Silicon foliar application attenuates the effects of water suppression on cowpea cultivars

Aplicação foliar de silício atenua os efeitos da supressão hídrica em cultivares de feijão-caupi

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ABSTRACT
Silicon and proline play important physiological, metabolic and functional roles in plants, especially under water deficit conditions. Their application can mitigate the adverse effects of stress in crops by increasing water use efficiency and antioxidant activity. The objective of this study was to evaluate silicon (Si) as attenuator of the effects of water deficit on cowpea cultivars, through physiological, biochemical and growth indicators. The experimental design was randomized blocks with four cultivars (BRS Guariba, BRS Itaim, BRS Aracê and BRS Rouxinol) and four irrigation treatments associated or not with Si application (W100 - 100% ETo; W50 - 50% ETo; W50+100Si and W50+200Si, with 100 and 200 mg L\(^{-1}\) silicon, respectively), in a 4 x 4 factorial scheme, with five replicates. Leaf water potential, proline concentration, antioxidant enzymes and growth indicators were evaluated in cowpea plants. Under water deficit conditions, all cultivars showed reductions in leaf water potentials, which compromised plant growth. However, Si applications of 200 mg L\(^{-1}\) in the cultivar BRS Guariba and 100 and 200 mg L\(^{-1}\) in the cultivar BRS Itaim minimized the effects of stress, by increasing leaf water potential and the activity of the enzyme ascorbate peroxidase, in both cultivars, besides increasing proline concentration in the former and reducing proline concentration in the latter, which ensured the maintenance of growth. Despite the no contribution to the increase in water potential, Si applications of 100 and 200 mg L\(^{-1}\) in BRS Rouxinol and BRS Aracê, respectively, reduced the deleterious effects of the stress on their growth by regulating the enzymatic metabolism and proline.

Index terms: Vigna unguiculata (L.) Walp; water deficit; antioxidant enzymes; osmotic adjustment.

RESUMO
Silício e prolina desempenham papéis fisiológicos, metabólicos e funcionais importantes nas plantas, principalmente sob déficit hídrico. Suas aplicações podem mitigar os efeitos adversos do estresse nas lavouras, aumentando a eficiência no uso da água e a atividade antioxidante. O objetivo do estudo foi avaliar o silício como atenuador dos efeitos do déficit hídrico sobre cultivares de feijão-caupi, através de indicadores fisiológicos, bioquímicos e de crescimento. O delineamento experimental foi em blocos casualizados com quatro cultivares (BRS Guariba, BRS Itaim, BRS Aracê e BRS Rouxinol) e quatro tratamentos de irrigação associados ou não à aplicação de silício (W100 - 100% da ETo; W50 - 50% da ETo; W50+Si100 e W50+Si200, com 100 e 200 mg L\(^{-1}\) de silício, respectivamente), em esquema fatorial 4x4, com cinco repetições. Avaliou-se o potencial hídrico foliar, a concentração de prolina, enzimas antioxidantes e indicadores de crescimento do feijão-caupi. Em condições de déficit hídrico, todas as cultivares apresentaram reduções nos potenciais hídricos foliares, que prejudicou o crescimento das plantas. Contudo, as aplicações de 200 mg L\(^{-1}\) de silício na cultivar BRS Guariba e de 100 e 200 mg L\(^{-1}\) na cultivar Itaim minimizaram os efeitos do estresse, através do aumento no potencial hídrico foliar e da atividade da enzima ascorbato peroxidase, em ambas as cultivares. Além do aumento da concentração de prolina na primeira e da redução na concentração de prolina na segunda, o que garantiu a manutenção do crescimento das plantas. Apesar de não contribuir com o aumento do potencial hídrico, as aplicações de 100 e 200 mg L\(^{-1}\) de silício nas cultivares BRS Rouxinol e BRS Aracê, respectivamente, reduziram os efeitos deletérios do estresse sobre o crescimento através da regulação do metabolismo enzimático e da prolina.

