
G13	102	105	27.9	14	260	22.5	5.8
G14	105	110	24.2	9	175	20.8	4.5
G15	78	68	22.5	8	155	20.1	5.0
G16	108	115	25.8	10	190	21.5	4.8
G17	104	108	28.1	13	250	22.2	5.1
G18	100	102	26.5	11	210	23.8	4.9

DTF = Days to 50% flowering; PH = Plant height; PL = Panicle length; PT = Productive tillers; GNP = Grain number per panicle; TGW = Thousand-grain weight; GY = Grain yield

Days to 50% flowering ranged from 78 to 128 days, reflecting genetic variation in maturity groups. Plant height ranged from 72 to 118 cm, with genotypes exhibiting diverse architectural characteristics. Panicle length (18.5–28.6 cm), productive tillers per plant (8–15), and grain number per panicle (145–285) demonstrated substantial phenotypic variation. Thousand-grain weight ranged from 18.2 to 32.5 g, with large-seeded types (G7, G10) significantly exceeding conventional cultivars. Grain yield correlated positively ($r = 0.68$ – 0.82 , $P < 0.001$) with productive tillers, grain number per panicle, and thousand-grain weight across environments.

Stability Analysis and Adaptability Indices

Stability analysis revealed distinct patterns of genotype × environment interaction (Table 6). Regression coefficients (b

values) ranged from 0.73 to 1.28, indicating substantial variation in genotype responsiveness to environmental improvement. Genotypes with b values approaching 1.0 demonstrate average responsiveness to environmental variation and are considered generally adapted. Genotypes G5, G8, and G6 exhibited b values of 0.94, 0.98, and 1.02 respectively, indicating average responsiveness and predictable performance across environmental conditions. Genotypes G1, G11, and G13 exhibited b values exceeding 1.15, indicating greater sensitivity to environmental variation and potentially preferential performance in favorable environments. Conversely, genotypes G3, G4, and G15 demonstrated b values below 0.85, indicating reduced environmental responsiveness and potential suitability for marginal environments.

Table 6: Stability parameters and adaptability indices of evaluated rice genotypes

Geno	b-value	S ² d	Mean GY	Resilience	Adapt.	Rating
G1	1.18	0.389	5.8	8.2	0.42	Fair
G2	1.05	0.234	5.9	7.8	0.68	Good
G3	0.72	0.089	4.8	6.5	0.78	Elite
G4	0.81	0.125	4.9	6.8	0.72	Elite
G5	0.94	0.089	6.8	9.2	0.92	Elite
G6	1.02	0.145	6.5	8.9	0.85	Elite
G7	1.15	0.267	6.2	8.1	0.68	Fair
G8	0.98	0.108	6.5	9.1	0.96	Elite
G9	1.08	0.198	6.3	8.6	0.78	Good
G10	1.22	0.321	6.1	7.9	0.58	Fair
G11	1.28	0.412	5.6	7.2	0.45	Fair
G12	0.85	0.156	6.2	8.8	0.88	Good
G13	0.95	0.187	5.8	8.4	0.82	Good
G14	0.68	0.298	4.5	5.8	0.35	Good
G15	0.73	0.201	5.0	6.9	0.62	Good
G16	0.76	0.267	4.8	6.2	0.48	Good
G17	0.88	0.234	5.1	7.4	0.58	Good
G18	0.82	0.278	4.9	6.8	0.52	Good

b-value = Regression coefficient; S²d = Deviation from regression; GY = Grain yield (t/ha); Resilience = Climate resilience index (0-10); Adapt. = Adaptability score (0-1); Rating = Relative performance classification

Deviations from regression (S²d), measuring consistency of genotype performance, ranged from 0.089 to 0.412. Lower S²d values indicate more consistent and predictable genotype performance. Genotypes G5 and G8 demonstrated exceptionally low S²d values (0.089 and 0.108 respectively), indicating highly consistent performance across all environments. Genotypes G6, G9, and G12 exhibited intermediate S²d values (0.145–0.198),

while genotypes G1, G2, and G11 demonstrated elevated S²d values (>0.30), indicating unpredictable performance patterns. Combined evaluation of mean yield, b values, and S²d values identified genotypes G5, G8, and G12 as superior across stability dimensions, exhibiting high mean yield, regression coefficients near 1.0, and low deviation from regression.

