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Full Length Article

Evaluating the Impact of Different Salt Stress on Growth and Nutritional Parameters of three Lablab-bean (*Lablab purpureus*) Genotypes

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Abstract

Lablab-bean (*Lablab purpureus*) is a potential forage crop for improving the productivity of livestock sector in salt-affected areas. Farmers usually do not have enough knowledge about the right varieties for their fields, which results in lower yields. In this study, genotypic variations in agronomic and physiological traits were analyzed in three *L. purpureus* genotypes (ILRI-6529T, ILRI-184T, Local) commonly grown in Ethiopia. The pot experiments were conducted under 5 soil salinity levels (0, 5, 10, 15, 20 dS m^{-1}). In general, agronomic traits (such as germination percentage, mean germination time, plant population, chlorophyll content, shoot and root length and biomass yield) and quality attributes (ash contents, CP, NDF, invitro organic matter digestibility and metabolizable energy) of all genotypes were negatively affected by increasing salinity levels. The maximum delay in germination was noted in ILRI-6529T genotype followed by ILRI-184T genotype and the local cultivar. The shoot and root lengths and chlorophyll content (SPAD) were higher in ILRI-6529T genotype than other two genotypes under all salinity levels. However, crude protein and metabolizable energy was higher in ILRI-184T genotype under higher salinity levels. The results indicate that ILRI-6529T and ILRI-184T genotypes performed superior than the local cultivar in terms of agronomic and quality parameters at all salinity levels. Therefore, these genotypes can be used to increase forage production and livestock productivity in salt-affected lands in Ethiopia due to its large dependence on livestock sector. © 2019 Friends Science Publishers

Keywords: Salt stress; Forage production; Livestock productivity; Smallholders; Degraded lands

Introduction

Increasing soil salinization is threatening the productivity of arable lands especially in marginal environments (Hussain *et al.*, 2018). Current estimates suggest that by the middle of 21st century, about 50% of lands will lose their production potential due to salinity and associated problems (Daba*et al.*, 2019). Soil salinity is a serious problem as it reduces the productivity of ecosystems and negatively affecting plant growth such as low seed germination, poor crop growth and low yields (Ahmad *et al.*, 2010; Farooq *et al*., 2015; Al-Dakheel and Hussain, 2016). Since drainage infrastructures are expensive to build and operate; therefore, biosaline approaches are becoming more popular to tackle salinity problems. This approach involves the selection of crops that can tolerate salt and water stresses such as halophytes (Koyro and Eisa, 2008).

In Ethiopia, approximately 11 million hectares (Mha) of land (*i.e*., 9% of the country's total landmass and 13% of the irrigated area) are salinized (Frew *et al.*, 2015). The growing occurrence of salt-affected soils is affecting the productivity of irrigated lands as well as farm and livestock productivity. To meet future food security challenges, the introduction of salt-tolerant forage grasses can increase fodder production in the degraded lands. One such crop is Lablab-bean (*Lablab purpureus*), which is a perennial forage legume (family: Fabaceae). It is good for nitrogen fixation and producing rhizomes. It is native in south and central America, India, China, south and south-east Asia and is suitable to grow under salt and water stress conditions (Munns, 2011). It is extensively used as an intercropping crop in India and Australia because it can suppress weeds, control soil erosion and improve soil retention due to its deep rooting system and quick response to rains and relative palatability (Maas *et al*., 2005). However, it is vulnerable against stresses such as high temperature, water and oxidative stresses and toxicity of heavy metals (Sima *et al.*, 2013).

Since salt-tolerant forage plants are variable in biomass production and nutritive value, they need to be tested locally for their (a) edible biomass production, (b) nutritive value of edible biomass (*i.e*., the response in animal production per unit of voluntary feeding intake), and (c) the use of micronutrients and nutraceutical properties. Unfortunately, this information is lacking for Ethiopian conditions,

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which is causing low productivity. Therefore, various varieties of *L. purpureus* need verification for local conditions before they can be recommended to farming communities for large scale adoption. This study was conducted to evaluate three Lablab forage legume (*L. purpureus*) genotypes for their tolerance against soil salinity, yield, and nutritional value. Considering wide range of salt-affected lands in Ethiopia, experiments were conducted for five different salt stress conditions. The results of this research will be beneficial to smallholder farmers in increasing their livestock productivity and to develop crop-livestock value chain systems. This will help in reducing poverty and improving livelihood of rural communities.

