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RESEARCH ARTICLE

Pearl Millet Mapping Population Parents: Performance and Selection Under Salt Stress Across Environments Varying in Evaporative Demand

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Abstract It is vital to screen the germplasm of crop plants for salt stress tolerance so as to utilize them in breeding programs. Accordingly, in the present study, twenty diverse inbred lines, parents of mapping populations of pearl millet were chosen to determine the phenotypic contrasts for seed yield, which can open the way for developing salt tolerance QTLs. Parents were grown in two summer seasons (late and early) with VPD ≥ 2 kPa, and one rainy season with VPD < 2 kPa, during flowering and grain filling under saline (150 and 200 mM) and non-saline (0 mM) conditions. Salinity delayed flowering time by a fortnight in the summer seasons but only 5–6 days in the low VPD rainy season. Salinity decreased grain yield by 86% in late-summer and 80% in early-summer, but less than 70% in rainy season. GY penalty was higher than vegetative biomass under saline conditions especially in summer season when the evaporative demand was very high. It appears that reproduction and grain filling are sensitive to high temperature that can compound the effect of salinity and high VPD. GY of inbreds under salinity was not better in comparison with non-saline conditions. DOF and grain density (thousand grain weight) were found as important correlated traits under salinity. Also, GY was affected significantly if VPD increased during flowering time.

Abbreviations

DAS	Days after sowing
DOF	Days of flowering
TGW	Thousand grain weight
GY	Grain yield
PW	Panicle weight
QTL	Quantitative trait loci
VPD	Vapor pressure deficit
h^2	Broad sense heritability

Significance statement The phenotypic trait information generated in parental pairs with varying saline doses and evaporative conditions would open the possibility to map salt tolerance QTLs in pearl millet.

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Introduction

More than 800 million ha of lands throughout the world are salt affected [FAO/UNESCO Soil Map 2008], out of which about 100 million ha are arable. These lands have accumulated salts over geological periods of time in arid and semi-arid zones, but also more recently from mismanaged irrigation [1]. Pearl millet (*Pennisetum glaucum* L. R. Br.) is a well-adapted crop to areas characterized by drought, low soil fertility, and high temperature. It is also classified as a moderately salt tolerant crop (threshold electrical conductivity of the soil saturation extract ECe = 3–6 dS m^{-1} [2]. Therefore, focusing on research to screen the germplasm and to improve the salt tolerance of

pearl millet could increase the food and feed production of salt affected dry areas. Salinity is a complex trait and breeding for salt tolerance would likely benefit a lot from marker assisted selection, and rapid progress is currently taking place in chickpea [3], rice [4], and sorghum [5]. Unfortunately, no molecular markers that map for salinity tolerance are known in pearl millet. So, the first step towards this would be to identify pearl millet lines that vary in salt tolerance levels, possibly using highly diverse and representative materials as was used previously in the reference collection of chickpea [6], for use in the development of mapping population. The highly heterozygous nature of cross-pollinated pearl millet and the non-availability of diverse and representative inbred material led us to assess possibly contrasting lines for salinity tolerance among parental lines of existing pearl millet mapping populations, where several recombinant inbred line populations and the genotypic data are available. Finding phenotypic contrast in any of these parental pairs would open the possibility to map salt tolerance QTLs, as has been carried in chickpea, using an existing mapping population where parental lines showed an important contrast in their yield response to salinity [3, 7].

Salinity induces water stress before ionic stress, but osmotic stress has a major negative effect on plant growth compared to ionic stress. Therefore, this raises the question whether changes in the evaporative demand, which could make the osmotic stress more severe by increasing the water potential gradients between soil and atmosphere can affect the genotypic response to salt stress. Recent evidence in maize [8], sorghum [9], and pearl millet [10] showed differences in the transpiration response to high VPD. Such reports indicate that a large genotype-by-environment variation for the response to salinity is expected, which must be tested before progressing further for the identification of salt tolerance QTLs. Therefore, the objective of the present study was (1) to screen a set of pearl millet inbred lines, parents of existing mapping populations, and select the most contrasting pairs of parental lines, (2) to assess the relative contribution of environment (VPD) to GY and its attributing traits under saline and non-saline (control) conditions, (3) and to identify the major traits involved in salinity tolerance.

Material and methods

Plant Material

Twenty-four genotypes were used for salinity screening, which consisted of nine diverse inbred pairs ($9 \times 2 = 18$), which are the parents of existing mapping populations,

along with two inbred forage pollinator controls (Tift 186, Tift 383) and four elite B-lines ICMB 89111-P2, ICMB 89111-P6, ICMB 90111-P2 and ICMB 90111-P6. These inbreds were paired on the basis of center of origin and 18 different parameters by ICRISAT (Table 1).

