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Seedling emergence of guar (*Cyamopsis tetragonoloba* L.) genotypes and their growth under irrigation water salinity

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Abstract

Salinity and water scarcity are threatening the sustainability of agriculture in the arid regions of the world. There is an urgent need to develop information on suitable crops that can tolerate salinity and drought to ensure the long-term viability of agriculture in these regions. Guar (*Cyamopsis tetragonoloba* L.) is considered a salt- and drought-tolerant crop that is economically viable. This study was conducted to evaluate the tolerance of 25 guar genotypes under three irrigation water salinities (3.0, 6.0, and 9.0 dS m⁻¹) at Sindh Agriculture University, Tandojam, Pakistan. Twenty-five seeds of each genotype were sown in sand-filled pots, irrigated with 1.5 L of treatment water, and arranged according to a split-plot design with three replications. Genotypes were categorized into four groups based on salinity tolerance index (STI) values of seedling emergence and composite biomass of shoot and root, and considered tolerant (>0.75), moderately tolerant (0.51–0.75), moderately sensitive (0.26–0.50), and sensitive (≤0.25) at various salinity levels. Across all genotypes, shoot and root lengths, shoot and root biomass, K⁺ concentrations, and the K⁺/Na⁺ ratio in shoot dry matter decreased, while Na⁺ and Cl⁻ concentrations in shoot dry matter increased with rising salinity levels. STI values at 9.0 dS m⁻¹ indicated that two genotypes (S-5932 and S-5881) were tolerant, while four were moderately tolerant based on seedling emergence. Additionally, one genotype (S-5981) was tolerant, and six (S-5612, S-5881, S-5932, S-6003, S-6052, and S-6067) were moderately tolerant, based on the composite biomass of shoot and root. The seedling emergence, growth, and dry biomass reduction with increasing salinity underscore the importance of our research in identifying guar genotypes that can thrive in regions with salinity issues.

Abbreviations: SAR, sodium adsorption ratio; SE, seedling emergence; STI, salinity tolerance index.

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1 | INTRODUCTION

Agriculture in arid and semiarid regions of the world is facing the dual threat of elevated salinity and water scarcity (Mekonnen & Hoekstra, 2016; Vörösmarty et al., 2010). To ensure the long-term viability of agriculture in these regions, there is a strong need to develop information on crops that are salt and drought tolerant. Guar (*Cyamopsis tetragonoloba* L.) crop is considered as drought resistant and has the potential to be economically viable for these regions (Ashraf et al., 2002; Shrestha, 2022). Guar has many uses, such as green vegetables, fodder, green manure, and, most importantly, guar gum, which has industrial applications (Douglas, 2005; MacMillan et al., 2021). The global market for guar gum was estimated at \$1.1 billion in 2023 and is projected to reach \$1.7 billion by 2030. It is estimated that the annual global production of guar is about 3.2 million metric tons, and most of it is produced in India and Pakistan. In Pakistan, guar is mainly produced in the southeastern region called Thar (Pathak, 2015; Suthar et al., 2018).

Available literature suggests that guar can tolerate drought stress (Francois et al., 1990). Guar requires reasonably low amounts of irrigation water and can complete its life cycle under limited moisture conditions (Singh et al., 2024). A relatively low water requirement compared to other crops makes guar a reliable and viable crop for dryland agriculture. However, salt tolerance of guar, especially at the germination stage under extremely arid conditions, is not well understood. Seedling emergence (SE) is a sensitive stage for most of the crop species and their genotypes, especially under salt stress conditions (Wang & Shannon, 1999). Plant population is ultimately a major production factor that depends on the seedling's emergence and its vigor. According to Awasthi et al. (2016), germination of most crop species is significantly delayed at elevated root zone salinity. Salinity, whether in soil or irrigation water, causes negative impacts on germination rate and delay in the emergence of seedlings and their growth (Bimurzayev et al., 2021). SE is mostly impacted by salt stress conditions due to osmotic effects and ion toxicity (Tlahig et al., 2021). According to Munns et al. (2002), salinity tolerance among different genotypes is believed to be associated with K^+/Na^+ discrimination.

Furthermore, genetic variability also plays a major role in salinity tolerance at germination (Ahmad et al., 2013; Basavaraja et al., 2021). Therefore, given the importance of adequate plant population for production, evaluating salt stress tolerance at the SE level of various guar genotypes under irrigation water salinity is important. This strategy of identifying salt-tolerant genotypes can be helpful in making the decision to select the genotype under the specific salt level in irrigation water. The Guar Research Station in Bahawalpur is actively working on producing new varieties and genotypes of guar in Pakistan. Recently, three new guar varieties have been released, such as Thal Guara, BR-2022, and BR-21.

