Journal BiNET
Scientific Publication

Published with Open Access at Journal BiNET
Vol. 12, Issue 02: 1054-1063

Journal of Bioscience and Agriculture Research

Journal Home: www.journalbinet.com/jbar-journal.html

Adaptability of dekoko (*Pisum sativum* var. *abyssinicum*) seedlings to salinity stress in vitro culture conditions

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ABSTRACT

Dekoko (Pisum sativum var. abyssinicum) is one of the most important food legumes grown in south Tigray and north Wollo, northern Ethiopia. It is one among the most important food legumes in terms of price and protein content. It grows mixed and alone with many cereal crops growing in north Ethiopia. This study was conducted with the objective of selecting adaptable and relatively high yielding P. sativum var. abyssinicum collections under different salt (NaCl) concentrations at laboratory conditions. Seeds for 30 collections were obtained from nine districts; two regional states of north Ethiopia with different attitudinal ranges 1300 m.a.s.l being the lowest and 3148 m.a.s.l the highest. Of the 30 on farm and wire house tested local collections, six vigorously growing local collections, three from Ofla (T-001/08OF, T-002/08OF and, T-003/08OF), one from Sirinka (TA-026/15Sr), one from Emba-Alaje (T-025/15E/A), and one from Endamohoni (T-023/08MW) were selected four this study; for salt stress resistances in controlled condition by priming in four salt treatment levels (5 dS/m, 7 dS/m, 9 dS/m and 15 dS/m). Distilled water (0 dS/m) was used as control. Fifty (50) surface sterilized seeds per petri dish were sown for the four salt treatments and the control. Collections T-001/08 from Ofla and T-023/08 from Endamohoni showed good growth performance at 5 dS/m. However, TA-026/15Sr from Sirinka and T-025/15E/A from Betmara (Emba-Alaje) respond positively upto 7dS/m. At higher salinity level growth features decreased with increasing salinity stress. But, T-023/08MW, T-001/08OF, TA-026/15Sr followed by T-025/15E/A from lower to the higher resistances respectively could withstand lower to medium concentrations of salinity as compared to the other collections.

Key Words: *Pisum sativum, Northern Ethiopia, High yielding, Resistance and Salinity level*

Cite Article: Gebreegziabher, B. G. and Tsegay, B. A. (2017). Adaptability of dekoko (*Pisum sativum* var. *abyssinicum*) seedlings to salinity stress in vitro culture conditions. Journal of Bioscience and Agriculture Research,12(02), 1054-1063. **Crossref:**https://doi.org/10.18801/jbar.120217.129

I. Introduction

It is known and approximated the incessantly growing human population will increase global food production requirement by more than half by 2050 that the land and water are unable to sustain them (Wild, 2003). Increasing food production of the staple crops through expansion of cultivation area will become challenging in the future. This is due to progressive urbanization, industrialization, soil

