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Effect of saline water irrigations on physiological, biochemical and yield attributes of dual purpose pearl millet (*Pennisetum glaucum*) varieties

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ABSTRACT

A split-plot experiment with four replicates on performance of dual purpose pearl millet (*Pennisetum glaucum* L.) under different saline water irrigation was conducted during *kharif* 2015 at ICAR-CSSRI experimental farm, Nain, Panipat. The experiment was conducted with 4 main-plot treatment consisting of different levels of saline irrigation water [Normal(~0.6 dS/m) and three levels of saline irrigation (ECiw 3, 6 and 9 dSm^{-1})] and two sub-plot treatments of varieties [ICMV-15111 and AVKB-19]. AVKB-19 variety was found robust and more stable than ICMV 15111 in terms of physiological attributes (relative water content, membrane injury and chlorophyll content) at all three periodic observations taken at 50 days after sowing (at 1st cut), 30 days and 60 days after 1st cut. Among biochemical attributes, total soluble sugars, proline, epicuticular wax content increased with increasing stress levels where as protein content decreased with increasing level of salinity stress. AVKB-19 variety accumulated significantly higher osmolyte content than ICMV-15111. Irrespective of salinity treatments, Na^+ content (shoot and root) was recorded gradually high with ICMV-15111 than AVKB 19 at all study stages. AVKB 19 variety presented significantly higher shoot and root K^+ content than ICMV 15111. Data on green fodder yield showed significant differences between two cultivars across different saline water levels. The AVKB-19 variety produced significantly higher mean green fodder yield (27.96 t/ha) as compared to ICMV-15111(25.51 t/ha). The AVKB-19 resulted into significantly maximum DM yield at the 1st as well as in 2nd cut (5.67 and 5.09 t/ha) over ICMV-15111. AVKB-19 variety produced significantly higher (16.26%) grain yield of 1.93 t/ha as compared to 1.66 t/ha in ICMV-15111. The magnitude of reduction (%) with increasing salinity over good quality irrigation water was observed 10.14, 20.74 and 37.33 with 3.0, 6.0 and 9.0 dS/m salinity water, respectively. Results concluded that AVKB-19 variety of pearl millet proved superior and may be adapted as a choice for getting higher grain as well as green fodder yield with better physiological and biochemical responses under saline environment.

Key words: Pearl millet, Physiological attributes, Saline water, Yield

Presence of salt affected soils on the earth surface is as old as the history of mankind (Sharma and Choudhari 2012). It has now become a very serious threat for crop production (Ashraf and Wu 1994, Rengasamy 2006) particularly in arid and semi-arid regions, which constitute about one third of the world's land surface. At present about 6.73 million hectare (Mha) salt affected soils exist in India. Out of total salt affected soils 2.96 Mha are saline and remaining 3.77 Mha are characterized as sodic soils (Sharma and Singh 2015). The scarcity of water in association with high salinity is now major problem hindering crop production in salty lands. Poor quality groundwater extensively occupied (32-84%) in arid and semi-arid parts of India (CGWB 1997) and its indiscriminate use poses a serious threat to sustainability of the natural resources and environment. States like Rajasthan and Haryana do not have sufficient surface water resources for meeting irrigation water requirement and thereby depend on poor quality ground water which is 68% and 55%, respectively in the two states (Lal *et al.* 2008). The farmers of these states have been over exploiting groundwater for supplementing the limited surface water resources. Soil and water salinity cause several physiological disorders in plants, connected with the abnormal concentration of ions in the rhizospheric environment; these can range from a cytotoxic and denaturizing effect of the ions themselves (Bernstein 1975) to osmotic stress (Greenway and Munns 1980, Yeo 1983) and alteration of the ion uptake balance (Rains 1972, Flowers and Lauchli 1983). Consequently, it has become

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imperative to search for suitable crop/genotype alternatives and develop ecologically sustainable and economically sound production systems that can use poor quality water and withstand drought on saline lands.

In this context, Pearl millet (*Pennisetum glaucum* L.) is a promising dual purpose, short duration, quick growing crop with good salinity tolerant characteristics, therefore has an advantage over others cultivated fodder in salt affected areas. Pearl millet has been reported to have high tolerance to salinity and drought thus it can serve as an important crop to ensure good quality fodder for animals in the arid and semi-arid regions of India and elsewhere in the world under similar agro ecologies (Kulkarni *et al.* 2006, Patel *et al.* 2008). Several researchers documented that Pearl millet showed minimum yield reduction under saline environment, thus demonstrating its tolerant nature towards salinity. Therefore, present study was conductively undertaken to access the physiological, biochemical and yield responses of pearl millet varieties. By understanding the traits, affecting the production potential will help to deliver a suitable alternative genotype that could be helpful to use the poor quality water without compromising the soil health and yield.

MATERIALS AND METHODS

The present study was carried out at ICAR-CSSRI experimental farm, Haryana, India. The climate of the area is semi-arid, with a mean annual rainfall of 678 mm (70-80% of which received during July-September) with the mean annual USWB-open pan evaporation (ET_p) of 1598 mm. The meteorological phenomenon occurred during study period (July-November) are presented in Table 1. The Initial Soil Status of experimental site and salinity build-up in each plot after harvest is presented in Table 2.

The experiment was designed in split-plot arrangements with four replications. The experiment was conducted with 4 main-plot treatment consisting of levels of irrigation water [normal (~0.6 dS/m) and three levels of saline irrigations (ECiw 3, 6 and 9 dS/m)] and two sub-plot treatments of varieties [ICMV-15111 and AVKB-19]. To supplement the total rainfall and to maintain 1.2 ID/CPE ratio, 240 mm water was applied through irrigation. To fulfil this purpose check basin method was adopted. Each experimental unit consisted of 4.5 m × 4.5 m plots. The field was deep ploughed by chisel plough to break the hard pan below the plough layer before start of the experiment. The pearl millet cv.