Termos para indexação: Vigna unguiculata (L.) Walp., déficit hídrico, enzimas antioxidantes, ajustamento osmótico.
INTRODUCTION

In the North and Northeast regions of Brazil, cowpea (*Vigna unguiculata* (L.) Walp.) constitutes one of the main components of the population diet, especially in the rural area, due to its high nutritional value (Silva et al., 2018). In these regions, it also stands out for its commercial value, representing one of the main sources of income for rural populations (Públio-Júnior et al., 2017). However, because of the low technological level used in the cultivation system in Brazil, the yield of this leguminous crop is still considerably low (400 kg ha⁻¹), mainly due to the reductions in its growth and development under water deficit (Saboya et al., 2013; Silva et al., 2019).

Water deficit is one of the most aggravating factors for yield losses and its effects vary mainly according to species, cultivar, time of exposure and edaphic factors (Carvalho et al., 2016). Under drought conditions, the loss of cell turgor causes physiological alterations, restricting cell elongation and division, reduction in leaf area and total dry matter, stomatal conductance, transpiration and leaf water potential, that cause photosynthesis disorders and affecting plant development and yield (Freitas et al., 2017). Disorders in the photosynthetic apparatus, caused by water restriction, lead to the accumulation of electrons in cell metabolism that easily bind to oxygen molecules, forming reactive oxygen species (ROS) (Campos et al., 2019). These species act as messengers of various cellular processes, including tolerance to different environmental stresses, which depend on the balance between their production and their elimination (Sharma et al., 2012; Martins et al., 2018). At high concentrations, ROS are responsible for causing oxidative stress, which may damage photosynthetic components such as chlorophylls, intensify lipid peroxidation in membranes, denature proteins, damage nucleic acids, among others (Maia et al., 2015).

However, different plant species, including cowpea, use mechanisms that contribute to the process of tolerance to water deficit. To eliminate ROS, plants have enzymatic and non-enzymatic antioxidant systems that constitute an important primary defense against free radicals. Activities of enzymes such as superoxide dismutase (SOD), catalase (CAT) and ascorbate peroxidase (APX), associated with the osmoprotective system, mediated by compatible solutes such as proline, contribute to cell homeostasis and guarantee the normal development of the crop (Lisar et al., 2012; Szabados; Savoure, 2010; Yang; Lan; Gong, 2009).

Additionally, the use of eliciting substances, such as silicon (Si), mitigates the effects of water deficit on plants and increases water use efficiency, besides promoting improvements in metabolic pathways, resulting in adaptations to environmental changes (Zhang et al., 2017b). Si deposition occurs in different parts of the plant, such as the epidermis of the aerial part, and effectively contribute to the increase in the absorption of nutrients, modification of the gas exchange mechanism, increase in the antioxidant defense system, modification of osmolytes and phytohormones, in addition to directly acting on the reduction of transpiration through its deposition in the leaf apoplast, including in legumes (Zhang et al., 2017b).

Studies on the action of Si on plant metabolism are still preliminary, but several papers report its participation in mitigating the effects of water deficit (Crusciol et al., 2013; Ferraz et al., 2014; Mccue et al., 2000; Souza et al., 2014; Teodoro et al., 2015). In cowpea, Merwad, Desoky and Rady (2018) verified that Si application improved leaf anatomical features, pigment content and increased the antioxidant activity of this crop under water deficit, suggesting that exogenous Si recovers the deleterious effects of stress.

From that, it has been increasingly necessary to identify and develop drought-tolerant cultivars that have high yields, besides evaluating the effects of attenuating agents in the mitigation of stress, facilitating their adaptation to different agroclimatic conditions. Thus, considering the importance of cowpea production in semi-arid regions and the need for improvement in water use efficiency for irrigated production systems, the objective of the present study was to characterize the effects of foliar Si application on cowpea cultivars subjected to water restriction, through their water status and biochemical and growth indicators.

