



## **Germination response of chemo-primed sorghum seeds to salinity**

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### **Abstract**

Salinity is one of the most important environmental elements affecting plant growth and development. The study hypothesised that priming sorghum seed with growth stimulators alleviates salinity stress. The seeds were chemically primed for 3 hours with 50, 150, and 250 ppm of Indole acetic acid (IAA) and Gibberellic acid (GA3), at the 3 concentrations each. After that, the seeds were subjected to sodium chloride (NaCl) stress at concentrations of 50, 150, and 250 mM NaCl. The time it took for seedlings exposed to 50 mM NaCl to germinate after priming with GA3 was shorter than the time it took for seeds in the control of distilled water (22.2 hours). Despite the use of primers, the results demonstrated that increased salinity concentrations were deleterious to seedling vigour, shoot and root growth e.g., root length decreased from 6.3 cm in the control to 0.90 cm when 150 ppm GA3 seeds were subjected to 250 mM NaCl. The findings imply that chemical priming with IAA and GA3 at concentrations of up to 150 ppm can hasten germination but other measures need to be employed to reduce deleterious effects of NaCl salinity >150 mM on plant growth and vigour.

Key words: Germination indices and time, gibberellic acid, indoleacetic acid, NaCl, seedling growth, vigour

### **Introduction**

Salinity is a global problem that affects around 7% of the world's total land surface, including 20% of all cultivated land and 33% of all irrigated land, resulting in 20% yield losses worldwide (Ikhajiagbe *et al.*, 2009; Jamil *et al.*, 2011; Ohanmu *et al.*,

2018). Because salinity is a global problem, it must be addressed in order to prevent plant yields from dwindling or perhaps disappearing. Salinised soil, which eventually impacts plants, is expected to ruin 10 million hectares of agricultural land every year (Pimentel *et al.*, 2004). Salinity is the saltiness or amount of salt dissolved in a body of water. Soils can acquire salinity due to mishandled irrigation (Jamil *et al.*, 2011). Salinity results from the building up Na<sup>+</sup>, K<sup>+</sup>, Ca<sup>2+</sup>, Mg<sup>2+</sup>, Cl<sup>-</sup>, HCO<sub>3</sub><sup>-</sup> and SO<sub>4</sub><sup>2-</sup> deposited by evaporating water (Herr, 2005; Ikhajiagbe *et al.* (2007a,b). In addition, in arid and semi-arid regions, the salinization process occurs because of high evaporation and inadequate amounts of precipitation for considerable leaching of salts that accumulate in the top layers of soil (Dai *et al.*, 2011). In plants, ionic stress and toxicity are caused by the absorption of excess Na<sup>+</sup> and Cl<sup>-</sup> ions from soils, which disrupt metabolic activities such as nucleic and protein metabolism, energy synthesis, and respiration (Mwando *et al.*, 2020).

The effect of salinity on plant growth is a complex trait that involves osmotic stress, ion toxicity, mineral deficiencies and physiological and biochemical perturbations (Seghatoleslami *et al.*, 2012). It limits the ability of plants to take up water as water gets into plant roots by osmosis controlled by the level of salt in the soil water and in the water contained in the plant (Sairam *et al.*, 2002). Elevated salinity generates reactive oxygen species (ROS), which damage macromolecules including proteins, carbohydrates, nucleic acids and lipids, or cellular structures like membranes (Ibrahim, 2016). The extent of unfavourable effects of salinity on plant growth and development range from inhibiting seed germination, to affecting leaf size, flowering, fruit set, maturation, and yield characteristics (Seckin *et al.*, 2009; Ratnakar and Rai, 2013). Thus, high salt concentration ultimately reduces crop yield and the quality of the produce (Sairam and Tyagi, 2004). According to Mbinda and Kintai (2019), the effect of the salt depends on the variety utilised and the level of salinity stress exerted. Seed germination is the start of the first developmental phase in the life cycle of higher plants and is key and susceptible stage of plant growth since the duration of this phase influences seedling establishment and future plant growth (Abo-Kassem, 2007; Hakim *et al.*, 2010; Rajjou *et al.*, 2012). For understanding the effects of salt, the stages of germination and emergence in sorghum (*Sorghum bicolor*) development have been deemed the most revealing stages of the plant's lifecycle (Krishnamurthy *et al.*, 2007). Sorghum, is a glycophyte and non-halophyte that is sensitive to salinity. Sorghum is a grass grown for its grain, which is used for human consumption, animal feed, and the production of ethanol. Sorghum is the world's fifth most significant grain crop, after rice, wheat, maize, and barley, with 59.34 million metric tons of annual global production in 2018 (FAOSTAT, 2020). Sorghum is also comparatively drought tolerant and is therefore deemed very essential in this era of climate change (). Therefore, is critical to improve its ability to survive in high-salt environments. To this effect, priming with plant growth stimulators such as indole