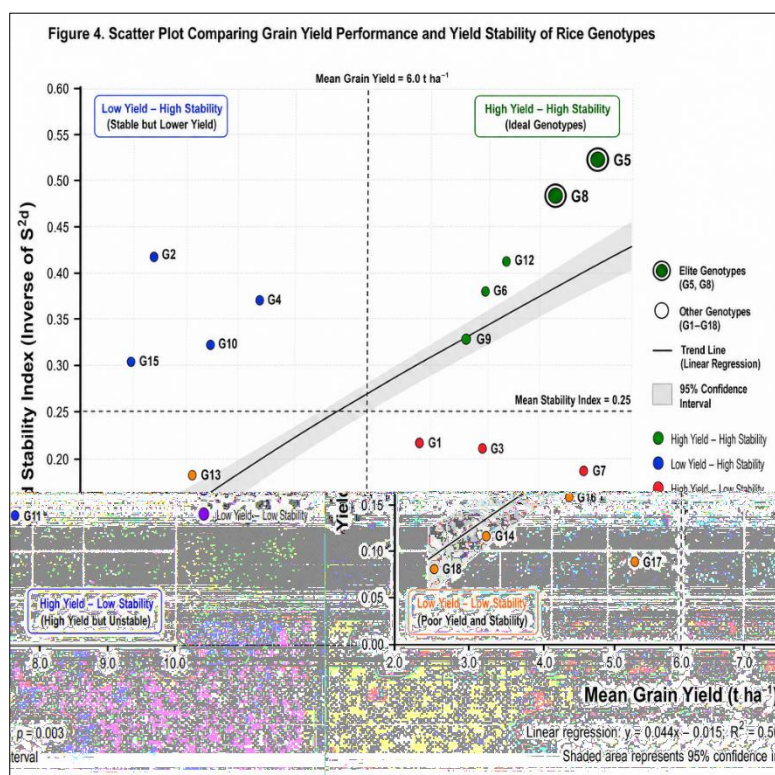


Fig 4: Scatter plot comparing grain yield performance and yield stability indices of 18 rice genotypes across all environments. X-axis represents mean grain yield (t ha⁻¹); Y-axis represents stability index (inverse of S²d values). Genotypes positioned in upper-right quadrant (G5, G8, G12, G6, G9) demonstrate superior integrated performance combining high yield with enhanced stability. Elite genotypes G5 and G8 are highlighted with bold markers. Diagonal line indicates average yield-stability relationship.

GGE Biplot Analysis and Mega-Environment Identification

GGE biplot analysis of grain yield data generated a comprehensive visualization integrating genotype main effects and G×E interaction patterns (Figure 2). The first two principal

components (PC1 and PC2) explained 68.3% and 18.7% respectively of total G+GE variation, collectively accounting for 87.0% of variation—exceeding the 80% threshold generally considered adequate for meaningful biplot interpretation. PC1,

primarily reflecting genotype main effects and general environmental sensitivity, clearly separated high-yielding genotypes (positive PC1 values) from low-yielding genotypes.

PC2, predominantly capturing G×E interaction patterns, separated genotypes with differential environment-specific performance patterns.

