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Proceeding Paper

Germination Performance of Different Sorghum Cultivars under Saline Conditions †

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Abstract: This study aimed to evaluate the influence of salinity in the germination of four sorghum cultivars. The experimental design used was completely randomized, in a 4 × 5 factorial arrangement, with four sorghum cultivars (BRS-373, BRS-380, BRS-658, and BRS-716) and five NaCl concentrations (0, 50, 100, 150, and 200 mM). The variables investigated were final germination percentage, first count, mean germination time, germination speed index, and sodium and potassium contents (radicle and aerial part). The findings demonstrated that the salinity levels interfered negatively in the germination performance of all cultivars, showing an expressive reduction, mainly in the highest concentration of NaCl.

Keywords: *Sorghum bicolor* (L.) Moench; abiotic stress; salinity; germination; seedlings

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1. Introduction

Salinity is one of the main abiotic factors that contribute to the reduced productivity of crops [1,2]. Due to low rainfall and high evaporation rates, semi-arid regions are the ones that most present this type of stress [3]. It is also enhanced by inadequate soil and water management, through the use of water containing high levels of salts [4], irrigation without a drainage system, and application of agricultural fertilizers with high salinity [3–5].

Changes in different biochemical and physiological processes occur in plant cells under saline stress [6]. The excess of salts present in the soil affects the germination, growth, and productivity of plants due to the osmotic, toxic, and nutritional effects that it provides, causing difficulties in water absorption and a series of imbalances in plant metabolism due to the accumulation of toxic ions, such as Na^+ and Cl^- [7,8].

Sorghum [*Sorghum bicolor* (L.) Moench], a grass that has its origin in Africa and Asia, presents high adaptability to semiarid regions [9], being characterized by its moderate tolerance to salt stress [10]. Due to the high production potential in dry regions and with problems with salinity, several studies have been carried out to select genotypes that are more tolerant to these conditions.

Larcher [11] affirms that the evaluation of the germination process would indicate the sensitivity of plants to salinity at later stages of growth and development. Hence, the assessment of sorghum cultivars during seed germination becomes necessary. Therefore, this study aimed to evaluate the effects of salinity in four sorghum cultivars during the germination phase.

2. Material and Methods

The experiment was conducted during August and September 2020 at the Seed Analysis and Plant Physiology Laboratories, belonging to the Department of Plant Science and Biochemistry and Molecular Biology, respectively, of the Federal University of Ceara (UFC), located in Fortaleza, Ceara, Brazil. Sorghum seeds of the cultivars BRS-373, BRS-380, BRS-658, and BRS-716, donated by Embrapa Maize & Sorghum, were used in this study. These seeds were submitted to conditions of salt stress and evaluated their germinative aspects and accumulation ratio of sodium (Na^+) and potassium (K^+) in the radicle and aerial part.

The most uniform seeds with intact integument of each cultivar were selected. Next, the seeds were disinfected in a 1% sodium hypochlorite solution for 1 min and then washed with distilled water. For the germination test, the seeds were uniformly placed between three sheets of blotting paper moistened with distilled water or sodium chloride solution (NaCl) in the proportion of 2.5 times the weight of the dry paper [12], obeying the different NaCl concentrations established (0, 50, 100, 150, and 200 mM).

For each treatment, 200 seeds were used, divided into four replicates of 50 seeds. Each group was distributed among three sheets of blotting paper substrate, using two sheets as a base and one to cover. Then, the three sheets were rolled, and the resulting rolls were placed inside polyethylene pots covered with transparent plastic. These materials were kept in a BOD-type germination chamber at a constant temperature of 25 °C [12], with variations adjusted to ±2 °C and under 12 h light/12 h dark photoperiod.

The evaluation of the percentage of germinated seeds (G) was carried out on the tenth day after sowing [12], being considered as a germination criterion the radicle emission with at least 2.0 mm in length in each treatment [13]. The first germination count (FGC) and the germination speed index (GSI) were performed together with the germination test. FGC was determined from the percentage of seeds germinated on the fourth day after installation of the trial [12] and GSI by the sum of the number of seeds germinated each day, divided by the number of days between sowing and germination [14]. Mean germination time (MGT) was also calculated, obtained through daily counts of germinated seeds until the tenth day after sowing [15], with the results expressed in days.