MATERIAL AND METHODS

This study was carried out in the Forest Nursery, located at 07° 12’ 42.99” S latitude and 35° 54’ 36.27” W longitude, at an altitude of 521 meters, belonging to the Paraiba State University, Campus I, Campina Grande-PB, Brazil.

The experimental design was randomized blocks (RBD) distributed in four cultivars and four treatments with irrigation depths associated or not with Si application, combined in a 4 x 4 factorial scheme, with five replicates, totaling 80 parcels with one plant each. The following cultivars, provided by Embrapa Mid-North, were used: BR5 Guaria, BR5 Itaim, BR5 Aracé and BR5 Rouxinol, under the following irrigation depths: control plants without water deficit (W100 - 100% of evapotranspiration,
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ET0) and with water restriction (W50 - 50% ET0). This second factor was associated with Si applications at concentrations of 100 mg L⁻¹ (W50 + 100Si) and 200 mg L⁻¹ (W50 + 200Si), in the form of potassium silicate (K₂SiO₃).

The experiment was carried out under field conditions in 25-L pots distributed in 10 rows, each row with 8 pots, in a total of 80 experimental plots. A spacing of 1 m between rows and 0.8 m between pots was used. The soil used in the experiment, classified as Neossolo Flúvico (Entisol), of sandy loam texture, was analyzed (Table 1) and corrected according to the needs of the crop.

**Table 1: Physical and chemical characteristics of the soil used for pot filling. Campina Grande, PB, 2018.**

| Physical characteristics | Tc | Bd | Pd | Pt | ECe |
|--------------------------|----|----|----|----|-----|
| Sand Silt Clay           | 86.63 | 9.04 | 4.33 | 1.44 | 2.72 | 47.06 | 1.41 |

| Chemical characteristics | Ca²⁺ | Mg²⁺ | Na⁺ | K⁺ | S | H⁺ | Al | OM | pH |
|--------------------------|------|------|-----|----|---|----|----|-----|----|
|                           | 1.42 | 1.50 | 0.84 | 0.21 | 3.07 | 0.64 | 0.00 | 0.88 | 6.11 |

Tc – textural class; Bd (g cm⁻³) – bulk density; Pd – particle density; Pt – porosity (%); ECe (nmhos cm⁻¹) – electrical conductivity; OM (%) – organic matter.

Cowpea seeds were screened in order to eliminate those with physical damage, biological damage and/or malformation. After screening, fungicide (Captan®) was applied at dose of 0.22 g 100 g⁻¹ seeds, remaining at rest for 24 hours.

After 30 days of soil correction, six seeds were sown in each experimental unit at a standard depth of 0.03 m from soil surface. Water replacement was performed based on the reference evapotranspiration, using the SoilControl® JR-120 evaporimeter, and calculated from the pot area, phenological stage of the crop and daily evaporation. Thinning was performed 15 days after plant emergence.

Potassium silicate was used as source of silicon, containing 12% silicon and 15% potassium, applied at the phenological stage V5 using a 20-L backpack sprayer, until the product dripped from the leaves.

Crop water status was measured by taking readings of petiole water potential (Ψw) in the fourth fully expanded trifoliate leaf of the main stem. Petiole Ψw was quantified at predawn, at the R8 stage, using a Scholander-type pressure chamber, model 3005 from Soilmoisture Equipment Corp., with values expressed in MPa (Scholander et al., 1965).

Growth variables were also evaluated at the R8 stage by the absolute growth rate (AGR) and absolute expansion rate (AER) of stem diameter, according to Benincasa (2003) and Floss (2004). Total leaf area was measured on a precision analytical scale (0.0001 g). Proline concentration was determined according to the methodology of Bates, Waldren and Teare (1973), with reading at 520 nm and expressed in µmol g⁻¹ fresh matter. The activity of SOD and APX enzymes was determined according to methodologies of Beauchamp and Fridovich (1971) and Nakano and Asada (1981), expressed in EU g⁻¹ FM and mmol Asc min⁻¹ g⁻¹ FM, respectively. For proline and enzyme analysis the youngest fully expanded trifoliar was used. The material was collected in the morning, stored in ice coolers and immediately taken to the laboratory for subsequent proline and enzyme extraction. The extracted material was stored in a freezer until analysis.