Plant Growth Conditions

Experiments were carried out at ICRISAT headquarters Patancheru, India, in three environments: (1) late-summer (Mar to late June), (2) early-summer (Feb to late May) and (3) rainy season (Aug to early Nov). The VPD (calculated based on average temp and RH % of day and night) was on an average above 2 kPa (T_{max} 41 °C) during summer season and was below 1 kPa (T_{max} 33 °C) during the rainy season, indicating that the evaporative demand was extremely different between summer and rainy seasons (Fig. 1). Plants were grown in 12-inch plastic pots containing 10 kg of red soil (Alfisol) from ICRISAT farm fertilized with di-ammonium phosphate at the rate of 0.3 g per kg of soil. Experiments were carried out in an open air equipped with rain-out shelters to protect from rain. Pots were buried 3/4th in soil to have a soil temperature closer to a field environment and to avoid pot wall heating. Saline treatment was applied as a 2.733 g NaCl kg^{-1} soil in late-summer and 2.049 g NaCl kg^{-1} soil in both early-summer and rainy seasons. These treatments were equivalent to the application of a salt solution of 200 mM (late-summer) and 150 mM (early-summer and rainy seasons) concentration, in sufficient amount to saturate the Alfisol used at field capacity (approximately 20% w/w). Salt treatment was applied in three equal split doses (0.911 in summer 2007 and 0.683 g NaCl kg^{-1} soil in the other two trials) within 15 days of sowing. The first dose was applied at sowing and dissolved in 2 L pot^{-1}. The second and third doses were applied at 8 and 15 DAS, and diluted in 0.5 L pot^{-1}. Thereafter, pots were watered with tap water containing no significant amount of NaCl and maintained close to field capacity to avoid an increase in salt concentration in the salt solution. Since the pots were sealed at the bottom to avoid salt leakage, utmost care was taken to avoid excessive and insufficient watering to prevent water logging and water stress. Non-saline (control) pots were initially brought to field capacity with normal tap water. The pots were opened at the bottom so as to drain out excess water. Four hills were planted with several seeds. Thinning was performed 5-days after complete salt treatment and two plants were maintained in each pot. Spacing of pots was such that population in the trial was about 15–18 plants m^{-2}. Trials were uniformly managed to maximize growth and grain yield. The size of pot was sufficient to allow plants to grow until maturity.

Table 1 Names, pedigrees, and related information for 18 inbred mapping population parental lines and 2 inbred forage pollinator controls (Tift 186, Tift 383) of pearl millet used