Core Ideas

- The guar is a potential crop for arid regions of the world under elevated salinity and hot climatic conditions.
- Identifying the appropriate guar genotypes for their salt tolerance at the seedling growth stage is important.
- We hypothesized that the salinity tolerance index (STI) can be an important mechanism for salt tolerance in genotypes.

TABLE 1 Average ambient environment conditions inside the net house at Sindh Agriculture University, Tandojam, Pakistan, during 15 days of the study on evaluation of guar seedlings emergence and their growth at different levels of salinity during the spring season of 2017.

Average temperature (°C)		Average relative humidity (%)	
Minimum	Maximum	Minimum	Maximum
24	38	73	81

These varieties need to be evaluated for salinity tolerance for wider acceptance. This study assessed the SE behavior of guar seeds under controlled salt stress conditions created through irrigation water salinity with an emphasis on genetic variation among genotypes.

The specific objectives of this study were to determine the SE behavior and early vegetative growth of 25 guar genotypes under various levels of saline water irrigation. The study aims to assess the impact of salt on SE and early growth and compare the response among guar genotypes to salinity.

2 | MATERIALS AND METHODS

This pot culture study was conducted during the spring season of 2017 in a net house at the Soil Science Department, Sindh Agriculture University, Tando Jam, Pakistan. The average temperature and relative humidity were recorded over 15 days of the experiment under ambient environmental conditions (Table 1). The experiment was laid out in a split-plot design, with salinity levels as the main plot and genotypes as subplots with three replications.

2.1 | Plant material, sowing, and nutrient supplement

Seeds of 25 guar genotypes included in this study are listed in Table 2. These include genotypes from the Guar Research Station (GRS), Bahawalpur, Pakistan, and the Agriculture Research Institute (ARI), Tandojam, Pakistan.

TABLE 2 Guar genotypes were collected and included in the study to evaluate salt tolerance.

Source	Guar genotypes
GRS	^a BR-90, ^a BR-99, 2/1, S-5981, S-5883, S-5985, S-6023, S-6005, S-6003, S-6052, S-6067, S-6128, S-6049, S-5881, S-5932, S-5876, S-8885, S-5609, S-5607, S-5611, S-5608, S-5612, S-5615, N-30
ARI	^a Thar bean

Abbreviations: ARI, Agriculture Research Institute; GRS, Guar Research Station.

^aGenotypes/cultivars commonly cultivated in the farmers' fields.

TABLE 3 Chemical properties of the canal and saline water used for irrigation in pots for evaluating guar genotypes for salt tolerance.

Parameter	Unit	S0	S1	S2	S3
EC _{iw}	dS m ⁻¹	0.60	3.00	6.00	9.01
pH	–	7.50	7.72	7.94	8.10
Na ⁺	meq	3.50	6.09	9.48	12.9
Ca ²⁺	L ⁻¹	0.79	1.43	2.26	3.10
Mg ²⁺		1.20	1.78	2.54	3.30
CO ₃ ²⁻	–	–	–	–	–
HCO ₃ ⁻		2.10	3.89	6.23	8.60
Cl ⁻		1.03	1.06	2.35	3.10
SAR	–	3.50	4.80	6.10	7.20
RSC	meq L ⁻¹	0.11	0.68	1.43	2.20

Abbreviations: EC_{iw}, electrical conductivity of irrigation water; RSC, residual sodium carbonate; S0, salinity (zero/control); S1, salinity level 1; S2, salinity level 2; S3, salinity level 3; SAR, sodium adsorption ratio.

Soil (pure sand) was collected from dunes about 9 miles from the experimental site, washed with canal water (0.6 dS m⁻¹), and left to dry. Note that 3 kg of dry sand was filled into round pots (25.4 cm in diameter and 10-cm deep) without drainage, and these pots were arranged as per the experimental design in the ambient environment.

For the nutrient supplement to the seeding of guar, Hoagland's complete nutrient solution, which typically includes all essential plant nutrients (Hoagland & Arnon, 1950), was applied in two splits at the time of sowing (150 mL) and again 7 days later (150 mL). In each pot, 25 healthy seeds of each guar genotype were sown 2.5 cm apart from each other.

2.2 | Preparation and application of saline water

Different saline water levels, 3.0, 6.0, and 9.0 dS m⁻¹, were prepared by dissolving the NaCl and CaCl₂ salts at a 20:1 molar ratio in canal water as a control (0.6 dS m⁻¹). The properties of these waters are given in Table 3. Before sowing, 1.5 L of irrigation water was applied in each pot per the designated treatment. The amount of saline water and Hoagland's solution applied to each pot was sufficient to maintain soil moisture for up to 12 days. After 12 days, the soil moisture

level in the pot was maintained with the application of canal water (a total of 1 cm per pot).