The data were subjected to analysis of variance (F test, P < 0.05) and the means of both the different cultivars in each treatment and the treatments with irrigation depths associated or not with Si were compared by Tukey test (P < 0.05), using SISVAR 5.6. software.

**RESULTS AND DISCUSSION**

The mean values of water potential, determined in the petiole of cowpea plants, decreased when plants were subjected to water deficit (Figure 1). Under the W50 treatment, there were reductions of 39, 104, 74 and 53% in the water potentials of the cultivars BRS Guariba, BRS Itaim, BRS Aracê and BRS Rouxinol, respectively, compared to their respective control treatments (W100) (Figure 1). Under the stress condition, all cultivars showed similar water potentials (P > 0.05), ranging from -0.73 to -0.86 MPa.

Water potential in plants has been accepted as an excellent indicator of water stress. In the present study, water restriction affected the water potential of cowpea plants, which may cause interference in the normal functions of cowpea plants by physiological and morphological changes, mainly due to the imbalance of the oxidoreduction system, which causes losses in the developing organs during growth as observed by Dutra et al. (2017).
After Si application of 100 mg L\(^{-1}\) in cowpea plants subjected to the water deficit treatment (W50), there was an increase of 25% in the water potential only in the cultivar BRS Itaim, compared to the treatment W50 (Figure 1). However, in the cultivars BRS Guariba and BRS Rouxinol subjected to the treatment W50 + 100Si, there were reductions of approximately 37%, compared to the treatment W50 (Figure 1). With Si application of 100 mg L\(^{-1}\), the cultivar BRS Itaim had the highest mean value of water potential (-0.64 MPa) among the cultivars exposed to the same treatment.

By contrast, Si application of 200 mg L\(^{-1}\) in the treatment W50 (W50 + 200Si) increased the water potential of the cultivars BRS Guariba (17%) and BRS Itaim (25%) compared to their respective W50 treatments (Figure 1). In the cultivars BRS Aracê and BRS Rouxinol, Si application in the treatment with water deficit (W50 + 200Si) caused reductions of 16 and 25%, respectively, when compared to the W50 treatment. The application of the treatment W50 + 200Si led to the highest water potential for the cultivars BRS Guariba (-0.62 MPa) and BRS Itaim (-0.64 MPa), among the cultivars evaluated in the same treatment.

Increases of leaf water potentials in the cultivars BRS Itaim, after Si applications of 100 and 200 mg L\(^{-1}\), and BRS Guariba, after Si application of 200 mg L\(^{-1}\), suggest the effect of Si deposition in the epidermis of their leaves. The accumulation of Si forms a physical barrier that hinders the loss of water through leaf transpiration, contributing to the initial osmotic adjustment (Keller et al., 2015; Rizwan et al., 2015). In addition, silicon application may contribute to increased synthesis of osmotic adjusters such as proline. Maghsoudi et al. (2018) suggest that silicon application favors increased expression of the P5CS gene in wheat, an important proline biosynthesis pathway gene. Gong and Chen (2012) and Amin et al. (2014) reported increases in the leaf water potential of wheat and maize plants, respectively, subjected to water deficit after Si application.

When cowpea plants were subjected to water deficit (W50), there were increments of 109, 163, 170 and 160% in the proline concentrations of the cultivars BRS Guariba, BRS Itaim, BRS Aracê and BRS Rouxinol, respectively, compared to their control treatments (W100) (Figure 2A).