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degradation including soil salinization and insufficient farmlands (Wild, 2003). Chemical fertilizations are highly used. Urban wastages are derived to water bodies. On the other hand, irrigation practices are the goals and strategies of this era to increase crop production particularly in developing countries. Agricultural areas become saline needing salt tolerant plants instead of introducing new crops. Salinity adversely outcome slow agricultural production and low income due to high production costs, and due to soil erosion and ecological imbalance (Hu and Schmidhalter, 2002). However, solutions for salt problems are available. One among, saline soil fertility can be improved by cultivating legume plants that enrich the soil with nitrogen due to their symbiotic capacity (Keneni *et al*., 2013). The demand for productivity and homogeneity in peas, as in other crops, has resulted in a limited number of high-yielding varieties at the price of the loss of heterogeneous traditional local varieties (landraces) through genetic erosion (Smýkal *et al*., 2015). Conversely, landraces preserve much of this diversity lost and comprise the genetic resources for breeding new crop varieties to help cope with environmental and demographic changes that enhance soil fertility (Esquinas-Alacazar, 2005; Smýkal *et al*., 2015). Grain legumes take up an exceptional position in world agriculture by virtue of their high protein content and capacity to fix atmospheric nitrogen. Field pea (*Pisum sativum* L.) is grown in many countries and currently ranks fourth among the pulses in the world with cultivated area of 6.33 million ha (FAO, 2012). Field pea is known to be grown in Ethiopia since antiquity (Keneni *et al*., 2013). Currently, the crop is the fourth most important pulse crop in Ethiopia, preceded only by faba bean, haricot bean and chickpea in terms of both area coverage and total national production (CSA, 2011; Berhane and Berhanu, 2016). There are two botanical cultivars of *Pisum sativum* cool season food legumes (CSFLs) known to grow in Ethiopia largely produced by subsistence farmers and serve as supplementary protein sources and soil fertility restorers by crop rotation or intercropping, namely *P. sativum* var. *sativum* and *P. sativum* var. *abyssinicum* (Keneni *et al*., 2013; Smýkal *et al*., 2015). Their yields are very low, mainly limited by soil fertility (Yemane and Skjelvåg, 2003). In Ethiopia, they are cultivated in poor soils, often without fertilization and in areas not conducive to staple crops, flood exposed. *P. sativum* var. *abyssinicum* is capable of producing seed yield of up to 1.95 t/ha under phosphorus fertilization. It is known for its high market price (more than double of the price of faba bean and field pea) and for its food preference even with its scarcityof production (Berhane and Berhanu, 2016). Many crops exhibit a broad spectrum of response to salt stress but numerous have shared common responses that their growth and eventually yield reduced under salinity (Munns and Tester, 2008). Salt stress reduces dry matter content, increases the root to shoot ratio and reduces the leaf size, which all lead to yield reduction (Munns and Tester, 2008). Survival of plants in unfavorable conditions requires human intervention to improve their metabolism, including accumulation of protective compounds such as compatible solutes (e.g. proline, salt) for osmotic adjustment, proteins and antioxidants for oxidative stress mitigation in this era of climate change scenario (Ashraf and Foolad, 2007). Understanding of mechanisms that alleviate the impact of salt on plant growth and mechanisms of salinity tolerance; at the whole-organism, organelle and molecular levels allows selection and improving salt tolerance of crops both in irrigable and non-irrigable lands of the world. Although, disregarded by breeders, *P. sativum* var. *abyssinicum* is characterized by high protein content in its cotyledons and seeds, low soil fertility requirements; and unusual among legume plants, tolerant to different pH and soil types especially to drought and moisture stress (Girmay *et al*., 2014, reviewed in Berhane and Berhanu (2016). These and other molecular features make *P. sativum* var. *abyssinicum* plant selective and alternative to other high-protein legumes such as peas or beans, which require better growing conditions. Beyond the health value it contributes, agronomical, *P. sativum* var. *abyssinicum* is a good nitrogen fixer and improves the yield of succeeding crops (Keneni *et al*., 2013). In the *in vitro* culture conditions, *P. sativum* var. *abyssinicum* is considered as less recalcitrant to regeneration of other similar plants contrasting to grass pea (Barpete *et al*., 2014; Tsegay and Gebreslassie, 2014). Even though *P. sativum* var. *abyssinicum* stands out next to grass pea with resistance to various abiotic stresses (Tsegay and Gebreslassie, 2014), mechanisms of adaptation to these stresses including salt stress at the physiological and molecular level are not as the studies for other peas. There is a start on the effect of salinity on seed germination, growth and development ontwo local collections of *P. sativum* var. *abyssinicum* seedlings (Tsegay and Gebreslassie, 2014). Therefore, the aim of this study was to identify possible adaptation growth responses of six on farm and wire house promising collections of *P. sativum* var. *abyssinicum* to salinity stress. By doing so, it is possible to select adaptable and relatively high yielding collections.

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II. Materials and Methods

Seed materials selection and salt treatments: This study was conducted using saline growth media to validate and select the on farm as well as wire house agronomically better thrives, adaptable and relatively high yielding *P. sativum* var. *abyssinicum*local collections under different salt (NaCl) concentrations at controlled condition. In addition to thrive, their traditional cultivation on different altitude and potential exposures to harsh environmental conditions were the criteria for selection. The seeds of 30 collections (Table 01) were obtained from nine districts; two regional states of Ethiopia at different attitudinal ranges. Of the 30 on farm and wire house investigated collections, six better thrives, three from Ofla (T-001/08OF, T-002/08OF and, T-003/08OF), one from Sirinka (TA-026/15Sr), one from Emba-Alaje (T-025/15E/A), and one from Endamohoni (T-023/08MW) were selected for this study in controlled condition primed in four salt treatment levels (5 dS/m, 7 dS/m, 9 dS/m and 15 dS/m). Distilled water (0 dS/m) was used as control. Fifty (50) surface sterilized seeds per petri dish were sown for the four salt treatments and the control tests.

Table 01. Description by sources of regions, districts and altitudial variation of the 30 Dekoko collections studied and the six better thrives selected for salt resistances