Materials and Methods

Experimental Design and Treatments

This pot study was conducted in the net house of Werer Agricultural Research Center (WARC), located in the Afar region of Ethiopia during 2016-18. The average annual precipitation in the study area is 570 mm, out of which 85% falls from February to September (Fig. 1). The average annual Class A Pan evaporation is 2700 mm, which makes irrigation necessary for sustainable crop production.

In each pot, 7.3, 14.3, 21.4 and 29.3 g of NaCl was mixed with 6 kg of soil packed to obtain salinity levels of 5, 10, 15 and 20 dS m^{-1}, respectively. In the control pot, no NaCl was added. Two Lablab genotypes (ILRI-184T; ILRI-6529T) along with one local cultivar were tested using completely randomized experimental design with three replications. Ten seeds of each genotype were directly planted in glazed pots. Irrigations were applied based on crop evapotranspiration ($ET_C = ET_O \times Kc$) demand. To meet leaching requirements, 10% additional good quality water was applied. The reference evapotranspiration (ETo) was calculated by the modified Penman-Monteith equation using climatic data collected from the study area. Each time, experiment continued for 90 days. The soils in the study area are very rich in N, P and K therefore no fertilizer is recommended for plant growth (Menaleshoa *et al.*, 2016). The average amount of water applied during each experiment was 293 mm.

Data Collection and Statistical Analysis

Ten seeds of each genotype were planted in each pot. The data regarding germination time, number of tillers and plant height was collected from all pots. *L. purpureus* legumes were sustained up to 90 days after planting and both shoot and root fresh weight was recorded at the time of harvesting. The germination count was done on the 5th and 15th day after plantation. The germination percentage (GP) was determined using the equation of Ashraf and Foolad (2005).

$$GP = \frac{Total\ germinated\ seeds}{Total\ number\ of\ seeds} \quad (1)$$

Mean germination time (MGT) was calculated using the expression of Ellis and Roberts (1981).

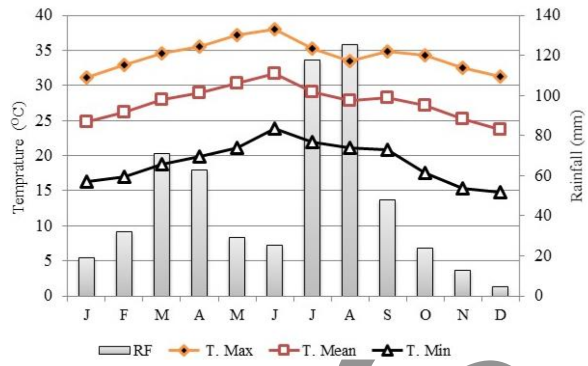

Fig. 1: Mean monthly rainfall and minimum and maximum temperatures at the trial site

$$MGT = \frac{(n5*D5)+(n10*D10)+(n15*D15)}{n5+n10+n15} \quad (2)$$

Where
n = Seeds germinated on day D, and
D = Days after germination
The metabolizable energy content was calculated according to Beever and Mould (2000).

$$ME\ (MJ\ kg^{-1}) = 0.15 \times IvDMDC \quad (3)$$

Where, *IvDMDC* = *In vitro* dry matter digestibility content

Chlorophyll content was measured using SPAD 502 plus Chlorophyll meter. Total ash content was determined using dry mineralization method and approximately 5 g of samples were burned in the quartz capsules for 4 hours at 650°C (Soest *et al.*, 1991; Kitcherside *et al*., 2000). The crude protein (CP) content was calculated by multiplying the Kjeldahl nitrogen concentration by 6.25 (Kacar and İnal, 2008). Neutral Detergent Fibre (NDF) were analyzed using the methods of Soest *et al.* (1991). Invitro dry matter digestibility content (IvDMDC) was analyzed using the method of Tilley and Terry (1963).

Statistical Analysis

The analysis of variance (ANOVA) technique was used for the data analysis for factorial CRD using S.A.S. 9.3 software (S.A.S. Institute, Cary, N.C.). The interaction effects of different treatments were evaluated at 5% significance level ($P < 0.05$) using least significant test method.

Results

Germination Percentage and Mean Germination Time

The interaction between salinity levels and genotypes had significant effect on germination percentage (GP) and mean germination time (MGT) of *L. purpureus* (Table 1). Increasing salinity levels, particularly higher salinity levels, decreased the GP along with higher MGT of all genotypes.

