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The Effects of Different Concentrations of NaCl and NaCl - $CaCl_2$ Combinations on Germination, Emergence Performance and Seedling Growth of *Sorghum bicolor* L. Moench

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Author's contribution

The sole author designed, analyzed and interpreted and prepared the manuscript.

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ABSTRACT

Aim: The study was carried out to investigate the effects of different concentrations of NaCl and NaCl - $CaCl_2$ combinations on germination, emergence performance and seedling growth of *Sorghum bicolor* L. Moench
Study Design: The experiment was set up in a completely randomized design (CRD).
Place and Duration of the Study: The study was conducted at Department of Biological Sciences, Ondo State University of Science and Technology, Okitipupa, Ondo State, Nigeria, between, January, 2016 and June, 2016.
Methodology: Seeds of *Sorghum bicolor* were germinated in Petri-dishes which had been lined with Whatman No 1 filter papers. Treatments were control, NaCl (30, 60 and 120 mM) and combinations of NaCl and $CaCl_2$. The filter paper in each of the Petri-dishes allocated to the control was moistened with 10 mL of water while those of the Petri-dishes allocated to the other treatments were moistened with 10 mL of the different concentrations of the salt solutions. The germination percentage, germination rate, water content of the seedlings and the growth parameters were determined according to conventional methods.

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Results: The germination and germination rate of the seedlings were not significantly reduced by 30 mM salt solution. Treatment with higher concentrations of salt solutions gave a significant ($P < 0.05$) reduction in germination and seedling growth. The lowest germination percentage was observed in the 120 mM NaCl regime. As salinity increased the germination and seedling growth decreased. Combinations of NaCl and $CaCl_2$ improved the germination and seedling growth of the test crop compared to NaCl regimes

Conclusion: The germination and seedling growth of NaCl treated *S. bicolor* was significantly inhibited. Combinations of NaCl and $CaCl_2$ improved the germination and seedling growth of the test crop compared to NaCl regimes.

Keywords: Sorghum bicolor; salinity; water content; germination; seedling growth.

1. INTRODUCTION

Sorghum (*Sorghum bicolor* (L.) Moench) is an annual grass which belongs to the family Poacea. It is a cereal important for grain, forage and bioethanol production [1]. According to Mahajan and Tuteja [2], increased salinity of agricultural land is expected to have destructive global effects. Introduction of salt tolerant plants is one of the ways to utilize the waste saline water and lands [3]. It is reported that plants growing under saline conditions are affected in three ways: reduced water potential in root zone causing water deficit, phytotoxicity of ions such as Na^+ and Cl^- and nutrient imbalance depressing uptake and transport of nutrients. Na^+ competes with K^+ for binding sites essential for cellular functions [4]. High intracellular concentrations of both Na^+ and Cl^- can inhibit the metabolism of dividing and expanding cells [5], retarding germination and even leading to seed death. Increased salinity caused a significant reduction in germination percentage, germination rate, and root and shoots length and fresh root and shoots weights [6]. In vegetative plants, salt stress causes reduced cell turgor and depressed rates of root and leaf elongation [7].

Among the stages of the plant life cycle, seed germination and seedling emergence and establishment are key processes in the survival and growth of plants [8]. Seed germination is a critical stage in the history of plants and salt tolerance during germination is crucial for the establishment of plants that grow in saline soils. Salinity has many-fold effects on the germination process: it alters the imbibitions of water by seeds due to lower osmotic potential of germination media [9], causes toxicity which changes the activity of enzymes of nucleic acid metabolism [10], alters protein metabolism [11], disturbs hormonal balance [12] and reduces the utilization of seed reserves [13]. Seed germination, seedling emergence, and their survival are particularly sensitive to substrate salinity [14]. High levels of soil salinity can significantly inhibit seed germination and seedling growth, due to the combined effects of high osmotic potential and specific ion toxicity [15]. Germination rate and the final seed germination slow down with the decrease of the water movement into the seeds during imbibitions [16]. Intolerance to salinity may result in physiological and biochemical disorders which prevent or delay germination or cause abnormal seedlings [17]. High salinity may inhibit root and shoot elongation due to the lower water uptake by the plant [18]. Demir and Arif [19] observed that the root growth was more adversely affected as compared to shoot growth by salinity. According to Zhang et al. [20] salinity can decrease root water uptake through its osmotic effect, and subsequently induce water stress. Jeannette et al. [21] reported that total fresh weight of root and shoot of cultivated accessions of cowpea was reduced with increased salt stress. Generally the growth of plant is reduced by salinity but may vary from species to species in their tolerance [22]. The objective of this study was to investigate the effects of different concentrations of NaCl and NaCl - $CaCl_2$ combinations on germination, emergence performance and seedling growth of *Sorghum bicolor* L. Moench.

