Paper ID: 1569
Theme 4. Biodiversity, conservation and genetic improvement of range and forage species
Sub-theme 4.1. Plant genetic resources and crop improvement

Salinity tolerance of forage range legumes during germination and early seedling growth

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Introduction

Salinity is one of the rising problems causing tremendous yield losses in many regions of the world especially in arid and semiarid regions. In India, about 5.95M ha areas were affected by salinity. Five states viz. Haryana, Punjab, Rajasthan, Gujarat and Andhra Pradesh accounts for 48% of the total salt affected soils of the country. Establishment of seedlings at early growth stages of crop plants as one of the most important determinants of high yield is severely affected by soil salinity. Increasing salinity levels significantly decreased germination parameters, shoot and root length, shoot and root fresh and dry weights of some forage sorghum cultivars (Kandil *et al.,* 2012).Therefore, in the present investigation three forage range legumes namely centro, clitoria and siratro were tested for their relative salt tolerance to increasing levels of salinity in those combinations of salts which nearly exist in the natural salt affected soils.

Materials and Methods

Centro (*Centrosema pubescens*), clitoria (*Clitoria ternatea*) and siratro (*Macroptilium atroperpureum*) seeds were collected from Indian Grass land and Fodder Research Institute, South Regional Research Station, Dharwad. The experiment was carried out under laboratory conditions in Department of Plant Physiology, University of Agricultural Sciences, in a completely randomized design with three replications. This study consists of experiment on seed inoculation with salinity as mixing the different salts (ratios 13:7:1:4) like NaCl, Na2SO_4, $MgCl_2$, and $CaSO_4$ at concentrations of 0 (distilled water as control), 4, 8, 12 and 16 ds m^{-1} on germination and early growth of three range legumes, differing in salt tolerance. The seeds were surface sterilized with 0.01% mercuric chloride (w/v) for 10 min to avoid fungal invasion, followed by washing with distilled water. For each plant species, 25 seeds for each of the five salt treatments were allowed to germinate on filter paper (Whatman No. 2) in a solution of the respective salt concentration. The number of germinant seeds was counted every day up to 10 days and the seeds were considered germinated when the radical emerged. Final germination percentage was calculated as follows (Azizi *et al*., 2011): Final germination percentage = S/T×100, Reduction of germination percentage = (1-The number of germinated seeds conditions salinity/ number of germinated seeds conditions control) ×100. At the fifth day, 5 seedlings were randomly selected and seedling and root length with fresh weight were measured, seed viability index was determined by the following equation: Seed viability index =Final germination percentage × average seedling [(shoot and root length (cm)] / 100. The fresh and dry biomasses of seedling were obtained after the length assessment.

Statistical design and analysis: Analysis of variance of data was performed with Microsoft Excel and SAS 9.3 statistical program using completely randomized design and mean values were compared using Duncan's Multiple Range Test (DMRT).

Results and Discussion

Final germination percentage, seed viability index, seedling and root length, seedling fresh and dry weight of centro, clitoria and siratro were strongly affected ($P< 0.05$) by all salt treatments (Table 1). Positive significant correlation was found between seed viability index and seedling shoot length and negative significant correlation was found between seed viability index, reduction of germination percentage at $p <0.01$ and 0.05 levels (Table 2)

The results of seed germination showed that germination percentage reduced with increasing salt concentration. Strong reduction was observed mainly at the higher level of salt concentration compared to control. Lowest mean germination percentage was observed in centro (5%) followed clitoria (46 %) while the highest mean value was measured in siratro (73%) (Table1). Average reduction of germination percentage in all three seed species showed the greatest reduction in salt concentration with 16 ds m^{-1} (Table 1). However at all salinity level, more reduction was found in centro (53%) and clitoria (44%) and least in siratro (15%). Germination response of centro at all salinity levels was not significantly different from control, may be because of inherent dormancy of the seeds of centro. Otherwise, highly significant differences were found in clitoria followed by siratro (Table 1).

Table 1: Effect of salt stress on the final germination, reduction of germination and seed viability index in three forage legumes

Variables	EC (ds/m)	Centro	Clitoria	Siratro
Final germination percentage (%)	0	9.3±3.9a	61.3±3.9a	82.7±3.9e
	4	4.0±1.9a	60.0±3.8b	78.7±2.9d
	8	6.7±1.1a	46.7±5.8c	76.0±1.9c
	12	1.3±1.1a	37.3±4.7cd	74.7±5.8b
	16	5.3±1.1a	25.3±2.9d	54.7±3.9a
	M	5.3	46.1	73.0
Reduction of germination percentage (%)	0	0±0a	0±0a	0±0b
	4	66.67±23.57a	47.09±3.88b	7.62±2.22b
	8	58.33±5.89a	43.02±16.89a	11.79±5.18b
	12	83.33±11.79a	59.79±8.87a	19.71±4.73ab
	16	54.17±14.73a	70.80±3.64a	34.91±7.82a
	M	52.50	44.14	14.81
Seed viability index (cm)	0	51.73±42.24a	677.15±41.61a	887.97±60.33a
	4	0.00±0.00a	533.15±25.29b	516.59±18.98b
	8	7.11±5.81a	254.85±28.97c	390.53±11.26c
	12	0.00±0.00a	130.99±18.63d	230.45±20.26d
	16	3.00±2.45a	53.20±5.50d	143.09±10.0d
	M	12.37	329.87	433.73

Means followed by the same letter (s) in column (s) are not significantly different at $P<0.05$. Standard error (±), N=3

Table 2. Correlations between different parameters in three forage crops under salt stress

