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Original Article

Phosphate fertilization, biofertilizer and *Bacillus* sp. in peanut cultivation under salt stress[1]

Adubação fosfatada, biofertilizante e *Bacillus* sp. no cultivo de amendoim sob estresse salino

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HIGHLIGHTS:

Inoculation with Bacillus *sp. in peanut is effective in mitigating salt stress.*
Organic fertilization with bovine biofertilizer is effective in the growth and gas exchange of peanut grown under salt stress.
Bacillus *sp. mitigates salt stress by increasing water use efficiency in the peanut crop.*

ABSTRACT: Peanut (*Arachis hypogaea*) is an important agricultural crop in Brazil, considered one of the most important oilseed crops cultivated. The use of fertilizer-solubilizing bacteria can mitigate salt stress. The objective of this study was to evaluate the gas exchange, growth, and water use efficiency of the peanut crop irrigated with brackish water under phosphate fertilization and inoculated with bacteria of the genus *Bacillus* sp. The experimental design used in this study was completely randomized, in a 4 × 2 × 2 factorial scheme, with 5 replicates, referring to the forms of fertilization (F1 - 0% phosphorus, F2 - 50% phosphorus, F3 - 100% phosphorus, and F4 - bovine biofertilizer), presence and absence of the inoculant and two levels of electrical conductivity of the irrigation water (ECw – 0.3 and 4.0 dS m^{-1}). Fertilization with organic fertilizer (100% biofertilizer) and mineral fertilizer (50 and 100% phosphorus) associated with *Bacillus* sp. mitigated the damage caused by salt stress and promoted greater water use efficiency, chlorophyll index, internal CO_2 concentration and stem diameter. The control treatment (without phosphate fertilization and without salt stress) and the application of *Bacillus* sp. promoted greater performance in net photosynthesis, transpiration rate, stomatal conductance, and plant height in peanut plants.

Key words: *Arachis hypogaea* L., plant nutrition, microorganisms

RESUMO: O amendoim (*Arachis hypogaea*) é uma importante cultura agrícola no Brasil, considerada uma das mais importantes oleaginosas cultivadas. O uso de bactérias solubilizadoras de fertilizantes pode mitigar o estresse salino. O objetivo deste estudo foi avaliar as trocas gasosas, o crescimento e a eficiência do uso da água da cultura do amendoim irrigado com água salobra sob adubação fosfatada e inoculada com bactérias do gênero *Bacillus* sp. O delineamento experimental utilizado neste estudo foi inteiramente casualizado, em esquema fatorial 4 × 2 × 2, com 5 repetições, referente às formas de adubação (F1 - 0% fósforo, F2 - 50% fósforo, F3 - 100% fósforo e F4 - bovino biofertilizante), presença e ausência do inoculante e dois níveis de condutividade elétrica da água de irrigação (CEa – 0.3 e 4.0 dS m^{-1}). As adubações com fertilizante orgânico (100% de biofertilizante) e mineral (50 e 100% de fósforo) associada a *Bacillus* sp. mitigou os danos causados pelo estresse salino e proporcionou maior eficiência no uso da água, índice de clorofila, concentração interna de CO_2 e diâmetro do caule. O tratamento controle (sem adubação fosfatada e sem estresse salino) e a aplicação de *Bacillus* sp., promoveu maior desempenho na fotossíntese líquida, taxa de transpiração, condutância estomática e altura das plantas em plantas de amendoim.

Palavras-chave: *Arachis hypogaea* L., nutrição de plantas, micro-organismos

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Introduction

Peanut (*Arachis hypogaea* L.) belongs to the Fabaceae family and is one of the main agricultural crops in Brazil and other countries, which makes it one of the most important cultivated oilseeds (Arruda et al., 2015). Peanuts are the second most cultivated oilseeds in Brazil, reaching a production of 893 thousand tons in the 2022/2023 harvest (CONAB, 2023).

In semi-arid regions, agricultural crops are subjected to the effects of abiotic factors, such as water and salt stress. The use of brackish water irrigation causes a reduction in the soil's osmotic potential and, consequently, in water and nutrient consumption, causing restrictions in physiological activities and reductions in crop yields (Barbosa et al., 2022; Silva et al., 2022).

An alternative way to mitigate salt stress is through the use of microorganisms, which colonize the roots and act by secreting various products such as phytohormones and antioxidants and improving the solubility and absorption of nutrients (Castaldi et al., 2023). Bacteria from the *Bacillus* genus such as *Bacillus subtilis* and *Bacillus megaterium* are capable of developing adaptive activities at higher salt concentrations and improving the availability of phosphorus to plants (Liu et al., 2020).

Mineral or organic fertilization is another way to mitigate salt stress. The use of phosphate fertilizer as a mineral source can promote greater action on root growth, fruit maturation, and the formation of grains and fruits (Prado, 2020). Studies carried out by Silva et al. (2022) revealed a positive effect on peanut cultivation under phosphate fertilization and salt stress. Likewise, Ribeiro et al. (2022b) described attenuation by phosphate fertilizer in cowpea (*Vigna unguiculata* L.) crops irrigated with brackish water. Bovine biofertilizer, as an organic source, can improve the chemical and microbiological properties of the soil and mitigate salt stress (Sousa et al., 2018; Souza et al., 2019).

With this in mind, the objective of the present study was to evaluate the gas exchange, growth, and water use efficiency of the peanut crop irrigated with brackish water under phosphate fertilization and inoculated with bacteria of the *Bacillus* genus.