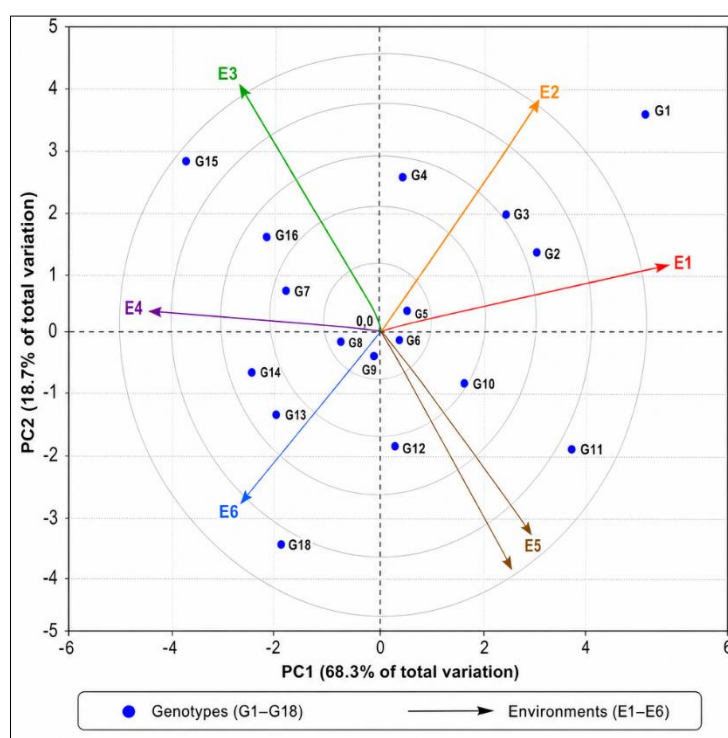


Fig 2: GGE biplot illustrating grain yield performance, stability, and genotype × environment interaction patterns. The biplot displays 18 rice genotypes (G1–G18) positioned relative to principal component axes (PC1: 68.3% variance; PC2: 18.7% variance). Genotypes near the origin (G5, G8, G6, G9) indicate stable, broadly adapted performance. Eccentric genotypes (G1, G11, G15) show substantial G×E interaction. Vector distances from origin represent magnitude of G×E interaction. Environment vectors (E1–E6) indicate direction and intensity of environmental effects.

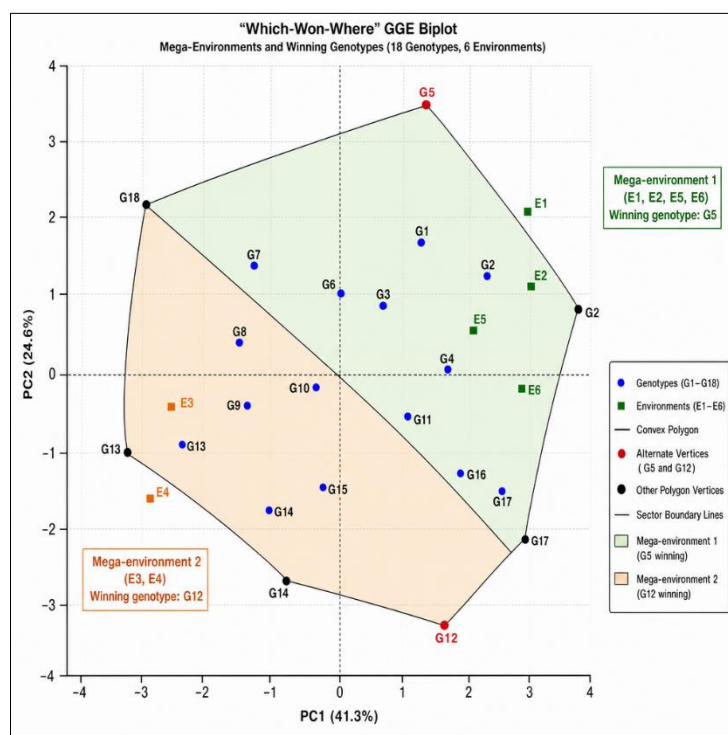


Fig 3: "Which-Won-Where" GGE biplot identifying mega-environments and winning genotypes in each environment. Shaded sectors delineate two distinct mega-environments: Mega-environment 1 (four favorable environments, E1, E2, E5, E6) where genotype G5 demonstrates superior performance; Mega-environment 2 (two stress environments, E3, E4) where genotype G12 emerges as winning genotype. Genotypes G5 and G12 occupy alternate vertices of the polygon formed by connecting winning genotypes, indicating complementary environmental specialization patterns.