Name	Pedigree/origin	Comments	Registration/references
(LGD 1)-B-10	Partial backcross d_2 dwarf, e_1 early (donor = Tift 756) derivative of a bold-seeded *Iniadi* landrace sample from Togo; bred at Tifton, GA, USA; reselected at ICRISAT-Patancheru	d_2 dwarf, e_1 photoperiod-insensitive early flowering	[11, 12]
(ICMP 85410)-P7	{[SC 14(M)-1] × [SD2 × EB 2 (D1088)]-1}-64	d_2 dwarf, late flowering	[12, 13]
(Tift 23D$_2$B$_1$)-P1-P5	Partial backcross d_2 dwarf derivative of forage seed parent maintainer line Tift 23B$_1$; bred at Tifton, GA, USA	d_2 dwarf, many tillers	[12, 14]
(IP 18292, WSIL)-P8	Genetic stock (*ws, d$_2$, y, gl*) with complex pedigree developed at ICRISAT-Patancheru	d_2 dwarf, long panicles	[12, 15]
(81B)-P6	Downy mildew resistance outcross derivative of Tift 23D$_2$B$_1$ selected from a mutation breeding program at ICRISAT-Patancheru	d_2 dwarf	[12, 16]
(ICMP 451)-P8	Downy mildew resistant restorer selection from ICMP 451 (LCSN 72-1-2-1-1)	Tall, long panicle bristles	[12, 16]
(ICMP 451)-P6	Downy mildew resistant restorer selection from ICMP 451 (LCSN 72-1-2-1-1)	Tall long panicle bristles	[12, 16]
(H 77/833-2)-P5(NT)	Off-type segregant from H 77/833-2	Short, many tillers, photoperiod-sensitive early flowering	[12]
H 77/833-2	Elite pollinator line from Haryana Agricultural University, Hisar, India	Short, many tillers, photoperiod-sensitive early flowering; seedling heat stress tolerant	[12]
PRLT 2/89-33	Inbred line bred at ICRISAT-Patancheru from the Bold Seeded Early Composite (largely based on *Iniadi* landrace germplasm and derived breeding materials), with the pedigree BSEC 8501-13-2-2-3-2	Medium tall, early flowering; seedling heat stress sensitive; terminal drought stress tolerant	[12]
(W 504)-1-P1	Breeding line from Indian Agricultural Research Institute, New Delhi, India	Tall, medium-late flowering	[17]
(P 310-17)-B	Stable source of downy mildew resistance selected at ICRISAT-Patancheru from germplasm line IP 6329 from Mali	Tall, late flowering	[15, 17]
(PT 732B)-P2	"Spontaneous" dwarf mutant in elite breeding line from Tamil Nadu Agricultural University, Coimbatore, India	d_2 dwarf, photoperiod-sensitive late flowering	[18]
(P 1449-2)-P1	IP 21168; stable source of downy mildew resistance selected at ICRISAT-Patancheru from germplasm line IP 5853 from Senegal	Tall, photoperiod-sensitive late flowering	[15]
(ICMB 841)-P3	Downy mildew resistant outcross of MS 5141B; developed at ICRISAT-Patancheru by pure line selection for disease resistance in a contaminated seed lot of MS 5141B	Medium tall, medium-early flowering	[18, 19]
(863B)-P2	Maintainer line developed at ICRISAT-Patancheru by selfing in a bold-seeded Iniadi landrace sample from Togo	Medium tall, medium-early flowering, drought tolerant	[19]
(IP 18293)-P152	d_2 dwarf, *P* purple foliage genetic stock with complex pedigree developed at ICRISAT-Patancheru		[15]
(Tift 238D$_1$)-P158	d_1 dwarf restorer of the A$_1$ cytoplasmic male-sterility system bred at Tifton, GA, USA	Late flowering	[20]
Tift 186	Forage pollinator bred at Tifton, GA, USA by selfing in a forage germplasm accession from South Africa	Tall, late flowering	[21]
Tift 383	d$_2$ dwarf forage pollinator bred at Tifton, GA, USA from Tift 186 × (Tift 239D$_2$B$_2$ × Tift 186)	d_2 dwarf, late flowering	[22]

Fig. 1 Vapor pressure deficit (VPD) conditions during the growing season of late summer (late March–July), early (Feb–May), and the rainy seasons (August 8 to early Nov). VPD was calculated on the basis of average of maximum and minimum temperatures (°C) and relative humidity (RH %) per day at ICRISAT, Patancheru, India
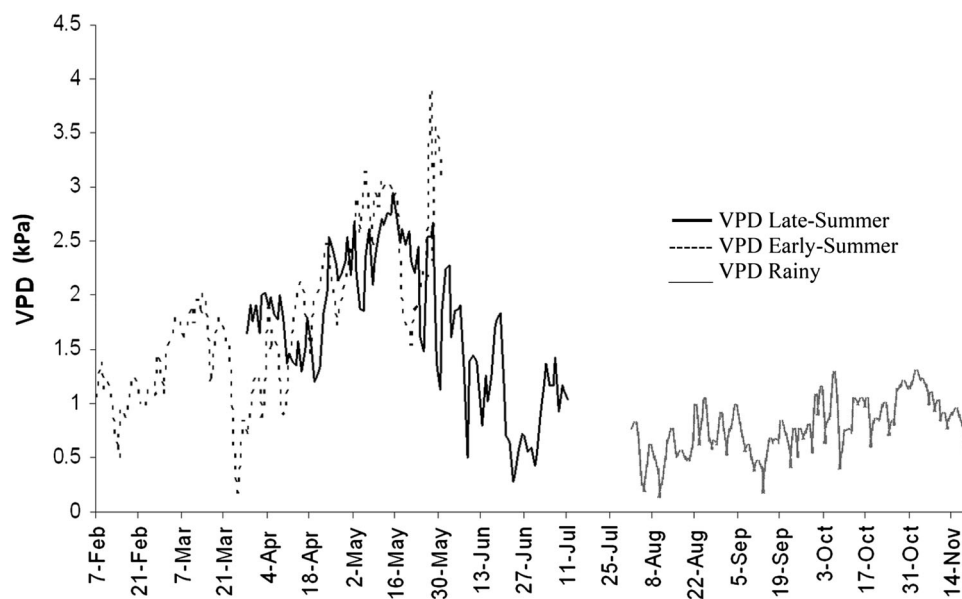
Measurements

Leaves, stems and panicles were separated after harvest and dried in oven at 60 °C for 72 h and dry weights were recorded. Panicles were held separately to recover grain yield. Parameters measured in all three environments included DOF (appearance of 50% of white stigma on main stem panicle), PW (g plant^{-1}), GY (g plant^{-1}), leaf and stem dry weights (g plant^{-1}) and shoot biomass (leaf and stem dry weights, g plant^{-1}). In late and early summer, alongside the above parameters, panicle length (cm), TGW (counted manually), plant height (cm) and number of productive tillers (tillers which produce fertile panicles) were recorded.

