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# Sorghum (*Sorghum bicolor*) physiology and phytomass in saline-sodic soil treated with amendments and single superphosphate

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### Abstract

This study aimed to evaluate the physiology and phytomass accumulation of sorghum plants, in saline-sodic soil treated with amendments and doses of single superphosphate. The experiment was conducted in the period from January to June 2013, in a greenhouse in the municipality of Pombal, Parafba, Brazil, using samples of a saline-sodic soil. The experimental design was randomized blocks, in a 2 x 5 factorial scheme, relative to two types of amendments of sodic soil and 5 levels of single superphosphate, with 3 replicates, totaling 30 experimental units. Each plot was represented by two pots with capacity for 2 dm<sup>3</sup> with one plant each. At 30 days after sowing, sorghum plants were evaluated for gas exchanges, chlorophyll *a* fluorescence and phytomass accumulation. Single superphosphate application positively stimulated the gas exchanges of sorghum plants at the doses of 97.9 mg dm<sup>-3</sup>, for the soil treated with gypsum, and 88.1 mg dm<sup>-3</sup>, for the soil treated with elemental sulfur. Soil treatment with agricultural gypsum promoted the highest levels of chlorophyll *a* fluorescence, in comparison to elemental sulfur treatment. Phytomass accumulation was favored by the single superphosphate, with highest accumulation when the dose of 75 mg dm<sup>-3</sup> was applied.

Keywords: phosphate, gas exchanges, fluorescence.

### Introduction

Sorghum [Sorghum bicolor (L.) Moench] is a type of sorghum with high forage potential. It resembles sugarcane, for example, because of the high levels of fermentable sugars contained in its stem. Sorghum has been a promising alternative for ethanol production and forage material in semi-arid regions, such as in Northeast Brazil. This fact is related to its good adaptability and moderate tolerance to salt stress (Laei et al., 2011; Kausar et al., 2012). Although the potentiality of sorghum has been identified, it should be pointed out that salinity is one of the abiotic factors that most reduce growth and yield of crops (Munns and Tester, 2008), notably sorghum (Laei et al., 2011; Kausar et al., 2012). The negative effects of salts on plant growth have been associated with the component of osmotic tension, caused by the decrease in soil water potential and, consequently, by the restriction of water absorption by roots (Oliveira et al., 2011). However, there may also be effects of ionic nature, with accumulation of toxic ions in the leaves and the occurrence of nutritional imbalance, besides multiple effects (Reichards, 1954; Syvertsen and Garcia-Sanchez, 2014). To make possible the use of areas with salinity issues, such as salinization and sodicity, recovery strategies can be employed through the application of amendments (Miranda et al., 2011; Sá et al., 2013; Sá, et al., 2015). Furthermore, the use of amendments can be associated with fertilizers to reduce salinity effects on the soil, for example, phosphorus (P). However, the positive results of the effect of P fertilization on these soils are not elucidated, since some crops do not respond to phosphate fertilization under saline conditions. such as jatropha (Sousa et al., 2012), requiring further research. According to Khalil et al. (1967), the positive responses are related to the increase in P availability, because of the poor root development of the plants under saline conditions. This fact can assist in the absorption of P, since it is a primordial constituent of the adenosine triphosphate (ATP), energy currency in all physiological activities inside plants, in addition to its functions in the photosynthetic process (Taiz and Zeiger, 2013). Phosphate fertilization combined with soil correction treatments can assist in the development of plants used for remediation, such as sorghum. One of the ways to evaluate the responses of plants to salinity is by monitoring characteristics directly related to photosynthesis, such as gas exchanges and chlorophyll a fluorescence, which has been reported as successful in some studies (Mendonça et al., 2010; Silva et al., 2014), considering the relationship between P and the photosynthetic process. Hence, this study aimed to evaluate the physiology and phytomass accumulation of sorghum plants in salinesodic soil treated with amendments and doses of single superphosphate.