Material and Methods

The experiment was conducted during February and April 2023 at the Auroras Seedling Production Unit (UPMA), located at the Universidade da Integração Internacional da Lusofonia Afro-Brasileira - UNILAB, Auroras Campus, Redenção, Ceará, Brazil. The city is located at a latitude of 04° 14' 53" S, a longitude of 38° 45' 10" W and an altitude of 240 m. The region's climate is of the BSh' type, as very hot temperatures and rainfall prevail in the summer and fall seasons (Alvares et al., 2013). The meteorological data obtained during the period of the experiment is shown in Figure 1.

The design used was completely randomized, in a 4 × 2 × 2 factorial scheme, with 5 replications, referring to the forms of fertilization (F1 - 0% phosphorus, F2 - 50% phosphorus, F3 - 100% phosphorus, and F4 - bovine biofertilizer), presence (PI) and absence of the inoculant (AI) and two levels of electrical conductivity of the irrigation water (ECw - 0.3 and 4.0 dS m^{-1}).

Peanut cultivar BR-1 was sown at a depth of 2 cm, in polyethylene pots with a capacity of 11 dm^3 containing as a substrate a mixture of arisco (light-textured sandy material normally used in constructions in Northeast Brazil), sand, and cattle manure in a ratio of 7:2:1 (v/v), respectively, whose chemical characteristics are shown in Table 1.

The seeds were inoculated with the commercial product BiomaPhos® as recommended by the manufacturer (100 mL of the product for every 60.000 seeds). The product contains a mixture of the bacteria strains BRM 119 (*Bacillus megaterium*) and BRM 2084 (*Bacillus subtilis*).

The enriched bovine biofertilizer was prepared under aerobic fermentation with a mixture of equal parts of fresh bovine manure and non-salty water (ECw = 0.3 dS m^{-1}). The mixture was stored in a 100 L container and underwent a

Table 1. Chemical characteristics of the substrate used before the treatments were applied

OM	N	P	K	Ca	Mg	Na	pH	ESP	ECse
(g kg^{-1})							H_2O	(%)	(dS m^{-1})
0.8	0.21	0.06	0.28	0.07	0.03	0.11	6.5	3.4	0.37

OM - Organic matter; ESP - Exchangeable sodium percentage; ECse - Electrical conductivity of the saturation extract

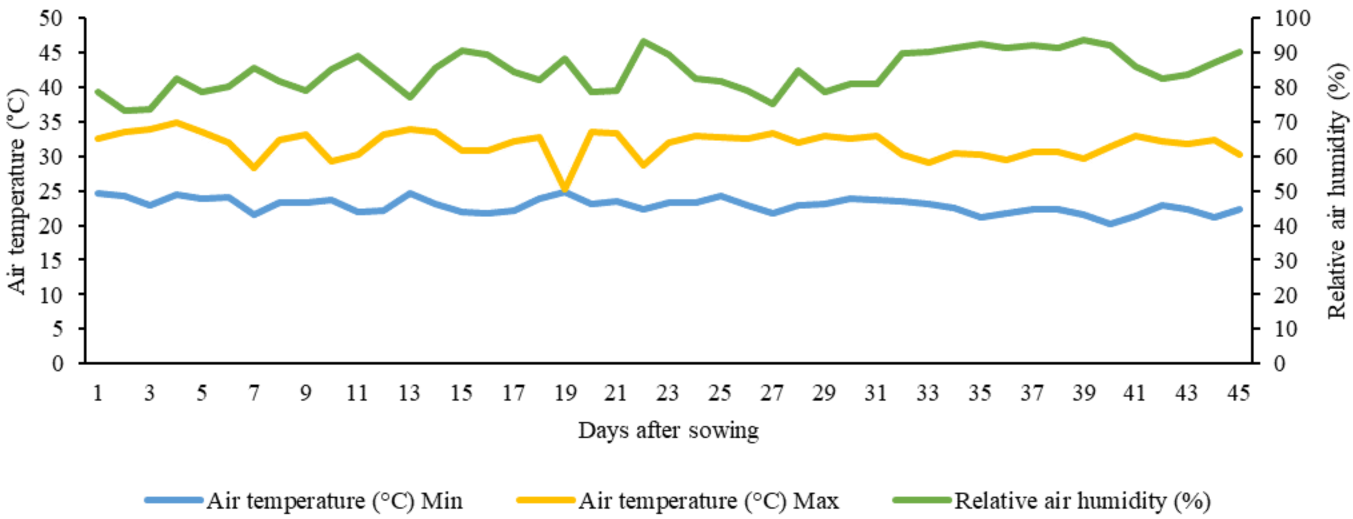


Figure 1. Mean values of minimum (Min) and maximum (Max) temperatures, and relative air humidity observed during the experimental period from February to April 2023

Table 2. Chemical analysis of the aerobically fermented bovine biofertilizer

N	P	K	Ca	Mg	Fe	Cu	Zn	Mn
(g dm^{-3})					(mg dm^{-3})			
0.82	1.4	1.0	2.5	0.75	141.6	1.92	68.2	14.72

fermentation process for 30 days, also receiving bone meal with 23% of P_2O_5. The results of the chemical analysis of the organic fertilizer are shown in Table 2.

The plants were fertilized based on the chemical analyses of the substrate and the bovine biofertilizer (Tables 1 and 2), and the mineral fertilization recommendation was based on the maximum chemical fertilization recommendation (Fernandes, 1993), which includes 15 kg ha^{-1} of N, 62.5 kg ha^{-1} of P_2O_5 and 50 kg ha^{-1} of K_2O. With a stand of 10,000 plants, the maximum dose per plant^{-1} in the cycle was: 1.5 g N, 6.25 g P_2O_5 and 5.0 g K_2O.