Data Analysis

Combined and individual analysis of data obtained from late, early-summer and rainy seasons were carried out with season (E), treatment (T) and genotypes (G) as main factors, using the software SAS PROC GLM (Version 9.2; SAS Institute, Inc., Cary, NC). To assess the range of genotype-by-treatment (G x T) interaction in each of the seasons, two-way ANOVA analysis was performed using genotype (G), treatment (T) and genotype × treatment (G × T) variance components. Since the environmental conditions varied between summer and rainy seasons, the range of genotype-by-season interaction was performed in each of the treatments. Initially, data from summer seasons for saline and non-saline treatments were analyzed separately to facilitate G × E interaction effect and found its interaction was smaller than the year effect (data not presented). Therefore, combined data from three seasons were

subjected independently for G, E, and genotype × season (G × E) analysis separately for saline and non-saline treatments.

Broad sense heritability was estimated for each treatment within each season separately using the formula

$$h^2 = \sigma_g^2 / \left(\sigma_g^2 + \sigma_e^2 \right).$$

Simple correlation between agronomic traits under saline and non-saline conditions were analyzed combining for summer, and separately for rainy season using SAS PROC CORR (Version 9.2) because the basic objective was to find out the differences in relationship among the measured parameters in different seasons.

Results and Discussion

Source of Variation

The two-way ANOVA conducted to assess the genotype (G), treatment (T), and G x T interaction effects independently for three seasons revealed that salinity affects ten measured traits in each season (Table 2). G and T effects were predominant over the G × T interactions for phenological attributes and DOF. However, for critical traits like GY and PW, G and G × T, interaction effects displayed similar magnitude in late-summer and rainy season implying that GY under salt stress is not predictable from non-saline performance. Independent analysis of G, E, G × E (genotype × season) under non-saline and saline conditions revealed that they affect the traits significantly except for panicle length where seasonal effect was not significant (Table 3). The magnitude of G × E interaction

Table 2 The magnitude and significance of mean sum of squares (MSS) of genotype (G), treatment (T) and their interaction (G × T) effects of ten agronomic traits for twenty inbred and four tester lines of pearl millet screened under non-saline and saline environment of late-summer season (200 mM NaCl), early-summer season (150 mM NaCl) and rainy season (150 mM NaCl)

Source of variation	df	Days of flowering (DAS)	Panicle weight (g plant^{-1})	Grain yield (g plant^{-1})	Leaves weight (g plant^{-1})	Stem weight (g plant^{-1})	Shoot biomass (g plant^{-1})	Panicle length (cm)	1000 grain weight (g)	Plant height (cm)	Productive tiller number
Late-summer season											
Treatment	1	9085.4***	15,328.4***	7272.5***	2605.4***	18,608.8***	35,120.9***	632.0***	266.2***	152,537.7***	46.3***
Genotype	23	21,578.1***	4902.9***	2466.5***	4004.5***	11,858.9***	20,327.6***	3272.1***	283.0***	103,694.0***	233.3***
G x T	23	2891.7***	4484.2***	2209.6***	1348.9**	4833.5***	8325.6**	1390.3***	59.9***	34,963.1***	43.2***
replication	3	2.33 ns	310 ns	188 ns	22 ns	478 ns	626 ns	4.57 ns	0.715 ns	960 ns	2.19 ns
Early-summer season											
Treatment	1	8397.6***	23,450.2***	10,807.5***	1118.0***	19,690.6***	30,192.5***	1098.7***	262.2***	196,796.8***	24.12***
Genotype	23	17,383.4***	10,777.2***	6561.0***	3356.9***	16,617.4***	29,377.2***	3213.7***	454.8***	117,897.8***	181.2***
G x T	23	2879.8***	5702.0***	2475.3**	9337*	10,036.3***	15,134.6***	452.0***	69.1*	30,373.4***	35.9 ns
replication	3	257 ns	576.2 ns	495.5 ns	118.5 ns	159.5 ns	496.8 ns	13.32	3.2 ns	1835 ns	0.74 ns
Rainy season											
Treatment	1	2364.1***	15,825.1***	5479.9***	7790.7***	32,148.2***	71,453.7***				
Genotype	23	6394.5***	5902.2**	3577.9**	3813.4**	19,501.4***	29,500.9***				
G x T	23	1408.2***	6666.1***	3916.9**	1063.7 ns	10,907.8**	13,697.8**				
replication	2	150.4 ns	1767.7 ns	765.8 ns	687.5 ns	1397.8 ns	3942.2 ns				

