



Assessment of Genetic Parameters and Multi-trait Selection for Morpho-physiological Characteristics in Drought-tolerant Maize

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10.18805/IJARE.AF-1067

ABSTRACT

Background: Drought tolerance in maize is essential for stabilizing yields amid increasingly erratic rainfall patterns. This study aimed to estimate the heritability of agronomic and morpho-physiological traits, predict genetic gain under water stress and identify top-performing hybrids across varying drought intensities.

Methods: The study evaluated 35 maize hybrids under drought at ICERI (Inceptisol) using an RCBD with three replications, applying 350 kg/ha NPK + 150 kg/ha urea at 10 DAP and 200 kg/ha urea at 30 DAP, with drought imposed by withholding irrigation from 40 to 80 DAP (CIMMYT protocol). 15 traits were measured and ANOVA, genotypic/phenotypic variance, broad-sense heritability, genetic advance and MGIDI index.

Result: ANOVA revealed significant genotypic differences ($p < 0.05$) for all traits, with leaf area ($h^2 = 0.60$), SPAD ($h^2 = 0.57$) and grain yield ($h^2 = 0.57$) showing high heritability and genetic advance (GA% up to 49.6%). Grain yield correlated strongly positively with ear length ($r = 0.74$), kernels/row ($r = 0.72$) and 1000 kernel weight ($r = 0.61$), but negatively with leaf angle ($r = -0.34$) and days to tasseling ($r = -0.37$). The MGIDI identified genotypes Gen 30, Gen 12 and Gen 6 with balanced drought performance.

Key words: Heritability, Maize, MGIDI, Morpho-physiology.

INTRODUCTION

Maize (*Zea mays* L.) provides essential nutrition to over a billion people, accounting for nearly 30% of caloric intake in sub-Saharan Africa and Latin America (FAO, 2022) and also serves as livestock feed and industrial raw material. However, climate change has increased drought frequency and intensity (IPCC, 2021), reducing maize yields by 30-40% in rainfed systems and disproportionately affecting small-scale farmers (Cairns *et al.*, 2013). Breeding for drought tolerance has advanced through phenotypic selection and molecular tools such as QTL mapping and GWAS (Tuberosa and Salvi, 2006); the drought tolerant maize for Africa project has produced hybrids yielding 25-30% more under drought stress (Bänziger *et al.*, 2000). Nevertheless, progress is hindered by the polygenic nature of drought tolerance, involving complex interactions among genetic, physiological and environmental factors (Edmeades, 2008).

Key agronomic and morpho-physiological traits support maize adaptation to drought. Root architecture, for instance, plays a pivotal role in water acquisition, with deeper root systems enhancing access to subsoil moisture (Comas *et al.*, 2013). However, the heritability of these traits varies significantly across environments. Traits like the anthesis-silking interval, which exhibits moderate heritability, have proven reliable for selection, whereas others, such as root depth, show context-dependent expression (Bolaños and Edmeades, 1996).

Among recent advances in selection methodologies, the multi-trait genotype-ideotype distance index (MGIDI) is a powerful tool for identifying superior genotypes based

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How to cite this article: Suwardi, Azrai, M., Riadi, M., Aqil, M., Rafiuddin, Efendi, R. and Laurence, R. (2026). Assessment of Genetic Parameters and Multi-trait Selection for Morpho-physiological Characteristics in Drought-tolerant Maize. *Indian Journal of Agricultural Research*. **60(10)**: 1490-1497. doi: 10.18805/IJARE.AF-1067.