Code and name of Accessions		Description of sources area			
No	Name of Accession	Name of region	Name of district	Agro-ecology	Altitude (m.a.s.l)
1	**T-025/15E/A**	Tigray	Emba-Alaje	highland	2116
2	**TA-026/15Sr**	Amhara	Sirinka	Midland	1868
3	**T-001/08OF**	Tigray	Ofla	highland	2457
4	**T-023/08MW**	Tigray	Endamohoni	highland	2100
5	**T-002/04OF**	Tigray	Ofla	highland	2457
6	**T-003/04OF**	Tigray	Ofla	highland	2457
7	TK-004/08AL	Tigray	Alamat	lowland	1178-3148
8	TK-005/08AL	Tigray	Alamata	lowland	1178-3148
9	TK-006/08AL	Tigray	Alamat	lowland	1178-3148
10	TK-008/08AL	Tigray	Alamat	lowland	1178-3148
11	T-022/08E/A	T igray	Emba-Alaje	highland	2116
12	T-024/08E/A	Tigray	Emba-Alaje	highland	2116
13	T-021/08H/W	Tigray	Hintalo-Wajerat	midland	1400-3050
14	T-007/08KO	Amhara	Kobo	lowland	1100-3000
15	T-009/08KO	Amhara	Kobo	lowland	1100-3000
16	T-010/08KO	Amhara	Kobo	lowland	1100-3000
17	T-017/08KO	Amhara	Kobo	lowland	1100-3000
18	T-018/08KO	Amhara	Kobo	lowland	1100-3000
19	T-019/08KO	Amhara	Kobo	lowland	1100-3000
20	T-020/08KO	Amhara	Kobo	lowland	1100-3000
21	T-012/08G/L	Amhara	Guba-lafto	highland	2061
22	TA-013/08Sr	Amhara	Sirinka	midland	1868
23	TA-014/08Sr	Amhara	Sirinka	midland	1868
23	TA-015/08	Amhara	Sirinka	midland	1868
25	T-011/08Ha	Amhara	Habru	lowland	700-1900
26	T-016/08hA	Amhara	Habru	lowland	700-1900
27	T-027/15Sr	Amhara	Sirinka	midland	1868
28	T-028/15MW	Tigray	Endamehoni/mychew	highland	2100
29	T-029/15MW	Tigray	Endamehoni/mychew	highland	2100
30	T-030/15E/A	Tigray	Emba- Alaje	highland	2116

Collections written in bold (1-6) were target collections for this salt resistance study.

Seed germination and seedling growth: Seeds were allowed to germinate and grow at (24±1) ^{0}C for 14 days. Randomized complete block design (RCBD) replicated four times was used throughout the culture. Fifteen grams of seeds from each collections of *P. sativum* var. *abyssinicum* were placed in petri dishes containing distilled water to determine water uptake of seeds necessary for germination.

Water uptake was measured as fresh weight percentage increase in seed weight. For each replication, 50 seeds of *P. sativum* var. *abyssinicum* were grown on Whatman no. 2 filter papers (Figure 1) with 50 ml from the prepared respective stock solution treatments. Growth media were put in a sealed polythene bags to prevent evaporation. Germination and seedling growth traits were evaluated. Germination percentage was estimated according to the International Rules for Seed Testing when their radicles extend longer than 2 mm (ISTA, 1985). Mean germination time was calculated for the rate of germination with the following formula: MGT = $\frac{\sum dn}{\sum n}$ (Kandil *et al.*, 2012), where n is the number of seeds which were germinated at day 'd', and d is the number of days counted from the beginning of sowing. Germination index (GI) was calculated as the product of number of days after sowing and number of germinated seeds divided to the total number of sown seeds for the correlation analysis. GI=$\frac{\sum diNi}{S}$ (Li, 2008), where 'di' is number of days after sowing of seeds, 'Ni' is number of germinated seeds , 'S' is total number of sown seeds. Seedling growth features such as shoot growth traits were measured. Root length, shoot length, and seedling fresh weights and the other traits in (Table 02, Table 03 and Table 04) and Figure 02 were measured on the 14th day after sowing.