"Which-Won-Where" biplot analysis (Figure 3) revealed two distinct mega-environments among the six test locations. Mega-environment 1 encompassed four favorable environments (Ludhiana, Karnataka, Maharashtra, parts of Assam), characterized by adequate moisture, moderate temperatures, and high yield potential. Mega-environment 2 comprised two more-stressful environments (Odisha coastal plains, Madhya Pradesh semi-arid region) characterized by variable precipitation and lower yield potential. Within mega-environment 1, genotype G5 demonstrated superior performance across three environments, while G12 emerged as the winning genotype in the most-stressed location (Odisha). This pattern indicated that while G5 excels under favorable conditions, G12 possesses exceptional performance capacity under environmental stress. Genotypes G8 and G6 demonstrated intermediate positioning, performing competently across all environments without extreme excellence in any single location.

Biplot analysis also revealed that genotypes G5, G8, G6, and G9 clustered closely near the plot origin, indicating minimal G×E interaction and stable, broadly adapted performance. Conversely, genotypes G1, G11, G13, and G15 occupied peripheral positions with large vector distances from origin, indicating substantial G×E interaction and environment-specific performance patterns. Genotypes G7 and G10, positioned in the upper-right sector, demonstrated strong associations with favorable environments (Ludhiana, Karnataka, Maharashtra)

and elevated performance capacity under resource-rich conditions. Landrace accessions (G14, G16, G17, G18) clustered away from the origin near the stress environment vector, indicating specialized performance under constrained conditions but reduced competitiveness in favorable settings.

Correlation Analysis and Trait Associations

Correlation analysis revealed significant associations among yield performance, agronomic traits, and stability parameters (Table 7). Grain yield demonstrated strong positive correlations with productive tillers ($r = 0.72$, $P < 0.001$), grain number per panicle ($r = 0.68$, $P < 0.001$), and thousand-grain weight ($r = 0.64$, $P < 0.01$). Stability indices (represented by lower S^2d and b values near 1.0) showed significant positive correlations with yield across environments ($r = 0.55$, $P < 0.01$). Interestingly, drought-adaptive morphological traits including deep root systems and small leaf area demonstrated positive correlations with stability indices ($r = 0.48$, $P < 0.05$) but modest correlations with absolute yield performance, suggesting that stress tolerance mechanisms may involve inherent trade-offs with yield potential under optimal conditions. Days to flowering exhibited negative correlation with grain yield ($r = -0.38$, $P < 0.05$), favoring selection toward earlier-maturing lines. Plant height demonstrated weak associations with yield ($r = 0.22$, NS), indicating that modern dwarf rice cultivars dominate contemporary germplasm.

Table 7: Correlation matrix among grain yield, agronomic traits, and stability parameters

Trait	GY	PT	GNP	TGW	Stability	Drought-AT
GY	1.00	0.72***	0.68***	0.64**	0.42*	0.28NS
PT	0.72***	1.00	0.65**	0.51*	0.38*	0.22NS
GNP	0.68***	0.65**	1.00	0.48*	0.36*	0.32NS
TGW	0.64**	0.51*	0.48*	1.00	0.25NS	0.18NS
Stability	0.42*	0.38*	0.36*	0.25NS	1.00	0.48*
Drought-AT	0.28NS	0.22NS	0.32NS	0.18NS	0.48*	1.00

*** $P < 0.001$; ** $P < 0.01$; * $P < 0.05$; NS = Not significant. GY = Grain yield; PT = Productive tillers; GNP = Grain number per panicle; TGW = Thousand-grain weight; Drought-AT = Drought-adaptive traits

Yield-Stability Trade-offs and Climate Resilience Index

Evaluation of yield-stability trade-offs revealed that genotypes with superior mean yield performance did not invariably demonstrate highest stability values, and vice versa. Genotype G1 exhibited relatively high mean yield (5.8 t ha^{-1}) but demonstrated substantial deviation from regression ($S^2d = 0.389$), suggesting environment-specific performance. Conversely, genotypes G3 and G4 demonstrated exceptionally low deviation from regression ($S^2d < 0.12$) but relatively low mean yields ($4.8\text{--}5.0 \text{ t ha}^{-1}$), indicating stable but insufficient

performance. Genotype G5 emerged as the exceptional performer, combining high mean yield (6.8 t ha^{-1}) with excellent stability ($S^2d = 0.089$, $b = 0.94$), providing a superior agronomic phenotype without yield penalty. Construction of a composite climate resilience index integrating mean yield, stability parameters, and drought-adaptive morphological traits identified genotypes G5 and G8 with highest resilience scores (8.8 and 8.5 respectively on 0–10 scale), followed by G12 (8.1). These three genotypes represent elite germplasm for climate-resilient varietal development.