acetic acid (IAA) and gibberellins (GA3) have been widely used (Abd - El – Samad *et al.*, 1998; Fusun *et al.*, 2004; Cavusoglu and Sulusoglu, 2015). Priming is an induced state in which a plant reacts more quickly and effectively to a stress. It can be defined as an induced condition in which a plant reacts faster and effectively to a stimulus (Balmer *et al.*, 2015). A faster emergence may help to improve competitiveness of cultivated plants against weed species (Jalali and Salehi, 2013). The goal of this research is to discover if chemo-priming sorghum seeds before sowing improves their germination and growth qualities.

## Materials and methods

### *Study location*

The experiment was carried out in the pedigree laboratory of the Department of Plant Biology and Biotechnology, University of Benin, Benin city, Nigeria in 2021. Sorghum seeds (var. SAMSORG 47) were bought at a seeds store at Oba market, Ring Road area in Benin City.

### *Preparation of study compounds*

Distilled water was used in preparing all the solutions for growth stimulators and for salinity stress. Indole acetic acid (Sigma-Aldrich, CAS number 13750) and gibberellins (Sigma-Aldrich, CAS number 77-06-5) were the growth stimulators used in this study. IAA and GA, each, were studied at concentrations of 50, 150 and 250 ppm were measured; i.e., 0.05 g of IAA, and GA, respectively, were dissolved in 1000 ml of distilled water to get 50 ppm. Likewise, 0.15 g of IAA, and GA, respectively, were dissolved in 1000ml of distilled water to get 150 ppm, and 0.25 g of IAA, and GA, respectively, were dissolved in 1000ml of distilled water to get 250ppm. Sorghum seeds were soaked in glass beakers containing the different concentrations of Indole acetic acid (IAA) and gibberellic acid (GA) for 3 hrs and stirred at intervals for imbibition of the growth stimulators by the seeds that allows for partial hydration without radicle protrusion. The seeds were submerged in the respective solutions in the beakers at the rate of 100 seeds in 100ml of the respective solutions. Thereafter, from each batch, 20 seeds were dried and placed in petri dish.

For salinity stress, the concentrations 50, 150 and 250 mM (millimolar) were taken. For 50 mM, 2.9 g of NaCl were dissolved in 1000 ml of distilled water; 8.7 g of NaCl in 1000 ml of distilled water for the 150 mM; and 14.61 g of NaCl in 1000ml of distilled water for the 250 mM. The process involved picking the solutes a spatula, weighing on a precision electric balance (Model, MINGYI HZY-A120), while avoiding atmospheric haze; the air conditioner and/or fan were turned off to prevent vibrations which may alter the weights. Thereafter, the measured quantities were

placed in the conical flasks, and then distilled water was added gently and stirred for homogeneity before adding more distilled water to 1000 ml.

#### *Experimental design*

Seeds primed with different concentrations of the growth stimulators were tested under different (NaCl) concentrations as detailed in section 2.2 above. A control of distilled water was included in the study. The trial was done in triplicate.

#### *Data collection and parameter computations*

Germination and growth parameters studied included germination percentage computed as the percentage of the number of seeds that germinated in each petri dish. The shoot length was determined as the distance between the point of rooting and the shoot apical meristem, and the root length was measured as the average of 15 randomly measured main roots per plant. A measuring tape was used to determine the length of the leaf blade as well as the length of the sheath.

Foliar chlorophyll content index (CCI) was measured with the aid of a chlorophyll content meter; CCM – 200 plus, a non – destructive chlorophyll content measuring meter, which exploits the distinct optical absorbance characteristics of the chlorophyll in order to determine its relative concentration. The average meter reading of three leaves per petri dish was taken as the CCI.

Germination indices calculated were done according the methods of Baki and Anderson (1972); AOSA (1983); Scott *et al.* (1984); ISTA (1993); Al-Mudaris (1998); ISTA (1993); Sadeghi *et al.* (2001); Josep and Maria (2002). The germination indices that were calculated are:

*First day of germination (or Germinability) (FDG)* = day for first germination event.

*Last day of germination (LDG)* = day for last germination event.