### **Results and Discussion**

There were significant responses for sorghum gas exchanges as a function of the different amendments and doses of single superphosphate (SSP) applied to the saline-sodic soil (Figs 1 and 2). In the soil treated with agricultural gypsum, the highest photosynthetic activities were observed when sorghum plants were fertilized with 97.9 mg dm<sup>-3</sup> of SSP, with value of 12.30  $\mu$ mol.m<sup>-2</sup>.s<sup>-1</sup> for CO<sub>2</sub> assimilation rate (Fig 1A). For the treatment with elemental sulfur, the highest net photosynthesis was observed at the SSP dose of 88.1 mg dm<sup>-3</sup>, with net CO<sub>2</sub> assimilation of 8.39  $\mu$ mol.m<sup>-2</sup>.s<sup>-1</sup>.

Thus, there were different responses of the plants to the studied amendments, in which the net CO2 assimilation was 31.8% higher in plants cultivated in soils treated with agricultural gypsum, in comparison to those cultivated in soils treated with elemental sulfur, requiring only 9.8 mg dm <sup>3</sup> more of SSP (Fig 1 A). These results are possibly related to the lower levels of sodicity in the soil treated with gypsum, in comparison to that treated with sulfur, especially due to the higher contents of calcium and magnesium present in the soil treated with gypsum (Miranda et al., 2011; Sá et al., 2013; Sá, et al., 2015), which can be observed in Table 3. As to the P levels, doses close to 100 mg dm<sup>-3</sup> promoted more promising results, which can be related to the availability of the nutrient in the soil, since single superphosphate is a fertilizer with low solubilization and, at doses higher than those, the nutrient was not satisfactorily released to the plants.

For the internal CO<sub>2</sub> concentration, sorghum plants cultivated in soil treated with gypsum also responded quadratically to the increase in SSP doses, which were reduced up to the dose of 111 mg dm<sup>-3</sup>, corroborating the increase in photosynthetic rate. These facts can be related to the low CO<sub>2</sub> flow into the substomatal chamber associated with the increase in RuBisCO activity, which can be identified with the evaluation of stomatal conductance (Fig 1B). Another point observed was that doses higher than this one promoted increase in Ci, even with the decrease of  $CO_2$ assimilation rate, which can be related to the low activity of the enzymes RuBisCO (Ribulose 1,5-Bisphosphate Carboxylase Oxygenase) (Machado et al., 2007) or PEP (Phosphoenolpyruvate Carboxylase), due to the imbalance caused by the inefficiency in the reduction of soil salinity observed for doses higher than 150 mg dm<sup>-3</sup> in the soil treated with gypsum, increasing the severity of the salt stress on the plants (Table 3).

Thus, SSP doses higher than 111 mg dm<sup>-3</sup> probably caused toxic effect on plants due to the increase in the content of calcium sulfate salts in the soil, which combined with sodium (Table 1), inhibits the photosynthetic process, through the reduction of  $CO_2$  concentration in the substomatal chamber, compromising the enzymes RuBisCO (Ribulose 1,5-Bisphosphate Carboxylase Oxygenase) and PEP (Phosphoenolpyruvate Carboxylase), due to the oxidative effect.

For plants cultivated in soil treated with sulfur, there was an increasing linear behavior, with increment of 0.188 ( $\mu$ mol m<sup>-2</sup> s<sup>-1</sup>) in *Ci* per unit increase in SSP doses (Fig 1 B). This behavior differed from that of the CO<sub>2</sub> assimilation rate,

which decreased at SSP doses higher than 81.1 mg dm<sup>-3</sup> applied to the soil (Fig 1 A). Considering that sorghum is a C4 species and that these plants are characterized by the presence of the PEP carboxylase enzyme, responsible for concentrating CO<sub>2</sub> in the stomatal chamber to improve the activity of RuBisCO (Taiz and Zaiger, 2013), this result allows to claim that the increase in *Ci* is mainly related to the activity of the PEP carboxylase.