Submitted: 02-06-2026 **Accepted:** 22-07-2026 **Online:** 17-09-2026

on overall performance across multiple traits (Olivoto and Nardino, 2021). By combining factor analysis with trait rescaling, MGIDI quantifies the distance between each genotype and an ideal ideotype, supporting balanced selection for complex traits like drought tolerance. In maize

breeding, MGIDI can be effectively integrated with genotype $\times$ environment (G $\times$ E) analyses, such as the additive main effects and multiplicative interaction (AMMI) and genotype plus genotype-by-environment (GGE) biplot models, to jointly identify high-yielding, stable and broadly adapted hybrids across diverse environments (Raj *et al.*, 2023). While AMMI and GGE characterize yield stability and environmental adaptation, MGIDI extends the selection framework by simultaneously optimizing multiple agronomic and physiological traits. MGIDI has been widely applied in various crops, including maize (Azrai *et al.*, 2023), rice breeding (Tushar *et al.*, 2025), rice lines with enhanced grain quality (Feizi *et al.*, 2025) and drought tolerant wild wheat accessions at early growth stages (Aboughadareh *et al.*, 2021; Silva *et al.*, 2023).

This study evaluates the heritability and genetic potential of drought-tolerant maize hybrids under water-stressed conditions using a multi-trait approach. The specific objectives are: (1) to estimate the heritability of agronomic and morpho-physiological traits associated with drought adaptation, (2) to predict genetic gains for yield and related traits under limited water availability and (3) to identify high-performing hybrids through multi-trait selection with stable yields across varying drought intensities.

MATERIALS AND METHODS

This study evaluated 35 single-cross maize hybrids under drought conditions at ICERI (119°50'E, -5°31'S) on Inceptisol soils during the July-October 2025 dry season. A randomized complete block design with three replications was used (4-row plots, 5 m length, 70 $\times$ 20 cm spacing). Fertilizer: 350 kg/ha NPK (15:15:15) + 150 kg/ha urea at 10 DAP, plus 200 kg/ha urea at 30 DAP. Drought stress was imposed by suspending irrigation at 40 DAP until the early hard-dough stage (~80 DAP), following CIMMYT protocols (Bänziger *et al.*, 2000). Measured traits included plant height, ear height, stem diameter, leaf area, leaf angle, days to tasseling, SPAD, ear length, ear diameter, kernel rows per ear, kernels per row, 1000-kernel weight, shelling percentage and grain yield.

To assess the genetic performance of the tested hybrids, statistical analysis was performed using analysis of variance (ANOVA). Calculations of genetic variability, heritability, genetic advance and trait correlations were conducted using the RStudio statistical environment. Estimates of genotypic and phenotypic variance components were derived from the ANOVA framework. Genotypic variance (σ^2g) was calculated as:

$$\sigma^2g = \frac{MSg - MSe}{r}$$

Meanwhile, phenotypic variance (σ^2p) was estimated using the expression:

$$\sigma^2p = \sigma^2g + (\sigma^2e/r)$$

Standard deviations for both genotypic and phenotypic variances were computed using the following formulas:

$$SD \text{ of genotypic variance: } SD(\sigma^2g) = \sqrt{[2/r^2 \times [(MSg^2/dfg + 2) + (MSe^2/dfc + 2)]}$$

$$SD \text{ of phenotypic variance: } SD(\sigma^2p) = \sqrt{[2/r^2 \times [(MSg^2/dfg + 2)]}$$

Broad-sense heritability (H^2) was estimated following the method of Johnson *et al.* (1955), while genetic advance (GA) was computed using Lush's (1940) equation:

$$H^2 (\%) = \frac{\sigma^2g}{\sigma^2p} \times 100$$

$$GA = H^2 \times S$$

$$\% GA = \frac{GA}{\bar{x}} \times 100$$

S= Selection differential.

x= The population mean.

Heritability estimates were categorized as high ($H^2 > 0.5$), moderate ($0.2 < H^2 < 0.5$) and low ($H^2 < 0.2$). Similarly, genetic advance was classified as low (0-10%), moderate (10-20%) and high (>20%).