For the analysis of inorganic solutes, samples from dry matter of the aerial part and root system of the seedlings were removed and macerated to form a fine powder, which was stored in properly identified Eppendorf tubes for later use. Crude extracts were prepared according to the method of Rinne et al. [16], using 50 mg of powder from the aerial part or root system to 5 mL of deionized water. In test tubes, the freeze-dried powders of leaves and roots were added separately to deionized water. The samples were then shaken vigorously and incubated at 85 °C for 30 min. After that, the samples were centrifuged at 4000× *g*, at 25 °C for 20 min, and the supernatant (extract) was collected, filtered, and stored in glass flasks at −20 °C for later use. The determination of K^+ and Na^+ contents was performed by readings in the flame photometer for each properly diluted extract.

The experimental design used was completely randomized, in a 4 × 5 factorial arrangement, referring to four sorghum cultivars (BRS-373, BRS-380, BRS-658, and BRS-716) and five NaCl concentrations (0, 50, 100, 150, and 200 mM), with four replications. Data were submitted to analysis of variance, and when significant at 1 and 5% by the F test, regression analyzes were performed. For the statistical analysis and plot of the graphics, the computer programs "R" v. 4.0.2 [17] and "SigmaPlot 11.0" (Copyright© 2014 Systat Software Inc., San Jose, CA, USA), respectively, were used.

3. Results and Discussion

The results of the analysis of variance are shown in Table 1. All variables were significantly influenced at the level of 1% probability by the F test, both for isolated factors (Cultivars and Saline Concentrations) and for the interaction between them.

Table 1. Summary of analysis of variance for germination percentage (G), first germination count (FGC), germination speed index (GSI), mean germination time (MGT), sodium and potassium ratio in the radicle (Na^+/K^+ Ra) and aerial part (Na^+/K^+ Pa) of four sorghum cultivars submitted to five NaCl concentrations. Fortaleza, Ceara, Brazil, 2020.

Sources of Variation	DF	Medium Square					
		G	FGC	GSI	MGT	Na^+/K^+ R	Na^+/K^+ AP
Cultivares (C)	3	481.65 **	3542.7 **	120.70 **	4.12 **	112.03 **	7.668 **
Concentrações Salinas (CS)	4	245.80 **	671.0 **	149.99 **	2.80 **	639.81 **	46.491 **
Int. C × CS	12	29.90 **	47.5 **	0.85 **	0.07 **	24.81 **	1.615 **
Resíduo	60	8.15	21.1	0.56	0.03	2.18	0.124
Total	79	-	-	-	-	-	-
CV%	-	3.13	5.68	4.85	4.99	19	11.7

DF = Degree of freedom; CV = Coefficient of variation; ** Significant by F test at 0.01.

Figure 1a shows the linear behavior of the four cultivars studied toward the final germination percentage (G) as a function of the five NaCl concentrations. It is possible to observe that with the increase in salt concentrations, there was a reduction in the G variable of all cultivars in a similar way, except for cultivar BRS-373, which, when comparing the 200 mM treatment with the control treatment, exhibited greater sensitivity, showing a decrease of 19.3% in its germination. When compared to the other cultivars at the concentration of 200 mM, the cultivar BRS-373 presented a reduction of about 16.8% in its final germination.