Table 2 Initial soil status and salinity build-up in each plot after harvest

Initial soil status		Depth wise (cm) salinity levels after harvest (ECe) (dS/m)		
Texture	Sandy loam	Control 1 (0.6)	0-15	5.31
Walkley-Black C (%)	0.3		15-30	5.32
Ece(dS/m)	6.65	3	0-15	6.17
pH	8.3		15-30	6.18
$KMnO_4$ Oxidizable N (kg/ha)	130.4	6	0-15	6.83
0.5 M $NaHCO_3$ extractable P (kg/ha)	11.6		15-30	6.85
1N NH_4OAC extractable K (kg/ha)	248.4	9	0-15	7.58
			15-30	7.61

AVKB-19 and ICMV-15111 were sown with a seed rate of 12 kg/ha during second fortnight of July in 2015 with a row spacing of 30 cm and plant to plant distance at 10 cm. A common dose of nutrients amounting 120 kg N + 60 kg P_2O_5 + 40 kg K_2O were applied in all treatments through urea [$CO(NH_2)_2$], di-ammonium phosphate($(NH_4)_2HPO_4$) and sulphate of potash (K_2SO_4). The 1/3rd N and whole P_2O_5 and K_2O was applied as basal, while remaining 2/3rd N was top dressed as urea in two equal splits at 1st cutting and 30 days after 1st cutting. In view of best weed management, two hands weeding at 20 DAS and 30 days after 1st cutting was done to control weeds. The first cut of crop was taken at (50 DAS) 8-10 cm above the ground level. Then the crop was left for grain production. The final cut was taken at ~110 DAS for grain purpose. Three observations for physiological parameters were studied, at 50 DAS (1st cut); second observation at 30 days after 1st cut (80 DAS), and third /final observation at 60 days after 1st cut (110 DAS) from representative (tagged) plants from each plot. *Relative water content* (RWC) was measured following the procedure described by Weatherley (1950). Third fully expanded leaf from the top of the plant sample was detached from the shoots between 9:00 to 10:00 AM, quickly sealed in humified polythene bags, and transported on ice to the laboratory. Sand was removed with the help of a soft brush. Then the leaves were weighed immediately to take their fresh weight. The leaves were kept in petri dishes

Table 1 Meteorological observations during study period

Month	Temperature (°C)		Relative humidity (%)		Total rainfall (mm)	Total evaporation (mm)
	Mean Maximum	Mean Minimum	Mean Maximum	Mean Minimum		
20-31 July	32.92	25.4	86.08	69.67	11.4	71.2
1-31 August	32.68	25.1	92.97	73.2	70.2	103.6
1-30 September	33.51	22.8	91.5	59.2	46.8	106.5
1-31 October	32.15	17.77	91.67	46.3	6.4	90.7
1-15 November	27.73	14.19	91.56	47.06	9.2	30.3

full of distilled water for 3 h.

After that the leaves (fully turgid) were weighed again and then kept in oven at 85°C for 72 h till a constant dry weight. These three weights were used to calculate RWC (%) of leaves according to the following formula.

$$\text{RWC (\%)} = \frac{\text{Fresh weight–Dry weight}}{\text{Turgid weight–Dry weight}} \times 100$$

Membrane injury was estimated according to the method of Dionisio-Sese and Tobita (1998). The leaf discs of 1 cm diameter were kept in 20 ml vials containing 10 ml deionized water at 25°C. After five hours, the electrical conductivity (EC) of the surrounding solution was measured by the Water Analysis Kit (Naina, India Ltd., NDC 732) and designated as ECa. Then the samples were kept in boiling water bath for 50 min to achieve total killing of the tissue. After cooling, the EC of the solution was again measured and designated as ECb. The relative stress injury (RSI)/ Membrane stability index was calculated as follows and expressed in percentage:

$$\text{RSI (\%)} = \frac{\text{ECa}}{\text{ECa + ECb}} \times 100$$

The *chlorophyll content* was determined using DMSO (Dimethyl sulphoxide) as described by Hiscox and Israelstam (1979) using dimethyl sulfoxide (DMSO). Third leaf from the top of plant was detached and weighed, 200 mg of this leaf material was transferred into a test tube containing 5 ml of DMSO. The test tube was then placed into oven at 60°C for about 2 h or more (if required) to facilitate the extraction of pigment. After 2 h and attaining the room temperature, the absorption was recorded at 645 and 665 nm on spectrophotometer. Absorbance at respective wavelengths was also recorded in DMSO and used as blank. Calculations for different pigments were made as per the procedure described by Welburn (1994).

Chl ‘a’ (µg/ml) = 12.19 A_{665} – 3.45 A_{645}

Chl ‘b’ (µg/ml) = 21.99 A_{645} – 3.32 A_{665}

Total chlorophyll = Chl ‘a’ + Chl ‘b’.

Proline content was estimated using the method of Bates *et al.* (1973). Three hundred mg of leaves was homogenized in 5 ml of 3% sulphosalicylic acid and then centrifuged at 5000 rpm for 15 minutes and supernatant was taken. Two ml of supernatant was taken and 2.0 ml reagent acid ninhydrin + 2.0 ml acetic acid was added. This mixture was then kept in boiling water bath for 1 hr at 100°C and thereafter reaction was terminated by keeping tubes in ice-bath. Then 4.0 ml of toluene was added. After vigorous shaking, the upper organic phase was taken after attainment of room temperature and absorbance was recorded at 520 nm by using toluene as blank. A standard curve was prepared by using graded concentration of proline in 3% sulphosalicylic acid. The proline content was expressed as µg/g FW.