In general, the proline content in the leaves increased with the reduction in water availability, in an attempt to balance the osmotic potential of the cytoplasm, suggesting that the production of proline is probably a common response of plant species under water deficit conditions (Merwad; Desoky; Rady, 2018).

For Goufo et al. (2017), proline accumulation in cowpea leaves may be related to the increase of plant resistance to water deficit. For these authors, the mechanisms by which cowpea modifies its metabolism to meet the demands of various functions of resistance, when exposed to water deficit, appear to be determined by the proline synthesis pathway.

Prisco and Gomes Filho (2010) emphasized that, when a plant is exposed to stress, first there will be the processing of the information that will induce changes in plant metabolism, such as proline synthesis, which culminate in a future response, such as the maintenance of the most favorable cellular osmotic gradient. In the present study, although the water potential of plants subjected to water deficit did not increase during the evaluated moment, it should be pointed out that high levels of proline allow the maintenance of cell turgor, guaranteeing adequate functioning of the normal plant metabolism, as described by Goufo et al. (2017) studying cowpea plants under water deficit.

The cultivar BRS Guariba increased by 161% the levels of proline after Si application of 100 mg L\(^{-1}\) in plants subjected to the treatment with stress (W50 + 100Si), compared to the treatment W50 (Figure 2A). The cultivar BRS Guariba stood out in terms of proline
production among the cultivars evaluated under W50 + 100Si, reaching the mean value of 1.08 µmol g⁻¹ FM, whereas the other cultivars had mean values between 0.41 and 0.50 µmol g⁻¹ FM of proline.

By contrast, Si application of 200 mg L⁻¹ (W50 + 200Si) in plants under stress increased their levels of proline by 72 and 130% in the cultivars BRS Guariba and BRS Rouxinol, respectively, compared to their treatments W50 (Figure 2A). Under W50 + 200Si treatment, the cultivar BRS Aracê had the highest mean value among the cultivars evaluated, reaching 1.19 µmol g⁻¹ FM of proline, while the cultivars BRS Itaim (0.30 µmol g⁻¹ FM) and BRS Rouxinol (0.52 µmol g⁻¹ FM) had the lowest mean values for proline concentration in the treatment W50 + 200Si.

Increments of proline levels in the cultivars BRS Guariba, after Si application of 100 and 200 mg L⁻¹, and BRS Aracê, after Si application of 200 mg L⁻¹, suggest the important role of Si in the increase of N metabolism, since Si increases the activity of nitrate reductase, an important enzyme in the amino acid synthesis pathway (Pereira et al., 2013). Specifically in the cultivar BRS Guariba, it is possible that the increase in proline concentration, after Si application, has contributed to a more efficient osmotic adjustment, resulting in the maintenance of the plant’s water potential. In studies with potato plants (Crusciol et al., 2009), wheat (Kaya; Tuna Higgs, 2006) and pepper (Pereira et al., 2013), Si application increased proline levels when plants were subjected to water deficit.

For the activity of the antioxidant enzymes under water deficit conditions (W50), there were increases of 106% in APX activity in the cultivar BRS Guariba, 42% in SOD activity in the cultivar BRS Itaim, 40 and 63% in SOD and APX activities in the cultivar BRS Aracê, besides 29 and 112% in the SOD and APX activities in the cultivar BRS Rouxinol, respectively, compared to their treatments controls (W100) (Figure 2B and C).

**Figure 2:** Proline (A), Superoxide dismutase (B) and Ascorbate peroxidase (C) concentrations of cowpea cultivars BRS Guariba, BRS Itaim, BRS Aracê and BRS Rouxinol submitted to control (W100) treatment and water deficit (W50) plus 100 mg L⁻¹ (W50 + 100Si) and 200 mg L⁻¹ (W50 + 200Si) Silicon. Means followed by uppercase letters compare the cultivars in the same treatment and the means followed by lowercase letters compare the treatments on the same cultivar by the Tukey test ($P \leq 0.05$), n = 5.
Water deficit causes imbalance between the production of ROS and their removal by antioxidant activity, which increases the effects of oxidative stress on the plant (Dutra et al., 2017). The increase in ROS levels compromises membrane permeability because it causes proteolysis and intensifies lipid peroxidation (Zhang et al., 2017a). It is possible that in the present study the low integrity of the membranes has hampered the retention of water in the cells, which favored the reduction of the water potentials in all cultivars studied, under water deficit conditions.

