



Evaluation of Morphological and Biochemical Traits for Salt Tolerance in Sorghum Genotypes

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(Received: 17 April 2025; Revised: 31 May 2025; Accepted: 27 June 2025; Published online: 15 July 2025)

(Published by Research Trend)

ABSTRACT: Sorghum (*Sorghum bicolor* (L.) Moench, $2n = 2x = 20$) is one of the most important cereal crops globally, recognized for its exceptional adaptability to harsh environments, nutritional richness, and economic relevance, especially in arid and semi-arid regions. The present investigation was carried out to assess the genetic variability, heritability, and trait associations among 50 sorghum genotypes under saline soil conditions. Twelve quantitative traits were evaluated, revealing significant genotypic differences for all traits studied. High genotypic and phenotypic coefficients of variation (GCV and PCV) were recorded for panicle weight, grain yield per plant, biological yield, harvest index, and flag leaf Na^+/K^+ ratio, indicating considerable genetic diversity and a low environmental influence. High heritability coupled with high genetic advance for these traits suggests the predominance of additive gene action, making them amenable to improvement through phenotypic selection. Principal component analysis (PCA) and biplot visualization further aided in identifying key traits contributing to genetic divergence and helped classify salt-tolerant genotypes. Genotypes such as SGP-GS-36, BP-53, and SGP-GS-74 were identified as superior performers under saline conditions. The study provides valuable insights for developing salt-tolerant sorghum varieties suitable for cultivation in the salt-affected regions of Gujarat and beyond.

Keywords: Sorghum, Salt, Variability, Phenotypic coefficient of variation, Heritability, Genetic advance.

INTRODUCTION

Sorghum (*Sorghum bicolor* (L.) Moench, $2n = 2x = 20$) is one of the most important cereal crops globally, recognized for its exceptional adaptability to harsh environments, nutritional richness, and economic relevance, especially in arid and semi-arid regions. Belonging to the *Poaceae* family, sorghum is also known as great millet, jowar, milo, and durra. The genus *Sorghum* comprises around 25 species, among which *S. bicolor* is the only widely cultivated species for food, feed, fodder, biofuel, and industrial purposes. *Sorghum bicolor* is an annual or short-lived perennial crop with diverse morphological forms that vary in plant height, panicle structure, grain colour, and utility. Though primarily self-pollinated, it exhibits a natural outcrossing rate of up to 30%, depending on the genotype and environmental conditions, contributing significantly to its genetic variability (Reddy *et al.*, 2006). This diversity forms the basis for crop improvement and stress resilience.

India ranks among the top sorghum-producing countries (Anonymous, 2021), cultivating the crop under both rainfed and irrigated conditions, mainly in Maharashtra,

Karnataka, Andhra Pradesh, and Tamil Nadu. It plays a critical role in food and nutritional security, particularly for resource-poor farmers in marginal ecosystems. Besides its use as a staple food, sorghum is utilised in poultry and livestock feed, alcoholic beverages, and increasingly as a gluten-free grain in health foods and processed products.

Sorghum grains are rich in carbohydrates, protein, dietary fibre, and micronutrients such as iron, zinc, and antioxidants. Some genotypes also contain bioactive compounds like phenolics, tannins, and flavonoids, contributing to therapeutic and functional food applications (Dicko *et al.*, 2006). The crop is also gaining international attention as a climate-resilient alternative to conventional cereals under the increasing threat of climate change.

However, sorghum productivity is often constrained by the use of traditional, low-yielding cultivars and exposure to multiple biotic (e.g., shoot fly, stem borer, grain mold) and abiotic stresses. Among these, salinity is a major abiotic stress that severely affects germination, growth, and yield in salt-affected soils. Therefore, evaluating sorghum germplasm for salinity tolerance is crucial for identifying resilient genotypes.

In this context, the present study was conducted under saline soil conditions to assess the genetic variability and potential of diverse sorghum genotypes for salinity tolerance using key morphological and biochemical traits.

The findings from such evaluations are essential for advancing breeding strategies aimed at developing salt-tolerant, high-yielding sorghum cultivars. Harnessing the inherent genetic diversity of indigenous germplasm holds the key to enhancing crop resilience and securing sustainable sorghum production under challenging agro-ecological conditions.