*Final germination percentage (FGP)* = germination percentage attained by the plant even beyond the time period.

*Peak period of germination (PPG) or Modal time of germination (MTG)* = time in which highest frequency of germinated seeds are observed and need not be unique.

*Median germination time (Days required for 50% germination)* = the number of days required for 50% germination

#### *Germination rate index (GRI)*

The percentage germination on each day of the germination period is indicated by GRI.

It is calculated as,

$$\text{GRI } P\% [\text{GP1}/1 + \text{GP2}/2 + \text{GP3}/3 + \dots + \text{GPn}/n]$$

where: GP1 is germination percentage on 1<sup>st</sup> day; GP2 is germination percentage at two days; GPn is the germination percentage at *n* days.

Speed of accumulated germination (SAG)

$$\text{SAG } P\% [(GP1/1 + (GP1+GP2)/2 + (GP1+GP2+GP3)/3 + \dots + (GP1+GP2+GP3+\dots+GPn)/n]$$

Where: GP1 is germination percentage on 1<sup>st</sup> day; GP2 is germination percentage at 2 days; GPn is germination percentage at *n* days.

Corrected germination rate index (GRI corrected)

$$\text{GRI corrected} = \text{GRI} / \text{FGP}$$

Timson's Index, (TI) or Germination Energy Index (GEI)

$$\sum n = \sum_{i=1}^t G_i$$

$$\text{GEI} = (\text{GP1} + \text{GP2} + \text{GP3} + \dots + \text{GPn})$$

Where: GP1, GP2 + ... + GPn are the germination percentages at days 1, 2, ...n, respectively.

Modified Timson's Index, (TI<sub>mod</sub>)

TI<sub>mod</sub> = Timson's index (TI) divided by the number of intervals (*t*).

$$T_{mod} = \frac{T}{t}$$

Seedling vigour

$$\text{SVI (I)} = \text{Seedling length} \times \text{FGP}$$

$$\text{SVI (II)} = (\text{Root length} + \text{Shoot length}) \times \text{FGP}$$

$$\text{SVI (III)} = \text{Seedling dry weight} \times \text{FGP}$$

Time spread of germination, or Germination distribution (TSG)

$$\text{TSG} = \text{LDG} - \text{FDG}$$

This is the time (in days) taken between the first and last germination events.

Germination index, GI

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$$GI = 10 \times (S_1 + S_2 + S_3 + \dots + S_n) / (1 \times S_1 + 2 \times S_2 + 3 \times S_3 + \dots + n \times S_n)$$

Where: S<sub>1</sub>, S<sub>2</sub>, S<sub>3</sub>, S<sub>n</sub> are number of seeds that germinated per lot (or petri dish) at day 1,

day 2, day 3 ... day n

Mean daily germination, MDG

MDG = FGP / d, where d is the number of days it took to first arrive at the FGP

Daily germination speed, DGS

DGS = 1 / MDG. This is the reciprocal of MDG

Mean germination time, MGT

$$T = \frac{\sum_{i=1}^k N_i T_i}{\sum_{i=1}^k N_i}$$

Where: S<sub>1</sub>, S<sub>2</sub>, S<sub>3</sub>, S<sub>n</sub> are number of seeds that germinated per lot (or petri dish) at day 1,

day 2, day 3 ... day n. The lower the MGT, the faster a seed population has germinated.

It is also called Length of Germination Time (LGT) or Germination Resistance (GR) or Sprouting Index (SI). It is the average length of time required for maximum germination of a seed lot.

Germination Value (Czabator)

$$GV = PV \times MDG$$

Where: PV is the peak value and MDG is the mean daily germination percentage from the onset of germination.

Mean germination rate, MGR

$$MGR = 1 / MGT$$

Coefficient of velocity of germination, CVG

$$CVG = [(G_1 + G_2 + G_3 + \dots + G_n) / (1 \times G_1 + 2 \times G_2 + 3 \times G_3 + \dots + n \times G_n)] \times 100$$

Where: G<sub>1</sub>, G<sub>2</sub>, G<sub>3</sub>, G<sub>n</sub> are germination percent per lot (or petri dish) at day 1, day 2, day 3

... day n. CVG gives an indication of the rapidity of germination.

Germination capacity, GC

$$GC = FGP / N$$

Where: N is number of seeds used in the bioassay

Synchronization Index

Or Uncertainty of the germination process

$$E = - \sum_{i=1}^k f_i \log_2 f_i$$

Where:  $f_i$  Is the relative frequency of germination.