Regarding the instantaneous carboxylation efficiency, there were linear reductions as a function of the increase in P doses on the order of 0.0014 for every increment of 1 mg dm<sup>-3</sup> of SSP in the soil containing sulfur (Fig 1C). However, this increase in *Ci* was positive up to the SSP dose of 88.1 mg dm<sup>-3</sup>, and sorghum plants cultivated in soils treated with sulfur reached maximum  $CO_2$  assimilation (Fig 1A). Possibly, doses higher than that caused nutritional imbalances in the plants, because of the concentration of calcium sulfate salts supplied via SSP associated with high contents of sodium present in the soil after the treatment, affecting the enzymatic activity and possibly the integrity of the chlorophyll, which can be observed in the evaluation of the fluorescence of the photosystem II (PSII).

On the other hand, it was observed that sorghum plants cultivated in soil treated with agricultural gypsum showed a quadratic behavior for EiCi as a function of the P doses, with maximum efficiency at the SSP dose of 100 mg dm<sup>-3</sup> (Fig 1C). These results corroborate the increase in  $CO_2$ assimilation rate, confirming that the addition of single superphosphate assists in the reduction of salt stress effects on plants, stimulating the performance of the photosynthetic activity. Since EiCi is a variable that relates the availability and consumption of CO<sub>2</sub>, Machado et al. (2007) report that the increase in the photosynthetic efficiency of the plants as a function of the P availability can be associated with the synthesis of the adenosine triphosphate (ATP) molecule, the energy currency required for the performance of the vital activities of the plant, especially in the photosynthetic process.

The stomatal conductance of sorghum plants showed a quadratic response as a function of the increase in SSP doses to the soil, regardless of the applied amendments (Fig 1 D). Maximum peaks of stomatal opening on the order of 0.075 and 0.042 (mmol of  $H_2O$  m<sup>-2</sup> s<sup>-1</sup>) were observed at the SSP doses of 100 and 75 mg dm<sup>-3</sup> for plants cultivated in soils treated with gypsum and sulfur, respectively (Fig 1 D). Hence, there was an increment of 44% in the stomatal opening of plants in soil with gypsum, in comparison to sulfur, because this variable is responsible for controlling all water losses of the plant and, consequently, it reflects the availability of water in the soil to these plants (Brito et al., 2013; Silva et al., 2014).

Thus, in the soil with gypsum, sorghum plants were probably under better water availability conditions due to the greater clay flocculation and soil aggregation, reductions in sodium contents and neutralization of carbonate in the soil (Sá et al., 2013; Sá et al., 2015), but a lower P availability, because phosphate decomposition occurs in acidic medium. However, in the soil with sulfur, this element acts in the reduction of soil pH, assisting the decomposition of SSP, releasing more phosphorus, calcium and sulfur into the soil than in soils treated with gypsum. Consequently, the reduction in the transpiration activity of the plants in the soil treated with sulfur can be related to the regulation of absorption of water and nutrients, due to their higher concentration in the soil.

As observed for stomatal conductance, there was also a quadratic behavior for the transpiration of sorghum plants as a function of the SSP doses. Plants cultivated in soil with gypsum showed 28% higher transpiration, compared with those cultivated in soil with sulfur, in relation to the maximum transpiration reached by the plants at the doses of 109 and 96 mg dm<sup>-3</sup>, respectively (Fig 1 E).

For stomatal conductance, although plants cultivated in soil treated with sulfur exhibited 44% less stomatal opening than those in soil treated with gypsum, their transpiration was only 28% lower in the comparison of maximum efficiency of stomatal conductance and transpiration (Figs 1 D and E). These results indicate that plants in soil treated with sulfur exhibited a greater water loss, possibly because of the necessity to regulate the internal temperature through the transpiration activity (Brito et al., 2013; Taiz and Zeiger, 2013), even under conditions of reduction in stomatal opening due to the salt stress or to the nutritional imbalance caused by the high levels of sulfur in the soil, from the amendment and SSP.