To identify superior genotypes under drought conditions, the MGIDI index was used as an integrative selection approach. Grain yield was assigned a double weight due to its critical importance under water-limited environments, while all other traits were weighted equally. To manage trait intercorrelations and simplify data interpretation, factor analysis (FA) was applied as follow:

$$F = Z (A^T R^{-1})^T$$

Genotype scores were calculated using rescaled trait values, factor loadings and the relationships between traits. An ideal genotype (ideotype) was then defined based on the best possible performance for all traits. The MGIDI for each genotype was measured as the euclidean distance between its scores and the ideotype, showing how close each genotype was to the ideal.

$$MGIDI = \sum_{j=1}^t [(\gamma_{ij} - \gamma_j)^2]^{0.5}$$

RESULTS AND DISCUSSION

Analysis of variance

ANOVA revealed significant genotypic differences ($p < 0.05$) for all traits, indicating substantial genetic variability among the evaluated genotypes that provides a basis for selection in drought tolerance breeding (McMillen *et al.*, 2022) (Table 1). Plant height and ear height showed significant genotypic effects ($F = 1.996$, $p = 0.008$; $F = 1.902$, $p = 0.012$), while height-to-ear ratio was highly significant ($F = 5.003$, $p < 0.001$), reflecting structural stability for stress resilience (Li *et al.*, 2015). Stem diameter and leaf area differed significantly ($F = 5.456$ for leaf area), suggesting improved light capture and canopy plasticity (Tardieu *et al.*, 2018). SPAD values varied markedly ($F = 4.976$), indicating chlorophyll retention during grain filling (Li *et al.*, 2022). Days to tasseling ($F = 3.635$) and leaf angle ($F = 2.694$) showed variability in developmental timing and canopy architecture for drought escape (Vadez *et al.*, 2025).

Yield traits-ear length, kernel rows, 1000-kernel weight- showed pronounced differences ($p < 0.01$) and grain yield was highly variable ($F = 5.039$, $p < 0.001$), linking drought tolerance to productivity (Cairns *et al.*, 2013).

Performance of agronomic and yield hybrids

Significant genetic variation was observed among 35 genotypes for PH, SD, area, LA and SPAD (Fig 1). Plant height ranged from 152.07 cm (Gen-12) to 203.27 cm

Table 1: Analysis of variance and 15 maize traits.

Trait	Sum Sq.	Mean Sq.		F value	Pr (>F)
		Genotype	Error		
Plant height (cm)	15443.616	454.224	227.581	1.996	0.008
Ear height (cm)	9237.442	271.689	142.847	1.902	0.012
Height to ear ratio	0.298	0.009	0.002	5.003	0.000
Stem diameter (cm)	187.435	5.513	1.760	3.131	0.000
Leaf area (cm ²)	328158.381	9651.717	1768.938	5.456	0.000
Days to tasseling	191.048	5.619	1.546	3.635	0.000
Leaf angle (degrees)	1123.178	33.035	12.261	2.694	0.000
SPAD value	56.842	1.672	0.336	4.976	0.000
Ear length (cm)	153.314	4.509	1.368	3.296	0.000
Ear diameter (cm)	917.600	26.988	12.038	2.242	0.002
Number of rows per ear	75.685	2.226	0.808	2.754	0.000
Number of kernels per row	1435.446	42.219	11.945	3.535	0.000
1000-Kernel weight (g)	107745.735	3168.992	1148.873	2.758	0.000
Shelling percentage	0.142	0.004	0.002	2.773	0.000
Grain yield (ton/ha)	67.950	1.999	0.397	5.039	0.000

