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Full Length Research Article

EFFECTS OF SALINITY AND ETHYLENEDIAMINE TETRA ACETIC ACID (EDTA) ON THE GERMINATION OF TOMATO *(SOLANUM LYCOPERSICUM* L.*)* SEEDS

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ABSTRACT
In this study, the effects of the combined treatment of salinity and ethylenediamine tetra acetic acid (EDTA) on the germination of tomato seeds in Petri-dishes were compared to sole salinity. The treatments consisted of seven concentrations of sodium chloride (NaCL): 0 (control), 10, 50, 100, 250, 500 and 1000 mM. The results from this study showed that percentage germination decreased from 86.7% in the control to 73.3% in 10 mM, 70 % in 50 mM and 23.3 % in 100 mM. Seeds treated with 250, 500 and 1 000 mM sodium chloride did not germinate. The addition of EDTA at concentration of 1.0 mM significantly ($p \leq 0.05$) reduced the salinity effect and enhanced germination of tomato seeds except for concentrations between 250 – 1000 mM where the effect of EDTA had no effect on the germination. Similarly, plumule and radicle lengths were significantly reduced with increase in concentration of NaCl. The results showed that higher concentration of NaCl significantly reduced the germination potential of tomato seeds and this can be reversed with the addition of EDTA when the concentration of NaCl was not above 100 mM.

Keywords: Ethylenediamine tetra acetic acid, Germination, Sodium Chloride, Tomato seeds.

INTRODUCTION

Tomato *(Solanum lycopersicum* Mill.) is annual crop belonging to Solanaceae family. It is one of the popular and widely grown vegetable crops in the tropics(Olayinka, *et al*., 2009). The plant is rich in Vitamin A, B and C, and iron (Awodoyin *et al*., 2007). It is also an excellent source of follate, potassium, vitamin C and E, flavonoids, chlorophyll, β-carotene and lycopene which are important for human health (Wilcox, *et al*., 2003).

Salinity is one of the environmental factors that severely limit crop production(Jamil *et al*., 2006; Mahmoodzadeh, 2009). The response of plant to salinity stress varies according to plant ontogeny. The first exposure of crop to salinity stress occurs at the germination stage in direct planting and transplanting (Demir and Mavi, 2008). Available literature had shown that high salinity impair seed germination, nodule formation, plant development and crop yield (Dan and Brix, 2007; Mujeeb-ur-Rahman *et al*., 2008). Zhu, (2002) had also found that high salinity and drought stress affect mostly all aspects of plant physiology and metabolism in terms of causing both hyper ionic and hyper osmotic stresses, which lead to plant demise. Shoji *et al.* (2006) demonstrated that high salinity affects cortical microtubule organization and helical growth in Arabidopis. Al-Karaki (2000), reported that high concentration of NaCl in nutrient solution adversely affected tomato shoot and roots, plant heights, K concentration, and K/Na ratio.

Ethylenediamine tetra acetic acid (EDTA) is a powerful chelating agent of metals with high affinity to form metal-EDTA complexes; it could deliberately be added to sequester metal ions. It has a wide range of applications due to its unique ability to form stable complexes with most metals over a wide range of environmental conditions (Mgbeze, *et al*., 2011). Addition of 1.0 mM EDTA has been found to reduced salinity stress on germination of two species of pepper (Mgbeze, *et al*., 2011).

Arising from the above, it becomes necessary to know the effect of this chelating compound on tomato plant by possible replacement reaction between Na^+ of NaCl and H^+ of EDTA. The fact that different plants vary in their response to salinity stress, the present investigation was carried out to determine the extent of salinity stress that could be tolerated by tomato and the ability of EDTA in ameliorating the impact of salinity stress on the germination and seedling growth of tomato.

MATERIALS AND METHODS

Seeds of tomato *(Solanum lycopersicum)* were obtained from National Horticultural Research Institute (NIHORT), Ibadan, Oyo State, Nigeria. The saline solutions were prepared by preparing 1M solution of NaCl. This was achieved by dissolving 55.44 g of NaCl in a final volume of 1liter or 1000 ml of distilled water. Since 1Mof NaCl solution is equivalent to 1000mM, dilutions were made to give 10, 50, 100, 250, 500, and 1000 mM sodium chloride. A control (0mM) was made with distilled water without NaCl.