$(f_i = \frac{N_i}{\sum_{i=1}^k N_i})$ ,  $N_i$  is the number of seeds germinated on the  $i$ th time and  $k$  is the last day of observation.

#### *Data analysis*

The data were analysed using SPSS version 20. ANOVA was used to test the germination response to different levels of growth hormone stimulators, and salt (NaCl). Means were separated with the Fisher's least significant difference test at 5% level of significance.

## **Results**

#### *Effect on germination time and percentage*

Time to last germination and peak period of germination were significantly influence by treatments ( $P < 0.01$ ). The treatments also had a marginal effect on time to first germination ( $P = 0.072$ ) (Table 1). Germination time as a result of chemo-priming in salt exposed seeds showed that it took an average of 22.2 hrs for germination to occur in the control. This was not significantly different from the time it took for all treatments with the growth stimulators in combination with 50mM, and 150 Mm of NaCl. However, when 50 ppm and 150 ppm of GA3, and 50 ppm IAA were exposed to 250mM NaCl, the first time to germination reduced significantly by 24.2-33.3hrs (Table 1). In the control, the last time to germination was 94.9 hrs, which was not significant from seeds primed with GA3 at 150 and 250 ppm studied at the 250mM NaCl level and IAA at 50 ppm in 50 mM NaCl. Other levels of GA3 and IAA treatments significantly reduced the germination period (Table 1). The trend in last time to germination was the same for peak period of germination (hr) (Table 1). Median germination time (hr) was not significantly influenced by treatments ( $P > 0.05$ ; Table 1).

With regard to germination percentage, it was final germination percentage that was significantly influenced by the treatments ( $P < 0.001$ ). The final germination percentage of seeds in the control was 95.9%, this was not significantly different from that of seeds primed with GA3 and IAA at all the studied levels treated with 50 or 150mM of NaCl. When salinity increased to 250 mM of NaCl, germination percentage of the seeds was significantly reduced regardless of GA and IAA levels (Table 1). Figure 1(a) shows growing seeds in different treatments at three days after initiation.

Table 1. Effects of treatments on germination time and percentage germination

Treatments	Time to first germination (hr)	Time to last germination (hr)	Median germination time (hr)	Final germination percentage (%)
Control	22.2	94.9	42.4	95.9
50 mM NaCl+50 ppm GA3	22.2	46.4*	35.3	100
50 mM NaCl+150 ppm GA3	19.2	70.7*	40.4	100
50 mM NaCl+250 ppm GA3	13.1	70.7*	18.2	100
50 mM NaCl+50 ppm IAA	19.2	94.9	40.4	100
50 mM NaCl+150 ppm IAA	15.1	70.7*	22.2	100
50 mM NaCl+250 ppm IAA	17.2	70.7*	35.3	100
150 mM NaCl+50 ppm GA3	22.2	46.4*	42.4	95.9
150 mM NaCl+150 ppm GA3	27.3	70.7*	43.4	95.9
150 mM NaCl+250 ppm GA3	25.2	70.7*	36.3	95.9
150 mM NaCl+50 ppm IAA	27.3	70.7*	35.3	85.8
150 mM NaCl+150 ppm IAA	27.3	46.4*	27.3	95.9
150 mM NaCl+250 ppm IAA	23.2	46.4*	32.3	95.9
250 mM NaCl+50 ppm GA3	46.4*	70.7*	36.3	70.7*
250 mM NaCl+150 ppm GA3	49.5*	94.9	40.4	60.6*
250 mM NaCl+250 ppm GA3	28.3	94.9	36.3	75.7*
250 mM NaCl+50 ppm IAA	55.5*	70.7*	46.4	50.5*
250 mM NaCl+150 ppm IAA	27.3	70.7*	35.3	80.7*
250 mM NaCl+250 ppm IAA	28.3	70.7*	36.3	75.7*
Mean	27.8	71.1	35.8	89.1
LSD <sub>(0.05)</sub>	16.3	19.7	16.7	13.6
P-value	0.072	<0.001	0.162	0.009

\*Means within the same column having an asterisk differed significantly from the control (P<0.05)

#### *Effect on selected germination indices*

Table 2 shows the different germination indices of the different treatments. The germination rate index, which sums the germination percentages per day for the studied period, for the control was 119.6. This was only exceeded by that of seeds primed with 250ppm of GA3, and 50 or 150 ppm of IAA; at 50mM of NaCl. For speed of accumulated germination, the control was at 217.5. Again this was only exceeded by seeds treated with 250 ppm of GA3 and 50 ppm of IAA, at 50mM of NaCl. The Timson's index for the control was 290, exceeded only by that of 50 ppm of GA3 at 50 mM NaCl, which was at 300(Table 2).