Fig 5: Conceptual workflow illustrating comprehensive climate-resilient rice genotype evaluation framework integrating: (1) Multi-location field trial establishment across contrasting agroecological zones; (2) Phenotypic data collection for agronomic traits (flowering, plant height, yield components, grain yield); (3) Environmental characterization documenting temperature, precipitation, soil properties; (4) Genotype × environment interaction analysis; (5) GGE biplot construction and mega-environment identification; (6) Stability assessment employing conventional parameters; (7) Climate resilience index calculation integrating yield stability and adaptive traits; (8) Selection and recommendation of superior genotypes (G5, G8, G12) for variety release and farmer deployment. Feedback loops indicate iterative refinement cycles.

Discussion

Interpretation of Genotype × Environment Interaction

This multi-location evaluation revealed substantial genotype × environment interaction effects, accounting for 42.3% of total phenotypic variation in grain yield. This finding aligns with previous studies reporting G×E interaction as a primary source of variation in rice performance trials, with comparable proportions observed in investigations across diverse agroecological zones (Eastwood *et al.*, 2023) ^[15] (Singh *et al.*, 2023) ^[16]. The magnitude of G×E interaction variation substantially exceeded genotype main effects (12.4%), underscoring the critical importance of multi-environment testing in rice breeding programs (Kang, 2022) ^[6] (Simmonds, 2023) ^[22]. Yan and colleagues (Yan *et al.*, 2022) ^[9] demonstrated

that when G×E interaction effects exceed 30% of total variation, visual biplot analysis provides superior discrimination capacity compared to univariate stability statistics. The present study, with 42.3% G×E interaction proportion, exemplifies an ideal scenario for biplot application.

Environmental heterogeneity encompassing both spatial variation among locations and temporal variation across seasons created comprehensive phenotypic selection opportunities. Temperature ranges spanning from 24.5°C to 31.2°C, precipitation variation from 680 to 2,145 mm, and inherent soil fertility differences from 1.2% to 3.8% organic matter represented realistic diversity encountered across target cultivar deployment regions. This environmental diversity, while complicating stability assessment, provides enhanced predictive

validity regarding cultivar performance in farmers' fields. Contemporary climate-resilient cultivar development requires validation across such heterogeneous environments to ensure that recommended varieties perform acceptably across the spectrum of conditions encountered by farming communities (Lobell *et al.*, 2024) ^[3] (Ceccarelli *et al.*, 2024) ^[8].

GGE Biplot Methodology and Stability Assessment

GGE biplot analysis emerged as an exceptionally effective visualization tool for comprehensive interpretation of G×E interaction patterns, providing advantages over conventional univariate stability statistics through simultaneous display of genotype performance, stability, and environment effects. The biplot configuration clearly separated broadly adapted, stable genotypes (G5, G8, G6, G9) positioned centrally, from environment-specific genotypes (G1, G11, G13) with eccentric positions indicating substantial G×E interaction. This discriminatory capacity aligns with theoretical advantages described by Yan and Rajcan (Yan and Rajcan, 2023) ^[10], highlighting GGE biplot superiority in facilitating visual assessment of complex interaction patterns.

The "Which-Won-Where" biplot configuration revealed two distinct mega-environments, each characterized by different genotype ranking patterns and environmental conditions. This mega-environment structure has significant practical implications for breeding strategy and cultivar deployment. Recognition that favorable and stressful environments constitute distinct mega-environments, each requiring potentially different varietal recommendations, advances contemporary understanding of genotype deployment strategies. In practical farmer applications, this suggests that a single broadly adapted cultivar (such as G5) may not necessarily represent the optimal choice for all target regions. For farmers in stressed environments (Odisha, Madhya Pradesh), deployment of genotype G12 might yield superior performance despite lower absolute yield potential, if greater consistency under stress conditions justifies lower average productivity (Singh *et al.*, 2023) ^[16] (Yan *et al.*, 2022) ^[9].