Colorimetric method (Ebercon *et al.* 1977) was used for *epicuticular wax* deposition. Plant leaf blade samples, were taken from different treatment for wax analysis. The colorimetric method is based on the color change produced due to the reaction of wax with acidic $K_2Cr_2O_7$ reagent. The reagent was prepared by mixing 40 ml deionized water and 20 g powdered potassium dichromate. The resulting slurry was mixed vigorously with 1 litre conc. sulfuric acid and heated (below boiling point) until clear solution was obtained. The individual sample of pearl millet was immersed in 15 ml redistilled chloroform for 15 second. The extract filtered and evaporated on a boiling water bath until smell of chloroform disappeared. After cooling, 12 ml of deionized water was added. Several minutes were allowed for colour development and cooling and then optical density of the sample was read at 590 nm.

Total soluble carbohydrate was determined with the method of Yemm and Willis (1954). The extraction of total soluble carbohydrate was done according to the method of Barnett and Naylor (1966). Fifty mg dry sample of leave was homogenized separately with 5 ml of 80% (v/v) ethanol. Like this, third extraction was done with 60% (v/v) ethanol. The supernatant from different extractions was pooled and evaporated to dryness in a boiling water bath. Then total volume of extract was made to 10 ml with distilled water. The extract so obtained was used for estimation of total soluble carbohydrate and free amino acids. The residue was used for total soluble protein extraction. Anthrone reagent was prepared by dissolving 0.2 g anthrone in 100 ml concentrated H_2SO_4. After that 0.5 ml aliquot of the extract was taken in a test tube and mixed with 4.0 ml of the anthrone reagent. The mixture was heated in boiling water bath for 10 minutes. After cooling, absorbance was recorded at 620 nm. Glucose (10-100 µg) was taken as standard.

Total soluble proteins were determined by Bradford (1976) method using supernatant. Dissolve 100 mg of Coomassie Brilliant Blue G-250 (CBBG-250) in 50 ml of 95% ethanol. Add to this solution in 100 ml 85% (w/v) phosphoric acid and make final volume 200 ml with double distilled water. Filter through whatman filter paper No. 1 (if any ppt. occurs), diluted to a final volume of 1 litre and stored at 4ºC in amber colour bottle. To 25 µl of the supernatant, 5 ml of the coomassie Brilliant Blue G-250 reagent was added and mixed either by inversion or vortexing. The optical density (OD) was measured at 595 nm after 2 minutes and before 1hr against reagent blank. Standard curve was prepared using graded concentration of bovine serum albumin.

For *Na^+ and K^+ content*, 100 mg of oven dried and well ground plant material was digested with 10 ml of HNO_3 : $HClO_4$ (3:1) di-acid mixture and readings were taken with flame photometer (PFP7, Jenway, Bibby Scientific, UK) using standard NaCl and KCl. Yields of 1x1 m^{-2} was recorded and converted in per hectare. All data recorded were analyzed with the help of analysis of variance (ANOVA) technique using the SAS (Version 9.3, SAS Institute Inc., Cary, NC, USA). The least significant test was used to decipher the main and interaction effects of treatments at 5% level of significance ($P<0.05$).

RESULTS AND DISCUSSION

Perusal of the physiological and biochemical data indicated significant differences among the varieties and treatments with respect to different parameters.

Relative water content (RWC) (%): It is the appropriate measure of plant water status in terms of the physiological consequence of cellular water deficit. Statistical analysis of data indicated that salt stressed pearl millet plants exhibited significant differences for RWC through increasing salinity of irrigation water (Fig 1). The decrease in relative water content (RWC) could be attributed to early symptoms of stress conditions. The maximum relative water content (%) was observed from good quality irrigation water (87.7, 86.135 and 79.32 at 50 DAS, 30 DA 1st cut and 60 DA 1st cut respectively) and the minimum (68.87, 61.65 and 58.04 at 50 DAS, 30 DA 1st cut and 60 DA 1st cut respectively) from irrigation water having 9.0 EC. The minimum significant percentage reduction 10.74, 11.19 and 5.29% was observed at 3.0 dS/m in comparison to good quality water at all periodic observations, whereas the maximum significant was noticed 21.08, 28.42 and 26.82% from irrigation water having 9.0 EC in comparison to good quality water at all sequential periodic observations. Irrespective of water quality, the mean maximum performance with respect to this parameter was found from genotype AVKB-19 at all sequential periodic observations than ICMV 15111 but the difference between genotypes was non-significant at all the three stages. This reduction of RWC in stressed plants might be associated with a decrease in plant vigour (Kumar *et al.* 2016). High salt concentration in root zone causes osmotic stress which restricts water absorption by the plants and causes cellular dehydration, seems to be primarily responsible for decrease in RWC (Greenway and Munns 1980). These results agree with the findings of Netondo *et al.* (2004) in sorghum, and Vijayalakshmi *et al.* (2012) in pearl millet.

Total Chlorophyll content (µg/ml of FW): Chlorophyll concentration has been known as an index for evaluation of source (imparts lush green colour to the fodder, improves radiation absorption and quality), therefore decrease in concentration can be considered as a stomata non-limiting factor under stress condition. The observations with respect to chlorophyll content as affected by different irrigation water salinity levels are presented in Fig 2. Mean performance of the genotype AVKB 19 (51.91µg/ml and 39.68 µg/ml at 30 DA 1st cut and 60 DA 1st cut, respectively) were found significantly higher than that ICMV-15111 (45.87 and 37.35 µg/ml) at 30 DA 1st cut and 60 DA 1st cut, respectively), but in case of 1st observation at 50 DAS, there was statistically no difference among both genotypes. The significantly maximum chlorophyll content was found with good quality irrigation water whereas minimum chlorophyll content recorded with EC 9.0 dS/m of irrigation water. Similarly significantly variation were noticed by every increasing levels of salinity at 50 DAS and 60 DA 1st cut. The effects of salt stress on chlorophyll degradation, presumably due to increased activity of the enzyme chlorophyllase (Kumar *et al.* 2016), were characterized as the yellowing of leaves which failed to produce the optimum amounts of photosynthate leading to reduced plant growth and vigour.