Table 2. Effects of treatments on germination indices

Germination parameters	Control	GA3			IAA		
		50 ppm	150 ppm	250 ppm	50 ppm	150 ppm	250 ppm
----- 50 mM NaCl -----							
Germination rate index	119.6	75	108.3	145.8	124.2	130.8	115.8
Speed of accumulated germination	217.5	87.5	163.3	231.7	224.2	204.2	176.7
Timson's index	290	125	220	260	300	245	230
Modified Timson's index	72.5	62.5	73.3	86.7	75	81.7	76.7
Time spread of germination	72	24	51	57	75	55	53
Germination index	3.5	5.6	4.3	4.7	3.5	4.5	4.4
Mean daily germination	23.8	50	33.3	33.3	25	33.3	33.3
Mean germination time	2.9	1.8	2.3	2.1	2.9	2.2	2.3
Mean germination rate	0.3	0.6	0.4	0.5	0.3	0.5	0.4
Germination capacity	4.8	5	5	5	5	5	5
Seedling vigour I at day 3	285	290	380	370	300	300	260
Seedling vigour II at day 7	414.8	324.3	424	424	253.3	333.8	323.8
Seedling vigour III	1.9	2	3	3	4	3	2
----- 150 mM NaCl -----							
Germination rate index	119.6	72.5	84.2	65	73.3	108.8	67.5
Speed of accumulated germination	217.5	85	120.8	134.2	105	194.6	77.5
Timson's index	290	120	190	200	265	285	115
Modified Timson's index	72.5	60	63.3	66.7	88.3	71.3	57.5
Time spread of germination	72	24	43	45	43	19	23
Germination index	3.5	5.6	4.1	4.2	4.1	3.4	5.5

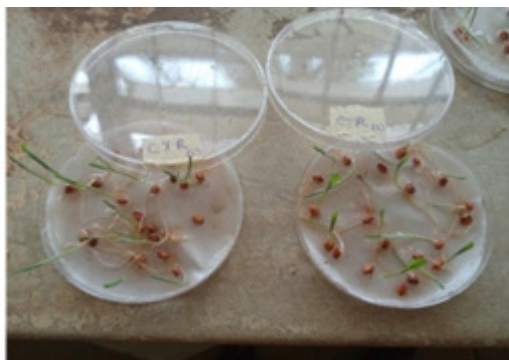
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Table 2. Contd.

Germination parameters	Control	GA3			IAA		
		50 ppm	150 ppm	250 ppm	50 ppm	150 ppm	250 ppm
Mean daily germination	23.8	47.5	31.7	31.7	28.3	23.8	47.5
Mean germination time	2.9	1.8	2.4	2.4	2.5	2.9	1.8
Mean germination rate	0.3	0.6	0.4	0.4	0.4	0.3	0.5
Germination capacity	4.8	4.8	4.8	4.8	4.3	4.8	4.8
Seedling vigour I at day 3	285	57	66.5	66.5	42.5	57	38
Seedling vigour II at day 7	414.8	69	78.4	59.5	45.1	49.9	50.4
Seedling vigour III	1.9	2.9	3.8	2.9	2.6	1.9	2.9
----- 250 mM NaCl -----							
Germination rate index	119.6	53.3	56.7	77.1	29.2	69.2	60
Speed of accumulated germination	217.5	73.3	98.3	135	37.5	99.2	80.2
Timson's index	290	130	160	210	75	155	140
Modified Timson's index	72.5	43.3	40	52.5	25	51.7	46.7
Time spread of germination	72	24	45	66	15	43	42
Germination index	3.5	3.9	3.3	3.3	3.8	4.1	4
Mean daily germination	23.8	23.3	15	18.8	16.7	26.7	25
Mean germination time	2.9	2.5	3.1	3	2.7	2.5	2.5
Mean germination rate	0.3	0.4	0.3	0.3	0.4	0.4	0.4
Germination capacity	4.8	3.5	3	3.8	2.5	4	3.8
Seedling vigour I at day 3	285	0	0	0	0	0	0
Seedling vigour II at day 7	414.8	1	0.9	1	0.8	1	1.1
Seedling vigour III	1.9	2.8	2.4	2.3	2	2.4	2.3

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(i) Control at day 3



(ii) Left – Right seeds primed with 50 ppm, 150 ppm and 250 ppm Gibberellic Acid exposed to 50mM Sodium Chloride at day 3



(iii) Left – Right seeds primed with 50 ppm, 150 ppm and 250 ppm Indole acetic Acid exposed to 50mM Sodium Chloride at day 3



(iv) Left – Right seeds primed with 50ppm, 150ppm & 250ppm Gibberellic Acid exposed to 150mM Sodium Chloride at day 3

Figure 1a. Germination of chemo-primed sorghum seed under different salt levels.