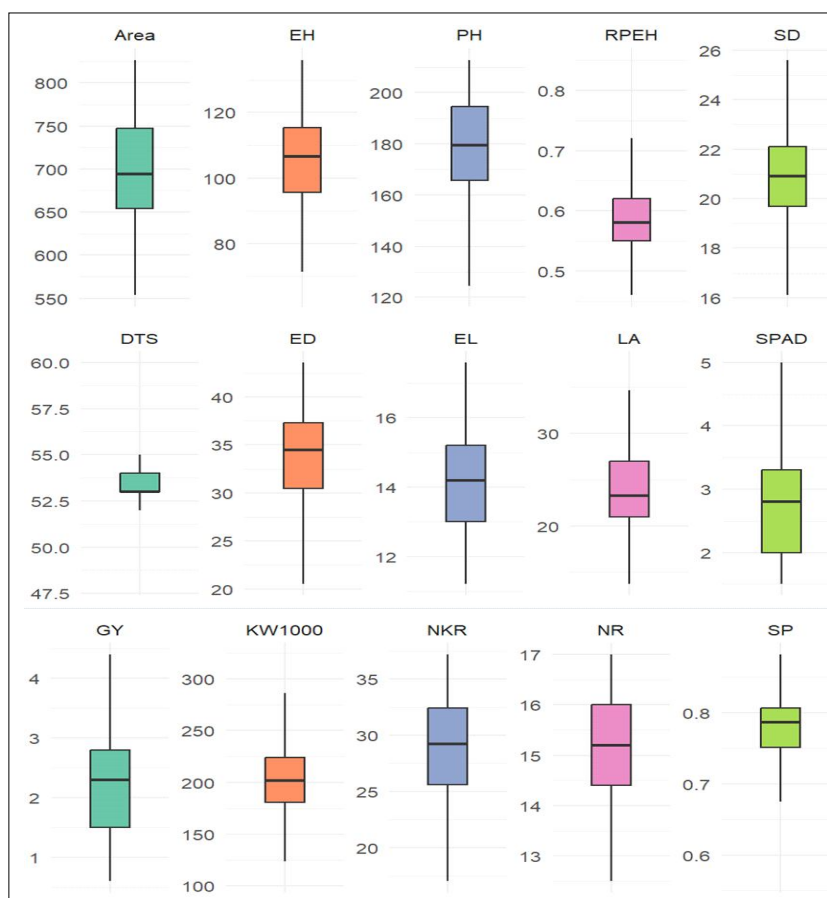


Fig 1: Boxplot of several agronomic traits and yield components from various maize genotypes.

(Gen-4), reflecting differences in vegetative growth strategies (Cooper *et al.*, 2014). Stem diameter varied from 18.13 mm (Gen-5) to 25.27 mm (Gen-21), indicating substantial variation in stalk morphology among genotypes. Greater stem diameter is generally associated with stronger stalks and improved resistance to stalk lodging in maize (Xue *et al.*, 2020). Leaf area spanned 576.80 cm² (Gen-18) to 793.03 cm² (Gen-31), enhancing photosynthesis but potentially increasing water loss. Leaf angle ranged from 18.23° (Gen-33) to 30.67° (Gen-22); upright leaves improve light penetration and drought avoidance (Dzievitz *et al.*, 2023). SPAD values ranged from 1.80 (Gen-21) to 5.00 (Gen-32), indicating chlorophyll content and photosynthetic efficiency under stress (Gao *et al.*, 2024). Boxplots are shown in Fig 2.

The yield components varied markedly among the 35 genotypes. Ear length ranged from about 11.9 cm to

16.8 cm, with Gen-33 showing the longest ears and Gen-34 the shortest. Ear diameter followed a similar pattern, from 28.5 mm up to 40.3 mm, again with Gen-33 at the top. Kernel weight showed an even wider spread, from roughly 136 g in Gen-35 to over 305 g in Gen-33. Shelling percentage, which reflects the proportion of grain harvested, varied from around 68% in Gen-11 to nearly 84% in Gen-16. Grain yield also differed greatly, from under 1 t/ha in Gen-26 and Gen-32 to almost 4 t/ha in Gen-4 and Gen-33.

Gen-33 shows good performance in ear length, diameter and kernel weight, making it a high-yield candidate. Gen-4 combines heavy kernels with top grain yield, another promising line. Gen-16's exceptional shelling percentage. For overall yield improvement, Gen-33 and Gen-4 show consistent multi-trait superiority.