Regarding water use efficiency, there were linear reductions in plants cultivated in soil treated with elemental sulfur, corresponding to 0.0073 [( $\mu$ mol m<sup>-2</sup> s<sup>-1</sup>) (mmol H<sub>2</sub>O m<sup>-2</sup> s<sup>-1</sup>)<sup>-1</sup>] for every increase of 1 mg of SSP in the substrate. The values decreased up to the dose of 200 mg dm<sup>-3</sup>, reaching 5.33 ( $\mu$ mol m<sup>-2</sup> s<sup>-1</sup>) (mmol H<sub>2</sub>O m<sup>-2</sup> s<sup>-1</sup>)<sup>-1</sup> at the dose of 200 (mg dm<sup>3</sup>)<sup>-1</sup> (Fig 1 F). This variable reflects the relationship between CO<sub>2</sub> assimilation and water loss by the plant (Silva et al., 2014), which leads to the deduction that plants subjected to these conditions showed high levels of photorespiration, possibly reducing their photosynthetic potential, which can be evidenced by the evaluation of phytomass accumulation.

Plants cultivated in saline-sodic soil treated with gypsum showed a quadratic behavior regarding water use efficiency as the SSP doses increased. Maximum WUE of 6.51 (µmol  $m^{-2} s^{-1}$ ) (mmol  $H_2O m^{-2} s^{-1})^{-1}$  occurred at dose of 130.5 mg dm<sup>-3</sup>, decreasing to 6.02 [(µmol  $m^{-2} s^{-1})$  (mmol  $H_2O m^{-2} s^{-1})^{-1}$ ] as the SSP dose increased to 200 mg dm<sup>-3</sup> (Fig 1 F). It represents a reduction of 7.5% in WUE, which is low but significant, considering the increase in the costs with fertilizers.

The result observed in WUE of sorghum plants in soil treated with sulfur is contrary to that of plants in soil treated with gypsum, because in both cases the treatments of soil correction promoted large reductions in soil salinity (Tables 1 and 3). The increase in P doses probably promoted reduction in WUE because of a condition of increase in conductance and transpiration, in detriment of a smaller increment of net photosynthesis, which caused WUE to be reduced from 6.79 to 5.3 between the lowest and highest studied levels. However, these values are considered as normal for sorghum plants, even under stress (Santos et al., 2014).

Fertilization with single superphosphate did not influence the variables relative to chlorophyll a fluorescence of sorghum plants. However, there were different responses of fluorescence with respect to the soil correction treatments, except for PSII efficiency, which was not significant for any of the studied treatments (Fig 2 A, B, C and D).

In the soil treated with agricultural gypsum, sorghum plants obtained the highest contents of initial, maximum and variable fluorescence, equal to 286, 1078 and 792 (electrons quantum<sup>-1</sup>), in comparison to plants cultivated in the soil treated with sulfur, which obtained values of 254, 921 and 667 (electrons quantum<sup>-1</sup>) for these same variables. This corresponds to a superiority of 11.2%, 14.6% and 15.8% for the initial, maximum and variable fluorescence, respectively, of the plants in soil treated with gypsum (Fig 2 A, B and C). The increase in initial fluorescence corresponds to the destruction of the PSII reaction center (P680) or decrease in

the capacity of transfer of excitation energy from the antenna to the PSII (Baker and Rosenqvst, 2004). Hence, despite the higher initial fluorescence observed in sorghum plants cultivated in soil with gypsum, these plants did not show restrictions to gas exchanges in comparison to those cultivated in soil treated with sulfur. Thus, the low photosynthetic yields of plants in soil treated with sulfur are related to stomatal factors, because the PSII was not compromised (Freire et al., 2014; Silva et al., 2014).

In addition, it should be pointed out that plants cultivated in soil treated with gypsum also obtained the highest levels of maximum fluorescence (Fig 2B). The maximum intensity of fluorescence (Fm) denotes the state in which the PSII reaction centers reached their maximum capacity, evidencing a reduced condition of all the quinone (QA) due to the electrons transferred from the P680 (Munns and Tester, 2008; Syvertsen and Garcia-Sanchez, 2014). These results in higher photosynthetic efficiency, evidenced by the higher  $CO_2$ assimilation rate observed in plants under this treatment.