To determine the fraction of nutrients in the substrate, the density of the substrate (1.3 kg dm^{-3}) was multiplied by the volume of substrate placed in each pot (11 dm^{-3}). Subsequently, the value found (14.3 kg) was multiplied by the amounts of N, P, and K obtained from the substrate analysis (Table 2).

Mineral fertilization was carried out following the recommendation of Fernandes (1993), with 15 kg ha^{-1} N, 62.5 kg ha^{-1} P_2O_5 and 50 kg ha^{-1} K_2O. For pot fertilization purposes, a stand of 10.000 plants ha^{-1} was considered, and plants fertilized with 100% mineral received 0.75 g N, 3.125 g P_2O_5 and 2.5 g K_2O, through the sources urea, single superphosphate, and potassium chloride, respectively, as the study went through the growth phase, with N being applied all as basal dose, and P and K being applied 50% as basal dose and 50% in top dressing (20 days after sowing – DAS).

For phosphate fertilization, the proportions were according to the treatments: F1 (0%), F2 (50%), and F3 (100%) of the recommended dose of phosphorus, with application of 0, 3.12, and 6.25 g cycle^{-1} plant^{-1} of single superphosphate, respectively. For organic fertilization, F4 (100%), 6.0 L plant^{-1} of bovine biofertilizer was applied via soil. The fertilizers were applied manually.

To prepare the water with electrical conductivity of 4.0 dS m^{-1}, the soluble salts NaCl, $CaCl_2.2H_2O$, and $MgCl_2.6H_2O$ were used in an equivalent ratio of 7:2:1 (Medeiros, 1992) between Na, Ca, and Mg, respectively, following the relationship between ECw and its molar concentration ($mmol_c$ L^{-1} = EC x 10).

Irrigation with water of higher electrical conductivity began at 10 DAS and continued until harvest. Irrigation was carried out on a daily basis and calculated according to the drainage lysimeter principle (Bernardo et al., 2019), in plants grown in an extra pot under conditions similar to those of the treatments, keeping the soil at field capacity, according to Eq. 1:

$$VI = \frac{(Vp - Vd)}{(1 - LF)} \quad (1)$$

where:

VI - volume of water to be applied in the irrigation, mL;
Vp - volume of water applied in the previous irrigation, mL;
Vd - volume of water drained, mL; and,
LF - leaching fraction of 0.15.

The following variables were analyzed at 45 DAS: number of leaves (NL) – determined by direct counting of the leaves; plant height (PH) - obtained with a measuring tape graduated in centimeters; stem diameter (SD) - obtained with a digital caliper, measured at the basal diameter of the plant stem at a height of approximately 2 cm from the soil surface; and leaf area (LA) - estimated by the non-destructive method (length versus width of leaves) and subsequently multiplied by the correction factor (0.875) proposed by Ribeiro et al. (2022a); photosynthesis (A, μmol CO_2 m^{-2} s^{-1}), transpiration (E, mmol H_2O m^{-2} s^{-1}), stomatal conductance (gs, mol H_2O m^{-2} s^{-1}), and internal CO_2 concentration (Ci, μmol CO_2 mol^{-1} air) using the Infrared Gas Analyzer (IRGA - LI 6400 XT, LICOR).

The gas exchange measurements were made in an open system with an airflow of 300 mL min^{-1} between 08:00 and 10:00 a.m. under natural conditions of air temperature and CO_2 concentration. This data was used to quantify instantaneous water use efficiency (A/E= WUE). The relative chlorophyll index (RCI) was measured on the same leaves using the non-destructive method with a portable meter (SPAD - 502 Plus, Minolta, Japan).

To assess the normality of the data, the variables were subjected to the Kolmogorov-Smirnov test ($p \leq 0.05$). The data were then subjected to analysis of variance, and the Tukey's test for comparing means ($p \leq 0.05$) was carried out using the software Assistat 7.7 (Silva & Azevedo, 2016).

Results and Discussion

The analysis of variance (Table 3) shows that transpiration and photosynthesis were influenced by the interaction between the forms of fertilization (FF) and the electrical conductivity of irrigation water (ECw). As for stomatal conductance, instantaneous water use efficiency, chlorophyll index and internal CO_2 concentration, there was an interaction between the factors FF, ECw, and inoculation (I). As for leaf area, there was an individual effect of FF. Stem diameter and number of leaves were influenced by the interaction between FF and ECw. As for plant height, there was an interaction between the factors FF, ECw, and I.