The panicle length, TGW, plant height and productive tiller number recorded only in summer seasons. Screenings were conducted in alpha design with four replications in each treatment (non-saline and saline) in both summer season and three replications in the rainy season

ns non-significant

* Significant at *P* = 0.05; ** significant at *P* = 0.01; *** significant at *P* = 0.001

Table 3 The magnitude and significance of mean sum of squares (MSS) of genotype (G), season (E) and their interaction (G × E) effects of ten agronomic traits for twenty inbred and four tester lines of pearl millet screened under non-saline and saline environment of late-summer season, early-summer season and rainy season

Source of variation	df	Days of flowering (DAS)	Panicle weight (g plant^{-1})	Grain yield (g plant^{-1})	Leaves weight (g plant^{-1})	Stem weight (g plant^{-1})	Shoot biomass (g plant^{-1})	Df	Panicle length (cm)	1000-grain weight (g)	Plant height (cm)	Productive tiller number
Non-saline												
Season (E)	2	335.7***	2245.6***	552.7***	2981.8***	4110.3***	13,396.2***	1	0.02 ns	33.7***	4254.1***	9.8***
Genotype (G)	23	558.1***	1149.1***	552.7***	363.3***	2588.0***	3870.8***	23	208.5***	21.4***	9600.9***	10.8***
G × E	46	36.83***	331.83***	623.4***	72.3*	377.2***	562.2**	23	11.1*	4.28***	537.4**	1.6**
R(season)	3	21.67 ns	222.3 ns	107.5 ns	127.0 ns	224.8 ns	279.7 ns	3	3.7 ns	1.2 ns	2.3 ns	0.43 ns
Saline												
Season (E)	2	1242.5***	294.2***	37.2***	420.4***	256.6***	1333.6***	1	37.5**	18.3***	7956.7***	11.2***
Genotype (G)	23	1028.1***	50.4***	21.2***	138.6***	162.4***	482.1***	23	110.6***	10.2***	2228.2***	5.8***
G × E	46	161.1***	32.7**	15.6**	18.1**	24.4***	56.4***	23	31.9***	4.24***	466.5***	2.9***
R(season)	3	36.5 ns	76.7 ns	25.6 ns	15.7 ns	25.4 ns	69.4 ns	3	2.13 ns	0.17 ns	96.4 ns	0.5 ns

The panicle length, TGW, plant height and productive tiller number recorded only in late-summer season and early-summer season. Screenings were conducted in alpha design in rainout shelter (ROS), ICRISAT, India

ns non-significant

* Significant at $P = 0.05$; ** significant at $P = 0.01$; *** significant at $P = 0.001$

effects were much lower than the G effects for flowering time, leaf, stem and shoot biomass, panicle length, TGW, tiller number and plant height under saline conditions. Even for the PW and GY, the G x E effects were smaller than the G effects, which mean that by and large, across seasons under salt stress the G effects dominated the differences. When analysis was restricted to two summer seasons, the G × E interactions were small for grain yield (data not presented).

Trait Expression in non-saline and Saline Conditions

Genotypes recorded wide variations for all measured traits under non-saline conditions (Table 4). Among the 24 genotypes, two-fold increase was noticed in panicle length, 2–3-folds in TGW, 4–6-folds in stem and shoot biomass, 6–8-folds in PW and leaf dry weight and about 15-folds in GY across the three seasons. The DOF for late and early-summer was close to 50 after sowing and slightly shorter in rainy season (46.7 DAS). Large variations for all traits were observed under saline conditions (Table 4). The DOF showed a three-fold variation in late (32.3–102.5 DAS) and early-summers (33.3–103 DAS); while a two-fold variation was recorded in rainy season (36–75 DAS). GY varied five and seven-folds in late and early-summers respectively and eight-folds in rainy season, and these variations were lower than those noticed under non-saline conditions. The mean flowering time was 64.9, 64.4 and 55.7 DAS for late and early-summers and rainy season respectively. Nine-day early flowering was noticed in the rainy in comparison with summer season. The mean PW and GY were 5.8 and 2.2 g in late-summer, 7.1 and 4.0 g in early-summer and 11.3 and 5.7 g in rainy season respectively. When the data on GY obtained in late-summer (200 mM NaCl) were compared to those of early-summer (150 mM NaCl), it was observed that addition of 50 mM salt in late-summer led to a 45% drop in GY compared to the early-summer. This could be partially related to VPD and temperature differences during the reproductive period in summer season, because the grain yields under non-saline conditions decreased only by 20% in late compared to early-summer. It has been pointed out that low temperature during germination and high temperature during flowering and grain development adversely affect the respective processes [23, 24]. Similarly, when GY under salinity was compared in early-summer and rainy seasons, both seasons receiving the same salt treatment, the GY in rainy season was 30% higher than that of summer, while the same under non-saline conditions was similar in both the seasons. This infers that response to salinity stress depended on VPD which was more severe in summer.