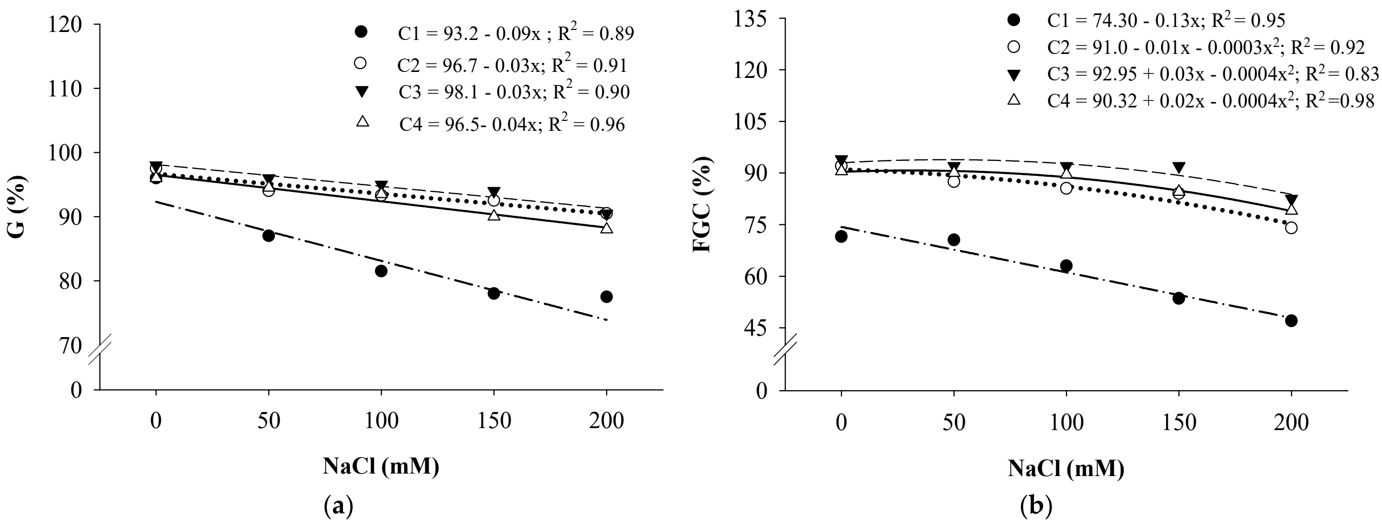

Figure 1. (**a**) Final germination percentage (G) and (**b**) First Germination Count (FGC) in seeds of four sorghum cultivars submitted to different saline concentrations. C1: BRS−373; C2: BRS−380; C3: BRS−658; and C4: BRS−716. Fortaleza, Ceara, Brazil, 2020.

The First Germination Count (FGC), shown in Figure 1b, presented similar behavior to the G, shown in Figure 1a. Likewise, cultivar BRS-373 exhibited results inferior to the other cultivars, being considerably more sensitive to the increase in salt concentration. Cultivar BRS-373 presented a 34.3% reduction in the FGC variable in the 200 mM treatment, while BRS-380, BRS-658, and BRS-716 had, respectively, a reduction of 19.6, 12.2, and 12.7%.

The same behavior was observed by Oliveira and Gomes Filho [18], who working with two forage sorghum cultivars (CSF 18 and CSF 20), found that germination is negatively affected by salt stress, which reduces the osmotic potential affecting the process. of imbibition and mobilization of seed reserves.

Initially, salinity causes water and nutrient deficiencies in the root that leads to metabolic changes in plants, especially with regard to germination, given that it is a water-dependent process for enzyme activation and reserve mobilization. Once there is this limitation of water, there is consequently a reduction in the speed of seedling development [6].

In line with the variables G and FGC, the Germination Speed Index (GSI) of the four cultivars, in Figure 2a, was also negatively affected by the increase in the number of salts. Nevertheless, the cultivar BRS-373 had lower GSI even in the 0 mM treatment, demonstrating that this delay in germination would be characteristic and associated with genetic factors. The cultivar BRS-380 was the most affected in the 200 mM treatment, revealing a 42.5% reduction in GSI, while BRS-373, BRS-658, and BRS-716 showed, respectively, reduction of 38.3, 38.0, and 35.8%.