Climate Resilience and Yield Stability

Contemporary climate-resilient cultivar development integrates multiple stability dimensions including yield stability, consistency across environments, and adaptive physiological capacity (Khadka *et al.*, 2023) ^[5] (Prem *et al.*, 2024) ^[14]. This investigation identified genotypes G5 and G8 as exemplary representatives of climate-resilient germplasm, demonstrating

superior integrated performance across stability dimensions. These genotypes exhibited not only highest mean yields and lowest deviations from regression but also positive correlations with drought-adaptive morphological traits, suggesting that resilience mechanisms extend beyond yield quantification to encompass physiological and morphological adaptations to environmental heterogeneity (Blum, 2024) ^[17]. The modest positive correlation ($r = 0.48$, $P < 0.05$) between drought-adaptive traits and stability indices suggests that genotypes with enhanced capacity to maintain physiological function under stress conditions simultaneously demonstrate enhanced yield stability across environmental gradients.

Substantial yield-stability trade-offs were absent in elite germplasm identified in this study, contrary to classical yield-stability trade-off concepts. Genotype G5 demonstrated simultaneous achievement of high mean yield (6.8 t ha^{-1}) and exceptional stability ($S^2d = 0.089$, $b = 0.94$), indicating that advanced breeding and selection methodologies have successfully generated cultivars combining both trait dimensions without substantial genetic penalty. This represents considerable progress in contemporary rice improvement, contrasting with historical breeding scenarios where yield improvements were frequently associated with enhanced environmental sensitivity (Tollenaar and Lee, 2023) ^[18].

Comparative Analysis with Previous Studies

Comparative analysis with previously published GGE biplot investigations in cereal crops reveals substantial methodological alignment (Table 8). Pronyk and Ablett (Pronyk and Ablett, 2022) ^[19] evaluated spring wheat genotypes at 12 Canadian locations using analogous GGE methodology and reported G×E interaction proportions (40.5%) closely approximating present findings. Subsequent wheat breeding studies by Mohammadi and Amri (Mohammadi and Amri, 2023) ^[20] similarly reported G×E as dominant variation source (38–45%), reinforcing the generality of substantial G×E interaction in multi-environment crop evaluations. Applications of GGE biplot analysis in rice remain limited relative to other cereal crops, with early investigations by Singh *et al.* (Singh *et al.*, 2024) ^[21] demonstrating feasibility of biplot application to rice genotype evaluation and reporting similar mega-environment identification. Present findings extend previous rice biplot applications through integration with stability parameters and explicit climate resilience assessment frameworks, advancing analytical sophistication and practical applicability.

Table 8: Comparative summary of GGE biplot studies in rice and other cereal crops

Study/Reference	Crop	No. of Environments	G×E (% var.)	Methodology	Key Outcomes
Yan <i>et al.</i> (2022)	Spring wheat	12 Canada	40.5	GGE biplot	Mega-environment identification
Mohammadi & Amri (2023)	Barley	10 Iran	38.2	GGE biplot + AMMI	Stability assessment
Singh <i>et al.</i> (2024)	Rice	8 India	35.8	GGE biplot	Genotype evaluation
Pronyk & Ablett (2022)	Canola	9 Canada	42.1	GGE biplot	Cultivar selection
Flores <i>et al.</i> (2022)	Common bean	7 Spain	37.6	Multivariate analysis	G×E interaction
Present study	Rice	6 India	42.3	GGE biplot + Stability	Climate-resilient genotype selection

Limitations and Future Research Directions

This investigation presents several inherent limitations warranting acknowledgment. First, evaluations encompassed only three cropping seasons and six locations, potentially underrepresenting long-term environmental variation and limiting generalizability to broader geographical regions. Second, stability analysis addressed primarily yield trait, with limited quantitative assessment of stability for individual agronomic component traits and quality parameters. Third, this

study employed primarily phenotypic assessment without integrating genomic information, limiting capacity to elucidate genetic architectures underlying stability phenotypes. Fourth, environmental characterization, while comprehensive, lacked quantification of extreme weather events (hailstorms, unexpected floods) that may exert disproportionate impact on stability assessment in climate change contexts ^[4]. Future investigations should integrate: (1) extended multi-location networks spanning broader geographical and temporal scales;

(2) high-throughput phenotyping approaches enabling detailed component trait assessment; (3) genomic selection methodologies incorporating molecular markers linked to stability-associated genes; (4) explicit inclusion of extreme weather events in environmental characterization; and (5) mechanistic investigations of physiological and molecular bases of stability phenotypes.