Saline treatment delayed flowering time by 20-days in late and by 15-days in early-summer. Contrarily, in rainy season, it was delayed only by 5-days. The relative decrease in shoot biomass accumulation from non-saline treatment was 45% in late, 37% in early-summer and 26% in rainy seasons. Vegetative growth decreased by 50% under high VPD conditions and overall, the relative decrease in biomass under salinity was more severe in summer than in rainy season. Stem dry weight experienced severe drop compared to leaves (Table 3). Salinity decreased the PW to 76% of the non-saline control in both summer seasons and 68% in rainy season. Similarly, salinity decreased GY by 86 and 80% in late- and early-summers respectively, while the decrease was 70% in rainy season. GY performance under salinity was decreased in comparison with non-saline conditions for each experiment separately (Fig. 2). There was no significant relationship except early-summer, which confirmed the high level of G × T interactions across late-summer and early-rainy experiments (Table 3). Panicle length displayed variation among genotypes under salinity and was closely related with GY, but was not affected by salinity. However, yield was reduced due to panicle tip sterility. Panicle/spikelet sterility has been found as the most common symptom under salinity and has been reported in maize [25] and rice earlier [26].

Heritability (h^2)

Variable h^2 was noticed between treatments in each season (Table 4). Overall, DOF, panicle length and plant height exhibited > 70% h^2 in all seasons whereas growth-related traits and TGW showed h^2 in the range of 45–80% across the treatments and seasons. PW and GY showed 30–65% h^2. DOF, plant height, and panicle length have been found as highly heritable traits followed by shoot biomass. TGW and GY recorded medium to high range of h^2 in different conditions. High broad sense h^2 was reported previously for GY per plant, biomass, panicle length and ear girth under salt stress in pearl millet [27].

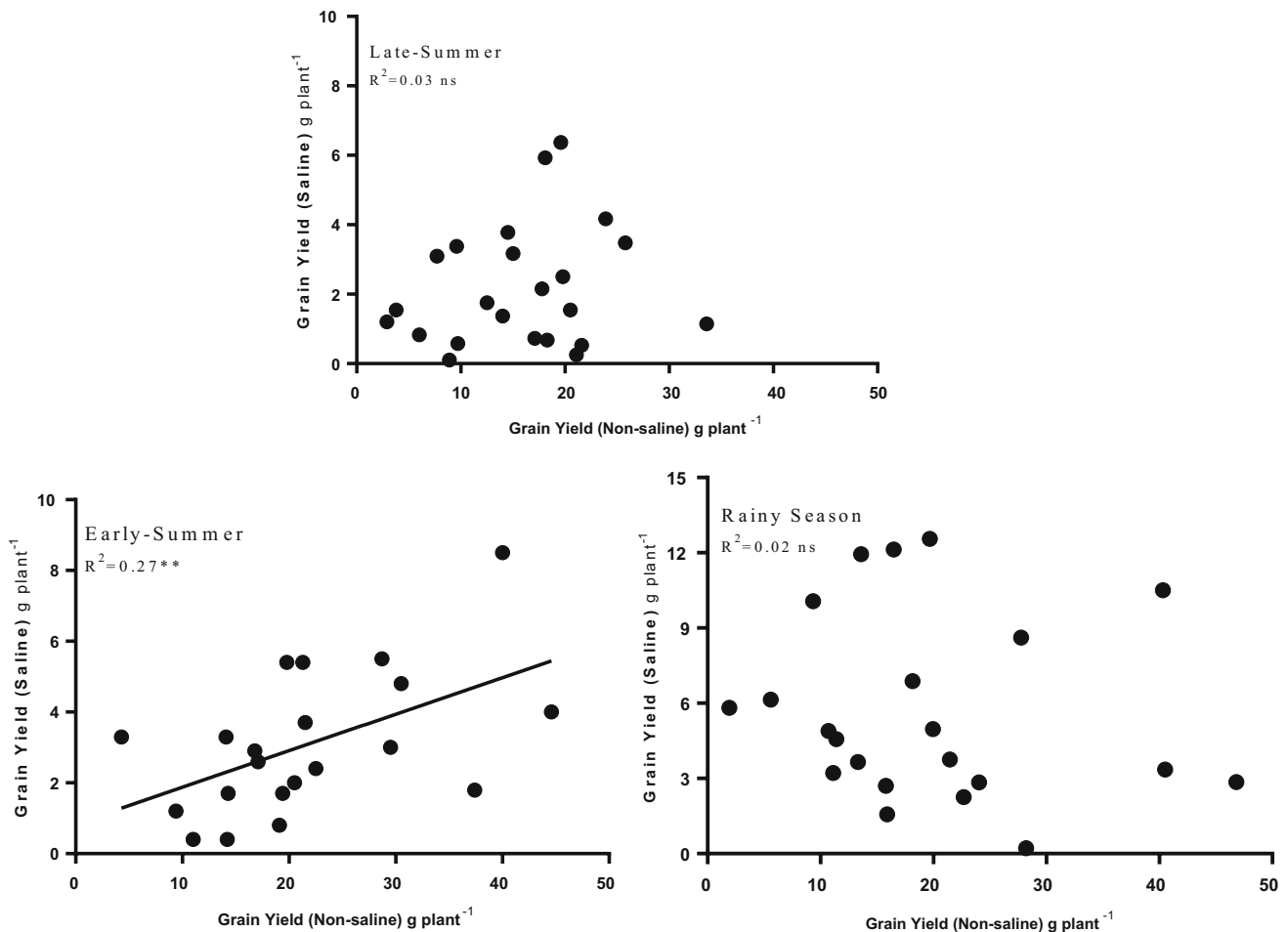## Correlation for Summer and Rainy Seasons