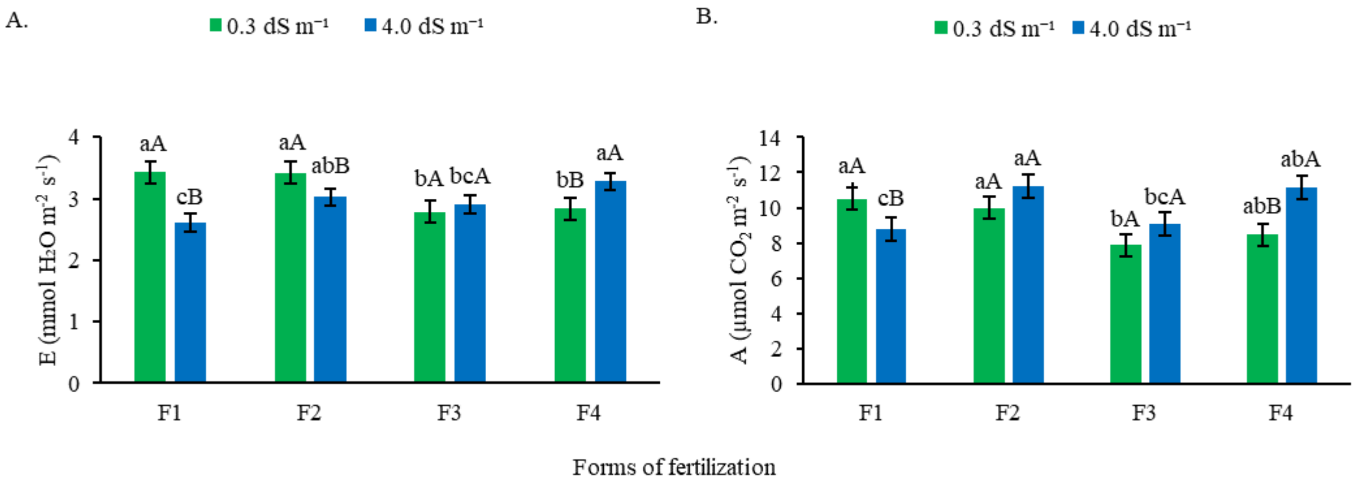
The lowest transpiration rates (E) were observed in treatments with F1 and F2 fertilizer methods irrigated with water with higher salinity, while in F3 there was no significant difference between the waters and in F4 the water of higher salinity was superior (Figure 2A). This reduction may be associated with a response mechanism to retain water, thus maintaining the water potential for easy absorption and regulating the amount of water absorbed as a response mechanism to minimize water loss (Rodrigues et al., 2022). Similarly, Barbosa et al. (2022) found a reduction in the transpiration of the peanut crop fertilized with NPK under salt stress.

However, the form of fertilization with bovine biofertilizer (F4) led to higher transpiration rates under salt stress (Figure 2A). This organic source may have contributed to

Table 3. Summary of the analysis of variance for transpiration (E), stomatal conductance (gs), photosynthesis (A), internal CO_2 concentration (Ci), relative chlorophyll index (RCI), instantaneous water use efficiency (WUE), number of leaves (NL), plant height (PH), stem diameter (SD), and leaf area (LA) of peanut plants cultivated under different forms of fertilization, electrical conductivity of irrigation water and with and without inoculation with Bacillus bacteria, 45 days after sowing

SV	DF	Mean squares									
		E	gs	A	Ci	RCI	WUE	NL	PH	SD	LA
Forms of fertilization (FF)	3	0.48^{**}	3.43^{ns}	15.34^{**}	217.44^{ns}	5.19^{ns}	0.35^{ns}	125.70^{*}	120.98^{*}	3.43^{ns}	32.59^{**}
Inoculant (I)	1	0.21^{ns}	3.46^{ns}	11.45^{ns}	6.05^{ns}	1.37^{*}	0.62^{ns}	14.45^{ns}	20.25^{ns}	3.46^{ns}	8.10^{ns}
Electrical conductivity of water (ECw)	1	0.54^{*}	13.58^{*}	14.23^{*}	775.01^{*}	138.07^{ns}	6.43^{**}	101.25^{*}	501.75^{*}	13.58^{ns}	18.19^{ns}
FF x I	3	0.06^{ns}	1.91^{ns}	8.08^{ns}	40.10^{ns}	5.84^{ns}	1.00^{*}	21.81^{ns}	48.45^{*}	1.91^{ns}	4.45^{ns}
FF x ECw	3	1.54^{**}	31.22^{**}	17.24^{**}	114.3^{ns}	1.71^{ns}	0.53^{ns}	33.68^{*}	52.29^{*}	31.22^{*}	1.93^{ns}
I x ECw	1	0.07^{ns}	29.07^{**}	18.31^{ns}	12.01^{ns}	4.56^{ns}	0.002^{ns}	80.00^{ns}	48.90^{ns}	29.07^{ns}	0.84^{ns}
FF x I x ECw	3	0.05^{ns}	9.97^{**}	1.39^{ns}	480.85^{*}	24.32^{**}	0.93^{*}	11.43^{ns}	50.68^{*}	9.97^{ns}	4.50^{ns}
Treatments	15	0.48^{**}	12.38^{**}	11.34^{**}	223.41^{ns}	17.01^{ns}	1.03^{**}	51.57^{**}	92.54^{**}	12.38^{ns}	10.50^{ns}
Residual	64	0.08	1.97	3.19	130.44	6.42	0.27	11.08	15.89	1.97	3.77
CV%	-	9.70	45.15	18.59	3.02	6.65	16.49	11.72	9.37	7.45	15.60

SV - Source of variation; DF - Degree of freedom; CV (%) - Coefficient of variation; *Significant at $p \leq 0.05$ by F test; **Significant at $p \leq 0.01$ by F test; ^{ns}Not significant



Same uppercase letters comparing the average values of ECw in each form of fertilization and same lowercase letters comparing the average values of the different forms of fertilization at the same level of ECw do not differ statistically from each other by Tukey's test ($p \leq 0.05$); vertical bars represent standard error (n=4)