2.3 | Irrigation water analyses

Irrigation water samples were tested for electrical conductivity (EC_{iw}) and pH using standardized digital EC and pH meters. The concentration of Na⁺ was measured with a flame photometer. Titration methods were employed to determine the levels of Ca²⁺ and Mg²⁺ using an ethylenediaminetetraacetic acid solution, CO₃²⁻ and HCO₃⁻ with H₂SO₄ (0.01 N), and Cl⁻ with AgNO₃ (0.005 N) (Jaiswal, 2006).

The sodium adsorption ratio (SAR) was calculated using the formula provided by Richards (1954).

$$\text{SAR} = \text{Na}^+ / [\text{Ca}^{2+} + \text{Mg}^{2+}]^{0.5}$$

2.4 | Agronomical data and final harvest

SE data were observed 15 days after sowing. The SE percentage was calculated using the following formula (Sun et al., 2013):

$$\text{SE \%} = \frac{\text{Total number emerged seedlings}}{\text{Total seed sown in pot}} \times 100$$

Seedlings and roots were carefully uprooted from the sand-filled pots after the 15th day of sowing. After that, the plant material was washed with distilled water and rinsed. The shoot height and root length were measured. To achieve the constant dry weight of all seedlings and their roots, the shoots and roots were separated and placed in an oven at 70 ± 1°C for 72 h. Data on the dry weight of the shoot and root were obtained.

2.5 | Plant tissue analyses

Plant material was digested and diluted 100X (Wolf, 1982). The extracted material was diluted and run on a flame photometer (Jenway PFP7) to analyze the concentration of Na⁺ and K⁺. The ratio of K⁺/Na⁺ was calculated by dividing the values of K⁺ by Na⁺ (Guo et al., 2025).

Chlorides were analyzed through AgNO_3 (Mohr's titration) using potassium chromate (K_2CrO_4) at 5% as an indicator (Wolf, 1982).

2.6 | Categorization of guar genotypes for saline-water tolerance

Guar genotypes were categorized based on salinity tolerance index (STI) values at >0.75 , $0.51\text{--}0.75$, $0.26\text{--}0.50$, and ≤ 0.25 and were considered as tolerant, moderately tolerant, moderately sensitive, and sensitive, respectively. These STI values were obtained based on each parameter, including SE and composite shoot and root biomass, at different salinity levels (3.0, 6.0, and 9.0 dS m^{-1}). This was calculated by the following formula (Bağcı et al., 2003):

$$\text{STI} = \frac{\text{Data at stress condition}}{\text{Data at control condition}}$$

2.7 | Statistical analysis

The data on the seedling's emergence percentage, shoot height, root length, dry matter of shoot and root, and concentrations of Na^+ , K^+ , K^+/Na^+ , and Cl^- in shoot dry matter were analyzed for analysis of variance, correlation matrix, and least significant difference. These data were used to analyze the significant effect of various salinity levels, guar genotypes, and their interaction at $p \leq 0.05$ using GenStat Ver. 4.

3 | RESULTS AND DISCUSSION

3.1 | Effect of salinity on the seedling emergence of 25 guar genotypes

The data in Figure 1a show the emergence percentage of seedlings significantly ($p < 0.05$) different in various salinity levels and genotypes. The maximum emergence was found under the control salinity level, and the least emergence was observed under the highest level of salinity (9.0 dS m^{-1}) in all the genotypes. However, significantly ($p < 0.05$) maximum SE was noted in the genotype S-5881 (74.7%), followed by S-5932 (70.7%) at a threshold level of salinity (9.0 dS m^{-1}). Five genotypes, including S-8885, S-6005, S-5615, S-5611, and 2/1 showed zero emergence at 9.0 dS m^{-1} . All genotypes showed $>50\%$ emergence at both 3.0 and 6.0 dS m^{-1} levels of salinity. At the same time, the maximum SE was found in genotype S-5612 (97.3%) at 3.0 dS m^{-1} and S-5881 (90.7%) at 6.0 dS m^{-1} of salinity.

These results were similar to the observation of different scientists that the percentage of guar seed germination

decreased with increasing root zone salinity levels (Teolis et al., 2009). Francois et al. (1990) found a significant delay in SE, but this did not affect SE up to 8.5 dS m^{-1} .