Membrane injury index (MI) (%): Electrolyte leakage (an indicator of membrane damage) increased with increase in saline irrigation. Leaves of pearl millet plants irrigated with good quality water exhibited lowest (8.18, 8.56 and 10.42% at 50 DAS, 30DA and 60 DA 1st cut, respectively) membrane injury than all higher level of saline water irrigation (Fig 3).

The maximum MI (24.11, 26.53 and 35.8%) was noticed from irrigation water having 9.0 EC at respective sequential observations. While investigating the genotypic potential for membrane stability, AVKB 19 was found hardier and more stable than ICMV-15111 because it had lower injury index at all three periodic observations taken at 50 DAS, 30 and 60 DA 1st cut. Salt stressed plants exhibit damage

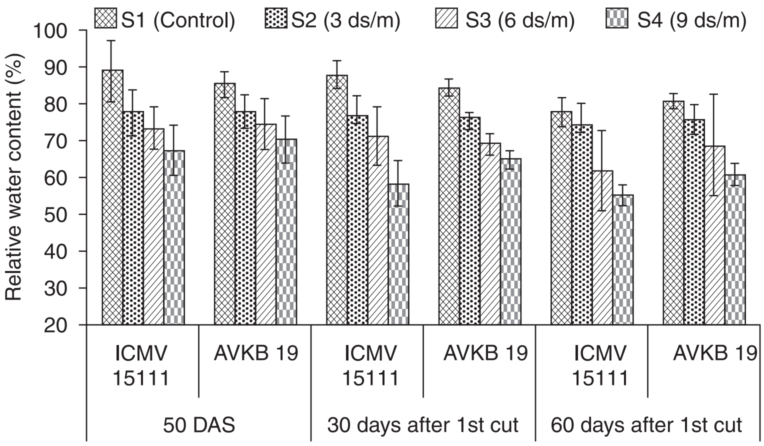

Fig 1 Effect of saline water irrigations on relative water content (%) in dual purpose pearl millet varieties.

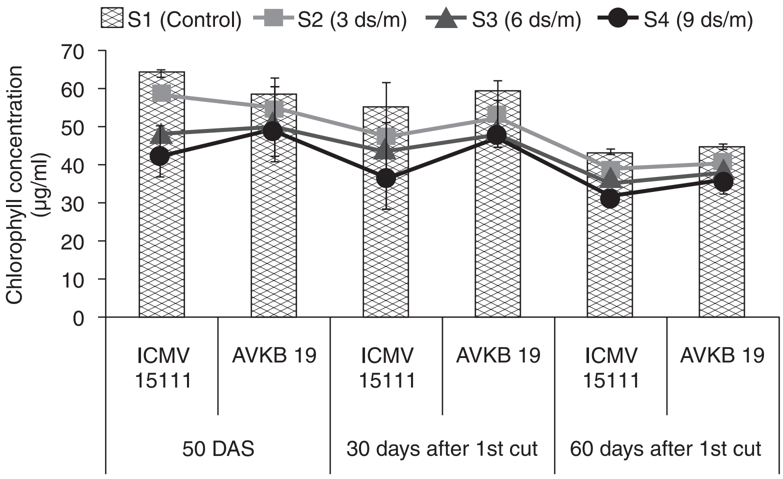

Fig 2 Effect of saline water irrigations on total chlorophyll concentration (µg/ml) in dual purpose pearl millet varieties.

of lipid membranes which often results in increased cell permeability and electrolyte leakage from cells (Kumar *et al.* 2015).

Biochemical attributes: The accumulation of compatible solutes such as soluble sugars, proline (key osmolytes that contribute to osmotic adjustment) etc. which are non-toxic at higher concentrations to cytoplasmic functions allow additional water uptake from the environment and turgor maintenance (Sairam *et al.* 2002, Misra and Gupta 2005).

The accumulation of soluble carbohydrates in plants has been widely reported as a response to salinity or drought, despite a significant decrease in net CO_2 assimilation rate (Murakezy *et al.* 2003).

Total soluble sugars (TSS) (mg/g DW): The mean minimum TSS was recorded with good quality water at all three periodic observations taken at 50 DAS (7.22 mg/g DW), 30 DA 1st cut (10.89 mg/g DW) and 60 (13.72 mg/g DW) DA 1st cut, whereas the mean maximum TSS was recorded with irrigation water having 9.0 EC at 50 DAS (22.43 mg/g DW), 30 DA 1st cut (25.41 mg g DW) and 60 (38.35 mg g DW) DA 1st cut (Fig 4). The significant increase in TSS was found at all three sequential periodic observations from 9.0 EC of irrigation water over good quality water. When comparing the varietal difference for TSS observations at 50 DAS and 60 DA 1st cut ICMV-15111 found to contain 13.68 mg/g DW and 30.91 mg/g DW TSS being significantly higher than AVKB-19 having 10.56 mg/g DW and 20.46 mg/g DW.

Sugars are the source of energy and carbons needed for adaptive or defensive responses to stress. The higher sugar content in the extreme salinity levels as an additional mechanism to prevent salt injury (Kumar *et al.* 2016, 2017). The changes in the carbohydrate are regulated primarily by K^+ and Cl^- ions. Similarly increasing sugar content at higher salinities has been reported in several halophytes (Prado *et al.* 2000, Ashraf and Harris 2004).