Recognising the nature and magnitude of genetic variability is fundamental for designing an efficient breeding program. Estimation of parameters such as the genotypic coefficient of variation (GCV) and phenotypic coefficient of variation (PCV) provides insights into the degree of variability present within a set of genotypes. A high GCV value for a trait typically indicates a greater scope for improvement through selection. However, GCV alone does not account for the proportion of variation that is heritable (Singh and Nandpuri 1974). For effective and precise selection, it is essential to focus on the heritable portion of variability (Burton, 1952).

Hence, the evaluation of heritable variation and associated genetic parameters such as GCV, heritability estimates, and genetic advance is vital for the genetic enhancement of complex quantitative traits. Among various measures, the range of variation observed for a given trait serves as a straightforward indicator of genetic diversity within the population under investigation. Additionally, Principal Component Analysis (PCA) can be employed to reduce dimensionality and identify the traits that contribute most to total variation among genotypes. PCA helps in grouping genotypes based on multivariate trait performance and supports the selection of superior and diverse genotypes for breeding programs.

MATERIALS AND METHODS

A. Phenotyping of germplasm

The present experiment was conducted during the *kharif* season of 2024 at the Coastal Soil Salinity Research Station, Navsari Agricultural University, Danti-Ubharat. The experimental field comprised black soil with a saline-alkaline nature, exhibiting an electrical conductivity (EC) ranging from 4 to 6 dS/m. A total of 50 diverse sorghum genotypes were selected for field evaluation and grown under Randomized Complete Block Design (RCBD) with three replications, using the dibbling method for sowing. Each genotype was sown in three rows of 4 meters in length, with a spacing of 45 cm between rows and 15 cm between plants. Standard agronomic practices and recommended plant protection measures were followed to ensure a healthy and uniform crop stand.

For morphological characterization, phenotyping was carried out on five randomly selected, healthy, and competitive plants per genotype in each replication. Data were recorded on 12 quantitative traits, including germination percentage (%), plant height (cm), panicle

length (cm), panicle weight (g), test weight (g), grain yield per plant (g), biological yield per plant (g), harvest index (%), flag leaf length (cm), and flag leaf Na^+/K^+ ratio. Observations for days to 50% flowering and days to maturity were recorded on a plot basis. This setup allowed for accurate assessment of genotypic performance under saline field conditions.

B. Data analysis

Analysis of variance and Least significant different test on phenotypic data of 12 morphological and biochemical characters were executed to identify the diverse genotypes. The genotypic and phenotypic components of variance, variability coefficients, heritability (in the broad sense), genetic advance, and genetic advance as a percentage of mean were examined. According to Sivasubramanian and Menon (1973), phenotypic (PCV) and genotypic coefficients of variation (GCV) were classified [$<10\%$ = low, $10\text{--}20\%$ = moderate and $>20\%$ = high]. Similarly, heritability (broad sense) [$<30\%$ = low, $30\text{--}60\%$ = moderate and $>60\%$ = high] and genetic advance as per cent of mean [$<10\%$ = low, $10\text{--}20\%$ = moderate and $>20\%$ = high] were classified according to Robinson *et al.* (1949); Johnson *et al.* (1955) respectively.

Principal Component Analysis (PCA) was performed to assess the multivariate variation among 50 sorghum genotypes based on 12 morphological and biochemical traits recorded under saline conditions. Standardized data were subjected to PCA using 'R' software to identify key traits contributing to overall diversity. Eigenvalues, factor loadings, and the percentage of variation explained by each principal component were computed. A biplot was generated to visualise genotype distribution and trait associations. Traits with high loadings on major components were considered influential in genotype discrimination.

RESULTS AND DISCUSSION

A. Mean and range

Mean is a relatively simple measure used in plant breeding to assess phenotypic variability and it serves as the basis for screening desirable genotypes. The mean values of 50 genotypes of sorghum for all the 12 quantitative characters, along with standard error of mean (S.E.m \pm), critical difference (C.D) and coefficient of variation (C.V%) are given in Table 1. Earliness in sorghum helps to escape from salinity stresses occurring during the crop period and hence days to 50% flowering and days to maturity are appropriate selection standards to identify early maturing and salt-tolerant genotypes. In current study, SGP-GS-11 (62.67 days) followed by SGP-GS-46 (63.00 days) and SGP-GS-56 (63.33 days) were matured early while, Gundari (88.00 days) followed by CSV 46 F (84.00 days) and GFS-6 (80.33 days) was detected as late-maturing genotype.