3.2 | Effect of salinity on the growth and dry biomass of 25 guar genotypes seedlings

The data in Figure 1b,c indicated that shoot height and root length decreased significantly ($p < 0.05$) with increasing levels of salinity. Of the emerged seedlings, the shortest seedlings and roots were found at the 9.0 dS m^{-1} salinity level, and the most extended shoots and roots were found in pots that were applied with Indus River water (0.6 dS m^{-1}). Within the threshold salinity level of 9.0 dS m^{-1} , the significantly ($p < 0.05$) longest shoots (5.3 cm) were found in genotype S-5881, whereas the shortest shoots (1.2 cm) were found in genotype S-5981 (Figure 1b). Similarly, under the same salinity level of 9.0 dS m^{-1} , the significantly ($p < 0.05$) shortest roots were found in genotype S-5609 (1.02 cm), and the longest roots were observed in genotype S-5881 (7.6 cm) (Figure 1c).

A similar trend was also observed in shoot and root dry weights in response to changes in salinity levels. The dry weights of shoots and roots decreased significantly ($p < 0.05$) with an increase in salinity level (Figure 2a,b). However, at the highest level of salinity 9.0 dS m^{-1} , among all those genotypes with emerged seedlings, the maximum shoot dry weight was recorded in genotype Thar bean (0.045 g/seedling). In contrast, the minimum shoot dry weight was recorded in genotype S-5981 (0.016 g/seedling) (Figure 2a). The root dry weights varied between 0.0005 g (S-5985) and 0.004 g (S-5612 and S-5876) among genotypes at 9.0 dS m^{-1} (Figure 2b).

These significant reductions in the results of growth and biomass could be due to the boosted root zone salinity level produced by applied saline water (Chaganti & Ganjegunte, 2022; Liu et al., 2020). Other scientists also reported the adverse effects of salt stress on the initial growth of various legume crops like faba bean and chickpea (Lavrenko et al., 2019; Tavakkoli et al., 2010). Parameters related to vegetative growth of guar crops decreased by 9.6% as per unit of salinity increase above 4.9 dS m^{-1} (Francois et al., 1990).

3.3 | Effect of salinity on shoot Na^+ , K^+ , and Cl^- concentration and K^+/Na^+ ratio of 25 guar genotypes

The concentration of Na^+ and Cl^- increased, while the concentration of K^+ and K^+/Na^+ ratio decreased significantly ($p < 0.05$) with an increase in salinity level across the 25 genotypes (Figure 3a–d). Among all genotypes at the 9.0 dS