Proline content (μg/g FW): It is a potent osmoregulator molecule and counteracts the adverse effects of toxic salt ions in cell vacuoles, contributes to membrane stability and mitigates the effect of NaCl on cell disruption. The mean maximum proline content was recorded from 9.0 EC of irrigation water 3.78, 3.59 and 4.47 at 50 DAS, 30 and 60 DA 1st cut, respectively in comparison to minimum of 0.92, 1.2 and 1.58 at 50 DAS, 30 and 60 DA 1st cut, respectively from good quality irrigation water. The increase in proline content was found non-significant up to 6.0 EC at 50 DAS and up to 3.0 EC of irrigation water at 30 DA 1st cut stage, whereas the increase in percentage was found significant from every increasing level of salinity at 60 DA 1st cut stage (fig 5). When comparing the varietal potential against irrigation water salinity, variety ICMV-15111 found containing significantly higher proline content than AVKB-19 at 30 DA 1st cut stage, but non-significantly higher at 50 DAS. In case of 60 DA 1st cut stage, variety

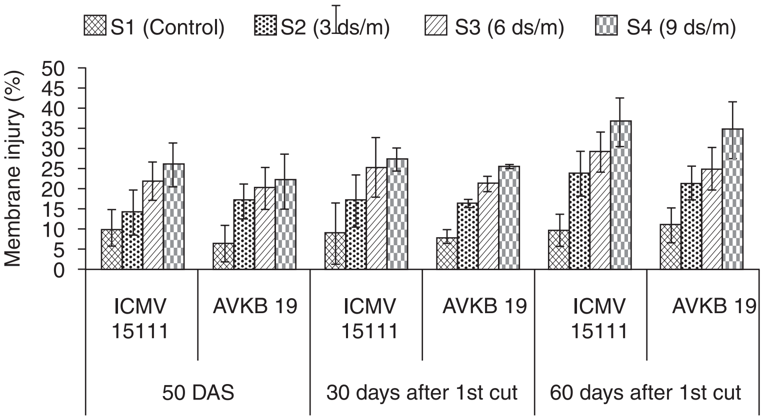

Fig 3 Effect of saline water irrigations on membrane injury (%) in dual purpose pearl millet varieties.

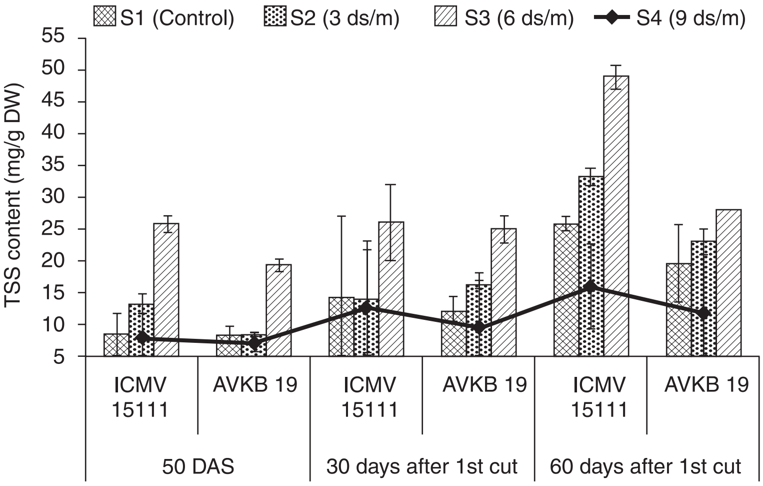

Fig 4 Effect of saline water irrigation on total soluble sugar content (mg/g DW) in dual purpose pearl millet varieties.

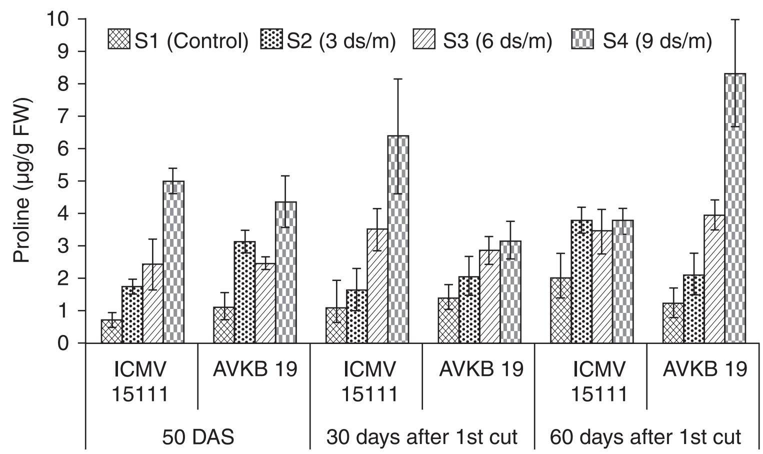

Fig 5 Effect of saline water irrigation on proline content (μg/g FW) in dual purpose pearl millet varieties.

AVKB-19 was found to contain significantly higher proline content than variety ICMV-15111. Accumulation of these osmolytes can also improve plant tolerance by protecting and stabilizing membranes and enzymes during stress conditions (Ashraf and Harris 2004, Singh *et al.* 2015, Lata *et al.* 2017).

Total soluble protein content(TSP)(mg/g): The proteins that accumulate under stress conditions may provide a storage form of nitrogen that is re-utilized in post-stress recovery and also play a role in osmotic adjustment. The mean maximum total soluble protein content (mg/g) from good quality water with 15.26, 29.68 and 37.84 at 50 DAS, 30 and 60 DA 1st cut, respectively, whereas mean minimum with 9.0 EC of irrigation water containing 5.99, 11.47 and 24.52 at 50 DAS, 30 and 60 DA 1st cut, respectively (Table 3). The reduction in TSP was found statistically significant from each increasing salinity level of irrigation water over good quality water irrigation at all three periodic observations. When comparing cultivars responses for total soluble protein content, AVKB-19 found slightly high than ICMV-15111 at all three observations. Several researchers showed in earlier findings that soluble protein contents of leaves decreased in response to salinity (Parida *et al.* 2002, Wang *et al.* 2000, Agastian *et al.* 2000, Kumar *et al.* 2016, 2017).