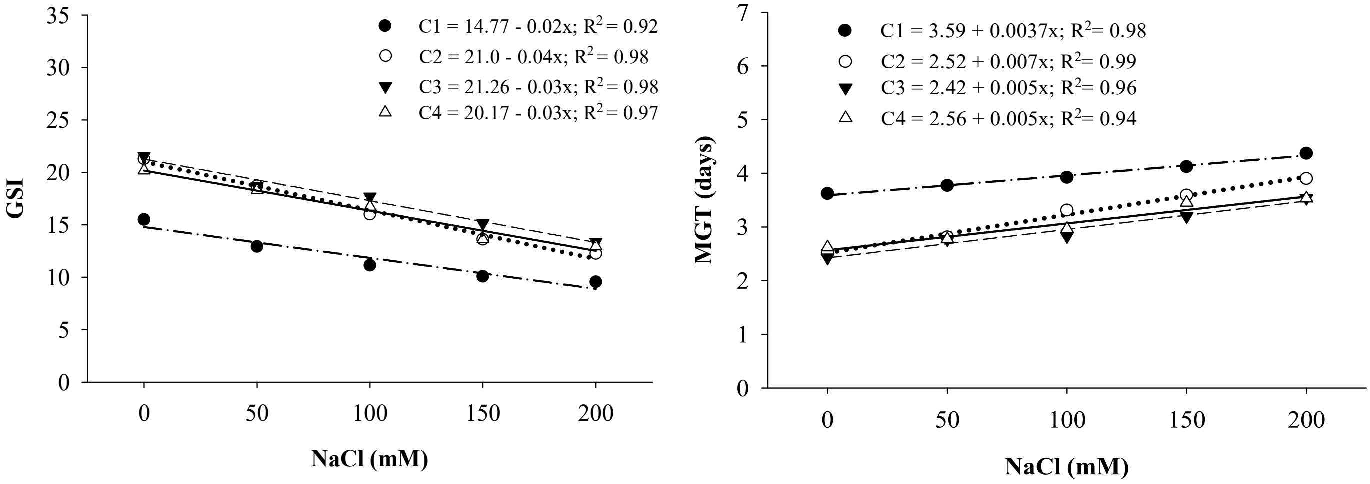

Figure 2. (**a**) Germination speed index (GSI) and (**b**) mean germination time (MGT) in seeds of four sorghum cultivars submitted to different saline concentrations. C1: BRS−373; C2: BRS−380; C3: BRS−658; C4: BRS−716. Fortaleza, Ceara, Brazil, 2020.

About the average germination time (MGT), it is shown in Figure 2b, that the cultivar BRS-380 presented the greatest delay in the germination process, reaching a 54.7% increase in the number of days compared to 200 mM treatment with the control, whereas BRS-373, BRS-658, and BRS-716 showed an increase of 20.7, 45.7, and 34.7%, respectively, in the number of days.

According to [19], the results of GSI and MGT might be influenced by the selected varieties, which could explain the discordant behavior of the cultivar BRS-373 regarding other cultivars BRS-380, BRS-658, and BRS-716, all similar to each other. Furthermore, [18] state that the germination speed index and the average germination time are among the variables most affected by salt stress, verifying that this promotes a reduction in vigor and germination speed of sorghum seeds, precisely because salt reduces the free water content near the seed and causes nutritional imbalances in developing organs through the action of potentially toxic specific ions, such as Na^+.

Figure 3 shows the Na^+/K^+ ratio in the radicle and aerial part of the seedlings. It is possible to note that as there is an increase in the salt concentration, there is a linear increase in the Na^+/K^+ ratio, showing, in turn, an effect competitive between both ions.