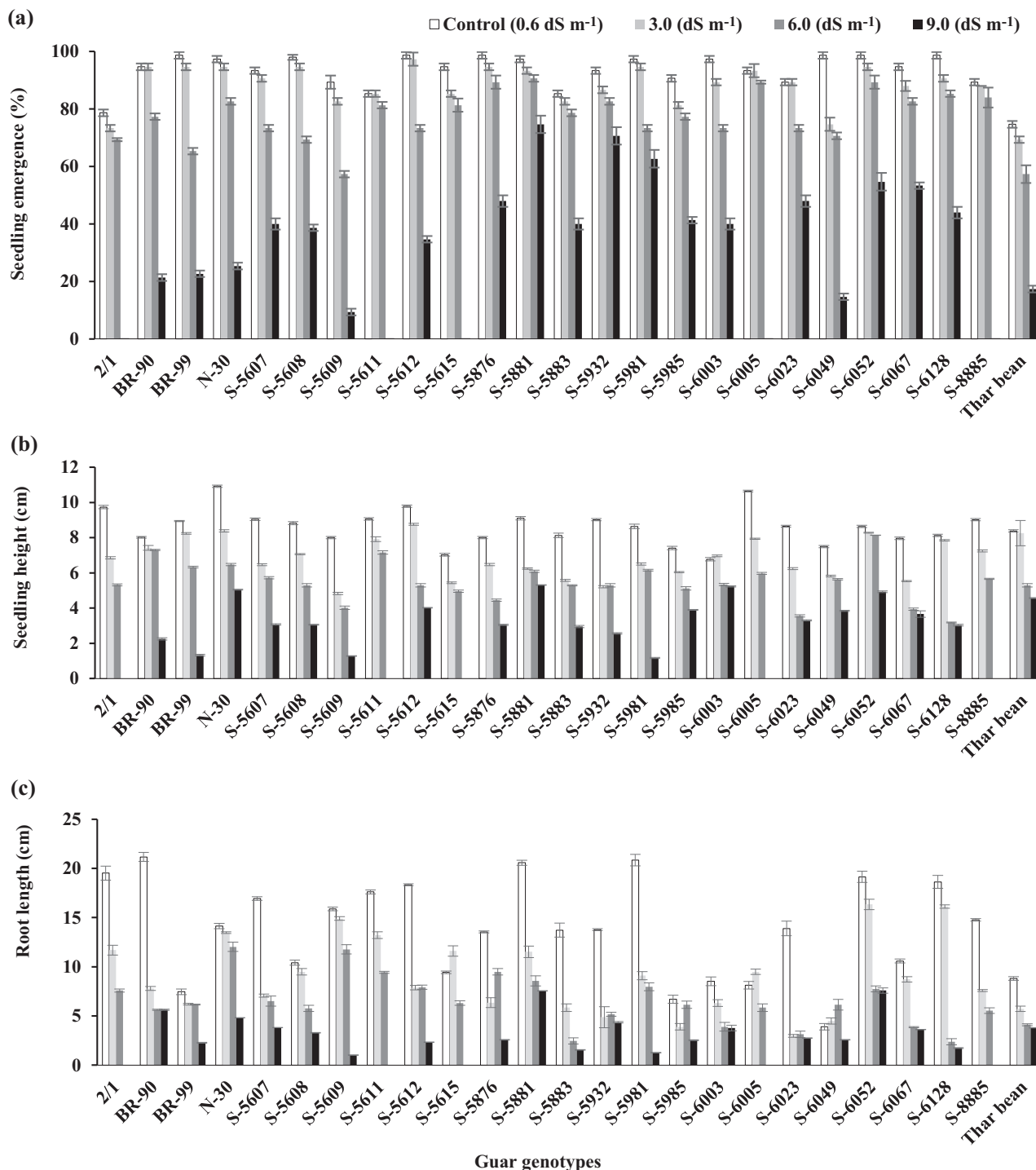


FIGURE 1 Seedling emergence (%) (a), seedling height (cm) (b), and root length (cm) (c) of 25 guar genotypes under control and varying salinity levels of irrigation water.

m⁻¹ salinity level, results showed the significantly ($p < 0.05$) highest concentration of Na⁺ (1.96%) in genotype S-5876 and Cl⁻ (1.65%) in genotype S-6003, while genotype S-6128 recorded the lowest Na⁺ (0.76%) and Cl⁻ (0.99%) concentration (Figure 3a,c). However, the converse was true for the uptake of nutrient element K⁺, at the high level of salinity of 9.0 dS m⁻¹, the shoot dry matter K⁺ concentration

was recorded as significantly lower at 0.026% in genotype S-5612. The ratio of K⁺/Na⁺ varied in direct proportion to K⁺ concentration and inverse proportion to Na⁺ concentration in the dry matter of the plant. As shown in Figure 3d, the highest K⁺/Na⁺ ratio was obtained at the control, while the lowest K⁺/Na⁺ ratio was obtained at a salinity level of 9.0 dS m⁻¹.