Despite the low cell water potential, the action of a complex antioxidant defense system was clear in the evaluated cowpea cultivars, which includes the participation of SOD and APX enzymes in the mitigation of the harmful effects of ROS through their removal. When cowpea plants were exposed to water deficit, their metabolism was altered and, in order to counterbalance the production and accumulation of ROS, there was an increase in SOD and APX enzymes in most of the cultivars evaluated. Similar results were discussed by other authors in leguminous crops.

Si application of 100 mg L⁻¹ in the treatment W50 (W50 + 100Si) reduced SOD activity in the cultivars BRS Itaim (34%) and BRS Rouxinol (31%), compared to their W50 treatments (Figure 2B). However, the same treatment with Si caused increments in APX activity in cultivars the BRS Guariba (33%), BRS Itaim (218%) and BRS Aracê (33%), compared to their respective W50 treatments (Figure 2C).

Under Si application of 100 mg L⁻¹, the cultivar BRS Aracê had the highest mean value for SOD activity (157.35 EU g⁻¹ FM), whereas the cultivars BRS Itaim (107.47 EU g⁻¹ FM) and BRS Rouxinol (107.54 EU g⁻¹ FM) had the lowest values of SOD activity. The cultivars BRS Guariba and BRS Itaim had the highest mean values of APX activity, reaching 0.25 and 0.27 mmol of Asc min⁻¹ g⁻¹ FM, respectively, while the cultivars BRS Aracê and BRS Rouxinol had the lowest values of APX activity, 0.2 mmol of Asc min⁻¹ g⁻¹ FM, for each cultivar.

The application of the treatment W50 + 200Si did not cause significant alterations (P > 0.05) in SOD activity. However, Si application of 200 mg L⁻¹ increased APX activity by 88 to 136% in the four cultivars investigated under the W50 treatment (Figure 2C). In this treatment, the cultivar BRS Itaim had the highest mean value of SOD activity (169.37 EU g⁻¹ FM), while the mean values of activity in the other cultivars ranged between 131 and 137 EU g⁻¹ FM. The cultivars BRS Guariba and BRS Rouxinol had the highest mean values of APX activity, 0.45 and 0.46 mmol of Asc min⁻¹ g⁻¹ FM, respectively. Conversely, the cultivar BRS Itaim showed the lowest APX activity under the treatment W50 + 200Si, with a mean value of 0.16 mmol of Asc min⁻¹ g⁻¹ FM.

For Rizwan et al. (2015), Si application reduces the oxidative stress by increasing the activities of antioxidant enzymes that alleviate the damage of plant membranes, resulting in increased growth and biomass, due to the adjustment in the relative water content in the leaf. In the present study, the same justification was confirmed in the cultivars BRS Guariba, after Si application of 200 mg L⁻¹, and BRS Itaim, after Si application of 100 and 200 mg L⁻¹. According to Rizwan et al. (2015), the reduction in the effects of oxidative stress due to the increase in the activity of antioxidant enzymes, mediated by Si, depends on the cultivars and growth conditions, as also observed in the present study.

The effects of Si application on the increase of antioxidant activity in plants subjected to water deficit have also been observed in wheat (Karmollachaab et al., 2013; Tale Ahmad; Haddad, 2011), rice (Kim et al., 2014), tomato (Shi et al., 2014), cowpea (Merwad; Desoky; Rady, 2018) and other leguminous crops (Zhang et al., 2017b).