The variable fluorescence followed the same patterns observed for initial and maximum fluorescence, and plants cultivated in soil treated with gypsum exhibited the highest levels of variable fluorescence, 15.8% more than those in soil treated with elemental sulfur (Fig 2C). Thus, it can be claimed that plants in soil treated with gypsum showed a larger range of light absorption in comparison to the treatments with sulfur, which possibly favored the higher photosynthetic potential in these plants (Fig 1 A).

The reduction of variable fluorescence in plants cultivated in soil treated with sulfur, in comparison to the treatments with gypsum, indicates inhibition of the photochemical activity in the leaves, possibly due to the beginning of toxicity caused by the high levels of sulfur applied to the soil. For Freire et al. (2014), the reduction in the variable fluorescence of chlorophyll a of the plants evidences that the analyzed abiotic conditions lead to damages in the photosynthetic apparatus, compromising the PSII over the time of exposure to the stress factor of the stress intensity. However, despite the behavior observed in gas exchanges and reduction in the fluorescence levels of plants in the soil treated with sulfur and gypsum, due to the salinity of the soils after the treatment, there was no significant fit for the quantum efficiency of PSII, but the Fv/Fm levels were below 0.75 (Fig 2 D), which denotes damages in the photosynthetic apparatus of the sorghum plants (Baker and Rosenqvst, 2004; Lucena et al., 2012; Santos et al., 2014; Silva et al., 2014).

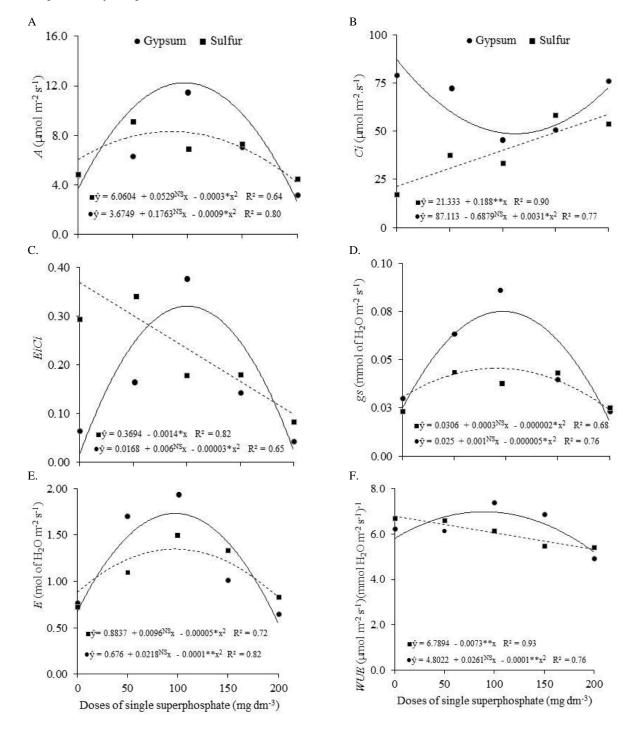
Regarding the total phytomass accumulation, there was a quadratic response as a function of the SSP doses, with maximum value at the dose of 75 mg dm<sup>-3</sup> (0.36g), indicating promising results of the mineral supplementation of the crop with single superphosphate in saline-sodic soils after soil correction treatment (Fig 3). These results indicate that soil correction, combined with mineral supplementation, can attenuate salt stress and increase the physiological activity of the crops, when properly performed, thus promoting higher assimilation of photoassimilates and greater conversion of biomass.