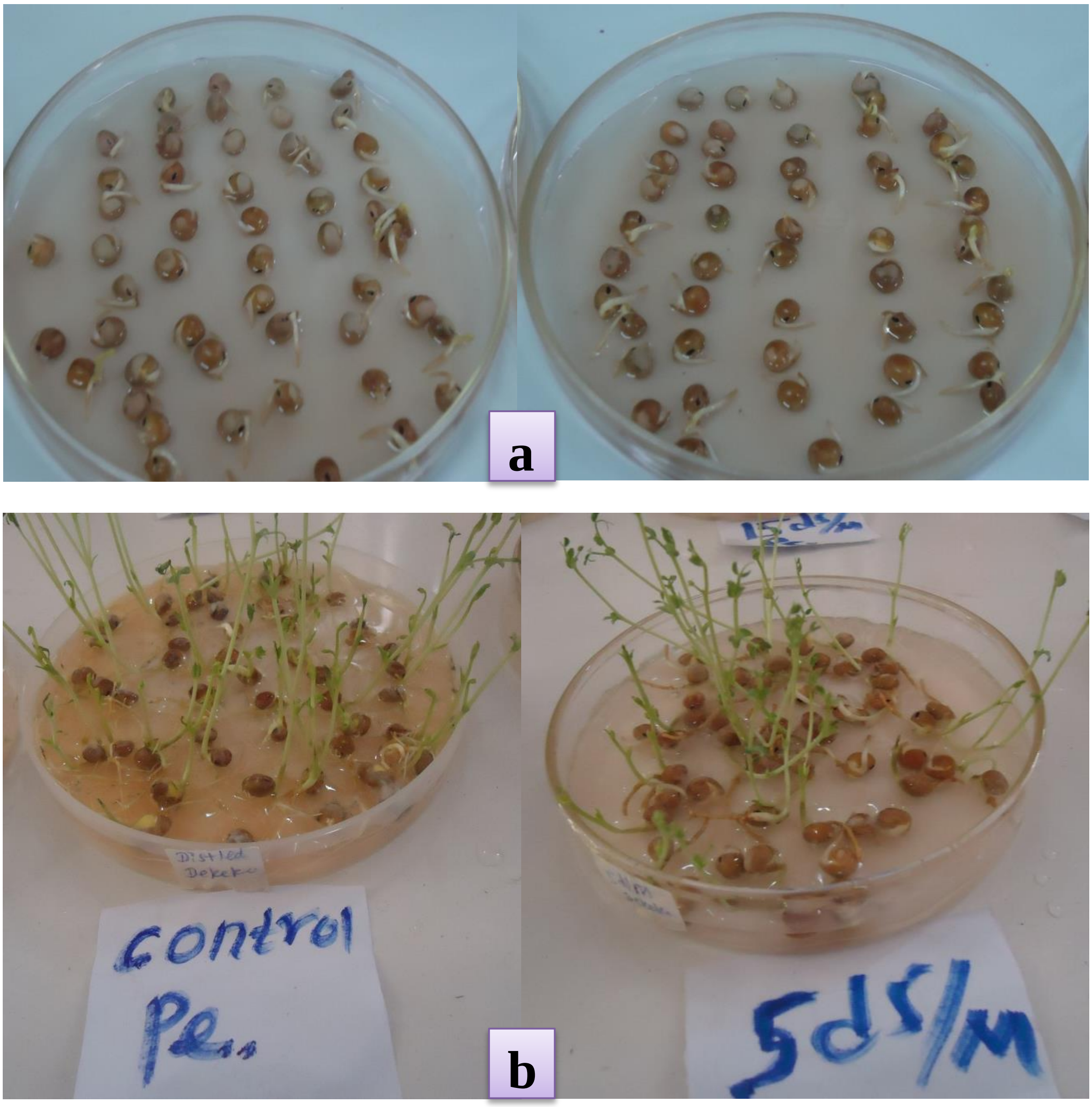

Figure 01(a). Fifty seeds of *P. sativum* var. *abyssinicum* for each collection plated in growth chambers (Petri dishes); 01(b). Seed recovery rate and seedling growth difference of the control and 5 dS/m salt treated seeds of T-025/15E/A collection.

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Recovery test of seed germination experiment: Determination of seeds recovery rate from salt stress inhibition was carried out with modification using the method of Almansouri *et al.* (2001). Fifty *P. sativum* var. *abyssinicum*seeds from each collection were imbibed for 10 hours in 7dS/m salt solution. Thereafter, the seeds were allowed to germinate on petri dishes washed and disinfected with alcohol and air driedon two layers of Whatman no. 2 filter paper moistened with 50 ml NaCl solution of the same concentration used for seed imbibition (Figure 01b). After recovery seedlings from 5 dS/m treatment were thicker and fewer in number of germinated seedlings than the control treatment (0 dS/m) as shown figure 01b above.

Data presented in the results are means from 4 replicates per treatment. Means were compared between salt treatments and collection types by Turkey Test at the 0.05 confidence level. Correlation analysis were used know relatedness of the seedling growth variables, salt treatment concentrations and the collection crops.