Regarding the growth indicators, it was observed that the water deficit (W50) restricted the development of all cultivars evaluated, compared to the control treatment (W100) (Figure 3). Water restriction caused reductions of 44 and 32% in the leaf area of the cultivars BRS Guariba and BRS Itaim, respectively (Figure 3A), and reduced by 68, 38, 36 and 34% the fresh matter of all cultivars evaluated, compared to the control treatments (Figure 3B). Also, the W50 treatment reduced by 47 and 60% the absolute expansion rate of the stem of BRS Guariba and BRS Rouxinol, respectively (Figure 3C), and reduced by 27, 22 and 24% the absolute growth rate of BRS Guariba, BRS Aracê and BRS Rouxinol, respectively, compared to the control treatments (W100) (Figure 3D).

During the initial growth of cowpea, water deficit is one of the main factors limiting the establishment of plants, causing disorders and malformation in the structures composing the morphological, physiological, biochemical and anatomical attributes (Merwad; Desoky; Rady, 2018). In the different cultivars evaluated, water restriction caused reduction of leaf water potential, which culminated in the inhibition of most growth indicators. Under drought
conditions, plant growth is reduced due to a decrease in cell expansion and division, associated with losses in metabolic activities, which hinder biomass production. Additionally, the supposed increase in ROS during the stress may have affected mainly the photochemical stage of photosynthesis, leading to the alteration of the growth processes (Maia et al., 2015; Melo et al., 2018).

Additionally, water restriction influences the availability and transport of nutrients from the soil to the plant, as it decreases the diffusion of nutrients and the mass flow of water-soluble substances, such as nitrate, sulfate, calcium, magnesium and silicon, which compromises the development of the plant (Selvakumar; Panneerselvam; Ganeshamurthy, 2012).

The Si applications of 100 and 200 mg L⁻¹ mitigated the effects of water deficit on the growth of most cultivars evaluated (Figure 3). After Si application of 100 mg L⁻¹, there were increments of 40% in the leaf area of BRS Guariba (Figure 3A) and 137, 26, 29 and 33% in the fresh matter of the cultivars BRS Guariba, BRS Itaim, BRS Aracê and BRS Rouxinol, respectively, when subjected to water deficit (W50 + 100Si), compared to the treatments under stress (W50) (Figure 3B). Also, the treatment W50 + 100Si led to increments of 72, 38 and 92% in the absolute expansion rates of the stem of the cultivars BRS Guariba, BRS Itaim and BRS Rouxinol (Figure 3C), while increasing the absolute growth rate of BRS Aracê and BRS Rouxinol by 33 and 41%, respectively, compared to the W50 treatments (Figure 3D).

Si application of 100 mg L⁻¹ favored the growth especially of the cultivars BRS Aracê and BRS Rouxinol, which reached the highest mean values of leaf area (182 and 217 cm²), fresh matter (12 and 11 g) and absolute growth rate (0.28 and 0.31 cm day⁻¹), respectively, compared to the other cultivars subjected to the same treatment.

![Figure 3: Leaf area (A), fresh matter (B), absolute expansion rate - AER (C) and absolute growth rate - AGR (D) of cowpea cultivars BRS Guariba, BRS Itaim, BRS Aracê and BRS Rouxinol submitted to control treatment (W100) and water deficit (W50), plus 100 mg L⁻¹ (W50 + 100Si) and 200 mg L⁻¹ (W50 + 200Si) of silicon. Means followed by uppercase letters compare the cultivars in the same treatment and the means followed by lowercase letters compare the treatments on the same cultivar by the Tukey test (P ≤ 0.05), n = 5.](image-url)