Genetic variability and heritability

Variance component analysis indicated distinct genetic architectures among traits (Table 2). Leaf area, 1,000-kernel weight and leaf angle exhibited high genetic variance proportions (60%, 37% and 36%, respectively), indicating strong potential for rapid improvement through selection. Leaf area (genetic variance= 2,628 vs. phenotypic variance= 4,396) and 1,000-kernel weight (673 vs. 1,822) are predominantly under additive genetic control, aligning with findings that such traits drive heritable gains in stress-resilient maize (Beyene *et al.*, 2015). Conversely, height-to-ear ratio and shelling percentage showed negligible genetic variance (0.001-0.002).

Most traits showed low to moderate heritability ($h^2 = 0.23-0.60$) and varied genetic advance, indicating different potentials for selection. Leaf area ($h^2 = 0.60$) and grain yield ($h^2 = 0.57$) combined high heritability with large genetic advance as a percentage of the mean (GA% = 11.8% and 49.6%, respectively). SPAD value also had high heritability (0.57) and very high GA% (36.3%), making it another excellent indirect selection trait. Traits like height-

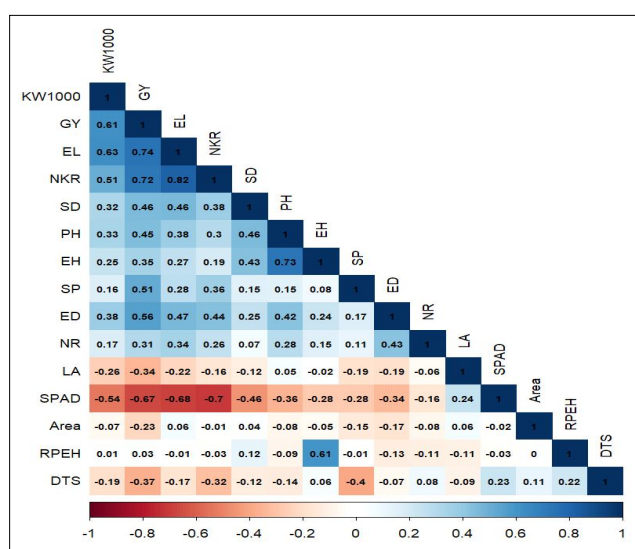


Fig 2: Correlation heatmap among traits.

Table 2: Variance components and standard deviations for evaluated traits.

Trait	σ^2_g	σ^2_p	σ^2_e	$\sigma_{\sigma_{2g}}$	$\sigma_{\sigma_{2p}}$
Plant height (cm)	75.55	303.13	227.581	8.692	17.411
Ear height (cm)	42.95	185.8	142.847	6.553	13.631
Height to ear ratio	0.002	0.004	0.002	0.048	0.064
Stem diameter (cm)	1.251	3.011	1.76	1.118	1.735
Leaf area (cm ²)	2628	4396.5	1768.938	51.26	66.306
Days to tasseling	1.358	2.904	1.546	1.165	1.704
Leaf angle (degrees)	6.924	19.186	12.261	2.631	4.38
SPAD value	0.445	0.781	0.336	0.667	0.884
Ear length (cm)	1.047	2.415	1.368	1.023	1.554
Ear diameter (cm)	4.983	17.022	12.038	2.232	4.126
Number of rows per ear	0.473	1.281	0.808	0.687	1.132
Number of kernels per row	10.09	22.036	11.945	3.177	4.694
1000-Kernel weight (g)	673.4	1822.2	1148.873	25.949	42.688
Shelling percentage	0.001	0.002	0.002	0.03	0.049
Grain yield (ton/ha)	0.534	0.931	0.397	0.731	0.965

to-ear ratio ($h^2 = 0.57$, $GA\% = 12.8\%$), number of kernels per row ($h^2 = 0.46$, $GA\% = 15.4\%$) and 1,000-kernel weight ($h^2 = 0.37$, $GA\% = 15.8\%$) showed moderate heritability with respectable genetic gains, so they can be improved through selection (Table 3). In contrast, plant height and ear height had low heritability ($h^2 = 0.25$ and 0.23) and low $GA\%$ (5-6%).