Taking the case of seedling vigour II [root length + shoot length]\*final germination percentage]at day 7, it was only GA3 at 150 and 250 ppm treatments at 50 mM of NaCl (both at 424) that had vigour higher than the control (414.8). However, with salinity over 50 mM, there was progressive reduction in seedling vigour to <1 readings at 250 mM NaCl. This was the also the case with seeding vigour I (seedling length\*final germination percentage) at 3 days, and seedling vigour III (seedling dry weight\*FGP) (Table 2). Generally, in spite of the growth stimulator that was used as the chemo-priming agent, the increased salinity still had its negative impairing effect on seedling vigour and germination percentage. Figure 1 (b) showcases a representation of the differences in shoot length at 5 day after initiation and the general decrease in growth with increasing salinity.

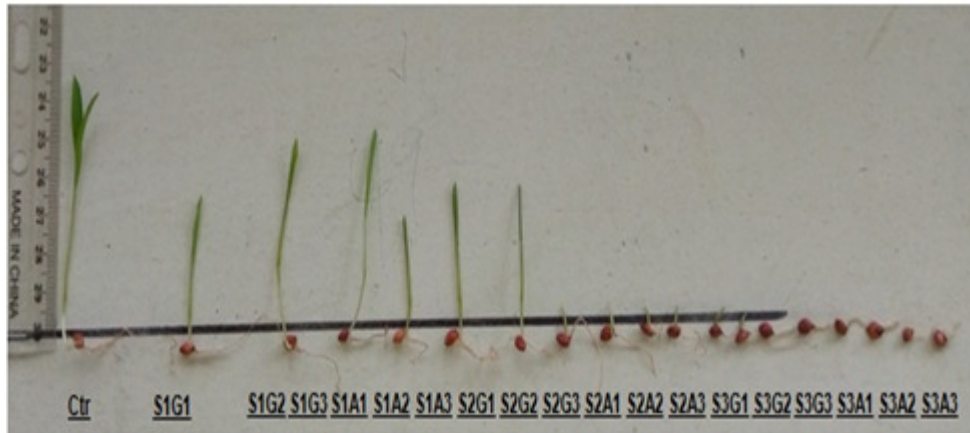


Figure 1b. Seedling growth of chemo-primed sorghum seed under different salt levels Where: Ctr: Control (water treatment), S1G1: 50 mM sodium chloride + 50ppm Gibberellic acid, S1G2: 50 mM sodium chloride + 150 ppm Gibberellic acid, S1G3: 50mM sodium chloride + 250 ppm Gibberellic acid, S1A1: 50mM sodium chloride + 50ppm Indole acetic acid, S1A2: 50mM sodium chloride + 150ppm Indole acetic acid, S1A3: 50 mM sodium chloride + 250ppm Indole acetic acid, S2G1: 150 mM sodium chloride + 50 ppm Gibberellic acid, S2G2: 150 mM sodium chloride + 150 ppm Gibberellic acid, S2G3: 150mM sodium chloride + 250 ppm Gibberellic acid, S2A1: 150 mM sodium chloride + 50ppm Indole acetic acid, S2A2: 150mM sodium chloride + 150 ppm Indole acetic acid, S2A3: 150 mM sodium chloride + 250 ppm Indole acetic acid, S3G1: 250 mM sodium chloride + 50 ppm Gibberellic acid, S3G2: 250 mM sodium chloride + 150ppm Gibberellic acid, S3G3: 250mM sodium chloride + 250 ppm Gibberellic acid, S3A1: 250mM sodium chloride + 50 ppm Indole acetic acid, S3A2: 250 mM sodium chloride + 150ppm Indole acetic acid, S3A3: 250 mM sodium chloride + 250 ppm Indole acetic acid.