In summer and rainy seasons, under non-saline conditions GY was correlated well with shoot biomass (summer 0.46; rainy 0.69), whereas no significant correlation was observed under stress (Table 5). TGW was strongly correlated with GY in saline (0.49) than in non-saline (0.31) conditions indicating that selection for high grain density could be beneficial under saline conditions. Under salinity, DOF showed negative correlation with GY (summer − 0.30; rainy − 0.39) and TGW (− 0.30). Contrarily, a positive correlation with shoot biomass (0.46) was

Table 4 Mean, range and broad sense heritability under non-saline and saline conditions for 20 inbred lines and 4 testers for DOF, GY and shoot biomass measured in three environment in late-summer season (salt stress = 200 mM), early-summer season (salt stress = 150 mM) and the rainy season (salt stress = 150 mM) whereas traits panicle length, 1000-grain weight, plant height and productive tiller number recorded only in summer seasons

Season	Days of flowering (DAS)	Panicle weight (g plant⁻¹)	Grain yield (g plant⁻¹)	Leaves weight (g plant⁻¹)	Stem weight (g plant⁻¹)	Shoot biomass (g plant⁻¹)	Panicle length (cm)	1000-grain weight (g)	Plant height (cm)	Productive tillers number
Non-saline										
Late-summer										
Mean	50.4	25	16.1	19.9	31.7	51.6	19.3	6.7	135.1	2.6
Range	28.5–67.5	6.1–46.7	2.9–33.6	5.4–34.2	11.9–52.1	19.0–79.2	11.3–30.3	4.7–10.7	68.8–194.4	1.0–5.6
h^2(bs)	0.93	0.45	0.44	0.46	0.57	0.47	0.76	0.67		
Early-summer										
Mean	50.6	30.2	20.3	13	28.8	41.8	19.3	7.5	125.7	3.1
Range	30.5–63.0	8.0–64.1	2.9–44.6	2.9–23.3	10.4–61.9	13.3–82.6	9.4–31.9	3.8–12.9	74.0–192.3	1.3–5.5
h^2(bs)	0.77	0.52	0.53	0.51	0.65	0.62	0.87	0.80	0.81	0.58
Rainy										
Mean	46.7	34.6	19.1	24.9	42.9	67.9				
Range	30.3–58.3	12.4–76.3	3.1–46.8	6.2–39.4	12.0–83.3	18.1–115.5				
h^2(bs)	0.90	0.56	0.45	0.24	0.71	0.57				
Saline										
Late-summer										
Mean	64.6	5.8	2.2	12.2	11.1	23.4	15.4	4.1	76.5	1.6
Range	32.3–102.5	1.7–11.1	0.8–3.8	2.8–21.3	0.9–21.2	4.5–40.4	7.3–29.2	1.6–7.7	33.7–125	0.5–5.4
h^2(bs)	0.98	0.54	0.65	0.70	0.84	0.80	0.81	0.97	0.87	0.81
Early-summer										
Mean	64.4	7.1	4	7.9	7.7	15.6	14.2	5.1	59.8	2.3
Range	33.3–103.0	2.9–13.8	1.24–8.5	0.7–15.3	0.5–18.0	1.1–32.5	7.5–23.7	2.9–9.9	34.02–93.7	1.0–4.0
h^2(bs)	0.81	0.42	0.28	0.65	0.62	0.62	0.71	0.45	0.71	0.50
Rainy										
Mean	55.74	11.3	5.7	8.8	9.1	17.9				
Range	36.0–75.0	4.2–22.0	1.51–12.6	1.0–22.3	1.30–22.3	2.3–43.7				
h^2(bs)	0.78	0.32	0.31	0.66	0.40	0.53				