About 1.0 mM EDTA was prepared by dissolving 1 mM of the salt in 1000 ml of distilled water. In each treatment, a Whatman filter paper was placed on a 9 cm Petri dish and ten seeds were arranged in a circular order in the Petri dish with two seeds at the centre to ensure adequate space between the seeds. Each level of sodium chloride was applied with the aid of a syringe and distilled water was applied to the control at ambient temperature. About 2.0 ml of sodium chloride and EDTA concentrations was applied at interval two days till the end of the experiment for 16 days for both salinity and EDTA treatments. EDTA (1 ml) was applied with 1ml of the saline solutions using a syringe. Data collected were germination count as indicated by the emergence of radicle. Plumule and radicle lengths were also measured using 10 cm meter rule at interval of 2 days. The layout of the petri-dishes followed complete randomized design (CRD) with three replication. Before planting the seeds on petri-dishes, seeds were soaked sterilized with 0.1% mercuric chloride and rinsed with several changes

Data were subjected to One Way Analysis of Variance. Duncan multiple range test was used to separate the means at 0.05 probability level using the Statistical Package for Social Science (SPSS) version 16.0.

RESULTS

The effect of salinity on the percentage germination at different periods is presented in Fig. 1. At all days studied, the highest percentage germination was recorded in the control and followed in decreasing order of magnitude by those treated with 10 mM, 50 mM and 100mM NaCl. Seeds treated with 250 mM, 500m M and 1 000 mM NaCl did not germinate at all days studied (Figure 1).

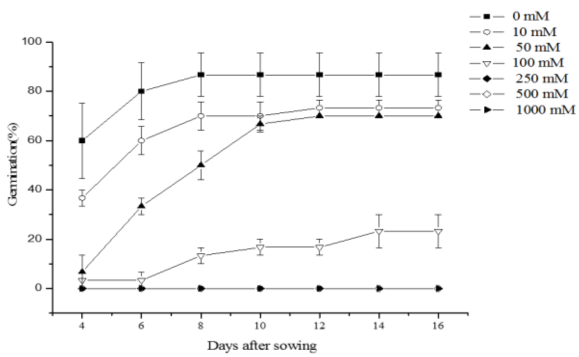

Figure 1: Effect of salinity on the germination of *Solanum lycopersicum*

Effect of salinity on the germination of *Solanum lycopersicum* seeds was significantly reduced when 1mM of EDTA was applied (Figure 2). At day 6, the highest percentage germination was observed in 50 mM which is in contrast to what was obtained when the seeds were treated with sodium chloride alone (Fig.1). This implies that germination of *Solanum lycopersicum* seeds was enhanced with addition of EDTA.

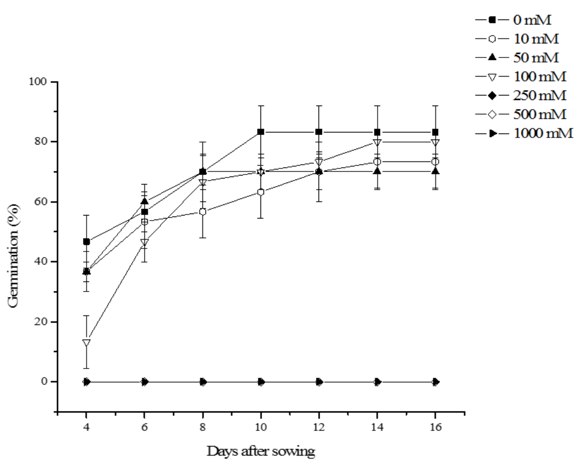

Figure 2: Effect of salinity and 1.0 mM EDTA on the germination of *Solanum lycopersicum* seeds

In the absence of EDTA, seedlings growth decreased with increase in concentration of NaCl (Table 1). As observed for germination, growth was arrested when the concentration of NaCl exceeded 100 mM with or without addition of EDTA (Table 1). The addition of EDTA significantly increased both the plumule and radicle lengths up to 100 mM NaCl (Table 1). This is evident by the Significantly highest plumule length recorded in 10 mM NaCl + 1.0 mM EDATA (6.83 ± 0.07) and followed in decreasing other of magnitude by those of the control (6.00 ± 0.58), 50 mM + 1.0mM EDTA (5.50 ± 0.50) and 100 mM + 1.0 mM EDTA (2.83 ± 0.17). The results of radicle length followed similar pattern as recorded for plumule except that significant differences were not recorded between control and 10mM of NaCl (Table 1).