Epicuticular wax load (EWL) (mg/g DW): It is an effective component of abiotic stresses resistance (avoidance mechanism) in pearl millet. A varying response in EW accumulation was seen among the genotypes (Table 4). When comparing the genotypic potential for epicuticular wax load, ICMV-15111 was seen to accumulate epicuticular wax in significantly very high amounts (2.58 mg/g DW, 3.3 mg/g DW and 3.76 mg/g DW at 50 DAS, 30 and 60 DA 1st cut, respectively) as compared to AVKB 19 with 1.72 mg/g DW, 2.91 mg/g DW, 2.63 mg/g DW at 50 DAS, 30 and 60 DA 1st cut respectively. The mean minimum quantity of EW was found when plants are irrigated with good quality water (0.87 mg/g DW, 1.29 mg/g DW and 1.68 mg/g DW at 50 DAS, 30 and 60 DA 1st cut, respectively, whereas the mean maximum amount was obtained (3.99 mg/g DW, 5.03 mg/g DW and 5.1 mg/g DW at 50 DAS, 30 and 60 DA 1st cut respectively). Present findings have shown that with progress of time pearl millet showed higher deposition of EW because under stress conditions, stomata closed and lead to decreased cuticular permeability of water loss with higher deposition of EW (Blum 1988). Kumar *et al.* (2016) also reported increased EW load with salt and sodic stress in halophytes.

Table 3 Effect of saline water irrigation on total soluble protein content (mg/g DW) in dual purpose pearl millet varieties

Irrigation water quality (dS/m)/Varieties	Total soluble protein content (mg/g DW) at 1st cut (50DAS)			Total soluble protein content (mg/g DW) 30 DA 1st cut			Total soluble protein content (mg/g DW) at grain harvest stage		
	ICMV 15111	AVKB 19	Mean	ICMV 15111	AVKB 19	Mean	ICMV 15111	AVKB 19	Mean
Control (0.6)	14.43	16.10	15.26	28.38	30.98	29.68	34.94	40.75	37.84
3	10.32	11.19	10.75	22.40	22.29	22.34	32.44	35.00	33.72
6	7.71	7.08	7.4	19.77	17.49	18.63	28.55	29.98	29.26
9	5.36	6.63	5.99	11.56	11.38	11.47	25.57	23.47	24.52
Mean	9.46	10.25		20.53	20.54		30.37B	32.3A	

Factor	T	V	T×V	V×T	T	V	T×V	V×T	T	V	T×V	V×T
CD (P=0.05)	2.74	NS	NS	NS	2.28	NS	NS	NS	3.94	1.63	3.26	4.57

Table 4 Effect of saline water irrigation on epicuticular wax content (mg/g DW) in dual purpose pearl millet varieties

Irrigation water quality (dS/m)/Varieties	Epicuticular wax content at 1st cut (50 DAS)			Epicuticular wax content at 30 DA 1st cut			Epicuticular wax content at 60 DA 1st cut		
	ICMV 15111	AVKB 19	Mean	ICMV 15111	AVKB 19	Mean	ICMV 15111	AVKB 19	Mean
Control (0.6)	1.12	0.63	0.87	1.31	1.27	1.29	1.3	2.06	1.68
3	2.08	1.08	1.58	2.66	2.05	2.35	3.24	1.96	2.6
6	2.80	1.53	2.17	4.31	3.16	3.73	4.64	2.95	3.8
9	3.56	3.63	3.99	4.90	5.15	5.03	5.85	4.34	5.1
Mean	2.58	1.72		3.3	2.91		3.76	2.63	

Factor	T	V	T×V	V×T	T	V	T×V	V×T	T	V	T×V	V×T
CD (P=0.05)	0.75	0.75	NS	NS	0.47	0.36	0.71	0 .69	0.47	0.36	0.71	0.69

*T = Treatments (Irrigation at ASM Level), V = Varieties, T × V = Treatments at the same level of Varieties and V × T = Varieties at same level of Treatments, CD Critical difference, DAS Days after sowing

Ionic relations

Sodium content (% DW): Perusal of data (Fig 6) on Na^+ content of shoots indicated that Na^+ content significantly increased with every increasing level of salinity at all periodic observations, except at 1st with good quality water treatment. The maximum Na^+ concentration was recorded with 9.0 EC of irrigation water (0.21, 0.64 and 0.94) whereas minimum with plot subjected to good quality water (0.06, 0.16 and 0.18) at 1st, 2nd and 3rd observation respectively. Similarly, the minimum root Na^+ was recorded with good quality water treatment containing (0.78, 2.9 and 3.6) and maximum recorded with irrigation water treatment of 9.0 EC (0.78, 2.9 and 3.6) at 1st, 2nd and 3rd observation respectively (Fig 6).

Potassium content (% DW)

The maximum K^+ content in shoot was recorded with good quality water treatment containing (2.98, 5.16 and 5.25) whereas minimum in irrigation water treatment of 9.0 EC (1.2, 1.79 and 1.5) at 1st, 2nd and 3rd observation respectively (Fig 7). The highest K^+ content in root was recorded from good quality water treatment having (4.13, 6.18 and 7.26), whereas minimum with irrigation water treatment of 9.0 EC containing (1.66, 3.37 and 3.96) at 1st, 2nd and 3rd observation respectively. A significant reduction was observed with every increasing salinity level of irrigation water. AVKB-19 variety had significantly higher shoot and root K^+ content than ICMV-15111 variety. The higher affinity for K^+ transporters may also act as lower affinity for Na^+ transporters under saline environment, which may reduce K^+ uptake. Our results are supported with the findings of Liu *et al.* 2000 and Radhouane *et al.* 2013.

Yield

Green fodder yield: The main effect of irrigation water salinity on green fodder yield of pearl millet has suggested that irrigation with good quality (0.6 dS/m) and low salinity (3.0 dS/m) water produced the maximum green fodder yield of 37.55 and 34.74 t/ha, respectively at 1st cut at 50 DAS (Table 5). It was minimum, i.e. 23.97 t/ha (36.17 % reduction from good quality water) with high salinity water (9.0 dS/m) irrigation.