However, the genotypes behaved differently and both ILRI-6529T and ILRI-184T genotypes observed higher GP along with lesser MGT at all salinity levels compared with control cultivar (Table 1). The GP values ranged between 96.7-100% at control, 96-98.7% at 5 dS m^{-1}, 90-93.3% at 10 dS m^{-1}, 70.7–86.7% at 15 dS m^{-1} and 43.3-63.3% at 20 dS m^{-1} (Table 1). At higher salinity levels of 15-20 dS m^{-1}, the maximum GP was observed in ILRI-6529T (Table 1). The MGT was increased with the increasing salinity levels in all genotypes and maximum increase was recorded in control cultivar compared with ILRI-6529T and ILRI-184T genotypes (Table 1).

Plant Population at Maturity and Chlorophyll (SPAD value) Content

Plant population at maturity of *L. purpureus* was negatively affected by the increasing soil salinity (Table 2). The average plant population for all genotypes decreased from 8.0-8.7 at control to 4.3–5.7 at 20 dS m^{-1}. The highest plant population at control was observed for ILRI-6529T genotype (8.7) and the lowest for all three tested genotypes was found at 20 dS m^{-1}. However, ILRI-6529T and ILRI-184T genotypes performed better at 20 dS m^{-1} compared to local cultivar.

The chlorophyll (SPAD value) content was also negatively affected with increasing salinity. The average chlorophyll (SPAD value) content of three genotypes was noted as 50.1, 48.6, 46.6, 37.3 and 35.4 at 0, 5, 10, 15 and 20 dS m^{-1}, respectively (Table 2). ILRI-6529T and ILRI-184T produced higher chlorophyll (SPAD value) content than the local cultivar. At all salinity levels, maximum SPAD value was observed for ILRI-6529T followed by ILRI-184T and the local cultivar.

Shoot and Root Length

Shoot and root lengths of three *L. purpureus* genotypes also showed a declining trend against increasing soil salinity (Table 3). The reduction in shoot length was 9.3 to 51.2% as the salinity increases from 5 dS m^{-1} to 20 dS m^{-1}. At control, the average shoot length of all genotypes was 110 cm, with a highest of 122 cm for ILRI-6529T. However, at 20 dS m^{-1}, the average shoot length was reduced to 73 cm. The highest root length for all salinity levels was achieved for ILRI-6529T trailed by ILRI-184T genotypes and the local cultivar. The reduction in root length was from 2.2 to 6.4 cm with increase in salinity from 5 to 20 dS m^{-1}.

Shoot and Root Dry Matter

The shoot and root dry matter yields also declined by inflated salinity values (Table 4). ILRI-6529T showed maximum shoot biomass (44 g/plant) at control, followed by ILRI-184T and local cultivar. However, shoot biomass for ILRI-6529T was reduced to 39 and 35 g/plant at higher salinity levels (15-20 dS m^{-1}). The local cultivar produced 35 g/plant at 15 dS m^{-1} and 30 g/plant at 20 dS m^{-1}. ILRI-6529T genotype proved best for salt-tolerance for above ground shoot biomass. For all salinity levels, ILRI-6529T produced highest root dry matter followed by ILRI-184T and local cultivar. The maximum root dry matter for ILRI-6529T and ILRI-184T genotypes was obtained at 5-10 dS m^{-1} salinity level.

Nutrient Composition of *L. purpureus* Genotypes

Nutritional value of forages is usually judged by the values of crude protein (CP) and Ash contents. Higher CP (20.9%) and Ash content (20.8%) values of *L. purpureus* genotypes were noted in ILRI-6529T at control whereas the lowest were recorded in the local cultivar (Table 5). Neutral detergent fiber (NDF) values follows similar trends as of CP for all genotypes (Table 6). The NDF values for ILRI-6529T genotype were the highest whereas the local cultivar gives lowest values for all salinity levels. The highest *In vitro* dry matter digestibility content (IvDMDC) was detected in ILRI-184T followed by the other two genotypes. The average metabolizable energy content was 9.5 MJ kg^{-1} DM.

Discussion

Salt-tolerant crops present necessary genetic variation to develop appropriate traits for improving agricultural productivity for degraded lands (Ikram *et al.*, 2010; Turi *et al.*, 2012). The forage legumes such as *L. purpureus* can increase forage production in the salt-affected lands due to their capacity to avert salinity build up by forming a mulching layer on the surface of the soil (D'Souza and Devaraj, 2010; Temel *et al.*, 2015). The soil mulching decreases upward water movement through capillary rise thereby forcing plants to fulfill their atmospheric demand by using water stored in the soil profile (Al-Dakheel *et al.*, 2015).