III. Results and Discussion

Increase in water uptake percentage of the *Pisum sativum* var. *abyssinicum* collections (Table 02) treated with salt (5 dS/m, 7 dS/m and 9 dS/m) compared with the control (0 dS/m) indicated that physiological activities are enhanced for salt stress resistances induced by various salt stresses (Gebreegziabher and Qufa, 2017). These processes ensure that the water balance that can provide a relative normal physiological environment for the study collections (Wang *et al.*, 2015). Even among the six collections, there experienced a difference in water uptake percentage. Better thrivers in germination percentage, germination rate, and root length required more water than the less growers; taking T-025/15E/A, TA-026/Sr, T-023/08MW, T-001/08OF the four medium salts treatment resistant collections in descending order (Table 02). This means there is a considerable difference in percentage of seedling survivorship among the collections (Wu and Lin, 2008). Increases in mean germination time of the salt stress resistant promising collections (T-025/15E/A, TA-026/Sr, T-023/08MW, T-001/08OF) is in relation to their germination stage tolerances to salt stress (Table 02). A negative relationship between germination percentage, and water uptake percentage and mean germination time was observed in the promising collections of the control and different salt treatments. Seeds did not emerge when the salinity exceeds 9 dS/m where no seeds were germinated at the 15 dS/m treatment. Increases in mean germination time of the resistant or tolerant collections are because of the adaptive features of root elongation for water uptake search and survivorship mechanisms of the germinated seeds after stress (Ramoliya and Pandey, 2003). Similarly, findings from Janagard *et al.* (2008) stated attributes associated to root and shoot to be the two most important features determining adaptation of plants to different salinity conditions. It is known that roots are the first organs that face the corrosion due to salinity affecting the whole growth features of the crop. Toxic effect of sodium chloride causes reduction in shoot growth through unbalanced nutrient uptake under the NaCl stress. With respect to root fresh weight and dry weight at germination the resistant collections of *Pisum sativum* var. *abyssinicum* have better value at the medium salinity stresses; 5 dS/m and 7 dS/m although salt stress is negatively affected growth and development of these collections as it increases above 7 dS/m and even at the medium salt treatments for the other collections (Table 02). This is related to the root biomass relationship of the crop and the salinity tolerance of collections increased because of the root Na^+ increments (Akinci *et al*., 2010). Results in Table 02 showthat there is statistically significant difference on the seedling growth traits among the six *P. sativum* var. *abyssinicum* collections. This indicates the existences of considerable variability among the collections for most of the growth parameters after salt treatments. This implies that, the collectionsevenmay belong to the same variety (species) but their cultivation in different areas (Table 01) force them to have different adaptation mechanisms for abiotic stress like in this case salinity. This might cause to different cellular adaptive responses such as accumulation of compatible solutes. The salt priming of seeds prior to sowing enhances a process used naturally by plants to minimize the movement of Na^+ to the shoot. Here, the complexity of interactions between stress factor or salt stress and various molecular, biochemical and physiological phenomena affect the different growth and developments among the collections (Hamdia and Shaddad, 2010; Tsegay and Gebreslassie, 2014; Gebreegziabher and Qufa, 2017). T-025/15E/A, TA-026/Sr, T-023/08MW, T-001/08OF perform betterin descending order for root growth features with root height reduction percentage of negative value at the medium salt concentrations (Table 02) indicating increases in root length from the untreated seeds for the increases of Na^{+} content of the root systems. This is the need to accumulate inorganic and organic solutes for resisting osmotic stress induced by various salt stress levels (Wang *et al.*, 2015).

Table 02. Effect of NaCl treatment on *Pisum sativum* var. *abyssinicum* growth parameters (root length, root fresh and dry weights) under salinity stress