Table 1: Effect of salinity with or without EDTA on the plumule and radicle lengths of *Solanum lycopersicum*

Treatment	Concentration (mM)	Plumule length (cm)	Radicle length (cm)
	0	6.00 ± 0.58^{b}	6.67 ± 0.88^{b}
	10	6.00 ± 0.76^{b}	6.83 ± 0.33^{b}
NaCl	50	3.17 ± 0.17^{d}	3.83 ± 0.44^{d}
	100	1.67 ± 0.33^{f}	2.00 ± 0.88^{f}
	250	0.00 ± 0.00^{g}	0.00 ± 0.00^{g}
	500	0.00 ± 0.00^{g}	0.00 ± 0.00^{g}
	1000	0.00 ± 0.00^{g}	0.00 ± 0.00^{g}
NaCl + EDTA	10	6.83 ± 0.17^{a}	7.67 ± 0.33^{a}
	50	5.50 ± 0.50^{c}	6.00 ± 1.00^{c}
	100	2.83 ± 0.17^{e}	3.17 ± 0.44^{e}
	250	0.00 ± 0.00^{g}	0.00 ± 0.00^{g}
	500	0.00 ± 0.00^{g}	0.00 ± 0.00^{g}
	1000	0.00 ± 0.00^{g}	0.00 ± 0.00^{g}
	Mean	2.69 ± 0.42	3.03 ± 0.47
	P-value	<0.001	<0.001

Values represented with the same superscripts along the column are not significantly different at $p<0.05$. Values are mean ± standard error of three replicates

DISCUSSION

High salinity had been variously reported as one of the environmental constraints that adversely affect crop productivity (Hamed *et al*., 2011; Ali and Gholam, 2014). The current results tend to confirm this statement in that germination of tomato seeds under relatively high concentration of NaCl (250-1000 mM) was completely hindered. However, germination of tomato seeds was not completely arrested when the concentrations of salt were between 10-100 mM. This showed that tomato seeds exhibited some degree of tolerance at these concentrations and this tolerance tends to attenuate with increase in concentration. The delayed in seeds germination with increase in the concentration most especially at 100 mM before germination was completely halted agreed with findings of Naseer and Sholi (2012) who reported decreased seed germination with increase in salinity most especially at the highest concentration of 150 mM Nacl in four cultivars of tomato. The delayed in seed germination under high salinity might be due to partial osmotic or ion toxicity which affect enzymes activity as pointed by other researchers (Croser *et al.,* 2001; Esa and Al-Ani, 2001). Zivkovic *et al*. (2007) who reported that salinity can affect germination by affecting the osmotic component, with the ionic component, involving accumulation of Na and Cl ions.

The results of the seedling growth in terms of plumule and radicle lengths followed similar trend as percentage germination. These results were in conformity with the results reported by several authors (Turhan *et al.,* 2009; Taffouo *et al.,* 2010; Naseer and Sholi, 2012) who found out that tomatoes grown in saline condition above 100 mM showed reduction in growth parameters.

In this study, the effect of salinity on germination and seedling growth of tomato was found to be ameliorated with the addition of

1.0 mM EDTA most importantly when the concentration did not exceed 100 mM. Similar results had been reported for pepper (*Capsicum annum* and *C. frutescens)* where addition of EDTA significantly reduced salinity stress by increasing percentage germination of the studied plants (Mgbeze *et al*., 2011). Aside from salinity stress, EDTA, has been reported to mitigate copper toxicity in *Brassica napus* by enhancing growth and biomass of this plant. The increased growth observed was traceable to ability of EDTA to bind copper ions (Habiba *et al.,* 2015). The mode of action of EDTA in reducing salinity stress as demonstrated in this study is due to its claw-like structure which binds and takes hold of metals to form a stable ring structure as earlier mentioned in the work of Mgbeze *et al*. (2011).

Conclusion

The present investigation had revealed that growth of tomato was grossly affected at higher saline conditions. Salinity stress as low as 10 mM and as high as 100 mM could be tolerated by the plant. Addition of EDTA had been found to be effective in promoting better establishment of the seeds most especially when tomato seeds were grown in saline condition not above 100 mM.

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