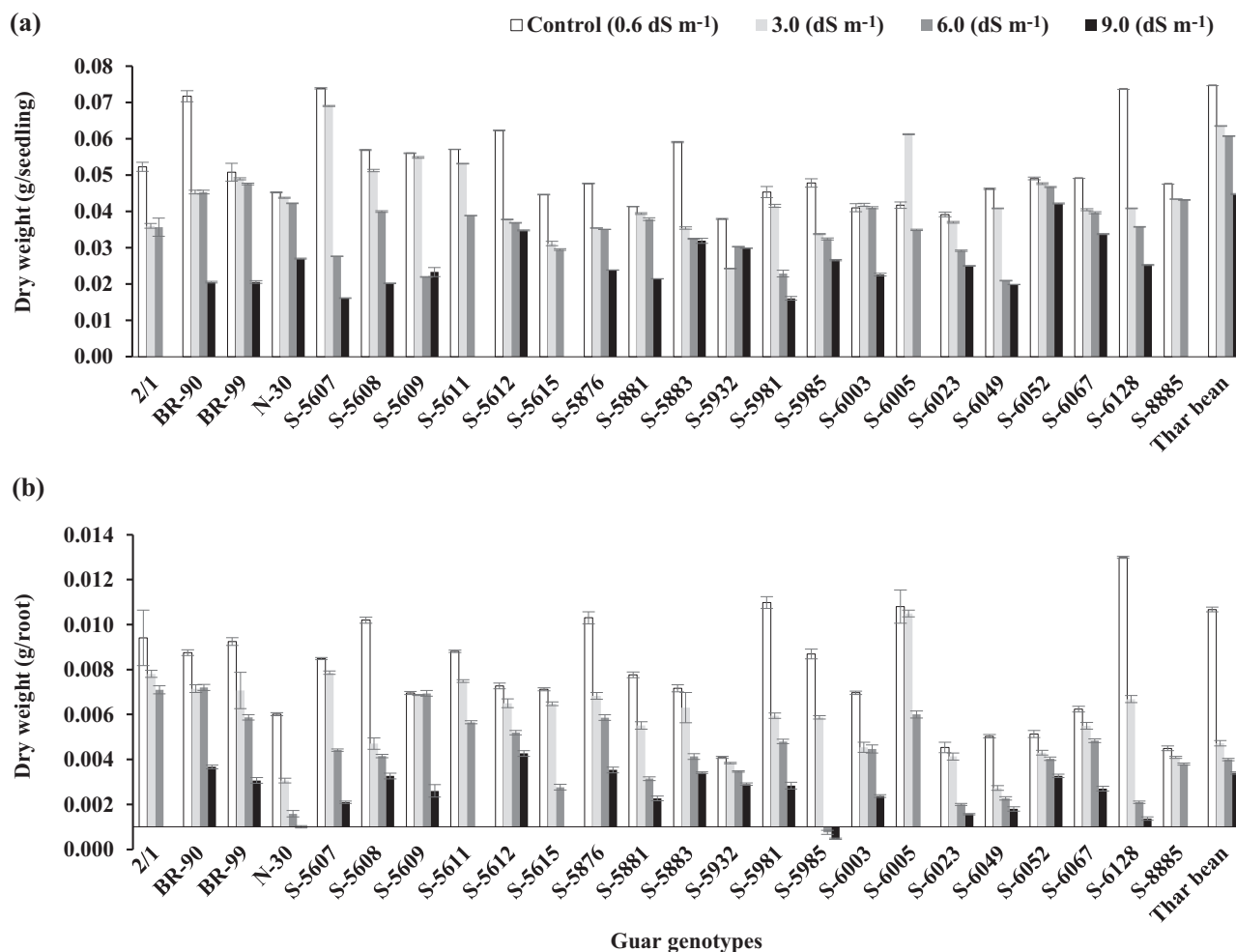


FIGURE 2 Dry weights (g) per shoot (a) and root (b) of 25 guar genotypes under control and varying salinity levels of irrigation water.

Various findings also showed that the Na^+ concentration is inversely proportional to the nutrient concentration, especially of K^+ , which was observed in plant tissue (wheat, Khan et al., 2000; Vicia faba, Abd El-Sattar & Abdelhameed, 2024). Different scientists also achieved similar results, indicating that plants having a higher K^+/Na^+ ratio in their tissues are also tolerant to the stress of salinity (Khan et al., 2009). Chloride concentration in leaf tissue increased along with the increasing level of root zone salinity (Hooks et al., 2019).

3.4 | STI evaluation of 25 guar genotypes at varying salinity levels and correlation matrix among different treatments

All 25 guar genotypes were evaluated and categorized as tolerant, moderately tolerant, moderately sensitive, and sensitive as per STI values ≥ 0.75 , 0.51–0.74, 0.25–0.50, and < 0.25 , respectively (Table 4). At 9.0 dS m^{-1} salinity, based on the STI for SE, two genotypes (S-5932 and S-5881) were classified as tolerant, while four were moderately tolerant. Similarly, based on the STI of composite biomass (root and

shoot), one genotype (S-5981) was found to be tolerant, six were moderately tolerant, and the remaining genotypes ranged from moderately sensitive to sensitive.

At 6.0 dS m^{-1} salinity, based on the STI of composite biomass, seven genotypes were identified as tolerant, 12 were moderately tolerant, and the rest were classified as moderately sensitive. In terms of SE at this salinity level, all genotypes were found to be moderately tolerant to tolerant.

At the lowest salinity level of 3.0 dS m^{-1} , 13 genotypes were identified as tolerant based on the STI of composite biomass, while the remaining genotypes were identified as moderately tolerant. Notably, all genotypes were classified as tolerant based on the STI values for SE at this level.

The correlation matrix between different treatments was significant (≥ 0.05) for all parameters except genotype (Table 5).

4 | CONCLUSIONS

The current study showed a variation in salt tolerance in the evaluated genotypes that can be used to identify and develop

TABLE 4 Tolerance levels of 25 guar genotypes based on salinity tolerance index (STI) values of seedling emergence and composite biomass of shoot and root, under three salinity levels (3.0, 6.0, and 9.0 dS m⁻¹).