Sorghum is valued both as a grain and fodder crop; therefore, higher values for grain yield per plant and biological yield per plant (g) are desirable, as they directly contribute to overall productivity. In the present study, twenty genotypes exhibited significantly higher grain and biological yields compared to the population mean, indicating their superior performance. Among

the evaluated traits, flag leaf length was notably higher in genotype BP-53, followed by SGP-GS-247 and SGP-GS-146. Flag leaf length is a critical trait for assessing salt injury at the reproductive stage. Additionally, the flag leaf Na⁺/K⁺ ratio proved to be a useful indicator for distinguishing salt-tolerant genotypes. Genotypes such as SGP-GS-36 (0.51), BP-53, SGP-GS-74 (0.55), and SGP-GS-101 (0.61) demonstrated lower Na⁺ accumulation in the flag leaf, suggesting their enhanced ability to maintain ionic balance under saline conditions.

The diversity was observed among sorghum genotypes for most traits, with many genotypes performing well

under saline conditions. This diversity enables the identification of promising genotypes, which can be utilized in developing salt-tolerant varieties suitable for the salt-affected soils of Gujarat and other regions of the country.

The Analysis of variance for studied quantitative traits depicted highly significant ($P < 0.01$) differences among the genotypes (Table 1) in the overall result of F tests which indicated the existence of ample variability in genotypes which can be exploited for chilli improvement.

Table 1: Mean performance, analysis of variance (ANOVA) and estimation of genetic variables for 12 characters of sorghum.

Trait	Mean performance					Source of variation and mean squares (ANOVA)		
	Mean	Range	S. Em.	C.D. at 5%	CV%	Replication (df = 02)	Genotypes (df = 49)	Error (df = 98)
Germination percentage	78.41	50.0-94.0	1.33	3.73	2.94	0.45	250.44**	5.30
Days to flowering	70.41	58.0-90.0	1.59	4.47	3.92	2.69	107.38**	7.62
Days to maturity	119.96	108.0-138.0	1.60	4.50	2.31	7.46	92.95**	7.70
Plant height (cm)	135.26	80.0-187.0	3.30	9.26	4.23	20.58	999.01**	32.70
Panicle length (cm)	21.77	11.0-35.0	1.06	2.97	8.42	0.69	50.33**	3.36
Panicle weight (g)	42.51	8.0-162.0	1.56	4.37	6.34	50.11**	2479.80**	7.28
Test weight (g)	22.97	12.0-34.0	1.07	2.99	8.05	2.75	43.39**	3.42
Grain yield per plant (g)	34.61	10.0-120.0	1.36	3.80	6.78	0.29	1568.55**	5.51
Biological yield per plant (g)	154.16	42.0-383.0	3.93	11.03	4.42	26.40	17883.6**	46.4
Harvest index (%)	22.78	8.82-53.12	1.41	3.95	10.71	5.63	220.90**	5.95
Flag leaf length (cm)	29.31	16.0-42.0	1.57	4.40	9.26	2.10	78.92**	7.37
Flag leaf Na ⁺ /K ⁺ ratio	1.17	0.45-2.54	0.07	0.21	10.86	0.01	0.66**	0.01

B. Genetic variability, heritability and genetic advance

Sorghum, being an often cross pollinated and inbreeding species, requires sufficient genetic variation for the improvement of quantitative traits. While analysis of variance (ANOVA) provides a measure of differences among genotypes, it may not fully reveal the extent of variability within the population. To better assess this, the phenotypic and genotypic variances are standardized using coefficients of variability.

In line with previous studies, higher values of phenotypic coefficient of variation (PCV) compared to genotypic coefficient of variation (GCV) were observed for all the traits studied (Sharma *et al.*, 2006; Berhanu and Abraha 2020; Deshmukh and Waghmare 2024;

Haile *et al.*, 2024). However, the differences between PCV and GCV were minimal (Table 2), indicating a limited environmental influence on trait expression. High GCV estimates (>20%) were recorded for traits such as panicle weight (g), grain yield per plant (g), biological yield per plant, harvest index, and flag leaf Na⁺/K⁺ ratio. Similar findings were also reported by Berhanu and Abraha (2020); Deshmukh and Waghmare (2024). The high GCV values, particularly for yield-related traits, and their closeness to corresponding PCV values suggest the presence of substantial genetic variability with minimal environmental effect, making these traits promising for selection and genetic improvement.