Treatments (NaCl)	Collections water uptake (%)						Collections germination (%)					
	T-001/08 OF	T-002/08 OF	T-003/08 OF	TA-026/15 Sr	T-025/15 E/A	T-023/08 MW	T-001/08 OF	T-002/08 OF	T-003/08 OF	TA-026/15 Sr	T-025/15 E/A	T-023/08 MW
0dS/m	55.43^{a}	56.24^{a}	54.79^{c}	60.9^{c}	63.89^{c}	59.16^{ab}	82.04^{a}	79.54^{b}	67.08^{a}	83.86^{a}	90.00^{a}	78.32^{a}
5dS/m	70.98^{a}	69.45^{a}	71.00^{b}	78.90^{b}	80.01^{b}	84.23^{ab}	57.68^{b}	48.54^{c}	37.87^{b}	81.22^{a}	88.78^{a}	81.44^{a}
7dS/m	78.67^{a}	75.06^{a}	73.46^{a}	82.20^{b}	86.96^{a}	75.05^{ab}	34.55^{c}	18.67^{d}	28.32^{c}	65.00^{b}	76.02^{b}	61.61^{b}
9dS/m	82.30^{a}	83.01^{a}	84.70^{a}	89.22^{a}	90.43^{a}	85.67^{ab}	17.95^{d}	01.65^{e}	25.22^{d}	55.08^{b}	65.54^{b}	49.00^{c}
Treatments (NaCl)	Collections germination rate						Collections mean germination time (days)					
	T-001/08 OF	T-002/08 OF	T-003/08 OF	TA-026/15 Sr	T-025/15 E/A	T-023/08 MW	T-001/08 OF	T-002/08 OF	T-003/0 8OF	TA-026/15 Sr	T-025/15E /A	T-023/08 MW
0dS/m	13.28^{a}	13.85^{a}	11.68^{a}	14.60^{a}	15.67^{a}	13.16^{a}	3.1^{c}	3.21^{c}	3.41^{c}	4.63^{a}	4.08^{a}	3.61^{b}
5dS/m	14.04^{a}	8.45^{b}	6.59^{c}	14.74^{a}	15.86^{a}	14.66^{a}	3.70^{b}	3.64^{b}	2.92^{c}	4.00^{a}	4.22^{a}	3.72^{b}
7dS/m	6.01^{c}	3.25^{e}	4.93^{c}	11.32^{b}	13.24^{b}	10.73^{b}	4.01^{a}	3.89^{b}	2.43^{d}	4.55^{a}	4.86^{a}	4.02^{a}
9dS/m	3.12^{d}	0.29^{d}	4.39^{d}	9.59^{b}	11.42^{b}	8.53^{b}	0.89^{d}	0.65^{d}	0.00^{e}	1.57^{d}	3.00^{b}	2.50^{c}
Treatments (NaCl)	Collections root length (cm)						Collections root height reduction (%)					
	T-001/08 OF	T-002/08 OF	T-003/08 OF	TA-026/15 Sr	T-025/15 E/A	T-023/08 MW	T-001/08 OF	T-002/08 OF	T-003/08 OF	TA-026/15 Sr	T-025/15 E/A	T-023/08 MW
0dS/m	9.31^{a}	9.46^{a}	11.03^{a}	12.11^{a}	12.59^{b}	9.87^{a}	0.00^{e}	0.00^{c}	0.00^{d}	0.00^{b}	0.00^{a}	0.00^{c}
5dS/m	10.04^{a}	9.73^{a}	7.93^{b}	13.23^{a}	14.22^{a}	10.32^{a}	-7.84^{d}	-2.85^{d}	28.11^{c}	-9.25^{d}	-12.95^{d}	-4.56^{d}
7dS/m	8.13^{b}	7.00^{b}	6.81^{b}	12.42^{a}	13.66^{a}	7.51^{b}	12.68^{b}	26.00^{b}	38.26^{b}	-2.26^{c}	-8.50^{c}	23.91^{b}
9dS/m	1.46^{c}	1.50^{c}	1.01^{c}	5.20^{b}	5.50^{b}	0.99^{c}	84.34^{a}	84.14^{a}	90.84^{a}	57.06^{a}	56.31^{b}	89.97^{a}
Treatments (NaCl)	Collections root fresh weight (g)						Collections root dry weight(g)					
	T-001/08 OF	T-002/08 OF	T-003/08 OF	TA-026/15 Sr	T-025/15 E/A	T-023/08 MW	T-001/08 OF	T-002/08 OF	T-003/08 OF	TA-026/15 Sr	T-025/15 E/A	T-023/08 MW
0dS/m	6.40^{b}	5.62^{b}	5.43^{b}	6.67^{b}	6.58^{b}	6.51^{b}	3.04^{b}	2.34^{b}	2.28^{a}	3.50^{a}	3.52^{b}	2.74^{a}
5dS/m	7.30^{a}	7.40^{a}	6.82^{a}	7.51^{a}	7.49^{a}	7.54^{a}	4.71^{a}	3.51^{a}	2.81^{a}	3.42^{b}	4.03^{a}	2.60^{a}
7dS/m	5.92^{c}	4.89^{c}	5.40^{b}	7.88^{a}	8.00^{a}	7.22^{a}	3.90^{a}	2.01^{c}	1.87^{b}	3.94^{a}	4.21^{a}	1.87^{b}
9dS/m	4.86^{d}	3.62^{d}	3.01^{c}	6.20^{b}	7.72^{a}	4.75^{c}	2.20^{c}	1.93^{c}	1.43^{c}	2.73^{c}	3.52^{b}	1.06^{c}

Subscriptions (a-e) with different letters indicate significant difference among means within the columns ($P<0.05$, using LSD test).

Shoot growth parameters such as shoot length, shoot height reduction, shoot fresh weight and shoot dry weight were assayed (Figure 02). T-025/15E/A followed by TA-026/15Sr and T-003/08OF with similar responses showed better seedling growth at 5 dS/m and the shortest shoot length at this salt treatment was observed in T-002/08OF. The highest shoot height reduction was observed from T-003/08OF collection at all the salt stress levels. This may be because of the collection is hampered by hyper-hydricity, a phenomenon of deformity in which the shoots become translucent during weight losingfrom their original form and physiology within a short period of time (Wu *et al.*, 2011). T-025/15E/A and TA-026/15Sr respectively are salt resistant collections in all the salinity treatments with respect to shoot length of the seedlings (Figure 02). The reason is that, these collections have lower concentration of Na^{+} and Cl^{-} concentrations in their shoots contrasting to the sensitive collections (Lahiri *et al.*, 2009).