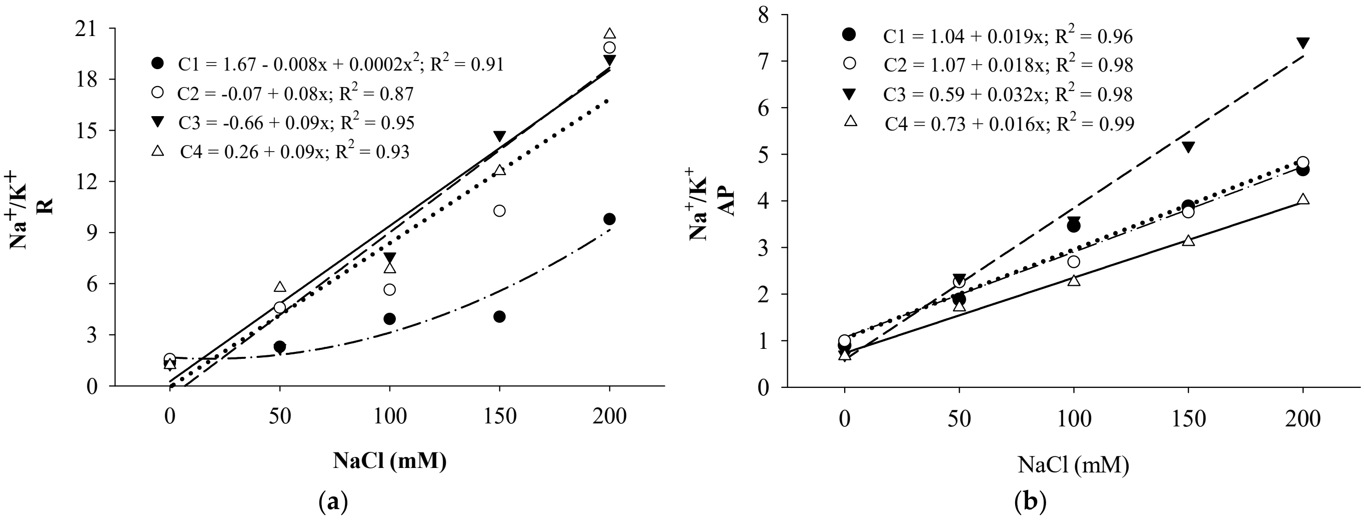

Figure 3. (**a**) Sodium and potassium (Na^+/K^+) ratio in the radicle and (**b**) in the aerial part of four sorghum cultivars submitted to different saline concentrations. C1: BRS−373; C2: BRS−380; and C3: BRS−658; C4: BRS−716. Fortaleza, Ceara, Brazil, 2020.

The Na^+ may potentiate the non-absorption or absorption in smaller amounts of K^+, considering that the two ions compete for the same transporter site, with the absorption of the ion that is in higher concentration [20]. Therefore, the Na^+/K^+ ratio tends to increase with increasing NaCl concentration, which can damage the ionic homeostasis of plant cells.

The Na^+/K^+ ratio is a very important factor for tolerance in plants under saline conditions [21]. Generally, salinity-tolerant cultures develop several physiological and biochemical mechanisms against this stress and one of them is the maintenance of a low Na^+/K^+ ratio in saline conditions through processes that aim at Na^+ exclusion, dilution, compartmentation and partitioning [22,23].

Cell ionic homeostasis is critical for physiology and the balance of K^+ and Na^+ is essential in saline stress situations in glycophytic cultures such as sorghum to improve salinity tolerance capacity [24]. When plants are exposed to saline stress, Na^+ competes with K^+ for absorption by plant roots, which can cause nutritional deficiencies of K^+ and trigger the impairment of all physiological processes dependent on this ion [25].

Figure 3, it is observed that, despite having accumulated sodium, the cultivars maintained the highest levels of sodium in the radicles, particularly, the cultivar BRS-716, which had the lowest values for the treatment of 200 mM for Na^+/K^+ ratio in the aerial part, indicating superior potassium levels to sodium. This result suggests that cultivar BRS-716 was efficient in compartmentalizing sodium in the radicle and not exporting it to the aerial part, reducing the effects of toxicity and competition.

A similar result was observed by [26], who worked with sorghum cultivars CSF 18 and CSF 20. They concluded that this sodium retention in the aerial part would prevent its accumulation in leaf tissues.

It is noteworthy that the cultivar BRS-373 had the lowest Na^+/K^+ ratio in the root and one of the lowest also in the aerial part, while BRS-658 had the highest values for this ratio, indicating the emergence of possible toxic effects by the Na^+ ion in the subsequent establishment of the plant.

4. Conclusions

The salinity levels interfered negatively in the germination performance of all cultivars, showing an expressive reduction, mainly in the highest NaCl concentration. Cultivar BRS-380 had the worst germination indices, with a delay in the germination process. Cultivar BRS-716 exhibited greater germination indices and the lowest sodium accumulation when compared to the potassium in the aerial part.

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