2. MATERIALS AND METHODS

The seeds of the test plant were selected randomly on the basis of uniformity of size and the seeds were then soaked for five minutes separately in 5% sodium hypochlorite to prevent fungal infection. Thereafter, they were rinsed for about five minutes in running tap water. Ten of the seeds were placed in each of the clean oven dried Petri-dish which had been lined with a Whatman No 1 filter paper. The filter paper in each of the Petri-dishes allocated to the control was moistened with ten millilitres of distilled

water while that of the Petri-dishes allocated to the other treatments were moistened with ten millilitres of the appropriate concentrations of the salt solution. The Petri-dishes were incubated at room temperature for two weeks. Emergence of one millimetre of the radicle was used as the criterion for germination.

Water uptake: Water uptake was recorded for 12 hours. Water uptake percent was calculated by the formula according to Mujeeb-ur-Rahman et al. [23].

$$WU = \frac{W2-W1}{W1} \times 100$$

W1 = Initial weight of seed

W2 = Weight of seed after absorbing water in a particular time.

Germination percentage (GP) was calculated according to the International Seed Testing Association (ISTA) method [24]

$$GP = \frac{\text{number of normally germinated seeds}}{\text{total number of seeds planted}} \times 100$$

Germination rate (GR) was calculated according to the method of Ellis and Robert [25].

$$GR = \frac{\sum N}{\sum nxg}$$

Where,

N: the number of germinated seeds; n: number of germinated seed on growth day and g: number of total germinated seeds.

The tissue water content (TWC) was calculated according to the formula of Black and Pritchard [26]

$$TWC = \frac{\text{Fresh weight} - \text{dry weight}}{\text{fresh weight}} \times 100$$

The plumule length, radicle length, radicle fresh weight and plumule fresh weight of each plant were determined. The fresh plants parts used for the fresh weight analysis were packaged separately in envelopes and dried to constant weight at 80℃ in a GallenKamp (Model IH-150) incubator. The dried plant parts were then weighed on a chemical balance (Mettler Toledo balance).

2.1 Statistical Analysis

A one way Analysis of Variance (ANOVA) was performed to determine significant ($P< 0.05$) effects. Means were compared by Duncan's test. *P*-values less or equal to 0.05 were considered statistically significant.

3. RESULTS AND DISCUSSION

Table 1 shows the effect of salinity on the germination of seeds of *Sorghum bicolor.* The percentage germination of the control was 76% while that of the 30 mM, 60 mM and 120 mM NaCl regimes were 75% 35% and 24% respectively. The percentage germination increased as the salt concentration decreased. The reduction of the germination of the seeds by the different salt solution was significant at $P < 0.05$. There was significant difference among the germination of the seeds in the 30 mM, 60 mM and 120 mM NaCl regimes. Decrease and delay in germination in saline medium have also been reported by Rahman et al*.* [27] and Mirza and Mahmood [28]. According to Umed- Ali- Soomro et al. [29], salinity affects germination in two ways: There may be enough salt in the medium that decrease the osmotic potential to such a point which retard or prevent the uptake of water necessary for mobilization of nutrient required for germination; the salt constituents or ions may be toxic to the embryo. Cramer et al. [30] suggested that ion ratios are important in determining relative toxicities of various ions and can provide insight into ion antagonisms. According to Wilson et al*.* [31], the increase in salinity shortens this ratio and probably caused injury to embryo. Water uptake by the seeds decreased with increase in salinity of the medium. The germination of the test crop was improved in mixtures of NaCl and $CaCl_2$ compared to NaCl regime (Table 2). The Ca^{2+} could have reduced the effect of the NaCl. According to Shabala et al.[32], the alleviating effect of Ca^{2+} in salinity stress is due, in part, to the ability of Ca^{2+} in decreasing the influx of Na^+ and the efflux of K^+ through the inhibition of non-selective cations and outward rectifying K^+ channels, respectively. This was consistent with the finding of Tuna et al. [33] who reported that supplemental Ca^{2+} can reverse the adverse effects of salinity on growth and membrane permeability of most plant species.

The plumule and radicle growth were inhibited by the application the different salt solutions. This

was consistent with the findings of [34] who stated that the growth of a plant is generally reduced by salinity. The result showed that the water content of the test crop in the treatment regimes was not significantly different from that of the control (Tables 3 and 4). This could be as a result of osmotic adjustment in the treated seedlings of *S. bicolor.* The effect of salinity on the biomass accumulation of *S. bicolor* is shown in Tables 5 and 6. The seedling fresh and dry weights were decreased by treatment with the salt solutions. This was in agreement with the findings of Gururaja et al. [35] who reported that salinity decreased biomass production.