Parameters such as germination, survival rate and seedling growth or biomass accumulation, have most commonly been used for selecting salt-tolerant plants (Khan *et al.*, 2006). In this study, the increasing salinity levels delayed the germination of all *L. purpureus* genotypes although there were differences among three genotypes. This could be due to the fact that increased salinity reduced cell elongation resulting from decreased turgor, cell volume, and cell growth. Similarly, crop genotypes may germinate effectively under salt stress conditions, however, their seedling growth is likely to be affected. The reduction in GP and MGT with increased salinity could be due to the reduced cell elongation resulting from decreased turgor, cell volume, and cell growth (Aloui *et al.*, 2014; Daba*et al.*, 2019). This phenomenon has also been confirmed for durum wheat and many other crops such as *tef* (*Eragrostis tef* (Zucc.)), lentils (*Lens culinaris*), pepper (*Piper nigrum*) and Rhodes grass (*Chloris gayana*) (Almansouri *et al.*, 2001;Asfaw and Intanna, 2009; Kagan *et al.*, 2010). The seedling growth is affected by increasing salt stress. We have found that ILRI-

Table 1: Effect of different salinity levels on germination percentage (GP) and mean germination time (MGT) of three *L. purpureus* genotypes

Parameters	Cultivars	NaCl salt level (dS m^{-1})					LSD values at $P \leq 0.05$
		0	5	10	15	20	
Germination (%)	Local cultivar	96.67	96.00	90.33	70.66	43.33	7.41
	ILRI 184T	98.33	96.90	90.00	76.66	60.00	
	ILRI 6529T	100.00	98.70	93.00	86.66	66.33	
Mean germination time (days)	Local cultivar	3.77	4.30	6.44	8.77	9.83	0.78
	ILRI 184T	5.66	6.00	7.00	8.50	8.88	
	ILRI 6529T	5.00	5.20	7.27	8.33	8.83	

(LSD = Least Significant Difference)

Table 2: Effect of different salinity levels on plant population at maturity and chlorophyll content of three *L. purpureus* genotype

Parameters	Genotypes	NaCl salt level (dS m^{-1})					LSD values at $P \leq 0.05$
		0	5	10	15	20	
Chlorophyll contents (SPAD value)	Local cultivar	46.86	46.18	44.52	35.62	34.06	2.70
	ILRI 184T	50.56	48.65	48.04	38.34	34.59	
	ILRI 6529T	52.90	50.78	47.15	38.72	37.55	
Plant population at maturity per pot	Local cultivar	8.33	7.66	7.00	6.66	4.33	0.96
	ILRI 184T	8.00	8.00	7.66	7.00	5.66	
	ILRI 6529T	8.66	8.33	8.00	7.33	5.66	

(LSD = Least Significant Difference)

Table 3: Effect of different salinity levels on shoot length and root length of three *L. purpureus* genotypes

Parameters	Genotypes	NaCl salt level (dS m^{-1})					LSD values at $P \leq 0.05$
		0	5	10	15	20	
Shoot length (cm)	Local cultivar	106.66	95.99	87.66	84.98	69.67	17.2
	ILRI 184T	100.33	97.30	97.66	89.66	75.33	
	ILRI 6529T	122.00	107.80	96.66	91.66	72.65	
Root length (cm)	Local cultivar	22.50	20.25	20.00	16.66	14.70	4.2
	ILRI 184T	21.00	18.90	19.69	17.30	16.33	
	ILRI 6529T	23.16	20.84	19.66	17.66	16.56	

(LSD = Least Significant Difference)

Table 4: Effect of different salinity levels on shoot dry matter yield (SDMY) and root dry matter yield (RDMY) of three *L. purpureus* genotypes

Parameters	Genotypes	NaCl salt level (dS m^{-1})					LSD values at $P \leq 0.05$
		0	5	10	15	20	
SDMY (g/pot)	Local cultivar	40.89	36.801	37.99	34.98	29.51	4.09
	ILRI 184T	43.97	40.47	40.03	38.06	32.13	
	ILRI 6529T	44.07	39.66	40.31	39.39	34.91	
RDMY (g/pot)	Local cultivar	4.41	3.97	3.89	3.36	2.97	0.46
	ILRI 184T	4.46	4.11	4.32	3.83	3.19	
	ILRI 6529T	5.03	4.63	4.27	4.02	3.78	

(LSD = Least Significant Difference)

6529T and ILRI-184T are more salt-tolerant and can establish themselves more effectively under different salinity conditions. Therefore, these traits can help reduce biomass yield losses through adequate crop establishment (Vicente *et al.*, 2004; Almodares *et al.*, 2007).