Tolerance levels		STI of seedlings' emergence			On average STI of shoot and root dry biomass		
Salinity levels (dS m ⁻¹)		3.0	6.0	9.0	3.0	6.0	9.0
Guar Genotypes	2/1	T	T	Sens.	MT	MT	Sens.
	BR-90	T	T	Sens.	MT	MT	MS
	BR-99	T	MT	Sens.	T	T	MS
	N-30	T	T	MS	MT	MT	MS
	S-5607	T	T	MS	T	MS	Sens.
	S-5608	T	MT	MS	MT	MT	MS
	S-5609	T	MT	Sens.	T	MT	MS
	S-5611	T	T	Sens.	T	MT	Sens.
	S-5612	T	MT	MS	T	T	MT
	S-5615	T	T	Sens.	T	MT	Sens.
	S-5876	T	T	MS	MT	MT	MS
	S-5881	T	T	T	MT	MT	MT
	S-5883	T	T	MS	MT	MS	MS
	S-5932	T	T	T	T	T	MT
	S-5981	T	MT	MT	MT	MS	T
	S-5985	T	T	MS	MT	MS	MS
	S-6003	T	MT	MS	T	T	MT
	S-6005	T	T	Sens.	T	MT	Sens.
	S-6023	T	T	MT	T	MT	MS
	S-6049	T	MT	Sens.	MT	MS	MS
	S-6052	T	T	MT	T	T	MT
	S-6067	T	T	MT	T	T	MT
	S-6128	T	T	MS	MT	MS	Sens.
	S-8885	T	T	Sens.	T	T	Sens.
	Thar bean	T	T	Sens.	MT	MT	MS

Note: T, tolerance (>0.75 STI value); MT, moderately tolerance (0.51–0.75 STI value); MS, moderately sensitive (0.26–0.50 STI value); and Sens., sensitive (≤0.25 STI value).

TABLE 5 Correlation matrix among salinity levels (S), genotypes (G), seedling emergence (SE), shoot length (SL), root length (RL), shoot dry weight (SDW), root dry weight (RDW), ions (Na⁺, K⁺, and Cl⁻) content, and K⁺/Na⁺ ratio in shoot dry matter of guar crop at seedling growth stage.

Parameters	S	G	SE	SL	RL	SDW	RDW	Na ⁺	K ⁺	Cl ⁻
G	0.02									
SE	-0.82	0.01								
SL	-0.85	-0.05	0.74							
RL	-0.70	-0.19	0.57	0.71						
SDW	-0.65	0.02	0.53	0.68	0.52					
RDW	-0.70	-0.21	0.55	0.64	0.58	0.63				
Na ⁺	0.81	-0.05	-0.68	-0.75	-0.62	-0.64	-0.59			
K ⁺	-0.76	0.04	0.54	0.60	0.51	0.52	0.51	-0.58		
Cl ⁻	0.77	0.12	-0.60	-0.65	-0.53	-0.44	-0.53	0.64	-0.47	
K ⁺ /Na ⁺	-0.72	0.48	0.48	0.61	0.66	0.50	0.54	-0.74	0.75	-0.53

Note: n = 300. r significant at 5% ≥ 0.12. r significant at 1% ≥ 0.22.