Table 2: Variability parameters for 12 characters of sorghum under saline conditions.

Characters	σ^2_g	σ^2_p	GCV (%)	PCV (%)	H^2_b (%)	GA % Mean
Germination percentage	81.71	87.02	11.53	11.90	93.90	23.01
Days to 50 % flowering	33.25	40.88	8.19	9.08	81.35	15.22
Days to 50 % maturity	28.42	36.11	4.44	5.01	78.68	8.12
Plant height (cm)	322.10	354.80	13.27	13.93	90.78	26.04
Panicle length (cm)	15.66	19.02	18.18	20.03	82.33	33.98
Panicle weight (g)	824.17	831.45	67.53	67.82	99.12	38.97
Test weight (g)	13.32	16.74	15.89	17.82	79.57	29.21
Grain yield per plant (g)	521.01	526.52	65.94	66.29	98.95	35.13
Biological yield per plant (g)	5945.73	5992.09	50.01	50.21	99.23	20.56
Harvest index (%)	71.65	77.60	37.16	38.67	92.33	28.31
Flag leaf length (cm)	23.85	31.22	16.66	19.06	76.39	30.00
Flag leaf Na ⁺ /K ⁺ ratio	0.22	0.23	39.75	41.21	93.08	29.60

The results indicate that phenotypic selection for the evaluated traits could be effective and beneficial for sorghum improvement programs. However, traits such as days to 50% flowering and days to 50% maturity exhibited low genotypic and phenotypic coefficients of variation (<10%), suggesting limited genetic variability and, consequently, a lower potential for improvement through selection. Similar findings were reported by Dhutmal *et al.* (2020); Haile *et al.* (2024), who also observed low GCV and PCV for these traits, indicating their relatively stable expression across genotypes.

Salinity disrupts the normal development of plants by causing the accumulation of salts in various plant parts. Saline soils adversely affect plant growth from the germination stage through to the reproductive phase, ultimately leading to a significant reduction in yield. In the present study, high values of genotypic and phenotypic coefficients of variation (GCV and PCV) were recorded for the flag leaf Na⁺/K⁺ ratio, indicating a substantial degree of variability among genotypes. This suggests that several genotypes possess the ability to maintain a lower Na⁺/K⁺ ratio, which supports normal panicle development and helps sustain both grain and biological yield under saline conditions.

Though, many advance genomic tools are available with breeder to deploy in sorghum breeding programs. But without knowledge of heritability and genetic gain selection program is ineffective. Heritability evident the inheritance of traits to filials from parents (Falconer, 1960).

Understanding heritability and genetic advance as a percentage of the mean is crucial for predicting the potential genetic gain through selection in any breeding program. According to the classification by Robinson *et al.* (1949), all the quantitative traits evaluated in the present study exhibited high heritability, with broad-sense heritability (H²) values exceeding 60% (Table 2). High heritability estimates for traits such as days to 50% flowering, days to 50% maturity, plant height, panicle length, test weight, grain yield per plant, biological yield per plant, and harvest index have also been reported by several researchers, including Subudhi *et al.* (2008); Reddy *et al.* (2015); Wagh *et al.* (2023). Higher value of heritability suggested that the environment has little influence on characters expression.

Genetic advance as a percentage of the mean (GAM) ranged from 8.12% for days to 50% maturity to 38.97% for panicle weight in the present study. Similar results have been reported in earlier studies, where high heritability was accompanied by high GAM for these traits (Subudhi *et al.*, 2008; Reddy *et al.*, 2015; Wagh *et al.*, 2023). Traits exhibiting both high heritability and high GAM are primarily governed by additive gene action and can be effectively improved through phenotypic selection with minimal effort (Table 3). Moreover, in the context of improving genotypes for salt tolerance, these traits are particularly valuable, as additive gene action plays a significant role in their inheritance and expression.

Table 3: Classification of variability parameters and selection criteria.