Table 1. Effect of NaCl on the Germination and Growth of *Sorghum bicolor*

Treatments	Germination %	Germination Rate	Plumule length (cm)	Radicle length (cm)
Control	76 [a]	3.52 [a]	8.21 [a]	6.70[a]
30 mM	75[a]	3.48 [a]	6.54 [b]	5.40[b]
60 mM	35[b]	1.47 [b]	4.28 [c]	3.65[c]
120mM	24[c]	1.10 [c]	2.50[d]	2.00[d]

Means followed by the same letters are not significantly different according to Duncan test at 5% level

Table 2. Combined Effect of NaCl and $CaCl_2$ on the Germination and Growth of *Sorghum bicolor*

Treatments	Germination %	Germination rate	Plumule length (cm)	Radicle length (cm)
Control	76 [a]	3.52[a]	8.21[a]	6.70[a]
NaCl 30 mM+ 10 mM $CaCl_2$	80 [b]	2.65b	7.58 [a]	5.11[b]
NaCl 60 mM+ 10 mM $CaCl_2$	50 [c]	1.98c	5.24 [b]	4.02[c]
NaCl 120 mM+ 10 mM $CaCl_2$	46 [d]	2.00 d	2.87 [c]	2.60[d]

Means followed by the same letters are not significantly different according to Duncan test at 5% level

Table 3. Effect of NaCl on the water absorption and tissue water content of *Sorghum bicolor*

Treatments	Water absorption %	Water content %
Control	69.40[a]	88.22[a]
30 mM	58.34[b]	89.26[a]
60 mM	46.51[c]	88.75[a]
120 mM	40.57 [d]	89.82[a]

Means followed by the same letters are not significantly different according to Duncan test at 5% level

Table 4. Combined effect of NaCl and $CaCl_2$ on the water absorption and water content of *Sorghum bicolor*

Treatments	Water absorption %	Water content %
Control	69.40 [a]	88.22[a]
NaCl 30 mM+ 10 mM $CaCl_2$	68.80 [ab]	89.14[a]
NaCl 60 mM+ 10 mM $CaCl_2$	68.61[ab]	89.60[a]
NaCl 120 mM+ 10 mM $CaCl_2$	67.80 [b]	88.30[a]

Means followed by the same letters are not significantly different according to Duncan test at 5% level

Table 5. Biomass accumulation in *Sorghum bicolor* as affected by the different concentrations of NaCl

Treatments	Root fresh weight (g)	Plumule fresh weight (g)	Root dry weight (g)	Plumule dry weight (g)
Control	0.88[a]	2.11 [a]	0.16 [a]	0.31[a]
30 mM	0.62[b]	1.72 [b]	0.07[b]	0.12 [b]
60 mM	0.41 [c]	1.38 [c]	0.02 [c]	0.06 [c]
120 mM	0.16 [d]	1.02 [d]	0.02 [c]	0.06 [c]

Means followed by the same letters are not significantly different according to Duncan test at 5% level

Table 6. Biomass accumulation in *Sorghum bicolor* as affected by the different concentrations of NaCl and $CaCl_2$

Treatments	Root fresh weight (g)	plumule fresh weight (g)	root dry weight (g)	plumule dry weight (g)
Control	0 .88[a]	2.11[a]	0.16 [a]	0.31[a]
NaCl 30 mM+ 10 mM $CaCl_2$	0 .67 [b]	1.55 [b]	0.08 [b]	0.15[b]
NaCl 60 mM+ 10 mM $CaCl_2$	0.47 [c]	1.34 [c]	0.06 [c]	0.15[b]
NaCl 120 mM+ 10 mM $CaCl_2$	0.47[c]	1.01 [d]	0.06 [c]	0.08[c]

Means followed by the same letters are not significantly different according to Duncan test at 5% level

4. CONCLUSION

This study demonstrated that germination and seedling growth of salt treated *S. bicolor* was significantly inhibited by NaCl salinity stress. Combinations of NaCl and $CaCl_2$ improved the germination and seedling growth of the test crop compared to NaCl regimes. Therefore supplemental Ca^{2+} can reverse the adverse effects of salinity on the growth of *S. bicolor.* Researches should be directed towards understanding of the physiological and biochemical responses of plants that grow in saline environment.

COMPETING INTERESTS

Author has declared that no competing interests exist.

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