Correlation

Correlation analysis indicated that grain yield had the strongest positive associations with ear length ($r = 0.74$), number of kernels per row ($r = 0.72$) and ear diameter ($r = 0.56$), suggesting that longer ears with more densely arranged kernels contribute significantly to yield (Fig 2). Thousand-kernel weight also showed a strong positive correlation with yield ($r = 0.61$), while shelling percentage ($r = 0.51$), stem diameter ($r = 0.46$) and plant height

($r = 0.45$) exerted moderate positive effects. In contrast, grain yield was moderately negatively correlated with leaf angle ($r = -0.34$), chlorophyll index ($r = -0.67$) and days to tasseling ($r = -0.37$), indicating that genotypes with narrower leaf angles, lower chlorophyll content and earlier flowering tend to allocate more resources toward grain production.

Principal component analysis and genetic contribution

The principal component analysis (PCA) showed that the first three principal components (PCs) captured a substantial portion of the total variance among the evaluated traits, explaining 41.6%, 13.8% and 12.2% of the variation, respectively, with a cumulative total of 67.6% (Fig 3). This indicates that a large proportion of the multivariate variability in the dataset can be effectively captured using only these three components, which is advantageous for dimensionality reduction and trait interpretation. PC1 contributed

Table 3: Grand mean, heritability, genetic advances of observed character.

Trait	Grand mean	Heritability	Genetic advance	GA (%)
Plant height (cm)	177.78	0.249	8.939	5.03
Ear height (cm)	104.79	0.231	6.491	6.19
Height to ear ratio	0.59	0.572	0.075	12.76
Stem diameter (cm)	20.92	0.415	1.485	7.09
Leaf area (cm ²)	693.41	0.598	81.63	11.77
Days to tasseling	53.10	0.468	1.641	3.09
Leaf angle (degrees)	24.24	0.361	3.257	13.44
SPAD value	2.86	0.57	1.038	36.31
Ear length (cm)	14.23	0.434	1.388	9.75
Ear diameter (cm)	34.17	0.293	2.488	7.28
Number of rows per ear	15.16	0.369	0.86	5.67
Number of kernels per row	28.86	0.458	4.428	15.35
1000-Kernel weight (g)	205.38	0.37	32.5	15.82
Shelling percentage	0.78	0.371	0.037	4.82
Grain yield (ton/ha)	2.30	0.574	1.14	49.60

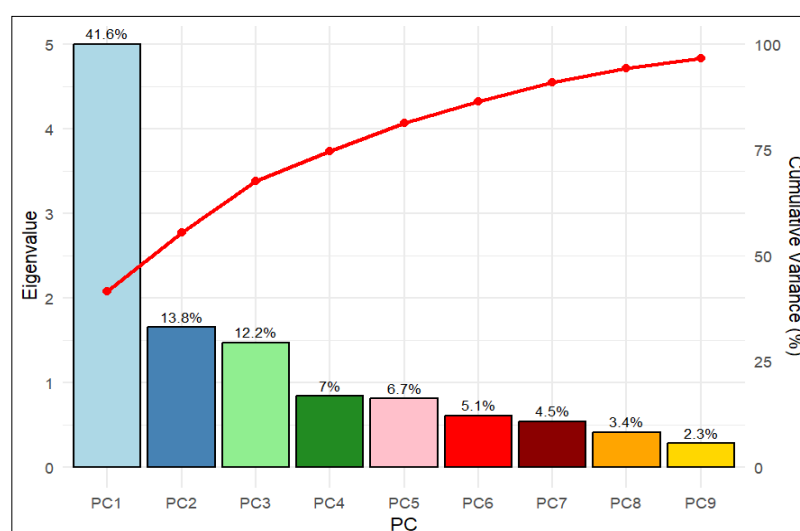


Fig 3: Principal component analysis of agronomic traits in maize under drought conditions.

the most to the observed variance, suggesting it encapsulates key patterns or trait groupings strongly influenced by the underlying genotype or environmental structure. Beyond PC3, each additional component contributed progressively less to the overall variance, with PCs 4 through 9 together accounting for only 29% of the total variability.