Characters	GCV (%)	PCV (%)	H ² _b (%)	GA % Mean	Gene effect	Selection remark
Germination percentage	Moderate	Moderate	High	High	Additive	✓
Days to flowering	Low	Low	High	Moderate	Additive	✓
Days to maturity	Low	Low	High	Moderate	Additive	✓
Plant height (cm)	Moderate	Moderate	High	High	Additive	✓
Panicle length (cm)	Moderate	Moderate	High	High	Additive	✓
Panicle weight (g)	High	High	High	High	Additive	✓
Test weight (g)	Moderate	Moderate	High	High	Additive	✓
Grain yield per plant (g)	High	High	High	High	Additive	✓
Biological yield per plant (g)	High	High	High	High	Additive	✓
Harvest index (%)	High	High	High	High	Additive	✓
Flag leaf length (cm)	Moderate	Moderate	High	High	Additive	✓
Flag leaf Na ⁺ /K ⁺ ratio	High	High	High	High	Additive	✓

C. Principal Component analysis

The number of components extracted in PCA is equal to the number of variables studied. This means that an analysis of twelve characters of sorghum genotypes produces twelve components, but only selective components account for a meaningful amount of total variance, so only these selective components are retained and used in subsequent analysis. The scree test criterion (scree plot) is used for retaining of principal component for the study.

In the present study, eigenvalues greater than one from the scree plot (Fig. 1) were used to retain principal components for analysis. The first two components, which had eigenvalues greater than one, together accounted for 46.10% of the cumulative variability among the 50 sorghum genotypes evaluated for 12

quantitative traits. Individual traits, such as germination percentage, days to flowering, days to maturity, test weight, and flag leaf Na⁺/K⁺ ratio, significantly contributed to Dim1, indicating that these traits have high variability and may precisely discriminate salt-tolerant genotypes in sorghum. Meanwhile, Dim2 highlighted traits such as plant height, panicle length and biological yield per plant. Similar result supported by Calone *et al.* (2020); Rajabi dehnnavi *et al.* (2024).

The PCA biplot visualizes the relationships between different variables (traits) and samples (genotypes). Here, the x-axis represents the first principal component (Dim1), which explains 31.0 % of the variation in the data, while the y-axis represents the second principal component (Dim2), explaining 15.1 % of the variation. Together, these two components capture 46.10 % of the

total variance, indicating a good summary of the dataset.

The variables were superimposed on the plot as vectors in the biplot; each arrow represents a variable (trait). The direction of the arrow indicates the direction of increase for that variable. The length of the arrow indicates the strength of the variable's contribution to the principal components. The relative length of the vector represents the relative proportion of variability. The genotypes that were farthest from the origin displayed more variation and less similarity with other genotypes.

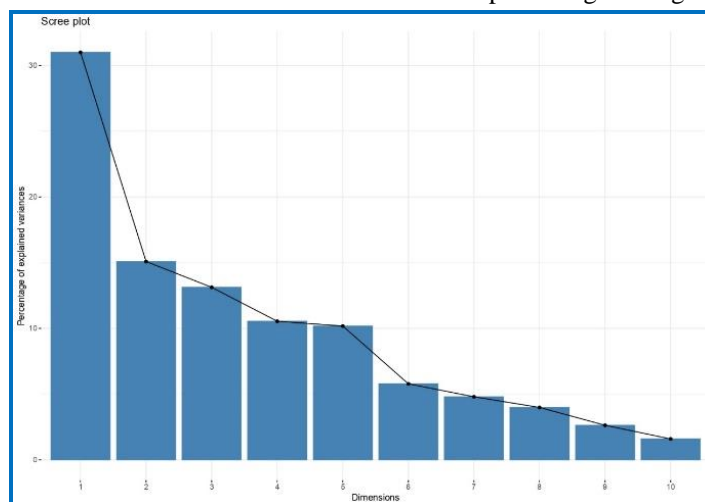


Fig. 1. Scree plot showing contribution of different components to total divergence.

The PCA biplot presents the multivariate relationships among fifty sorghum genotypes and key agronomic traits under saline soil conditions. The traits such as grain yield per plant (GYPP), biomass yield per plant (BYPP), and harvest index (HI) are closely clustered, reflecting a strong positive correlation. Similarly, days to flowering (DF) and days to maturity (DM) appear together, indicating a shared influence on plant

The pointing of in the vectors same direction have a strong positive correlation. The days to maturity and days to flowering were strongly positively correlated, indicating that an increase in both flowering and maturity occurred simultaneously. The panicle weight, biological yield per plant and grain yield per plant were positively correlated. The vectors pointing in opposite directions indicate strong negative correlations. Flag leaf Na^+/K^+ ratio is negatively correlated with harvest index, flag leaf length and grain yield per plant, indicating susceptible genotypes having a high Na^+/K^+ ratio and producing lower grain yield per plant.

development. The plant height (PH), panicle length (PL), and panicle weight (PW) showed moderate association with yield-related traits, while flag leaf length (FLL) and the Na^+/K^+ ratio in the flag leaf (FNaK) were oriented away from the yield components, suggesting either a negative impact or independent variation.