#### *Effect on leaf growth and quality parameters*

Experimentation was terminated 12 days after germination initiation. At this time, plants in 250 mM NaCl had died and were not included in ANOVA. Therefore, the treatments of 50 mM and 150 mM NaCl only were used, and compared with the control. Shoot length, leaf blade length, sheath length and chlorophyll content were all significantly influenced by treatments ( $P < 0.05$ ; Table 3). Shoot breadth did not vary with treatments. Shoot length, leaf blade length and sheath length were highest in the control; and generally decreased with increasing salinity irrespective of growth stimulator with the setting of 150 mM especially producing very low values (Table 3). Chlorophyll content followed a different trend. Only GA3 at 150 ppm at 50 mM NaCl had distinctly lower, and IAA at 50 ppm at 150 mM NaCl higher than the control.

Table 3. Leaf growth and quality parameters on termination of experiment

Treatments	Shoot length (cm)	Leaf blade length (cm)	Leaf breadth (cm)	Sheath length (cm)	Chlorophyll content (CCI)
Control	10.08	5.25	1.16	6.51	2.312
50 mM NaCl+50 ppm GA3	7.56	3.78	0.92	4.83	1.894
50 mM NaCl+150 ppm GA3	9.45	4.41	0.88	4.62	2.181
50 mM NaCl+250 ppm GA3	10.08	4.2	1.17	4.83	2.131
50 mM NaCl+50 ppm IAA	5.46*	3.36	1.06	3.36	3.152
50 mM NaCl+150 ppm IAA	7.98	3.57	0.88	5.46	2.142
50 mM NaCl+250 ppm IAA	7.98	3.99	0.95	5.25	2.736
150 mM NaCl+50 ppm GA3	3.78	0.63*	0.51	1.05*	2.142
150 mM NaCl+150 ppm GA3	2.52*	0.41*	0.23	1.26*	2.205
150 mM NaCl+250 ppm GA3	3.36*	0.57*	0.21	0.84*	2.314
150 mM NaCl+50 ppm IAA	3.15*	0.55*	0.42	0.84*	2.941
150 mM NaCl+150 ppm IAA	3.36*	0.34*	0.37	0.84*	2.562
150 mM NaCl+250 ppm IAA	3.15*	0.21*	0.34	1.26*	2.52
Mean	5.99	2.41	0.7	3.15	2.39
Variance	8.58	3.75	0.13	4.7	0.14
LSD (0.05)	3.21	2.42	1.87	3.21	0.38
P-value	0.008	<0.001	0.204	0.002	0.066

\*Means differ from the control ( $P < 0.05$ )

Plants in 250 mM NaCl died before termination of experiment they were not included in the ANOVA

Results on root length are shown in Figure 2. The control had significantly the longest roots at 63 cm. Root length decreased with increasing NaCl levels. Treatments within the same salinity class were not significantly different in root length irrespective of the priming compound.

The uncertainty of the germination process is usually represented by the synchronisation index. The rule is, a lower synchronization index, when compared with the control, implies that germination may not necessarily occur, whereas, high synchronization index implies the possibility of germination to occur at higher frequencies. Results show that the synchronization index in the control was 31.8. The synchronization index in seeds primed in GA at 150 ppm and exposed to 50 mM of NaCl had a synchronization index of 36.8, which was more than the control. Similarly, when seeds were primed IAA at 150 ppm and exposed to 50 mM of NaCl, had a

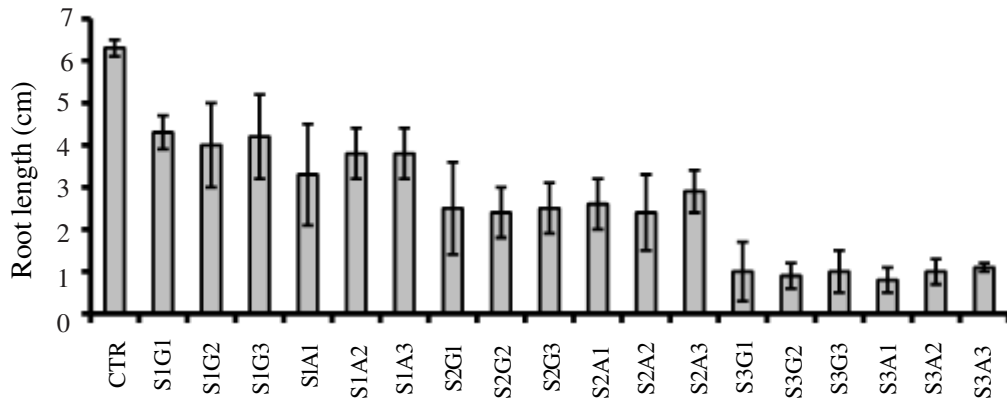


Figure 2. Root length of germinants in the different treatments.

synchronisation index was 55.87, also higher than the control. Synchronisation indices higher than those of the control implied the capacity for enhanced germinability and germination characteristics of the plants in spite of exposure to stress condition when chemically primed with growth stimulators like GA and IAA at moderate concentrations.