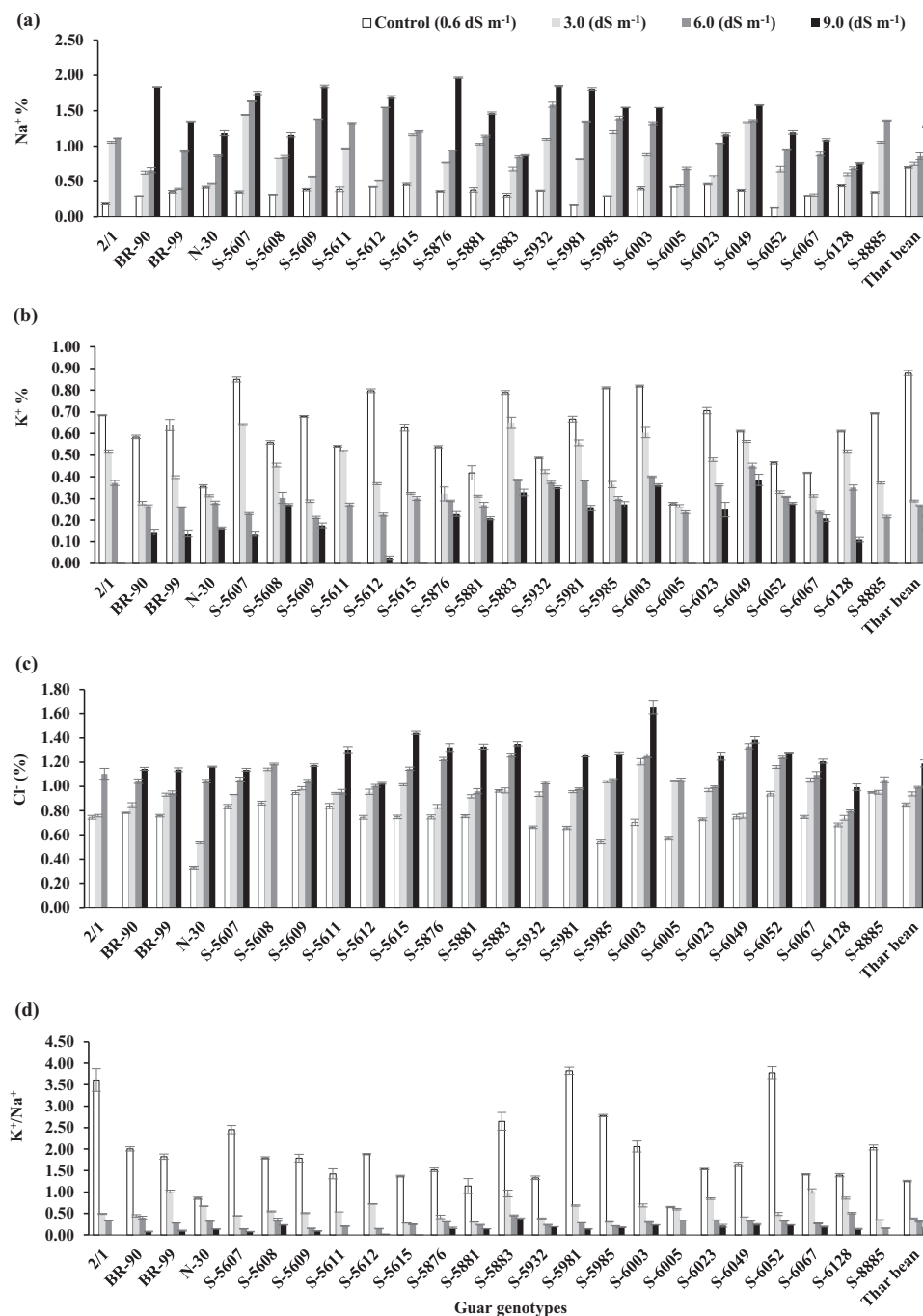


FIGURE 3 Na⁺ % (a), K⁺ % (b), and Cl⁻ % (c) concentrations and K⁺/Na⁺ ratio (d) in shoot dry matter of 25 guar genotypes under control and varying salinity levels of irrigation water.

salt-tolerant guar varieties. In general, SE rate, shoot height, root length, and dry biomass of shoot and root decreased with increasing salinity. Among the 25 guar genotypes evaluated, two (S-5932 and S-5981) were moderately tolerant based on the STI of SE. However, based on the STI of composite biomass of shoot and root, genotype S-5981 was considered tolerant, and six genotypes (S-5612, S-5881, S-5932, S-6003, S-6052, and S-6067) were moderately tolerant at the highest salinity level of 9.0 dS m⁻¹ tested in the current study. At the

second highest salinity level of 6.0 dS m⁻¹, all genotypes were moderately tolerant to tolerant at STI of SE, and similar results were found on STI of composite biomass of shoot and root, but only six genotypes (S-5607, S-5883, S-5981, S-5985, S-6049, and S-6128) were moderately sensitive. At 3.0 dS m⁻¹, however, all 25 genotypes were categorized as moderately tolerant to tolerant. At the threshold salinity of 9.0 dS m⁻¹, S-5881 and S-5981 genotypes showed the longest (5.3 cm) and shortest (1.2 cm) shoots; S-5881 and S-5609 recorded the longest

(7.6 cm) and shortest (1.0 cm) roots, respectively. After 15 days, the highest shoot dry weight ($0.045 \text{ g plant}^{-1}$) was noted for the genotype “Thar bean,” and the highest root biomass of $0.004 \text{ g plant}^{-1}$ was observed for three genotypes (BR-90, S-5612, and S-5876). An increased ratio of K^+/Na^+ was found in plant tissues of genotypes with high salt tolerance, indicating differential uptake of these ions as a possible mechanism for salt tolerance in guar.

AUTHOR CONTRIBUTIONS

Jhaman Das Suthar: Conceptualization; formal analysis; investigation; methodology; writing—original draft. **Inayat-ullah Rajpar:** Conceptualization; methodology; supervision; writing—review and editing. **Zia-ul-Hassan:** Conceptualization; methodology; supervision; writing—review and editing. **Girisha K. Ganjegunte:** Conceptualization; methodology; resources; supervision; writing—review and editing. **Kulbhushan K. Grover:** Formal analysis; methodology; writing—review and editing.

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CONFLICT OF INTEREST STATEMENT

The authors declare no conflicts of interest.

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