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Additionally, Si application of 200 mg L−1 in treatments with water deficit (W50 + 200Si) also increased the leaf area of the cultivars BRS Guariba (124%), BRS Aracê (37%) (Figure 3A) and the fresh matter of BRS Guariba (209%), BRS Itaim (46%), BRS Aracê (34%) and BRS Rouxinol (48%), compared to their W50 treatments (Figure 3B). The absolute growth rate increased by 47 and 66% in the cultivars BRS Guariba and BRS Aracê, respectively, compared to the W50 treatment (Figure 3D). Si application of 200 mg L−1 led to the highest mean values of leaf area (267 cm²) and absolute growth rate (0.35 cm day⁻¹) for the cultivar BRS Aracê, highest values of fresh matter for the cultivars BRS Guariba (13 g) and BRS Aracê (12 g) and BRS Rouxinol (12 g), besides the highest absolute expansion rate of the stem for BRS Itaim (0.03 cm day⁻¹).

Notably, in the present study foliar application of Si, in the different cultivars of cowpea, promoted beneficial effects on plant organism, mainly by acting in the optimization of biochemical and physiological processes, which influenced the growth indicators evaluated. According to Merwad, Desoky and Rady (2018), Si promoted improvements in leaf anatomy, guaranteeing a good translocation of assimilates and absorbed nutrients to the cells to be used in different metabolic processes, which positively resulted in vigorous growth and satisfactory yield under water deficit conditions.

Rizwan et al. (2015) affirm that there is a natural tendency for improvement in plant growth processes after Si application, due to increased levels of antioxidant enzymes, photosynthetic capacity and osmotic adjustment so that crops continue to grow and develop fully. In a study with cotton, Ferraz et al. (2014) found significant effects on plant height with foliar application of up to 94.3 mg L⁻¹ of Si. Ibrahim, Merwad and Elnaka (2018), evaluating the effect of five Si concentrations (0, 140, 280, 420 and 560 mg L⁻¹) and irrigation depths on rice, observed that Si stimulated its growth under drought conditions, increasing plant height by 38% under the application of the highest Si concentration.

Maghsoudi, Emam and Ashraf (2016) studied the attenuating effect of Si on four wheat cultivars under water deficit and reported a significant increase in leaf area after the application of 6 mM of Si. Tale Ahmed and Haddad (2011) investigated sorghum plants subjected to 200 mg L⁻¹ of potassium silicate and detected larger leaf area compared to the control. The results of these studies corroborate the findings of the present study regarding the efficiency of Si in mitigating the effects of water stress on cowpea and contribute to elect Si application as an alternative for cowpea cultivation of in semiarid environment.

In general, the cowpea cultivars showed different responses, regardless of the variable analyzed and the treatment imposed to each one of them. Rivas et al. (2016) point out that cowpea cultivars showed distinct performance under abiotic stress due to different physiological, biochemical and anatomical characteristics, which are specific to each cultivar.

**CONCLUSIONS**

Under water deficit conditions, all cowpea cultivars evaluated showed reductions in leaf water potential, which resulted in losses in most of the growth indicators evaluated. However, the increase in the activity of antioxidant enzymes and in the osmoregulating agent proline, in almost all cultivars, suggests important modifications in the metabolism of tolerance against water deficit. Application of 200 mg L⁻¹ silicon minimized the deleterious effects of water deficit on the cultivar BRS Guariba, through the increase in leaf water potential, mediated by the increase in proline concentration and activity of the enzyme ascorbate peroxidase, which culminated in the maintenance of the growth of this cultivar. The cultivar BRS Itaim positively responded to silicon applications of 100 and 200 mg L⁻¹, with increments in leaf water potential and ascorbate peroxidase activity, which alleviated the effects of stress represented and resulted in the maintenance of growth of this cultivar. Despite not contributing to the increase in water potential, silicon applications of 100 and 200 mg L⁻¹ in the cultivar BRS Rouxinol and 200 mg L⁻¹ in the cultivar BRS Aracê reduced the deleterious effects of stress on their growth, since it reduced proline concentrations and superoxide dismutase activity in the former and increased proline concentration and ascorbate peroxidase activity in the latter.

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