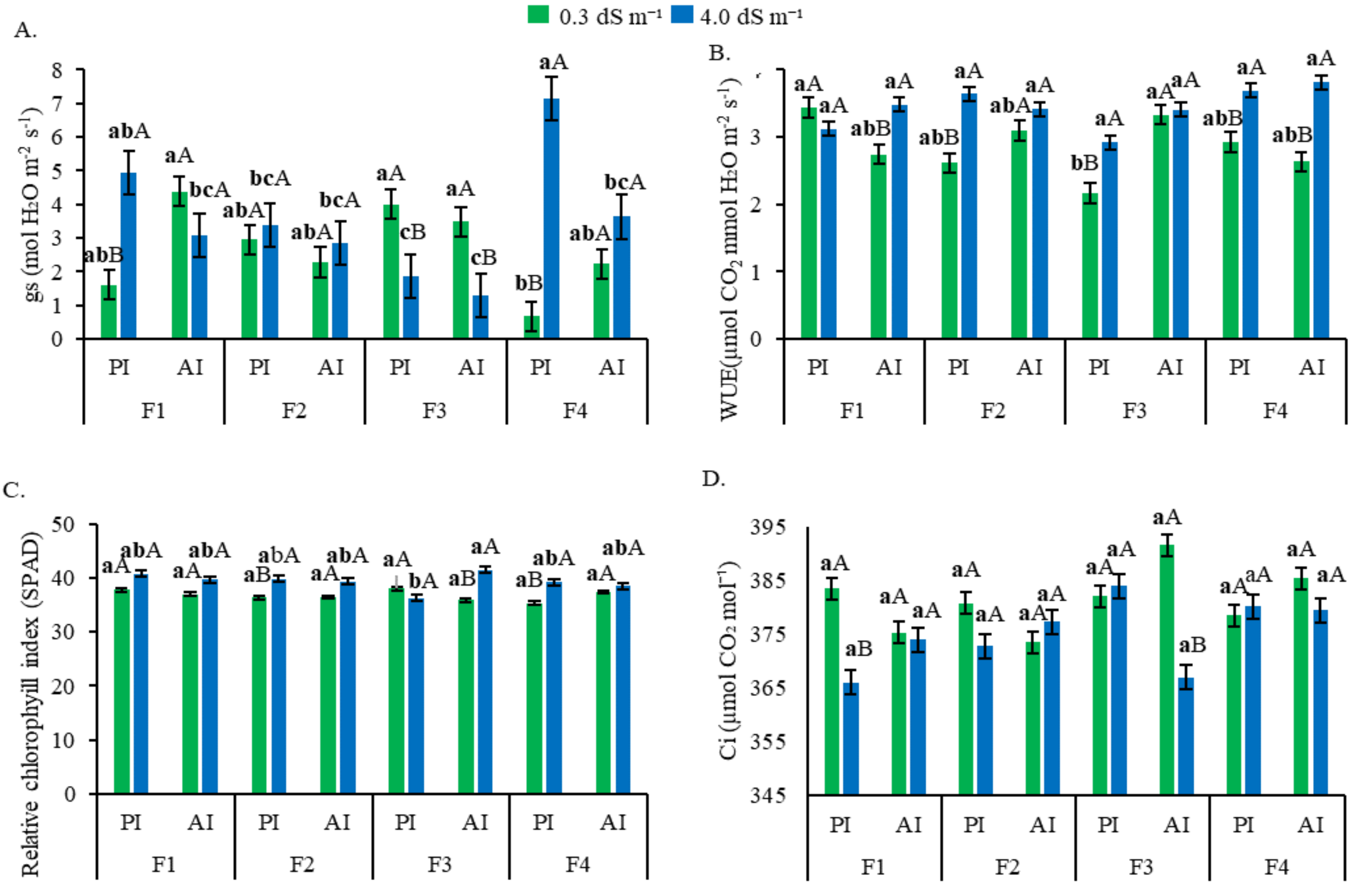
Figure 2. Transpiration - E (A) and photosynthesis - A (B) of peanut plants under different forms of fertilization (F1 - 0% phosphorus, F2 - 50% phosphorus, F3 - 100% phosphorus and F4 - bovine biofertilizer) and different levels of electrical conductivity of the irrigation water (ECw - 0.3 and 4.0 dS m^{-1})

increasing the K^+/Na^+ and Ca^{2+}/Na^+ ratios and consequently reducing the effect of salt stress, as the reduction of Na^+ in plants reduces nutritional imbalance and ionic toxicity. The greater accumulation of K^+ at the expense of Na^+ is important in stomatal regulation, and at adequate concentrations it promotes greater stomatal opening, allowing greater entry of CO_2 into the cells and consequently a higher photosynthetic rate (Sousa et al., 2018). Similarly, Souza et al. (2019) found that lima bean (*Phaseolus lunatus* L.) transpiration was higher in the presence of bovine and goat biofertilizer compared to the treatment without the organic input. The biofertilizer may have contributed to higher transpiration rates resulting from the stimulation of the action of proteins and organic solutes, leading to better nutritional conditions of the soil, and consequently allowing the maintenance of water absorption and cellular turgidity of the plant (Sousa et al., 2021b; Taiz et al., 2017).

The photosynthetic rate (A) was lower in the treatment without phosphorus (F1) associated with salt stress (Figure 2B). The presence of salts in the soil solution causes partial closure of the stomata, affecting plant metabolism and causing physiological changes with the absorption of toxic ions such as Na^+ and Cl^-, which, at high concentrations, accumulate in the chloroplasts of the cells and directly affect plant photosynthesis (Taiz et al., 2017).