	GP	RGP	SVI	SSL	SRL	SFWT	SDWT
GP	1	-.680**	.817**	0.387	-0.089	0.37	0.068
RGP		1	-.690**	-.655**	-0.372	-0.11	0.209
SVI			1	.786**	0.223	0.426	0.131
SSL				1	.719**	0.34	0.117
SRL					1	0.053	-0.086
SFWT						1	.885**
SDWT							1

**,* significant at the 0.01 and 0.05 levels respectively. (GP-germination percentage, RGP-Reduction of germination percentage, SSL-seedling shoot length, SRL -seedling root length, SVI- Seed viability index, SFWT- seedling fresh weight, SDWT-seedling dry weight)

Means comparison showed that at all salinity levels, loss of seed viability index was observed in all three species as compared to control treatment (0 ds m^{-1} salinity) (Table 1). Highest mean seed viability index was found in siratro (434 cm), followed clitoria (330 cm) and centro (12.37 cm), seedling length showed a strong inhibition with the increasing level of salt solution particularly at high salt levels (8, 12 and 16 ds m^{-1}). There were no significant differences among all the forage species for seedling fresh and dry weight. The fresh and dry weight of seedling of all three species was strongly affected by all salinity levels. Seedling fresh and dry weight were reduced at high salinity levels (8 and 16 ds m^{-1}), whereas fresh seedling weight was reduced more as compared to dry weight of seedling at increasing level of salinity (Fig 1), as the turgidity of the shoot portion might have been affected due to lowering of water potential at higher salinity levels. This trend was more prominent in clitoria than centro and siratro at all salt levels except 8 ds m^{-1} in clitoria. Highest seedling (+root) fresh weight was recorded in clitoria than siratro and centro. The osmotic potential usually decreases at higher salinity levels to such lower value at which water uptake is retard or prevented which is necessary for imbibitions and mobilization of nutrient required for germination (Kollar and Hades). The dry weight was not more affected in all three forage legumes at increasing salinity levels (4, 8, 12 and 16 ds m^{-1}) as compared to control (Fig 1) indicating that water related processes were more affected due to salinity than light and temperature controlling processes during germination and seedling growth of all the crop seeds.

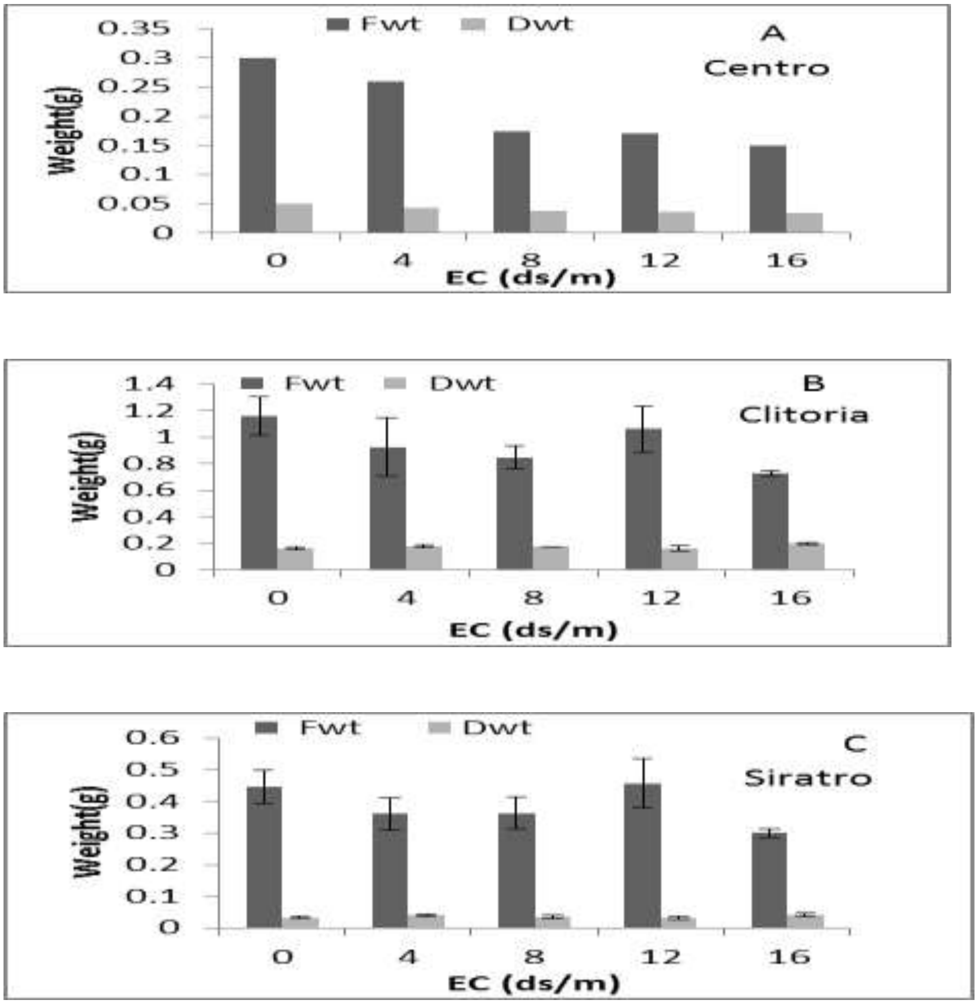

Fig 1: Effect of salt stress on the fresh and dry weight (g) in three forage legumes (Fwt- fresh weight, Dwt-dry weight). Standard error ($\pm$), N=3

Conclusion

The current study indicated that increasing salinity levels affected germination percentage, reduction of germination percentage, seedling viability index, and seedling fresh and dry weight of three range legumes. Among the three fodder species tested, siratro was adjudged tolerant to the salinity than centro and clitoria

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