In addition, the stacked bar chart indicating the proportional contributions of genetic and residual components to the phenotypic variance observed in agronomic traits of maize genotypes subjected to drought conditions is shown in Fig 4. Traits such as plant height (PH), leaf area (LA) and number of roots (NR) exhibit a pronounced genetic contribution to their phenotypic variance, suggesting a strong heritable basis and indicating that these traits are amenable to effective selection within breeding programmes aimed at genetic improvement. The high genetic proportion for these traits underscores their potential as reliable targets for enhancing drought resilience through breeding strategies. In contrast,

traits like ear diameter (ED) and SPAD chlorophyll content demonstrate a greater influence from residual components, indicating a higher degree of environmental sensitivity or potential variability in measurement. For these traits, non-genetic factors play a more significant role in shaping their expression, which may present greater challenges for genetic improvement efforts.

Factor loading

Varimax rotation of twelve agronomic traits under drought identified three factors (Table 4), explaining most variation (mean 0.68; communalities 0.50-0.92; uniqueness 0.08-0.50). Factor 1 (structural-physiological) showed negative loadings for stem diameter, kernel weight, ear length and grain yield, but positive for SPAD, indicating a photosynthesis vs. yield-structure trade-off. Factor 2 (morphology/development) had negative loadings for plant height and leaf area and positive for ear diameter and row number. Factor 3 (reproductive efficiency) had positive loadings for days to silking and shelling percentage, which showed minimal cross-loading. Grain yield's high communality (0.92) confirms its integrative nature across multiple trait domains.

A multi-trait genotype-ideotype distance index (MGIDI)

A multi-trait genotype-ideotype distance index was employed to identify superior maize genotypes under drought stress, incorporating multiple morphology and physiology traits into a single selection criterion. The circular radial plot illustrates the ranking of 36 genotypes based on their proximity to the ideotype (ideal genotype), with lower MGIDI values indicating genotypes with more desirable trait combinations (Fig 5). From the analysis, ten genotypes (Gen 30, Gen 12, Gen 21, Gen 9, Gen 15, Gen 13, Gen 6, Gen 10, 27 and Gen 29) were identified as superior and thus selected for further consideration. These genotypes exhibited the smallest distances from the ideotype, suggesting a more favorable balance of traits under drought conditions. They are visually represented in red, distinctly separated from the remaining genotypes (non-selected), which possess comparatively higher MGIDI values.

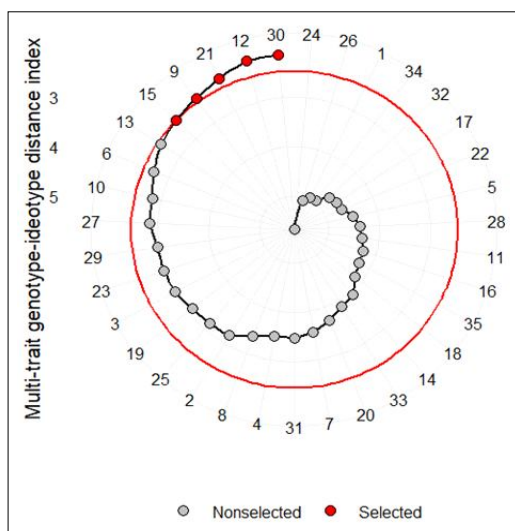


Fig 4: Selected genotypes based on MGIDI selection under drought conditions.

Table 4: Factor loadings and shared variances from the analysis of factor.