The absence of phosphate fertilization (F1) together with salt stress caused a reduction in photosynthesis. In other words, the lack of phosphorus possibly reduced the absorption of magnesium, as it has a synergistic effect with this nutrient and consequently negatively affected photosynthesis, as reported by Ribeiro et al. (2022b). A study similar to this one was carried out by Barbosa et al. (2022) on the peanut crop with 100% phosphate fertilization. These same authors found that salt stress negatively affected the photosynthetic rate.

In treatment F4, salt stress in the presence of bovine biofertilizer as an organic source led to higher stomatal conductance (gs) values compared to plants that were not under salt stress. Possibly this organic input enabled the release of humic substances, reducing the deleterious effects of salts, enabling greater nutritional balance for plants and, as a consequence, improving photosynthetic activity and the production of photoassimilates (Sousa et al., 2018). The highest stomatal conductance values were found in inoculated plants when irrigated with higher ECw water (Figure 3A).



Same lowercase letters comparing the forms of fertilization with and without inoculation within the same ECw and same uppercase letters for the different ECw within each form of fertilization do not differ statistically from each other by Tukey's test ($p \leq 0.05$); vertical bars represent standard error (n=4)

Figure 3. Stomatal conductance - gs (A), instantaneous water use efficiency - WUE (B), relative chlorophyll index - SPAD (C), and internal CO_2 concentration - Ci (D) of peanut plants under different forms of fertilization (F1 - 0% phosphorus, F2 - 50% phosphorus, F3 - 100% phosphorus, and F4 - bovine biofertilizer), in presence and absence of the inoculant (PI and AI) and different levels of electrical conductivity of the irrigation water (ECw - 0.3 and 4.0 dS m^{-1})

For F2 there was no difference between the salt levels, in the presence and absence of the inoculant. In F3, the water with the lower salinity was superior to the water with the higher salinity. It can also be seen that inoculation favored greater stomatal opening in plants under stress in the absence of phosphate fertilization. This response may be associated with the ability of *Bacillus* to regulate factors involved in stomatal limitation, especially when plants are under nutritional and salt stress, as greater stomatal opening occurs with the increased demand for water and CO_2 (Akram et al., 2018). In the study conducted by Valle-Romero et al. (2023) with strawberry cultivation (*Fragaria x ananassa*), up to 60 days after the start of treatments, lower stomatal conductance values were reported for plants that were not inoculated and did not receive phosphate fertilizer. However, the authors found no difference between plants inoculated in the absence of phosphate fertilization and non-inoculated plants, regardless of fertilization after 80 days of treatment. For these same authors, the lower conductance values in non-inoculated and unfertilized plants may be associated with increased CO_2 limitation. Phosphorus limitation affects photosynthesis as it is necessary for ATP biosynthesis, thus being involved in stomatal physiology. Murguse et al. (2020) detected greater stomatal apertures in melon (*Cucumis melo* L.) plants inoculated with phosphate-solubilizing bacteria compared to non-inoculated plants.

In addition, fertilization with bovine biofertilizer (F4) may have generated a better nutritional balance with greater accumulation of K^+ in the guard cells and contributed to the transport of water within the cells, increasing cell turgidity and resulting in greater opening of the stomata (Taiz et al., 2017). Souza et al. (2019), when evaluating salt stress and the use of biofertilizer from bovine and caprine sources in lima bean cultivation, obtained better stomatal conductance in the presence of organic biofertilizers.

The instantaneous water use efficiency (WUE) in peanut plants, except for F1, those with more forms of fertilization that received inoculation, was higher under conditions of salt stress (Figure 3B). High rates of water use efficiency can reduce salt absorption and alleviate water deficit caused by salinity (Sousa et al., 2022).

Plants inoculated with microorganisms under conditions of salt stress can adapt through the production of phytohormones which, in addition to helping to resist permanent salt stress, contribute to the balance of physiological adaptations. This result is not similar to that observed by Valle-Romero et al. (2023), who obtained higher water use efficiency values in inoculated strawberry plants that received phosphate fertilization. According to the authors, phosphorus is involved in fundamental processes such as carbon assimilation and energy transfer, thus favoring the stages of carbon assimilation and energy transfer efficiency by plants and consequently contributing to water use efficiency.

Sousa et al. (2021b), evaluating the instantaneous water use efficiency in peanut plants under salt stress and forms of organomineral fertilization, found higher WUE averages when plants were irrigated with water of 5.0 dS m^{-1} at 40 DAS. With the reduction in water availability caused by changes in the osmotic potential of the soil, osmotic adjustment was maintained as a strategy to guarantee water absorption and maintain cell turgidity (Sousa et al., 2021a).

The relative chlorophyll index in peanut plants showed a reduction under irrigation with lower salinity water in treatments F2 and F4 with inoculation and in treatment F3 in the absence of inoculant (Figure 3C). In environments under stress caused by excess salts, Abdelmoteleb & Gonzalez-Mendoza (2020) observed that inoculation with *Bacillus megaterium* and *Bacillus cereus* in bean plants mitigated salt stress and increased chlorophyll index.

El-Esawi et al. (2018) reported a similar trend to that found in this study, where inoculation of soybean (*Glycine max* L.) plants with *Bacillus firmus* mitigated the effects of salinity on the plants by promoting chlorophyll biosynthetic pathways. For the authors, inoculation may have contributed to nutrient absorption and induced pigment biosynthesis, mitigating the effect of stress on the photosynthetic machinery.