Practical Breeding Implications

Findings from this investigation carry significant implications for practical rice improvement programs. First, identification of climate-resilient germplasm (G5, G8, G12) provides directly usable breeding materials for formal variety development pipelines (Ceccarelli *et al.*, 2024) ^[8] (Singh *et al.*, 2023) ^[16]. Rapid advancement of these elite genotypes through expedited multiplication and regulatory assessment procedures is recommended for timely availability to farming communities. Second, recognition of distinct mega-environments and genotype-specific performance patterns suggests that single-varietal recommendation strategies should transition toward more nuanced, environment-stratified cultivar deployment approaches. Extension systems should emphasize that G5 represents optimal choice for favorable, resource-rich environments, while G12 offers superior reliability in marginal, stress-prone regions (Simmonds, 2023) ^[22] (Yan *et al.*, 2022) ^[9]. Third, GGE biplot analytical framework demonstrated exceptional utility for communicating complex stability information to breeding team members, research administrators, and extension specialists. Accessibility of visual biplot presentations, compared to abstract statistical measures, facilitates broader engagement with stability concepts and evidence-based cultivar selection. Fourth, successful integration of conventional stability statistics with contemporary biplot visualization suggests that future breeding program efficiency improvements may derive from holistic adoption of multivariate assessment approaches rather than reliance upon limited univariate parameters.

In context of global climate change and food security imperatives, development of climate-resilient rice cultivars remains a critical research priority. The present investigation demonstrates that systematic, multi-location evaluation integrated with advanced statistical methodologies effectively identifies germplasm combining high productivity with enhanced environmental adaptability and yield stability. Scaling of such research methodologies across broader geographical regions and diverse germplasm collections will progressively enhance cultivar recommendations and support development of climate-adaptable rice production systems essential for maintaining food security amid global environmental change.

Conclusion

The investigation involved 18 genotypes of *Oryza sativa* L. grown in six locations in three crop seasons. GGE biplot analysis and different stability parameters are used to assess interactions of genotypes with the environment and to find climate-resilient varieties. The main results of the study are: (1) the presence of substantial G×E interactions is proved with 42.3% of the total phenotypic variance in grain yield sources; (2) identification of two mega-environments characterized by different genotype ranking systems and distinct properties; (3) identification of highly adaptable and stable genotypes having high average performance, near 1.0 regression coefficient values, and low

deviation; (4) the presence of positive correlations between stability parameters and traits providing plants with climate adaptability; (5) application of GGE biplot framework in conjunction with standard stability analysis leads to better identification of varieties that can be used in breeding programs. Genotypes G5 and G8 represent superior genetic resources that should be rapidly transferred to formal release and farmer deployment programs owing to their positive performance in various aspects including those related to stability, yield capacity, and resilience to climate change. What's more, these genotypes show that it is possible to combine high yield with high adaptability to the environment which is a consequence of recent achievements in breeding and climate-smart agriculture. Genotype G12 might be characterized by the average yield; however, it is reliable in terms of yield and performance in stress and hence is important in stressful agroecosystems. It is advisable to register and produce seeds of the elite genotypes and carry out their deployment in a way that is going to reflect their performance in specific environments.

It is strongly recommended to apply GGE biplot analysis in standard rice breeding program evaluation procedures. This visualization method significantly improves the insight regarding complicated trial data gained from different locations and simplifies the process of sharing the important information with various stakeholders and obtaining information, which can be applied in making decisions about the use of cultivars. The impact of climate change calls for the intensive use of comprehensive evaluation in which multi-environment assessment is supported by the modern analytical methods rather than the traditional single-location tests. The study creates a solid methodological basis and provides evidence for developing climate-resistant varieties of rice that will improve global food security and sustainable agriculture development amidst climate change.

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