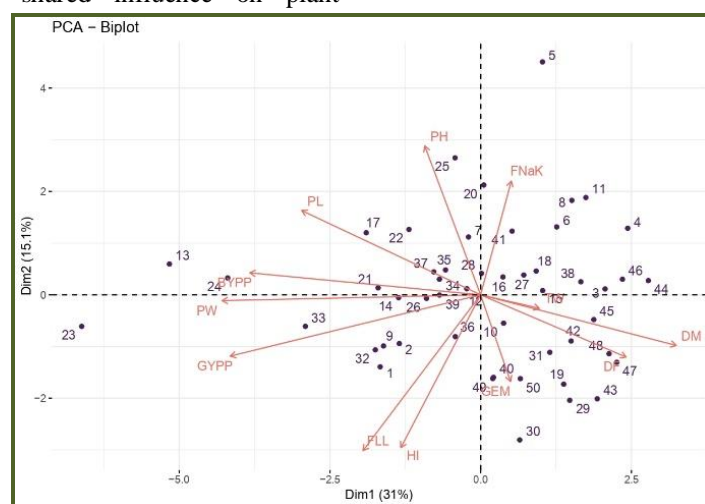


Fig. 2. GT biplot based on different characters of sorghum genotypes under saline conditions.

The genotypes were distributed across all four quadrants, indicating a wide range of genetic diversity in response to salinity. Those positioned in the direction of yield-enhancing traits are likely to exhibit better performance and salt tolerance, making them strong candidates for selection and breeding. The genotypes near DF and DM may provide adaptability to varied

growing periods. Individuals near the origin reflect average performance, while those aligned with FNaK and distant from yield traits may be salt-sensitive, potentially accumulating more sodium.

This biplot serves as a powerful visual tool to distinguish high-performing, tolerant genotypes from sensitive ones. It enables effective classification into

tolerant, moderately tolerant, and sensitive categories, facilitating strategic selection in breeding programs aimed at improving sorghum resilience under saline conditions.

CONCLUSIONS

The present study revealed substantial genetic variability among 50 sorghum genotypes under saline conditions, particularly for key traits such as panicle weight, grain yield, biological yield, and flag leaf Na^+/K^+ ratio. High heritability and high genetic advance for these traits indicate the predominance of additive gene action, suggesting that phenotypic selection would be effective for improvement. Genotypes such as SGP-GS-36, BP-53, and SGP-GS-74 demonstrated superior salt tolerance by maintaining lower Na^+/K^+ ratios. PCA analysis further supported the identification of salt-tolerant genotypes by highlighting the major contributing traits. Traits with low GCV and PCV, like days to flowering and maturity, showed limited scope for improvement. Overall, the findings provide a strong foundation for selecting and developing salt-tolerant sorghum varieties suitable for cultivation in saline soils of Gujarat and similar agro-ecological regions.

FUTURE SCOPE

The current investigation provided the information regarding the variability in terms of genotypic and phenotypic components, heritability and genetic advance as % of mean. In future selection of genotype based on these traits is useful to develop elite genotypes of sorghum with higher production of grain yield under saline soil conditions. The selection for those particular traits identified with a high heritability and high genetic advance as percent of mean can directly be helpful in improving the yield of sorghum and sustain against various abiotic stresses.

Acknowledgement. Authors acknowledge Research supervisor for proper guidance and host institute (Navsari Agricultural University, Gujarat, India) for providing necessary facilities to conduct the experiment and Confederation of Indian Industry (CII), Anusandhan National Research Fellowship (ANRF) and Western Agri Seeds Limited jointly providing PM Fellowship to the author for financial support during his research.

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How to cite this article: Sunil K. Patel, B.K. Davda, Ketan Kanjariya and Haimil Joshi (2025). Evaluation of Morphological and Biochemical Traits for Salt Tolerance in Sorghum Genotypes. *Biological Forum*, 17(7): 155-160.