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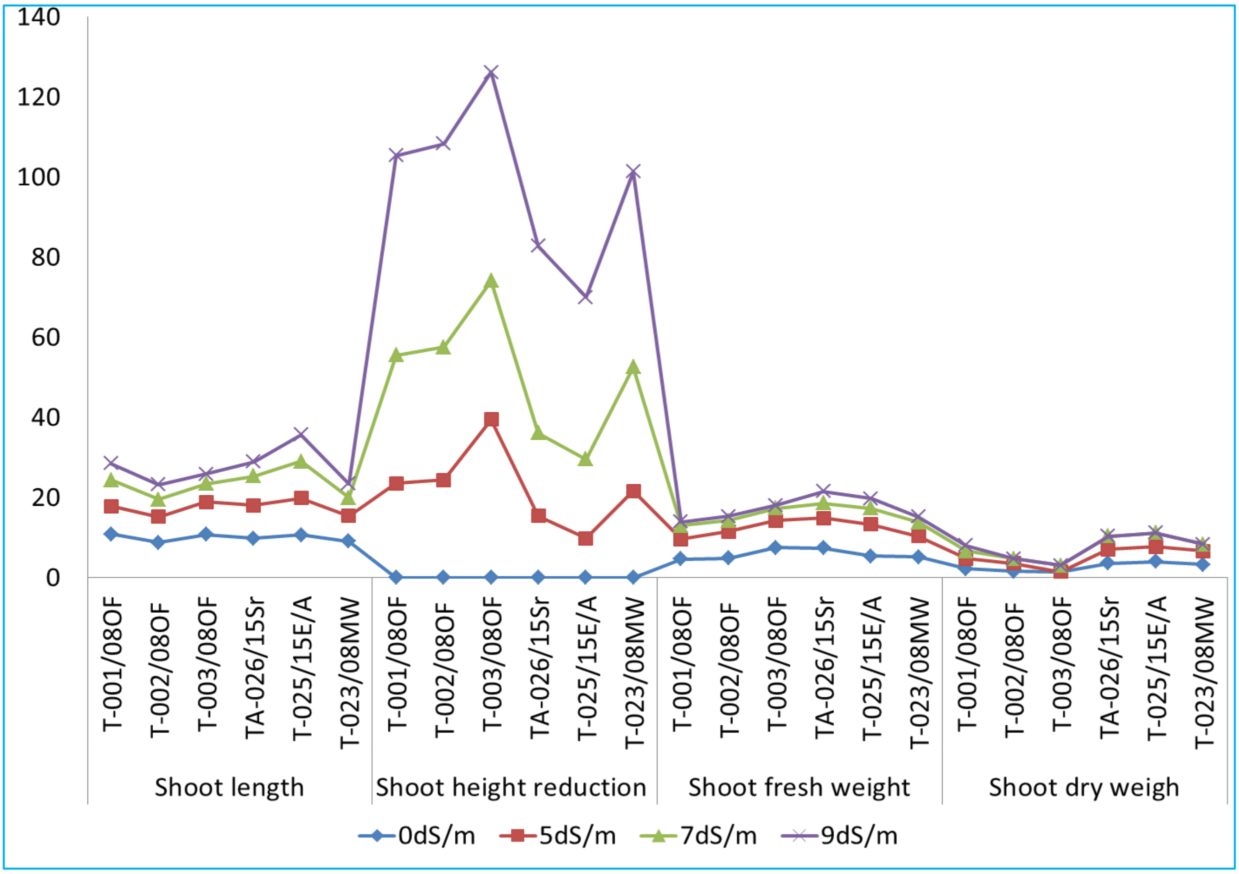

Figure 02. Effect of salinity on shoot growth features of different Dekoko collections.

Table 03. Pearson correlation coefficient between collections and salt concentration for *P. sativum* var. *abyssinicum* shoots growth parameters

	2	3	4	5	6	7	8	9
1	0	0.217	-0.049	-0.049	-0.047	-0.249	0.008	-0.242
2		0.109	-0.926**	-0.962**	-0.952**	-0.818**	-0.901**	-0.807**
3			-0.173	-0.173	-0.205	-0.105	-0.212	-0.219
4				1.00**	0.973**	0.880**	-0.790**	0. 818**
5					0.973**	0.880**	-0.790**	0. 818**
6						0.865**	-0.814**	0.811**
7							-0.776**	0.869**
8								-0.734**

(1) Collections; (2) Salt concentration (dS/m); (3) Water uptake percentage; (4) Seed germination; (5) Mean Germination time (days); (6) Shoot length (cm); (7) Shoot height reduction percentage; (8) Shoot fresh weight (gm); (9) Shoot dry weight (gm) after oven dry. **. Correlation is significant at the 0.01 level (2-tailed).