Traits	FA1	FA2	FA3	Communality	Uniqueness's
PH	0.32	-0.64	-0.06	0.51	0.49
DTS	-0.08	-0.18	0.88	0.82	0.18
LA	-0.65	0.27	-0.02	0.5	0.5
SD	-0.78	-0.05	0.08	0.62	0.38
Area	-0.14	-0.64	-0.44	0.62	0.38
SPAD	0.87	-0.02	-0.16	0.79	0.21
EL	-0.87	0.14	0.11	0.78	0.22
ED	-0.34	0.69	0.06	0.59	0.41
NR	-0.02	0.72	-0.21	0.56	0.44
KW1000	-0.76	0.28	0.16	0.68	0.32
SP	-0.31	0.18	0.76	0.71	0.29
GY	-0.73	0.37	0.51	0.92	0.08

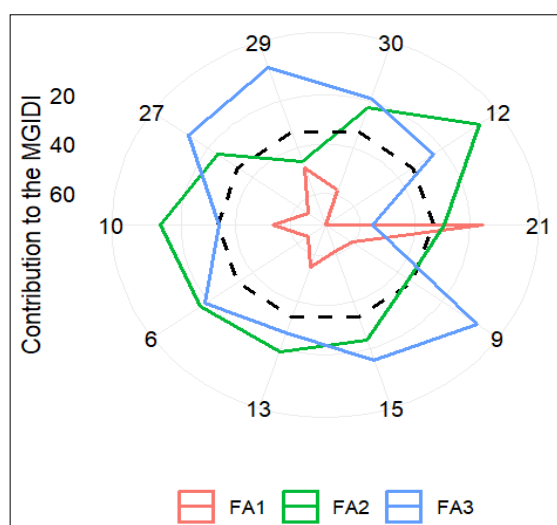


Fig 5: Strength and weakness of the selected genotypes based on MGIDI under drought conditions.

The radar plot illustrates the contribution of three latent factors (FA1, FA2 and FA3) to the MGIDI for the ten genotypes selected under drought conditions (Fig 5). Each colored line represents a different factor: FA1 in red, FA2 in green and FA3 in blue, while the black dashed circle serves as a reference threshold for average contribution. Because grain yield in maize is a complex quantitative trait governed by multiple interrelated agronomic characters, evaluating the relative contribution of different latent factors provides a comprehensive basis for identifying superior genotypes (Lal *et al.*, 2025). Consequently, the factor-specific contributions shown in the radar plot reveal the trait groups that most strongly influenced the selection of drought-tolerant genotypes.

Under drought, genotypes 30 and 12 showed strong performance across all trait groups (low FA1, FA2, FA3 contributions), aligning well with the ideotype for drought resilience, grain yield and phenological stability. Gen 6, 10 and 13 also had favorable balance with moderate contributions across factors, indicating solid multi-trait adaptability. Gen 15 and 27 showed strengths in FA1 and FA2 (physiological/morphological responses). Conversely, genotype 21 had a pronounced FA1 contribution (weakness in yield or stress efficiency). Genotypes 9 and 29 showed elevated FA3 contributions (less stable in reproductive/structural traits), though compensated by other trait groups. The MGIDI radar plot effectively differentiated performance, supporting genotypes 30, 12 and 6 as potential parents for drought-prone breeding.

CONCLUSION

Evaluation of 35 maize genotypes under drought revealed high genetic variation. Leaf area, chlorophyll index and thousand-kernel weight showed strong genetic control. Grain yield correlated positively with ear length, kernel number, ear diameter, kernel weight, shelling percentage and stem diameter; negatively with leaf angle, chlorophyll

index and days to tasseling. MGIDI identified ten superior genotypes (Gen 30, 12, 21, 9, 15, 13, 6, 10, 27, 29) with balanced drought performance.

ACKNOWLEDGEMENT

The authors thank Department of Agronomy, Faculty of Agriculture, Hasanuddin University for supporting and providing the necessary facilities.

Disclaimers

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Conflict of interest

The authors declare that there are no conflicts of interest regarding the publication of this article. No funding or sponsorship influenced the design of the study, data collection, analysis, decision to publish, or preparation of the manuscript.

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