The statistical analysis of results on performance of pearl millet cultivars showed that AVKB-19 produced 32.74 t/ha green fodder yield which was significantly higher as compared to 30.65 t/ha in case of ICMV-15111 on 1st cut taken at 50 DAS. Data on green fodder yield showed significant differences between two cultivars across different saline water levels. The variety AVKB-19 produced significantly higher mean green fodder yield (27.96 t/ha) as compared to ICMV-15111(25.51t/ha). The interaction between water quality and varieties was non-significant for both cuttings. Ghadiri *et al.* (2005) reported reduced uptake of water by salinity due to the high osmotic potential of the soil solution and high concentrations of specific ions that may cause physiological disorders in the plant tissues and reduced yields. The primary effect of high salinity of irrigation water generally renders less water available to plants in spite of the fact that some is still present in the root zone. This is because of the fact that osmotic pressure of the soil solution becomes more negative as the salt concentration increases. Apart from the osmotic effect of salts in the soil solution, excessive concentration and absorption of individual ions such as Na^+ may prove toxic to the plants and/or may retard the absorption of other

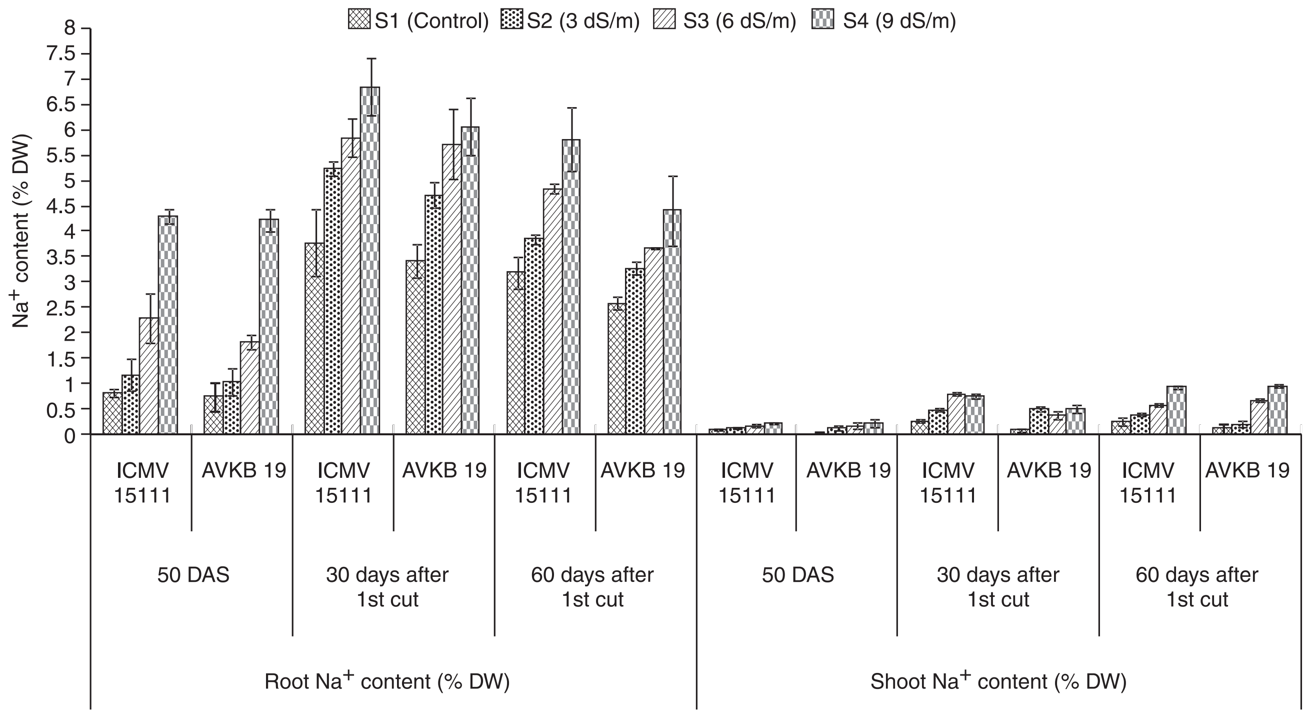

Fig 6 Effect of saline water irrigations on root and shoot Na^+ content (%) in dual purpose pearl millet varieties.