For this variable, no significant effect of the applied amendments was observed, indicating that, despite the alterations in the photosynthetic activity they were not sufficient to affect biomass conversion. It also confirmed by the results of PSII efficiency. However, as observed for variable fluorescence, the prolongation of the cultivation in soil with elemental sulfur under high SSP doses would cause negative effects on sorghum physiology (Fig 2C). Therefore, based on the total dry phytomass, SSP application at doses

**Table 1.** Characterization of the saturation paste of the sample of saline-sodic soil before and after correction with agricultural gypsum and elemental sulfur according to the methodology of Richards (1954).

Soil	pH	EC	Ca <sup>+2</sup> +Mg <sup>+2</sup>	$Na^+$	$K^+$	SAR	ESP
		dS m <sup>-1</sup>	mmol <sub>c</sub> L	1		$(\text{mmol}_{c} \text{ L}^{-1})^{0.5}$	(%)
Initial*	9.40	39.90	7.00	289.7	0.30	489.7	87.8
Gypsum*	8.17	5.32	19.60	27.3	0.40	8.71	10.39
Sulfur*	8.31	5.45	9.40	28.2	0.48	13.01	15.20

\*Initial soil sample; Soil after correction with gypsum; Soil after correction with elemental sulfur; EC = Electrical conductivity; SAR = Sodium adsorption ratio; ESP = exchangeable sodium percentage.



**Fig 1.** CO<sub>2</sub> assimilation rate, *A* (A), internal CO<sub>2</sub> concentration (*Ci*) (B), instantaneous carboxylation efficiency, *EiCi* (C), stomatal conductance (*gs*) (D), transpiration, *E* (E), and water use efficiency, *WUE* (F) of sorghum plants in saline-sodic soil treated with gypsum (—), elemental sulfur (---) and SSP doses. \* And \*\* = significant at 0.05 and 0.01 probability levels (p <0.05 and p <0.01); <sup>NS</sup> = not significant.

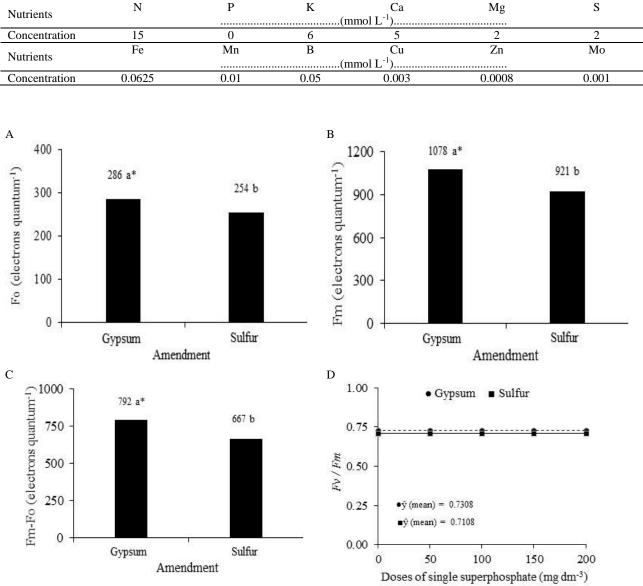


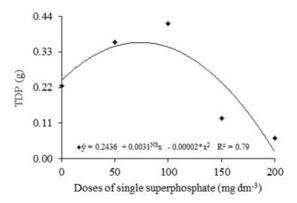
Table 2. Concentration of nutrients in the nutrient solution proposed by Hoagland and Arnon (1950), adjusted to the experiment.

**Fig 2.** Initial fluorescence, Fo (A), maximum fluorescence, Fm (B), variable fluorescence, Fm-Fo (C) and quantum efficiency of PSII, Fv/Fm (D) of sorghum plants in saline-sodic soil treated with gypsum (—), elemental sulfur (---) and SSP doses. Equal letters do not differ by Tukey test at 0.05 probability level.

Table 3. Chemical	characterization of	the soil after the	experiment.