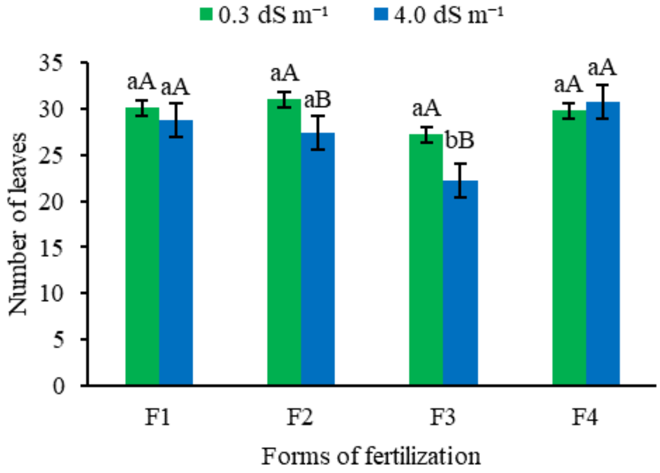
The internal CO_2 concentration (Figure 3D) was lower in peanut plants irrigated with higher salinity water and fertilized in F1 and F4, indicating the detrimental effect of salts on physiological and biochemical processes in plants (Zafar et al., 2018). For F2 there was no significant difference, and in F3 the water with lower salinity was superior to the one with higher salinity.

Under conditions of exposure to high concentrations of salts, microorganisms can act by modulating genetic expressions to maintain ionic homeostasis, helping to increase the level of K^+ and maintain the K^+/Na^+ ratio by removing excess Na^+ in plants under stress (Abdel Latef et al., 2020). The mitigating effect of salt stress on plants inoculated in F2, F3, and F4 fertilization forms may be associated with this mechanism.

The reduction in internal CO_2 concentration is a consequence of stomatal closure and is one of the main mechanisms in reducing the rate of CO_2 assimilation. The greater stomatal opening favors the entry of CO_2 from the atmosphere into the leaf mesophyll, and in case of high stress there are limitations in the entry of CO_2 (Taiz et al., 2017).

Barbosa et al. (2022) also observed a reduction in the internal concentration of CO_2 as the concentration of salts in the irrigation water increased in peanut plants fertilized with 100% of the recommended dose of phosphorus. In a similar way, Sousa et al. (2021b) also obtained a reduction in the internal concentration of CO_2 in peanut plants irrigated with higher salinity water and fertilized with bovine biofertilizer.

Figure 4 shows that the increase in salt concentration in the irrigation water reduced the number of leaves for fertilization methods F2 and F3. Under conditions of high salt concentrations, plants are capable of altering their morphological and anatomical characteristics, for instance reducing leaf production, causing

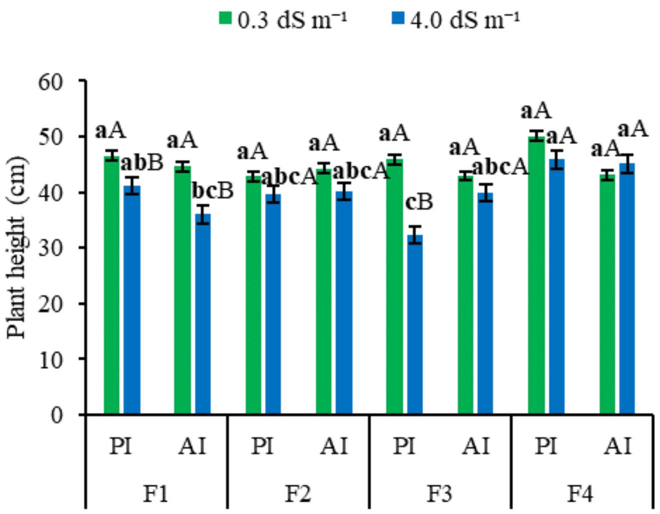


Same uppercase letters comparing the average values of ECw in each form of fertilization and same lowercase letters comparing the average values of the different forms of fertilization at the same level of ECw do not differ statistically from each other by Tukey's test ($p \leq 0.05$); vertical bars represent standard error (n=4)

Figure 4. Number of leaves of peanut plants under different forms of fertilization (F1 - 0% phosphorus, F2 - 50% phosphorus, F3 - 100% phosphorus and F4 - bovine biofertilizer) and different levels of electrical conductivity of the irrigation water (ECw -0.3 and 4.0 dS m^{-1})

stomatal closure and a consequent reduction in CO_2 availability (Sousa et al., 2021a). Silva et al. (2022) obtained a reduction in the number of leaves of peanut plants irrigated with water of higher electrical conductivity. Barbosa et al. (2022) found a reduction in the number of peanut leaves as the concentration of salts in the irrigation water increased.

The height of the plants (Figure 5) under the F1 and F3 fertilization forms, in the presence of the inoculant, showed a difference between the electrical conductivity levels of the water, and water with lower salinity was superior to water with high salinity. For the water with lower electrical



Same lowercase letters comparing the forms of fertilization with and without inoculation within the same ECw and same uppercase letters for the different ECw within each form of fertilization do not differ statistically by Tukey's test ($p \leq 0.05$); vertical bars represent standard error (n=4)

Figure 5. Plant height of peanut plants under different forms of fertilization (F1 - 0% phosphorus, F2 - 50% phosphorus, F3 - 100% phosphorus and F4 - bovine biofertilizer), in presence (PI) and absence (AI) of the inoculant and different levels of electrical conductivity of the irrigation water (ECw - 0.3 and 4.0 dS m^{-1})

conductivity, there were no differences between fertilization and inoculation.