Collected data was also subjected to total correlation analysis based on all the relevant variables. Total correlation analysis for salt stress adaptation showed no significant linear association among collections, interaction between collections and salt concentration, and interaction between collections and growth features (Table 03). Salt concentration is negatively correlated with seed germination, shoot length, shoot height reduction, shoot fresh weight and shoot dry weight i.e., as the salt concentration increased from 5 dS/m to 15 dS/m, the aforementioned variables decreased linearly. The longest shoot length was obtained at all treatments from T-025/15E/A followed by the T-001/08OF and TA-026/15Sr collections (Figure 02). Shoot length was severely influenced by salt stress with complete inhibition and no shoots grown at 15 dS/m NaCl stress in most of the collections but of with dwarf shoot from T-025/15E/A and TA-026/15Sr. This is because; increasing in osmotic potential (more negative) decreases the movement of nutrients and sap up and down the stem of the cropcauses major reductions in crop productivity and quality. Salinity effects are the results of complex interactions among morphological, physiological, and biochemical processes including seed germination, plant growth, and water and nutrient uptake (Shrivastava and Kumar, 2015). Compared to control, each increase in NaCl concentration resulted in remarkable decrease in shoot fresh weight for all collections (Figure 02). The highest shoot fresh weight (100 g/plant) was observed from T-025/15E/A followed by accession TA-026/15Sr and T-003/08OF at the control and 5 dS/m the lowest salt treatment (Figure 02). However, the distributions demonstrated a very different picture among the collections; where TA-026/15Sr a midland crop is comparative with T-025/15E/A and T-003/08OF the highland crops (Table 01) that need to be investigated. As shown in table 04, the effect of salt treatment is negatively correlated with vigor index, leaf branch, germination index and relative NaCl injury rate. The relative measure of standardized variables of *P. sativum* var. *abyssinicum* seedling growth traits showed a decreased trend with increasing NaCl salt concentration. The highest coefficient of correlation was observed from vigor index followed by leaf branches. Decrease in number of branches per plant of *P. sativum* var. *abyssinicum* may be to decrease the leaf injury by adjusting high leaf k^+/Na^+ ratio and having fewer branches instead of branches that aggravate leaf injury due to its lose husk. This is in line with the finding of Shahid *et al.* (2013) where number of branches decreased with salt treatment increase in pea cultivars. From overall correlation analysis, the correlation was significant ($P<0.01$ and $P < 0.05$) between NaCl salt treatments and *P. sativum* var. *abyssinicum* seedling growth traits. NaCl stress has negatively affected shoot length, shoot height reduction, shoot fresh weight, and shoot dry weight after oven dry as a result of other seedling growth traits such as vigor index, leaf branch per plant, germination index and relative NaCl injury rate.

Table 04. Pearson correlation coefficient of salt concentration, shoot growth with other *P. sativum* var. *abyssinicum* seedling growth traits

	2	3	4	5	6	7	8	9
1	-0.952**	-0.818**	-0.901**	-0.807**	-0.888**	-0.880**	-0.902**	-0.759**
2		0.865**	-0.814**	0.811**	0.856**	0.874**	0.845**	0.765**
3			-0.776**	0.869**	0.856**	0.858**	0.757**	0.635**
4				-0.734**	-0.784**	-0.789**	-0. 947**	-0.801**
5					0.897**	0.683**	0. 693**	0.645**
6						0.754**	0.715**	0.716**
7							0.827**	0.553*
8								0.781**

(1) Salt concentration (dS/m); (2) Shoot length (cm); (3) Shoot height reduction (%); (4) Shoot fresh weight; (5) Shoot dry weight after over dry; (6) Vigor index; (7) Leaf branch per leaf; (8) Germination index; (9) Relative NaCl injury rate. **. Correlation is significant at the 0.01 level (2-tailed); *. Correlation is significant at 0.05 levels (2-tailed).

IV. Conclusion

Collections of *Pisum sativum* var. *abyssinicum* collected from different attitudinally variable agro-ecologies have different adaptive responses to salt stress. This indicates the same subspecies growing in different areas vary to abiotic stress responses such as salinity. Moreover, *Pisum sativum* var. *abyssinicum* is salt tolerant more at germination stage particularly of root length than seedling stage. It is clear that seed germination, root length, shoot length have significant association with salt stress; the more the salinity, the more reduction in these growth performances. But, T-023/08MW, T-001/08OF, TA-026/15Sr followed by T-025/15E/A from lower to the higher resistances respectively could withstand lower to medium concentrations of salinity as compared to the other collections similar to what has been found from the field and wire-house trial of these collections.

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EISSN: 2312-7945,

Acknowledgments

The author is grateful to Bahir Dar University and Woldia University. The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.

Competing Interests

The authors declare that they have no competing interests.

V. References

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HOW TO CITE THIS ARTICLE?

Crossref: https://doi.org/10.18801/jbar.120217.129

APA (American Psychological Association)

Gebreegziabher, B. G. and Tsegay, B. A. (2017). Adaptability of dekoko (*Pisum sativum* var. *abyssinicum*) seedlings to salinity stress in vitro culture conditions. Journal of Bioscience and Agriculture Research, 12(02), 1054-1063.

MLA (Modern Language Association)

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Gebreegziabher, B. G. and Tsegay, B. A. "Adaptability of dekoko (*Pisum sativum* var. *abyssinicum*) seedlings to salinity stress in vitro culture conditions". Journal of Bioscience and Agriculture Research, 12 no.02(2017): 1054-1063.