Broad sense heritability $h^2(bs): \sigma_g^2 / \left(\sigma_g^2 + \sigma_e^2\right)$

Fig. 2 Regression representation of grain yield in saline and non-saline condition in late, early summers and rainy seasons

Table 5 Correlation coefficient under non-saline and saline conditions for 20 inbred lines and 4 testers for days of flowering, grain yield, shoot biomass and 1000-grain weight presented combine for summer seasons (late and early) and separately for the rainy season

Traits	Summer season				Rainy season		
	Days of flowering	Grain yield	Shoot biomass	1000-grain weight	Days of flowering	Grain yield	Shoot biomass
Days of flowering	1.00	− 0.01 ns	0.18*	− 0.06 ns	1.00	0.19 ns	0.04 ns
Grain yield	− 0.30***	1.00	0.46***	0.31***	− 0.39***	1.00	0.51***
Shoot biomass	0.46***	− 0.05 ns	1.00	− 0.04 ns	0.49***	− 0.07 ns	1.00
1000 grain weight	− 0.30**	0.49***	− 0.1 ns	1.00			

Thousand-grain weight not recorded in rainy season

ns non-significant

* Significant at $P = 0.05$; ** significant at $P = 0.01$; *** significant at $P = 0.001$

recorded indicating that late flowering reduced the GY under salinity-promoted vegetative growth. The delay in flowering time negatively affected GY under salinity independent of the season. The delay in flowering under P deficiency or Na^+ toxicity lead to high yield, which could offer screening large number of entries [28].

Selection of Contrasting Parents

Based on agronomic assessment (DOF, GY, shoot biomass and TGW) under saline and non-saline conditions, mapping population-inbred parent pairs having the maximum contrast for GY under saline conditions were selected

(Table S1). Besides these criteria, pairs of parents were chosen with a small range of flowering time (mean ± SD), since this trait displayed a negative relationship with GY. Based on these parameters, following contrasting pairs were selected for further studies; PRLT 2/89-33 and H 77/833-2, ICMB 841B-P3 and 863B-P2 and W 504-1-P1 and P 310-17-B along with two elite B lines 89111B-P6 and ICMB 90111-P2. However, the pair of inbred parent ICMB 841B-P3 and 863B-P2 was found flowering early with high GY but lesser than PRLT 2/89-33 and H 77/833-2 under non-saline conditions. This pair showed three-fold difference for GY in late-summer and two-fold in remaining two seasons under salinity. Interestingly, 863B-P2 displayed low reduction in GY and shoot biomass under salinity compared to non-saline conditions and higher TGW than 841B-P3. The study lead to the identification of salt tolerant lines such as ICMB 841B-P3, PRLT 2/89-33, 863B-P2 and W 504-1-P1 which may be further utilized for developing QTLs. But, Dewey [29] cautioned that a genotype whose yield is not much affected by salinity may still outperform by a high yielding genotype which may lose 50% of its yield under saline conditions. If salt tolerant genotype is intrinsically low-yielding, then it is unlikely to impress a farmer unless the absolute yield is adequate [30], therefore genotypes should be judged by the performance under salinity in relative terms with non-saline conditions.

Conclusions

Large variations were noticed in diverse mapping population parents developed at the ICRISAT and sufficient enough for the selection of mapping population parents for QTL analysis. Inbred performance under salinity has been found not related with performance under non-saline conditions for GY. Therefore, screening based on vegetative growth does not provide the correct picture of genotypic performance in field. DOF and GY were found as important and correlated traits under salinity. Evaporative demand of environment is affecting the response to salinity. In the present study, lines ICMB 841B-P3, PRLT 2/89-33, 863B-P2 and W 504-1-P1 were found as salt tolerant. Testing the recombinant inbred lines of these parents may lead to identification of some important salinity tolerance loci.

Compliance with Ethical Standards

Conflict of interest The authors declare no conflict of interest for publication of this manuscript.

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