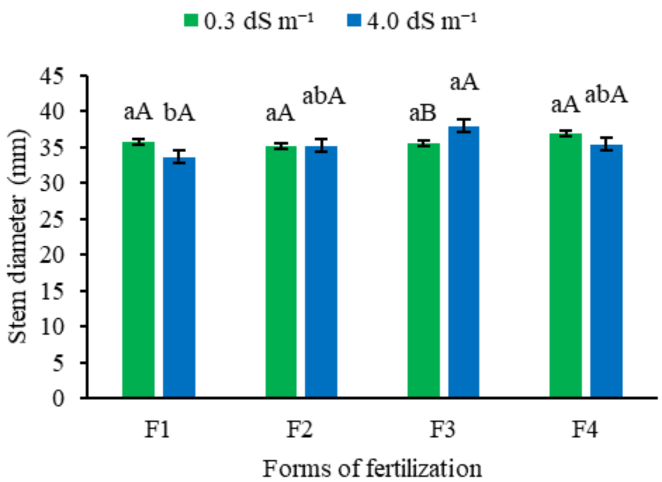
The accumulation of salts in the root zone causes a reduction in water absorption by the plant due to the osmotic effect, affecting the processes of cell multiplication and expansion and, as a consequence, causing a reduction in plant height (Sá et al. 2018). A similar result was observed by Ribeiro et al. (2022b) in a study with cowpea and saline water.

The effect of the microorganism as a mitigant of salt stress is due to its important role in the excretion of phytohormones, since plants under these conditions may not produce sufficient quantities and act by increasing the accumulation of proline and indole-3-acetic acid (IAA, Hidri et al., 2019). Regarding the action of the organic source, Sousa et al. (2018), in soybean plants, obtained a reduction in the height of plants subjected to salt stress and fertilized with bovine biofertilizer.

For stem diameter, there was no difference between the levels of electrical conductivity of the water in the F1, F2, and F4 fertilization forms (Figure 6). However, when considering water of 4 dS m^{-1}, fertilization with 100% of the phosphorus dose (F3) led to greater stem diameter.

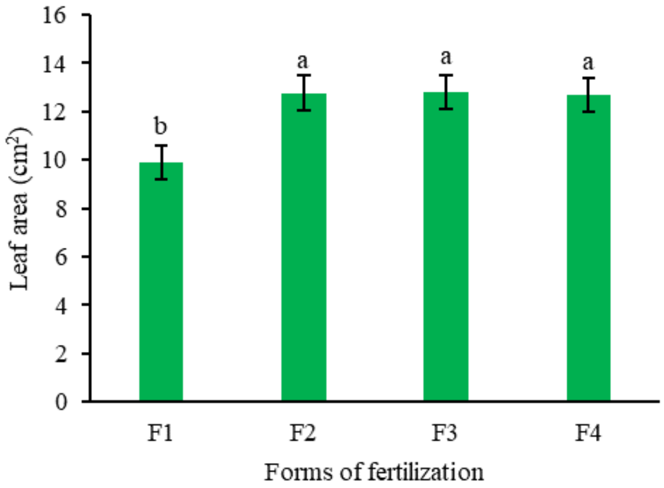
Phosphorus acts in the transfer of energy in the cell in the form of ATP and participates in processes such as respiration and photosynthesis, besides being directly related to plant growth (Taiz et al., 2017). Under conditions of reduced P availability, there is direct interference in plant development, as reported in the study of Ribeiro et al. (2022b), who observed a reduction in stem diameter in cowpea plants under stress when fertilized with 50% of the P dose.

Silva et al. (2022) observed that peanut plants which received bovine biofertilizer did not have a reduction in stem diameter when irrigated with water of higher electrical conductivity. In this study, the presence of bovine biofertilizer was also able to attenuate salt stress.



Same uppercase letters comparing the average values of ECw in each form of fertilization and same lowercase letters comparing the average values of the different forms of fertilization at the same level of ECw do not differ statistically from each other by Tukey's test ($p \leq 0.05$); vertical bars represent standard error (n=4)

Figure 6. Stem diameter of peanut plants under different forms of fertilization (F1 - 0% phosphorus, F2 - 50% phosphorus, F3 - 100% phosphorus and F4 - bovine biofertilizer) and different levels of electrical conductivity of the irrigation water (ECw - 0.3 and 4.0 dS m^{-1})



Same lowercase letters compare the average values between forms of fertilization using the Tukey test ($p<0.05$); vertical bars represent standard error (n=4)

Figure 7. Leaf area of peanut plants subjected to different forms of fertilization (F1 - 0% phosphorus, F2 - 50% phosphorus, F3 - 100% phosphorus and F4 - bovine biofertilizer)

Figure 7 shows that F1 had the smallest leaf area, with a reduction of around 22% compared to the other treatments. Fertilizer treatments F2, F3 and F4 did not differ from each other. The superiority of the treatments with mineral and organic fertilization is related to the greater availability of nutrients present in the soils that were fertilized, causing an increase in leaf expansion.

Similar results were also observed by Rodrigues et al. (2022) when they studied sources of mineral and organic fertilization in sunflower (*Helianthus annuus* L.) cultivation. These authors found that fertilization with NPK and goat biofertilizer were superior to the control.

Conclusions

1. Fertilization with organic fertilizer (100% biofertilizer) and mineral fertilizer (50 and 100% phosphorus) associated with *Bacillus* sp. mitigated the damage caused by salt stress and promoted greater water use efficiency, chlorophyll index, internal CO_2 concentration, and stem diameter.
2. The control treatment (without phosphate fertilization and without salt stress) and the application of *Bacillus* sp. promoted greater performance in net photosynthesis, transpiration rate, stomatal conductance, and plant height in peanut plants.

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