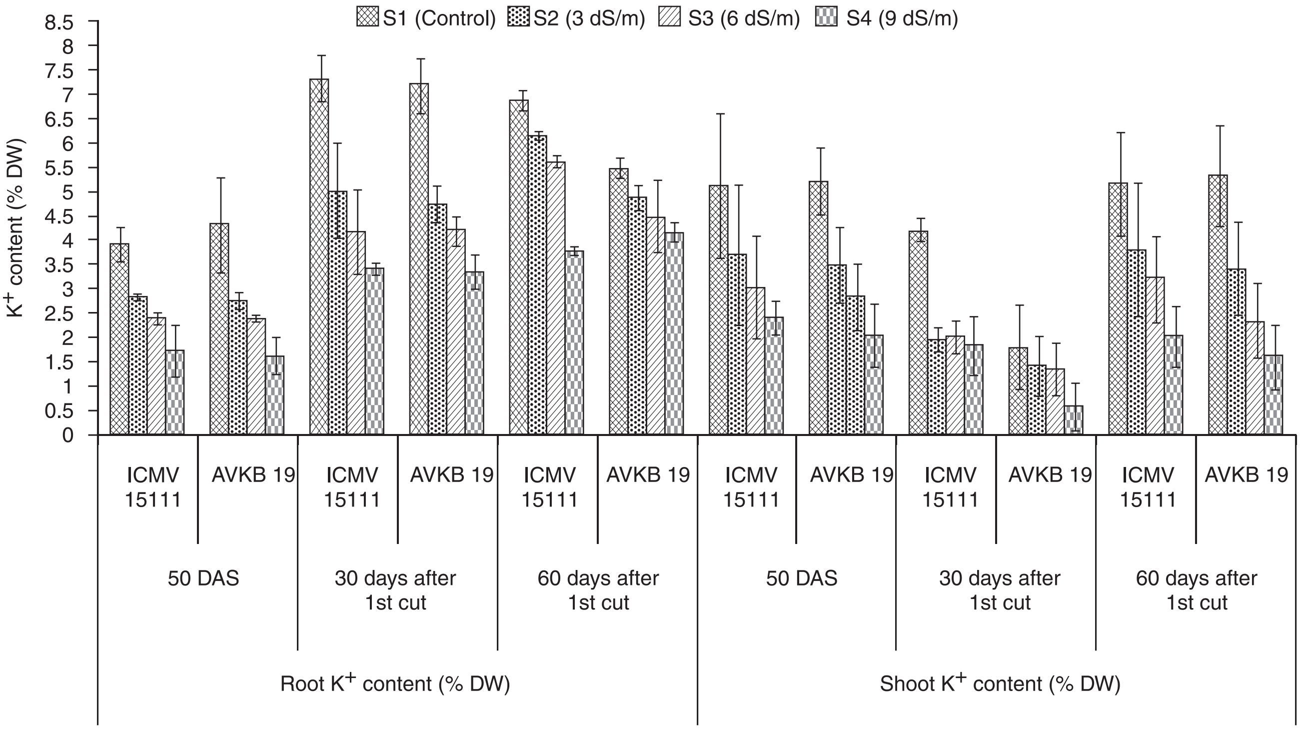

Fig 7 Effect of saline water irrigations on root and shoot K^+ content (%) in dual purpose pearl millet varieties.

essential plant nutrients (Qadir *et al.* 2008). Yadav *et al.* (2004), Nadaf *et al.* (2010), and Zhapayev *et al.* (2015) also reported similar findings in different crops.

Dry Matter yield: The mean maximum DM yields at 1st (5.89 t/ha) and 2nd cut (5.25 t/ha) were recorded with good quality irrigation water. Application of good quality water resulted into 23.34 and 13.69% higher DM yield at 1st and 2nd cutting, respectively over irrigation water of EC 9.0 dS/m. However, the irrigation water upto EC 3.0 dS/m at 1st cut and upto EC 6.0 dS/m at 2nd cut were found at par with good quality irrigation water (Table 5). This may be due to the fact that combined effect of rainfall and irrigation after 1st cut diluted the salt concentration and helped in leaching the excess salts from the root zone up to irrigation water EC 6.0 dS/m. The AVKB-19 resulted into significantly maximum DM yield at the 1st (5.67 t/ha) as well as 2nd cut (5.09 t/ha) over ICMV-15111. The supply of good quality water significantly enhanced the uptake of macro and micro nutrient by the plants which ultimately lead to higher DM yield due its beneficial effect on the photosynthetic efficiency of plants. Whereas the higher load of salt with increased level of salinity of irrigation water restricted the root growth of plants that in turn reduced the uptake of nutrients leading to leaf chlorosis that reduces the photosynthetic potential of crops which ultimately leads to lower DM yield. The similar results of lower DM yield with

Table 5 Effect of saline water irrigation on yield of pearl millet varieties

Treatments	Green fodder yield (t/ha)		Dry matter yield (t/ha)		Grain yield (t/ha)
	1st cut	2nd cut	1st cut	2nd cut	At 110 DAS
Salinity levels (dS/m)					
Control (0.6)	37.55a	31.50a	5.89a	5.25a	2.17a
3	34.74b	28.61b	5.69a	5.13a	1.95a
6	30.52c	25.55c	5.25b	4.88a	1.72b
9	23.97d	21.30d	4.51c	4.53b	1.36b
SEm (±)	0.99	0.94	0.18	0.19	0.12
CD (P=0.05)	2.06	1.95	0.38	0.40	0.24
Varieties					
ICMV-15111	30.65a	25.51a	5.00b	4.79b	1.66b
AVKB-19	32.74b	27.96a	5.67a	5.09a	1.93a
SEm (±)	0.70	0.66	0.13	0.14	0.08
CD (P=0.05)	1.46	1.38	0.27	0.29	0.17

increasing salinity levels were also reported by Kumari *et al.* 2014 and Farissi *et al.* 2018.

Grain yield: Variety AVKB-19 produced significantly higher (16.26%) grain yield of 1.93 t/ha as compared to 1.66 t/ha in ICMV-15111, and thus has potential to grow in saline environment. Significantly highest (2.17 t/ha) mean grain yield was recorded with good quality water in comparison to 1.95, 1.72 and 1.35 t/ha achieved with irrigation using 3.0, 6.0 and 9.0 dS/m salinity water, respectively (Table 5). The magnitude of reduction (%) with increasing salinity over good quality irrigation water was observed 10.14, 20.74 and 37.33 with 3.0, 6.0 and 9.0 dS/m salinity water, respectively. Decrease in grain yield by increasing salinity of irrigation water may be due to more negative water potential of soil solution causing reduced water and nutrient uptake consequently lower leaf area development in turn reduced net assimilates. Shani and Dudley (2001) also related the yield loss to reduced photosynthesis, high energy and carbohydrate expenses in osmoregulation, and interference with cell functions under saline conditions. Similar results for decreasing grain yields under saline environment were reported in pearlmillet by Meena *et al.* (2012).

Conclusion

Present results suggest that AVKB-19 genotype of pearlmillet proved superior and may be adapted as a choice for getting higher grain as well as green fodder yield with better physiological response as compare to the ICMV-15111 under saline environment in north-western region of India and elsewhere under similar agro-climatic conditions.

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