Doses (mg dm <sup>3</sup> ) <sup>-1</sup>	Gypsum*				
Doses (ing dili )	EC	pH	SAR	ESP	
0	2.24	8.31	10.68	12.56	
50	7.34	5.96	7.11	8.41	
100	6.64	5.98	4.14	4.61	
150	6.33	5.82	4.72	5.39	
200	7.95	6.08	7.25	8.58	
$D_{1}$ ( 1 <sup>3</sup> )-1	Sulfur*				
Doses $(mg dm^3)^{-1}$	EC	pH	SAR	ESP	
0	0.88	8.13	7.49	8.88	
50	5.95	5.35	4.75	5.42	
100	5.88	5.44	4.50	5.08	
150	6.19	6.10	5.13	5.90	
200	6.08	5.55	5.09	5.88	

\*EC= electrical conductivity, dS m<sup>-1</sup>; SAR= Sodium Adsorption Ratio ((mmol L<sup>-1</sup>)<sup>-0.5</sup>); ESP= Exchangeable Sodium Percentage (%).



**Fig 3.** Total dry phytomass (TDP) of sorghum plants in saline-sodic soil treated with doses of single superphosphate. \* = significant at 0.05 probability level (p <0.05) and <sup>NS</sup> = not significant.

higher than 75 mg dm<sup>-3</sup> in saline-sodic soils treated with elemental sulfur is not indicated for sorghum cultivation.

### **Materials and Methods**

# Location, experimental procedure, treatments and plant material

The experiment was carried out in a greenhouse at the Center of Science and Agri-food Technology (CCTA) of the Federal University of Campina Grande (UFCG), Campus of Pombal, Paraíba, using samples of a saline-sodic soil. The samples were randomly collected at the depth of 0-40 cm, in a lot belonging to the Sector of Fruticulture of the Federal Institute of Education, Science and Technology of Paraíba (IFPB-PB), located in the Irrigation District of São Gonçalo, 10 km away from the municipality of Sousa.

After air-dried, pounded to break up clods and passed through a 2.0-mm-mesh sieve, the soil samples were taken to the Laboratory of Soils and Plant Nutrition of the CCTA/UFCG, where they were classified regarding salinity, using the values of pHse, ECse (pH and electrical conductivity of the saturation extract) and the soluble contents of Ca, Mg, K and Na, and ESP (Exchangeable sodium percentage), according to Richards (1954) (Table 1). Based on the results of the soil analysis, the soil was corrected through the application of agricultural gypsum or elemental sulfur, according to the treatment. The dose of agricultural gypsum (NG) was calculated based on the content of exchangeable sodium in the soil, to decrease the ESP to 15%, while the amount of elemental sulfur (S) was calculated using the analytical product (AP), whose dose was calculated based on the determined dose of gypsum, to provide the same amount of sulfur supplied by the gypsum.

After applying the amendments, the soils were maintained for 30 days with moisture corresponding to 70% of field capacity. The next step corresponded to the washing of the soils, through the application of a volume of water (EC < 0.3 dS.m<sup>-1</sup>) equivalent to two times the total soil porosity. After washing, 300 g of soil were collected and a new chemical characterization was performed, according to the previously described procedures (Table 1).

After the new chemical characterization of the soil, the second factor was applied, which corresponded to 5 doses of single superphosphate:  $P_1 = 0.0$ ;  $P_2 = 50.0$ ;  $P_3 = 100.0$ ;  $P_4 = 150.0$  and  $P_5 = 200.0$  mg dm<sup>-3</sup> of single superphosphate (18%  $P_2O_5$ ; 18% Ca<sup>+2</sup>; 10% SO<sub>4</sub><sup>-</sup>). The combination of the factors, in a randomized block design, resulted in 10 treatments (2

soil correction treatments (Cor) x 5 doses of single superphosphate (P)), with 3 replicates, totaling 30 experimental units, with each plot represented by two pots.

### Establishment and management of the experiment

After the doses of single superphosphate were applied, the soil was placed in pots with capacity for  $2 \text{ dm}^3$  and incubated for 20 days with moisture content close to the maximum retention capacity, before sowing.