The morphological and physiological traits of all genotypes were reduced under different salinity levels (McConnell *et al.*, 2008; Kumari *et al.*, 2017). There was a significant reduction in different agronomic parameters of *L. purpureus genotypes* due to increased salinity stress. This can be explained by the fact that the excessive energy spent by plants to get required amount of water and nutrients from soil under saline conditions negatively affect root length, crop yield and quality of the produce. (Robinson *et al.*, 2004; Masters *et al.*, 2007; Qadir *et al.*, 2008; D'Souza and Devaraj,2010; Sima *et al.*, 2013; Kumari *et al.*, 2017).

The higher values of chlorophyll content for Lablab accelerated the photosynthetic rate which resulted in more biomass production and higher productivity (Khan *et al.*, 2009). The reduction in chlorophyll contents under saline conditions had also been confirmed by many studies (Ashraf and Foolad, 2005; Iqbal *et al.*, 2006; Khan *et al.*, 2009).

The quality parameters of forage legumes such as crude protein fiber and total digestibility were reduced with the increasing salinity levels for all three genotypes. This causes reduction in the root area and nitrogen uptake by plants (Al-Dakheel *et al.*, 2015; Daba*et al.*, 2019). The lower NDF values for all legumes indicate that the digestibility of forages improved with the increasing salinity levels (Suyama *et al.*, 2007). The metabolizable energy generally followed similar trends with the *in vitro* dry matter digestibility. Lower NDF values are usually desirable

Table 5: Effect of different salinity levels on ash, crude protein (CP) and neutral detergent fiber (NDF) contents of three selected *L. purpureus* genotypes

Parameters	Genotypes	NaCl salt level (dS m^{-1})					LSD values at $P \leq 0.05$
		0	5	10	15	20	
Ash (%)	Local cultivar	19.16	18.98	18.20	17.16	16.45	2.45
	ILRI 184T	20.08	19.98	19.44	18.32	17.43	
	ILRI 6529T	20.76	20.08	19.4	18.29	16.55	
CP (%)	Local cultivar	18.49	18.31	17.34	16.52	15.93	2.31
	ILRI 184T	19.67	18.98	18.35	18.21	17.18	
	ILRI 6529T	20.91	19.20	18.15	17.84	16.77	
NDF (%)	Local cultivar	69.05	68.79	64.42	62.13	59.76	3.87
	ILRI 184T	74.97	73.23	69.96	66.72	62.16	
	ILRI 6529T	75.93	74.01	70.49	66.11	64.41	

(LSD = Least Significant Difference)

Table 6: Effect of different salinity levels on *In vitro* dry matter digestibility (*IvDMDC*) content and metabolizable energy (ME) of three selected *L. purpureus* genotypes

Parameters	Genotypes	NaCl salt level (dS m^{-1})					LSD value at $P \leq 0.05$
		0	5	10	15	20	
IvDMDC (%)	Local Cultivar	66.68	66.09	63.04	58.9	54.02	3.78
	ILRI 184T	69.19	68.86	65.14	60.16	57.44	
	ILRI 6529T	67.33	68.15	64.07	61.33	56.53	
ME (MJ kg^{-1})	Local Cultivar	10.00	9.91	9.46	8.84	8.10	1.53
	ILRI 184T	10.38	10.33	9.77	9.02	8.62	
	ILRI 6529T	10.10	10.22	9.61	9.20	8.48	

(LSD = Least Significant Difference)

for fodder as well as grains. The lower NDF values for all legumes indicate that the digestibility of forages improved with the increasing salinity levels (Suyama *et al.*, 2007). The metabolizable energy generally followed similar trends with the *in vitro* dry matter digestibility.

Conclusions

The agronomic and nutritional composition of three *L. purpureus* genotypes (ILRI-6529T, ILRI-184T, local cultivar) showed a declining trend with the growing salt stress. The results of this study indicate that the ILRI-6529T and ILRI-184T genotypes performed superior compared to the local cultivar in terms of all agronomic and nutritional parameters at all salinity levels. Therefore, these two genotypes can be grown in salt-affected areas to increase productivity of livestock sector in Ethiopia and other African countries with similar conditions.

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