## Discussion

The addition of growth stimulants shortened the germination period, implying that the sorghum seeds germinated more quickly than they would have if the growth stimulators had not been added especially under imposed stress. Indole acetic acid and gibberellic acid (GA3) stimulate seed germination through mechanisms involving plant hormones, they are important for seed endosperm weakening, reserve mobilisation, cell division, and cell elongation during germination (Sachs 1887; Sung *et al.*, 2008). On the other hand, according to Bailly *et al.* (2008), oxidative stress was essential for triggering germination. Reactive oxygen species (ROS) are produced continuously during seed development, from embryogenesis to germination. They also play a dual role in the physiology of seeds by participating in cell signalling pathways. Several components of the ROS-mediated signaling pathways are activated during the first hydration phase of the priming process. All in all, priming treatments, mostly used before seed sowing, are anticipated to activate physiological and molecular pathways that allow the seed to react more quickly and/or more strongly after exposure to an environmental stress factor. Priming may thus afford a higher level of energy over a short time to sustain final germination (Nascimeto *et al.*, 2013).

Nevertheless, management of oxidative stress is an important component of resistance to a wide range of stress. The ultimate stress resistance in the seedlings may then be

linked to the persistence of the antioxidative defenses after final germination. Thus, the benefits of chemo-priming may be limited to decreasing germination time and but do not extend to enhancing plant growth and vigor after germination.

Extreme salinity (250 mM NaCl) reduced percentage germination and seedling vigour, even at high levels of chemo-priming. This indicated that the positive effects of chemo-priming did not hold as increasing levels of salinity. The absorption of excess Na<sup>+</sup> and Cl<sup>-</sup> ions from growth medium creates ionic stress and causes toxicity which contributes to disruption in biochemical processes including nucleic and protein metabolism, energy production and respiration (Maathuis *et al.*, 2014; Mwando *et al.*, 2020). High concentration of Na<sup>+</sup> in plant tissue inhibits uptake of K<sup>+</sup> which is an essential macronutrient for plant growth and development that results in low productivity, and may even cause death of the plant (Kronzucker *et al.*, 2013; Gupta and Huang 2014).

Leaf and root growth parameters also reduced with increase in salinity irrespective of chemo-priming with growth stimulators. These results concur with those of Jamil *et al.* (2006), who reported that increasing salinity significantly reduced germination percentage and rate, root and shoot length, and fresh and dry weights of the exposed plants. Other studies have echoed the fact that salt stress has a negative impact on growth and associated metabolites, as well as the final yield (Abdel-Lateef, 2005; Munns and Tester, 2008; Stanley *et al.*, 2016). Salinity affects photosynthesis mainly through a reduction in leaf area, chlorophyll content and stomatal conductance, and to a lesser extent through a decrease in photosystem II efficiency (Netondo *et al.*, 2004). These alterations in plant growth and related metabolites could be due to decrease in natural growth hormones in plant tissues (Fusun *et al.*, 2004). Exterogenous hormonal treatment were found to affect stress (Abd - El-Samad and El-Komy, 1998); in fact, Azooz *et al.* (2004) reported that IAA and gibberellic acid (GA3) stimulate growth in sorghum under stress conditions. Seed priming was proposed to trigger a sequence of physiological development that would increase performance of plants grown in salt stress, including the introduction of antioxidants systems (Varier *et al.*, 2010; Eisdand *et al.*, 2010). However, adding the growth stimulators did not succeed in counteracting the effects of salt stress on plant growth in this study.

## Conclusion

The study showed that sorghum seed primed with Indole acetic acid or gibberellic acid at concentrations of 50-250 ppm reduced the germination time under salt (NaCl) stress conditions in comparison to the control of distilled water. However, the growth stimulators did not bring appreciable advantage in germination percentage and seedling vigour; and there was a marked progressive growth impairment in leaf and root

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parameters under increasing salt stress conditions. Salinity was deleterious to the developing plants and chemical seed priming with growth stimulators could not counteract the effect.

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### **Conflicts of interest**

The authors declare no conflicts of interests.

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