Water replacement during the incubation and irrigations were based on the drainage lysimetric method, and the applied water depth was added of a leaching fraction of 15%. The volume applied (Va) per pot was obtained by the difference between the previous water depth (Lp) minus the mean drainage (D), divided by the number of pots (n), as indicated in equation 1:

 $Va = \frac{Lp-D}{n(1-LF)}$ 

Eq. 1

After incubation, three seeds were planted in each pot, totaling 6 seeds per plot. Thinning was performed when the plants showed two true leaves, maintaining only one plant in each pot. The substrate for plant development was the corrected soil mixed with the respective doses of single superphosphate. Supplementary fertilization was applied using the modified solution of Hoagland and Arnon (1950) at the proportion of 5% of the substrate volume at 10 days after sowing, when the total emergence of the plants was achieved (Table 2).

### Traits measured

Thirty days after sowing, sorghum plants were evaluated for gas exchanges, based on the measurement of CO<sub>2</sub> assimilation rate (*A*) (µmol m<sup>-2</sup> s<sup>-1</sup>), transpiration (*E*) (mmol of H<sub>2</sub>O m<sup>-2</sup> s<sup>-1</sup>), stomatal conductance (*gs*) (mol of H<sub>2</sub>O m<sup>-2</sup> s<sup>-1</sup>) and internal CO<sub>2</sub> concentration (*Ci*) (µmol m<sup>-2</sup> s<sup>-1</sup>) on the third mature leaf counted from the apex. These data were used to calculate the water use efficiency (WUE) (*A/E*) [(µmol m<sup>-2</sup> s<sup>-1</sup>) (mmol H<sub>2</sub>O m<sup>-2</sup> s<sup>-1</sup>)<sup>-1</sup>] and instantaneous carboxylation efficiency (*EiCi*) (*A/Ci*) (mol.m<sup>-2</sup>.s<sup>-1</sup>) (Silva et al., 2014), using the portable photosynthesis meter "LCPro+" (ADC Bio Scientific Ltda.).

On the same leaves where gas exchanges were analyzed, leaf clips were placed and, after a period of 30 minutes of adaptation to the dark, the following variables were determined: initial fluorescence (Fo) (electrons quantum<sup>-1</sup>), maximum fluorescence (Fm) (electrons quantum<sup>-1</sup>), variable

fluorescence (Fv= Fm-Fo) (electrons quantum<sup>-1</sup>) and the quantum efficiency of photosystem II (Fv/Fm) (Mendonça et al., 2010), using the device PEA - Hansatech.

Also 30 days after sowing, plants were collected for the determination of total dry phytomass (TDP) (g). The material was collected, placed in a forced-air oven at 65 °C for drying (72 h) and then weighed on analytical scale.

At the end of the experiment, the substrates were collected and analyzed for salinity, to verify the saline character of the soil, based on the evaluation of electrical conductivity (EC dS  $m^{-1}$ ), pH, sodium adsorption ratio (SAR) ((mmol L<sup>-1</sup>)<sup>-0.5</sup>) and exchangeable sodium percentage (ESP) (%) of the saturation paste, according to the methodology of Richards (1954) (Table 3).

### Statistical analysis

The obtained data were evaluated through analysis of variance by F test. In cases of significance, the F test was conclusive for the factor 'amendment' and the polynomial regression analysis (linear and quadratic) was applied for the factor 'doses of single superphosphate', using the program SISVAR (Ferreira, 2011).

#### Conclusion

Single superphosphate application positively stimulated the gas exchanges of sorghum plants at the doses of 97.9 mg dm<sup>3</sup>, for the soil treated with gypsum, and 88.1 mg dm<sup>3</sup>, for the soil treated with elemental sulfur. Soil treatment with agricultural gypsum promotes the highest levels of chlorophyll *a* fluorescence, in comparison to elemental sulfur. Phytomass accumulation was favored by the doses of single superphosphate, with highest accumulation when the dose of 75 mg dm<sup>3</sup> was applied.

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